STUDIES ON THE PHYLLOPLANE MYCOFLORA OF OAK (*Quercus serrata* Thunb.) WITH SPECIAL REFERENCE TO THE REARING PERFORMANCE OF OAK TASAR SILK WORM (*Antheraea proylei* Jolly.) IN DIMA HASAO, DISTRICT, ASSAM.

A

Thesis submitted to

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By

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CERTIFICATE

This is to certify that Shri Ananda Krishna Gogoi has carried out this reaserch work entitled "Studies on the Phylloplane mycoflora of Oak (*Quercus serrata* **Thunb.**) with special reference to the rearing performance of Oak Tasar Silk Worm (*Antheraea proylei* **Jolly.**) in Dima Hasao, District, Assam" under my supervision and guidance. The results incorporated in his thesis are original and to the best of my knowledge, he has not submitted this thesis work or any part thereof for any degree in any institute or university. Shri Ananda Krishna Gogoi has fulfilled all the requirements as laid down by the University of Science and Technology, Meghalaya (USTM) for submission of Ph.D. Thesis. This thesis may be accepted for adjudication .

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DECLARATION

I hereby declare that this Thesis is my original work and my acknowledgements have been made all those persons who helping me during the investigation. To the best of my knowledge that this has not been submitted previously for any diploma or degree at any other University including this University or other Institutions.

(Ananda Krishna Gogoi)

DEDICATION

This work is dedicated to my parents, family members and others well wishers .

Date: (Ananada Krishna Gogoi)

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Date (Ananda Krishna Gogoi)

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ABBREVIATION

^{0}C	:	Degree Centigrade
N	:	North
E	:	East
NE	:	North East
FYM	:	Farm Yard Manure
NPK	:	Nitrogen Phosphate Potash
cm	:	Centimeter
m	:	Meter
nm	:	Nano Meter
min	:	Minutes
ml	:	Milliliter
mm	:	Millimeter
gm	:	Gram
Kg	:	Kilogram
Con	:	Concentration
etal	:	Etalia
µg	:	Microgram
DFL	:	Disease free layings
ERR	:	Effective rate of rearing
Fig	:	Figure
P.P.No	:	Photo Plate No.
Viz	:	Namely

V/V	:	Volume per volume
W/V	:	Weight per volume
ppm	:	part of per million
SR	:	Shell Ratio
S.S.P.	:	Single Super Phosphate
M.O.P.	:	Murate of Photash
RS	:	Rhizosphere
NRS	:	Non rhizosphere
RP	:	Rhizoplane
PDA	:	Potato Dextrose Agar
NBFL	:	Non Breakable Filament Length

ABSTRACT

Title of the thesis: "**Studies on the Phylloplane mycoflora of Oak** (***Quercus serrata*** **Thunb.**) **with special reference to the rearing performance of Oak Tasar Silk Worm** (***Antheraea proylei*** **Jolly.**) **in Dima Hasao**, **District**, **Assam".**

Oak (*Quercus serrata* **Thunb.**) is belong to family Fagaceae and one of the important forest forming trees in temperate Himalayas. A total 35 Oak species are reported in India, Nepal and Bhutan. 7 Oak species are used as host plant of oak tasar (temperate tasar) silkworm distributed in North Eastern to North Western Himalayan. They are *Quercus serrata, Q. griffithii, Q. acutissima* and *Lithocarpus dealbata* in North-East Himalayan and *Q.leucotrichophora, Q. floribunda* and *Q. semecarpifolia* in North-West Himalayan. Leaf of *Q.serrata* is used for Oak tasar silkworm rearing and is considered to be the primary host plant of Oak tasar silk producing silkworm *Antheraea proylei* (**Jolly.**) in the North Eastern India. In Assam 24000 hectares area are under Oak flora among them 2000 hectare area are exploitable in Karbi-Along and Dima Hasao district. There are two crops of Oak tasar silkworm i.e. Spring and Autumn crop. In the present study it has been found that a numbers of micro organism are present in the rhizoplane and phylloplane and in other habitants in the studied host or in close proximity of the host. The root exudates of the host plants were found to have stimulatory effects on the growth and sporulation of different types of micro-organisms, which is helpful in the development of the phylloplane of the host plant by providing special type of nutrition, and development of the different varieties of leaf surface micro organisms. These micro floras may be playing a very important role in supplying different types of nutrients to the growth of the plants as well as the silkworms which will be helpful for heartening the growth and development of the plant and ultimately has a great impact on the raw silk production *i.e.* Oak tasar silk. By adaption of Oak tasar culture in hilly region may also help prevention of jhum cultivation and also save the environment and encouraging economic upliftment of poor rural people residing in hilly region.

The nature and type of microbial population in the rhizoplane and phylloplane, of Oak plants particularly of economic/cash crops have received

considerable attention during last few years. Till date no serious attempt has been made to work out the microbial complexes of rhizosphere and phylloplane of Oak plants (*Q.serrata*) at Umrangso Research Extension Centre Farm, and their effect/impact on the growth and development of Oak tasar silkworm, which was not carried out till date.

Average Leaf yields of per plant and per hectare of *Q.serrata* in different treatments *i.e.* (a) Control (Without application of Farm Yard Manure and Nitrogen. Phosphate and Potash.), (b) Application of FYM and N.P.K., (c) Application of FYM during Spring and Autumn crop during 2013 and 2014 and Rearing performance of *Antheraea proylei* **Jolly.** The leaf yield per plant /per hectare of *Q.serrata* 1.53 Kg/10355.00Kg in Spring and 0.630Kg/4236.00 kg in Autumn crop when FYM and NPK. The rearing performance recorded ERR 64.5% ,SR 10.15% in Spring crop and ERR 31.6% ,SR 9.54% in Autumn crop .The Statistical analysis had been done for leaf yield per plant and effective rate rearing in the seasons during the investigation.

Different fungal species were found in the soil and rhizosphere of Oak plants at Umrangso REC Farm. They are *Alternaria alternata, Aspergillus flavus, Aspergillus niger, Aspergillus fumigatus, Curvularia* sp, *Cladosporium clodosporides, Cladosporium herbarum, Colletotrichum gloeoporiodes, Fusarium solani, Fusarium oxysporium Mucor* sp, *Penicillum* sp, *Trichoderrma harizanum,* and Sterile mycelia were isolated during the period of investigation. The increasing trend of microbial population was observed from rhizosphere of Oak seedling to mature Oak plant and also phylloplane of different status of leaves in Spring and Autumn season. The Rhizosphere microbial population was higher than rhizoplane and non-rhizosphere soil it was also observed that the fungal population was higher on mature leaves on lower surface than semi- mature and tender leaf upper surface in Autumn season. Seasonal variation of microbial population was observed in the study *i.e.* higher in Autumn season than Spring season. *Penicillum* sp was observed in Autumn season only.

Eleven fungal species were isolated and identified in the phylloplane of Oak plant (*Q.serrata)* in different ages of leaves namely tender, semi-mature and mature. *Alternaria alternata Aspergillus niger, Aspergillus fumigatus, Aspergillus flavus,*

Curvularia sp., *Mucor* sp, *Penicillum* sp, *Verticilium, Fusarium* sp, *Colletotrichum* sp, *Cladosporium* sp, during the two rearing season Spring and Autumn. Tender leaves shown lowest population on the upper surface and showed higher population on the lower surface and nos of fungal population increasing like tender < semi-mature < mature leaves. The seasonal variation chemical constitution of *Quercus serrata* leaves where plants were applied with (NPK+FYM),in Spring season moisture, crude protein, crude fibre, crude fat, ash, carbohydrates and ERR% were recorded as 68.98%, 10.28%, 6.78%, 2.34%, 1.88%, 10.80% and 64.50% respectively and during the Autumn season leaf moisture 57.23%,crude protein 5.13%, crude fibre 7.09% crude fat 1.94%, ash 1.90% carbohydrates 21.33% and ERR 31.60% were recorded. Higher leaf moisture, crude protein and ERR% has been recorded in Spring season; while in Autumn season less % of moisture and ERR% but higher % of carbohydrates were recorded in leaves in Autumn season may be impact on higher population fungal on mature leaves of *Quercus serrata*.

The studied height of the *Quercus serrata* plants were 1.50 m which will be more conducive for rearing in the both seasons (*i.e.* Spring and Autumn). Fungal species were isolated from air over *Quercus serrata* plantation during Spring and Autumn season in 2013 and 2014, at height 0.75 m and 1.50 m at Umrangso REC farm; so air samples were collected from at height of 0.75 m and 1.50 m two different heights.

In the study a total eleven fungal species were isolated and they were *Alternaria alternata. Aspergillus flavus, Aspergillus fumigatus, Aspergillus niger, Cladosporium* sp , *Colletotrichum* sp, *Curvularia* sp, *Fusarium* sp, *Mucor* sp, *Penicillum* sp and *Sterile mycelia*. But no different species were isolated from air during different seasons (Spring and Autumn) at the two status of height, climatic condition prevailing over the season influence the fungal population.

Physcio-chemical character of soil under *Quercus serrata* plantation at R.E.C. Umrangso Farm were done. In fertilized soil pH 4.51, Organic Carbon 2.23%, average Nitrogen 331.90 ppm, Phosphate 32.00 ppm and potash 75.00 ppm. But in non fertilized soil pH 4.61, was little higher than fertilized soil Organic Carbon 1.56%, which was laser then fertilized soil availability of Nitrogen 231.14 ppm which was very much less than fertilized soil. Availability of Phosphous was

observed slightly more *i.e.* 36.00 ppm but availability of Potash was observed 150.00 ppm which was almost double than fertilized soil.

Reeling parameter of *Antheraea proylei* **Jolly.** Cocoons.

1. Filament length –Length of reeled silk filament per cocoon in meters.
2. Denier filament -Weight in gm of 9000 meter of reeled filament length.
3. Recovery%-Filament (weight length/cocoon shell weight) x100.
4. Reelability-1/(Number of ends feeding /cocoon) x 100.
5. NBFL-(Filament length (Meter) x Reelability %) /100.

The reeling parameter of *A.proylei* **J.** cocoons were taken from three different treatments

A. Cocoons, harvested from the rearing where FYM and NPK not applied in Oak plants.
B. Cocoons, harvested from the rearing where applied FYM and NPK in Oak plants.
C. Cocoons harvested from the rearing where Oak plants were applied FYM only.

Average Filament length 588.92 m, Denier 6.288, Non Breakable Filament Length (NBFL) 298.60 m, Silk recovery 62.20% and Reelability 41.14% were observed from the treatment (A) cocoons. Similarly, cocoons from treatment (B) average Filament length were 662.030 m, Denier 6.148, NBFL 330.40 m, Silk recovery 70.15% and Reelability 46.27%. From cocoons treatment(C) average filament length 627.14 m, Denier 6.225, NBFL 314.30m,Silk recovery 65.130% and Reelability 43.20%. Hence reeling parameter comparative better from the treatments like this B>C>A.

The fungal population isolated from leaf, soil and air were found in the present investigation and it is indicates mycoflora present in the cyclic pattern of appearance in air, phylloplane and soil. The mycoflora population density observed was the highest in rhizosphere soil and lowest in air of *Quercus serrata* plantation.

The present investigation deal with the fungal population of rhizosphere and phylloplane of (*Quercus serrata*) Oak host of temperate tasar and their effect on the growth and development of Oak tasar silk worm (*Antheraea proylei* **Jolly.**) at Umrangso, District Dima Hasao, Assam.

INTRODUCTION

Silk is a very unique fabric, and a luxury product, signifies liberty, life, sensuality, pleasure, tenderness, warmth, purity, serenity nature and universalism. Silk is queen of textile and natural protein fibers comprising sericin and fibron used for weaving of fibers. India stands at a unique place for producing all commercially valued natural silk viz. Mulberry, Tropical Tasar, Temperate Tasar (Oak Tasar), Eri and Muga. All these five types of commercially important silks are obtained from different species of silkworms which feeds on different of food plants.

Temperate (Oak) tasar silk is produced by silkworm (*Antheraea proylei* **Jolly.)** in India and (*Antheraea pernyi* **Guerin-Meneville**) in China which feed on the leaves of various oak leaves. In India, Oak tasar culture, is a new culture; it is produced in Jammu and Kashmir, Himachal Pradesh, Uttarakhand in North -West and Assam, Arunachal Pradesh, Manipur, Meghalaya, Mizoram and Nagaland in North -Eastern sub-Himalayan Oak belt.

Antheraea proylei **Jolly.** temperate tasar silkworm feeds on *Quercus serrata, Quercus acutissima, Quercus griffithii, Lithocarpus dealbata* in North-Eastern Himalayas and *Quercus glauca, Q. leucotrichophora, Q.floribunda* and *Q.semecarpifolia* in North–Western Himalayan. Leaf of *Quercus serrata* is used for Oak tasar (temperate tasar) silkworm rearing and is considered to be the primary host plant in North- Eastern Indian states.

Table 1. Distribution and acreage of Oak species used as host plants of *A.proylei* in India.

Sl no.	States	Area under exploitable (Hac)	Exploitable OakArea (Hac)	Exploitable species of Oak tasar rearing
1	Arunachal Pradesh	12,25,000	5,000	*Quercus griffithii* 900-1800 mASL
2	Assam	24,000	2,000	*Q.serrata* 600-1200 mASL *Q.acutissima* 600-1200 mASL *Lithocarpus dealbata* 600-750 mASL
3	Manipur	40000	20000	*Q.serrata* 600-1800 mASL *Q.acutissima* 600-1800 mASL *Q.griffithii* 900-1800 mASL *Q.semiserrata* 600-900 mASL *Lithocarpus dealbata* 600-750 mASL
4	Meghalaya	23,000	500	*Q.serrata* 600-1800 mASL *L.dealbata* 600-750 mASL
5	Mizoram	15,000	5000	*Q.serrata* 600-1800 mASL *Q.acutissima* 600-1800 mASL *Q.griffithii* 900-1800 mASL
6	Nagaland	15,000 -20,000	5,000	*Q.serrata* 600-1800 mASL *Q.acutissima* 600-1800 mASL *Q.griffithii* 900-1800 mASL *Q.semiserrata* 600-900 mASL *L.dealdata* 600-750 mASL
7	Jammu and Kashmir	55,000		*Q. leucotrichophora* 1200-2000 mASL *Q.floribunda* 2100-2700 mASL *Q.semecarpifolia* 2000-3500 mASL
8	Uttara-Khand	3,05,000	500	*Q.glance* 1200-1600 mASL *Q.leucotrichophora* 1200-2000 mASL *Q.floribunda* 1900-2200 mASL *Q.semecarpifolia* 2000-3500 mASL
9	Himachal Pradesh	1,39,500	3,000	*Q.leucotrichophora* 1200-1600 mASL *Q.semecarpiflolia* 2000-3500 mASL

Source: Technical Bulletin of Regional Tasar Research Station, Central Silk Board, Ministry of Textiles, Government of India, 2007-2008.

Table 2: State-wise Oak tasar raw silk production in India (MetricTone).

Year State	2007-08	2008-09	2009-10	2010-11	2011-12	2012-13	2013-2014	2014-15
Arunachal Pradesh	0.03	0.1	0.1	0.1	0.34	-	-	-
Manipur	3.00	3.00	3.5	2.0	2.45	2.80	4.00	4.00
Mizoram	0.02	0.10	0.02	0.40	0.93	0.72	0.70	0.02
Nagaland	0.16	0.50	0.50	0.30	0.06	0.21	0.21	0.10
Jammu and Kashmir	0.01	0.50	0.10	-	-	-	-	0.02
Uttarakhand	0.50	0.50	0.10	-	-	-	0.10	0.02
Total(MT)	3.71	4.21	5.30	3.00	3.78	3.73	5.00	4.14

Source**:** Regional Tasar Research Station; Central Silk Board Ministry of Textile Govt. of India.

In Assam 24,000 hectares area are under Oak flora and exploitable area 2,000 hectares, in Karbialong and Dima Hasao District. The Oak tasar silkworm rearing conducted during March-April as Spring crop and September-October as Autumn crop.

In the year 1969 crossing between *Antheraea pernyi* from China and its male counter *Antheraea roylei* of Indian wild silk moth gave birth to fertile hybrid *Antheraea proylei* **Jolly.** The commercial rearing of silkworm *A.proylei* was established in 1973-74 in India. *Antheraea proylei* is the source of Indian Oak tasar silk, a rough, course and nubby silk usually with natural shade of beige. Origin of the Oak-based tasar silk production is documented in China at least to the Han Dynasty (206 BC–AD220). Since then, this culture is so far the exclusive monopoly of the People Republic of China. The rearing of silkworm has been rationalized and year old occupation by the Chinese people. Oak tasar industry is a new culture in India. Dr M.S. Jolly was then Director of Central Tasar Research and Training Institute, Ranchi had been awarded Central Silk Board's prestigious "Seth Baldeodas Shah" award in recognition for the discovery of a hybrid *Antheraea proylei* and development of the new field of tasar culture on Oaks in Manipur, and afterwards in other states of North East India. This activity is a kind of livelihood and provides gainful employment to several communities in rural areas in North Western and North- Eastern Himalayans states of India especially weaker section of the society, which will be help for the economic upliftment of hilly people.

More than 90-95% of Oak tasar silk and seed production is generated from the Spring crop. The seed cocoons are obtained from the rearing of Spring crop rearing is preserved for at least 8 months (May to January next year) in semi dark seed cocoons preservation halls under properly ventilated condition. During the long period preservation of seed cocoons high loss of silk moth biomass is observed. To sustain the production and also to make a profitable venture second crop (Autumn crop) rearing is indispensable for sericulturist.

The Oak tasar silkworm rearing is carried out with the dfls (Disease free layings) prepared from erratically emerged mother moths or by inducing photoperiodic treatment of the silk cocoons. *Antheraea proylei* being weak voltinism, the diapausing behaviour is easily broken by fluctuating meteorological factors and observed loss of more than 20-30% during seed cocoon preservation due to stray emergence of mother moths. Loss is also contributed due to pupal mortality during the long period of seed cocoon preservation. To compensate the loss and also to meet the demand of seed requirement for commercial/seed crop rearing of first crop (Spring crop) during the next crop. So it is very much important to

conduct second crop rearing during Autumn season. The second crop rearing is conducted on the plantations where Spring crop rearing was not done, and light pruning/clipping were done for sprouting of new foliages. Due to minimized stray emergence of moths during the seed cocoon preservation from Autumn to next Spring crop. During the period pupal mortality is low due to short period of seed cocoons preservation. The performance of Autumn to Spring season preservation with respect to grainage behaviour such as emergence, coupling, fecundity and cocoon dfls ratio are better than Spring to Spring preservation lot.

For every terrestrial ecosystem fungi are most important ecosystem for the success and health and very must essential to the sustainability of biodiversity. Fungi are present everywhere and colonize, multiply and survive habitats performing a wide variety of various activities and play a very important role in nutrient cycling and plant health development (Bridge and Spooner, 2001, Thorn, 1997). Some of fungi are known to cause of plant diseases others are known to antagonize plant pathogens, decompose plant residues, provide nutrient to plants and stimulate plant growth. Approximates 1.5 million fungal species are present on the earth of which only about 70,000 have been described till-date (Hawksworth and Rossman, 1997). The number of fungi recorded in India about 27,000 species, the largest biotic community after insects (Sarbhoy *et al.*, 1996).It is become gradually evident that a good number of fungi do not exist in nature individually, but numbers of micro-organisms are present in the rhizosphere, rhizoplane, phyllosphere, phylloplane and in other habitants in the host or in close proximity of the host.

The term “Rhizosphere” was introduced in (1904) by the L. Hiltner, (Campbell and Greaves, 1990) to denote that region of the soil which is subject to the influence of the plant root. The word rhizosphere is derived from the Greek word “rhiza” meaning root and shere meaning field of influence, rhizosphere effect indicates the overall influence of plant roots on soil micro-organisms. It is now clearly established that Greater of bacteria, fungi and actinomycetes are present in the rhizosphere soil than in non-rhizosphere soil.

The Rhizosphere as the zone of soil immediately adjacent to legume roots that support high levels bacterial activity (Morgan *et al.*, 2005, Egambardiyeva, 2006). According to Rovira (1965) rhizosphere represents a poorly defined zone of

the soil with a microbiological gradient in which maximum changes to the microflora, which is occur in soil adjacent to the root and decline with distance away from it. Snell and Dick (1971) considered that rhizosphere as "the region in the vicinity of the root in connection with the mycorrhizae", (Davennport, 1976).The term has now been broadening to include the volume of soil influenced by the root tissues colonized by micro-organisms (Pinton *et al.,* 2001).

The region of soil surrounding and including the plant root is of crucial and very importance for the plant health and nutrition (Marschner,1995).The rhizosphere is characterized by the increased microbial activity stimulated by the leakage and exudation of organic substances from the root (Graystone *et al*., 1996). Plant roots exude simple sugars, amino acids, many other compounds in the rhizosphere region which are available for the microorganisms (Campbell 1989; Klein, 1992). The Rhizosphere studies are fascinating and interesting leading to many beneficial consequences though some microbes have harmful effects. The biotic interactions in the rhizosphere are very complex and variable, and the rhizosphere study has got very special attention of microbiologists, molecular biologists and biochemists all over the world.

The relationship between plants micro-organisms are very clearly defined so far as diseases and symbiotic nature are concerned. The interactions between plants and saprophytic rhizosphere organisms are somewhat intricate. The significance of these microorganisms to the plant may range from the competition and amensalism to mutualism depending on the environmental conditions (Trolldenier,1979). Many of the micro organisms of the root zone are probably harmless commensals without any significance for the plant. The plant rhizosphere is a very dynamic environment in which many other factors may be affect the structure and species composition of the microbial communities that colonize the roots (Yang and Crowley, 2000). The Microbial communities treated with the rhizosphere vary depending on the plant species (Graystor *et al*., 1998), the soil type (Campbell *et al.*, 1997) and cultural practices (Lupwayi *et al*., 1998). The root exudates of the plants have stimulatory effects on the growth and sporulation of different types of micro-organisms, which is help on the development of the phylloplane of the host plant by providing special type of nutrition.

The micro-habitat has been recognized and has been defined as the "rhizoplane or the root surface. The term "rhizoplane" proposed by Clark in 1949, is the external surface of plant roots together with tightly adhering soil particles or debris (Sharma and Sinha, 1974). The rhizoplane is often regarded as a part of the rhizosphere (Alexandar, 1977; Lawrence 2000). As the roots grow they cast off dead cells and navigate around soil particles making the rhizoplane highly irregular. The rhizoplane is the site of water and nutrient uptake which is released of exudates into soil.

Plant parts specially leaves are exposed to dust and air currents resulting in establishment of a typical flora on their surface aid by the cuticle, waves and appendages which help in the anchorage of microorganisms. These microorganisms may die or survive or proliferate on one leaf depending on the extent of influence of the materials in leaf diffusates or exudates. Leaf diffusates or leachates have been analysed for their chemical constituents. The principal nutritive factors are amino acids, glucose, fructose and sucrose. If the catchment areas on the leaves or leaf sheaths are significantly substantial, such specialized habitats may provide niches for nitrogen fixation and secretion of substances capable of promoting the growth of plants (Rao, 2008).

The leaf surface has been term phylloplane and the zone on leave inhabited by microorganisms as phyllosphere and the inhabitants are called epiphytes (Lindow and Brandl, 2003). Epiphytic communities are dynamic and non-uniformly distributed in time and space of leaf surfaces (Thomson *et al.,* 1993; Hirano and Upper, 1991). The term "phyllosphere" was proposed independently by Last (1955) and Ruinen (1961) and subsequently by Dickinson (1965) who used the "Phylloplane" for the study of leaf surface microflora. The phylloplane studies of various plant species attracted the attention of the many different workers (Dickinson 1967, 1971, 1976; Lamb and Brown 1970; Sinha 1971; Mishra and Srivastava 1971b, 1974; Mishra and Tiwari 1976b; Sharma and Mukerjii 1976; Rajkumar and Gupta 1980; Mishra and Dickinson 1981; Breeze and Dix 1981; Cabral 1985; Sahu and Tiwari 1985, 1988; Vardavakis 1988; Adhikari 1990; Thompson *et al.*, 1993; Barua and Bora 1995; Inacio *et al.,* 2002; Osono *et al.,* 2004; Singh and Shukla 2005).

Plant parts, especially leave surface carries a heterogenous population of the microbes which grow, reproduce and multiply on the leaf surface in dynamic equilibrium with the existing micro and macro environment (Devi *et al*., 2003). The leaves constitute a very large microbial habitat, and it is also estimated that, the terrestrial leaf surface area that might be colonized by microbes is about $6.4x10^8$ Km^2 (Morris and Kinkel 2002). The Microbial populations on the leaf surface vary in size and diversity depending on the influence of several biotic and abiotic factors which affect their growth and survival (Bakker *et al.*, 2002). These factors include leaf age, external nutrients, the interactions between populations of different micro-organisms (Blakeman 1985), temperature, relative humidity, duration at leaf wetness, light intensity, wind speed and presence of air pollutants and pesticides (Dix and Webster 1995). Due to these factors, the phylloplane microhabitat is in a state of continuous flux (De Jager *et al.,* 2001). The study of qualitative and quantitative composition of epiphytic micro-organisms occurring on the leaf surface as well as the investigation of their activities consist a very important problem concerning the interrelation between plants and microorganisms. The leaf surface is well known to all that to harbor a definite microbial community by virtue of the presence of leachates (Godfrey 1976; Irvine *et al.*, 1978). The leaf leachates or exudates which are greatly influence by the quality and quantity of micro-organisms occurring on the leaf surfaces (Tukey 1971; Tyagi and Chauhan 1982). The different microbial populations in such a community interact with one another by competing for the space and nutrients by production of secondary metabolites and also antibiotics (Fokkema 1973; Hudson 1978). This is an important aspect of phylloplane is the production of self inhibitory and self stimulatory compounds as leaf exudates or by the micro-organisms Further, a large number of nitrogen fixing organisms are present on the leaves of plants growing on plants growing on nitrogen deficient soil (Pillai and Sen, 1966).The supplied with moisture from the atmosphere and nutrients in the form of leaf exudates, such organisms may be fix considerable amount of nitrogen. Many workers have studied on this aspect and are try to find out different micro-organisms which might play a very important role in the growth and nitrogen nutrition of the host plants and in turn may be increase the yield of plants (Blasco and Jordon 1976; Remacle 1977; Banarjee and Chandra 1978; Nandi and Sen 1981; 1982; Sadykov 1981; Sengupta *et al.,* 1981).

The fungal populations on the leaf surface make a very interesting study. The living leaf can act as a landing stage for fungal spores and other propagules in the air, whether they deposited by gravity, boundary layer exchange, and the impactation or water splash. Once on the leaf, unless they are washed off by rain, they derive benefit by diffusion of nutrients from leaf, and from algae and pollen grains which are present on the leaf surface. When leaves are infested with aphids, the honey dew forms an abundant source of food (Pugh and Buckley 1971). There are two group of phylloplane fungi, they are residents and casuals (Norse, 1972) The resident can multiply on the healthy leaves surface without noticeably affecting the host tissues, whereas the casuals land on the leaf surface but unable to grow (Leben 1965). The occurrence of many fungi on the aerial surface may be directly related to inoculations from the atmosphere, which in turn is related to the production of deciduous propagules elsewhere, to their successful release into the atmosphere and to their survival and dispersal in this environment. The rain may also play a very significant role in dispersal and in some instances fungi may move from plant to plant in splash droplets (Dickinson 1976).

The phylloplane studies have been concerned mostly with pathogens or non-pathogenic fungi of crops or economically important trees (e.g Dickinson 1967, 1973; Pugh and Willams 1968; Mishra and Srivastava 1971b, 1974; Bainbridge and Dickinson 1972; Godfrey 1974; Mishra and Tiwari 1976b; Mishra and Dickinson 1981; Cabral 1985; Vardavakis1988; Carries 1992; Barua and Bora 1995; De Jager *et al*., 2001; Sharma 2004; Singh and Shukla 2005; McGrath and Andrew 2006).

So far as study of the fungal population associated with of *Quercus* sp is concerned, very a little works has been reported so far. Oak plants have an added aspect to it, since, the Oak tasar silkworm *i.e.* temperate tasar (*Antheraea proylei* **Jolly.**) feed on its leaves and, the growth and development of Oak tasar silkworm largely depends on the quality of leaves they feed upon. A few reports on the study of phylloplane of *Quercus* sp may be mentioned. Adhikari and Tiwari (1991) some experimental studies of phylloplane and litter fungi of *Quercus semecarpiflolia.* Gupta and Khuble (1991) studies on decomposition of Oak leaf litter by fungi in the forest of Almora Hills. Such study has not been reported from Umrangso in Dima Hasao District of Assam. Further, a study on both rhizoplane and phylloplane myco-fungi of *Quercus serrata* plants during different Oak tasar silkworm rearing seasons

are the lacking *i.e.* on Spring and Autumn season considering the crucial role played by the micro-organisms in nutrient cycling and there by supporting plant growth, as well as the aspect that the Oak tasar silkworm (*Antheraea proylei* **Jolly.**) while feeding on the leaves of *Quercus serrata* probably consume all the micro-organisms and their metabolic by products present on the leaves. The present investigation was undertaken to study the fungal population of the rhizoplane and phylloplane associated with *Quercus serrata* plants during different seasons related to various crop seasons rearing (Spring and Autumn) crop. This study has been carried out to find out, the qualitative and quantitative nature of the fungal population of rhizosphere and phylloplane, An attempt has been made to investigate the fungal population of air and soil during different season in order to find the cyclic pattern of occurrence of fungal population over Oak (*Quercus serrata*) plantation at Umrangso in Dima-Hasao District in Assam, and has also been made to study the co-relation between the performance of the temperate tasar crops, climatic conditions and the micro-fungal association, as some fungi are known to be pathogenic to Oak plants; while others cause disease in Oak tasar silkworm. Study of some microbial secretion of Oak leaves (*Quercus serrata*) has also been estimated through biochemical assay in two different rearing seasons *i.e.* Spring and Autumn crop. It is also made effort to study reeling parameter of Oak tasar cocoons which is very important for Oak tasar industry to generated employment in rural sector.

The present effort has also been made to study the effect of rearing *Antheraea proylei* **Jolly.** its impact on, their growth and development. This may boost up the Oak tasar industry in India by evolved superior breed of Oak tasar silkworm as well as superior varieties of *Quercus serrata*.

The aim of the present investigation is to study of qualitative and quantitative nature of Air, Soil, Rhizosphere and Phylloplane myco-flora with special reference to various Fungi along with their seasonal variation from Oak Tasar food plant and their effects on the growth and development of Oak Tasar silkworm (*Antheraea proylei* **Jolly**.) in Dima Hasao district of Assam for the production of oak tasar raw silk .

- It encompasses the following objectives.
- To isolate and identify different myco-flora from Phylloplane, Rhizosphere, Rhizoplane of soil and Air from oak tasar plantation..
- To study rearing performance of oak tasar silk worm *Antheraea proylei* **Jolly.** in different seasons.
- To study biochemical constituents of *Quercus serrata* leaf in different seasons.
- To study reeling parameter of *Antheraea proylei* **Jolly**. cocoons.

