FOOD BIORESOURCES AND ETHNIC FOODS OF MANIPUR, NORTH EAST INDIA

Thangjam Anand Singh
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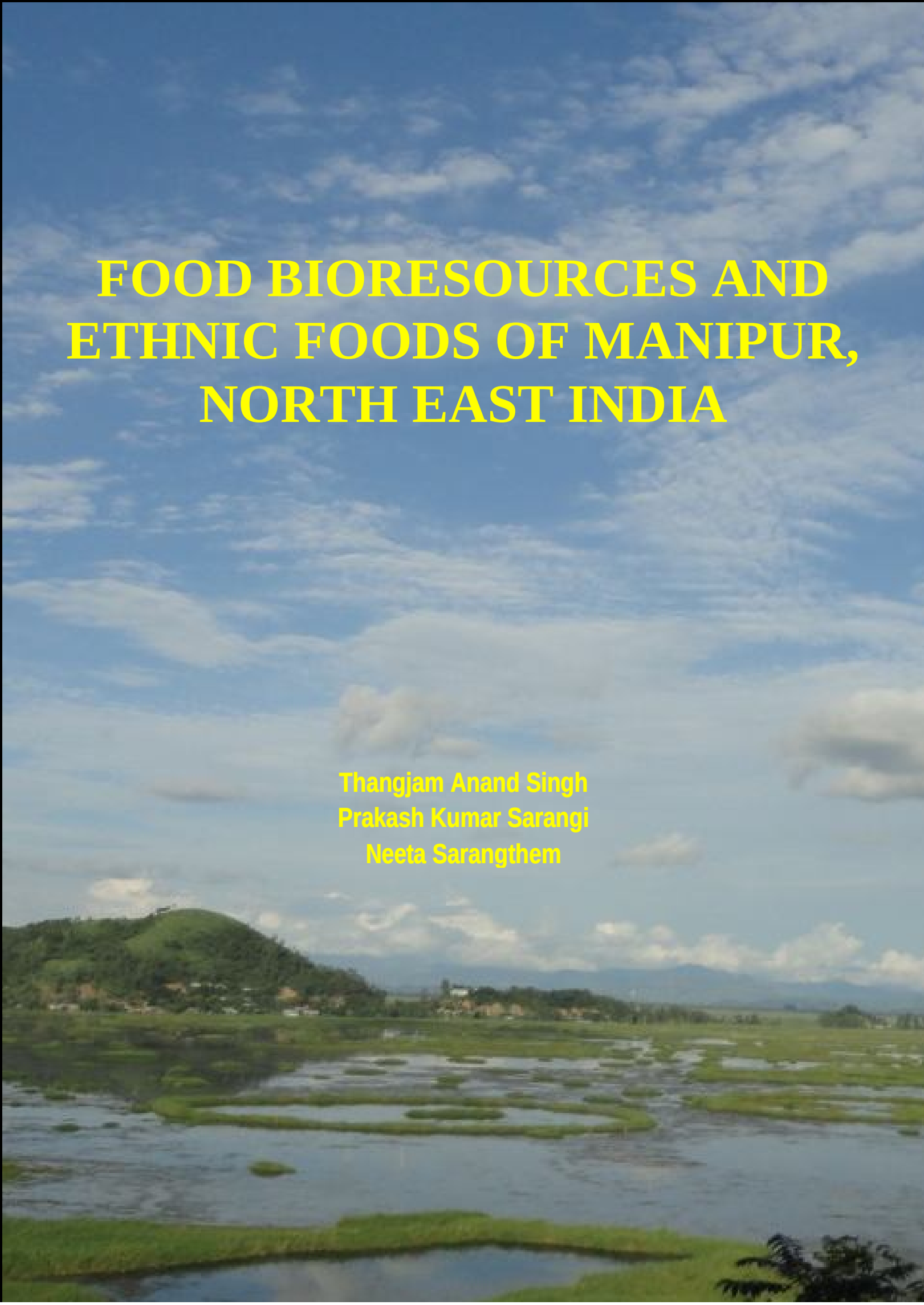

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Editors

About The Editors

Dr. Thangjam Anand Singh is a Scientist working with the Directorate of Research, Central Agricultural University in Imphal, India. He had his Ph.D. degree in broad area of therapeutic Enzymes associated with fermented foods of Northeast India from Biotechnology Department, Gauhati University, Guwahati, India although his major bulk of the PhD research was carried out at Institute of Bioresources and Sustainable development (IBSD), Imphal, India; M. Sc. Degree Biochemistry from Manipur University, Canchipur, India. During his research career he had worked with many renown Institutes in the Northeastern Hill Region (NEH) region of India including, North Eastern Regional Institute of Science and Technology (NERIST), Nirjuli, Arunachal Pradesh, Indian Council of Agricultural Research (ICAR), NEH region of India at Barapani, Meghalaya, IBSD, Imphal and have firsthand knowledge of Region during many field trips reaching nook and corners of this remote area. Dr. Singh has been working on traditional ethnic foods of the region. His research is focused on ITK, fermented foods, traditional food processing, ethno biology, food microbiology, nutraceutical and functional aspects of ethnic food and post-harvest engineering & technology. He has more than 15 years of teaching and research experiences in medicinal and aromatic plant, Microbial Biotechnology, Downstream Processing, Food biochemistry, Food quality and Molecular Biology. He also published more than 35 research articles in peer-reviewed international and national journal and authored in more than 5 book chapters and reviewed articles for many international journals. He is a member of Association of Microbiologists of India and Indian Science Congress Association.

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Preface

This book ***FOOD BIORESOURCES AND ETHNIC FOODS OF MANIPUR, NORTHEAST, INDIA*** is primarily aim at documenting the rich heritage of foods of ethnic people of State of Manipur, India. The State of Manipur which have about 90% hilly region and less than 10% valley area with altitude ranges from 180 msl to 2800 msl covering tropical, subtropical and alpine type of climate with average rainfall of 1200 cm of rainfall per annum. The forest cover is about 60%; limiting agricultural areas to less than 30% of the land mass. The dependence on natural forest for food and other needs leads to identification and consumption of wild edible plants and unconventional foods like insects, frogs, snakes etc. The valley had vast wetland areas which provide ample scope for fishing from wild.

The seasonal foods particularly, wild edible plants and fishes were processed and stored for yearlong consumption during offseason. The traditional processing were simple like fermentations, sun drying, roasting, boiling etc were commonly practiced. Here, traditionally salt was consider a luxurious item thus it was consciously missing during fermentation and other traditional processing. In this book various food bioresources of Manipur were documented with its nutritional medicinal and social values along with perceived values. Apart from farm produces, wild edible plants, aquatic plants, wild edible macrofungi, bamboo shoots, fish, insects and meat and milk were detailed in depth. It is believed that this book will help immensely to those foodies looking for exotic foods, researchers, anthropologist and lastly to the future generation who were alienated from their traditional foods due rapid urbanisation and fast life style.

Manipur is one of the north eastern states of India, surrounded by Nagaland in the north, Mizoram in south, Assam in west and eastern and southern part of the state is bounded by the border of Myanmar . The state lies at latitudes of 23°83'N – 25°68'N and longitudes of 93°03E – 94°78'E. This state has an area of 22327 sq km and population of about 3 millions. Wheat, pulses, paddy, maize, potato and mustard are the major crops grown in this state. Pineapple, papaya, banana, passion fruit, orange, lemon, mango are the major fruits and major vegetables include cabbage , cauliflower , pea , French bean, tomato. Major forest products include Oak, Teak, Pine, Cane Bamboo, Leihao , Uningthou etc. Bamboo shoot, Ginger, Pineapple, Mushroom are the major wild edible plant food products of this state. Glutinous rice is cultivated in plain area or valley as well as in riverbank of hilly areas.

There are 32 recognised tribes in Manipur included in two major tribe i.e Naga and Kuki. Naga tribes occupied northern and north western parts of Manipur. Rice is a staple food. They used metal, earthen pot or bamboo tube for preparing meal. During summer, they use to collect young bamboo shoots and dried it and they usually take dried bamboo shoot along with the rice. Kukis also known as Khongais occupy south western and south eastern hills of Manipur. Most of them usually live in the district of Churachandpur, Tengnoupal, Kangpokpi districts in northern Manipur. Rice is their staple food. There is certain restriction in the consumption of animal flesh. Rice beer (yu) is the most important alcoholic beverage as they use it in all the festivals of their community. Most of the ethnic tribes take dried fish, sometimes they also consume locust, dogs and different kinds of birds, frog and most of the domesticated animals though cat is seldom consumed.

Meitei, Meitei pangal are the Valley dweller communities of Manipur which do not belong to any schedule tribe. Most of the Meitei settle in plain areas and fish is one of the most important in their diet. Rice is their staple food and wheat is usually taken in the form of roti or chapatti. They do not take wheat as their staple food. Meitei pangals follow Islamic religion and mostly settled in Thoubal District of Manipur though sporadic settlement in Imphal East, Bishenpur Districts of Manipur. They provide exotic mosaic, rich in the tapestry of colour, rhythm and movement. Most of the ethnic groups in hills and plain areas prefer to plant wild plants for food slowly introducing these plants for cultivations. At present, the availability, variety and quantity of wild plants has been decreased significantly. The cultivated plants has gradually but indubitably replacing the wild plants thereby plummeting the number of wild plants and its significance.

All the different tribes of this state has its own traditional food and beverages. traditional food habit of different tribal group is simple and is related to ritual and festival, thus forming a mosaic of ethnic cultural combination. Traditional knowledge exist among different tribes has been practicing since time immemorial eg. Preparation of boiled food, fermented food, beverages, and nutritionally rich traditional products prepared from indigenous plants, forest products and meat of wild or domesticated animals. These foods are very important in their social life. There are many traditional foods which can prevent or cure many disease and disorder. In recent times, Ethnic diets are influenced by many factors and other cultures, due to various socio-political reasons. Sanskritisation is the most important factor that influences diet eg. Meeitei society were meat eating society after accepting Hinduism, vegetarian emerge in our society. Traditional ethnic food of different tribes get attention from several Anthropologies, Ethno botanists. Due to rapid urbanisation, there is less consumption of traditional food and consumption of fast food and ready to eat food increased, that may lead to heart disease, anaemia among pregnant and lactating women. Even though the new generation have adapted new food habits they need to document the traditional food products and their importance of different tribes.

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Table of Contents

Exploring indigenous flora of Manipur (North East India) as potential source of nutrients and dietary antioxidants

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Abstract
Manipur (North East India), being an integral part of Indo-Burma biodiversity hotspots has rich source of indigenous food plants. Ethnic communities of the state have comprehensible knowledge about the use of wild edible plants for sustainable and diverse diet. However, majority of such plants remain neglected or known to certain communities and existing data on their nutrient composition is limited. The present study was conducted to evaluate the nutrient and antioxidant composition of some of these indigenous floras to assess their potential role in bringing food and nutritional security of ethnic communities. Fourteen indigenous vegetables of Manipur were analysed for their nutrient and dietary antioxidant profiles. The proximate composition ranged from 6.35% to 18.00% for protein, 6.35% to 17.64% for total carbohydrate, 0.72% to 5.12% lipid, 9.69% to 36.35% dietary fibre, 5.40% to 14.80% ash and calorific value of 85.93 Kcal/100g to 157.52 Kcal/100g. The studied plants are found to be rich in protein, dietary fibre and ash. Dietary antioxidants were also studied in terms of total phenolics and flavonoid content, total antioxidant capacity and DPPH assays. The total phenolics and flavonoid content ranged from 9.62 –22.94 GAE/g and 8.42 - 22.68 RE/g respectively. Total antioxidant capacity ranged from 96.09 to 833.04 AAE/g and IC_{50} value ranged from 10.48 to 409.54 µl/mg. The present study revealed that Indigenous vegetables of Manipur have the potential to be good source of nutrients and might play significant role in overcoming nutrient deficiency in local diet.

Keywords: Manipur, wild edible plants, nutrient, antioxidants, ethnic, food security.

Introduction

Since time immemorial ethnic communities all over the world are using wild edible plants (WEPs) for various purposes- food, medicine, livelihood, cash income, cultural and religious purposes. Presently, about 90% of world food production comes from less than 30 species. By implication, the nutrition of the world's population depends on this limited number of food sources and only three three crops (rice, wheat, and maize) dominate as staple crops (Collins and Hawkin, 1999). In addition to this, growing climate change in present times pose a serious challenge to major agricultural crops, putting at risk the food security of a country in case of any major crop failure. Therefore, food and nutritional security is still a major concern despite phenomenal advancement in civilization. It is in this context, that wild food plants (WFPs) are receiving renewed interest and attention from researcher's and policy planners.

Wild food and medicine are plant resources that grow in natural conditions and which are harvested or collected for the purpose of human consumption and used as food, dietary supplements and medical treatment (Smith Hall *et al*., 2012; Price, 2006). Wild food knowledge is context specific in the sense that very different wild food species are used in distinct regions and use of the same species can differ widely from one region to another (Rivera *et al*., 2006). They are part of local people's identity and traditions (Heinrich *et al*., 2006). WEPs provide staple food for indigenous people; serve as complementary food for non-indigenous people and offer an alternative source of cash income (Gemedo-Dalle *et al*., 2005; Teklehaymanot *et al*., 2010). They also provide the much needed diet diversity of many ethnic communities and bring about household food security. WEPs are important source of nutrients, vitamins and mineral supplements for indigenous population (Ogle and Grivetti, 1985; Ali-Shtayeh *et al*., 2008) and hence play an important role in reducing food insecurity of local communities and act as a buffer in times of food shortage (Balemie and Kebebew, 2006; N'danikou, 2011). Many WEPs are reported to have to nutritional quality which is comparable or sometimes superior to many cultivated vegetables (Alfoyan and Jimoh, 2009; Sundriyal and Sundriyal, 2001). Apart from their rich nutritive values, they have therapeutic value due to the presence of biologically active compounds, and are referred to as functional food (Pereira *et al*., 2011; Sanchez-Mata *et al*., 2012). Therefore, incorporation of WEPs in the diet could be beneficial to nutritionally marginal population in eliminating micronutrient malnutrition and prevention of degenerative diseases. The use of wild edible food plants can also contribute to community health and conservation for sustainable utilization (Kalita *et al*., 2014).

Wild species can be used for the development of new crops through domestication and by providing plant breeders with a broad pool of potentially useful genetic resources for crop improvement (Hajjar and Hodgkin, 2007; Pandey *et al*., 2007). The conservation of these genetic resources is essential for the maintenance and improvement of agricultural production and for sustainable development. These genetic resources and associated cultural characteristics are important assets of the rural poor that need to be safeguarded (IPGRI, 2000) as neglect of WFPs and minor food crops may compromise food and nutritional security (FAO, 1998; Fahey 1998) of a particular region.

However, WEPs remained widely neglected despite being important nutritional source and is restricted to certain communities or regions. Ethnobotanical studies indicated that hundreds of wild species are still unexplored in each country, representing an enormous wealth of agrobiodiversity with potential to contribute to improved income, food security and nutrition, combating hidden hunger. Inventory of wild food resources on their diversity, availability, status, accessibility and usage along with nutritional evaluation can establish native species as alternative to achieve food and nutritional security. However, there are challenges with limited data reported on the nutritional and phytochemical composition of indigenous species (Konsam *et al*., 2016). Identification of wild edible species, which are believed to be nutritious, coupled with nutritional and phytochemical composition, may help in selection of potential and unique species. A study by Royal Botanical Garden, Kew (England) revealed that only 3.5% of the recorded species have been studied scientifically for their chemical composition and utility. With this gap there is a growing apprehension that many valuable wild food plants may be lost even before they are investigated scientifically. Therefore, there is a need for scientific investigation and documentation of the nutritive values, phytochemical activities and horticultural potential of indigenous species considering their long history of consumption by local communities.

Studies on WEPs from North East India with special reference to Manipur

The Northeast India, which is a part of both Himalaya as well as Indo-Burma biodiversity hotspot, is one of the global biodiversity hotspots recognized and is blessed with a broad range of physiographic and eco-climatic conditions. This region is at the confluence of the Indian Malayan, Indo-Chinese and Indian bio-geographical realms are unique in providing a profusion of habitats which accounts for its high biodiversity and rich traditional knowledge. It has the richest reservoir of plant diversity in India supporting 50% of India's biodiversity (Mao, 2000) and is one of the major centres for potential plants. Out of 3895 economically significant plant species reported from North East India, nearly 7.34% are used as wild vegetables, fruits and ethno-medicine (Sarma *et al*., 2010). Tribal communities of the region have comprehensible knowledge on the use of wild edible plants and highly dependent on them for their livelihood since ancient times. As many as 118 wild edible plants are reported to be used by different indigenous groups of Arunachal Pradesh (Angami *et al*., 2006). In the Sikkim Himalaya region, about 190 wild edible species have been known to possess food value, of which 45 are used on a large scale (Sundriyal and Sundriyal, 2004). Over 155 wild edible plants have been used as supplementary and emergency food by the tribes of Assam (Borthakur, 1996).

Manipur, a region of north-eastern India lies at a latitude of 23°27N'- 25°41' N and longitude of 93°61E'-94°48' E. It comprises of an area of 22, 327 km^2 with a population of 2,570,390 (2011 census) and is administratively divided into 16 districts. The state is rich in both cultural and biological diversity, having populated by diverse ethnic, linguistic and religious groups including 33 indigenous tribes. Agriculture is the single largest occupation in Manipur and the mainstay of the state's economy. Though comparatively small in area, it is home to about 13,500 plant species of which 7000 are endemic (Chatterjee *et al*., 2006). Some 2500 species of flowering plants have been reported from Manipur (Chakraborty *et al*., 2012). Forests account for 67% of the total land area of the state and serve as a prime habitat for many wild species that fulfill the needs of the communities throughout the year. The local communities in this region have rich traditional knowledge of plants and a large number of plant species including algae, fungi and angiosperm are reported to be consumed by them. Several studies undertaken in this regard have substantiated the importance of wild edible plants among different ethnic communities (Pfoze *et al*., 2011; Salam *et a l*., 2012; Gangte *et al*., 2013; Rajkumari *et al*., 2013; Singson *et al*., 2015). Devi *et al*. (2010) reported 45 wild edible plants; Salam *et al*. (2012) and Konsam *et al*. (2016) have respectively reported 46 and 68 species of wild leafy vegetables being sold in the local markets of Manipur.

Fig-1: Physical Map of Manipur (source: www.mapsofindia.com)

Nutritive potentials of wild edible plants (WEPs)

Human beings require sufficient food for growth, development and to lead a healthy life and it depends upon the quality and quantity of food taken by them. The quality of food depends upon the presence of relative concentrations of various nutrients such as proteins, fat, carbohydrates, fibre, vitamins and minerals. Carbohydrates, fats and proteins form the major portion of the diet and are also referred to as proximate principles and, while minerals and vitamins form comparatively a smaller part and play important role in the regulation of the metabolic activity of the body (Gopalan *et al*., 2004). Wild edible plants are perceived as being the healthiest for humans as they grow naturally without man's intervention and consequently are likely to contain the highest amounts of nutrients and beneficial substances (Sansanelli and Tassoni, 2014). WEPs are good sources of nutrients, minerals, fibre, essential fatty acids and enhance taste and colour in diet and many have the potential of broadening the narrow food base of the human (Aletor and Aladetimi, 1989; Janick and Simon, 1990). They are also potential sources of unusual colours and flavours, bioactive compounds, dietary supplements or functional foods. It is implicated that nutritional success is linked to diversification of the food base, therefore it is important to preserve the traditional uses of wild food plant as an alternative among the variety of vegetables already known to us. For all these reasons, the advancement in knowledge about their chemical composition is regarded as an important issue in nutritional and phytotherapy research (Salvatore *et al*., 2005; Ansari *et al*., 2005).

Wild food plants are generally characterized by high nutritional and low energy values (Trichopoulou *et al*., 2000). In comparison to cultivated species, wild food plants have higher fibre content (Leonti *et al*., 2006), are rich in antioxidants and flavonoids (Pieroni *et al*., 2002) and contain very low amount of lipids. Many were proven to have important beneficial effects in preventing several chronic diseases of modern society such as age-related diseases, heart pathologies, diabetes and different types of cancer (Trichopoulou *et al*, 2000; Finkel and Holbrook, 2000; Maritim *et al*., 2003). Wild edible species are sources of minerals, fibres, vitamins, pigments, essential fatty acids, other secondary metabolites such as alkaloids, polyphenolic compounds, gums and resins etc. Also they have anti-microbial, anti-inflammatory, hepatoprotective, anti-carcinogenic properties due to the presence of biologically active compounds and therefore can be considered as food-medicine (Sanchez Mata *et al*., 2012; Pereira *et al*., 2011). For example., *Borago officinalis* is a source of linoleic acid and other fatty acids that are precursor to mediators of the inflammatory response (Pereira *et al*., 2011). Wild and semi-wild vegetables such as amaranth (*Amaranthus* spp.) and black jack (*Bidens pilosa L.*) are more valuable sources of vitamin C, vitamins A and E, and iron and zinc than cabbage (*Brassica oleracea*) and Swiss chard (*Beta vulgaris*) (Modi et al., 2006). *Amaranthus viridis* contains 29.45% crude protein, 6.15% carbohydrate, 5% lipid, 4.90% crude fibre and 16.87% ash. Several reserachers like Orech *et al*. (2005), Rashid and Anand (2009) and Gelmesa (2010) have discussed the specific importance of such plants in preventing diseases caused by the nutrient deficiency during the periods of food crises.

Indigenous vegetables as source of antioxidants

Antioxidants are substances that inhibit oxidation, especially one used to counteract the deterioration of stored food products or remove potentially damaging oxidizing agents in a living organism. They are agents, which scavenge the free radicals and prevent the damage caused by them. Some of these compounds are exogenous in nature and are obtained from food; called the dietary antioxidants. Antioxidants include dozens of food based substances such as carotenoids like a-tocopherol, b-carotene, lycopene, ascorbic acid and some micronutrient elements. They many health benefits such as – improvement of overall health, longevity, detoxification, protection against heart diseases, reduced risk of cancer and cognitive problems, slower sign of ageing etc. Providing modern healthcare to rural people is still a far-reaching goal, due to economic constraints (Grover *et al*., 2002). Therefore, it becomes necessary that we resort to traditional forms of medicine that mainly depend on the locally available plant materials to cure various health disorders. Indigenous vegetables may be rich source of various phytochemicals that contribute to antioxidant activity in the diet (Uusiku *et al*., 2010) and thus providing strong protective effect against diseases associated with oxidative damage (Kaur and Kapoor, 2002). They are also among the most important sources of natural antioxidants for pharmaceutical applications against chronic diseases related to free radicals production. For example, *Dioscorea communis* and *Humulus lupulus* contain high amount of citric acid, malic acids, antioxidants which are beneficial to health due to their ability to chelate metals

(Sanchez Mata *et al*., 2012). The importance of antioxidant constituents of plant materials has been established in the maintenance of health by acting against stress related diseases such as infections, diabetes, cancer and coronary heart disease (Idowu *et al*., 2006). Several epidemiological studies have consistently shown that the consumption of fruits and vegetables is associated with reduced risk of chronic and neurodegenerative diseases, probably due to the presence of phenolic compounds and tocopherols (antioxidants) that are involved in the delay or prevention of oxidative reactions (Gerber *et al*., 2002; Di Matteo and Esposito 2003).

These beneficial effects of green leafy vegetables are attributed partly to antioxidants. Hence, attention should be given to the wild edible species, many of which are underexploited, but possess a tremendous potential to help people overcome the deadly diseases of modern society. The daily requirements for bio-available micronutrients and phytochemicals are obtained through the consumption of indigenous leafy vegetables (Uusiku *et al*., 2010). The consumption of these vegetables plays a key role in maintaining a balanced diet and helps to avert the chronic effects of malnutrition. Epidemiological studies indicate that a high intake of plant products in the diet is associated with reduced risk of a number of chronic health conditions such as cancer, neurodegenerative and cardiovascular diseases (Yahia, 2010). This could be partially attributed to the biological activities of phytochemical constituents such as phenolic compounds, pro-anthocyanidins, vitamins, carotenoids, flavonoids, saponins and iridoids (Francis *et al*., 2002; Podsedek, 2007). These phytochemicals are responsible for a multitude of biological effects, including antioxidants, anti-inflammatory, antimicrobial, anti-cancer activities. In particular, phenolic compounds have become biomarker of nutritional quality of food because of their antioxidant activity (Mertz *et al*., 2009). Phenolic compounds are known to exert therapeutic activities because of their antioxidative and anti-inflammatory properties. They play vital role in the stability of food products, as well as in the antioxidative defence mechanism of biological systems (Nijveldt *et al*., 2001). The indigenous leafy vegetables consumed in southern Africa had higher levels of phytochemicals and also exhibited more potent antioxidant activity compared to the commercial varieties (Moyo *et al*., 2013). Several underexploited green leafy vegetables are known as potent source of antioxidant and other phytoconstituents as revealed by Subhasree *et al*. (2008). For example, *Centella asiatica* has been found to possess chemical constituents like vallarine, centellin, asiaticin, centellicin, pentacyclic triterpenes, tannins and saponins (Oyedeji and Afolayan, 2005). *Sauropus androgynus*, an underexploited shrub, is an excellent source of vitamins A, B and C.

In recent years, the use of natural antioxidants present in food and other biological materials has attracted considerable interest due to presumed safety, nutritional and therapeutic value (Ajila *et al*., 2007). They are in high demand for application as nutraceuticals, bio-pharmaceuticals as well as food additives because of consumer preference.

Naturally occurring constituents like polyphenolics, terpenoids and provitamins are replacing as alternative therapeutic agents to fight against various oxidative stress-induced diseases. Researchers are attempting to isolate powerful and non-toxic natural antioxidants from native species not only to prevent autoxidation and lipid peroxidation, but also to replace synthetic antioxidants. Searching wild sources may bring new natural products into the food industry with safer and better antioxidants that provide good protection against oxidative damage, which occurs both in the body and our daily food. This could help unearth plant species that could influence health sector in the future as there is lack of information on the dietary intake of plant-derived bioactive phytochemicals, especially in developing countries (Hervert-Hernandez *et al*., 2011).

Materials and methods

Extensive market surveys were conducted in all districts of Manipur from August 2012- March 2014 throughout different seasons for availability of WEPs from time to time collecting about 69 WEPs. To prioritised the species, an Integrated Assessment (Konsam *et. al*, 2016) was performed based on certain criteria viz.- taste, availability, edible part, frequency of use, community status, market potential, medicinal value etc. Each species is scored against these criteria and integrated values (Wujisguleng and Khasbagen, 2010) were calculated. Based on this assessment some of the highest scoring species are selected for further analysis.

The samples selected for the study are as follows – *Gnaphalium indicum* L., *Viola pilosa* Blume, *Cissus adnata* Roxb, *Amomum* sp., *Cycas pectinata* Buch.-Ham, *Antidesma diandrum* (Roxb.) Heyne.ex.Roth, *Elatostema lineolatum* Wight, *Piper pedicellatum* DC, *Ficus bengalensis* L., *Ficus palmata* Forssk, *Rhynchotechum ellipticum* A. DC., *Wendlandia glabrata* DC., *Zanthoxylum budranga* DC, *Clerodendrum colebrookianum* Walp.

Methods

1. Proximate Composition

Biochemical analysis of the samples was carried out on dry weight basis. The major nutritional components analysed were crude protein, total carbohydrate, total lipid, dietary fibre and total mineral in the form of ash content. Crude protein, total lipid and ash were determined following the standard methods of AOAC (2000). Crude protein was determined from the total nitrogen content in the sample estimated by micro-Kjeldahl method (AOAC) by multiplying by a factor of 6.25. Crude lipid was estimated by soxhlet-extraction method using petroleum ether (bp 40-60° C). Total ash content was determined by dry ashing in muffle furnace at 630° C for 3-4 hrs. Dietary fibre was determined by acid-base digestion (Sadasivam and Manickam, 2007). Total carbohydrate content was determined by Anthrone method (Sadasivam and Manickam, 2007). And the calorific values were calculated in

Kilocalories (Kcal) by multiplying the values for carbohydrate, fat, and protein by 4, 9 and 4 respectively and adding up the values. Energy, E= [(9×fat) + (4×carbohydrates) + (4× protein)] Kcal /100g**.**

2. Estimation of Antioxidants

Antioxidants potential of the selected species were determined in terms of- Total phenolics content (TPC), total flavonoids content (TFC) and DPPH radical scavenging activity assay and Total antioxidant capacity (TAC).

Total Phenolic Content (TPC) was determined by the Folin-Ciocalteu method (Singleton and Rossi, 1965) with the slight modification. Total Flavonoid Content (TFC) was measured by the Aluminium chloride calorimetric method of Park *et al*. (2008). The total antioxidant capacity of the methanolic fraction was determined by phosphomolybdate method (Rai *et al.*, 2011) using ascorbic acid as a standard. DPPH radical scavenging activity was estimated following the method of Barros *et al*. (2010). The radical scavenging activity (RSA) was calculated as a percentage of DPPH discoloration using the following equation.

$\% RSA = [(A_C - A_S) / (A_C)] \times 100$

Where, A_C = absorbance of control

A_S = absorbance of sample

The antiradical activity was expressed as IC_{50} (µg/ml), the concentration required to cause 50 % DPPH inhibition. The IC_{50} was calculated from the graph of % RSA against extract concentration. Ascorbic acid (AA) was used as a positive control in all assays. All the experiments were done in triplicates and the results were expressed as mean ± SEM.

Result and Discussion

Analysis of nutritional composition of selected WEPs

The proximate composition of selected wild edible plants is presented in Table 1. The crude protein content of the analysed vegetables ranged from 6.35% in *Viola pilosa* to 18.00% in *Amomum* sp. According to the Food and Nutrition Board (2001), food plants that provide more than 12% of their calorific value of protein are a good source of protein. In that context, eight species viz. *Antidesma diandrum* (16.06%), *Gnaphalium indicum* (15.68%), *Amomum* sp. (18%), *Cycas pectinata* (13.87%), *Ficus bengalensis* (13.91%), *Piper pedicellatum* (15.16%), *Clerodendrum colebrookianum* (16.77%), *Rhynchotechum ellipticum* (15.53%) have high protein content. This is comparable to the protein values ranging between 13.25- 26.44 % in the leafy vegetables of South Africa as reported by Afolayan and Jimoh (2009). Study by the same author also reported that lack of protein, either in quality or quantity, leads to low body mass, growth retardation in children and developmental deficiency during pregnancy etc. which are the major nutritional concerns among ethnic communities of Africa and Asia. Protein-calories malnutrition is still a major factor

responsible in nutritional pathology (Roger *et al.*, 2005). The high crude protein content of these wild vegetables may encourage their use to prevent protein malnutrition.

The total carbohydrate content of the samples varies considerably and from 6.37% in *Ficus bengalensis* to 17.84% in *Elatostema lineolatum.* Total carbohydrate in other wild species have been found to be similar such as *Cucurbita moschata* has 9.65%, *Perilla frutescens* 11.52%, *Diplazium esculentum* 5.06%, *Amaranthus viridis* 6.15% and *Dryopteris cochleate* 9.86 % of total carbohydrate (Handique, 2003). The recommended dietary allowance (RDA) for carbohydrates is 130 g (FAO, 1998). Therefore, the present study indicates low carbohydrate content in indigenous vegetables and thus can be recommended to people with specific health conditions who need a low-carbohydrate diet.

Lipid content has been found to be relatively low in the range of 0.72% to 5.12% with a mean value of 2.66%. The lowest value is observed in *Piper pedicellatum* and highest value is observed in *Rhynchotechum ellipticum.* The finding bears similarity with the vegetable species of Mardan, ranging between 0.3% - 6.1% (Hussain *et al.*, 2011). This indicates that Wild leafy vegetables are poor source of lipid. Due to this, consumption of wild vegetables in large amount is a good dietary habit especially for people suffering from overweight and obesity.

A. *Gnaphalium indicum*

B. *Viola pilosa* Blume

C. *Cissus adnata* Roxb.

D. *Amomum* sp.

E.*Cycas pectinata*

F.*Antidesma diandrum*

G.*Elatostema lineolatum*

H. *Piper pedicellatum*

I.*Ficus bengalensis*

J. *Ficus palmata*

K. *Rhynchotechum ellipticum*

L. *Wendlandia glabrata*

M. *Zanthoxylum budranga*

N. *Clerodendrum colebrookianum*

Fig. 2 Pictures of the analysed wild edible plants in the study

The crude fibre content has been found to be very impressive and considerable variations in the range of 9.69% to 36.35%. The highest fibre content is found in *Rhynchotechum ellipticum* (36.35%), followed by *Ficus bengalensis* (24.83%), *Amomum* sp. (23.73%), *Elatostema lineolatum* (22.62%) and *Cycas pectinata* (20.44%) etc. As many as ten of the fourteen species have high dietary fibre content above 15% which is remarkable. The values were comparable to the values of 8.50 - 20.90% and 16.08 - 23.08 % reported in the wild leafy vegetables of Nigeria (Isong and Idiong, 1997) and South Africa (Afolayan and Jimoh, 2009). High level of dietary fibre is advantageous for their role in regulation of intestine, increasing dietary bulk and faecal consistency (Jenkin, 1986). It also has beneficial physiologic effects and health benefits such as lowering cholesterol level, risk of colon cancer and losing weight (Jones and Varady, 2008) in human beings. The low lipid and high fibre content of WEPs could be particularly useful in combating lifestyle diseases of modern times.

Total mineral in the form of ash content has been found to be very impressive and the range of variation is 5.40 % to 14.80 %. The lowest ash in content was found *Cycas pectinata*, and the highest ash content in *Elatostema lineolatum*. Other species with high ash content are *Viola pilosa* (11.49%), *Amomum* sp. (11.45%), *Rhynchotechum ellipticum* (10.52%), *Gnaphalium indicum* (10.51%) etc. Similar ash values were reported in wild edible plants of Iran and India (Aberoumand and Deokule, 2009). The analyzed wild greens also contain higher ash content than some commonly consumed vegetables like cabbage (0.6%), cauliflower (3.2%), spinach (1.7%), shepu (2.2%), lettuce (1.2%) etc. (Gopalan *et al*., 1996). The results indicate that these non-conventional vegetables are good source of minerals. By comparision these indigenous vegetables are superior in ash content and therefore should be promoted in diet for alleviating problem of micronutrient deficiency.

The calorific value ranged from 85.93 Kcal/100g in *Zanthoxylum budranga* to 157.52 Kcal/100g in *Rhynchotechum ellipticum*, followed by *Clerodendrum colebrookianum* (156.77 Kcal/100g), *Gnaphalium indicum* (138.33 Kcal/100g), *Cycas pectinata* (135.61 Kcal/100g) etc . The variations in total carbohydrate, lipid, crude protein, ash, dietary fibre and moisture content of the wild edible plants were found to be highly statistically significant ($p < 0.05$). Variations in the chemical constituents may be due to species differences as all these species belong to different families. Difference in agro-climatic conditions, different age and stage of the plants may also play a role in it.

Antioxidant analysis

The total phenolic content in the studied plants showed wide variations from 9.62 mg GAE/g in *cycas pectinata* to 22.94 mg GAE/g in *Antidesma diandrum*. Four other species viz- *Ficus palmata* (21.74 mg GAE/g), *Zanthoxylum budranga* (21.62 mg GAE/g), *Wendlandia glabrata* (21.21 mg GAE/g), *Clerodendrum colebrookianum* (20.27 mg GAE/g), showed promising phenolics content. The findings revealed that such species can be explored as source of phenolic compounds. Morales *et al*. (2012) had reported a much higher values of total phenolics in Spanish wild leafy vegetables (26.72- 80.47 mg GAE/g extract. However, the total phenolic content of unconventional leafy vegetables consumed in southern Africa is significantly lesser (2.68 - 5.16 mg GAE/g) from present study (Moyo *et al*. 2013).

The Total flavonoid content of the leafy vegetables was found to be comparable to the total phenolics ranging between 8.42 -22.68 mg RE/g. The highest flavonoid content is found in *Clerodendrum*

colebrookianum (22.68 mg RE/g) followed by *Gnaphalium indicum* (17.84 mg RE/g), *Cissus adnata* (15.61 mg RE/g), *Zanthoxylum budranga* (15.10 mg RE/g), while the least is found in *Piper pedicellatum* (8.42 mg RE/g).The resulted values are comparable with the flavonoid content of wild Spanish vegetables ranging between 6.09-21.65 mg CE/g extract (Morales *et al*., 2012).

Table-1: Proximate composition of selected wild edible plants of Manipur

Sl. no.	Species	Edible part	Moisture content % ± SEM	Crude Protein % ± SEM	Total Carbohydrate % ±SEM	Total Lipid % ± SEM	Ash content % ± SEM	Crude Fibre % ± SEM	Energy kCal/100g
1.	*Antidesma diandrum* (Roxb.) Heyne.ex.Roth	Leaf	87.74±0.06	16.06±0.15	10.04 ±0.09	1.89±0.07	7.40± 0.02	**9.69±0.12**	121.41
2.	*Gnaphalium indicum* L	Whole part	85.65±0.05	15.68±0.01	11.59±0.22	3.21±0.11	10.51±0.00	18.55±0.06	137.97
3.	*Amomum* sp.	Inflorescence	85.80±0.03	**18.00±0.06**	12.54 ±0.28	1.04±0.04	11.45±0.00	23.73±0.03	131.28
4.	*Cycas pectinata* Buch.-Ham	Leaf	**89.19±0.02**	13.87±0.07	9.07 ±0.08	4.85±0.21	**5.40± 0.00**	20.44±0.23	135.41
5.	*Viola pilosa* Blume	Leaf	88.08±0.02	**6.35±0.08**	**17.64±0.22**	2.18±0.09	11.49±0.00	17.61±0.37	115.58
6.	*Cissus adnata* Roxb.	Leaf	**85.27±0.08**	9.85±0.14	9.24 ± 0.16	3.4±0.08	8.10±0.02	18.75±0.14	106.96
7.	*Ficus bengalensis* L.	Inflorescence	87.22±0.05	13.91± 0.16	**6.37 ± 0.19**	3.08±0.02	6.39± 0.00	24.83±0.14	108.84
8.	*Wendlandia glabrata* DC.	Inflorescence	87.93±0.01	11.20±0.10	7.30 ± 0.20	1.98±0.06	6.96± 0.01	11.88±0.07	91.82
9.	*Zanthoxylum budranga* DC	Leaf	88.22±0.02	6.39 ±0.14	11.47±0.24	1.61±0.05	7.63± 0.00	14.78±0.17	**85.93**
10.	*Elatostema lineolatum* Wight	Leaf	86.76±0.03	9.67±0.14	**17.86 ±0.15**	1.66±0.05	**14.80±0.00**	22.62±0.25	125.06
11.	*Piper pedicellatum* DC	Leaf	86.06±0.04	15.16±0.10	9.85 ±0.12	**0.72±0.01**	10.48±0.00	18.79±0.16	106.80
12.	*Ficus palmata* Forssk	Leaf	88.20±0.01	7.03± 0.13	8.97 ±0.09	3.81±0.01	7.78± 0.04	16.44±0.23	98.29
13.	*Clerodendrum colebrookianum* Walp.	Leaf	88.65±0.01	16.77± 0.19	16.10 ±0.24	2.81±0.01	5.67± 0.01	13.10±0.09	156.77
14.	*Rhynchotechum ellipticum*	Leaf	88.69±0.04	15.53±0.47	10.08±0.21	**5.12±0.12**	10.52±0.04	**36.35±0.25**	**157.52**
	Mean value		87.39	12.53	10.64	2.66	8.89	19.11	119.97
	F ratio value		855.56	527.17	33.17	162.76	1417.53	1200.92	273.34

The interspecific variation is significant at $p<0.05$. Values are expressed as mean of 3 replicates. The values are % of dry weight.

