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Distribution, Adaptation and Cytotype Diversity of Cynodon Dactylon (L.) Pers. In Different Bio-Ecological Zones of Bangladesh

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DISTRIBUTION, ADAPTATION AND CYTOTYPE DIVERSITY OF *CYNODON DACTYLON* (L.) PERS. IN DIFFERENT BIO-ECOLOGICAL ZONES OF BANGLADESH

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BY

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Dedicated

To

My Parents

DECLARATION

I hereby declare that the research work embodied in this thesis entitled **"DISTRIBUTION, ADAPTATION AND CYTOTYPE DIVERSITY OF *CYNODON DACTYLON* (L.) PERS. IN DIFFERENT BIO-ECOLOGICAL ZONES OF BANGLADESH"** has been carried out by me for the degree of Doctor of Philosophy under the guidance of **Professor Dr. S. M. Shahinul Islam** (Principal Supervisor), Institute of Biological Sciences, University of Rajshahi and **Professor Dr. Md. Hasan Tarique** (Co-supervisor), Department of Agronomy and Agricultural Extension, University of Rajshahi, Bangladesh.

I also declare that the result presented in this dissertation is my own investigation and any part of this thesis work has not been submitted to elsewhere for any degree/diploma or for similar purpose.

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CERTIFICATE

This is to certify that **Saika Kabir Nitu** worked under our supervision as a Ph.D Fellow, Roll No: P-1612184524, Session: 2015-2016, Institute of Biological Sciences, University of Rajshahi, Bangladesh. It is our great pleasure to forward her thesis entitled **"DISTRIBUTION, ADAPTATION AND CYTOTYPE DIVERSITY OF *CYNODON DACTYLON* (L.) PERS. IN DIFFERENT BIO-ECOLOGICAL ZONES OF BANGLADESH"** which is a bonafide record of research in the Institute of Biological Sciences, University of Rajshahi, Bangladesh.

This work is original and has not been submitted so far in part or in full, for the award of any degree or diploma by any other institute in home or abroad. **Saika Kabir Nitu** has fulfilled all the requirements for submission of the thesis for the award of the degree of **Doctor of Philosophy**.

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The Authoress

CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

The following abbreviations have been used through the text:

Acc.	:	Accessions
AFLP	:	Amplified fragment length polymorphisms
ANOVA	:	Analysis of variance
APX	:	Ascorbate peroxidase
A-T	:	Adenine- thymine base pair
B	:	Boron
C_4	:	C_4 plants are that use the C_4 pathway or Hatch-slack pathway during the dark reaction
Ca^{2+}	:	Calcium ion
CAT	:	Catalase
Cl^-	:	Chloride ion
cm	:	Centimeter
CMA^+	:	Chromomycin A3
CRD	:	Complete randomized design
Cu	:	Copper
DAPI	:	4, 6-diamidino-2-phenylindole
dm	:	Decimeter
DMRT	:	Duncan’s multiple range test
DNA	:	Deoxyribonucleic acid
et al.	:	Et alia (and others)
etc.	:	Et cetera is the latin expression that means “and other similar things”

F-value	:	The F-value is a value on the F distribution which is used in analysis of variance
F_1	:	F_1 hybrid is the first filial generation of offspring of distinctly different parental types
FAA	:	Formalin acetic acid alcohol
Fe	:	Iron
Fig.	:	Figure
g	:	Gram
hrs	:	Hours
i.e.	:	The Latin phrase id est, which means "that is"
ILRD	:	International livestock Research Institute
K^+	:	Potassium ion
km	:	Kilometer
m	:	Meter
Med.	:	Medium
Mg^{2+}	:	Magnesium ion
mm	:	Milimeter
mm	:	Milimeter
mm^2	:	Millimeter square
Mn	:	Manganise
n	:	Haploid number of chromosome
Na^+	:	Sodium ion
NV	:	Nuclear volume
OM	:	Ocular micrometer

OM	:	Organic matter
Opt.	:	Optimum
P	:	Phosphorus
p	:	Probability value
Pbf	:	Present band frequency of an individual with known ploidy level
pg	:	Picogram
pH	:	Potentiality of hydrogen ion
POD	:	Peroxidase
r	:	Correlation coefficients is a measure of the strength of the straight line or linear relationship between two variables
RNA	:	Ribonucleic acid
S	:	Sulfur
SM	:	Stage micrometer
SO	:	Sulfur monoxide
SO_2	:	Sulfur dioxide
SOD	:	Superoxide dismutase
SP.	:	A single species
SPSS	:	Statistical package for the social sciences
TS	:	Transverse section
TCA	:	Trichloroacetic acid
USA	:	United states
var.	:	Variety
VH	:	Very high
viz.	:	Videlicet (namely)

VL	:	Very low
Zn	:	Zinc
μl	:	Micro liter
μm	:	Micro meter
%	:	Percent sign
°C	:	Degree celsius
°F	:	Degree fahrenheit
μ	:	Micron
2n	:	The total number of chromosomes in diploid cells
4x	:	Tetraploid

ABSTRACT

The objectives of the present investigation on *Cynodon dactylon* were to examine the role of ecological factors on intraspecific variation based on their distribution, to make an analysis of morphological variations of the ecotypes from botanic as well as agronomic points of view, and to determine the physico-chemical properties of the soil of original habitat and experimentation field, to focus the qualitative and quantitative leaf epidermal characteristics with special reference to stomatal features, to study anatomy of root and stem which includes distribution and arrangement of different tissue and tissue system qualitative and quantitatively, and to determine the nuclear phenotype along with the estimation of ploidy level.

To build up the above mentioned objectives extensive amount of information on *C. dactylon* in relation to its life cycle, phenology, deletorious characteristics, ecotypes, genetics, environmental requirements and also the viewpoints pertaining to habitat, dissemination, soil tolerance, anatomical change, cytotypic variations etc. were taken under consideration reviewing a good number of literature.

Among 64 districts of Bangladesh, 23 districts plus one location of Saint Martin's Island belonging to different bio-ecological zones of Bangladesh were selected as the study area. The accessions as plant material named Bermuda grass (*C. dactylon*) and their habitat soil were collected for conducting the present investigation. A total 24 accessions of this grass species were transplanted in the experimentation field of the Institute of Biological Sciences, University of Rajshahi, Bangladesh. To exonerate the objectives of the present investigation different experiments were conducted separately following the update methods. The findings obtained from different experiments were analyzed statistically except that about distribution.

At present Bangladesh comprises 64 districts and the grass species *C. dactylon* is found with an outstanding spreading ability all over the country. In these districts, 24 locations were considered as the study sites. A keen study was made on the intensity of growth, growth duration and tolerance to drought/salinity stress. A matrix of the studied accessions was tabulated regarding the above mentioned characters. All these accessions were collected mainly from roadside/in the vicinity of lawn or homestead. They were found to grow in crop lands/thoroughfares/sidewalks/in vacant lots/on summer dry soils of foothills.

The accessions showed variations in many of the morphological characters, which are of very much taxonomic importance. The statistical analysis revealed highly significant variation among the characters of shoot, root, flower, seed, leaf, plant height and stolon. In this chapter physico-chemical properties of the soil from the original habitat along with the experimentation field were estimated. The soil test values based on critical limits, magnitude of physico-chemical properties of the habitat as well as experimentation field were interpreted as low, very low, very high, medium, optimum etc.

Findings on leaf epidermal anatomical characteristics in relation to ecotypic adaptation of *C. dactylon* were illustrated both qualitative and quantitatively. Qualitative study was conducted with stomata type, shape of subsidiary cells, long cell margins, types of silica bodies, presence of macro and micro-hairs, prickle angular and hook. On the other hand, number, length and width of long cell, frequency and index of stomata, number, length and breadth of epidermal cell, stomata length and breadth with guard cell, number of silica bodies, prickles angular, hooks, macro-hair etc. in both abaxial and adaxial surface of leaves were measured and expressed in different units. All the characters were analyzed using DMRT and analysis of variance, and except stomatal index all of them were found to show significant variations at 0.01 and 0.001 probability levels.

Anatomical features of stem and root of 24 accessions of *C. dactylon* revealed epidermis, hypodermis, the ground tissue system along with vascular bundles of stem, and epidermis, cortex, endodermis, pericycle, vascular bundle, conjunctive tissues and pith very clearly. All these characters were evaluated quantitatively. The recorded values were tested using DMRT and further analysis of variance was applied for findings the significant differences among the accessions. Except sclerenchyma thickness in case of root all the sources were found to show statistical difference.

All the accessions of *C. dactylon* exhibited chromocentric nuclear organization and chromocenter numbers were found to range from 11.4 to 17.6, which were not found to be corresponding with their somatic chromosome number. Heterochromatin percentage were expressed per nuclear area and the values were very much variable. Nuclear as well as interphase chromosome volume of all the accessions were determined. In this case also no such relation was observed between chromosome number and interphase chromosome volume. Among all the accessions chromosome number ranged from 12 to 40 and only nine were found to be diploid, one tetraploid and rest of them were aneuploid. In this study no such effect was found apparently when stomatal frequency and stomatal index were predicted both in case of euploid and aneuploid. However, finally it has been tried to discourse the overall findings with the help scholarly research works.

Chapter I

Introduction

1. INTRODUCTION

Cynodon dactylon (L.) Pers. is a typical warm season turfgrass belongs to the family Poaceae and this grass species is widely adapted to various environments of tropical and sub-tropical regions around the world (Zheng *et al*. 2017). It spreads by scaly rhizomes and flat stolons. This plant species is a C_4 grass species, which has been included in the Global Compendium of Weeds (Randall 2012). It has also been listed as one of the most serious agricultural and environmental weeds in the world (Holm *et al.* 1977). This grass species has different vernacular names. In Bengali, it is known as Durba, Dub, Dubla, Doorva, Neel Doorva (Asthana *et al.* 2012). In English it is termed as Couch grass, Bahama grass, Bermuda grass, Dun grass, Doab grass etc. mentioned common name for all the East African rhizomatous species of *Cynodon* as Bermuda grass Harlan (1970) and Burton and Hanna (1985). However, Harlan and de Wet (1969) mentioned that *Cynodon dactylon* has become a ubiquitous cosmopolitan weed.

Cynodon dactylon can tolerate different types of soil and its growth becomes healthy in case of heavy clay soil compared to that of light sandy soil in dry regions, which might be due to water holding capacity of clay (Burton and Hanna 1985). However, alkaline soils are tolerated nicely by this grass species than acidic ones. Drought conditions, high temperature, intensive sunlight and alkaline soils are found to be best requirements for healthy growth of this grass. *Cynodon dactylon* shows direct competitive behaviour for space and nutrients by its rapid growing ability and adaptive capacity. *Cynodon dactylon* is found together with underground rhizomes and ground runners (Cabrera 1968, Covas and Salavi 1972). The rhizomes penetrate to the soil at a depth of above 30 cm (Perez and Labrada 1985, Philips and Moaisi 1993). The rhizomes may be extensively wide than the runners and this is found to be variable characters in their populations. Runner or rhizome nodes generally bear viable buds. Leaves show alternate distal pattern of distribution along the runners. Leaf blades are unhaired but the ligule is found with a conspicuous fringe of white hairs. Leaf blades are green to dull green, lanceolate, finely parallel-ribbed on both the surfaces, without midrib (Rosengurt *et al.* 1960). The inflorescence is supported on a culm and consisted of a single whorl of 3-7 racemes. Spikelets are closely appressed to the rachis. Glumes are one nerved, lemma is silky pubescent on the keel and palea is glabrous. Caryopses are sub-eliptical, compressed and brownish in colour (Kissmann 1991).

Cynodon dactylon shows some sort of social impact, as its pollen has been found to cause allergic symptoms in asthmatics in Malaysia (Sam-Choonkook *et al.* 1998) and Brazil (Kissmann 1991). The use of *Cynodon dactylon* in different religious ceremonies has been studied by Dubey *et al.* (2000). This grass species is used as medicinal plant. Its antioxidant has been reported extensively by Auddy *et al.* (2003) for treatment of neurodegenerative diseases. This plant species is used as pasture grass in many countries, since it contains many soils of chemical components like proteins, carbohydrates, mineral constituents, phosphorous, calcium, potassium, vitamin C, carotene, fats, palmitic acid etc. Paranjpe (2001) reported a good amount of crude protein, fiber and total ash. *Cynodon* species have been categorized as noxious weeds, especially *C. dactylon*, which is very invasive (Fernaudez 2003). This species is suitable for erosion control and soil stabilization. It is also observed that mechanical tillage of weed-infested fields makes an effect on fragmentation and dispersal of stolon and rhizome propagules, and thereafter infestation is increased.

However, Van de Wouw *et al.* (2009) stated that the International livestock Research Institute (ILRD) collected some commercial cultivars and accessions of *Cynodon* species from wild habitats. These collections covered four different species of *Cynodon viz*. *C. aethiopicus*, *C. dactylon*, *C. incompletus* and *C. nlemfuensis* along with hybrids of *C. dactylon* and *C. transvaalensis*. The concerned investigators determined the amount of morphological variation present in the collections, based on what the users can select the appropriate accessions for their further experiment. The present study is supposed to make an analysis of morphological variation of *C. dactylon* ecotypes from botanic as well as agronomic points of view, and the determination of physico-chemical properties of their habitat soils. According to Stace (1984) and Jones (1986) leaf epidermis is generally considered as an important aspect for the classification and delimitation of species and genera, and for sorting out the evolutionary and phylogenetic problems. Ahmed *et al.* (2011) stated that the leaf epidermal characters have significance in grass systematics and classification of ambiguous groups which are not properly adjusted within grasses, particularly at subfamily and tribe level. Therefore, it is imperative to make any attempt to study the epidermal characters of taxonomic importance. It is evidenced now that leaf epidermal features can help to elucidate many ecological parameters. In Poaceae leaf epidermal anatomy shows variations with a higher degree of specialization than that of any other family and provides extensive features of taxonomic importance. Leaf epidermal anatomical features like stomata and trichomes along with some other qualitative and quantitative characters are very much useful in perspective of morphological, ecological, physiological and taxonomical studies. The morphology and ontogenies of taxa are considered to be more important in case of intra-generic and intra-species systematics due to diversity of stomata types. On the contrary, the most frequent stomata type is considered as taxonomic character.

Plant anatomical traits are good index of habitat quality, since they display variability in relation to microclimatic conditions (Barber *et al.* 2004). Stomatal traits like stomatal density, stomatal apparatus and guard cell architecture respond to environmental and physiological cues (Nadeu and Sack 2002, Gitz and Baker 2009). The present study aims to identify the leaf epidermal features which may focus on stomatal characters because of their importance in many ecological aspects of grass species. *Cynodon dactylon* has paracytic type of stomata (Abid *et al.* 2007) and they are arranged in parallel rows with silica bodies on the epidermal surface. Paracytic type is considered to be primitive and their arrangement reflects the developmental process. In *Cynodon dactylon* stomata may be present on both side (adaxial and abaxial) of the expanded leaf, but in many grass species they are exclusively abaxial. Leaf epidermal traits like epidermal cells, stomata and micro-hairs have been proved valuable in identification and differentiation of different taxa (Stenglein *et al.* 2003). Macro-hairs are also found to vary in size, shape and wall thickness, and these are of great importance in grass systematic. The shape of silica bodies varies among different grass species from round or oblong to linear, crescent or dumbbell shaped, nodular, sinuous and shaddle or cross shaped (Chaudhary *et al.* 2001, Ahmad *et al.* 2012, Chaudhari *et al.* 2014).

Stomatal size, number and shape are of great ecological significance (Jian *et al.* 2012) and extremely important in stress tolerance (Xu and Zhou 2008, Zheng *et al.* 2013). Stomatal size and shape regulate water use efficiency. In small stomata less turgour is required for their opening and closing (Tufail *et al.* 2017). Stomatal frequency improves the photosynthetic efficiency of plant species. The present study is focused on the qualitative and quantitative leaf epidermal characteristics with special reference to stomatal features to find out the impact of environmental conditions on *Cynodon dactylon*.

Many grass species succeed from their tolerance of grazing herbivores and some other disturbances, but they survive by different mode of reproduction and by their functional photosynthesis. In case of flowering plants, where new growth starts in the aerial plant body, occurs at the shoot tips only. Anyhow, if the tip is removed, buds in the axils of lower leaves may start growing, but at the same time the original shoot stops growing. Meristems of grasses die at the base of each and every stem between the leaves and thus, regrowth may be possible even after removal of the tip by any means. *Cynodon dactylon* is a creeping grass and its exhibits tough and rough texture. Its root grows, wherever, a node touches the ground and forms a thick mat. The stems are slightly flattened and surface is hard and longitudinally furrowed. Asthana *et al.* (2012) states that it has a sweet gelatinous taste and stems of this grass range from erect to prostrate.

Cynodon dactylon is distributed abundantly in tropical and temperate areas (Chaudhary 1989). Natural population of this grass species may have considerable genetic variation for tolerance to soil temperatures, salinity and drought (Speranza 1995). It can tolerate relatively high salinities (Mass and Hoffman 1977). Morphological and anatomical modifications in plant body acts for minimizing detrimental effects of salt stress (Poljakoff-Mayber 1988, Grigore and Toma 2017). Salt tolerant species shows a range of anatomical adaptive features due to increased succulence (both in root and stem), thick cuticle and deposition of wax, salt secretory trichomes, thick and many layered epidermis and well developed water storing tissues in the cortex, widening of casparian bands and enhanced development of root endodermis (Akram *et al.* 2002, Wahid 2003). The effect of salinity on anatomical structures were discussed by Gadalla (2009) and Younis *et al.*

(2014b). From these points of view, the present study deals the measurement of the ecotypic variability and degree of adaptation in *Cynodon dactylon* collected from different ecological habitats. Stem anatomy of *C. dactylon* as seen through transverse section includes description, distribution and arrangement of different tissue and tissue system alongwith qualitative characterization and quantitative measurement of different anatomical traits.

Cytological feature like interphase nuclear phenotype and chromosomal characterization are very useful parameters in distinguishing the cytotypes, accessions (Acc.) and even germplasm of a plant species (Huang *et al.* 2014). The information on chromosome number of *C. dactylon* is somewhat limited to the determination of ploidy level. On the basis of previous research work, this grass species has the basic chromosome number x = 9 (Dhaliwal and Gupta 2011, Zhi-Yun *et al.* 2013). However, this species has been reported with different ploidy level such as triploid (2n = 3x = 27), tetraploid (2n = 4x = 36), few pentaploid (2n = 5x = 45) and hexaploid (2n = 6x = 54) according to Harlan *et al.* (1970) and Wu and Taliaferro (2009). In contrast, Hunter (1934), Hurcombe (1947), Moffett and Hurcombe (1948) and Rochecouste (1962) recorded different chromosome number such as 2n = 40 for this species with probable basic number of x = 10. However, information on aneuploidy of this species is insufficient and it seems like that real differences in chromosome number along with variation might be related to ecological condition. Parvin (2002) reported aneuploidy in *C. dactylon*. Thus, the goal of this study was to discriminate the different accessions of *C. dactylon* collected from different ecological habitats of Bangladesh.

Structural organizations in plant cell nuclei are two types and they are namely chromocentric and reticulate (Lafontaine 1974). Interphase nuclear phenotype of plants is species specific and it indicates several important features regarding chromosomal arrangement (Patankar and Raujekar 1984). Its reasons are unknown although the role of nuclear DNA content and repetitive DNA sequence has been suggested to be associated with those features (Lafontaine 1974, Nagl and Fusenig 1979). They also suggested that chromocentric nuclear organization is assumed to be governed by small size of chromosomes and low DNA content. As chromocenters correspond to heterochromatin (Nagl and Fusenig 1979), percentage of heterochromatin values may be obtained by determining the area of nucleus and of chromocenters by planimetry (Kabir and Singh 1989). Keeping these points in mind interphase nuclear phenotype along with chromosome counts of *C. dactylon* accessions have been determined in this study.

However, the objectives of the present study on *Cynodon dactylon* growing in different bio-ecological zones of Bangladesh were to:

i. Examine the role of ecological factors on intraspecific variation based on its distribution,

ii. Make an analysis of morphological variations of the ecotypes from botanic as well as agronomic points of view, and to determine the physico-chemical properties of the soil of original habitat and experimentation field,

iii. Focus the qualitative and quantitative leaf epidermal characteristics with special reference to stomatal features,

iv. Study anatomy of root and stem which includes distribution and arrangement of different tissue and tissue system qualitative and quantitatively, and

v. Determine the nuclear phenotype along with the estimation of ploidy level.

Chapter II
Review of Literature

2. REVIEW OF LITERATURE

Cynodon dactylon (L.) Pers. is an invasive and competitive grass species. The large intra-specific variability in this grass species is represented by several varieties, which have been spread to other countries from East Africa and most notably *Cynodon dactylon* var. *dactylon* is referred. This grass species grows well throughout the warmer regions of both hemisphere and needs warm temperature to thrive long periods of freezing weather or short duration of extremely low temperature are detritmental to the plants. Besides, it tolerates a wide range of soil types and conditions. Growth is found to be better on heavy clay soils than on light sandy soils in dry regions and this may be due to greater water holding capacity of the clay. The drought and alkali tolerance along with sunlight requirement of this grass makes its growth successfully. Its residues and growing parts may create a direct threat to the growth of neighbouring plants particularly herbs. Various extensive number of information on *Cynodon dactylon* is available. Its life cycle, phenology, deletorious characteristics, ecotypes, genetics, environmental requirements etc. are almost well represented. But, information pertaining to habitat, dissemination, soil tolerance, anatomical change, cytotypic variations etc. are not so well documented particularly in the last 20-25 years. Thus, the literature, which of them are very much imported even in the 18^{th} and 19^{th} century also and which are available upto 2000, and thereafter a good number of effective recent articles (upto 2019) are reviewed here under three subheads such as distribution, adaptation and cytotype diversity.

2.1 Distribution

The genus *Cynodon* consists systematically a distinct group in the tribe Chlorideae. The cosmopolitan species *C. dactylon* (L.) Pers. has received a lot of attention by researchers because of its economic value as a turf grass and for grazing and hay production, and because of its abundance and wide spread distribution as a weed (Harlan *et al.* 1970). The genus as a whole, however, has been rather neglected on the contrary. Since a new revision of the genus has now been published (Clayton and Harlan 1970, Harlan and de Wet 1969, Harlan *et al.* 1969), it would appear oportune to record what is currently known of the geographic distribution of the several species (Harlan *et al.* 1970). They further stated that older classifications failed to recognize certain taxa in East Africa and resulted in considerable confusion. It is likely that much more information on distribution and ecological behaviour will emerge as these discrete entities are better known.

Cynodon dactylon, commonly known as Bermuda grass was probably originated in tropical Africa; however, Australia, Eurasia, the Indo-Malaysian area, and the Bengal region of India / Bangladesh have also been proposed as its home (Holm *et al.* 1979). Although many early workers believed that this grass species started in India and most improved strains of *Cynodon* were developed from African stock (Kingsbury 1964). *C. dactylon* appears to be native in Australia, though some evidence suggests that it might have been introduced (Everist 1979). This grass species was probably introduced into the United States during the middle 1700s and part of reportedly it was first imported in 1751 into Savannah, Georgia by Governor Henry Ellis (Callahan and Engel 1965). Its rapid propagation and wide distribution in the Southern United States were noted in 1807 by James Mease in his Geological Account of the United States (Mease 1807). Frederick Pursh, who collected from Maine to the Carolinas during the first ten years of the 19^{th} century, also this grass as frequently growing on roadsides and in the cultivated ground (Pursh 1814). In 1880, Sereno Watson reported Bermudagrass near San Bernardino and San Josp and mentioned that it was a troublesome weed in cotton fields. He also stated that this grass has never been known in the Eastern States to perfect its seed, and it can only propagate by cuttings, an assertion which has long been disproved. By 1915, this grass species was established sufficiently in California to rank as a top weed in Imperial, Napa (Newman 1915), and Kern Countries (Knowlton 1915). The oldest specimens in California herbarium include five specimens.

The genus *Cynodon* includes a total of eight tropical and warm climatic species (Mabberley 1987) and several varieties, mostly in Africa and Australia; *C. dactylon* is widely distributed throughout the world. Mabberley (1987) also stated that the genus includes several pasture and lawn grasses, mainly strains of the species *dactylon*. Members of the grass are perennial with creeping stolons or rhizomes, short blades, and several slender spikes digitate at the summit of the upright culms (Mitich 1989). *C. dactylon* is the type species of the genus (Hitchcock 1950). Species are difficult to define and distinguish, and there is a great variability within species, particularly in *C. dactylon* become complex (Hansen 1918), and arises much confusion of nomenclature in the literature (Everist 1979). African ugandagrass, *C. transvaalensis* have become naturalized in the United States (Mabberley 1987). This fine leaved ugandagrass species is often crossed with *C. dactylon* to produce turf hybrids (Madison 1971). *C. bradleyi* is another African species Watt and Breyer-Branwijk (1962). *C. hirsutus* is a common weed in Argentina, and *C. plactostachyum* is also a common weed in Australia and Kenya (Holm *et al.* 1977). *C. nlemfuensis* and *C. incompletes* have been reported as native to both Australia and Africa (Everist 1979, Harlan *et al.* 1969).

C. dactylon grows throughout the tropical and sub-tropical areas of the world, from latitude 45°N to 45°S and in eastern Africa it is distributed from sea level to 2,200 m (Holm *et al.* 1979). In USA, *C. dactylon* is found in open ground, grassland, fields and waste places, from Maryland to Oklahoma, south to Florida and Texas, and west to California and it occasionally grows in north of this region- from Massachusetts to Michigan, and Oregon (Hitchcock 1950). It thrives in warm or hot weather and usually does not survive heavy freezes, although it had leaved through temperature of 10°F in the vicinity of the district of Columbia (Hoover *et al.* 1948). This grass species grows on way moderately in well drained soil, either acid or alkaline, provided moisture and plant fruit nutrients are adequate (Philips Petroleum Company 1958). This grass is drought resistant and tolerant of alkali. It was reported that patches of *C. dactylon* near California, although submerged in the salt on sea for over two years, were still alive and making new growth from the stems when that body of water finally evaporated to a lower level (Robbins *et al.* 1970). *C. dactylon* creeps extensively by means of scaly rhizomes or by strong flat stolons (Hitchcock 1950). It can produce vegetatively, from both rhizomes and stolons, and by producing seeds. The extensive network of rhizomes and stolons makes this grass particularly difficult to eradicate (Crampton 1974). It is introduced into new areas in many ways; the seeds are a common impurity in commercial seeds; plants bearing seeds are carried in hay, in packing, in bedding for livestock in feed stuffs; the seeds are carried by wind and irrigation water (Robbins *et al.* 1970).

Worldwide, *C. dactylon* is the most serious weed of the grass family and it is reported as a problem in 40 crops in over 80 countries (Holm *et al.* 1979). It is a problem in essentially every cropping area except northern Europe, and appears in rotation crops, perennial crops, grassland, and waste places (Hafliger and Scholz 1981). It is not only common in crops, but along throughfares, and alongside walks or in vacant lots in cities. *Cynodon* is a genus in the grass family and native in warm temperate to tropical regions of the world as wellbeing cultivated and naturalized in new world and on many oceanic islands. Probably native to East Africa where it is widely distributed from sea level to 2,160 m altitude. In temperate zones, it grows along sea coasts; in tropics, most commonly in areas with 670-1750 mm rainfall and in arid zones, along rivers and on irrigated land. In India it can be found up to 2600 m altitude. Ranging from Cool Temperature Steppe to Wet through Tropical Desert to Wet Forest Life Zones, *C. dactylon* is reported to tolerate annual precipitation of 0.9 to 4.9 dm (mean of 84 cases = 12.8), annual temperature of 5.9 to 27.8°C (mean of 84 cases = 19.5) and pH of 4.3 to 8.4 (mean of 74 cases = 6.4) as reported by Duke (1978).

Bermudagrass is thought to have originated around the Indian Ocean Basin, from East Africa to India. It was introduced to all tropical and subtropical areas. It is found as far as 50°N in Europe and down to 37°S in the southern hemisphere. This grass species can be found at high altitudes up to 2600 m in the tropics and 4000 m in the Himalayas (Ecoport 2012, FAO 2012). Bermudagrass is common in grasslands, lawns and pastures (FAO 2012). It is dominant in uncultivated areas: roadsides, sea-coast sandy-dumes, or along rivers and irrigated land (Ecoport 2012). It does well on overgrazed and tranpled areas (FAO 2012, Ecoport 2012, Cook *et al.* 2005). It grows in areas where average rainfall temperatures range within 6-28°C, though it does better where daily temperatures are in the range of 17-35°C. Under 15°C it stops growing and foliage is killed at -2 to -3°C but the stand recovers from the rhizomes. The grass species requires 625 - 1750 mm annual rainfall, but moisture level as low as 550 mm and as high as 4300 mm are acceptable. Cook *et al.* (2005) stated that because of its deep rhizome, bermudagrass is tolerant to both dry and flooding conditions. Though this grass species prefers deep and well drained fertile soils, it can adapt to a wide range of soils including those that are relatively fertile with a pH ranging from 4.3 to 8.4 (optimum >5.5). It responds positively to N and K (Coblentz *et al.* 2004). It has some salt tolerance; hence its ability to be grown on coastal areas or on irrigated land (Cook *et al.* 2005, Ecoport 2012, FAO 2012). According to Cook *et al.* (2005) and Hanna 1992) this grass species is sensitive to shade and even to many pests and diseases.

Blair *et al.* (2014) said about the global distribution of grasslands, which is extensive, with wide spread representation of grasslands on every continent except Antarctica. Although grasslands are currently absent from Antarctica, a grass species (Antarctic hair grass, *Deschampsia antarctica*) does occur on the Antarctic peninsula and surrounding islands, where recent warming is thought to be promoting the spread of this native grass. Major grasslands in temperate regions of the world include the steppes of Eurasia, the velds of southern Africa, the pampas of Argentina, and the prairies of North America (Archibold 1995). However, grassland environments are the basis for major agricultural areas word wide. The conversion of grasslands to agriculture continues now-a-days (especially with increased demand of biofuels) as mentioned by Fargione *et al.* (2008), some of the most significant losses of grasslands now are related to changing land management coupled with other global phenomena. While the loss of native grasslands due to agricultural conversion and urbanization is ongoing in many locations around the word, another major threat is the dramatic increase in shrubs and trees now occurring in many types of grassland (Briggs *et al.* 2005).

In a study of differential response of the edaphic ecotypes in *Cynodon dactylon* to soil calcium Ramakrishnan and Singh (1966) found a close relation which was discernible for three populations with the habitats they occupy in nature. Three populations of *Cynodon dactylon* occupied very diverse habitats with markedly different levels of exchangeable calcium. These populations showed a varying capacity to accumulate different minerals in the leaf tissue and these differences are maintained even when the three populations are cultivated on neutral substratum. Snaydon and Bradshaw (1961) suggest that the induction of heavy metal toxicity at the lowest calcium levels may be one of the factors for poor development of roots. However, the results obtained in their study indicate clearly that the differences between the three populations are as marked as those that they may be found for different species with completely different edaphic tolerance ranges.

Paul *et al.* (2012) stated that *Cynodon dactylon* is found to grow in open areas where there are frequent disturbances of grazing animals, flood, fire amongst other calamites. This species is growing out most other grasses and invading other habitats and has become hard to eradicate. Nasiri *et al.* (2012) also stated that *Cynodon dactylon* is widely distributed in diverse habitats of Iran. It is a perennial herb and its function in soil stabilization and conservation are noticeable (Roudsari and Pishdar 2007), protecting the soil erosion and also its nature as weed are remarkable (Mahanta *et al.* 2007, Oad *et al.* 2007). It is widely used as warm season turf and forage species in temperate and tropical regions (Li and Qu 2004). The populations of this grass species harbour many useful physiological traits such as tolerance to high temperature and drought (Bethel 2005).

Juska and Hanson (1964) stated that adaptation of *Cynodon dactylon* to the northern limits of its distributions is largely dependent on cold hardiness. Information has not been readily available, however, on the relative cold hardiness of many named varieties and selections. In order to meet this need, comparative tests were conducted to evaluate bermudagrasses for winter survival and turf quality. Varieties, introductions and selections were compared under all combinations of two mowing heights and two levels of nitrogen. Data submitted by several agricultural experiment stations are included for comparative analysis. They further stated that winter hardiness is major concern in determining the adaptation of bermudagrass in the transition zone. Recovery rate after growth starts in the spring is an excellent measure of winter hardiness, as major recovery is inversely proportional to winter damage.

Zhou *et al.* (2010) made a screening for drought resistance among a large number of Australian *Cynodon* ecotypes during canopy establishment. Australia wide exposure to water deficit is the most important problem adversely affecting growth and quality of *Cynodon* turfgrasses. Green cover defixed as the percentage of green leaves in a turf plot and leaf relative water content during the drought period have been used as criteria to select drought resistant genotypes (Huang *et al.* 1997, Richardson *et al.* 2008). Morphological traits such as internode length and branching habit may influence evapo-transpiration through their effects on leaf density and leaf area (Ebdon and Petrovic 1998). However, the finding of Zhou *et al.* (2010) suggests that there is potential to find *Cynodon* ecotypes with drought resistance for every state and territory of Australia.

It is said that *Cynodon dactylon* has originated in Africa but now occurs worldwide in both tropical and subtropical regions including Asia, North, Central and South America, the Caribbean Islands and the Islands in the Pacific Ocean. It also spreads into temperate areas of Europe and North America but it is limited by sensitively to prolonged frost. *C. dactylon* invades almost all kinds of crops and modified ecosystems, including urban areas and circulation paths (road and railroad tracks) in many regions. The distribution of *Cynodon dactylon* almost all over the world are shown in Table 1, following the summary table available as Invasive Species Compendium (Last modification 20 November, 2019) in accordance with website https://www.cabi.org/isc/datasheet/17463# to pictures, omitting two columns regarding last and first report since there was no information.

Table 1: Distribution of *Cynodon dactylon* almost all over the world (https://www.cabi.org/isc/datasheet/17463# to pictures).

Continent/Country/ Region	Distribution	Origin	Invasive/ Naturalized	Reference	Notes
Africa					
Algeria (datasheet/108415)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Angola (datasheet/108357)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Benin (datasheet/108375)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Botswana (datasheet/108385)	Present, Widespread	Native	-	CABI(Undated) (datasheet/17463#REF-DDB-27)	Original citation: Gibbs Russell *et al.* (1990)
Burkina Faso (datasheet/108371)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Burundi (datasheet/108374)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Cabo Verde(datasheet/108406)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Cameroon(datasheet/108397)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Central African Republic(datasheet/108391)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Chad(datasheet/108576)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Congo, Democratic republic of the(datasheet/108615)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Congo, Republic of the(datasheet/108392)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-

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Cote d'lvoire(datasheet/108394)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Djibouti(datasheet/108411)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Egypt(datasheet/108418)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Equatorial Guinea(datasheet/108442)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Eritrea(datasheet/108420)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Eswatini(datasheet/108573)	Present, Widespread	Native	-	CABI (Undated) (datasheet/17463#REF-DDB-27)	Original citation: Gibbs Russell *et al.* (1990)
Ethiopia(datasheet/108422)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Gabon(datasheet/108430)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Gambia(datasheet/108439)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Ghana(datasheet/108436)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Guinea(datasheet/108440)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Kenya(datasheet/108470)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Lesotho(datasheet/108488)	Present	Native	-	CABI(Undated) (datasheet/17463#REF-DDB-27)	Original citation: Gibbs Russell *et al.* (1990)

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Liberia (datasheet/108487)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Libya (datasheet/108492)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Madagascar (datasheet/108498)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Malawi (datasheet/108512)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Mali (datasheet/108502)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Mauritius (datasheet/108510)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Rodrigues (datasheet/108547)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Morocco (datasheet/108493)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Mozambique (datasheet/108515)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Namibia (datasheet/108516)	Present	Native	-	CABI (Undated) (datasheet/17463#REF-DDB-27)	Original citation: Gibbs Russell *et al.* (1990)
Niger (datasheet/108518)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Nigeria (datasheet/108520)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Reunion (datasheet/108546)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-

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Rwanda (datasheet/108551)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Saint Helena (datasheet/108558)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Senegal (datasheet/108564)	Present	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Seychelles (datasheet/108554)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
-Aldabra Islands (datasheet/108364)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Sierra Leone (datasheet/108562)	Present	Native	-	CABI (Undated) (datasheet/17463#REF-DDB-27)	Original citation: Hopper (1972)
South Africa (datasheet/108613)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Sudan (datasheet/108555)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Tanzania (datasheet/108591)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Togo (datasheet/108579)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Tunisia (datasheet/108584)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Uganda (datasheet/108594)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Western Sahara (datasheet/108419)	Present	Native	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-

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Zambia (datasheet/108614)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Zimbabwe (datasheet/108616)	Present, Widespread	Native	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Asia					
Afghanistan (datasheet/108351)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Armenia (datasheet/108355)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652)	-
Azerbaijan (datasheet/108366)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652)	-
Bahrain (datasheet/108373)	Present, Widespread	Introduced	-	Chaudhary *et al.* (1981) (datasheet/17463#REF-DDB-22483)	-
Bangladesh (datasheet/108369)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Bhutan (datasheet/108383)	Present, Widespread	Introduced	-	Parker (1992) (datasheet/17463#REF-DDB-59579)	-
British Indian Ocean Territory					
-Chagos Archipelago (datasheet/108365)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272)	-
Brunei (datasheet/108378)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Cambodia (datasheet/108472)	Present	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
China (datasheet/108398)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-

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-Beijing (datasheet/108668)	Present	Introduced	Naturalized	Kairo *et al.* (2003) (datasheet/17463#REF-DDB-110168)	Naturalized
-Fujian (datasheet/108670)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Gansu (datasheet/108672)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Guangdong (datasheet/108671)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Hainan (datasheet/108675)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Hubei (datasheet/108676)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Jiangsu (datasheet/108683)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Shaanxi (datasheet/108694)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Shanxi (datasheet/108693)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Sichuan (datasheet/108691)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
-Yunnan (datasheet/108698)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated

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-Zhejiang (datasheet/108699)	Present, Widespread	Introduced	-	Flora of China Editorial Committee (2014) (datasheet/17463#REF-DDB-166467)	Cultivated
Cocos Islands (datasheet/108389)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272)	-
Georgia (datasheet/108433)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652)	-
Hong Kong (datasheet/108678)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
India (datasheet/108459)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
--Andaman and Nicobar Islands (-datasheet/108720)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Assam (datasheet/108723)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Himachal Pradesh (datasheet/108733)	Present	-	-	Urvi-Sharma *et al.* (2018) (datasheet/17463#REF-DDB-196281)	-
-Tamil Nadu (datasheet/108751)	Present	-	-	Devi *et al.* (2015) (datasheet/17463#REF-DDB-199875)	-
-Uttar Pradesh (datasheet/108753)	Present	-	-	Rashtra Vardhana (2017) (datasheet/17463#REF-DDB-200959)	-
-Uttarakhand (datasheet/108754)	Present	-	-	Dangwal *et al.* (2011) (datasheet/17463#REF-DDB-198971)	-
-West Bengal (datasheet/108755)	Present	Introduced	-	Duary and Mukherjee (2013) (datasheet/17463#REF-DDB-195592)	-

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Indonesia (datasheet/108455)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Java (datasheet/108714)	Present, Widespread	Introduced	Invasive	CABI (Undated) (datasheet/17463#REF-DDB-27)	Original citation: Soerjani *et al.* (1987)
-Lesser Sunda Islands (datasheet/108717)	Present, Widespread	Introduced	Invasive	CABI(Undated) (datasheet/17463#REF-DDB-27)	Original citation: Soerjani *et al.* (1987
-Maluku Islands (datasheet/108716	Present, Widespread	Introduced	Invasive	CABI (Undated) (datasheet/17463#REF-DDB-27)	Original citation: Soerjani *et al.* (1987
-Sumatra (datasheet/108719)	Present, Widespread	Introduced	Invasive	CABI(Undated) (datasheet/17463#REF-DDB-27)	Original citation: Soerjani *et al.* (1987
Iran (datasheet/108462)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174	-
Iraq (datasheet/108461)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174	-
Israel (datasheet/108457)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174	-
Japan (datasheet/108467)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Honshu (datasheet/108761)	Present	Introduced	-	Numata and Yoshizawa (1975) (datasheet/17463#REF-DDB-10306)	-
-Kyushu (datasheet/108762)	Present	Introduced	-	Numata and Yoshizawa (1975) (datasheet/17463#REF-DDB-10306)	-
-Ryukyu Islands (datasheet/108763)	Present	Introduced	-	Numata and Yoshizawa (1975) (datasheet/17463#REF-DDB-10306)	-

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Shikoku (datasheet/108764)	Present	Introduced	-	Numata and Yoshizawa (1975) (datasheet/17463#REF-DDB-10306)	-
Jordan (datasheet/108466)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Kazakhstan (datasheet/108480)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652)	-
Kyrgyzstan (datasheet/108471)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652)	-
Laos (datasheet/108481)	Present	Introduced	-	Waterhouse (1993) (datasheet/17463#REF-DDB-57459)	-
Lebanon (datasheet/108482)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Malaysia (datasheet/108514)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Peninsular Malaysia (datasheet/108765)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Sabah (datasheet/108766)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Sarawak (datasheet/108767)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Myanmar (datasheet/108503)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Nepal (datasheet/108524)	Present	Introduced	-	Moody (1989) (datasheet/17463#REF-DDB-41941)	-
North Korea (datasheet/108476)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-

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Oman (datasheet/108529)	Present, Widespread	Introduced	-	Chaudhary *et al.* (1981) (datasheet/17463#REF-DDB-22483)	-
Pakistan (datasheet/108537)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Philippines (datasheet/108535)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Saudi Arabia (datasheet/108552)	Present	Introduced	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Singapore (datasheet/108557)	Present, Widespread	Introduced	Invasive	Waterhouse (1993) (datasheet/17463#REF-DDB-57459)	-
South Korea (datasheet/108477)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Sri Lanka (datasheet/108485)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Taiwan (datasheet/108590)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Tajikistan (datasheet/108581)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652	-
Thailand (datasheet/108580)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Turkey (datasheet/108587)	Present, Widespread	Introduced	Invasive	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Turkmenistan (datasheet/108583)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652	-
United Arab Emirates (datasheet/108350)	Present, Widespread	Introduced	-	Chaudhary *et al.* (1981) (datasheet/17463#REF-DDB-22483)	-

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Uzbekistan (datasheet/108599)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652	-
Vietnam (datasheet/108604)	Present	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Yemen (datasheet/108609)	Present, Widespread	Introduced	-	Chaudhary *et al.* (1981) (datasheet/17463#REF-DDB-22483)	-
Europe					
Albania (datasheet/108354)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Austria (datasheet/108361)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Belarus (datasheet/108386)	Present	Introduced	-	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	Casual alien
Belgium (datasheet/108370)	Present	Introduced	Naturalized	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	Naturalized
Bulgaria (datasheet/108372)	Present, Widespread	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Cyprus (datasheet/108408)	Present, Widespread	Introduced	-	AMERICANOS (1972) (datasheet/17463#REF-DDB-4067)	-
Czechia (datasheet/108409)	Present	Introduced	Invasive	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	-
Czechoslovakia (datasheet/108403)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Federal Republic of Yugoslavia (datasheet/108610)	Present, Widespread	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
France (datasheet/108429)	Present, Widespread	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-

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-Corsica (datasheet/108704)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Germany (datasheet/108410)	Present, Localized	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Greece (datasheet/108443)	Present, Widespread	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Hungary (datasheet/108454)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Italy (datasheet/108464)	Present, Widespread	Introduced	Invasive	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
-Sardinia (datasheet/108758)	Present	Introduced	-	Nannini *et al.* (2009) (datasheet/17463#REF-DDB-1198484	-
Latvia (datasheet/108491)	Present	Introduced	-	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	Casual alien
Liechtenstein (datasheet/108484)	Present	Introduced	-	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	Casual alien
Luxembourg (datasheet/108490)	Present	Introduced	-	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	-
Netherlands (datasheet/108522)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Portugal (datasheet/108542)	Present, Widespread	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
-Azores (datasheet/108776)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Romania (datasheet/108548)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-

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Russia (datasheet/108550)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Russian (Europe) (datasheet/108784)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
-Russian Far East (datasheet/108785)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652	-
-Siberia (datasheet/108787)	Present	Introduced	-	Tselev and Fedorov (1983) (datasheet/17463#REF-DDB-177652	-
Serbia (datasheet/108549)	Present, Widespread	Introduced	Invasive	Nestorovic and Konstantinovic (2011) (datasheet/17463#REF-DDB-174739)	-
Spain (datasheet/108421)	Present, Widespread	Introduced	Invasive	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
-Balearic Islands (datasheet/108701)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Switzerland (datasheet/108393)	Present, Widespread	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
Ukraine (datasheet/108592)	Present	Introduced	-	DAISIE (2014) (datasheet/17463#REF-DDB-150689)	-
United Kingdom (datasheet/108431)	Present	Introduced	-	Tutin *et al.* (1980) (datasheet/17463#REF-DDB-144827)	-
North America					
Antigua and Barbuda (datasheet/108352)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Aruba (datasheet/108363)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-

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Bahamas (datasheet/108382)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-
Barbados (datasheet/108368)	Present	Introduced	-	CABI (Undated) (datasheet/17463#REF-DDB-27)	Original citation: May and Baker, 1964
Belize (datasheet/108387)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
British Virgin Islands (datasheet/108602)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-
Canada (datasheet/108388)	Present		-	CABI (Undated) (datasheet/17463#REF-DDB-27)	Present based on regional distribution
-British Columbia (datasheet/108654)	Present	Introduced	-	USDA-NRCS (2014) (datasheet/17463#REF-DBB-150757)	-
Cayman Islands (datasheet/108479)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-
Costa Rica (datasheet/108402)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Cuba (datasheet/108405)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Curacao (datasheet/108401)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-
Dominica (datasheet/108413)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Dominican Republic (datasheet/108414)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-

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El Salvador (datasheet/108571)	Present	Introduced	-	Clayton *et al.* (2014) (datasheet/17463#REF-DBB-169521)	-
Grenada (datasheet/108432)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Guadeloupe (datasheet/108441)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Guatemala (datasheet/108445)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Haiti (datasheet/108453)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-
Jamaica (datasheet/108465)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Mexico (datasheet/108513)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Montserrat (datasheet/108508)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Netherlands Antilles (datasheet/108356)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Nicaragua (datasheet/108521)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Panama (datasheet/108530)	Present	Introduced	-	Clayton *et al.* (2014) (datasheet/17463#REF-DBB-169521)	-
Puerto Rico (datasheet/108541)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Saint Kitts and Nevis (datasheet/108475)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-

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Saint Lucia (datasheet/108483)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Saint Pierre and Miquelon (datasheet/108539)	Present	Introduced	-	USDA-NRCS (2014) (datasheet/17463#REF-DBB-150757)	-
Saint Vincent and the Grenadines (datasheet/108600)	Present	Introduced	-	Broome *et al.* (2007) (datasheet/17463#REF-DDB-168959)	-
Trinidad and Tobago (datasheet/108588)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
U.S. Virgin Islands (datasheet/108603)	Present	Introduced	-	Acevedo-Rodriguez and Strong (2012) (datasheet/17463#REF-DDB-151259)	-
United States (datasheet/108597)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Alabama (datasheet/108796)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Arizona (datasheet/108798)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Arkansas (datasheet/108797)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-California (datasheet/108799)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Colorado (datasheet/108800)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Connecticut (datasheet/108801)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-

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-Delaware (datasheet/108803)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-District of Columbia (datasheet/108802)	Present	Introduced	-	USDA-NRCS (2014) (datasheet/17463#REF-DBB-150757)	-
-Florida (datasheet/108804)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Georgia (datasheet/108805)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Hawaii (datasheet/108806)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	Noxious weed
-Idaho (datasheet/108808)	Present, Localized	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Illinois (datasheet/108809)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Indiana (datasheet/108810)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Iowa (datasheet/108807)	Present. Localized	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Kansas (datasheet/108811)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Kentucky (datasheet/108812)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-

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-Louisinia (datasheet/108813)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Maryland (datasheet/108815)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Massachusetts (datasheet/108814)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Michigan (datasheet/108817)	Present, Localized	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Mississippi (datasheet/108820)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Missouri (datasheet/108819)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Montana (datasheet/108821)	Present	Introduced	-	USDA-NRCS (2014) (datasheet/17463#REF-DBB-150757)	-
-Nebraska (datasheet/108824)	Present, Localized	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Nevada (datasheet/108828)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-New Hampshire (datasheet/108825)	Present, Localized	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-New Jersey (datasheet/108826)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-

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-New Mexico (datasheet/108827)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-New York (datasheet/108829)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-North Carolina (datasheet/108822)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Ohio (datasheet/108830)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Oklahoma (datasheet/108831)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Oregon (datasheet/108832)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Pennsylvania (datasheet/108833)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Rhode Island (datasheet/108834)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-South Carolina (datasheet/108835)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Tennessee (datasheet/108837)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Texas (datasheet/108838)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-

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-Utah (datasheet/108839)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Vermont (datasheet/108841)	Present, Localized	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Virginia (datasheet/108840)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-Washington (datasheet/108842)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
-West Virginia (datasheet/108844)	Present	Introduced	-	Lorenzi and Jeffery (1987) (datasheet/17463#REF-DBB-36903)	-
Oceania					
American Samoa (datasheet/108360)	Present	Introduced	-	Whistler (1983) (datasheet/17463#REF-DDB-18174)	-
Australia (datasheet/1083620	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-New South Wales (datasheet/108620)	Present, Widespread	-	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	Probably native
-Northern Territory (datasheet/108619)	Present	Introduced	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	-
-Queensland (datasheet/108621)	Present, Widespread	-	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	Probably native

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-South Australia (datasheet/108622)	Present	Introduced	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	-
-Tasmania (datasheet/108623)	Present	Introduced	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	-
-Victoria (datasheet/108624)	Present	Introduced	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	-
-Western Australia (datasheet/108625)	Present	-	-	Queensland Department of Primary Industries and Fisheries (2011) (datasheet/17463#REF-DDB-181620)	-
Christmas Island (datasheet/108407)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Cook Islands (datasheet/108395)	Present	Introduced	-	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Federated States of Micronesia (datasheet/108427)	Present	Introduced	-	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Fiji (datasheet/108425)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
French Polynesia (datasheet/108533)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Guam (datasheet/108446)	Present	Introduced	-	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Kiribati (datasheet/108473)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-

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Marshall Islands (datasheet/108499)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Nauru (datasheet/108526)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
New Caledonia (datasheet/108517)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
New Zealand (datasheet/108528)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Niue (datasheet/108527)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Norfolk Island (datasheet/108519)	Present	-	-	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Northern Mariana Islands (datasheet/108505)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Palau (datasheet/108543)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Palau New Guinea (datasheet/108534)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Samoa (datasheet/108608)	Present	Introduced	-	Whistler (1983) (datasheet/17463#REF-DDB-18174)	-
Solomon Islands (datasheet/108553)	Present	-	-	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Tonga (datasheet/108585)	Present	Introduced	Invasive	Whistler (1983) (datasheet/17463#REF-DDB-18174)	-
US Minor Outlying Islands (datasheet/108596)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-

Contd......

-Wake Island (datasheet/108607)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Vanuatu (datasheet/108605)	Present	Introduced	-	PIER (2014) (datasheet/17463#REF-DDB-167272	-
Wallis and Fatuna (datasheet/108606)	Present	Introduced	Invasive	PIER (2014) (datasheet/17463#REF-DDB-167272	-
South America					
Argentina (datasheet/108359)	Present, Widespread	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Bolivia (datasheet/108379)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Brazil (datasheet/108381)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Alagoas (datasheet/108627)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Amazonas (datasheet/108628)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Bahia (datasheet/108630)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Ceara (datasheet/108631)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Espirito Santo (datasheet/108632)	Present, Widespread	Introduced	Invasive	13N-Brazil (2014) (datasheet/174633REF-DDB-178843)	-
-Goias (datasheet/108634)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Maranhao (datasheet/108635)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-

Contd......

-Mato Grosso (datasheet/108638)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Mato Grosso do Sul (datasheet/108637)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Minas Gerais (datasheet/108636)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Para (datasheet/108639)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Paraiba (datasheet/108640)	Present, Widespread	Introduced	Invasive	13N-Brazil (2014) (datasheet/174633REF-DDB-178843)	-
-Parana (datasheet/108643)	Present, Widespread	Introduced	Invasive	13N-Brazil (2014) (datasheet/174633REF-DDB-178843)	-
-Pernambuco (datasheet/108641)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Piaui (datasheet/108642)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Rio de Janeiro (datasheet/108644)	Present	Introduced	Invasive	13N-Brazil (2014) (datasheet/174633REF-DDB-178843)	-
-Rio Grande do Norte (datasheet/108645)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Rio Grande do Sul (datasheet/108648)	Present, Widespread	Introduced	Invasive	13N-Brazil (2014) (datasheet/174633REF-DDB-178843)	-
-Rondonia (datasheet/108646)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Santa Catarina (datasheet/108649)	Present, Widespread	Introduced	Invasive	13N-Brazil (2014) (datasheet/174633REF-DDB-178843)	-

Contd......

-Sao Paulo (datasheet/108651)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
-Sergipe (datasheet/108650)	Present	Introduced	-	Lorenzi (1982) (datasheet/17463#REF-DDB-23540)	-
Chile (datasheet/108396)	Present, Widespread	Introduced	Invasive	IABIN (2014) (datasheet/17463#REF-DDB-166628)	Also invasive on J. Fernandez Islands
-Easter Island (datasheet/108666)	Present, Widespread	Introduced	Invasive	IABIN (2014) (datasheet/17463#REF-DDB-166628)	-
Colombia (datasheet/108399)	Present, Widespread	Introduced	Invasive	IABIN (2014) (datasheet/17463#REF-DDB-166628)	-
Ecuador (datasheet/108416)	Present	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
-Galapaos Islands (datasheet/108700)	Present	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
French Guiana (datasheet/108434)	Present	Introduced	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Guyana (datasheet/108448)	Present	Introduced	-	Clayton *et al.* (2014) (datasheet/17463#REF-DDB-169521)	-
Paraguay (datasheet/108544)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Peru (datasheet/108532)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Suriname (datasheet/108568)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Uruguay (datasheet/108598)	Present	Introduced	Invasive	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-
Venezuela (datasheet/108601)	Present, Widespread	Introduced	-	Holm *et al.* (1979) (datasheet/17463#REF-DDB-18174)	-

The above shown Table 1 reveals that *C. dactylon* has high potential for further spread in many areas where it is still absent, and thus eco-physiological and genetic traits coupled with both forms of propagation give this species a high score for success in almost any ecosystem. However, Nitu *et al.* (2019) stated that distribution as well as adaptation of *Cynodon dactylon* to different ecological zones of Bangladesh is mainly dependent on weather and adaphic factors of their respective habitats. Generally, it is a warm season perennial grass species that initiates growth in the vernal season and its growth continues rapidly when moisture is adequate and they find the alkaline soil as their habitat. From this point of view, this study was aimed at analyzing the morphological variations of *C. dactylon* ecotypes along with determining the physicochemical properties of soils from their particular habitats.

2.2 Adaptation

Adaptation of *Cynodon dactylon* to different geographical and ecological zones of the world depends mainly on weather and adaphic factors of their particular habitats. If this grass species is well adapted and properly established, it stabilizes the soil and prevents erosion by wind and water, reduces tracking of mud, cools the surroundings and serves as a recreational area. This plant species all over the world have wide differences in their ranges of adaptation. If its adaptation characteristics are known well, professional landscapers can make their right choice for any particular area. However, certain adaptations are basic to survival of this grass species under specific environmental conditions and cannot be violated. Other characteristic such as colour, texture, density etc. are also the matter of adaptation. In addition, full sunlight and light shade, salt tolerance, drought tolerance, water tolerance, establishment rate, maintenance, nitrogen requirements etc. matters the adaptation of this grass species.

Cynodon dactylon is an invasive and competitive weed and its extensive stolon and rhizome system provide a means of rapid expansion. This grass requires high temperature and high light levels to thrive. They are drought tolerant but tend to grow where water is available. Although wide spread, this grass species thrives only under extreme disturbance and does not invade natural grassland or forest vegetation (Harlan and de Wet 1969). Growth of *C. dactylon* is greater on heavy clay soils than on light sandy soils, and this may be due to the greater water holding capacity of the clay (Burton and Hanna 1985). They also stated that this grass can even survive long periods of flooding, but little to no growth occurs without adequate soil aeration. It grows on soil with a wide range of pH values, however alkaline soils are tolerated more than acidic ones and large amount of nitrogen is required for maximal growth above ground.

Rhizomes are the primary over-wintering structure (Holm *et al.* 1977). The rhizome system of the grass species is superficial as well as deep, which may *Cynodon* species very much able to infest in both arable and waste lands. In a variety of conditions this grass species may show concentric growth, outward from the original rhizome sprout, and of the rhizomes which correspond to the circular above ground growth pattern (Horowitz 1972a). Dormancy period is not found in rhizomes and sprouting occurs once when apical suppression is relieved by fragmentation (Horowitz 1972b). Stolons are formed newly at the basal node of shoots which developed from rhizomes. The initial stolons are more away from the central shoot followed by secondary stolons growing in all directions to form dense circular mats of sod. Roots developing from rhizomes and stolons are much longer and more abundant than those close to the original stem (Rochecouste 1962). The rhizomatous and stoloniferous growth lead to the extensive spreading capabilities of *C. dactylon* and these natures along with some other morphological features may make all types of accessions to be well adapted to the soil of different parts of world indicating the functional qualities like rigidity, resiliency, elasticity etc. of this grass species.