REVIEW OF LITERATURE

The serieigenous insect *Antheraea proylei* **Jolly.** is crossed between *Antheraea pernyi* from China and its male counterpart *Antheraea roylei* of India in 1969 and the fertile hybrid *A.proylei* was established in 1973-74 in India as a commercial rearing which help the upliftment of rural areas in North-Eastern and North-Western in Himalayan states where the Oak plantation grow abundantly in Natural forest. *Antheraea proylei* **J.** temperate tasar silkworm feed on *Quercus serrata, Quercus acutissima, Quercus griffithii, Lithocarpus dealbata* in North-Eastern Himalayans and *Quercus leucotrichophera, Quercus floribunda, Quercus semecarpifolia* in North-Western Himalayans. Leaf of *Quercus serrata* is used for Oak tasar silkworm rearing and is considered to be the primary host plant of Oak tasar silkworm producing in North-Eastern Indian States *i.e.* Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland. The *Quercus serrata* plant is an erect, deciduous tree and attains a height of about 25-30 meters and above when full grown with a trunk up to1.5 meters diameter. The Oak plants regulated water cycle; conserve soil moisture and environmental/ecological stability of the fragile mountain ecosystem. In *Quercus serrata* foliar variations (Srivastava *et al.*,2004) like lanceolinear, lanceolate, elliptic, oblong, obovate, oblanceolate, spathulate, cuneate. Texture deep or light green, both sides glabrous or glabrous above, pubescent beneath, thick medium or thin (papery). Leaf margin, awn tipped and undulate/serrata/dentate. Leaf apex acute/obtuse/acuminate/mucronate and 10-20 pairs lateral veins. Sprouting and Flowering in February-March and fruiting in May-June, ripening in October-November, and seeds collection in the month of December. Four foliar fungal diseases caused by Rust (*Cronartium quercum*), Powdery mildew (*Phyllactinia corylea*), Sooty mould (*Chaetophorma quercifolia*), Leaf blister (*Taphrina carulescence*) (Das and Pandey, 1991, Ghose *et al.,* 1992), and four foliar fungal diseases and Leaf curl disease caused by virus (Srivastava *et al.*, 2004). The inter-specific hybridization in *Quercus* sp is very common due to open pollination. It has been realized that replacement of Oak forest with Pine in the Himalayas affects the nitrogen cycle (Singh *et al.*, 1984) and causes heavy landslides leading to heavy and damage. Pine has got less soil holding capacity and no coppicing capacity. Oak tree hold soil more strongly than pine and have more coppicing power which should be publicized in order to encourage plantation of

more Oak trees by Non Government Organization and Govt. Department like (Forest and Sericulture) *Antheraea proylei* **J.** (Temperate tasar silkworm) is biovoltine in nature Spring crop during (March-April) and Autumn crop during (September-October). Spring crop is commercial crop as well as seed crop and Autumn crop is seed crop only. Seasonal factors greatly influence the growth and development of temperate tasar silkworm. The silkworms require nutritious leaf for healthy and uniform growth of silkworm and its effect production of quality cocoons.

According to Singh and Tikko, (1989) Pandey, Rath and Goel (1989) *Q. serrata* plants were pruned in December/January for Spring crop rearing and light pruning/clipping in August for Autumn crop rearing one month before rearing. According to Das and Pandey (1991) pruning schedule to support rearing one of the important steps in the growth of Oak tasar industry, *Q.serrata* light pruning followed by fertilizer application during winter (December-January) helps to rejuvenate the plant for maximum flush of new leaves during spring season. When pruning was conducted during October-November the newly sprouted buds gave rise to thin, small and yellowish foliage, which showed signs of stunted growth, may be due to unfavorable season for the plant to grow. Therefore, pruning during October and November is not recommended. The effect of pruning on the nutritive value of the leaves of *Quercus serrata* in a significant increase in the level of soluble protein, total sugar, total free amino acids etc which gave better performance of rearing as compared to rearing of *A.proylei* on un pruned plants (Ghosh and Srivastava, 1996).

Leaf yield of *Q.serrata* per hectare in economical plantation of 1.2 X 1.2 m spacing (8000Kg without any input *i.e.* Farmyard manure and Nitrogen, Phosphous, Potash, which is increasing to 13000Kg) when applied 7000Kg FYM and N_{150Kg} P_{50Kg} K_{38Kg} per hectare.

Source**:** Regional Tasar Research Station**,** Imphal**,** Manipur (1998).

Sinha and Jolly(1971) analysed the foliage constituent of *Quercus serrata and Q.incana.* Quality of leaves may adversely affect the physiology of individual insect (Sinha *et al.,* 1986), Fenny (1970) has reported the trends in the spring leaves of *Q.robusta.* Chinese tasar silkworm, *Antheraea pernyni* on Oak during the spring season presence of lesser amount of tannins content, which is increasing in the

following seasons as a result leaves unsuitable for silk worm (Fenny,1969). The studies on foliar constituents of Oak tasar silk worm host plants were also reported Banerjee *et al.*, (1993) Ghosh *et al.*, (1995, 1996). In spring season higher crude protein% and less crude fibre % and also resulting higher % Effective Rate of Rearing Oak tasar silkworm (*Antheraea proylei* **J.**). The quality of leaves has got direct influence on the healthy growth and survival of silkworm Sinha *et al.*, (1986). Better quality of leaves greater possibilities of obtaining good cocoon harvest. Therefore, the selection of the food plants processing superior nutritive value could be utilized for the healthy development of silkworm for obtaining good crop. According to Pandey and Goel (1991) crude protein contents of young leaves were higher than old leaves in 3 Oak species were *Q.serrata* showed maximum 28.92%, *Q. semecarpifolia* 20.77% and *Q. incana* 16.47% but old leaves contained less protein contents than young leaves. Pandey(1995) observed seasonal changes in the leaf composition of *Q.serrata,* where leaf protein were 6.81% in March and 7.89% in April which were decreasing 4.74% in October and ash 2.23% in April 1.95%, as result the leaf quality of March and April month was found most suitable for rearing of *A.proylei.* A strong positive correlation was found between leaf content and larval body weight. The higher survival of Oak tasar silkworm during Spring season may be due to higher protein content of leaves during April. Leaf quality for many lepidopteron larvae is determined on protein content basis (Mattson, 1980). The Autumn crop of Oak tasar not fully success may be due to decline in protein content and excess of relative humidity. Ponnuvel *et al.,* (1996) leaf moisture percentage of *Quercus serrata* leaves decreasing from February to November (71.9% to 56.78%) and crude protein decreasing from March(10.17%) maximum and minimum in September (5.39%) and October (5.07%).In Spring crude fibre less (0.09%) in February and 5.28% in March), but in Autumn season crude fibre 6.85% in September and 7.66% in October. Carbohydrates contents of leaves observed low in Spring season and increasing in Autumn season. Rana *et al.,* (1987) studies food consumption, utilization and rate of growth of *A.proylei* feeding with *Quercus serrata* leaf. In a period of about 36 days of its larval life, the average cumulative consumption was 72.88 gm. The quality of food consumption increased with increase in age of the worm and reached its peak in the fifth instars. The assimilation and tissue growth were positively correlated to the amount of food consumed.

Sericulture and silk weaving is the part and parcel of cultural heritage of the people of NE India (Unni *et al.,* 2009). Tasar silk production is one of the major agro-based industries playing an important role in the rural economy of NE states. The Oak tasar (*Antheraea proylei* **J**.) silkworm, the larvae of which feed on leaves of Oak tree *Quercus* sp, is an important source of tasar silk, a rough, coarse and nubby silk usually with natural shade of beige (Singh and Singh 1998). The hard and compact Oak tasar cocoons can't be satisfactorily softened by boiling in plain water unlike the mulberry (*Bombyx mori* **L.**) silk cocoons (Jolly, Sonwalker and Prasad, 1979) due to presence of relatively low amounts of sericin and high amount of protein tannin complexes in the form of pro-anthocyanidins and are thus difficult to reel (Pandey, 1990) The proteolytic activity of the pineapple extract helps in partial solubilisation of the proteinaceous silk gum(sericin). Binding the silk (fibroin) stands together in silkworm cocoon. *Quercus serrata* fed Oak tasar cocoons which contain relatively more sericin, are easily cooked with alkaline method in the North-East India. The reelability of *A.proylei* cocoons was found 53.43% and tannin in the cocoon shell, less reelability (Pandey,1990).

Soaking in boiling water has been found to significantly reduce tannin content. The reduction is due to leaching out of tannins. Cooking and treatment with alkalis (NaOH, KOH, Na_2CO_3, $NaHCO_3$). Oak tasar cocoon cooking with 1% sodium carbonate solution gives good results in respect of reelability, yield per 1,000 cocoons and other technological properties. Reelability, production/8 hrs/reeler and yield of raw silk per 1000 cocoons stand at,204gm and 347 gm respectively which are at almost at par with that of Biopril-50 (Tikoo and Goel,1987). Temperate tasar cocoon were cooked using 0.5% Sodium Carbonate an 0.5% Sodium Silicate at 90ºC for 30 minutes (Rajkhowa,1998).Cocoon cooking by the standardized enzymatic procedure developed *i.e.* pressure cooking of Oak tasar cocoons for 30 minutes and soaking in pineapple extract for 12 hours at room temperature gives a very high reeling performance as compared to traditional method (Devi,*et al.,* 2012).

The fungi presence on the leaf surfaces was recognized by De Bary in 1886 and described that *Dematium pullulans* as a fungus commonly occurring under such situation. It was known that population of pathogenic microorganisms as well as non-pathogenic microorganisms may also grow on the surface of living leaves and

other aerial parts of plants. The term "Phyllosphere" was proposed by Last (1995 a) and Ruinen (1956) to describe the milieu of leaf surfaces. Dickinson (1965), Last and Deighton (1965) restricted this term to the zone near leaves and used "Phylloplane" to actual leaf surfaces. The populations of microorganisms colonizing on leaf surface form an ecology niche which can be studied either as the phylloplane or the phyllosphere (Davenport,1976). The aerial habitat colonized by microbes is term phyllosphere and the inhabitants are called epiphytes (Lindow and Brandle, 2003). Several earlier workers viz. Ruinen (1961); Last and Deighton (1965); Leben (1965); Sinha (1965); and Sharma and Mukerji (1974) had done extensive investigation on the epiphytic micro flora of living leaves. In mid 1950's the study of microorganisms of leaf surface has become a recognized field of investigation, and in 1970 it was felt to hold an international symposium on the subject and the proceedings were published in 1971 (Preece and Dickinson,1971). The increasing research being carried out in the field was reflected in a second symposium after five years (Dickinson and Preece, 1976). In July 2005, the Centre of Ecology and Hydrology, Oxford hosts the 8th International Symposium on the Microbiology of Aerial Plant surfaces at St Catherine's College (Bailey *et al.,* 2006). and its reflects the extensive progress made in the field of this study, and to mention a few of the workersof this field are, Diem (1974); Fokkema (1981); Mishra and Dickinson (1981); Sharma *et al.*,(1984); Cabral(1985); Fokkema and Van Den Heuvel (1986); Legault *et al.*,(1989); Adhikari (1990); Kinkel (1997); Andrews and Harris (2000); Yang *et al.*,(2002); De Jager *et al.*,(2001); Andrew *et al.*,(2002); Osono(2002); Lindow and Brandle (2003); Osono *et al.*, (2004).

Phylloplane mycoflora.

Last (1995a) studied seasonal incidence of sporobolomyces on leaves of different three crop viz. spring and winter sown wheat and spring sown barley. Ruinen (1961) observed that maximum population of microbes' senescent foliage. She had also observed that olingonithrophilic and nitrogen fixing bacteria as first colonizers on the leaf surface. Kerling (1964) studied the leaf surface fungi on rye and strawberry and observed that the population of *Botrytis cinera* increased rapidly as the leaves of strawberry approached to senescence. Hudson (1962) termed the active microorganisms present on the leaf as colonizers, were classified into two categories: (i) Common primary saprophytes which are also present in the air, so

referred them as field fungi viz. *Aspergillus* sp, *Penicillum* sp, (ii) Restricted primary saprophytes which are specific to the host plant. Last and Deighton (1965) reported that bacteria and yeast like organisms were more abundant on the leaves than the hyphomycetes, and members of Sporobolomycetaceae dominated on the surface of diseased leaves infested by fungi, nematodes and mites etc.

According to Dickinson (1965) distinguished three different groups of leaf fungi viz, the transient fungi present on the leaf surface including yeasts and other propagules which are capable of sporulating on leaf surface but are not isolated from washed discs. The second groups of fungi dominated by *Cladosporium herbarum* were recorded from both leaf surface washings and washed discs, and third group consisted of forms growing vegetativly on leaf surface but pycnidia were formed only on moribund leaves. Leben (1965) distinguished the epiphytic leaf microorganisms as 'casuals' and 'residents', the first one remaining inactive or developing only on organic debris fallen on the leaf from elsewhere, the later growing actively on the leaf surface and using nutrients excreted by the leaf. There were an intermediate may also exist in addition to 'casuals' and 'residents' microorganisms. Hogg and Hudson (1966) recognized three distinct patterns of fungal distribution on Birch leaves. According to them *Cladosporium herbarum* is a primary colonizer. Holoman (1967) investigated leaf surface microflora of three potato varieties where *Aureobasidium pullulans* and *Cladosporium herbarum* were the usual inhabitants. According to Dickinson (1967) forms like *Cladosporium*, *Stemphylium* and *Alternaria* were very frequently observed on the leaves of *Pisum sativum* and point out that not all the fungi recorded by moist chamber technique were phylloplane saprophytes but several species viz, *Penicillum* sp, and *Aspergillus fumigatus* constitute casual inhabitants of the phylloplane and their presence may be reflect the relative abundance of their spores in the atmosphere which was very important during the investigation.

Lamb and Brown (1970), they were also confirmed that the microflora present on leaf surface may be divided into two groups (i) the residents group which are actively growing in saprophytic form (ii) the transient species which are deposited on the surfaces of leaves as a wind borne propagules. The residents are the nature of epiphytic organisms which grow and reproduce as saprophytes on the leaf surface whereas transient organisms are present merely by chance on the surface and

active growth and reproduction do not occur in such forms. According to Mishra and Srivastava (1971 b) however, differentiated the phyllosphere and phylloplane and retained both the terms to denote in the two different regions of leaf surface as rhizosphere and rhizoplane regions of root. The recent years workers use either of the term, and they consider them more or less synonym (Dickinson, 1976). Diem (1973) reported *Cladosporium* sp to be the only important colonizer of Barley phyllosphere. Tiwari and Sahu (1987) isolated 26 fungal species belonging to sixteen genera at various stages from the leaf surface of *Brassica campestris* **L**.

The colonization of leaf surfaces by fungi presents a very interesting study with regard to substrate relationship (Pugh and Buckley, 1971). Occurance of the microbial pattern of the phyllosphere is characteristic of a particular plant species (Pillai and Sen, 1966; Garg *et al.*, 1978), the host species themselves play an important role in microbial colonization by providing different niches to the microorganisms (Dickinson, 1981; Hirano and Upper, 2000; Mercier and Lindow, 2000; Lindow and Brandl, 2003; Chmiel, 2004). The nature and size of the phyllosphere microbial community which is a reflection of the host plant leaf characteristics together with environmental fluctuations of particular area (Dickinson, 1986). The microbial communities of leaves are diverse and variations in population sizes are caused in great part by the large fluctuations in the physical and nutritional conditions characteristic of the phyllosphere (Lindow and Brandl, 2003).

The numbers of microorganism increasing with advancing age of leaf which were observed by (Ruinen, 1956; Pugh, 1958; Hudson, 1962; Kerling, 1964; Sinha, 1971; Kumar and Singh, 1981; Narula and Mehrotra, 1981; Cabral, 1985). De Jager *et al.*, 2001 reported that gradual increase of filamentous fungal and yeast densities from foliole stage, through flash and juvenile to mature leaf stage in Mango (*Magnifera indica*) phylloplane and the most common fungal genera isolated were *Cladosporium* sp and *Alternaria* sp. They also observed higher densities and diversities of bacteria and the filamentous fungi over the abaxial leaf surface.

Hirano and Upper (1991) Thompson *et al.*, (1993) observed that the epiphytic communities are dynamic, but non-uniformly distributed in time and space of leaf. The physical surface of the leaf is highly dynamic (McGrath and Andrews,

2006). The cuticle erodes progressively which can change the topography, the wettability of the surface, exudation of nutrients and retention of microbes (Mechaber *et al.*,1996; Schonerr and Baur, 1996; Schreiber *et al.*,2001; Andrew *et al.*, 2002). The non-pathogenic fungi which inhabit the phyllosphere depend upon nutrients extruded from the leaf or those deposited from the atmosphere (Belanger and Avis 2002; Inacio *et al.*, 2002). According to Lindow and Brandle (2003) availability of carbon containing nutrients on leaves is a major determinant factor of epiphytic colonization. Several studies have revealed that small amounts of nutrients can be washed from the leaves. The simple form of sugars such as glucose, fructose, and sucrose are the dominant carbon sources on the plants that have been examined and are thought to simply leach from the interior of the plant (Tukey, 1971; Mercier and Lindow, 2003). In addition to nutrient levels, for the growth and abundance of phylloplane fungi is also influenced by environmental conditions such as availability of water (moisture), ultraviolet radiation and temperature (Breeze and Dix, 1981;Newsham *et al.*,1997; Zak,2002).The microbial community dynamics are most influenced by the external factors such as micro-climate (Andrews *et al.*,1980; Jacques *et al.*,1995), anatomical features (Andrew and Kennerly, 1980; Jacques *et al.*,1995), the physical variations (Ishimaru *et al.*,1991), the environmental changes (Dickinson, 1965; Collins and Hayes, 1976; Irvine *et al.*,1978 Breeze and Dix, 1981; Thompson *et al.*,1993; Lindow and Anderson, 1996) and the use of agrochemicals (Gibbs, 1972; Andrews and Kennerly, 1978; Blakeman, 1985).

The factors like moisture, pH, temperature, and wax deposition on leaf and height affect on microbial colonization on the phylloplane (Holloway, 1971; Hallam and Juniper, 1971; Forester, 1977; Barlocher *et al.*, 1978; Dwivedi and Kumar, 1981; Merall, 1981). A few investigators have also correlated climatic factors such as atmospheric temperature, humidity, wind velocity, light and rain (Gregory, 1961; Hirst and Stedman; (1963) Kumar and Singh, 1981; Sharma *et al.*, 1984; Adhikari, 1990). The qualitative composition of the epiphytic microorganisms depends on many environmental factors especially at the beginning of spring season, as in this stage new leaves sprouting (Burlaga and Garbolensha, 2006). Pandey *et al.*, (1989); Sahu and Tiwari (1985, 1988); Tiwari and Sahu 1987, 1989, 1991); Sahu *et al.*, (1986, 1988); Thompson *et al.*, (1993). The environmental factors are the most important physical factors, which affect the total microbial population present on the

leaf surface. The microbial species composition also changes over time, and some inhabitant may impact on the other inhabitants by producing inhibitory compounds (McCormack *et al.*,1994; Andrews and Harris, 2000 Koitabashi., 2002).

Air mycoflora:

According to (Mishra and Tiwari, 1976 a) there is very close co-relation between the air spora of one locality and the leaf surface fungi of plants growing in the area. The fungal colonizers on the leaf surface which come from air or from soil. The investigation in the aerobiology in India, was started by Cunningham (1873) who reported the changes in atmospheric spore content of Calcutta prison . The atmosphere consists of the different gaseous component as well as living organisms and non-living agents which is either fly or which depended on the wind for their dispersal. Among the latter are the microscopic forms which form the air spora (Gregory, 1973). Grainer (1954) reported higher concentration of *Helminthosporium avenae* in air at lower level of infected Oat crop. According to Last (1955 b) in his study of air spora within and above mildew infected cereal crops found higher population near the ground. Gregory (1957, 1961) also observed that a direct correlation between air-spora and the micro-flora of leaf surface. Pady *et al.,* (1967) reported that many of the air borne fungal hyphae are conidiophores which are probably released by the wind current from dead leaves of crops. Pady (1971) also pointed out that the leaf is thus a most suitable site for the both saprophytic and parasitic fungi and under favorable conditions spores are produced in large numbers and released into the air. The spores of various obligate pathogens such as rusts, smuts are usually present in the large numbers in the air and they exhibit a characteristic release pattern (Gregory, 1961; 1971). And he also suggested three main ways for the arrival of spores *i.e.* (i) dry wind borne route, (ii) in rain drops and (iii) in rain splash droplets. The studies on air spora in India have been done by several workers in time to time they were Konger and Baruah (1958); Rajan *et al.*, (1952); Sreeramalu (1967, 1970); Baruah and Bora (1965); Sreeramalu and Ramalingam (1965); Subbareddy (1970); Tilak (1974); Singh and Baruah (1979):Ramalingam (1971); Rajkumar and Gupta (1976).

The many fungi present on the aerial surface may be directly related to inoculation from the atmosphere which in turn is related to the production of

deciduous propagules elsewhere (Dickinson, 1976). Studies on myco-organic component of air over crop fields are important in understanding the mode of dissemination of airborne plant pathogens and in establishing the forecasting systems for the disease control (Kamal and Singh, 1974). It is a clear understanding of the nature, periodicity and density of the fungal propagules in the air is helpful in making a forecasting regarding the presence of fungal diseases and the quantum of viable pathogenic propagules likely to cause infection (Mishra and Tiwari, 1976 a). The leaf surface releases a fungal spore which is largely contributed to the air spora of the locality. Mishra and Srivastava (1971 a), a kind of cyclic phenomenon exists between fungal spores of air, soil and plant surface and some forms, maintain their specificity in the special environment. Many researchers {Lamb and Brown (1970); Mishra and Srivastava (1971a, 1972); Sinha (1971); Burrage (1976); Mishra and Tiwari (1976a); Kumar and Gupta (1980); Dixit and Gupta (1980); Sahu and Tiwari (1988);} have contributed to the comparative studies between phylloplane and air spora of the different fields to get a clear picture in this tropic.

In the atmosphere presents a tremendous diversity of the airborne spores with a high concentration, frequently occurring from spring to fall in temperate areas of the world (Gregory, 1973; Levetin, 1995). Numbers of spores in the air usually differ depending upon the pattern of rainfall of that area, and number of conidia decrease dramatically just after rain, side by side at that time ascospores increase (Alexopoulos *et al.*,1996). The air spora constitutes both the source of fungi that colonize on the leaf surface and the sink of spores released from the leaf surface by various dispersal mechanisms (Pedgley, 1991; Kinkel, 1997; Aylor, 2002). The airborne spores impact on the leaf surfaces and may adhere due to structural or chemical features of the epidermis and the spore (Andrews and Buck, 2002). The release of spore from many fungi inhabiting the phylloplane is passive through the action of wind or rain splash; however, other spores are actively propelled into the atmosphere by the various mechanisms (Kinkel, 1997; Aylor, 2002; Levetin, 2002). Singh *et al.,* (1990) reported that predominance of *Cladosporium* during winter, *Alternaria* during summer and *Penicillum* species during autumn season. Fang *et al.,* (2005) reported that a high frequency of air borne fungi in regions with high vegetation coverage in summer season in Beijing and also mentioned that most of the airborne fungal spores came from vegetation rather than from soil. According to

Huang *et al.,* (2002) airborne fungi were higher in winter season than other seasons at municipal landfill sites in Southern Taiwan and ascribed it to the geographic characteristics of the sampling area. The microorganisms which are found on the surface of plants either as pathogens or as saprophytes also get suspended in air. The species of microorganisms which were found in air vary according to the different geographical location. The microbial flora of air of a particular area always depends on the environmental conditions of that region (Basumatary *et al.,* 2002).

Levetin and Dorsey (2006) during their study of airborne spores on the roof of a building on the University of Tulsa campus with leaf surface fungi collected from *Ulmus americana* and *Quercus palustris* trees from the same campus reported that 19% of the fungal population isolated from leaves are found in air and suggested that some leaf surface fungi are major contributors to the air spora. The most abundant taxa in air samples recorded by them are *Cladosporium* followed by *Alternaria*. Air pollution is one of the most serious problems to human health. The Fungi are among the most important natural pollutants which can be pathogenic under specific circumstance (Nourian *et al.,* 2007) . The Geographical location, climate, and short-term meteorological conditions are mainly responsible for outdoor types and levels of fungal spores (Codina *et al.,* 2008). This air-borne fungal flora, is so called opportunistic fungi, they change the spectrum of fungal diseases (Singh, 2001).

Rhizosphere and non-rhizosphere (soil) mycoflora:

The microbial association and their activity amply evidence are soil, rhizosphere, rhizoplane and phylloplane are the specialized ecological niches (Manoharachary and Mukerji, 2006). The plant roots support the growth and activities of a wide variety of microorganisms that may have a profound effect on the growth and or health of plants (Ladygina, 2005). The term “rhizosphere” was first used in 1904 by Lorenz Hiltner, a soil bacteriologist and professor of agronomy at the Technical college of Munich, (Campbell and Greaves, 1990) to describe the interaction between bacteria and legume roots. At present rhizosphere is recognized as a biologically active zone of the soil around plant roots that contains different living organisms such as soil-borne microbes including actinomycetes, bacteria, fungi, microalgae, protozoa, invertebrates (collembolans, nematodes, earthworms) in

their abiotic environment (Kennedy 1998).According to Pinton *et al.*, (2001), the term rhizosphere includes both the volume of soil influenced by the root and the root tissues colonized by micro-organisms. The Rhizosphere is a soil ecological region where soil is subjected to specific influence by plant roots due to exudates from root cells and sloughing of root tissues (Giddens and Todd, 1984; Curl and Truelove, 1986). Rhizosphere represents a poorly defined zone of soil, with a microbiological gradient, in which maximum changes in the population of microflora in soil, which is evident adjacent to root and decline with distance away from it (Newman, 1978; Bowen, 1991; Mukerji, 2002).

The Rhizoplane is more closely or narrowly defined and describes the surface of the plant root itself along with tightly adhering soil particles (Curl and Trueloved, 1986; Boltton *et al.*, 1992). The term "rhizoplane" was first proposed by Clark (1949) and cited by Sharma and Singha (1974). The root surface or the rhizoplane who support a relatively high biological activity and it reflects more sensitivity than the rhizosphere (Bruehl, 1987). The rhizosphere can be divided into several distinct zones (Lynch, 1987).These also include the endo-rhizosphere (root tissue including the endodermis and cortical layers), the rhizoplane (the root surface with the epidermis and mucilaginous polysaccharide layer), and the ecto-rhizosphere (the soil immediately adjacent to the root).

The physical, chemical, and biological properties differing of the root associated soil, compared with those of the root-free bulk soil, which are responsible for changes in microbial diversity and for increased numbers and activity of microorganisms in the rhizosphere micro-environment (Kennedy,1998). The great array of root-microbe interactions results in the development of a dynamic environment known as the rhizosphere where microbial communities also interact (Barea *et al.*,2005).

The most of earlier workers like Katzneelson, 1946; Agnihothrudu, 1955; Parkinson, 1957; Sadasivan1965; Sorenson, 1997; have reported the phenomenon of accumulation of microorganisms around the root zone. Parkinson *et al.*, (1963) investigated that colonization of roots of barley, cabbage and dwarf bean by fungi and opined that initial root colonization may be by awide range of soil fungi, but that this mixed population rapidly gave way to a stable and typical root surface mycoflora dominated by such fungi as *Fusarium* sp, *Cylindrocarpon radicicola*, *Gliocaldium* sp and *Penicillum* sp. Parkinson ad Pearson (1967) reported that a

marked decrease of total fungal population in increasing depth of soil. They however observed that the sterile dark forms of fungi increased in frequency of occurrence with increasing depth. Parkinson and Thomas (1969) reported that there is close similarity between the rhizosphere and non rhizosphere mycofloras and really marked qualitative changes in the rhizosphere mycoflora accompanying increasing the plant age.

The rhizosphere micro-floras of crop plants increase during the maturity stage have been observed by several workers (Katznelson, 1965). Mehrota and Claudius (1974) reported that a general increase in the microbial population of rhizosphere soil as compared to non rhizosphere soil. They also reported that microflora of the rhizoplane was stimulated to a lesser degree than the corresponding rhizosphere. Mall (1975) who studied the root region mycoflora of coriander and reported that the fungal species which appeared in rhizosphere of four day old plants changed with the age of the plant and in sampling after maturation of seeds, the pattern was completely different. In the rhizoplane the fungal population increased with increase in age of the plants and it was maximum during flower initiation and during flowering with decreased at seed stage but again showed manifold increase after seed maturation.

According to Subrahmanyan and Rao (1977) the peaks in rhizosphere populations of *Arachis hypogeal* **L**. when the plants were at flowering stage and again at maturity and *Penicillum* sp followed by *Aspergillus* sp has been reported as dominant groups from the rhizosphere in Kharif season, while in Rabi season, *Aspergillus flavus* and *Aspergillus* sp. were most dominant. El-Amin and Saadabi, (2007) reported that significant variation in total number of fungal colonies and percent abundance of fungal species in the rhizosphere soil of Sugarcane obtained from the various study sites and increased fungal activities with plant age. The highest number of fungi reported in the rhizosphere of *Hibiscus esculentus* each week followed by rhizoplane and lastly by the non-rhizosphere soil (Oyeviola, 2009). He also reported that *Aspergillus niger* and *Aspergillus clavatus* were predominant in both rhizosphere soil and the rhizoplane, while *Penicillum oxalicum* and *Alternaria herbarum* were predominant in rhizosphere soil only.

It is now well established that the quantitative and qualitative differences between soil and rhizosphere populations depend on the mineral nutrition of the plants and the oxygen content of the root environment (Troldenier, 1979).

According to (Grayston *et al.*, 1996), it is the loss of carbon compounds from the roots which drives the development of enhanced microbial populations in the rhizosphere when compared with the bulk soil. After germination of seeds the roots grow through the soil and the loss of organic material provides the driving force for the development of active microbial populations around the root and it is, known as the rhizosphere effect (Whipps, 1990; Morgan and Whipps, 2001). Pandey and Palni (2007) investigated on the rhizosphere effect in the trees of the Indian Central Himalayans and reported that (i) the microbial population and the corresponding R: S ratio in long duration plants (*e.g*. perennial trees) are considerably lower in comparison to short duration annual crops. (ii) The microbial population and the rhizosphere effect would appear to decrease when increasing the altitude and (iii) under cold and harsh climatic conditions of sub-alpines, the tree root exudates tend to become more acidic, and exert a negative influence on the microbial population.

The microbial communities that colonize the roots due to many factors may affect the structure and species compositions (Yang and Crowley, 2000). The fungal flora associated with plant roots is subject, either directly or indirectly, to the influence of a number of factors such as soil type, and pH and the fungal flora of the root zone changes as the plant grows with certain fungi assuming predominance (Peterson, 1961). The factors such as soil type, soil moisture, pH, temperature, plant age, relative humidity and several other factors are known to influence rhizosphere effect (Manoharachary and Mukherji, 2006; Gangopadhyay and Banerjee, 1987). Parkinson *et al.*,(1963)reported that the incidence of any fungus in a soil is dependent on the structure and past biotic history of that soil, but how the influence of soil type operates in relation to root colonization which is not clear.

The interactions between the plants and the rhizosphere organisms are governed by the properties of that habitat, such as the presence of the growing plant, soil structure, partial pressure of oxygen, water content, temperature and plant nutrients (Trolldenier, 1979). The soil factors particularly moisture, physical structure and nutrient level, which influence the amount of exudation and hence colonization of roots (Whipps and Lynch, 1986). The variety of abiotic and biotic factors shapes soil and plant associated habitats and modifies the compositions and activities of their microbial communities, which in turn bear upon the quality of their environment, the growth of plants, and the production of root exudates (Bever *et al.*,

1997). The microbial activity in the rhizosphere affects the rooting pattern and the supply of available nutrients to plants, thereby modifying the quality and quantity of root exudates (Bowen and Rovira, 1999; Barea, 2000). The plant root exudates contain simple carbon substrates, including the primary metabolites, such as sugars, amino acids, and organic acids, in addition to a diverse array of secondary metabolites that are released into the rhizosphere and surrounding soil (Jones *et al.*, 2004). The microorganisms that colonize the rhizosphere help plants to acquire Phosphate(P) and Potash (K), and some enhance Nitrogen(N) uptake from the soil by their effect on root morphology and physiology (Coking, 2003).

The root exudates influence the growth of bacteria and fungi that colonize the rhizosphere by altering the soil chemistry in the vicinity of the plant roots and by serving as selective growth substrates for soil microorganisms. The microorganisms in turn influence of the composition and quantity of various root exudates components through their effects on root cell leakage, cell metabolism, and plant nutrition (Yang and Crowley, 2000). The rhizosphere microbial communities differ between plant species (Batten *et al.*,2006; Inns *et al.,* 2004; Westover *et al.,* 1997) and between different developmental stages of a given plant (Mougel *et al.,* 2006). Rahman *et al.,* (2003) screened microbial population of *Machilus bombycina* and *Litsea polyantha* from Goalpara district, Assam, during summer and winter season. They also reported highest occurrence of *Fusarium* sp from both rhizosphere and non rhizosphere soil of *Machilus bombycina* during summer season and *Aspergillus* sp winter season. The highest occurrence of *Penicillum* sp was found by them in rhizosphere of *Litsea polyantha* while in non rhizosphere *Aspergillus* sp was the highest. According to Pandey and Palni, (2007), conifers of subtropical and temperate locations, namely *Cedrus*, *Pinus* and *Taxus* support relatively higher microbial population in the rhizosphere in comparison to non-coniferous species.