The total antioxidant capacity is expressed as Ascorbic acid equivalent, mg AAE/g extract. The total antioxidant capacity of the wild vegetables ranged from 96.59 to 833.34 mg AAE/g. *Gnaphalium indicum* has the highest total antioxidant capacity (833.34), followed by *Clerodendrum colebrookianum* (818.07), *Piper pedicellatum* (738.19), *Ficus palmata* (626.73), *Rhynchotechum ellipticum* (526.26), *Antidesma diandrum* (492.11) while the remaining species have values less than 286.01 mg AAE/g. These species also have higher total phenolics content compared with the rest. There may be a positive co-relation between high phenolics content and antioxidant capacity.

DPPH radical scavenging assay is expressed in terms of IC_{50} value, which is the concentration of antioxidant necessary to decrease the initial DPPH concentration by 50%. The lower the IC_{50}, the higher is the antioxidant power. The IC_{50} value of the WEPs analyzed ranged from 10.48 to 409.54 µg/ml. *Ficus palmata* (10.48 µg/ml), *Rhynchotechum ellipticum* (10.58 µg/ml) and *Clerodendrum colebrookianum* (10.67 µg/ml) have the lowest IC_{50} value and the most potent antioxidant activity. Besides these three species, *Zanthoxylum budranga* (33.33µg/ml), *Gnaphalium indicum* (39.47 µg/ml), *Antidesma diandrum*

(46.38 µg/ml), *Piper pedicellatum* (50.20 µg/ml) also possess high radical scavenging activity. These species also have high total phenolics content and total antioxidant activity which agrees with their low IC_{50} value. The resulted IC_{50} values are comparable with those of green leafy vegetables ranging between 85 - 435 µg/ml as reported by Subhasree *et al*. (2009).

Table-2: Total phenolics, flavanoids, antioxidant capacity assay and IC_{50} values of WEPs of Manipur

Sl.no	Species name	Total Phenolics (mg GAE/g fraction)± SEM	Total flavonoid (mg RE/g fraction) ± SEM	TAC mg AAE/g extract ± SEM	DPPH radical scavenging assay IC_{50} ug/ml
1.	*Antidesma diandrum*	**22.94 ±0.03**	13.92 ± 0.10	492.11± 4.37	46.38
2.	*Gnaphalium indicum*	15.11 ± 0.06	17.84 ± 0.09	**833.04± 2.66**	39.47
3.	*Amomum* sp.	9.84 ±0.09	11.89 ± 0.06	**96.09± 1.09**	**409.54**
4.	*Cycas pectinata*	**9.62 ±0.07**	11.42 ± 0.01	104.19± 1.14	87.42
5.	*Viola pilosa*	13.14 ±0.13	10.50 ± 0.03	241.71± 2.84	124.54
6.	*Cissus adnata*	12.84 ±0.07	15.61 ± 0.04	198.66± 2.22	53.06
7.	*Ficus bengalensis*	11.94 ±0.14	13.76 ± 0.13	166.03± 2.43	74.27
8.	*Wendlandia glabrata*	21.21 ±0.15	13.69 ± 0.05	286.01± 3.05	121.19
9.	*Zanthoxylum budranga*	21.62 ±0.11	15.10 ± 0.12	134.75± 0.99	33.33
10.	*Elatostema lineolatum*	10.02 ±0.05	11.91 ± 0.09	139.17± 1.79	106.96
11.	*Piper pedicellatum*	13.40 ±0.07	**8.42± 0.10**	738.19±2.77	50.20
12.	*Ficus palmata*	21.74±0.05	12.73 ± 0.15	626.73± 2.42	**10.48**
13.	*Clerodendrum colebrookianum*	20.27 ±0.20	**22.68 ± 0.04**	818.07± 1.67	**10.67**
14.	*Rhynchotechum ellipticum*	16.98 ±0.04	14.51 ± 0.06	526.26± 3.68	**10.58**
	Mean value	15.76	13.85	385.78	84.14
	F ratio value	2148.96	1442.33	12354.72	5909.73

The interspecific variation is significant at $p<0.05$

In this study, *Clerodendrum colebrookianum* showed highest TAC value with 818.07 mg AAE/g while DPPH assay also showed it to be among the best with IC_{50} value of 10.67 µg/ml. Moreover its phenolics and flavonoid content were also highest or among the highest which strongly indicate a positive correlation between the two. Likewise, *Amomum* sp. recorded lowest TAC value of 96.09 mg AAE/g; while it was also found to exhibit the weakest IC_{50} value of 409.25 µg/ml; at the same time its phenolics and flavonoid contents were among the lowest. Other two species *Ficus palmata* and *Rhynchotechum ellipticum* exhibited strongest antioxidant activity based on DPPH assay; but based on TAC assay their antioxidant activity were found to be average. There are diverse reports about correlation between antioxidant content and in-vitro antioxidant activity. Guleria *et al*. (2011) working with 20 medicinal plants found significant and linear positive correlation between phenolics and antioxidant activity; but the degree of antioxidant activity based on FRAP and DPPH were quite different. The non-identical result between two or more assay method appear to be due to difference in reaction mechanism and principle involved as well as phytochemical diversity (Subhasree *et al*., 2009).

Conclusion

The NE region of India which falls in the Indo-Burma hotspot support considerable biodiversity. Wild edible plants (WEPs) are widely consumed in the daily diet of the local people. The diverse use of wild plant resources for food, medicine, income and socio-cultural purposes by the ethnic communities of Manipur revealed the high dependence on them for their sustenance. However, WEPs received a little attention in research activities, economic development, biodiversity conservation and sustainable management. Many are largely ignored and remained unexplored. Information on their chemical composition is also scanty. With a view of reducing the gap in traditional knowledge and tapping the hidden potential resources for proper utilization, exploitation, and sustainable management of WEPs are crucial.

Nutrient evaluation of selected species shows its potential as important source of proteins, fibre, and total mineral content for incorporation in human diet. Nutritional values of the studied plants are comparable or sometimes even superior to commonly consumed vegetables. Wild vegetables can be promoted into diets as the most practical and sustainable way to prevent common diseases and ensure food and health security of local population. The investigated plants also had varying concentration of phytochemical constituents like phenolics, flavonoids etc., which are cumulatively responsible for the antioxidant activity and may be responsible for good human health. These species have potent antioxidant activity which finds a good motivation for exploration of horticultural potential of indigineous vegetables. Further research should focus on the identification and quantification of individual phytochemical constituents as well as quantification of daily dietary intake of these species for the local population. The studied plants could be selected because of their nutritional interest and high antioxidant activity to be used as functional food or as potential functional ingredients in food industry.

The high nutritional quality and unique taste of these unconventional vegetables, which make up an integral component of traditional dishes of the local population, are likely to be neglected in the near future. Therefore, it is now imperative that a nutritional, anti-nutritional and medicinal database of these wild species should be set up to retain the information of these unique species. Documentation and conservation of highest priority species would ensure they are available for use in genetic improvements of crop species as a contribution towards food and nutritional security.

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Ichthyofaunal Resources of Manipur

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Abstract

Manipur, also popularly known as "Jewel of India", is the part of the north eastern India situated at latitude of 23°83'N and 25°68'N and longitude of 93°03'E and 94°78'E, with a total area covered of 22,347 km². The state is considered to be the meeting place of the eastern Himalaya and the Burmese region and feeding with two majour river systems viz. the Barak-Surma-Meghna system and the Chindwin-Irrawady system. Manipur is blessed with 218 species of fishes belong to 92 genera under 32 families and 11 orders. Fifity five species of fish have been discovered in the last two decades. The Chindwin River basin is represented with 25% of threatened species inwhich 21% of species are under vulnerable category while the Barak River basin is with 10% threatened species in which 5% are undef vulnerable category. The species under least concern are more in the Barak River with 66% of species than the Chindwin River with only 38% of species. The fishes of the region are facing threats due to expansion of developmental process in the catchment areas, destructive fishing including explosives, poisoning, dynamiting, electrofishing, as well as overexploitaion using fish barrages. A regular monitoring of habitat condition and population trends of fishes should be undertaken for conservation of such valuable natural resources and its sustainable utilization.

Keywords: Ichthyological resources, Manipur, Chindwin River, Barak River, Inrernatoinal Union of Conservation of Nature and Natural resources Status, Conservation, Sustainable utilization.

Introduction

Fishes exhibit enormous diversity in their morphology and biology and occupy diverse habitats. Unlike the other commonly recognized vertebrates, fishes are a heterogeneous assemblage of aquatic vertebrates with branchial respiration, distributing every corner of the aquatic medium (Forese & Pauly, 1998). They constitute one half of the total number of vertebrates in the world with over 35,200 species (Frick *et al.*, 2019). Of these, about 41 per cent are freshwater species and 1 per cent move back and forth between salt and freshwater. Freshwater biodiversity contributes a vitally important component of the planet, with a species richness that is relatively higher compared to both terrestrial and marine ecosystems (Gleick, 1996).

Ichthyofaunal diversity refers to the variety of fish species in respect of the context and scale, to alleles or genotypes within fish population, per cent to species of life forms within a fish community and to species or life forms across aqua regimes (Burton *et al.*, 1992). Biodiversity is essential for stabilization of ecosystems, protection of overall environmental quality for understanding intrinsic worth of all species on the earth (Ehrlich & Wilson, 1991).

Fish forms the most important wetland product on a global scale, and is certainly the most utilized wetland resource. Asia accounts for 70.8 per cent of total fish production (Chang *et al.*, 2017) and fish accounts for more than 71 percent of the typical diet across Asia as a whole (FAO, 2018). It has great cultural and psychological value to human beings. Freshwater fishes are defined as those that spend all or a critical part of their life cycle in freshwaters. There are an estimated 13,000 freshwater fish species in the world. IUCN Red list assessment of freshwater fishes of the Eastern Himalaya shows that about 2 per cent of fishes of the region are at high risk of extinction (Vishwanath *et al.*, 2010).

The Northeastern India, a part of the Eastern Himalaya, is a land of Blue Mountains, Green Valleys and Red River which lies between 21°57' and 29°23' N and between 87°58' and 97°09' E. It extends from Sikkim eastwards embracing the Darjeeling hills of West Bengal to Arunachal Pradesh and to Mizoram in the south-east. The region covers over an area of about 2,62,230 sq. km and comprises of the eight sister states, viz, Arunachal Pardesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland, Tripura, and Sikkim.

The Northeastern India has five major river systems like the Brahmaputra, the Barak-Surma-Meghna, the Karnafuli, the Koladyne (Kaladan), and the Chindwin-Irrawaddy with their countless tributaries. The region accounted for 11 per cent of country's total (1,95,210 km) rivers and canal water resources and 8 per cent of other water resources for aquaculture, which include reservoirs, tanks, lakes, floodplain lakes and derelict water and brackish water. These water bodies' harbours diversified native ornamental fishes with large economic importance under natural condition.

Ichthyological diversity of northeast India

Northeast India which forms a part of the Eastern Himalaya falls within the tropical monsoon climate with an average annual rainfall of 200 cm, containing the rainiest locality of the world, i.e. the Mawsynram – Cherrapunjee area. Besides north east India has one of the densiest drainage network in the world, due to essentially high rainfall and the character of the topography with hills, mountain and flat land just opposite together. It lies between 21°57′ and 29°23′N and between 87°58′ and 97°09′ E. The region extends over an area of about 2,62,230 sq. km. It comprises of the eight sister states, viz., Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland and Tripura and Sikkim. The area covers the middle Brahmaputra and parts of the Upper Brahmaputra, the Himalayan foothills, the Ganges delta and plain, the Chin-Arakan coast and the Sittaung-Irrawaddy ecoregions (Abell *et al.* 2008). The region is drained by four major important drainages viz., the Brahmaputra, the Barak-Surma-Meghna, the Kaladan, and the Chindwin. The Chindwin–Irrawaddy drainage is entirely separated by high mountain ranges from the watersheds of the Barak–Brahmaputra (Chaudhuri, 1919), and the Koladyne (Kaladan).

Drainages of Manipur

Manipur, also popularly known as "Jewel of India", is located in the north eastern part of India. The state lies at latitude of 23°83'N and 25°68'N and longitude of 93°03'E and 94°78'E. The total area covered by the state is 22,347 km². The capital lies in an oval-shaped valley of approximately 700 square miles (2,000 km^2) surrounded by blue mountains and is at an elevation of 790 metres above the sea level. The slope of the valley extends from north to south. The origin and evolution of the valley may be the result of

tectonics activity and neotectonism remarkably influenced by a long history of fluvialacustrine process (Singh & Singh, 1994). The state is considered to be the meeting place of the eastern Himalaya and the Burmese region. The state is drained by two majour river systems viz. the Barak-Surma-Meghna system and the Chindwin-Irrawady system (Figure 1).

The Barak-Surma-Meghna system

The Barak-Surma-Meghna River system drains the western half of Manipur. It occupies 50% of the state total catchment area (Vishwanath, 2000). It arises in the southern spurs of Mt. Japvo at Liyai-Khunou in Senapati District. It follows a south westerly course passing through National Highway 37 (Imphal-Dimapur road) at Karong then turns rowards the northwest corner of the state from where flows southwards on the eastern side of the Vangai range and then makes a U-turn at Tipaimukh where it is joined by the Tuivai River which originates from the Chin Hills in Chin State in the northwestern Myanmar, flowing westward between Manipur and Mizoram. The Barak River then flows northward on the western side of the Vangai range and then enter the Cachar district of Assam and finally enter Bangladesh to join the Surma which in turn joins the Meghana near Bhairab Bazar forming the Barak-Surma-Meghna drainage system.

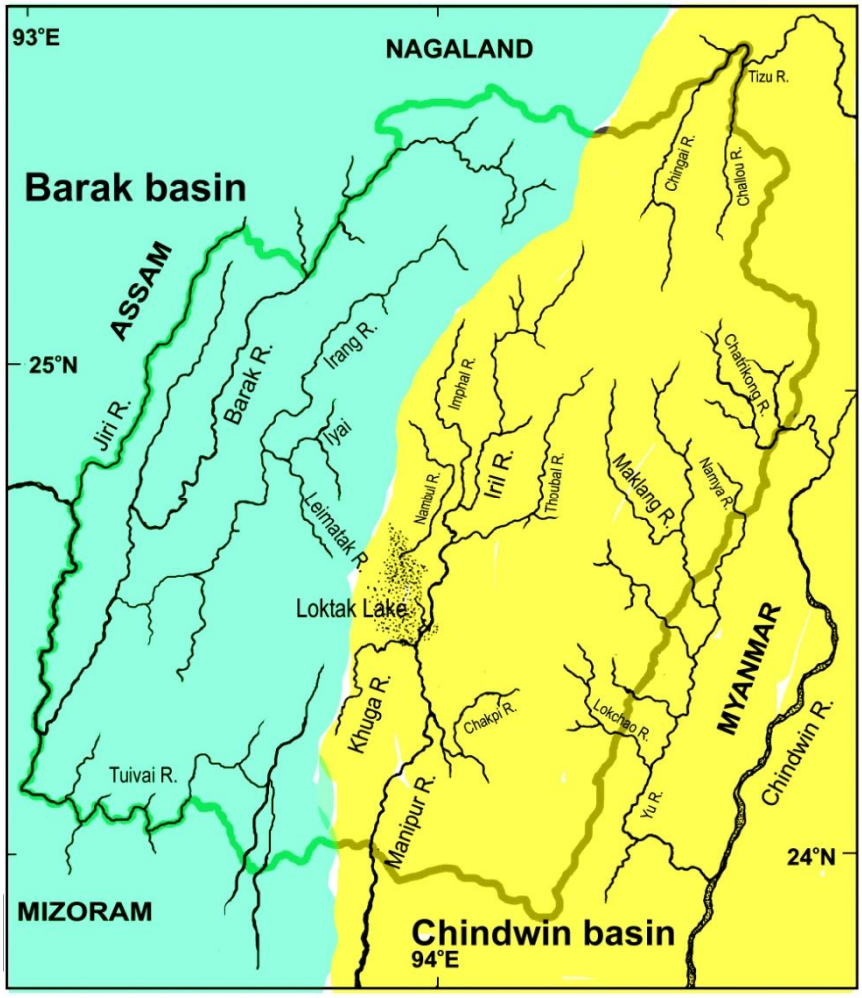

Figure-1: A map showing two river basin in Manipur with its tributaries.

The Chindwin-Irrawady system

The Chindwin-Irrawady system of Manipur consists of the Imphal River and its tributaries, the lakes and marshes lying in the valley and the tributaries of Yu River in Ukhrul, Tengnoupal and Chandel districts which drain into the Chindwin River of Myanmar. It covers over 24000 sq. km in the eastern margin of northeast India, with only about 2200 km^2 of plain land represented by the Manipur basin. The four important tributaries through which this drainage has been set up are the Tizu, Chatrickong, Yu and

Manipur river system. The rivers Tizu, Chatrickong and Yu retain steep hilly character while the main headstream of Manipur River, viz., Iril, Imphal, Thoubal have graded course over the basin.

The Manipur river complex encompasses an area of approximately 7,000 sq. km in the state. It drains the central and southern parts of Manipur including the low lying lakes and marshes lying in the valley. The Imphal River, also known as Turrel Achouba (Principal River) originates from the north of Kangpokpi and flows southward to the eastern side of the Loktak Lake. The principal tributaries of the river in the valley are the Iril, Thoubal, Nambul and Nambol. The Iril and Thoubal rise in the eastern side and meet the Imphal River. The Heirok and Sekmai are small brooks which join the Imphal on the northern sides of the Ikop pat and Kharung pat respectively. The Nambul and Nambol rises in the western hills and discharge into Loktak Lake. The Imphal do not fall into the Loktak, but receives the outflow of the lake and forms a short course called the Khordak. Another stream called the Khuga originates in the hills of Churachandpur District and joins the Imphal at Ithai from south. Now the bigger river is called the Manipur River. It makes its way towards the south. At Sherou, another river called Chakpi, originates from the Tengnoupal areas of Chandel District, and joins the Manipur River from the eastern side. The principal river then follows a southward course in the boundary line between the Churachandpur and Chandel Districts leaving north east India to meet Myittha, a tributary of Chindwin in Myanmar.

The Tizu River forms an important drainage system. It drains the south eastern part of Nagaland and north eastern part of Manipur. It originates from the central part of the state and runs through a northeast direction flows through Zunheboto, Phek Districts and empties itself in the Chindwin River of Myanmar. In Manipur, the northern part of the Ukhrul District is drained by the Tizu River and its tributaries at an altitude of about 500–2000 m asl. The important tributaries of tizu include the Challou or Chingai or Chammu and Laniye. Both the rivers originate from the Sirohi hills. Laniye meets the Zerry lok, which comes from the eastern side of Senapati District and flows north eastward meet Tizu near Jessami (116 kms of north east Ukhrul town). Challou River originates from the northern watersheds of Sirohi hills, joins by the Riori River and then flows northward towards Tizu. The Tizu meets the Challou River at Akash-bridge forming a boundary between Manipur and Nagaland.

The Chatrickong River is one of the important water heads of Chindwin river basin in Manipur which originates from the Khunukhong stream from the western part of the Shiroi peak. It flows south east at the foothills of the highest peak Khayangphung (2833 m asl) of Ukhrul District near the indo-Myanmar border. The stream flows southward for about 100 km through villages' viz., Khayang Chamu, Chatric and flows into the river Khunukong at a place Dha-ado. The river after receiving the two streams become bigger and flows as Chatrickong for about 5 km east and enters inside Myanmar directly to falls into the mighty Chindwin.

The Yu River system lies extending between latitudes 25°15′ and 25°45′N and longitudes 94°14′ and 94′30′E. The Yu with its headwaters drains the eastern margin of Manipur and then flows south meeting the Chindwin finally. Some of the major tributaries of Yu include the Maklang, Sanalok, Namya, Taret, Lokchao, Dutah and other eastward flowing small streams of Ukhrul and Chandel districts at an altitude of about 50–1000m asl.

Ichthyofaunal Inventory

The studies on the Indian freshwater fish fauna started with a more scientific bent and focus on the needs of modern taxonomy only from the 19th century. Beginning with Hamilton's (1762–1829) account on the fishes of Ganges (1822), followed by Heinrich Kuhl (1796–1821), John M′Clelland (1839), col. W. sykes (1841), T.C Jerdon (1842), Blyth (1858, 1860), the vast array of fish found in this area came to light. All these pioneer researchers laid a solid foundation for Indian systematic ichthyology. Francis Day, though an assistant surgeon in the service of the east India company, brought out for the first time, the monumental treaties ″Fish Fauna″ (1875–1878) embodying his own extensive observations and the results of the earlier workers. Day included in his work (including the supplement), 1418 species found within the boundaries of present India, Pakistan (including Afghanistan), Bangladesh, Myanmar and Sri Lanka. Besides the text, a separate volume of illustrations with 195 plates was also issued. Though Day's work had its own limitations, his monograph is irreplaceable even today considering the wide coverage and compactness.

Albert Günther's (1857–70) catalogue of fishes in the British Museum is also worthy to mention. It includes a number of taxa of the region but without many illustrations as Day has done.

The 20th century placed Indian ichthyology on a universal pedestal through indomitable researches of the late Dr. Sunder Lal Hora (1920–1955). For any student of Indian ichthyology the very first source of reference and further basic information, the publications of Hora are indispensible. Hora in his lifetime published 440 papers and established 3 families, 28 genera (including subgenera) and 139 species, many of which are still valid (Jayaram, 1976). Hora's associate Dev Dev Mukerji in his short span of life made several noteworthy contributions to the Indian and Burmese fish fauna. In a similar manner the late K. S Misra (1949, 1953 & 1954) published a series of checklists and aids to the identification of the common commercial fishes of India and Pakistan. Talwar & Jhingran (1999) present more recent compilations for India and Burma. Sen (1985) compiled monograph of the fishes of northeast states of India and in 2000, she had reported a total of 267 species. Vishwanath *et al.* (2014) reported 357 species of 120 genera and 35 families from north east India.

In the core of the nineteenth century, Pemberton (1835) wrote on the fishes of Manipur state and compared with those of Bengal. He noted 18 common varieties. The earliest ever scientific publication on the fishes of Manipur was that of Chaudhuri (1912), who described two new species from hill streams of Ukhrul District viz., *Danio naganensis* (now *Devario nagaensis*) and *Nemacheilus manipurensis* (now *Schistura manipurensis*). Hora (1921) reported a comprehensive account on the fish and fisheries of Manipur including descriptions of new species, viz., *Barilius dogarsinghi* (now *Opsarius dogarsinghi*), *Lepidocephalichthys irrorata*, *Nemacheilus kanjupkhulensis*, *N. prasadi* (now *Physoschistura prasadi), N. sikmaiensis* (all *Nemacheilus* now *Schistura*). He also remarked that Rev. William Pettigrew collected the types of these fishes in 1910. His reports included some part of Nagaland also. The report provided a lists of 56 fishes which was collected from four localities, viz., i) Loktak Lake, ii) Sluggish streams in the Manipur Valley, iii) streams with rocky bed in the southern watershed and iv) streams with rocky beds in the northern watersheds of the Naga hills. The northern watershed of the Naga hills as referred in his report covered only small streams at various points on the road between the Manipur Valley and Dimapur (Nagaland) which belongs to the Brahmaputra drainage (Dayang valley) and Chindwin drainage (Tizu valley) of Nagaland. Later Hora & Mukerji (1935) reported 44 species of fishes from the Naga Hills (hill areas of present Manipur and Nagaland), sorted fishes of Brahmaputra and Chindwin basins and described *Psilorhynchus homalptera*. Hora (1936) then reported fishes from the Barak River and its tributaries of Naga Hills, Manipur. Based on collections made by Mr. S. J. Duncan, Sub-Divisional Officer, Hora (1937) further reported fishes from Chindwin drainage viz., Khunukhong, Chahong Khullen, Namya, Lokchao and Chakpi rivers of Ukhrul and Chandel Districts respectively. Menon (1952) listed 19 species collected from the following areas: i) Kanglatombi, and ii) Kanglatombi bazaar, in the Imphal–Dimapur road, about 26 and 19 km north of Imphal, iii) Imphal market, iv) water pools and streams in the Imphal-pallel road between 29.0 and 35.4 km south of Imphal. Later, Menon (1954) made extensive collections from the Loktak Lake, Imphal River and the Barak River at different localities. Vishwanath & Tombi (1985) provided lists of 35 fishes including some new records from Chandel District. Vishwanath & Tombi (1986) remarked that fishes of the further deeper hill areas are yet to be explored. Description of several new taxa, revision of certain genera and families were carried out by subsequent workers eg. Manojkumar & Vishwanath (1996) on the Manipur River and its system, Selim & Vishwanath (1997) on the Chatrick River of Ukhrul District, Kosygin & Vishwanath (1997) on the Tizu River, Vishwanath *et al*., (1998) reported 116 species of 67 genera and 25 families from the Chindwin basin of the state, Nebeshwar (2002) on the Barak River system of the state, Vishwanath (2000) reported 165 species of 84 genera and 27 families from Manipur, Linthoingambi & Vishwanath (2005) on the Iril River.

Some of the recent scientific publications on the description of new species from the Chindwin Basin in Manipur includes *Garra elongata* (Viswanath & Kosygin, 2000), *Opsarius chatricensis*, *O. ngawa* (Vishwanath & Selim 2002, Viswanath & Manojkumar, 2002), *Pethia meingangbii* (Arunkumar & Tombi, 2003), *Pethia ornata, Rasbora ornata, Garra nambulica* (Vishwanath & Laisram, 2004a, 2004b, 2005), *Schistura khugae, Garra paralissorhynchus* (Vishwanath & Shanta, 2004, 2005), *Schistura reticulata* (Vishwanath & Nebeshwar, 2004), *Glyptothorax granulus, G. ngapang, Pethia atra, P. khugae, Amblyceps*

tuberculatum, *A. torrentis* (Linthoingambi & Vishwanath 2007a, 2007b, 2007c, 2007d, 2008a, 2008b), *Physoschistura chindwinica*, *P. trigrina* (Lokeshwar & Vishwanath, 2002a, 2002b), *Garra namyaensis*, *Psilorhynchus chakpiensis*, *P. maculatus*, *P. ngathanu*, (Shangningam & Vishwanath, 2014), *Schistura phamhringi* (Shangningam, Lokeshwor & Vishwanath, 2014), *Garra chakpiensis, G. cornigera*, *G. triloba*, *G. ukhrulensis, Pethia lepidomaculata, Devario deruptotalea*.

In the later part of the 20th century and in the beginning of the 21st century, scientists from all over the world have focused their research on the ichthyofaunal exploration in south-east Asia. Many workers from different parts of the world are also interested in the fish diversity of the state. But owing to difficult hilly terrain, transport problems and security reasons, they are not freely accessible to the fish specimen of the place. However, the data on the fishes, especially those in the Chindwin basin are very important to fill up the gaps and establish phylogeny of several freshwater fish taxa of the region. Abell *et al*., (2008) noted that there are more endemic species in the Sittaung-Irrawaddy region than in any of the Ganga-Brahmaputra subregions. According to Hora (1921) the endemic element is chiefly confined to the hill streams and strictly speaking is isolated. He also remarked that the Chindwin fauna consists of Burmese and endemic Manipur elements. The variety of torrential hill streams, rivers and lake support rich fish diversity and the degree of endemism though not fully known is expected to be high in the upland streams (Vishwanath, 2000). The fauna of rapids is known to have a very high rate of endemism, but it is one of the least well known habitats. Hundreds of species probably still await discovery. For obvious reasons, sampling of these habitats can be difficult and dangerous, and they are under sampled in most countries (Kottelat & Whitten, 1996).

Ichthyofaunal Diversity of Manipur

The fish fauna of Manipur comprises representatives of the both Assamese and Burmese elements (Hora & Mukerji, 1935). The distinctive piscine fauna of Manipur is attributed to: the drainage pattern comprised of two entirely different river systems viz., Barak drainage system and the Chindwin–Irrawaddy system; unique mode of its geological formation and the formation of a geographically isolated central valley at a high altitude; unique climatic factors like variations in temperature, rainfall, elevations, etc.

The Barak drainage system drains the western of Manipur and occupies 50% of the state total catchment area (Vishwanath, 2000). The Barak arises from Lai Leeyai area of Senapati District. Some of the important tributaries include Irang, Jiri, Makru and Tuivai.

Fishes are of immense value to human communities since the dawn of civilization. Freshwater biodiversity is an important issue with tremendous economic, social, and environmental impacts. It impinges on human welfare in terms of food, nutrition, other resources, purification of water, recreation, health of aquatic ecosystem, and control of infectious organisms. Most attention has focused on fish as food because of their overwhelming economic importance.

Hora (1921) and Vishwanath (2000) has mentioned the importance of fish in Manipur. Being easy to catch without much sophisticated gear is a common avocation for many in the villages and towns of Manipur. There is hardly any body of water which does not have some variety or other of fish. As such the biodiversity within freshwater ecosystem in Manipur is both high and of great regional importance to livelihoods and economies. However, the State Fisheries Department has given no prominence on the exploration of state's extremely rich and large endemic ichthyofauna. However, importance has been given to the culture and production of the high yielding exotic varieties.

At present, Manipur is blessed with 218 species of fishes belong to 92 genera under 32 families and 11 orders (Table 1). Fifity five species of fish have been discovered in the last two decades. Cypriniformes forms the largest order with 127 species followed by Siluriformes with 56 species, Perciformes with 19 species and Synbranchiformes with six species. Order Osteoglossiformes, Clupeiformes and Cypriodontiformes are represented by two species each. Order Anguilliformes, Mugiliformes, Beloniformes and Tetraodontiformes are with a single species each. Family Cyprinidae under order Cypriniformes represents maximum number of species with 65 species under 30 genera (Table 2).

Table 1. Systematic index of the Fishes of Manipur showing their distribution in the two river basins. Symbol + represents present – Absent. EN-Endangered; VU-Vulnerable; NT- Near threatened; LC-Least concern; DD-Data deficient; NE-Not evaluated; BR-Barak River basin; CR-Chindwin River basin; IUCN-Inrernatoinal Union of Conservation of Nature and Natural resources status

Fish	BR	CR	IUCN
Phylum **CHORDATA**			
Subphylum **VERTEBRATE (CRANIATA)**			
Super class **GNATHOSTOMATA**			
Class **ACTINOPTERYGII**			
Division **TELEOSTEI**			
Subdivision ELOPOMORPHA			
Order ANGUILLIFORMES			
Family ANGUILLIDAE Rafinesque			
1. Genus *Anguilla* Garsault			
1. *A. bengalensis* (Gray)	+	+	LC
Subdivision OSTEOGLOSSOMORPHA			
Order OSTEOGLOSSIFORMES			
Family NOTOPTERIDAE Bleeker			
2. Genus *Chitala* Flower			
2. *C. chitala* Hamilton	+	–	NT
3. Genus *Notopterus* La Cepède			
3. *N. notopterus* (Pallas)	+	+	LC
Subdivision CLUPEOMORPHA			
Order CLUPEIFORMES			
Family CLUPEIDAE Flower			
4. Genus *Tenualosa* Fowler			
4. *T. ilisha* (Hamilton)	+	–	LC
5. Genus *Gudusia* Fowler			
5. *G. chapra* (Hamilton)	+	–	LC
Subdivision OSTARIOCLUPEOMORPHA			
Order CYPRINIFORMES			
Family CYPRINIDAE Rafinesque			
Subfamily Cyprininae			
6. Genus *Chagunius* Smith			
6. *C. chagunio* (Hamilton)	+	–	LC
7. *C. nicholsi* (Myers)	–	+	LC
7. Genus *Cyprinus* Linnaeus			
8. *C. carpio* Linnaeus	+	+	NE
8. Genus *Gibelion* Haekel			
9. *G. catla* (Hamilton)	+	+	LC
9. Genus *Neolissochilus* Rainboth			
10. *N. hexagonolepis* (McClelland)	+	–	NT
11. *N. hexastichus* (McClelland)	+	–	NT
12. *N. stracheyi* (Day)	–	+	LC
10. Genus *Osteobrama* Heckel			
13. *O. belangeri* (Valencienes)	–	+	NT
14. *O. cotio* (Hamilton)	+	–	LC
15. *O. cunma* (Day)	–	+	LC
16. *O. feae* Vinciguerra	–	+	LC
11. Genus *Systomus* McClelland			
17. *S. burtoni* (Mukerji)	–	+	LC
18. *S. clavatus* (McClelland)	+	–	NT

19. *S. sarana* (Hamilton)	+	+	LC
12. Genus *Semiplotus* Bleeker			
20. *S. corrhosus* Chaudhuri	+	–	DD
21. *S. semiplotus* (McClelland)	+	+	VU
13. Genus *Tor* Gray			
22. *T. mosal* (Hamilton)	+	–	NE
23. *T. progenius* McClelland	+	–	NT
24. *T. putitora* (Hamilton)	+	–	EN
25. *T. tor* (Hamilton)	+	–	NT
Sub family Barbinae			
14. Genus *Hypsibarbus* Rainboth			
26. *H. myitkyinae* (Prasad & Mukerji)	–	+	LC
15. Genus *Pethia* Pethiyagoda *et al.*			
27. *P. atra* (Linthoi. & Vish.)	–	+	VU
28. *P. conchonius* (Hamilton)	+	–	LC
29. *P. khugae* (Linthoi. & Vish.)	–	+	VU
30. *P. manipurensis* (Menon *et al*.)	–	+	EN
31. *P. ornata* (Vish. & Juliana)	–	+	VU
32. *P. stoliczkana* (Day)	+	–	DD
33. *P. meingangbii* (Arunkumar & Tombi)	–	+	NE
34. *P. yuensis* (Arunkumar & Tombi)	–	+	VU
16. Genus *Puntius* Hamilton			
35. *P. chola* (Hamilton)			
36. *P. sophore* (Hamilton)	+	+	LC
17. Genus *Schizothorax* Heckel	+	+	LC
37. *S. chivae* Arunkumar & Alphonsa	–	+	NE
Subfamily Labeoninae			
18. Genus *Bangana* Hamilton			
38. *B. dero* (Hamilton)	+	–	LC
39. *B. devdevi* (Hora)	–	+	LC
19. Genus *Cirrhinus* Oken			
40. *C. cirrhosus* (Bloch)	+	+	VU
20. Genus *Garra* Hamilton			
41. *G. abhoyai* Hora	–	+	NE
42. *G. chakpiensis* Nebesh. & Vish.	–	+	NE
43. *G. compressa* Kosygin & Vish.	–	+	VU
44. *G. cornigera* Shang. & Vish.	–	+	NE
45. *G. elongata* Vish. & Kosygin	–	+	NT
46. *G. gravelyi* (Annandale)	–	+	NT
47. *G. lissorhynchus* (McClalland)	+	–	LC
48. *G. litanensis* Vishwanath	–	+	VU
49. *G. manipurensis* Vish. & Sarojnalini	–	+	VU
50. *G. naganensis* Hora	+	–	VU
51. *G. nambulica* Vish. & Joyshree	–	+	VU
52. *G. namyaensis* Shang. & Vish.	–	+	NE
53. *G. nasuta* (McClelland)	+	–	LC
54. *G. paralissorhynchus* Vish. & Shanta	–	+	VU
55. *G. ukhrulensis* Nebesh. & Vish.	–	+	NE
21. Genus *Gymnostomus* Heckel			
56. *G. ariza* (Hamilton)	+	–	LC
22. Genus *Labeo* Curvier			
57. *L. bata* (Hamilton)	+	–	LC
58. *L. boga* (Hamilton)	+	–	LC

59. *L. calbasu* (Hamilton)	+	+	NE
60. *L. dyocheilus* (M'Clelland)	+	+	LC
61. *L. gonius* (Hamilton)	+	–	LC
62. *L. pangusia* (Hamilton)	+	+	NT
63. *L. rohita* (Hamilton)	+	+	LC
23. Genus *Tariqilabeo* Mirza & Saboohi			
64. *T. burmanicus* (Hora)	–	+	NE
65. *T. latius* (Hammilton)	+	–	LC
Subfamily Squaliobarbinae			
24. Genus *Ctenopharhyngodon* Steindachner			
66. *C. idella* (Valenciennes)	+	+	NE
Subfamily Xenocyprinae			
25. Genus *Hypopthalmichthys* Bleeker			
67. *H. molitrix* (Valenciennes)	+	+	NE
68. *H. nobilis* (Richardson)	+	+	NE
Subfamily Rasborinae (Danioninae)			
26. Genus *Amblypharyngodon* Bleeker			
69. *A. mola* (Hamilton)	+	+	LC
27. Genus *Barilius* Hamilton			
70. *B. barila* (Hamilton)	+	–	LC
71. *B. bendelisis* (Hamilton)	+	–	LC
72. *B. lairokensis* Arunkumar & Tombi	–	+	NT
28. Genus *Cabdio* Hamilton			
73. *C. morar* (Hamilton)	+	–	LC
74. *C. ukhrulensis* (Selim & Vishwanath)	–	+	NE
29. Genus *Danio* Hamilton			
75. *D. dangila* (Hamilton)			
76. *D. quagga* Kullander	+	–	LC
30. Genus *Devario* Heckel	–	+	NT
77. *D. acuticephala* (Hora)			
78. *D. aequipinnatus* (McClelland)	–	+	VU
79. *D. naganensis* (Chaudhuri)	+	–	LC
80. *D. yuensis* (Arunkumar & Tombi)	–	+	VU
31. Genus *Esomus* Swainson	–	+	VU
81. *E. danrica* (Hamilton)			
32. Genus *Laubuka* Bleeker	+	+	LC
82. *L. laubuca* (Hamilton)			
33. Genus *Opsarius* M′Clelland	+	–	LC
83. *O. barnoides* (Vinciguerra)			
84. *O. dogarsinghi* (Hora)	–	+	LC
34. Genus *Raiamas* Jordan	–	+	VU
85. *R. guttatus* (Day)			
35. Genus *Rasbora* Bleeker	+	–	LC
86. *R. daniconius* (Hamilton)			
87. *R. ornata* Vish. & Laishram	+	–	LC
36. Genus *Salmostoma* Swainson	–	+	VU
88. *S. phulo* (Hamilton)	+	–	LC
89. *S. sladoni* (Day)	–	+	LC
Family PSILORHYNCHIDAE Hora			
37. Genus *Psilorhynchus* M'Clelland			
90. *P. amplicephalus* Arunacha. et al.	+	–	DD
91. *P. balitora* (Hamilton)	+	–	LC
92. *P. breviminor* Conway & Mayden	–	+	DD

93. *P. chakpiensis* Shang. & Vish.	–	+	NE
94. *P. homaloptera* Hora & Mukherji	+	–	LC
95. *P. maculatus* Shang. & Vish.	–	+	NE
96. *P. microphthalmus* Vish. & Manoj.	–	+	EN
97. *P. ngathanu* Shang. & Vish.	–	+	NE
98. *P. rowleyi* (Hora & Misra)	–	+	NE
Family BOTIIDAE Berg			
38. Genus *Botia* Gray			
99. *B. almorahae* Gray	+	–	LC
100. *B. dario* (Hamilton)	+	–	LC
101. *B. histrionica* Blyth	–	+	LC
39. Genus *Canthophyrs* Swainson			
102. *C. gongata* (Hamilton)	+	–	LC
40. Genus *Synchrossus* Blyth			
103. *S. berdmorei* (Blyth)	–	+	NT
Family COBITIDAE Fitzinger			
41. Genus *Acantopsis* van Hasselt			
104. *A. spectabilis* (Blyth)	–	+	NT
42. Genus *Lepidocephalichthys* Bleeker			
105. *L. berdmorei* (Blyth)	–	+	LC
106. *L. guntea* (Hamilton)	+	–	LC
107. *L. irrorata* Hora	–	+	NE
43. Genus *Pangio* Blyth			
108. *P. pangia* (Hamilton)	–	+	LC
Family BALITORIDAE Swainson			
44. Genus *Balitora* Gray			
109. *B. brucei* Gray	+	–	NT
110. *B. burmanica* (Hora)	–	+	LC
45. Genus *Homalopteroids* Fowler			
111. *H. rupicola* (Prashad & Mukherji)	–	+	LC
Family NEMACHEILIDAE Regan			
46. Genus *Paracanthocobitis* Grant			
112. *P. botia* (Hamilton)	+	–	LC
113. *P. zonalternans* (Blyth)	–	+	LC
47. Genus *Neonoemacheilus* Zhu & Guo			
114. *N. morehensis* Arunkumar	–	+	DD
115. *N. peguensis* (Hora)	–	+	DD
48. Genus *Physoschistura* Banarescu & Nalba.			
116. *P. prasadi* (Hora)	–	+	VU
117. *P. chindwinensis* Loke. & Vish.	–	+	NE
118. *P. trigrina* Lokeshwor & Vish.	–	+	NE
119. *P. tuivaiensis* Lokeshwor et al.	+	–	NE
49. Genus *Schistura* M′Clelland			
120. *S. chindwinica* (Tilak & Hussain)	+	–	LC
121. *S. fasciata* Lokeshwor & Vish.	+	–	NE
122. *S. ferruginea* Lokeshwor & Vish.	+	–	NE
123. *S. kangjupkhulensis* (Hora)	–	+	EN
124. *S. khugae* Vish. & Shanta	–	+	VU
125. *S. manipurensis* (Chaudhuri)	–	+	NT
126. *S. minutes* Vish. & Shantakumar	+	+	EN
127. *S. nagaensis* (Menon)	–	+	VU
128. *S. phamhringi* Shangningam *et al.*	–	+	NE
129. *S. reticulata* Vish. & Nebeshwar	–	+	EN