Rochecouste (1962b) observed growth response of the biotypes of *C. dactylon* when treated with trichloroacetic acid and 2, 2- dichloropropionic acid. The findings indicated that the tetraploid biotypes were more resistant than the triploid, and that biotypes of the same chromosome number were also found to differ in their response to these herbicides. Investigations on the effects of adding non-ionic, cationic and anionic wetting agent to aqueous solutions of the herbicides in which the leaves of plants were immersed showed that, in general the wetting agents did not significantly increase the phytotoxicity of both the herbicides. The author of this research work also stated that bud dormancy responded to uptake of TCA and dalapon via the roots was through the transpiration stream. But in tetraploids the dormant period induced by the acids was shorter than in triploids. With reference to TCA, Barrons and Hummer (1951) considered foliar application to be probably unimportant for a systemic effect. Otis (1951) and Stahler (1950) found that major effects were through the roots and that foliar contact has little effect, while Peters and Willard (1951) found that foliar applications of TCA were effective and that translocation occurred basipetally. Referring to dalapon, a judgement may be made that although the main effects are from foliar transport, root transport also takes place (Barrons 1953, Crafts 1957). However, the present study has established the existence of biotypes of *C. dactylon* differing both in their morphological and anatomical characters. It is also clear that in *Cynodon* complex both tetraploid and triploid races may occur and these races may differ in their response to the herbicides.

Ramakrishnan and Gupta (1973) observed different type of response to nitrogen, phosphorus and potassium for the three edaphic ecotypes in *Cynodon dactylon*. The soils for the three sites differed markedly in respect of available nutrients and the three populations differed also from each other regarding uptake of various nutrients. It was concluded by the investigators that besides calcium the availability of nitrogen, potassium and also phosphorus played important role for restriction of the three ecotypes to their respective habitats. However, Ramakrishnan and Sing (1966) reported that the three edaphic ecotypes of *Cynodon dactylon* showed direct response to calcium and also showed a differential response to three other nutrients. The investigators of this study postulated that if wide fluctuations in phosphorous availability occur in a non-calcareous soil then a wide range of tolerance for the population of *C. dactylon* naturally occurring in this soil is to be expected and this was found to be the case. Therefore, such a differential response of the populations of *C. dactylon* is well related to the phosphate availability in the soils. Besides the calcium and phosphorous interaction and similar interactions among nitrogen, phosphorous, and potassium may also influence the nutrition of the ecotypes of *C. dactylon*. The need for balance between these nutrients has been shown by Richards and Rees (1962) in case of crop species.

Naturally adapted salt tolerant populations provide a valuable material for exploring the adaptive components of salt tolerance. Under this aspect two populations of *Cynodon dactylon* were subjected to salt stress in hydroponics by Hameed and Ashraf (2008). It is known that the evolution of a particular character depends mainly on two determinants occurring together for a considerable length of time i.e; the existence of a great magnitude of appropriate genetic variation and the occurrence of appropriate natural selection (Ashraf *et al.* 1986, Ashraf 1994). Mechanism of adaptation to saline environments can be very specific for different ecotypes. In the ecotype of *C. dactylon* from the salt range, shoot dry weight was slightly affected by increasing the salt level, but its root weight was markedly increased. Gulzar *et al.* (2003) and Marcum (1999) also reported increased root weight under saline conditions in relatively salt tolerant grasses. Salt tolerant plants differ from salt sensitive ones in having a low rate of Na^+ and Cl^- transport to leaves, and the ability to compartmentalize these ions in vacuoles to prevent their build-up in cytoplasm or cell walls and thus, avoid salt toxicity (Munns 2002). Due to these phenomena *C. dactylon* from the salt range thrived well under saline conditions, because it accumulated much much less concentrations of both Na^+ and Cl^- at high external salt levels. Shoot or root K^+ and Ca^{2+} contents varied inconsistently under salt stress in both ecotypes. There are several reports on considerable inhibition of both K^+ and Ca^{2+} uptake under salinity as reported by Gonzalez *et al.* (2000) and Netondo *et al.* (2004). Their study reveals considering salt tolerance of a natural population of *C. dactylon* from salt range confirms that impact of natural selection on this population has been consistent and considerable from physiological and biochemical adaptations aspect. Another example of physiological adaptation of *C. dactylon* was performed by Tan *et al.* (2010). From enzymatic protection mechanism, their data were consistent with an integrated pathway involving CAT, SOD, POD, GR and APX for protection against detrimental effects of activated oxygen species under submerged stress. These data suggest that the antioxidative activities of roots play an important role in enduring long term and deep sub-emergence of *C. dactylon*.

Nutrient acquisition in differentially adapted populations of *Cynodon dactylon* and *Cenchrus ciliaris* under drought stress was studied by Akram *et al.* (2008). They used two populations of each of two grass species in their experiment. One population of each of two grass species was collected from drought-hit area and other from often irrigated field and both of them were subjected to three different levels of water stress. Populations of both grasses collected from drought-hit area were better than the irrigated field. The authors reported that a significant reduction in fresh and dry biomass of two populations of each of two grass species was observed when 88 days old plants were subjected to water stress for a period of 30 days. Their findings may be supported by Ashraf *et al.* (1986) who found genetic variation for drought tolerance among various grass species. It was prominent that populations from drought-hit area accumulated greater amount of K^+ than those collected from irrigated area. According to the investigators, naturally occurring drought prone areas impose a considerable selection pressure on the plant species occurring there and selectively allow some of plant populations to grow and produce offspring there. Drought-hit area is generally unproductive due to low rainfall and brakish ground water there is rich in salinity (Afzal *et al.* 1999, Ahmad *et al.* 2002). Therefore, the plant species inhabiting the drought-hit area may experience severe saline conditions since the time they started growing there.

The effect of shade on the growth and development of *Cynodon dactylon* was determined by Juraimi *et al.* (2004) under glasshouse conditions. Results have shown that continuous shading severely reduced the number of tillers, leaf area, leaves and stem dry weight, root dry weight and total dry weight of this grass species. Shading at later stages also reduced growth of this grass but early shading followed by full light at later stages had less shade effect. Light is important to plant growth based on the photosynthesis process and phytochrome reactions (Willium and Joseph 1982). The result of this study demonstrate the considerably decreased growth of *Cynodon dactylon* at all levels of shade treatment which was supported by the reports of Schmidt and Blaser (1969). But researchers of this study found increased total nitrogen in the stolons of *C. dactylon*. That might be due to limited nitrate reduction and inhibited nitrogen utilization, which resulted in poor production of forage and underground parts of *C. dactylon*. Burton *et al.* (1959) reported almost similar results. Semba (1996) found reduced irradiance which results lower dry matter production, net photosynthetic rate, relative growth rate, net assimilation rate and larger leaf area ratio when spring barley and five weed species were grown in a growth chamber at three light levels over a four weeks period. The overall responses of *C. dactylon* to shading indicate that the canopy shading typical of many row crops will reduce vegetative growth significantly (Keeley and Thullen 1978). This type of shading sequence might be expected to occur in a row crop in which the weed usually emerges soon after the emergence of the crop.

Anatomical adaptations of *Cynodon dactylon* were studied by Hameed *et al.* (2010). They evaluated root and stem anatomical modifications of naturally adapted salt tolerant population of *C. dactylon* from highly saline soils. They collected a population of the non-saline soil for comparison. Both populations were subjected to salt stress hydroponically. They found that the salt range population showed specific root and stem anatomical adaptations for its better survival under harsh saline environments. Increased exodermis and sclerenchyma, endodermis, cortex and pith parenchyma in roots were critical for checking water loss and enhancing water storage capability. In stem, succulent stem area, increased epidermis and sclerenchyma thickness, increased cortex thickness, and increased also number and area of vascular tissue which seemed to be crucial for its better survival under harsh saline environments.

Grasses reside in the earth in abundance than other analogous faction, some plants are present in hot, moist and tropical climates, while other plants inhabited in polar areas, where sunlight is not present. From marketable and dietary point of view Poaceae is very important family of plants for the welfare of human beings, as its many members are known as staple food and fodder for livestock (Jones 1999). Considering application of leaf epidermal anatomical technique for identification of some grass species Ishtiaq *et al.* (2018) selected five grass species for their study. *Cynodon dactylon* was one of them, which showed stomatal index of 15.0. Their study revealed that all the selected species contained paracytic type of stomata in which guard cells were accompanied by two subsidiary cells and guard cells were dumbbell shaped. Two years before of this work Ahmad *et al.* (2016) studied ecotypic adaptations in *Cynodon dactylon* for altitudinal stress tolerance. Differential response of all the three ecotypes in terms of adequate structural modifications to different elevation levels was an evident to confirm the hypothesis that plants inhabiting different altitudes show variation in structure (internal modifications) and strategic alteration (response) due to heterogenity in environmental gradients. Soil at top hill was more acidic and displayed significant increase in ionic content and total nitrogen. High elevation had severe impact on morpho-anatomical and physiological attributes. Chlorophyll content was found to decline significantly at high elevations. Anatomical alterations (increased leaf thickness, intensive sclerification around the vascular bundle and pith area) were observed. Leaf morphological variables (leaf width, length, thickness, area, weight, stomatal density and specific leaf area) varied consistently with increasing altitude (Kofidis *et al.* 2007, Fatemeh *et al.* 2011). The decreasing trend in case of leaf length with increased altitude may be under the control of environmental conditions rather than genetic control (Hovenden and Schoor 2004). Therefore, it appears that for *C. dactylon* at least, morphological characters in response to altitude are largely environmentally controlled. Thus, Royer (2001) and McElwain (2004) stated that leaf morphological characters may well serve as useful substitution measurement of climate.

Banan *et al.* (2019) conducted a study on anatomical adaptation in species belonging to the family Poaceae growing in Al-Ha'ir region of Riyadh, Saudi Arabia. Aim of their work was to determine the anatomical adaptations of leaves and stems of four species of this family and they were *Cynodon dactylon* L. Pers., *Chloris barbata* SW., *Setaria verticillata* L.P. Beauv., and *Panicum coloratum* L. Cross-sections of the leaves showed that epidermal cells were spherical to oval and formed a layer with a thick cuticle as well as numerous bulliform cells in a fan shape and prickles and ground tissue system was found to be consisted of chlorenchyma cells. In *C. dactylon* and *C. barbata* a large vascular bundle were found to be surrounded by two bundle sheaths, while in case of *S. verticillata* and *P. coloratum* one bundle sheath sclerenchyma was found to be surrounded by vascular bundle. At the same time one bundle sheath with chlorenchymatous cells was surrounded by small vascular bundles. The researchers of this study also observed cross section of stems to exhibit an epidermis consisted of single layer of cells with spherical to oval shaped and had a thick cuticle. Their findings showed ground tissue to contain strands of chlorenchyma cells followed by sclerenchyma tissue surrounding vascular bundles, making a continuous cylinder. They found scattered vascular bundles in the ground tissue and each vascular bundle was found to be surrounded by a single sclerenchymatous bundle sheath. Their findings indicated that these four plant species showed such an anatomical adaptations which enhanced drought tolerant capabilities and facilitating survival in both arid and semi-arid regions.

Duble (2001) stated that temperature is the main environmental factor that limits the adaptability of *C. dactylon* to tropical and subtropical areas of the world. At the onset of low temperature, this grass species began to discolour, change of protein fractions in composition and reserve carbohydrates increased in the stems and rhizomes. Leaves and stems of *C. dactylon* remained dormant until average daily temperature rises and the roots and rhizomes of bermudagrass continued to grow several weeks after the leaves and stems stop growth. This grass developed into a semidormant state during very dry conditions, but had the capability of surviving extreme droughts. Generally, diploid or tetraploids of *C. dactylon* have the deepest root and rhizome penetration and better withstand prolonged drought periods. The diploid (common) species also has the characteristic of producing seed heads under stress conditions such as drought. Thus, the seeds provide another method by which the species can survive extreme drought.

Ecotypic adaptations in *Cynodon dactylon* in relation to growth performance and stomatal behaviour was studied by Tufail *et al.* (2017). They stated that evolution has great ecological significance in term of plant morphological and stomatal characteristics that must have been genetically fixed during the long evolutionary period. Impact of environmental conditions on growth and stomatal features of twelve ecotypes of *C. dactylon*, collected from ecologically different habitats were evaluated. There was a huge variation in all morphological characters and these were due to heterogenity of environmental conditions to which these ecotypes were originally adapted. Since these ecotypes were evaluated under controlled conditions, the characteristics were genetically expressed (Mojica *et al.* 2012, Paccard *et al.* 2013), and they are representatives of their respective habitats. The collected ecotypes from stressful condition when grown in normal climatic conditions and showed stimulated growth (Bita and Gerates 2013, Hu *et al.* 2015), whereas those from non-stressed environments responded normally. Huge number of leaves might certainly improved the photosynthetic efficiency of a plant (Weraduwage *et al.* 2015), but smaller leaves increased water use efficiency by controlling transpiration rate (Medrano *et al.* 2015), which is a vital object to survive in harsh saline desert conditions. In relation to these features stomatal density improved the photosynthetic capability of a plant species and thus, sometimes ecotypes of *C. dactylon* might be rather as the better. Stomatal shape, however, was found to be different in different ecotypes.

Under stress condition plants are greatly restricted in growth field and may evolve many mechanisms to adapt rapidly to drought stress condition for keeping growth and productivity (Yamaguchi-Shinozaki and Shinozaki 2006, Zhu 2002). Now more attentions are being paid to mechanisms of plant drought stress tolerance. Shi *et al.* (2012) evaluated the natural variation of drought tolerance among nine *C. dactylon* varieties by measuring physiological responses after drought stress treatment through withholding water. Three groups differing in drought tolerance were identified, including two tolerant, five moderately tolerant and two susceptible varieties. *C. dactylon* is a warm-season creeping grass and has natural variation in the drought response (Kim *et al.* 2009, Zhao *et al.* 2011). The upper parts of *C. dactylon* die off under drought stress condition, but the grass keeps growing from its rhizome. This grass species reproduces rapidly and thus, some fundamental specifications make this grass a potential target for genetic engineering of stress tolerant plants. *C. dactylon* is found to be adapted to cultivation in a wide range of climatic condition (Chan and Shi 2015). Analysis of natural variations of drought stress tolerance revealed that different *C. dactylon* cultivars exhibited varied tolerance to drought stress (Lu *et al.* 2007, Shi *et al.* 2012, Shi *et al.* 2014). Plants develop different approaches to cope with adverse environmental conditions and thus abiotic stress tolerance may be considered as a complex trait. Based on these observations Chan and Shi (2015) proposed a possible model that is depicting now *C. dactylon* to abiotic stress responses.

Expansion of *Cynodon dactylon* has become limited in Italy due to lack of suitable species for cultivation in the Mediterranean climate. With this view, Viggiani *et al.* (2015) developed a research project with the main purpose to find out an agronomically characterised native turfgrass species of Southern and Central Italy and to compare them with some commercial cultivars. During the first step of the research, 11 sites from 6 regions of Southern and Central Italy were identified. In these sites 24 ecotypes of *Cynodon dactylon* were collected, and their habitus and phenology plus some biometric parameters were determined. During the two years of research both botanic and agronomic characterization of the collected ecotypes of *C. dactylon* and their comparison with three commercial cultivars were carried out. In this study some native accessions showed behaviour similar to commercial cultivars while an ecotype from the Abruzzo region showed better results when compared to the commercial cultivars for several quality indices. Majority of the commercially available turfgrasses used in Italy are of foreign and thus, a great threat can be the use of this alien plant material into natural ecosystem (Potenza *et al.* 2012). The use of native warm-season grasses would be preferred due to their reduced threat to native ecosystems and their greater resistance to high temperature and water deficit stress (Kenna 2008, Erdogan *et al.* 2011). However, Leto *et al.* (2008) stated that the native ecotypes of *C. dactylon* of the Mediterranean region are of remarkable quality.

Chaudhary *et al.* (2014) studied foliar epidermal anatomy of twenty nine grass species belonging to 10 different tribes of Thal desert. Among them *Cynodon dactylon* was of significant systematics. Leaf epidermis of *C. dactylon* was found to be important for sorting out the evolutionary and phylogenetic importance. Almost all epidermal cells in *C. dactylon* were sinuous or wavy similar to the findings of Ahmad *et al.* (2010). Like almost all grasses stomata in *C. dactylon* were paracytic type earlier proved by Abid *et al.* (2007). According to Metcalfe (1960), Chaudhary *et al.* (2001b) and Ahmad (2009) silica bodies in *C. dactylon* are saddle shaped, which were again confirmed be Chaudhary *et al.* (2014). In *C. dactylon* subsidiary cells were found to be triangular at abaxial side and high dome shaped at adaxial side. Chaudhary *et al.* (2001) reported that subsidiary cells in this grass species to be only triangular in shape and no variation was found by them. Micro hair was present either on adaxial, abaxial or both sides (Chaudhary *et al.* 2001, Freire *et al.* 2005, Ahmad 2009). But no micro hair was found adaxially or abaxially by Chaudhary *et al.* (2014) in his study, which might be due to dictatorial environmental conditions. These findings prove that *C. dactylon* may adapt themselves against any unfavourable environmental conditions.

Planting of appropriate grass species in severe SO_2 polluted areas is very critical to achieve better air quality and landscape. *Cynodon dactylon*, a widely used grass species has good SO_2 tolerant ability. From this point of view Wang *et al.* (2014) selected 9 out of 38 *C. dactylon* accessions from South West China as representative of high and intermediate SO_2 tolerant, and SO_2 sensitive accessions to comparatively analyze their physiological differences in leaves under SO_2 treated and untreated conditions. Their findings revealed that SO_2 tolerant *C. dactylon* accessions showed higher soluble sugar, proline, and chlorophyll a content both under SO_2 treated and SO_2 untreated conditions along with few other behaviour. Finally, their findings indicated that SO_2 tolerance of *C. dactylon* might be largely related to soluble sugar, proline, chlorophyll a contents and SO enzyme activity. Their previous study indicated that growth rate of *C. dactylon* was affected and visible symptoms appeared on leaves under SO_2 stress condition; although, this species has much better SO_2 tolerant ability among warm season turf grasses (Wang *et al.* 2013). *C. dactylon* is widely distributed in tropical and subtropical regions of the world and displays abundant genetic diversities worldwide (Assefa *et al.* 1999, Kang *et al.* 2008, Farsani *et al.* 2012, Ling *et al.* 2012). Soluble sugars and proline, as two major compatible solutes in cytoplasm and organelle, play important roles under both of stress condition, drought and salinity (Shi *et al.* 2012, Uddin *et al.* 2012). However, increased soluble sugar and proline contents in SO_2 tolerant *C. dactylon* accessions are not likely involved in osmotic pressure but more likely involved in maintaining cell membrane stability, synthesis of other compounds, supply of energy, action as regulators of gene expression, and signal molecules based on their multiple functions (Mohammadkhani and Heidari 2008). However, soluble sugar and proline contents can be considered as marker for selection of *C. dactylon* variations with SO_2 tolerability. At the same time, it indicates that *C. dactylon* accessions are more adaptable to this chemical hazardous environment compared to that of other grass species.

Hameed *et al.* (2013) examined anatomical modifications of the leaves of a naturally adapted salt tolerant population of *Cynodon dactylon* collected from a heavy salt affect soil in the vicinity of a natural salt lake. An ecotype of this grass was also collected from a non-saline habitat and both the ecotypes were subjected to salt stress in hydroponics. The prominent adaptive features in salt tolerant ecotypes were found to increase the development of vesicular hairs for exclusion of the toxic ions through leaves as well as affected parenchymatous tissue and that might be due to salt stress. Additionally, some adaptations reflected were the development of xeromorphic characteristics essential for checking the undue water loss. Based on various growth attributes measured by Hameed and Ashraf (2008), the ecotype of *Cynodon dactylon* from the salt range was categorized as highly tolerant to salt stress. One of the major anatomical modifications in ecotype of *C. dactylon* from the salt range was increased hairness, which had been considered a characteristic feature in grass leaves at highly saline habitat (Cheng and Chou 1997). Reduction in stomatal density and size might also be an efficient feature of checking undue water loss via transpiration during limited water availability under high salinities as reported by Walsh (1990), and Bray and Reid (2002). The salt range ecotype showed also decreased stomatal area and density on the adaxial leaf surface, which indicated that it can be regarded as the best adapted ecotype against high saline environment.

Morphological variation in *Cynodon dactylon* and its relationship with the environment along a longitudinal gradient were determined by Wang *et al.* (2020). The objectives of their research were- i) analyze the variability in 10 quantitative traits of *C. dactylon* collected at sites representing 13 different longitudes, ii) explore the pattern of morphological variation among and within populations, and iii) evaluate the relationship between morphological traits, soil, and climate at the different longitudes. All of the 13 different longitudinal sites were divided into three groups based on morphological traits by cluster analysis. The major sources of diversity at the different longitudes were leaf length and width of the erect shoot, and the internode lengths of the erect shoot and stolon were determined by principal component analysis. Correlation analysis indicated that longitude was significantly and negatively correlated with these traits as well. Average rainfall was significantly correlated with leaf length and internode lengths of the erect shoot and stolon. Average temperature was only significantly correlated with internode length. Average sulfur showed significant correlation with internode length, plant height and reproductive branch height, while Ca was significantly correlated with internode length and stolon. Soil pH showed similar results. From their findings it can be said that longitude is an important factor that affects morphological trait variation in *C. dactylon*, and the leaves of the erect shoot and the internode length enlarged significantly with the collection sites moving from east to west. Previous studies have shown that morphological variations are significantly affected by geographical locations (Anway 1972, Cooke *et al.* 2012, Soolanayakanahally *et al.* 2013, Naima *et al.* 2013, Antigoni *et al.* 2018). A study of 260 accessions of bermudagrass germplasm indicated that there is an extensive morphological variation in *C. dactylon* populations along longitudinal gradients, which has been found in some other studies (Potenza *et al.* 2014, Akbari *et al.* 2018, Reasor *et al.* 2018). In a different study Wang *et al.* (2020) stated that their research explored the evolutionary trends in morphological trait variation in wild bermudagrass populations along a longitudinal gradient, and provides abundant wild resources for breeding in bermudagrass.

However, based on the above mentioned findings by different research workers in different times it can be said that *Cynodon dactylon* is an invasive and competitive weed. By means of extensive stolon and rhizome system this grass species provides a way of rapid expansion in spite of anatomical, physiological and biochemical constraints. In general, this grass species requires high temperature and high light intensity to thrive and grows also in disturbed areas. Edaphic factor along with some other environmental conditions are very much important for proper growth and development of this grass species. Although extremely drought tolerant, *C. dactylon* leads to grow where water is somewhat available. This plant is not frost or shade tolerant and the rhizomes and stolons are susceptible to desiccation. Complete eradication of this plant species has not been possible and thus, they are found more adaptable to many sorts of environmental condition that may be from one continent to another of the word. This grass can even survive long periods of flooding, but little to no growth occurs without adequate soil aeration. It grows well on soil with a wide range of pH, alkaline soils are more tolerated than acidic ones and a large amount of nitrogen is required for its maximal growth above ground.

2.3 Cytotype diversity

Cynodon dactylon, one of the best fodder grass is equally valued as a lawn grass. This grass species has been investigated extensively and various chromosome number such as 2n = 18, 18 + 1-3B, 30, 36, 36 + 1-2B, 40 and 54 are known for this species (Brown 1950, Gould 1966 & 1970, Malik and Tripathi 1968, Hurcombe 1946 & 1947, Mitra and Datta 1967, Mehra *et al.* 1968). A meiotic number of n = 18 has been reported with nearly 100% pollen fertility by Mehra and Sharma (1975). In 1978 Sharma *et al.* made a preliminary cytological screening of 19 grasses of coastal Orissa of India and found *C. dactylon* to show haploid chromosome number of n = 18. Their findings were found to be compared with the reports of Federov (1974), Christopher and Abraham (1974), Brown (1950) and Hurcombe (1947).

Chromosome number in *C. dactylon* in relation to ecological conditions was reported by De Silva and Snaydon (1995). They collected populations of *C. dactylon* from three habitats (roadsides, lawns and paddy-fields) within each of five climatic regions (arid, dry, intermediate, wet and hill) in Sri Lanka. Further populations were collected by them from forests and grass lands in the hill country. Most of the populations contained only tetraploid plants (2n = 36), but populations from roadsides and lawns in the wet region, and from forests in the hilly country, contained only diploid plants (2n = 18). Populations from paddy fields in the wet region contained both tetraploid and diploid plants. No triploid plant was found in any environment, despite the fact that *C. dactylon* predominantly reproduces vegetatively by both stolons and rhizomes. They seem that soil acidity may be the main determining factor for distribution of the two cytotypes. However, Gupta and Srivastava (1970) reported naturally occurring triploids (2n = 27) in *C. dactylon* for the first time. They stated that since, besides diploids, tetraploids are also known to occur in the species and the triploids were presumably the result of a natural cross between diploid (2n = 18) and tetraploid (2n = 36). They thought about another manner in which the triploids could arise was the fertilization of an unreduced gamete by a reduced gamete. Although, the triploid hybrid was artificially produced by Forbes and Burton (1963) by crossing 2x and 4x races of *C. dactylon*, where frequency of trivalents indicated homology between the diploid and tetraploid genomes.

Dissimilarly, Moffett and Hurcombe (1948), and Rochecouste (1962) recorded chromosome numbers of 2n = 40 in *C. dactylon* and considered the basic number to be x = 10. But Clayton and Harlan (1970) considered the basic number to be x = 9, when they obtained counts of 2n = 18. It seems that real differences or dissimilarities in chromosome number exist within the species and this sort of variation might be related to ecological conditions. In order to uncover patterns and processes of segregation of the co-existing cytotypes, Hulber *et al.* (2009) investigated a zone in the eastern Alps (Austria) where diploid and polyploid individuals of the alpine herb, *Senecio carniolicus* were found to occur. They stated that compared to diploids, hexaploids were found in more species and denser communities. They stated that this might be due to their better competitive ability and lower tolerance of abiotic stress compared to the diploids. However, polyploid is significantly considered as major driving force for speciation and it was reported that all angiosperms had at least an ancient polyploid origin (Soltis *et al.* 2009). Weiss *et al.* (2003) and Baack (2004) stated that differences at ploidy level do not only occur between species, but also within them. Some research works have focused on the population biology of polyploids and specially the origin of tetraploids and their interactions with their diploid progenitors (Lewis 1980, Thompson and Lumaret 1992). This perspective has arisen as a result of research works that suggests that polyploids may arise repeatedly in diploid taxa (Soltis *et al.* 1989, Wolf *et al.* 1990, Soltis and Soltis 1993).

Maguire (1977) stated that missing chromomeres may reflect failure of condensation or differential aggregation during precipitation of chromatin subunits. This may account for the variability observed in chromomere patterns of *C. dactylon*, and would therefore indicate that no intrachromosomal morphological differences may have existed in the materials. Ho and Kasha (1974), and Gillies (1972) found that differences in contraction rate changed relative lengths of chromosome and arm ratios. The differences in contraction rate might be due to differential rates of contraction in euchromatic and heterochromatic regions. A chromosome with euchromatic regions in both arms may contract faster than one with an arm that is largely heterochromatic. However, there is still the possibility that some differences, such as the difference in arm ratios for particular two chromosomes could be due to chromosomal biotypes. Rawal and Chedda (1971) concluded from cytomorphological studies that varieties *afghanicus* and *aridus C. dactylon* evolved from a common non-rhizomatous ancestor and that the barriers to gene flow are primarily ecological and geographical rather than chromosomal. The similarity of the pachytene morphology noted in the two *Cynodon dactylon* studies suggests that the genomes in both cases are homologous. This is in agreement with Harlan and de Wet (1969) concluded that only one genome exists for all crossable species in the genus *Cynodon*. However, Brilman *et al.* (1982) studied pachytene chromosome morphology of diploid *C. dactylon*. The chromosomes were identified by their arm length ratios, relative length, and position and number of prominent chromomeres. They found a very little variability in chromosome morphology among the clones. Chiavegatto *et al.* (2016) stated that *Cynodon* species showed high morphological variability in a large number of varieties and cytotypes, hampering identification. Their study aimed to determine the karyotype asymmetry index among accessions of *Cynodon* to discriminate between them. They reported basic number for the genus as x = 9 and found for two diploid accessions (2n = 2x = 18), one triploid accession (2n = 3x = 27), four tetraploid accessions (2n = 4x = 36) and one pentaploid accession (2n = 5x = 45). They divided the accessions into two groups on the basis of the karyotypic asymmetry indices and said that the data facilitated a determination of the degree of proximity between the accessions.

As mentioned earlier, the variation in ploidy levels (from diploid to hexaploid) in *Cynodon* has been described by several authors, among them Dhaliwal and Gupta (2011) described three cytotypes of *Cynodon dactylon* elaborately. All these findings indicate that there is a vast genetic variability in the group and that is expressed as morphological, phenotypical and biochemical variability. These are important characteristics for the origin and evolution of species (Schifno-Wittmann 2004) and the development of improved cultivars (Wu and Taliaferro 2009). The origin of *Cynodon dactylon* var. *dactylon* (tetraploid) has been associated with the union of the unreduced gametes of the ancestor *C. dactylon* var. *aridus*. *C. dactylon* var. *polevansii* is also strictly described as tetraploid (2n = 4x = 36). In a study by Anderson *et al.* (2009) *C. polevansii* were identified as triploid, tetraploid and pentaploid. Wu (2011) stated that pentaploid can occur through crosses between hexaploid and tetraploid cytotypes. Chiavegatto *et al.* (2016) have stated further that regarding the accession's relationship, the methodologies used for determining chromosome asymmetry revealed that *Cynodon* species was closely linked to *C. dactylon* var. *polevansii*. The other accessions which were identified as *Cynodon* sp., were closer to each other. Their observations can be extrapolated to *C. dactylon* var. *dactylon* and *C. transvaalensis*, which had the same chromosome number, ploidy, DNA content and karyotype formula. The diploid accessions *C. incompletes* var. *hirsutus* and *C. nlemfuensis* had the same cytogenetical characteristics but they differed in case of karyotype formula and chromosomal asymmetry. *C. nlemfuensis* was found to be close to the triploid *Cynodon* sp. and the pentaploid *C. dactylon* var. *polevansii.*

Genetic analysis of Chinese *Cynodon* accessions by flow cytometry and amplified fragment length polymorphism (AFLP) markers were made by Wu *et al.* (2006). Their findings revealed four polyploid cytotypes among the Chinese accessions. Tetraploid (2n = 4x = 36) accessions were more prevalent (88%), with nuclear genome sizes ranging from 1.96 - 2.30 pg/2L nucleus^{-1} respectively. Seven hexaploid (2n = 6x = 54), three pentaploid (2n = 5x = 45) and six triploid (2n = 3x = 27) accessions had respective nuclear size of 2.90 to 3.13, 2.37 to 2.49, and 1.55 to 1.65 pg/2L nucleus^{-1}. They grouped the accessions into five clusters based on 466 polymorphic AFLP bands. The close genetic relatedness of three different cytotypes indicated their common ancestry. It is evident that hexaploid and pentaploid are rare in *Cynodon*. Powell *et al.* (1968) reported a hexaploid clone from a cross of tetraploid × diploid parents, and speculated that doubling of chromosome number at an early zygote probably accounted for its occurrence. Another hexaploid plant was identified in the progeny of a self-pollinated plant of tetraploid *C. dactylon* by Felder (1967). On the other hand, Johnston (1975) found three pentaploid plants among progeny from a hexaploid population which might be due to derivation from interspecific hybridization of *C. barberi* and *C. dactylon* and thereafter crossed with a tetraploid *C. dactylon* plant. Burton *et al.* (1993) reported 'Tifton 85' pentaploid, as F_1 interspecific hybrid from a *C. dactylon* × *C. nlemfuensis* cross. However, Wu *et al.* (2006) said that tetraploid *C. dactylon* is prevalent among *Cynodon* germplasm indigenous to the eleven provinces of China. Southeast China may represent a unique geographic area for the occurrence of hexaploid and pentaploid *C. dactylon* cytotypes frequently.

Kang *et al.* (2008) conducted a study on genetic diversity among Korean *Cynodon* spp. The objective of their study was to evaluate the genetic diversity of this grass species at morphological, cytological and molecular level. They observed morphological parameters, the nuclear DNA content and ploidy levels in 43 *Cynodon* ecotypes. They evaluated AFLP markers to define the genetic diversity and chromosome counts to confirm the inferred cytotypes. Triploid, tetraploid, pentaploid and hexaploid cytotypes were recorded based on the genomic number x = 9. Majority (81%) of the collections were tetraploid (2n=4x=36). The fast growing fine textured ecotypes had lower ploidy levels, while the pentaploids and hexaploids were coarse types. Hanna *et al.* (1990) reported similar results about hexaploid *Cynodon* which showed coarse textured leaves of a dark green colour and rapid establishment rate. *Cynodon* ecotypes collected from Korea showed not only diverse phenotypic characteristics but also extra chromosomes (Kang *et al.* 2008), which seem to be B chromosomes with distinctive morphology and an irregular inheritance (Jones 1995). This type of chromosome results showed numerical variations. Nitu *et al.* (2019) studied 19 accessions of *Cynodon dactylon* and found eight diploid, one tetraploid and rest of them aneuploid whose chromosome numbers were 12, 14, 16, 20, 22, 22, 24, 26, 32, 40 etc. They stated that the preponderance of aneuploidy throws light on the evolutionary tendency which might go hand to hand in the phylogenetic evolution due to their different adaptive values. When one of the characters might put a type or accession of *C. dactylon* at highest level of advancement then the others may be still in original stage. However, aneuploids play an important role in the breeding programme and thus, the main objective of forage breeding programme may be an important way to obtain those genotypes with desirable traits.

Gulsen *et al.* (2009) said that for developing a better understanding of associations among ploidy level, geographic distribution and genetic diversity of *Cynodon* accessions many ideas could be taken under consideration for bermudagrass breeding programmes. That may enhance our understanding of the evolutionary biology of this warm season grass species. Their study was initiated to- (1) determine ploidy analysis of *Cynodon* accessions collected from Turkey, (2) investigate associations between ploidy level and diversity, (3) determine whether geographic and polyploid distribution are related to nuclear genome variation, and (4) correlate among four nuclear marker systems for *Cynodon* accessions genetic analysis. They found that increased ploidy levels were associated strongly with higher band frequency ($r = 0.62$). The band frequency of an individual with known ploidy level can be estimated using the formula $Pbf = 0.154 + 0.125x - 0.01x^2$. As Wendel (2000) suggested, this response could be due to gain of new effective maintenance of original structure or function, and silencing point mutations. However, the ploidy levels of *Cynodon* accessions are morphologically indistinguishable, which is also found in case of buffalo grass (Shearman *et al.* 2004). It was suggested by Leitch and Leitch (2008) that increased ploidy levels likely result in increased diversity, which gives plant species significant advantages for adaptation to variable environments.

Polyploidy is often assumed to increase the spread and thus, the invasive success of alien plant species is noticeable, but few empirical studies still exist (Treier *et al.* 2009). They tested this hypothesis with *Centaurea maculosa*, a species native to Europe and introduced into North America approximately 120 years ago, where it became highly invasive. The research workers analyzed the ploidy level of more than 2000 plants from 93 native and 48 invasive *C. maculosa* populations and found a pronounced shift in the relative frequency of diploid and tetraploid cytotypes. Several workers have tried to identify the factors which can make a plant species invasive (Mack *et al.* 2000, Keane and Crawley 2002, Callaway and Ridenour 2004). Since the proportion of polyploids is higher among invasive than native species in some regional floras, few researchers have said that polyploidy may constitute a favourable attribute for the success of allien plants (Verlaque *et al.* 2002, Pandit 2006, Pandit *et al.* 2006). Shifts in cytotype frequency within individual taxa may simply result from stochastic processes operating during range expansion (Kliber and Eckert 2005). Evidence for increased ecological tolerance of polyploids comes from Stebbin's (1971) observation that tetraploids tend to have larger geographic ranges than diploids. Soltis *et al.* (2007) stated that ecological differences between cytotypes are increasingly recognized, even under auto-polyploidy and different ploidy types may thus be considered as distinct biological entities.

Cynodon dactylon has different ploidy levels and thus, Chaves *et al.* (2018) conducted a quantitative and descriptive study of the leaf epidermal structure in different accessions of this grass species, to investigate the consequences of the ploidy level. In their findings, ploidy level increased, with an increase in stomata dimension and a decrease in stomatal density. They found diploid and triploid accessions easily differentiated in relation to polar diameters and equatorial diameters, which indicated it an indirect method for determining the variation in ploidy level. The variables stomatal density and stomatal index were not efficient to discriminate the accessions. Khan *et al.* (2017) mentioned that *C. dactylon* accessions had epidermal cells with sinuous cell walls and that is related to the increase of the surface for higher light uptake (Castro *et al.* 2009). The siliceous cells were classified as saddle like and they are often associated to defense mechanism against herbivory (Massey *et al.* 2006, Huitu *et al.* 2014). *C. dactylon* accessions showed structures similar to salt glands as mentioned by Parthasarathy *et al.* (2015). For the accessions with a higher ploidy level, the differentiation may be fulfilled based on equatorial diameter of adaxial face and the polar diameter of abaxial face. These results indicate gigas effect on the morphometry of stomata, which can be used as an indirect method to determine variations in ploidy level, as previously reported in other plants (Omidbaigi *et al.* 2012, Feng *et al.* 2017). Tufail *et al.* (2017) observed stomata with small dimensions in ecotypes of *C. dactylon*, which could be related to a more efficient physiological regulation since less turgor is required for the opening and closing of the ostiole.

Dhaliwal *et al.* (2018) studied male meiosis of 17 species under 11 genera of the family Poaceae. They reported three grass species as new cytotypes of which *Cynodon dactylon* was found to show the gametic number n = 8. Another five species including *C. dactylon* showed anomalous meiotic behaviour with the presence of cytomixis, chromatin bridges, laggards, secondary associations and inter bivalent connections. However, Gupta *et al.* (2017) made cytological studies on grasses of Haryanna and adjoining Shiwalik hills, the same habitat from where Dhaliwal *et al.* (2018) collected their working materials. In case of *C. dactylon*, three cytological races with n = 8, 9 and 18 were detected from the study area. The diploid cytotype with 2n = 16 is a new chromosome report for the species with normal meiosis, whereas the other cytotypes with 2n = 18 and 36 confirms the previous reports. They also reported meiotic course of the tetraploid cytotype and found the presence of laggards and chromatin bridges. *C. dactylon* showed well developed polyploid (2x, 3x, 4x and 6x) and aneuploid cytotypes.

Cynodon dactylon is characterized by taxonomic and systematic complexity, and polyploidy is one of the factors responsible for its genetic and morphological diversity. Based on this view points, Chaves *et al.* (2019) made a comparative karyotype analysis among cytotypes of *C. dactylon* with the help of fluorescent banding and nuclear DNA content. Nine accessions of this grass species were evaluated and the chromosome number of the accessions ranged from 2n = 2x = 18 to 2n = 5x = 45. Chromosomal polymorphism was also observed based on the distribution and number of heterochromatic bands, with CMA^+ bands located in the pericentromeric position and DAPI bands mainly in the terminal position. Triploid accession (2n = 3x = 27) and tetraploid accession (2n = 4x = 36) had the highest and lowest number of DAPI bands, respectively. The number of CMA^+ bands was stable, as only three pentaploid accessions (2n = 5x = 45) showed variation. There was no direct correlation between an increase in the ploidy level and an increase in the percentage of heterochromatic regions, mainly in relation to A-T rich blocks. The chromosomal banding variation found reinforces the notion of allopolyploidy occurrence in *C. dactylon* and demonstrates the genomic complexity of this species regard to repetitive DNA content. Morphologically, all accessions exhibited predominantly metacentric chromosomes, which agreed with the study by Chiavegatto *et al.* (2016). However, chromosomal polymorphism of *C. dactylon* cytotypes is a possible indicator of allopolyploidy occurrence. Elucidation of chromosomal and genomic organization can assist the classification and taxonomic identification and in understanding the evolutionary events involved during speciation (Chaves *et al.* 2019). Due to economic importance of *Cynodon*, it is essential to understand relationship between species and cytotypes.

However, it can be said after reviewing the literature that polyploidisation is one of the most important mechanism in the evolution of *Cynodon.* As in many other genera, formation of polyploids has significant contribution to diversification of *Cynodon dactylon*. Ploidy level and genome size variation in this grass species can be explored based on taxonomic and comprehensive ecological samplings.

Chapter III
Materials and Methods

3. MATERIALS AND METHODS

Geographical location of Bangladesh in South Asia is between 20°34′ to 26°38′ North latitude and 88°01′ to 92°41′ East longitude. Maximum extension is about 440 km in E-W direction and 760 km in NNW-SSE direction. Under delineation process, Bangladesh has been divided into twelve bio-ecological zones, some of which have been divided into sub-zones. The zones flow from North to South in orderd to make it user- friendly as suggested by Nishat *et al.* (2002). Their works are presented in Fig.1 and Table 2 with certain modifications based on other online information. The map of Bangladesh attached herewith showing 64 districts along with a location of Saint Martin’s Island. (Fig. 2). Bangladesh is a land of deltaic fluvial and alluvial system accomplished a sub-tropical monsoon climate and characterized by variations in temperature, humidity, rainfall, soil type, soil pH etc. Physico-chemical properties of soil have been considered in the present study for justifying the habitat of bermudagrass. However, the above mentioned characteristics are shown in Table 3, which reveals somewhat the climatic conditions of Bangladesh at a glance.

3.1 Plant materials and habitat soils

Among 64 districts of Bangladesh, 23 districts plus one location of Saint Martin’s Island belong to different bio-ecological zones were selected at random as the study area (solid circle in Fig. 2). The plant material bermudagrass (*Cynodon dactylon*) and their habitat soil were collected for conducting the present investigation. All the collected accessions of *C. dactylon* were transplanted in the experimentation of the Institute of Biological Sciences, University of Rajshahi, Bangladesh (Fig. 3). To exonerate the objectives of the present study, different experiments were conducted separately and those are described here experimentwise methodically.

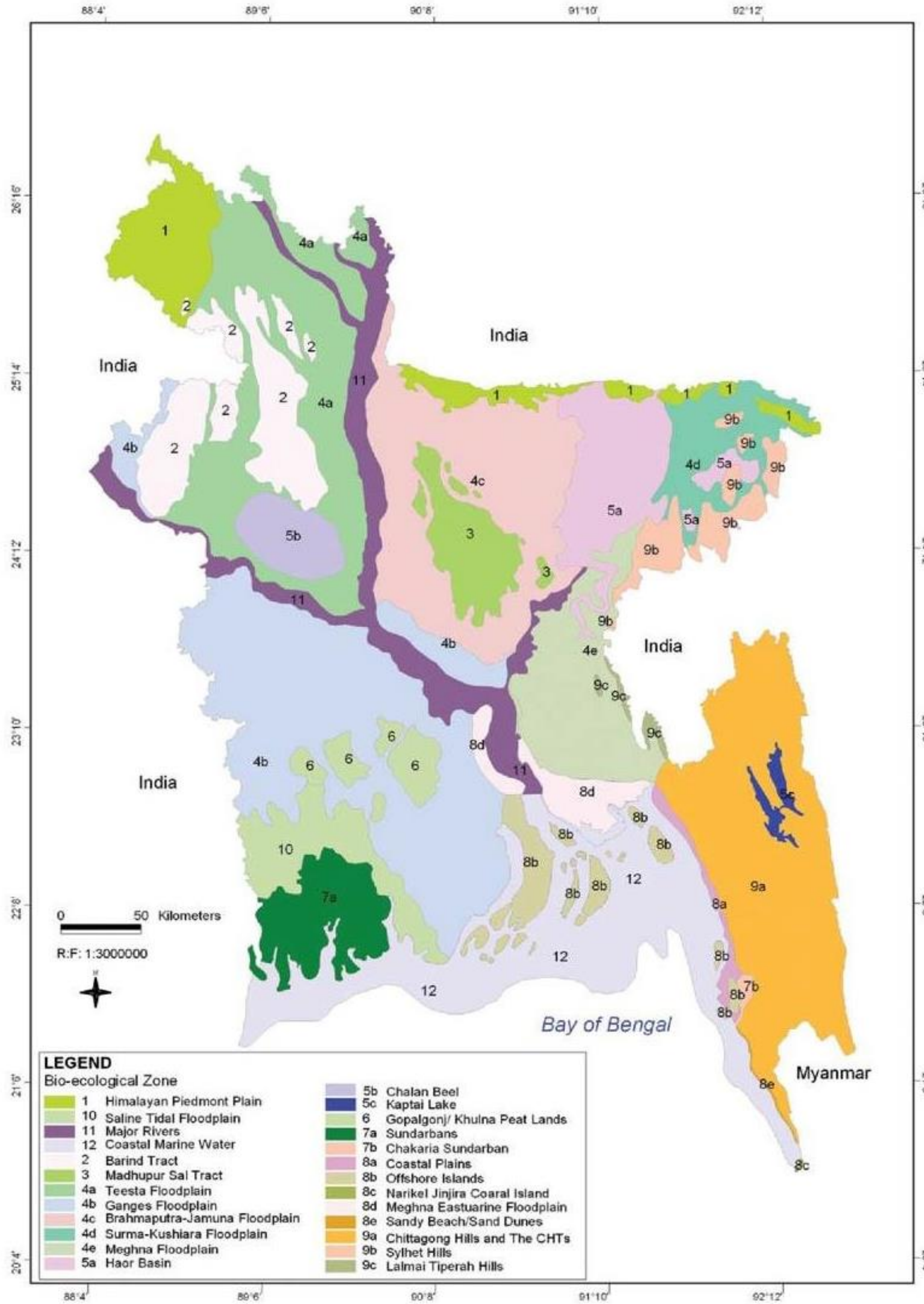


Fig. 1: Map showing bio-ecological zones of Bangladesh (Source: IUCN 2002).

Table 2: Bio-ecological zones of Bangladesh.

Sl. No.	Zones	Locations
1	Himalayan Piedmont Plain	Panchagarh, Thakurgaon, Mymensingh and parts of Netrokona, Sherpur, Sunamganj, Sylhet
2	Barind Tract	Naogaon, parts of Rajshahi, Nawabganj, Joypurhat, Dinajpur, Gaibandha
3	Madhupur Shal Tract	Gazipur, Tangail, Mymensingh, partly Narshingdi
4a	Teesta Floodplain	Nilphamari, Rangpur, Gaibandha, Lalmonirhat, Kurigram
4b	Ganges Floodplain	Jessore, Kushtia, Faridpur, Gopalganj, Barishal, Meherpur, Manikganj, Rajbari, Munshiganj, Magura, Jhenaidah, Chuadanga
4c	Brahmaputra-Jamuna Floodplain	Mymensingh, Dhaka, Jamalpur, Tangail, Pabna, Siranjganj, Narayanganj
4d	Surma-Kushiyara Floodplain	Sylhet Basin, the core haor area
4e	Meghna Floodplain	West of the Madhupur Sal Tract
5a	Haor Basin	Sunamganj, Habiganj, Sylhet, Kishoreganj, Moulvibazar, Netrokona
5b	Chalan Beel	Ullapara, Raiganj, Tarash thanas of Sirajganj district, Singra and Gurudashpur thanas of Natore districts, Chatmohar, Bhanga and Faridpur thanas of Pabna district, Bogura
5c	Kaptai Lake	Forest valleys and arable land in Chittagong, and Bandarban, Khagrachari and Rangamati Chittagong Hill Tracts (CHTs) districs
6	Gopalganj / Khulna Peat Lands	The South of Faridpur region, the adjoining parts of Khulna and Jessore districts, Madaripur, Narail
7a	Sundarbans	Mangrove forest along the Harinbhanga-Raimangal-Kalindi river system in the west and Baleswar river in the east
7b	Chakaria Sundarban	Newly formed grassy islands, river channels, tidal creeks, aquaculture ponds, mangrove forests and inter-tidal mudflats, located in the estuarine system of Matamuhuri river and several smaller rivers in the Bay of Bengal.

Contd...

Sl. No.	Zones	Locations
8a	Coastal Plains	Khulna, Satkhira, Bagerhat, Gopalganj, Shariatpur, Chandpur, Pirojpur, Jhalakathi, Barguna, Barishal, Patuakhali, Bhola, Lakshmipur, Noakhali, Feni,Chittagong, Cox's Bazar
8b	Offshore Islands	Bhola, Hatiya, Ghasiar char, Moulvir char, Shahebanir char, Char bata, Char kukri mukri, Nijhum dweep, Manpura
8c	Narikel Jinjira Coral Island	Saint Martin's Island separated by a channel named the Naaf estuary
8d	Meghna Estuarine Floodplain	Meghna estuary, Gumti canals, situated in the districts of Noakhali and Lakshmipur
8e	Sandy Beach / Sand Dunes	Narrow coastal plains, and banded from the mouth of Bakkhali river, continues south to the Rengaduma khal
9a	Chittagong Hills and the CHTs	Southeastern hill forest of the country, continues uptothree districts of CHTs

Sl. No.	Zones	Locations
9b	Sylhet Hills	The hill ranges peering Dinajpur-Shatgaon, Bhanugach- Raikandi, Haraganj-Singla, Pratapgarh-Duhatia, Sorrispur-Siddeshwar, Bramnbaria
9c	Lalmai-Tipperah Hills	Few miles west of Comilla, Tipperah surface, Eastern low hill ranges (Dupitilla and Dihing formation, Surma and Tipam formation
10	Saline Tidal Floodplain	Satkhira, Khulna, Bagerhat, Jhalakathi, Barguna, Bhola, Noakhali, Feni
11	Major Rivers	Across the delta of four major rivers: the Ganges- Padma, Brahmaputra-Jamuna, Meghna and Teesta
12	Coastal and Marine Waters	The coastal area comprising delta of the Ganges-Brahmaputra-Meghna river system flowing through the country on its way to Bay of Bengal subjected to coastal dynamics by river flow and tidal wind actions, particularly the coastal part of Khulna, Barshal, Noakhali, Chittagong, Cox's Bazar

Fig. 2: Map of Bangladesh showing the study area, where solid circle indicates the particular habitat considered in the present study.

Table 3: Climate and soil characteristics in 64 districts plus one location of Bangladesh.

Sl. No.	Locations/ Habitats	Climatic conditions				Soil characteristics	
		Annual average temperature (°C)		Average relative humidity (%)	Annual average rainfall (mm)	General soil types	Soil pH
		Minimum	Maximum				
1	Panchagarh*	10.10	30.20	67	2931	Non-calcareous Alluvium soil	6.6
2	Thakurgaon*	10.50	33.50	66	2536	Non-calcareous soil	6.5
3	Nilphamari	11.20	32.30	67	2931	Non-calcareous Alluvium soil	5.4
4	Lalmonirhat*	11.20	32.30	65	2931	Non-calcareous Alluvium soil	5.4
5	Kurigram	20.15	29.74	67	1425	Non-calcareous Alluvium soil	5.2
6	Dinajpur*	10.50	33.50	64	2536	Non-calcareous Alluvium soil	7.1
7	Rangpur*	21.60	30.33	65	1593	Non-calcareous Alluvium soil	7.2
8	Gaibandha*	26.00	36.00	66	1037	Non-calcareous Alluvium soil	6.5
9	Joypurhat	11.20	35.00	57	1576	Non-calcareous soil	5.8
10	Sherpur*	12.00	33.30	76	2174	Non-calcareous & calcareous soil	6.8
11	Naogaon*	21.64	31.06	54	1132	Non-calcareous Alluvium & calcareous soil	7.0
12	Bogura	20.30	30.30	57	1762	Non-calcareous Alluvium & calcareous soil	6.2
13	Jamalpur	26.00	36.00	67	1174	Non-calcareous	6.3
14	Netrokona	12.27	33.30	67	2174	Non-calcareous & peat soil	6.5
15	Sunamganj	13.60	33.20	66	3334	Non-calcareous	5.2

Contd...

16	Sylhet	21.85	31.37	74	3244	Non-calcareous & peat soil	5.3
17	Nawabganj	11.20	37.80	65	1862	Non-calcareous, Calcareous, Grey & Clay soil	7.8
18	Sirajganj	11.90	34.60	71	1610	Non-calcareous Alluvium, Grey & Clay soil	6.4
19	Mymensingh*	20.60	29.90	57	2249	Non-calcareous & Grey soil	7.8
20	Kishoreganj	21.55	30.34	63	1289	Non-calcareous	7.7
21	Habiganj	25.00	34.00	77	2135	Non-calcareous, Grey & peat soil	5.4
22	Maulvibazar	21.16	30.66	70	1641	Non-calcareous & peat soil	5.5
23	Rajshahi*	19.98	31.06	61	1211	Non-calcareous, Calcareous & Clay soil	8.1
24	Natore	11.20	37.80	66	1862	Non-calcareous & calcareous soil	6.5
25	Tangail	20.89	31.14	73	1872	Non-calcareous, Terrace & Grey soil	6.2
26	Pabna*	9.60	36.80	75	1872	Non-calcareous Alluvium, Calcareous & Clay soil	7.8
27	Gazipur*	26.00	34.00	67	1474	Non-calcareous, Terrace & Clay soil	6.4
28	Narsingdi*	12.70	36.00	75	1620	Non-calcareous, Terrace & Clay soil	8.5
29	Brahmanbaria	10.00	32.00	66	1650	Non-calcareous Alluvium, Terrace, Grey Pediment & Clay soil	6.7
30	Meherpur	11.20	37.10	67	1857	Non-calcareous & calcareous soil	7.5
31	Kushtia	11.20	37.80	67	1467	Non-calcareous & calcareous soil	8.0

Contd...

32	Manikganj	12.70	36.00	73	2376	Non-calcareous & calcareous soil	5.9
33	Dhaka	11.50	34.50	74	1931	Non-calcareous, calcareous, peat, clay& Terrace soil	6.0
34	Rajbari	12.60	35.80	67	2105	Calcareous soil	7.7
35	Narayanganj	12.70	36.00	65	2376	Non-calcareous	5.9
36	Chuadanga	21.20	31.98	56	1246	Non-calcareous & calcareous soil	7.6
37	Jhenaidah*	11.20	37.10	74	1467	Non-calcareous & calcareous soil	8.0
38	Faridpur*	21.18	39.00	67	1263	Non-calcareous & calcareous soil	7.9
39	Munshiganj	12.70	36.00	74	2376	Non-calcareous & calcareous soil	5.1
40	Magura	20.30	30.70	56	1681	Calcareous soil	5.2
41	Comilla	21.00	30.10	68	2295	Non-calcareous, calcareous & Clay soil	5.4
42	Shariatpur*	10.00	32.00	63	2293	Non-calcareous & calcareous soil	8.4
43	Jessore*	15.40	34.60	57	1537	Non-calcareous	8.2
44	Madaripur	21.20	31.43	74	1659	Non-calcareous & calcareous soil	7.6
45	Chandpur	12.70	34.30	65	2551	Non-calcareous Alluvium & calcareous soil	5.3
46	Narail	11.20	37.10	74	1467	Calcareous & Peat soil	8.0
47	Gopalganj	21.00	30.70	67	1809	Non-calcareous, calcareous, peat soil	7.8
48	Lakshmipur	10.00	32.00	67	2753	Non-calcareous & calcareous soil	7.1
49	Feni	14.40	34.30	74	3302	Non-calcareous, calcareous & Brown Hill soil	6.5

Contd...

50	Khulna*	21.60	31.10	76	1809	Non-calcareous, calcareous, peat soil	7.5
51	Barisal	21.30	30.70	84	2070	Non-calcareous & calcareous soil	7.1
52	Noakhali	14.40	34.30	77	3302	Non-calcareous & calcareous soil	8.0
53	Satkhira	12.50	35.50	71	1710	Non-calcareous & calcareous soil	7.8
54	Pirojpur	12.50	35.50	74	1710	Non-calcareous & calcareous soil	7.5
55	Bagerhat	12.50	33.50	83	1710	Non-calcareous, calcareous & peat soil	7.1
56	Jhalokati	12.10	33.30	70	2506	Non-calcareous & calcareous soil	7.1
57	Bhola	22.05	31.28	75	2192	Non-calcareous	6.4
58	Patuakhali	12.10	33.30	74	2506	Non-calcareous	7.2
59	Barguna*	12.10	33.30	70	2506	Non-calcareous & calcareous soil	7.8
60	Khagrachari*	13.00	34.60	45	3031	Non-calcareous & Grey Piedmont soil	5.1
61	Rangamati*	21.40	30.20	79	2549	Grey Piedmont & Hill soil	6.7
62	Chittagong	21.70	30.40	79	2890	Non-calcareous Alluvium, calcareous, Grey Piedmont, clay, hill & peat soil	5.3
63	Bandarban*	14.00	33.00	66	2925	Grey Piedmont & Hill soil	6.7
64	Cox's Bazar*	16.10	34.80	80	3524	Non-calcareous Alluvium, calcareous & Hill soil	4.9
65	St. Martin's Island*	22.44	30.85	77	3432	Calcareous Alluvium soil	7.3

* = Habitat considered in present study. Sources: Internet, Books, Journals (2017-2020)

3.2 Morphological characteristics and physico-chemical properties of soil

3.2.1 Study sites

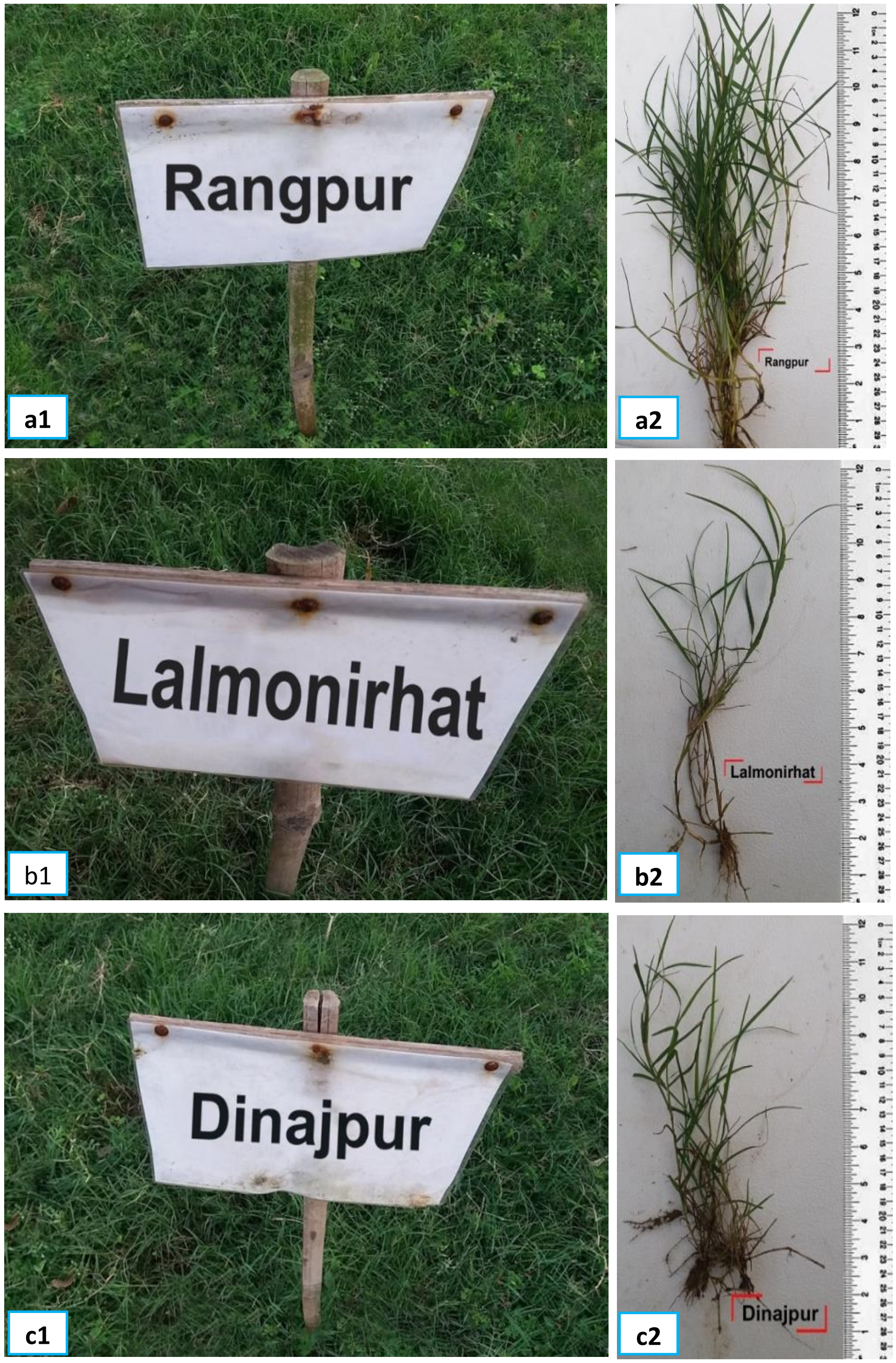
A total of 24 accessions of *Cynodon dactylon* (L.) Pers. were collected from different ecological habitats in Bangladesh. Selections were based on environmental conditions of the collection site. The selective habitats / locations are presented in Table 4.