According to Jones and Darrah, (1996) plants can directly control levels of carbon within the rhizosphere and thereby control the size of the microbial population. Free living microorganisms can also enhance plant growth through the suppression of soil borne plant pathogenic microbes and deleterious soil microbes (Kloepper, 1992). The region of soil surrounding and including the plant root is of crucial importance for the plant health and nutrition (Marschner, 1995). The microbes living in the complex region of rhizosphere influence crop health and also

yield. The area of this zone depends on the soil type and the host under study and soil environmental conditions (Manoharachary and Mukerji, 2006) First of all the Rhizosphere is a unique hot spot in the soil at the viewpoint of microbial ecology as soil microorganisms are considerably stimulated in the vicinity of the roots, as a consequence of release by roots of a range of Carbon (C) compounds (Jones *et al.*, 2004). The Rhizosphere is relatively nutrient rich because 40% of the photosynthesis moving into the roots are lost to the soil in the form of soluble exudates, mucilage, and shed. It is a habitat for a vast interactive community of rhizotrophic microorganisms whose activities largely determine the physico-chemical properties of the rhizosphere soil. The microbial composition in the rhizosphere very often differs greatly from that of the surrounding soil and from one plant species to another, as a result of diverse plant microbe interactions are observed (Egambrediyeva, 2006).

The soil is a rich habitat for the growth of diverse and interacting population of the microbes, and fungi are one of the most important functional groups of soil microbes and which are critical to nutrient cycling, Transport of nutrients to plants, plant growth, and disease suppression (Christensen, 1989; Thorn, 1997). In the beginning of the study of soil fungi was made as early as 1886 when Adamtz in Germany isolated several fungi species and in course of his biochemical studies on soils (Saksena and Sarbhoy, 1964). A very wide range of soils under many different types of vegetation and from many different geographical areas have been examined for the presence of fungi by many of investigators (Saksena, 1955; Mishra, 1966; Gams and Domsch, 1969; Rama Rao, 1970; Manoharachary, 1977; Bisset and Parkinson, 1979; Behera and Mukerji, 1985; Hawksworth, 1991; Zou *et. al*, 2000; Azaz and Pekel, 2002; Manoharachary *et al.*, 2005; Rane and Gandhe, 2006; Sagar *et al.*, 2007, Shukla and Tripathi, 2007).

According to Saksena and Sarbroy (1964) observed periodicity of fungi throughout the year, maximum number of micro fungi being present during the winter and the rainy seasons during their investigation of fungi in different soils of Allahabad. They isolated 19 species of *Aspergillus* which were reported to be encountered in every season. Sundaram, (1977) while studying by three different methods, fungal flora of rice field soils, found most of the belonging to well known genera of *Aspergillus*, *Penicillum*, *Fusarium*, *Curvularia*, *Dreschlera*, *Chaetomium*. Hashem, (1993) reported twenty four species of nine different genera of fungi from

four different places and the predominant genus reported as *Aspergillus* from all soils during the investigation.

According to Zou *et al.,* (2000) for the same kind of soil, the number and species of the microorganisms in the surface layer were the, and it declined with the increase of the depth. They also observed that the number of microorganisms increases with the increase of atmospheric temperature. The seasonal variations in forest soil mycoflora was observed by Mishra,(1966),Rama Rao,(1970),Persiani *et. al*,(1998) and many workers Christensen (1981) compared 33 micro-fungal communities from several different environments and found a clear correspondence between fungal species composition and vegetation type and concluded that soil micro-fungi are remarkable indicators of environmental similarity. The differences in micro-fungal assemblages between coniferous and deciduous forest in southern Qubec was observed by Widden (1986). Rane and Gandhe (2006) isolated maximum number of fungi during winter season a minimum number during rainy season. According to Waldrop and Firestone (2006), change in microbial community composition seasonally is due in large part to soil temperature. The population and activity of microbes in soils are influenced by a variety of factors such as climate, soil fertility stats and vegetation (Ross 1987; Sarathchanda *et al.,*1988; Okano *et al.,* 1991).According to Tiwarii, Jagrati and Verma (2011) *Aspergillus niger* and *Trichoderma viride* having potential as bio-control agents of wood decay fungi. Trans,N.Ha (2010) Using *Trichoderma* species for bio-control of plant pathogens in Vietnam.

The seasonal patterns in microbial community compositon and it is not clear whether there are predictable seasonal patterns in microbial community composition. The microbial community composition from season to season are largely random and unpredictable (Bardgett, *et al.,*1999) or seasonal community dynamics are so small as to be unimportant (Waldrop and Firestone 2006). Shukla and Tripathi (2007), studied distribution of micro fungal communities in forest soil and observed that the density of fungal propagules had a close inverse relationship with the pH of the soil. The actual community composition was highly related to environmental conditions, which seems to be more important for the presence or absence of microbial species than incubation time or tree species.

MATERIALS AND METHODS

LOCATION: The experimental site of Research Extension Centre Farm is situated above 627 mASL and its lies 24.98º N Latitude and 92.83º E Longitude at Dima Hasao (North Cachar Hills) district of Assam.

Location Map of Umrangso, District. Dima Hasao, Assam

Figure 1: Map of Dima Hasao (North Cachar Hills)
Source: Satellite Map

MATERIALS AND METHODS

3(a).1 Materials

Quercus serrata 300 number of plants each of the same age (20 years), height and girth were taken at Research Extension Centre. farm of Umrangso, Dima Hasao district, Assam for Spring and Autumn season leave yield during the year 2013 and 2014 respectively.

3(a).2 Methods

3(a).2.1 Studies of leaf yield of *Q.serrata*

Pruning and pollarding of *Q. serrata* plants were done during the first week of December followed by application of Farm Yard Manure and Chemical Fertilizer for spring season and light pruning, removal of 1/3 biomass/ clipping off of leave during the second week of August for Autumn season followed by application of Farm Yard Manure and Chemical Fertilizer to rejuvenate the plant for maximum flush of new leaves.

Agronomical practices: Application of FYM 10 kg / Plant and Urea= 48.0 gm, Single Super Phosphate= 46.5 gm and Murate of potash= 9.3 gm per plant (Nitrogen : Phosphous : Potash =150:50 : 38 and Farm Yard Manure= 7000 kg/hectare) in economical plant spacing of 4´x4´ (6724 no. of plants/hectare).

Leave yield/hectare =Average leaf yield per plant (Kg) x actual no of plant.

100 plants were taken in each treatment (Spring and Autumn crop in 2013 and 2014)

Treatment A : Control
Treatment B : Application of NPK+FYM
Treatment C : Application of FYM

3(b).1 Materials

A total 100 x 3= 300 plants taken from three different treatment *i.e.*. A,B and C which mentioned in 3(a).1 and introduced of *A.proylei* silkworms 100 x10=1000 per treatment of newly hatched silkworm.

3(b).2 Methods

3(b).2.1 Rearing of *Antheraea proyeli* Jolly. during the period 2013 and 2014 in Spring and Autumn crop.

100 x10=1000 (100 silkworms were brushed in one plant) for each treatments as cellular rearing under nylon net in spring and autumn crop in 2013 and 2014.

1. Mature worm weight: Mature silk worm weight was taken just after discharge of the excreta by each male and female larvae.

2. Cocoon weight: Removal of dry leaves after 6th day of spinning by each larva. Ten numbers of silk cocoon male and female each selected randomly for cocoon analysis.

3. Shell weight: Male and female each cocoons considered for taking shell weight after removal of pupa from the cocoon.

4. Shell ratio: Shell ratio of each cocoon was calculated following the formula:

$$\text{Percentage of shell ratio (SR\%)} = \frac{\text{Shell weight}}{\text{Cocoon weight}} \times 100$$

5. Effective rate of rearing: Effective rate of rearing is a performance of assessment indicator for a siik worm rearing. It is calculated as follows:

$$\text{Effective rate of rearing (ERR\%)} = \frac{\text{Total no.of cocoon harvested}}{\text{Total no.of silkworm}} \text{ x } 100$$

6. Cocoon harvested: The total numbers of cocoons spanned for each treatment was counted as cocoon harvested.

7. Larval period: The duration of larva was calculated from the first day of feeding *i.e.*. 1st day of 1st instar till the starting day of spinning for silk cocoon.It is calculated in numbers of days.

3(c).1 Materials

Tender, semimature and mature of *Quecus serrata* leaves to be collected randomly from REC, Umrangso farm. The method of sampling of leaves as described by Kamal and Singh(1970) was followed during the collection of leaves.

3(c).2 Methods

3(c).2.1 Isolation and identification of species from phylloplane of *Q.serrata*. during spring and autumn season in 2013 and 2014.

Serial washing technique of Kamal and Singh (1970),leaf discs were cut out from different categories of leaves with the help of sharp sterilized cork borer. Pieces of different categories of leaves were placed separately in 20ml of sterilized distilled water in 250 ml of erlenmyer flasks and were shaken for 20 minutes at 120 rpm.The extract of the detachable fungal propagules from the leaf surface was determined by plating 1 ml solution from washing to the Petri plates containing PDA media. The cut-out leaf discs upper and lower surface were impringed on the surface of Petri dishes containing PDA media. The Petri dishes were incubated at 28 ± 1^{o}C for 4 days and then the plates are examined for the development of fungal colonies. The experiment was conducted in Spring and Autumn season The isolated fungi were Identified with the help of by Barnatt H.L (1960) "Illustrated genera of Imperfect fungi" and Gilman (1961) "A manual of soil fungi".

3(d).1Materials

Study samples of rhizosphere soil and air samples were collected during different Oak tasar silkworm rearing seasons.

3(d).2Methods

3(d).2.1 Isolation of Fungi from Rhizosphere, Non-rhizophere, Rhizoplane and Air Mycoflora.

Root sample with adhering soil were dug out carefully from matured plants of Oak and one year seedlings, collect in polyethylene bags and brought to the laboratory .Soil samples were collected from a depth of 0-15 cm from five different spots of the farm. The collected samples were mixed thoroughly and composite

samples obtained from each plot.The samples were then brought to the laboratory immediately in polyethylene bags and stored in refrigerator at 4° C. Five samples were collected in each case of study.

3(d).2.2 For isolation of Fungi from Rhizosphere

Serial dilution plating method (Atlas R M and Parks L C 1997: Mukherjee and Subba Rao 1982) was followed. After removal of superfluous soil, the root system along with the adhering soil (approx 1 gm) was placed in a conical flask containing 10 ml sterile distilled water shaken vigourly. Stock solution 1ml aliquot was transferred with a sterilized pipette to another flask containing 9ml sterilized distilled water and shaken vigorously to obtain a dilution of 10^{-1}, 10^{-2} , $10^{-3}$and 10^{-4} were prepared.

One ml aliquot from dilution of 10^{-3} and 10^{-4} was transferred a septically into sterile Petri-plates with 10^{-12} ml of melted PDA media (15 mltreox). With 0.5 ml 250mg *Streptomycin* to suppress bacterial growth. Five petri plates were provided for each solution. Five replicates of petriplate were maintained for each inoculation. The whole process was carried out in a Laminar Air Flow Chamber. The petridishes were incubated upside down for 5-7 days at 28± 1º C in B.O.D incubator. Fungal population was estimated by counting the number of colonies. Total number of fungi in rhizosphere was calculated on dry weight basis. Pure culture of fungi were maintained on slants of PDA media in culture tubes for identification.

Gilman (1995), Subramanian (1971) and Barnett and Hunter (1972)were consulted for identification of fungi. The following formula was used for determination of relative abundance of a fungal species.

$$\text{Relative abundance (\%)} = \frac{\text{Total no of colonies of individual species}}{\text{Total no of all species}} \times 100$$

3(d).2.3 Isolation of fungi from Rhizoplane:

Roots were washed in running tap water and 1cm root pieces were cut out from tap and lateral roots with sterilized scissor. The roots were then washed in sterile distilled water thrice serially. Root pieces were transfer to PDA media plates supplemented with 0.5 ml streptomycin (0.2 gm/lit). under aseptic condition. The plates were incubated upside down at 28 ± 1º C in a BOD Incubator for 5 days.

3(d)2.4 Isolation of fungi for non Rhizosphere soil.

Soil samples were collected randomly from 5 different spots of Oak plantation of rearing site of Umrangso in 2013 and 2014.One kg soil samples included plant debris were collected with a soil at a depth of 0-15cm from the root region of Oak plants using the conventional sampling method (Johnson and Curl,1972). The samples were placed in clean plastic bags. Five soil sample (four at corner and one at centre) taken from each study site were mixed thoroughly in one composite sample.

1.0gm soil samples were processed for isolation of soil mycoflora. For isolation of soil mycoflora, serial soil dilution plating technique was followed (Johnson and Curl, 1972).Isolation was done within 24hours of collection.1.0 gm soil was transferred to a 250 ml conical flask containing 90ml sterilized distilled water to make a total of 100ml.The suspension was shaken vigorously for 30 minutes to obtain a homogenous solution. Stock solution 1.0ml was pipetted aseptically and dispensed in dilution test tube with 9.0ml sterilized distilled water. Series of soil dilutions of 10^{-1}, 10^{-2} ,$10^{-3}$and 10^{-4} were prepared.1.0ml aliquots from dilution of 10^{-3} and 10^{-4} was transferred aseptically into sterile Petri-plates with 10.0-12.0 ml of melted PDA media suppleted with 0.5ml of 250 mg Streptomycin to s suppress bacterial growth.5 petriplates were provided for each solution. These plates were incubated in BOD incubator for 28 ± 1ºC for 5-7 days. Grown fungi were isolated and identified.

3(d)2.5 : Isolation of Air mycoflora:

3 petriplates containing sterilized PDA media containing streptomycin (0.2gm/lit)were exposed in the air over the Farm area for 15 minutes against the air

current at two different height(0.75metre and 1.50metre) 3 replication for each height. Petriplates brought laboratory incubated for 5 days

3(e).1Materials

Soil of Oak (*Quercus serrata*) vegetation of Research Extension Centre Farm of Umrangso.

Physico-chemical nature of soil of Oak vegetation in the farm of Research Extension Centre, Umrangso, District Dima Hasao (North- Cachar Hills), Assam.

3(e).2 Methods

3(e).2.1 Determination of Soil pH:

10.0 gm of soil was taken in a beaker and it was mixed with 50ml of distilled water. The soil water mixture was stirred for 20 minutes on a magnetic stirrer. Then pH of soil was measured through an electronic digital pH meter.

3(e).2.2 Estimation of Organic Carbon:

Walkey and Black's (1934) rapid titration method was followed for determination of organic carbon. One gm air-dried and sieved (0.2mm) soil was taken in a dry and clean 500 ml conical flask, 10ml of 1N $K_2Cr_2O_7$ was added to the flask was swirled for a while followed by addition of 20 ml conc. H_2SO_4. The flask was swirled again and allowed to stand for 30 minutes. 10ml of 85% phosphoric was added to it and titrated with 1N $FeSO_4$ solution using diphenylamine as indicator. Same determined as given below.

Organic carbon (%)= $\frac{B-S\times 0.003\times 100}{W}$

Where B=Volume of FeSO4

S=Volume of K2Cr2O7

W=Weight of soil sample(gm)

3(e).2.3 Estimation of Nitrogen:

Nitrogen estimation by Alkaline Potassium permanganate ($KMnO_4$) method (Subbiah and Asija 1956): 20gm of soil was taken in a distillation flask and add 20ml of water and 100ml of 0.32% $KMnO_4$ pipette out 25ml of N/50 H_2SO_4 in a conical flask. And add 2-3 drops of methyl red indicator and dip the end of the delivery tube into it. Pour 100ml of 2.5% NaOH solution into the flask and cork it immediately. Distil the ammonia gas from the distillation flask and collect in H_2SO_4 solution. Continue distillation till the evolution of ammonia ceases completely (test by bringing a moist red litmus paper near the outlet of the condenser, which will turn blue as long as ammonia is being evolved).Titrate the excess of H_2SO_4 against N/50 NaOH and note the volume of NaOH used. The end point is reached when the color changes from pink to yellow.

1. Weight of soil taken=20gm

2. Volume of N/50, H2SO4 taken =25 ml

3. Volume of N/50, NaOH used (titrated value) = X ml

4. Volume of N/50 acid used for NH3 absorption (25-X)ml

(1ml of N/50 H2SO4=0.02 meq. of N 0.28mg N=0.00028 gm N)

Calculation:

a. Percentage of available N = (25-X) x 0.00028 x100/20

b. Available N in the soil (ppm)=(a) x 10,000

c. Available N in the soil (kg/ha)=(b) x 2.24

3(e).2.4 Estimation of Phosphate:

Estimation of available phosphate Bray's method (Bray and Kurtz,1945)

This procedure is primarily meant for soils for soils which are moderately to strong acids (pH around 5.5 or less).This method gives results highly correlated with the crop response to phosphate fertilization.

1.5.0gm soil was taken in 100ml conical flask.

2. Add 50ml of extractant solution to the soil.

3. Shake the contents of the flask for exactly 5 minutes, and filter through Whatman No.42 filter paper.

4. Prepare a blank in which all the reagents are added similarly, except the soil.

3(e).2.5 Determination of Available Potassium in soil (K_2O)

Procedure:

1. 5.0 gm soil in a 150ml of conical flask.

2. Add to it 25 ml N NH_4O Ac solution.

3. Shake the contents of the conical flask on an electric shaker for 5 minutes and filter.

4. Feed the filtrate in the atomizer of the flame photometer, 100 of which has been set with 40 ppm K solution and recorded the reading.

3(f).1 Materials

Leaf samples *Quercus serrata* of different types (tender, semi-mature and mature) were collected from R.E.C. Umrangso farm, in different treatments and in spring and autumn season during the year 2013 and 2014.

3(f)2 : Methods

3(f)2.1 Foliar constituents *Quercus serrata* in spring and autumn season during 2013 and 2014.

Leaves of different types namely tender, semi-mature and mature were collected from plants/trees. For spring crop pruning and pollarding were done during the 1st week of December and for autumn crop light pruning/clipping were done in 2nd week of August. The agronomical practices, application FYM and NPK were done for better quality and quantity of leaves for rearing seasons.

3(f)2.1.1 Determination of moisture content.

Moisture content of the leaves was determined by the method of A.O.A.C (1984).One hundred gram of *Q.serrata* leaves(tender, semi-mature, and mature)were collected separately of three different clean properly with clean cloth to removed dirt and dried separately at 60º C for 24 hours and powdered.

$$\text{Moisture}(\%)=\frac{\text{Fresh weight of leaves} - \text{Dry weight of leaves}}{\text{Fresh weight of leaves}} \times 100$$

3(f)2.1.2 Estimation of Crude Protein:

The total soluble protein content was estimated by using the method of Lowry *et al.*, (1951)

Regents:

i) 2% $NaCO_3$ in 0.1NaOH

ii) 0.5% $CuSO_4.5H_2O$ in1% sodium citrate or sodium potassium tartrates

iii) Alkaline copper solution: 1 ml of reagent 'b' mixed with 50ml of reagent 'a'.

iv) 1N Folin-ciocalteau reagent (Commercial reagent) dilultated with water to give a solution 1N in acid.

Extraction:

The residues left after 80% acetone extraction was hydrolyzed in 5.0ml of 1N NaOH for overnight and centrifuged at 5000rpm for 20 minutes. Supernatant was kept aside and residue was again extracted with 5.0 ml of 1N NaOH for 1 hour and then centrifuged. Both the supernatants were pooled and made the volume 10.0 ml.

Procedure:

A 0.5 ml aliquot was taken in test tube and mixed with 5ml of reagent (c) solution allowed to stand for 10 minutes. Thereafter, 0.5 ml of reagent (d) was added with instant mixing.

After 30 minutes absorbance was recorded at 570 mm through spectrophotometer (Model SL.177) against reagent blank. Standard curve was prepared with a graded concentration of bovine-serum albumin.

3(f)2.1.3 Estimation of crude fibre content:

The crude fibre content was determined by the method of A.O.A.C (1984).Four grams of moisture and fat free sample was digested with 200 ml of 1.25 per cent sulphuric acid (H_2SO_4) for 30 minutes. The acid solution was decanted and the material was with hot water to remove the acid. The acid free residue was treated with 200ml of 1.25 per cent sodium hydroxide (W/V) solution for 30 minutes. After decantation of top layer, solid material was filtered through previously weighed filter paper. The residue was made free from alkali by repeated washing with hot water and then washed with alcohol and finally with ether. The material was then dried in an oven at 100 º C for five hours and weighed (W_e).The was transferred to a crucible, heated in a muffle furnace (Make: INSIF) at 600 ºC for three hours, cooled and weighed again (W_a).The difference in weight (W_e -W_a) represents the weight of crude fibre.

$$\text{Crude fibre content (\%)} = \frac{(We - Wa)}{\text{Weight of leaf sample}} \times 100$$

The crude fibre content was expressed in percentage of moisture and fat free sample on dry weight basis.

↓

2gm sample +2.5 gm ConH_2SO_4=200ml D/W (acidic treatment)

↓

After boiling (30 minutes),the samples are filtered in a muslin cloth and washed it in D/W

↓

Drying samples extract+2.5gmNaOH pelletsP+200m D/W(alkaline treatment)

↓

Again boiling (30 minutes),the samples are filtered in a muslin cloth and washed it inB/W

The extra water is removed by washing alcohol and kept in oven for 24hours at 600^0C

↓

The samples is weighted note down

↓

The acid and alkali treatment, oxidative hydrolytic degradation of the cellulose and degradation of lignin occur. The loss is weight give the fiber content.

3(f).2.1.4 Estimation of Crude fat content:

The crude fat content in the leaf samples was estimated by the A.O.A.C method (1984). Crude fat content was determined by extracting the fat from the sample using a solvent,then determining the weight of the fat recovered. As lipids/fats are relatively non-polar molecules, they were pulled out of a sample using relatively non-polar solvents. With a non-polar solvent, only non-polar molecules in the sample dissolved while polar ones do not.

3(f).2 Methods

Weigh 2-3 gm of the dried food (leaf) sample into extraction thimble.

↓

Place the thimble inside the Soxhlet Apparatus.

↓

Connected a dry pre-weighted solvent flask beneath the apparatus and added required quantity of solvent and connect the condenser.

↓

Adjusted the heating rate to give a condensation rate of 2-3 drops and extract for about 16 hours.

↓

After completing the extraction removed the thimble and reclaim ether using the apparatus.

↓

Complete removal of ether on a boiling bath and dry flask at 105 ºC for 30 minutes. Cooled in a desiccator and weighed.

Calculation: Crude Fat (% of Dry matter) = (weight of fat x/weight of sample) × 100

3(f)2.1.5 Determination of Ash content by A.O.A.C method (1984)

For estimation of total ash %, 1.0gm of leaf sample was dried in a nickel crucible and heated it on a low flame till the organic matter turn to burn.

The crucible were placed in a muffle furnace and heated it 600ºC and stopped when grayish white ash formed. The residue represented the total ash percentage.

3(f)2.1.6 Estimation of Carbohydrate content:

The carbohydrate content were estimated by Anthrone method (Sadasivam and Manickam, 2005).In this method, carbohydrates are first hydrolysed into simple sugars using dilute hydrochloric acid. In hot acidic medium, glucose is dehydrated to hydroxymethyl furfural. This compound forms with anthrone a green coloured product with an absorption maximum at 630 nm.

50mg of the leaf sample was taken into a boiling tube and hydrolysed it by keeping in boil water bath for 1 hour with 2.5 ml of 2.5 N-HCl and cool to room temperature. Neutralized it with solid sodium carbonate till the effervescence ceases. Made up the volume to 50 ml and centrifuged it. The supernatant was collected and 0.5 and 1ml aliquots were taken for analysis. The standards were prepared by taking 0.0, 0.2, 0.4, 0.6, 0.8 and 1.0ml of the working standard (0 serves as blank)and make up the volume to 1.0ml in all tubes including the sample tubes by adding distilled water. Then 2.0ml of anthrone reagent was added. Heated for ten minutes in a boiling water bath. Cooled rapidly and read the green to dark green color at 630nm.

50 mg sample+2.5ml of 2.5N HCL in to a test tube

↓

Hydrolysed by keeping it in a boiling water bath for 1.0 hour

↓

Neutralized it with Na_2CO_3 until effervescence ceases

↓

Made up the volume to 50 ml and centrifuged

↓

Collected the supernatant and taken aliquots for analysis

↓

Prepared the working standard Zero served as blank

↓

Made up the volume to 1.0 ml in all tubes

↓

Added 2.0ml of Anthrone reagent and heated for 10 minutes

↓

Cooled rapidly and read the absorbance at 630 nm

↓

Carbohydrates hydrolyzed in to sugar using dil HCl. In acidic medium glucose

↓

Dehydrated to hydroxymethyl furfural which formed a green colored Anthrone.

3(g).1.Materials

Oak tasar cocoons (*Antheraea proylei* **Jolly**.) were taken from harvested three different treatments for observed the reeling parameter.

3(g).2 Methods.

Oak tasar cocoons were hot air stifled for 6-7 hrs at 70° C and stored for 2-3 months were used in this study.

Thirty oak tasar cocoons (ten replication of three cocoons each) were wrapped in a coarse cotton cloth and subjected to 30 minutes pressure cooking at 1.05kg/cm pressure.

The cocoons were then soaked in pineapple extract at room temperature (26-31^0C) for 12hrs and cocoons along with the wrapper were taken out from cooking medium and then washed repeatedly with tap water until the associated brown colour and proteinase activity were washed out (Devi *et al.*,2012).

The cocoons were then removed from the wrapper, semidried on blotting papers, deflossed and then subjected to single filament reeling on an eppouvette machine.

Statistical analysis:

All the observed data were analyzed statistically using the technique of analysis of variance. The significant of treatment difference was judged by 'F' test as outlined by Cochran (1977).

The standard error of the differences SED ± was calculated by using following expression.

$$\text{SED} \pm = \sqrt{\text{Error mean square} \times 2} \,/\, \text{pooled number of application}$$

The critical differences (CD) was calculated to test the significance of differences of the treatments. Critical differences (CD) was calculated by using following formula:

$$\text{CD } (5\%) = (\text{SED} \pm) \times \text{'t'}$$

' Where, the =5% tabulated value of the 't' at error degree of freedom.

EXPERIMENTAL FINDINGS (RESULTS)

RESULTS:

4. Average Leaf yield of per plant (*Quercus serrata*) and per hectare with application of FYM and NPK

The experiment finding of the present investigation on average leaf yield of per plant of *Quercus serrata* and per hectare under three different treatments in Spring and Autumn season in 2013 and 2014.

(A) Control without any input (without application of FYM and NPK)

(B) Application of NPK and FYM.

(C) Application of FYM

4.1. Leaf yield of per plant /per hectare of *Q.serrata.* under different treatments During Spring 2013 and 2014, the average yield per plant of *Quercus serrata* recorded .

Treatment A(Control)-1.15 Kg per plant and 7752.00 Kg per hectare when not applied any input.

Treatment B – 1.53 Kg per plant and 10355.00 Kg per hectare when applied 48.0 gm Urea, 46.5gm SSP and 9.3 gm MOP and 10Kg FYM per plant.

Treatment C- 1.320 Kg per plant and 8875.00 Kg per hectare when applied 10Kg FYM only.

During Autumn 2013 and 2014, the average yield per plant of *Quercus serrata* recorded .

Treatment A - (Control)- 0.420 Kg per plant and 2824.00Kg per hectare when not applied any input.

Treatment B - 0.630 Kg per plant and 4236.00 Kg per hectare when applied 48.0 gm Urea, 46.5gm SSP and 9.3 gm MOP and 10Kg FYM per plant.

Treatment C - 0.530 Kg per plant and 3563.00 Kg per hectare when applied 10Kg FYM only.

Average leaf yield was found in Spring season per plant of *Quercus serrata* (1.53Kg) and per hectare 10355.00 Kg and Autumn season 0.630Kg per plant and per hectare 4236.00Kg when applied Chemical fertilizer and FarmYard Manure. The leaf yield found more other than treatment. Hence, to increasing more quantitative of leaf in per plant and per hectare plantation of *Q.serrata* was found very much essential to increasing rearing capacity of silkworm rearing per unit area.

Table 3: Average Leaf yield of per plant (*Quercus serrata*) and per hectare with application of FYM and NPK in 2013 and 2014 in Spring and Autumn season:

Season	Treatment	Average Leaf yield per plant			Average Leaf yield per hectare Kg.(6724 plant /hectare)
		Average. Leaf yield per plant (kg)2013	Average Leaf yield per plant (kg)2014	Average Leaf yield per Plant 2013 and 2014	
Spring	(A) Control(without FYM and NPK)	1.176	1.13	1.15	7752.00
	(B) N P K FYM 150 50 38 7000 Kg hectare (Urea- 48.0gm; SSP-46.5gm; MOP-9.3gm;FYM-10 Kg per plant)	1.56	1.50	1.53	10355.00
	(C) N P K FYM 0 0 0 7000 Kg (FYM-10Kg per plant)	1.328	1.312	1.32	8875.00
Autumn	(A) Control(without FYM and NPK	0.423	0.412	0.42	2824.00
	(B) N P K FYM 150 50 38 7000 Kg hectare (Urea- 48.0gm; SSP-46.5gm; MOP-9.3gm;FYM-10 Kg per plant)	0.642	0.618	0.63	4236.00
	(C) N P K FYM 0 0 0 7000 Kg (FYM-10Kg per plant)	0.531	0.516	0.53	3563.00

N- Nitrogen (Urea) ; P- Phosphate(Single Super Phosphate); K- Potash (Murate of Potash) ; FYM- Farm Yard Manure

Photo Plate No 1: (a)Showing *Quercus serrata* plantation to study leaf yield, *A.proylei* **J.** silkworm rearing, biochemical analysis of leaf, study myco-flora from phylloplane, rhizosphere, non-rhizosphere, rhizoplane of soil and from over air. (b) Seedlings for study myco-flora from rhizosphere and non rhizosphere soil.

4.1.2. Average leaf yield of per plant of *Q.serrata.*

Table: 4 Average yield of per plant (*Quercus serrata)* in Spring and Autumn season in 2013 and 2014 under different treatments.

Treatment / Replication	Spring 2013 and 2014			Autumn 2013 and 2014		
	Control (Kg)	NPK+FYM (Kg)	FYM (Kg)	Control (Kg)	NPK+FYM (Kg)	FYM (Kg)
R1	1.18	1.59	1.321	0.41	0.65	0.55
R2	1.05	1.52	1.36	0.45	0.61	0.53
R3	1.22	1.48	1.28	0.43	0.63	0.51
R4	1.10	1.45	1.34	0.44	0.67	0.50
R5	1.13	1.61	1.30	0.41	0.60	0.52
R6	1.22	1.53	1.32	0.40	0.62	0.57
R7	1.20	1.52	1.42	0.38	0.63	0.48
R8	1.18	1.64	1.20	0.425	0.61	0.53
R9	1.18	1.43	1.34	0.38	0.65	0.58
R10	1.07	1.53	1.32	0.425	0.63	0.53
Total	11.53	15.30	13.20	4.15	6.30	5.30
Mean	1.15	1.53	1.320	0.42	0.63	0.53
SED(±)	0.00209			0.00032		
CD(5%)	0.03262			0.00056		

Leaf yield of *Q. serrata* in the Spring and Autumn season statistically analysis. In Spring season SED(±) 0.00209 and CD(5%) 0.03262 and Autumn season SED(±) 0.00032 and CD(5%) 0.00056.

The leaf yield of *Q. serrata* in both Spring and Autumn season during 2013 and 2014. However, leaf yield was found to be much higher in Spring than in Autumn season. The highest average leaf yield (1.53 Kg) was recorded in plants applied with FYM and NPK.

4.2. The Meteorological data recorded during the rearing seasons of *Antheraea proylei* **Jolly.** in Spring and Autumn season in 2013 and 2014 .