130. *S. sikmaiensis* (Hora)	–	+	LC
131. *S. singhi* (Menon)	–	+	VU
132. *S. tigrinum* Vish. & Nebeshwar	+	–	EN
Order SILURIFORMES			
Family AMBLYCIPITIDAE Day			
50. Genus *Amblyceps* Blyth			
133. *A. mangois* (Hamilton)	+	–	LC
134. *A. torrentis* Linthoi. & Vish.	–	+	DD
135. *A. tuberculatum* Linthoi. & Vish.	–	+	DD
Family AKYSIDAE Gill			
51. Genus *Akysis* Bleeker			
136. *A. manipurensis* (Arunkumar)	–	+	DD
137. *A. prashadi* Hora	–	+	LC
Family SISORIDAE Bleeker			
52. Genus *Bagarius* Bleeker			
138. *B. bagarius* (Hamilton)	+	–	NT
139. *B. yarrelli* (Sykes)	–	+	NT
53. Genus *Erethistes* Müller & Troschel			
140. *E. hara* (Hamilton)	+	–	LC
141. *E. pussilus* Müller & Troschel	+	–	NE
54. Genus *Exostoma* Blyth			
142. *E. barakensis* Vish. & Joyshree	+	–	DD
55. Genus *Gagata* Bleeker			
143. *G. cenia* (Hamilton)	+	–	LC
144. *G. dolichonema* He	–	+	LC
56. Genus *Gogangra* Robert			
145. *G. viridecsens* (Hammilton)	+	–	LC
57. Genus *Glyptothorax* Blyth			
146. *G. burmanicus* Prasad & Mukerji	–	+	LC
147. *G. cavia* (Hamilton)	+	–	LC
148. *G. clavatus* Rameshori & Vish.	+	–	NE
149. *G. granulus* Vish. & Linthoi.	–	+	LC
150. *G. maceriatus* Ng & Lalramliana	+	–	NE
151. *G. manipurensis* Menon	+	–	VU
152. *G. dorsalis* Vinciguerra	+	–	LC
153. *G. ngapang* Vish. & Linthoi.	–	+	LC
154. *G. scrobiculus* Ng & Lalramliana	+	–	NE
155. *G. senapatiensis* Premananda et al.	+	–	NE
156. *G. ventrolineatus* Vish. & Linthoi.	–	+	LC
58. Genus *Myersglanis* Hora & Silas			
157. *M. jayarami* Vish. & Kosygin	–	+	VU
59. Genus *Pseudocheneis* Blyth			
158. *P. ukhrulensis* Vish. & Darshan	–	+	VU
60. Genus *Sisor* Hamilton			
159. *S. barakensis* Vish. & Darshan	+	–	VU
160. *S. rabdophorus* (Hamilton)	+	–	LC
Family SILURIDAE Cuvier			
61. Genus *Ompok* La Cepède			
161. *O. bimaculatus* (Bloch)	–	+	NT
162. *O. pabda* (Hamilton)	+	–	NT
163. *O. pabo* (Hamilton)	+	–	NT
62. Genus *Pterocryptis* Peters			
164. *P. barakensis* Vish. & Nebeshwar	+	–	EN

165. *P. berdmorei* (Blyth)	–	+	LC
63. Genus *Wallago* Bleeker			
166. *W. attu* (Schneider)	+	+	NT
Family CHACIDAE Bleeker			
64. Genus *Chaca* Gray			
167. *C. chaca* (Hamilton)	+	–	LC
Family CLARIIDAE Bonaparte			
65. Genus *Clarias* Scopoli			
168. *C. gariepinnus* (Burchell)	+	+	NE
169. *C. magur* (Hamilton)	+	+	EN
66. Genus *Heteropneustes* Muller			
170. *H. fossilis* (Bloch)	+	+	LC
Family SCHILBEIDAE Bleeker			
67. Genus *Ailia* Gray			
171. *A. coila* (Hamilton)	+	–	NT
68. Genus *Eutropiichthys* Bleeker			
172. *E. burmanicus* Day	–	+	NE
173. *E. murius* (Hamilton)	+	–	LC
174. *E. vacha* (Hamilton)	+	–	LC
Family BAGRIDAE Bleeker			
Subfamily Bagrinae			
69. Genus *Hemibagrus* Bleeker			
175. *H. microphthalmus* (Day)	–	+	LC
176. *H. peguensis* (Boulenger)	–	+	LC
70. Genus *Mystus* Scopoli			
177. *M. bleekeri* (Day)	+	–	LC
178. *M. cavasius* (Hamilton)	+	–	LC
179. *M. falcarius* Chakrabarty & Ng	–	+	LC
180. *M. ngasep* Darshan, Vish. & Mahan.	–	+	NE
181. *M. pulcher* (Chaudhuri)	–	+	LC
182. *M. rufescens* (Vinciguerra)	–	+	LC
183. *M. tengara* (Hamilton)	+	–	LC
71. Genus *Sperata* Holly			
184. *S. acicularis* Ferraris & Runge	+	–	LC
185. *S. aor* (Hamilton)	–	+	LC
Subfamily Batasinae			
72. Genus *Batasio* Blyth			
186. *B. affinis* Blyth	–	+	NE
Subfamily Olyrinae			
73. Genus *Olyra* M′Clelland			
187. *O. horae* (Prashad & Mukerji)	–	+	DD
Subfamily Ritinae			
74. Genus *Rita* Bleeker			
188. *R. rita* (Hailton)	+	–	LC
Series MUGILIMORPHA			
Order MUGILIFORMES			
Family Mugilidae Cuvier			
75. Genus *Sicamugil* Fowler			
189. *S. cascasia* (Hailton)	+	–	LC
Subdivision EUTELEOSTEI			
Superorder ACANTHOPTERYGII			
Series ATHERINOMORPHA			
Order BELONIFORMES			

Family BELONIDAE Collette			
76. Genus *Xenentodon* Regan			
190. *X. cancila* (Hamilton)	+	+	LC
Order CYPRINODONTIFORMES			
Family APLOCHEILIDAE M′Clelland			
77. Genus *Aplocheilus* M'Clelland			
191. *A. panchax* Hamilton	+	+	LC
Family POECILIIDAE Garman			
78. Genus *Gambusia* Poey			
192. *G. affinis* (Baird & Girard)	+	+	LC
Series PERCOMORPHA			
Order SYNBRANCHIFORMES			
Family SYNBRANCHIDAE Swainson			
79. Genus *Monopterus* La Cepède			
193. *M. cuchia* (Hamilton)	+	–	LC
194. *M. javanensis* La Cepède	–	+	LC
Family MASTACEMBELIDAE Swainson			
80. Genus *Macrognathus* La Cepède			
195. *M. aral* (Schneider)	+	–	LC
196. *M. morehensis* Arunkumar & Tombi	–	+	LC
197. *M. pancalus* Hamilton	+	–	LC
81. Genus *Mastacembelus* Scopoli			
198. *M. armatus* (La Cepède)	+	+	LC
Order PERCIFORMES			
Family AMBASSIDAE Khunzinger			
82. Genus *Chanda* Hamilton			
199. *C. nama* Hamilton	+	+	LC
83. Genus *Johnius* Bloch			
200. *J. coitor* (Hamilton)	+	–	NE
84. Genus *Parambassis* Bleeker			
201. *P. lala* (Hamilton)	+	–	NT
202. *P. ranga* (Hamilton)	+	–	NE
203. *P. waikhomi* Geetaku. & Basudha	–	+	NE
Family BADIDAE Barlow, Liem &Wickler			
Subfamily Badinae			
85. Genus *Badis* Bleeker			
204. *B. badis* (Hamilton)	+	–	LC
205. *B. tuivaiei* Vish. & Shanta	+	–	EN
Family NANDIDAE Bleeker			
86. Genus *Nandus* Valenciennes			
206. *N. nandus* (Hamilton)	+	–	LC
Family CICHLIDAE Bonaparte			
87. Genus *Oreochromis* Günther			
207. *O. mossambica* (Peters)	+	+	NE
Family GOBIIDAE Cuvier			
88. Genus *Glossogobius* Gill			
208. *G. giuris* (Hamilton)	+	+	NE
Family ANABANTIDAE Bonaparte			
89. Genus *Anabas* Cloquet			
209. *A. testudineus* (Bloch)	+	+	DD
Family OSPHRONEMIDAE van der Hoeven			
90. Genus *Trichogaster* Bloch in Schneider			
210. *T. chuna* (Hamilton)	+	–	LC

211. *T. fasciata* Blotch	+	+	LC
212. *T. labiosa* Day	+	+	LC
Family CHANNIDAE Fowler			
91. Genus *Channa* Scopoli			
213. *C. gachua* (Hamilton)	+	+	LC
214. *C. marulius* (Hamilton)	+	+	LC
215. *C. punctata* (Bloch)	+	+	LC
216. *C. stewartii* (Playfair)	+	–	LC
217. *C. striata* (Bloch)	+	+	LC
Order TETRAODONTIFORMES			
Family TETRAODONTIDAE Linnaeus			
92. Genus *Leiodon* Swainson			
218. *L. cutcutia* (Hamilton)	+	+	LC

Table-2: Distribution table of fish species of Manipur with respective orders and number of represented Families, Genera and Species.

Order	Families	Genera	Species
ANGUILLIFORMES	1	1	1
OSTEOGLOSSIFORMES	1	2	2
CLUPEIFORMES	1	2	2
CYPRINIFORMES	6	36	127
SILURIFORMES	8	25	56
MUGILIFORMES	1	1	1
BELONIFORMES	1	1	1
CYPRINODONTIFORMES	2	2	2
SYNBRANCHIFORMES	2	3	6
PERCIFORMES	8	10	19
TETRAODONTIFORMES	1	1	1
Total	34	84	218

Status of Fishes in Manipur

According to the IUCN redlist assessment of eastern Himalayas (2010), the majority of threatened fishes are in the Chindwin Basin in Manipur. Many of the species endemic to the Chindwin basin in Manipur are assessed as Endangered, Vulnerable and near threatened. There are 11 species of fishes which under Endangered category in which Six species are from the Barak River and five from the Chindwind River in Manipur (Table 3 & 4; Plate I). The Chindwin River basin is represented with 25% of threatened species inwhich 21% of species are under Vulnerable category. The Barak River basin is with 10% threatened species in which 5% are under vulnerable category (Figure 3). The species under least concern are more in the Barak River with 66% of species than the Chindwin River with only 38% of species. The remaining species are at data deficient caregory.

Table-3: IUCN Status of Fishes of the Barak River basin

Endangered species (EN):

1. *Tor putitora* (Hamilton)
2. *Schistura minuta* Vish. & Shantakumar
3. *S. tigrinum* Vish. & Nebeshw.
4. *Clarias magur* (Hamilton)
5. *Badis tuivaiei* Vish. & Shanta
6. *Pterocryptis barakensis* Vish. & Nebeshw.

Vulnerable species (VU):

1. *Semiplotus semiplotus* (McClelland)
2. *Cirrhinus cirrhosus* (Blotch)
3. *Garra nagaensis* Hora
4. *Schistura chindwinica* (Tilak & Hussain)
5. *Glyptothorax manipurensis* Menon
6. *Sisor barakensis* Vish. & Darshan

Near Threatened species (NT):

1. *Chitala chitala* Hamilton
2. *Neolissochilus hexagonolepis* (McClelland)
3. *Neolissochilus hexasticus* (McClelland)
4. *Systomus clavatus* (McClelland)
5. *Tor progenius* McClelland
6. *Tor tor* (Hamilton)
7. *Labeo pangasia* (Hamilton)
8. *Balitora brucei* Gray
9. *Bagarius bagarius* (Hamilton)
10. *Ompok pabda* (Hamilton)
11. *Ompok pabo* (Hamilton)
12. *Wallago attu* (Schneider)

Table-4: IUCN Status of Fishes of the Chindwin River Basin.

Endangered species

1. *Clarias magur* Hamilton
2. *Pethia manipurensis* (Menon *et al.*)
3. *Psilorhynchus microphthalmus* Vish. & Manoj.
4. *Schistura kangjupkhulensis* (Hora)
5. *Schistua reticulata* Vish. & Nebes.

Vulnerable species

1. *Devario acuticephalus* (Hora)
2. *D. naganensis* (Menon)
3. *D. yuensis* (Arunkumar & Tombi)
4. *Garra compressa* Kosygin & Vish.
5. *G. litanensis* (Vish.)
6. *G. nambulica* (Vish. & Joyshree)
7. *G. paralissorhynchus* Vish. & Shanta
8. *Laubuka laubuca* (Hamilton)
9. *Myerglanis jayarami* Vish. & Kosygin
10. *Opsarius dogarsinghi* (Hora)
11. *O. barnoides* (Vinciguerra)
12. *Pseudecheneis ukhrulensis* Vish. & Darshan
13. *Pethia atra* (Linthoingambi & Vish.)
14. *P. khugae* (Shanta & Vishwanath)
15. *P. ornata* (Vish. & Juliana)
16. *P. yuensis* (Arunkumar)
17. *Physoschistura prasadi* (Hora)
18. *Rasbora ornata* Vish. & Juliana
19. *Schistura khugae* Vish. & Shanta
20. *Schistura nagaensis* (Menon)
21. *Schistura singhi* (Menon)

Near Threatened species:

1. *Bagarius bagarius* (Hamilton)
2. *Bagarius yarrelli* (Sykes)
3. *Barilius lairokensis Arunkumar*
4. *Garra elongata* Vishwanath & Kosygin
5. *Garra gravelyi* (Annandale)
6. *Neolissocheilus hexagonolipis* (McClelland)
7. *Ompok bimaculatus* (Bloch)
8. *Ompok pabo* (Hamilton)
9. *Osteobrama belangeri* (Valenciennes)
10. *Schistura manipurensis* (Chaudhuri)
11. *Syncrossus berdmorei* (Blyth)
12. *Tor tor* (Hamilton)
13. *Wallago attu* (Schneider)

Table-5: Exotic Fish species in Manipur

1. *Cyprinus carpio* Linnaeus
2. *Cetenopharhyngodon idella* (Valenciennes)
3. *Hypopthalmichthys molitrix* (Valenciennes)
4. *H. nobilis* (Richardson)
5. *Clarias gariepinnus* (Burchell)
6. *Gambusia affinis* (Baird & Girard)
7. *Piaractus brachypomus (Cuvier)*
8. *Oreochromis mossambica* (Peters)

According to the IUCN (2010), assessment on the Eastern Himalaya, maximum richness and endemicity of fish species are found in the northeast India. The most threatened species are also found in the Chindwin basin in Manipur and the adjoining areas (Vishwanath *et al*., 2010). Of the 15 endangered species, *Pethia manipurensis*, *Schistura kanjupkhulensis* and *Psilorhynchus microphthalmus* are endemic to Manipur Valley while *Schistura reticulata*, is endemic to the Eastern hill streams of Manipur draining into the Yu River.

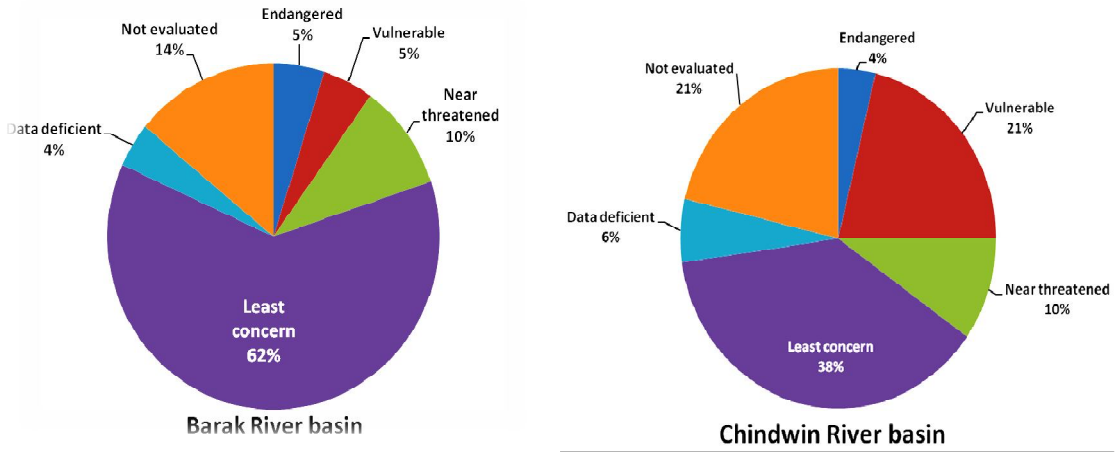

Figure-2: Pie chart showing the percentages of fish under different IUCN threat categories in the two river basin of Manipur.

Many of the species endemic to the Barak River basin of Manipur are *Budis tuvaiei*, *Pterocriptis barakensis*, *Schistura minutus* and *S. tigrinum* (Vishwanath *et al.*, 2010). They are confined to a restricted to upper reaches of this river basin. The proposed Tipaimukh dam across the Barak River in Manipur has the potential to result in the loss of habitat and impact on migratory species, as well as other down stream impacts (Vishwanath *et al.*, 2010). The construction of this dam will cause habitat fragmentation of the distribution of *B. tuivaiei.*

Pethia manipurensis, Schistura reticulata, S. kanjupkhulensis and *Psilorhynchus microphthalmus* are facing habitat loss due to sand and gravel mining for construction and urban development near their habitats (Shangningam, 2014). It is certain that these species does not occur in other tributaries of the Chindwin on the Myanmar side.

Kottelat (2001) remarked that the impacts of introduced species may include the degradation of the host environment, the disruption of the host community, and the genetic degradation of host stock by hybridisation, the introduction of diseases and parasites, and socio economic hardships for surrounding fishing communities. The effect of introduced species of fishes into the natural watershed of Manipur has a

negative effect on indigenous fish community. Hora (1921) reported *Osteobrama belangeri* from Manipur Valley. Menon (1989) referred the species as the Loktak fish of Manipur and reported the fish to be disappearing fast due to the introduction of *Cyprinus carpio* in the Lake. The species is known to migrate from the Ayeyarwaddy to Manipur Valley where they breed and grow. With the construction of the Ithai barrage across the Manipur River for the Loktak hydro-electric project in the early 1980s, the route of the species to the valley has been disrupted. The species listed by Hora (1921) are no longer present. Many weed and introduced fishes, such as Chinese carps and *Oreochromis mossambica* appear to have replaced the endemic species. There are as many as eight exotic fishes have been introduced legally or illegally (Table 5; Plate II). One of the major concerns on the introduction of exotic fishes in the region is *Piaractus brachypomus, commonly called Redbelly Pacu or Penba manbi or rupchanda, illegally in Barak River basin. It is need to be* questioned whether this fishes are a good choice for the typical aquarist. Tough they are not aggressive carnivores like the piranha, their crushing jaw system, used primarily for eating seeds and nuts, can be hazardous. If a large population of pacu enters an ecosystem to which it is not native, the fish can have a very adverse effect. It is now recognized by all authorities that no further introductions should be contemplated anywhere without exhaustive research into the potential effects.

Northeast India is regarded as the country future powerhouse. The proposal of hydroelectric power project in several regions of northeast India by NHPC will create a great pressure to the freshwater biodiversity affecting the fish fauna. Habitats of several hill stream fishes will be disturbed and creat habitat fragmentation of upstream and downstream parts of the river. All major projects that could impact freshwater systems should be subject to an independent and transparent EIA. The extensive development across much of the region, both current and planned, of hydropower and irrigation dams should be analysed by appropriate environmental impact assessments with the consideration of impacts on migratory and commercially valuable fish species, and the environmental flow requirements of all species. The restoration of natural flow regimes should be adopted by dam and river management authorities, and technologies to mitigate the impact of barrages to migratory species implemented.

Conclusions

Fresh water harbours the greatest concentration of life and also faces a crisis of historical and planetary proportions. Biodiversity is besieged. Extinction is the gravest aspect of the biodiversity crisis, it is irreversible. The human intervention was observed as one of the significant causes for the environmental status of the river basin. IUCN (2010) has reported the freshwater of the Eastern Himalaya region are under anthropogenic pressures and a total of 70% species (13.9%) are considered threatened, while 46 (8.8%) are Near Threatened. The deteriorating ecological situation of the aquatic water bodies and their catchments due to various anthropogenic activities are the major concern that caused tremendous pressure to many indigenous fish populations and aquatic biodiversity in general. Fishes also support the lifelihoods of many, especially rural, households and communities thus need to take necessary steps to conserve fish genetic resources and their habitats on the other hand, and to develop rational and efficient utilization and management of fish stocks on the other.

There is an urgent need for the conservation of fish species across the region for both economic and ecological regions. The fishes of the region are under threats due to destructive fishing including explosives, poisoning, dynamiting, electrofishing, as well as overexploitaion using fish barrages. With the development of Moreh and Tamu townships for Indo–Myanmar trade, great increase in human population and developmental activities has impacted the aquatic environment, particularly in the Lokchao River in Manipur and Yu River basin in Myanmar (Vishwanath *et al.*, 2010). The construction of railway track and tunnel along the Barak baisn in Manipur causes tremendous discharge of silt to Barak River causing increase fluctuation of the physic-chemical paramenter of water disturbing both physiology and reproductive ground of many of the endemic species.

Therefore, a regular monitoring of habitat condition and population trends of fishes should be undertaken. There should be prevention of the destructive fish harvesting techniques viz. dynamiting poisoning and electric fishing and laws on catching brooders formulated at the national and transboundary level. Research and training in fish raxonomy and conservation should be promoted. The available fish resources should be

utilised sustainably. The livelihood development program should be undertaken to the catchment area of the river basin to find out alternative conserve this valuable natural gift to us.

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PLATE-I

Endangered species of fish in Manipur

Tor putitora (Hamilton)

Pethia manipurensis (Menon *et al.*)

Psilorhynchus microphthalmus Vish. & Manoj

Schistura reticulata Vish. & Nebesh.

Schistura minuta Vish. & Shantaku.

Schistura kangjupkhulensis (Hora)

Schistura tigrinum Vish. & Nebesh.

Clarias magur (Hamilton)

Badis tuivaiei Vish. & Shanta

Pterocry ptis barakensis Vish. & Nebesh.

PLATE-II

Exotic fish species in Manipur

Cyprinus carpio Linnaeus

Piaractus brachypomus (Cuvier)

Hypopthalmichthys molitrix (Valenciennes)

Hypopthalmichthys nobilis (Richardson)

Clarias gariepinnus (Burchell)

Gambusia affinis (Baird & Girard)

Cetenopharhyngodon idella (Valenciennes)

Oreochromis mossambica (Peters)

Edible insects consumed by different ethnic people in Manipur and its potential use in food and feed

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Abstract

Edible insects, the ethnic food are rich sources of protein, vitamins and minerals. It has been reported for having more nutritional contents than the other conventional foods. Looking into the fact of high nutritional profiles, it will substitute the protein source in present day crisis of insufficient protein in diet. More than 300 ethnic groups inhabited in the world consumed insect as their conventional food, among Indian states, the state of Manipur is not behind, with 30 ethnic groups being consumed. A total of 41nsect species belonging to 8 orders under 24 families and 36 genera were recorded as edible insect in Manipur. Utilization of protein rich edible insect, silk worm pupae in particular in the form of Protein concentrate powder has been conducted in human food and animal feed to value added the products as nutrient supplement.

Keywords: Edible insects, fish feed, Manipur, silkworm pupa, pupae protein concentrate

Introduction

The practice of eating insects, entomophagy, is rooted in human evolutionary history (Fontaneto et al., 2011). They have been found to play a major role in the history of human nutrition in many parts of the world. Over 1500 species of edible insects have been recorded in 300 ethnic groups from 113 countries (MacEvilly, 2000). Some important groups include grasshoppers, caterpillars, beetle grubs, winged termites, bees, worms, ant brood, cicadas, and a variety of aquatic insects. Many species of insects have served as traditional foods among indigenous peoples and the insects have played an important role in the history of human nutrition (Defoliart, 1995). The insects are high in protein and many are rich sources of vitamins and minerals. Edible insects have been reported to have more nutritional content than the other conventional foods (Lokeshwari and Shantibala, 2010). It is estimated that, in 2050 the world population would be more than 9 billion people, resulting in an additional need for food specially protein. Conventional protein sources will be insufficient and there will be need for alternative sources. In this context, promoting the idea of insect as food to help increase consumer is suggested (Vantomme, 2015). Insects are also being discussed as an innovative ingredient for animal feeds, including feed for conventional livestock, fish and pets. As a result, insect base animal feeds are high in protein especially when processed and transformed into insect flour (van Huis et al., 2013). Although insect based feeds appear to be a viable option, further studies are needed regarding the nutritional, environmental and economic benefits of using them to reduce and replace conventional animal feeds (Gasco & van Huis, 2018).

Entomophagy depends upon insect palatability, taste, availability and suitability, nutritional value, food taboo restrictions, local traditions and religious customs (Chakravorty, 2011). This phenomenon is well observed where there is availability of insects around the dweller. Being a distinctive part of the Indo-Burma biodiversity hotspot region, the state of Manipur enjoys a rich diversity of insect fauna. There is a long traditional history of association of insects with the varied ethnic people of the state. The insects have been in use in varied ways such as for edible, medicinal, industrial and cultural purposes, among which insects are more popular as edible items. The 30 ethnic communities inhibiting the state have distinct identity, culture and food habit. Hence, the paper aims to signify the practice of entomophagy prevalent among the different tribes of Manipur. Further, the presented research work aims to focus the potential uses of edible insect for nutrients supplement in human and animal food for sustainable value addition for the benefit of the society as whole.

Utilization of different edible insects by ethnic inhabitants of Manipur

Almost all the 30 different ethnic groups inhibited in Manipur consumed insect as food. A total of 41 insect species belonging to 8 orders under 24 families and 36 genera were recorded as edible insect in Manipur (Table 1).The species composition in different orders comprises as follows, 1 species each in Dictyoptera and Isoptera, 4 species each in Lepidoptera, 7 species in Orthoptera, 5 species in Coleoptera, 7 species in Odonata, 7 species in Hymenoptera, and 10 species in Hemiptera. Some important insect species consumed by each ethnic community is presented in Figure 1.Study revealed that five ethnic groups namely Meitei, Tarao, Tangkhul, Chothe and Thadou consumed 28-30 species in comparison to 9-26 species consumed by other ethnic groups. Depending on the type of insect species, insects are prepared into different form such as curry, roasting, frying and even in raw form. Hard bodied insects are eaten in roasted or fried form whereas soft bodied insects are eaten as curry or raw. All the species of bee and wasps are customary cuisine of the Tangkhul community relating with their culture. Spring season, the main season of bee species was mainly coincided with the special festival of Tangkhul community such as “the Siroy lily festival” where the bee larva and pupae cuisine is the special and compulsory and also sold in a very high price. Hence, edible insects are culturally important traditional food of ethnic people in various part of Manipur. Therefore, for wider aspect and utilization of this nutrient rich unconventional food item, attempt has been to produce as value added products like pupae protein concentrate (PPC) for human food and animal feed.

Table-1: List of documented Edible insect species of Manipur

SL.NO.	COMMON NAME	SCIENTIFIC NAME	ORDER: FAMILY
1.	True water beetle	*Hydrophilus olivaceous* (Fabricius)	Coleoptera: Dytiscidae
2.	True water beetle	*Cybister confuses* Shp.	Coleoptera: Dytiscidae
3.	Rhino beetle	*Oryctes rhinoceros* (L.)	Coleoptera: Dynastidae
4.	Weevils	*Crytotrachelus* sp.	Coleoptera: Cucurlionidae
5.	Beetle	*Anoplophora glabripennis* (Motchulsky)	Coleoptera: Cerambycidae
6.	Giant water bug	*Belostoma indica* Lep & Serv.	Hemiptera: Belostomatidae
7.	Water bug	*Diplonychus rusticus* (Fabricius)	Hemiptera: Belostomatidae
8.	Water bug	*Ranatra virepes virepes* Stal.	Hemiptera : Nepidae
9.	Water bug	*Hydrometra greeni* Kirkaldi	Hemiptera: Hydrometridae
10.	Nepas	*Laccotrephes maculatus* F.	Hemiptera: Nepidae
11.	Bug	*Corius sp.*	Hemiptera: Denidoridae
12.	Stink bug	unidentified species	Hemiptera: Pentatomidae
13.	Stink bug	unidentified species	Hemiptera: Pentatomidae
14.	Bug	*Udonga montana* Distant	Hemiptera: Pentatomidae
15.	Backswimmer	*Notonecta* sp	Hemiptera: Notonectidae
16.	Rice grasshopper	*Oxya hyla hyla* Serville	Orthoptera: Acrididae
17.	Mole cricket	*Gryllotalpa orientalis* Beauvois	Orthoptera: Gryllotalpidae
18.	Grass hopper	*Mecapoda elongata* Linn.	Orthoptera: Tettigoniidae
19.	Grasshopper	*Shistocerca gregaria*	Orthoptera: Tettigoniidae
20.	Locust	*Gryllus* sp.	Orthoptera : Acrididae
21.	Grasshopper	*Acridium melanocorne*	Orthoptera: Acrididae
22.	Dragon fly	*Pantala flavescens* (Fabricius)	Odonata: Libellulidae
23.	Dragon fly	*Acisoma panorpoides* Rambur	Odonata: Libellulidae
24.	Dragon fly	*Crocothermis servillia* Drury	Odonata: Libellulidae
25.	Dragon fly	*Orthetrum triangulare* (Selys)	Odonata: Libellulidae
26.	Dragon fly	*Rhyothemis variegata* (Linnaeus)	Odonata: Libellulidae
27.	Dragon fly	*Diplocodes trivalis* (Rambur)	Odonata: Libellulidae
28.	Dragon fly	*Rhyothermes* sp	Odonata: Libellulidae
29.	Preying mantis	*Heirodula* sp.	Dictyoptera :Mantidae
30.	Honey bee	*Apis millifera*	Hymenoptera: Apidae
31.	Honey bee	*Apis cerana indica* Fabr	Hymenoptera: Apidae
32.	Honey bee	*Apis dorsata*	Hymenoptera: Apidae
33.	Wasps	*Vespa basalis*	Hymenoptera: Vespidae
34.	Wasps	*Vespa tropicalis*	Hymenoptera: Vespidae
35.	Yellow jacket wasps	*Vespula vulgaris* (Linnaeus)	Hymenoptera: Vespidae
36.	Paper wasps	*Prophelis sp.*	Hymenoptera: Vespidae
37.	Silk moth	*Bombyx mori*	Lepidoptera : Bombycidae
38.	Silk moth	*Samia cynthia ricini*	Lepidoptera : Saturnidae
39.	Silk moth	*Antheraea proylei*	Lepidoptera : Saturnidae
40.	Bamboo worm	*Omphisa fuscidentalis*	Lepidoptera : Pyralidae
41.	Termite	*Odontotermes* sp.	Isoptera : Termitidae

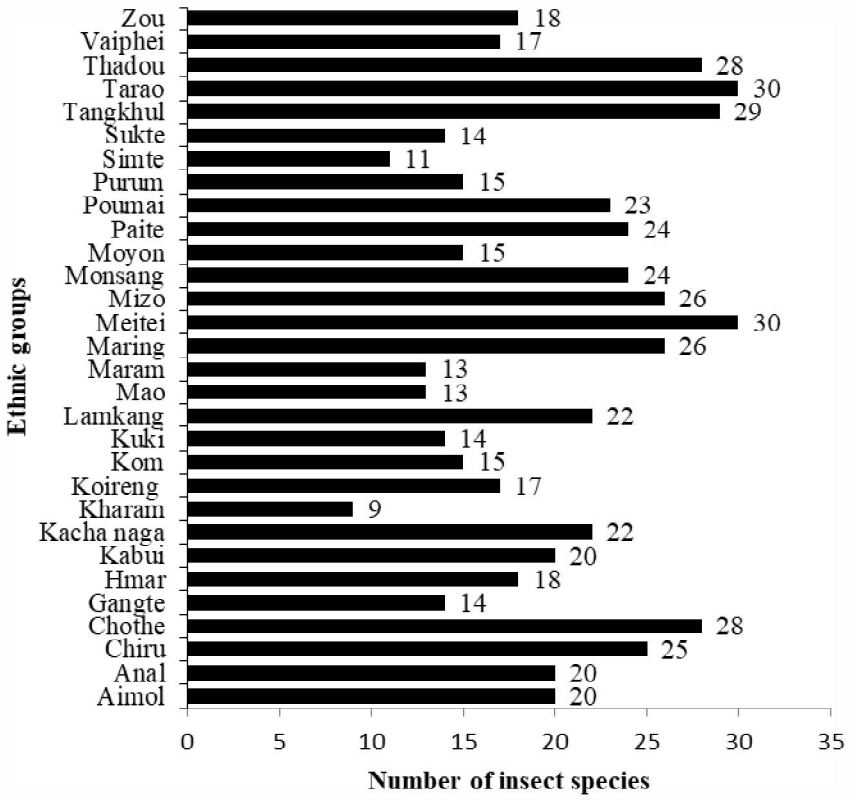

Figure1: Utilization of different edible insect species among 30 different ethnic inhabitants.

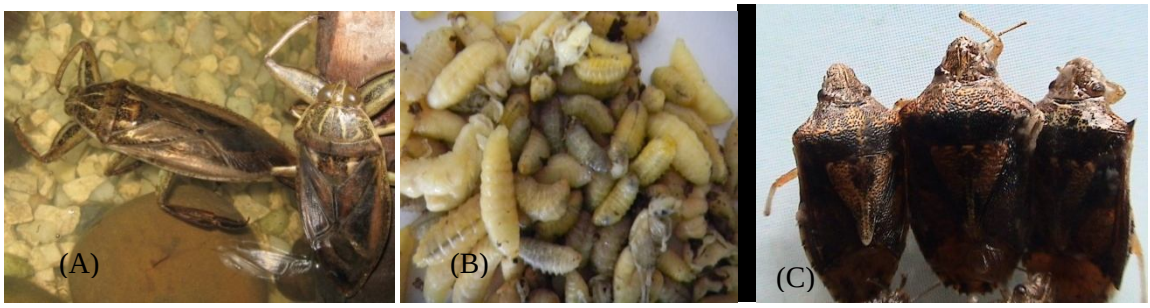

Figure 1a: Important edible insects (A) *B. indica* (B) Pupa of wasps, *V. basalis* (C) Stink bug,*U. Montana*

A. Utilization of Edible insect, *Bombyx mori* silkworm pupae for preparation of Pupae Protein Concentrate (PPC):

Generally, insect-based food is not socially supportive. Entomophagy is practised mainly by tribal communities in the states of Kerala, Odisha, Jharkhand, Karnataka, Tamil Nadu, Chhatisgarh, Madhya Pradesh and North East India (Gahukar, 2018). Thus, conversion of silkworm pupa material into edible quality pupae protein concentrate (PPC) and its value added products will be valuable items for protein supplement in human diet. An ideal product will be PPC which is white or near-white with a protein content of about 80 per cent and less than 1 per cent of fat, which is odour-less and at the same time, possesses a bland taste. PPC of desired nutritional quality would make an excellent supplement to the dietary of all sections of the population and especially to the vulnerable groups in areas where protein malnutrition is extensively prevalent.

Fresh spent silkworm pupae were collected and dried in the oven till constant weight is achieved. There is more than 80% reduction in the weight of the pupa after drying (Figure 2). Silkworm pupa protein concentrate (PPC) was prepared in laboratory using different solvents (ethanol, hexane and hexane + ethanol) and various techniques so as to obtain a standard PPC.

Table-2: Nutritional profile of *Bombyx mori* pupae powder

Sample	Protein (%)	Carbohydrate (%)	Lipid (%)	Moisture content (%)	Ash content (%)
Bombyx mori	22.1±0.23	4.16±0.43	29.02±0.34	3.32±2.56	5.51±0.02

Table-3: Characteristics of silkworm pupa protein concentrate

Solvent extract used	colour	Texture	Odour	Protein content (%)	Lipid content (%)
Ethanol	Cream	Granular	Odourless	82.5±0.07	0.52±0.03
Hexane	Light cream	Amorphous	Pungent	88.4±0.05	0.45±0.04
Hexane and ethanol	Light cream	Amorphous	odourless	93.6±0.05	0.25±0.08

In its crude form, the spent silkworm pupa has good nutritional quality (Table 2). The PPC obtained by running soxhlet with ethanol is less amorphous with irregular texture. It is darker in colour with a slight smell of ethanol. It also takes a longer period for PPC preparation. The PPC obtained running with hexane takes comparatively shorter period (4hrs as compared to 2 days in ethanol). The PPC thus obtained is more amorphous and lighter in colour but possesses a characteristic off smell (Table 3). Hence, there arises a need to remove the off smell, so the PPC obtained by running with hexane is again run with ethanol as solvent for around 4 hrs. Thus, an amorphous, white and odourless PPC is obtained. Thus, the protein concentrate recover can be used as protein supplement and value added product as such.

***In-vitro* digestion of PPC**

In-vitro analysis of the PPC with Pepsin digestibility, Trypsin digestibility and Pepsin - Trypsin digestibility showed gradual increases in digestibility, maximum being the combine digestibility with 51.17%, indicating good amount of protein digestible capability (Figure 3).

B. Utilization of pupae protein concentrate of silkworm pupa in fish feed formulation as nutrient supplement

The preference of particular species of insect mainly depends on taste and custom but biomass of the insect is also an important factor. Considering the consuming preference of silkworm as important edible insect among all the northeast ethnic groups in term of wide acceptability and biomass availability, it is selected as a valuable edible insect to be utilized in value added products for nutrient supplement. Being a leading state in the field of sericulture, Manipur produces all the four traded species of silkworm viz. *Samia cynthia ricini* (Eri), *Bombyx mori* (Mulberry), *Antheraea assamensis* (Muga) and *A. proylei* (Tasar) with a total annual production of 615.45 MT (2017-18). During the course of silk production, large biomass of silkworm pupa is also discarded as seri-waste every year (about 70% of silkworm cocoon biomass). The spent pupae are also complete nutritional package rich in protein (~41%), micro nutrients such as Iron (111 mg/100gm), Magnesium (622 mg/100gm), Calcium (~30.51 mg/100gm) etc and high antioxidant property (IC_{50} %, 68µg/ml, eri pupae) (Lokeshwari et al., 2019). Thus, seri-pupae are considered as nutritional goldmine that can be used for animal feed, especially in aquaculture, pet food industry or poultry feed.

