Chapter 1

Introduction

and

Objectives of the study
The sedge family (Cyperaceae) is with about 5000 species under 104 genera distributed worldwide except Antartica, making it the 3rd largest family of monocots (Goetghebeur, 1998). The cosmopolitan genus Carex L. represented about 1,800 species mainly in temperate and cold regions of the world (Mabberley, 2008). The genus Carex L. of Cyperaceae is with the largest number of species, about 2000 according to Goetghebeur (1998). However, Bernard (1990) had estimated more than 2000 species distributed worldwide. Although its main centers of diversity are in North America and East Africa (Starr et al., 1999), and is also well represented in South East-Asia and India, especially in high lands. Carex L. is also one of the most important genera in this family Cyperaceae. Although the species are extending throughout the world, but most of them are occurring in Northern temperate zones (Jermy and Tutin, 1968). Different species are found to occur in wide range of habitats (Schutz, 2000). Most of the species are able to spread laterally by rhizomatous growth and as a result, large clonal patches / tufts may be formed (Bouzille, 1992). The genus Carex L. placed under tribe Cariceae Pax comprises roughly 40% of the family by species making it one of the largest genera of angiosperms (Reznicek, 1990; Mabberley, 2008). Carex species are economically important members of flood plain forests, dry prairies, alpine meadows, peat lands, swamp forests, sedge meadows and a wide range of other communities (Reznicek, 1990). The genus is having diverse biogeographical patches as Gondwanaland, Arcto-Tertiary and Bipolar (Nelmes, 1949; Ball, 1990). It is also remarkable within Cyperaceae because it is easy to recognize as all the species of Cariceae Pax are distinguished by the presence of consistently unisexual flowers where a naked gynoeicum is surrounded by a flask-like prophyll known as a perigynium or utricle (Blaser, 1944; Gehrke et al., 2012).

Although the genus is easily defined and presumably natural, the infra-generic taxonomy of the genus is complex and still unresolved. The floral structures are highly reduced (Clarke, 1893-1894; Kukenthal, 1909; Metcalfe, 1971). The anatomy is generally uniform (Metcalfe, 1969 and 1971; Reznicek, 1990) and species are often distinguished by only a single character or by many small quantitative differences (Kukenthal, 1909; Hamlin, 1959; Naczi, 1992). Moreover, due to cosmopolitan distribution having its enormous diversity make it very difficult to study the entire genus. Most studies are regional and focused on a
portion of sectional diversity only (Mackenzie, 1935; Nelmes, 1951; Standley, 1987; Dai Lunkai et al., 2010). The regional basis study provides that in the family Cyperaceae there are 865 species under 33 genera (326 species endemic and 5 species introduced) distributed only in China (Dai Lunkai et al., 2010). Further it is also mentioned that out of about 2000 species of the genus Carex 527 species are found in China including the estimation of 260 endemic species. The species are included under 3 subgenera and 69 sections (2 endemic) in China. Thus within South-East Asia including China Carex L. is with highest diversity of species.

Clarke (1893-94) had reported 142 species of Carex L. from the then British India of which 69-72 are found to be restricted in North-Eastern India. Rao and Verma (1982) reported 49 species of Carex found in North-Eastern India and 33 species in the rest part of India. Moreover, North-Eastern India harbours the largest number of endemic species of the genus. Other species are found to be restricted to the rest part of India and then also in China, Japan and South-East Asia. Interestingly all the species investigated here in this study are also present in China (Dai Lunkai et al., 2010). Accordingly Clarke’s sub-area is with the total of 46 species of Carex L. found in the then Assam as a phytogeographical region of India which is now considered as Assam, Meghalaya, Nagaland, Manipur, Mizoram and Tripura. Of course, there are 7 species which are found common to the Western Himalaya, 5 to Indian desert, 12 to Peninsular India and Sri Lanka, 1 to Gangetic plain and 26 to Eastern Himalayas (Rao and Verma, 1982). Karthikeyan et al. (1989) had reported about 163 species of the genus in the present day India. Sikkim and from the Darjeeling hills of West Bengal are the main habitats of different species of the genus Carex L. (Rao and Verma, 1962; Karthikeyan et al., 1989). The study of Noltie (1994) had reported 73 species of Carex L. from Bhutan, Sikkim and from the Darjeeling Himalayas along with several infra-specific taxa as subspecies and varieties. Further more, according to Noltie (1994) 55 species out of these 73 are found in Sikkim and the Darjeeling Himalayas and 26 species are found only in Darjeeling Himalayas. Hazra and Verma (1996) have reported 62 species of Carex L. along with several varieties from Sikkim and Darjeeling Himalayas. Dai Lunkai et al. (2010) in Flora of China had reported 530(of which three species under taxa incertae sedis) species under 69 sections of which about 66 species (one species treated as taxa incertae sedis) found in India. The taxonomy of the genus Carex L. in general and sections of Carex L. in particular, both are controversial issue. It has stirred emotions since the mid 18th century i.e.
since the description of *Carex muricata* Linn. by Linnaeus (1753). For more than two centuries no universal, unchanging key or taxonomic classification has been created.