- In 2013(Spring season) average temperature maximum and minimum was recorded 31.06° C and 18.05°C, relative humidity maximum 70.86 % , minimum 55.41% and rainfall 427mm in 9 days.
- In 2013 (Autumn season) average temperature maximum 31.81^0 C, minimum 22.46 ^{0}C, relative humidity maximum 83.03%,minimum 55.79%, rainfall 817 mm. in 22 days.
- In 2014 (Spring season) average temperature maximum 29.43°C, minimum 17.50°C, relative humidity maximum 64.98% , minimum 53.57%,rainfall 273 mm. in 5 days.
- In 2014 (Autumn season) average temperature maximum 27.62°C,minimum 23.28°C, relative humidity86.57%, minimum 54.14%, rainfalls 1071 mm. in 16 days.
- In Autumn season minimum temperature and relative humidity was recorded slightly higher than Spring season and rainfalls also recorded more in millimeter and increased nos days.
- The minimum temperature, less maximum humidity and less rainfalls in the Spring season was recorded which got positive impact on silkworm rearing

Table 5: Meteorological record of Oak tasar crop at Umrangso during Spring and Autumn season in 2013 and 2014.

Crop season	Temperature. °C		Relative humidity %		Total rainfalls (millimeter)	No. of rainy days
	Maximum	Minimum	Maximum	Minimum		
2013 - Spring March-April	31.06	18.05	70.86	55.41	427	9
2013 – Autumn Sept-Oct	31.81	22.46	83.03	55.79	817	22
2014 - Spring March-April	29.43	17.50	64.98	53.57	273	5
2014 – Autumn Sept-Oct	27.62	23.28	86.57	54.14	1071	16

4.3 Rearing performance of *A.proylei* J. under different treatment in 2013 and 2014.

4.3.1 In Spring season rearing performance of *A.proylei* **J.** without NPK and FYM application(Control) larval period 36-43days, mature larval weight male14.8gm,female 15.8 gm, cocoon weight male 4.88 gm, female 5.60 gm, shell weight male 0.44 gm, female 0.50 gm, silk ratio of male 9.0%, female 8.83% and average silk ratio(SR)8.98% was found. In Autumn season larval period 38-46 days, mature larval weight male 14.76 gm, female 15.7gm,ERR 16.2%, average cocoon weight male 4.82gm, female 5.55 gm, average shell weight 0.44 gm, female 0.48 gm, silk ratio male 9.22%, female 8.65 gm, average SR 9.18% .

Table 6: Rearing Performance of *Antheraea proyeli* Jolly. without NPK and FYM application (Control) in 2013 and 2014.

Crop	No. of Worm brushed	Larval period (days)	Mature larval weight (gm)		Average cocoon weight (gm)		Average Shell weight (gm)		Shell ratio (%)		Average Shell ratio (%)
			♂	♀	♂	♀	♂	♀	♂	♀	
Spring 2013	1000	36-43	14.82	15.90	4.86	5.49	0.44	0.49	9.05	8.92	8.98
Spring 2014	1000	36-43	14.77	15.82	4.91	5.71	0.44	0.50	8.96	8.75	8.84
Average	36-43		14.8	15.86	4.88	5.60	0.44	0.50	9.00	8.83	8.98
Autumn 2013	1000	38-46	14.78	15.71	4.83	5.36	0.44	0.48	9.11	8.95	9.03
Autumn 2014	1000	38-46	14.74	15.80	4.82	5.75	0.45	0.48	9.34	8.35	9.34
Average	38-46		14.76	15.75	4.82	5.55	0.44	0.48	9.22	8.65	9.18

4.3.2. In Spring season rearing performance of *A.proylei* **J.** with application of NPK and FYM, larval period 34-38 days, average mature larval male15.70gm,female 17.01 gm, cocoon weight male 5.52gm, female 6.97 gm, shell weight male 0.535 gm, female 0.705 gm, silk ratio male10.15%,female 10.12% ,average SR 10.15% were recorded. In Autumn season larval period 34-40 days, mature larval male 15.66 gm, female 17.01gm,cocoon weight 5.04 gm, female 6.76 gm , shell weight male 0.495 gm, female 0.635 gm, silk ratio male9.79% female 9.40%, average silk ratio SR 9.60%.

Table 7: Rearing Performance of *Antheraea proyeli* Jolly. with application of NPK and FYM in 2013 and2014.

Crop	No. of Worm brushed	Larval period (days)	Mature larval weight (gm)		Average cocoon weight (gm)		Average Shell weight (gm)		Shell ratio (%)		Avg Shell ratio (%)
			♂	♀	♂	♀	♂	♀	♂	♀	
Spring 2013	1000	34-38	15.72	17.10	5.26	7.00	0.534	0.71	10.20	10.14	10.20
Spring 2014	1000	34-38	15.69	16.92	5.25	6.95	0.533	0.70	10.10	10.10	10.10
Average	34-38		15.70	17.01	5.25	6.97	0.535	0.705	10.15	10.12	10.15
Autumn 2013	1000	34-40	15.67	17.07	5.03	6.90	0.49	0.64	9.74	9.30	9.52
Autumn 2014	1000	34-40	15.65	16.96	5.05	6.63	0.50	0.63	9.84	9.50	9.67
Average	34-40		15.66	17.01	5.04	6.76	0.495	0.635	9.79	9.40	9.60

4.3.3. In Spring rearing performance of *A.proylei* **J.** with application of FYM, larval period 34-40 days, mature larval weight male15.24 gm, female 16.26 gm, cocoon weight male 4.97 gm, female 6.57 gm, shell weight male 0.48 gm, female 0.625 gm. silk ratio male 9.65% ,female 9.51% ,average SR 9.60% were found. In Autumn season larval period 38-45 days , mature larval weight male 15.08 gm, female 16.12gm, cocoon weight male 4.92 gm, female 6.48 gm, shell weight 0.48 gm, female 0.595 gm., silk ratio male 9.75%,female 9.17,average SR 9.46% were recorded.

Table 8:Rearing Performance of *Antheraea proyeli* Jolly. with application of FYM in 2013 and 2014.

Crop	No. of Worm brushed	Larval period (days)	Mature larval weight (gm)		Average cocoon weight (gm)		Average Shell weight (gm)		Shell ratio (%)		Average Shell ratio (%)
			♂	♀	♂	♀	♂	♀	♂	♀	
Spring 2013	1000	34-40	15.19	16.32	5.02	6.59	0.48	0.63	9.60	9.56	9.60
Spring 2014	1000	34-40	15.30	16.22	4.92	6.55	0.48	0.62	9.71	9.46	9.60
Average	34-40		15.24	16.26	4.97	6.57	0.48	0.625	9.65	9.51	9.60
Autumn 2013	1000	38-44	15.13	16.18	4.97	6.57	0.48	0.60	9.66	9.13	9.4
Autumn 2014	1000	38-46	15.04	16.06	4.88	6.40	0.48	0.59	9.84	9.22	9.53
Average	38-45		15.08	16.12	4.92	6.48	0.48	0.595	9.75	9.17	9.46

Among all the treatment rearing performance was found better (ERR 64.5% and SR 10.15% in Spring season) and in Autumn season (ERR 31.6% and SR 9.60%) when with application of NPK and FYM. and it was found that larval period short, mature larval weight, cocoon weight, shell weight more significant.

Photo plate No:**7(a)** cellular rearing and different larval stage of *Antheraea proylei* **Jolly. 7(b)** 1st instar, **7(c)** 2nd instar, **7(d)** 3rd instar, **7(e)** 4th instar and**7(f)** 5th instar), **8(a)** silkworm rearing of *Antheraea proylei* **Jolly. 8(b)**yellow, **8(c)**green and **8(d)**blue; **9**Silkworm rearing of *Antheraea proylei* **Jolly.** yellow colour), **10** silkworm moths **10(a)** male moth **10(b)** female moth, **10(c)** egg, DFL and **10(d)** cocoons of *Antheraea proylei* **Jolly.**)

Table: 9 Effective rate of rearing(ERR%) of *A.proylei* during 2013 and 2014 under different treatments.

Treatment Replication	Spring 2013 and 2014			Autumn 2013 and 2014		
	Control (%)	NPK+FYM (%)	FYM (%)	Control (%)	NPK+FYM (%)	FYM (%)
R1	36.0	63.5	40.5	18.0	36.0	25.0
R2	31.0	64.5	47.5	16.0	34.5	26.5
R3	30.0	60.5	44.5	17.0	31.5	24.5
R4	35.0	68.5	43.5	14.0	34.0	20.5
R5	38.0	70.5	43.0	12.5	32.5	22.0
R6	32.0	57.0	46.0	16.5	30.0	20.0
R7	30.0	56.5	48.0	18.5	30.0	23.5
R8	26.5	64.5	42.5	18.0	28.5	28.0
R9	24.0	68.5	43.5	15.5	31.5	22.5
R10	32.5	71.0	36.0	16.0	28.0	26.0
Total	315.00	645.00	435.00	162.00	316.50	238.50
Mean	31.50	64.50	43.50	16.2	31.65	23.85
SED(±)	10.203			2.56		
CD(5%)	17.692			4.435		

Effective rate of rearing(ERR%) of *A.proylei* during 2013 and 2014 under different treatments without application of FYM and NPK(Control) is 31.5% , application of NPK and FYM is 64.5% and the application of FYM is 43.5% in spring season. The effective rate of rearing application of FYM and NPK (Control) is 16.2%, application of NPK and FYM is 31.65% and the application of FYM is 23.85% in autumn season. The ERR % showed highly significant difference amongst in both Spring and Autumn crop rearing 2013 and 2014. ERR% was found

much higher in Spring season rearing than in Autumn crop rearing. Highest average ERR% of *A. proylei* **J**. was recorded 64.50% where the rearing was conducted those plants applied with Farm yard manure and chemical fertilizer NPK.

4.4 Isolation and identification of species from Phylloplane, Rhizosphere, Non-rhizosphere, Rhizoplane and Air Mycoflora of *Q.serrata* during Spring and Autumn in 2013 and 2014.

4.4.1 Qualitative and quantitative study of phylloplane Mycoflora of *Q.serrata* leaves during Spring and Autumn season of Oak tasar silkworm rearing (*Antheraea proylei* **Jolly.**) at Umrangso, Research Extension Centre, Farm. Eleven fungal species were isolated from the leave surface of *Q.serrata*. The types of fungi which colonized the leaves at different stages of maturation viz. Tender, Semi-mature and Mature leaves, on the both side of leaves in the Spring and Autumn season.

i) <u>Spring season 2013 and 2014</u>

In spring season on the upper surface of tender leaves fungal species were isolated *i.e. Aspergillus niger*) 70.50 – 72.50% (P.P.No 2a and 2b, *Alternaria alternata* 17.0 – 18.0%(P.P.No 3c), *Mucor* sp 10.5 – 11.5%(P.P.No 5a) , and on lower surface *Aspergillus niger* 65.5 – 66.5%, *Alternaria alternata* 12.5 – 13.5%, *Mucor* sp 10.5 – 11.5%, and *Curvularia* sp 9.5 – 10.5%(P.P.No 4a) . On the upper surface of semi-mature leaves *Aspergillus niger* 60.5 – 61.5%, *Alternaria alternata* 15.5 – 17.5%, *Mucor* sp 12.5 – 14.5%, and *Curvularia* sp 8.5 – 9.5%. were isolated, like that on lower surface of the semi-mature leaves *Aspergillus niger* 57.5 - 58%, *Alternaria alternata* 15.5 – 16.5%, *Mucor* sp 10.5 – 12.5%, and *Curvularia* sp 8.5 – 9.5%. and *Fusarium* sp 5.0 – 6.5% (P.P.No 4c) on upper surface of mature leaves *Aspergillus niger* 54.0 – 55.5%, *Alternaria alternata* 22.5 – 23.5%, *Mucor* sp 10.5 – 12.5%, and *Curvularia* sp 3.5 – 4.0%. and *Fusarium* sp 6.0 – 8.0% were isolated but on the lower surface of mature leaves more number of fungal species were isolated *i.e. Aspergillus niger* 44.0 – 45.5%, *A.fumigatus* 16.0 – 16.5% (P.P.No 3a and 3b) *A.flavus* 3.5 – 5.0% (P.P.No 2c and 2d) ,*Alternaria alternata* 12.0 – 12.5%, *Mucor* sp 8.0 – 10.0%, and *Curvularia* sp 5.5 – 7.5% and *Fusarium* sp 6.5 – 7.5%.(Table:10,Fig:2).

ii) Autumn season 2013 and 2014

Upper surface of tender leaves *Aspergillus niger* 55.0 – 55.5%, *A.fumigatus*14.5 – 16.5%, *Alternaria alternata* 15.5 – 16.5%, *Mucor* sp 12.5– 14.0%, and lower surface of mature leaves *Aspergillus niger* 51.0 – 52.5%, *A.fumigatus* 18.0 – 19.5%, *Alternaria alternata* 15.5 – 17.0%, *Mucor* sp 6.5– 7.5%, *Fusarium* sp 6.0 – 6.5%, on the upper surface of semi-mature leaves *Aspergillus niger* 51.5 – 52.5%, *A.fumigatus*16.0 – 17.5%, *Alternaria alternata* 13.5 – 14.0%, *Mucor* sp 7.5– 8.5%, *Penicillum* sp 8.0 – 10.0% (P.P.No 4d and 4e), and on the lower surface *Aspergillus niger* 45.5 – 47.5%, *A.fumigatus*14.0 – 15.5%, *Alternaria alternata* 12.5%, *Mucor* sp 6.0– 6.5%, *Penicillum* sp 6.5 – 8.0%, *Curvularia* sp 8.0– 9.0%, *Fusarium* sp 4.0 – 4.5%, were isolated.

On the upper surface of mature leaves *Aspergillus niger* 44.0-45.5%, *A.fumigatus* 9.5-11.0%, *A.flavus* 4.0-4.5%, *Alternaria alternata* 15.5-16.5%, *Curvularia* sp 4.5-5.5%, *Penicillum* sp 5.0-6.0%, *Fusarium* sp 4.5-5.5%, *Verticillium* sp 4.5-5.0%, *Mucor* sp 4.0-4.5% and on the lower surface of mature leaves a total eleven numbers of fungal species were isolated *i.e. Aspergillus niger* 44.5-45.5%, *A.fumigatus* 6.0-6.5%, *A.flavus* 3.5-4.0%, *Alternaria alternata* 14.5-15.5%, *Mucor* sp 3.0-4.0%, *Curvularia* sp 4.0-4.5%, *Penicillum* sp 5.5-7.0%, *Verticillium* sp 3.0-3.5%,*Fusarium* 4.5-5.5%, *Colletotrichum* sp 3.5-4.5%(P.P.No 5b) , *Cladosporium* sp 3.5-4.0% (P.P.No 3d and 3e) (Table:11 and Fig:3).

Photo Plates No(P.P.No): **2 (a)** *Aspergillus niger* culture in petridish **2(b)***Aspergillus niger* spore with conidiophores, **2 (c)** *Aspergillus flavus* culture in petridish , **2(d)***Aspergillus flavus* spore with conidiophores., **3(a)** *Aspergillus fumigatus* culture in petridish, **3(b)** *Aspergillus fumigatus* conidiophores, **3(c)** *Alternaria alternata* conidia ,**3(d)** *Cladosporium* sp culture in petridish, **3(e)** *Cladosporium* spores, **4(a)** *Curvularia* sp conidiophores, **4(b)** *Fusarium solani* culture in petridish, **4(c)** *Fusarium* sp spore,**4(d)** *Penicillum* sp culture in petridish and **4(e)***Penicillum* sp conidiophores.; **5(a)** *Mucor* sp culture in petridish, **5(b)** *Colletotrichum* sp spore. **6(a)** *Rhizopus* sp culture in petridish **6(b)** *Rhizopus* sp spore ; showing different fungal isolated from phylloplane, rhizosphere, non-rhizosphere, rhizoplane soil, over air of *Quercus serrata* plantation and fungal isolated from rhizosphere and non-rhizosphere soil of *Quercus serrata* seedlings.

Table 10: Fungal isolates of leaf surface of *Quercus serrata* at different status of age during Spring crop (March –April) 2013 and 2014.

Status of leaves	Surface	No. of Sampling	Fungal Isolates	Relative abundance %	
					2014
Tender	Upper	10 nos	*Aspergillus niger*	70.50	72.50
			Alternaria alternata	18.0	17.0
			Mucor sp	11.5	10.5
	Lower	10 nos	*Aspergillus niger*	65.5	66.5
			Alternaria alternata	12.5	13.5
			Mucor sp	11.5	10.5
			Curvularia sp	10.5	9.5
Semi-mature	Upper	10 nos	*Aspergillus niger*	61.5	60.5
			Alternaria alternata	15.5	17.5
			Mucor sp	14.5	12.5
			Curvularia sp	8.5	9.5
	Lower	10 nos	*Aspergillus niger*	57.5	58.0
			Alternaria alternata	15.5	16.5
			Mucor sp	12.5	10.5
			Curvularia sp	9.5	8.5
			Fusarium sp	5.0	6.5
Mature	Upper	10 nos	*Aspergillus niger*	55.5	54.0
			Alternaria alternata	22.5	23.5
			Mucor sp	12.5	10.5
			Curvularia sp	3.5	4.0
			Fusarium sp	6.0	8.0
	Lower	10 nos	*Aspergillus niger*	45.5	44.0
			A.fumigatus	16.5	16.0
			A.flavus	3.5	5.0
			Alternaria alternata	12.5	12.0
			Mucor sp	10.0	8.0
			Curvularia sp	5.5	7.5
			Fusarium sp	6.5	7.5

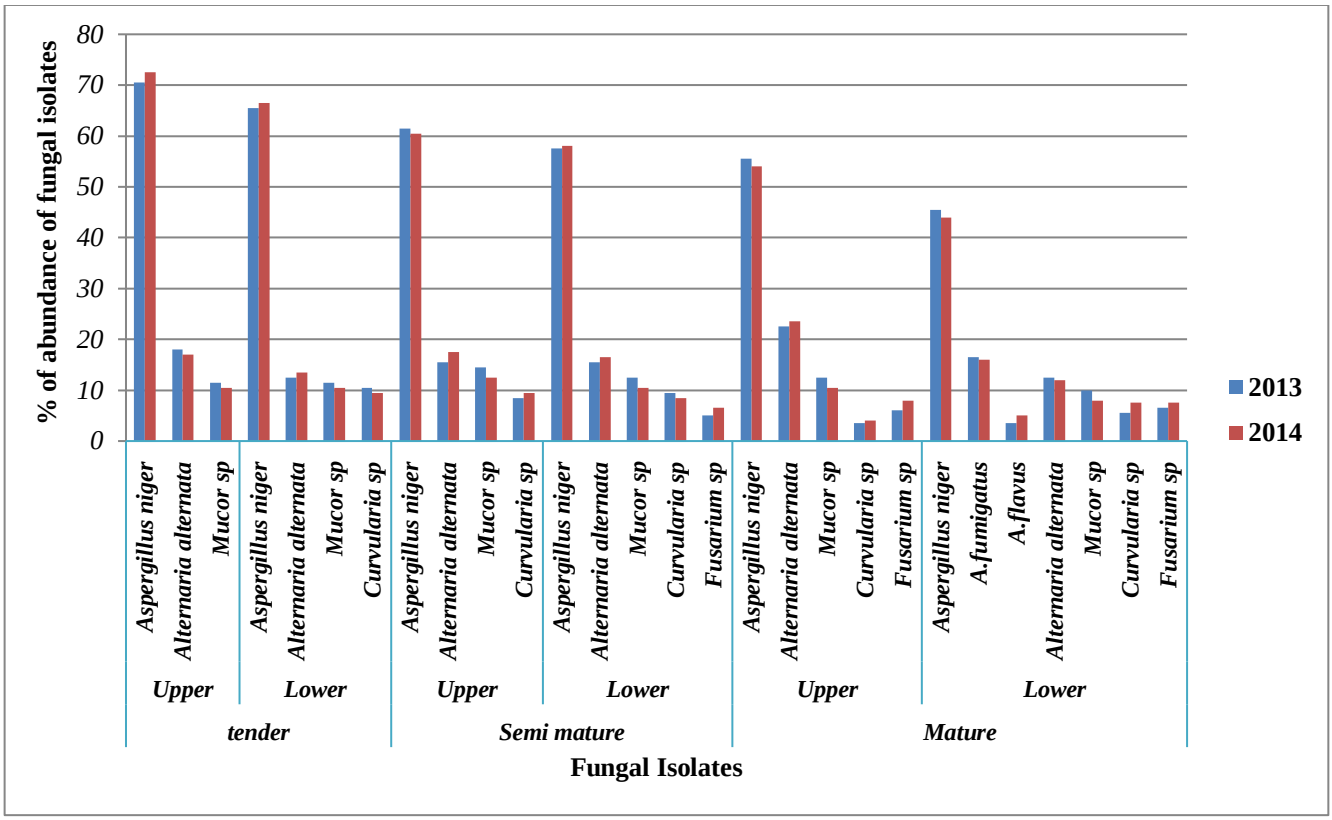

Fig 2: Fungal isolates of leaf surface of *Quercus serata* at different status of age Spring crop (March –April) 2013 and 2014.

Table 11:Fungal isolates of leaf surface of *Quercus serrata* at different status of age during Autumn crop (Sept-Oct.)2013 and 2014.

Status of leaves	Surface	No.of Sampling	Fungal Isolates	Relative abundance %	
				2013	2014
Tender	Upper	10 nos	*Aspergillus niger*	55.5	55.0
			A.fumigatus	16.5	14.5
			Alternaria alternata	15.5	16.5
			Mucor sp	12.5	14.0
	Lower	10 nos	*Aspergillus niger*	52.5	51.0
			A.fumigatus	19.5	18.0
			Alternaria alternata	15.5	17.0
			Mucor sp	6.5	7.5
			Fusarium sp	6.0	6.5
Semi - mature	Upper	10 nos	*Aspergillus niger*	52.5	51.5
			A.fumigatus	17.5	16.0
			Alternaria alternata	13.5	14.0
			Mucor sp	8.5	7.5
			Penicillum sp	8.0	10.0
	Lower	10 nos	*Aspergillus niger*	45.5	47.5
			A.fumigatus	15.5	14.0
			Alternaria alternata	12.5	12.5
			Mucor sp	6.5	6.0
			Penicillum sp	6.5	8.0
			Curvularia sp	9.0	8.0
			Fusarium sp	4.5	4.0
Mature	Upper	10 nos	*Aspergillus niger*	45.5	44.0
			A.fumigatus	9.5	11.0
			A.flavus	4.5	4.0
			Alternaria alternata	15.5	16.5
			Curvularia sp	5.5	4.5
			Penicillum sp	5.0	6.0
			Fusarium sp	5.5	4.5
			Verticillium sp	4.5	5.0
			Mucor sp	4.5	4.0
	Lower	10 nos	*Aspergillus niger*	45.5	44.5
			A.fumigatus	6.5	6.0
			A.flavus	3.5	4.0
			Alternaria alternata	14.5	15.5
			Mucor sp	4.0	3.0
			Curvularia sp	4.5	4.0
			Penicillum sp	5.5	7.0
			Verticillium sp	3.5	3.0
			Fusarium sp	5.5	4.5
			Colletotrichum sp	3.5	4.5
			Cladosporium sp	3.5	4.0

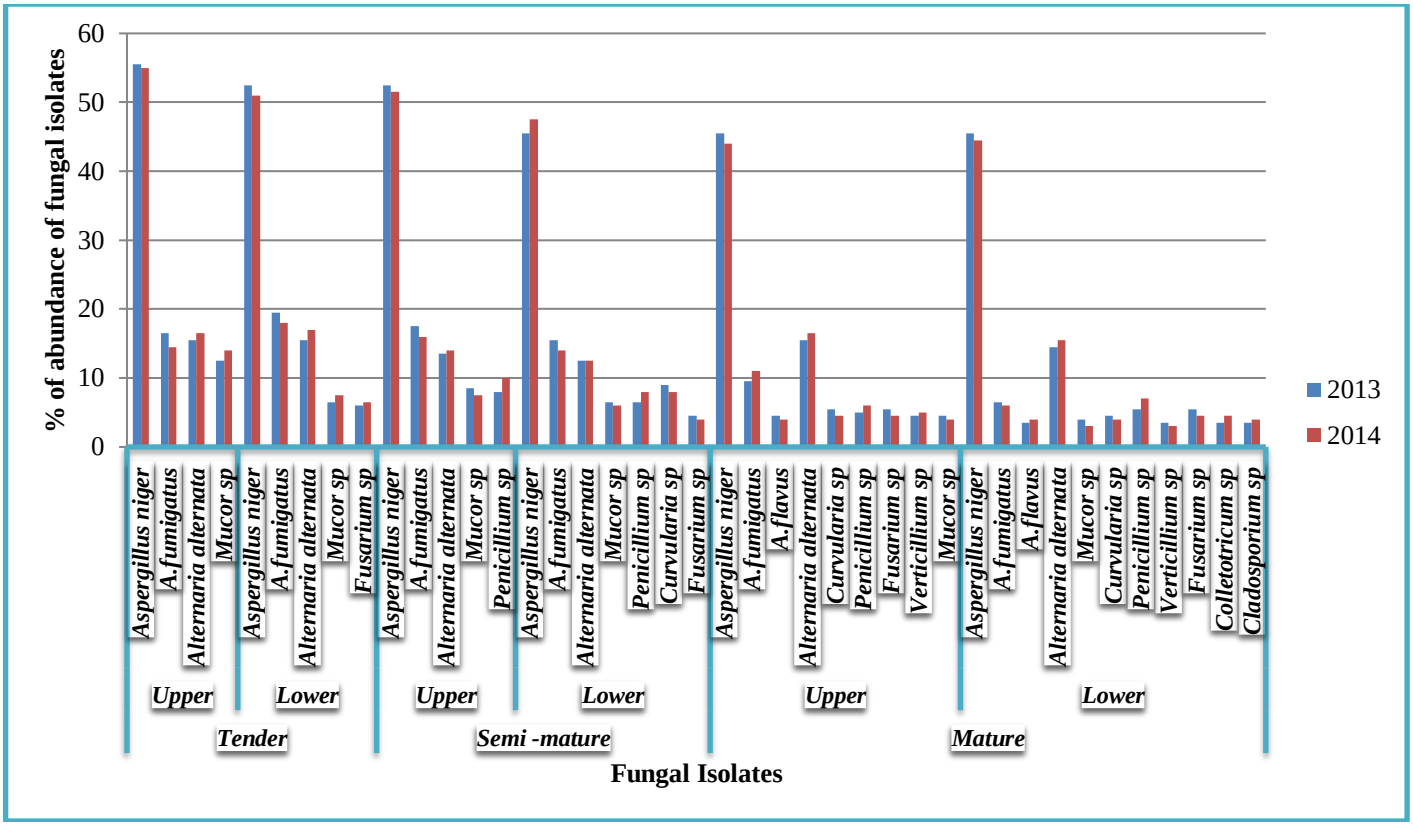

Fig 3: Fungal isolates of leaf surface of *Quercus serata* at different status of age during Autumn crop (Sept-Oct.)2013 and 2014.

The fungal species of *Aspergillus*, particularly *Aspergillus niger* was found to be most dominant in the three stages of growth during all seasons. *Aspergillus fumigatus* was found co-dominant in lower surface of mature leaves in Spring season and both surface of tender and semi-mature leaves in Autumn season. It was also found that *Alternaria alternata* co-dominant in the both leaf surface of tender and semi-mature leaves and upper surface of mature leaves in Spring season. *Penicillum* sp was found in the both side of semi-mature and mature leaves in Autumn season.

4.4.2. Qualitative and quantitative estimation of fungal population of soil from *Quercus serrata* seedlings at Umrangso REC Farm during 2013 and 2014.

Seedling soil: Ten fungal species were isolated from the soil of *Quercus serrata* seedling during the investigation period. (Table:12). The seedlings were 6 month and 1 year old raised in seedling beds. It takes 21-28 days for the *Quercus serrata* seeds to germinate under the climatic conditions of Umrangso. The seeds were sown in 4th week of February to 2nd week of March and same ages of seedling are taken for investigation during Spring and Autumn seasons. Among the isolated species *Alternaria alternata, Aspergillus flavus, Aspergillus niger, Fusarium oxysporium, Penicillum* sp and *Trichoderma* sp was found in both rhizosphere and non rhizosphere soil throughout the study period while *Fusarium solani* observed in rhizosphere soil. *Cladosporium* sp, *Collectotrichum* sp observed in rhizosphere and non rhizosphere soil during Autumn season (September-October), but *Mucor hiemalis* found only in rhizosphere soil in autumn season (P.P.No :2-6). *Aspergillus niger* was found dominant in both rhizosphere and non rhizosphere soil during Spring and Autumn season. *Aspergillus flavus* was found co-dominant followed by *Alternaria alternata, Fusarium solani, Fusarium oxysporium* and *Trichoderm* sp were co-dominant during Spring season in rhizosphere soil but *Aspergillus niger* and *Penicillum* sp *was* found dominant in non- rhizosphere soil and *Aspergillus flavus, Alternaria alternata, Fusarium oxysporium* and *Trichoderm* sp were found co-dominant during Spring season. *Fusarium solani* and *Fusarium oxysporium* were found co-dominant followed by *Aspergillus flavus* and *Mucor hiemailis*, than *Alternaria alternata, Cladosporium* sp, *Penicillum* sp and *Trichoderma* sp in rhizosphere soil in Autumn season. *Colletotrichum* sp showed lower occurrence in both season in both rhizosphere and non-rhizosphere soil.

Table 12: Relative abundance% Fungal isolates of RS and NRS of (*Quercus serrata)* seedlings during Spring and Autumn season during 2013 and 2014.

Sl. No	Fungal Isolates	Relative abundance%								REMARKS
		2013				2014				
		March –April Spring		Sept.-Oct Autumn		March –April Spring		Sept.-Oct Autumn		
		RS	NRS	RS	NRS	RS	NRS	RS	NRS	
1	*Alternaria alternata*	11.5	14.5	8.0	14.0	11.5	14.3	8.5	14.3	
2	*Aspergillus flavus*	18.0	14.5	8.5	15.0	20.0	14.3	8.5	14.3	
3	*Aspergillus niger*	29.5	21.0	21.5	19.5	29.5	21.5	21.0	19.0	Dominant
4	*Cladosporium* sp	0.0	0.0	8.0	9.5	0.0	0.0	8.5	9.5	
5	*Colletotrichum* sp	0.0	0.0	4.5	4.5	0.0	0.0	4.0	4.8	
6	*Fusarium solani*	11.5	0.0	12.5	0.0	10.5	0.0	12.0	0.0	
7	*Fusarium oxysporium*	11.5	14.5	12.5	9.5	10.5	14.3	12.0	9.5	
8	*Mucor hiemalis*	0.0	0.0	8.5	0.0	0.0	0.0	8.5	0.0	
9	*Penicillum* sp	6.5	21.0	8.0	14.0	6.5	21.5	8.5	14.3	
10	*Trichoderma* sp	11.5	14.5	8.0	14.0	11.5	14.1	8.5	14.3	

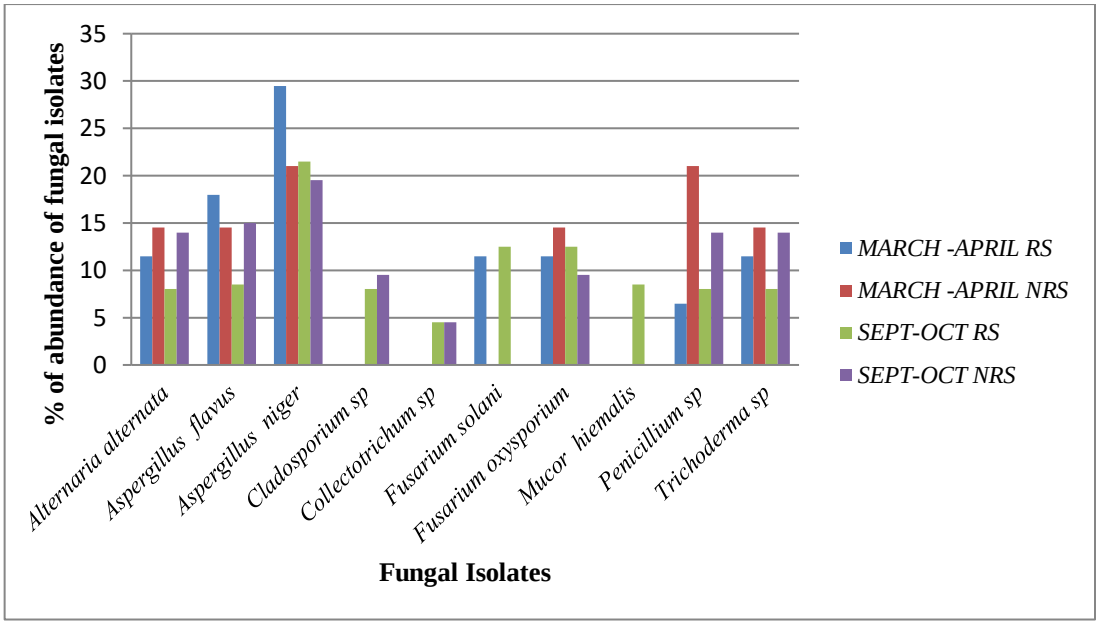

Fig 4: Showing fungal isolates of RS and NRS of *Quercus serrata* seedlings during Spring (March-April) and Autumn (September-October)2013.