Figure 2: (A) Cocoon of *B.mori* (B) reeling of silk fibre (C) spent pupa with a thin cover (D) spent pupa (E) drying of pupa (F) pupae protein concentrate

Fish being one of the culturally important cuisines in Manipur, huge demand of about 52,000 MT is required annually in the state (Anonymous, 2018). Only 32,000 metric tons (MT) of fish is produced annually having shortfall of about $2/5^{th}$ portion of fish requirement in the state. To fill up the huge gap, fish is being imported annually from other states like Andhra Pradesh and West Bengal to meet the deficiency of 20,000 MT in various forms such as fresh/frozen, dried and semi-fermented. However, availability of fish feed is a limiting factor in aquaculture development despite of having huge scope and absolutely dependent to outside state for feed supply. Concerted efforts are required in fish production to ensure nutritional security of the people. Geo-physical handicap on the national highways, high transportation cost are some of the factors affecting feed import to the state. Fish farming will be sustainable only when fish feeds are formulated using locally abundant feed ingredients. Low cost nutrient rich feed can be prepared using local ingredients such as protein rich seri-pupa (65-75%), mustard oil cake, rice brand, aquatic biomass of plants and incorporation of vitamins and minerals. As outside import dependent fish feed cost covers 60-80% of the total production cost where protein cost accounting about 15% of feed cost in livestock farming, therefore, utilization of protein gold mine, waste silkworm pupae in the low cost feed formulation will be convenient and cheaper than the conventional protein feed sources such as groundnut cake, fishmeal and soybean meal which does not permit profit maximization in aquaculture ventures. Above all, fishmeal is the only conventional animal protein source for fishery and that fish meal is scarce and expensive (Karimi, 2006). Henceforth, efficiency of, varying proportion of PPC of spent silkworm pupae waste incorporated fish feed formulations with locally available agro-byproducts such as Rice Bran (RB), Mustard oil cake in addition with mineral and vitamins fish feed formulation were evaluated.

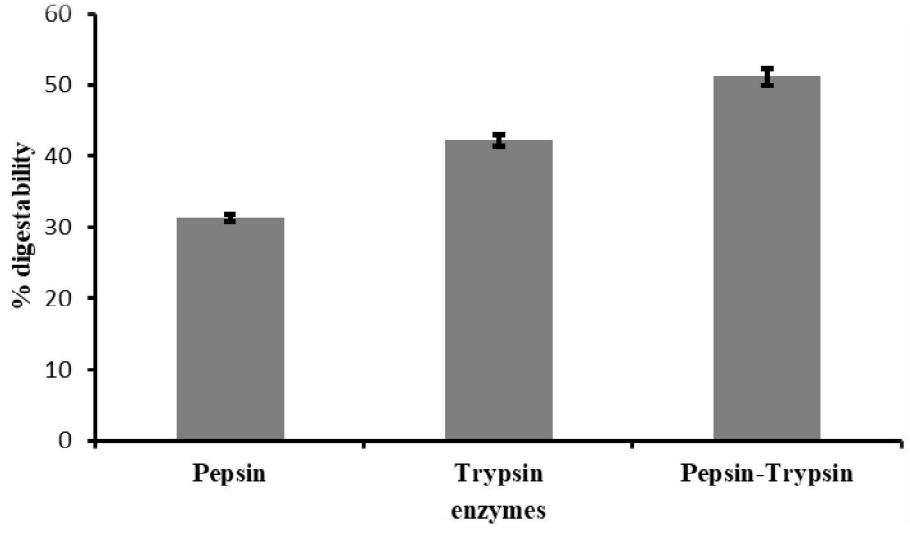

Figure-3: Protein digestibility percentage of PPC z

Formulation of silkworm pupae (PPC) incorporated different feeds and efficacy evaluation

Three different experimental feed formulations and one conventional control feed were prepared 1) T1=RB + (MOC: SWP; 1:1); 2) T2=RB + (MOC: SWP; 1:2); 3) T3=RB + (MOC: SWP; 1:3) and 4) C =RB + MOC; 1:1 as control. In all the diets (control and tests) binder mix (Tapioca flour + Maida + Rice Flour; 6+3+1) and Vitamins & minerals mix were incorporated @ 5% and 2% respectively.

Determination of feeding efficacy of three formulated feeds against the control feed were evaluated with carp fingerlings (*Labeo rohita*) of 15-16gm in weight at the Aquatic laboratory of Institute of Bioresources and Sustainable Development, Imphal, Manipur. Twelve FRP Tanks were stocked with 10 fingerlings per tank (at a stocking density of 6000/ha) and fed with the control and test feed formulations for 120 days @ 5% of the body weight maintaining triplicates. Before supplying each feeding to the tank, faecal matter and uneaten feed were siphoned out and 20% water exchange was done daily. At every ten days interval, sampling was conducted to record fish growth and to adjust the feed quantity. At the end of the experiment, all the fish was collected, counted and weighed and feed conversion ratio (FCR), periodic weight gain, relative growth rate, protein efficacy ratio (PER), specific growth rate (SGR) and survival rate were determined to find out the best formulation.

At present, only conventional feed consisting of rice bran and mustard oil cake (50:50) is used in fish culture and use of formulated fish feed is still in a nascent stage due to high cost involved. Production of low cost fish feed is a necessity for increasing the productivity to meet the requirement of the state. Silkworm pupae meal (SWPM) is a protein-rich feed ingredient of animal origin with a high nutritional value. On dry matter (DM) basis its crude protein content ranges from 50% to more than 80% (defatted meal). Waste silkworm pupae (SWP) generate vast resources of nutrients for livestock and fish farming and considered to be one of the unconventional top class proteins (65-75%) and lipid feed. Among many alternative protein sources, SWP are considered as an important dietary protein source for fish culture. Therefore, with the scope to utilize the seri-pupa waste of the state, present study evaluated efficacy of three different fish feed formulations (T1, T2 & T3) incorporated with silkworm pupae at varying level on carp fingerlings (*L. rohita*). Defatted silkworm pupae waste (DSPW) was incorporated at 25%, 50% and 75% as replacement of MOC respectively. The feeding experiments were conducted for three months and their efficacy was compared against the control feed (C) which is farmer's conventional feed where no DSPW is incorporated (rice bran: mustard oil cake @ 1:1) (Figure 4). Among the test feeds, T1, where MOC and SWP are incorporated at same ratio, showed non-significant difference on growth rate with C. However, T3 showed significant effective growth rate indicating more than double the size (36.60±0.56gm) then the initial day (16.66±0.56gm) at the end of 90days experiment. Relative Growth Rate (RGR) of different formulated feeds indicated that with respect to the proportion of DSPW incorporation, approximately 45-

50% increase in relation to the proportion of SWP incorporated in feed formulations (Figure 5). Hence, experiment on the *in-vitro* utilization of DSPW as protein supplement along with mustard oil cake (MOC) and rice bran in fish feed revealed double the relative growth rate of fish in three months compare to control feed. Sasmal et al, (2018) also revealed that used of different proportion of silkworm pupae as protein sources (25%, 30%, 35%, and 40%) with rice bran, mustard oil cake-based control diet (25% protein), showed the best performance in the fish with the diet having 40% protein level incorporation. Specific growth rate (SGR), feed conversion (FCR), protein efficiency ratio (PER) and protein and lipid deposition in the muscle showed highest with the maximum incorporation of silkworm pupae. In an earlier study conducted by Nandeesha, et al. (2000), it was shown that feeding common carp with diets containing upto 30% silkworm pupae resulted in progressive increase in growth with increasing level of pupae as compared to a fishmeal based 30% protein diet and the highest weight was recorded at 30% of pupae incorporation. Further, it was also mentioned that the cost of production was lowest with silkworm pupae incorporated feed than control without silkworm pupae. Therefore, these types of low cost fish feed formulation with locally available materials will be cheaper than the conventional protein feed sources in maximizing aquaculture ventures. Hence, silk industry waste of Manipur can be an alternative low-cost protein supplement for effective fish feed formulation of the state.

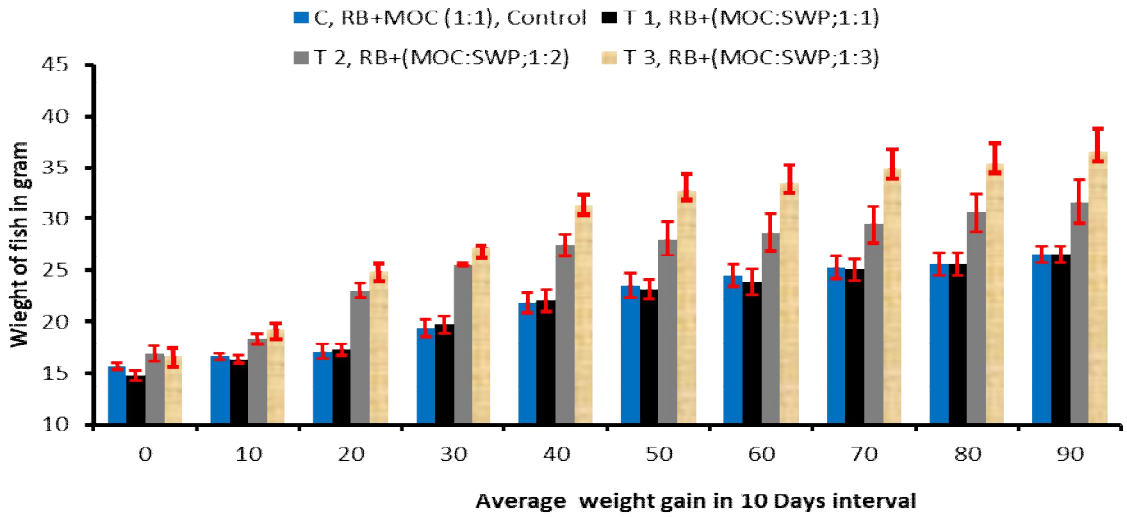

Figure-4: Efficacy of three different seri-pupae fish feed formulations against control feed on the growth of *Labeo rohita* fingerlings.

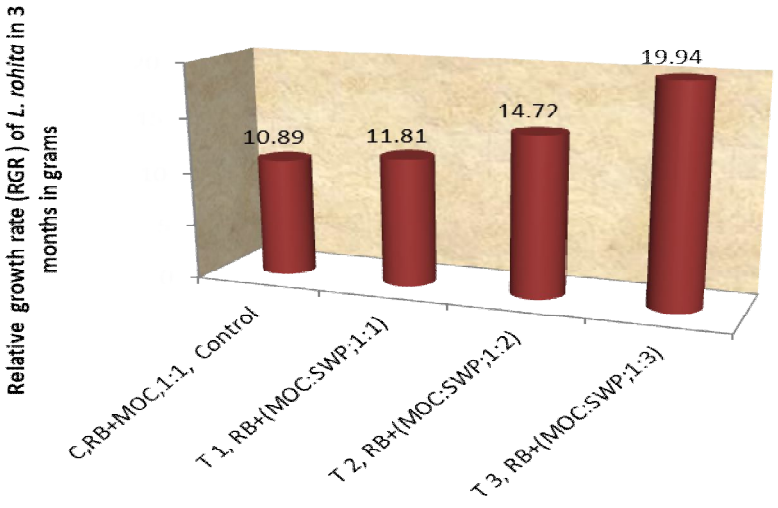

Figure-5: Relative Growth rate of carp fingerlings fed with three different feed formulations and conventional control feed.

Conclusion

In India, entomophagy is more prevalent in North-East India where insects are readily available. North-east India can be treated as epicentre of entomophagy. Eventually, more research is needed to understand the prevailing entomophagy in the North East region where indigenous communities enjoy nutritious food with insects as sustainable ingredient in main dish or as supplement. More down line studies on production of value added products are needed at this hour. In this juncture, researchers are required to go in collaborative work with food industries and entrepreneurs. Once the hope of incorporating the benefits of edible insects into the food resources is successful, it will definitely combat malnutrition and undernourishment.

Acknowledgement

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Bamboo as a resource of nutraceuticals: an analysis of bioactive component

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Abstract

Bamboo, the wonderful 'Green gold' plant is nature's most valuable gifts to mankind. The emerging fresh young delicate bamboo shoots and leaves of some bamboo species are known as one of the important and culturally significant food items and at present are being promoted as a new health food and as a potential nutraceuticals. Bamboo leaves which contain high fiber, antioxidant and other nutrients are beneficial to the health. Although the different parts of the bamboo are used as a food resource, a systematic conclusive study on its nutritional and nutraceuticals significance is not available. The present paper explores the various bioactive components in different bamboo species along with its nutraceutical potential. Both fresh and fermented bamboo shoots contain high level of phytosterols. Significant differences were observed in the phytosterol content of different species (125.23 –287.55 mg/100 g dry wt.). The representative physterols in bamboo shoots were β-sitosterol, campesterol and stigmasterol. These phytosterol act as neutraceuticals and are precursors of many pharmaceutically active steroidal drugs. Phytosterols are effective in lowering cholesterol by inhibiting the absorption of cholesterol from the small intestine. Hence Bamboo shoots and bamboo leaves may be promoted as health enhancing food due to its rich nutritive bioactive components which are of great interest in both food and pharmaceutical industries.

Keywords: Bamboo shoots / bamboo leaves//bioactive component/ phytosterols/ neutraceuticals

Introduction

Neutraceuticals are products derived from food sources that are purported to provide extra health benefits, in addition to the basic nutritional value found in foods (Maher, 2003). Nutraceuticals may be considered as health bio-actives that may be used in functional foods and one of the most researched groups of nutraceuticals in the area of cardiovascular diseases is phytosterols (Zawistowski, 2002). Plant sterols, known generally as phytosterols, are essential components of the membrane lipid bilayer (Schuler, 1997). There is considerable interest in phytosterols as dietary supplements as they are reported to lower cholesterol levels (Ostlund et al., 2003) and also have a positive impact on cardiovascular diseases (Patel and Thompson,2006; Weingartner et al., 2009). Again, bioactive compounds are used as natural product-derived therapeutic agents or as disease-preventing nutrients(Ajikumar, 2008). Over the last few decades, natural bioactive compounds with potential for the treatment and prevention of human diseases have attracted much attention in many laboratories and industries and bamboo is one such source of nutraceuticals. Three predominant phytosterols have been identified in bamboos, β-sitosterol, campesterol, and stigmasterol (He and Lachance 1998; Lachance and He 1998; Sarangthem and Singh 2003; Lu et al.,2010) but some minor sterols have also been reported. The bamboo shoots are reported to have anti-hypertensive, anti-tumour and anti-oxidant properties. Bamboo leaves and bamboo salt made from the culm of bamboo are used as a medicinal food in many Asian countries. Bamboo leaves are also used as fodders and it serves as the sole food source for Giant Pandas. However, there is a general lack of information available on the nutrient value of leaves. Due to its various uses, bamboo is now being recognized for economic and environmental development, thus leading to a change its image from being called 'poor man's timber to "Green Gold". In addition to its multifarious uses, bamboo shoots are now being promoted as health food (INBAR 1997; Ogunwusi, 2011).

The emerging fresh young bamboo shoots which are used in numerous Asian dishes are available in markets in various sliced forms like fresh, fermented and even canned version (Tai, 1985; Fu et al, 1987; Midmore, 1998). At present over two million tons of edible bamboo shoots are consumed in the world in each year (Yang et al. 2008) mainly in the Southeast Asain region. In India, the northeastern states located at the foothills of the Himalaya are rich in flora and many wild species of plants are consumed in the local diet as wild edible plants, amongst which, bamboo features prominently. In Manipur, one of the north eastern states of India, fresh and fermented bamboo shoot consumed in large scale are highly prized vegetable items. More than 700,000 culms are extracted every year in Manipur (Statistical bulletin of Manipur forest, Govt. of Manipur, 1999-2000). Young delicate bamboo shoots are of particular favorite because of its high fiber content and its delicacy (Fuchigami, 1990). An increase in dietary fiber is reported to reduce the blood pressure (Anderson and Strong 1983)while according to George et al (1982), dietary fiber is useful in the management of hypertension and obesity through its effect on energy density of food and the extent of interference with the nutrients of bioavailability. With the increasing studies and validation of the usefulness of including dietary fibre, plants with high fibre content like bamboo are naturally more preferred. Not only dietary fibre but bamboo are also rich in minerals, have adequate amount of glucose, are low in fat and is tender yet crunchy, delicious and nutritive (Yamaguchi and Kusama, 1976; Yamaguchi, 1983; Nirmala et al., 2008; Park and John, 2009). It also contains many secondary metabolites which can be used as precursors of many pharmaceutical industries (Sarangthem and Singh, 2003). In spite of their high nutritive value one drawback of bamboo shoots consumption is that they are found to contain cyanogenic glycosides. These cyanogens glycosides on being disrupted by slicing or maceration, releases hydrogen cyanide which is toxic to human being. The cyanogenic glycoside in bamboo is taxiphyllin which is a p-hydroxylatemandelo-nitrile triglochinin (Nahrstedt, 1993; Hunter and Yang, 2002; Pandey and Ojha, 2013; Schwarzmaier, 1977). Taxiphyllin is a bitter compound (Ke-jun et al., 2005) making some bamboo shoots taste bitter to eat. Processing and fermentation of the bamboo shoots decreases the toxicity of the cyanogenic glycoside (Sarangthem and Hoikhokim, 2010).

This chapter delves into the analysis of the type of bioactive component and phytosterols content in bamboo shoots in both fresh and fermented form (traditional and scientifically modified fermentation) as well as in bamboo leaves so that it may be promoted as health enhancing neutraceutical foods.

Bamboo shoot fermentation

Bamboo shoot fermentation processes are usually simple although different regions have different traditional methods. Preservative methods of the fresh succulent bamboo shoots are done in large-scale in Manipur by traditional methods of fermentation process. The fermented bamboo shoot slices are locally called *soibum* and *soidon*. Bamboo shoots of many species like *Bambusa tulda, B. balcooa, Dendrocalamus hamiltonii, D. giganteus, D. hookeri, Pseudostachyum polymorphum, Schizostachyum dulloa* and *Thyrsostachys oliveri* are used for fermentation of *soibum.* In the traditional method the *soibum* is prepared by storing thin slices of fresh succulent and soft bamboo shoots in certain containers/chambers for 2-3 months. The fermented chambers are either made of bamboo planks or roasted earthen pots. The inner surface of bamboo chambers are lined with banana leaves and a thin polythene sheets. The upper surface is sealed with polythene sheet and weights are then put on top for proper pressing. At the initial stage of fermentation the exudates is leached/drained out of the tilted side of the bamboo plank chamber. After fermentation is completed, which is indicated by the smell, colour and texture, *soibum* can be stored up to one year.

In controlled fermentation for laboratory analysis, a modified form of the traditional method of fermentation is adopted which involves inoculating thin slices of succulent bamboo shoots with the exudates obtained from already fermented slices of bamboo shoots (traditionally fermented) under aseptic condition using a Laminar flow. After inoculation, the samples were kept in an incubator at 30+2^{o}C for a period of 90 days.

For the emerging young fresh succulent bamboo shoots (Fig 1) of *Bambusa tulda* Roxb, *B. balcooa* Roxb., *Dendrocalamus hamiltonii* Nees & Arn.ex Munro, *D. giganteus* Munro, *D. Hookeri* Munro, *Pseudostachyum polymorphum* Munro, *Schizostachyum dulloa* Gamble *and Thyrsostachys oliveri* Gamble are collected during the growing season (May- July) in different districts of Manipur, India for fermenting. Collection of the bamboo shoots are usually made and processed the same day. The outermost scale portions of the fresh succulent bamboo shoots are normally discarded and the inner soft delicate shoots which are edible is taken and kept separately for the processing.

Biochemical analysis

For biochemical analysis the sliced shoots were oven dried at 60-70°C in an air oven to dry for 12h and the dried samples of the delicate shoot apex were then crushed to powder form using a grinder and kept for the analysis.

Leaf samples were also collected in the same growing season (May- July) with approximately 50 g of fresh leaf tissue per bag from field-grown plants. Samples were placed into paper bags and their contents were weighed immediately following collection. Paper bags with leaf samples were then placed at 70°C in an air oven to dry. Dry samples were grounded using a grinder and kept ready for analysis.

To assess the nutritional values, bioactive component and phytosterol content of the mixture of fresh bamboo shoot slices each of *Bambusa tulda , B. balcooa, Dendrocalamus hamiltonii, D. giganteus, D. hookeri , Pseudostachyum polymorphum, Schizostachyum dulloa* and *Thyrsostachys oliveri* and fermented samples of these succulent bamboo shoots, different parameters of biochemical analysis were conducted such as mineral element, crude fibre, total phenols, total flavonoids and tannins, total phytosterols

Fig-1: Young emerging bamboo shoot

Mineral elements

Leaf samples were analyzed for K, Ca, Mg, Fe, Mn, Zn, Co,and Cu concentrations using recommended digestion procedures (Perkin Elmer). Prior to the elemental analysis, leaf samples were processed using the same methodology for protein analysis. Leaf samples were heated at 70°C in an oven for 24 hours and placed in desiccators. Three replicates of 1g tissue from each sample were weighed in porcelain crucibles and incinerated at 500°C for 4 hours and allowed to cool overnight. The incinerated samples were digested with 20ml of 33% HCl until 10ml of solution remained in the crucible. After digestion, each sample was filtered through Whatman filter paper (No. 541) into a 100ml volumetric flask using hot distilled water. After cooling down, the sample was adjusted to 100ml by adding water and then employed for nutrient analysis using Perkin Elmer atomic absorption spectrophotometer, Analyst AA-200 Spectrometer. The emission signal of samples was obtained by developing calibration curves. The results for triplicate samples were averaged and standard deviation for each element was calculated.

From the investigation the mineral content of the fresh bamboo shoots slices sample analyzed are shown in Table 1. Elements like copper, zinc, cobalt, ferric, calcium, manganese, magnesium and potassium were found with variation in amount in all the samples analyzed. For calcium it ranges from 1.29mg/100g dry wt. to 39.10mg/100g dry wt. with *Thyrsostachys oliveri* shoot the highest and lowest in *Dendrocalamus giganteus.* For magnesium, the highest range was observed in *Schizostachyum dullooa* (8.81mg/100g dry wt.) and lowest in *Dendrocalamus hookeri* (1.96mg/100g dry wt.) and manganese concentration ranges from 0.42mg/100g dry wt. to 1.20mg/100g dry wt. with *Bambusa balcooa* the highest and *Dendrocalamus hamiltonii* the lowest (Table 1). Amount of zinc, copper, iron and cobalt were also detected in fresh bamboo shoots. Zinc content was highest in *Dendrocalamus giganteus* (1.8mg/100g dry wt.) and lowest in *Dendrocalamus hookeri* (0.49mg/100g dry wt.) as in Table 1. Copper concentration ranges from 0.08mg/100g dry wt. to 0.29mg/100g dry wt. Cobalt content was the highest in the shoots of *Bambusa tulda* (0.167 mg/100g dry wt.) and lowest in the shoots of *Thyrsostachys oliveri* (0.20mg/100g dry wt.). Ferric concentration was the highest with *Bambusa tulda* shoot (1.043mg/100g dry wt.) and lowest in *Schizostachyum dullooa* shoot (0.394 mg/100g dry wt.) as in Table 1. Potassium content in the fresh shoots ranges from 400mg/100g dry wt. to 4940mg/100g dry wt. with *Thyrsostachys oliveri* having the highest and lowest with *Schizostachyum dullooa*

Table-1: Mineral elements in edible bamboo shoots of different bamboo species (mg/100g dry wt.)

Name of species	Cu	Zn	Co	Fe	Ca	Mg	Mn	K
1. *Bambusa balcooa*	0.35 ±0.02	0.76 ±0.02	0.21 ±0.03	0.78 ±0.01	16.12 ±0.06	6.35 ±0.03	0.82 ±0.01	423.33± 1.2
2. *Bambusa tulda*	0.325 ±0.02	1.65 ±0.02	0.167 ±0.01	1.043 ±0.01	1.61 ±0.06	3.15 ±0.06	1..20 ±0.01	415.213 ±1.2
3. *Dendrocalamus giganteus*	0.257 ±0.04	1.80 ±0.06	0.110 ±0.003	0.292 ±0.015	1.29 ±0.10	2.70 ±0.06	0.45 ±0.01	435.67± 1.5
4. *Dendrocalamus hamiltonii*	0.025 ±0.004	0.80 ±0.02	0.142 ±0.02	0.522 ±0.01	15.73 ±0.01	2.60 ±0.06	0.42 3±0.01	423.27± 1.4
5. *Dendrocalamus hookeri*	0.568 ±0.06	0.49 ±0.01	0.08 ±0.01	0.487 ±0.06	25.46 ±0.04	1.96 ±0.06	0.81 ±0.02	401.33± 1.2
6. *Thyrsostachys oliveri*	0.751 ±0.03	0.74 ±0.01	0.20 ±0.01	0.441 ±0.01	39.10 ±0.03	2.65 ±0.01	0.62 ±0.04	494.00± 1.2
7. *Schizostachyum dullooa*	0.123 ±0.01	1.51 ±0.03	0.29 ±0.01	0.394 ±0.06	28.11± 0.04	8.81 ±0.06	0.54 ±0.01	400. ±0.1
8. *Pseudostachyum polymorphum*	0.273±0 .008	0.71± 0.11	0.045±0 .047	0.925±0 .010	18.37± 0.04	7.49± 0.22	0.030±0 .010	452.61± 1.4

* Data presented as mean ± SD.

Crude fiber content

Estimation of total crude fiber content was done following Sadasivam and Manikam (1992). 2 g of the grinded sample was extracted in petroleum ether to remove fat. The extracted sample was then air dried. The dried sample was then boiled with 200ml of H_2SO_4 (1.25%) for 30min. Filtered through muslin cloth and washed repeatedly (2-3 times) with boiling water until washings are free of acid. The residue was again boiled with 200ml of NaOH (1.25%) for 30minute. Filtered through muslin cloth and again washed repeatedly (2-3 times) with 25ml of boiling H_2SO_4 (1.25%), three 50ml portions of water and 25ml of alcohol. The residue was then removed and transfer to a pre- weighted ashing dish (w_1). Then the residue was again dried for 2hour at 130 ± 2^0C, cool in desiccators and weighted (w_2). Ignited for 30minute at 600 ± 15^0C in a muffle furnace, then cool in desiccators and reweighted (w_3). Total crude fibre is then calculated using the following formula

$$\text{Crude fiber content (\%)} = \frac{\text{Loss in weight on ignition } (W_2 - W_1) - (W_3 - W_1)}{\text{Weight of the sample}} \times 100$$

Total phenolics, flavonoids and tannins

The dried powdered bamboo shoot samples were extracted in 10 ml of methanol by intermittent maceration up to 48 h, centrifuged and the supernatants were used for the estimations of total phenols. Total phenolic contents were determined by folin-ciocalteu method with sodium carbonate solutions following McDonald *et al.* (2001). The absorbance was measured at 765 nm using chlorogenic acid as the standard. Flavonoids content were determined by Aluminium chloride method following Chang *et al.* (2002). The calibration curved was prepared by different concentrations of Quercetin in methanol. The absorbance was measured at 415 nm in a spectrophotometer. Tannin contents were determined by Folin-Denis method (Sadasivam and Manickam, 1992) which is based on the non-stoichiometric oxidation of the molecules containing a phenol hydroxyl group. Tannin like compounds reduced phosphotungstomolybdic acid in alkaline sodium carbonate solutions to produce highly blue coloured solutions. The intensity of which is proportional to the amount of tannin. The absorbance was measured at 700 nm using tannic acid as the standard compound.

Table 2 shows the bioactive component in young bamboo shoots of different bamboo species. Bamboo shoots contain the bioactive component of total phenols ranging from 256 to 798.23 mg/100g fresh wt. and flavonoids content ranges from 128.76 to 798.23 mg/100g fresh wt. and tannin content ranges from 0.86 to 45.49 mg/100g fresh wt. (Table 2). The phenolic and flavonoids compounds have been reported to exert multiple biological effects including antioxidant, free radical scavenging, anti-inflammatory, anticarcinogenic and antiviral activities (Miller, 1996 ;Oboh 2008) Sevral studies indicated that the antioxidant activities of some plants are highly correlated with their phenolic contents (Palav et al. 2006; Oboh 2008; Gupta et al. 2010). Therefore, the bamboo shoots can also be used for formation of natural anti-oxidants. Pandey et al. (2011) also reported that phenolic acids have a correlation with antioxidant properties. Crude fiber content of the fresh shoots of the bamboo species ranges from 18.65% to 32.12% dry wt. with Bamboo leaves containing the highest (32.12% dry wt.) and lowest with that of *Dendrocalamus giganteus* shoots(18.65% dry wt.) as in Table 2.

Moisture content were determined using the ISTA methods (1996) as follows-

$$\text{Moisture content (\%)} = \frac{\text{original weight- oven dry weight}}{\text{Original weight}} \text{ x100}$$

The results in Table 3 shows that fresh bamboo shoot slices mixture each of *Bambusa tulda , B. balcooa, Dendrocalamus hamiltonii, D. Hookeri Pseudostachyum polymorphum, Schizostachyum dulloa* and *Thyrsostachys oliveri* shows 79.47 per cent of water content; 6.08 pH value 97.73 mg/100g fresh wt. of phenol; 51.11 mg/100g fresh wt. of flavonoid; 31.49 mg/100g fresh wt. of tannin content.

Table-2: Bioactive component in young bamboo shoots of different bamboo species

Sl. No.	Name of the bamboo Species	Phenol (mg/100g fresh wt.)	Flavonoid (mg/100g fresh wt.)	Tanins (mg/100 g fresh wt)	Phytosterols (mg/100g dry wt.)	Crude fiber (%)
1.	*Bambusa balcooa*	278.32 ± 1.4	128.76 ± 1.4	31.49 ±1.50	287.55 ± 5.0	28.25 ±1.20
2.	*B. tulda*	412.23 ± 1.6	412.23 ± 1.6	35.49 ±0.57	267.72 ± 3.3	26.35 ±0.06
3.	*Dendrocalamus giganteus*	798.00 ±1.4	798.00 ±1.4	41.23 ±1.60	173.10 ± 1.8	18.65 ±1.20
4.	*D. hamiltonii*	786.32 ± 1.2	786.32 ± 1.2	45.49 ±0.67	248.35 ± 5. 02	21.47 ±0.14
5.	*D. Hookeri*	798.23 ± 1.5	798.23 ± 1.5	24.46 ±0.57	225.00 ± 5.0	23.98 ±1.63
6.	*Thyrsostachys oliveri*	691.03 ± 1.2	691.03 ± 1.2	28.53 ±1.21	178.07 ± 3.24	25.68 ±1.14
7.	*Schizostachyum dulloa*	720.42 ± 1.4	720.42 ± 1.4	22.16 ±0.04	125.23 ± 4.10	25.720 ± 1.02
8.	*Pseudostachyum polymorphum*	402.12 ± 1.2	402.12 ± 1.2	12.86 ±0.59	189.12 ± 5.12	24.52 ±1.23
9.	Bamboo leaves	256.00 ± 1.8	260.76 ± 1.6	0.86 ±0.59	118.15 ± 5.12	32.12 ±1.60

*Data presented as mean ± SD.

Table-3: Bioactive component present in fresh edible bamboo shoots (mixture of the eight bamboo species shoots) and fermented bamboo shoot slices (Scientifically modified laboratory fermented and traditionally fermented for three months)

Parameters	**Fresh bamboo shoot slices**	**Laboratory fermented sample (3 months old fermentation)**	**Traditionally fermented sample (3 months old fermentation)**
1. Moisture (%)	79.47±1.20	68.92±0.18	73.36±1.38
2. pH value	6.08±0.22	4.18±0.07	4.41±0.34
3. Total phenol (mg/100g fresh wt.)	97.73±1.40	204.91±3.80	190.65±1.48
4. Flavonoid (mg/100 fresh wt)	51.11±1.77	56.73±0.86	63.96±0.41
5. Tannin (mg/100 fresh wt)	31.49±1.50	52.00±1.58	68.21±0.55

*Data presented as mean ± SD.

Antioxidant properties

There are many methods used to determine antioxidant properties of food out of which the most widely used method due to its simple and fast nature while being inexpensive at the same time involves the use of free radical DPPH (2,2-Diphenyl-1 picrylhydrazyl). DPPH radical is scavenged by antioxidants through the donation of a hydrogen atom, forming the reduced DPPH. The colour changes from purple to yellow colour after reduction and this is quantified by its decrease in absorbance at 517nm. Here, the DPPH method is adopted and preparation of the extracts is processed as follows - 10g of the powdered samples were extracted in a Soxhlet apparatus with 100 mL of ethanol (60ºC for 12 h). The samples were filtered through Whatman No.1 paper in Buchner funnel, the filtrate was freeze dried and weighed. 2, 2-Diphenyl-1-picrylhydrazyl (DPPH) and quercetin were obtained and the method used by Fogliano et al. (1999) was adopted. The absorbance of the colour developed was measured at 517 nm by a spectrophotometer. Data

were processed using excel and the concentrations that caused 50% reduction in absorbance (RC50) were calculated. Percent inhibition of DPPH was calculated by following equation (Lee et al., 1998)

% Inhibition = 1- (A1/A2) × 100

where A1 is the absorbance of the test samples and A2 the absorbance of control reaction.

In the present study all the methanolic extracts of the fresh and fermented samples exhibited antioxidant activity in dose-dependent manner. The percentage of inhibition and IC_{50} are given in Table 4. Highest radical scavenging activity as % of DPPH inhibition is 89.70% at 50µg/ml with fresh bamboo leaves (Table 4). Antioxidant of bamboo leaves (AOB), an extract from *Phyllostachys pubescens*, has been reported to exhibit multiple biological activities, such as scavenging oxygen radicals and anticancer, antibacterial, and antiviral activitiy, and is especially known for its antioxidant activity (Park and Jhon,2010; Saki and Maeda,2010). Several studies indicated that the antioxidant activities of some plants are highly correlated with their phenolic contents (Palav et al., 2006; Oboh 2008; Gupta et al. 2010). Pandey et al. (2011) also reported that phenolic acids have a correlation with antioxidant properties.

Table-4: Comparison of the anti-oxidant properties in leaves, fresh edible bamboo shoots and fermented bamboo shoot slices

Sample	% of inhibition of DPPH (µg/ml)				
	10µg ml^{-1}	20µg ml^{-1}	30µg ml^{-1}	50µg ml^{-1}	IC_{50}
Leaves of bamboo	28.25±1.25	48.37±0.57	6.70±0.85	89.70±0.14	8.10±0.05
Fresh bamboo shoot slices	22.85±1.36	40.37±0.69	66.70±0.85	79.70±0.64	2.10±0.05
Laboratory fermented bamboo shoot slices	17.57±1.27	33.82±1.18	40.49±3.04	77.50±0.64	3.33±0.01
Traditionally fermented shoot samples	22.99±1.50	37.84±1.16	52.56±1.13	68.03±0.99	5.30±0.24
Quercetin (standard sample)	27.16±1.27	45.09±0.27	77.02±1.43	86.022±1.2	8.56±0.07

*Data presented as mean ± SD

Total phytosterol

For the analysis of total phytosterols, delicate bamboo shoot apex was sliced and oven dried at 60°C± 2°C for 12h. The dried samples of the delicate shoot apex were then crushed to powder form. The powder was used for determination of total phytosterols using Liebermann-Burchard reaction (Katayama *et al*. 1974).To purify phytosterols, the dry fermented samples were taken and extracted in a 1litre clevenzer apparatus using benzene, petroleum ether and 2N ethanolic KOH(10:5:1) as the refluxing solvent (Sarangthem and Srivastava,1997). After selective solubilization of the crude phytosterols with acetonitrile, the crude phytosterols were then subjected to TLC (Stahl, 1969). TLC was performed on silica gel-G plates (0.25mm thick, 20x20 cm) using solvent pairs hexane: ethyl acetate (3:1). Detecting reagent were acetic anhydride and sulphuric acid (30:1). For obtaining crystallized form of the phytosterols isolated from fermented shoot samples, preparative TLC was conducted. The phytosterols (tentatively identified as β- sitosterol, stigmasterol, diosgenin and campesterols) resolved on TLC and confirmed with standard samples were scraped and eluted in chloroform for analysis. The UV spectral analysis for the crystals obtained after preparative TLC (Stahl 1969) as well as control authentic samples (Sigma Chemicals, USA) were measured from 225 to 400 nm on a Beckmann DU-64-spectrophotometer. The fractions obtained after preparative TLC were further purified by refluxing with acetonitrile and the precipitate so obtained along with β-sitosterol, Stigmastero l& campesterols (Sigma Chemicals) were analysed by High performance liquid chromatography(HPLC). Methanol /water/acetic acid (750:240:10) was as the mobile phase ; flow rate 1ml/min run time 20min,column temperature 30^0c , Injection temperature 280°C,inj. 30µl.column used – Zorbax SB-C18

Further analysis of IR, NMR and Mass spectral analysis were done at CDRI, Lucknow for confirmation of the compound in comparison with control authentic samples (Sigma Chemicals, USA).

The level of total phytosterols in the succulent shoot samples of different species of bamboo ranges from 0.15 per cent dry wt. in fresh shoot slices (Table 5) to 0.65 per cent in traditionally fermented shoots i.e. the level of phytosterols increases to four times or more in fermented bamboo slices than fresh shoots. Fermentation increases the accumulation of certain by-products as a result of breaking down of the raw organic molecules (polymers) by the activity of microorganisms. The crude phytosterols extracted from the bamboo shoot slices when subjected to selective solubilization yielded different amount of phytosterols in various fractions, which on further analysis by Co-chromatography with TLC, HPLC with standard samples revealed that the fractions were β–sitosterol, stigmasterol and campesterol (Fig 3). This was further identified by analysis of its melting point, molecular wt. and mass spectral analysis at CDRI, Lucknow. The UV and IR spectral data of the compound showed similarity with those obtained with the authentic samples of β–sitosterol, stigmasterol and campesterol (Sigma Chemicals,USA). Park and Jhon have pointed out that bamboo shoots could reduce the serum total cholesterol, low-density lipoprotein cholesterol (Park and Jhon, 2009; 2010). Phytosterols have received particular attention due to their capability to lower serum cholesterol levels in humans, resulting in significant reduction of the risk of cardiovascular diseases (Plat & Mensink, 2001). Furthermore, they were also regarded as a kind of natural product with anti-inflammatory (Bouic, 2002), anti-bacterial (Ovesna et al., 2004), and anti-carcinogenic properties (Awad et al., 2000).

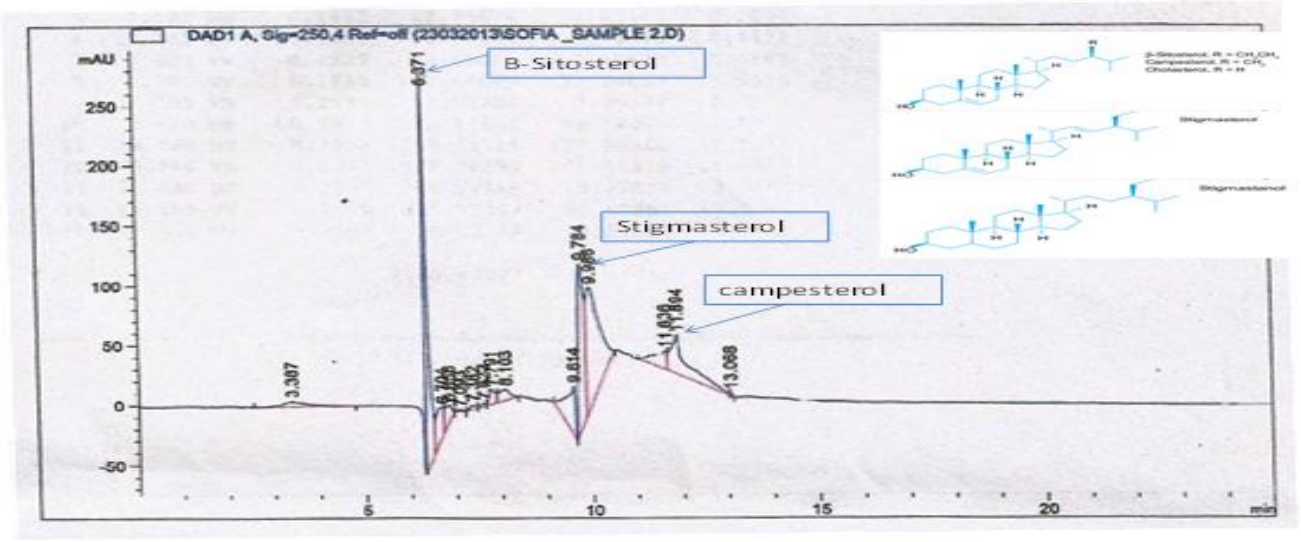

Fig-3: HPLC chromatogram of the phytosterol isolated from bamboo shoot

Table-5: Phytosterols content in fresh bamboo shoots, traditionally fermented bamboo shoots (*soibum*) and scientifically modified laboratory fermented samples.