Phylogenetic relationships within *Carex* L. are poorly understood. Traditional sectional arrangements based on morphology (Kukenthal, 1909; Mackenzie, 1931-1935) are generally recognized as being inadequate (Reznicek, 1990; Ball and Reznicek, 2002) to know the details of all. The number of species involved and their broad global distribution has hampered efforts to produce a vigorous modern revision. Starr *et al.* (1999) represents one of the earliest efforts to develop a molecular phylogeny at the sectional level. Their study, based on molecular sequences, produced strong evidence for the morphology of *Phyllostachys*, but they didn’t sample enough to allow for confident interpretation of sectional arrangements. Yen and Olmstead (2000) sampled two genes regions (*ndhE* and *trnL* intron /trnL-trnF spacer) from more representative sets of species from across the genus (and tribe). However, with only 33 taxa their work was also preliminary in nature. Their data is also strongly supported the traditional arrangements of *Carex* L. as part of the tribe *Cariceae*, but allowed only limited resolution of intra-generic relationships. Roalson *et al.* (2001) included 100 species of *Carex* L. to explain the phylogenetic signals in ITS and *trnL*-T-F spacer regions. Even with this expanded sampling effort they focused primarily on the phylogenetic position of a single section *Acrocystis*. The obtained results show that this section to be a polyphyletic one, also confirmed some of the results of earlier studies as the subgenus *Vignea* is monophyletic; the other genera of the tribe *Cariceae* are nested within the genus *Carex* L. In an effort to classify and to show phylogenetic relationships within *Acrocystis*, Roalson and Friar (2004) expanded their views to include ITS and ETS sequences from 22 taxa from that section. However, the resulting phylogeny was poorly supported. Waterway and Starr (2008) sampled three or more gene regions (nuclear ETS and ITS and several non coding chloroplast regions) from over 100 *Carex* species. This work produced much better support for clade within *Carex* L. than any of the other works.

**Objectives of the study**

There are different counts or the estimation of the species of *Carex* L. that are distributed in Sikkim and Darjeeling and other part of the North-eastern Himalayas. So, proper taxonomic interpretation, identification and distribution pattern of *in-situ* population is needed. In this study it has been tried to identify the collected specimens not only on the basis
of morphological study but also with the vegetative anatomy as well as fruit epidermal study including the presence of silica bodies that have been used to delimit species and sections of *Carex* L. Although frequent conflicts with traditional classifications have led many authors to question whether they can be used to interpret evolutionary relationships (Starr and Ford, 2001).

Morphological features are not a clear criterion for delimiting the individual taxa as expressed by Magdalena and Klimko (1999). In agreement with the suggestions of many researchers, anatomical features have been taken into account, as they are relatively constant and are not as strongly modified as morphological characters. Never the less, even they can be variable to some extent within the genus. In this study an attempt was made to examine if these features are variable within a much narrower taxonomic category. Therefore, the major aim of this work was to compare and to analyze similarities and differences in the anatomical features of both vegetative organs as well as reproductive organs of individual taxon. It can be noted that no comparative studies of the anatomical structure of different species of the genus *Carex* L. from this region had been studied before. The objective of this investigation was to study the anatomical characteristics of the different species found to occur in this area.

It has been expected that from the present study the data from both the morphological and the anatomical characters will eventually come to play an important role in the understanding of the systematics of different species of *Carex* L.