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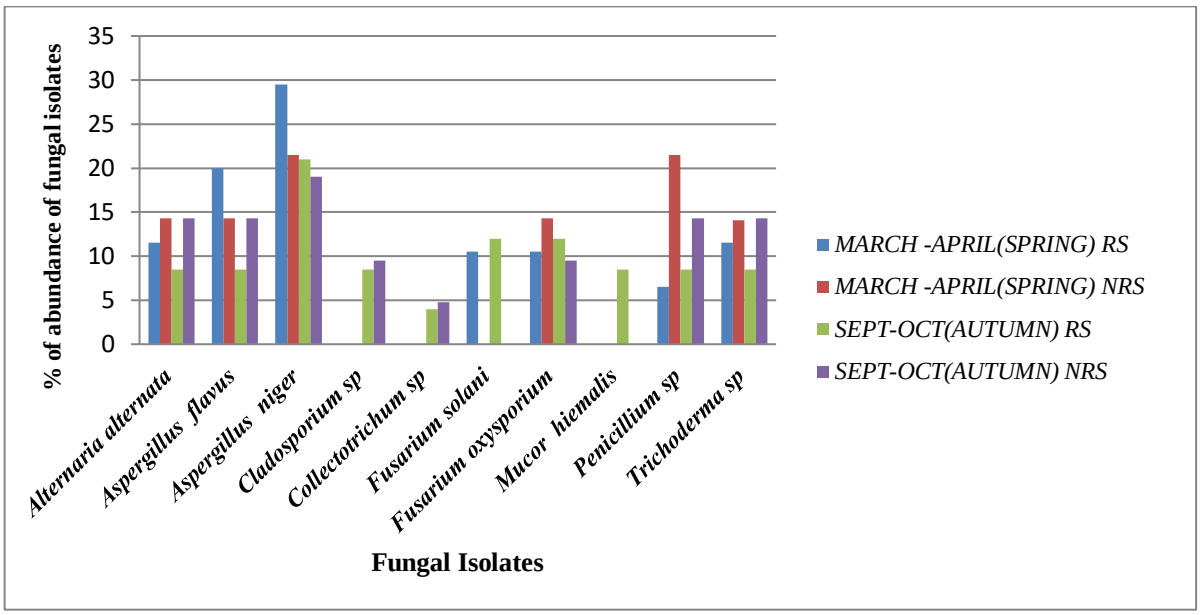


Figure 5: Showing fungal isolates of RS and NRS of *Quercus serrata* seedlings during Spring (March-April) and Autumn (September-October) 2014

4.4.3. Qualitative and quantitative estimation of fungal population of soil from *Quercus serrata* plantation at Umrangso REC Farm.

Mature plants soil: A total of sixteen fungal species were isolated from the soil of *Quercus serrata* plantation at Umrangso REC Farm during 2013 and 2014 (Table:13,14) In Spring season *Aspergillus flavus, Aspergillus fumigatus, Aspergillus niger, Alternaria alternata, Curvularia* sp. *Cladosporium cladosporides, Cladosproium herbarum, Colletotrichum gloeosporiodes, Fusarium solani, Fusarium oxysporium, Trichoderma harizanum Aureobasidium pullulans, Mucor* sp, *Penicellium* sp and Sterile mycelia in *rhizosphere* soil. *Aspergillus flavus. A.fumigatus , A.niger, Alternaria alternata, A.pullulans, Curvularia* sp *Cladosporium clodosporides, Cladosporium herbarum, Fusarium* sp, *Penicillum* sp *Trichoderma harzianum, Mucor* sp, *Gliocladium* sp and *Sterile mycelia* in non–rhizosphere soil. But *Cladosporium herbarum* and *Gliocladium* sp were not found in rhizoplane soil. *Aspergillus* sp was dominant in all rhizosphere, non-rhizosphere and rhizoplane soil in spring season, followed by *Fusarium* sp.

A total eighteen fungal species were isolated from the soil of *Q.serrata* plantation in autumn season. *Aspergillus flavus, A.fumigatus, A.niger, A.terreus, Aspergillus* sp, *Alternaria alternata, Alternaria* sp, *Aureobasidium pullants, Curvularia* sp, *Cladosporium clodosporides, Cladosporium herbarum, Fusarium solani, Fusarium oxysporium, Mucor* sp, *Penicillum* sp, *Rhizopus* sp (P.P.No 6a and 6b), *Trichoderma harizanum* and Sterile mycelia in RS soil, *Cladosporium clodosporides* in NRS and *Cladosporium herbarum* were not found in RP soil. *Aspergillus* was dominant in all rhizosphere, non-rhizosphere and rhizoplane, followed by *Fusarium*> *Alternaria*> *Penicillum* sp> *Trichoderma, etc.*

Table 13: Relative abundance% Fungal isolates of RS, NRS and RP soil of *Quercus serrata* plantation during Spring season 2013 and 2014.

Sl.No.	Fungal isolates	Relative abundance %						Remarks
		2013			2014			
		RS	NRS	RP	RS	NRS	RP	
1	*Alternaria alternata*	9.0	6.0	5.5	9.0	6.6	5.6	
2	*Aspergillus flavus*	13.5	10.0	11.0	13.0	10.0	11.2	
3	*Aspergillus fumigatus*	7.0	8.0	8.5	7.0	6.6	8.3	
4	*Aspergillus niger*	13.5	18.0	14.0	15	16.8	13.9	Dominant
5	*Aureobasidium pullulans*	4.5	4.0	5.5	4.5	3.4	5.6	
6	*Curvularia* sp	2.5	4.0	8.5	2.0	3.4	8.3	
7	*Clado sporium clodosporides*	4.5	4.0	3.0	4.5	3.4	2.7	
8	*Clado sporium herbarum*	4.5	10.0	0.0	4.5	10.0	0.0	
9	*Collectotrichum gloeosporiodes*	4.5	7.0	5.5	4.5	6.6	5.6	
10	*Fusarium solani*	9.0	0.0	11.0	9.0	0.0	11.2	
11	*Fusarium oxysporium*	7.0	7.0	8.5	7.0	6.6	8.3	
12	*Mucor* sp	4.5	4.0	8.5	4.5	3.4	8.3	
13	*Penicillum* sp	4.5	4.0	5.5	4.5	6.6	5.6	
14	*Gliocladium* sp	0.0	4.0	0.0	0.0	3.4	0.0	
15	*Trichoderma harizanum*	7.0	6.5	2.5	7.0	6.6	2.7	
16	*Sterile mycelia*	4.5	3.5	2.5	4.0	6.6	2.7	

- **RS: Rhizosphere; NRS: Non. Rhizosphere** ; **RP: Rhizoplane**

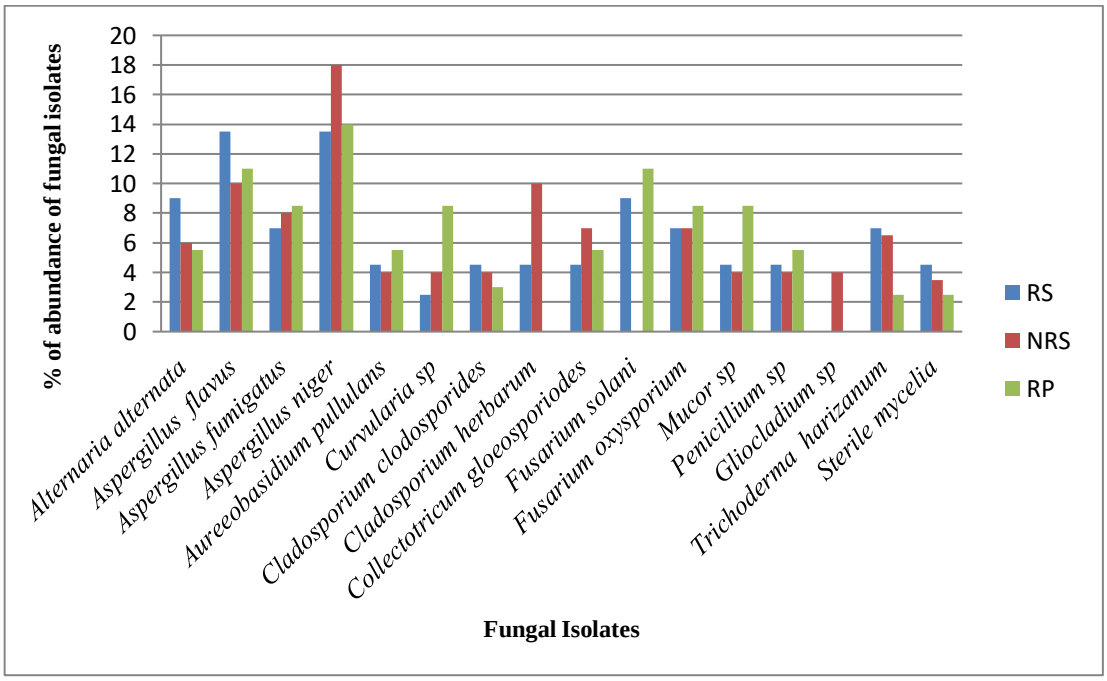


Fig 6: Fungal population density of plantation (*Quercus serrata*) soil in Spring (March-April) in 2013.

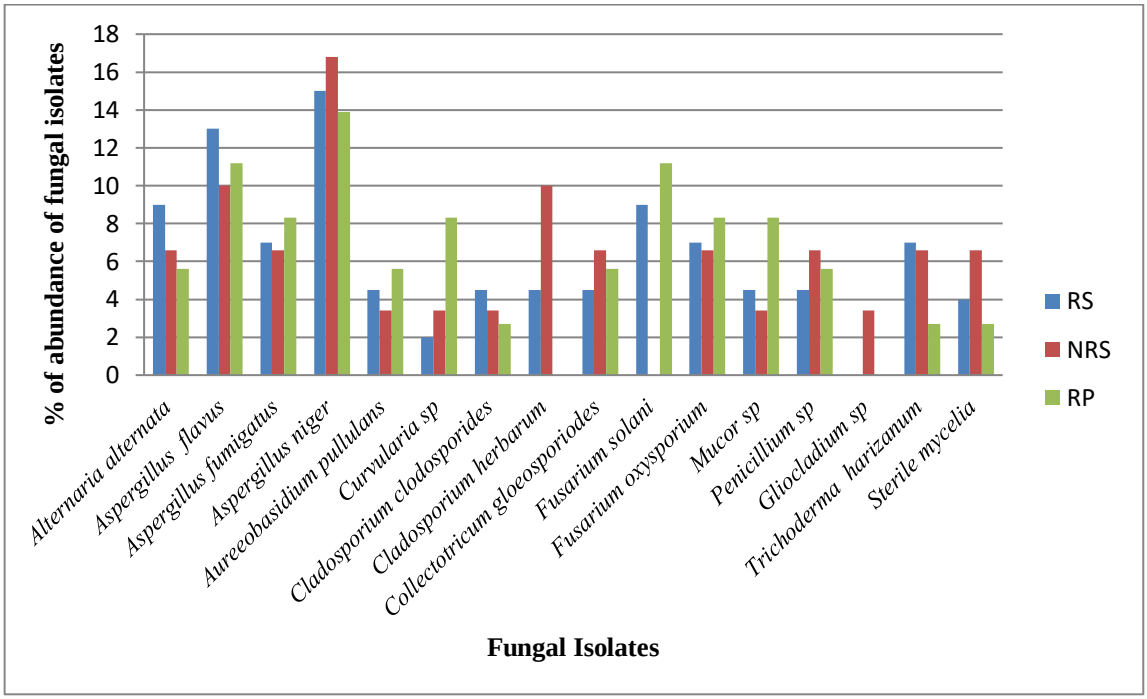
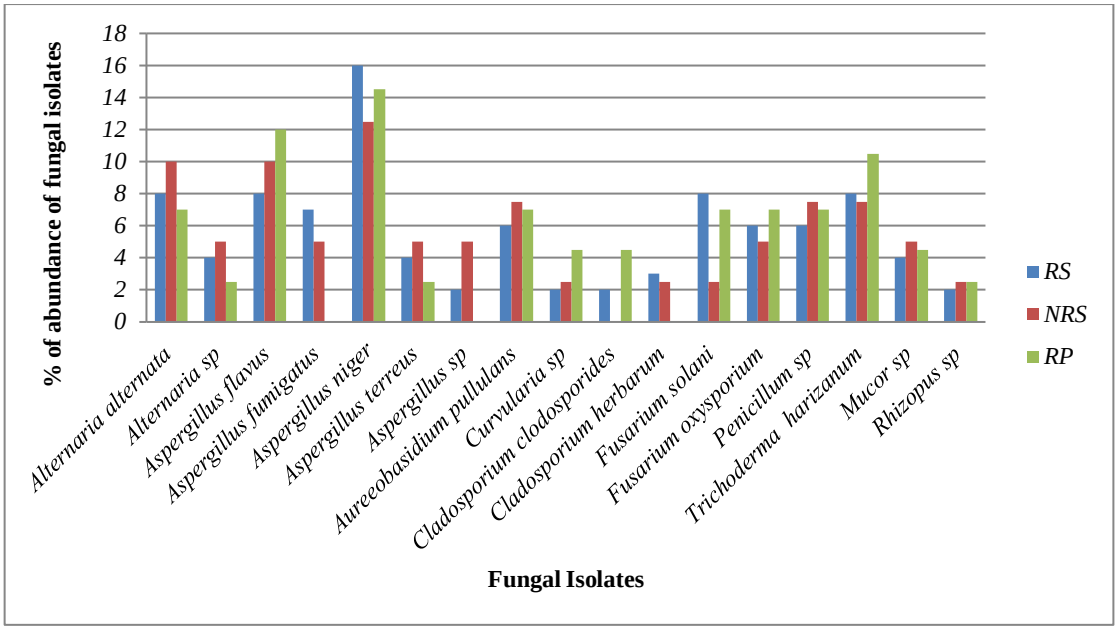


Fig 7: Fungal population density of plantation (*Quercus serrata*) soil in Spring (March-April), 2014.

Table 14: Relative abundance% Fungal isolates of RS, NRS and RP soil of *Quercus serrata* plantation during Autumn season (September-October), 2013 and 2014.

Sl.No.	Fungal isolates	Relative abundance %						Remarks
		2013			2014			
		RS	NRS	RP	RS	NRS	RP	
1	*Alternaria alternata*	8.0	10.0	7.0	7.5	10.5	9.5	
2	*Alternarea* sp	4.0	5.0	2.5	4.0	5.0	2.5	
3	*Aspergillus flavus*	8.0	10.0	12.0	12.0	10.6	14.5	
4	*Aspergillus fumigatus*	7.0	5.0	-	7.5	7.8	7.5	
5	*Aspergillus niger*	16.0	12.5	14.5	16	13.0	16.5	Dominant
6	*Aspergillus terreus*	4.0	5.0	2.5	2.0	2.5	2.5	
7	*Aspergillus* sp	2.0	5.0	-	4.0	5.0	-	
8	*Aureobasidium pullulans*	6.0	7.5	7.0	5.9	5.0	4.5	
9	*Curvularia* sp	2.0	2.5	4.5	4.0	5.0	2.5	
10	*Cladosporium clodosporides*	2.0	-	4.5	2.0	-	4.5	
11	*Cladosporium herbarum*	3.0	2.5	-	2.0	2.5	-	
12	*Fusarium solani*	8.0	2.5	7.0	7.6	2.5	7.5	
13	*Fusarium oxysporium*	6.0	5.0	7.0	5.9	7.8	7.5	
14	*Penicillum* sp	6.0	7.5	7.0	5.8	7.8	4.5	
15	*Trichoderma harizanum*	8.0	7.5	10.5	5.8	5.0	6.5	
16	*Mucor* sp	4.0	5.0	4.5	4.0	2.5	4.5	
17	*Rhizopus* sp	2.0	2.5	2.5	2.0	2.5	2.5	
18	*Sterile mycelia*	4.0	5.0	2.5	2.0	5.0	2.5	

RS: Rhizosphere; NRS: Non Rhizosphere ; RP: Rhizoplane



RS: Rhizosphere; NRS: Non Rhizosphere ; RP: Rhizoplane

Fig 8: Fungal population density of plantation (*Quercus serrata*) soil in Autumn (Sept-Oct), 2013.

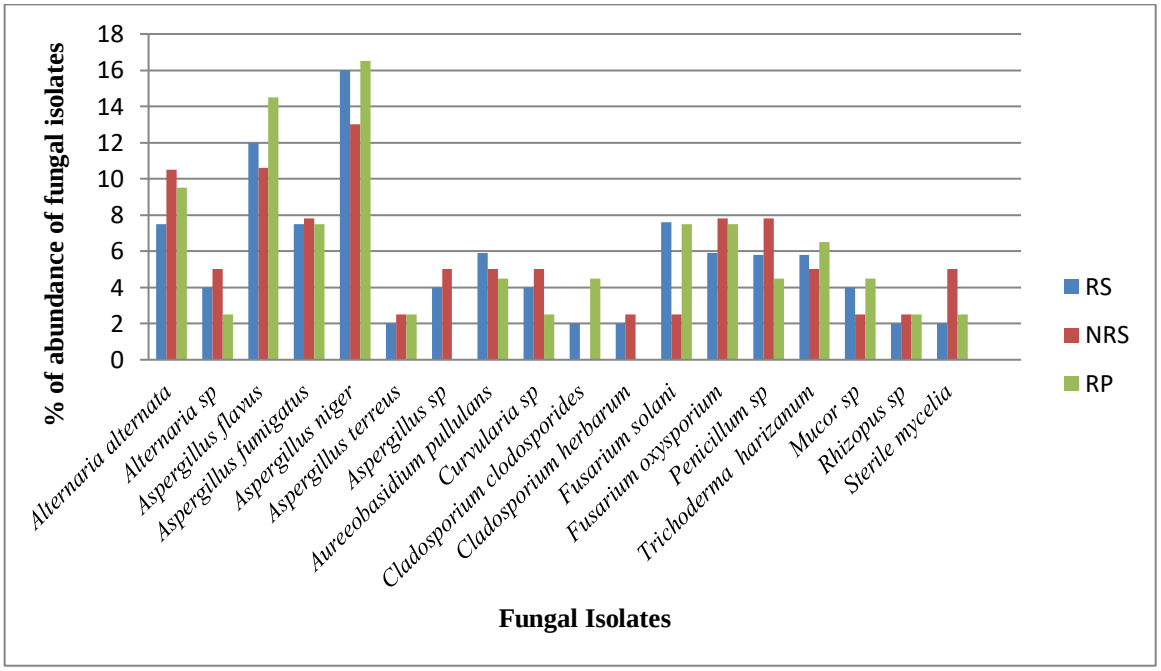


Fig 9: Fungal population density of plantation (*Quercus serrata*) soil in Autumn (Sept-Oct), 2014.

4.4.4. Qualitative and quantitative study of mycoflora of *Quercus serrata* plantation during Spring and Autumn season.

A total eleven fungal species were isolated from air over *Quercus serrata* plantation during Spring and Autumn season during 2013 and 2014 at 0.75 meters at 1.50 meters height(Table:15,16). *Aspergillus flavus, A.fumigatus, A.niger, Alternaria alternata, Cladosporium* sp, *Colletotrichum* sp, *Curvularia* sp *Mucor* sp *Penicillum* sp, Sterile mycelia were found in Spring and Autumn season, in additional *Fusarium* sp found. In autumn season only in 0.75 meters height *Aspergillus* was dominant, followed by *Alternaria*> *Cladosporium*> *Penicillum* autumn season. *Penicillum* sp and *Fusarium* sp were not found in spring season but found in autumn season at 1.50 meters height.

Table 15: Relative abundance% Fungal isolates from air over (*Quercus serrata* Plantation during Spring (March-April) and Autumn season (September-October), 2013 and 2014 at 0.75meter height.

Status of height 0.75 meter	Fungal isolates	Relative abundance%			
		2013		2014	
		March- April	Sept-Oct	March- April	Sept-Oct
	Alternaria alternata	12.0	10.0	12.0	10.00
	Aspergillus flavus	14.0	12.0	12.5	15.5
	Aspergillus niger	20.0	22.0	20.0	20.0
	Aspergillus fumigatus	12.0	10.0	5.0	7.0
	Cladosporium sp	10.0	8.0	8.0	7.0
	Colletotrichum sp	5.0	6.0	5.0	4.0
	Curvularia sp	5.0	6.0	8.0	8.0
	Fusarium sp	0.0	4.0	0.0	3.5
	Mucor sp	8.0	8.0	12.5	7.5
	Penicillum sp	8.0	10.0	9.0	12.0
	Sterile mycelia	6.0	11.0	8.5	5.5

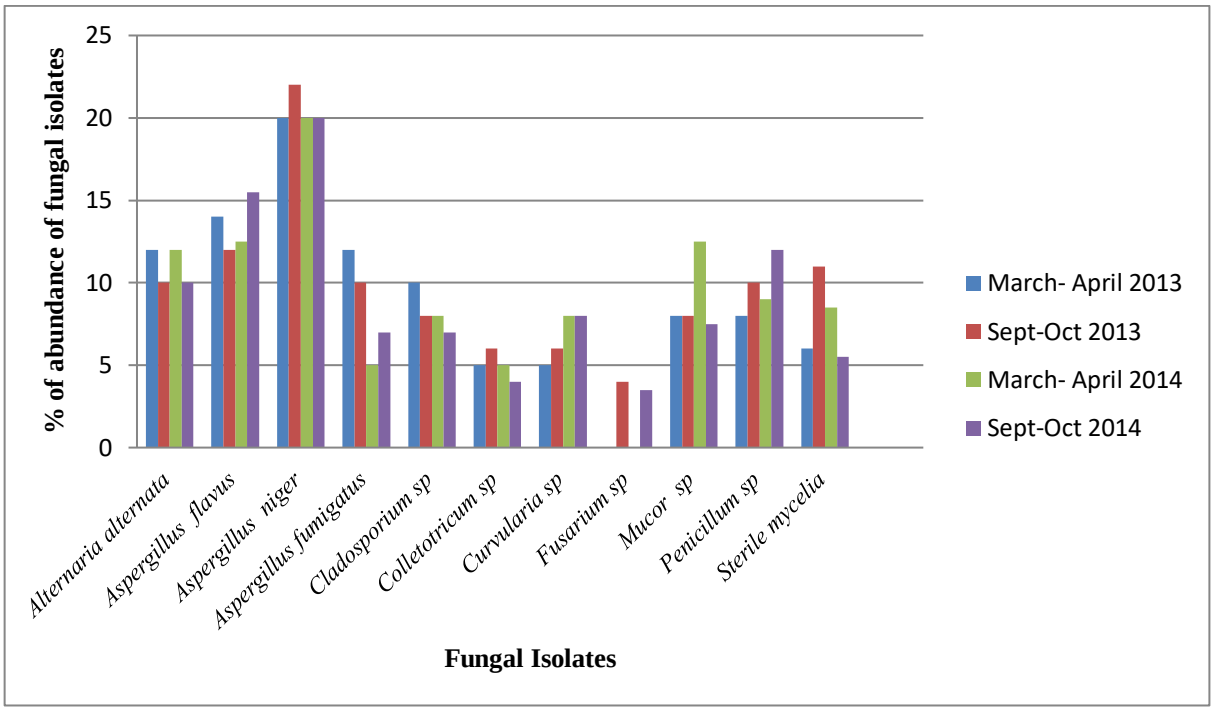


Fig 10: Fungal population density of from air over (*Quercus serrata)* plantation during Spring (March-April) and Autumn season (September-October), 2013 and 2014 at 0.75 meter.

Table 16: Relative abundance% Fungal isolates from air over (*Quercus serrata)* plantation during Spring (March-April) and Autumn season (September-October), 2013 and 2014 at height 1.5 meters.

Status of height meter	Fungal isolates	Relative abundance%			
		2013		2014	
		March- April	Sept-Oct	March- April	Sept-Oct
1.50 meter	*Alternaria alternata*	12.0	10.00	13.0	10.00
	Aspergillus flavus	15.0	14.0	16.0	14.0
	Aspergillus niger	22.0	20.0	24.0	23.0
	Aspergillus fumigatus	8.0	4.0	8.0	4.5
	Cladosporium sp	10.0	8.0	11.0	9.0
	Colletotrichum sp	8.0	8.0	5.5	12.5
	Curvularia sp	8.0	6.0	5.5	4.5
	Fusarium sp	0.0	6.0	0.0	0.0
	Mucor sp	6.0	8.0	5.5	8.0
	Penicillum sp	0.0	10.0	0.0	10.0
	Sterile mycelia	11.0	6.0	11.5	4.5

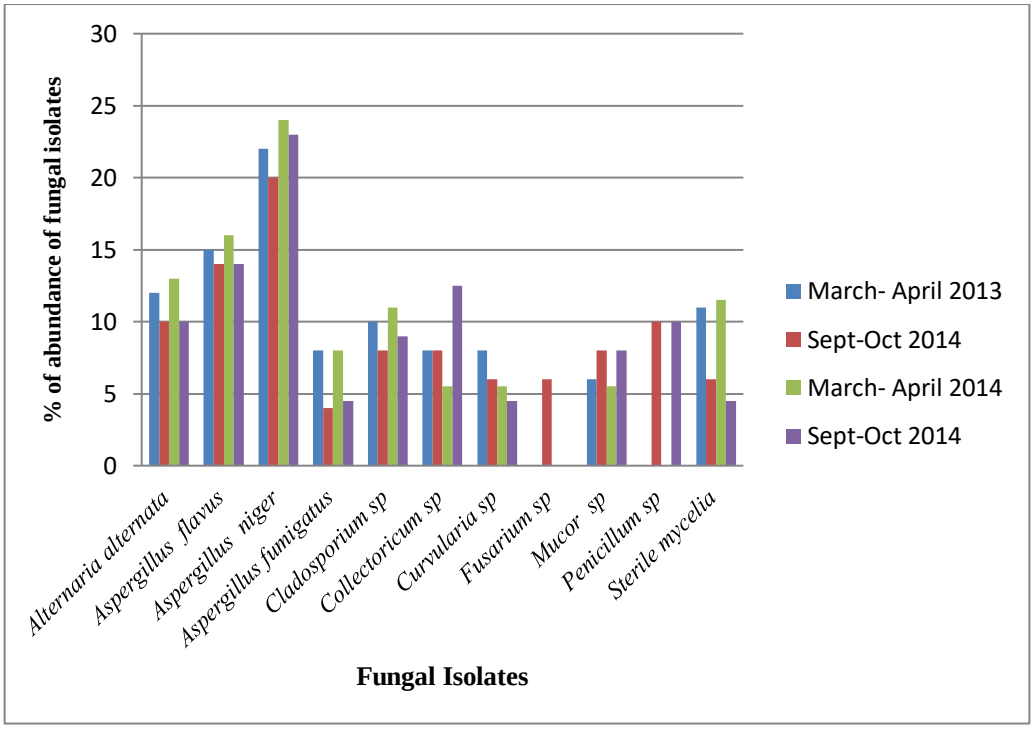


Fig 11: Fungal population density of from air over (*Quercus serrata* plantation during Spring (March- April) and Autumn season (September-October), 2013 and 2014 at 1.5 meters.

4.5 Physico-chemical characters of soil under *Quercus serrata* plantation at R.E.C.Umrangso Farm.

Organic carbon percentage and Nitrogen (ppm) of the soil is more when applied FYM+NPK than control. But Phosphous and Potash (ppm) is more in control where any FYM or NPK not applied.

Analysis of soil from Umrangso Research Extension Centre, farm were conducted (Table:17). In fertilized soil pH 4.51, Organic Carbon 2.23%, average Nitrogen 331.90 ppm, Phosphate 32.00 ppm and Potash 75.00 ppm. But in non fertilized soil pH 4.61, was little higher than fertilized soil Organic Carbon 1.56%, which was laser than fertilized soil availability of Nitrogen 231.14 ppm which was very much less then fertilized soil. Availably of Phosphous was observed slightly more *i.e.* 36.00 ppm but availability of Potash was observed 150.00ppm which was almost double than fertilized soil.

Table 17: Physico-chemical characters of soil under *Quercus serrata* plantation at R.E.C.Umrangso Farm.

SL No.	Particulars	pH	O.C.(%)	Av.N_2 (ppm)	Av.P (ppm)	Av.K (ppm)
1	Fertilizer	4.51	2.23	331.90	32.00	75.00
2	Non- fertilizer	4.61	1.56	231.14	36.00	150.00

4.6 Foliar constituents of Oak leaf *(Quercus serrata)* 2013 and 2014 under different treatments.

4.6.1 Treatment A (Control without application of NPK and FYM):

Foliar constitution of ***Quercus serrata*** where plants were without application of NPK and FYM (Control) in Spring (March-April) and Autumn (September-October) in 2013 and 2014 in Spring season average leaf moisture 60.96%, crude protein 7.185%, Crude Fibre 4.685%, Crude Fat 1.765%, Ash 1.42%, Carbohydrates 6.46% and ERR 31.50%.

In Autumn Season Leaf Moisture 51.10%, Crude Protein 4.10%, Crude Fibre 4.545%, Crude Fat 1.605%, Ash 1.41%, Carbohydrates 14.85% and ERR 16.20% were found.

Table 18: Foliar constituents of Oak leaf (Q*uercus serrata*) 2013 and 2014 plants were treated without FYM and NPK.

Season	Moisture%	Crude protein%	Crude fibre%	Crude fat%	Ash%	Carbohydrates %	ERR%
Spring 2013	61.01	7.20	4.757	1.83	1.44	6.5	31.50
Spring 2014	60.962	7.2	4.70	1.70	1.40	6.431	
Mean	60.96	7.185	4.685	1.765	1.42	6.46	
Autumn 2013	51.10	4.10	4.60	1.61	1.41	14.80	16.20
Autumn 2014	51.10	4.10	4.49	1.60	1.415	14.9	
Mean	51.10	4.10	4.545	1.605	1.41	14.85	

4.6.2 Treatment B: Foliar Constituents application of ***Quercus serrata,*** where plants were with application NPK and FYM in Spring and Autumn Season in 2013 and 2014. Application per plant 48.0gm urea, 46.5gm single super phosphate 9.3gm murate of potash and 10kg FYM.

In the Spring season average leaf moisture 68.98%, Crude Protein 10.28%, Crude Fibre 6.78%, Crude Fat 2.34%, Ash 1.88%, Carbohydrates 10.80% and ERR 64.50% were found, but in Autumn season found less Leaf Moisture 57.23%, Crude Protein 5.13%, a little more Crude Fibre 7.09%, Crude Fat 1.94%, Ash 1.90%, but Carbohydrates more 21.33% almost double than Spring Season ERR 31.65% which was less than Spring Season.

Table 19: Foliar constituents of Oak leaf *(Quercus serrata)* in 2013 and 2014 where plants were applied with FYM and NPK.

Season	Moisture%	Crude protein%	Crude fibre%	Crude fat%	Ash%	Carbohydrates %	ERR%
Spring 2013	68.62	10.27	6.76	2.32	1.88	10.83	64.50
Spring 2014	69.35	10.30	6.80	2.36	1.88	10.78	
Mean	68.98	10.28	6.78	2.34	1.88	10.80	
Autumn 2013	57.20	5.08	7.08	1.95	1.92	21.00	31.65
Autumn 2014	57.27	5.18	7.10	1.93	1.88	21.66	
Mean	57.23	5.13	7.09	1.94	1.90	21.33	

4.6.3. Treatment C : Foliar Constituents application of ***Quercus serrata,*** where plants were with application FYM in Spring and Autumn Season in 2013 and 2014. Application per plant 10kg FYM.