Table 4: Twentyfour habitats / locations of the accessions of *Cynodon dactylon* in Bangladesh.

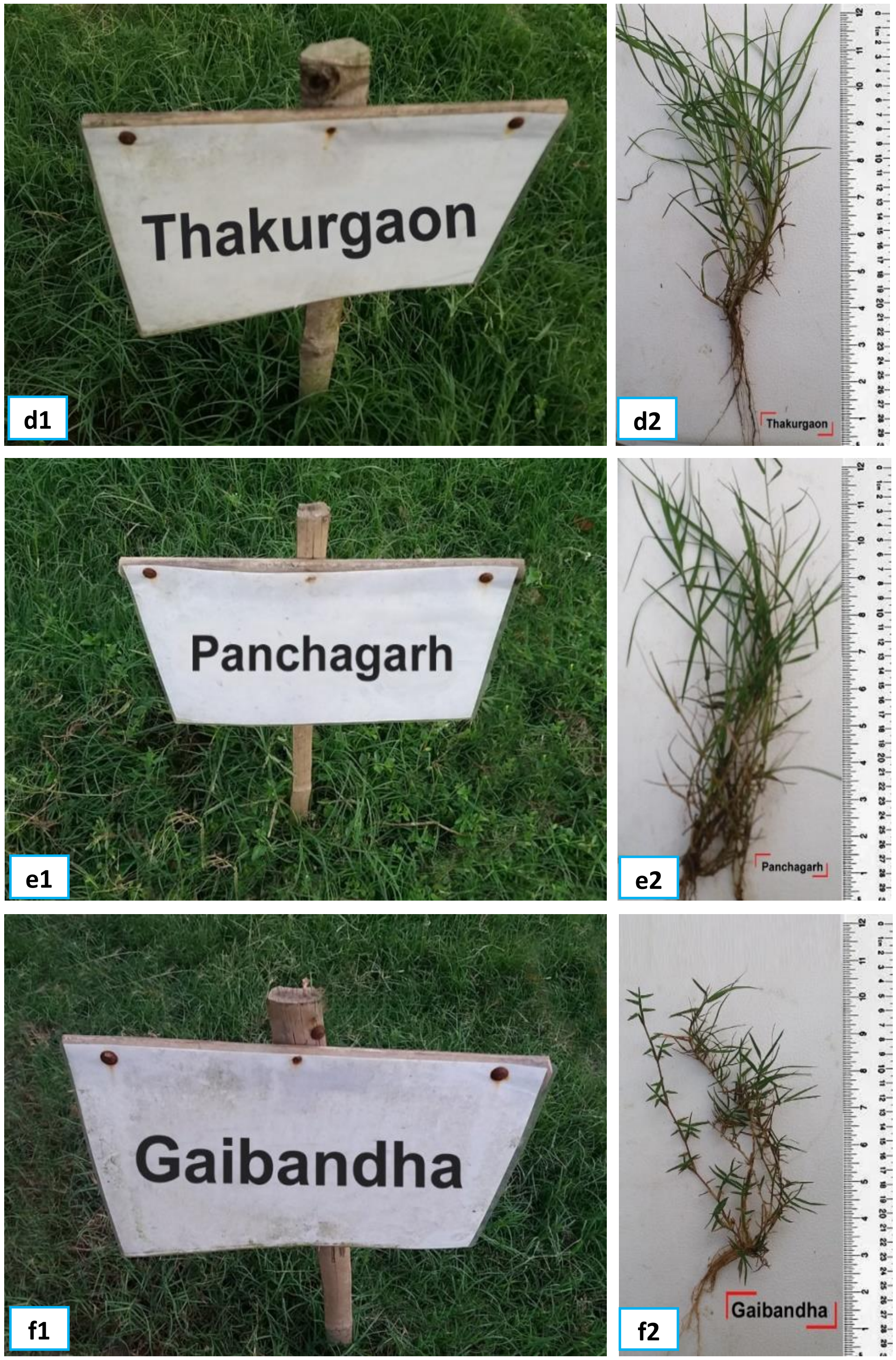
Serial No.	Habitats / locations	Serial No.	Habitats / locations
1	Rangpur	13	Mymensingh
2	Lalmonirhat	14	Khulna
3	Dinajpur	15	Jessore
4	Thakurgaon	16	Jhenaidah
5	Panchagarh	17	Faridpur
6	Gaibandha	18	Shariatpur
7	Rajshahi	19	Barguna
8	Naogaon	20	Khagrachari
9	Pabna	21	Bandarban
10	Gazipur	22	Rangamati
11	Narsingdi	23	Cox's Bazar
12	Sherpur	24	Saint Martin’s Island

The study was carried out in at the research field of Institute of Biological Sciences of Rajshahi University, Bangladesh. The experiment was laid out in a randomized complete block design (RCBD) with three replication (Gomez and Gomez 1984). The site has an annual average rainfall of 661.2 mm of which the majority falls during the main rainy season (July-September). No fertilizers were used in the experimentation field. The soil of the research field is loamy. All (24) the accessions of *C. dactylon* were transplanted from root splits in 1.5 m × 1.4 m plots, collected from 24 habitats. Data were recorded on 26 characters. The characters were selected based on their agronomic/taxonomic relevance and expected variation among accessions. The morphological characters were observed within the first 4 months after cutting before most accessions had started flowering. Owing to the rhizomatous and stoloniferous nature of *Cynodon dactylon* it was difficult to identify individual plants. So, observations were made randomly within the plot. Six randomly selected areas of 25 cm × 25 cm in the plot were chosen for rhizome observations and six randomly selected areas of 200 cm × 50 cm at the end of the plot for stolon observations. Plants were removed from three areas at the time of cutting and after 7 weeks the numbers of rhizomes or stolon in the selected areas were counted. The collected ecotypes were grown under same environmental conditions in research field of the Institute of Biological Sciences of Rajshahi University, till their acclimatization to evaluate genetically fixed characteristics. Photographs of all the twentyfour accessions of *C. dactylon* just after uprooting from the experimentation field are shown in Figs. 3 - 4 (a1 - x1 & a2 - x2). Plants were uprooted carefully and washed with distilled water. For dry weight, plant samples were oven-dried at 65°C for 48 hours and constant dry weight was achieved. Data were recorded of the following plant parts and their mean values were tabulated.

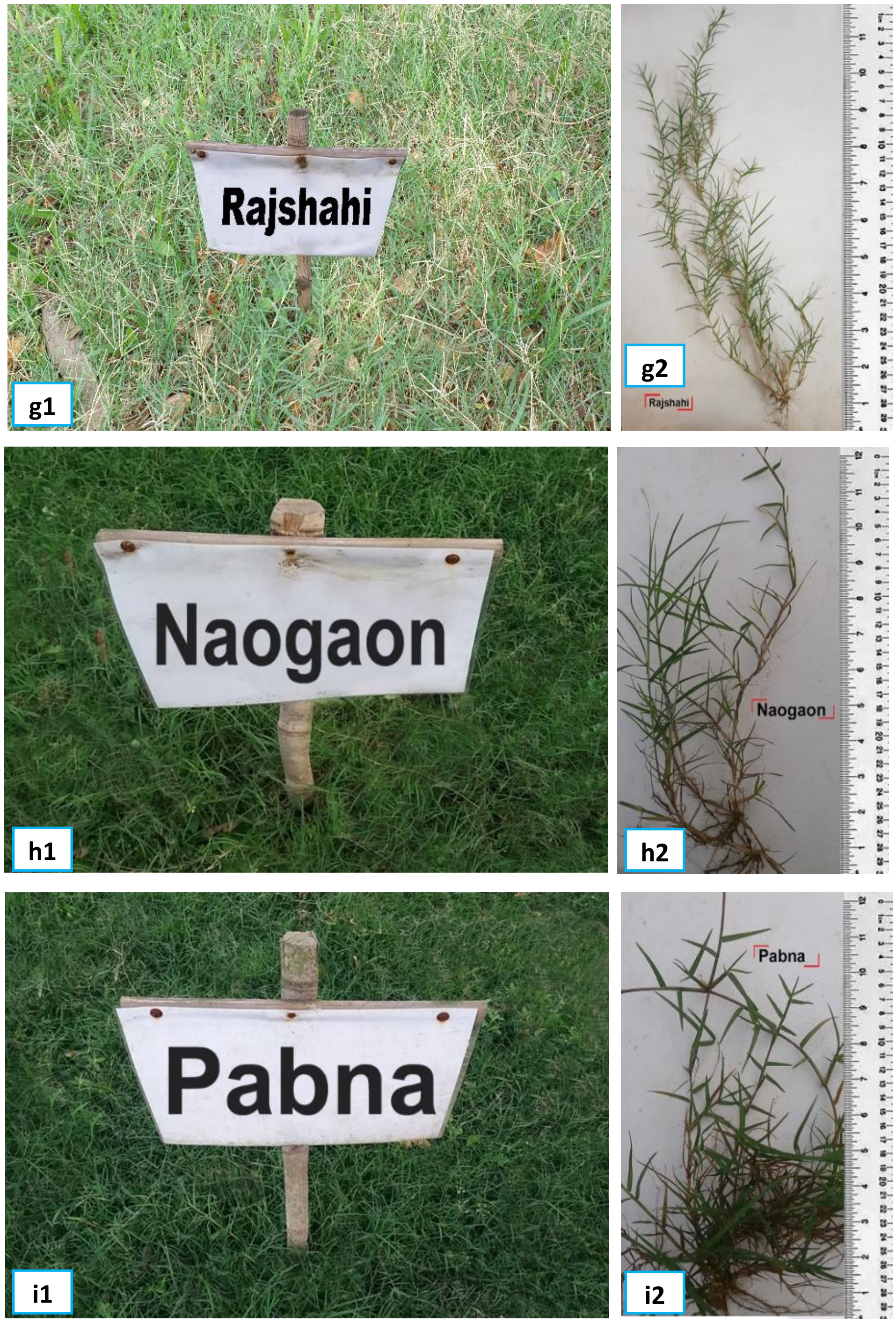
Fig. 3: Transplanted accessions of *Cynodon dactylon* in experimentation field of the Institute of Biological Sciences, University of Rajshahi, Bangladesh.



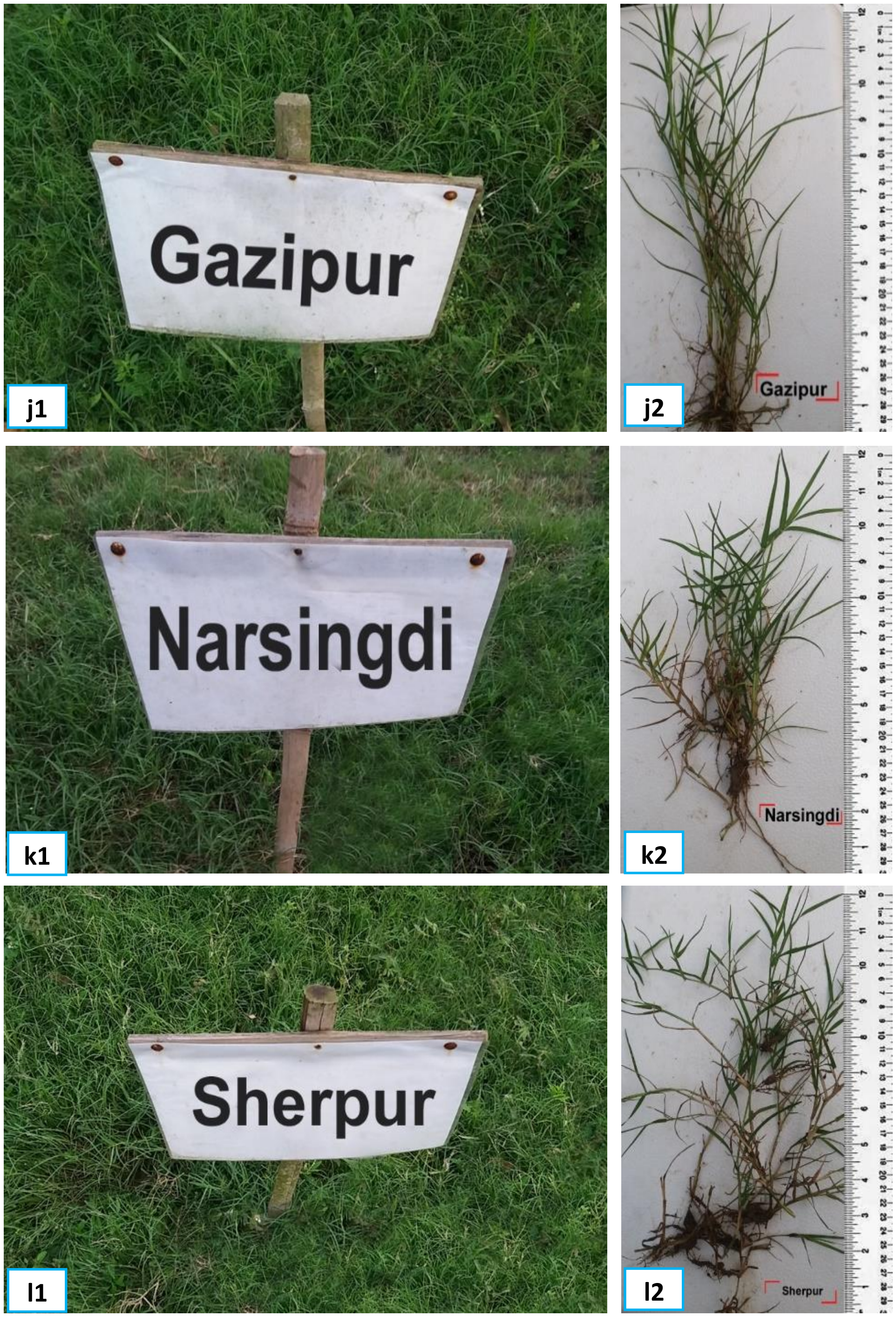
Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **a1 & a2** = Rangpur, **b1 & b2** = Lalmonirhat, **c1 & c2** = Dinajpur.



Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **d1 & d2** = Thakurgaon, **e1 & e2** = Panchagargt, **f1 & f2** = Gaibandha.

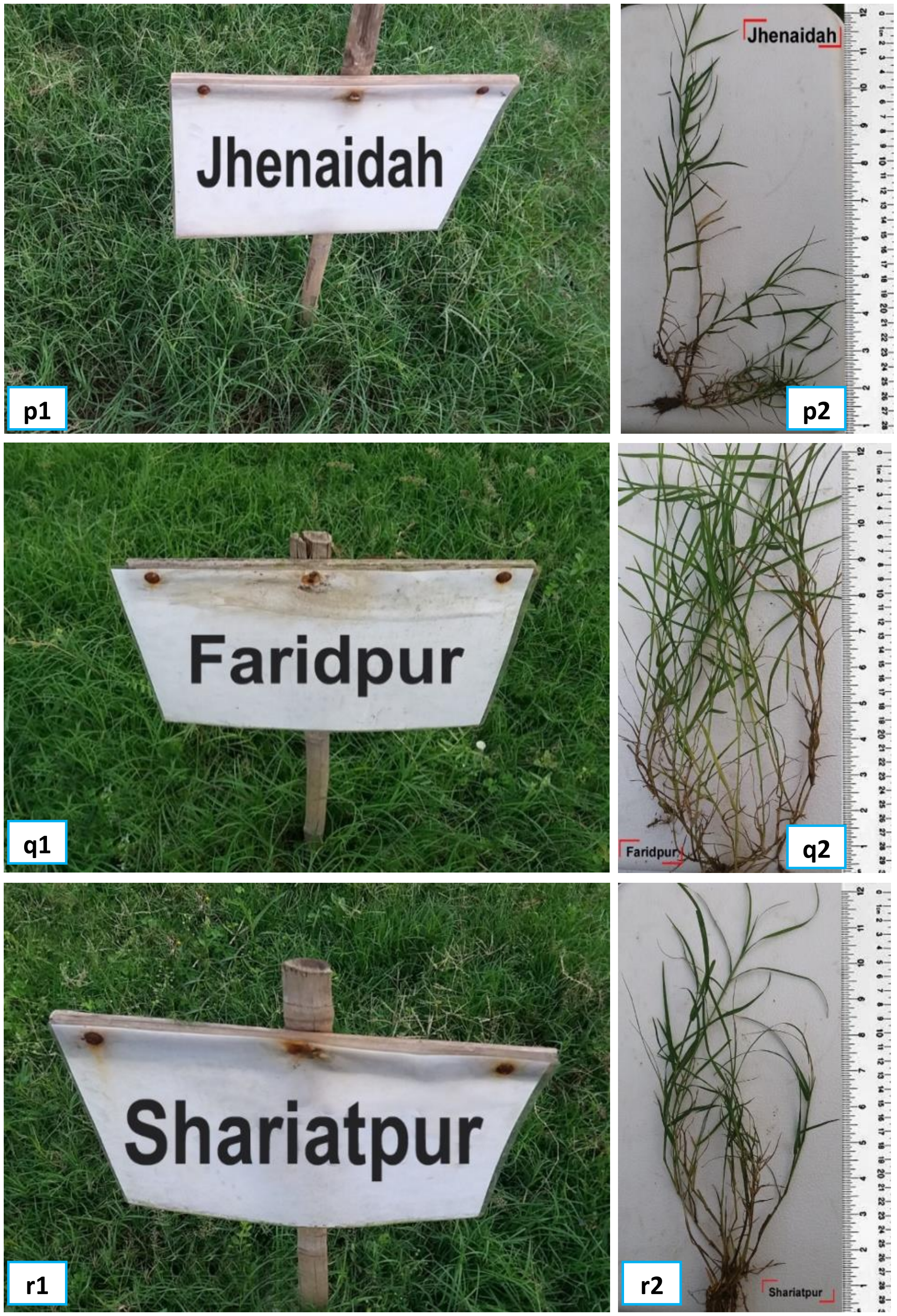


Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **g1 & g2** = Rajshahi, **h1 & h2** = Naogaon, **i1 & i2** = Pabna.

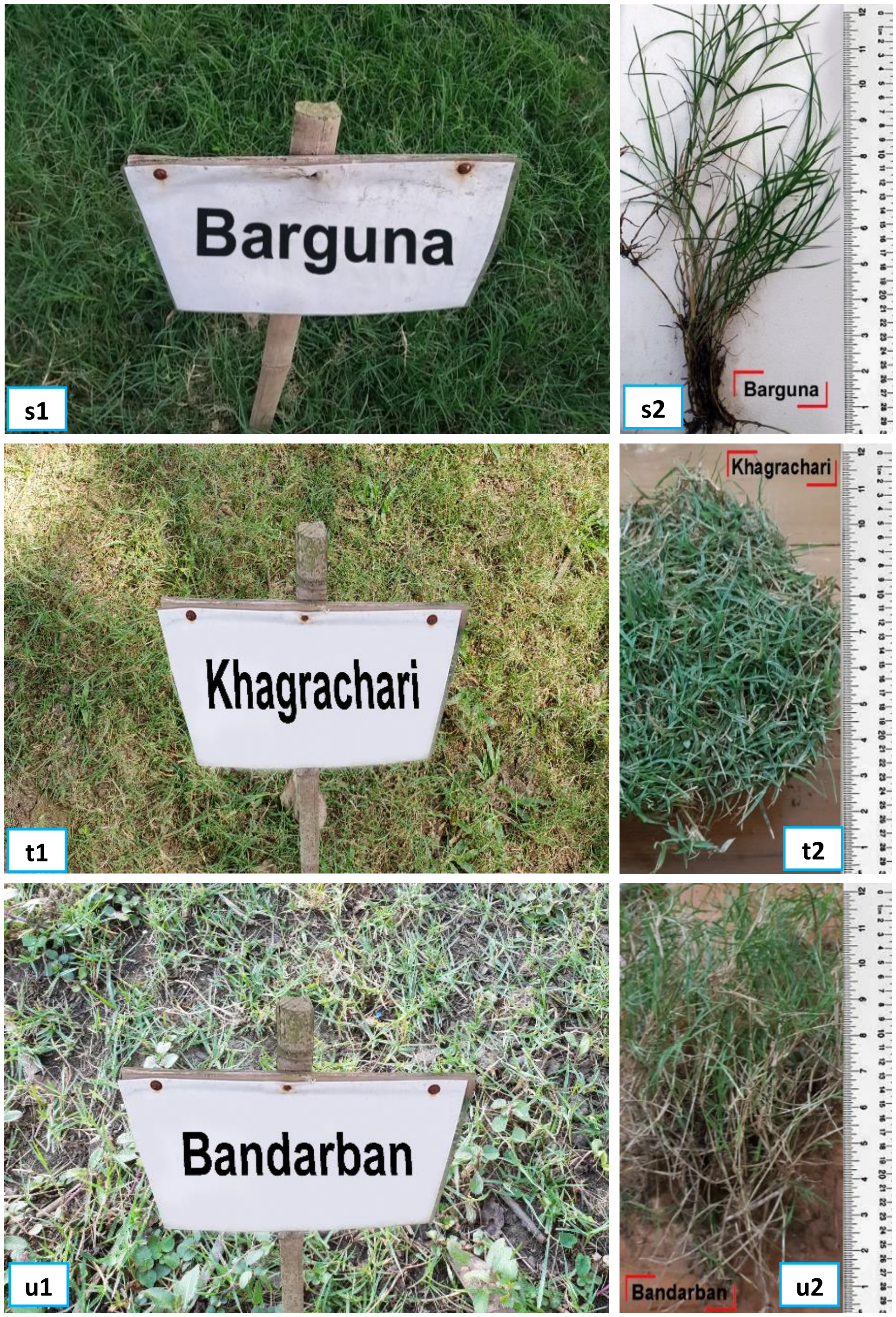


Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **j1 & j2** = Gazipur, **k1 & k2** = Narsingdi, **l1 & l2** = Sherpur.

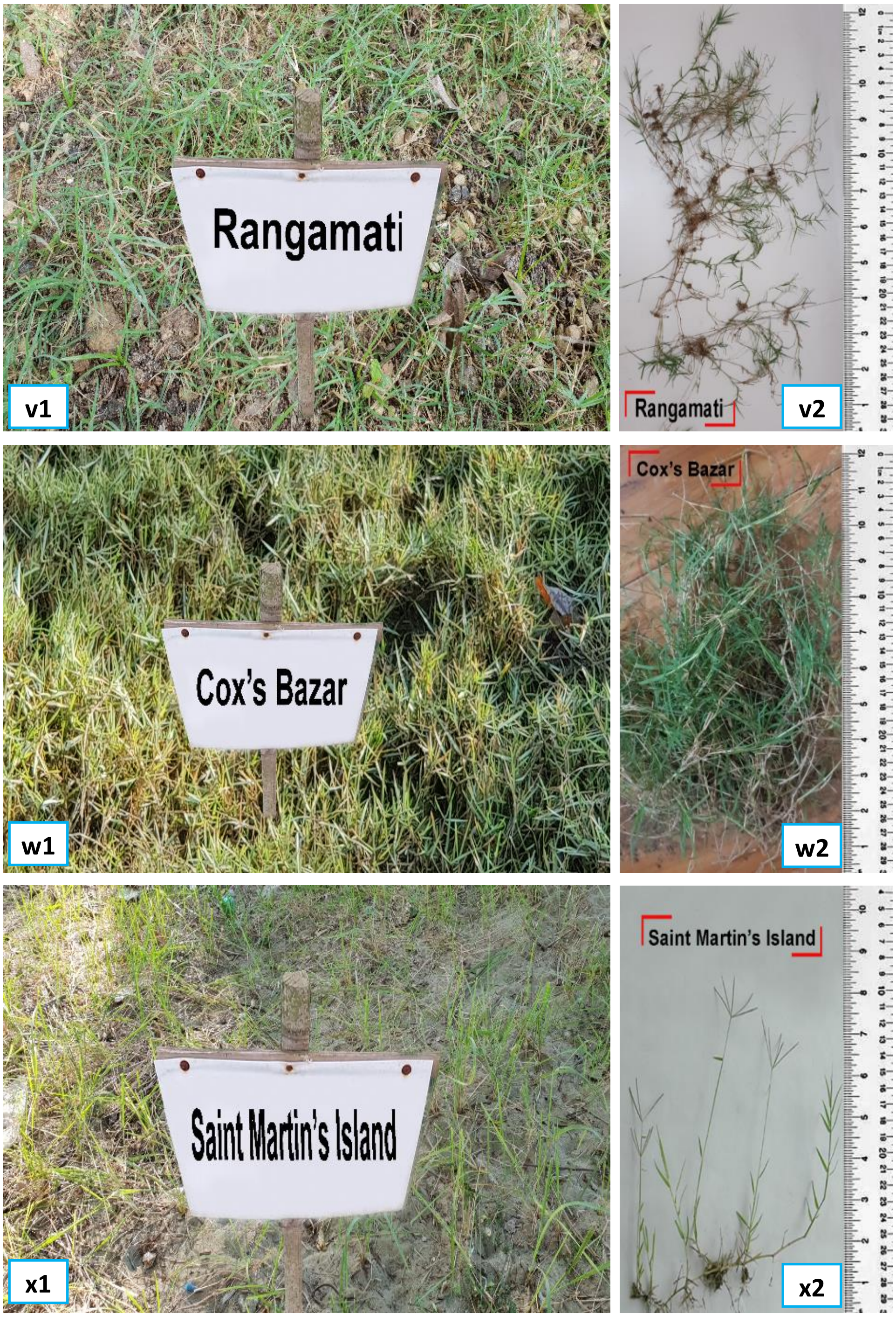
Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **m1 & m2** = Mymensingh, **n1 & n2** = Khulna, **o1 & o2** = Jessore.



Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **p1 & p2** = Jhenaidah, **q1 & q2** = Faridpur, **r1 & r2** = Shariatpur.



Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **s1 & s2** = Barguna, **t1 & t2** = Khagrachari, **u1 & u2** = Bandarban.



Figs. 4 (a1 - x1 & a2 - x2): Growing *Cynodon dactylon* after transplantation in experimentation field collected from different habitats and morphology of more than one respective accession of this grass species. **v1 & v2** = Rangamati, **w1 & w2** = Cox's Bazar, **x1 & x2** = Saint Martin's Island.

3.2.2 Quantitative characters

Twentysix different quantitative characters of each accession considered for recording from both vegetative and reproductive parts.

1. Number of shoots/plant
2. Shoot length (cm)
3. Number of nodes/plant
4. Internode length (cm)
5. Root length
6. Number of roots/plant (cm)
7. Number of leaves/plant
8. Single leaf length (cm)
9. Single leaf width (mm)
10. Total leaf area/plant (cm^2)
11. Number of rhizomes/plant
12. Shoot fresh weight/plant (g)
13. Root fresh weight/plant (g)
14. Total leaves fresh weight/plant (g)
15. Shoot dry weight/plant (g)
16. Root dry weight/plant (g)
17. Total leaves dry weight/plant (g)
18. Inflorescence number/plant
19. Length of racemes (cm)
20. No. of racemes/plant
21. Seed number/shoot
22. Seed number/plant
23. Maximum height (cm)
24. Minimum height (cm)
25. Number of stolons
26. Stolon length

For determination of the leaf area single segment of leaf was cut off with the help of a sharp knife, weighted after oven drying and then it was calculated by using the following formula:

$$\text{Leaf area} = \frac{\text{Area of segment} \times \text{weight of leaves}}{\text{Weight of segment}}$$

3.2.3 Analysis of soil samples

Soils from rhizosphere at 16 cm depth taken from each habitat were to analyze the physico-chemical characteristics. The soil extract was used to determine the pH, organic matters %, potassium(K^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), total nitrogen, phosphorus (P), sulfur (S), boron(B), copper (Cu), iron (Fe), manganese (Mn) and zink (Zn) of soil. The collected soils were air-dried for 7 days and then sieved using a 2 mm mesh to remove debris. Air dried and sieved soils (250 g) of each habitat were used for soil analysis. Soil pH was measured by HANNA HI 92210N ATC pH meter. Total organic matter was determined following the wet oxidation method as described by Walkley and Black (1934) and the micro Kjeldahl procedure was used for the determination of total nitrogen (Bremnerv and Mulvaney 1982). Available phosphorus, sulfur and boron were measured by using spectrophotometer at 890 nm. The values for Magnesium, Calcium, Manganese, Copper, Iron and Zinc were obtained using atomic absorption spectrophotometer (Perkin-Elmer Model 403, Shelton, Connecticut, USA). Available Potassium was measured by using a flame photometer. Phosphorus was determined following the Olsen method. Neutral, acidic and alkaline, and the level of organic matter were estimated as follows:

pH = 6.6 – 7.3 (Neutral)

6.5> (Acidic)

7.4< (Alkaline)

Organic matter (OM) = below 1- Very low

1-1.7 [Low]

1.8-3.4 [Medium]

3.4< [High]

A change of pH and oxidation state can increase or decrease the potential bioavailability of metals in soil. Metals such as Zn, Cu, and Mn are essential for living organisms at low concentrations, but became toxic due to increasing concentrations.

3.2.4 Statistical analysis

A statistical comparison of means of different accessions and their morphological quantitative characters were carried out using analysis of variance (ANOVA) and by Duncan's Multiple Range Test (DMRT). Significance level was set at $p< 0.05$. The data analysis was done by using SPSS version 20.0 for Windows. Graphs were drawn by Excel software.

3.3 Leaf epidermal anatomy in relation to ecotypic adaptation

3.3.1 Materials

Leaves, sharp blade, soft camel hair brush, forceps, petri dishess, glass slides, cover slips, needles, forceps, safranin, glycerin, distilled water, blotting paper, light microscope, ocular meter, stage micrometer etc.

3.3.2 The features considered for this study were as follows

Stomata type, shape of subsidiary cells and silica bodies, types of long cell margins, presence of macro hair, micro hair, prickle angular and hooks, number of epidermal cells, long cells, silica bodies, prickles angular, hooks and macro hair, length of long cell, stomata and epidermal cells, width of long cells, breadth of stomata and epidermal cells, stomatal frequency and stomata index on both abaxial and adaxial surface of the leaves of *Cynodon dactylon*.

3.3.3 Calibration of ocular and stage micrometer

i) Stage micrometer (SM) was a calibrated having 100 division of 1 mm and therefore, 1 division is equal to 0.01 mm or 10 µm. The SM was placed on the stage of the microscope and focused it at given magnification e. g. 40x.

ii) The ocular micrometer (OM) was placed in one of the eye pieces of microscope. While observing through the eye pieces, the scale of OM was aligned with that of SM.

iii) The divisions of OM coinciding with divisions of SM were counted.

For example: 8.0 divisions of OM = 1 divisions of SM i.e., 1.0 µm

So, 1 division of OM = 0.125 µm

iv) Following the above three steps, the ocular scale was calibrated for magnification as well i.e., 150X

v) Once the ocular scale was calibrated for required magnification, SM was removed and slide having sample material was placed for observations and measurements.

3.3.4 Preparations of slides

Leaves of *Cynodon dactylon* were collected from the study area and washed them gently with running water to remove the dust and debris. The foliar epidermal peels were taken from the middle of both surfaces of mature leaves. The specimens were irrigated with water holding it downwards from the end. The epidermis above the desired surfaces was scrapped carefully with a sharp razor blade. Loose cells were washed away from the epidermal peel with the aid of soft camel hair brush. The uniform peels were taken on glass slides and a drop of 1% safranin stain was added, kept for 1 minute, washed with water and then mounted in a drop of 10% glycerin to prevent the drying. A cover-slip was placed and excess of water was blotted with the help of botting paper. Both abaxial and adaxial sides of leaves were prepared and observed under (15 × 10) X and phase contrast microscope for detailed analysis and obtaining better pictures as well as measuring the length and breadth of stomata including guard cells. When the abaxial epidermis was prepared, the leaf was placed on a tile adaxial surface upper most. When a preparation of the adaxial epidermis was to be made, the leaf was placed with abaxial side uppermost. The measurements were obtained from analysis of ten leaves (five for each epidermal face) with five fields of view and five stomata/field randomly in Laboratory of Ecology, University of Rajshahi, Bangladesh.

3.3.5 Determinations of stomatal frequency

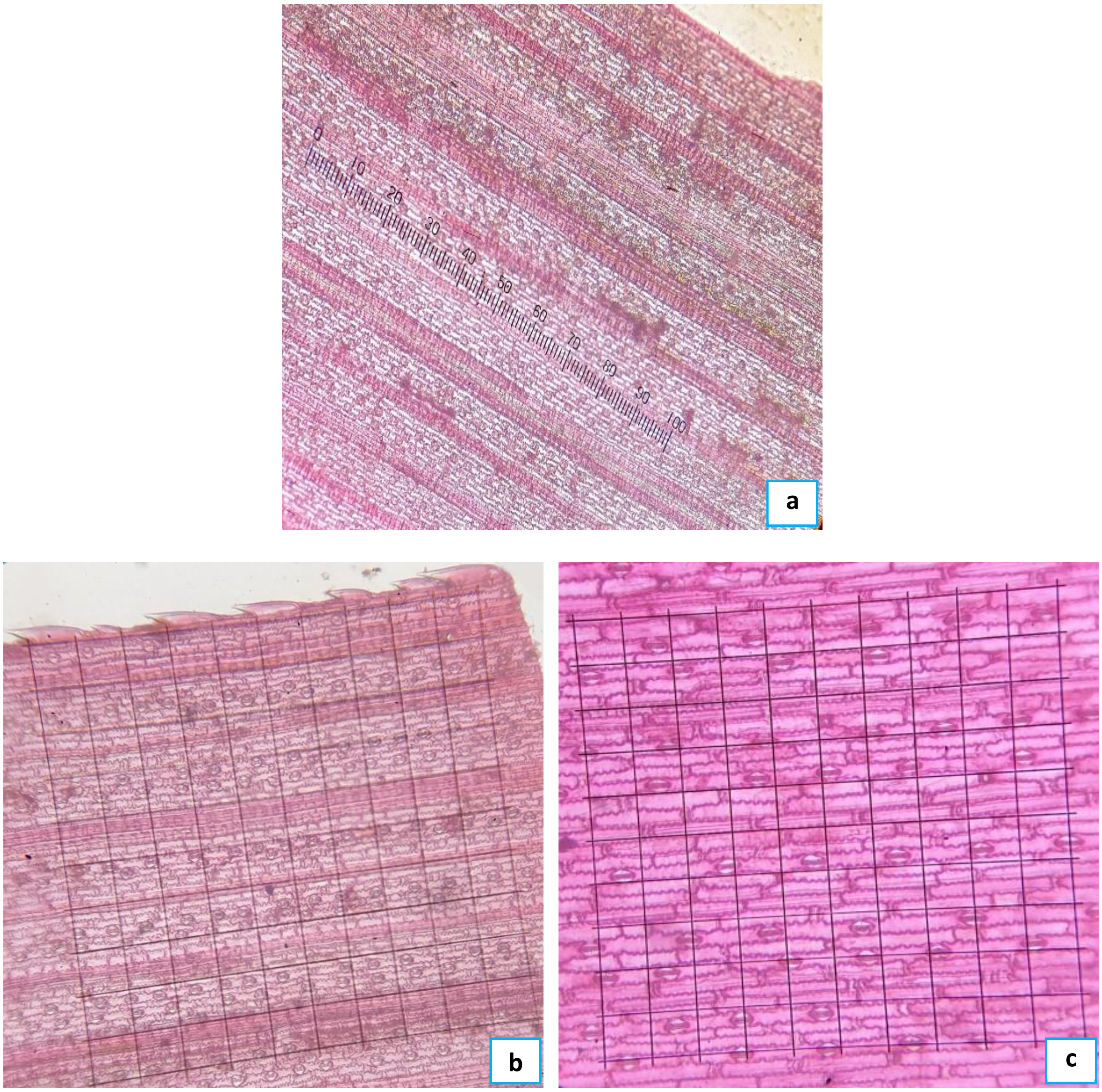
Observations for number of stomata present in microscopic view field was made for recording and for calculating the stomatal frequency and thereafter, expressed in terms of stomata/mm^2. The diameter of view field was calculated by ocular scale (Fig. 5a). At a given magnification the total number of stomata was counted as visible by square grid scale under microscope (Figs. 5b & 5c). The square grid was composed of 100 identical small squares.

3.3.6 Determination of stomatal index

The stomatal index (SI) was calculated using the formula SI = (S/S+E) × 100 where, S and E are the number of stomata per unit area and number of epidermal cells per unit area, respectively in microscopic view field and the values were expressed in percentage (%).

3.3.7 Length and breadth of long cells, stomatal cells and epidermal cells

Morphometric measurements for different cells (long cells, stomatal cells, epidermal cells) were taken under suitable magnification by using calibrated ocular micrometer (Fig. 5a).



Figs. 5 (a-c): Estimation of leaf epidermal characters using a) ocular sacle and b & c) square grid scale under compound microscope.

3.3.8 Microscopic analysis

The stained peels were examined under a compound microscope (SWIFT Instruments International, S.A No.760090). The best micro-photographs of the mounted materials were taken using a digital camera (model: C-B5, Brand: Optica) fitted on the electrical light microscope (Model: XSZ-107T, Brand: Novel) with total magnification of 15 × 10X in Laboratory of Phycology and Limnology, University of Rajshahi, Bangladesh. These micro-photographs were useful for identification and differentiation of epidermal cells on the basis of microscopic features.

3.3.9 Statistical analysis

A statistical comparison of means of different accessions and leaf epidermal quantitative characters was carried out by analysis of variance (ANOVA) and by Duncan's Multiple Range Test (DMRT). Significance level was set at $p < 0.05$. The data analysis was done using SPSS version 20.0 for Windows. Graphs were drawn by Microsoft Office Excel software.

3.4 Study of root and stem anatomy

3.4.1 Materials

Roots and stems of 24 accessions of *Cynodon dactylon,* sharp blade, soft camel hair brush, forceps, petri dishes, glass slides, cover slips, needles, forceps, safranin, glycerin, FAA solution, acetic-alcohol solution, fast green, watch glass, dropper, distilled waters, blotting paper, tissue paper, light microscope, ocular meter, stage micrometer etc.

3.4.2 The features considered for the study of root and stem were as follows

The anatomical characteristics recorded for stems were epidermis thickness, cortex thickness, sclerenchyma thickness, vascular bundle area, vascular bundle number, metaxylem area and phloem area. The anatomical characteristics recorded for roots were epidermis thickness, cortex thickness, sclerenchyma thickness, endodermis thickness, metaxylem area, phloem area, pith thickness and pith cell area.

3.4.3 Preparations of slides

Plants *Cynodon dactylon* were removed from the soil, their stems and roots were separated and washed with distilled water to remove soil dust and other contaminants. For anatomical study, free hand transverse sections were cut and thin sections were separated with the help of research microscope. Transverse sections of stem and root of 10 individual plants per accessions were made using sharp blade from the fresh materials. For root, some potato blocks were used. The root pieces were put inside the block then free hand transverse sections were cut and at least three thin sections were selected for each root with the help of microscope. The materials were fixed in FAA solution (formalin 10%, acetic acid 5%, ethyl alcohol 50% and distilled water 35%) for 48 h and then transferred to acetic-alcohol solution (1:3) ratio for root and stem anatomical studies. Double staining dehydration procedure (safranin and fast green) was used for the preparation of permanent slides (Ruzin 1999) to study various cells and tissues of root and stem in Laboratory of Ecology, University of Rajshahi. The stained sections were observed under a light microscope at 15 × 10X magnification and photographed. The anatomical characters observed under microscopes were recorded. Measurements were taken with a light microscope, using an ocular micrometer.

3.4.4 Microscopic analysis

The stained transverse sections of root and stem were examined under a compound microscope (SWIFT Instruments International, S.A No.760090). The best photo micrographs of the mounted materials were taken using a digital camera (model: C-B5, Brand: Optica) fitted on the electrical light microscope (Model: XSZ-107T, Brand: Novel) with total magnification of 15 × 10X in Laboratory of Phycology and Limnology, University of Rajshahi, Bangladesh. These photomicrographs were useful for identification and differentiation of various cells and tissues of root and stem on the basis of microscopic features.

3.4.5 Statistical analysis

A statistical comparison by means of Duncan's Multiple Range Test (DMRT) was made. For root and stem's quantitative anatomical characters of different accessions was made statistically by analysis of variance (ANOVA). Significance level was set at $p \leq 0.05$. The data analysis was done using SPSS version 20.0 for Windows. Graphs were drawn by Microsoft Office Excel software.

3.5 Nuclear phenotype and chromosome count

Cynodon dactylon accessions of 24 habitats were used as experimental material in the present study and from at least ten plants of each accession root tips were collected from the experimentation field of the Institute of Biological Science, University of Rajshahi, Bangladesh. When these were grown well in the experimental field, their root tips (approximately 1-1.5 cm in length) were treated with saturated solution of paradichlorobenzene at 10°C in the refrigerator for 4-4.5 hrs. Then the root tips were washed with distilled water and fixed in 1:3 aceto-alchohol for 48 hours at room temperature. After fixation they were washed again with distilled water and preserved in 70% ethanol and kept in the refrigerator till used.

For cytological study the root tip cells were stained with 0.5% haematoxylin according to the method of Haque *et al.* (1976). Stained meristematic zone was taken on a clean slide, squashed with a drop of 0.5% acetocarmine and a gentle pressure was applied by thumb over the cover glass. For chromosome counting, photomicrography was made from metaphase plate by using 40 × 10 magnification and at the same time for clear visualization camera lucida drawings were made.

For measuring Nuclear Volume (NV) from interphase ocular micrometer was used and the thereafter values were converted in micron (µ) with the help of stage micrometer, using the formula, $NV = 4/3\ \pi r^3$ (Nayar *et al.* 1971). Nuclear volume was measured and the values were expressed in μm^3. These values divided by somatic chromosome number gave the interphase chromosome volume. Chromocenters were counted and heterochromocentric values were determined by planimetry using tracing paper and mm graph paper following the procedure as suggested by Kabir and Singh (1989).

3.5.1 Statistical analysis

A statistical comparison by means of Duncan's Multiple Range Test (DMRT) was made. For different parameters of interphase nuclear structure of different accessions was made statistically by analysis of variance (ANOVA). Significance level was set at $p \leq 0.05$. The data analysis was done using SPSS version 20.0 for Windows. Graphs were drawn by Microsoft Office Excel software.

Chapter IV
Results

4. RESULTS

The cosmopolitan grass species *Cynodon dactylon* has received a lot of attention because of its economic importance as a turf grass, for grazing, hay production and also because of its abundance and widespread distribution. This grass species as a whole, however, has been neglected until recently, particularly in Bangladesh. It is likely that much more information on distribution and ecological behaviour will emerge which may be probable aspects for the living beings. From this point of view, the findings in the present study may be considered as preliminary information, which may be useful for revising and amplifying the future works at various levels. However, the findings obtained from this investigation are presented and described under different heads and sub-heads as follows.

4.1 Distribution

Bermuda grass (*Cynodon dactylon* (L.) Pers.) is thought to have originated around the Indian Ocean Basin, from East Africa to India. Before 1947 Bangladesh was an integral part of India and that time a term was used to say and that is Greater Bengal, which means Bangladesh plus West Bengal (a province now of India). From that point of view, it may be said that the Bangladesh is also a center of origin of the bermudagrass, which is called or known as Durba in Bengali in both parts of the divided Bengal. This grass species is found to grow well in tropical and sub-tropical parts of the world. This grass is also found as far as 50 N in Europe and down to 37 in the southern hemisphere. It can be found at high altitudes, upto 2600 m in the tropics, and 4000 m in the Himalayas.

At present Bangladesh comprises 64 districts and this grass species is found with an outstanding spreading ability all over the country. In the present study 23 districts plus one location of high salinity under different bio-ecological zones were considered as the study area. Physico-chemical properties of the habitat soils were analyzed and the data are presented in this chapter partially and also elaborately in the next chapter of adaptation. However, a keen study was made on the intensity of growth, growth duration and tolerance to drought / salinity stress of *Cynodon dactylon* collected from 24 locations/ habitats. A matrix of 24 accessions of this grass species to these said parameters is shown in Table 5.

In almost all the sites or locations selected in the present study *C. dactylon* accessions were found to creep extensively by means to scaly rhizomes or by flat stolons. Some of them were found to produce seeds side by side reproducing vegetatively and also that from rhizome or stolon. It was also observed that in case of plain land they were growing moderately on well-drained soil, either acidic or alkaline. They need mainly soil moisture alongwith adequate nutrients and sunlight.

Table 5: Matrix of growth intensity, production time, soil pH and drought / salinity stress of *Cynordon dactylon*.

Sl. No.	Habitats	Growth intensity	Season of production	Soil pH	Drought / Saline tolerant
1	Rangpur	++	Round the year	7.2	DT
2	Lalmonirhat	+	”	5.4	N
3	Dinajpur	++	”	7.1	DT
4	Thakurgaon	+	”	6.5	N
5	Panchagarh	+	”	6.6	N
6	Gaibandha	+	”	6.5	N
7	Rajshahi	+++	”	8.1	DT
8	Naogaon	++	”	7.1	DT
9	Pabna	+++	”	7.8	N
10	Gazipur	+	”	6.4	N
11	Narsingdi	+++	”	8.5	N
12	Sherpur	++	”	6.8	N
13	Mymensingh	+++	”	7.8	N
14	Khulna	++	”	7.5	N
15	Jessore	+++	”	8.2	N
16	Jhenaidah	+++	”	8.0	N
17	Faridpur	+++	”	7.9	N
18	Shariatpur	+++	”	8.4	N
19	Barguna	+++	”	7.8	ST
20	Khagrachari	+	”	5.1	N
21	Bandarban	+	”	6.7	N
22	Rangamati	+	”	6.7	N
23	Cox’s Bazar	+	”	4.9	ST
24	St. Martin’s Island	++	”	7.3	ST

+ = Low, ++ = Medium, +++ = High, N = Normal, DT = Drought tolerant, ST = Saline tolerant.

Accessions of *C. dactylon* used in the present study were collected mainly from roadside or in the vicinity of lawn or homestead. Besides they were found to grow in the crop lands. They were found to grow abundantly along thoroughfares or sidewalks or in vacant lots or on summer dry soils of foothills. In some of the locations this grass was found as pasture and playing fields.

Table 5 reveals growth intensity of *C. dactylon* from low to high. Nine accessions showed low intensity, six showed medium and rest of them showed high intensity of growth. According to opinion of the local peoples all the 24 accessions were found to show their duration/period/ season of production round the year. Local people's opinions were recorded by the authoress herself and by the accession collectors. High intensity of growth was found in those locations where the soils were highly alkaline and those particular habitats were at Rajshahi, Pabna, Narsingdi, Mymensingh, Jessore, Jhenaidah, Faridpur, Shariatpur and Barguna. On the contrary low intensity of growth was observed in case of Lalmonirhat, Thakurgaon, Panchagarh, Gaibandha, Gazipur, Khangrachari, Bandarban, Rangamati and Cox's Bazar where the soils were found to be acidic. Soil pH of all the habitat soils were measured in the present study. Among these soils pH at 24 locations were found to range from 4.9 to 8.5 and almost all the alkaline soils showed the growth intensity of *C. dactylon* as medium or high. Among 24 accessions four were found to be drought tolerant and three were found to be saline tolerant, and rest of them were normal i.e., any type of stress free. The present findings indicate that in less than 50% locations of Bangladesh *C. dactylon* is growing enormously as variable perennial grass. So undoubtedly this grass species has become a truly cosmopolitan herbaceous plant species all over the country and exhibits a wide ecological tolerance due to its great plasticity and capability of growing in different soil types under varied climatic conditions.

4.2 Morphological characteristics and physico-chemical properties of habitat soil and soil of experimentation field

Taxonomic evaluation of key morphological characters observed in all the 24 accessions of *Cynodon dactylon* was found to be species specific and constant. Similarity in growth habit and spikelet structure was also uniform. The accessions showed variation in many of the morphological characters (Tables 6-10). Shoot number, shoot length, node number, internode length, fresh and dry weight of shoot were found with higher values in the accessions collected from Cox's Bazar, Khagrachari, Rangamati and for the last three characters in Bandarban, respectively (Table 6). On the contrary, lower values of shoot length and internode length were found in the accessions collected from Panchagarh, Dinajpur, and shoot number and number of nodes in Panchagarh, and for last two characters in Shariatpur, subsequently.

Table 6: Morphological variation of 24 accessions of *Cynodon dactylon* shoot collected from different habitats of Bangladesh.

Habitats	Parameters of shoot (Mean ± SE)						
	Number of shoot	**Length (cm)**	**No. of node**	**Internode length (cm)**	**Colour**	**Fresh weight (g)**	**Dry weight (g)**
Rangpur	8.50±0.1097j	14.72±0.0208o	6.00±0.0757gh	2.52±0.0252j	Green	4.31±0.0529n	1.4±0.0115o
Lalmonirhat	12.00±0.0153c	16.13±0.0306k	4.50±0.0321n	2.73±0.0200i	Green	4.83±0.0321l	1.82±0.0173kl
Dinajpur	7.67±0.0643l	10.66±0.0551t	4.17±0.0306o	1.44±0.0208n	Green	2.95±0.0306t	1.05±0.0306q
Thakurgaon	6.83±0.0321m	19.45±0.0569g	4.83±0.0416m	2.80±0.0361i	Green	8.11±0.0416e	3.48±0.0404e
Panchagarh	5.67±0.0058q	13.92±0.0473q	3.83±0.0473p	3.23±0.0306h	Green	3.27±0.0361r	1.12±0.0058q
Gaibandha	8.17±0.0361k	14.36±0.0520p	4.50±0.0404n	2.72±0.0321i	Green, Gray	5.89±0.0586i	1.91±0.0231jk
Rajshahi	11.83±0.0231d	20.02±0.0265f	9.17±0.0100e	2.77±0.0231i	Green	3.99±0.0503p	1.91±0.0153jk
Naogaon	9.33±0.0757h	14.66±0.0100o	4.17±0.0252o	2.28±0.0173k	Green	4.14±0.0603o	1.60±0.0289n
Pabna	9.50±0.0451g	17.11±0.0115i	5.67±0.0231ij	2.12±0.0351lm	Green	5.30±0.0404j	2.01±0.0173i
Gazipur	6.50±0.0153o	16.41±0.0153j	4.83±0.0265m	4.13±0.0577d	Green	4.20±0.0153no	1.29±0.0252p
Narsingdi	8.50±0.0737j	12.48±0.0751r	4.17±0.0361o	2.23±0.0115kl	Green	5.86±0.0252i	1.94±0.0208ij
Sherpur	8.50±0.0666j	17.42±0.0681h	6.67±0.0379f	3.48±0.0346g	Green	5.10±0.0624k	1.69±0.0115mn
Mymensingh	9.00±0.0569i	16.12±0.0513k	6.17±0.0153g	2.73±0.0173i	Green, Gray	6.64±0.0473g	2.65±0.0513g
Khulna	7.67±0.0551l	15.72±0.0361m	5.50±0.0451jk	2.42±0.0153j	Green	4.64±0.0666m	1.75±0.0231lm
Jessore	6.67±0.0231n	15.56±0.0208n	4.67±0.0503mn	2.08±0.0351m	Brown	3.79±0.0208q	1.65±0.0153n
Jhenaidah	5.83±0.0416p	16.15±0.0306k	5.33±0.0252kl	2.78±0.0100i	Green	4.58±0.0252m	1.44±0.0252o
Faridpur	10.67±0.0265e	16.33±0.0656j	4.17±0.0115o	2.15±0.0306lm	Green	3.11±0.0231s	1.10±0.0551q
Shariatpur	8.50±0.0624j	12.03±0.0321s	5.23±0.0306l	2.50±0.0379j	Green	2.57±0.0265u	0.81±0.0404r
Barguna	9.67±0.0643f	15.88±0.0458l	5.83±0.0208hi	3.32±0.0603h	Green	6.17±0.0321h	2.13±0.0208h
Khagrachari	8.50±0.0404j	73.25±0.0702a	23.00±0.0321b	5.53±0.0808b	Green	10.50±0.0289c	4.20±0.0635c
Bandarban	9.40±0.0379gh	70.10±0.0493b	19.80±0.0436c	6.10±0.0451a	Green	11.30±0.0173a	4.50±0.0058a
Rangamati	7.75±0.0115l	58.38±0.0289d	25.50±0.0551a	3.65±0.0503f	Green	9.60±0.0451d	3.81±0.0231d
Cox's Bazar	18.60±0.0153a	62.98±0.0265c	20.00±0.2974c	4.62±0.0379c	Light Green	11.00±0.0781b	4.39±0.0289b
St. Martin's Island	14.80±0.0551b	33.16±0.0173e	12.20±0.1557d	3.78±0.0551e	Light Green	7.68±0.0681f	3.07±0.0404f

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

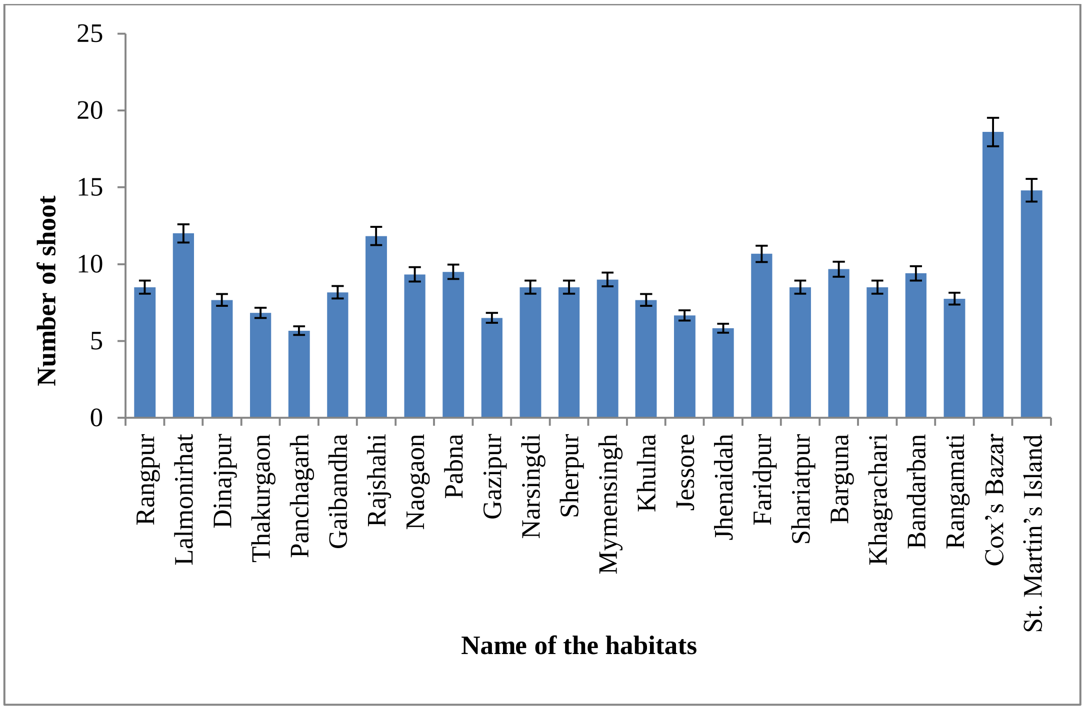


Fig. 6: Number of shoots of the accessions of *C. dactylon* collected from 24 habitats.