Name of the sample	Concentration of total phytosterol (% dry wt.)
1.Fresh bamboo shoot samples	0.15±0.08
2.Traditionally fermented bamboo shoot samples	0.65±0.21
3. Scientifically modified laboratory fermented shoot samples	0.33.±0.04

Data presented as mean ± SD.

Cyanogenic glycoside

Cyanogenic glycosides estimation was done using the technique of Picrate impregnated paper according to Bradbury *et al*., 1999. The liberation of HCN is indicated by the colour change of picrate paper from yellow to reddish or red brown colour in proportion to the amount of HCN released. Absorbance was measured at

510nm and the total cyanide content was determined by preparing standard curved from potassium cyanide. Changes in the cyanogenic content during fermentation of the bamboo shoot slices were conducted. The weekly analysis on the hydrogen cyanide content assessed in the laboratory fermentation (90 days) with the bamboo shoot slices of *Bambusa balcooa* shows a decreasing trend of hydrogen cyanide level (Fig.2). In all fermentation it shows a degradation of HCN content with the advance of fermentation. Since HCN are highly volatile, the loss of HCN during the fermentation processes like peeling, slicing, cutting, repeated washing (3-4 times) is quite rapid. This may explain the reason that though bamboo shoots may contain significantly higher levels of HCN, however, the HCN content is reduced substantially to non toxic level for safe consumption.

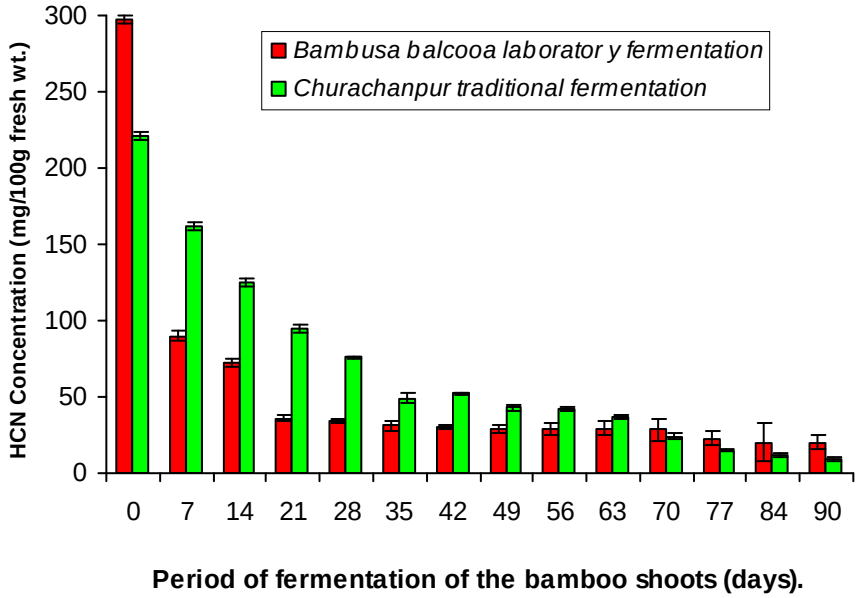

Fig-2: Changes in the cyanogenic glycosides (HCN) content during fermentation of the bamboo shoot slices of *Bambusa balcooa.* (Laboratory fermentation and traditional fermentation)

Conclusions

The present study concludes that bamboo shoots both fresh and fermented form can be of good nutraceutical source as they contain good amount of phenolic , flavonoids, tannin and phytosterols and other nutritive food value . They are low in calories and cyanogens content and have significant amount of phenolics, flavonoids, anti-oxidant properties. Several studies indicated that the antioxidant activities of some plants are highly correlated with their phenolic contents and have a correlation with antioxidant properties. Therefore, the bamboo shoots can also be used for formation of natural antioxidants. Bamboo shoots have high phytosterol contents , hence bamboo shoots are regarded as potential sources of sterols. Phytosterols have received particular attention due to their capability to lower serum cholesterol levels in humans, resulting in significant reduction of the risk of cardiovascular diseases. Furthermore, they were also regarded as a kind of natural product with many health benefits such as anti-inflammatory, anti-bacterial and anti-carcinogenic properties etc. Bamboo shoots were a kind of phytosterol-rich health food and the representative compounds found in bamboo shoots were β-sitosterol, campesterol, stigmasterol as the major sterol. According to these and above findings bamboo shoots can be portrayed as a rich potential source of nutaceuticals which will be of interest to the food and pharmaceutical industries.

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Morphological, phytochemical and therapeutic efficacy of *Citrus macroptera* Montruz

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Abstract

Citrus macroptera Montruz*., wild orange, is locally called as heiribob in Manipur, North East India. They are rich source of flavonoids, ascorbic acid, and antioxidant. They are traditionally used for curing stomach pain and alimentary disorder. Other local therapeutic uses, bioactivities and genetic diversity analysis are also reviewed. This wild fruit happens to be one of the most income generating fruit under Citrus family in Manipur. As this plant is less studied scientifically, biotechnological interventions can be applied in different aspects. Various compounds have been reported from the essential oil of C. macroptera but is not reported from extract yet. They have antidiabetic, antimicrobial, cardioprotective, lipid lowering and neuroprotective potential. Their cardioprotective, hepatoprotective, and neuroprotective efficacy hints the safety of Citrus macroptera Montruz for use as dietary supplement and phytomedicine. Further extensive researches on wider range on prospecting for therapeutic resources are encouraged.*

Keywords: Citrus macroptera, therapeutic, ethnopharmacological uses, phytochemistry, economic importance

INTRODUCTION

Citrus macroptera Montr. is a semi-wild species of the Rutaceae family and the citrus genus (Dreyer and Huey, 1973), Fig 1A and B.There are two widely used classification systems in Citrus (Swingle, 1943; Swingle and Reece, 1967). The genus Citrus L. belongs to the subtribe Citrineae, the tribe Citreae within the subfamily Aurantioideae of the Rutaceae family (Webber, 1967). The Swingle system included 16 species under two subgenera – Citrus and Papeda, while the Tanaka system recognized 162 species under the subgenera Archicitrus and Metacitrus. Advanced studies based on biochemical and morphological characterization, suggest that there are only three basic species, i.e. citron (*C. medica* L.), mandarin (*C. reticulate* Blanco), and pummelo (*C. maxima* (Burm.) Merr.) within the subgenus Citrus and that the other edible citrus, e.g. lemon, lime, sour orange, sweet orange, grapefruit, etc. are apomictically perpetuated biotypes with probable hybrid origin (Scora, 1975; Barrett and Rhodes, 1976). Citrus is thus believed to have its primary center of origin in North east India, China, Malaysia and Australia (Swingle and Reece, 1967; Scora, 1975; Gmitter and Hu, 1990)

Citrus flavo-noids have potential antioxidant (prevents aging), anti-cancer, antiviral, anti-inflammatory activities, effects on capillarity, and cholesterol-lowering ability (Stone, 1985). Citrus fruits are well-known for their dietary, nutritional, medicinal and cosmetic properties and are also good sources of citric acid, flavonoids, phenolics, pectins, limonoids, ascorbic acid, etc. (Dugo and Di Giacomo, 2002). *Citrus* fruits, including oranges, lemons, limes and grapefruits, are a principal source of such important nutrients, which are suggested to be responsible for the prevention of degenerative disease. These include vitamins C, folic acid, carotenoids, dietary fibres, potassium, selenium and a wide range of phytochemicals (Silalahi, 2002). Mabberley, (2004) treated Citrus in a broader sense by merging three of its closely allied genera – Fortunella Swingle, Eremocitrus Swingle and Microcitrus Swingle within it.

Most of the literature says plant extracts having anti-oxidant activities have health promoting effects, anti-ageing effects and used for various metabolic and chronic disease like cancer, liver diseases, inflammation, diabetes, arthritis, stroke (Willcox *et al*., 2004; Fusco *et al.*, 2007). A great number of epidemiological studies have shown that *Citrus* fruit consumption is protective in a variety of human cancers. Citriculture as a garden industry existed for centuries in India. The indigenous genetic resources of Citrus have great utility in citriculture and citrus industry. Unfortunately, like in many other crop plants, the genetic base of indigenous and wild species of Indian Citrus is being eroded due to habitat destruction, introduction of new exotic cultivars/varieties, and lack of appropriate conservation and management strategies (Kumar *et al*., 2010). It comprises the third largest fruit industry after mango and banana and occupies about 7.5% of land under fruits (Sidana *et al.*, 2013).

Sidana reviewed (Sidana *et al*., 2013) on Citrus and reported that polymethoxylated flavones of Citrus have been shown in numerous *in vitro* studies to exert strong anti-proliferative action against cancer cells, antigen activated T lymphocytes, gastric cancer cells, prostate cancer cells, squamous cell carcinoma, and ant metastatic actions against human breast cancer cells, protective cardiovascular, ant hyperglycemic, anti-inflammatory, anti-allergic, analgesic, anti-feedent, antioxidant, antibacterial, antifungal, antiviral activities. Citrus species are small to medium-size shrubs or trees that are cultivated throughout the tropics and subtropics regions of India and southern China to northern Australia and New Caledonia (Asif, 2014).

In the United States, citrus is suggested as part of a healthy diet because of its high vitamin C content and its lycopene and flavonoids, which are known to reduce prostate and breast cancer risk, reduce viral effects and inflammation, and improve capillary activity and choles-terol levels, recent research has focused on the biological activity of compounds found in citrus species, including compounds called flavanoids, carotenoids and limonoids, especially in terms of their effects on citrus palatability and anti-cancer activity (Asif, 2014). Lyco-pene-containing fruits and vegetables have been shown to contribute to a significant reduction in prostate and mammary cancer risk (Asif, 2014). Citrus fruit juices can be given in colds and fever as a refrigerant and as a source of vitamin C. The fruit juices are also helpful as an astringent for sore throats.

CLASSIFICATION AND MORPHOLOGICAL CHARACTERISATION

C. macroptera Montruz. – Melanesian Papeda were reported to occur in the subtropical forests of North-east India and the foot hills of the East Himalayas (Tanaka, 1937; Bhattacharya and Dutta, 1956). *Citrus macroptera* (Var annamensis) belongs to the family of Rutaceae and it is native to the regions of Southeast Asia mainly Myanmar, Thailand, Indonesia Malaysia, Papua New Guinea, Sylhet Division of northeastern Bangladesh and northeastern India mainly Manipur and Assam, local in Bengali it is called "hatkora" or "shatkora" and in English known as Wild orange (Carpenter and Reece 1969; Dreyer and Huey 1973). It is called as heiribob in Manipuri. It is a semi wild species and used as medicine by local tribes of Assam, India (Ghosh, 1990). Nair and Nayar, (1997); Sharma *et al.*, (2004) reported that *C. macroptera* Montr. occur in the subtropical forests of North-east India and the foot hills of the East Himalayas.

Classification

Kingdom	Plantae
Division	Tracheophyta
Subdivision	Spermatophytina
Class	Magnoliopsida
Order	Sapindales
Family	Rutaceae
Genus	*Citrus*
Species	*macroptera*

Morphological characteristics

Leaf

Division	simple
Lamina Shape	orbicular
Lamina Length	6.19 cm
Lamina Width	4.5 cm
Lamina Attachment	longipetiolate
Margin	entire
Apex	acuminate
Petiole Length	9.22 cm
Petiole Wing	present
Petiole Wing Wide	broad
Petiole Wing Shape	obovate
Junction Between Petiole and Lamina	articulate

Fruit

Weight	578.26 gm
Diameter	8.33 cm
Length	9.32 cm
Shape	Spheroid
Base Shape	Convex
Apex Shape	rounded
Skin Texture	smooth

Segment	17 nos.
Segment Shape Uniformity	Present
Rind Colour	White
Rind Thickness	1.08 cm
Pulp Colour	white
Pulp Colour Intensity	light
Axis	solid
Axis Shape	oval
Axis Diameter	0.67 cm
Oil Density (sq.cm)	86.25
Oil Gland Nature	moderately conspicuous
Areola	absent
Areola Diameter	0 cm
Style Scar	absent

Seed

Shape	semi-deltoid
Surface	wrinkle
Colour	White
Length	1.57 cm
Width	0.66 cm
Cotyledon Colour	white
Average Seeds	15-23

Adapted from Sanabam *et al*., 2012.

There is no report of extensive research on *C. macroptera* about its Phytochemistry, pharmacological and therapeutic properties, although some leiterature is available and reviewed. In Manipur this Wild Orange (*C. macroptera*) grows well with good quality fruiting as location specific crop of Chandel District and Jiribam Sub-Division of Imphal East District, although all Citrus are primarily valued for the fruit, which is either eaten alone (sweet orange, tangerine, grapefruit, etc.) as fresh fruit, processed into juice, or added to dishes and beverages (lemon, lime, etc.); however this Wild Orange has wide range of uses viz. the dried rind of the fruit as flavouring spice in preparation of meat dishes, the juice of the fruit as medicine for treatment of stomach ailments as well as digestive enzyme, the fruit pulp as washing detergent, not the least the most important is the essential oil from the leaves which otherwise wasted. (http://manipursfac.com/wild-orange-citrus-macroptera/ 1/.)

TRADITIONAL AND LOCAL USES

In Guam, Stone (Stone, 1985) noted that the pulp was used for washing clothes and hair The rinds of ripe citrus fruits can be used in decoction for coughs, colds, indigestion and diarrhea. A decoction of the leaves is used as a bath to induce sweating in patients with fever. The macerated pulp and leaves of wild orange were used as a shampoo in Guam, Samoa, and Fiji (Walter and Sam, 2002). The seeds are also said to have pain-relieving effects. The fruit of *Citrus macroptera* (var annamensis) is edible and popular among the people of Bangladesh, Meghalaya and Assam of India as green matured fruits, used in cooking for flavoring curry mainly meat dishes, pickle preparation and oil is used in perfume production (http://gsl.articlealley.com/an-introduction-of-satkara-1932990.html).

Malik *et al.*, 2006 reported *Shatkara* as an endangered wild species, used by locals in Northeast India as medicine for stomach pain and alimentary disorder. The traditional healers of Manipur give peels of this

fruit to the person suffering from epilepsy to smell to recover from epileptic state. *Citrus macroptera* Montr. was used in remedies against ringworm, complex remedies against sicknesses; epilepsy-like symptoms, antiprotozoal and nematocidal activities of some medicinal plants from New Caledonia (Desrivot *et al*., 2007). In Bangladesh the Laleng (Patra) historical indigenous community call *C. macroptera* as Jamir uses: loc. Kushirgool: and their fruits are edible (Partha, 2014). The fruit is typically used during cooking and for pickle preparation and is popular for its medicinal purposes in Assam, India (Bose and Mitra 1990).

In Manipur this Wild Orange (*C. macroptera*) grows well with good quality fruiting as location specific crop of Chandel District and Jiribam Sub-Division of Imphal East District, although all Citrus are primarily valued for the fruit, which is either eaten alone (sweet orange, tangerine, grapefruit, etc.) as fresh fruit, processed into juice, or added to dishes and beverages (lemon, lime, etc.); however this Wild Orange has wide range of uses viz. the dried rind of the fruit as flavouring spice in preparation of meat dishes, the juice of the fruit as medicine for treatment of stomach ailments as well as digestive enzyme, the fruit pulp as washing detergent, not the least the most important is the essential oil from the leaves which otherwise wasted. (http://manipursfac.com/wild-orange-citrus-macroptera/ 1/.). This species grow in sloppy and high altitude area. The peel is dried and kept in every household as essential spice ingredients for cooking. The juice is used for treating and flushing stone from Kidney.

RANDOM AMPLIFIED POLYMORPHIC DNA (RAPD) ANALYSIS

Citrus macroptera Montr. (Melanesian papeda) is one of the wild and endangered economically important species of northeast India. Chaudhury and Bhat (2013) used RAPD markers to evaluate genetic diversity and inter-relationship among 30 accessions of *C. macroptera* collected from Tripura and Mizoram states under Indian cintinent. They generated a total of 92 bands in *C. macroptera* based on 12 RAPD primers, of which 91 bands (98.8%) were polymorphic; a pair-wise genetic similarity value between the accessions ranged from 0.26 to 1.00 with an average value of 0.57. Their study indicates the existence of high level of genetic diversity among the accessions of *C. macroptera* collected from different geographical regions. Unweighted pair-group method for arithmetic mean dendrogram separated all the accessions of *C. macroptera* into two major clusters with similarity value of 0.53 and various sub-clusters. Grouping of accessions was broadly based on genetic similarity among the accessions rather than geographical distribution. Based on the marker analysis, accessions collected from Mizoram were found to be more diverse as compared to those collected from Tripura. The RAPD markers confirmed the distinction between close accessions, inter-relationship and ability to detect genetic variation among the accessions of *C. macroptera*, which has implications for genetic improvement of citrus germplasm (Chaudhury and Bhat 2013).

COMPOUNDS OF *CITRUS MACROPTERA* Montr

There was report on isolation of coumarins like bergamottin, psoralen, marmin, severine and geiparvarin (Dreyer and Huey, 1973). Bergamottin and analogues are also constituents of non-commercial citrus fruits such as *Citrus macroptera*, a species native to islands of the S. Pacific (Dreyer and Huey, 1973). The distillation and subsequent extraction of the distillate obtained from the fresh peels of the fruits of *Citrus macroptera* var. *annamensis* afforded 120 mg of oil (yield 0.12 %). Previous phytochemical investigations resulted in the isolation of alkaloids like (+) ribalinine and isoplaty desmine, (Gaillard *et al*., 1995) aromatic compounds like cinnamic acid, syvingaldehyde, vanilline and methyl vanillate (Gaillard *et al*., 1995). The two alkaloids (Kokusaginine and rybalinine) are of the quinolone type and have not been found in Ruta species, although they have been found in other plants, e.g. *Haplophyllum patavinum* and *Citrus macroptera* Montr. (Gaillard *et al*., 1995). The structures are presented in Fig.2. The isolation and structure elucidation of the Lupeol and Stigmasterol by using spectroscopic techniques and the preliminary antioxidant activities of the organic extractives are being reported (Chaudary *et al*., 2008).

The leaves and flowers together with those of *Colubrina asiatica* and *Citrus macroptera* were used to make shampoo in Samoa (Thomson and Thaman 2008). Chowdhury *et al*., (2008) reported the presence of Lupeol and Stigmasterol in leaves of *C. macroptera*. GC/MS Analysis of the *C. macroptera* leaves essential oil allowed the identification of 35 compounds. The oil was mainly constituted of monoterpenes (96.3%), among which b-pinene (33.3%), a-pinene (25.3%), p-cimene (17.6%), (E)-ocimene (6.7%), sabinene

(4.8%), g-terpinene (3.1%), and limonene (2.4%). Moreover, spathulenol (0.6%) and caryophyllene oxide (0.5%) were the major oxygenated sesquiterpenes present in the oil. Other sesquiterpenes such as a-cardinol (0.2%), viridiflorol (0.2%), and a-muurolol (0.1%) were also identified (Waikedre et al 2010). Analyses both by gas chromatography (GC) and gas chromatography- mass spectrometry (GC-MS) of the sample resulted in the identification of 25 terpenoids, predominantly monoterpene hydrocarbons, accounting for 97.0 % of the total oil, and limonene (73.5 %) as the major component (Miah *et al*., 2010). Their leaves contain mainly terpenoids like limonene and aromatic hydrocarbons (Waikedre *et al*., 2010; Rana and Blazquez 2012). The oil of *C. macroptera* contained limonene (55.3%), beta-caryophyllene (4.7%) and geranial (3.5%) as main compounds (Rana and Blazquez, 2012).

A paste of *C. macroptera* leaves are applied to the navel for children, and pills prepared from leaf paste are taken orally to cure helminthiasis (Nahar *et al*., 2013).

A **B**

Fig-1: **A.** Leaves, fruits and seeds of *Citrus macroptera*. Fig. 1 **B.** Rind of *Citrus macroptera*

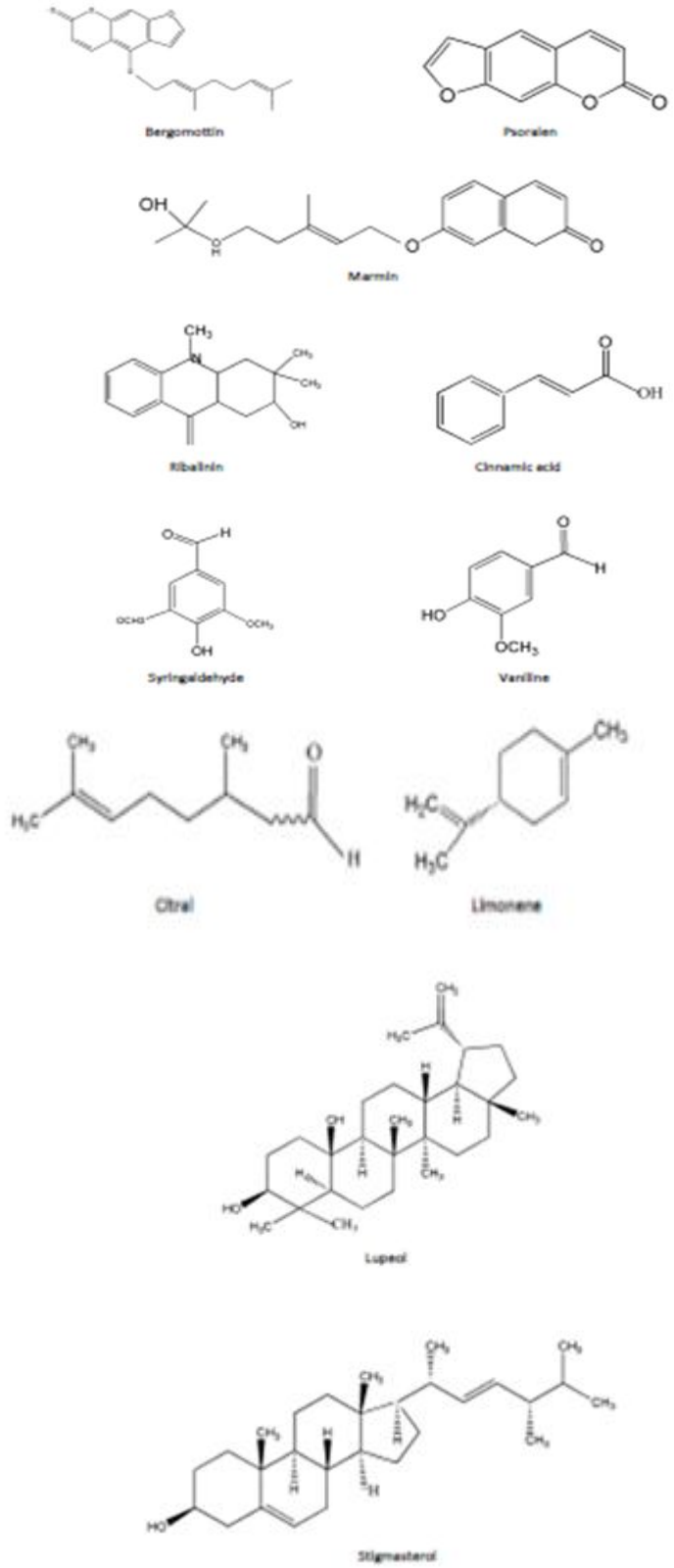

Fig-2: Compound report in *Citrus macroptra montruz*

BIOLOGICAL ACTIVITIES

Antioxidant activity

It is reported that stem bark of *Citrus macroptera* posses antioxidant activity (Chowdhury *et al*., 2008). The hot methanol extract of the stem bark of *Citrus macroptera* showed potential antioxidant activity with the

IC50 value of 178.96 µg/ml whereas the cold methanol and the dichloromethane extracts showed moderate activity with the IC50 of 242.78 µg/ml and 255.78 µg/ml respectively. The n-hexane extract showed mild activity (IC50: 422.94 µg/ml) against DPPH free radical. It is evident that all possess antioxidant activity (Chowdhury et al., 2008). Miah *et al*., (2010) reported that the oil did not exhibit any *in vitro* free-radical-scavenging (DPPH). Both time and solvent extractions played a vital role in the extraction of phenolic contents and their antioxidant properties of *C. marcoptera* (Gope *et al*., 2014). Rahman *et al*., (2014a) reported that *Citrus macroptera* fruit peels posses anti-oxidant activities. The order of activities were ethanol> n-hexane> chloroform extracts of *Citrus macroptera* fruit peels. Ethanolic extract of *Citrus macroptera* fruit peels (EECM) in DPPH scavenging activity (IC50 281.11 µg/ml), Hydrogen Peroxide scavenging activity (IC50 216.49 µg/ml), Nitric Oxide scavenging activity (IC50 182.89 µg/ml) were comparable with standard Ascorbic acid. The total Phenolic content was highest in EECM (142.5±3.29 mg/gm Eq of Gallic Acid) and Total Flavanoid content was highest in EECM (333.0±36.06 mg/gm Eq of Quercetin.

Hepatoprotective activity

Liver function was evaluated by determining the activities of various serum enzymes (ALT, AST, ALP, GGT and LDH), metabolites (TB) and plasma proteins (TP and ALB) (Paul et al., 2015). During hepatic injury, enzymes tend to leak out into the blood stream because of their cytoplasmic location, which facilitates their release into circulation in response to damage of liver structural integrity (Afroz *et al*., 2014). The extract tended to produce different effects on liver enzymes. There were no significant changes in plasma ALT and GGT levels, indicating that the extract did not exert any toxic effects in the liver. The highest dose (1000 mg kgG1) was even found to significantly decrease AST enzymatic activity, indicating that the fruit may exert protective effects in the liver (Paul et al., 2015). Citrus macroptera also possessed nephroprotective activity against Acetaminophen-Induced Hepatorenal Toxicity in Rats (Paul et al., 2016)

Cardioprotective activity

Dose-dependent reductions were observed in serum levels of total cholesterol, triglyceride, very low density lipoprotein cholesterol and low density lipoprotein cholesterol in response to exposure to the fruit pulp, suggesting its lipid lowering activity and cardioprotective effect (Uddin *et al*., 2014b). Additionally, a dose-dependent incremental change in serum HDL-C was observed. Because a high level of high density lipoprotein cholesterol in plasma exerts a protective effect via reverse cholesterol transport because it scavenges excess cholesterol from the peripheral tissues of the body (Uddin *et al*., 2014a), the effects of Citrus macroptera pulp would certainly be beneficial. Therefore, these findings provide strong support in favor of the cardioprotective effect of the fruit (Paul et al., 2015).

Hypoglycemic activity

The therapeutic effects of methanol extract of *Citrus macroptera* fruit in α-amylase inhibitory activity (*in vitro*) and hypoglycemic activity in normal and glucose induced hyperglycemic rats (*in vivo*) were investigated and presence of saponin, steroid, terpenoid were identified. The fruit extract showed moderate α-amylase inhibitory activity as compared to acarbose (Uddin *et al*., 2014a). *Citrus macroptera* extracts have been reported to exert anti-diabetic effects, possibly by reducing fasting blood glucose and serum insulin levels, which has been shown to alleviate hyperglycemia-associated oxidative stress in experimental type 2 diabetic rats (Zheng *et al*., 2012). These effects are thought to occur via the saponins, steroids and terpenoids that have been identified in *C. macroptera* (Uddin *et al*., 2014a).

Neuroprotective activity

There is little published work on the neuroprotective effects of herbal extracts or natural phytochemicals having anti-oxidant activities. Phytochemicals containing flavonoid polyphenols and organosulfur compounds have neuroprotective effects, as shown experimentally in cell and animal studies (Parihar and Hemnani 2003; Heo and Lee 2005). There is little literature available for neuropharmacological activities of *Citrus macroptera* (Varannamensis). Ethanolic extract of *Citrus macroptera* fruit peels (EECM) found to posses anti-depressant and anxiolytic activity, it was found to protect from oxidative stress in brain and it was found to protect brain antioxidant enzyme levels in *in-vivo* (Rahman *et al*., 2014b). The phytoconstituents of *C. macroptera*, such as polyphenols, flavonoids and organosulfur compounds have also been reported to contribute

to its neuropharmacological effects (Yip and Dallman, 1988; Dallman *et al*., 1980).

It can be concluded that neuroprotective activity of *Citrus macroptera* fruit peels may be due to protection from oxidative stress and it may prove the tradition uses of *Citrus macroptera* fruit peels for anxiety and depression in Assam (Rahman *et al*., 2014b).

Antimicrobial activity

According to the literature, many monoterpenoids identified in the essential oils of *C. macroptera* such as limonene or p-cymene, are known as anticandidals (Pauli, 2006). Essential oil obtained from leaves posses antimicrobial activity and traditionally fruits as appetite stimulant activity (Waikedre *et al*., 2010). It showed considerable antibacterial activity against *Bacillus cereus*, *B. subtilis*, *Escherichia coli* and *Staphylococcus aureus* with the MIC values ranging from 1.25 to 5.0 mg/mL (Miah *et al*., 2010). The MIC values of ketoconazole and the *C. macroptera* essential oil against *T. mentagrophytes* var. interdigitale (filamentous fungus) were 5 and 12.5 mg/ml, respectively, and against *Candida albicans* were 40, 75 mg/ml, respectively (Waikedre *et al*., 2010). As contrast to the finding of Mia *et al*., (2010), the essential oils of leaves of *C. macroptera* have no antibacterial activity against all five bacteria viz., *Staphylococcus aureus* ATCC 6438, *Staphylococcus epidermidis* ATCC 12228, and *Bacillus subtilis* ATCC 6633, and two Gram-negative strains, viz., *Escherichia coli* ATCC 10536 and *Klebsiella pneumoniae* ATCC 13883 (Waikedre *et al*., 2010).

Lipid lowering activity

In vivo studies confirmed that *Citrus macroptera* pulp possessed a significant lipid lowering activity that occurred in a dose dependent manner and that it caused beneficial changes in several other biochemical parameters. Additionally, a significant diminution of lipid peroxidation in liver and kidney tissues was observed.

The toxic effects of the methanol extract of *Citrus macroptera* Montr. Fruit was assessed via Biochemical and Hematological Evaluation in Female Sprague-Dawley Rats and the evaluated non-toxic effect suggests a wide margin of safety for therapeutic doses (Uddin *et al*., 2014b). The low dose of the fruit extract reduced creatinine levels; however, no significant changes were observed with the administration of higher doses, indicating that the fruit extract is not renal toxic. The extract also did not significantly affect other kidney parameters, such as urea and uric acid, even when it was administered at high doses. Therefore, this fruit extract was considered to be nontoxic and it exhibited no destructive effects on normal kidney functions (Paul et al., 2015).

ECONOMIC IMPORTANCE

Every year, Bangladesh earns handsome amount of foreign currency by exporting these fruits especially to UK, USA and Middle East countries, like other citrus species Sat Kara is also propagated conventionally by means of seeds, grafting and budding methods (Miah *et al*., 2002). During the season per fruit cost Rs.15-20 but when fruiting season is over the cost is doubled or sometimes more than double (http://manipursfac.com/wild-orange-citrus-macroptera/ 1/). Due to present demand of this fruit in both local and foreign markets it is necessary to develop a suitable protocol for mass propagation from existing elite cultivars (Miah *et al*., 2002). It is a costly species, the price ranges upto Rs.100 per fruit in off seasons in Manipur, a state in North East India. The cultivation of *C. macroptera* is the sole livelihood income generator of some tribal areas and the economy of some villages entirely depended on it.

CONCLUSION

C. macroptera Montr. has limited information on bioactivities. Till date, they are reported to be having antioxidant, antidiabetic, antimicrobial, cardioprotective, lipid lowering potential. As already proven to be associated with antioxidant activities and it does not exert toxic effect on liver and kidney as evident from the cited research works, thus they are safe for human consumption. Thus, *C. macroptera* Montr. may be a good plant candidate for bioprospecting the therapeutic agents for anticancer, anti-Alzheimer's and anti-inflamatory resources with extensive research on wider range.

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Spirulina platensis as a Nutritional Supplements

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Abstract

Blue-green algae are among the most primitive life forms on Earth. Their cellular structure is a simple prokaryote. They share features with plants, as they have the ability to perform photosynthesis. They share features with primitive bacteria because they lack a plant cell wall. Interestingly, they also share characteristics of the animal kingdom as they contain on their cellular membrane complex sugars similar to glycogen. Among blue-green algae, both edible and toxic species adapted to almost any of the most extreme habitats on earth. Edible blue-green algae, including Nostoc, Spirulina and Aphanizomenon species have been used for food for thousands of years. Spirulina are multicellular and filamentous blue green algae that has gained considerable popularity in the health food industry and increasingly as a protein and vitamin supplement to aquacultures diets. It grows in water, can be harvested and processed easily and has very high macro and micro nutrient contents. Several animal studies in vivo and in vitro followed by human trial have proved the importance of Spirulina to be commercialized and sold for therapeutic purposes. Spirulina appear to have considerable potential for development especially as a small scale crop for nutritional enhancement, livelihood development and environmental mitigation.

Keywords: Spirulina platensis, Zarrouk's medium, Mass cultivation, therapeutic

INTRODUCTION

This little microscopic organism has attracted people and scientists from all over the world due to its special properties. *Spirulina* has found wide applications in agriculture, food, pharmaceuticals, perfumeries, medicine and science. It is also used as a food supplement and is marketed in the form of pills, capsules and powder or incorporated into various types of food like cakes, biscuits, noodles and health drinks, etc. (Hayashi *et al*., 1996). The microorganism called "*spirulina*" was so named because of its spiral filament under the microscope, classified as cyanobacterium. *Spirulina*, now named *Arthrospira*, is a blue-green alga with an old history. Its name derives from the spiral or helical nature of its filaments (Belay, 2002).

Spirulina (Arthrospira) has a high protein content and excellent nutrient value (Spolaore et al., 2006) and has gained worldwide popularity as a food supplement (Colla et al., 2007). It has an amino acid content of 62% and it is a rich natural source of vitamins A, B1, B2, B12, as well as phytopigments, including carotenoids and xanthophylls (Richmond, 1988). In addition, it has a considerable amount of essential fatty acids and linolenic acid, which cannot be synthesized by humans (Becker, 1994); thus, more attention has been given to the cultivation of *Spirulina*. The world production of *Spirulina* for human consumption exceeds 1000 metric tons, annually (Ciferri and Tiboni, 1985). The world's largest *Spirulina* producer is Hainan Simai Enterprising, which is located in China and has an annual algal powder production of 200 tons. There are 20 countries in the world that produce *Spirulina*-based products, such as tablets and powder. Among these countries, the USA ranks first in *Spirulina*-based products, primarily in the form of pills and spray-dried powder, followed by China, Israel, Japan, Mexico, Taiwan and Thailand (Spolaore et al., 2006). From microalgae, a wide variety of nutraceuticals are available and marketed for sale. For example, the Myanmar-*Spirulina*-factory (Yangon, Myanmar) produces tablets, chips, pasta and liquid extracts. Similarly, Cyanotech (Hawaii, USA) produces pure powder under the name ''*Spirulina pacifica*". Phycocyanin, extracted from *Spirulina* and commercially known as 'lina blue', is used as a blue colorant for food and cosmetics. *Spirulina* also acts as a functional food, feeding beneficial intestinal flora, including *Lactobacillus* and Bifidus (Ciferri, 1983).

In the future, these health foods are expected to be a stable market, including products such as *Spirulina* liquid CO_2-extracted antioxidant capsules (Belay et al. 1993). One gram of *Spirulina* contains one-half of the adult daily requirements of Vitamin A. According to human studies, total serum cholesterol was lowered by consuming *Spirulina* (Gonzalez de Rivera et al., 1993). The purified PUFAs from *Spirulina* are added into infant milk formulas in European countries for health promoting purposes.

Spirulina has been used as a complementary dietary ingredient of feed for fish, shrimp and poultry and increasingly as a protein and vitamin supplement to aqua feeds (Orio Ciferri and OrsolaTinoni, 1985). China is using this micro alga as a partial substitute of imported forage to promote the growth, immunity and viability of shrimp. There has also been comprehensive research on the use of *Spirulina* as aquaculture feed additives in Japan. During the sixtieth session of the united nations general assembly, a revised draft resolution on the "Use of *Spirulina* to combat hunger and malnutrition and help achieve sustainable development" was submitted by Burundi, Cameroon, Dominican Republic, Nicaragua and Paraguay. As follow up of this resolution, FAO was requested to prepare a draft position paper on *Spirulina* so as to have a clearer understanding on its use and to convey FAO position on this. *Spirulina platensis* has been used as food for centuries by different populations and only rediscovered in recent years. Once classified as the "blue-green algae", it does not strictly speaking belong to the algae, even though for convenience it continues to be referred to in that way. China is using this micro alga as a partial substitute of imported forage to promote the growth, immunity and viability of shrimp. There has also been comprehensive research on the use of *Spirulina* as aquaculture feed additives in Japan. During the sixtieth session of the United Nations general assembly, a revised draft resolution on the "Use of *Spirulina* to combat hunger and malnutrition and help achieve sustainable development" was submitted by Burundi, Cameroon, Dominican Republic, Nicaragua and Paraguay. As follow up of this resolution, FAO was requested to prepare a draft position paper on *Spirulina* so as to have a clearer understanding on its use and to convey FAO position on this. It grows naturally in the alkaline waters of lakes in warm regions (Pelizer et al., 2002). Measuring about 0.1mm across, it generally takes the form of tiny green filaments coiled in spirals of varying tightness and number, depending on the strain. Its impressive protein content and its rapid growth in entirely mineral

environments have attracted the attention of both researchers and industrialists (Mitchell et al., 1990). *Spirulina* are unicellular and filamentous blue-green algae that has gained considerable popularity in the health food industry and increasingly as a protein and vitamin supplement to aquaculture diets. It has long been used as a dietary supplement by people living close to the alkaline lakes where it is naturally found. *Spirulina* has been used as a complementary dietary ingredient of feed for fish, shrimp and poultry. Among the various species of *Spirulina*, the blue green alga *Spirulina platensis* has drawn more attention because it shows a high nutritional content characterized by 70% protein content and by the presence of minerals, vitamins, amino acids, essential fatty acids etc (Kawata et al., 2004).

Spirulina platensis is naturally found in tropical regions inhabiting alkaline lakes (pH 11) with high concentration of NaCl and bicarbonates. These limiting conditions for other microorganism allow cultivation of microalgae in opened reactors (Harriet et al., 2008). In cyanobacteria, the light harvesting pigments include chlorophyll-a, carotenoids and phycobiliproteins. The later are proteins with linear tetrapyrrole prosthetic groups called according to their structure: phycocyanin, phycoerythrin and allophycocyanin. Among the various microorganisms used as sources of SCP, the blue green algae, *Spirulina* is considered as the best source. The composition of the biomass, including the high protein content, low content in nucleic acids, occurrence of high concentrations of vitamins and other growth factors and the presence of cell wall that is more easily digestible than that of other microbes indicate that *Spirulina* is a promising source of food or feed. *Spirulina* is commonly found in aquatic ecosystems like lakes, ponds and tanks. It is one of the nature's first photosynthetic organisms capable of converting light directly for complex metabolic processes. *Spirulina* is used for food from time immemorial by tribes living around Chad Lake in Africa. The predominant species of phytoplankton of the lake is *Spirulina platensis*. The algae *Spirulina* was eaten in Mexico under the names 'Tecuitlatl' (Farrar 1996). *Spirulina* grows optimally in pH range of 9-11 and there is least chance of contamination of other microbes (Supramaniyan and Jeeji Bai 1992). *Spirulina* can play an important role in human and animal nutrition, environmental protection through wastewater recycling and energy conservation.