In the Spring season average leaf moisture, Crude Protein, Crude Fibre, Crude Fat, Ash, Carbohydrates, and ERR were recorded 64.14%, 8.10%, 5.18%, 2.09%, 1.64%, 8.26%, and 43.50% respectively in Autumn season found leaf moisture, Crude Protein, Crude Fibre, Crude Fat, Ash, Carbohydrates, and ERR were recorded 54.10%, 4.17%, 5.17%, 1.80%, 1.61%, 18.20%, and 23.85%.

Table 20: Foliar constituents of Oak leaf *(Quercus serrata)* in 2013and 2014 where plants applied with FYM.

Season	Moisture%	Crude protein%	Crude firbe%	Crude fat%	Ash%	Carbohydrates %	ERR%
Spring 2013	61.198	8.09	5.197	2.089	1.638	8.233	43.50
Spring 2014	64.078	8.11	5.159	2.101	1.643	8.285	
Mean	64.14	8.10	5.18	2.09	1.64	8.26	
Autumn 2013	54.088	4.162	5.164	1.809	1.598	18.18	23.85
Autumn 2014	54.115	4.177	5.186	1.789	1.617	18.23	
Mean	54.10	4.17	5.17	1.80	1.61	18.20	

The result of foliar constituents of *Quercus serrata* under different of 2013 and 2014 in Spring and Autumn season (Table;18,19,20). In Spring season more leaf moisture %,crude protein and higher effective rate of recorded but in Autumn season carbohydrates % more and crude protein % and effective rate of rearing was less recorded in all three different treatment of *Quercus serrata* leaves. The better results were recorded in treatment (B) *i.e.*. leaf moisture, crude protein, crude fibre, crudefat, ash, carbohydrates and Effective Rate of Rearing (ERR%) 68.98%,10.28%,6.78%,2.34%,1.88%,10.80% and 64.50% respectively and during Autumn season leaf moisture 57.23%,crude protein 5.13%,crude fibre 7.09%,crude fat 1.94% ash 1.90%,carbohydrates 21.33% and ERR 31.60%.When in *Q.serrata* plants were applied recommended dose FYM and NPK got better quality and quantity leaves and better silk rearing performance.

Table 21: Leaf constituents of *Q.serrata* under different treatments Spring Crop 2013 and Statistical analysis.

Treatment Replication	Moistue(%)			Crude protein			Crude fibre			Crude fat(%)			Ash			Carbohydrate		
	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM
R1	61.1	68.2	64.5	7.35	10.27	8.24	4.85	6.76	5.25	1.85	2.32	2.10	1.45	1.88	1.65	6.50	10.83	8.25
R2	61.15	68.50	64.15	7.2	10.35	8.15	4.8	6.70	5.10	1.80	2.88	2.05	1.42	1.90	1.60	6.45	10.86	8.30
R3	60.80	68.45	64.30	7.25	10.25	7.92	4.82	6.82	5.24	1.82	2.36	2.12	1.40	1.86	1.62	6.50	10.80	8.24
R4	60.85	68.75	64.18	7.28	10.23	8.1	4.88	6.65	5.15	1.86	2.30	2.20	1.43	1.88	1.65	6.48	10.78	8.32
R5	60.80	68.30	63.85	7.10	10.30	7.96	4.75	6.68	5.18	1.82	2.35	2.05	1.45	1.86	1.68	6.45	10.85	8.35
R6	61.20	68.20	64.20	7.12	10.27	7.90	4.72	6.85	5.12	1.80	2.30	2.10	1.48	1.94	1.62	6.54	10.86	8.18
R7	60.75	68.65	64.75	7.15	10.35	8.10	4.68	6.82	5.18	1.88	2.25	2.05	1.40	1.86	1.64	6.50	10.82	8.10
R8	61.30	69.15	64.35	7.20	10.30	8.15	4.65	6.80	5.20	1.84	2.32	2.08	1.45	1.84	1.70	6.60	10.80	8.15
R9	61.25	68.48	64.48	7.25	10.25	8.24	4.70	6.75	5.35	1.80	2.40	2.08	1.46	1.86	1.62	6.35	10.80	8.24
R10	60.90	69.10	63.42	7.10	10.15	8.18	4.72	6.65	5.2	1.82	2.32	2.06	1.48	1.92	1.60	6.48	10.90	8.20
Total	610.10	686.20	641.98	72.00	100.70	80.94	47.57	67.48	51.97	18.29	23.20	20.89	14.42	18.80	16.38	64.85	108.30	82.33
Mean	61.10	68.62	64.19	7.20	10.07	8.09	4.757	6.748	5.197	1.829	2.320	2.089	1.442	1.880	1.638	6.485	10.830	8.233
SED(±)	0.041			0.003			0.003			0.001			0.000			0.002		
CD(5%)	0.072			0.006			0.005			0.001			0.001			0.004		

In Spring season 2013, the leaf Moisture of *Q.serrata* under different treatment SED(±) 0.41 and CD (5%) 0.072 which was not significant. Crude Protein SED(±) 0.003 and CD(5%) 0.006 which was not significant. Crude Fibre SED(±) 0.003 and CD(5%) 0.005, which was not significant. Crude Fat of *Q.serrata* under different treatment SED(±) 0.001 and CD (5%) 0.001, Ash SED(±) 0.000 and CD(5%) 0.001, Carbohydrate SED(±) 0.002 and CD(5%) 0.004 which was not significant among the different treatment.

Table 22: Leaf constituents of *Q.serrata* under different treatments Autumn Crop 2013 and Statistical analysis.

Treatment Replication	Moistue(%)			Crude protein			Crude fibre			Crude fat(%)			Ash			Carbohydrate		
	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM
R1	51.20	57.30	57.20	4.10	5.08	4.15	4.60	7.08	5.1	1.62	1.95	1.82	1.40	1.92	1.60	14.50	21.80	18.50
R2	50.85	57.00	54.10	4.08	5.04	4.20	4.58	7.10	5.16	1.60	1.93	1.78	1.42	1.94	1.62	14.80	21.50	17.80
R3	50.78	57.50	54.24	4.05	5.10	4.12	4.55	7.06	5.08	1.56	1.98	1.80	1.40	1.90	1.58	14.60	20.80	18.20
R4	50.60	57.20	54.30	4.15	5.06	4.18	4.52	7.05	5.14	1.64	1.96	1.85	1.38	1.90	1.56	14.80	21.20	18.00
R5	51.35	57.00	53.85	4.08	5.12	4.16	4.56	7.08	5.10	1.62	1.92	1.84	1.42	1’92	1.54	14.50	21.30	18.40
R6	51.40	56.80	54.20	4.06	5.06	4.08	4.48	7.12	5.18	1.56	1.94	1.78	1.42	1.90	1.60	15.00	21.20	18.00
R7	51..30	57.20	53.80	4.10	5.10	4.25	4.52	7.10	5.20	1.64	1.90	1.84	1.44	1.96	1.64	15.20	20.60	18.30
R8	51.40	57.50	53.75	4.05	5.04	4.14	4.58	7.12	5.24	1.60	1.98	1.76	1.40	1.94	1.62	14.80	20.80	18.60
R9	50.80	56.80	54.25	4.12	5.08	4.16	4.64	7.08	5.20	1.60	1.94	1.80	1.38	1.92	1.58	15.00	21.20	17.80
R10	51.30	57.70	54.15	4.08	5.12	4.18	4.60	7.01	5.24	1.64	2.0	1.82	1.40	1.90	1.64	14.50	20.40	1`8.20
Total	510.98	572.00	540.88	40.87	50.80	41.62	45.63	70.80	51.64	16.10	19.50	18.09	14.060	19.20	15.98	14.77	210.00	181.80
Mean	51.098	57.20	54.088	4.087	5.080	4.162	4.56	7.08	5.164	1.61	1.95	1.809	1.406	1.92	1.598	1.477	21.00	18.18
SED(±)	0.040			0.005			0.001			0.000			0.00017			0.048		
CD(5%)	0.069			0.0008			0.002			0.001			0.00030			0.089		

In Autumn season 2013,Leaf Moisture of *Q.serrata* under different treatment SED(±) 0.040 and CD (5%) 0.069, Crude Protein SED(±) 0.005 and CD(5%) 0.0008, Crude Fibre SED(±) 0.001 and CD(5%) 0.002 which were not significant among the different treatments. Crude Fat of *Q.serrata* under different treatment SED(±) 0.000 and CD (5%) 0.001, Ash SED(±) 0.00017 and CD(5%) 0.00030, Carbohydrate SED(±) 0.048 and CD(5%) 0.089 which were not significant among the different treatment.

Table 23: Leaf constituents of *Q.serrata* under different treatments Spring Crop 2014 and Statistical analysis.

Treatment Replication	Moistue(%)			Crude protein			Crude fibre			Crude fat(%)			Ash			Carbohydrate		
	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM
R1	61.25	69.40	63.50	7.25	10.35	8.14	4.75	6.85	5.15	1.65	2.38	2.15	1.48	1.89	1.58	6.80	10.76	8.35
R2	61.12	69.30	64.25	7.15	10.25	8.25	4.60	6.80	5.12	1.70	2.34	2.08	1.45	1.87	1.70	6.40	10.80	8.30
R3	60.50	69.25	64.30	7.20	10.30	7.90	4.72	6.75	5.25	1.62	2.36	2.10	1.42	1.85	1.64	6.35	10.74	8.45
R4	60.55	69.45	64.28	7.18	10.25	8.15	4.68	6.70	5.10	1.76	2.34	2.18	1.25	1.88	1.65	6.28	10.82	8.30
R5	60.70	69.30	63.65	7.15	10.35	7.95	4.65	6.85	5.15	1.62	2.40	2.15	1.35	1.86	1.62	6.25	10.72	8.35
R6	61.25	69.40	64.30	7.14	10.40	7.92	4.70	6.90	5.10	1.70	2.35	2.10	1.38	1.85	1.68	6.50	1o.80	8.20
R7	60.35	69.20	64.55	7.18	1O.20	8.15	4.62	6.80	5.14	1.68	2.35	2.06	1.45	1.89	1.64	6.70	10.74	8.15
R8	61.30	69.50	64.25	7.12	10.25	8.25	4.68	6.75	5.25	1.64	2.36	2.08	1.42	1.84	1.66	6.40	1o.82	8.20
R9	61.20	69.30	64.38	7.20	10.35	8.20	4.71	6.85	5.15	1.75	2.35	2.05	1.36	1.90	1.62	6.25	1o.78	8.25
R10	60.8	69.40	63.32	7.15	10.30	8.15	4.73	6.75	5.18	1.80	2.37	2.06	1.40	1.87	1.64	6.38	10.82	8.30
Total	609.02	693.50	640.78	71.72	103.00	81.060	46.84	68.00	51.590	16.92	23.60	21.010	13.960	18.70	16.430	64.310	107.80	82.850
Mean	60.902	69.35	64.078	7.172	10.30	8.106	4.684	6.80	5.159	1.692	2.360	2.101	1.396	1.87	1.643	6.431	10.78	8.285
SED(±)	0.050			0.004			0.001			0.001			0.001			0.008		
CD(5%)	0.086			0.007			0.002			0.002			0.002			0.0013		

Leaf Moisture of *Q.serrata* under different treatment SED(±) 0.050 and CD (5%) 0.086, Crude Protein SED(±) 0.004 and CD(5%) 0.007, Crude Fibre SED(±) 0.001 and CD(5%) 0.002 which were not significant. Crude Fat of *Q.serrata* under different treatment SED(±) 0.001 and CD (5%) 0.002, Ash SED(±) 0.001 and CD(5%) 0.002, Carbohydrate SED(±) 0.008 and CD(5%) 0.0013 which were not significant among the different treatment.

Table 24:Leaf constituents of *Q.serrata* under different treatments Autumn Crop 2014 and Statistical analysis.

Treatment Replication	Moistue(%)			Crude protein			Crude fibre			Crude fat(%)			Ash			Carbohydrate		
	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM
R1	51.35	57.30	54.15	4.15	5.20	4.25	4.50	7.10	5.15	1.64	1.93	1.72	1.45	1.90	1.65	14.80	21.68	18.80
R2	50.65	57.25	54.10	4.18	5.16	4.20	4.68	7.14	5.20	1.62	1`.91	1.75	1.40	1.84	1.60	14.60	21.60	18.00
R3	50.48	57.20	54.25	4.02	5.18	4.18	4.45	7.08	5.10	1.50	1.96	1.80	1.42	1.90	1.58	14.50	22.10	18.20
R4	50.65	57.25	54.30	4.05	5.15	4.15	4.62	7.06	5.15	1.65	1.88	1.86	1.35	1.92	1.60	14.80	21.60	18.50
R5	51.15	57.20	53.65	4.12	5.20	4.25	4.46	7.12	5.16	1.52	1.92	1.82	1.32	1.88	1.64	14.60	21.70	18.40
R6	51.45	57.20	54.20	4.08	5.22	4.08	4.40	7.08	5.10	1.56	1.94	1.78	1.45	1.86	1.67	15.20	21.75	18.20
R7	51.36	57.30	53.85	4.10	5.16	4.15	4.32	7.14	5.20	1.54	1.91	1.80	1.48	1.84	1.64	15.40	21.60	18.50
R8	51.45	57.26	54.10	4.08	5.14	4.20	4.28	7.06	5.25	1.65	1.93	1.74	1.46	1.85	1.62	14.80	21.67	18.40
R9	50.60	57.10	54.35	4.06	5.24	4.16	4.54	7.10	5.30	1.62	1.94	1.82	1.36	1.87	1.60	15.40	21.20	17.80
R10	51.50	57.64	54.20	4.08	5.15	4.15	4.62	7.12	5.25	`1.64	1.98	1.80	1.46	1.94	1.62	14.80	21.80	17.50
Total	510.64	572.70	541.150	40.920	51.80	41.77	44.87	71.00	51.860	15.94	19.30	17.890	14.150	18.80	16.170	148.90	216.70	182.30
Mean	51.064	57.27	54.115	4.092	5.18	4.177	4.487	7.10	5.186	1.594	1.930	1.789	1.415	1.88	1.617	14.89	21.67	18.23
SED(±)	0.036			0.0007			0.003			0.001			0.00071			0.052		
CD(5%)	0.063			0.0013			0.005			0.002			0.00123			0.090		

In Autumn season 2013,leaf Moisture of *Q.serrata* under different treatment SED(±) 0.036 and CD (5%) 0.063, Crude Protein SED(±) 0.0007 and CD(5%) 0.0013, Crude Fibre SED(±) 0.003 and CD(5%) 0.005 which were not significant among the different treatments. Crude Fat of *Q.serrata* under different treatment SED(±) 0.001 and CD (5%) 0.002, Ash SED(±) 0.00071 and CD(5%) 0.00123, Carbohydrate SED(±) 0.052 and CD(5%) 0.090 which were not significant among the different treatment.

4.7 The reeling parameter of *A.proylei* **Jolly.** cocoons were taken from three different treatments

A.Cocoons, harvested from the rearing where FYM and NPK not applied in Oak plants.

B.Cocoons, harvested from the rearing where applied FYM and NPK in Oak plants.

C.Cocoons harvested from the rearing where Oak plants were applied FYM only.

Average Filament length 588.92 m, Denier 6.288, Non Breakable Filament Length (NBFL) 298.60 m, Silk recovery 62.20% and Reelability 41.14% were observed from the treatment (A) cocoons. Similarly, cocoons from treatment (B) average Filament length were 662.030 m, Denier 6.148, NBFL 330.40 m, Silk recovery 70.15% and Reelability 46.27%. From cocoons treatment(C) average filament length 627.14 m, Denier 6.225, NBFL 314.30m,Silk recovery 65.130% and Reelability 43.20%. Hence reeling parameter comparative better from the treatments like this B>C>A.

Table 25:Reeling Parameter of *A.proylei* Jolly.cocoon

Filament Treatment	Average Filament length	Denier	NBFL in metres	Recovery %	Reelability %
A	585.92	6.288	298.6	62.20	41.14
B	662.30	6.148	330.4	70.15	46.27
C	627.14	6.225	314.3	65.13	43.20

Photo plate No:**11(a)**: Showing Reeling and **11(b)** Spinng machine, **11(c)**Preparation of Ghicha, **11(d)** Weaving loom, **12 (a)** Reeled yarn **12 (b)** Spun yarn, **12 (c)** Ghicha yarn, **12(d)** Different types oak tasar silk yarns, **13(a)** Oak tasar saree **13(b)** Fabrics (waistcoat, bag and coat)

Table 26: Reeling parameter of *A.proylei* cocoons under different treatments with Statistical analysis.

Treatment Replication	Filament length in meters			Deniar			NBFL			Silk recovery(%)			Reelability (%)		
	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM	Control	NPK+ FYM	FYM Control
R1	590.80	670.50	640.50	6.36	6.05	6.20	298	338	308	62.6	70.5	65.2	41.4	46.5	43.2
R2	590.6	660.80	625.60	6.30	6.18	6.14	306	330	315	62.2	70.2	65.6	41.2	46.2	43.5
R3	565.2	665.60	610’50	6.45	6.12	6.16	292	336	312	62.5	70.8	66.2	40.8	46.6	43.8
R4	595.5	658..80	618.20	6.25	6.24	6.25	296	324	318	62.6	70.2	64.8	40.6	46.4	43.6
R5	580.8	648.20	622.50	6.28	6.15	6.32	303	328	306	62.2	70.6	65.6	40.5	46.8	43.4
R6	588.5	668.40	630.20	6.32	6.10	6.28	301	340	310	62.8	71.2	64.5	41.5	46.2	43.2
R7	605.3	662.50	632.50	6.20	6.12	6.25	296	328	315	61.8	71.5	65.4	41.2	46.4	43.5
R8	570.2	668.80	635.20	6.24	6.20	6.20	294	320	320	61.5	68.8	65.2	41.4	45.8	42.8
R9	596.8	662.50	630.80	6.30	6.14	6.18	298	335	324	61.6	68.5	64.6	41.6	45.6	42.6
R10	575.5	654.20	625.40	6.18	6.18	6.27	302	325	315	62.2	69.2	64.2	41.2	46.2	42.4
Total	5859.20	6620.30	6271.40	62.88	61.48	62.250	2986	3304	3143	622.0	701.50	651.30	411.4	462.70	432.0
Mean	585.92	662.30	627.14	6.288	6.148	6.225	298.6	330.4	314.3	62.20	70.15	65.13	41.14	46.27	43.20
SED(±)	32.002			0.003			16.178			0.158			0.086		
CD(5%)	55.491			0.004			28.988			0.274			0.149		

The filament length SED(±) 32.002 and CD at 5% (55.492) under different treatment which was highly significant. The Deniar SED(±) 0.003 and CD at 5% (0.004) was found not significant. The NBFL SED(±) 16.178 and CD at 5% (28.988 which was highly significant. Silk recovery % SED(±) 0.158 and CD at 5% (0.274%) which was not significant. Reelability % SED(±) 0.086 and CD at 5% (0.149) which was not significant.

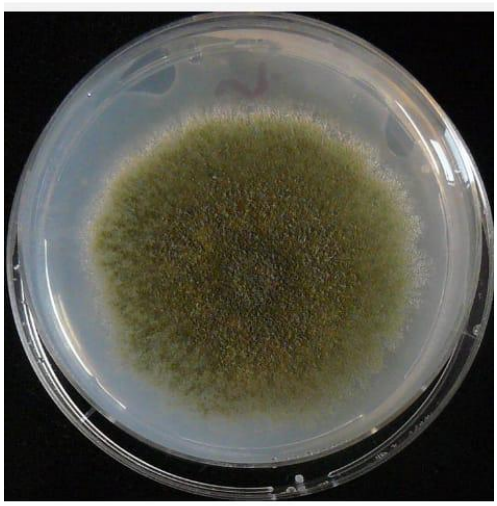
Photo Plate: 1

1(a) Plantation of *Quercus serrata*
at REC,Umrangso,food plant of *Antheraea proylei* J.

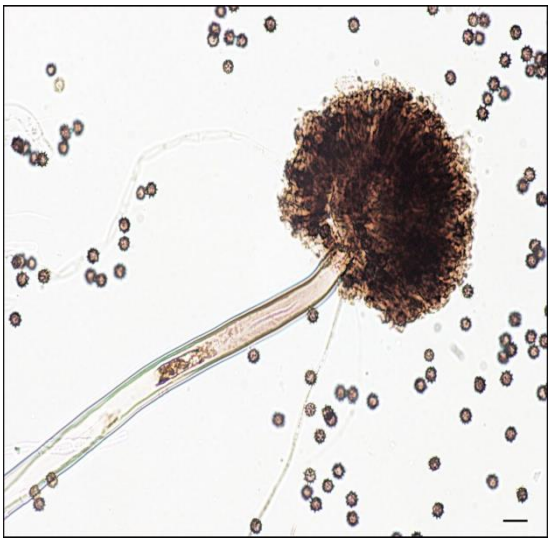
1(b) *Quercus serrata* seedlings.

Plantation of *Quercus serrata and Quercus serrata* seedlings

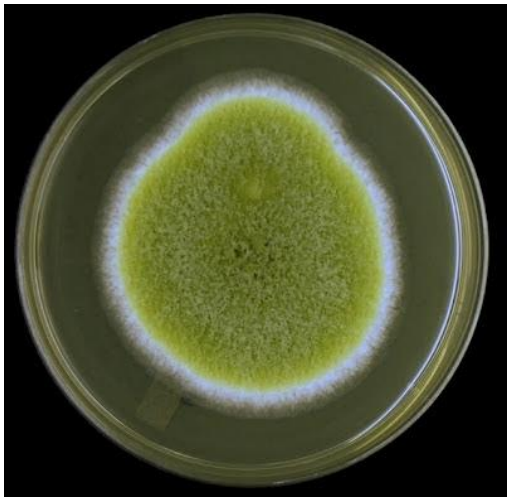
Photo Plate : 2



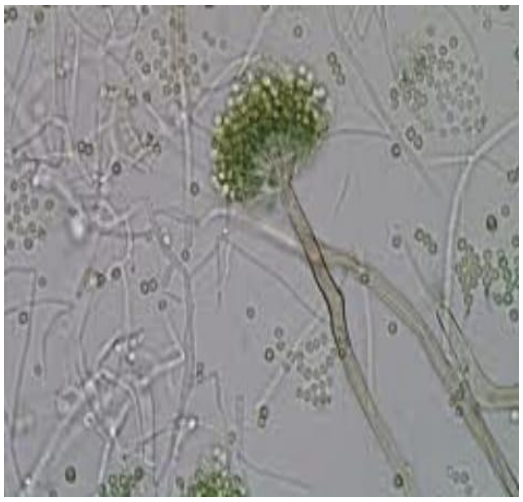
2(a) *Aspergillus niger* culture in petridish



2(b) *Aspergillus niger*

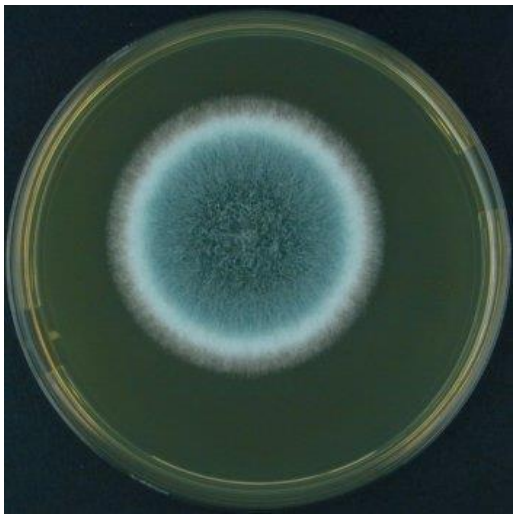


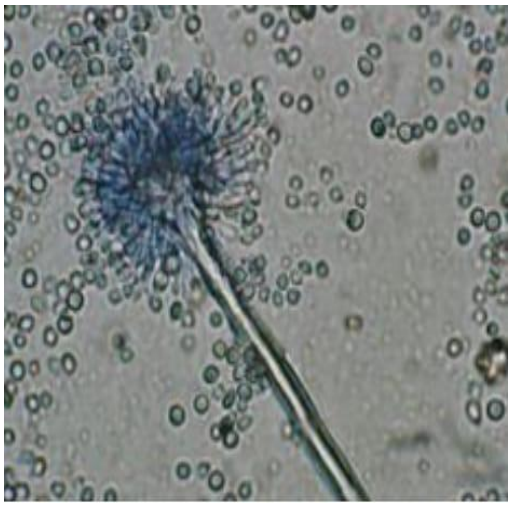
2(c) *Aspergillus flavus* culture in pertridish



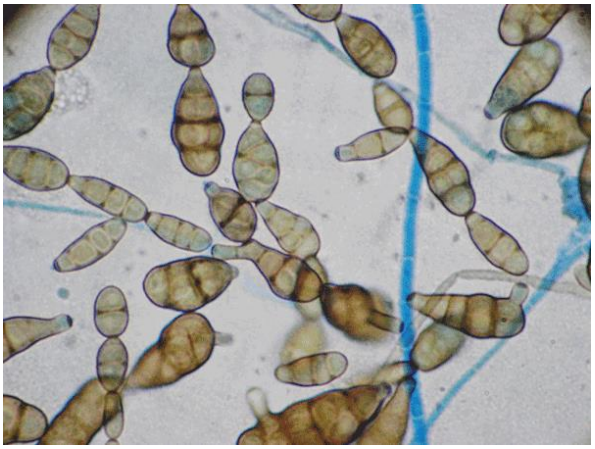
2(d) *Aspergilus flavus* spore with conidiophores

Photo Plate: 3

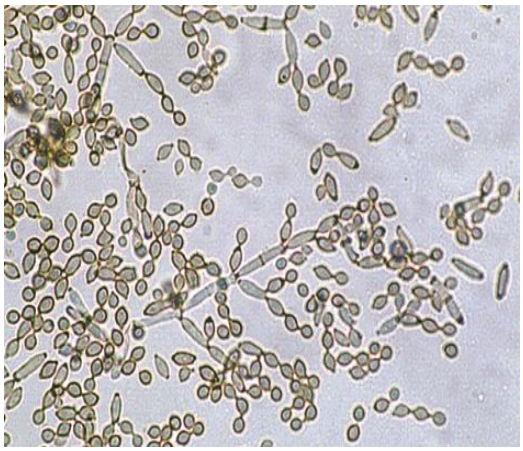




3(a)*Aspergillus fumigatus* culture in petridish **3(b) *Aspergillus fumigatus* conidiophores**

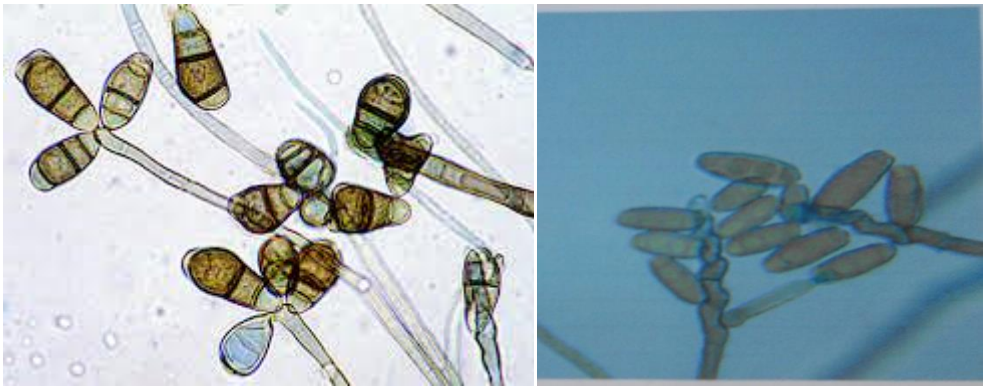


3(c) *Alternaria alternata* conidia

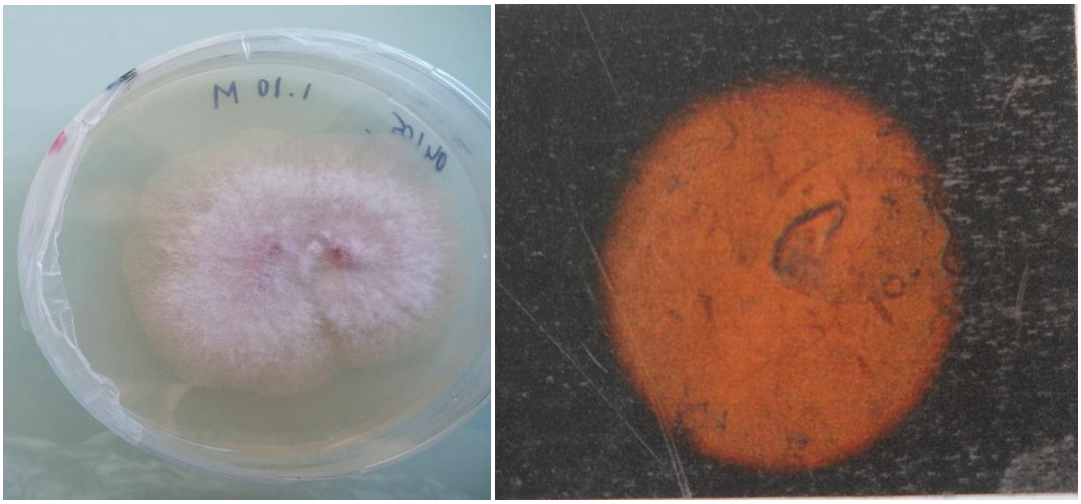


3(d) *Cladosporium* sp culture in petridish **3 (e) *Cladosporium* spores**

Photo Plate :4



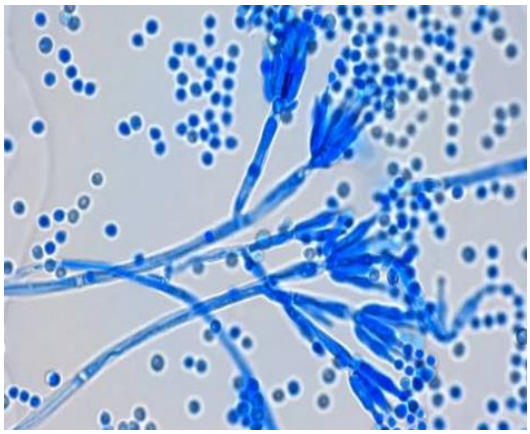
4(a) *Curvularia* sp conidiophore



4(b) *Fusarium solani* culture in petridish

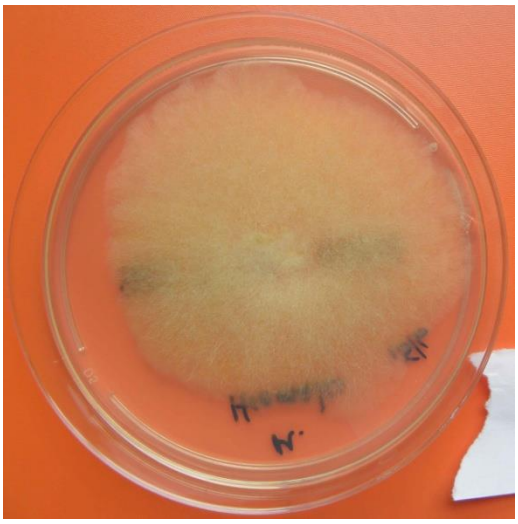
4(c) *Fusarium* sp spore

4(d) *Penicillum* sp culture in petridish

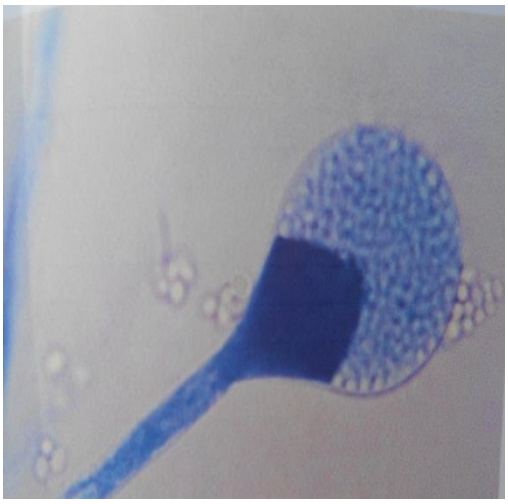


4(e) *Penicillum* sp conidiophore

Photo plate: 5



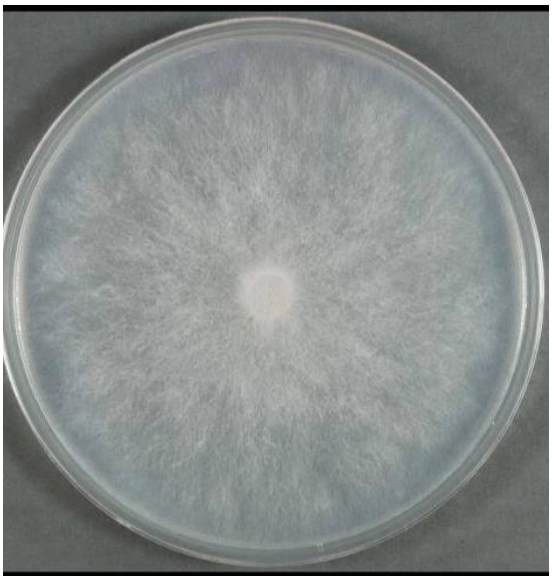
5(a) *Mucor* sp culture in petridish



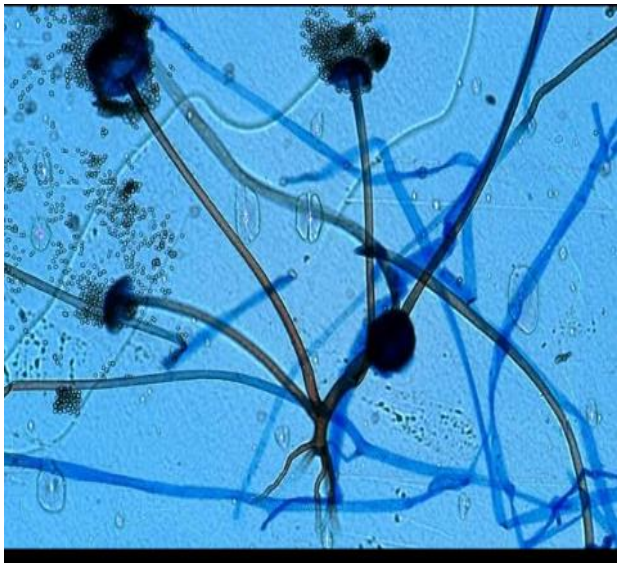
5(b) *Mucor* sp

5(c) *Colletotrichum* sp

Photo Plate: 6



6 (a) *Rhizopus* sp culture in petridish



6(b)*Rhizopus* sp conidiosphore

Photo Plate: 7

7(a) Cellular Rearing (*A proylei*) under nylon net

7(b) 1st instar (*A proylei*)

7(c) 2nd Instar (*A proylei*)

7(d) 3rd Instar (*A proylei*)

7(e) 4th Instar (*A proylei*)

7(f) 5th Instar (*A proylei*)

Photo Plate: 8




8(a) Silkworm rearing of *Antheraea proylei* **Jolly**.