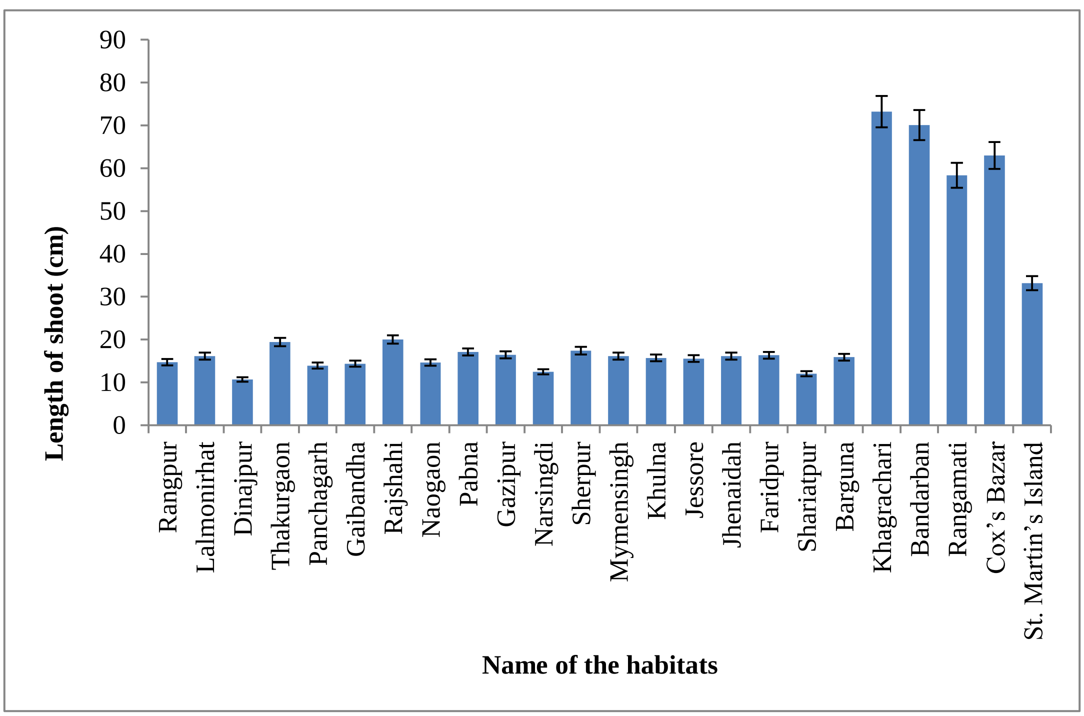


Fig. 7: Length of shoot of the accessions of *C. dactylon* collected from 24 habitats.

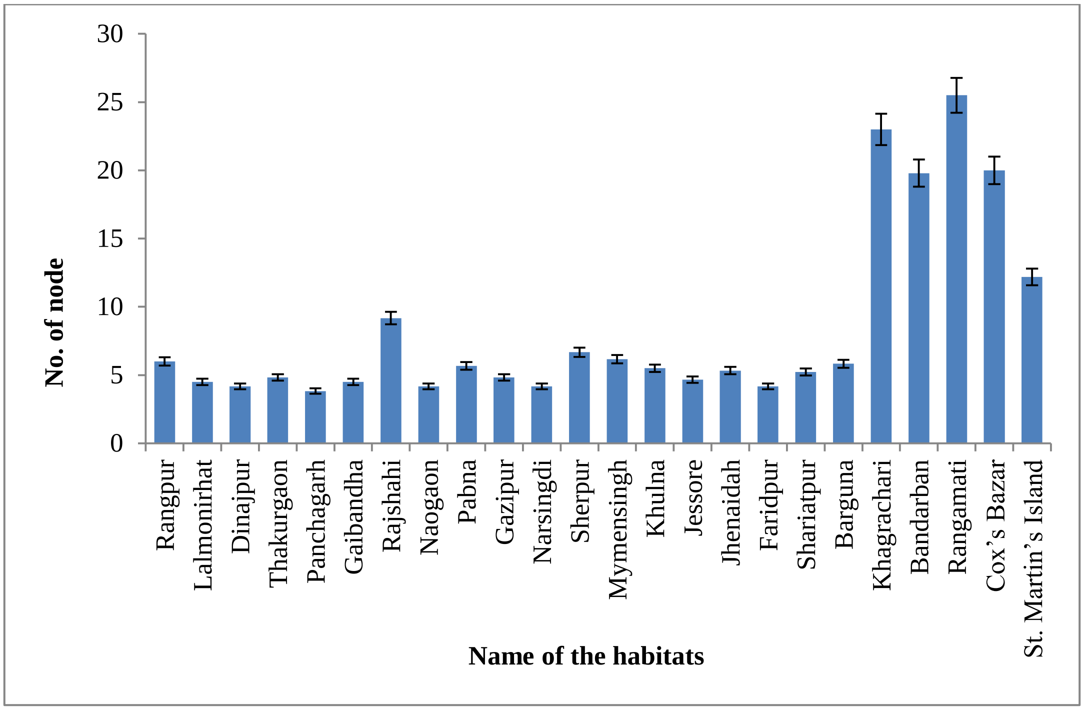


Fig. 8: Number of nodes of the accessions of *C. dactylon* collected from 24 habitats.

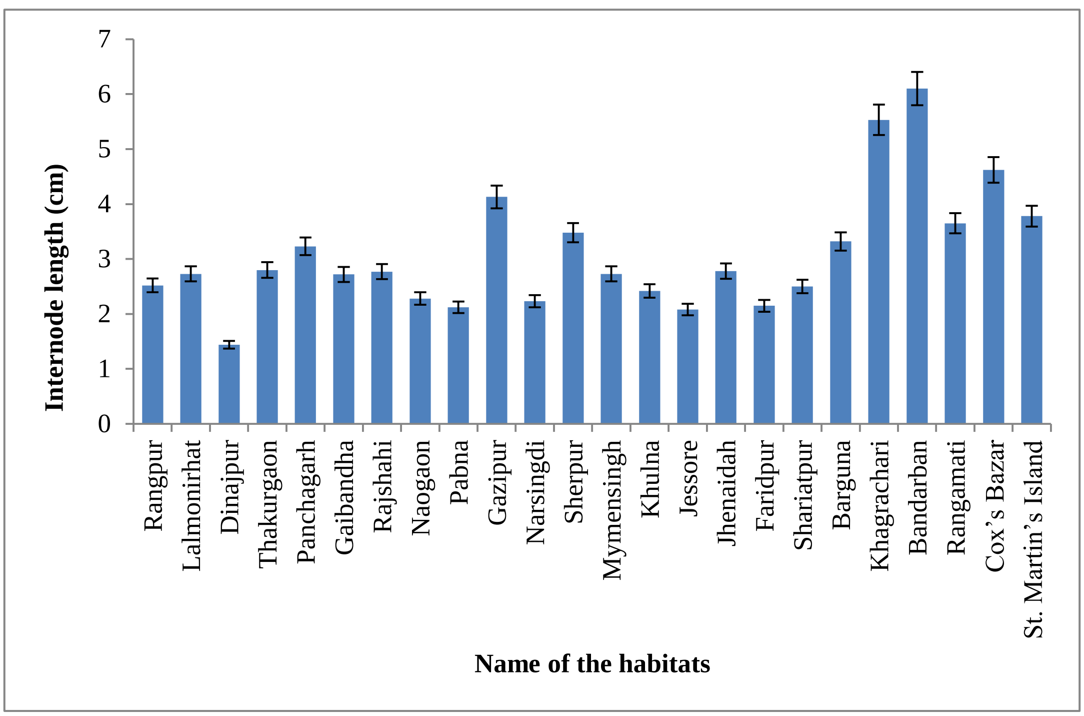


Fig. 9: Internode length of the accessions of *C. dactylon* collected from 24 habitats.

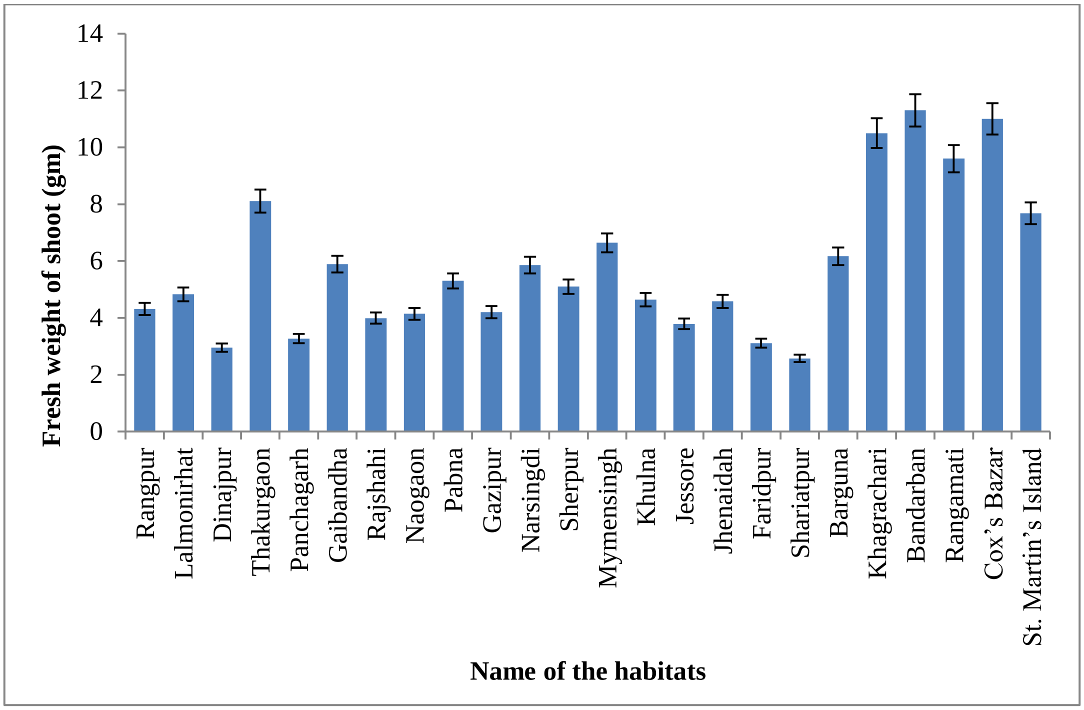


Fig. 10: Fresh weight of shoot of the accessions of *C. dactylon* collected from 24 habitats.

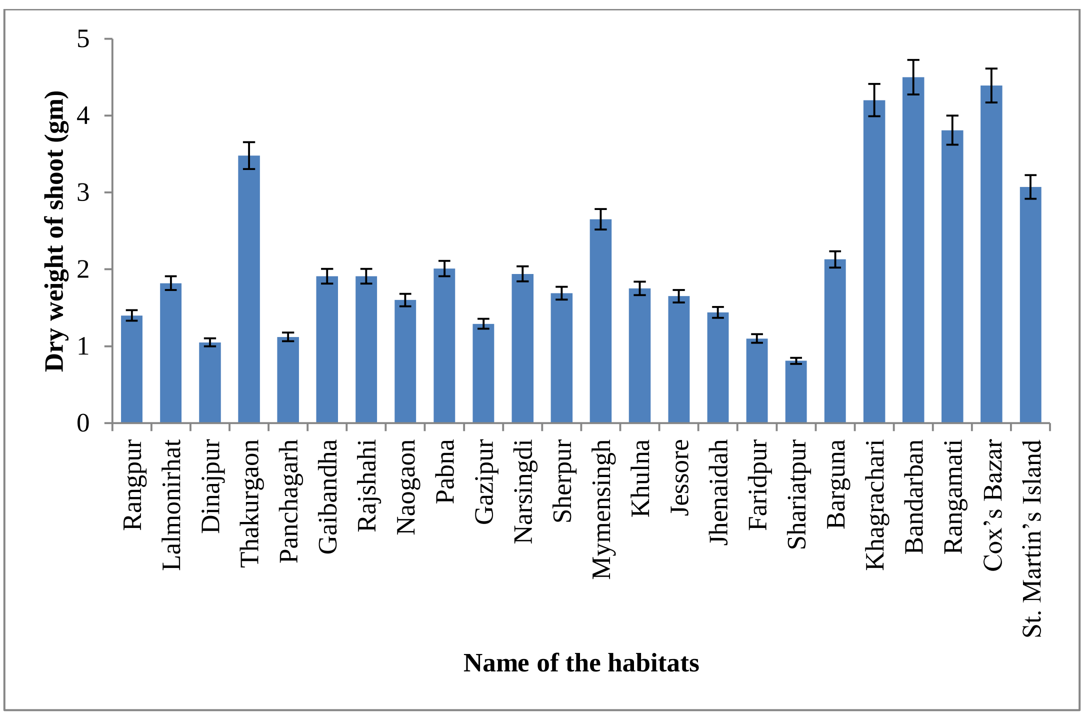


Fig. 11: Dry weight of shoot of the accessions of *C. dactylon* collected from 24 habitats.

Table 7 reveals higher values for root number and root length in case of Cox's Bazar; fresh and dry weight of root in case of Rajshahi and for rhizome number in case of the accessions collected from Khulna. Lower values were obtained for root number in accessions of Gaibandha, for root length in case of Rajshahi, fresh weight in case of Shariatpur, dry weight in case of Gazipur and for rhizome number in the accession collected from Rangamati.

Table 7: Morphological variation of 24 accessions of *Cynodon dactylon* root collected from different habitats of Bangladesh.

Habitats	Parameters of Root (Mean ± SE)				
	Number of root	Length (cm)	Fresh weight (g)	Dry weight (g)	Number of rhizome
Rangpur	14.17±0.1732i	10.77±0.0058h	1.93±0.1793efg	0.71±0.0265efgh	4.00±0.0289o
Lalmonirhat	15.00±0.2887h	10.25±0.0115j	1.60±0.1528ghi	0.76±0.0513defg	4.33±0.0404m
Dinajpur	9.50±0.2309o	8.50±0.0173n	1.28±0.1000j	0.58±0.0529ghij	9.00±0.0462c
Thakurgaon	14.17±0.1155i	11.10±0.0231g	2.29±0.0500d	0.85±0.0503de	9.17±0.0346b
Panchagarh	9.67±0.0052o	8.92±0.0029m	1.71±0.0153gh	0.72±0.0153defgh	3.33±0.0058q
Gaibandha	8.00±0.0040p	6.65±0.0176p	2.08±0.0252def	0.82±0.0306def	5.20±0.0346j
Rajshahi	14.83±0.0866h	4.92±0.0404q	4.61±0.0321a	2.37±0.1000a	7.50±0.0577f
Naogaon	11.50±0.1270l	11.05±0.0462g	1.73±0.0493gh	0.63±0.0252ghij	7.33±0.0115g
Pabna	11.00±0.0635m	8.50±0.0577n	1.69±0.0586gh	0.73±0.0635defg	8.83±0.0666d
Gazipur	13.50±0.0046j	10.25±0.0635j	1.22±0.0866j	0.47±0.0814j	4.00±0.0751o
Narsingdi	15.00±0.0520h	6.98±0.0751o	1.30±0.0029ij	0.62±0.0500ghij	6.33±0.0577h
Sherpur	16.00±0.0058g	9.30±0.0866l	1.63±0.0208gh	0.67±0.1127efghi	3.00±0.1155r
Mymensingh	11.50±0.0173l	10.68±0.0924hi	1.48±0.0635hij	0.64±0.0700fghij	4.17±0.0635n
Khulna	16.67±0.0231f	8.92±0.0087m	1.61±0.0361ghi	0.59±0.0404ghij	11.50±0.0289a
Jessore	13.50±0.0346j	6.88±0.0052o	1.25±0.0404j	0.53±0.0351hij	5.33±0.0173j
Jhenaidah	12.83±0.0404k	10.00±0.0064k	1.80±0.2887fgh	0.49±0.0252ij	4.50±0.0173l
Faridpur	11.33±0.0289l	12.77±0.0110e	1.49±0.1258hij	0.61±0.0794ghij	8.33±0.0693e
Shariatpur	11.17±0.0693lm	11.48±0.1097f	0.91±0.0794k	0.61±0.0473ghij	5.50±0.0115i
Barguna	10.67±0.0751n	10.53±0.0693i	2.15±0.0889de	0.90±0.0651d	5.33±0.0751j
Khagrachari	18.75±0.0006e	13.35±0.0808d	2.20±0.1000de	0.90±0.0529d	3.75±0.0577p
Bandarban	33.00±0.0924c	16.42±0.0981b	3.61±0.1323b	1.46±0.0557c	6.20±0.0635h
Rangamati	29.00±0.0981d	14.08±0.1039c	3.17±0.0586c	1.30±0.0493c	2.00±0.0520s
Cox's Bazar	73.20±0.1039a	16.78±0.0121a	4.50±0.0379a	1.84±0.0451b	4.75±0.0751k
St. Martin's Island	38.00±0.1443b	13.26±0.0144d	3.33±0.0850bc	1.36±0.0361c	4.00±0.0289o

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

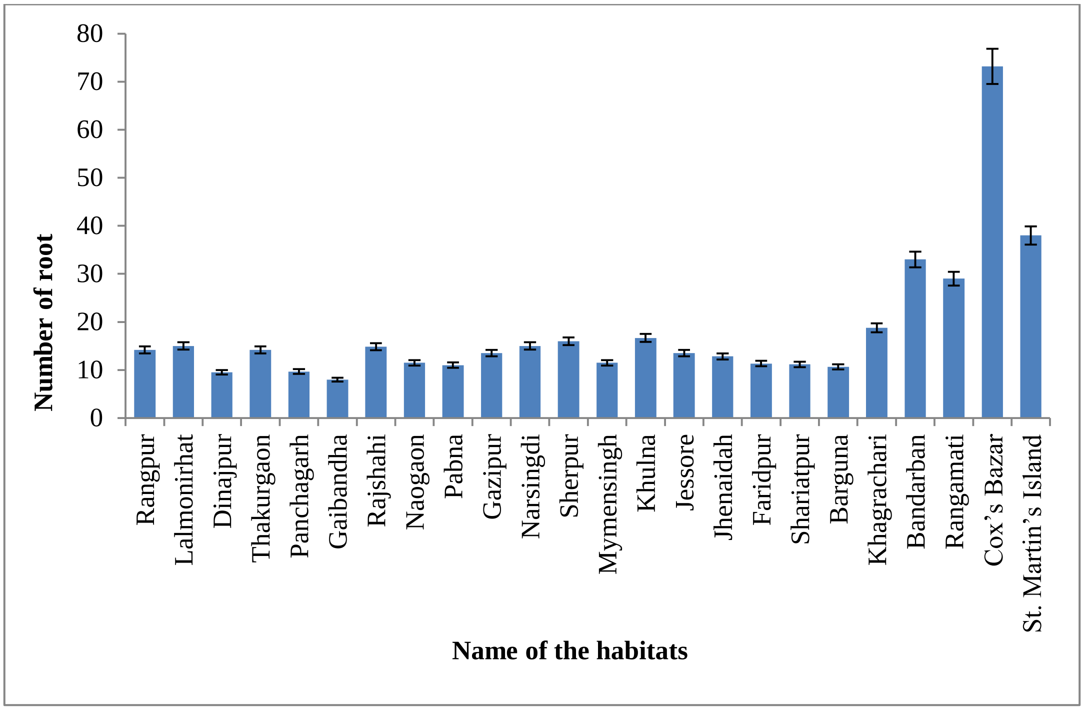


Fig. 12: Number of roots of the accessions of *C. dactylon* collected from 24 habitats.

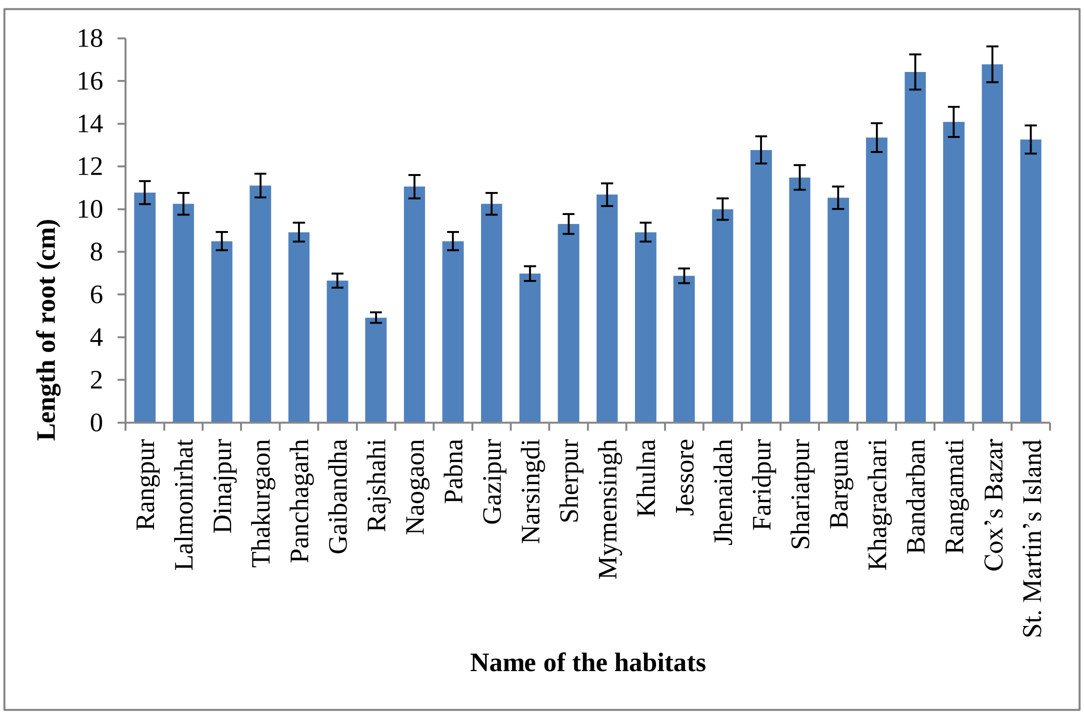


Fig. 13: Length of root of the accessions of *C. dactylon* collected from 24 habitats.

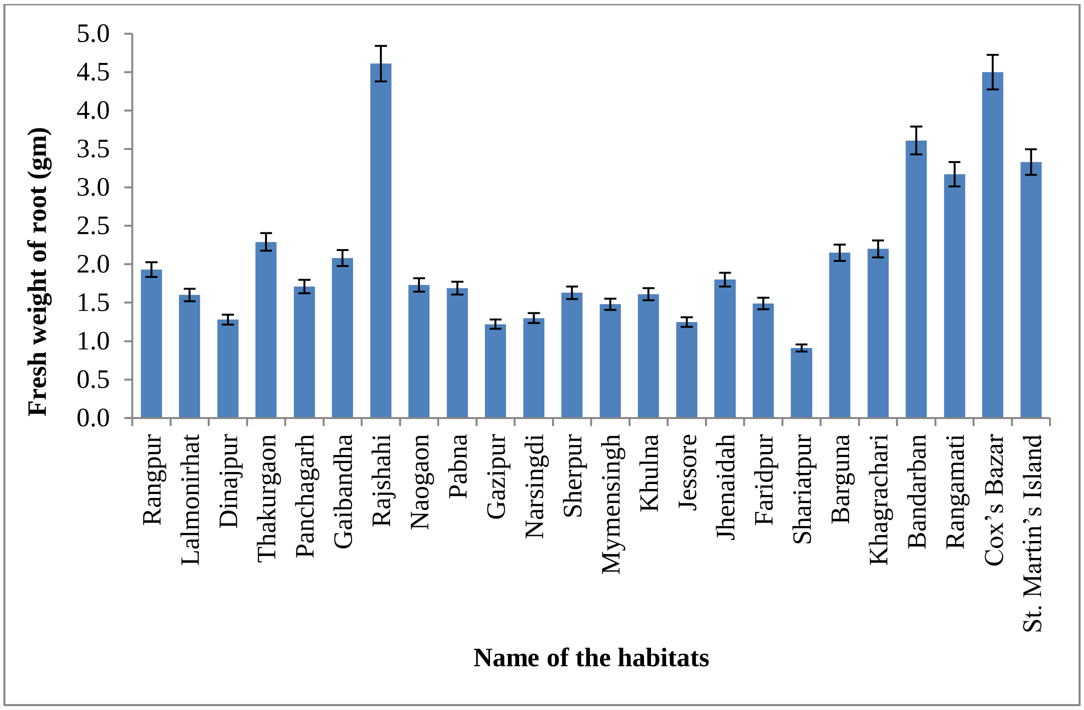


Fig. 14: Fresh weight of root of the accessions of *C. dactylon* collected from 24 habitats.

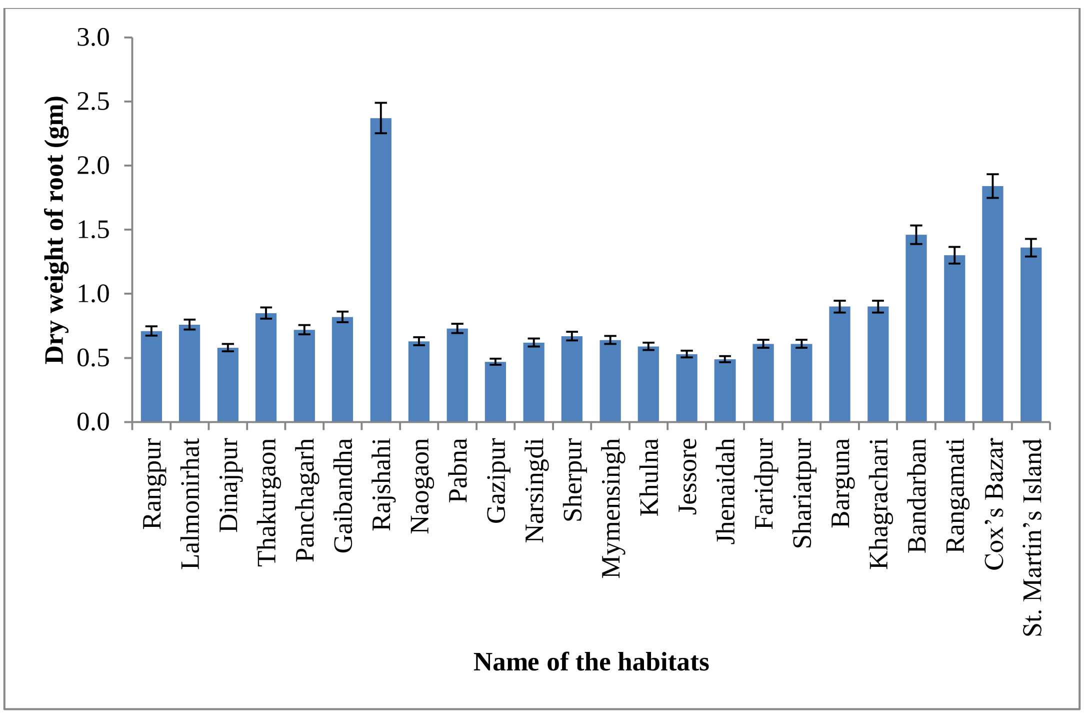


Fig. 15: Dry weight of root of the accessions of *C. dactylon* collected from 24 habitats.

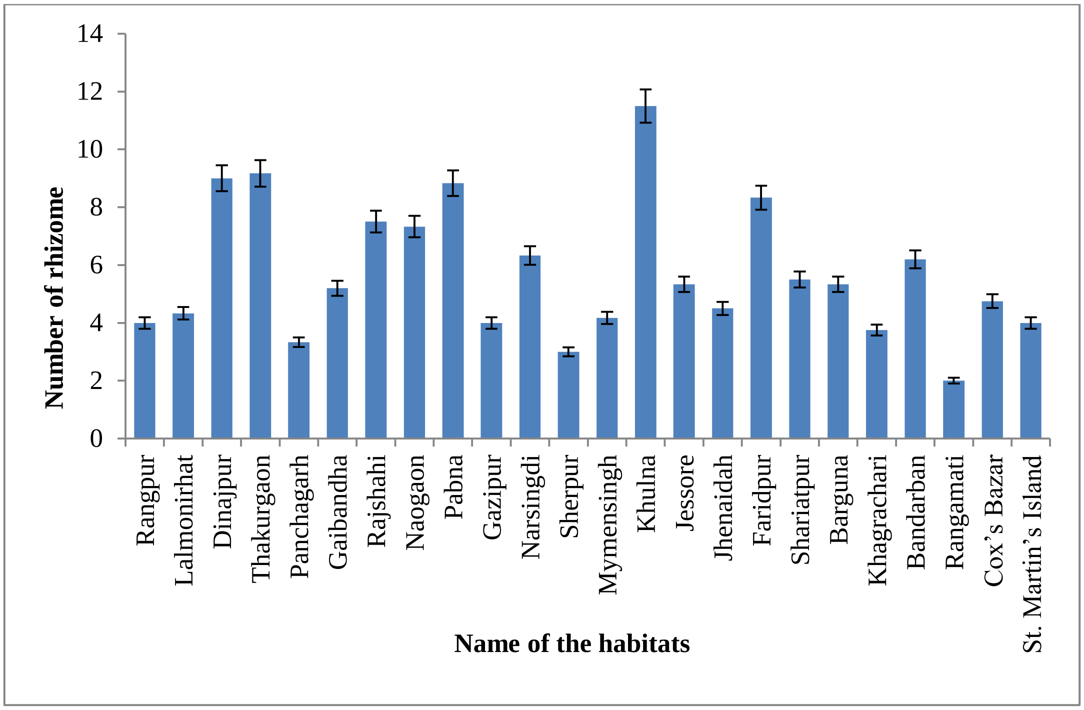


Fig. 16: Number of rhizomes of the accessions of *C. dactylon* collected from 24 habitats.

In case of flower characters spikelet colour were found to vary (Table 8). Highest value for number of inflorescences were found in accession collected from Cox's Bazar and lowest in that of Panchagarh and Saint Martin's Island. Highest and lowest values for number of racemes were found in the accessions collected from Cox's Bazar and Lalmonirhat, respectively. For length of racemes highest and lowest values were found in the accessions of Cox's Bazar and Rajshahi, respectively. Number of seeds per shoot and number of seeds per plant were found to be highest and lowest in the accessions collected from Cox's Bazar and Saint Martin's Island, subsequently.

Table 8: Morphological variation of 24 accessions of *Cynodon dactylon* flower and seed collected from different habitats of Bangladesh.

Habitats	Parameters of flower (Mean ± SE)				Parameters of seed (Mean ± SE)	
	No. of inflorescence / plant	No. of racemes	Length of racemes (cm)	Spikelet colour	No. of seeds / shoot	No. of seeds / plant
Rangpur	3.6±0.1793i	4.02±0.1762ghijk	4.00±0.1732cd	Brown	90.40±0.0265g	325.60±0.0289i
Lalmonirhat	5.0±0.1528g	3.73±0.1155l	3.42±0.1443hijkl	Green, Brown, DB	73.60±0.0513q	338.80±0.0404h
Dinajpur	3.6±0.1000i	3.88±0.0577jkl	3.07±0.0866mn	LG, Brown	73.40±0.0529r	264.00±0.0462p
Thakurgaon	4.8±0.0500g	4.18±0.0208fghi	3.54±0.0436ghij	Brown, LG	77.48±0.0503o	319.80±0.0346l
Panchagarh	2.0±0.0153l	4.10±0.0115ghij	4.27±0.0153bc	Brown	88.90±0.0153h	177.80±0.0058v
Gaibandha	2.8±0.0252jk	3.80±0.0173kl	3.60±0.0208fghi	Brown	66.10±0.0306w	182.20±0.0346u
Rajshahi	3.8±0.0321hi	5.95±0.0289d	2.77±0.0346n	Green, Brown	88.28±0.1000i	318.60±0.0577m
Naogaon	3.0±0.0493j	4.05±0.0058ghijk	3.67±0.0265efgh	Brown	85.44±0.0252k	244.00±0.0115s
Pabna	4.0±0.0586h	3.92±0.0520jkl	3.17±0.0666lm	LG, Brown	86.39±0.0608j	325.00±0.0635j
Gazipur	2.8±0.0866jk	3.93±0.0929ijkl	4.47±0.0781b	Brown	83.70±0.0814l	214.60±0.0751t
Narsingdi	3.0±0.0029j	4.25±0.0247fgh	3.84±0.0076defg	B, GB, Dark Brown	99.06±0.0500e	290.60±0.0577o
Sherpur	5.6±0.0208f	4.41±0.0321ef	3.30±0.0153ijklm	Brown, LG, DB	83.51±0.1127m	470.80±0.1155d
Mymensingh	7.2±00.0693e	4.12±0.0551ghij	3.03±0.0608mn	LG, Brown	71.53±0.0700s	525.40±0.0635c
Khulna	2.6±0.0361k	4.00±0.0289hijk	4.47±0.0265b	Brown	95.28±0.0404f	247.20±0.0289r
Jessore	3.6±0.0404i	4.01±0.0346hijk	3.93±0.0306de	Brown, LB, LG	113.72±0.0351b	392.00±0.0173f
Jhenaidah	2.8±0.2309jk	4.40±0.0462ef	4.24±0.3062bc	Brown, DB, LG	105.70±0.0252d	259.00±0.0173q
Faridpur	5.0±0.1258g	4.51±0.1443e	3.90±0.1153def	Brown, Light Green	109.32±0.0794c	537.80±0.0693b
Shariatpur	4.8±0.0794g	4.04±0.0693ghijk	2.98±0.0896mn	Brown, Light Green	82.96±0.0458n	393.80±0.0115e
Barguna	2.6±0.0889k	4.27±0.0872fg	3.26±0.0850jklm	Brown, Green, LG	68.10±0.0651u	172.40±0.0751w
Khagrachari	8.00±0.1000d	6.12±0.1155d	3.50±0.0929hijk	Brown	74.00±0.0529p	349.00±0.0577g
Bandarban	9.00±0.1323c	7.51±0.1323c	3.20±0.1300klm	Light Green, Brown	67.00±0.0557v	317.00±0.0635n
Rangamati	11.00±0.0586b	8.00±0.0586b	3.30±0.0608ijklm	Light Green, Brown	69.00±0.0493t	321.00±0.0520k
Cox's Bazar	13.00±0.0379a	10.25±0.0058a	6.10±0.0208a	Light Green	129.00±0.0451a	597.00±0.0751a
St. Martin's Island	2.00±0.0850l	4.00±0.0755hijk	2.85±0.0666n	Brown	28.00±0.0361x	130.00±0.0289x

B = Brown, G = Green, DB = Dark brown, LG = Light green.

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

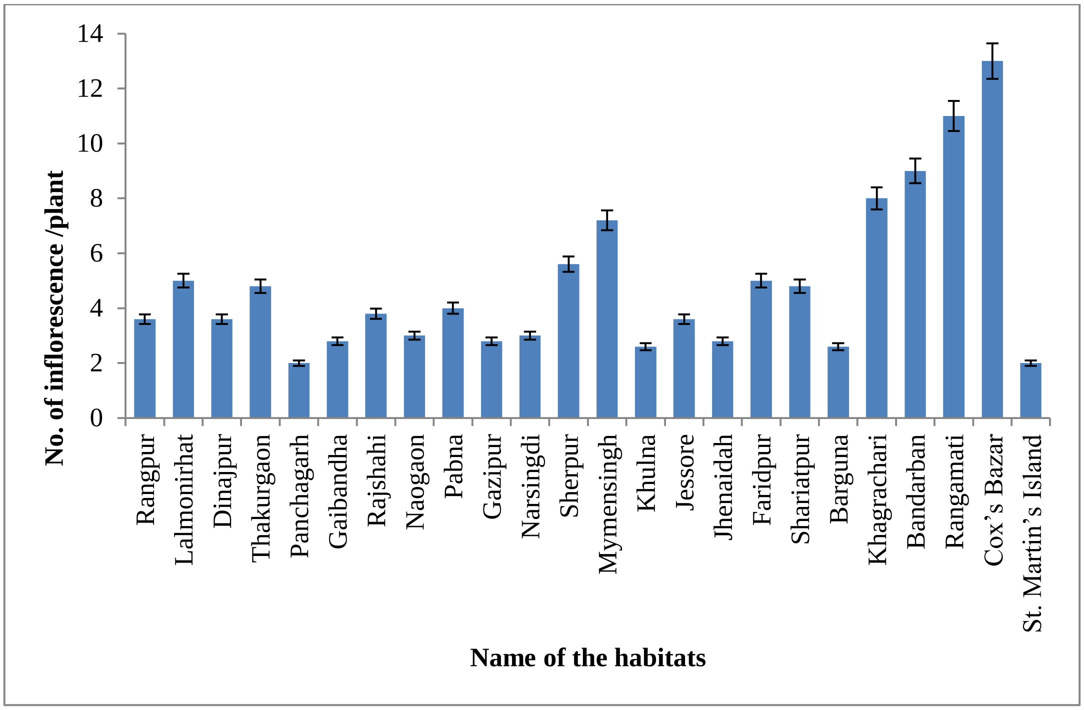


Fig. 17: Number of inflorescences of the accessions of *C. dactylon* collected from 24 habitats.

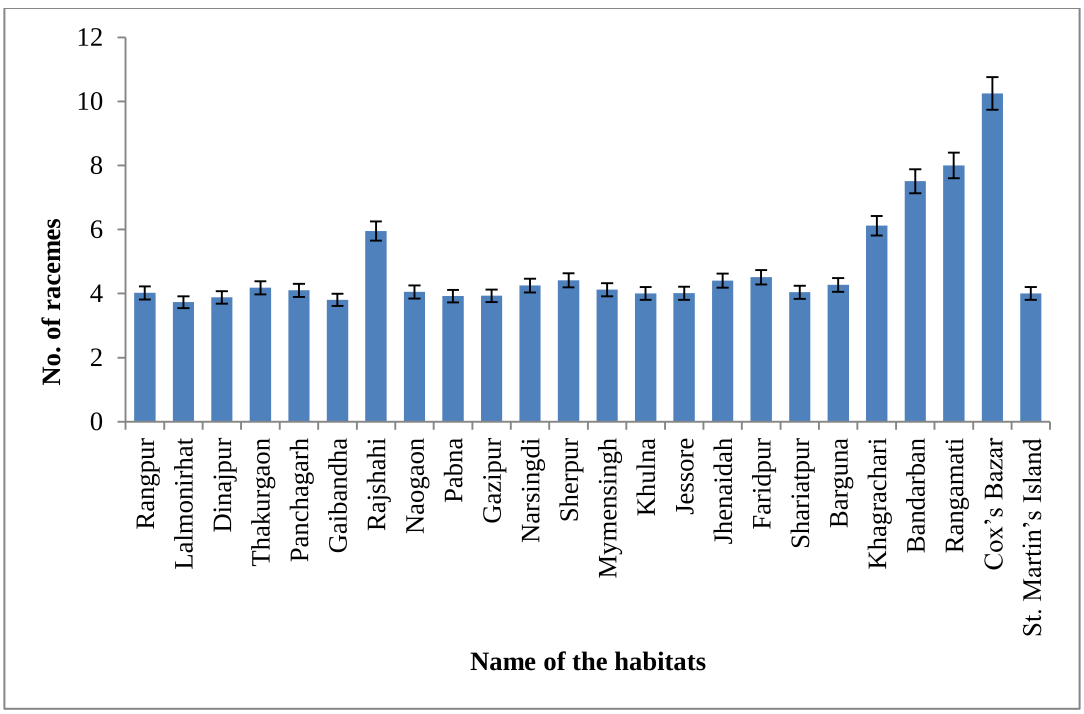


Fig. 18: Number of racemes of the accessions of *C. dactylon* collected from 24 habitats.

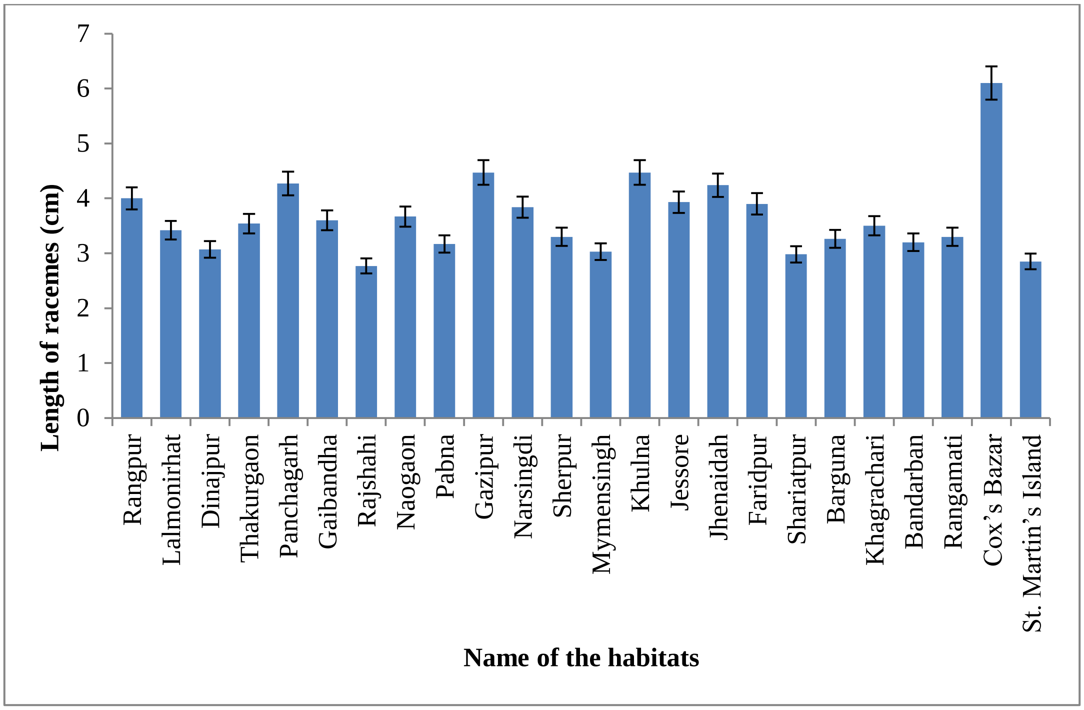


Fig. 19: Length of racemes of the accessions of *C. dactylon* collected from 24 habitats.

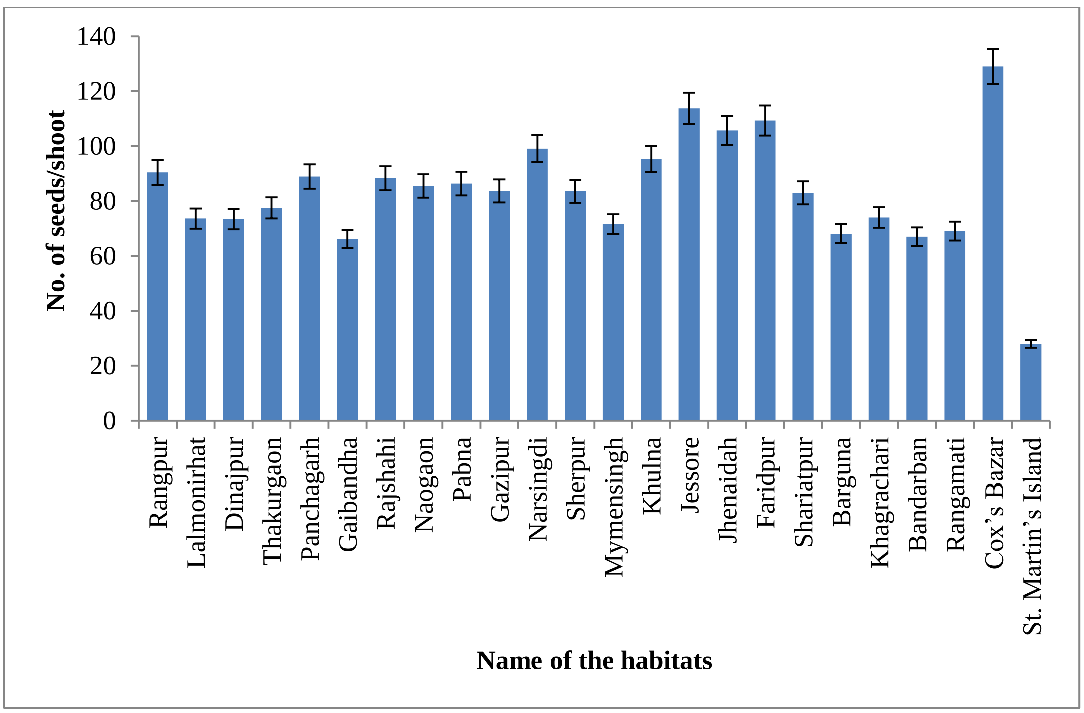


Fig. 20: Number of seeds per shoot of the accessions of *C. dactylon* collected from 24 habitats.

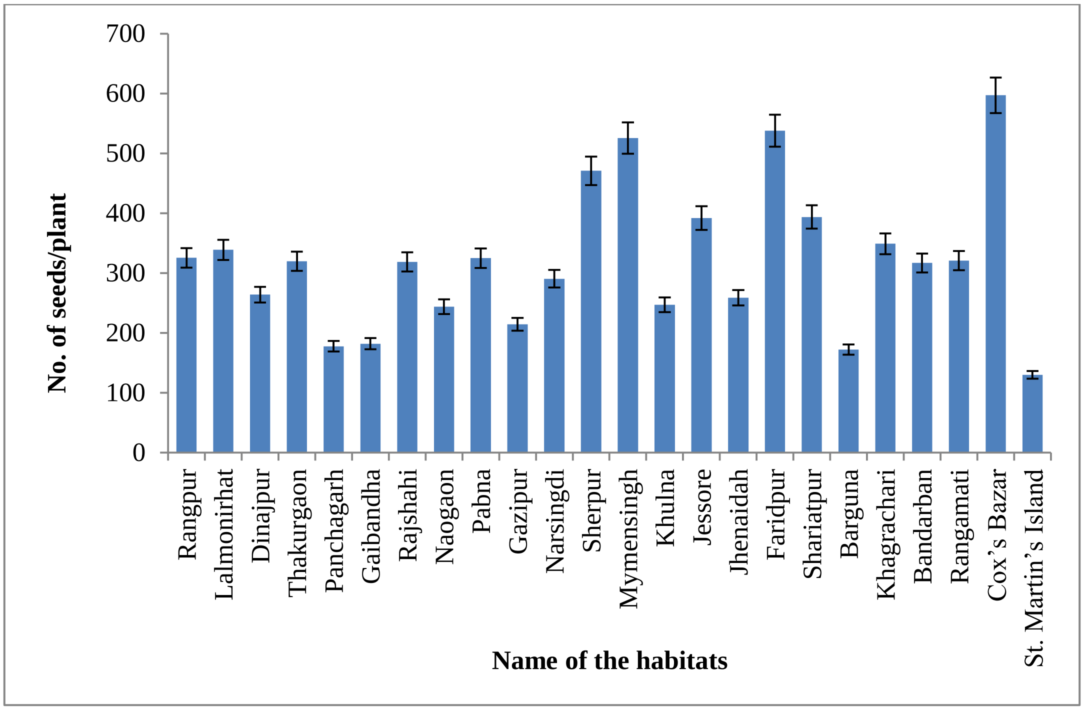


Fig. 21: Number of seeds per plant of the accessions of *C. dactylon* collected from 24 habitats.

In case of leaf characters (Table 9) number of leaves, total leaf area, total leaves fresh weight and total leaves dry weight were found with highest values in the accession collected from Bandarban and highest values for leaf length and leaf width were found in case of Cox's Bazar. On the other hand, lowest values for number of leaves, leaf length and leaf width were found in case of Faridpur, Rangamati and Khagrachari, and lowest values for total leaf area, leaves fresh weight and total leaves dry weight were found in case of Jessore and Shariatpur, consequently.

Table 9: Morphological variation of 24 accessions of *Cynodon dactylon* leaf collected from different habitats of Bangladesh.

Habitats	Parameters of Leaf (Mean ± SE)						
	No. of leaves / plant	Leaf length (cm) (single)	Leaf width (mm) (single)	Total leaf area (cm^2)	Total leaves fresh weight / plant (g)	Total leaves dry weight / plant (g)	Leaf nature (soft/stiff)
Rangpur	22.67±0.1732p	4.78±0.1762i	1.82±0.1850fg	224.25±0.0265m	1.73±0.0289jkl	0.39±0.0265efgh	Soft
Lalmonirhat	39.50±0.1323g	4.90±0.1155hi	2.00±0.1443def	242.45±0.0513k	2.24±0.0404g	0.37±0.0436fghi	Soft
Dinajpur	20.00±0.1041q	4.77±0.0577i	2.03±0.0866def	200.10±0.0529p	1.53±0.0462mno	0.31±0.0493hij	Stiff
Thakurgaon	23.50±0.0361n	5.68±0.0153d	2.07±0.0462def	154.30±0.0503u	1.61±0.0346lmn	0.24±0.0473ijk	Soft
Panchagarh	27.83±0.0153i	4.73±0.0115i	2.30±0.0173d	346.60±0.0153g	2.49±0.0058f	0.48±0.0153ef	Soft
Gaibandha	27.67±0.0208i	4.33±0.0173j	2.02±0.0231def	161.12±0.0306t	1.89±0.0346hij	0.31±0.0265hij	Soft
Rajshahi	80.33±0.0252e	5.40±0.0289ef	2.67±0.0346c	414.70±0.1000e	2.39±0.0577fg	0.51±0.0666e	Soft
Naogaon	13.50±0.0289u	4.82±0.0058hi	2.28±0.0265d	345.40±0.0252h	2.33±0.0115fg	0.49±0.0115ef	Soft
Pabna	15.67±0.0551t	3.75±0.0520k	2.02±0.0635def	249.50±0.0608j	1.79±0.0635ijk	0.45±0.0781efg	Soft
Gazipur	25.17±0.1172l	4.72±0.0981i	2.02±0.0808def	188.50±0.0814s	1.69±0.0751klm	0.30±0.0153hij	Soft
Narsingdi	19.33±0.0132r	4.88±0.0189hi	2.03±0.0076def	238.35±0.0500l	1.61±0.0577lmn	0.38±0.0666efgh	Soft
Sherpur	25.33±0.0200l	5.23±0.0115fg	1.58±0.0153ghi	211.15±0.1127o	1.70±0.1155klm	0.42±0.0529efgh	Soft
Mymensingh	17.17±0.0115s	5.05±0.0551gh	2.20±0.0635de	196.20±0.0700q	1.52±0.0635no	0.33±0.0551ghij	Soft
Khulna	27.33±0.0321j	5.37±0.0289ef	2.00±0.0231def	280.65±0.0361i	1 .70±0.0289klm	0.46±0.0361efg	Soft
Jessore	23.17±0.0361o	4.48±0.0346j	1.77±0.0208fgh	73.02±0.0208x	1.09±0.0173p	0.16±0.0208k	Soft
Jhenaidah	25.67±0.1044k	5.13±0.0462g	1.88±0.3002efg	118.05±0.0252v	1.17±0.0173p	0.23±0.0306jk	Soft
Faridpur	9.50±0.1210v	6.38±0.1443c	2.17±0.1153de	189.55±0.0794r	2.00±0.0693h	0.29±0.0153hij	Soft
Shariatpur	24.67±0.0693m	5.53±0.0693de	1.78±0.0896fgh	91.35±0.0458w	1.37±0.0115o	0.16±0.0173k	Soft
Barguna	29.50±0.1012h	5.47±0.0872de	2.18±0.0808de	223.60±0.0651n	1.91±0.0751hi	0.34±0.0115ghij	Soft
Khagrachari	139.50±0.0964b	4.35±0.1155j	1.45±0.0781i	720.16±0.0503b	10.65±0.0577b	1.76±0.0473b	Soft
Bandarban	177.80±0.1270a	5.68±0.1155d	1.60±0.1300ghi	800.00±0.0557a	11.73±0.0635a	2.44±0.0208a	Soft
Rangamati	136.25±0.0608c	2.50±0.0635l	1.50±0.0608hi	603.38±0.0608d	9.56±0.0520d	1.41±0.0520c	Soft
Cox's Bazar	134.00±0.0379d	10.08±0.0058a	3.82±0.0208a	650.12±0.0451c	10.26±0.0751c	1.67±0.0265b	Soft
St. Martin's Island	72.80±0.0681f	9.85±0.0755b	3.10±0.0666b	375.88±0.0153f	5.94±0.0289e	1.27±0.0153d	Soft

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

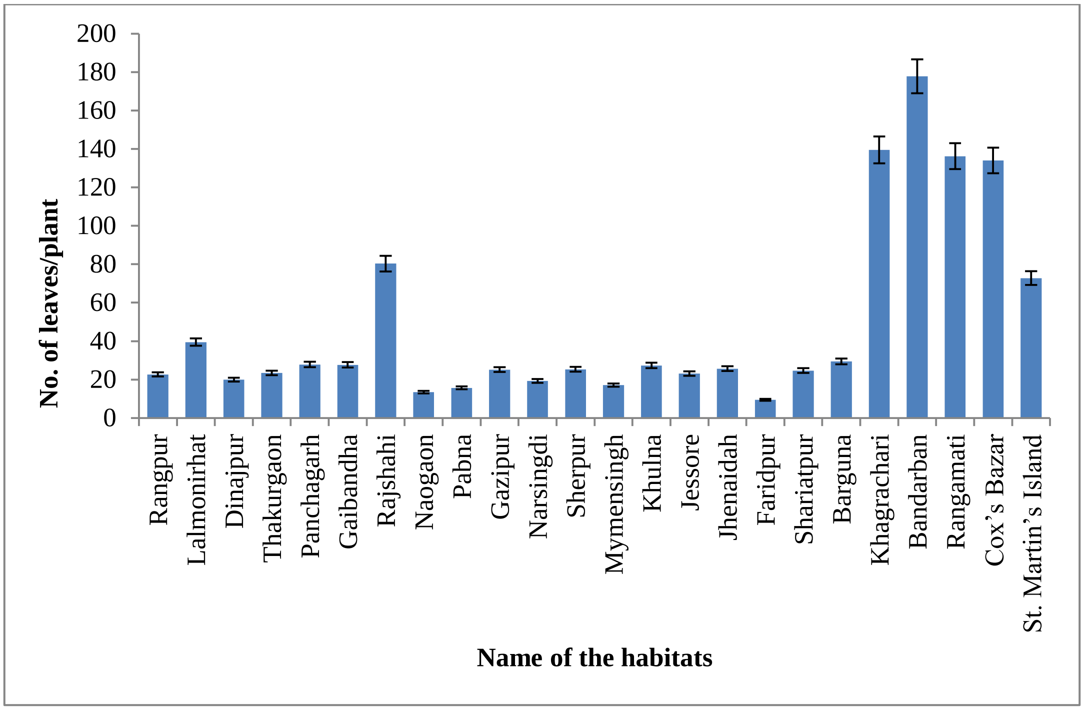


Fig. 22: Number of leaves per plant of the accessions of *C. dactylon* collected from 24 habitats.

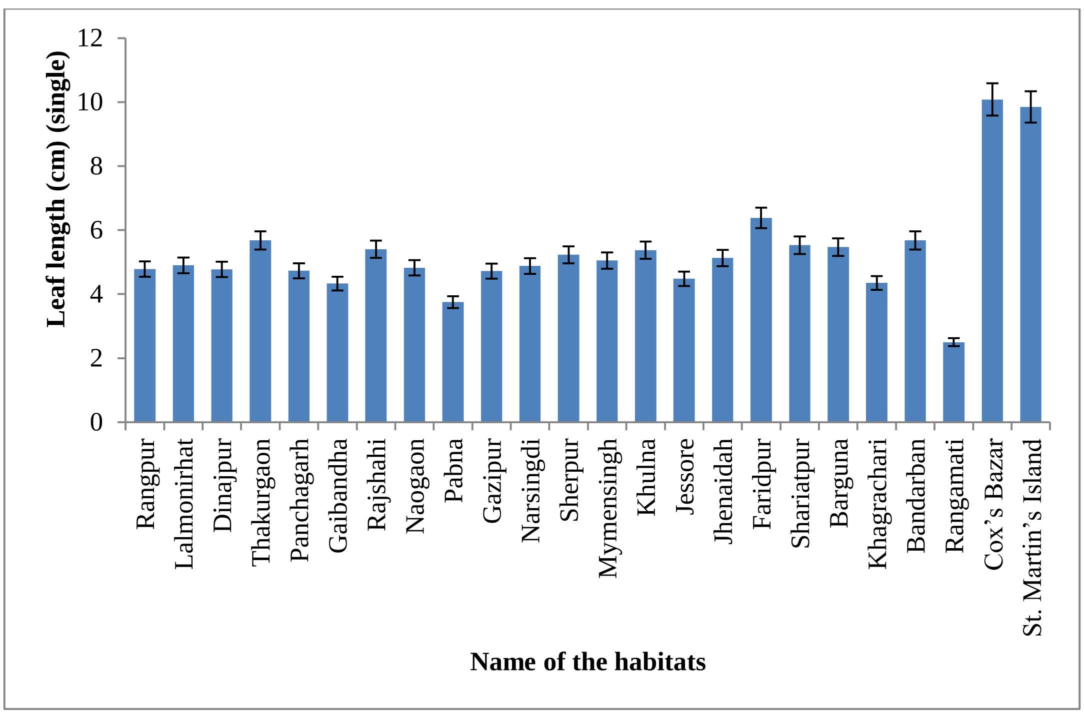


Fig. 23: Single leaf length of the accessions of *C. dactylon* collected from 24 habitats.