Spirulina is rich in proteins (60-70%), vitamins and minerals used as protein supplement in diets of undernourished poor children in developing countries. One gram of *Spirulina* protein is equivalent to one kilogram of assorted vegetables. The amino acid composition of *Spirulina* protein ranks among the best in the plant world, more than that of soya bean. Gamma-linolenic acid contained in this alga have been reported to stimulate prostaglandin synthesis and induction of the regulation of blood pressure, cholesterol synthesis, inflammation and cell proliferation (Venkataraman, 1993; Borowitzka and Borowitzka 2010).The generic name "single cell protein (SCP) was designed in 1996 during an international meeting held at MLT, USA to include protein source from unicellular or multicellular microbes like bacteria, yeast, fungi and algae. The concept of utilizing SCP is not completely new, as protein are already being used in foods and feeds in different regions of the world at varying levels. However, it is a new development *Spirulina* provides all essential nutrients without excess calories and fats. It is recommended to control obesity and premenstrual stress. Athletes take *Spirulina* for instant energy. Many herbal cosmetics like face creams biolipstick, hair lotion have been formulated from phycocyanin pigment found in *Spirulina*. The beta carotene and other carotenoids are having a suggested role in the control of cancer in human and enhancement of pigmentation of eggs, meats and coloration of ornamental fish. The mass cultivation of *Spirulina* is achieved both in fresh water and waste water. *Spirulina* grown in clean waters and under strictly controlled conditions could be used for human nutrition. The micro alga grown in waste water is used as animal feed and provide a source of the fine chemicals and fuels. The waste water system is highly applicable in populated countries like India where wastes are generated in high quantities and pose environmental problem. Large scale production of *Spirulina* is feasible in tropical conditions in developing countries, where land costs and labour are comparatively cheaper. The micro alga can be exploited as a potential source of food, feed and fuel. The generic name "single cell protein (SCP) was designed in 1996 during an international meeting held at MLT, USA to include protein source from unicellular or multicellular microbes like bacteria, yeast, fungi and algae. The concept of utilizing SCP is not completely new, as protein are already being used in foods and feeds in different regions of the world at varying levels.

Morphology and taxonomy

Holmgren *et al*.,1971 observed cross walls in *Spirulina* major under electron microscope and suggested the transfer of *Spirulina* major into the genus *Arthrospira* on the criterion of the presence of cellular septation. *Spirulina* and *Arthrospira*, the non heterocyst unbranched filamentous genera of order *Nostocales* and family Osillatoriaceae wear identified. Presence of two filaments in a single unit and absence of cellular septation under light microscope wear the main point which differentiated *Spirulina* from *Arthrospira* (Gupta and Chagwal 1992). Hintak, 1985 described a *Spirulina fusiformis* from the Kenya with a fusiform trichome construction. He mentioned that a relatively small, inconspicuous, broadly rounded calyptra was occasionally formed with or without an accompanying tapering of the trichome. *Spirulina* consisted multicellular, filamentous, unbranched and helicoidel trichomes wear formed by a single spirally twisted cell. Motile structure like flagella and heterocysts which are generally present in many blue green algae wear absent. The filaments wear called 'trichome'. The cells wear cylindrical and the spiral wear loose. The cells exhibited active rotary movements. The helical shape of the trichome was characteristic of the genus but the helical parameters varied with the species even within the same species (Somasekaran, 1987). Hintak, 1985 described a *Spirulina fusiformis* from the Kenya with a fusiform trichome construction. He mentioned that a relatively small, inconspicuous, broadly rounded calyptra was occasionally formed with or without an accompanying tapering of the trichome. *Spirulina* consisted multicellular, filamentous, unbranched and helicoidel trichomes wear formed by a single spirally twisted cell. Motile structure like flagella and heterocysts which are generally present in many blue green algae wear absent. The filaments wear called 'trichome'. The cells wear cylindrical and the spiral wear loose. The cells exhibited active rotary movements. The helical shape of the trichome was characteristic of the genus but the helical parameters varied with the species even within the same species (Somasekaran, 1987). *Spirulina platensis* is a filamentous, photoautotrophic, alkaliphilic cyanobacterium that belongs to the Family Oscillatoriaceae, Division Cyanophyta. This cyanobacterium blooms in bicarbonate-rich environments and has gained a significant position in recent years as a source of proteins and pigments in the food, pharmaceutical, and cosmetic industries (Cifferi, 1983). Pelizer *et al*., 2002 reported that *Spirulina platensis* is generally produced in open ponds in liquid culture but there is recent production possible also In solid-state cultivation system but the estimation of cell growth is made difficult in separating cells from the cultivated medium incase of solid-state cultivation systems.

Nutritive value of *Spirulina platensis*

Nutrient value of *Spirulina* is one of the natural sources containing the highest amount of protein-five times that of meat. *Spirulina* provides the majority of essential and nonessential amino acids. It has a fairly well-balanced amino acid pattern and contains the highest amount of beta-carotene, a precursor of vitamin A. It is the only vegetable source of vitamin B12 having two and half times the amount in liver. It is also the source of the essential fatty acid γ-linolenic acid which is the precursor of hormones involved in regulation of body functions. The constituents of *Spirulina* include protein (50-70%) including all essential amino-acids, essential fatty acids, polysaccharides, B vitamins particularly vitamin B12, beta-carotene and minerals particularly iron (Khan et al., 2005). *Spirulina* has been demonstrated to be an effective dietary source of vitamin A. An investigation in India on preschool children with vitamin A deficiency demonstrated that the bioavailability of carotenes from. *Spirulina* was comparable to that from other sources such as carrots and green leafy vegetables thereby suggesting the potential use of *spirulina* as a dietary source of vitamin A (Annapurna et al., 1991).Clement *et al*. reported that *Spirulina maxima* were an alga rich in organic nitrogenous constituents, used for food in Chad Republic (Africa). Amino acids, vitamins and nutritive value were determined for a strain of the algal growth in an open air pilot production unit. It contained 62 per cent protein, high digestibility and vitamins like β-carotene, β1, β2, β6, β12 and C. Drum dried preparations of *Spirulina platensis* were fairly similar in quality to that of casein sample with methionine was reported by Omstedt *et al*. Wahal *et al*. reported that water-soluble sugars constituted the major carbohydrates of *Spirulina*. The low amount of starch was due to the high activity of α and β amylase in *Spirulina*. The *in vitro* digestibility of *Spirulina* had been reported using an amylase enzyme. A sufficient amount of protease activity indicated that the enzyme was mainly involved in protein turn over rather than in storage hydrolysis. Po Chung *et al*. studied that the production and nutritive value of *Spirulina platensis* on swine wastes. The alga contained 55 to 61 per cent crude protein. Three indoor culture ponds 0.65m^2

were designed and built under the light intensity of 500 foot candles. Natives of the chad region still supplements their diet with *Spirulina* harvested from lakes. They fed *Spirulina* to nourished adults. It showed positive nitrogen balance with no harmful effects. Ansuya Devi *et al*., reported the characteristics of which nearly 9.9 per cent was non-protein nitrogen. Total protein was extracted by three successive extractions with water. The isoelectric point was found to be 3.0. The polyacrylamide gel electrophoretic pattern showed seven bands. The *in vitro* digestibility was found to be 85 per cent when assayed with a pepsin pancreatin system. Krishna Kumari *et al*.1981 studied the protein of *Scendesmus acutus* and *Spirulina platensis*. These were fed to the rats upto the dosage of 800 mg kg^{-1} of body weight. The absence of gastrointestinal disorders such as diarrhea indicated that these algae were tolerated by animals even at 800 mg kg^{-1}. Application of these algae into albino rats did not elicit any skin allergy. Weight gains, PER and nitrogen balance studies with *Spirulina fusiformis* supplementation to poor rice diets showed significantly higher values of all parameters over casein supplementation (Krishnakumari, 1982).Orio Ciferri and Tinoni studied the biochemistry and industrial potential of *Spirulina*. The composition of the biomass recorded, high protein content, low nucleic acids content and the presence of cell wall that was more easily digestible than that of yeasts or eukaryotic algae. It's consumption by human populations indicated a lack of toxic activities. It could be used as a source of chemical and other basic commodities like enzymes, vitamins, lubricants, pigments, etc. Experiments on humans over an eight week period of *Spirulina* intake (4.2 g d^{-1}) showed greater reduction in serum cholesterol level, especially in subjects with higher initial levels and improved the atherosclerosis index. (Nahaya, 1986). Bonotto 1988 reported that many species of microalgae and blue green bacteria under suitable conditions grew faster and produced large biomass with high protein content. They constituted a source of single cell protein and the algal biomass supplemented or even replaced animal proteins, thus short circuiting the rather inefficient animal food chain. Mitchell *et al*. demonstrated that *Spirulina maxima* significantly altered the storage and utilization of vitamins A and E. These were investigated by feeding diets containing 0, 2.7, 10.7, 18.7 and 26.7 per cent protein or by a mixture of them. Growth results indicated that rats did not utilize the diets containing *Spirulina maxima* as well as the case in control diet. The ingestion of *Spirulina maxima* caused a significant increase in dry matter and chloroform extractable crude fat in the faeces.

Table-1: Phytonutrient of *Spirulina* (Koru, 2012)

SN	Phytonutrient	g/100g^{-1}
1	Cis-beta-carotene	0.073
2	Trans -beta- carotene	0.26
3	Phycocyanin	12
4	Chlorophyll-a	1

Clement *et al*. 1967 reported that *Spirulina maxima* were an alga rich in organic nitrogenous constituents, used for food in Chad Republic (Africa). Amino acids, vitamins and nutritive value were determined for a strain of the algal growth in an open air pilot production unit. It contained 62 per cent protein, high digestibility and vitamins like β-carotene, β1, β2, β6, β12 and C. Rats were fed six consecutive days on a diet containing 20 per cent protein level. Lyophilized *Spirulina platensis* had a nutritional quality between that of wheat gluten and casein supplemented with methionine. Drum dried preparations of *Spirulina platensis* were fairly similar in quality to that of casein sample with methionine was reported by Omstedt *et al* 1973.Wahal *et al*., 1974 reported that water-soluble sugars constituted the major carbohydrates of *Spirulina*. The low amount of starch was due to the high activity of α and β amylase in *Spirulina*. The *in vitro* digestibility of *Spirulina* had been reported using an amylase enzyme. A sufficient amount of protease activity indicated that the enzyme was mainly involved in protein turn over rather than in storage hydrolysis. Po Chung *et al*., 1978 studied that the production and nutritive value of *Spirulina platensis* on swine wastes. The alga contained 55 to 61 per cent crude protein. Natives of the chad region still supplements their diet with *Spirulina* harvested from lakes. They fed *Spirulina* to nourished adults. It showed positive nitrogen balance with no harmful effects. Ansuya Devi *et al*. 1981 reported the characteristics of the protein of fresh water mass cultures *Spirulina platensis*. The total protein was 50-65 per cent of which nearly 9.9 per cent was non-protein nitrogen. Total protein was extracted by three

successive extractions with water. The isoelectric point was found to be 3.0. The polyacrylamide gel electrophoretic pattern showed seven bands. The *in vitro* digestibility was found to be 85 per cent when assayed with a pepsin pancreatin system. Krishna Kumari *et al*. 1981 studied the protein of *Scendesmus acutus* and *Spirulina platensis*. These were fed to the rats upto the dosage of 800 mg kg^{-1} of body weight. The absence of gastrointestinal disorders such as diarrhea indicated that these algae were tolerated by animals even at 800 mg kg^{-1}. Application of these algae into albino rats did not elicit any skin allergy. Weight gains, PER and nitrogen balance studies with *Spirulina fusiformis* supplementation to poor rice diets showed significantly higher values of all parameters over casein supplementation.

Trace metal and micronutrient supplement: Trace metals are essential nutrients required in very small amounts in the daily diet which play key roles in various activities of the cell. One of the important trace metals is selenium involved in immune function, reproduction, cardiovascular disease, cancer, viral infection control and metal toxicity. Another essential trace element is iodine, whose deficiency affects thyroid function, cardiovascular function, IQ and other brain disorders. *Spirulina platensis* has the potential to be used as a matrix for the production of selenium- and iodine-containing compounds (Desai and Sivakami, 2004). The relative proportion (Ca:P) of these micronutrients is compatible with the preservation of bone health since it reduces decalcification risk (Table 2). Moreover, as it was previously stated, the cyanobacteria of interest is an oxalate-free plant food, thus as with iron it provides calcium with high availability, thus it improves its absorption (Craig and Mangels, 2009). It is also a great source of many other micronutrients, such as vitamin B6, vitamin C, vitamin D and vitamin A. It is also a source of potassium, chromium, copper, magnesium, phosphorus, manganese, selenium and zinc.

Table 2: Minerals composition of *Spirulina* microalgae (Otles and Pire, 2001)

SN	Minerals	mg $100g^{-}$
1	Zinc	3
2	Sodium	900
3	Potassium	1400
4	Phosphorous	800
5	Manganese	5
6	Magnesium	400
7	Iron	100
8	Copper	1.2
9	Calcium	700

Protein source of malnutrition

Spirulina has no cellulose in its cell walls, being composed of soft mucopolysaccharides. This makes it easily digested and assimilated. It is 85 to 95% digestible making it important for people suffering from intestinal malabsorption, typically, older people. They find *spirulina's* protein easy to digest (Branger et al., 2003). *Spirulina* is effective for victims of malnutrition diseases like kwashiorkor, where the ability of intestinal absorption has been damaged. For malnourished children, it is more effective than milk powders because milk's lactic acid can be difficult to absorb Kelly et al. (2011). *Spirulina* is 65% protein which is extremely high for a plant, and one of the reasons it is becoming more popular (Table 3). It also contains sources of all 8 essential amino acids. It's an easy, healthy way to boost your protein intake and a great source of protein for vegetarians. Furthermore, this protein is highly absorbable (Sharoba, 2014).

Table-3: Protein composition of *Spirulina* microalgae (Colla et al., 2007)

SN	Amino acids	g/100g
1	Leucine	4.94
2	Valine	3.51
3	Isoleucine	3.20
4	Tryptophan	0.93
5	Methionine	1.15
6	Phenylalaline	2.78
7	Threonine	2.97
8	Lysine	3.02

Biological activities

Antiviral activity

There is a strong therapeutic research involving *spirulina*, its strong antibacterial and antimycotic substance which is isolated from *S. platensis*. These results demonstrate excellent in vitro inhibition of HIV-1 virus both in human T-cell lines and in human monocytes. The therapeutic index of the extract is reported to be over 100 and concentrations as low as 5-10 μg/ml evidently reduce the production of virus in any system and hence proves to be highly antiviral in activity. Ingestion of *spirulina* contributes to the functional preservation of the intestinal epithelium which acts as a first line of mucosal barrier against infections (Sheahan et al., 2003). Inhibition of humoral immune response, cell mediated immune response (delayed type hypersensitivity) and TNF-alpha was noticed in a dose-dependent manner in mice. (Shklar et al.,1988). Spirulina has also been found to protect against hay fever (Simpore et al., 2005).

Cures eye disease

Spirulina is rich in beta-carotene that can overcome eye problems caused by vitamin A deficiency. It provides the daily dietary requirement of beta-carotene which can help prevent blindness and eye diseases (Seshadri, 1993). The protein and B-vitamin complex makes a major nutritional improvement in an infant's diet. It is the only food source other than breast milk containing substantial amounts of essential fatty acid, essential amino acids and GLA that helps to regulate the entire hormone system (Ramesh et al., 2013).

Anti-Inflammatory effects

Recent research reveals that free bilirubin functions physiologically as a potent inhibitor of NADPH oxidase activity. The chromophore phycocyanobilin (PCB), found in blue-green algae and cyanobacteria such as *Spirulina*, also has been found to be a potent inhibitor of this enzyme complex, likely because in mammalian cells it is rapidly reduced to phycocyanorubin, a close homolog of bilirubin (Helliwell, 2011). Orally administered *Spirulina* or phycocyanin (the *Spirulina* holoprotein that contains PCB) can exert a wide range of anti-inflammatory effects. Until PCB enriched *Spirulina* extracts or synthetically produced PCB are commercially available, the most feasible and least expensive.

Radiation protective effects

Radiation protection offered by *Spirulina* may be due to the phytopigments (carotenoids, chlorophyll, phycocyanin) and polysaccharides. *Spirulina* can elevate the activity of all the antioxidant related enzymes viz., superoxide dismutase, catalase, glutathione peroxidase and glutathione reductase significantly. The effect may be due to the high phytopigments (carotenoids, chlorophyll, phycocyanin) in *Spirulina* (Hirahashi, 2002). Feeding children subjected to low level of radiation over a long period of time with 5 grams of *Spirulina* a day resulted in the reduction of Cesium-137 in urine by 50%. The c-phycocyanin and polysaccharide extracts of *Spirulina* stimulate recovery of white blood cells and bone marrow cell counts. The anaemic condition induced by irradiation was also reduced (Karkos et al., 2008).

Role of *Spirulina* in immunity

Spirulina helps in building immunity and improving resistance to viral infections, enhance components of the mucosal and systemic immune system as it activates the cells of innate immune system. *Spirulina* has

also been shown to activate macrophages, T and B cells (Swartz et al., 1987). Sulfolipids derived from *spirulina* have also proved effective against HIV. Extracts from *Spirulina* biomass have also been found active against herpes virus, cytomegalovirus, influenza virus, etc. *Spirulina* extracts have also been shown capable of inhibiting carcinogenesis (Blinkova, 2001). *Spirulina* use leads to higher levels of natural killer cells, interferon gamma and more potent production of interleukins (Hirahashi et al 2002).

Immune system strengthening

Several experiments have shown that *spirulina* has a favorable regulatory effect on the immune system (Borchers, 2007). It stimulates the activation of macrophages, as well as the activity of T cells and NK cells. This process induces the release of interferon-gamma (IFN-γ), which can eventually lead to virus inactivation. These actions are thought to be mediated by polysaccharides (Borchers 2009).

Antioxidant and nutrient profile

Spirulina contains phenolic acids, tocopherols and ß carotene which are known to exhibit antioxidant properties. Miranda et al., (1998) evaluated the antioxidant capacity of a *Spirulina* extract indicated that *Spirulina* provides some antioxidant. Accumulative data from those studies concluded that *Spirulina* ingestion significantly relieved or totally prevented the oxidative stress or inflammation and their associated pathological damages induced by insulting compounds (Deng and Chow, 2010). Although those studies were not directly investigating *Spirulina's* effects on cardiovascular conditions, the findings clearly demonstrated the antioxidant and anti-inflammatory activities of *Spirulina*. 180% more calcium than whole milk, 670% more protein than tofu, 3100% more beta carotene than carrots, 5100% more iron than spinach, more antioxidant and anti-inflammatory activity in 3 g of *Spirulina* than in five servings of fruits and vegetables (Moorhead et al 2005). *Spirulina* is also very high in calcium, meaning it is excellent for growing children to help their bones and teeth develop. It contains over 26 times the calcium in milk.

***Spirulina* in anaemia**

Spirulina possibly enhances red cell production and function. Over a 12-week study period, there was a steady increase in average values of mean corpuscular haemoglobin with *spirulina* intake. Older women benefitted more rapidly from *Spirulina* supplements (Mohan et al., 2014). Levels of anaemia also decreased in children when their diet was supplemented with *Spirulina* (Branger et al., 2003). *Spirulina* is a fantastic source of iron, meaning it is excellent for women during pregnancy.

Anticancer effects

Many researchers find phycocyanin to be a truly potent antioxidant, anti-inflammatory and anti-cancer properties pigment found only in *Spirulina* and other species of blue green algae. (Subbashini et al., 2004). It has been argued that the combined antioxidant and immune modulation characteristics of *Spirulina* may have a possible mechanism of tumor destruction and hence play a role in cancer prevention. The effects of *Spirulina* on oral carcinogenesis, in particular leukoplakia showed tumor regression after topical application or enteral intake of *Spirulina* extract (Shaklee and Schwartz 1988). Jalaja et al., 2011 studied the effect of *Spirulina* on chemoprevention of cancer in tobacco chewers in Kerala, India. It was found supplementation with *Spirulina* at 1 g/day for 1 year resulted in complete regression. If *Spirulina* proves to have such effect, it can easily be incorporated in the daily diet as a therapeutic agent (Baley, 2002).

Commercial production and importance of microalgae in diet

In most developed countries, people consume high caloric food items due to the modern life style, which leads to health problems, such as obesity, heart diseases and diabetics. A balanced nutritional diet is needed for health and should contain vitamins, minerals, PUFAs, etc. Microalgae are considered as a remarkable but poorly explored natural source for a healthy diet. Several species of microalgae are identified as rich in carbohydrates, proteins, lipids and nutritionally valuable components. Becker (2004) reported that microalgae are an abundant source of vitamins and minerals, such as vitamins A, B1, B2, C and E; nicotinate; biotin; folic acid; pantothenic acid; niacin; iodine; potassium; iron; magnesium and calcium. Interestingly, the lack of polysaccharides in the cell wall of cyanobacteria makes their biomass a more easily digestible material and therefore more acceptable for human consumption (Richmond and Preiss, 1980). The Chinese first used microalgae (*Nostoc* sp.) (over 2000 years ago) as a food and later, the commercial forms of microalgae (*Chlorella* sp. and *Spirulina* sp.) were consumed as healthy foods in

Japan, Taiwan and Mexico (Tamiya, 1957; Durand-Chastel, 1980; Soong, 1980). Currently, most of the commercialized products of microalgae are available in markets as a health food, in the forms of tablets, capsules and liquids (Pulz and Gross, 2004) and their products are mixed with pastes, snacks, candy, gums, noodles, wine, beverages and breakfast cereals (Yamaguchi, 1997; Lee, 1997; Liang et al., 2004). *Aphanizomenon flos-aquae*, *Chlorella* sp., *Dunaliella salina* (*D. salina*), *Dunaliella tertiolecta* (*D. tertiolecta*) and *Spirulina platensis* (*S. platensis*) are some of the microalgae species widely used as a human food source because they are rich in protein content and have high nutritive value (Soletto et al., 2005; Rangel Yagui et al., 2004). Among these microalgae, *Spirulina* (*Arthrospira*) and *Chlorella* are currently dominating the microalgal market. *Spirulina* (*Arthrospira*) has a high protein content and excellent nutrient value (Spolaore et al., 2006) and has gained worldwide popularity as a food supplement (Colla et al., 2007). It has an amino acid content of 62% and it is a rich natural source of vitamins A, B1, B2, B12, as well as phytopigments, including carotenoids and xanthophyll (Richmond, 1988). In addition, it has a considerable amount of essential fatty acids and linolenic acid, which cannot be synthesized by humans (Becker, 1994); thus, more attention has been given to the cultivation of *Spirulina*. The world production of *Spirulina* for human consumption exceeds 1000 metric tons, annually (Ciferri and Tiboni, 1985). The world's largest *Spirulina* producer is Hainan Simai Enterprising, which is located in China and has an annual algal powder production of 200 tons. There are 20 countries in the world that produce *Spirulina*-based products, such as tablets and powder. Among these countries, the USA ranks first in *Spirulina*-based products, primarily in the form of pills and spray-dried powder, followed by China, Israel, Japan, Mexico, Taiwan and Thailand (Spolaore et al., 2006). From microalgae, a wide variety of nutraceuticals are available and marketed for sale. For example, the Myanmar-*Spirulina*-factory (Yangon, Myanmar) produces tablets, chips, pasta and liquid extracts. Similarly, Cyanotech (Hawaii, USA) produces pure powder under the name ''*Spirulina pacifica*". Phycocyanin, extracted from *Spirulina* and commercially known as 'lina blue', is used as a blue colorant for food and cosmetics. *Spirulina* also acts as a functional food, feeding beneficial intestinal flora, including *Lactobacillus* and *Bifidus* (Ciferri, 1983). In the future, these health foods are expected to be a stable market, including products such as *Spirulina* liquid CO_2-extracted antioxidant capsules (Belay et al. 1993). One gram of *Spirulina* contains one-half of the adult daily requirements of Vitamin A. According to human studies, total serum cholesterol was lowered by consuming *Spirulina* (Gonzalez de Rivera et al., 1993). The purified PUFAs from *Spirulina* are added into infant milk formulas in European countries for health promoting purposes.

CONCLUSION

Spirulina shows potent immune stimulating effects, shows antiviral activity against a variety of harmful viruses. It shows promise as a cancer preventative agent and in the treatment of tumors. *Spirulina* shows far ranging cardiovascular benefits including improvement of blood lipid profiles, prevention of atherosclerosis and control of hypertension. In addition to high levels of provitamin A, dried micro algae can provide various other nutrients including proteins, minerals, vitamins and antioxidants. World production of consumable algae and algae products to be used as dietary supplements, food additives, functional foods and medicines has reached thousands of tons per year after research which has proved its importance.

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Pineapple as potential crop resource: Perspective and value addition

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ABSTRACT

Pineapples are among the major fruits abundantly available in Northeast Hill region of India particularly in Manipur where it is available for almost 8 months a year . The two most-sought-after varieties of Pineapple, viz. Kew (big size) and Queen (small size) are produced on commercial scale. The Queen variety available here is most suitable for preparation of juice, pulp and squash. The 8-month availability of pineapples in Manipur makes the State highly advantageous for processing and export. The specialty of Manipur is the Kew variety available here known for its distinctive taste and flavour. This Kew variety is most suited for making canned fruit products thus Canned Pineapples could form a part of the major exports in processed fruit industry from the Northeast Hill region. Processing of pineapple can invite the entrepreneurs of this region as good source of income generation. Entrepreneurs occupy a central position in a market economy activating and stimulating all economic activities. serving as the spark plug in the economy's engine. Different value added pineapple products like juice, squash, jelly, jam, osmo-dehydrated rings, bar having great market potential can attract such marketing personnel for their good business initiation.

Keywords: Pineapple, Juice,, Jam, osmo-dehydrated rings, value-addition

Introduction

The pineapple (*Ananas comosus*) is the most important horticultural produce of the family *Bromeliaceae* positioning as the important fruits in the world. The plant develops to a cone-shaped juicy and fleshy fruit with crown at the top (Morton, 1987; Tran, 2006) growing up to a height of 75-150 cm. The major pineapple growing countries in the world are Brazil, Thailand, Philippines, Costa Rica, China and India. The total area under pineapple cultivation in the world is 909.84 thousand ha with production around 19412.91 thousand tons. Apart from the use as fleshy raw fruit, these are also used as processed food items like juices, concentrates, and jams. After orange and apple juices, the pineapple juice occupies the third most preferred position worldwide (Cabrera *et al*., 2000). Pineapple (*Ananas comosus*) is one of the commercially important fruit crops of India. It is abundantly grown in almost entire North East region, West Bengal, Kerala, Karnataka, Bihar, Goa and Maharashtra states. There is sizeable increase in acreage and production of pineapple in India. In acreage, there is an increase from 87 thousand ha in 2006-07 to 89 thousand ha in 2010-11.Similarly the production has increased from 1,362.00 thousand tons in 2006-07 to 1,415.00 thousand tons in 2010-11. Maximum area under pineapple cultivation is in Assam (14 thousand ha), where productivity is of medium scale. Total production is higher in West Bengal, where productivity is also high. North-Eastern India having total geographical area of 2.62 million km^2, comprising total 8 states has a high potential for pineapple cultivation. This region gets immense climatic diversity, fertile and organic soil, sufficient rainfall thereby having scope for cropping variation, About more than 40% of the total pineapple production of the country was made from the NE region. and 90 to 95% of the produce is organic. ‘Giant Kew’ and ‘Queen’.are the common cultivars grown this region. About 95 % of pineapple produce from this region are regarded as organic in nature. NE is considered as the major producer of pineapple in India about more than 40% of national production. The fruits are getting high attraction due to high TSS and have less fiber content thus having much sweeter than other region. Various scientific and academician have much attention towards the exploration of processing and preservation of such an important produce of this region.

Entrepreneurship has been described as the "capacity and willingness to develop, organize and manage a business venture along with any of its risks in order to make a profit. An entrepreneur is an entity which has the ability to find and act upon opportunities to translate inventions or technology into new products: "The entrepreneur is able to recognize the commercial potential of the invention and organize the capital, talent, and other resources that turn an invention into a commercially viable innovation. In this sense, the term "Entrepreneurship" also captures innovative activities on the part of established firms, in addition to similar activities on the part of new businesses. In this context value addition of pineapple through different scientific process technologies is to be emphasized in this region to explore new process industries. Different products like pineapple like juice, squash, jelly, jam, osmo-dehydrated rings, bar can be utilized towards great market values for sustainable utilization of pineapple thereby putting importance to income generation and entrepreneurship development.

PINEAPPLES - NATURE'S HEALING FRUIT

Pineapples are nutritionally packed members of the bromeliad family. This delightful tropical fruit is high in the enzyme Bremeliaceae and the antioxidant vitamin C, both of which plays a major role in the body's healing process. Bremeliaceae is a natural anti-inflammatory that has many health benefits and encourages healing. Pineapple fruit is very low in Saturated Fat, Cholesterol and Sodium. It is a good source of Dietary Fiber. Some important health benefits of pineapple are:

- Pineapples are packed full of vitamin C and fiber – important for the immune and digestive systems.
- Pineapples have anti-inflammatory effects which are good for those long hard days and those heroic sporting injuries.
- They contain the enzyme Bromelain which is thought to aid digestion.
- It is regulates the gland and found to be helpful in cases of goiter (enlargement of the thyroid gland).

Importance of Value Addition

Value addition from commodities point of view, is processing raw product into a branded product that consumers are willing to pay for than the raw product. For example, a food processor could purchase fruit from farmers and process into fruit juice. This juice has added value and becomes a branded good. Basically unprocessed raw products such as, meat carcasses, and fresh pineapple fruit and cotton bolls have no value added.

The importance of value addition in pineapple processing

- It allows firms to prolong the shelf life of products.
- It emphasizes strength of brand that promote commodity.

- ❖ Helps firms increase their unit selling price (USP) for extra income
- ❖ Product achieves higher competitiveness in the market.
- ❖ It allows products to become more convenient for handling by the producer and the consumers
- ❖ It allows product to become more attractive to the consumer (buyer)
- ❖ It allows producers to make product to the specification and taste of the consumer.

PROCESSING OPPORTUNITIES FOR PINEAPPLE

Pineapples must be harvested at the right stage of maturity (60-80% yellow colouration) early in the morning to ensure quality fruits in terms of firmness, flavour and taste. Pineapple can be processed to:

A. PINEAPPLE GINGER RTS

B. PINEAPPLE CINNAMON RTS

C. PINEAPPLE CARDAMOM RTS

D. OSMO-DEHYDRATE PINEAPPLE RING

E. OSMO-DEHYDRATE PINEAPPLE TIT-BIT

F. PINEAPPLE JAM

G. PINEAPPLE JAM with GINGER FLAVOUR

H. PINEAPPLE BAR

A. PINEAPPLE GINGER RTS

Procedure

1. Raw fresh pineapple
2. Peeling and cutting
3. Extraction the juice & filter the juice by the mosline cloth
4. Get the 500ml real pineapple juice
5. Add 5-10gm crushed ginger with 1.7 lit distilled water & boil for 2 min at 85ºC and filter
6. Add 325gm sugar and 3 gm citric acid with filtered water and cool it
7. Then add 500ml read pineapple juice and mix well (final brix 16º)
8. Then put in sterilized glass bottle and capping well
9. Then autoclave the bottle at boiling water for 20 min or in autoclave machine.
10. Cool it at room temperature

Yield
1 kg of pineapples yield around 300-350 ml of juice

Shelf-life
4 months at ambient conditions

B. PINEAPPLE CINNAMON RTS
Procedure
1. Raw fresh pineapple
2. Peeling and cutting
3. Extraction the juice & filter the juice by the mosline cloth
4. Get the 500ml real pineapple juice
5. Add 5-10gm crushed cinnamon with 1.7 lit distilled water & boil for 2 min at 85ºC and filter it
6. Add 325gm sugar and 3 gm citric acid with filtered water and cool it
7. Then add 500ml read pineapple juice and mix well (final brix 16º)
8. The put in sterilized glass bottle and capping well
9. Autoclave the bottle at boiling water for 20 min or in autoclave machine.
10. Finally cool it at room temperature

Yield
1 kg of pineapples yield around 300-350 ml of juice

Shelf-life
4 months at ambient conditions

C. PINEAPPLE CARDAMOM RTS
Procedure
1. Raw fresh pineapple
2. Peeling and cutting
3. Extraction the juice & filter the juice by the mosline cloth
4. Get the 500ml real pineapple juice
5. Add 2-3 pieces crushed cardamom with 1.7 lit distilled water & boil for 2 min at 85ºC and filter it
6. Add 325gm sugar and 3 gm citric acid with filtered water and cool it
7. Then add 500ml read pineapple juice and mix well (final brix 16º)
8. The put in sterilized glass bottle and capping well
9. The autoclave the bottle at boiling water for 20 min or in autoclave machine.
10. After that cool it at room temperature

Yield
1 kg of pineapples yield around 300-350 ml of juice

Shelf-life
4 months at ambient conditions

D. OSMO-DEHYDRATE PINEAPPLE RING
Dehydrated pineapples are dried slices of pineapple which can be used as snacks or incorporated in other food preparations. They have a semi-hard and feathery texture and a sweet taste with the characteristic

flavour and colour of the fruit. Dehydration preserves most of its nutritive value compared to other processed products such as jam or candied/crystallised fruit. The sugar content is less than other products.

Principles of dehydration
The basic principle of dehydration is to remove moisture from the fruits by soaking them in a sucrose solution, followed by drying. This process may be used to reduce the number of micro organisms and browning of fruits during drying and storage.

Procedure
1. Raw fresh pineapple
2. Peeling and cutting into ring shape (1-1.5 cm thickness)
3. Make different Brix of sugar solutions (40º, 50º, 60º) and add 0.3-0.5 gm KMS on each
4. Then put the pineapple rings on this sugar solution for overnight (all rings should be dissolved on the solution)
5. After that remove the rings and stay for drain the solution
6. Then put on tray for drying (55º C for one day on tray dryer)
7. Check out the ring for proper drying (it should be turn another side)
8. After drying it should be properly packed.
9. Storage at room temperature

Process control
Fruits which are at ¾ colour break should be selected.

Slices should have uniform thickness; otherwise the fruit slices will not dry uniformly to the same final moisture content. Temperature and duration of drying are important parameters to be monitored to obtain a stable and properly dried fruit.

Packaging and storage
Fruits can be vacuum packed (55% vacuum level) and stored in a dry place, away from direct light.

Yield
1 kg of fresh pineapples yield about 100 g of dehydrated slices.

Shelf life
6 months in 55% vacuum bags and aluminum packs.

E. OSMO-DEHYDRATE PINEAPPLE TIT-BIT
Procedure
1. Raw fresh pineapple
2. Peeling and cutting into cube or try-ange shape (1.5 -2 cm thickness)
3. Make deferent Brix of sugar solution (40º, 50º, 60º) and add 0.3-0.5 gm KMS on each

4. Then put the pineapple ring on this sugar solution for overnight (all rings should be dissolved on the solution)
5. After that remove the ring and stay for drain the solution
6. Then put on tray for drying (55º C for one day on tray dryer)
7. Check out the ring for proper drying (it should be turn another side)
8. After drying we can use icing sugar on surface area for removing the stickiness and then it should be properly packed.
9. Storage at proper packing and temperature.

Yield
1 kg of fresh pineapples yield about 100-150 g of dehydrated tit-bits.

Shelf life
6 months in 55% vacuum bags and aluminum packs.

F. PINEAPPLE JAM
Jams are fruit based products preserved mainly with a high concentration of sugar and acid. To ensure a safe product which can be stored at room temperature for more than a year, it must contain at least 65% sugar. Jam is used as spread and are also used in the baking and confectionary industry.

Pineapple is naturally high in acidity but poor in pectin and as such commercially available pectin must be added to ensure setting of jams.

Procedure
1. Raw pineapple
2. Peeling and cutting and extraction the juice and pulp
3. Take 300 gm of pineapple pulp and mixed with 600gm of sugar and 8-10 gm pectin (pectin should be mixed with sugar previously) and add 3-5 gm citric acid.
4. Heat slowly and mix continuously and check the brix untill reach up to 65°-68° and add 0.3-0.5 gm KMS
5. Stop heating and stay for worm condition and put in to the sterilized container
6. Close the container properly and autoclave at 30min at boiling water.
7. Storage at refrigerated condition.

Yield
1 kg of fruits yield around 550 g jam

Shelf-life
10-12 months

G. PINEAPPLE JAM with GINGER FLAVOUR

Procedure

1. Raw pineapple
2. Peeling and cutting and extraction the juice and pulp
3. Take 300 gm of pineapple pulp and mixed with 600gm of sugar and 8-10 gm pectin (pectin should be mixed with sugar previously) and add 3-5 gm citric acid.
4. Make 10gm of ginger finely crushed and add with pulp
5. Heat slowly and mix continuously and check the brix untill reach up to 65°-68° and add 0.3-0.5 gm KMS
6. Stop heating and stay for worm condition and put in to the sterilized container
7. Close the container properly and autoclave at 30min at boiling water.
8. Storage at refrigerated condition.

Yield

1 kg of fruits yield around 550 g jam with ginger flavour

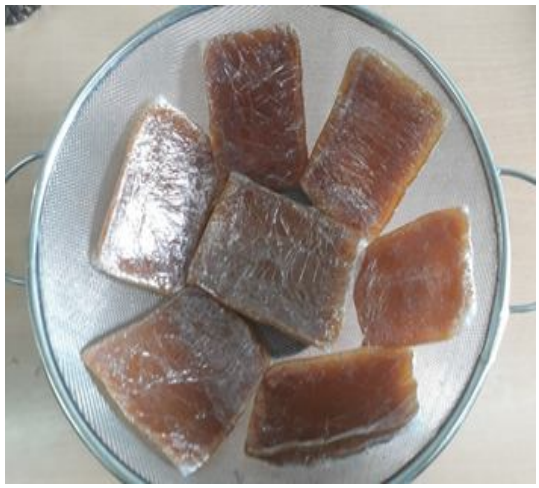
Shelf-life

10-12 months

H. PINEAPPLE BAR

Pineapple fruit bar is made from pineapple pulp, sugar and pectin. It has a sweet taste and a characteristic flavour. Fruit paste is more nutritious than common sweets made solely from sugar and artificial flavours.

1. Raw pineapple
2. Peeling and cutting and extraction the juice and pulp
3. 500gm pulp and 150 gm sugar mix properly
4. Slow heat for 10- 15 min with mixing sugar and mixed continuously until Brix will be 18°
5. Keep it worm or room heat condition and add 0.3 gm KMS
6. Speared on a aluminum foil properly and put into dryer
7. at 55°C for 2 days
8. Final brix 70°- 75° after dying
9. After that cutting into proper shape & size and packing with shrink packing
10. Keep it in the cool condition.