8(b)*A.proylei* (yellow) **8(c) *A.proylei* (green)** **8(d)*A.proylei* (blue)**

Fig: Silkworm rearing of *Antheraea proylei* **Jolly.** and different coloured silkworm of *Antheraea proylei* **Jolly**.

Photo Plate: 9

Silkworm rearing of *Antheraea proylei* **Jolly.**(yellow)

Photo Plate : 10

10(a) Male moth **10(b) Female moth**

Moths of *Antheraea proylei* **Jolly**.

10(c) Eggs of *Antheraea proylei* **Jolly**.(silkworm eggs *i.e.*Disease free lyings)

10(d) Cocoons of *Antheraea proylei* **Jolly**.

Photo Plate : 11

11(a)Reeling machine

11(b)Spinning machine

11(c) Preparation of Ghicha on earthen pot

11(d) Loom(production for fabric)

Reeling and Spinning machine, Preparation of Ghicha and Loom.

Photo Plate: 12

12(a) Reeled yarn

12(b) Spun yarn

12(c) Ghicha yarn

12(d) Different types of yarns

Different types of Oak tasar Silk yarn.

Photo Plate: 13

13(a) Oak tasar Saree

13(b) Waist coat, bag, and coat

(Product diversification of Ghicha yarns)

Oak tasar fabrics.

DISCUSSION

The North Eastern region of India is one of the 35 biodiversity hot spots declared in the world and occupies an important position in sericulture map of India due to its unique faunal and floral wealth. Four varieties of silk viz. Mulberry, Eri, Muga and Oak tasar are produced in N.E. region. Sericulture and silk weaving is the part and parcel of cultural heritage of the people of N. E. India. The oak tasar (*Antheraea proylei* **Jolly.**) silkworm fed on leaves of Oak tree *Quercus* species, is an important source of tasar silk, a rough coarse and nubby silk usually with natural shade of beige. The Oak tasar track extends from Jammu and Kashmir in West to Manipur in the East, Himachal Pradesh, Uttrakhand, Assam, Arunachal Pradesh, Meghalaya, Mizoram and Nagaland. The production of oak tasar raw silk 3.0-5.0 metric ton (2007-08 to2014-15 Table:2) and it is necessary to production of oak tasar silk to earning foreign exchange as well as creating employment generation in rural sector. A Little scientific input to add to its knowledge would open a new avenue for its improvement which will be help for upliftment of rural society.

Research Stations of Central Silk Board in the country are engaged in study relating to overall improvements the quality of silkworm and its host plant to increase raw silk productions, the microorganisms associated with the phylloplane and rhizoplane of Oak tasar host plants is yet to be recognized fully, in its role to improve the nutrition of the silkworms. The Fungi play a very important role in nutrient cycling and plant health development (Thorn, 1997; Bridge and Spooner, 2001). Many workers have studied the nutritional status of the leaves of host plants but the rhizosphere mycoflora of the host plants have not been taken care. Now it is a recognized fact that the fungal spores and mycelia spread over the leaf surface may contribute critically on the nutrition of the silkworms and the mycoflora present in the root surface or rhizosphere indirectly may influence it by improving the nutritional status of the host plants in the soil. The region surrounding and including plant root is of crucial importance for plant health and nutrition (Marschner, 1995).

The present investigation dealing with the rhizoplane and phylloplane of *Quercus serrata* and their role in growth and development of Oak tasar silkworm at Umrangso, Dima Hasao district of Assam has many limitations and therefore, it is

not exhaustive, but, rather a prelude to opening new avenues of study relating to the mycoflora associated with rhizoplane and phylloplane of host plants of oak tasar silkworm and their role in critical nutrition of silkworm and thereby their growths, development of silkworm and raw silk production efficiency.

The physico-chemical characters of the soil of the investigation area (Table:17) shows that soil of the area is acidic in nature with high organic content in soil. Soil moisture stress has been found to enhance and decrease root exudation (Grayston *et al.,* 1996). Soil pH is one of the most important factors affecting soil fertility (Foth and Ellis, 1988) and the ideal soil for most plants is slightly acidic to neutral since in this state most of the compounds containing the plant nutrients have their most ideal solubility (Kellogg, 1998). Fungal population is most positively co-related with organic matter content in soil (Noverriza and Quinmio, 2004). The variable quantities of different nutrient level in soil during this investigation may probably be due to the use of manures, fertilizers and other cultural practices in the soil and indicates the fertility level of the soil.

The plant rhizosphere is dynamic environments in which many factors may be affect the structure and species composition of the microbial communities that colonize the roots. The microbial communities associated with the rhizosphere vary depending up on the plant species (Grayston *et al.,* 1998), soil type (Campbell *et al.,* 1997), and cultural practices (Lupwayi *et al.,* 1998). The notable findings from the present investigation of soil mycoflora of seedling (Table:12) and mature plants (Table:13,14) of *Quercus serrata* are that microbial population are higher in mature plants as compared to the seedling. Rhizosphere soil contained greater spectrum of fungal species than either rhizoplane or non rhizosphere soil. Root exudates they stimulate microbial activity selectively in rhizosphere and rhizoplane regions (Bansal and Mukkherji, 1996). Mali (1975) reported that increased fungal population in the rhizoplane of Coriander with increasing age. El-Amin and Saabadi (2007) reported that significant variation in total number of fungal colonies and percent abundance of fungal species in rhizosphere soil of Sugarcane, which is increased with plant age. They also reported that high occurrence of *Aspergillus, Rhizopus, Penicillum, Fusarium and Curvularia* from Sugarcane rhizosphere. The Rhizosphere contains lots of organic substrates which harbour a high count of microorganisms, especially fungi (Noveriza and Quimic, 2004). The soil organic

content and as well as its acidic nature probably influenced the isolated population from the rhizosphere.

Most commonly the isolated fungi in both seedlings and mature plants during the investigation were *Aspergillus niger* and *Aspergillus flavus*, *Fusarium* sp, *Alternaria* sp, *Cladosporium* sp, *Trichoderma harzianum.* Abdel Hafez, (1982), reported that *Aspergillus niger, Aspergillus flavus* and *Fusarium solani* among the most frequently isolated fungi from the rhizosphere of *Tritichum vulgare* .Noveriza and Quimio (2004) also reported that *Aspergillus flavus* and *Aspergillus niger* most frequently from Black Pepper rhizosphere. *Aspergillus niger* was also found to be the most dominant species during this investigation. Domsch *et al.,* (1980) reported that *Aspergillus niger* was found in soils with pH range of 4-8. This study also shows similar result. The abundance of *Aspergillus* sp, during this investigation may be due to their high sporulating ability and tolerance for different physico-chemical conditions of the soil and also their ability to utilize the available nutrients more readily over other species.

Some of species like *Aureobasidium pullulans* showed seasonal appearance in matured plant during this investigation. *Curvularia* sp was common throughout the study in all soil in matured plants. *Fusarium solani* showed distinctive absence in non rhizosphere soil. The differing physical, chemical, and biological properties of the root associated soil, compared with those of the root free bulk soil, which are responsible for changes in microbial diversity and for increased numbers and activity of microorganisms in the rhizo sphere micro environment (Kennedy, 1998. Gams and Domsch (1969), Hawkworth (1991), Persiani *et al.,* (1998) the study of seasonal variation in soil fungi pointed out that fungal population differ from season to season in a particular soil (Rane and Gandhe; 2006). Bisset and Parkinson (1979) also indicated that the major sources of variation in micro-fungal species composition are attributable to differences among sites, which largely are determined by vegetation.

Soil fungi show marked periodically throughout the year, (Warcup, 1957) the maximum number of micro-fungi being present during winter and the rainy seasons. Microbial community seasonal maxima in the wet winter months and seasonal in dry summer month have been reported from Oak canopy soil compared with open

grassland soil by Waldrop and Firestone (2006). Rane and Gandhe (2006) also reported that maximum diversity of during winter from Pal forest soil. In the present investigation also shows maximum fungal diversity during autumn season. The occurrence of different fungal species depends upon soil type, moisture content, mineral nutrients, and soil temperature (Vanvuurde and Schippers, 1980; Shukla *et al.,* 1989). Isolated fungal species were most common to both rhizosphere and non-rhizosphere expects a few species. A few researchers like Dwivedi (1966), Dkhar and Mishra (1987) they discussed seasonal variation of fungal population in some soil types and concluded that changes in soil, organic contents, water holding capacity, temperature and pH of the respective season are the probable factors associated with fungal population. According to Pandey and Palni (2007) they were observed that a concomitant decrease in the pH of the rhizosphere soil where ever an inhibitory effect on the rhizosphere microbes was observed. Shukla and Tripathi (2007) during their study in the distribution of micro-fungal communities in forest soil observed that the density of fungal propagules had a close inverse relationship with the pH of the soil which was the one of the most important point.

The fungal isolates during the present investigations most probably, have been influenced by the soil characters, like moisture content, pH of the soil, age of the plants, nature of exudates from the roots, and the status of nutrient availability, cultural practices in plantation, location of study area as well as environmental condition governing the seasons.

The biochemical constituents of the leaves of *Quercus serrata* from the study area show that, irrespective of seasons moisture content of leaves are more in spring season than autumn season in three different treatments-mature (Table:18-24.). Bhuyan (2002) observed that from different types of host plant of Muga, and (Pathak, 1988) in Eri food plants. Moisture content of the leaves in *Persea bombycina* has a direct bearing on the health of silkworm, and high moisture content in the leaves favourable effects on the palatability and availability of nutrients and serves as a criterion in estimating leaf quality (Parpiev, 1968).

Total carbohydrates content is high on mature leaves of *Quercus serrata* than semi-mature and tender leaves reported by (Sinha *et al.,* 1986, Pandey and Goel; 1991, Pandey; 1995, Ponnuvel *et al.,* 1996). Crude protein content was more in

tender leaves of *Quercus serrata* than semi-mature and mature leaves which was found during the investigation. Higher leaf protein was found to be significant for better silk production (Verma and Kushwaha, 1970), Bhuyan, (2002), also observed that tender leaves contained more protein than mature leaves. Of som. It was also found that in present study the leaf moisture and crude protein more in Spring season which declining trend in the leaves of *Quercus serrata* in Autumn season(Table:18,19 and 20). It was also found that less carbohydrates in Spring season which increasing in Autumn season. Ponnuvel *et al.*,(1996) rerorted leaf moisture 69.49% and 67.03%,crude protein 10.17 %and 9.88%, carbohydrate 10.84% and 10.81% during March and April respectively.The Effective Rate of Rearing % was found higher in Spring season than Autumn season(Table:9)which was similar reported by Ponnuvel *et al.*,(1996) ERR 72.4%and 60.4% during March and April(Spring season) respectively. Pandey(1995) reported ERR 66.60% ,Raja Ram *et al.*,(1998)also reported ERR % of *A.proylei* **Jolly.**on *Q.semicarpifolia* of seven different morphotype range 54-81.4% in Spring season.

Eleven fungal species were isolated from the leaf surface of *Quercus serrata* (Table:10,11 and photo plate no. 2-5) the types of fungi which colonized the leaves at different stages of maturation, viz. Tender, semi-mature and mature leaves are more or less same. The species of *Aspergillus*, particularly *Aspergillus niger* was found to be the most dominant in the three stages of leaf growth during all the season. *Aspergillus fumigatus* was found co-dominant in lower surface of mature leaves in spring season and both leaf surface of tender and semi-mature leaves in autumn season. It was also found that *Alternaria alternata* co-dominant in both leaf surface of tender and semi-mature leaves and upper surface of mature leaves in spring season. Baruah *et al.*,(1998), Bhuyan (2002) reported dominance of *Aspergillus fumigatus* from three different host plants of Muga, viz. Som (*Machillus bombycina*) Soalu(*Litseae polyantha*) and Meejankari (*Litseae citrata*).They also reported that *Alternaria* sp , *Aspergillus fumigatus, Aspergilus flavus ,Aspergillus candidus, Curvularia* sp. *Mucor* sp,*Phoma* sp from the leaf surface of som plant. Gupta and Khube (1991) isolated from the oak leaf litter different fungal species were *Aspergillus flavus, Penicillum* sp, *Fusarium solani, Mucor hiemalis, Phoma humicola* etc.

The dominance of *Aspergillus* sp in all the seasons may be due to richness of *Aspergillus* species over *Quercus serrata* (Oak) plantation field and their ability to colonize the Phylloplane of Oak plants. *Aspergillus* sp dominance was also reported by Bhuyan (2002), Singh and Baruah (1979) and Mishra and Shukla (1989).

The fungal increased colonization of old leaves is due to super-infection of the leaves over time, air-borne inoculums (Suryanarayan and Thennarasan, 2004). Sharma (2004) has also reported that occurrence of some fungi in all stages of growth, while others with restricted representation in Sugarcane leaves.

According to De Jager *et al.,* (2001) reported that gradual increase of filamentous fungal and yeast densities from follicle stage, through flush and juvenile to mature leaf stage in Mango Phylloplane and the most common fungal genera isolated were *Cladosporium cladosporioides* and *Alternaria alternata*. They also reported that isolation of *Candia, Aspergillus, Colletotrichum, Curvularia, Drechslera, Nigrospora, Mucor, Penicillum* and *Rhizopus* along with many other filamentous fungi from Mango phylloplane. More fungal species were found in lower surface of leaves than upper surface of leaves of Oak leaves, and it's also observed more fungal species in mature leaves lower surface in autumn season than spring season. It may be due to the leaf surface anatomy, availability of nutrients, compatibility between the microbes present on the leaf surface and environmental factors like temperature, relative humidity around the phyllosphere, atmospheric precipitation, intensity of UV (ultra violet) radiation etc. Bhuyan (2002) also reported more fungal flora on the lower surface of Som leaves. The persistence of *Cladosporium* and *A.alternata* is ascribed to their excellent adaption to the Phylloplane (Dickinson, 1976). And Levetin and Dorsey (2006) also reported greater number of fungal colonies from the abaxial surface of *Quercus* and *Ulmus* leaves. Suryanaryanan and Thenarasan, (2004) have reported *Colletotrichum* sp and *Phyllostica* sp from *Plumeria rubra* leaves during the wet period. Das *et al.,* (2005) have reported *Colletotrichum gloeosporioides* as the casual organism of Leaf blight disease of *Persea bombycina* plants with peak in June-July and *Phyllosticta perseae* as causal organism of leaf spot disease of the same plant occurring from May to

August. Isolation of the two genera in healthy leaves of *Persea bombycina* may be due to abundance of their spore during rainy season or rain splash of the spores from the diseased leaves. *Phyllosticta perseae* was found during Spring to Rainy season and *Collototrichum gloeosporioides* was found from Spring to Autumn.

Qualitative and quantitative diversity of fungal species may be related mainly due to environmental factors, as well as cultural operation practiced in the plantation to suit Oak tasar silkworm rearing. During the Spring season effective rate rearing is higher than Autumn season may be due to presence of more *Aspergillus* sp abundance percent in Spring along with more leaf moisture and crude protein. Diem (1974) reported that presence of water on leaves was found necessary for the development of myco-flora, which is one of the most important factor. The reason for low diversity of fungi on the leaf surface during the rainy season, which is probably due to high precipitation in the area, due to which the fungal spores are washed out to the ground. In addition to nutrient level, growth and abundance of phylloplane fungi are also influenced by environmental conditions such as water availability, UV radition, and temperature (Breeze and Dix, 1981; Sundin 2002; Zak, 2002).

The occurrence of different fungal species during this investigation therefore may be attributed mainly to the local environment factors, leaf surface morphology, nutrient exudates of leaves, cultural practices like pruning to suit Oak tasar silkworm rearing in the farm, and more relative abundance of the reported fungi in the air over the plantation field etc. Eleven fungal species were isolated from air over the study area at Umrangso REC farm (Table: 15,16 and photo plate no: 2-6) *Aspergillus niger* was the most dominant species throughout the investigation. *Aspergillus* sp, was found to be the most predominant in air at Raipur (Tiwari and Sahu, 1988 and Sahu, 1998). Highest concentration of *Aspergillus* sp. was also observed from different plants of Aurangabad (Tilak and Sriivasulu, 1967), Raipur (Tiwari and Jadav, 2004). Basumatary *et al.,* (2002) reported isolation of *Alternaria* sp, *Aspergillus* sp, *Cladosporium* sp, *Fusarium* sp, *Helminthosporium* sp, *Mucor* sp, *Nigrospora* sp, *Penicillum* sp, *Rhizopus* sp, and *Torula* sp, from atmosphere of different environments from Goalpara District, Assam. Kamal and Singh (1974) reported *A.niger* as one of the *Aspergillus* sp , constantly occurring during all seasons of the year from sugarcane field at Gorakhpur (U.P.). Singh, (1981) reported

prevalence of spores of *Aspergillus* sp, *Cladosporium* sp, *Alternaria* sp, and *Curvularia* sp. as most frequently isolated from Shillong atmosphere. In this investigation also *Aspergillus* sp. *Cladosporium* sp and *Alternaria* sp. has been isolated in both Spring and Autumn season. The dominance of *Aspergillus* sp in the both season during Spring and Autumn season in this investigation may be due to richness of *Aspergillus* sp. in the air over *Quercus serrata* plantation of Umrangso REC Farm. Last (1956 b), in his study of air spora within and above mildew infected cereal crops found higher population near ground. Similar observation is made in this study. The richness of isolated fungal species in the bottom layer may be due to nearness to the soil and lifting of dry soil particles from soil during different periods. Uddin (2005) from West Bengal, Pund and Tidke (2005) from Amravati reported that total fungal species increased in Autumn and Winter season. The atmosphere contains tremendous diversity of airborne spores with high concentration frequently occurring from spring through fall in temperature areas of the world (Gregory, 1973; Levetin, 1995)

According to Huang *et al.,*(2002) the airborne fungi were higher in winter than other seasons in the municipal landfill sites of Taiwan and ascribed it to the geographic characteristics of the sampling area. Fang *et al.,* (2005) reported that a high frequency of airborne fungi in regions with high vegetation coverage in summer in Beijing and also mentioned that, most of the airborne fungal spores came from vegetation rather than soil. Hariri *et al.,* (1978) reported that the most prevalent airborne fungi in Ahvaz-Iran were *Penicillum, Alternaria* and *Aspergillus.* Abdel-Hafez (1984) showed that the maximum airborne fungi were in Winter and minimum in the Summer. A few more fungal diversity in air, during Autumn than Spring season in this investigation can also be also be attributed to various meteorological factors particularly moisture content of the atmosphere. Minimum fungal diversity during Rainy season may be due to high precipitation recorded by several workers.

Meteorological data recorded in the area during rearing period s of Umrangso. The rearing performance of Oak tasar silkworm reared during Spring (March-April) and Autumn (September-October) season. The effective rate of rearing (ERR%)and SR% was more in Spring season than Autumn season. It was observed that ERR% was very much influence of the environmental conditions

prevailing over the season and optimum conditions prevail over the seasons. The analyzed data on the economic traits of Oak tasar silkworm rearing performance reveal that shell weight of the cocoons during Spring season was significantly higher than Autumn crops and also silkworm weight. This clearly indicates that these characters were influence by the nutritional status of leaves rather than the environmental factors alone.

Rana *et al*.,(1987) studied food consumption, utilization and rate of growth of *Antheraea proylei* feeding with *Quercus serrata* leaf. The quality of food consumption increased with increase in age of the worm and reached its peak in the fifth instar. The assimilation and tissue growth were positively correlated to the amount of food consumed. The quality of leaves had got direct influence on the health, growth and survival of Oak tasar silkworm (Sinha *et al.,* 1986). Studies on foliar constituents of Oak tasar silkworm host plants were reported (Sinha and Jolly, 1971; Pandey 1993, Banerjee *et al.,* 1993; Ghose and Srivastava, 1996; Ghose *et al.,* 1995; Ponneuvel *et al.,* 1996). The difference in nutritional quality of host leaves was an important factor for the success and partly success. For instance, the predominant occurance of lepidopterous pests and feeding Chinese tasar silkworm, *Antheraea pernyi* on oak during the Spring season had been attributed to the presence of lesser amount of tannins,but increase of tannin in the following seasons the leaves become unsuitable for silk. In Spring season higher crude protein% and lesser crude fibre % resulting higher ERR% reported the influence of crude protein and amino acids on cocoon characters of Oak tasar silkworm *Antheraea proylei* **J.** .The quality of leaves had got direct influence on the healthy growth and survival of silkworm(Sinha *et al.*,) Better the quality of leaves, greater possibilities of obtaining good cocoon harvest. Therefore, the selection of the food plants possessing superior nutritive value could be utilized for the healthy development of silk for obtaining good cocoon crop. According to findings of Pandey and Goel(1991) crude protein contents of young leaves were higher than old leaves in three Oak species where *Quercus serrata* showed maximum *28.92%Q.semecarpifolia 20.77%,*and *Q.incana* 16.47%. but in old leaves contained nearly half of total protein contents of young leaves. The young leaves contained less crude fibres, old leaves had nearly double the fibre content.Pandey(1995) observed seasonal changes in the leaf composition of *Q.serrata,* where leaf proteins were 6.81% in March and 7.89% in April which were

decreasing 4.74% in October,and ash 2.23% which was increased from March 1.95%, as result, the leaf quality of March and April month was found most suitable for rearing of *A.proylei* **J**..A strong positive correlation was found between leaf content and larval body weight. The higher survival of Oak tasar silk worm during Spring season may be due to higher protein content of the leaves during April. Leaf quality for many lepidopteron larve is determined on protein content basis(Mattson *et al.,* (1980).The Autumn crop of Oak tasar not fully success may be due to decline in protein content.Ponnuvel *et al.,*(1996) leaf moisture percentage of *Quercus serrata* leaves trends decreasing from February to November (71.9% to56.78)hey found that leaves containing higher amount of crude protein and amino acid resulted producing higher amount single cocoon and shell weight, cocoon yield per hundred larvae filament length and fibroin content of the cocoon shell. In this investigation leaf moisture and crude protein observed more in the Spring season than Autumn season and *Aspergillus niger* were also found more on the leaf surface in spring season than autumn season which may play an important role better (ERR%) cocoon weight, shell weight and more filament length.The reeling parameter of *A.proylei* **J.** cocoons under three treatments (Table:25) in present investigation result similar with other investigators.Devi *et al.,*(2012) reported Filament length 699.6 meters, Denier 6.6(D) ,NBFL 333.0,Recovery 74.4% and Reelibility 47.6%. .Tikko and Goel(1987) reported cocoon cooking with 1% sodium carbonate solution gives good results in respect of reelabity 60.28% and 61.83% with Biopril-50.

In this investigation shows that the fungal population of soil, leaf and air are highly influence by the environmental factors like light, temperature, humidity and rain fall. Low humidity and high precipitation adversely affect the air, leaf surface and soil mycoflora and low precipitation favours the population of saprophytic microorganisms. According to Gregory and Hirst (1957); Sharma *et al.,* (1984) and Thomspson *et al.,* (1993), the season is a dominating factor determining the qualitative and quantitative composition of the leaf surface mycoflora, and further variation in the total number of isolates indicates changes in density of the active population. These results are similar to the result obtained in the present investigation were done during 2013 and 2014 at Umrangso on *Quercus serrata* plantation.

It is also clear evident from the present result that phylloplane mycoflora isolates were also present in the soil and in air and vice-versa. Therefore it seems a logical probability that there was a continuous process of distribution of the mycoflora of soil, leaf surface and air and vice-versa. The situation can be explained as follows in a cyclic pattern.

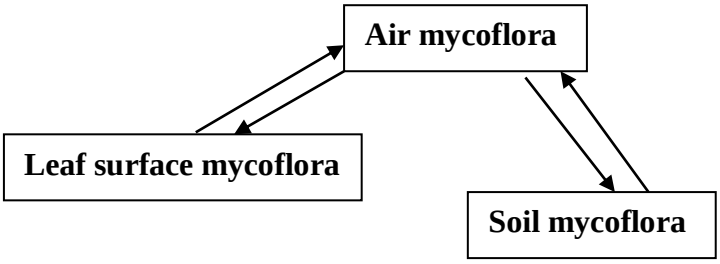


Fig 12: Cyclic pattern of mycoflora leaf to air and air to soil and vice versa.

The seasonal fluctuation in the population of leaf, soil and air mycoflora seems to depend upon the climatic factors *i.e.* temperature, humidity, wind velocity, phenology of the local vegetation and its associated fungi and precipitation. Further, the qualitative and quantitative variation of the total population of mycoflora in the soil may be due to inadequate soil moisture and nutrients, which affect the population (Rama Rao, 1970) and activities of the microorganisms, Waid (1960) suggested that fertility of soil and the fungal population of mycoflora, which seems to be a logical conclusion.

The present investigation is relating to the fungal population of rhizosphere and phylloplane of Oak tasar host plant (*Quercus serrata*) and their effect on the growth silkworm, development of cocoons and raw silk production may open new avenues of wider studies as no attempt had been made earlier to include both the rhizoplane and phylloplane fungal population relationship including that of air spora relating to Oak tasar host plant. The impact of fungal spores may probably due to the fact that the fungal populations of the soil provide essential nutrients for the plant growth and development. And these nutrients are translocated to the leaf and impacts upon the foliage quality by increasing or decreasing its moisture retention capacity, protein content, sugar and various other essential trace elements. The silkworm obtains its nutrition for the growth and development by feeding on leaves. It may be possible to exploit the situation once we understand in depth, the

microbial correlation ship of the soil, phylloplane as well as of the air and the seasons and how best the beneficial and nutritive fungal spores for silkworm growth can be trapped and increased. Potential of *Aspergillus niger* and *Trichoderma viride* as bicontrol agents of wood decay fungi(Tiwari,2011).*Trichoderma* species are using for biocontrol` of plant pathogens in Vietnam(Tran, N.Ha 2010) will be to boost oak tasar culture in country India.

In the present investigation indicates that the fungi population particularly those present on the leaf surface have a direct bearing on the rearing of silkworm and therefore, further study relating to the association of not only fungi but also bacteria and other microorganisms with the leaves and also rhizosphere and their impact on the health of the silkworms and their development which help the raw silk production is essentially required. Although the present study involves only one host plant *i.e. Quercus serrata,* there are other varieties of hosts and the microbial association of these varieties may be different. Hence, the development of specific types of rhizosphere and phylloplane fungi depends up on many environmental factors as well as on the physiology of the plants and their interaction of the micro-organisms themselves in their spheres of occurrence. If scientifically exploited, such microbial relationships would be very beneficial to the silk industry. The present investigation is only an attempted to draw attention to the new but prospective areas of work relating to Oak tasar silkworm and silk production. The health of the plants, as well as nutritional status of the foliage produced by the plants and also the nutritional requirements of Oak tasar silkworms essentially needs exhaustive study. Further identification of season specific of food plants and silkworm breeds their improvement through breeding is necessary which may help to cope with the various seasonal problems associated with Oak tasar silkworm rearing.

The microbiology of all the host plants is still obscure, although their biology is known. Hence a thorough understanding of the microbiology of the different plants parts like leaves, flower, stem and roots of these plants needs to be studied in future. The present study suggests that there is certain role of fungal spores on the growth and development of Oak tasar silkworm, its cocoon and raw silk production, the microbiological aspects which is related to the rearing of Oak tasar silkworm must be looked from wider perspectives and it is also a very important that to evolving high yielding varieties host plants and using green manure and bio-

fertilizer for soil health and rearing of high yielding varieties of Oak tasar silkworm and production more Oak tasar raw silk and earning more foreign exchange which will be boost up the rural economic upliftment and generating more employment which may help utilization of naturally grown oak flora and can be prevent the jhum cultivation in the hilly region which is less remunerative as compared to Oak tasar culture. Oak plants regulate water cycle; conserve soil moisture and environmental/ ecological stability of the fragile mountain ecosystem. It is a time to prepare an appropriate project to popularize and develop moment of Oak tasar culture in India by the government departments like sericulture, forest and non government organization. It will encouraging the conservation of Oak flora and generated more employment in the hilly region of sub Himalayans belt.

SUMMARY AND CONCLUSION

Sericulture is an agro base, industry; India has the distinction of cultivating all the five commercially known verities of silk, namely Mulberry, Eri, Muga and Tasar (Tropical and Temperate).The temperate tasar silk is produced by silkworm *Antheraea proylei* **Jolly.** In India in Jammu and Kashmir, Himachal Pradesh, Uttarkhand in Northwestern and Assam, Arunachal Pradesh, Manipur, Mizoram and Nagaland in North eastern India in sub-himalayans belt.

China is the homeland of sericulture, the technique of silk production was first introduced in China by Hoshomin the Queen of China. In India silk was treated as pure material used for sacred and ritual purpose. The temperate tasar silkworm feds on *Quercus serrata .Q.accustisima, Q.graffthii, Lithocarpus dealbata* in Northeastern Himalayas and *Quercus serrata* is used for Oak tasar silkworm rearing and is considered to be the primary host plant of *Antheraea proylei* **J.** in Northeastern India. Its leaves provide a unique environment to their surfaces occupants and the typical leaves exudates influence the growth and development of the varieties of micro-organisms. These micro-floras play an important role in supplying different types of nutrients to the plants as well as the silkworms.

It is an evident numbers of fungi do not exist in nature individually but the microorganisms are present in the rhizosphere and phyllosphere and in other habitats in the host or in close proximity of the host and root exudates of the host stimulate the growth and sporulation of the different types of microorganisms which on the other hand influence in developments of the phylloplane of the host by providing type of nutrition. The leaves constituent the major part of exposed plant surface and which are open to infection or spprotrophic colonization by air dispersed or splash dispersed microflora. These micro-organisms of the leaf surface play a very important role in supplying micronutrients to the plant and in protecting the leaf surface from the pathogenic infections.

In the present investigation the following observation were deals with the isolation of fungi on phylloplane, rhizosphere, non-rhizosphere and rhizoplane.