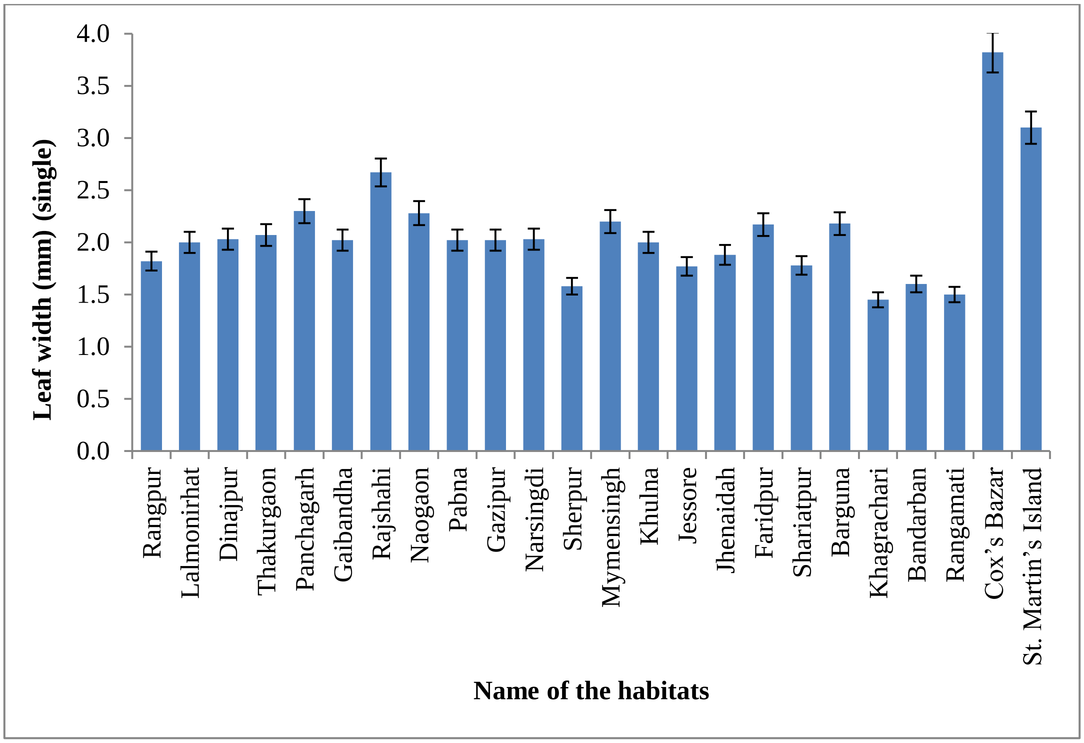


Fig. 24: Single leaf width of the accessions of *C. dactylon* collected from 24 habitats.

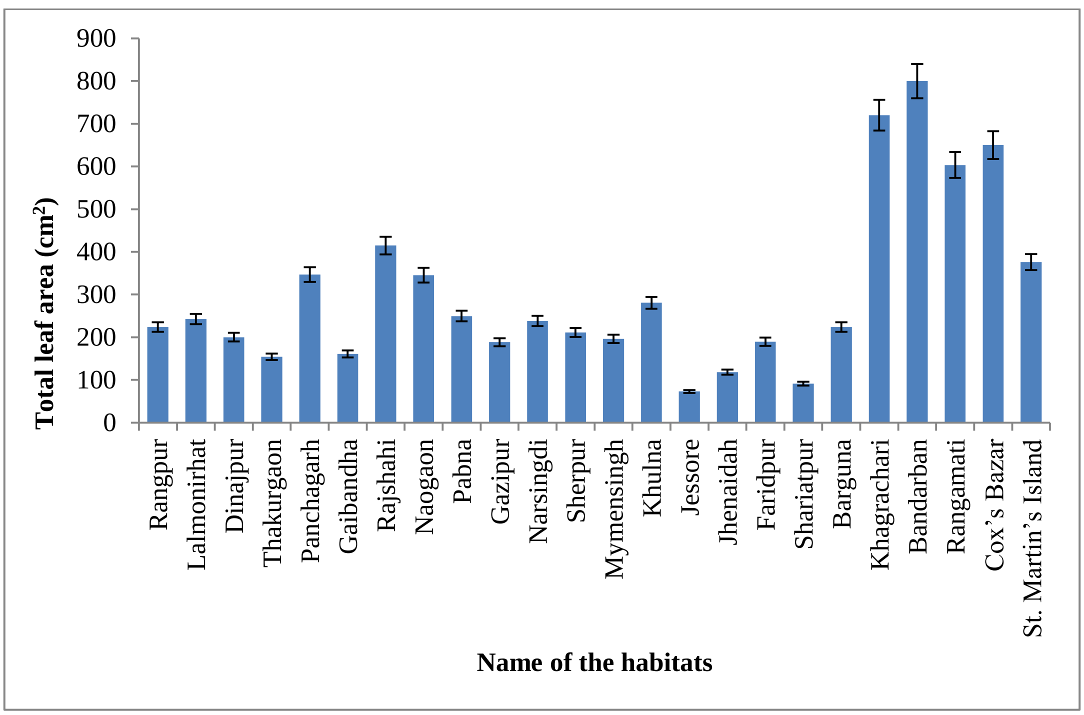


Fig. 25: Total leaf area of the accessions of *C. dactylon* collected from 24 habitats.

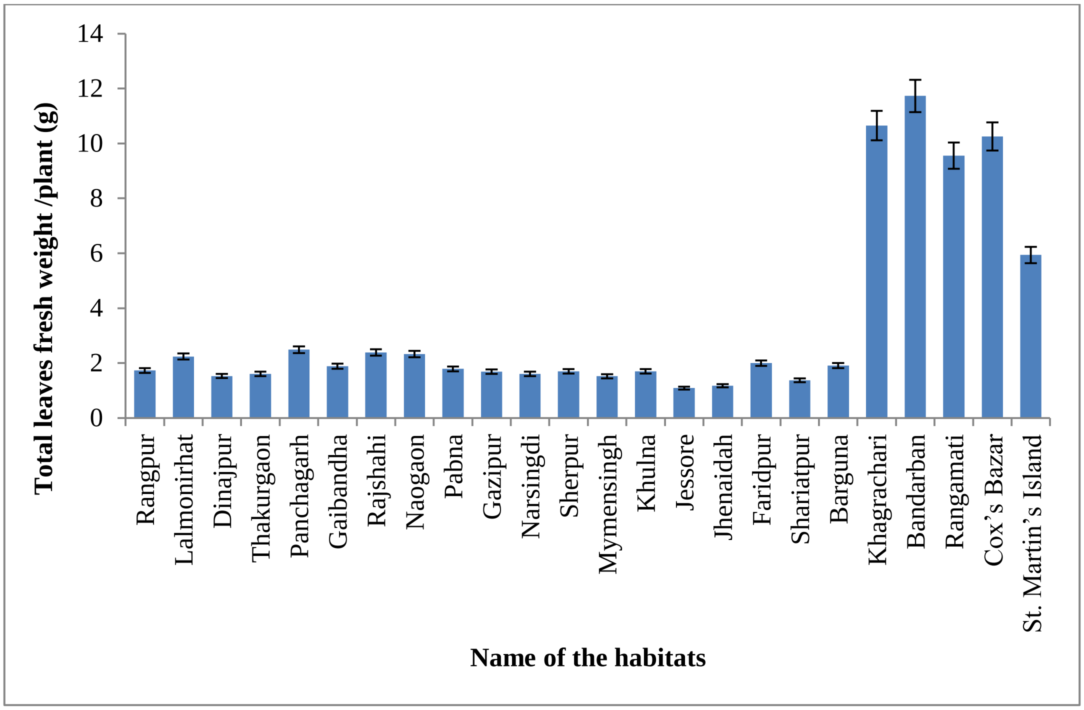


Fig. 26: Total leaves fresh weight per plant of the accessions of *C. dactylon* collected from 24 habitats.

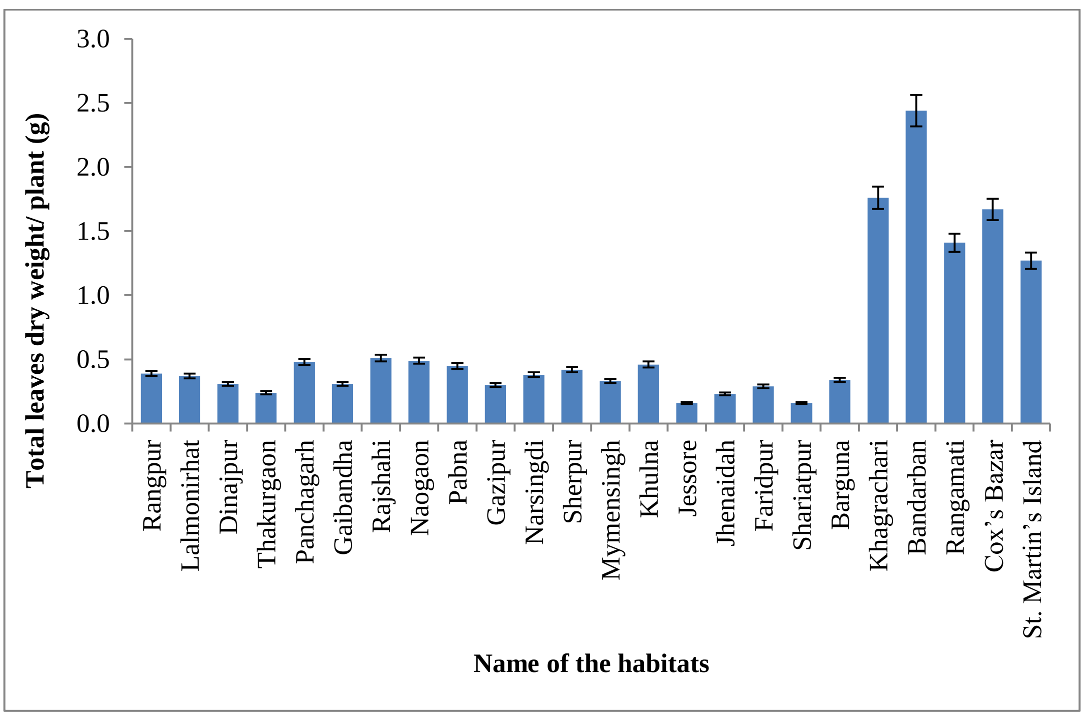


Fig. 27: Total leaves dry weight per plant of the accessions of *C. dactylon* collected from 24 habitats.

Table 10 reveals plant height (maximum and minimum), and number of stolons per plant and stolon length. Plant height were found with maximum and minimum values in accession of Bandarban and Gaibandha. Values for stolon per plant were found to be highest and lowest in the accessions of Cox's bazar and Khagrachari, respectively and highest and lowest value was found for stolon length in the accession of Khagrachari and Faridpur. Growth habit of all the 24 accessions showed erect and prostrate nature based on angle of stem to ground. Among all the accessions only eight accessions showed their both the growth habit in the ratio of 1:1 or 2:1, and rest of them were either erect or prostrate.

Table 10: Morphological variation of 24 accessions of *C. dactylon* plant height, stolon and growth habit collected from different habitats of Bangladesh.

Habitats	Parameters of plant height (cm) (Mean ± SE)		Parameters of stolon (Mean ± SE)		Growth habit: Angle of stem to ground (prostrated/erect)
	Maximum height (cm)	Minimum height (cm)	No. of stolon/plant	Stolon Length (cm)	
Rangpur	24.03±0.1825l	16.47±0.1848h	7.00±0.0379cd	2.94±0.0265j	Erect
Lalmonirhat	26.40±0.1343g	16.28±0.1212hi	5.00±0.1443i	2.72±0.0513klm	Prostrated
Dinajpur	17.97±0.1217t	13.35±0.0608m	4.67±0.0866jk	2.77±0.0529jklm	Erect
Thakurgaon	29.13±0.0404e	17.17±0.0153f	6.83±0.0462d	3.30±0.0503h	Erect: Prostrated 2:1
Panchagarh	22.23±0.0200p	14.53±0.0115k	6.83±0.0173d	3.21±0.0153hi	Erect
Gaibandha	21.33±0.0231r	10.50±0.0173p	6.20±0.0231gh	2.54±0.0306n	Erect: Prostrated 1:1
Rajshahi	27.02±0.0379f	16.75±0.0289g	7.17±0.0346c	3.33±0.1002h	Prostrated
Naogaon	24.18±0.0321l	16.12±0.0058i	4.33±0.0231l	2.88±0.0252jkl	Erect
Pabna	23.35±0.0586n	12.63±0.0520n	6.00±0.0635h	2.83±0.0608jklm	Erect: Prostrated 1:1
Gazipur	26.17±0.1159gh	13.88±0.0981l	6.72±0.0808de	3.12±0.0839i	Erect
Narsingdi	19.02±0.0132s	13.43±0.0189m	6.50±0.0087ef	3.28±0.0436hi	Erect
Sherpur	25.87±0.0153ij	13.57±0.0115m	4.50±0.0153kl	2.90±0.1155jk	Erect: Prostrated 1:2
Mymensingh	26.05±0.0153hi	16.42± 0.0635h	9.67±0.0635b	2.83±0.0723jklm	Prostrated
Khulna	23.62±0.0265m	15.07±0.0289j	4.83±0.0231ij	4.41±0.0289f	Erect: Prostrated 1:1
Jessore	21.93±0.0115q	11.37±0.0346o	6.71±0.0173de	2.70±0.0173lmn	Erect: Prostrated 2:1
Jhenaidah	25.70±0.1012jk	16.83±0.0462g	4.50±0.3002kl	2.85±0.0265jkl	Erect
Faridpur	25.60±0.1193k	13.83±0.1443l	7.00±0.1153cd	2.66±0.0839mn	Erect: Prostrated 1:1
Shariatpur	22.48±0.0586o	17.15±0.0693f	6.33±0.0866fg	3.32±0.0458h	Erect: Prostrated 2:1
Barguna	23.73±0.1044m	14.93±0.0872j	6.33±0.0808fg	3.55±0.0569g	Erect
Khagrachari	96.67±0.0872a	48.00±0.1155b	3.25±0.0781n	14.70±0.0557a	Prostrated
Bandarban	96.80±0.1582a	54.14±0.1155a	4.60±0.1328jkl	12.36±0.0473c	Prostrated
Rangamati	74.13±0.0777c	39.05±0.0635c	4.00±0.0608m	10.38±0.0058d	Prostrated
Cox's Bazar	80.28±0.0265b	33.84±0.0058d	12.75±0.0200a	8.05±0.0529e	Erect
St. Martin's Island	51.24±0.0681d	29.02±0.0603e	3.80±0.0666m	12.96±0.0173b	Erect

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

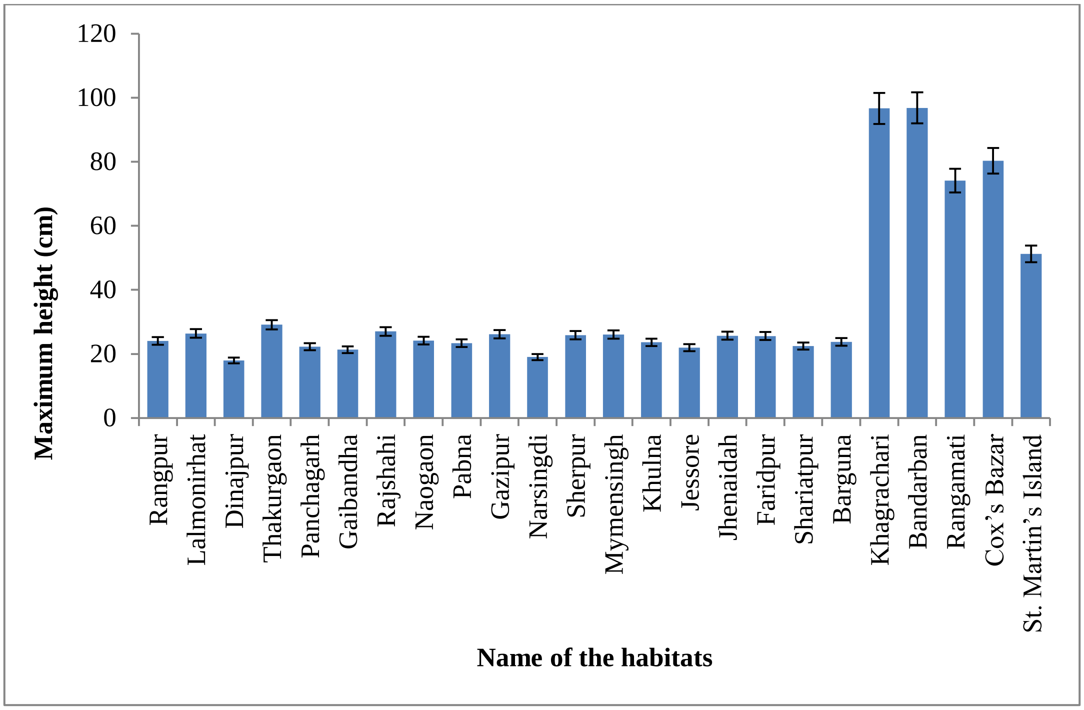


Fig. 28: Maximum height of the accessions of *C. dactylon* collected from 24 habitats.

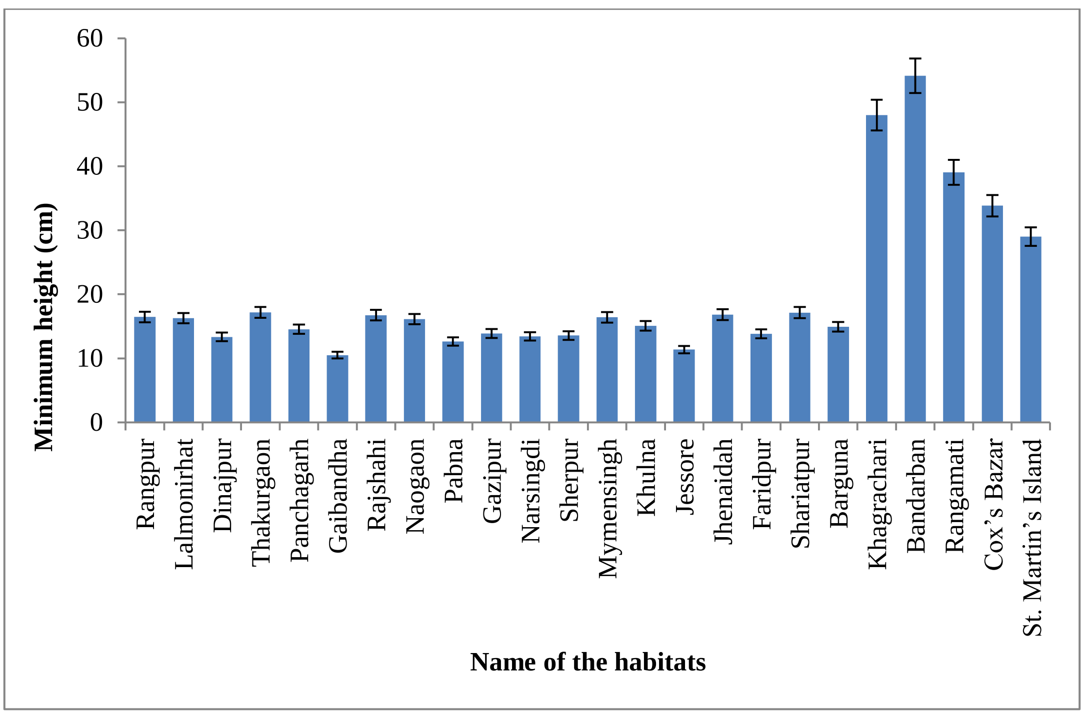


Fig. 29: Minimum height of the accessions of *C. dactylon* collected from 24 habitats.

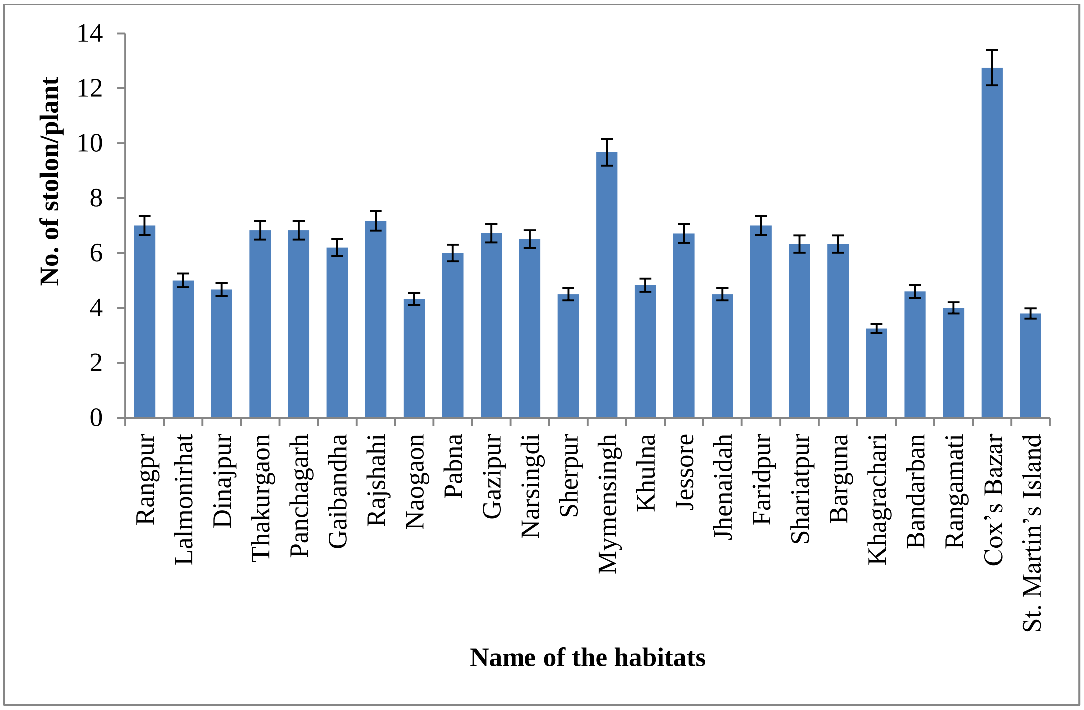


Fig. 30: Number of stolons per plant of the accessions of *C. dactylon* collected from 24 habitats.

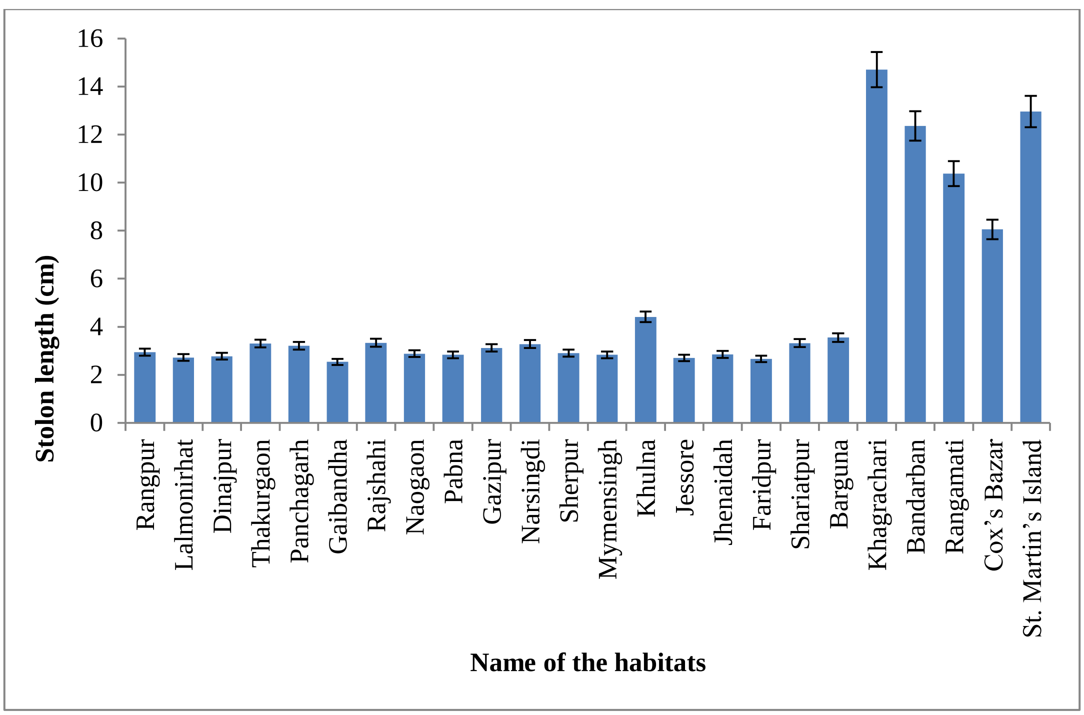


Fig. 31: Stolon length of the accessions of *C. dactylon* collected from 24 habitats.

The ANOVA (Tables 11-15) for shoot, root, flower, seed, leaf, plant height and stolon parameters showed the highly significant variation.

Table 11: Analysis of variance subjected to variation of different parameters of shoot ability in *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Shoot	No of shoot	Habitat	23	24.840	3.263***
		Error	48	0.008	
	Shoot length	Habitat	23	1145.351	1.995***
		Error	48	0.006	
	No. of nodes	Habitat	23	130.820	7.290***
		Error	48	0.018	
	Internode length	Habitat	23	3.680	843.567***
		Error	48	0.004	
	Shoot fresh weight	Habitat	23	20.230	3.393***
		Error	48	0.006	
	Shoot dry weight	Habitat	23	3.717	1.332***
		Error	48	0.003	

*** = significant at $p \leq 0.001$.

Table 12: Analysis of variance subjected to variation of different parameters of root ability in *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Root	No of root	Habitat	23	579.651	1.685***
		Error	48	0.034	
	Root length	Habitat	23	25.574	2.605***
		Error	48	0.010	
	Root fresh weight	Habitat	23	3.041	99.867***
		Error	48	0.030	
	Root dry weight	Habitat	23	0.649	65.587***
		Error	48	0.010	
	No. of rhizomes	Habitat	23	16.436	1.884***
		Error	48	0.009	

*** = significant at $p \leq 0.001$.

Table 13: Analysis of variance subjected to variation of different parameters of inflorescence and seed ability in *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Inflorescence	No. of inflorescence	Habitat	23	24.761	924.021***
		Error	48	0.027	
	No. of raceme	Habitat	23	7.936	444.448***
		Error	48	0.018	
	Length of racemes	Habitat	23	1.550	52.673***
		Error	48	0.029	
Seed	Seeds no. /shoot	Habitat	23	1200.911	1.221***
		Error	48	0.10	
	Seeds no. /plant	Habitat	23	42775.330	4.931***
		Error	48	0.009	

*** = significant at $p \leq 0.001$.

Table 14: Analysis of variance subjected to variation of different parameters of leaf ability in *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Leaf	No. of leaf	Habitat	23	7035.092	3.577***
		Error	48	0.020	
	Leaf length	Habitat	23	7.789	448.552***
		Error	48	0.017	
	Leaf width	Habitat	23	0.791	27.189***
		Error	48	0.029	
	Total leaf area	Habitat	23	118365.718	1.222***
		Error	48	0.010	
	Total leaves fresh weight / plant	Habitat	23	34.716	4.002***
		Error	48	0.009	
	Total leaves dry weight / plant	Habitat	23	1.090	223.663***
		Error	48	0.005	

*** = significant at $p \leq 0.001$.

Table 15: Analysis of variance subjected to variation of different parameters of shoot ability in *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Plant height	Maximum height	Habitat	23	1812.945	8.417***
		Error	48	0.022	
	Minimum height	Habitat	23	414.614	2.325***
		Error	48	0.018	
Stolon	No. of stolon	Habitat	23	12.391	495.804***
		Error	48	0.025	
	Stolon length	Habitat	23	42.288	4.549***
		Error	48	0.009	

*** = significant at $p \leq 0.001$.

Tables 16 and 17 reveals physico-chemical properties of the soil of 24 original habitats along with the experimentation field of IBSc (01) = 25 with values and their magnitude of organic and inorganic matters, respectively. The soil of experimentation field was found to be loamy. The pH value of all the habitats were also determined and its highest mean value 8.5 (alkaline) was found in the soil of Narsingdi, which was somewhat higher than that of experimentation field along with the original habitat of Rajshahi, Jessore, Jhenaidah and Shariatpur. The acidic habitats were found in case of Lalmonirhat, Thakurgaon, Panchagarh, Gaibandha, Gazipur, Sherpur, Khagrachari, Bandarban, Rangamati and Cox's Bazar. Rest of the habitats were somewhat alkaline.

The highest values for organic matter, potassium, calcium and magnesium were found in the soil of Khulna, Dinajpur, Rajshahi and Dinajpur, respectively. The total nitrogen was found to be highest in the soil of Bandarban. The highest values for phosphorus, boron, copper and zinc were found in the soil of Pabna and the values for sulfur was highest in case of Cox's Bazar. On the other hand, highest amount of iron and manganise were found in the soil of Rangpur and Dinajpur, respectively.

Table 16: Determination of soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

Soil of different habitats	P^H	OM (%)	K	Ca	Mg	Total Nitrogen (%)	P	S	B	Cu	Fe	Mn	Zn
			C mol (+)/kg				Microgram/g						
IBSc Field	8.2	1.77	0.13	34.60	1.70	0.10	11.70	1.50	0.54	1.06	18.0	14.8	1.26
Rangpur	7.2	1.09	0.19	4.37	1.10	0.06	60.0	2.00	0.73	1.55	248.6	12.7	0.71
Lalmonirhat	5.4	2.11	0.34	0.92	0.25	0.12	62.9	3.33	0.28	0.36	137.3	9.6	2.86
Dinajpur	7.1	1.77	1.88	39.10	7.10	0.10	68.6	1.80	0.55	0.86	163.6	45.6	4.84
Thakurgaon	6.5	0.85	0.61	3.20	1.03	0.05	10.3	16.20	0.41	1.25	45.2	17.7	0.52
Panchagarh	6.6	1.71	0.57	33.70	1.83	0.10	20.0	1.50	0.53	0.44	53.4	4.0	0.61
Gaibandha	6.5	1.62	0.11	8.07	2.42	0.09	55.8	7.00	0.43	1.78	190.9	30.6	2.71
Rajshahi	8.1	1.33	0.17	42.90	1.01	0.08	10.0	9.10	0.70	2.67	15.6	6.0	2.95
Naogaon	7.0	1.29	0.62	5.15	1.65	0.08	26.6	1.90	0.58	1.18	91.0	18.6	1.85
Pabna	7.8	2.42	0.40	5.04	1.57	0.14	116.2	57.40	1.07	4.06	86.8	36.9	10.81
Gazipur	6.4	2.73	0.11	7.78	1.89	0.16	13.6	1.61	0.39	1.04	146.7	36.6	8.31
Narsingdi	8.5	0.52	0.10	12.10	0.66	0.03	28.4	28.67	0.52	1.98	24.2	6.8	0.87
Sherpur	6.8	1.55	0.29	5.00	1.98	0.09	67.4	1.44	0.88	1.88	130.2	12.4	1.48
Mymensingh	7.8	1.33	0.24	10.11	2.90	0.08	27.4	23.43	0.41	0.26	87.1	16.9	2.19
Khulna	7.5	2.70	0.53	17.99	5.50	0.16	17.8	33.85	0.52	0.80	59.9	30.0	0.94
Jessore	8.2	0.49	0.10	26.70	4.20	0.03	12.1	5.20	0.73	0.48	12.3	5.6	1.03
Jhenaidah	8.0	1.20	0.30	17.84	2.98	0.07	26.8	2.45	0.38	1.94	18.9	14.0	1.00
Faridpur	7.9	2.43	0.12	5.33	2.34	0.14	21.3	2.00	0.95	1.78	41.6	9.2	10.60
Shariatpur	8.4	1.25	0.28	22.37	2.76	0.07	16.4	5.26	0.29	2.68	31.0	7.5	0.43
Barguna	7.8	0.69	0.62	5.07	1.62	0.04	17.0	41.40	0.70	1.39	40.4	12.6	0.55
Khagrachari	5.1	1.42	0.71	6.39	2.00	0.08	7.6	3.94	0.18	0.24	33.6	41.4	0.95
Bandarban	6.7	4.03	0.48	13.24	1.67	0.23	9.3	7.39	0.30	0.90	86.3	32.5	6.47
Rangamati	6.7	2.06	1.18	9.70	3.08	0.12	8.5	5.94	0.20	0.41	29.9	27.0	2.14
Cox's Bazar	4.9	0.35	0.36	3.58	3.65	0.02	2.0	123.16	0.10	0.24	56.0	32.8	7.00
St. Martin's Island	7.3	0.77	0.08	12.21	0.80	0.04	4.8	21.50	0.15	0.23	29.3	7.0	0.62

OM = Organic matter, K= Potassium, Ca= Calcium, Mg = Magnesium, P = Phosphorus, S = Sulfur, B = Boron, Cu = Copper, Fe = Iron, Mn = Manganise, Zn = Zinc.

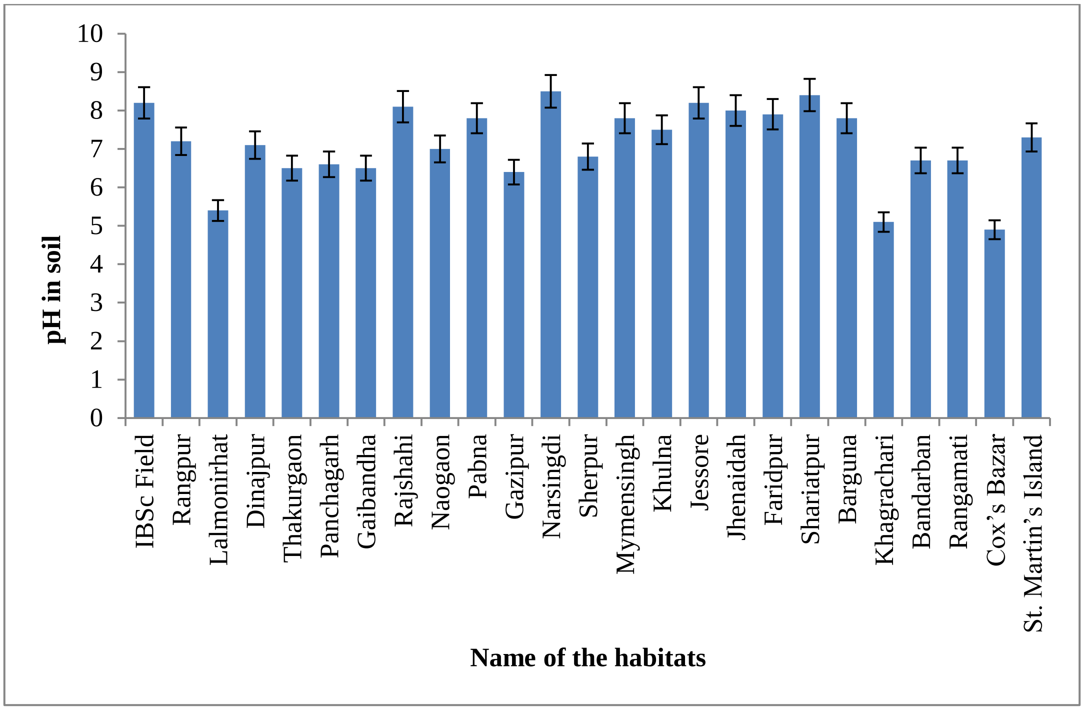


Fig. 32: Determination of pH in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

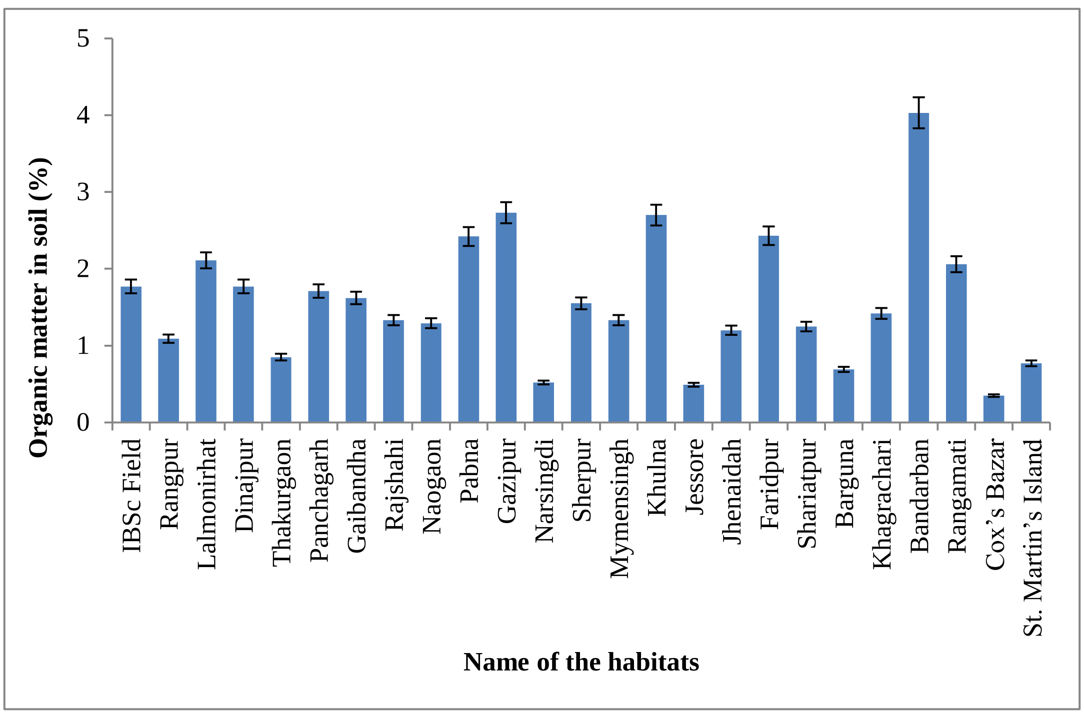


Fig. 33: Determination of organic matter in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

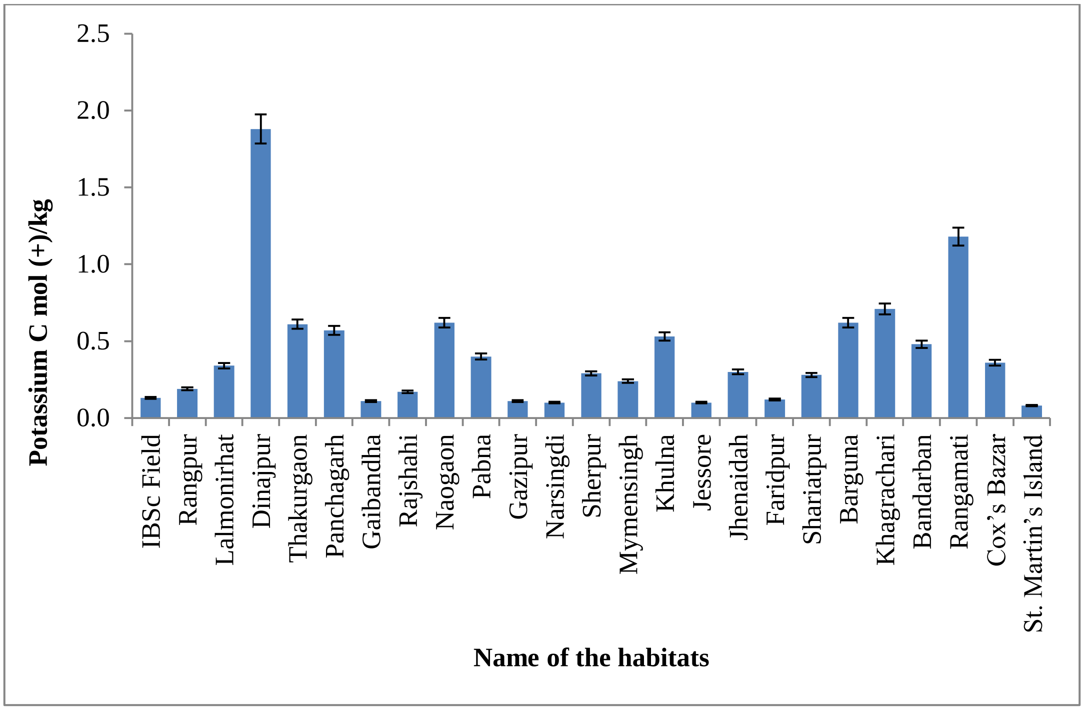


Fig. 34: Determination of potassium in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

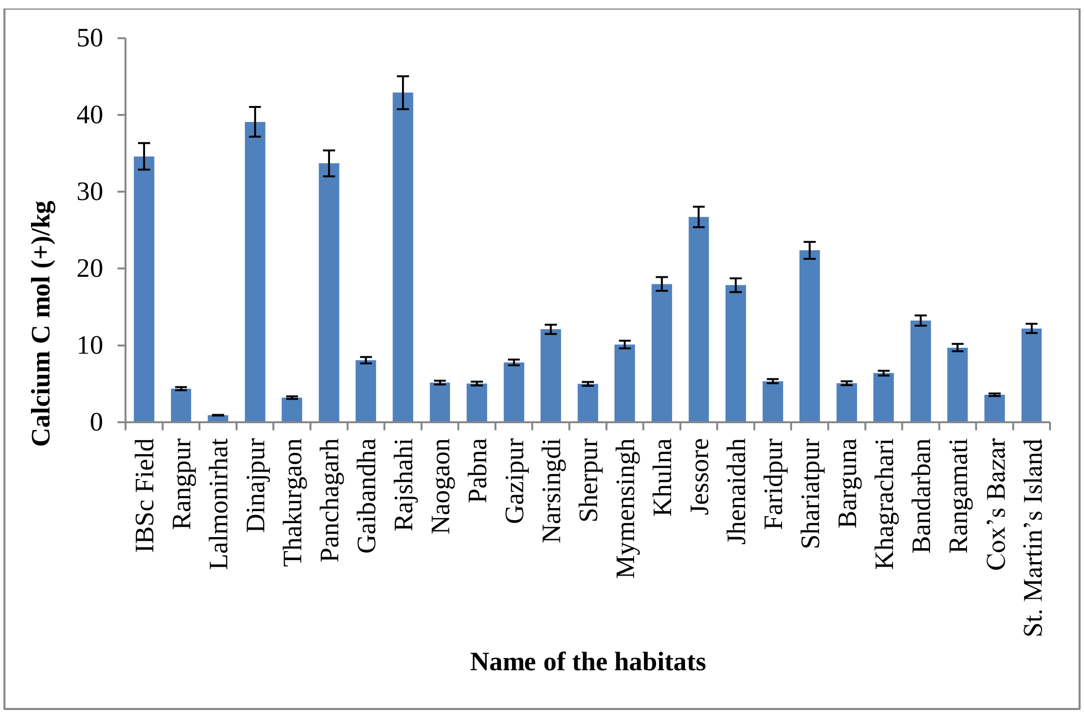


Fig. 35: Determination of calcium in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

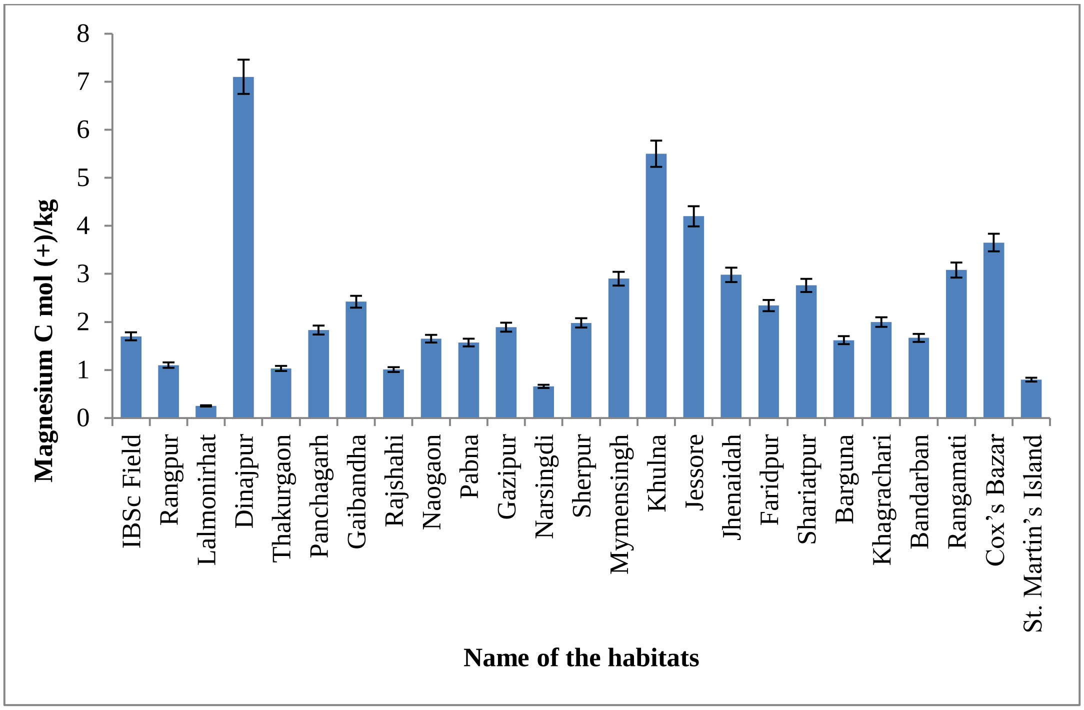


Fig. 36: Determination of magnesium in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

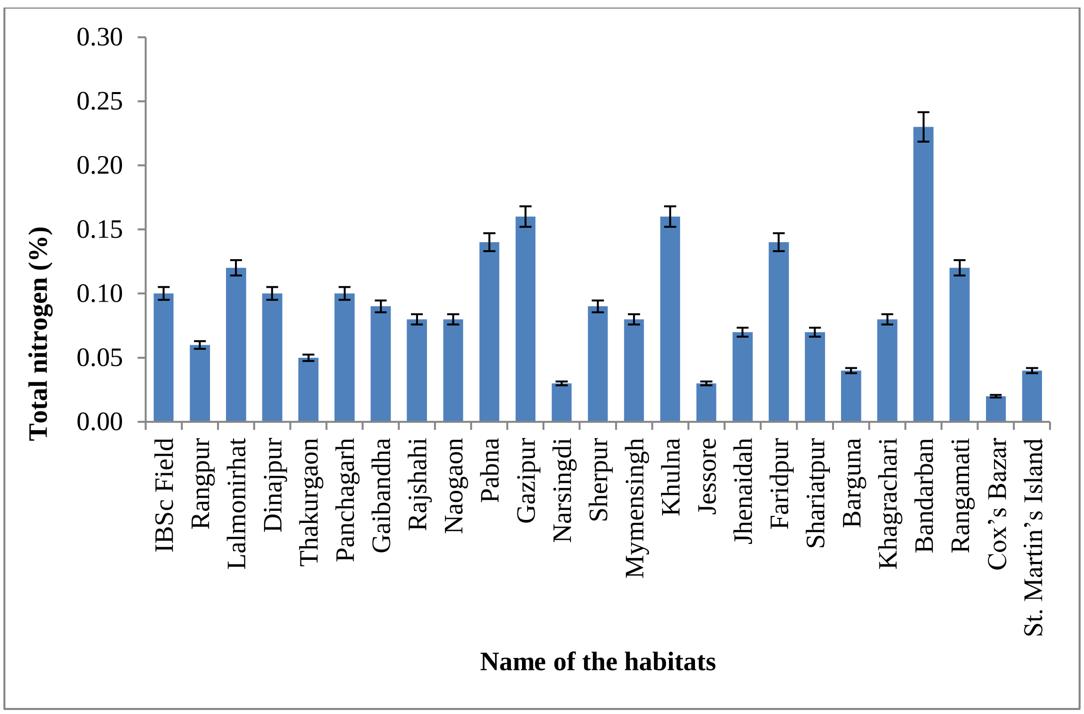


Fig. 37: Determination of total nitrogen in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

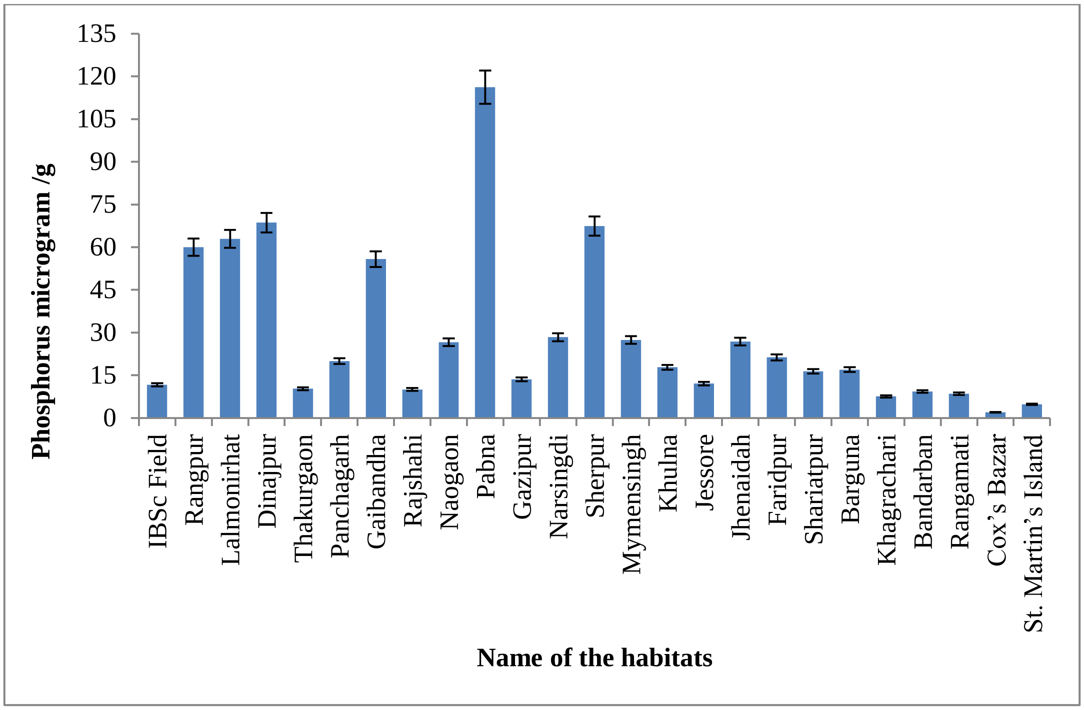


Fig. 38: Determination of phosphorus in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

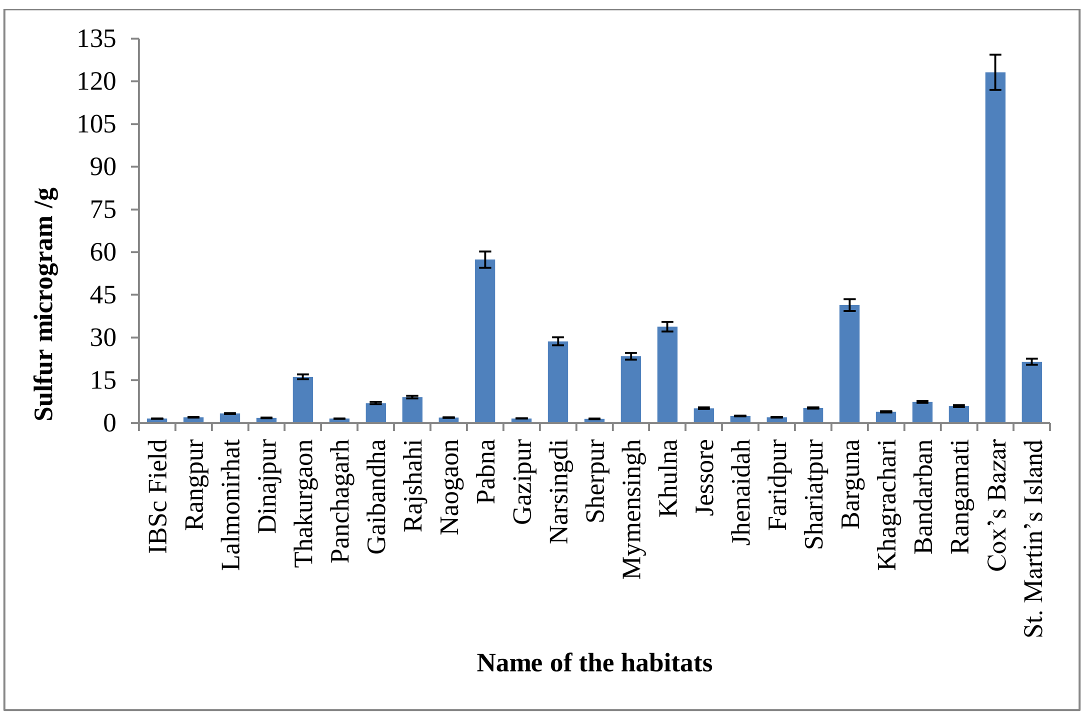


Fig. 39: Determination of sulfur in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

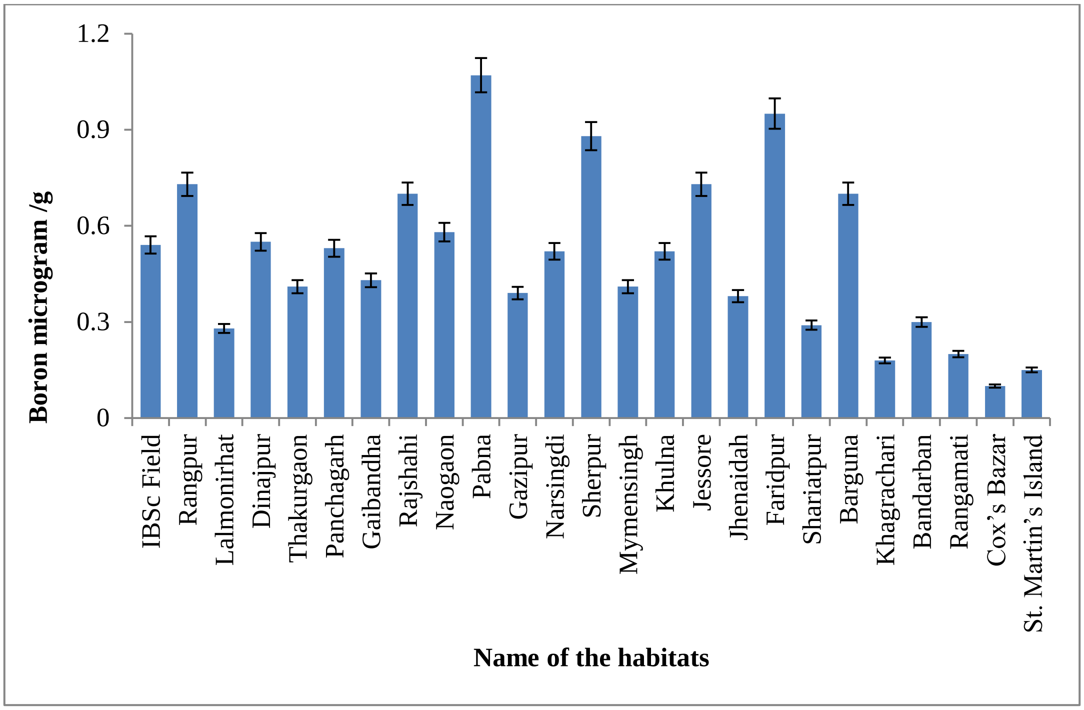


Fig. 40: Determination of boron in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

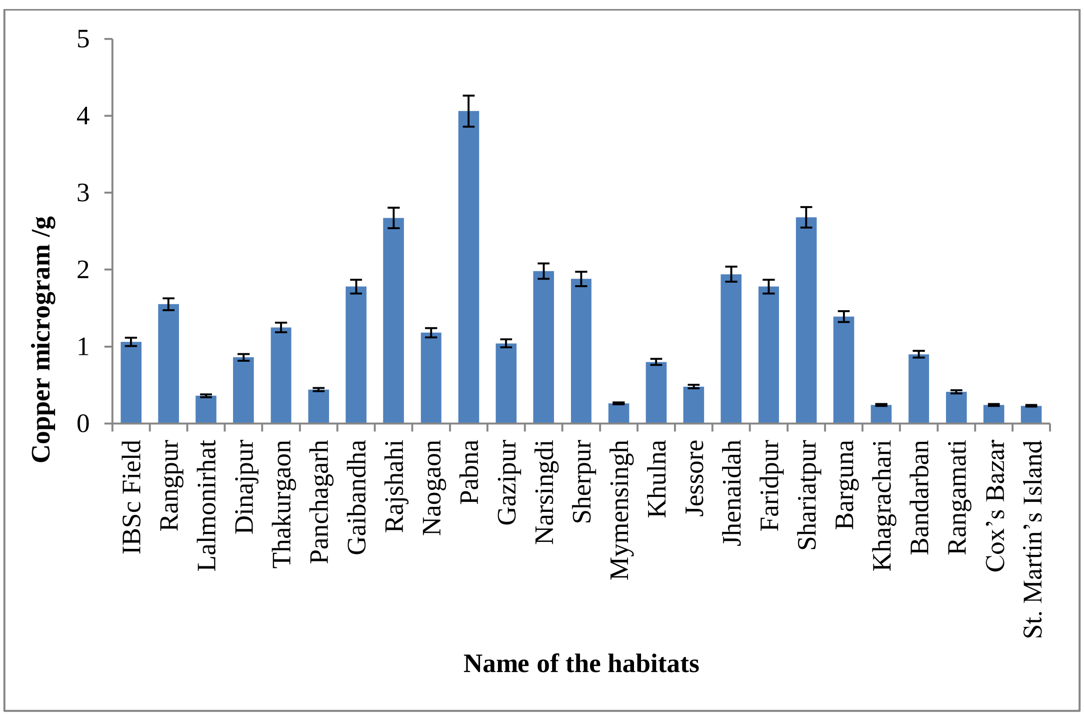


Fig. 41: Determination of copper in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

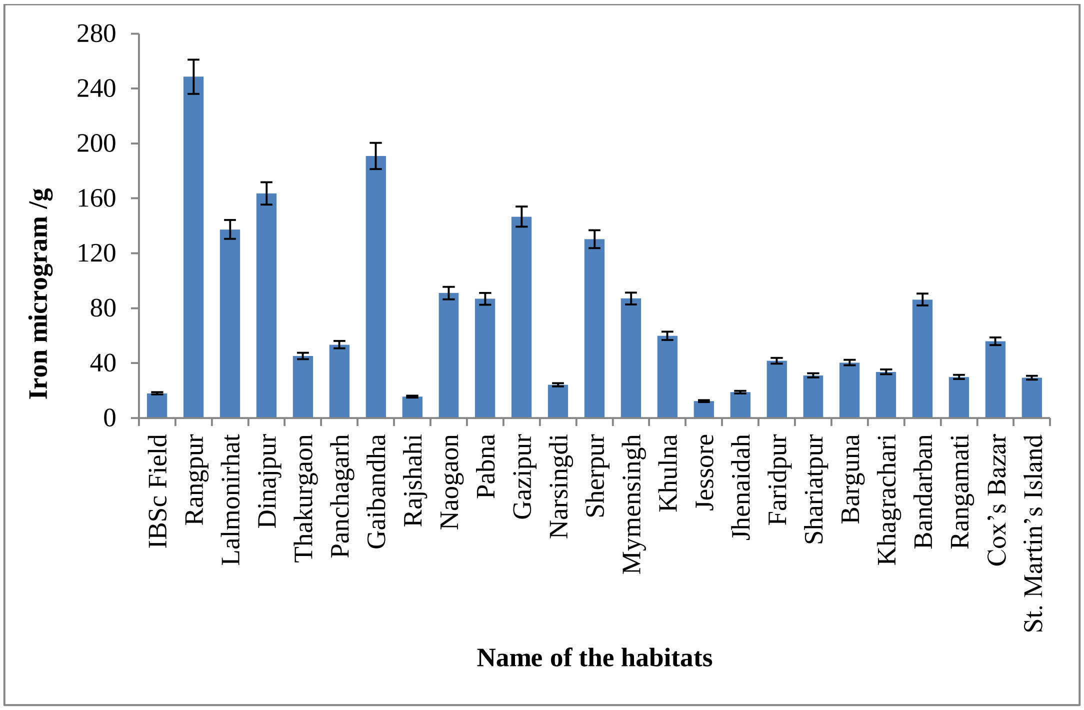


Fig. 42: Determination of iron in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

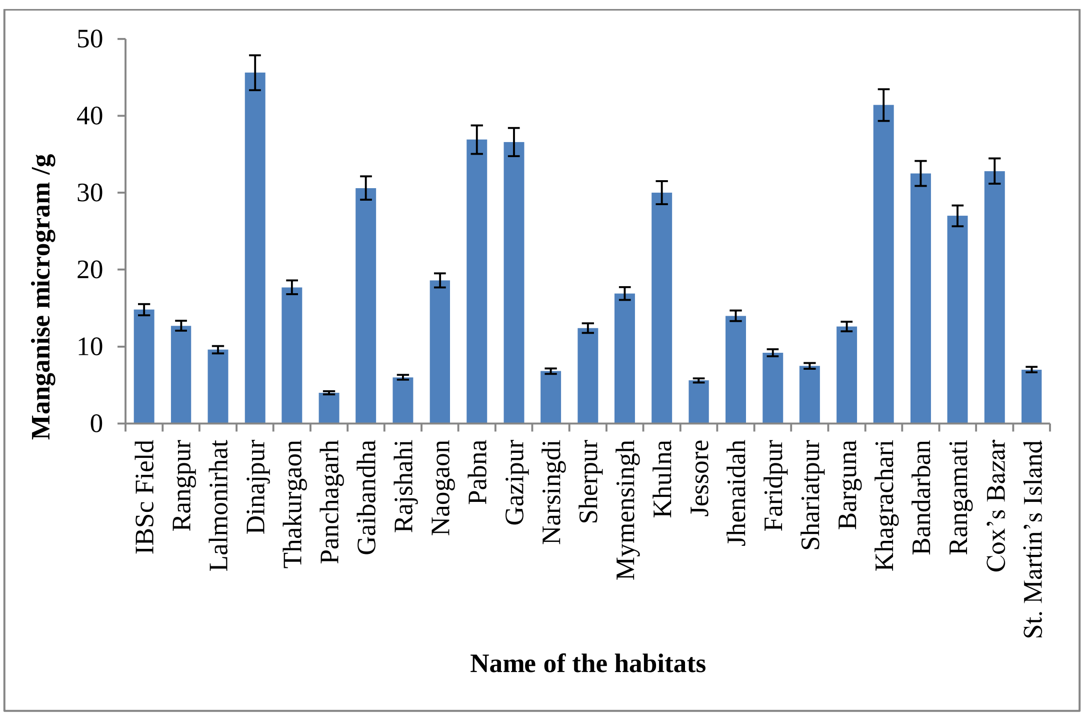


Fig. 43: Determination of manganise in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

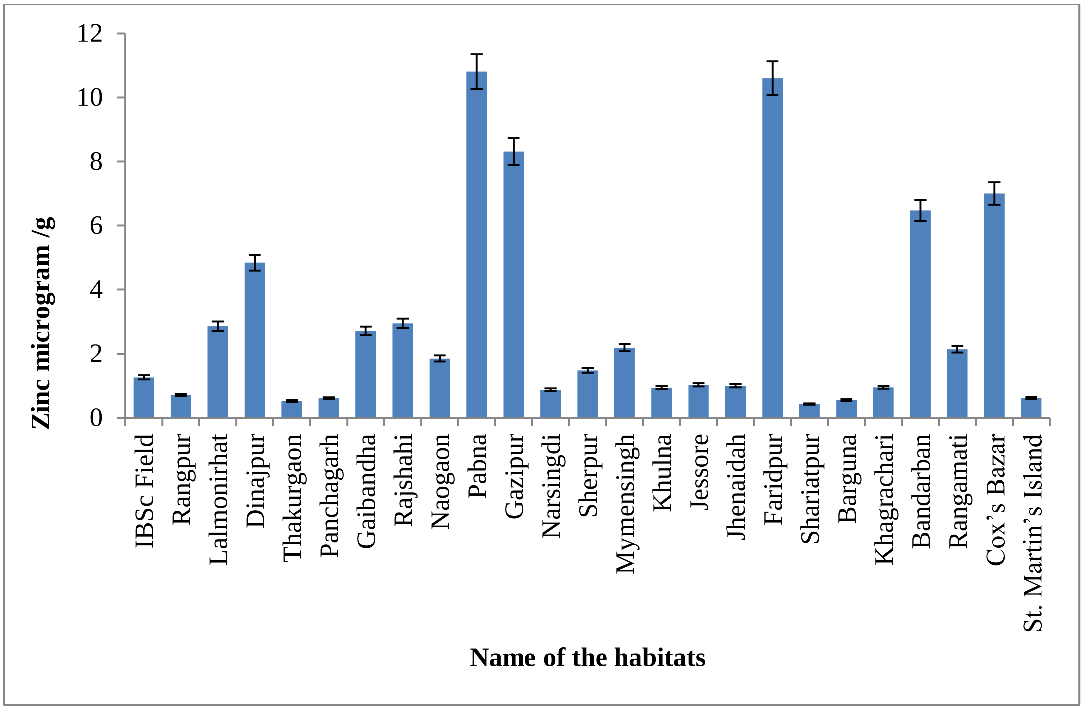


Fig. 44: Determination of zinc in soil for physico-chemical properties through soil analysis of 25 habitats of Bangladesh.