Yield
1kg of pineapple yield around 350g of pineapple paste

Shelf-life
10 -12 months

Conclusion
The food processing industry in this region is mainly unorganized and works on a smaller scale. The huge presence of the unorganized sector can be attributed to the fact that most of the units in the unorganized sector are less capital intensive, and the easy availability of raw materials makes it more attractive to the small entrepreneurs for seasonal processing. The high presence of own account enterprises does serve to mitigate unemployment to a large extent, but this sector is also responsible for the low level of income generation, and low level of productivity in the region. Most of the pineapples are sold as fresh and less than two percent of the total production is being processed for other value-added products like jam, jelly, squash, sauce, pickle, dehydrated product and wine. There are about 53 cold storage facilities and 112 processing units spread throughout the region. However, all the units suffer heavy losses every year mainly due to the absence of sales promotion, sales organized at individual level, inefficient production process, poor management system, high overhead cost, non-utilization of by-products. The main reason for this losses maybe lack of skilled manpower, Inefficient utilization of processing unit as most of the units are non-functional throughout the year, absence of adequate transportation facilities, inadequate number of cold storage and processing facilities, poor quality standards of produce and its control methods, lack of multi fruits or vegetable processing unit, inadequately linkages between R&D labs and industry and high cost of packaging material and poor marketing infrastructure. Hence, this scientifically processed pineapple products will look forward to demonstrate the efficient handling and scientific preservation suitable for the region therby leading to best source for income generation and entrepreneurship development.

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Value addition of Wild edible plants (WEPs) for Sustainable Utilisation and socioeconomic development of rural areas of Manipur, India

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Abstract

In spite of being a small state of India, Manipur has huge biodiversity that provide seasonal wild edible plants(WEPs) to the ethnic people. Gradual increase in population and decreasing forest areas exert huge anthropogenic pressure to these WEPs. Sustainable utilisations and conservation are the need of the hour. Domestication and processing of the WEPs were purposed for sustainability but domestication being a complex process, the value addition of WEPs has been considered more viable in the rural setup of Manipur. Traditionally, WEPs were processed by sun drying, fermentation, boiling and cooking for longer shelf life and unique flavour. Making Jam, jelly, RTS etc ensure not only the longer shelf life but also it made sure that these WEPs were also available in off seasons in process form. The value addition and proper packaging will undeniably enhance the sustainability in terms of economic viability, the role of Government and other organised sector need to put exemplary efforts to uplift the socio economic condition of the rural masses through production of value added products of WEPs.

Introduction

Manipur is a small state that lies in the slopes of the south flowing Sub-Himalayan ranges in the north east region of India at the latitude 23.83ºN to 25.68ºN and longitude 93.03ºE to 94.78º E. It is surrounded by hill ranges in fold forming a valley in the middle of the state. The two main physical features of Manipur are river valley and the western mountainous region. The climate of Manipur can be broadly divided into Temperate, Sub-tropical and Tropical which are influenced by the topography of the hilly region. The state with its varied climatic zones offers an array of huge biodiversity of flora and fauna including hordes of wild edible vegetables, fruits and medicinal plants all of which comes under the Wild edible plants (WEP) category. FAO defines them as plants that spontaneously in self maintaining populations in natural or semi natural ecosystem and can exist independently of direct human actions (Heywood et al 1999). In simpler terms, WEP are those plant species which are neither domesticated nor cultivated but available from the natural resources and used as food. These WEPs contribute appreciably in the global food stock though its importance remained unpretentious to its more appealing cultivated counterparts (FAO 2004). Studies have amply showed their role in complementing the diet of about 1million people worldwide especially for tribal and rural communities where diet is intricately woven with their culture . The prospective use of WEP in ensuring food security and reducing poverty by increasing local income generation while being a source of nutrition has also been emphasized by many studies (Burlingame 2000, Saha et al 2014, Jana & Chauhan 1998, Samant & Dhar 1997)

Manipur has been the home of more than thirty ethnic groups in historical times, with varying responses to the changing socio-cultural environment, forming a heterogeneous population including both tribal and non tribal population with a total population of 27,21,756 (Census of India, 2011). Besides the cultural differences, there are differences in lifestyle and food habits. From time immemorial the ethnic people of the state have been depending on the naturals forest produces for their livelihood as the forest provides ample opportunities for collecting the produces which are naturally abundant in those vast resources for their livelihood by supplementing their own food intake as well as by selling these produces to the local market. Many of these wild edible plants were sold in market and locally consumed by the ethnic people of the state. All the local market in the state sold these wild edible vegetables or fruits in any given month of the year. With gradual increase of population leading to ever increasing needs, exploitation of natural resources reached an unprecedented level. In today's context with the shortcoming of conventional food basket for the escalating population looming large, the WEPs provide sanguinity to these indigenously consumed foods needs. Hence, thorough study on the type and frequencies of WEP usage as well as their proper conservation strategies are required. In this regard, the present status, problem and prospects of these wild edible plants of Manipur state is discussed in the article.

Current Status, Problem and Prospects of wild Edible plants of Manipur

The highly diverse WEPs have significant role in the food security of the ethnic people of Manipur. Traditionally, the seasonal WEPs provide the major sources of nutrition in the remote villages of the state. Not many studies were carried out on the nutritional quality and food processing prospect of the resources though limited studies revealed high nutritional and nutraceutical properties in many of the common WEPs (Sundriyal et al., 2001, Rajkumari et al., 2013). Particularly, high content of vitamins, minerals and high antioxidant activities were found to be excellent or far ahead of commercial crops (Sharma et al, 2013, Pereira et al., 2011, Guleria et al., 2011). The Region offer a wide range of climatic variation in the state as a result huge biodiversity is bestowed upon it. The indigenous people of the state have long tradition of utilising the forest produces and harbours good knowledge of the value of the flora and fauna. Folk medicine is generally practice in all ethnic communities (Deb et al., 2015) of the state. A rough estimate from all the major markets in four districts of the state indicates yearly consumption of about 90 tonnes of wild fruits and 90-200 tonnes of wild vegetables. Many of these fruits and vegetables are believed to be either health promoting factor or medicinal properties (Rajkumari et al., 2013). More than 250 different species of wild plants are use as food in the state (Devi et al., 2010). Wild edible vegetable and fruits constitutes at least 40% of the marketed vegetables and fruits in any time of the year in local markets.

Due to unprecedented population growth and increasing demand for these resources made human to evade the uncharted territory of wild animals. What once used to be for local or domestic consumption are now

being increasingly used for commercial sale. With improved infrastructure in this remote areas leading to better connectivity and facilities to access main trading centres, the demand has definitely escalated comparable to the demand for conventional vegetables. This one sided high demand with no sustainable source of supply has led to excessive foraging and over harvesting. As there is no definite control in the collection of the plants from the forest, the collector usually harvest almost all available vegetables or fruits while they find these plants in the thick forest without much consideration of the survival of the plants. Time and again branches are chopped off for collecting the fruits, edible roots are uprooted, whole plants are removed entirely etc. The over extraction of wild fruits and selling them in the market made it impossible for many of the higher altitude fruits (Sundriyal et al., 2001) as there is very less seeds left in the natural habitat for its propagation. The collectors treat the forest as an unending resource. Above all many anthropogenic activities such as encroaching of forest land for human dwelling, dams, construction of other developmental activities destruct the natural habitat (FAO 2004).

Although there are clauses in the forest acts for regulating collection of NTFP (Non Timber Forest Products), implementation and timely action has been lacking or the laws are inadequate today given the situations are not the same as when they were enacted. This warrants a new role for the government to not only regulate but involve the welfare of the communities for a sustainable production of the WEP's (KIT 2003). It becomes imperative to make people aware that these resources need to be utilised sustainably. Otherwise, the over-extraction and over-exploitation of these resources would make them vanish from the surface of the earth.

Conservation strategy of WEPs: domestication and value addition through processing

Collecting the WEPs from their natural habitats for sale involves no production cost, less manpower and no investment. However, there will be shortage and even extinction of such plants in future due to rampart and indiscriminate collection of the wild edible plants. Domestication and processing of the WEPs are two conservation strategies for sustainable utilisation of the WEPs.

Domestication of WEP's is one of the obvious and logical solutions for conservation of wild threatened species especially for WEP's sustainability. It is reported that only 0.5% of plant species has been domesticated out of the total known wild species of plants. And the problem of domestication by cultivation was found to depend on the plants and not on the people (Diamond 2000). Successful or suitable agro-techniques (for most WEP) are not understood properly and the plant species, being wild ones, presents its own difficulties for cultivation. (Kala 2006).

In Manipur, some of the constraints of domestication can be attributed to small land holdings. As most WEP's come from the hill areas, cultivation area would possibly be a shortcoming. Though few commercially important WEPs like *Thangjing (Eurayle ferox), Kaokha (Saggitaria sagitifolia), Shougri (Hibiscus canabinus)* have been tried with certain degree of success, in the flat plains, most WEPs are still collected from the wild. Studies in the districts of Manipur (Pfoze et al 2012, Gante et al 2013) showed that the factors determining the selection of plants for domestication over others depend mainly on the commercial value, palatability and shelf life. Shorter duration of availability as well as rarity of the species is observed to increases the commercial value.

Another method of conservation technique is to increase the shelf life of these foods through processing. Almost all wild edible fruits and vegetables have very less shelf life therefore the right price of the produces are not obtained by the collector as they have to sell the goods within limited time. Thus, the collectors which are mostly from lower socio-economic strata would have to harvest more for their subsistence needs. There lies the need for technological intervention. And simplest intervention is the processing of available wild edible plants to appropriate finished products and its value added products to get maximum price out of the produces. This not only improves the socio-economic standard of the villagers but also will help in sustainable extraction of the same. There are a lot of advantages in processing these wild foods. Not only adding value to the products and increasing shelf life, it could overcome the seasonality as well as perishability. This can effectively substitute the imported food products flooding the local markets and expand the market. Food processing enterprises established in rural Manipur have potential to generate off farm employment and decrease dependence on natural resources only. Post harvest processing, handling

and marketing, increases food availability at household and community levels, and thus contributes to food security. Value addition was done to a few wild edible species in the WEP's of Sikkim and cost-benefit analysis has showed that the income from the plants could be increased by at least 3–5 times after due processing (Sundriyal and Sundriyal 2004). This was only for the edible fruit but it still indicates that if processing for other WEP's like leafy vegetable, root or seeds can be standardised, the income generation rate can be much higher.

Simple process like juicing and pasteurisation, drying and pickle making in the villages can be done without much investment for some of the WEPs. The fruits can be made into chutney, pickles, squash, wine, RTS jam, jelly etc. Vegetables may be dried, blanch, store in brine or fermented to enhance its nutritive value and flavour. Such methods will improve its shelf life and fetch much better price in the market. Proper packaging will also not only help in longer shelf life but also will be more presentable though scientific packaging may not be cost effective for individual or marginal farmers/collectors. With gradual development in agriculture and import of vegetable, the dependence on the WEPs have been reduce though demand is higher than before. It is high time to plan and execute specific conservation strategies to preserve this valuable and indispensable food resource of the State. Multiplication, area expansion and domestication of highly potential WEPs should be tried and encouraged.

Processed WEPs in local market

Very few process WEPs were marketed in local market. The most common technique of traditional food processing in Manipur is sun drying and natural fermentation along with some methods of pickling. Notable among processed WEPs are sundried wild mushrooms like *Kangla yen* (*Schizophyllum commune*), *U-yen* (*Lentinula edodes*), *Uchina* (*Auricularia auricularia*). Among the wild fruits *Heimang* (*Rhus* sp.) was widely used to make candy with brown sugar. *Heining* (*Spondias pinnata*) was used in pickling. *Chorphon* (*Eleocarpus floribundus*), *Heikru* (*Emblica officinalis*), *Heitup* (*Micrococus paniculata*) and *Heibung* (*Gracinia penduculata*) were boiled and sundried or made candy or mouth freshener. Starchy WEPs like *Koukha (Sagittaria sagitifolia), Heikak (Trapa bispinous), Lemphu (Nymphaea rubra), Kaothum* (*Cyperus escluntus*) etc were boiled and sold as ready to eat items. Fermented Bamboo shoots *soibum* (mostly *Dendrocalamus hamiltonii*) and *soidon* (*Teinostachyum wightii*), fermented *shougri (Hibiscus cannabinus)* seed known as *Gankhiangkhui* and fermented wild *Brassica* sp. (*Ankam thu*) are major fermented WEPs in the local markets of Manipur. The common fermented foods marketed in Manipur include *Hawaijar, Ngari, Hental, Soibum, Soidon, Zianshang* (Jeyaram et al., 2009). These traditional fermented foods harbours GRAS (generally regarded as safe) microorganisms with potential source of therapeutic enzyme (Singh et al., 2014) and other health benefits.

Some other unique local methods of preservation in the form of chutneys are also reported like *Arisaema leschenaultia* which is first roasted, pounded and soaked in ash water for three nights , then washed off with ash water and preserved. This is then mixed with chillies and salt and consumed as chutney (Gangte et al 2013). There are also many other such unique methods whose mode of preservation has been handed down from generations to generation but unknown to mainstream due to the knowledge being confined in a single community. Studies need to be conducted especially to unearth such information which would otherwise be lost. The following tables list some of the WEP's which are traditionally processed in Manipur.

Heimang heingan

Bori

Laloo

Kaothum

Soidon ***Uyen*** ***Heikak*** ***Gankhiangkhui***

Fig-1: Some process and fortified food items from wild edible plants in Markets of Manipur

Table-1: List of some WEP's which are simply sundried or dried after boiling

Plant scientific name	Family	Parts used
Bambusa nutans Roxb.	Poaceae	Tender shoots
Bambusa arundinaceae (Retz.) Wild.	Poaceae	Tender shoots
Citrus latipes (Swingle). Tanaka.	Rutaceae	Rind of fruit
Colocasia esculenta (Linn.) Schott.	Araceae	Leaves and petioles
Elsholtzia communis	Lamiaceae	Inflorescence
Eurya acuminata DC. Fl. Br.	Theaceae	Leaves
*Hibiscus cannabinus L*inn	Malvaceae	Leaves
Meriandra strobilifera Benth	Lamiaceae	Leaves
Osimum americanum Linn.	Lamiaccae	Leaves
Vangueria spinosa Roxb	Rubiaceae	Fruits
Vigna sinensis Savi ex Hassk.	Papilionaceae	Leaves

Table-2: List of some fermented WEP's of Manipur

Plant Scientific name	Family	Parts used
Anisomeles indica Linn	*Lamiaceae*	Seeds
Bambusa nutans Roxb.	*Poaceae*	Tender shoots
Bambusa arundinaceae (Retz.) Wild.	*Poaceae*	Tender shoots
Dendrocalamus hamiltoniii	*Bambusaceae*	Tender shoots
Teinostachyum wightii	*Bambusaceae*	Tender shoots
Hibiscus sabdarifa	*Malvaceae*	Fruits & seeds
Brassica sp	*Brassicaceae*	Leaves

Table-3: List of some wild edible fruits traditionally preserved as candy, jam, chutney or pickles

Scientific name	Family	Local name
Rhus hookerii Sahni & Bahadur.	Anacardiaceae	*Heimang*
Garcinia pedunculata Roxb.	Clusiaceae	*Heibum*
Prunus armeniaca L.	Rosaceae	*Malhei*
Ficus cunia, Buch. Ham. Ex. Roxb.	Moraceae	*Heirit*
Elaeagnus umbellate Thunb.	Myrtaceae	*Heiyai*
Morus nigra Linn.	Moraceae	*Heijampet*
Syzgium cumini (L.) Skeels	Myrtaceae	*Jam*
Microcos paniculata Linn.	Tiliaceae	*Heitup*
Ficus glomerata Roxb.	Moraceae	*Heibung*
Elaeocarpus floribundus	Elaeocarpaceae	*Chorphon*
Ziziphus mauritiana Lam.	Rhamnaceae	*Boroi*

Phyllanthus emblica (L.)	Euphorbiaceae	*Heikru*
Averrhoa carambola Linn.	Averrhoaceae	*Heinajom*
Prunus crasoides D. Don.	Rosaceae	*Chumbrei*
Spondias mangifera Willd.	Anacardiaceae	*Heining*
Horsfieldia amygdalnia (Wall) Warb.	Myristicaceae	*Nonganghei*
Dillenia indica Linn	Dilleniaceae	*Heigree*
Baccaurea sapida (Roxb) Muell.	Euphorbiaceae	*Motok- hei*
Aphanomixis polystachya (Wall) Parker.	Meliaceae	*Heirangkhoi*
Artocarpus chaplasa Roxb.	Moraceae	*Heirikothong*
Flacourtia jangomas (Lour.) Raeusch.	Flacourtiaceae	*Heitroi*

Constraints in conservation of WEPs in Manipur

It is well accepted now that WEP can provide local income and nutrition where they are consumed and this has also been supported by many studies as discussed above. Here, the sustainable consumption of WEP needs to start with cultivation. But especially in Manipur, two major constraints are faced here. The first constraints is at the grass root level with the indigenous people of rural and tribal areas from where almost all WEP are sourced. Studies have stated the lack of information of the dwindling population of plant species due to overexploitation especially to the uneducated local which has reduced the regeneration capacity of most WEP's (Salam et al 2012). These people are also not eager to start cultivation as it involves land, some monetary as well as physical inputs. To them, it is easier to collect than to cultivate. These mindsets have hindered the efforts of introducing sustainable usage of WEP's.

The second constraint faced is in the infrastructure capacity of Manipur as processing industry here is in nascent stage. In general there is no adequate technology for processing and hence inconsistent quality and quantity of the limited produces affect marketability. There are issues in safety and quality as well as storage and packaging. To top this there is always uncertainty of market of the processed WEPs. The role of the government to support R&D and infrastructure may be emphasized here as vital to any setup of an industry, however small it may be, as it requires investment. Reviewing the standards of the produce and establishing certain norms for such products will do a world of good in such endeavours.

Conclusion

The future of sustainable usage of WEP's with value addition is inextricably woven with the ability and effort of conservation strategies that needs to be performed at all levels, from the locals to the government. Cultivation method should be standardised for selected high commercial value species so that the income generation provide momentum for continued cultivation. Value addition techniques requiring low inputs and low technology needs to be promoted with incentives for standard quality and production as food processing and packaging of this wild edible plants with appropriate low cost technologies along with sustainable management could evolved as a viable small scale industry in the state. This will not only ensured better nutritional security but also provides job opportunities to many unemployment and underemployed youth of the state. R&D in the field should be encouraged to upgrade and verify the existing traditional knowledge associated with WEPs. Despite the inherent lack of infrastructure associated with remote hill areas of the country, the WEPs of the state could be converted to a sustainable tool for socio-economic development of the backward areas.

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Traditional Fermented Foods of ethnic people of Manipur, India

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Abstract

Manipur one of the north eastern state of India, is characterized by its rich culture, indigenous food habits and diverse population of over thirty different tribal and non-tribal ethnic communities. Different ethnic communities have their unique food habits. Rice is their staple food and is consumed with fresh or boiled vegetables with a variety of fermented foods. Indigenous fermented foods form an inherent traditional diet system and the cultural integrity of all ethnic communities. These fermented products are unique in terms of the substrate used, the traditional indigenous knowledge of preparation and the culinary practices. The ethnic fermented foods of Manipur are fermented bamboo shoots (soidon, , soibum, thunkheng, thunbin, thunkhengkang, thunbinkang), fermented fish (ngari, hentak, ithiitongba), fermented soybean (hawaijar, khuichang, bethu, bekanthu, theisui), fermented mustard leaves (inziangsang, ziangdui, ziangsang, ankamthu, ganang tamdui), fermented sesame seed (shithu), fermented mustard/rapeseed seed (hangammaru hawaichar), fermented roselle seed (gankhiangkhui), fermented milk (sangom aphamba) and fermented meat (sathu, saayung, guaighi kang, sahro, bongkarot, aithu). This chapter discusses and focuses on the traditional indigenous knowledge, socio-economic and ethical values, microbiology, and health and nutritional benefits of different traditional ethnic fermented foods of Manipur.

Introduction

Fermentation is the simplest and effective method of food processing due to its technical and economic advantages. It increases the shelf life and sensory quality, improves food digestibility, promotes removal of anti-nutritional factors and detoxification of toxic components. The traditional fermented foods are associated with unique functional microorganisms that enhance its nutritional, health promoting and therapeutic values (Nirmala et al., 2014, Rai and Jeyaram, 2015). This has served as a sustainable method of health food production, which has become an intricate part of the world's diet. It is the diversity of the ethnicity, the preparation process, the fermenting raw materials, the organoleptic properties, and the nutritional value and health benefits that make traditional fermented foods a popular choice among the global foods.

Manipur, a culturally rich state in the North East India, has been the home of more than thirty ethnic groups, with varying responses to the changing socio-cultural environment, forming a heterogeneous population including both tribal and non-tribal communities. The indigenous fermented foods and beverages occupy an indispensable position in the traditional diet system and the cultural integrity of all the ethnic communities, and contribute to a large proportion of the daily food consumption in the state (Jeyaram et al., 2009).

The traditional knowledge behind the production of ethnic fermented foods and beverages and their socio-ethical and therapeutic values have existed among the different communities since time immemorial. However, production and consumption of such ethnic foods have always been localized within the ethnic communities. Adaption to new food habits and lifestyle changes accompanied by decrease intake of traditional foods has resulted in an increased incidence of life-style associated diseases (Devi and Kumar et al., 2012). With the increased realization of the nutritional and health benefits of indigenous fermented food products, their production and commercialization with improve quality, safety, and bioactive ingredients have become increasingly important. The documentation of traditional indigenous knowledge of ethnic fermented foods, their socio-economic importance, ethical values, microbiology and health, and nutritional benefits is, therefore, imperative, which will form the basis for future development of fermented functional foods. This chapter will discuss the history and culture of different ethnic groups of Manipur with a focus on the above aspects of different traditional ethnic fermented foods and beverages of the state.

Food habit of Manipur

Cuisines of Manipur are very seasonal, prepared from the seasonal vegetables, are simple, tasty, organic and healthy. Thus, every season has their own specialty cuisines. Various aromatic vegetables and herbs such as tree bean (*yongchaak*), ginger lily (*loklei*), culantro (*awaphadigom*), haory basil (*mayangton*), fishwort (*tuningkhok*), *Allium odorosum* (*maroinakuppi),Allium hookeri* (*maroinapakpi*) and many others, which are not cultivated in mainland India are widely used in Manipur. The principal food items of *Meiteis* are fish, meat and vegetable, while other communities consume a variety of meats. Most of the *Meitei* diet are influenced by many other cultures due to various social political reason. Sanskritization is the most important factor that influence diet. After accepting Hinduism, vegetarian emerge in our society. Usually Manipuri eat two meals a day and take rice as the main course with vegetables, either fresh or boiled, and fermented items. In Manipur, breakfast is very light. *Chabon* (boiled rice water) was taken earlier but now it is replaced with milk, tea, coffee and snacks. Meitei people is fond of *eromba*, an ethnic delicacy prepared by smashing boiled potato and vegetables such as brinjal, tomato, banana pseudostem, banana flower, tree bean, corm of *Colocasia esculenta*, seeds of *Euryale ferox*, etc. and mixing with abundant amount of chilies and steamed or roasted ethnic fermented fish. Apart from vegetables, fermented bamboo shoots are also used to prepare delicious *eromba*. *Morokmetpa* is a spicy chutney prepared with a good amount of chilies and fermented fish and garnished with fresh aromatic herbs. *Singju* is a finely shredded fresh vegetables salad mixed with chili, fermented fish and crushed powder of roasted pea or roasted gram powder. *Kangshu* is a dry preparation of finely shredded boiled wild vegetables or herbs (such as *Centella asiatica*, bamboo shoots, etc.) mixed with fermented fish and chili. All the ethnic groups prefer wild plants as foods to cultivated plants. From time immemorial the ethnic people of the hill areas have been depending on the naturals forest produces for their livelihood as the forest provides naturally abundant resources for collecting the produces and supplement their own food intake as well as earn income by selling these

produces to the local market. The traditional food habits of different ethnic groups are simple and exhibit cultural ethnicity. The peculiarity of the ethnic foods is the dominant preparation and consumption of many traditional fermented items. Different ethnic communities have their own traditional fermented food and beverages that are unique in the traditional knowledge of preparation and the organoleptic characteristics of the fermented products (Devi and Kumar et al., 2012). Traditional foods of Manipur including the fermented foods and beverages have certain nutritional values and curative properties for diseases and disorders.

Types of ethnic traditional fermented foods of Manipur

The ethnic communities of Manipur prepare and consume various kinds of fermented foods and beverages as an indispensable part of their traditional diet. Some are acidic while some are alkaline; some are long period fermented food and some are short time fermented food. It is difficult to classify the traditional fermented foods but based on the raw material used and nature of the fermented product, the ethnic fermented foods can be broadly classified as (i) fermented vegetables (fermented bamboo shoots and fermented mustard leaves), (ii) fermented legume and pulses (fermented soybean, fermented roselle seeds, fermented sesame seeds, fermented mustard/rapeseed seeds), (iii) fermented fish, (iv) fermented meat (fermented pork fat, fermented pork meat, fermented, etc.), (v) fermented milk.

Ethnic fermented vegetables

Ethnic fermented bamboo shoots

Bamboo shoots are the young, tender saplings of bamboo plant, the giant grass belonging to subfamily Bambusoideae of family Poaceae. Globally, over 1250 species of bamboo across 75 genera are reported, to which India contributes 136 species belonging to 23 genera (Forest Survey of India 2017, Singh et al., 2003). They have great potential as food bioresources; however, less than 100 species are commonly known to utilize for consumption of bamboo shoots as food (Chongtham et al., 2011; Waikhom et al., 2013; Nongdam and Tikendra, 2014). North East region of India is considered as a treasure house of edible bamboo shoots as it harbors more than 50% of India's bamboo species (Forest Survey of India 2017, Nongdam and Tikendra, 2014). Manipur has a vast bamboo resource with 54 species under 11 genera that provides almost unimaginable amount of edible bamboo shoots. (Bhatt et al. (2004) reported that 5078 tonnes of fresh bamboo shoots were harvested annually for consumption in the region, with Manipur taking the highest consumption of 2188 tonnes per year. The edible bamboo shoots in Manipur are from *Bambusa balcooa* (locally called *leewa*/*ching saneibi*), *B. cacharensis* (*moriang wa*), *B. kingiana* (*watangkhoi*), *B. manipureana* (*uma*), *B. nana* (*khokwa*), *B. nutans* (*saneibi*), *B oliveriana* (*khokwa*), *B. tulda* (*utang*/*watang*), *B. tuldoides*, *Chimonobambusa callosa* (*laiwa*), *Dendrocalamus brandisii* (*wamu*), *D. flagellifer* (*longwa*), *D. giganteus* (*maribob*), *D. hamiltonii* (*wanap*/*unap*), *D. hookeri* (*watangkhoi*/*utangkhoi*), *D. latiflorus* (*wui*), *D. longispathus* (*uil*/*unap manbi*/*chingwa*), D. *manipureanus* (*ooe*/*oei*/*ui*), *D. strictus* (*unan*), *Melocanna baccifera* (*moubi wa*), *Schizostachyum dullooa* (*phisautong*/*tolluwa*), *S. munroi* (*naat*), *S. pergracile* (*pungsang*/*pongshang*) and *Thyrsostachys oliveri* (*kabo wa*). The total import and export economy of edible bamboo shoots in the global market is about US$ 1.2 billion, while the Indian edible bamboo shoot economy is estimated to be US$ 0.8 million indicating its great potential for growth and share in the international market. In North East India, a net annual income of US$ 0.7 million is estimated from bamboo shoot trade (Nongdam and Tikendrajit, 2014). Consumption of young succulent bamboo shoots is becoming popular in the modern world because of their exotic taste, flavor, nutritive value and numerous health benefits. Bamboo shoots are rich in dietary fibers, carbohydrates, minerals (potassium, phosphorus, magnesium, calcium and iron), vitamins and phytochemicals (polyphenols and phytosterols), and possess antioxidant, anti-inflammatory, neuroprotective, antiapoptotic, antimicrobial, antifungal, anticancer, antihypertensive and cholesterol lowering properties (Chongtham et al., 2011; Singhal et al., 2013; Nirmala et al., 2014). The bulk seasonal availability only during the monsoon months (May to September), and the highly perishable nature (shelf life of 3 - 4 days) of fresh bamboo shoots necessitate their preservation to ensure availability throughout the year. Age-old indigenous practices of traditional fermentation have been widely practiced in Asian countries to preserve these edible bamboo shoots. In Manipur, fermented bamboo shoot is an important part of the traditional cuisine, and various ethnic communities produced varieties of indigenous fermented bamboo shoots that differ in terms of the raw materials used, preparation process, texture and flavor.

Traditionally, fermentation of bamboo shoot is done in especially designed earthen pots or bamboo baskets/chambers in solid or submerged states via spontaneous fermentation or backslopping approach. Fresh shoots of *B. balcooa*, *B. tulda*, *D. hamiltonii*, *D. giganteus*, *M. baccifera* and *S. munroi* are exclusively used for preparing the ethnic fermented foods (Jeyaram et al., 2009, 2010, Romi et al., 2015). They are mainly produced by the Meitei community and commonly known as *soibum*, *soidon* and *soidon mahi*. Other lesser known fermented bamboo shoots are *thunkheng*, *thunbin*, *thunkhengkang* and *thunbinkang* of the Naga and Kuki communities. Based on the duration of fermentation, fermented bamboo shoots can be broadly categorized into two types: short-duration (2 - 7 days) fermented bamboo shoots, which comprised of *soidon* and *soidon mahi/soijin/soijim*, and long-duration (6 - 12 months) fermented bamboo shoots, which comprised of *soibum*, *thunkheng* and *thunbin*. Based on the difference in the traditional preparation methods and their common practice in specific region of Manipur, the fermented bamboo shoots is further classified into 5 types: Kwatha type, Noney type, Kakching type and Andro type, which comprise of *soibum*, *thunkheng* and *thunbin*, and Lamangdong type, which includes only *soidon*.

Soibum

Soibum is the most common and sought after fermented bamboo shoot of Manipur. It is a non-salted, acidic and long-duration ethnic fermented product produced by the Meitei community. It is prepared exclusively from the young succulent shoot sprouts of *D. hamiltonii* (*wanap*/*unap*), *D. giganteus* (*maribob*), *B. balcooa* (*leewa/ching saneibi*), *B. tulda* (*utang/watang*) and *M. baccifera* (*moubi wa*). It is consumed as an important constituent of the traditional diet. It is similar to *mesu* of Sikkim.

Soidon

Soidon is a non-salted, sour, short-duration fermented bamboo shoot product of the Meitei community of Manipur. It is exclusively produced in the Bishnupur (old name: *Lamangdong*), which is known as the 'place of *soidon* origin'. In earlier times, *soidon* was known as *lamangdong soidon*. The main production of *soidon* is from the small scale traditional production centres of Bishnupur. Unlike all other fermented bamboo shoots of North East India and other Asian countries, *soidon* is unique in the sense that it is prepared exclusively from the apical shoot meristem of *Schizostachyum munroi*, locally known as *naat*. *S. munroi* is endemic to the Ngariyan hill range of Bishnupur district. The commercial production of *soidon* stills depends on the age-old practice of indigenous traditional fermentation (Romi et al., 2015).

Thunkeng and Thunbin

Thunkheng and *thunbin* are minor ethnic fermented bamboo shoots of Manipur, which is prepared and consumed by the Zeliangrong (Naga) and Kuki tribes of Tamenglong and Noney hill districts. *Thunkeng* is the fermented whole succulent bamboo shoot, while *thunbin* is the fermented bamboo shoot slices. They are exclusively from *D. hamiltonii*, *D. sikkimensis*, *D. giganteus*, *M. bambusoides*, *B. tulda* and *B. balcooa*. The traditional preparation for the fermentation of bamboo shoots (locally called *thun*) is carried out during May to June when new shoots sprout during the monsoon. The process is similar to that of Kwatha *soibum* production, but is unique in the sense that the fermentation is carried out inside a pit in the soil. This preparation method commonly known as Noney-type fermentation. A bamboo basket (locally called *khoupak*) in the shape of the pit is made inside it and the inner wall is lined with banana or wild *Colocasia* leaves to keep airtight during fermentation. A hole is made at the bottom for draining the fermentation liquid. The harvested whole tender bamboo shoots are defoliated, cleaned and packed inside the basket for *thunkeng* preparation, while longitudinally sliced, thin small pieces of the shoots are used for the preparation of *thunbin*. The upper portion of the basket is then covered with banana leaves and stones are put on it as weight to initiate the natural spontaneous fermentation. Though the fermentation is carried out for months, *thunkeng* and *thunbin* are sold and used for making curry within 5-10 days of fermentation because of the preference of high degree of sourness by the local tribes. For longer storage, *thunkeng* and *thunbin* are sundried to produce *thunkengkang* and *thunbinkang*, respectively . These dried fermented shoots are kept in a basket to use in off-season. The fermented bamboo shoots are consumed as traditional delicacies by these tribes after boiling with pork, meat and vegetables. *Thunkhengkang* are graded according to size for preparing different dishes. Microbiology, nutritional composition and potential health benefits of these products are unknown though lactic acid bacteria are likely to be the functional

microorganism. (Singh et al., 2018b; Jeyaram et al., 2009). *Thunkeng* and *thunbin* are similar to *ekung* of Nyishing tribes and *hirring* of Apatani tribes of Arunachal Pradesh, respectively.

Inziangsang

Inziangsang, a sour, non-salted ethnic fermented mustard (*Brassica* sp.) leaves of the Naga communities of Manipur. Mainly the womenfolk produce them. It is similar to *gundruk* of Nepali communities of Sikkim. *Inziangsang*, is prepared during the winter season when mustards are grown and available in surplus amount. Withered mustard leaves (locally known as *hanggam*) (2 to 3 days old) are crushed using traditional wooden mortar and pestle and soaked in warm water. After squeezing to remove excess water, the crushed leaves are packed in airtight container and kept undisturbed in the ambient environment condition for 7-10 days semi-solid spontaneous fermentation. The fermented mustard leaves are called *inziangsang*. The fermented liquid is extracted from *inziangsang* by squeezing with hands to get *ziangdui*. *Ziangdui* in further concentrated by boiling to produce a condensed paste known as *ziangsang*. For future consumption and long term storage (> 1 year), *inziangsang* (freshly prepared as well after *ziangdui* extraction) is sundried for 4-5 days and stored at room temperature, while *ziangsang* is stored in traditional bamboo container. (Jeyaram et al., 2009; Tamang and Tamang, 2009).

Ankamthu

Ankamthu is the ethnic fermented mustard green extract prepared and consumed by the tribes (Chiru, Chin, Gangte, Hmar, Paite, Kom, Mate, Lushai, Simte, Thadou, Vaphei and Zou) of the Kuki-Mizo community of Churachandpur district of Manipur. It mainly prepared by womenfolk for off-season consumption and to supplement the economic income of the ethnic inhabitants. It is similar to *ziangsang* of Naga communities of Manipur. (Hoikhokim, 2017). *Ankamthu* is prepared from the extract of mustard green leaves (mainly *Brassica juncea*, *Brassica campestris*). In the generations-old preparation process, bulk amount of mustard green leaves are plucked, washed in tap water and wilted under the sun by spreading over a traditional bamboo winnowing tray (locally known as *pheh*). After crushing the wilted leaves by pounding using a traditional wooden mortar and pestle, the mesh is transferred to an airtight bamboo basket with a lid and kept form somedays. The dark colour leave exudate is then extracted by hand squeezing and boiled in a pot until the extract become condensed. The condensed extract is then filled in batches inside hollow bamboo internodes with one end sealed with the nodes. After sealing the other ends with banana leaves, the bamboo containers are kept near traditional kitchen fire or under the sun for 3-5 days for spontaneous fermentation.

Ganang Tamdui

Ganang tamdui is the fermented mustard leave extract of the Zeliangrong (Naga) and Kuki tribes of Tamenglong and Noney hill districts of Manipur. It is similar to *ankamthu* of Kuki-Mizo community. In the tradition preparation process, mustard leaves harvested from jhum field are wilted until the leaves turn yellow. The leaves are pressed inside a bamboo culm, which is then sealed with banana leaves and mud. The packed culms are kept undisturbed until a pungent smell is released. The fermented leaves are removed and squeezed with both hands to extract the dark color liquid. The extract is concentrated by boiling in a pot to form a condensed liquid known as *ganang tamdui.* It can be preserved in a tightly closed container up to a year. *Ganang tamdui* is mainly used in the preparation of *tam*, an ethnic soup. It is used as a taste and flavor enhancer in various local dishes. Its microbiology, nutritional composition and potential health benefits are unknown.

Ethnic fermented legume and pulses

Almost all ethnic people in Manipur produce and consume some types of fermented seed products. Most important among fermented seed product is the fermented soybean. It is locally known as *hawaijar* in Meitei, *khuichang* in Zeliangrong, *bethu* in Thadou and Kuki, *bekanthu* in Hmar and *theisui* in Tangkhul. Other fermented seed products include *shithu* (fermented sesame seeds), *hangammaru hawaichar* (fermented mustard/rapeseed seeds) and *gankhiangkhui* (fermented Indian roselle seed) of Hmar, Meitei and Zeliangrong communites, respectively.

Hawaijar

Hawaijar is a sticky, naturally fermented ethnic soybean food of Meitei people in Manipur. It is alkaline and slightly ammoniacal making it slightly pungent. It is non-salted, mucilaginous and sticky in nature and

has a unique flavour, but not very soft in texture. The color of *hawaijar* ranges from light grey yellow to tan yellow. It is produced in small quantity in household level and sold in local market by woman vendors. *Hawaijar* is produced and consumed primarily by the Meitei community of Manipur residing in the central valley areas. Traditionally, *hawaijar* is produced by ladies of Meitei Brahmin families, all Meitei ladies know the art of production of *hawaijar* and they usually prepare it for their household consumption from time to time. During the traditional production of *hawaijar* the soybean seeds (*Glycine max*) (locally known as *nunghawai*) are washed and soaked in excess water overnight (usually 8-10 h). Next day, soaked soybeans are properly cooked in a pot or pressure cooker, which is checked by crushing the seeds between fingers and is also indicated by a characteristic aroma released from the broth after boiling several hours. The cooked seeds are washed using boiled water and allowed to drain excess water. The drained seeds are wrapped with the leaves of *Ficus hispida* (locally called *asiheibong*) while they are still warm and kept in a traditional bamboo basket called *lubak*. It is then wrapped with gunny bags or old cloths and placed undisturbed near fireplace or buried in paddy for spontaneous natural fermentation. It takes 2-3 days during summer and 5-7 days during winter for the fermentation to complete. Release of the characteristic ammoniacal, pungent odour with sticky mucilage indicated the proper fermentation of *hawaijar*. *Hawaijar* is then packed in banana leaves in small quantities for sales in local market. A medium to small seed variety of soybean is preferred for *hawaijar* fermentation over large seed variety. *Hawaijar* has a very less shelf life (2-3 days) but it can be stored for about a week in refrigeration without changing its taste and other characters. For longer storage, it is dried for 2-3 days in sunlight by spreading on a bamboo mat or in fireplace. The dried *hawaijar* can be stored for several months to even one year. Similar products are prepared in hill areas by different ethnic tribal communities. In the western hills of Manipur, the Zeliangrong people produces fermented soybean known as *khuichang*, which is very similar with *hawaijar* in production and consumption except that the incubation is slightly longer (3-5 days during summer and 7-9 days during winter). In the eastern hills of Manipur, the Tankhul tribes produce fermented soybean called *theisui*, which is similar to *hawaijar* but ash is added during incubation like the *kinema* of Sikkim and Darjeeling. In the south western hills of Churachandpur district, the Hmar and Kuki ethnic tribes prepare fermented soybean known as *bekanthu* and *bethu*, respectively. (Jeyaram et al., 2008b, 2009; Singh et al., 2018b).

Gankhiangkhui

Gankhiangkhui is an alkaline ethnic fermented *Hibiscus canabinus* seeds (locally known as *shougri/gankhiang*) prepared and consumed by the Zeliangrong and Kuki tribes of Tamenglong district of Manipur. It is consumed as flavour enhancer and protein supplement. The mature seeds are harvested during August after *H. canabinus* plant no longer gives edible leaves. The seeds are harvested by trashing the plants, and then sun dried and kept in tin containers for storage until it is required to prepare *gankhiangkhui*. The process of preparation of *gankhiangkhui* is similar to preparation of *hawaijar*. The seeds were first cleaned, sorted and soaked overnight. In the morning the seeds were boiled for about 3-4 hours until it is soft and easily crushed between fingers. The cooked seeds are drained, crushed lightly, packed in an earthen pot and allowed to ferment (primary) for 3-4 days during which the texture of seeds changes and the colour of the seeds get darkened. During packing to the pot sometimes ashes obtained after burning rice straw are also added. The fermented seeds are then taken out and wrapped in banana leaves in smaller packs and kept in fireplace for 2-3 days until the peculiar odour is released from the incubated products. Thus fermented *shougri* seeds are ready to eat (Singh et al., 2018b).