Sixteen fungal species in spring season and eighteen in autumn season were isolated from the soil of mature *Quercus serrata* plant. Ten fungal species were

isolated from seedling soil. *Aspergillus* species was most dominant and *Fusarium* species co-dominant in both seedling and mature plant soils. *Aspergillus niger* was found most dominant species among *Aspergillus. Alternaria alternata, Fusarium* sp *Penicillum* sp and *Trichoderma* sp were also found abundant in seedlings soil in spring season, but in autumn season *Alternaria alternata, Aspergillus flavus, Aspergillus niger, Cladosporium* sp, *Colletotrichum* sp, *Penicillum* sp, and *Trichoderma* sp.

During spring season in rhizosphere soil *Alternaria alternata, Aspergillus flavus, Aspergillus niger, Fusarium solani. Fusarium oxysporium, Penicillum* sp and *Trichoderm* sp were isolated and non-rhizosphere soil *A.alternata, Aspergillus. flavus ,A.niger, Fusarium oxysporium, Penicillum* sp *,Trichoderma* sp were isolated. *A.niger was* dominant *Fusarium* spp co-dominant. In non-rhizosphere soil *A.alternata. A.flavus, A.niger, Cladosporium, Colletotrichum, F. oxysporium, Penicillum, Trichoderma. Aspergillus* spp dominant, and co-dominant. *A. alternata, Penicillum, Trichoderma* sp followed by *Cladosporium, F. oxysporium* and *Colletotrichum. But Fusarium solani, Mucor* sp were not found.

The rhizosphere soil harbors maximum diversity of fungi comparison to rhizoplane and non-rhizophere. The diversity of fungi also increased with increase in age of the plant. Higher fungal diversity was observed during autumn season September-October and lower during the spring season March-April. Soil moisture content, pH of soil, age of the plants, nature of exudates from the roots, status of nutrient availability, cultural practices in the plantation, location of study area as well as environmental conditions governing the seasons may have influenced the occurrence of the fungal species. Soil of the area is acidic in nature and observed more fungi species in autumn season it may be due to more rainfall in autumn season. The soil characteristics and the nutritional status of the soil, presumably influence the microbial composition of the soil during this investigation.

Eleven fungal species were isolated from the leave surface *Aspergillus* spp dominated in the both season *i.e.*. Spring and Autumn. *Alternaria alternata,* co-dominant. *Aspergillus niger, A.flavus, A.fumigatus, Alternaria alternata, Mucor* sp, *Curvularia* sp and *Fusarium* sp in the Spring season, but in Autumn season in addition to *Penicillum* sp, *Verticillium* sp.

Colletotrichum sp, *Cladosporium* sp. The presence of different fungal species on the surface in this investigation, probably, were influenced by factors such as leaf surface morphology, nutrient exudates of the leaves. local environmental factor viz. temperature, humidity and precipitation, moisture and nutrient content of the leaves, competition between the different microbes, cultural practices like pruning to suit Oak tasar silkworm rearing during spring and autumn season, abundance of spore load in the air over the plantation field, intensity of ultraviolet radiation etc. More fungal species were observed on the lower surface than upper surface of leaves.

The seasonal variation of chemical constitution of *Quercus serrata* leaves showed higher leaf moisture, crude protein and ERR% in Spring season while in Autumn season less leaf moisture, crude protein and ERR% but higher carbohydrates % in Autumn season were recorded. Higher carbohydrates in mature leaves in Autumn season may be impact on higher fungal population on *Q.serrata* mature leaves.

Eleven fungal species was isolated from air over *Q.serrata* plantation during the investigation. *Aspergillus* species were dominant among them *Aspergillus .niger* was the most dominant in the both season *i.e.* Spring and Autumn, *Alternaria alternata, Aspergillus .flavus, A. fumigatus, A.niger, Cladosporium* sp, *Colletotrichum* sp, *Curvularia* sp, *Fusarium* sp, *Mucor* sp, *Penicillum* sp and Sterile mycelia were observed.

Average leaf yield of per plant and per hectare in Spring season was recorded more than Autumn season (Table:3). By application of Farm Yard Manure and Chemical fertilizer Nitrogen, Phosphate and Potash gave better leaf yield and ERR%. It is also observed that higher ERR% in Spring season than Autumn season. Crude protein 10.28%, leaf per plant1.53kg, ERR64.5%. *Aspergillus niger* 70.50-72.50%, 61.5-60.5% and54.0-55.5% respectively found on upper surface of tender, semi-mature and mature leaf. Like that 66.5-66.5%, 57.5-58.0% and54.0-55.5% (Table: 10, 11). *Aspergillus niger* were observed on lower surface of *Quercus serrata* leaf and found that dominant in spring season which may help the growth of oak tasar silkworm. The reeling parameter of *Antheraea proylei* cocoons showed better performance cocoons were harvested from where rearing was conducted on plants where recommended FYM and NPK applied to got quality and quantity leaves. Supply of chawki worm to farmer also encouraging which may required motivating in new area of Oak tasar culture.

The fungal population were isolated from leaf, soil and air in the present investigation, indicates that mycoflora present in a cyclic pattern of appearance in air, phylloplane and soil. The mycoflora population density observed highest in rhizosphere soil and lowest in air of *Quercus serrata* plantation.

The present investigation deal with fungal population of rhizosphere and phylloplane of (*Quercus serrata)* host plant of temperate tasar silk worm and their effect on the growth and development of *Antheraea proylei* **Jolly.** at Umrangso, District Dima Hasao, Assam. By adaptation of Oak tasar culture in the hilly region may be prevent the Jhum cultivation, in forest area by deforestation of Oak trees has led to the erosion of top soils, landslides and ecological imbalance. Lot of plantation of *Quercus* species for proction of fragile, Himalayan ecosystem. Now it is very most essential to popularized oak tasar culture for the economic upliftment of hilly people, because jhum cultivation being less remunerative as compared to Oak tasar culture. At present it has been realized that replacement of Oak forest with pine in the Himalayas affects the nitrogen cycle (Singh *et al.*,1984) and caused heavy landslides to heavy toll and damage. Pine trees have less soil holding capacity and no coppicing capacity. Oak trees hold soil more strongly than pine and having more coppicing power which is required to publicized in order to encourage plantation of more and more oak trees by Non-Government Organizations, Central Silk Board and Forest departments. An appropriate project should be required to prepared to popularize and development of oak tasar culture. Attempts being made by Appropriate Technology of India in Uttarkhand needs to be appreciated and it should be copied by other government and non-government organization. Regional Tasar Research Station, Imphal, Manipur. Evolved of superior breeds of Oak Tasar Silkworm is one of the most important areas of breeding of research for the development of Oak Tasar Industry in India. A new breed having Blue colour in the larvae is isolated from the segregating progenies of the backcross involving the parents *Antheraea roylei* and *Antheraea proylei* has shown improvement in most of the yield contributing characters over that of *A.proylei* (Singh, *et al.*, 2008). The superior breed cocoons yield range 44 to 92 cocoons per dfl ,ERR ranges 48.85% to 80.84%, cocoon weight ranges 6.43gm to 7.03gm: cocoon shell weight ranges 0.65 to 0.75 gm and filament length ranges 657 meters to 755 meters . The superior breeds may be exploited for commercial production of Oak tasar raw silk. Singh *et*

al., (2010) suggested to supply chawki-worms to popularized of oak tasar culture. One hectare of *Quercus serrata* (oak) plantation can support a rearing of 1.0 Kg dfl (disease free laying) of this worm producing 25000-30,000 cocoons thus earning a revenue of Rs.30,000-40,000 within 50days in the Spring crop. The reelers and weavers can engaged to reeling Oak tasar cocoons and diversity product fabrics to get additional income for their family.

FINAL CONCLUSION

The leaf yield of *Quercus serrata* showed highly significant differences among in the Spring and Autumn season and leaf yield was found to be much higher in Spring season than in Autumn season. The highest average leaf yield was recorded in plants where applied with input Farm Yard Manure (FYM) and NPK Chemical fertilizer(Urea, Single Super Phosphate and Murate of Potash).

The Effective Rate of Rearing%(ERR%) showed highly significant difference amongst in both Spring and Autumn crop rearing and ERR% was found much higher in Spring crop rearing than in Autumn crop rearing.

Eleven fungal species were isolated from the leaf surface of *Quercus serrata* .the types of fungi which colonized on the leaves at different stages of maturation viz. Tender. Semi-mature and mature leaves. In Spring season observed less nos of fungal species than Autumn season. *Aspergillus* species was dominant in the both season but *Penicillum* sp was found in Autumn season.

Ten fungal species were isolated from the soil (Rhizosphere and Nonrhizosphere) of *Quercus serrata* seedlings during Spring and Autumn season. Sixteen fungal species were isolated from the soil(Rhizosphere, Non-rhizosphere and Rhizoplane) of *Quercus serrata* plantation in Spring season in RS soil *Gliocladium* spp absent, *Fusarium solani* absent in NRS soil *Cladosporium herbarum, Gliocladium* spp were not found in RP soil.

Eighteen fungal species were found in soil(RS,NRS,RP) in Autumn season *Cladosporium clodosporides,* was not found in NRS, some *Aspergillus* spp and *Cladosporium herbarum* were not found in RP.

Eleven fungal species isolated from air over *Quercus serrata* plantation during Spring and Autumn season at 0.75m and 1.50m height. *Aspergillus* spp, *,Alternaria alternata, Cladosporium* sp, *Colletotrichum* sp, *Curvularia* sp, *Mucor* sp, *Penicillum* sp, Sterile mycelia were found in both season(Spring and Autumn) and addition *Fusarium* sp found in Autumn season. *Penicillum* sp and *Fusarium* sp were not found in Spring season but found in Autumn season at 1.50 metres height.

The foliar constitution of *Quercus serrata* in Spring season moisture %,crude protein, was found more and Autumn season carbohydrates % found more than Spring season.

In fertilized soil Organic Carbon and Nitrogen (ppm) was found more than unfertilized soil, but Phosphate(ppm) was a little more and Potash(ppm) was found double in unfertilized soil than fertilized soil at REC. farm of Umrangso. Hence, application of FYM and NPK fertilizer are required for production quality and quantitative of leaf in hectare better production oak tasar cocoons and yarns.

The reeling yarn of Oak tasar, Filament Length and Non-Breakable Filament Length was highly significance but Denair, Silk Recovery%, and Reelability% was not significant.

From the present study will help to production of Oak tasar silk production in Spring season *Aspergillus niger* was dominant on phylloplane of *Quercus serrata* and leaf moisture % , crude protein % was found more than Autumn season. As result the Effective Rate of Rearing of *Antheraea proylei* **Jolly.** was also found better ,which will be help to encouraging the Oak tasar industry in India.

By adaptation of Oak tasar culture in the hilly region may be prevent the jhum cultivation , because jhum cultivation being less remunerative as compared to Oak tasar culture. Systemic plantation of *Quercus* species for protection of fragile, Himalayan ecosystem and evolved more superior varieties of Oak tasar silkworm for rearing with technology for silkworm rearing and produce lot cocoons. Modernization of post-cocoon sector reeling, spinning weaving, dyeing etc and created market for domestic and international buyers.

Shed Oak dry leaves can be used for mulching of agriculture field, preparation of leaves compost and vermin-composting, wood for construction house, make wooden charcoal for cooking, branches can used for mushroom cultivation, The barks and leaves are used in the preparation of user friendly dyes. Oak tasar silk pupae are as food for poultry, fish and pigs, the oil extracted from the pupae can be utilized in the preparation of cream, soap, shampoo etc.

An appropriate project should be required to be prepared to popularize and development of Oak tasar culture by assistant of Government Organization through Nongovernment Organization, for employment generation and it will be help for the upliftment of rural hilly people.

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List of papers published

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2.Gogoi,A.K.Baruah,P.K. and Ray,M.K.(2017)Studies of the Phylloplane mycoflora of Oak Tasar plant (*Quercus serrata* **Thunb**.) And their impact on rearing performance of Oak Tasar Silk *Antheraea proylei* **Jolly** wjpls,3(6): 204-207

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1. Studies on the Phylloplane Mycoflora of Oak Tasar food plant (*Quercus serrata* L) and their impact on rearing performance of Oak tasar silkworm (*Antheraea proylei* Jolly) A.K Gogoi, P.K Baruah. (International conference on Harnessing Natural Resources for Sustainable Development: Global Trends, 29th -31st January,2014 Cotton College, Guwahati, Assam, India.
2. Isolation of different morphotypes of *Quercus serrata* and their effect on the rearing of *Antheraea proylei* Jolly. A.K Gogoi, G.P Singh and P.K Baruah, National Seminer on Recent Trends of Research in Science and Technology(59th Annual Technical Session of Assam Science Society) 29th March,2014. Cotton College, Cotton College State University, Guwahati, Assam

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Seasonal Variation in Foliar Constituents of Oak Leaf (*Quercus serrata* Thunb.) and its Impact on Oak Tasar Silkworm (*Antheraea proyeli* Jolly.) Rearing

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ABSTRACT

Quercus serrata Thunb. is the primary food plant of oak tasar silkworm Antheraea proyeli Jolly. The chemical constituents of leaf i.e. crude protein, crude fibre, crude fat, carbohydrates were estimated in different seasons i.e. spring and autumn. Bioassay were also conducted in spring and autumn season. In spring leaf moisture, crude protein, crude firbe, crude fat, ash, carbohydrates and ERR% were recorded 68.98%, 10.28%, 6.78%, 2.34%,1.88%, 10.80% and 64.50% respectly. During autumn season the leaf moisture 57.23%, crude protein 5.13%, crude fibre 7.09%, crude fat 1.94%, ash1.90%, carbohydrates 21.33% and ERR 31.60% were recorded from Oak leaves. In spring season higher leaf moisture, crude protein and ERR% were observed while in autumn season less % leaf moisture and ERR% but higher % of carbohydrates were recorded. There were no significant different observed for crude fibre, crude fat and ash% content in both the season.

Key words: *Chemical constituents, Quercus serrata, Antheraea proylei.*

INTRODUCTION

Antheraea proylei Jolly. is mainly reared on naturally grown forest of Oak (*Quercus serrata* Thunb.) in North East India for the production of oak tasar silk. The Rearing of oak tasar silkworm is successful using spring season[6]. Spring crop is seed crop as well as commercial crop. Seed cocoons were preserved from one spring season to next spring season and were lost due to erratic emergence (20-30%) during the preservation period. So it is highly essential to conduct rearing in autumn crop as seed crop to get additional seed cocoons for the next spring crop grainage for production of more diseases free layings. In the present study seasonal variation of major leaf constituents of *Q. serrata* & nutritional indices of oak tasar silkworm in both spring and autumn season were estimated. Moreover an attempt has been made to study the cause of autumn season partly favourable for oak tasar crop.

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MATERIALS AND METHODS

Oak leaves of different types namely tender, semimature and mature were collected from plants/trees of Research Extension Centre, Umrangso field , Dima Hasao district of Assam during Spring and Autumn season in 2013 and 2014. For spring crop pruning and pollarding were done during the 1st week of December and for Autumn crop light pruning/clipping were done in 2nd week of August. The agronomical practices, application FYM and NPK were done for better quality and quantity of leaves for rearing seasons. The collected oak leaves (tender, semimature and mature) were dried separately at 60°C for 24 hours and powdered. The moisture content was estimated using the following formula.

$$\text{Moisture (\%)} = \frac{\text{Fresh weight} - \text{Dry weight}}{\text{Fresh wieght}} \times 100$$

Leaf constituents such as moisture, crude protein, crude fibre, crude fat, ash and carbohydrate were estimated according to A.O.A.C (1984) methods, at Central Muga Eri Research & Training Institute, Lahdoigarh, Jorhat, Assam. Nitrogen content of the leaves was estimated by Micro-kjeldahl method and crude protein was calculated by multiplying the estimated value of nitrogen content by 6.25. NPK and FYM in recommended dose per plant 48 gm Urea, 46.5 gm SSP, 9.3gm MOP, and 10 Kg FYM were applied. The pruning/pollarding, light pruning/clipping were done which help in maintaining optimum height of plant suitable for rearing and results to increase in the of soluble proteins, total sugar and free amino acids etc. besides reducing fibre contents. The seasonal change in the leaf constituents of *Quercus serrata* are present in Table.1,Table.2 and Table.3.

Table 1: Foliar constituents of Oak leaf *(Quercus serrata)* in 2013

Season	Leaf	Moisture %	Crude protein %	Crude firbe %	Crude fat %	Ash %	Carbohydrates %	ERR %
Spring 2013	Tender	73.50	12.30	5.25	1.80	1.65	8.85	65.10
	Semi mature	70.10	10.25	6.65	2.35	1.90	10.90	
	Mature	62.25	8.25	8.40	2.80	2.10	12.75	
	Mean	68.62	10.27	6.76	2.32	1.88	10.83	
	S.E ±	3.335	1.705	0.912	0.289	0.3535	1.128	1.756
	C.V	8.41	19.72	23.34	21.59	25.02	18.015	8.522
Autumn 2013	Tender	60.05	5.65	5.60	1.7	1.60	16.50	32.40
	Semi mature	57.15	5.10	6.75	1.90	1.85	20.35	
	Mature	54.40	4.5	8.90	2.25	2.30	26.30	
	Mean	57.2	5.08	7.08	1.95	1.92	21.00	
	S.E ±	1.625	0.3394	0.9683	0.161	0.205	2.854	1.046
	C.V	4.91	11.34	23.66	14.28	18.474	23.513	10.204

Table 2: Foliar constituents of Oak leaf *(Quercus serrata)* in 2014

Season	Leaf	Moisture %	Crude protein %	Crude firbe %	Crude fat%	Ash %	Carbohydrates %	ERR %
Spring 2014	Tender	73.60	12.5	5.20	1.85	1.60	8.80	63.90
	Semi mature	70.15	10.4	6.70	2.40	1.85	10.85	
	Mature	64.30	8.30	8.50	2.82	2.15	12.70	
	Mean	69.35	10.30	6.80	2.36	1.87	10.78	
	S.E ±	2.72	1.22	0.955	0.281	0.159	1.128	1.72
	C.V	6.78	20.424	24.294	20.614	14.73	18.098	8.493
Autumn 2014	Tender	60.10	5.75	5.75	1.65	1.60	16.55	30.80
	Semi mature	57.20	5.20	6.60	1.95	1.80	20.40	
	Mature	54.50	4.60	8.95	2.20	2.25	26.45	
	Mean	57.27	5.18	7.10	1.93	1.88	21.67	
	S.E ±	1.62	0.3325	0.96	0.159	0.1924	2.91	0.83
	C.V	4.89	11.104	23.38	14.27	17.71	23.229	8.493

Table 3: Foliar constituents of Oak leaf *(Quercus serrata)* 2013& 2014

Season	Moisture %	Crude protein %	Crude firbe %	Crude fat %	Ash %	Carbohydrates %	ERR %
Spring 2013	68.62	10.27	6.76	2.32	1.88	10.83	65.10
Spring 2014	69.35	10.30	6.80	2.36	1.88	10.78	63.90
Mean	68.98	10.28	6.78	2.34	1.88	10.80	64.50
Autumn 2013	57.20	5.08	7.08	1.95	1.92	21.00	32.40
Autumn 2014	57.27	5.18	7.10	1.93	1.88	21.66	30.80
Mean	57.23	5.13	7.09	1.94	1.90	21.33	31.60

RESULT AND DISCUSSIONS

The study shows that in spring season leaf moisture, crude protein, crude firbe, crude fat, ash, carbohydrates and ERR% were recorded 68.98%, 10.28%, 6.78%, 2.34%,1.88%, 10.80% and 64.50% respectly and during autumn season leaf moisture 57.23%, crude protein 5.13%, crude fibre 7.09%, crude fat 1.94%, ash1.90%, carbohydrates 21.33% and ERR 31.60% were recorded from Oak (*Quercus serrata* Thunb.) leaves. In spring season higher leaf moisture, crude protein and ERR% were observed while in autumn season less % leaf moisture and ERR% but higher % of carbohydrates were recorded. There were no significant different observed for crude fibre, crude fat and ash% content in both the season. The difference in nutritional quality of host leaves is an import factor for the success and partly success for silkworm rearing in different seasons. For instance,the predominant occurance of lepidopterous pests and feeding Chinese tasar silk worm, *Antheraea pernyi* on oak during the spring

season had been attributed to the presence of lesser amount of tannins, but increase of tannin in the following seasons the leaves become unsuitable for silkworm. In spring season higher crude protein% and lesser crude fibre % resulting higher % of ERR. of oak tasar silkworm(*Antheraea proylei*.). The quality of leaves has got direct influence on the healthy growth and survival of silkworm Sinha *et al*[9]. Better the quality of leaves, greater possibilities of obtaining good cocoon harvest. Therefore, the selection of the food plants possessing superior nutritive value could be utilized for the healthy development of silkworm for obtaining good cocoon crop. According to findings of Pandey & Goel[4], crude protein contents of young leaves were higher than old leaves in 3 oak species where *Q. serrata* showed maximum 28.92%, *Q. semecarpifolia* 20.77%, and *Q. incana* 16.47% but old leaves contained nearly half of total protein contents of young leaves. The young leaves contained less crude fibres, old leaves had nearly double the fibre content. Again, Pandey[6] observed seasonal changes in the leaf composition of *Q. serrata*, where leaf proteins were 6.81% in March and 7.89% in April which were decreasing 4.74% in October, and ash 2.23% in April which was increased from March 1.95% ,as result, the leaf quality of March and April month was found most suitable for rearing of *A. proylei*. A strong positive correlation was found between leaf content and larval body weight. The higher survival of oak tasar silk worm during spring season may be due to higher protein content of the leaves during April. Leaf quality for many lepidopteron larvae is determined on protein content basis[3].The autumn crop of oak tasar not fully success may be due to decline in protein content. Ponnuvel *et al.*[7] leaf moisture percentage of *Quercus serrata* leaves decreasing from February to November (71.9% to 56.78%) and crude protein decreasing from March (10.17%) maximum and minimum in September and October (5.39% and 5.07% respectly). In spring leaves contained less crude fibre (0.90% in February and 5.28% in March) but in autumn crude fibre 6.85% in September and 7.66% in October. Carbohydrates contents in leaves low in spring season and increasing in autumn in mature leaves.

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STUDIES ON THE PHYLLOPLANE MYCOFLORA OF OAK TASAR FOOD PLANT (QUERCUS SERRATA THUNB.) AND THEIR IMPACT ON REARING PERFORMANCE OF OAK TASAR SILKWORM ANTHERAEA PROYLEI JOLLY

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ABSTRACT

The aerial surface of Oak tasar food plant (Quercus serrata Thunb.) belong to the family Fagaceae, growing under natural conditions in Umrangso area of Dima Hasao district of Assam , boarding the hill regions and usually covered with large and varied populations of micro-organisms. The Oak tasar silkworm (Antheraea proylei Jolly.) is a bivoltine .Oak tasar food plant (Quercus serrata Thunb.) is a primary food plant of oak tasar silkworm. Its leaves provide a unique environment to their surface occupants and the typical leaves exudates influence the growth and development of the varieties of leaf surface micro-organisms. These microflora play an important role in supplying different types of nutrients to the plants as well as the silkworms.The present study deals with the isolation of the leaf surface mycoflora of Oak tasar leaves in different ages of leaves namely-tender,semi-mature, and mature leaves during the two rearing seasons i.e spring and autumn. 11 fungi have been isolated and indentified from the Oak tasar leaves during the two seasons. Aspergillus niger was the dominant species and Alternarea alternata was the co-dominant species in both the seasons while Penicillum species was prevalent during autumn. Studies on the rearing Oak Tasar silkworm during the corresponding seasons revealed better performance during spring season with a higher ERR(65.1%)and SR(10.17%) as compared to the autumn season.

KEYWORDS: phylloplane mycoflora, Quercus serrata, Antheraea proylei, ERR, SR.

INTRODUCTION

The leaf surface has been termed "Phylloplane" and the zone on leaves inhabited by microorganisms as "phyllosphere" by various workers.(Last,1955; Ruimen,1956 and Kerling, 1958). It is now well established that a large of micro-organisms inhabit the phylloplane of crop plants (Leben,1965; Preece and Dickinson,1967). The importance of assessing the microbial ecology, the aerial plant surface has now been recognized (Dickinson,1967; Pandey et al., 1989; Hollowman (1967); Kumar and Gupta (1976); Rai and Singh (1977). Tiwari and Sahu (1986;1987;1989;1991) have reported different kinds of mycoflora in different types of plants. In the present investigation, an attempt has been made to isolate and identify microfungi from the tender, semimature and mature leaves of Oak (Quercus serrata) plant which is a primary food plant of oak tasar silkworms(Antheraea proylei Jolly.).

MATERIALS AND METHOD

Tender semimature and mature oak leaves were collected randomly from REC Farm at Umrangso during 2013 in different seasons i.e spring and autumn during oak tasar silkworm rearing. The method of sampling of leaves as described by Kamal and Singh (1970) was followed during the collection of leaves. Serial washing technique of Kamal and Singh (1970) was used in which leaf discs were cut out from different categories of leaves with the help of sharp sterilized borer. Pieces of different categories of leaves were placed separately in 20ml of sterilized distilled water in 250 ml of erlenmeyer flasks and were shaken for 20 minutes at 120 rpm. The extract of the detachable fungal propagules from the leaf surface was determined by plating 1ml solution from washing to the Petri plates containing PDA media. The cut out leaf discs upper and lower surface were impringed on the surface of petridishes containing PDA media. The Petri dishes were incubated at 30±1°C for 4 days and then the plates are examined for the development of fungal colonies. The Experiment was conducted in two seasons viz spring and autumn. The isolated fungi were identified with the help of 'A manual of soil fungi by Gilman(1965) and "Illustrated genera of Imperfect fungi" by H.L.Baranatt (1960). Observation of the fungal isolates from Phylloplane of oak tasar food plant during

different seasons is presented in table 1 and 2. rearing data and economic parameters viz. effective rate of rearing cocoon weight , shell weight, silk ratio SR% were assessed during the two rearing seasons are shown in table 3.

Table 1: Fungal isolates from the phylloplane of oak tasar food plant Quercus serrata Thunb. During Spring (March- April, 2013) at Umrangsu, Dima Hasao, Assam.

Climatic factors	Status of leaf	Type of surface	No. of samples	Fungal isolates	% of occurence
Temp. (°C) Max. 31.06 Min. 18.05 RH Max. 70.86 Min. 55.41 Rainfall 427 mm (9days)	Tender	Upper	10	Aspergillus niger	70.50
				Alternaria alternata	18.00
				Mucor sp.	11.50
		Lower	10	Aspergillus niger	65.50
				Alternarea alternata	12.50
				Mucor sp.	11.50
				Curvularia sp.	10.50
	Semimature	Upper	10	Aspergillus niger	61.50
				Alternaria alternata	15.50
				Mucor sp.	14.50
		Lower	10	Curvularia sp.	8.50
				Aspergillus niger	57.50
				Alternaria alternata	15.50
				Mucor sp.	12.50
				Curvularia sp.	9.50
				Fusarium sp.	5.00
	Mature	Upper	10	Aspergillus niger	55.50
				Alternaria alternata	22.50
				Mucor sp.	12.50
				Curvularia sp.	3.50
				Fusarium sp.	6.00
		Lower	10	Aspergillus niger	45.50
				Aspergillus fumigatus	16.50
				Aspergillus flavus	3.50
				Alternaria alternata	12.50
				Mucor sp.	10.00
				Curvularia sp.	5.50
				Fusarium sp.	6.50

Table 2: Fungal isolates from the phylloplane of oak tasar food plant Quercus serrata Thunb. During Autumn (Sept –Oct, 2013) at Umrangsu, Dima Hasao, Assam.

Climatic factors	Status of leaf	Type of surface	No. of samples	Fungal isolates	% of occurence
Temp. (°C) Max. 31.81 Min. 22.46 RH Max. 83.03 Min. 55.79 Rainfall 817 mm (9days)	Tender	Upper	10	Aspergillus niger	55.50
				A.fumigatus	16.50
				A.alternata	15.50
				Mucor sp.	12.50
		Lower	10	A. niger	52.50
				A.fumigatus	19.50
				A.alternata	15.50
				Mucor sp.	6.50
				Fusarium sp.	6.00
	Semimature	Upper	10	A.niger	52.50
				A.fumigatus	17.50
				A.alternata	13.50
				Mucor sp.	8.50
				Penicillium sp.	8.00
		Lower	10	A.niger.	45.50
				A.fumigatus	15.50
				A.alternata	12.50
				Mucor sp.	6.50

				Penicillium sp.	6.50
				Curvularia sp.	9.00
				Fusarium sp.	4.50
	Mature	Upper	10	Aspergillus niger	45.50
				A.fumigatus	9.50
				A.flavus	4.50
				A.alternata	15.50
				Curvularia sp.	5.50
				Penicillium sp.	5.00
				Fusarium sp.	5.50
				Verticillium sp.	4.50
				Mucor sp.	4.50
		Lower	10	Aspergillus niger	45.50
				Aspergillus fumigatus	6.50
				Aspergillus flavus	3.50
				Alternaria alternata	14.50
				Mucor sp.	4.00
				Curvularia sp.	4.50
				Penicillium sp.	5.50
				Verticillium sp.	3.50
				Fusarium sp.	5.50
				Colletotrichum sp.	3.50
				Cladosporium sp.	3.50

Table 3: Rearing Performance of Antheraea proyeli Jolly.

Crop	Worm brushed	Larval period(days)	Wt of mature larva		Coccons harvested	ERR %	Av cocoon wt		Av shell wt		SR%		SR% (Av)
			♂	♀			♂	♀	♂	♀	♂	♀	
Spring 2013	1000	34-38	15.72	17.15	651	65.1	5.24	7.0	0.534	0.71	10.2	10.14	10.17
Autmn 2013	1000	34-40	15.67	17.07	324	32.4	5.03	6.9	0.49	0.65	9.74	9.42	9.58

RESULT AND DISCUSSIONS

Eleven number of fungi were isolated and identified from the leaf of oak tasar plant (Q. serrata L.) in different ages of leaves namely tender, semimature and mature leaves during the two rearing seasons spring and autumn.They were Aspergillus niger, A. fumigatus, A. flavus, Alternarea alternata, Curvularia sp., Mucor sp., Penicillium sp., Verticillium sp., Fusarium sp., Colletotricum sp., Cladosporium sp. The dominant fungal species which were isolated by Gupta and Khulbe(1991) throughout the year from the oak leaf litter inculed Mucor hiemalis, Aspergillus flavus, Penicillium spp, Fusarium solani, Phoma humicola etc. The fungi population showed increasing ability to colonise the leaves in order to their maturity. Aspergillus niger was the dominant species and Alternaria alternata was the co-dominant species in the both seasons while Penicillum species was prevalent during autumn season. It is observed that the environmental factors, atmospheric temperature, relative humidity, and rainfall seems to play a detrimental role in the quality and quantity of leaf surface mycoflora. Maximum number of fungi were recorded when the temperature was 31.81°C and the relative humidity was 83.03 %. The minimum number of mycoflora occurred during the spring season due to relatively low temperature (31.06°C) and relative humidity (70.86). According to Gregory (1961), Kumar and Gupta (1976), Pandey et al(1998), Sahu and Tiwari(1988), Tiwari(1977), Tiwari and Sahu(1989,1987), Sahu et al.,(1986) environmental factors are most important physical factors which affect the occurrence of micro-organisms on the leaf surface. The dominancy of Aspergillus spp. was also reported by Rajan et al.(1952), Singh and Baruah (1979) and Mishra and Shukla(1989). This may be due to richness of Aspergillus in the air over oak plantation field and their ability to colonise the leaf surface of the oak plants more easily than by others. Berustein and Feinberg (1947), Al Doory (1970), Agarwal et al. (1969), recorded marked seasonal periodicity of Aspergillus spp. This is however contrary to the findings of the Dickinson (1967), Pandey et al(1989) Sahu and Tiwari(1988), Sahu et al(1986) Tiwari (1977) and Tiwari and Sahu (1986,87,91) who reported Alternaria and Cladosporiium as dominant spp in Raphanus sativa ,Brassi campestris and Datura alva leaf surface. The dominance of Aspergillus fumigates over the leaf surface of the oak plants recorded in the present investigation may be due to leaf surface

morphology nutrient exudates of the leaves , local environmental factors, presence of more spores of this fungus in the air over the plantation, etc. Better performance was observed in spring season.ERR 65.1% and SR 10.17%.

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