Table 17: Determination of soil for physico-chemical properties through soil analysis of 25 habitats in Bangladesh.

Soil of different habitats	pH	OM (%)	K	Ca	Mg	Total Nitrogen (%)	P	S	B	Cu	Fe	Mn	Zn
			C mol (+)/kg				Microgram/g						
IBSc Field	Alkaline	Low	Low	VH	High	Low	Low	VL	Opt.	VH	VH	VH	Med.
Rangpur	Neutral	Low	Med.	Med.	Med.	VL	VH	VL	High	VH	VH	VH	Low
Lalmonirhat	Acidic	Med.	Opt.	VL	VL	Low	VH	VL	Low	Med.	VH	VH	VH
Dinajpur	Neutral	Low	VH	VH	VH	Low	VH	VL	Opt.	VH	VH	VH	VH
Thakurgaon	Acidic	VL	VH	Med.	Med	VL	Low	Med.	Med.	VH	VH	VH	Low
Panchagarh	Neutral	Low	VH	VH	High	Low	Med.	VL	Opt.	Med.	VH	VH	Low
Gaibandha	Acidic	Low	Low	VH	VH	VL	VH	VL	Med.	VH	VH	VH	VH
Rajshahi	Alkaline	Low	Low	VH	Med	VL	Low	Low	High	VH	VH	VH	VH
Naogaon	Neutral	Low	VH	Opt.	High	VL	Opt.	VL	Opt.	VH	VH	VH	High
Pabna	Alkaline	Med.	High	Opt.	High	Low	VH	VH	VH	VH	VH	VH	VH
Gazipur	Acidic	Med.	Low	VH	VH	Low	Low	VL	Med.	VH	VH	VH	VH
Narsingdi	Alkaline	VL	Low	VH	Low	VL	Opt.	Opt.	Opt.	VH	VH	VH	Low
Sherpur	Neutral	Low	Opt.	Opt.	VH	VL	VH	VL	VH	VH	VH	VH	Opt.
Mymensingh	Alkaline	Low	Med.	VH	VH	VL	Opt.	Opt.	Med.	Low	VH	VH	High
Khulna	Alkaline	Med.	VH	VH	VH	Low	Med.	High	Opt.	VH	VH	VH	Med.
Jessore	Alkaline	VL	Low	VH	VH	VL	Low	VL	High	Opt.	High	VH	Med.
Jhenaidah	Alkaline	Low	Opt.	VH	VH	VL	Opt.	VL	Med.	VH	VH	VH	Med.
Faridpur	Alkaline	Med.	Low	Opt.	VH	Low	Med.	VL	VH	VH	VH	VH	VH
Shariatpur	Alkaline	Low	Opt.	VH	VH	VL	Med.	VL	Low	VH	VH	VH	VL
Barguna	Alkaline	VL	VH	Opt.	High	VL	Med.	VH	High	VH	VH	VH	Low
Khagrachari	Acidic	Low	VH	High	VH	VL	Low	VL	Low	Low	VH	VH	Med.
Bandarban	Neutral	High	VH	VH	High	Med.	Low	VL	Low	VH	VH	VH	VH
Rangamati	Neutral	Med.	VH	VH	VH	Low	Low	VL	Low	Med.	VH	VH	High
Cox's Bazar	Acidic	VL	Opt.	Med.	VH	VL	VL	VH	VL	Low	VH	VH	VH
St. Martin's Island	Neutral	VL	VL	VH	Med.	VL	VL	Med.	VL	Low	VH	VH	Low

OM = Organic matter, K = Potassium, Ca = Calcium, Mg = Magnesium, P = Phosphorus, S = Sulfur, B = Boron, Cu = Copper, Fe = Iron, Mn = Manganise, Zn = Zinc, Opt.= Optimum, Med.= Medium, VH = Very high, VL = Very low.

Soil test values based on critical limits (Table 18) magnitude of physico-chemical properties (Table 17) of the soil of different habitats considered in the present study were interpreted.

Table 18: Interpretation of soil test values based on critical limits (loamy soil for upland crops).

Nutrient element	Very low	Low	Medium	Optimum	High	Very high
N%	≤0.09	0.091-0.18	0.181-0.27	0.271-0.36	0.361-0.45	>0.45
P (µg/g soil)	≤7.50	7.51-15.0	15.1-22.5	22.51-30	30.1-37.5	>37.5
S (µg/g) soil	≤7.50	7.51-15.0	15.1-22.5	22.51-30	30.1-37.5	>37.5
K (meq/100 g)	≤0.09	0.091-0.18	0.181-0.27	0.271-0.36	0.361-0.45	>0.45
Ca (meq/100 g)	≤1.50	1.51-3.0	3.1-4.5	4.51-6.0	6.1-7.5	>7.5
Mg (meq/100 g)	≤0.375	0.376-0.75	0.751-1.125	1.126-1.5	1.51-1.875	>1.875
Cu (µg/g)	≤0.15	0.015-0.3	0.31-0.45	0.451-0.6	0.61-0.75	>0.75
Zn (µg/g)	≤0.45	0.451-0.9	0.91-1.35	1.351-1.8	1.81-2.25	>2.25
Fe (µg/g)	≤3.0	3.1-6.0	6.1-9.0	9.1-12.0	12.1-15.0	>15.0
Mn (µg/g)	≤0.75	0.756-1.5	1.51-2.252	2.256-3.0	3.1-3.75	>3.75
B (µg/g)	≤0.15	0.151-0.3	0.31-0.45	0.451-0.6	0.61-0.75	>0.75

Ref. Fertilization Recommendation Guide (FRG 2012).

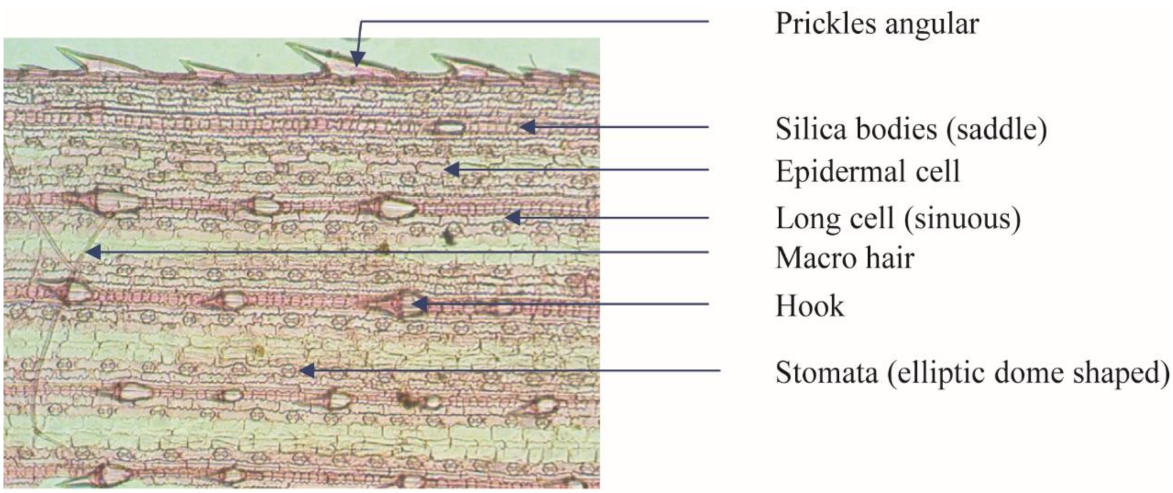
4.3 Leaf epidermal anatomical characters

Findings on leaf epidermal anatomical characteristics in relation to ecotypic adaptation of *Cynodon dactylon* are described as well as illustrated under two subheads as follows:

4.3.1 Qualitative leaf epidermal characters

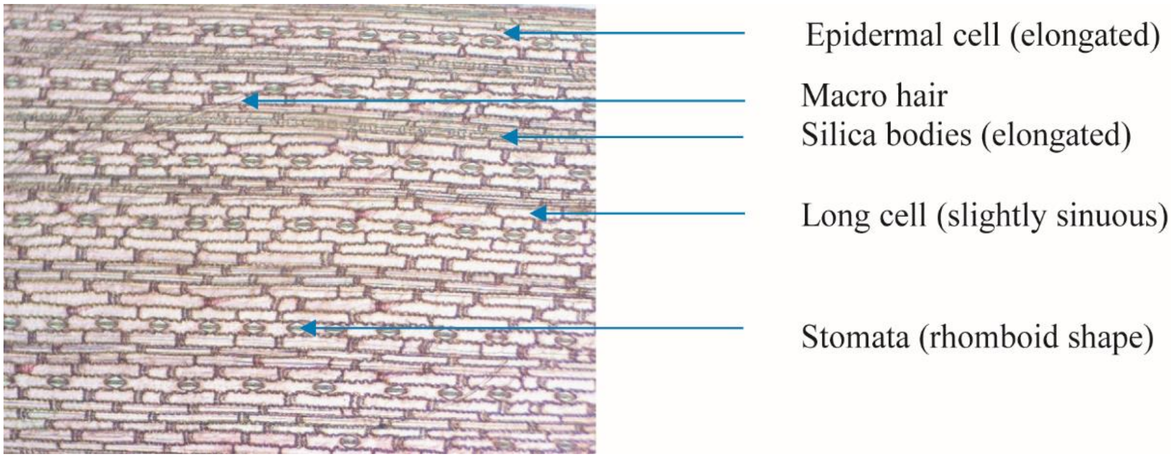
Under this subhead the findings are described based on Figs. 45-48 and Table 19. The leaves were found to be amphistomatic. The stomata were paracytic and dumbbell shaped with two subsidiary cells placed parallel to the pore. Two guard cells were found with two subsidiary cells lateral to the guard cells. Subsidiary cells were found to be dome shaped at both abaxial and adaxial surface of the leaves of all 24 accessions of *Cynodon dactylon.* The epidermal cells were elongated and arranged in vertical rows parallel to the long axis of the leaf. All epidermal cells in *C. dactylon* were found to be sinuous. The long cell margins were found to show almost sinuous in all the accessions except three. In case of the accessions of Barguna, Cox's Bazar and Saint Martin's Island, the long cell margins were found to be slightly sinuous.

Silica bodies were saddle shaped and found in accessions collected from Rangpur, Lalmonirhat, Dinajpur, Thakurgaon, Panchagarh, Gaibandha, Rajshahi, Naogaon, Gazipur and Jhenaidah. Cross shaped silica bodies were found in the accessions of Pabna, Sherpur Mymensingh, Khulna, Shariatpur, Khagrachari, Bandarban and Rangamati. Both saddle and cross shaped silica bodies were found in the durba grass of Narsingdi, Jessore and Faridpur. Horizontally elongated silica bodies were found in the studied material of Barguna, Cox's Bazar and Saint Martin's Island. Macrohairs were present in all the accessions of *C. dactylon* and microhairs were not found in any of the accessions abaxially or adaxially. Prickles angular were found as pointed form at the tip in all the accessions except that from Barguna, Cox's Bazar and Saint Martin's Island. Similar results were also found in case of hooks, which were not found in accessions of *C. dactylon* from Barguna, Cox's Bazar and Saint Martin's Island but in other 21 accessions hooks were found to be present.



Thakurgaon (non-saline habitat)

Fig. 45: Foliar epidermal structure of *C. dactylon* collected from non-saline habitats of Bangladesh.



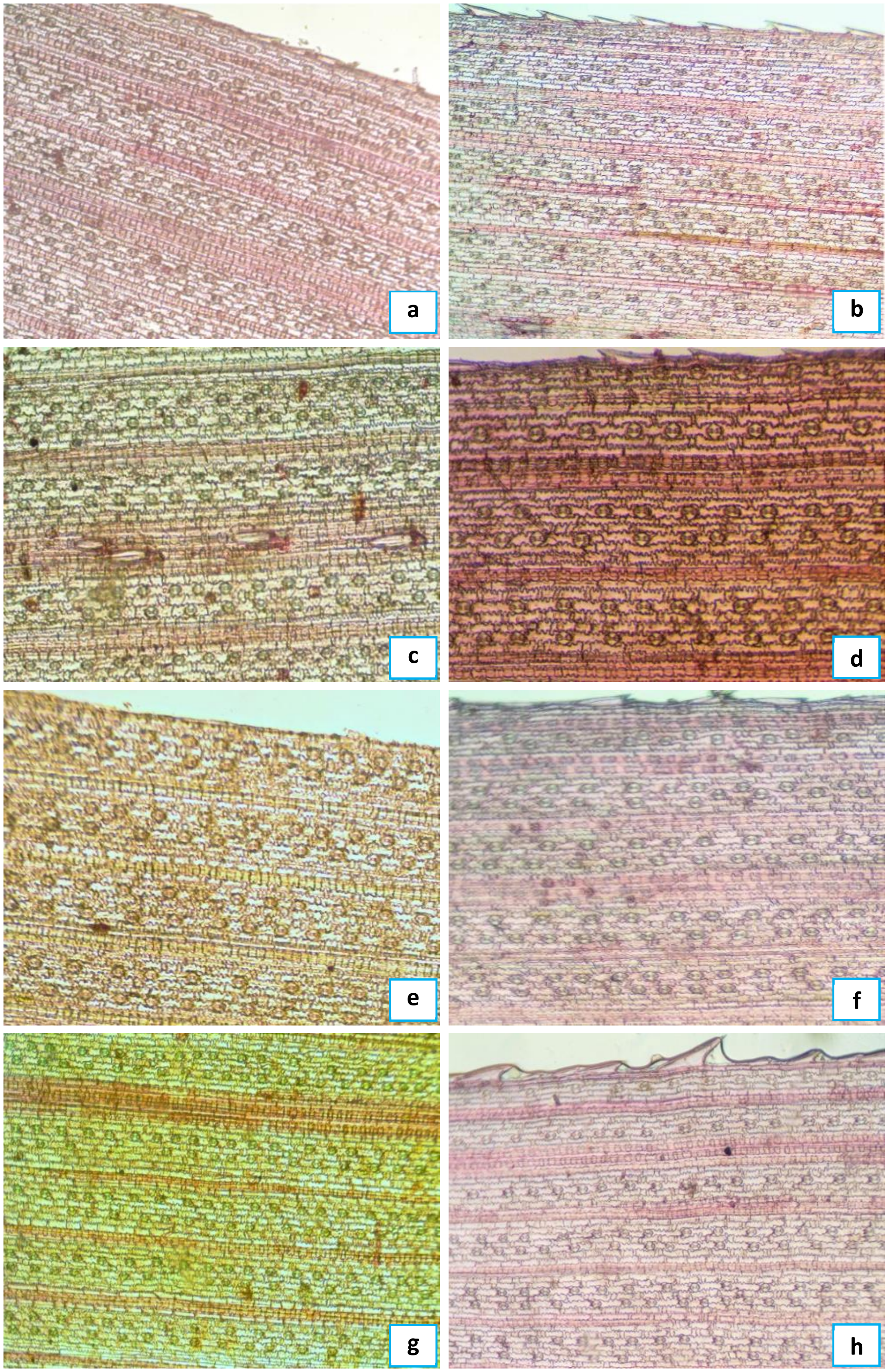
Cox's Bazar (saline habitat)

Fig. 46: Foliar epidermal structure of *C. dactylon* collected from saline habitats of Bangladesh.

Table 19: Qualitative leaf epidermal characteristics on both abaxial and adaxial surfaces of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Stomata type	Shape of subsidiary cells	Long cell margins	Types of silica bodies	Macro hair	Micro hair	Prickles angular	Hook
1	Rangpur	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
2	Lalmonirhat	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
3	Dinajpur	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
4	Thakurgaon	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
5	Panchagarh	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
6	Gaibandha	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
7	Rajshahi	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
8	Naogaon	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
9	Pabna	Paracytic	Dome	Sinuous	Cross	+	-	+	+
10	Gazipur	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
11	Narsingdi	Paracytic	Dome	Sinuous	Saddle, Cross	+	-	+	+
12	Sherpur	Paracytic	Dome	Sinuous	Cross	+	-	+	+
13	Mymensingh	Paracytic	Dome	Sinuous	Cross	+	-	+	+
14	Khulna	Paracytic	Dome	Sinuous	Cross	+	-	+	+
15	Jessore	Paracytic	Dome	Sinuous	Saddle, Cross	+	-	+	+
16	Jhenaidah	Paracytic	Dome	Sinuous	Saddle	+	-	+	+
17	Faridpur	Paracytic	Dome	Sinuous	Saddle, Cross	+	-	+	+
18	Shariatpur	Paracytic	Dome	Sinuous	Cross	+	-	+	+
19	Barguna	Paracytic	Dome	Slightly sinuous	Horizontally elongated	+	-	-	-
20	Khagrachari	Paracytic	Dome	Sinuous	Cross	+	-	+	+
21	Bandarban	Paracytic	Dome	Sinuous	Cross	+	-	+	+
22	Rangamati	Paracytic	Dome	Sinuous	Cross	+	-	+	+
23	Cox's Bazar	Paracytic	Dome	Slightly sinuous	Horizontally elongated	+	-	-	-
24	St. Martin’s Island	Paracytic	Dome	Slightly sinuous	Horizontally elongated	+	-	-	-

+ = Present, - = Absent



Figs. 47 (a-h): Foliar epidermal structure on abaxial surface of *C. dactylon* accessions collected from different habitats; a) Rangpur, b) Lalmonirhat, c) Dinajpur, d) Thakurgaon, e) Panchagarh, f) Gaibandha. g) Rajshahi, h) Naogaon.

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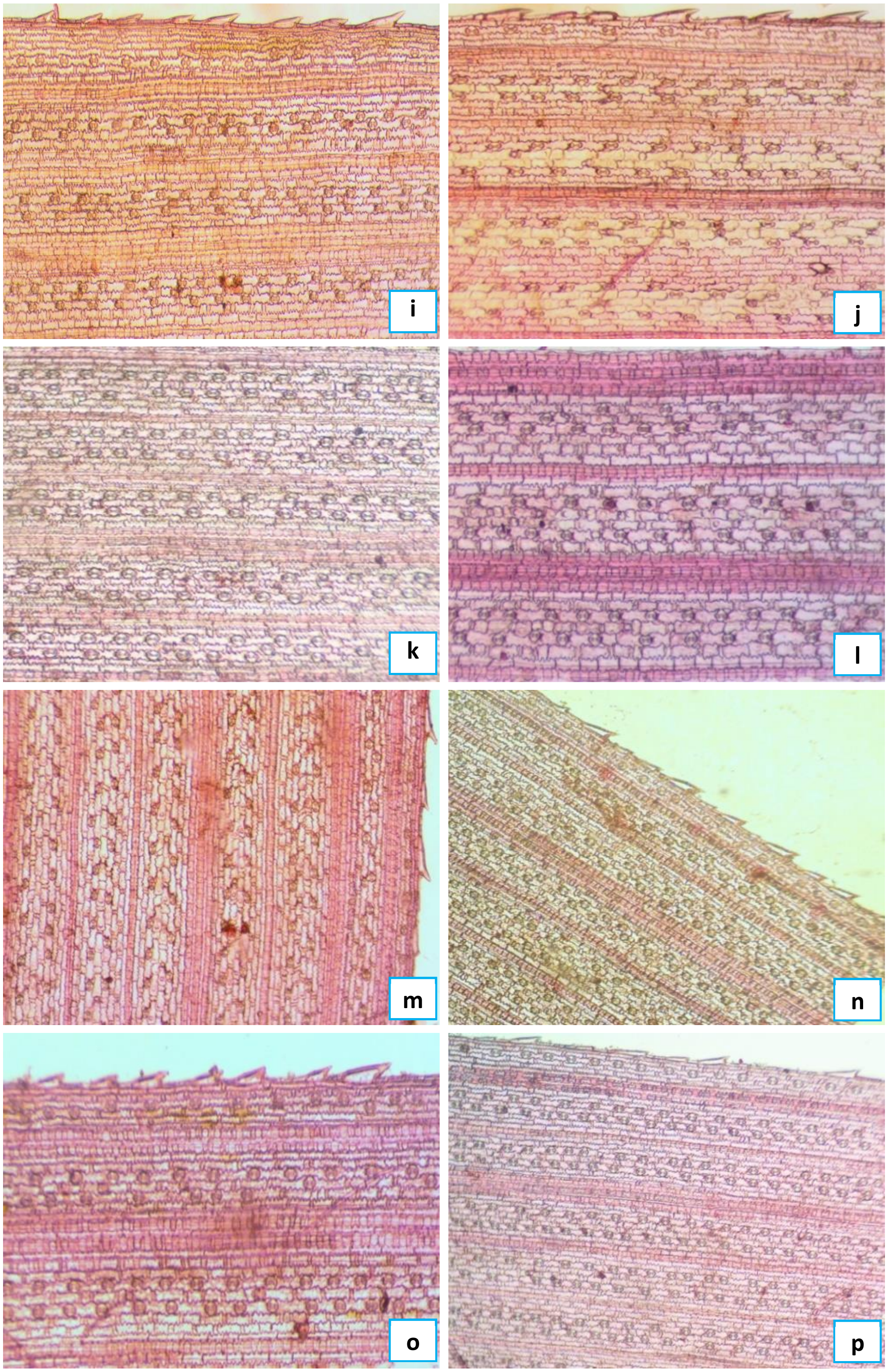

Figs. 47 (i-p): Foliar epidermal structure on abaxial surface of *C. dactylon* accessions collected from different habitats; i) Pabna, j) Gazipur, k) Narsingdi, l) Sherpur, m) Mymensingh, n) Khulna, o) Jessore, p) Jhenaidah.

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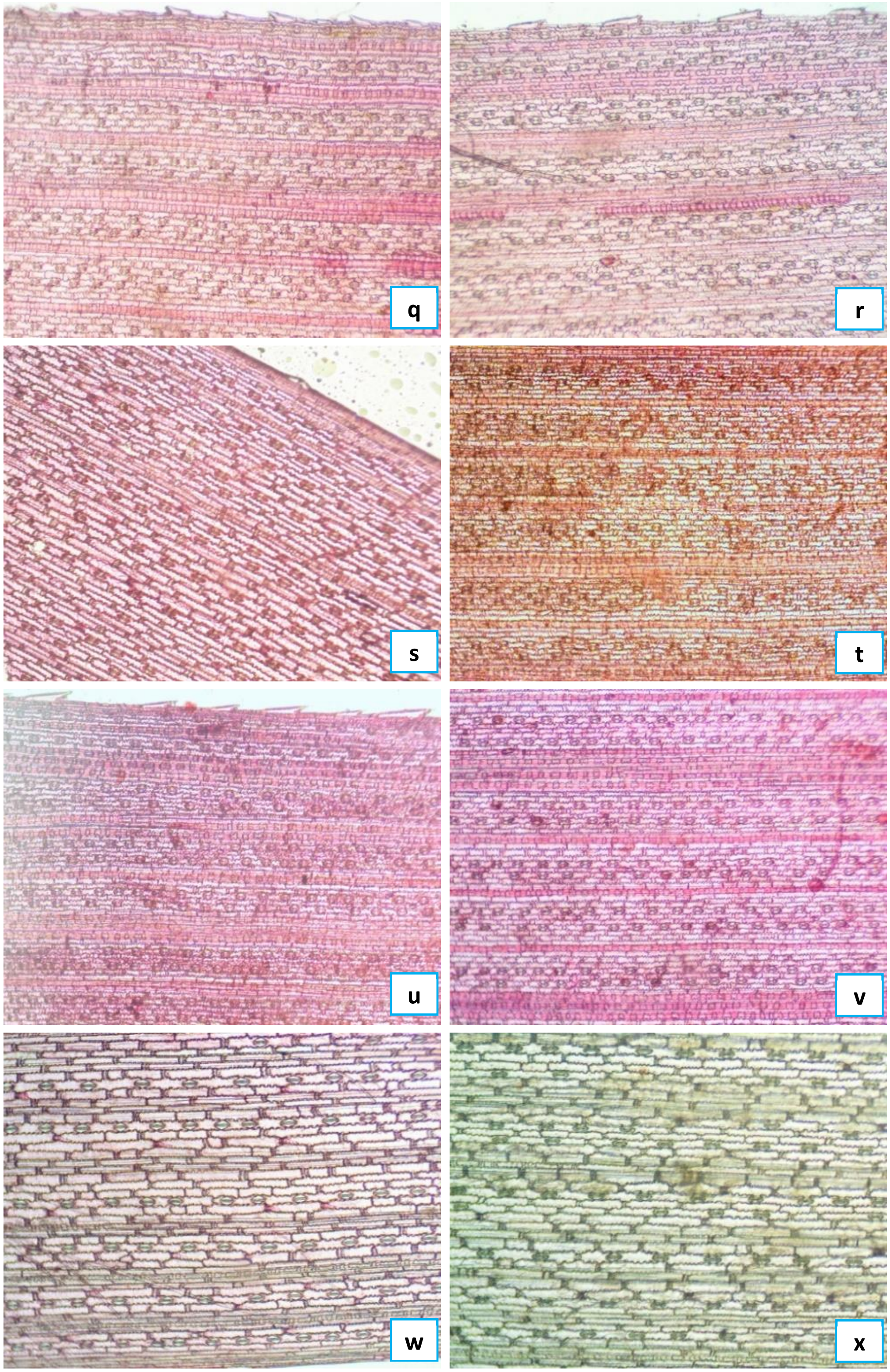
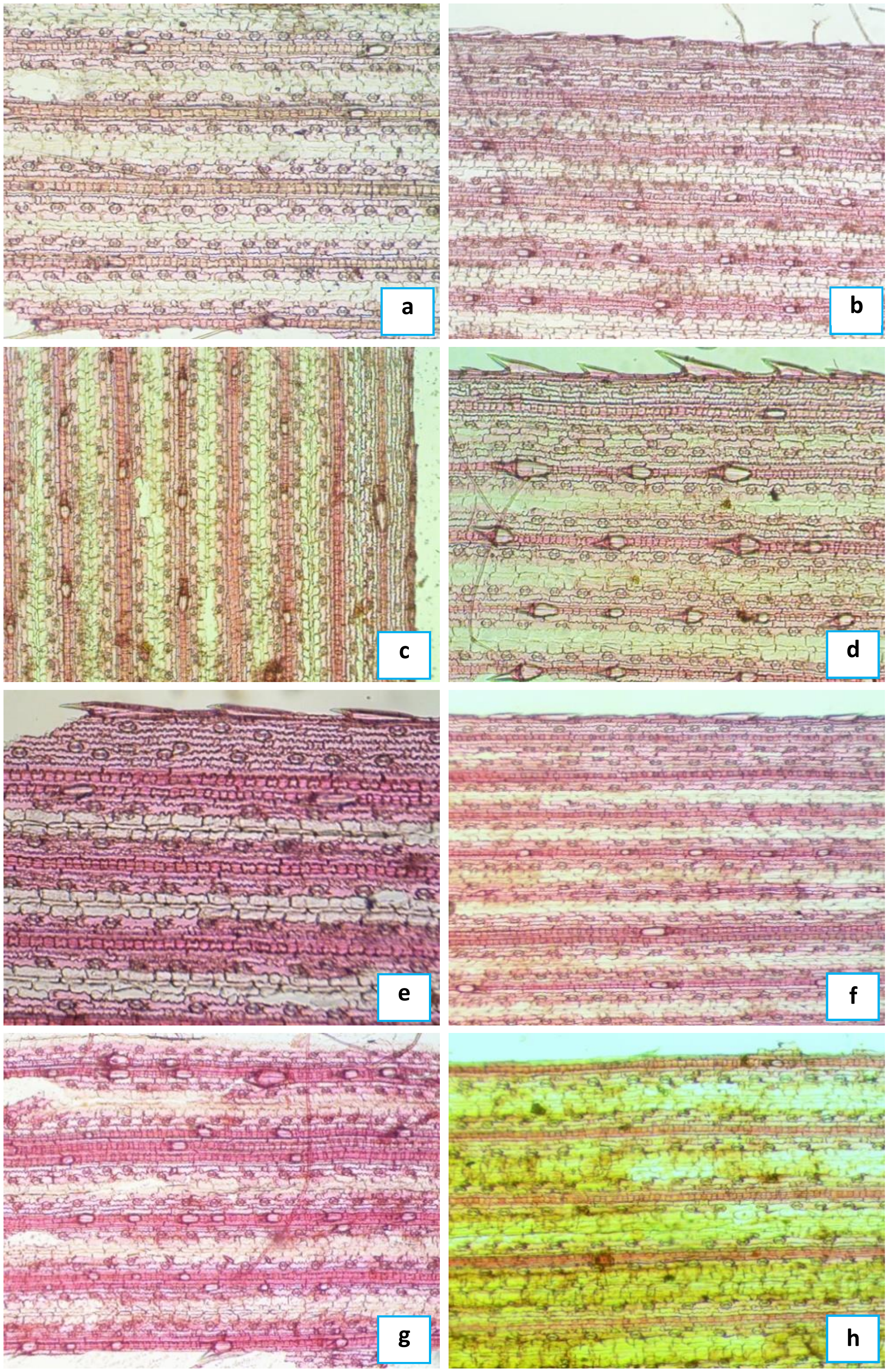


Figs. 47 (q-x): Foliar epidermal structure on abaxial surface of *C. dactylon* accessions collected from different habitats; q) Faridpur, r) Shariatpur, s) Barguna, t) Khagrachari, u) Bandarban, v) Rangamati, w) Cox's Bazar, x) Saint Martin's Island.



Figs. 48 (a-h): Foliar epidermal structure on adaxial surface of *C. dactylon* accessions collected from different habitats; a) Rangpur, b) Lalmonirhat, c) Dinajpur, d) Thakurgaon, e) Panchagarh, f) Gaibandha, g) Rajshahi, h) Naogaon.

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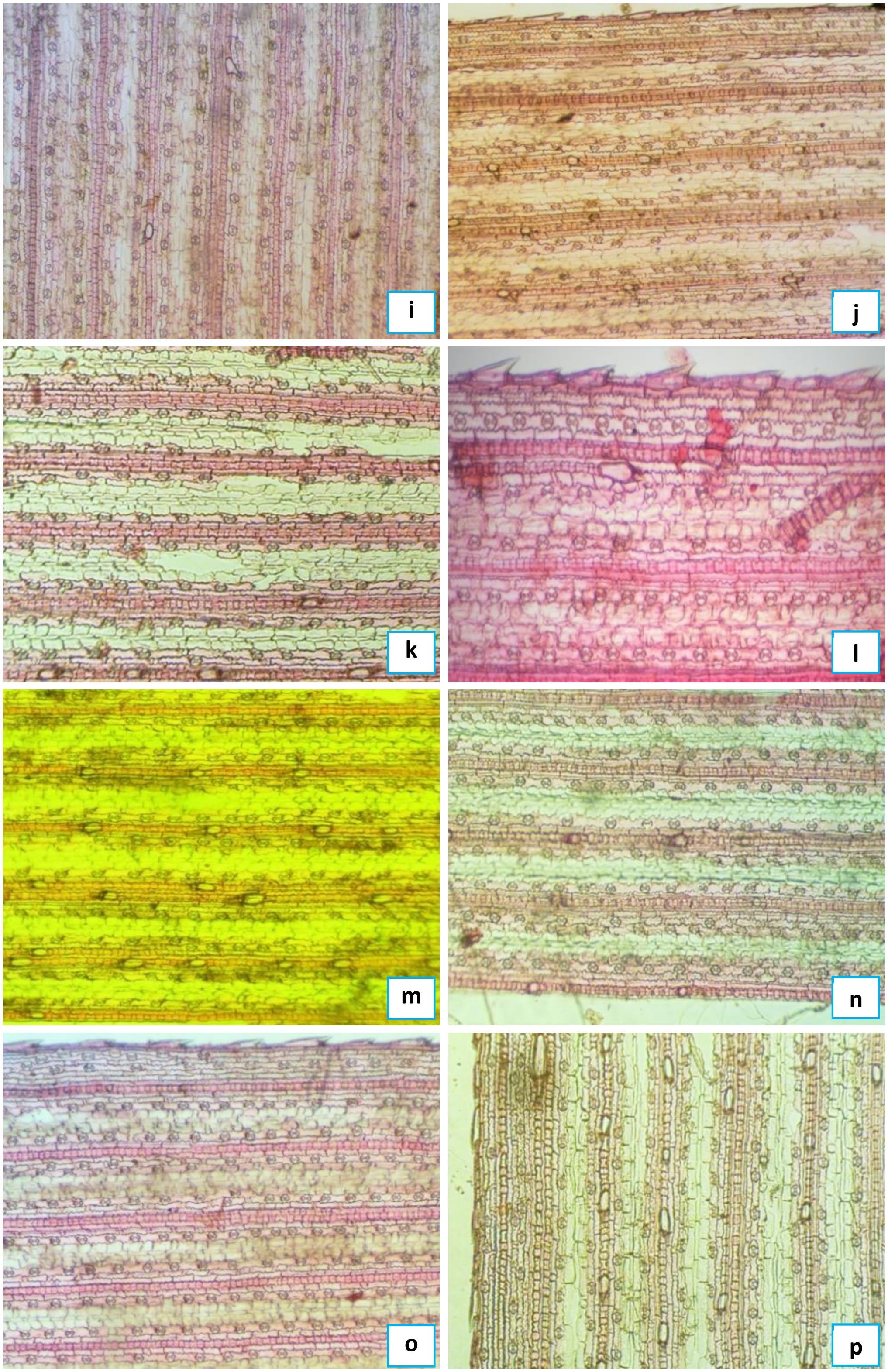

Figs. 48 (i-p): Foliar epidermal structure on adaxial surface of *C. dactylon* accessions collected from different habitats; i) Pabna, j) Gazipur, k) Narsingdi, l) Sherpur, m) Mymensingh, n) Khulna, o) Jessore, p) Jhenaidah.

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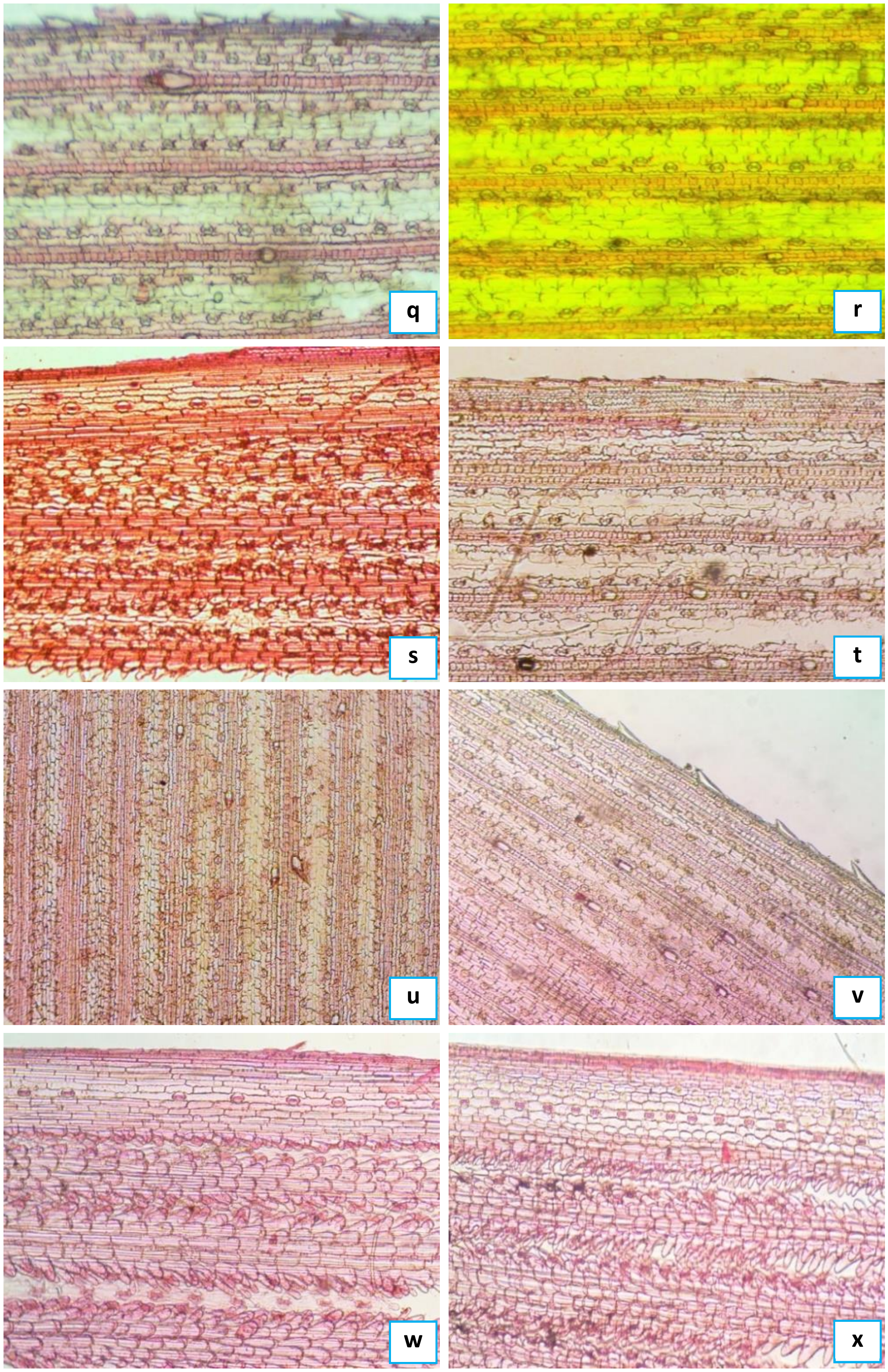


Figs. 48 (q-x): Foliar epidermal structure on adaxial surface of *C. dactylon* accessions collected from different habitats; q) Faridpur, r) Shariatpur, s) Barguna, t) Khagrachari, u) Bandarban, v) Rangamati, w) Cox's Bazar, x) Saint Martin's Island.

4.3.2 Quantitative leaf epidermal characters

Results regarding quantitative leaf epidermal characters are described here as shown in Figs. 47-48 and presented in Tables 20-26. Highest mean value (7.72) for long cell numbers per mm^2 was found in accession collected from Khulna and lowest mean value (2.14) was found in case of Saint Martin's Island in abaxial surface of leaves (Table 20). Highest mean value (9.65) for long cell numbers per mm^2 was found in accession collected from Shariatpur and lowest mean value (3.31) was found in Barguna in adaxial surface of the leaves (Table 21). Long cell length was highest (110.26 µm) in case of Saint Martin's Island and lowest (32.20 µm) in case of Cox's Bazar in abaxial surface of the leaves (Table 20). Long cell length was highest (90.76 µm) in case of Barguna and lowest (31.39 µm) in case of Shariatpur in adaxial surface of the leaves (Table 21). Long cell width was highest (8.82 µm) in case of Saint Martin's Island and lowest (4.03 µm) in case of Cox's Bazar in abaxial surface the leaves (Table 20). Long cell width was highest (12.33 µm) in Cox's Bazar and lowest (3.40 µm) in case of Rangamati in adaxial surface of the leaves (Table 21).

Table 20: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Abaxial surface of leaves		
		Long cell no. per mm^2	Long cell length (μm)	Long cell width (μm)
		Mean±SE	Mean±SE	Mean±SE
1	Rangpur	7.66 ±0.15cd	33.91 ± 2.39hi	6.43 ±0.26abcd
2	Lalmonirhat	5.69 ±0.13hijk	42.95 ± 2.39efg	6.42 ±0.17abcd
3	Dinajpur	5.76 ±0.10ghij	32.29 ± 2.26i	8.02 ±0.17ab
4	Thakurgaon	4.75 ±0.08m	40.29 ± 2.30efgh	8.02 ± 0.22a
5	Panchagarh	5.17 ±0.13jklm	42.29 ± 2.26efg	8.03 ± 0.26ab
6	Gaibandha	6.19 ±0.08efghi	39.94 ± 1.61efgh	8.02 ± 0.13abc
7	Rajshahi	5.16 ± 0.10jklm	39.86 ± 2.00efgh	6.69 ± 0.13abcd
8	Naogaon	7.07 ±0.08bc	40.20±1.56efgh	6.42±0.17abcd
9	Pabna	5.56±0.10ijkl	38.88±1.65fghi	8.03±0.26abcd
10	Gazipur	7.09±0.13bc	44.29±2.30def	4.43±0.22cd
11	Narsingdi	5.01±0.13lm	40.23±1.78efgh	8.02±0.17abc
12	Sherpur	6.37±0.13defg	40.37±1.61efgh	7.02±0.13abcd
13	Mymensingh	5.10±0.15klm	32.95±2.21i	7.70±0.13abcd
14	Khulna	7.72±0.10a	58.90±2.00c	6.82±0.13abcd
15	Jessore	6.09±0.13fghi	49.63±2.30d	7.22±0.13abcd
16	Jhenaidah	6.61±0.13cdef	37.64±2.34ghi	4.83±0.22d
17	Faridpur	5.04±0.10klm	37.02±1.74ghi	6.17±0.26abcd
18	Shariatpur	6.01±0.13fghi	45.87±2.08de	8.03±0.26a
19	Barguna	2.57±0.13n	96.28±2.17b	8.43±0.26ab
20	Khagrachari	7.62±0.15ab	33.59±2.00hi	7.48±0.13abcd
21	Bandarban	6.32±0.10efgh	34.31±2.43hi	6.68±0.26abcd
22	Rangamati	6.81±0.13cde	34.22±1.69hi	6.70±0.26bcd
23	Cox's Bazar	2.47±0.08n	32.20±1.56i	4.03±0.22cd
24	St. Martin’s Island	2.14±0.15n	110.26±2.00a	8.82±0.17a

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

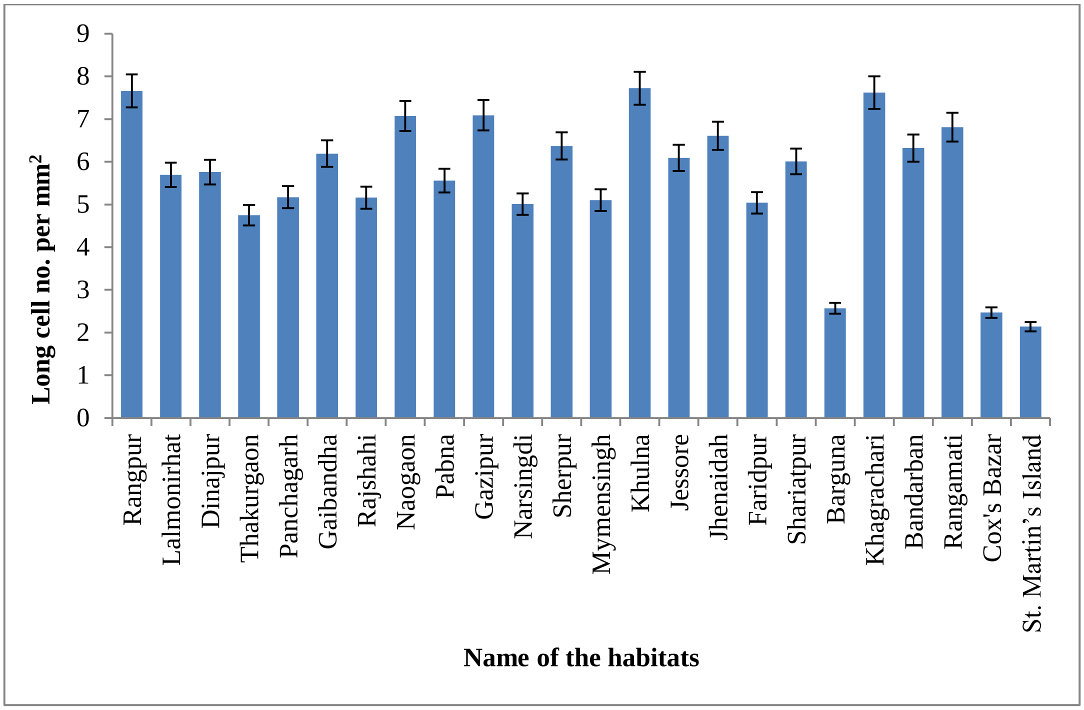


Fig. 49: Long cell number on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

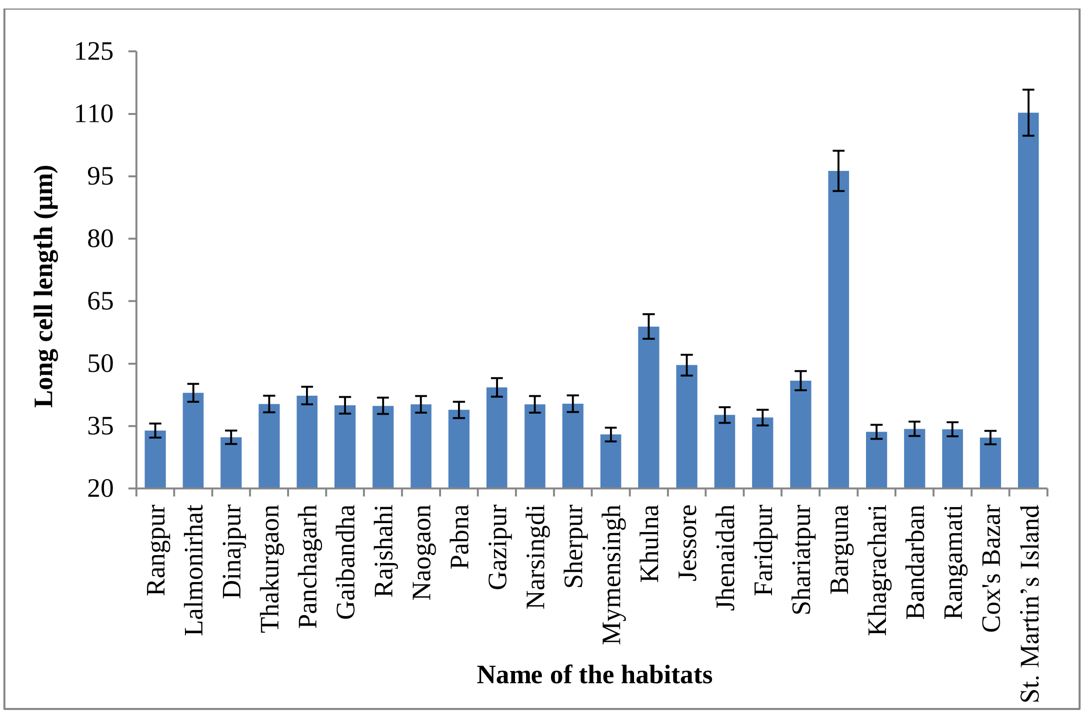


Fig. 50: Long cell length on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

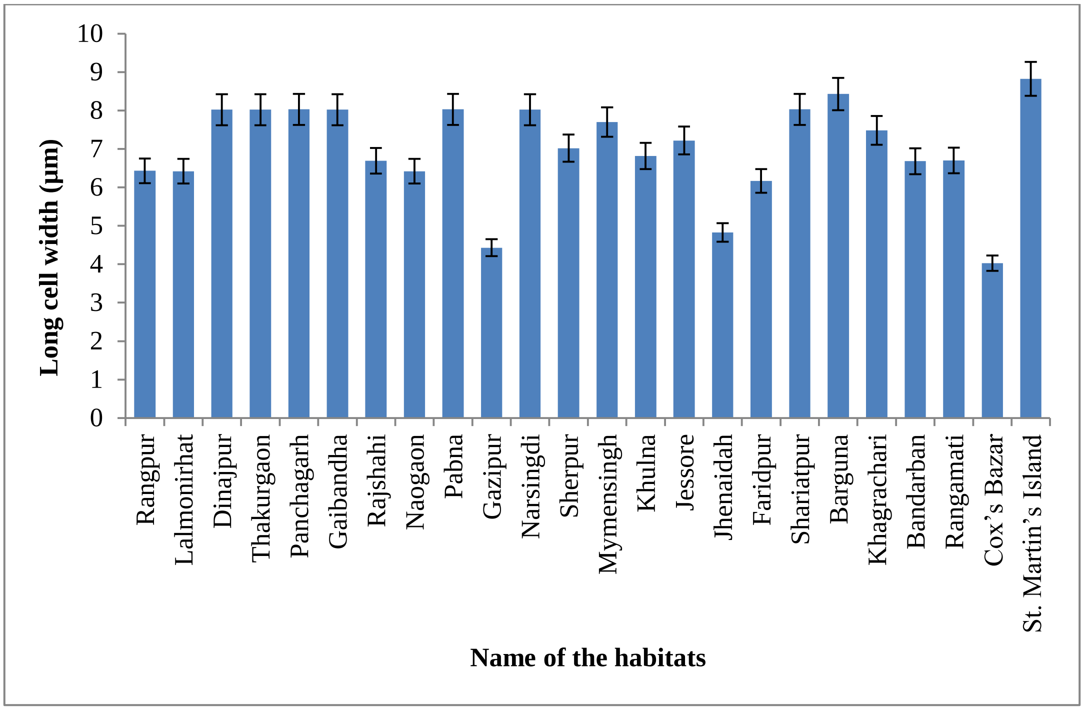


Fig. 51: Long cell width on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

Table 21: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Adaxial surface of leaves		
		Long cell no. per mm^2	Long cell length (µm)	Long cell width (µm)
		Mean±SE	Mean±SE	Mean±SE
1	Rangpur	5.88±0.50efghij	37.72±0.82mn	6.63±0.42cd
2	Lalmonirhat	5.33±0.52hij	50.12±0.70g	7.73±0.24bc
3	Dinajpur	5.15±0.20ij	48.85±0.76g	7.39±0.34bc
4	Thakurgaon	9.20±0.44a	72.69±0.61b	7.68±0.44bc
5	Panchagarh	9.01±0.30a	41.98±0.87jk	6.53±0.24cd
6	Gaibandha	8.46±0.32ab	41.16±0.67kl	7.40±0.36bc
7	Rajshahi	6.81±0.60cdef	36.05±0.76n	7.51±0.56bc
8	Naogaon	7.50±0.40bc	44.09±0.79hi	6.95±0.56bc
9	Pabna	7.43±0.42bc	55.46±0.73ef	6.72±0.58cd
10	Gazipur	6.32±0.58cdefghi	33.06±0.58o	7.73±0.24bc
11	Narsingdi	7.17±0.52bcde	43.55±0.79hij	6.61±0.38cd
12	Sherpur	4.61±0.38j	54.26±0.82f	5.39±0.34de
13	Mymensingh	5.75±0.20fghij	49.95±0.84g	7.10±0.54bc
14	Khulna	6.75±0.34cdefg	64.69±0.61c	4.62±0.40ef
15	Jessore	6.34±0.54cdefghi	56.66±0.58e	6.63±0.42cd
16	Jhenaidah	6.60±0.28cdefgh	59.53±0.79d	6.94±0.26bc
17	Faridpur	5.92±0.36defghij	42.03±0.61ijk	6.64±0.44cd
18	Shariatpur	9.65±0.52a	31.39±0.64p	4.92±0.22e
19	Barguna	3.31±0.20k	90.76±0.67a	8.28±0.50b
20	Khagrachari	5.43±0.42ghij	54.45±0.76f	7.92±0.58bc
21	Bandarban	6.92±0.36cdef	44.98±0.87h	7.02±0.40bc
22	Rangamati	6.62±0.40cdefgh	39.32±0.58lm	3.40±0.36f
23	Cox's Bazar	3.33±0.24k	72.95±0.84b	12.33±0.60a
24	St. Martin’s Island	7.28±0.44bcd	58.98±0.87d	7.85±0.60bc

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

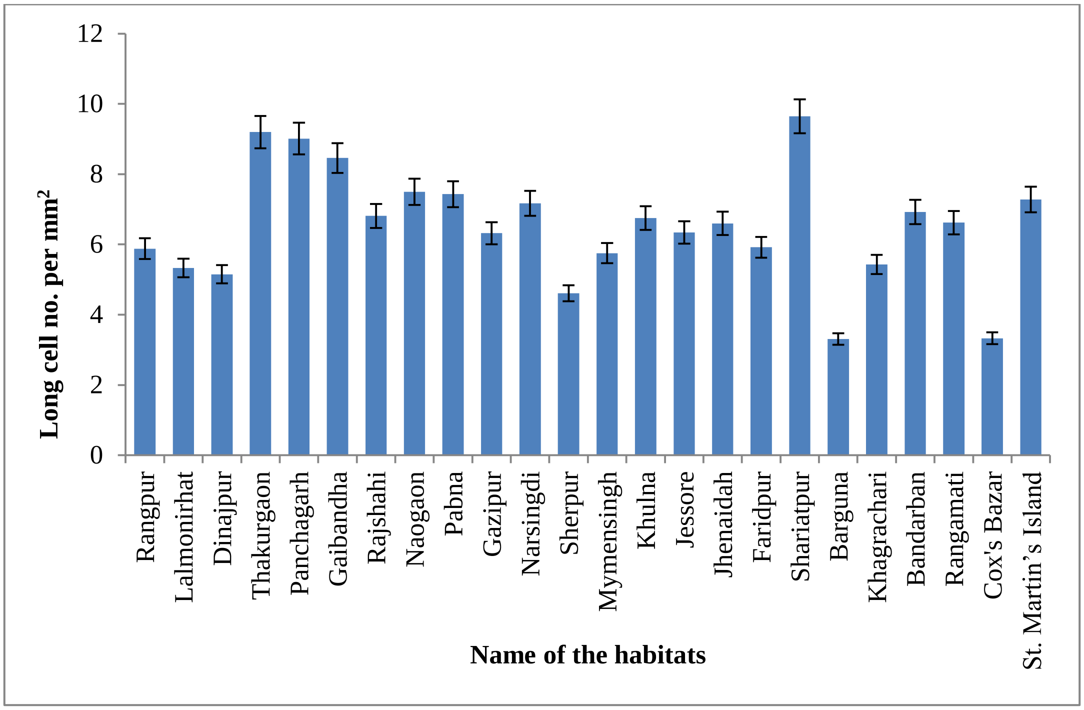


Fig. 52: Long cell number on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

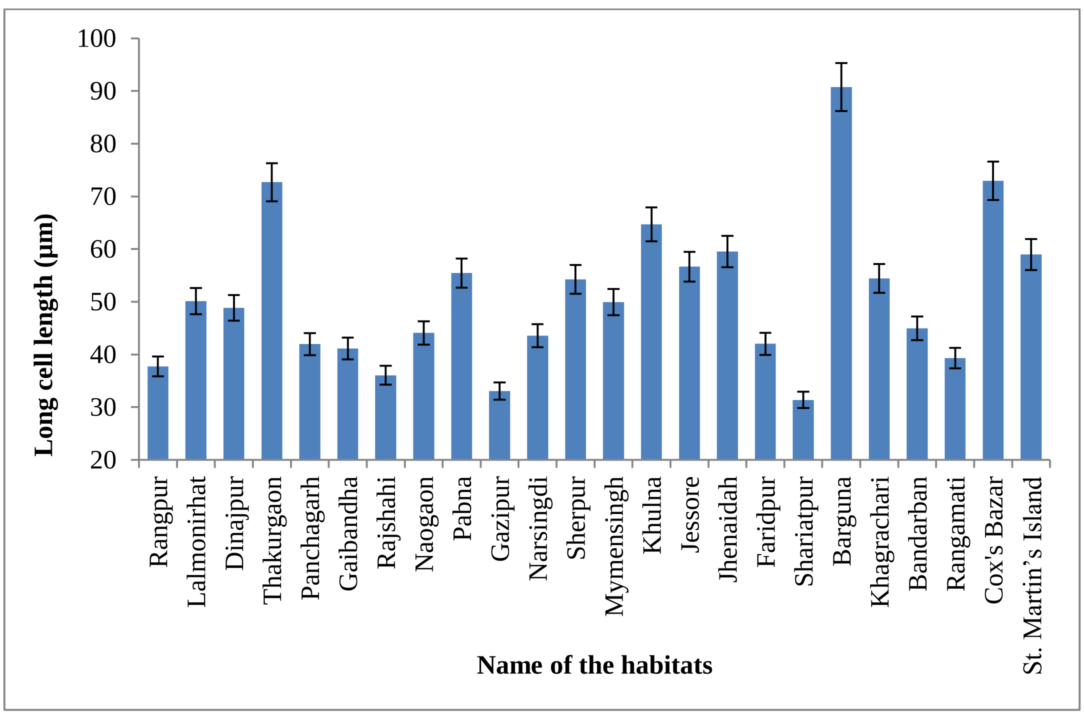


Fig. 53: Long cell length on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

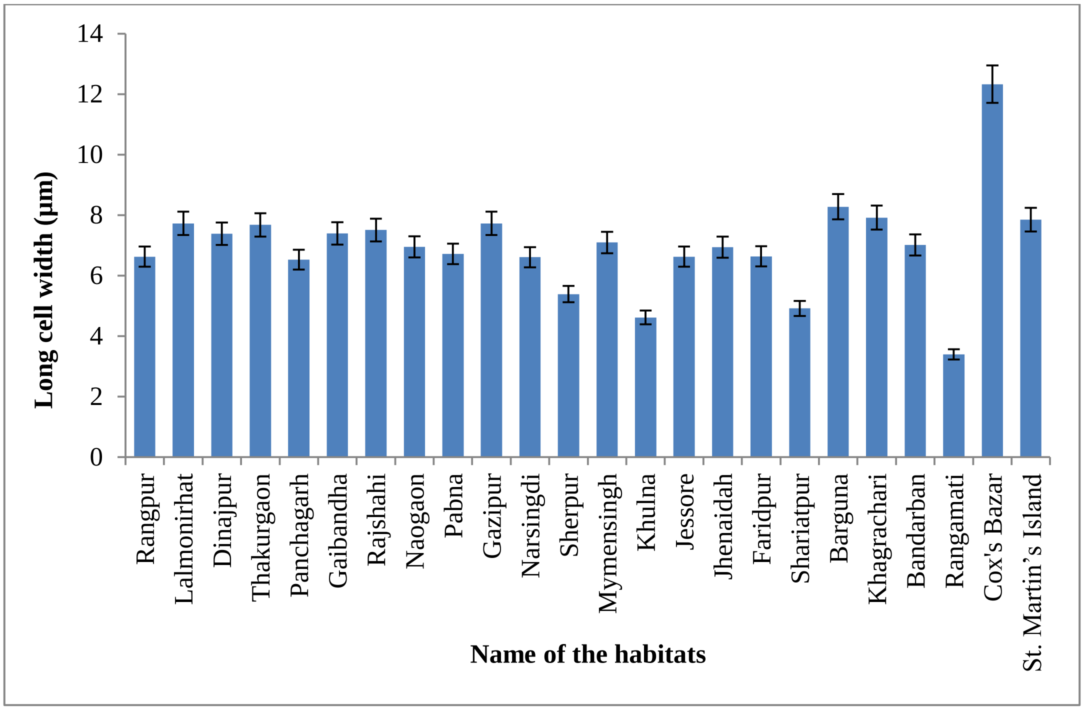


Fig. 54: Long cell width on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

Stomatal frequency was highest (5.56 mm^{-2}) in case of Gazipur and lowest (1.78 mm^{-2}) in case of Saint Martin's Island in abaxial surfaces of the leaves (Table 22). Stomatal frequency was highest (6.61 mm^{-2}) in case of Shariatpur and lowest (3.02 mm^{-2}) in case of Barguna in adaxial surfaces of the leaves (Table 23). Epidermal cell number per mm^2 was highest (23.89) in case of Gaibandha and lowest (4.5) in case of Saint Martin's Island in abaxial surface of the leaves (Table 22). Epidermal cell number per mm^2 was highest (19.99) in case of Shariatpur and lowest (9.28) in case of Barguna in adaxial surface of the leaves (Table 23). Stomatal index was highest (28.43 %) in case of Jessore and lowest (25.36 %) in case of Jhenaidah in abaxial surface of the leaves (Table 22). Stomatal index was highest (28.76 %) in case of Khulna and lowest (24.97 %) in case of Jhenaidah in adaxial surface of the leaves (Table 23).