Sithu

Sithu is a traditional fermented product of *sith* (*Sesamum indicum* L.) with characteristic odour and flavour. It is produced and consumed by Kuki-Chin-Mizo ethnic communities of Churachandpur district of Manipur. It is produced in household level and consumed regularly as food item. The product is prepared and kept in every household throughout the year for its addition in almost every vegetable item to make them tenderer and impart a characteristic flavour to the dishes. The seed of *sith* (preferably black variety) is cleaned by removing unwanted materials after by spreading in a bamboo matt. It is roasted till it produced a distinctive smell which indicates the proper roasting of the seeds. Then, it is pounded by a wooden crusher with addition of some water to make a thick sticky colloid. This is then transfer to an air tight container and place near fireplace for 4-5 days to undergo fermentation.

Hangammaru hawaichar

The Meitei people, particularly belonging to *Chakpa* community, prepare and consume *hangammaru hawaichar*, which is fermented mustard/rapeseed seed. It is strongly pungent and produces a distinctive taste. This product is produced during the months of March-April when the rapeseeds are harvested. The traditional method of preparation is similar to *Kahudi* of Assam. The seeds locally known as *hangammaru* were cleaned, washed and sundried. It is then roasted lightly on a pan before pounding with addition of unripe mango or *Garcinia*. The mixture forms a thick paste, which is transferred in a earthen pot whose inner part is coated with mustard oil. It is covered with banana leaves and allowed to ferment for a week or up to ten days. This product does not require cooking and consume directly from the storing container. It is usually taken as side dish along with other dishes.

Ethnic fermented fish

Ngari

Ngari is a non-salted fermented fish intrinsically bound with the diet of Meitei people and hardly any meal goes without *Ngari*. It is part of the traditional diet that forms an important part of the cultural identity of Meitei community. It is prepared in the valley region by the Meitei community but relished by almost all communities of Manipur. *Ngari* is traditionally prepared from sun-dried *Puntius sophore* (locally *phabou*) caught from streams and rivers and fermented in traditional pots. *Utong-nga* is a similar type of fermented fish, which is prepared in bamboo trunks instead of earthen pot but this method is not used anymore. *Ngari* is generally prepared during October to January as the fish are abundantly available during this period. Due to the non-availability of *phabou* in Manipur for large scale production, it is imported from Assam, Bangladesh and Gujarat (Soibam and Ayam, 2018; Das and Deka, 2012; Tamang, 2010; Jeyaram et al., 2009). The traditional method of *ngari* preparation and its associated indigenous knowledge passed through different generations are properly documented. Briefly, the sun dried whole fish is washed in a porous bamboo basket and the water allowed to drain off for 12-24 hours . Next, the washed fish is spread on a polythene sheet or gunny bags and pressed by legs (which are already covered with gunny bags) or mechanicalrollers to remove excess water and also to break the heads and bones of the fishes. The pressed fish is then packed inside the *kharung* (earthen pot) in batches of about 5kg until it is completely filled. *Kharung* is the traditional round bottom and narrow neck earthen pots with 45-50 kg capacity used in *Ngari* preparation. Before packing, the inner surface of the pot is coated with mustard oil in order to establish anaerobic environment inside the chamber and it also prevents fish from sticking to its surface. Generally, pre-used *kharungs* are preferred for *Ngari* preparation as they need to be oil-coated only once before fermentation while the new pots have to be coated at least 8-10 times intermittently for efficient fermentation. This packing is done by skilled artisans using legs or wooden rods. During packing, the pots are kept in pits lined with moist gunny bags or straw so that the pots remain stable and withstand the pressure of packing. The pots are then finally sealed with a dough mixture prepared by mixing trash fish, oil slurry, sand and cow dung slurry. When the sealed layer becomes dry and starts cracking after 3-4 days, the slurry mixture is applied again to maintain anaerobic conditions. These sealed pots are stacked together and incubated in room temperature for 6-12 months. After the fermentation is completed, the sealing layer is removed and the top layer of the fish known as *phumai* is taken out. *Phumai* is considered to be of low quality and sold with less price compared to *Ngari*, which sells for 500-600 per kg. The shelf life of *Ngari* is about 12-18 months. (Jeyaram et al., 2009; Tamang, 2010; Devi and Kumar, 2012; Das and Deka, 2012; Keishing and Banu, 2015; Thapa, 2016; Soibam and Ayam, 2018; Singh and others, 2003).

Hentak

Hentak is a non-salted fermented fish paste prepared by mixing fresh water fishes (*Esomus danricus*, locally *Ngasang*) and *Alocasia macrorhiza*. It is prepared and consumed by the Meitei community in the valley region of Manipur. It is believed that *hentak* was used commonly before *Ngari* became popular. (Jeyaram et al., 2009; Tamang, 2010; Das and Deka, 2012; Devi and Kumar, 2012) For preparing *hentak*, the sun-dried *Esomus danricus* is grinded into powder and kept aside. Similarly, fresh and chopped petioles of *Alocasia macrorhiza* are washed and sundried for few hours. In the next step, equal proportion of the fish powder and the chopped petioles are mixed together and grinded in a traditional mortar and pestle to form a thick paste. This paste is kneaded into small balls and kept in sealed earthen pots or bamboo tubes and kept at room temperature for natural fermentation. After 10-14 days of fermentation, the fish balls attain its

unique aroma and desirable texture and are ready for consumption. The fish balls become hard on long storage and needs to be kneaded with water and few drops of mustard oil for proper storage and to retain its taste. Although *hentak* can also be prepared with onions instead of *A. macrorhiza*, it results in inferior quality *hentak* with a shorter shelf life. (Jeyaram et al., 2009; Tamang et al., 2012; Kakati and Goswami, 2013; Soibam and Ayam, 2018)

Ithiitongka

Ithiitongka is a unique type of fermented fish, prepared and consumed by the Moyon tribe of Chandel district (hill region) in Manipur. For *ithiitongka* preparation, small fishes caught from local rivers and streams are cleaned and washed thoroughly. These fishes are put into a young bamboo trunk and the mouth sealed tightly with turmeric leaves. This sealed bamboo is kept in the fire till its external layer is partially burnt. It is then allowed to cool off and kept above a fire place to prevent it from spoilage. It can be stored up to two weeks by sealing the bamboo trunk tightly and keeping it over the fireplace. This fermented fish with unique flavour and taste is mainly prepared for household consumption when the fish catch is large. It is not available commercially and no investigation on its microbial profile and nutritional composition is reported. (Wanglar and Tarao, 2018).

Ethnic fermented meat

All the ethnic community in Manipur consume meat traditionally but with strong influence of Hinduism, people particularly, Meitei in valley, stop eating meat although fish is consumed except for few highly religious people who have converted to vegetarian. But the majority of tribal populace in hills districts who have embrace Christianity consume all kinds of meat. Chicken, pork and beef being most common meat consumed by them and these animals are being reared for meat. The hill people occasionally hunt for wild animals for food although it is restricted nowadays by the state government. Every dish has one or other meat item in preparations. Pork being the most cherished of the lot. The meats were required to store and therefore, it is usually processed for longer shelf life. Drying in sunlight and fire place and smoking are commonly practiced in the hills of Manipur but in fewer cases the meats and other parts of the animal carcass is fermented. Most common fermented meat products are fermented pork fat, cow or buffalo fat, fermented cow or buffalo skin, fermented pork meat and fermented bone marrow.

Sathu

Many tribal ethnic communities in Manipur produced and consumed fermented pork fat as traditional delicacies. Kuki, Hmar, Baite and Vaiphei communities called the fermented pork fat as *sathu.* It is slightly acidic (pH 6.3-6.5), have strong odour and oily in nature. It is usually used as taste enhancer in many of the traditional dishes. The boiled pork fats are fermented for 3-5 days in an airtight container to get the product. *Sathu* is similar to *sa'um* of Mizoram, India and similarly it could be infested with *Proteus mirabilis*. Not much study has been done on this product and hence the microbiology and nutritional composition is unknown. Similar product called *gwag ruum* is produced and consumed by Rongmei community in Western hills of Manipur. (Singh *et al*,. 2014). Similar products are also prepared and consumed by these communities using cow or buffalo fat and it called *bongthu.* The consumption and use of the product is similar to *sathu,* but *sathu* is more preferred than the other.

Saayung

Saayung is the fermented pork meat commonly produced by Tangkhul community during the late December, usually after Christmas festival. The excess pork meats during festivals are prepared into *saayung* and store for months for later consumption. The process of fermentation gives a peculiar taste and also made the meat tendered. The product is prepared by boiling and adding salt as well as spices. The product is given to pregnant woman and lactating mother due to the traditional belief that it gives more energy and nutrition to such women.

Guaighi kang

Guaighi kang is a dried fermented skin of cow or buffalo prepared and consumed by Rongmei community in western hills of Manipur. This product is specially used for preparation of special dishes called *tam.* It is prepared by staking the skin of the slaughter animals in an airtight container and keeping for about 7 days to allow fermentation process. The product is taken out from the container and the hairs are scrapped off

completely using a knife. Then it is either sundried or kept near fireplace. It can be stored in ambient temperature for 6-8 months. It is soaked in water to soften before preparation and cooking as the dried skin is very hard

Sahro

Sahro is a valued meat product of Hmar community. During the preparation of *sahro,* any meat but usually pork is half cooked to remove oil from the meat. It is spread in the sun for another 7 days during which fermentation take place. Then the meat is place near fire place. The speciality of this method is it removes more than half of the oil from the meat and it is usually given to old and small kids.

Bongkarot

Bongkarot is a fermented product made from the cow legs. Vaiphei, Baite and Hmar communities prepare this product to enhance its shelf life and for the unique taste. It is prepared by boiling few pieces of cow leg until the meat get tender. *Sathu* is also added during its cooking to give its aroma. After cooking for half an hour the meat become very sticky, during this time, the broth is cool down and packed in a bamboo culm, which is then covers tightly using banana leaves. It is believe that the broth spoils if it is touched by bare hand. It is kept for future use and could be store one month.

Aithu

Aithu is fermented crab, slightly alkaline and pungent food product of Kuki-Chin-Mizo communities of Manipur. This item is a delicacy and serves to special guest. It is not a main course of any meal but a side dish and tastes maker in many vegetarian and non-vegetarian traditional dishes. This product is similar to *Japangangnagtsu* of Ao Naga of Nagaland. *Aithu* is prepared by grinding the already washed and clean crab and mixed with ground *sith.* The mixture is wrapped tightly using banana leaves and kept in an earthen pot. The pot is kept near fireplace or under the sun until characteristic smell comes from the product, which usually takes 3 -5 days.

Ethnic fermented milk (*Sangom aphamba*)

Traditionally, the ethnic people of Manipur do not prefer milk product. In Manipur, traditionally milk is consumed fresh and seldom process. The only known fermented milk is curd locally called *sangom afamba* of Meitei community. The production of *sangom afamba* is simple and similar to *dahi* preparation. During its preparation, fresh raw milk is kept in a traditional earthen pot and a spoonful of curd from previous batch is added and pot is covered with a muslin cloth locally called *sana*. It is allowed to stand for 2-3 days to get the *sangom afamba*. Traditionally it is not used in any local dish preparations but usually taken as raw. It is also taken with steam rice. It serves as one of the essential requirement for performing rituals in certain traditional religious ceremony such *na-hutpa* (ear-piercing ceremony of children).

Thunbin *Soibum* *Thunkheng* *Soidon*

Fig-1: Ethnic fermented Bamboo shoot products of Manipur

Hawaijar *Khuichang* *Bekangthu* *Gankhiangkhui*

Fig-2: Ethnic fermented seed food products of Manipur

Sathu *Guaighi kang* *Phabou* *Ngari*

Fig-3: Ethnic fermented animal and fish food products of Manipur

Conclusion

The ethnic people of Manipur havetheir own unique diet patterns. As such, culture, traditions, ethics and food habit can't be separated or looked separately as they are all interrelated. Nowadays, their approaches to life have changed totally and it is not easy to find the age-old simple life style even in the villages. Traditional foods are still a favourite item in the food preparation. The advent of modern civilization has adversely affected the age-old tradition and thus the younger generations of tribal people of the region are not exposed to traditional practices. As the rich nutritional values of traditional foods and its healing properties are well known, there should be purposeful efforts to revive and promote the traditional food habit systems within villagers. The nutritional and microbiological aspects of traditional foods are required to be investigated in future.

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Prospects of Agriculture and allied Entrepreneurship Development in North-East India

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ABSTRACT

Entrepreneurship is the dynamic process of creating incremental wealth and that permeates an individual's business in an innovative manner that takes risks beyond security, seeks opportunities and having the tenacity to push an idea through to reality. Entrepreneurship to be successful requires proper skills and techniques to manage an enterprise. It involves management just as it is required in managing any other organization. Therefore a proper knowledge of management and its functions can very well be useful for entrepreneurs to be successful. The entrepreneur to be successful has to be a good manager. Entrepreneurship development are (i) Maximization of yield of all component enterprises to provide steady and stable income at higher levels (ii) Rejuvenation/amelioration of systems productivity and achieve agro ecological equilibrium (iii) Control the built-up of insects-pest, diseases and weeds population through natural cropping system management and keep them at low level of intensity (iv) Reducing the use of chemical fertilizer and other harmful agrochemicals and pesticides to provide pollution free, healthy produce and environment to the society at large (v) Increase in natural resource use efficiency by early recycling of nutrients (vi) Mitigation of negative impact of agriculture or livestock on environment. In north east India, efforts were made to develop low cost farming systems suitable for Indian conditions, based on the principles of productive utilization of farm wastes and fuller utilization of available resources and manpower. The sustained research efforts have resulted in the development of integrated farming systems involving fish culture, livestock rising and agriculture. The packages of practices for fish-cum-pig, fish-cum-duck and fish-cum-poultry farming have been developed and verified extensively for economic viability and feasibility at the farmers' level. The manure of livestock contains a substantial amount of nutrients and biomass and its application in the pond helps in growth of Phytoplankton and Zooplankton, which act as a source of nutrients for fish. Indigestible feed ingredients present in manure directly consumed by the fish. The wasted feed from feeding through also can be recycled through fish culture. Time to time the bottom soil layer of the pond is removed and applied in the field for crop or vegetable farming which is rich in biomass.

Keywords: Entrepreneurship development, Integrated Farming System, Livelihood Development and Indigenous Technological Knowledge.

INTRODUCTION

Good Entrepreneur can create a strong economy. Entrepreneurship is an important facet of industrial growth and development of a nation. It is the backbone of a nation that sets its eyes on maximizing its performance in every field. The spirit of entrepreneurship brings about enthusiasm, persistence and the ability to seek entrepreneurial opportunities that lead to success. A nation's ability to generate a steady stream of business opportunities can only come about when its people take to entrepreneurial activities. Entrepreneurs are essentially the engines of growth for a nation. There are several factors that go into making a successful entrepreneur, and he or she need not necessarily possess a strong business and financial background. On the contrary, well-conceived and well directed training can always produce an outstanding entrepreneur. In addition, today's world with its burgeoning population offers limited avenues of employment.

The North East of India comprising of the 8 states of Assam, Arunachal Pradesh, Manipur, Meghalaya, Mizoram, Nagaland, Tripura and Sikkim, is a reservoir of rich natural resources and a beautiful amalgamation of different people and cultures. It surely is a region waiting to happen. Blessed with biodiversity, huge hydro-energy potential, oil and gas, coal, limestone, forest wealth, fruits and vegetables, flowers, herbs and aromatic plants, rare and rich flora and fauna, NE India has all the potential to transform into a commercial hub and tourist paradise.

The region shares borders with China in the north, Bangladesh in the South-West, Bhutan in the North-West and Myanmar in the East. This makes the North-East a prospective hub of international trade and commerce. Unlike the rest of India, North East India has an added demographic advantage, in the sense that it occupies 7.8 per cent of the country’s total land space but has a population of 3.8 crore, which makes it approximately 3.73 per cent of the country’s population. This is also a huge untapped, emerging market, which should prove to be of interest to large domestic and international investors.

The area is a vibrant source of energy rich in oil, natural gas, coal, limestone and India’s largest perennial water system, the River Brahmaputra and its tributaries, which can be tapped for energy, irrigation and transportation. The fertile soil around the valley of the River Brahmaputra is a veritable storehouse of horticultural products/plantation crops/vegetables/spices and rare forest products. Subsidies on transport, capital investment, interest on working capital, excise duty refund, income tax exemptions etc. are available for industries in the region, as declared in the new North East Industrial and Investment Promotion Policy 2007 (NEIIPP). Attractive incentive structures should attract new domestic and foreign investments into the region, which, despite having several advantages, has not witnessed the kind of fast-paced growth and development that it should have experienced in the past years.

North East India also offers huge opportunities in sectors of strategic importance like energy and infrastructure; oil, natural gas and hydrocarbons; agro, food processing and horticulture; floriculture; IT and IT S; cement; defenses, etc. Tourism is another potential high growth industry. Manipur has huge potential in sectors like power, agro-based industries, etc.; a greater private sector involvement in the state economy will ensure local job creation and entrepreneurship development. The State Government of Manipur lays a major thrust on Agro and Food Processing; an Agric Export Zone for Passion Fruit is being developed in the state.

The literacy rate in Mizoram is above 90%, which is the 2nd highest in India. A well-educated and hardworking young population is an advantage for Foreign and also domestic corporate; sectors like IT and ITeS, agro processing, floriculture and bamboo are some areas that offer huge opportunities to investors. The state also grows fine quality of grapes, which can be utilized for large-scale wine production. The State Government of Manipur is pro-active and industry-friendly, and investors can be assured of single-window clearances. Assam provides huge investment opportunities in sectors like hydrocarbons, oil and natural gas, bamboo, handicrafts and tea. Assam tea is well known for its distinctive quality, and exported to the developed markets of Europe. Sectors like IT, hospitality and tourism, power, agro and food processing are poised for impressive future growth - companies like Infosys and Reliance have already shown interest in investing in Assam, with Tata Consultancy Services even having a BPO unit in Guwahati. All the North-

Eastern Indian States have distinct advantages, and provide immense economic and trade opportunities to domestic and international corporate.

WHO IS AN ENTREPRENEUR?

The entrepreneur as a person brings in overall change through innovation for the maximum social good. Human values remain sacred and inspire him to serve the society. He has firm belief in social betterment and he carries out this responsibility with conviction. In this process, he accelerates personal, economic as well as human development. The entrepreneur is a visionary and an integrated man with outstanding leadership qualities. With a desire to excel, he gives top priority to Research and Development. He always works for the well-being of the society. More importantly, entrepreneurial activities encompass all fields / sectors and foster a spirit of enterprise for the welfare of mankind.

OBJECTIVES OF ENTREPRENEURSHIP

- To create entrepreneurial awareness to the young generation and motivate them to establish small and medium enterprises
- To trained up the prospective new and young entrepreneurs to maintain and run their enterprises successfully.
- To enabling the entrepreneurs about the global production and market strategy in context of market economy
- To encourage the entrepreneurs to establish pollution free enterprises

IMPORTANCE OF ENTREPRENEURSHIP

- Entrepreneurship promotes capital formation by mobilizing the idle saving of the public.
- It provides immediate large-scale employment. Thus it helps to reduce the unemployment problem in the country, i.e., the root of all socio-economic problems.
- Entrepreneurship promotes balanced regional development.
- Entrepreneurship helps reduce the concentration of economic power.
- It stimulates the equitable redistribution of wealth, income and even political power in the interest of the country.
- It encourages effective resource mobilization of capital and skill which might otherwise remain unutilized and idle.
- It also induces backward and forward linkages which stimulates the process of economic development in the country.
- Entrepreneurship also promotes the country's export trade, which is an important ingredient to economic development.

ENTREPRENEURSHIP IN INDIA

"Entrepreneur is one who always searches for change, responds to it, and exploits it as an opportunity". Entrepreneurs innovate and innovation is a specific instrument of entrepreneurship. It creates resource because there is no such thing as a 'resource' until man finds a use for something and endows it with economic value. India is second among all nations in Total Entrepreneurship Activity as per the Global Entrepreneurship Monitor Report 2002. Over the years India has concentrated more on the development of the institutions that support private enterprise by building a stronger infrastructure to support it. The Government has encouraged entrepreneurship by providing training and also the facilities to succeed, particularly in the rural areas. Moreover, in India, the post-liberalization and globalization era has brought with it a growing middle class - roughly estimated to be 250 million - and rising disposable incomes. This presents a huge potential, which if tapped can be a veritable gold mine. Entrepreneurs can make the best of this by catering to various demands of this segment.

QUALITIES OF AN ENTREPRENEUR

The important qualities of a successful entrepreneur are as follows,

- Total commitment, determination and perseverance
- Drive to achieve and grow
- Opportunity and goal orientation
- Taking initiative and personal responsibility
- Persistent problem-solving
- Realism and a sense of humor
- Seeking and using feedback
- Internal focus of control
- Calculated risk taking and risk seeking
- Low need for status and power
- Integrity and reliability

CHALLENGES TO THE ENTREPRENEUR IN INDIA

Entrepreneurship plays a vital role in economic development through creation of utilities and generation of employment within a short period. It has been accepted that entrepreneurship is an economic venture by which the people can be changed to move upwards within a short period of time, especially from the point of view of employment generation. Out of the economically active population of 550 million at present only 27.53 million people have had the benefit of employment in the organized sector. 249.76 million are either employed or under employed in the unorganized sector, while the services of the remaining 272.71 million remain unutilized by the entrepreneur. As a result over 29 % of the population continues to suffer under the clutches of poverty and hunger.

The founding of new firms has always been a focal research issue in the entrepreneurship research tradition. In order to support the founding of new firms, it is important to know who starts new firms, in what kinds of situations, and for what kinds of reasons. The early research on entrepreneurship strove to identify the psychological characteristics, or traits, of those people who start new firms. One problem of this 'trait' line of research was that it focused on ex post situations, interviewing entrepreneurs who already had started a firm.

Prospects of Agric and allied Entrepreneurship Development Opportunities in North-East India

- **Pig Production for the Development of NE**

Table-1: Human and pig population in the NES of India

S.No.	Arunachal Pradesh	Assam	Manipur	Meghalaya	Mizoram	Nagaland	Sikkim	Tripura	NE India
Area (KM)	89743	78438	22327	22429	22081	16579	7096	10486	263179
Population (000)	1203	29564	2374	2540	973	2180	593	3505	42932
Pigs (000)	330	1543	415	419	218	644	38	209	3816

Source: North Eastern Development finance Corporation Databank (http://www.db.nedfi.com)

Table-2: Per capita consumption of pork (Kg. per annum) in urban and rural areas and for rural social groups in three northeastern states

State	Rural	Urban	Scheduled Tribes	Schedules Castes	Other Backward Caste	Others
Assam	0.02	0.61	2.26	0.44	0.49	0.21
Meghalaya	3.26	2.04	2.14	0.00	2.26	0.15
Nagaland	9.54	7.18	7.45	1.61	4.14	1.80

Source: National Sample Survey Organization (2003)

Assam and Nagaland were selected for the appraisals because Nagaland has the highest pig density of the eight states (0.6 pigs/person; table 1) and Assam has by far the largest human population (about 70% of the region total; table 1) and is the largest market for pork.

North East States of India has much higher pork consumption that the rest of the country. Table 2 shows that consumption for the three States, of these states; Nagaland has the highest per capita consumption. The tribal population in particular appears to consume more pork on average than other groups.

- **Mushroom Production for the Development**.

In the hill states of North-eastern region, mushrooms are a highly coveted item of food. The different kinds of edible and non-poisonous mushroom that are consumed in the region grow wild. There have been regular systematic plans at present to promote domestic cultivation of mushrooms. The horticulture department of almost all states do have scheme for promotion of mushrooms. And yet, commercial production of mushroom to a significant extent has not taken place so far. The efforts in this direction continue, both by the Government agencies and by voluntary social service organizations. These efforts must continue.

The region is not easily accessible by normal surface or water transport, which is one of the reasons which inhibit the rapid economic development of this region. Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland and Tripura constitute the so-called NE, but for development programmes, Sikkim has also been added. As the industry is almost non-existent and agriculture is of subsistence in kind in most parts, unemployment and unemployment prevail. Any program for economic development of this region might take into account the profitability of the venture, employment generation suitability of the climate and linking of the people.

A low volume-high value commodity venture like mushroom production is perhaps a highly suitable agro-industrial venture. It is not only highly profitable but is also labour-intensive, and above all not dependent on arable land. Mushrooms are grown indoors and rooms or huts can be raised on otherwise idle land such as slopes. The venture, being indoor highly suited to womenfolk. Being highly profitable and labour intensive, it can attract the youth.

Advantage NE

- Varied climate (5-35^{0}C) suitable for all mushroom
- Abundant agro wastes (raw materials)
- Excellent domestic (local) market
- Mushrooms as food very popular with the NE hill people
- High humidity (always above 60 per cent)

- **Poultry Production for Development of North-East of India**

Around 70 percent of Indian population is depending on agriculture and animal husbandry of which 30 percent are landless. Poultry farming provides employment to five million people of the country with a population of 30.1 million poultry. India rank 5th in egg production in the world producing about 38.5 billion eggs (2002). Its broiler production was 500 million tones and ranks 19th in broiler production in the world.

Backyard Poultry Production

It is an age old system is that or customary system of poultry farming in the rural condition of North-east region (NER). From the study it is observed that more than 80 percent of the poultry farming is done under this system in NER.

Advantage of such system is that there is less expenditure in chick feed and medicine, besides taste of local fowl is considered most protein us. Eggs are also highly tasty because of the deep yellow colour of the egg yolk. Plenty of beta-carotene is available from such egg.

Kitchen waste, broken rice, paddy, grain, leafy vegetables, etc. are used as ideal feed for the backyard poultry. In addition to this, bird itself can find out worms and insects by perching on the soil etc. Hence, it saves from heavy expenditure. Market price of poultry feed at Imphal (Rs/kg) is as follows:

- Pre-starter 28-30
- Starter (broiler) 28
- Grower mash 27-28
- Layer mash 26

Local cock and hen weigh on an average 1.44-2.2 kg and 1.3-1.8 kg, respectively. Average laying ability is 30-40 eggs per lying, and quickly brooded. Hatchability under natural condition is 80 percent, i.e. out of eggs, 11 chicks are able to hatch out. There is no expenditure for hatching.

Housing is very simple with some laying basket kept on top of the house/Bayer for night hostage. Disease resistance is another quality of local bird and can resist many diseases including Ranikhet, infectious bursal disease, foul pox, etc.

Improvement suggested

Better breed like Giriraja, vanaraja, Dahlem red, naked neck gene line, dwarf chicken (coloured), Grama priya, etc should be introduced. In Manipur Giriraja is found suitable and Central Agricultural University, Imphal is presently keeping 3000 parent stocks, and has distributed at least 84,000 chicks to Manipur and Neighbouring states

Fermented Bamboo Shoot for Development in NE of India

SOIBUM (FERMENTED BAMBOO SHOOT)

Soibum is a whitish fermented product of succulent bamboo shoots having characteristic flavor and taste, produced through the main process of fermentation without the addition of salt and others. Because of high demand of the fermented bamboo shoots the succulent and soft shoots are converted into another acidic food through the process of fermentation. The spontaneous fermentation of succulent bamboo shoots helps in the preservation of the fermented products which is known as the Soibum by the local people of Manipur.

The traditional processing of Soibum is undertaken without salt treatment. The formation is found to the conducted in different ways, and has been adopted according to the skill of the practitioners. The traditional way of fermentation can be broadly divided into two ways:

i. Noney type and
ii. Andro type.

Nonay type of fermentation of Soibum is found to be very common and popular. The fermentation in Nonay type is mostly conducted in chamber made of plank and bamboo and has no specific size, and the chamber is built up according to the convenience of the practitioners. The bottom of the chamber is perforated and the upper surface has one lid. The walls of the chamber on all sides except the upper one are first lined with the leaves of a specific plant. The leas enclosure is then followed by the lining of polythene sheet, and this polythene occupies the innermost lining.

The thin slices of the succulent bamboo shoots are then pressed compactly into the chamber. There are after filling the chamber with the slices to the capacity the upper surface scaled first with the polythene sheet and then with the leaves. The weights are then put on the mash for proper pressing. It is reported that production of good quality soibum can be achieved with adequate pressing of the mash. It has been reported that with the introduction of polythene sheets, it is used as the innermost lining of the chamber. It may be said in the primitive method of manufacturing soibum, the polythene sheets was not used Andro type of fermentation has been undertaken in another design. The practitioners adopted roasted earthen pot as the chamber of fermentation. The pot has bulky body with narrow neck. The succulent shoots which are then prepared into slices. Neither the leaf nor polythene enclosure is used for this purpose. As portion of the pots are filled with the slice. After the fermentation has been commenced as notices from the evolution of the characteristics smell of soibum, another mash of raw material are added. The procedure continues till the pot is filled either the bamboo slices.

The Nonay type soibum has more acidic as compared to Andro type. Moreover, there is no elaborate change in the colour of the succulent shoot. The Andro type is associated with production of brownie soibum.

Extensive biochemical analyses conducted in soibum indicate the contents of protein, free amino acid, reducing sugar, non-reducing sugar, total sugars, as carbic acid and thiamine.

- **Fermented Fish Production for Development in NE of India**

NGARI (FERMETED FISH) DRY

Ngari or Utonga- fermented dry dish locally known as ngari or utonga is a fermented by 3-6 months fermentation of sundried small sized fish-*Puntius sophore*, *P. ticto* in air tight containers.

Ngari is usually taken in vegetable or meat curry preparations, chilly preparations to have the flavor. Through ngari is consumed widely and is produced and sold in a large scale.

Preparation of 'Ngari'

The main steps or the process for the production of fermented fish, 'ngari' involve a brief washing of the sun-dried fishes, followed by drainage and then left dry for 24-48hrs.This makes the fish body rofter.The fishes were pressed hard using stone roller to breakdown head and bones before filling the fishes, a thin layered of mustard oil was applied to the inner wall of the pot'kharung' to cheek porosity. Then the pots were fixed half to the ground using sand to avoid from movement. Fishes were pressed hard mechanically inside the pots using wooden stick. The pots and the final product obtained is ready for consumption.

Preparation of 'Ngari' under laboratory condition

Sun dried *P. sophore* was also fermented under laboratory condition following the traditional method. In the laboratory fish pots were prepared in glass bottles instead of earthern pots used in natural(traditional) mehod. The fish pots were then stored at different temperature (20^{o}c, 30^{0}c and 40^{0}c) using BOD incubator .

ENTREPRENEURIAL OPPORTUNITIES

1) Packaging for Exports

Packaging standards, techniques etc. training programmes on packaging for exports are organized in various parts of the country. Role of packaging for exports has gained much significance in view of trends in the world markets. The need for better and scientific packaging for exports from small sector was recognized long back. These programmes are organized in association with Indian Institute of Packaging which has requisite expertise on the subject. Basic objective of these programmes is to generate the much needed consciousness in the industry and to educate the entrepreneurs about the scientific techniques of Packaging.

2) Technical & Managerial Consultancy Services

Technical & Managerial Consultancy Services to the entrepreneurs is provided through a net work of field offices of SIDO so as to ensure higher level of production and generation of higher exports.

3) National Awards for Quality Products
With a view to encourage the small scale units for producing Quality goods, National Awards for Quality Products are given to the outstanding small scale units, who have made significant contribution for improving quality of their products. National Awards encourage entrepreneurs to produce quality goods which further enable them to enter into export market.

4) Marketing Development Assistance
Marketing Development Scheme is being operated by Ministry of Commerce under which MDA is given to exporters through FIEO and Export Promotion Councils/ Commodity Boards to plan their marketing strategy for export growth.

5) Promotional Schemes
To meet the challenges of international competition and to promote exports of entrepreneurs, following promotional schemes are also being implemented.

1. Technology Development and Modernization Fund Scheme

2. Quality Awareness Scheme

3. Subsidy for obtaining ISO-9000 quality Certification

4. Process-cum-Product Development Centre

6) Small Industry Cluster Development Program
A new scheme for technology up gradation for industrial clusters has been started recently. 10 clusters of industries producing different groups in various parts of country have been selected. The scheme aims at diagnostic study of the clusters, identification of technological needs, technological intervention and wider dissemination of information and technology within the clusters. The expenditure involved on pilot plants etc. is to be met on 50:50 cost sharing basis by the Government and the concerned Industry Association of the clusters. The scheme is flexible and provides for smooth sourcing of technology even from abroad.

7) Commercialization of potential fruit crops
Manipur has vast potential for commercial plantation of fruits like pineapple, passion fruit, lemon, lime, orange, banana, amla, olive, etc. Both the two most-sought-after varieties of pineapple viz, Kew and Queen are produced in commercial scale and the availability for eight months make the State highly advantageous for processing and export. Passion fruit, both as fresh and in processed form like juice and concentrates has excellent market world-wide because of its unique taste and flavour. Besides, many other fruits like the famous Kachai Lemon, Khasi mandarin etc. are in high demand for its high quality. Manipur also has a great advantage of being linked with Myanmar and South-East Asian countries for exporting its produces.

8) Vast scope for area expansion under fruit crops:
Manipur has plenty of scope and potential to grow various horticultural crops because of varied agro-climatic condition. However, so far only 14.5 per cent of the identified potential areas for horticultural crops have been covered. With 9/10 of the geographical area covered with hills, there is ample scope for bringing more land under fruit cultivation in the hill areas. Fruits can be grown on uneven and undulating lands and in this way it gives additional benefits as forest cover for such lands which are liable to run off and erosion. Fruit like Cashewnut can be grown successfully even in cultivable wasteland.

9) Scope for increasing productivity:
There is enough scope for increasing farm returns and productivity through the adoption of improved package of practices like INM, IPM, good irrigation system, use of quality seeds and planting materials, high density planting etc. Based on the experimental finding about suitable planting density of pineapple for the foot hills of Manipur.

1. Scope for organic farming of fruits
With increased awareness and health consciousness among the people, organic food products, especially the fruits and vegetables are slowly gaining momentum in the country and in foreign markets like USA, Europe and Japan and fetching premium prices. Trends indicate that the organic food market would grow

substantially in most of the European countries USA and Japan. Currently, the demand out places the supply, Manipur has a big advantage of developing organic farming of fruits since more than 60 % of arable land is under traditional agriculture, where no synthetic inputs are being used. APEDA has set up one model organic farming project for passion fruit at Mao of Senapati District by transferring the existing farm in about 10 hectares through one NGO, namely "Good Samaritan Social Service".

2. Exploration of natural resources

Manipur has vast natural resources which are unique and has the potential of becoming the cynosure at the world stage and thereby becoming the continuous income and employment generating source and of course the human resource of the State.

3. Increasing market for processed fruit products

With changing food habit, the demand for processed and value added fruit products is increasing rapidly. At present, many of such processed fruits are imported to Manipur from other States and neighbouring countries through Myanmar. But, it has opened a vast opportunity for developing food processing industries in Manipur with the establishment of various processing units like Exotic Juice Limited, Mao, Pineapple Processing and Cold Storage Unit, Matai, Heiron Food Processing Centre, Moirang to name a few.

4. Development of fruit processing industries

Food processing industry is a major thrust area in the State of Manipur as it plays a significant supportive role in the diversification and commercialization with value addition of agro-horticultural products. Blessed with suitable agro-climatic conditions, Manipur abounds in varieties of raw material resources for food processing industries. The State has tremendous potential for further development of food processing industries based on locally available materials like pineapple, orange, Lemon, banana, passion fruit, papaya, guava etc.

CONCLUSION

Per capita availability of land in India has declined from 0.5 ha in the year 1950-51 to 0.15 ha in the year 2000-01. Due to conversion of valuable irrigated agricultural lands for non-agricultural purposes viz. residential houses, industrial and business establishments and subdivision and fragmentation of holdings, the per capita availability of land is declining day by day. Therefore, no single farm enterprise is able to meet the growing demands of food and other necessities of the small and marginal farmers. A judicious mix of cropping systems with associated enterprises like fruits, vegetables, flowers, dairy, poultry, duckery, piggery, goatary, fishery sericulture etc. suited to the given agro-climatic conditions and socio-economic status of the farmers shall be able to generate additional employment and income for the small and marginal farmers both under rainfed and irrigated conditions. Improved agricultural technologies even when considered technically sound for individual component of the farming system are of limited value if they are not adopted by the farming community.The farming system, as a concept, takes into account the components of climate, soil, water, crops; farm wastes livestock, land, labour, capital, energy and other resources with the farm family at the centre managing agriculture and related activities. This type of research is most appropriately carried out by the interdisciplinary team of scientists who in association with the Extension Officers continuously interact with the farmers in the identification of the problems and finding their solutions. Farming system approach to agricultural research and development efforts would accelerate agricultural growth of the country and thereby providing leverage for transforming poverty prone rural India to a prosperous India by strengthening rural economy. Certainly this will play a key role in agricultural revolution in the 21st Century, which is very much important to make India a developed nation.

FUTURE STRATEGIES FOR DEVELOPMENT OF FRUIT GROWING:

- Development of agro-techniques and improved varieties to increase productivity.
- Evaluation of high yielding varieties of fruit crops for Manipur condition and identification of resistant rootstocks.
- Gradual expansion of area under recommended varieties using cultivable waste land.
- Sequential replanting of unproductive orchard through participatory approach.

- Area expansion of potential crops like coconut, Cashewnut, litchi etc. in valley and peach, pear, plum, kiwi etc. in hill region.
- Popularization of technology like high density planting, mulching, drip and sprinkler irrigation systems etc.
- Research and extension on different models of fruit based cropping system for the State.
- Research and demonstration on Integrated Nutrient Management, Integrated Pest and Disease Management in Horticulture Based Cropping System.
- Development of basic infrastructure like transport, pre-cooling and grading unit, storage, pack house, refrigerated containers etc.
- Development of community based fruit processing industries
- Strengthening of effective linkage between State Government and ICAR along with CAU and other organizations like NGOs, NHB, NABARD and other funding agencies.
- Strengthening of Growers' Cooperatives, SHGs etc.
- Strategic advertising by using print, electronic and web media for marketing and export promotion of fruit crops with registered TRADEMARKS like "Manipur Orange" or "Kachai Lemon".

About the Book

This book ***FOOD BIORESOURCES AND ETHNIC FOODS OF MANIPUR, NORTHEAST, INDIA*** is primarily aim at documenting the rich heritage of foods of ethnic people of State of Manipur, India. The State of Manipur which have about 90% hilly region and less than 10% valley area with altitude ranges from 180 msl to 2800 msl covering tropical, subtropical and alpine type of climate with average rainfall of 1200 cm of rainfall per annum. The forest cover is about 60%; limiting agricultural areas to less than 30% of the land mass. The dependence on natural forest for food and other needs leads to identification and consumption of wild edible plants and unconventional foods like insects, frogs, snakes etc. The valley had vast wetland areas which provide ample scope for fishing from wild.

The seasonal foods particularly, wild edible plants and fishes were processed and stored for yearlong consumption during offseason. The traditional processing were simple like fermentations, sun drying, roasting, boiling etc were commonly practiced. Here, traditionally salt was consider a luxurious item thus it was consciously missing during fermentation and other traditional processing. In this book various food bioresources of Manipur were documented with its nutritional medicinal and social values along with perceived values. Apart from farm produces, wild edible plants, aquatic plants, wild edible macrofungi, bamboo shoots, fish, insects and meat and milk were detailed in depth. It is believed that this book will help immensely to those foodies looking for exotic foods, researchers, anthropologist and lastly to the future generation who were alienated from their traditional foods due rapid urbanisation and fast life style.

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