Prior to the findings of Metcalfe (1969, 1971) less attention had been given by the anatomists for the anatomy of the Cyperaceae in general and *Carex* in particular and thus there were very less scattered information. Still now the information on anatomy of *Carex* in comparison to the other genera of Cyperaceae is less though the genus is more or less distributed throughout and over 2400 species had been reported. It is, further, seen that the anatomical works of Indian species of *Carex*, the scenario is very very poor though more than 150 species of *Carex* had been reported by different authors at different times which were mostly identified on the basis of reproductive morphology only with a few very scanty exceptions. Thus the anatomical works of the genus *Carex* L. of Cyperaceae have been taken as a problem of this study along with the morphological study of both vegetative and reproductive structures. Thus it is expected that both the studies can provide a proper picture of the systematic of different taxa of *Carex*. This work can provide some anatomical
information for the species from North-east India particularly from Darjeeling and Sikkim Himalayas and can be used as additional information for those works conducted by or to be conducted near feature by some other contemporary workers of the native country on the species from the same eco-geographical regions.

Different samples of same species were collected from different altitude and climate and studied both morphologically and anatomically to see altitudinal as well as ecological variations.

Site selection of this study as North-eastern India, particularly the Darjeeling and Sikkim Himalayas is simply due to availability of the specimens as more than 75% of the species of the genus are confined in this regions, some of which are in very critical condition and often referred as endemic. So proper identification of these critically occurred species is very much needed. In this regard it may be mentioned that the enumeration of species by Noltie (1994), Karthikeyan et al. (1989), Hazra and Verma (1996), and Verma and Rao (1984) were consulted during this study and there by referred as basic information after the work of Clarke (1893-1894) in Flora of British India done more than one century ago. Though these works were conducted totally on the basis of studies conducted on limited deposition of different herbarium specimens in India. So the in-situ population study, geographical distribution, their present status of occurrence, reproductive biology and lastly the anatomical aspects of in-situ population of the genus itself will provide the true systematics of this genus Carex.

Classification of the genus Carex L. and interrelationships within the other related genera of the sedge family was done by different authors. Kukenthal (1909) divided the genus into 4 subgenera as Primocarex, Vignaea, Indocarex and Eucarex. Mackenzie (1931, 1936) had segregated the genus into 71 sections and placed the genus Carex L. in the tribe Cariceae of the family Cyperaceae along with Kobresia, Uncinia and Cymophyllus. The systematics of the genus subsequently done by Bruhl (1995) based on cladistics study. On the basis of reproductive morphology Reznicek (1990), Goetghbeur (1998), Egorova (1999) had classified Carex L. into 5 subgenera. Later Starr et al. (1999), Yen and Olmstead (2000), Starr (2004), Roalson et al. (2000), Muasya et al. (2000) also done systematic study of the genus Carex on the basis of molecular characters. Thus various interpretations have been published regarding the systematic and phylogenetic interrelationships of different taxa of the genus Carex. However, still now the systematics of the genus Carex is considered as unsatisfactory.
Moreover, till now there is no information in the classification of the genus *Carex* on the basis of anatomical parameters. The sedge taxonomy is a very challenging view for the uses of anatomical features rather than the other floral biological features. Thus this anatomical work may provide little ray of hope for the systematics of *Carex* to the Cariologists. Furthermore, South-east Asia is considered to be one of the centres of biodiversity of *Carex* mainly in China having 524 species and then in Japan and India as compared to the other countries of Asia, Africa, Europe and Australia. Thus it is the opportunity to get and to study more representative species of the genus and their relationships.

As many reports have been stated about mycorrhizal association of Cyperaceae and being mycorrhizal. Hence to confirm if there is really any mycorrhizal colonization present in Cyperaceae or not. To confirm the presence of VAM in Cyperaceae, in sedges this study was also conducted for this genus *Carex*. Since sedges are mostly found in disturbed habitats whereby survival would be much dependent on external factors hence to examine if sedges are less or not dependent on VAM association. This study was concluded mainly concentrating on *Carex* regarding dependency upon VAM. There have been a lot of reports on VAM colonization in the sedges, whereas from this region there were no reports on VAM association in the sedges. Hence representing Cyperaceae, which is one of the pioneers in sedges this genus *Carex*, it is the first report on VAM association from this region of Eastern Himalayas.