Table 22: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Abaxial surface of leaves		
		Stomatal frequency	Epidermal cell no. per mm^2	Stomatal index
		Mean±SE	Mean±SE	Mean±SE
1	Rangpur	4.52±0.10fghi	11.92±0.95cdef	26.36±0.49ab
2	Lalmonirhat	4.59±0.08efgh	13.28±0.91bcdef	26.34±0.62ab
3	Dinajpur	4.82±0.15cdef	12.61±1.30cdef	26.47±0.36ab
4	Thakurgaon	3.71±0.08k	10.81±1.30ef	25.46±0.32ab
5	Panchagarh	4.26±0.15hij	9.60±1.21f	28.00±0.91ab
6	Gaibandha	4.29±0.13hij	23.89±0.69a	25.40±0.82ab
7	Rajshahi	4.03±0.08jk	12.37±1.30cdef	25.61±0.88ab
8	Naogaon	4.93±0.13cde	12.59±1.48cdef	26.40±0.61ab
9	Pabna	4.82±0.15cdef	13.78±0.78bcde	25.78±0.52ab
10	Gazipur	5.56±0.10a	16.21±1.04b	25.80±0.61ab
11	Narsingdi	4.19±0.08ij	11.33±1.35def	26.25±0.30ab
12	Sherpur	4.32±0.10ghij	13.29±1.30bcdef	25.67±0.96ab
13	Mymensingh	4.77±0.13cdef	10.61±1.04ef	27.42±1.40ab
14	Khulna	5.40±0.10ab	15.53±1.04bc	27.78±1.31ab
15	Jessore	5.06±0.15bcd	10.48±0.65ef	28.43±1.55a
16	Jhenaidah	4.83±0.08cdef	13.63±1.17bcde	25.36±0.30b
17	Faridpur	4.68±0.10defg	10.32±0.91ef	27.64±1.51ab
18	Shariatpur	4.69±0.13cdefg	11.06±1.08ef	27.36±1.78ab
19	Barguna	2.13±0.13l	5.54±1.39g	26.26±0.43ab
20	Khagrachari	4.56±0.10efghi	12.3±0.78cdef	25.98±0.29ab
21	Bandarban	5.07±0.08bc	14.86±1.08bcd	26.13±0.71ab
22	Rangamati	4.52±0.10fghi	12.98±1.39bcdef	26.17±0.56ab
23	Cox's Bazar	2.13±0.13l	5.56±1.56g	26.31±0.30ab
24	St. Martin’s Island	1.78±0.15m	4.5±0.78g	26.59±0.78ab

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

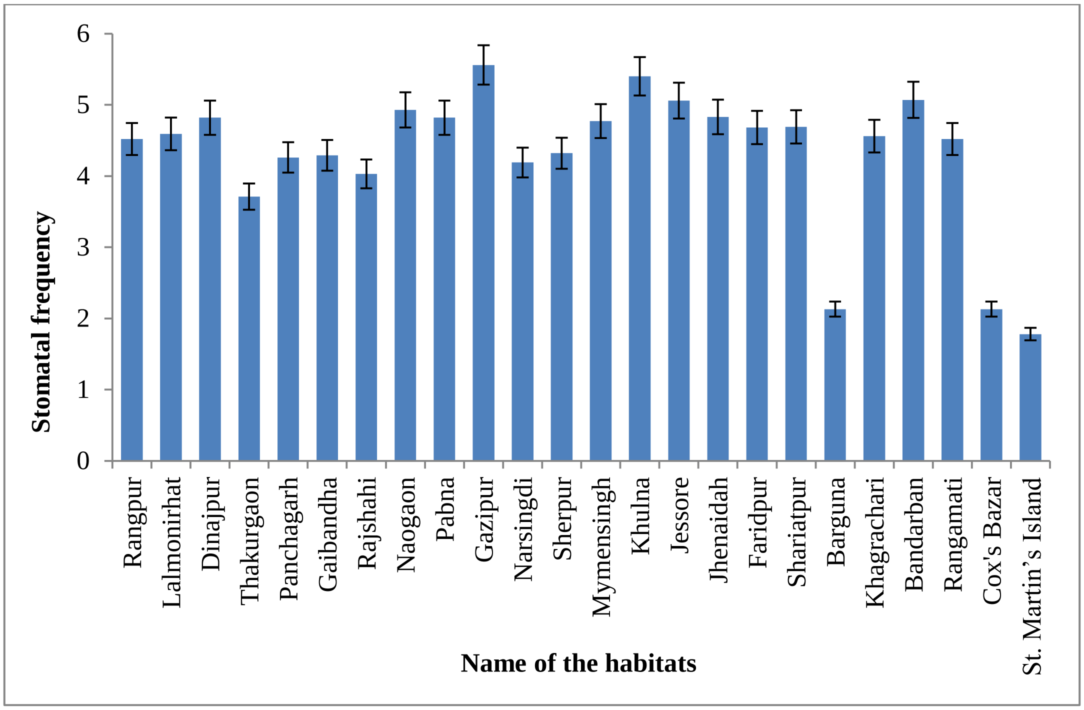


Fig. 55: Stomatal frequency on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

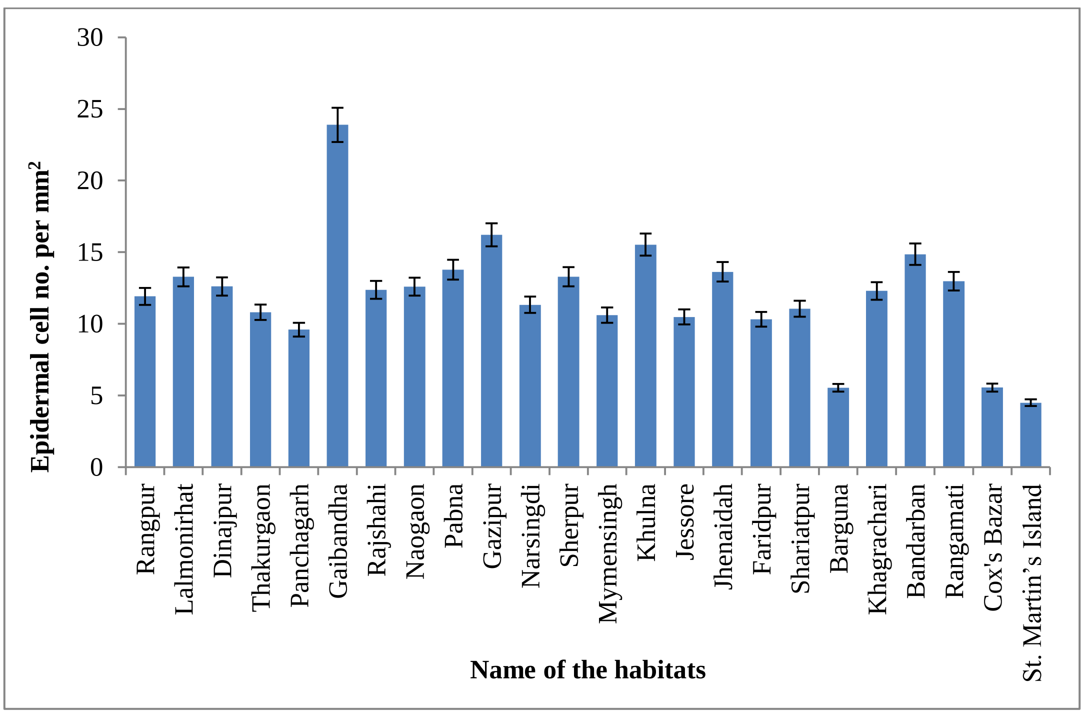


Fig. 56: Epidermal cell number on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

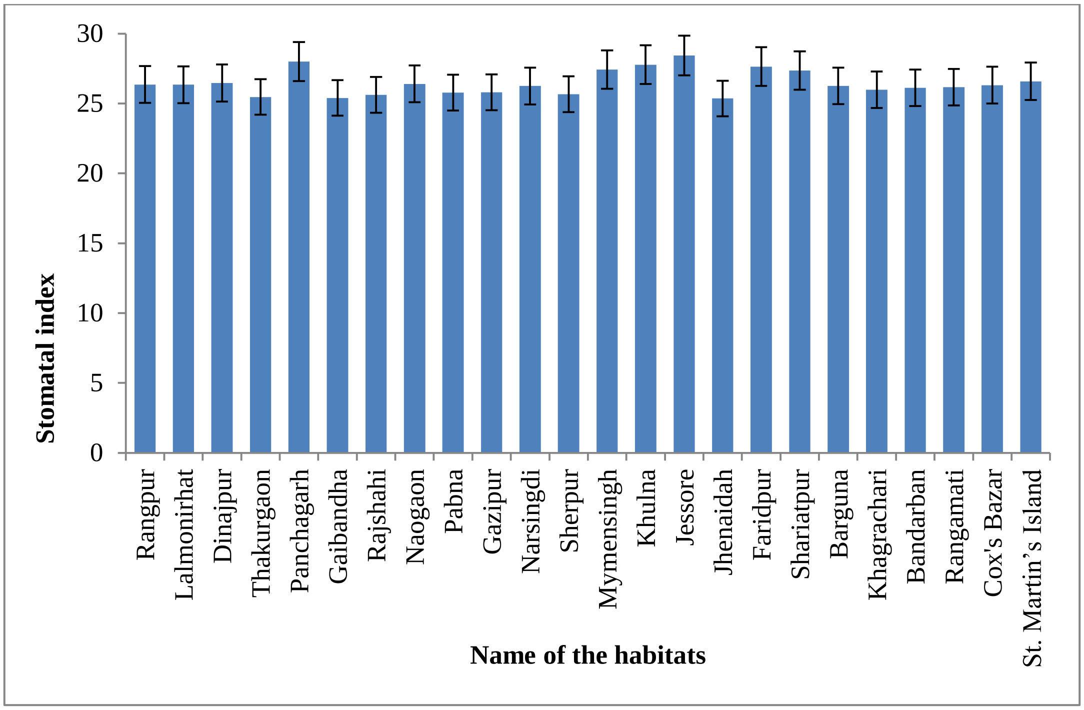


Fig. 57: Stomatal index on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

Table 23: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Adaxial surface of leaves		
		Stomatal frequency	**Epidermal cell no.per mm^2**	**Stomatal index**
		Mean±SE	**Mean±SE**	**Mean±SE**
1	Rangpur	5.21±0.24bcde	16.06±0.73cdef	25.77±0.59bc
2	Lalmonirhat	4.59±0.42defg	13.20±0.82hi	26.67±0.36b
3	Dinajpur	5.15±0.56bcde	14.95±0.70efgh	26.07±0.46bc
4	Thakurgaon	6.29±0.38ab	19.03±0.70ab	25.43±0.33bc
5	Panchagarh	5.44±0.22abcde	16.05±0.79cdef	26.81±0.64b
6	Gaibandha	6.31±0.56ab	18.85±0.76ab	25.73±0.56bc
7	Rajshahi	5.29±0.38bcde	15.98±0.73cdef	25.93±0.64bc
8	Naogaon	5.76±0.28abcd	17.69±0.79bcd	25.60±0.46bc
9	Pabna	4.61±0.38cdefg	13.57±0.76ghi	26.07±0.28bc
10	Gazipur	3.85±0.24fghi	11.85±0.61ij	25.80±0.61bc
11	Narsingdi	5.92±0.22abc	17.92±0.82abc	25.87±0.33bc
12	Sherpur	4.36±0.22efgh	13.04±0.64hi	26.24±0.48bc
13	Mymensingh	4.86±0.26cdefg	14.89±0.79efgh	25.78±0.43bc
14	Khulna	5.23±0.20bcde	13.89±0.76fghi	28.76±0.43a
15	Jessore	5.10±0.54bcdef	14.76±0.82efgh	26.55±0.61bc
16	Jhenaidah	6.40±0.44ab	19.57±0.61ab	24.97±0.28c
17	Faridpur	4.96±0.22cdef	15.14±0.55efgh	25.80±0.61bc
18	Shariatpur	6.61±0.38a	19.99±0.70a	25.40±0.31bc
19	Barguna	3.02±0.32i	9.28±0.82k	26.05±0.53bc
20	Khagrachari	5.37±0.60abcde	15.38±0.58defgh	25.61±0.31bc
21	Bandarban	5.58±0.32abcde	16.49±0.61cde	26.27±0.61bc
22	Rangamati	5.57±0.60abcde	15.66±0.55cdefg	26.37±0.41bc
23	Cox's Bazar	3.34±0.40hi	9.72±0.67k	26.26±0.33bc
24	St. Martin's Island	3.70±0.32ghi	10.48±0.64jk	27.04±0.41b

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

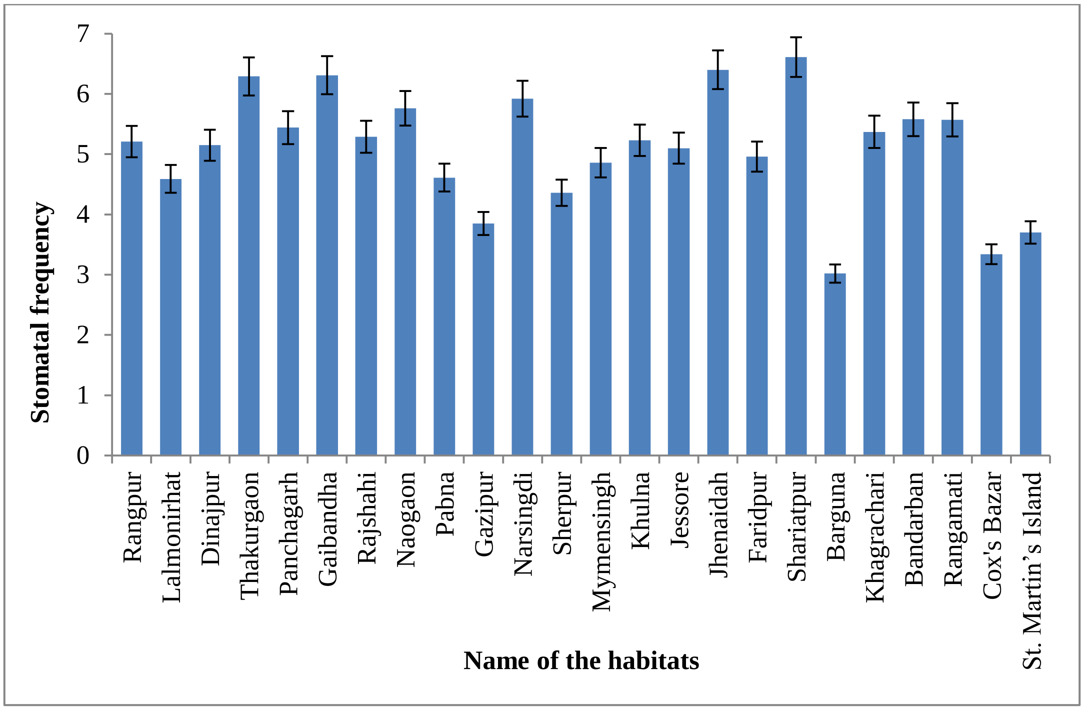


Fig. 58: Stomatal frequency on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

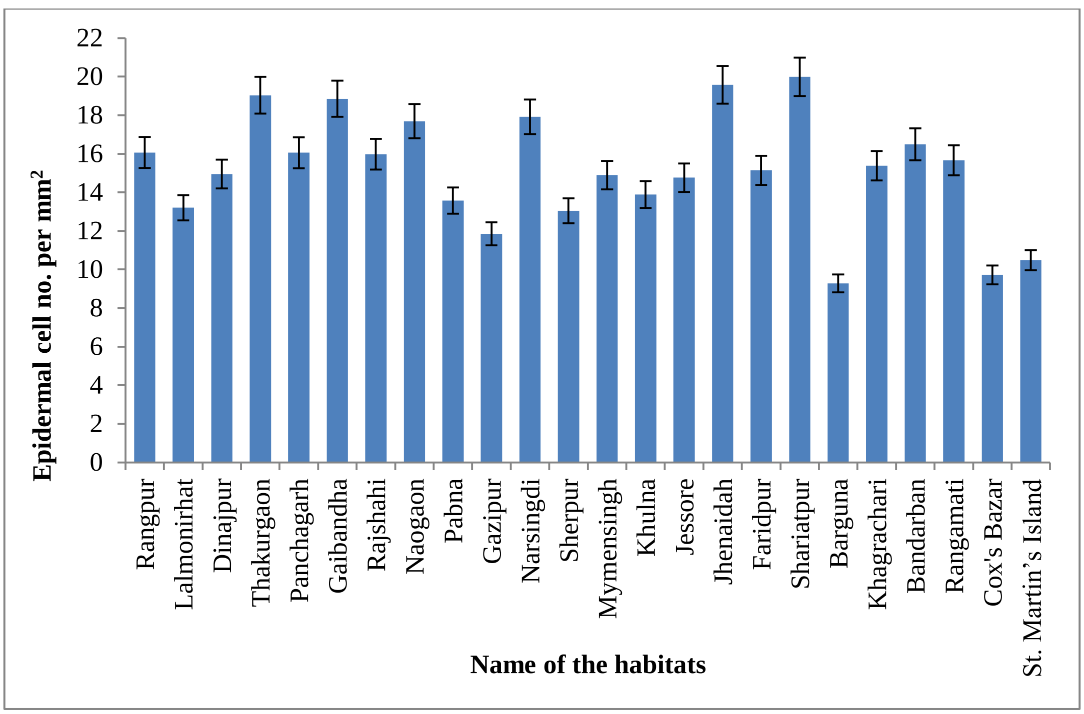


Fig. 59: Epidermal cell number on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

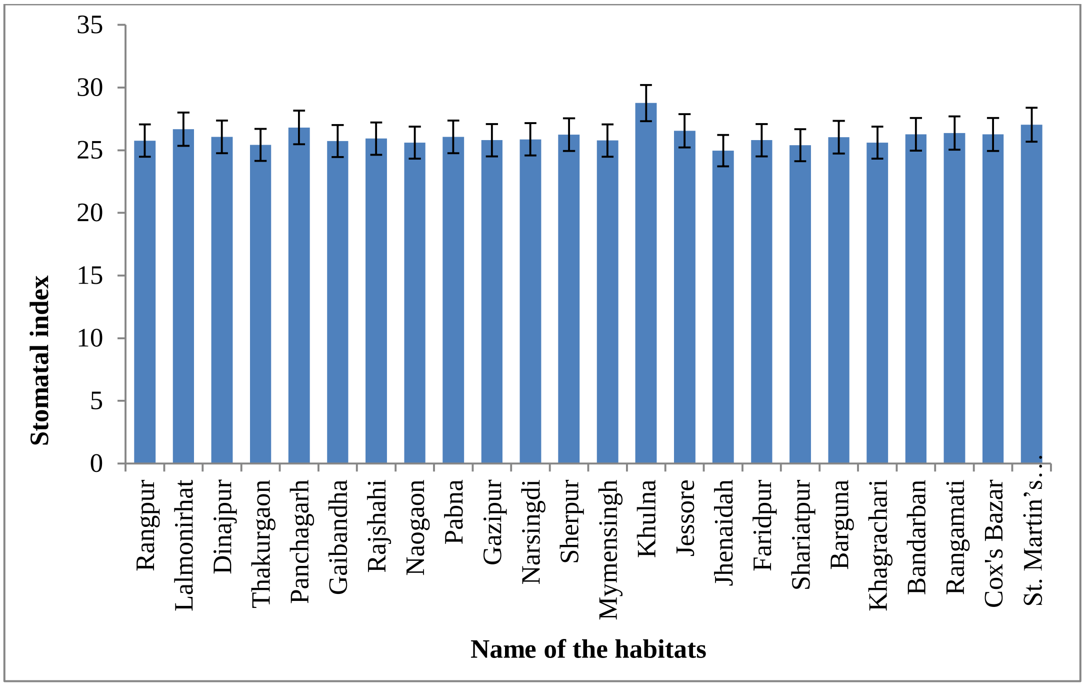


Fig. 60: Stomatal index on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

Stomatal length with guard cell was highest (21.50 µm) in case of Cox’s Bazar and lowest (11.50 µm) in case of Mymensingh in abaxial surface of the leaves (Table 24). Stomatal length with guard cell was highest (17.59 µm) in case of Saint Martin’s Island and lowest (11.56 µm) in case of Mymensingh in adaxial surface of the leaves (Table 25). Stomatal breadth with guard cell was highest (13.71 µm) in case of Saint Martin’s Island and lowest (8.11 µm) in case of Khulna in abaxial surface of the leaves (Table 24). Stomatal breadth with guard cell was highest (15.62 µm) in case of Barguna and lowest (8.11 µm) in case of Khulna in adaxial surface of the leaves (Table 25).

Epidermal cell length was highest (80.06 µm) in case of Saint Martin’s Island and lowest (23.07 µm) in case of Faridpur in abaxial surface of the leaves (Table 24). Epidermal cell length was highest (72.06 µm) in case of Barguna and lowest (28.76 µm) in case of Khulna in adaxial surface of the leaves (Table 25). Epidermal cell breadth was highest (21.74 µm) in case of Cox’s Bazar and lowest (7.31 µm) in case of Khulna in abaxial surface of the leaves (Table 24). Epidermal cell breadth was highest (12.20 µm) in case of Cox’s Bazar and lowest (7.10 µm) in case of Faridpur in adaxial surface of the leaves (Table 25).

Table 24: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Abaxial Surface of leaves			
		Stomata length with guard cell (µm)	Stomata breadth with guard cell (µm)	Epidermal cell length (µm)	Epidermal cell breadth (µm)
		Mean±SE	Mean±SE	Mean±SE	Mean±SE
1	Rangpur	12.82±0.66f	9.11±0.87cd	32.12±0.95h	8.14±1.13cd
2	Lalmonirhat	16.39±0.33cd	9.48±1.13bcd	35.06±0.95fg	8.41±1.13cd
3	Dinajpur	16.75±0.38c	8.26±0.43d	38.12±0.95de	8.09±0.74cd
4	Thakurgaon	15.75±0.28cde	9.07±0.87cd	32.13±1.00h	8.10±0.78cd
5	Panchagarh	16.12±0.59cde	9.38±0.78bcd	39.29±0.74d	10.12±0.95c
6	Gaibandha	15.60±0.51cde	9.02±0.48cd	36.10±0.78ef	9.73±1.04cd
7	Rajshahi	15.86±0.66e	8.22±0.43d	24.06±0.43k	8.06±0.43cd
8	Naogaon	16.05±0.64cde	9.37±0.69bcd	36.07±0.56ef	8.10±0.78cd
9	Pabna	15.95±0.38cde	9.58±1.13bcd	36.07±0.52ef	8.07±0.56cd
10	Gazipur	15.70±0.66e	10.66±0.43bcd	38.75±0.87d	7.79±0.43cd
11	Narsingdi	16.30±0.56cde	11.81±0.61ab	29.41±0.56i	8.06±0.43cd
12	Sherpur	12.57±0.53f	8.61±1.00cd	34.85±1.04fg	8.23±0.52cd
13	Mymensingh	11.50±0.46f	10.40±1.04bcd	33.32±0.95gh	8.14±1.13cd
14	Khulna	14.95±0.28e	8.11±0.87d	27.00±0.48j	7.31±0.82d
15	Jessore	12.23±0.28f	8.14±1.08d	36.11±0.82ef	8.14±1.08cd
16	Jhenaidah	15.35±0.59e	10.21±1.04bcd	29.40±0.48i	7.73±1.04cd
17	Faridpur	15.87±0.59de	8.14±1.08d	23.07±0.52k	8.46±0.43cd
18	Shariatpur	15.30±0.36de	12.07±0.56ab	28.12±0.91ij	8.78±1.08cd
19	Barguna	19.53±0.33b	9.83±0.56bcd	72.25±0.61b	10.26±1.00c
20	Khagrachari	16.2±0.51cde	8.86±0.48cd	40.08±0.61d	8.07±0.52cd
21	Bandarban	14.79±0.33e	8.12±0.91d	32.12±0.91h	8.09±0.69cd
22	Rangamati	15.74±0.36cde	8.93±1.04cd	26.07±0.52j	8.67±1.04cd
23	Cox's Bazar	21.5±0.36a	11.29±0.74abc	70.09±0.69c	21.74±1.13a
24	St. Martin’s Island	20.63±0.48ab	13.71±0.87a	80.06±0.43a	16.25±0.91b

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

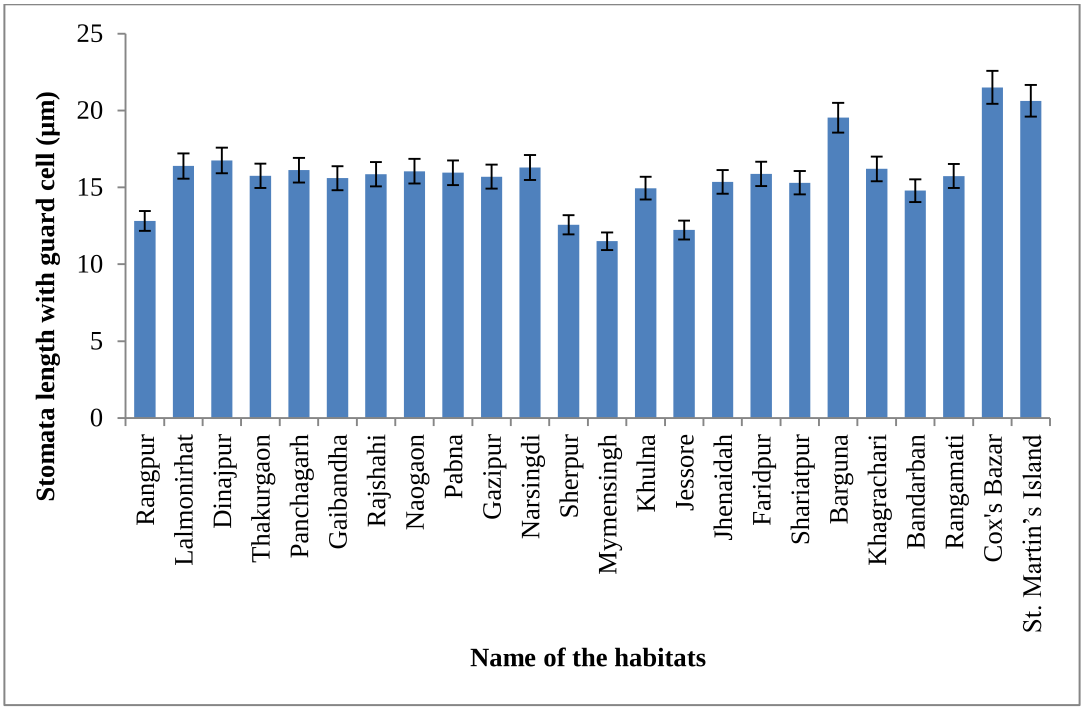


Fig. 61: Stomata length with guard cell on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

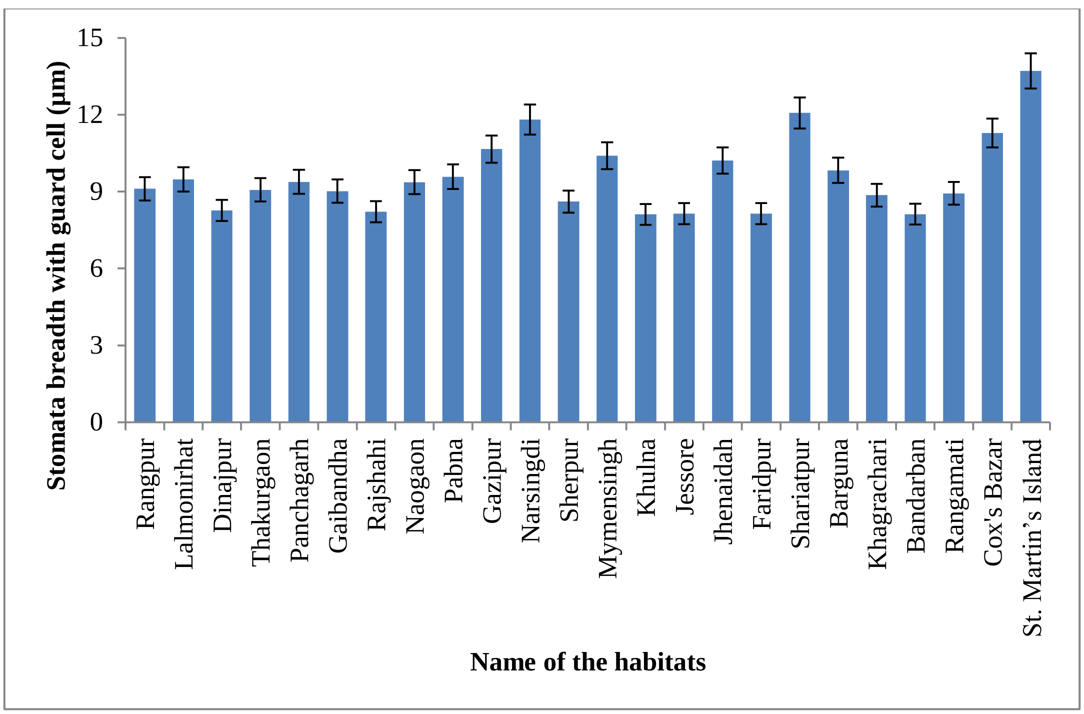


Fig. 62: Stomata breadth with guard cell on abaxial surface of leaves of *C. dactylon* accessions in different of habitats.

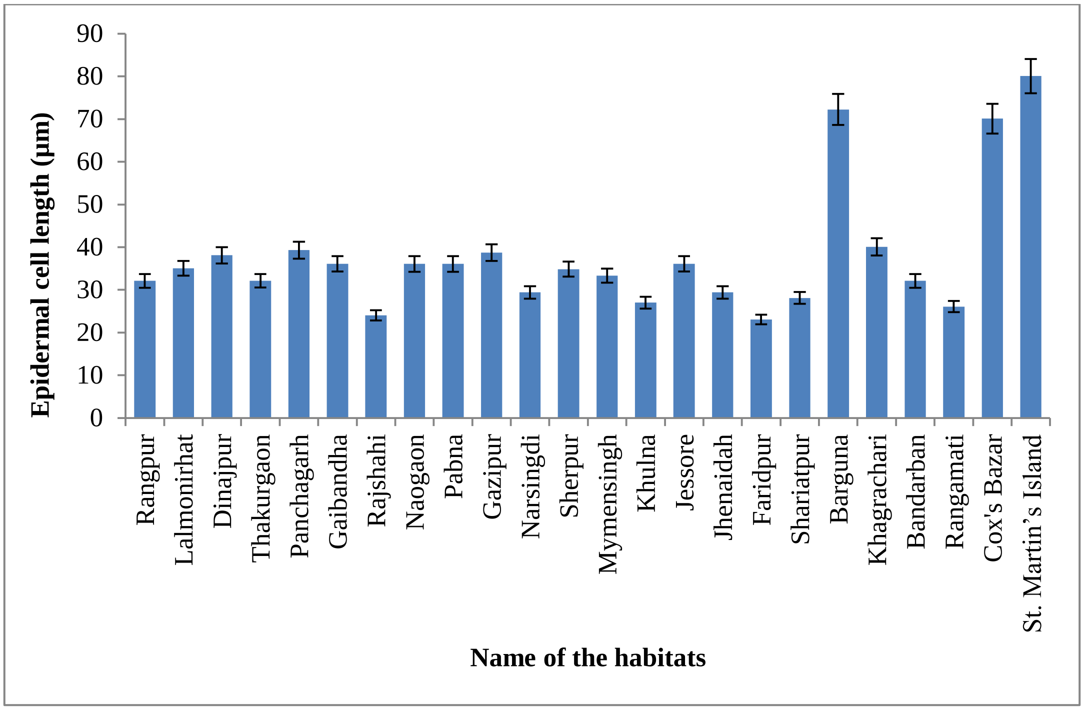


Fig. 63: Epidermal cell length on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

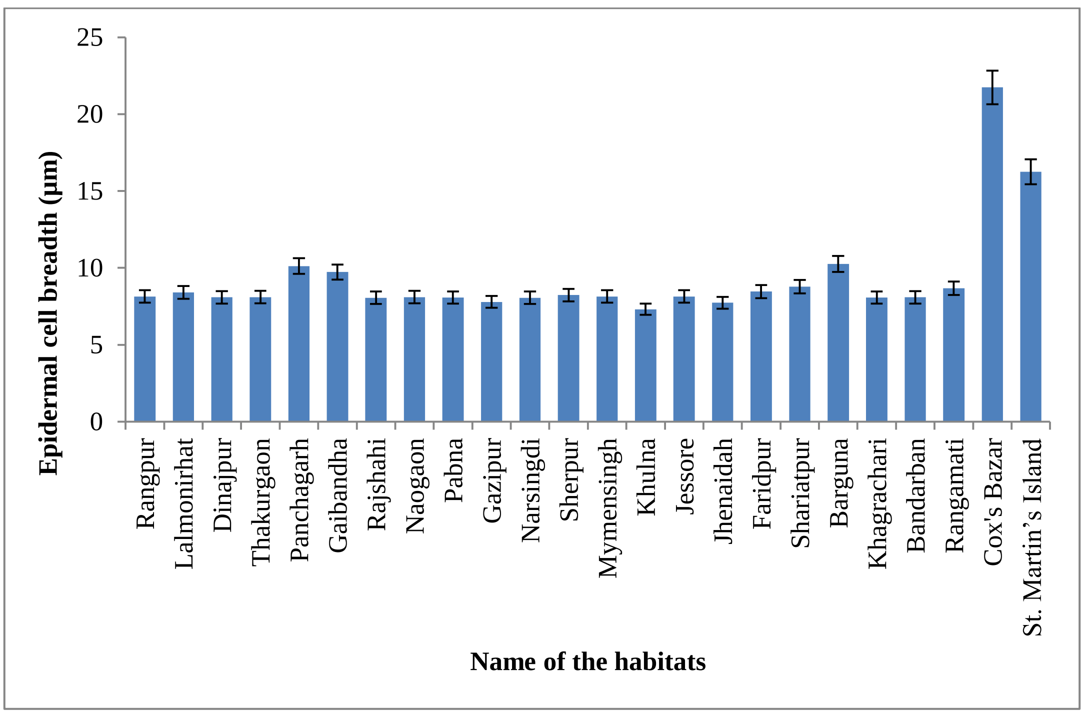


Fig. 64: Epidermal cell breadth on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

Table 25: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Adaxial Surface of leaves			
		Stomata length with guard cell (µm)	Stomata breadth with guard cell (µm)	Epidermal cell length (µm)	Epidermal cell breadth (µm)
		Mean±SE	Mean±SE	Mean±SE	Mean±SE
1	Rangpur	12.42±0.73ef	8.77±0.30fgh	38.79±0.70i	8.33±0.60defgh
2	Lalmonirhat	14.02±0.87cde	10.73±0.24c	34.16±0.73k	8.43±0.42defgh
3	Dinajpur	12.50±0.84ef	9.81±0.38cdef	38.22±0.79ij	7.53±0.60gh
4	Thakurgaon	13.69±0.55cdef	9.35±0.52defgh	42.69±0.61gh	8.19±0.34efgh
5	Panchagarh	15.41±0.76abcd	9.77±0.60cdef	47.29±0.55e	8.17±0.30efgh
6	Gaibandha	13.42±0.73def	9.79±0.34cdef	37.45±0.70ij	8.11±0.56efgh
7	Rajshahi	15.29±0.61abcd	8.83±0.30efgh	33.15±0.84k	9.11±0.56bcdef
8	Naogaon	15.66±0.58abcd	12.19±0.34b	37.72±0.82ij	10.18±0.56bc
9	Pabna	12.66±0.73ef	9.83±0.42cdef	29.52±0.64m	8.33±0.60defgh
10	Gazipur	14.09±0.67cde	8.80±0.28fgh	38.62±0.55i	9.81±0.38bcd
11	Narsingdi	14.29±0.61cde	8.37±0.38gh	33.85±0.76k	8.38±0.20defgh
12	Sherpur	15.57±0.61abcd	9.38±0.32defgh	40.85±0.76h	8.74±0.46cdefg
13	Mymensingh	11.56±0.67f	8.78±0.32fgh	51.79±0.70d	7.99±0.34fgh
14	Khulna	12.62±0.55ef	8.11±0.20h	28.76±0.67m	8.11±0.20efgh
15	Jessore	13.42±0.73def	9.41±0.38defgh	54.69±0.61c	8.31±0.56defgh
16	Jhenaidah	14.22±0.73cde	8.70±0.30fgh	44.95±0.84f	8.59±0.58defgh
17	Faridpur	15.96±0.82abc	10.12±0.58cde	36.36±0.67j	7.10±0.40h
18	Shariatpur	14.65±0.76bcde	9.50±0.54cdefg	30.72±0.64lm	8.12±0.22efgh
19	Barguna	16.72±0.64ab	15.62±0.28a	72.06±0.64A	8.31±0.56defgh
20	Khagrachari	15.34±0.55abcd	9.50±0.30cdefg	32.66±0.58kl	9.60±0.36bcde
21	Bandarban	12.57±0.79ef	9.42±0.40defgh	36.72±0.64ij	8.61±0.52defgh
22	Rangamati	12.86±0.58ef	8.54±0.26fgh	32.66±0.58kl	8.20±0.36efgh
23	Cox's Bazar	17.14±0.87a	14.93±0.60a	58.96±0.67B	12.20±0.36a
24	St. Martin's Island	17.59±0.70a	10.40±0.36cd	43.96±0.67fg	10.39±0.42b

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

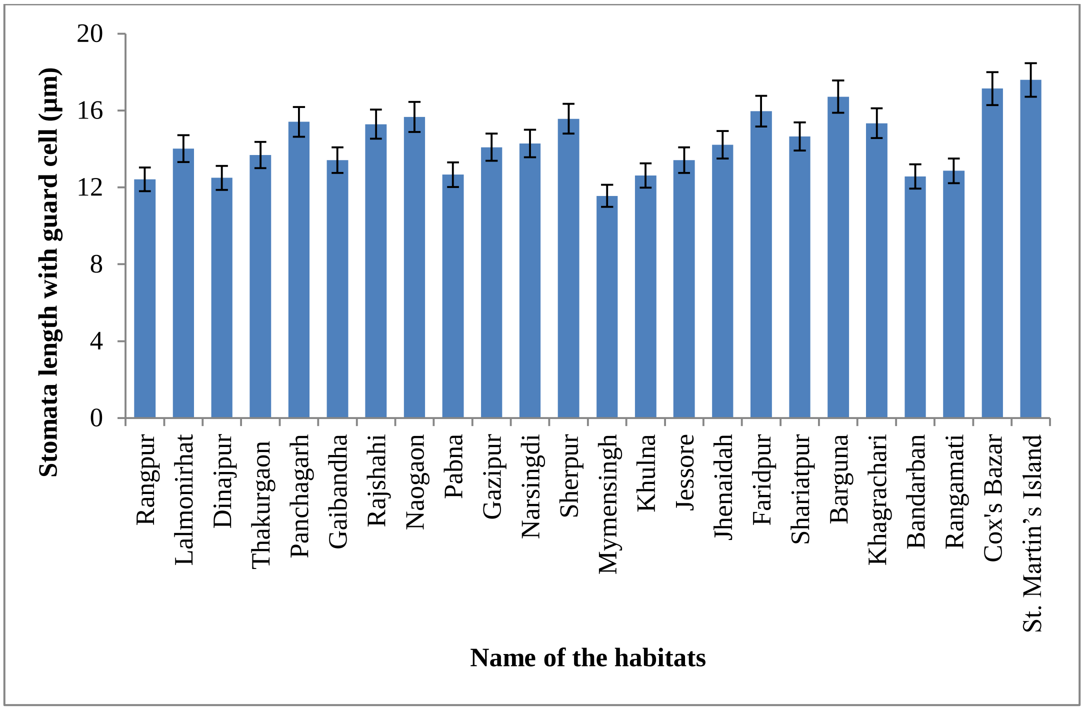


Fig. 65: Stomata length with guard cell on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

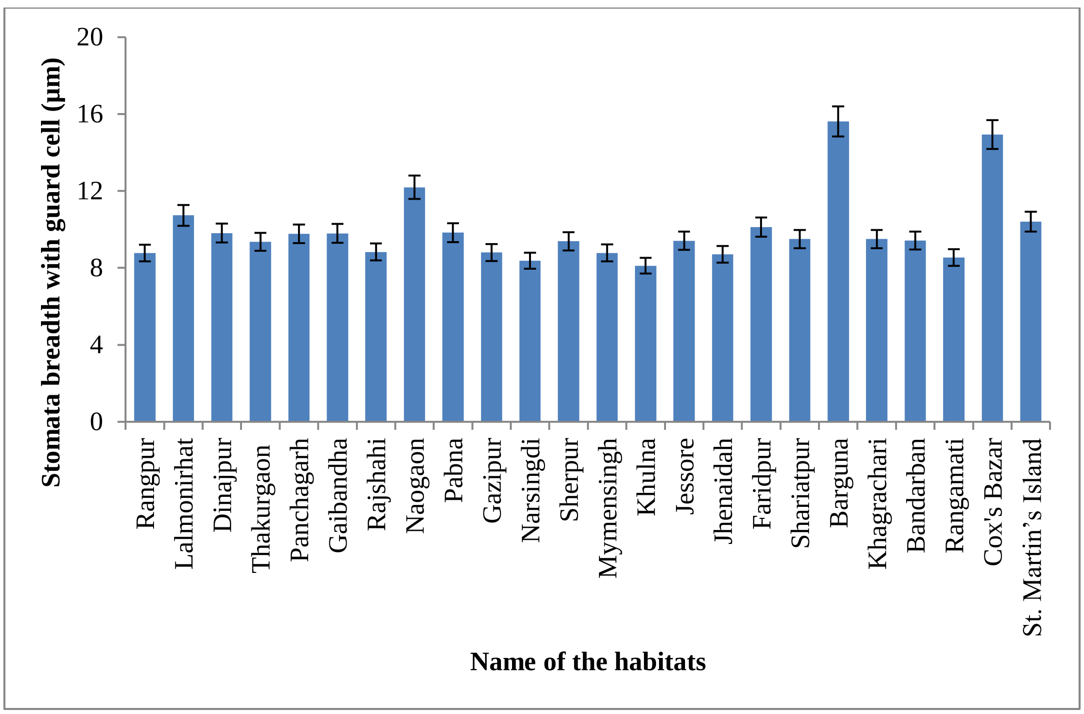


Fig. 66: Stomata breadth with guard cell on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

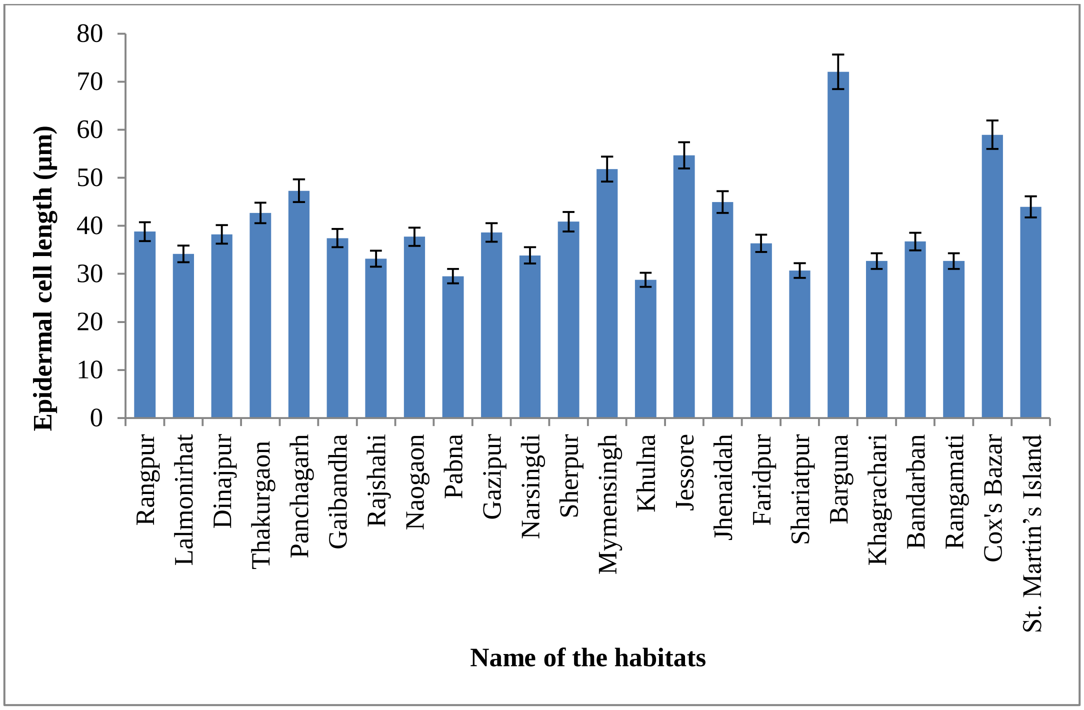


Fig. 67: Epidermal cell length on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

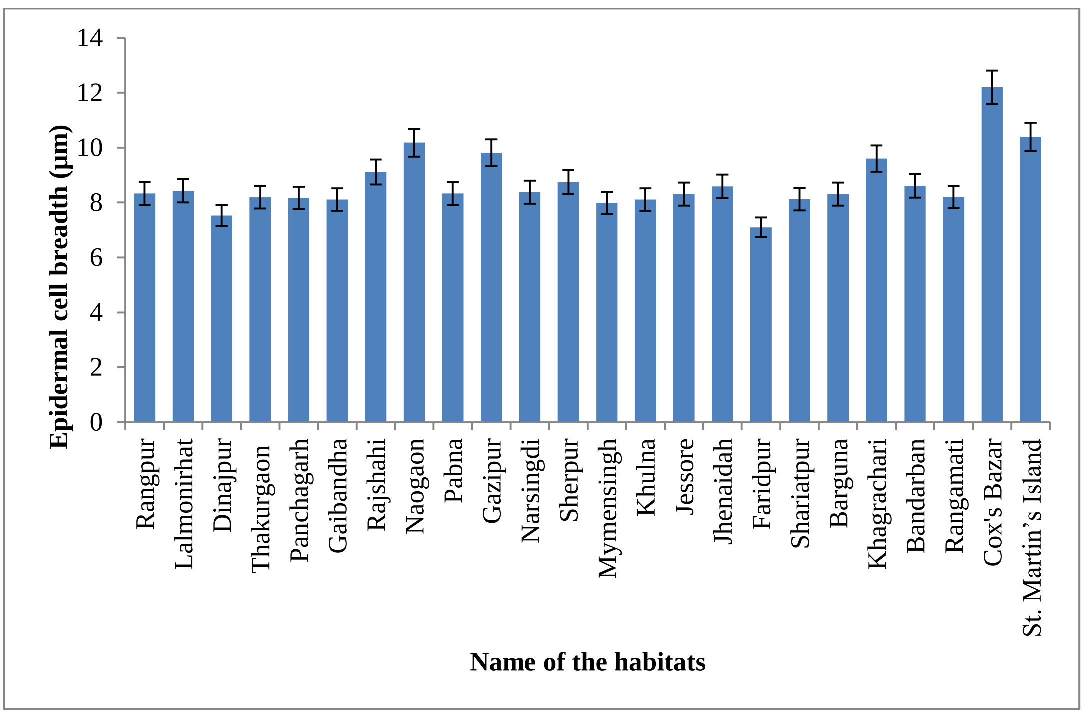


Fig. 68: Epidermal cell breadth on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

Number of silica bodies per mm^2 was highest (10.26) in case of Bandarban and lowest (2.13) in case of Saint Martin's Island in abaxial surface of the leaves (Table 26). Number of silica bodies per mm^2 was highest (7.95) in case of Naogaon and lowest (2.00) in case of Cox's Bazar in adaxial surface of the leaves (Table 27). Prickles angular numbers per mm^2 was highest (0.32) in case of Gaibandha and lowest (0.19) in case of Mymensingh and no prickles angular was found in case of Barguna, Cox's Bazar and Saint Martin's Island in abaxial surface of the leaves (Table 26). Prickles angular numbers per mm^2 was highest (0.42) in case of Thakurgaon and lowest (0.18) in case of both Pabna and Rangamati and no prickles angular was found in case of Barguna, Cox's Bazar and Saint Martin's Island in adaxial surface of the leaves (Table 27). Number of hooks per mm^2 was highest (0.96) in case of Rajshahi and lowest (0.02) in case of Thakurgaon, Sherpur and Saint Martin's Island in abaxial surface of the leaves (Table 26). Number of hooks per mm^2 was highest (1.69) in case of Lalmonirhat and lowest (0.02) in case of Barguna in adaxial surface of the leaves (Table 27). In case of Cox's Bazar, hook was absent both abaxial and adaxial surfaces of the leaves (Tables 26 & 27). Macro hair number per mm^2 was highest (0.26) in case of Cox's Bazar and lowest (0.02) in case of Khagrachari in abaxial surface of the leaves (Table 26). Macro hair number per mm^2 was highest (0.52) in case of Cox's Bazar and lowest (0.02) in case of Gazipur in adaxial surface of the leaves (Table 27). No macro hair was found in adaxial surface in case of Pabna (Table 27).

Table 26: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Abaxial Surface of leaves			
		Silica bodies no. per mm^2	Prickles angular no. per mm^2	Hooks no. per mm^2	Macro hair no. per mm^2
		Mean±SE	Mean±SE	Mean±SE	Mean±SE
1	Rangpur	7.05±0.53cdefgh	0.22±0.02bcd	0.15±0.03fgh	0.04±0.00fg
2	Lalmonirhat	5.28±0.31gh	0.23±0.03bcd	0.26±0.02 de	0.15±0.03bcd
3	Dinajpur	7.61±0.33bc	0.26±0.02abcd	0.18±0.02fg	0.14±0.02bcde
4	Thakurgaon	5.79±0.38gh	0.22±0.02bcd	0.02±0.02jk	0.09±0.01cdefg
5	Panchagarh	5.01±0.33h	0.29±0.01ab	0.28±0.04cd	0.07±0.03defg
6	Gaibandha	6.66±0.46efgh	0.32±0.04a	0.04±0.00ijk	0.14±0.02bcde
7	Rajshahi	6.45±0.43fgh	0.26±0.02abcd	0.96±0.04a	0.10±0.02bcdefg
8	Naogaon	8.29±0.43bcd	0.30±0.02ab	0.43±0.03b	0.12±0.04bcdef
9	Pabna	7.24±0.61bcdefg	0.22±0.02bcd	0.29±0.01cd	0.10±0.02bcdefg
10	Gazipur	7.66±0.46bcde	0.31±0.03a	0.47±0.03b	0.06±0.02efg
11	Narsingdi	5.92±0.51gh	0.26±0.02abcd	0.31±0.03cd	0.10±0.02cdefg
12	Sherpur	7.01±0.64efgh	0.23±0.03bcd	0.02±0.02jk	0.07±0.03defg
13	Mymensingh	6.39±0.38cdefgh	0.19±0.03d	0.35±0.03c	0.11±0.03bcdefg
14	Khulna	8.46±0.46ab	0.31±0.03a	0.29±0.01cd	0.14±0.02bcde
15	Jessore	7.36±0.51bcdef	0.23±0.03bcd	0.16±0.00fg	0.11±0.03bcdefg
16	Jhenaidah	7.17±0.64defgh	0.28±0.00abc	0.30±0.02cd	0.12±0.04bcdef
17	Faridpur	5.16±0.41h	0.23±0.03bcd	0.11±0.03ghi	0.09±0.01cdefg
18	Shariatpur	8.10±0.56bcdef	0.20±0.00cd	0.18±0.02fg	0.18±0.02b
19	Barguna	2.57±0.64i	0.00±0.00e	0.03±0.03jk	0.10±0.02cdefg
20	Khagrachari	9.75±0.28a	0.24±0.00abcd	0.09±0.01hij	0.02±0.02g
21	Bandarban	10.26±0.66a	0.27±0.03abc	0.19±0.03ef	0.07±0.03defg
22	Rangamati	8.87±0.59ab	0.20±0.00cd	0.08±0.04hijk	0.05±0.01fg
23	Cox's Bazar	2.92±0.61i	0.00±0.00e	0.00±0.00k	0.26±0.02a
24	St. Martin's Island	2.13±0.53i	0.00±0.00e	0.02±0.02jk	0.16±0.04bc

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

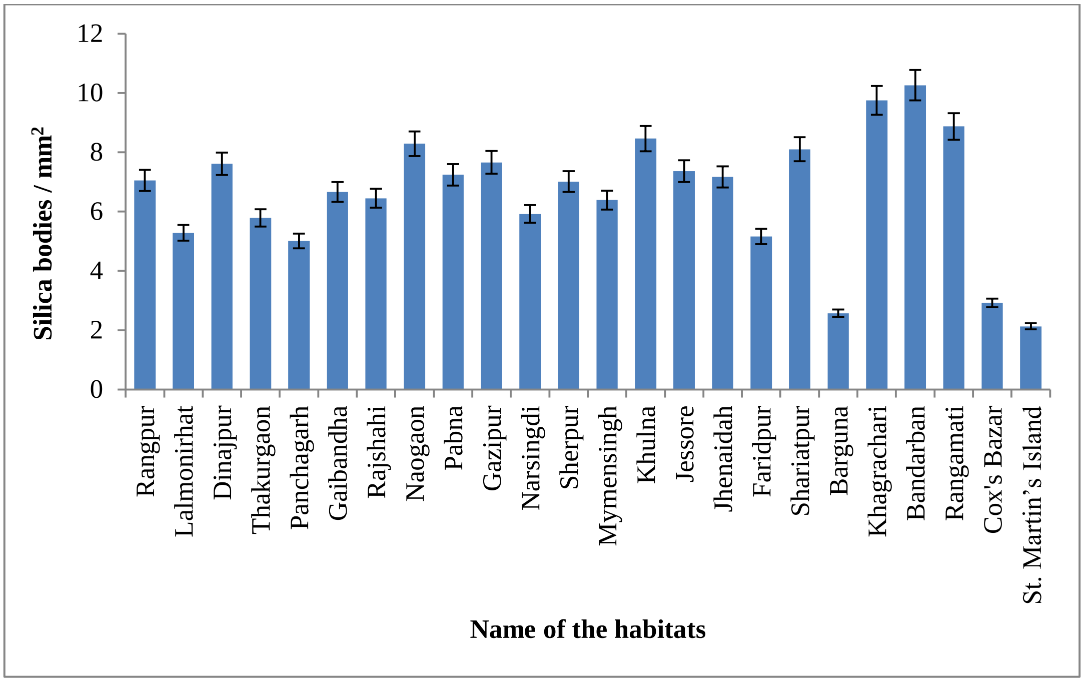


Fig. 69: Silica bodies number on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

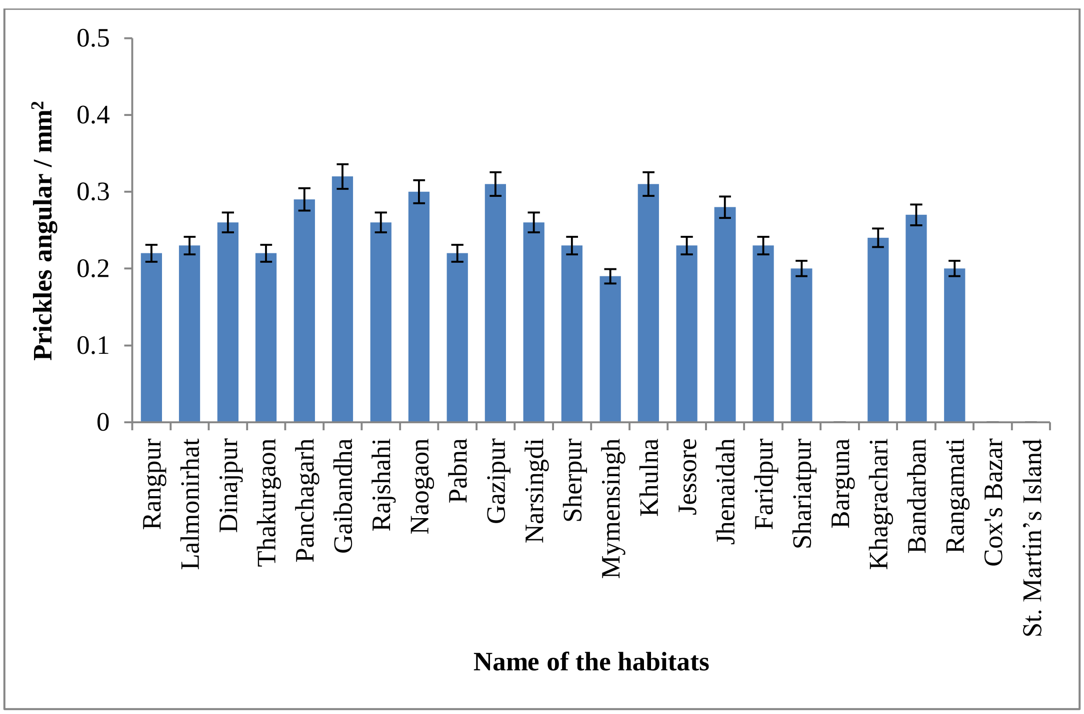


Fig. 70: Prickles angular number on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

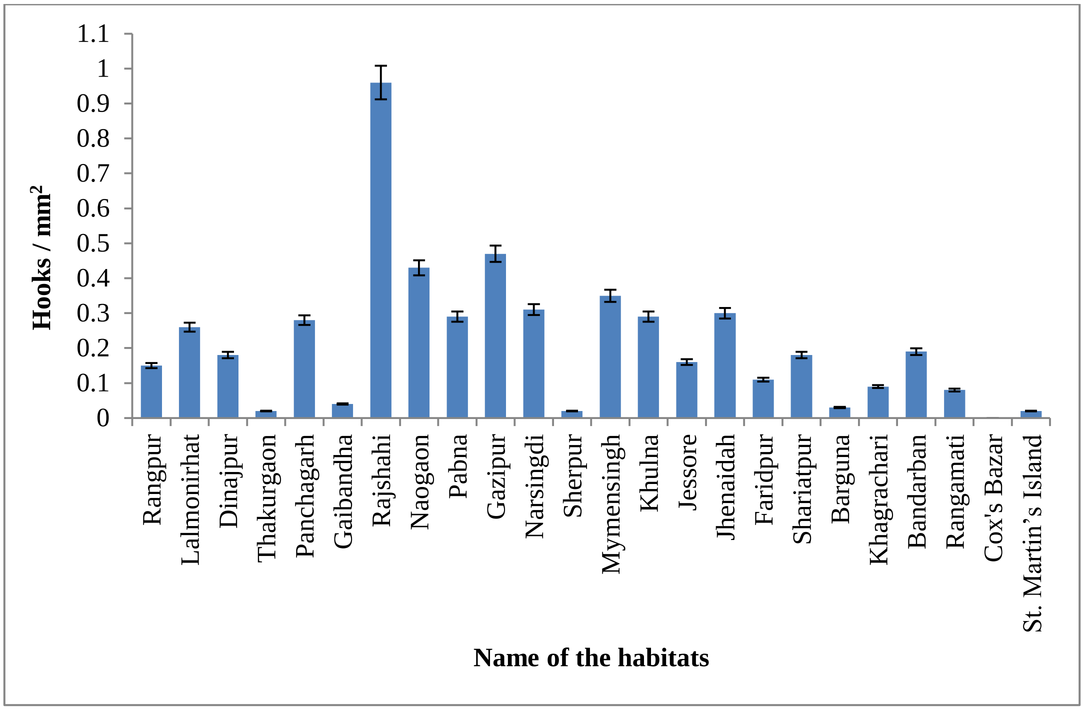


Fig. 71: Hooks number on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

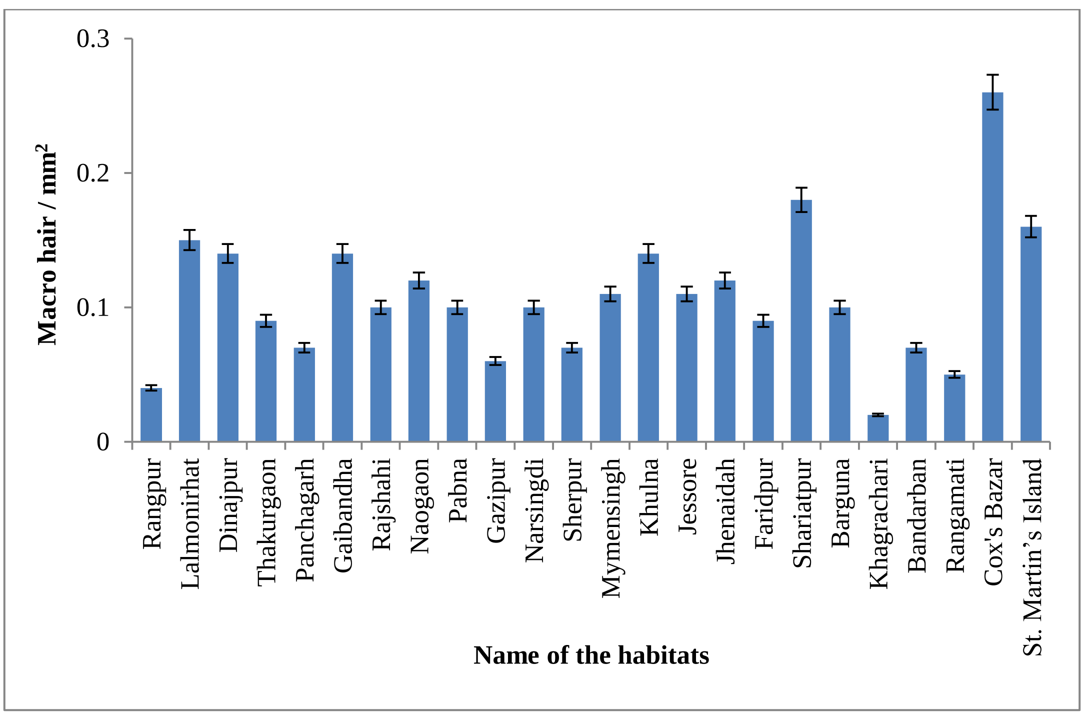


Fig. 72: Macro hair number on abaxial surface of leaves of *C. dactylon* accessions in different habitats.

Table 27: Quantitative leaf epidermal characteristics of *Cynodon dactylon* accessions collected from different habitats.

Sl. No.	Habitats	Adaxial surface of leaves			
		Silica bodies no. per mm^2	Prickles angular no. per mm^2	Hooks no. per mm^2	Macro hair no. per mm^2
		Mean±SE	**Mean±SE**	**Mean±SE**	**Mean±SE**
1	Rangpur	5.69±0.30cd	0.30±0.02cdef	0.52±0.00f	0.11±0.03de
2	Lalmonirhat	4.89±0.52d	0.39±0.03ab	1.69±0.01a	0.24±0.04bc
3	Dinajpur	5.73±0.30cd	0.22±0.02fg	0.98±0.02d	0.16±0.04cd
4	Thakurgaon	6.77±0.60abc	0.42±0.02a	1.54±0.02b	0.11±0.03de
5	Panchagarh	5.09±0.52d	0.24±0.04defg	0.32±0.04j	0.06±0.02efg
6	Gaibandha	6.48±0.44bc	0.28±0.04cdef	0.48±0.04fgh	0.04±0.00efg
7	Rajshahi	5.69±0.38cd	0.24±0.04defg	1.62±0.02a	0.25±0.01b
8	Naogaon	7.95±0.20a	0.30±0.02cde	0.79±0.03e	0.09±0.01defg
9	Pabna	5.89±0.38cd	0.18±0.02g	0.42±0.02ghi	0.00±0.00g
10	Gazipur	5.86±0.46cd	0.27±0.03cdef	1.08±0.00c	0.02±0.02fg
11	Narsingdi	6.68±0.44abc	0.26±0.02defg	0.75±0.03e	0.08±0.04defg
12	Sherpur	4.83±0.34d	0.32±0.00bcd	0.36±0.04ij	0.06±0.02efg
13	Mymensingh	5.65±0.24cd	0.30±0.02cdef	1.11±0.03c	0.11±0.03def
14	Khulna	6.87±0.20abc	0.22±0.02efg	0.46±0.02fgh	0.05±0.01efg
15	Jessore	5.75±0.42cd	0.35±0.03abc	1.11±0.03c	0.13±0.01de
16	Jhenaidah	6.56±0.58bc	0.39±0.03ab	1.05±0.01cd	0.10±0.02def
17	Faridpur	5.79±0.34cd	0.35±0.03abc	0.42±0.02ghi	0.12±0.00de
18	Shariatpur	6.88±0.28abc	0.26±0.02defg	0.43±0.03ghi	0.03±0.03efg
19	Barguna	2.38±0.32e	0.00±0.00h	0.02±0.02k	0.08±0.04defg
20	Khagrachari	6.89±0.30abc	0.41±0.01a	1.52±0.04b	0.16±0.04cd
21	Bandarban	6.15±0.34cd	0.26±0.02defg	0.50±0.02fg	0.07±0.03defg
22	Rangamati	7.67±0.20ab	0.18±0.02g	0.40±0.04hi	0.11±0.03de
23	Cox's Bazar	2.00±0.58e	0.00±0.00h	0.00±0.00k	0.52±0.04a
24	St. Martin’s Island	2.55±0.56e	0.00±0.00h	0.04±0.04k	0.24±0.04bc

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

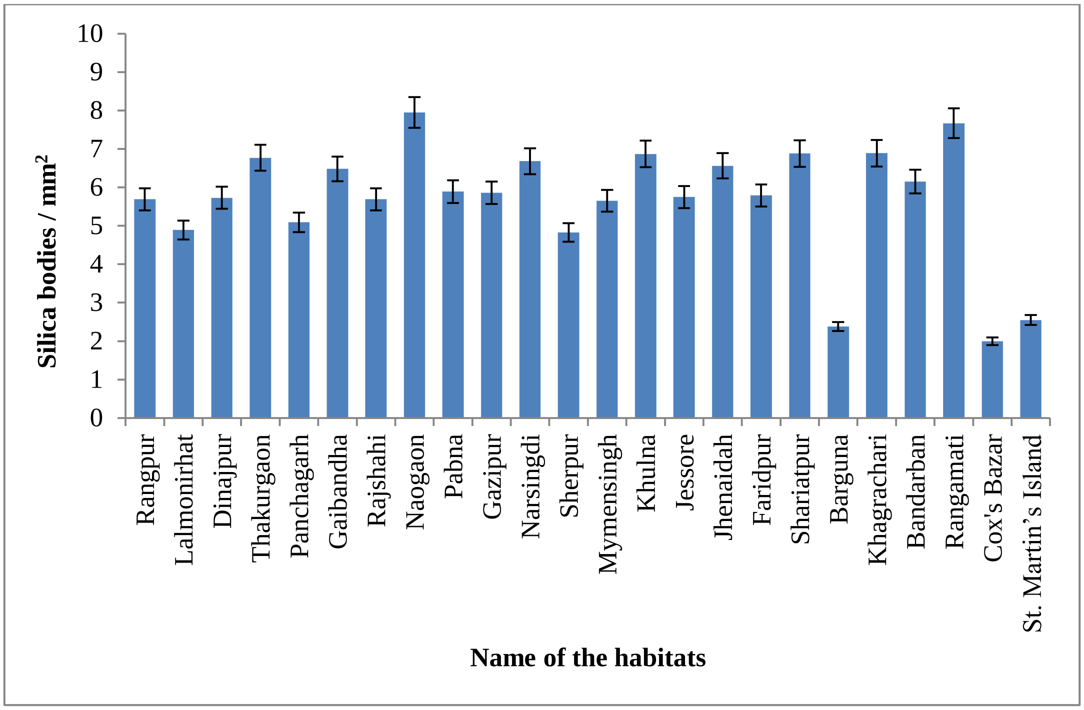


Fig. 73: Silica bodies number on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

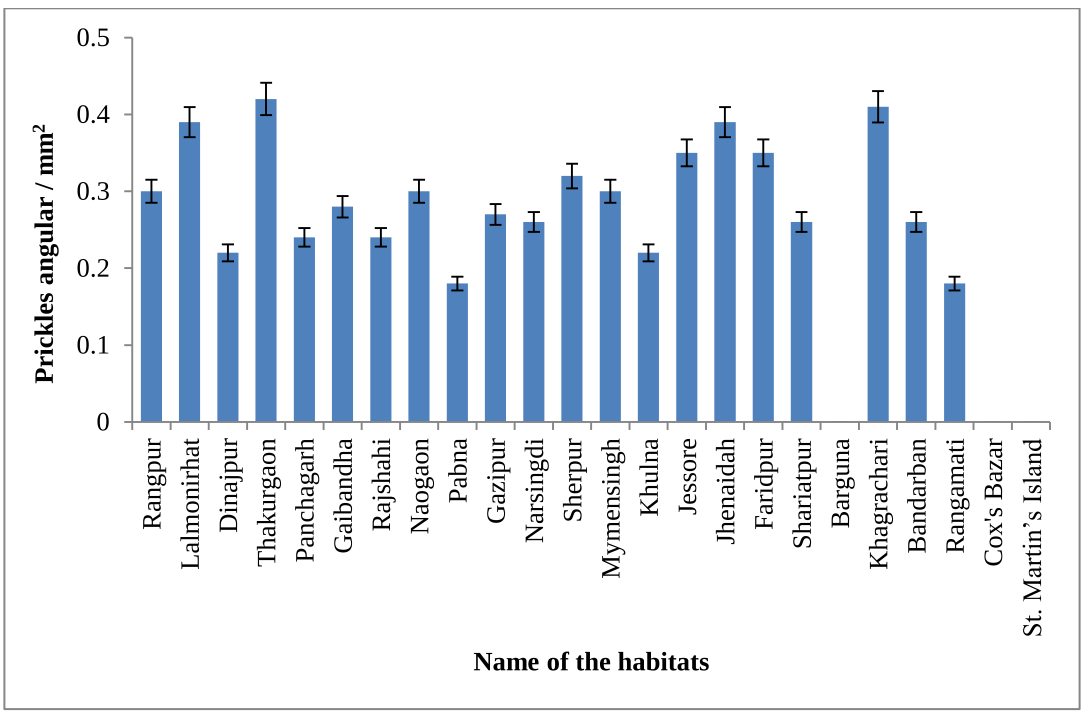


Fig. 74: Prickles angular number on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

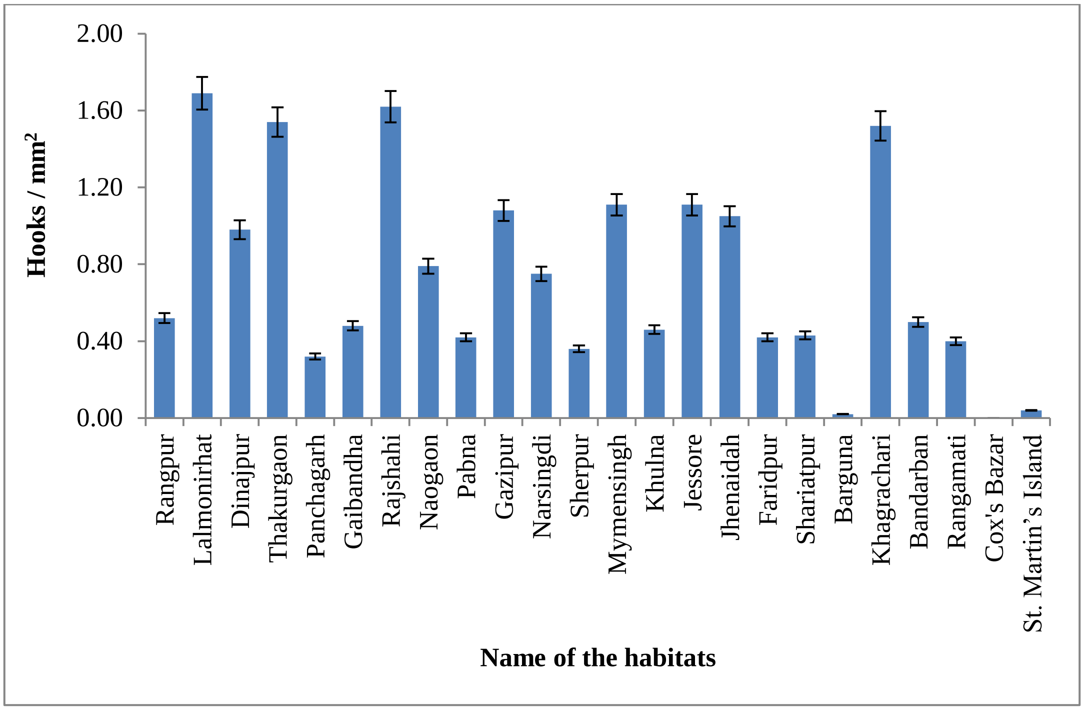


Fig. 75: Hooks number on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

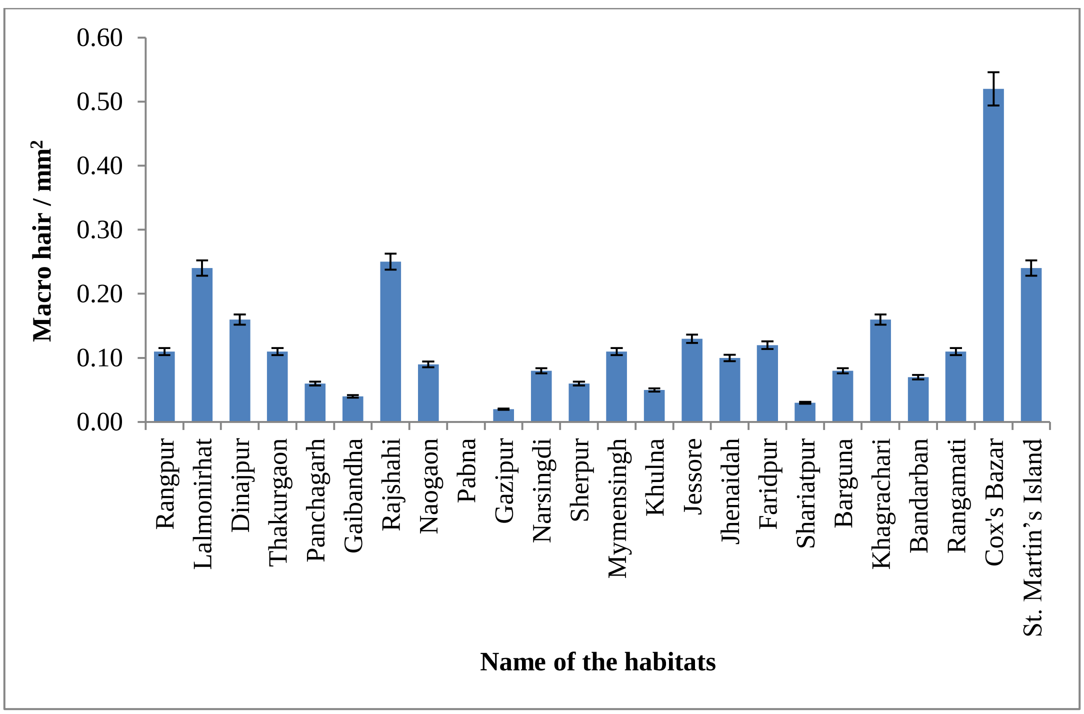


Fig. 76: Macro hair number on adaxial surface of leaves of *C. dactylon* accessions in different habitats.

The ANOVA (Tables 28 & 29) for stomatal characters on abaxial and adaxial surface of leaves, subsequently showed the highly significant variation except stomatal index on abaxial surface.

Table 28: Analysis of variance subjected to variation of different parameters of stomata on abaxial surface of leaves of *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Abaxial surface of leaves	Long cell number	Habitat	23	7.101	166.376***
		Error	48	0.043	
	Long cell length	Habitat	23	1091.251	86.863***
		Error	48	12.563	
	Long cell width	Habitat	23	4.655	39.247***
		Error	48	0.119	
	Stomatal frequency	Habitat	23	2.889	71.886***
		Error	48	0.040	
	Epidermal cell number	Habitat	23	45.104	11.707***
		Error	48	3.853	
	Stomatal index	Habitat	23	2.257	0.974^{ns}
		Error	48	2.318	
	Stomata length with guard cell	Habitat	23	16.224	23.394***
		Error	48	0.694	
	Stomata breadth with guard cell	Habitat	23	6.392	4.248***
		Error	48	1.505	
	Epidermal cell length	Habitat	23	590.137	35.454***
		Error	48	16.645	
	Epidermal cell breadth	Habitat	23	30.400	13.531***
		Error	48	2.247	
	Silica bodies number	Habitat	23	12.801	17.068***
		Error	48	0.750	
	Prickles angular number	Habitat	23	0.023	13.975***
		Error	48	0.002	
	Hooks number	Habitat	23	0.130	75.276***
		Error	48	0.002	
	Macro hair number	Habitat	23	.008	4.133***
		Error	48	0.002	

*** = significant at $p \leq 0.001$ and ns = non-significant.

Table 29: Analysis of variance subjected to variation of different parameters of stomata on adaxial surface of leaves of *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Adaxial surface of leaves	Long cell number	Habitat	23	7.835	15.332***
		Error	48	0.511	
	Long cell length	Habitat	23	597.194	365.079***
		Error	48	1.636	
	Long cell width	Habitat	23	7.845	13.763***
		Error	48	0.570	
	Stomatal frequency	Habitat	23	2.706	6.073***
		Error	48	0.445	
	Epidermal cell number	Habitat	23	26.014	17.194***
		Error	48	1.513	
	Stomatal index	Habitat	23	1.624	2.403**
		Error	48	0.676	
	Stomata length with guard cell	Habitat	23	8.008	5.414***
		Error	48	1.479	
	Stomata breadth with guard cell	Habitat	23	10.349	22.710***
		Error	48	0.456	
	Epidermal cell length	Habitat	23	315.999	223.038***
		Error	48	1.417	
	Epidermal cell breadth	Habitat	23	3.458	5.562***
		Error	48	0.622	
	Silica bodies number	Habitat	23	6.949	14.120***
		Error	48	0.492	
	Prickles angular number	Habitat	23	0.040	20.159***
		Error	48	0.002	
	Hook number	Habitat	23	0.787	376.989***
		Error	48	0.002	
	Macro hair number	Habitat	23	0.034	15.172***
		Error	48	0.002	

** = significant at $p \leq 0.01$ and *** = significant at $p \leq 0.001$.

4.4 Stem and root anatomical features

Anatomical features of stem and root of 24 accessions of *Cynodon dactylon* are described here based on the Figures (77-78) for stem and Figures (86-87) for root and recorded data in Tables 30 & 32 as follows:

4.4.1 Stem anatomy

Anatomical attributes are illustrated here based on transverse section of stem Figs. (77- 78) and Table 30.

4.4.1.1 Epidermis

The transverse section (TS) of epidermis reveals that the structure was comprised of a single layer of epidermis with thick cuticle apparently. The TS of the entire stem were looked like oval shaped. Epidermis, the outer most layer of the stem was found to be made up of compactly arranged transparent, elongated and rectangular- barrel shaped living parenchyma cells. The cells of the epidermis were radially broader than tangential direction. Outer periclinal wall was thick compared to inner periclinal wall. Due to the presence of cuticle the outside of the outer wall, the epidermal cells looked much thicker. No visual differences were found in length of epidermal cells of all the internodes. Hairs were found to be absent. Highest mean value (2110 µm) for epidermis thickness of stem was found in accession collected from Lalmonirhat and lowest mean value (380 µm) of it was found in accession of Cox's Bazar.

4.4.1.2 Hypodermis

Hypodermis was found to be made up of thick walled lignified sclerenchyma cells. Cell size were small, which acts as heat screen and provides rigidity and mechanical strength to the stem. The overall transverse sections were oval shaped but wavy in look due to the formation of small ridges, where small sized vascular bundles were found. Greatly increased sclerification outside vascular bundles were observed in stem of Barguna, Cox's bazar and Saint Martin's Island. In other accessions low sclerification outside vascular bundles of stem were seen. Sclerenchyma thickness of stem was highest (0.71 cm) in accession of Rangpur and that was lowest (0.07 cm) in case of both Cox's Bazar and Saint Martin's Island.

4.4.1.3 Ground tissue system

In monocot the 3rd tissue system is the ground tissue which does not show distinction into cortex, endodermis, pericycle, pith and pith rays. The ground tissue is made up with parenchyma cells and occupies the whole stem interior. Two types of cells were found to be present in the stem of *Cynodon dactylon* (L). Outer cells were small and angular towards the hypodermis but became large and oval in the inner region. Pith region was large and consisted of parenchyma cells. Abundant intercellular spaces were present in the ground tissue. These spaces communicate with exterior through the stomata present in the epidermis. Down to hypodermis, vascular bundles were found to show some pattern of arrangement. Outer vascular bundles were small and arranged in a complete circle. But the inner vascular bundles had a ring like arrangement somewhat scatteredly. Vascular bundles were found to distribute scatteredly in the cortical areas and those are smaller but more numerous toward the outside than towards the center. The vascular bundles of center were much larger than those next to hypodermis. Parenchyma cells around the vascular bundles were smallest in size and in the pith cells they were larger. Vascular bundles nearest to the epidermis were thickly arranged whereas the cortical bundles were thinly distributed. Hypodermal vascular bundles were of two different sizes, large and small, arranged almost alternately with equal spacing. These vascular bundles were always attached with hypodermal thick wall cells with rare exception. Twin vascular bundle found at the hypodermis layer in *Cynodon dactylon* (L.) Pers. was an exceptional feature.

Cortex was parenchymatous usually having thin wall cells and these cells surrounding a vascular bundle gradually sclerified into thick walled cells. These sclerified cells looked like bundle sheath extensions compared to distal internodes from the ground. In basal internodes chlorenchyma cells gradually transformed into thick wall cells of the chlorenchyma were frequently irregular in shape and arrangement, and exhibited no characteristic pattern. Highest mean value (1.45 cm) for cortex thickness of stem was found in accession collected from Rajshahi and lowest mean value (0.26 cm) was found in accessions of Saint Martin's Island and 2nd lowest mean value (0.31 cm) was found in case of Cox's Bazar.

Cortical cells around the vascular bundles were smaller in size and became sclerified with the age of the culm internode. Basal internodes had greater amount of sclerified cells compared to distal internodes. Pith region was limited to center of the culm, where usually no vascular bundles were found. The cells of the pith were usually large and had intercellular spaces in them.

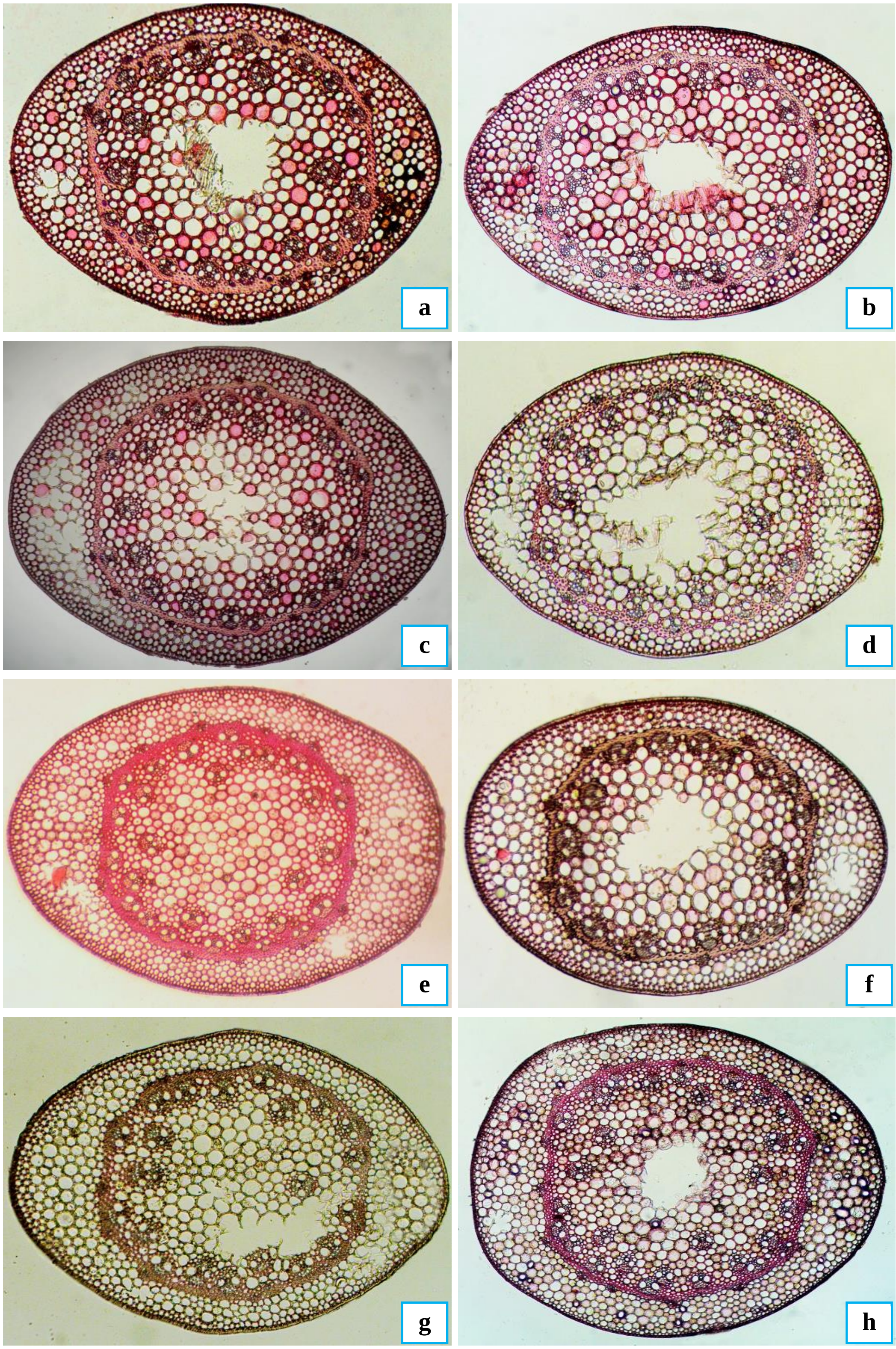
4.4.1.4 Vascular bundles

The vascular bundles were rounded in outline and they contained both phloem and xylem. Phloem lies towards the outside and the xylem on the inner side. Cambium was absent as the whole pro-cambium was consumed in the formation of vascular tissues. The vascular bundles were, therefore, conjoint, collateral but closed. Each vascular bundle was surrounded by a bundle sheath. The bundle sheath was more developed on the outer and the inner sides. Hypodermis and bundle sheaths were found to be coalesced in some of the outer vascular bundles. Phloem was consisted of sieve tubes, companion cells and a few phloem fibers. Phloem parenchyma was absent. Vascular bundle area of stem was highest (8690 μm^2) in accession of Rajshahi and the lowest (480 µm) in accession of Khagrachari. Vascular bundle number per transverse section of stem was highest (43) in accession of Cox's Bazar and 2nd highest value (35) in Barguna and lowest value (17) in accession of Gaibandha.

Xylem was observed in the form of letter Y and it is termed as endarch, i.e., protoxylem lies towards the center of the stem. Xylem was made up of vessels, tracheids, xylem parenchyma and a few xylem fibers. Metaxylem generally was consisted of two large oval or rounded vessels lying at the upper two angles of xylem. The metaxylem vessels had pitted walls. The two vessels were connected with each other by polygonal tracheids having pitted thickenings. The individual vascular bundles were usually more or less circular or oblong to elliptical in transverse section. The bundles were collateral, with the phloem at the pole of the bundle towards the periphery of the culm, where it was usually embedded in a fibrous tissue. The phloem was found to made up of sieve elements and phloem parenchyma, where the sieve elements were remarkable for its diameter. Companion cells were frequent and sieve tubes had thick walls.

Protophloem was visible in many cortical vascular bundles. In most of the cortical and large hypodermal vascular bundles the metaxylem were characterized by two vessels of which the vascular bundle was composed. In rare occasion, more than two metaxylem vessels were noticed but with less diameter. The protoxylem, lying at the opposite pole of the bundle to that at which the phloem was situated, usually had a solitary vessels (true for hypodermal bundles) or short radial row of vessels (true for cortical bundles) that had much shorter diameter than the other vessels. There were many vascular bundles scattered in this heavily sclerified area in accessions of Barguna, Cox's Bazar and Saint Martin's Island.

Metaxylem area of stem was highest (2320 μm^2) in accession of Rajshahi and lowest (480 μm^2) in accession of Saint Martin's Island and 2nd lowest (800 μm^2) in case of Cox's Bazar. Phloem area of stem was highest (4620 μm^2) in accession of Rajshahi and lowest (990 μm^2) in accession of Saint Martin's Island. Phloem and xylem were found to be separated from each other either by fibrous cells or by trachiary elements. Xylem parenchyma and phloem parenchyma with sclerified walls were noticed in most of the bundles. Protoxylem had lie at lower angle of xylem. Xylem parenchyma and a few fibers were found just outside them. Some of the protoxylem vessels and xylem parenchyma cells were found to be dissolved or separated during the rapid growth of the stem to form a cavity called protoxylem cavity or lacuna.



Figs. 77 (a-h): Stem anatomy of *C. dactylon* collected from different habitats of Bangladesh; a) Rangpur, b) Lalmonirhat, c) Dinajpur, d) Thakurgaon, e) Panchagarh, f) Gaibandha, g) Rajshahi, h) Naogaon.

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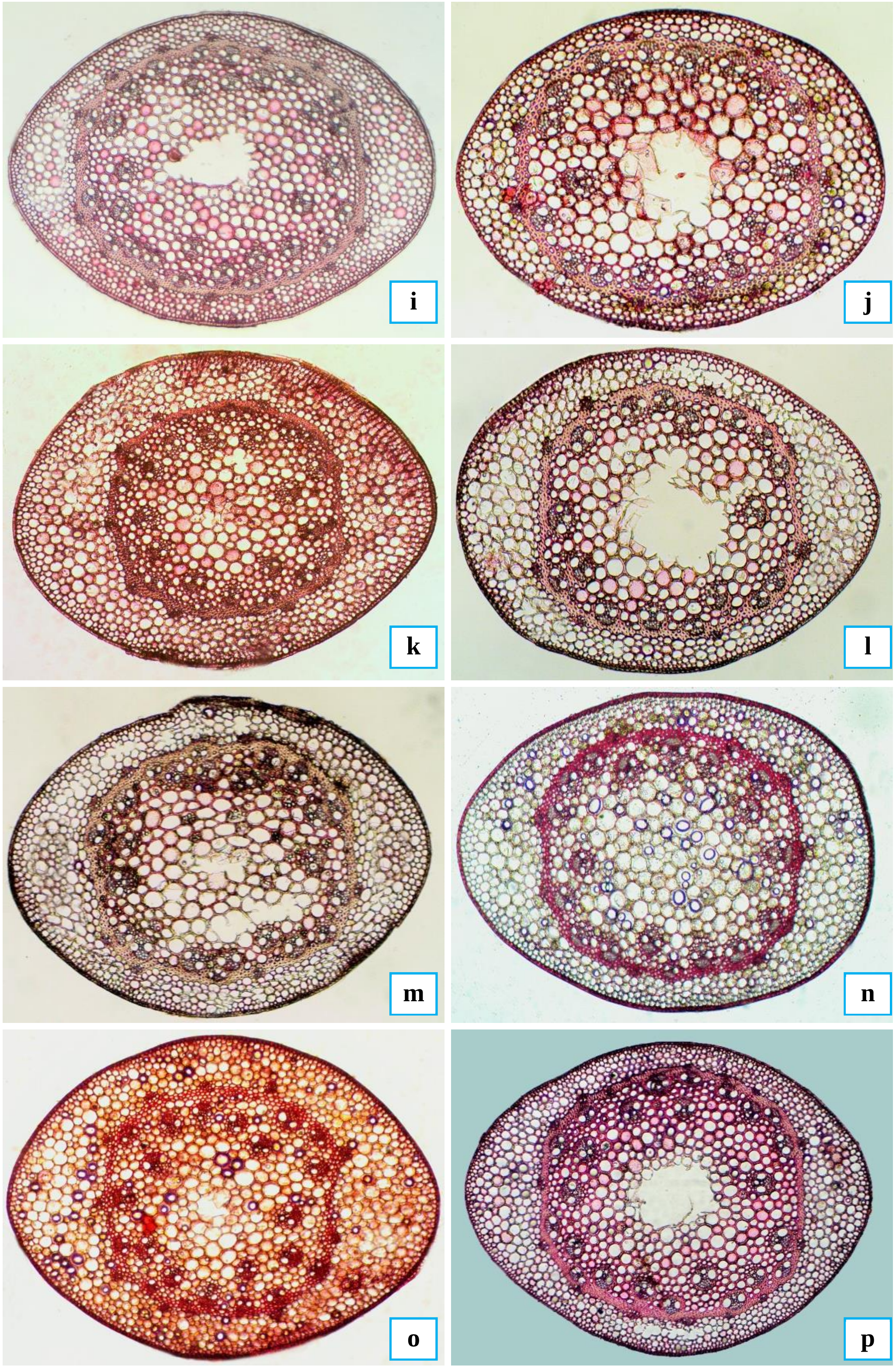

Figs. 77 (i-p): Stem anatomy of *C. dactylon* collected from different habitats of Bangladesh; i) Pabna, j) Gazipur, k) Narsingdi, l) Sherpur, m) Mymensingh, n) Khulna, o) Jessore, p) Jhenaidah.

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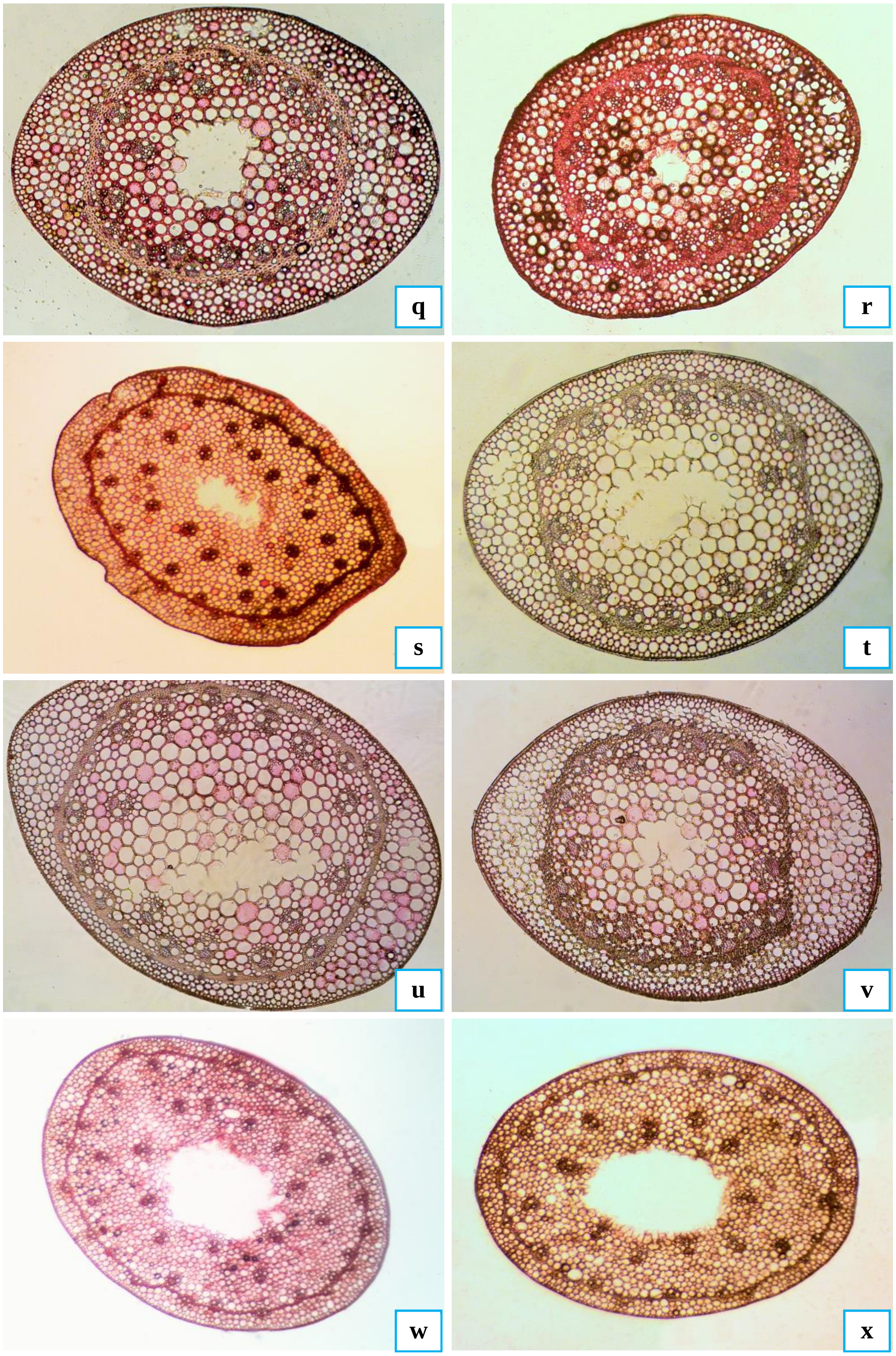
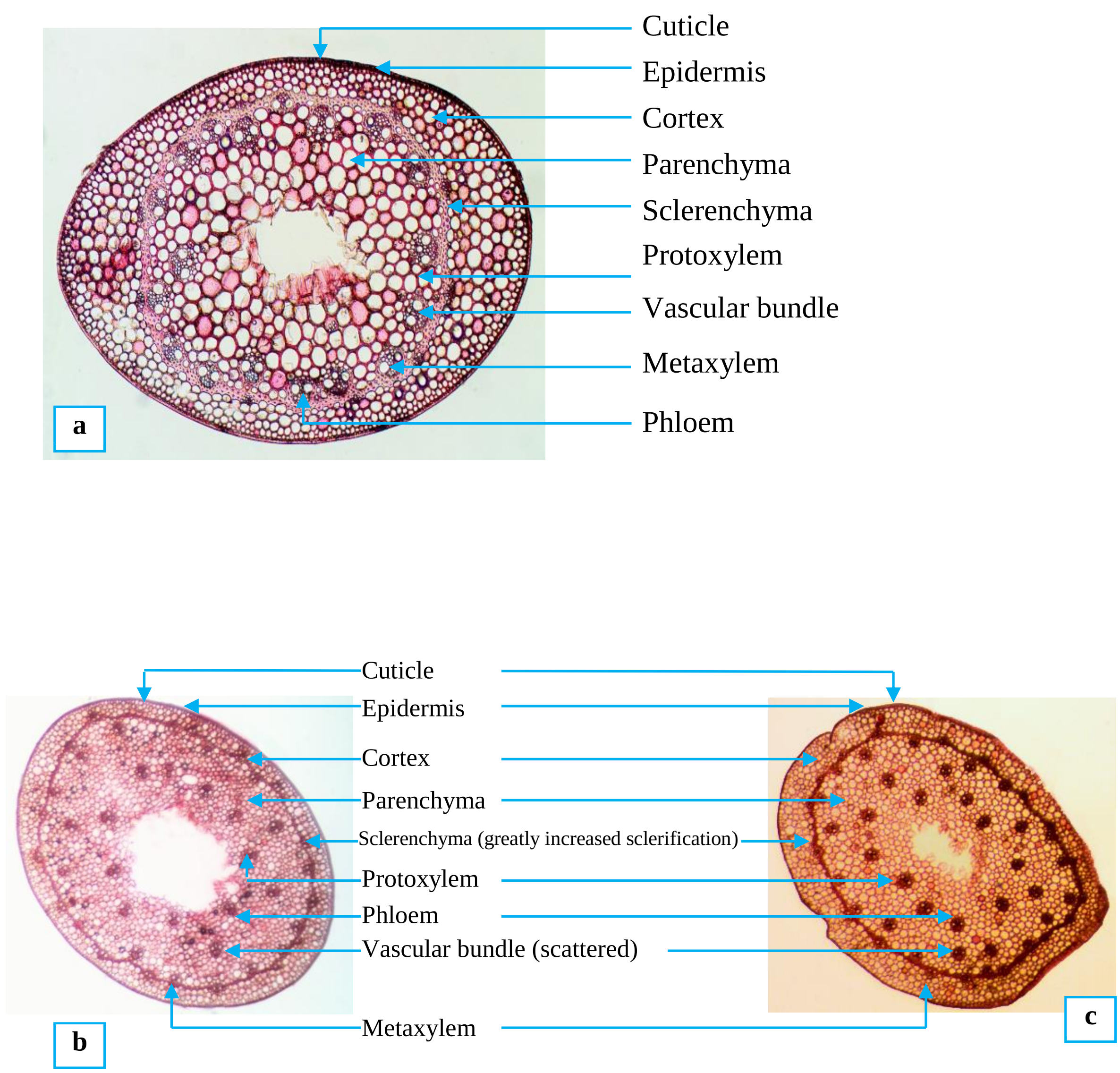


Figs. 77 (q-x): Stem anatomy of *C. dactylon* collected from different habitats of Bangladesh; q) Faridpur, r) Shariatpur, s) Barguna, t) Khagrachari, u) Bandarban, v) Rangamati, w) Cox's Bazar, x) Saint Martin's Island.



Figs. 78 (a-c): Stem anatomy of *C. dactylon* accession collected from saline and non-saline habitat of Bangladesh; a) Lalmonirhat (non-saline habitat), b) Cox's Bazar (saline habitat), c) Barguna (saline habitat).

Table 30: Stem anatomical attributes of different accessions of *Cynodon dactylon* collected from different habitats.

Sl. No.	Name of the habitats	Epidermis thickness (µm)	Cortex thickness (cm)	Sclerenchyma thickness (cm)	Vascular bundle area (µm^2)	Vascular bundle numbers / T.S. of stem	Metaxylem area (µm^2)	Phloem area (µm^2)
		Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE
1.	Rangpur	1480±0.58f	0.89±0.06fgh	0.71±0.01a	5660±0.43l	21±0.14gh	1160±0.36p	3330±0.06d
2.	Lalmonirhat	2110±0.29a	1.36±0.03ab	0.22±0.02bc	6880±0.07d	18±0.20jk	1930±0.06d	4310±0.11b
3.	Dinajpur	1390±0.14i	0.85±0.03gh	0.15±0.03bc	5770±0.09h	24±0.01f	1530±0.43k	4140±0.15c
4.	Thakurgaon	1230±0.58k	0.59±0.03i	0.19±0.01bc	4520±0.13s	19±0.05ij	1150±0.07q	3270±0.54e
5.	Panchagarh	870±0.46q	1.11±0.01cde	0.24±0.03bc	5070±0.16q	27±0.13d	1650±0.57i	1600±0.07q
6.	Gaibandha	1560±0.20d	0.86±0.01gh	0.15±0.06bc	4980±0.17r	17±0.33k	1210±0.07o	3200±0.33f
7.	Rajshahi	1470±0.09g	1.45±0.08a	0.33±0.06b	8690±0.18a	20±0.41hi	2320±0.07a	4620±0.53a
8.	Naogaon	1650±0.35c	1.25±0.05bc	0.22±0.07bc	5850±0.20g	30±0.34c	1570±0.11j	2660±0.15h
9.	Pabna	1360±0.29j	0.97±0.02efg	0.24±0.02bc	5170±0.23o	30±0.23c	1450±0.22k	1660±0.15p
10.	Gazipur	840±0.06r	0.41±0.03j	0.17±0.00bc	4480±0.24t	19±0.57ij	1070±0.40t	1810±0.40m
11.	Narsingdi	1120±0.14l	1.24±0.06bc	0.22±0.08bc	5490±0.28m	22±0.03g	1270±0.08n	1770±0.19n
12.	Sherpur	1790±0.10b	1.05±0.02def	0.35±0.02b	6430±0.06f	18±0.04jk	1800±0.08f	3010±0.41g
13.	Mymensingh	1550±0.34e	1.22±0.04bc	0.30±0.12b	8630±0.58b	23±0.52f	1730±0.12h	2000±0.14l
14.	Khulna	1400±0.32h	1.20±0.05bcd	0.32±0.10b	6790±0.08e	19±0.05ij	2120±0.11c	2120±0.42k
15.	Jessore	890±0.07p	1.01±0.03efg	0.18±0.05bc	3370±0.58u	21±0.44gh	1270±0.45n	1600±0.46q
16.	Jhenaidah	670±0.03u	0.95±0.05efg	0.19±0.09bc	5760±0.47i	24±0.37f	1890±0.15e	1670±0.15o
17.	Faridpur	670±0.19u	0.96±0.09efg	0.23±0.02bc	5740±0.55j	20±0.41hi	1080±0.56s	1520±0.57t
18.	Shariatpur	910±0.13n	0.84±0.02gh	0.22±0.11bc	5230±0.13n	19±0.45ij	1090±0.50r	1530±0.14s
19.	Barguna	690±0.26s	0.72±0.03hi	0.15±0.05bc	5120±0.27p	35±0.50b	1440±0.44l	2160±0.31j
20.	Khagrachari	900±0.15o	0.76±0.16h	0.19±0.05bc	480±0.59x	21±0.37gh	1400±0.09m	1590±0.51r
21.	Bandarban	990±0.34m	0.99±0.05efg	0.32±0.01b	7280±0.50c	21±0.32gh	2180±0.55b	2000±0.16l
22.	Rangamati	680±0.16t	0.83±0.05gh	0.24±0.10bc	5670±0.25k	20±0.28hi	1760±0.39g	2210±0.07i
23.	Cox's Bazar	380±0.06w	0.31±0.06j	0.07±0.03c	3120±0.06v	43±0.44a	800±0.15u	1270±0.06u
24.	St. Martin's Island	480±0.17v	0.26±0.03j	0.07±0.01c	2270±0.17w	26±0.35e	480±0.14v	990±0.41v

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

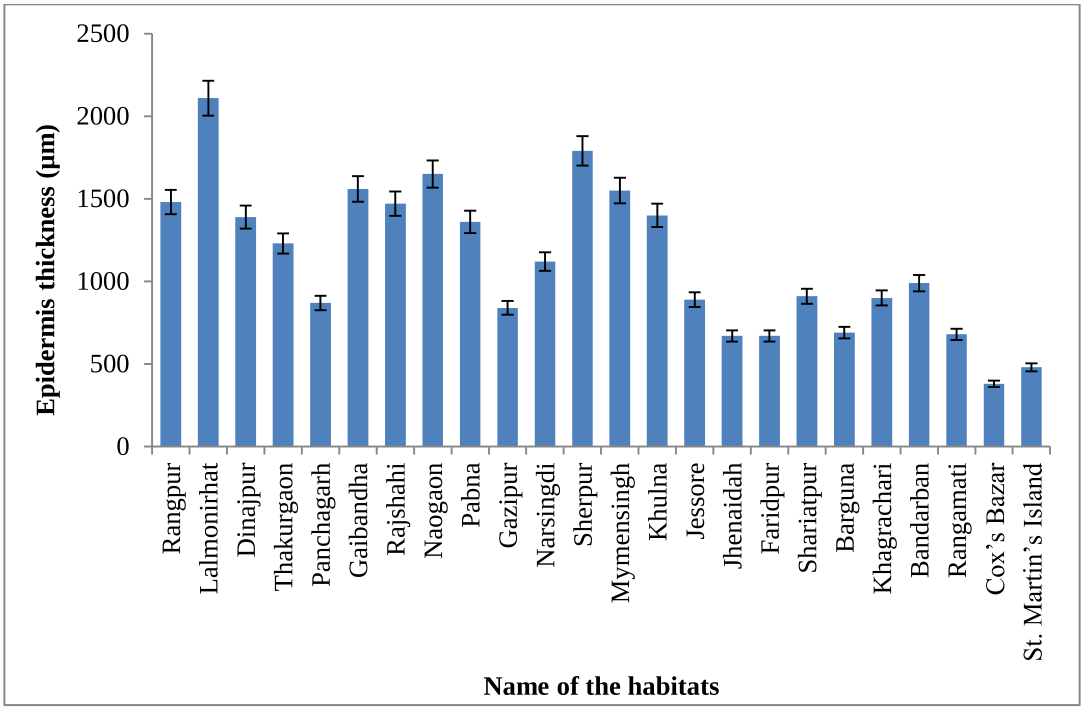


Fig. 79: Epidermis thickness in stem of *C. dactylon* accessions collected from different habitats.

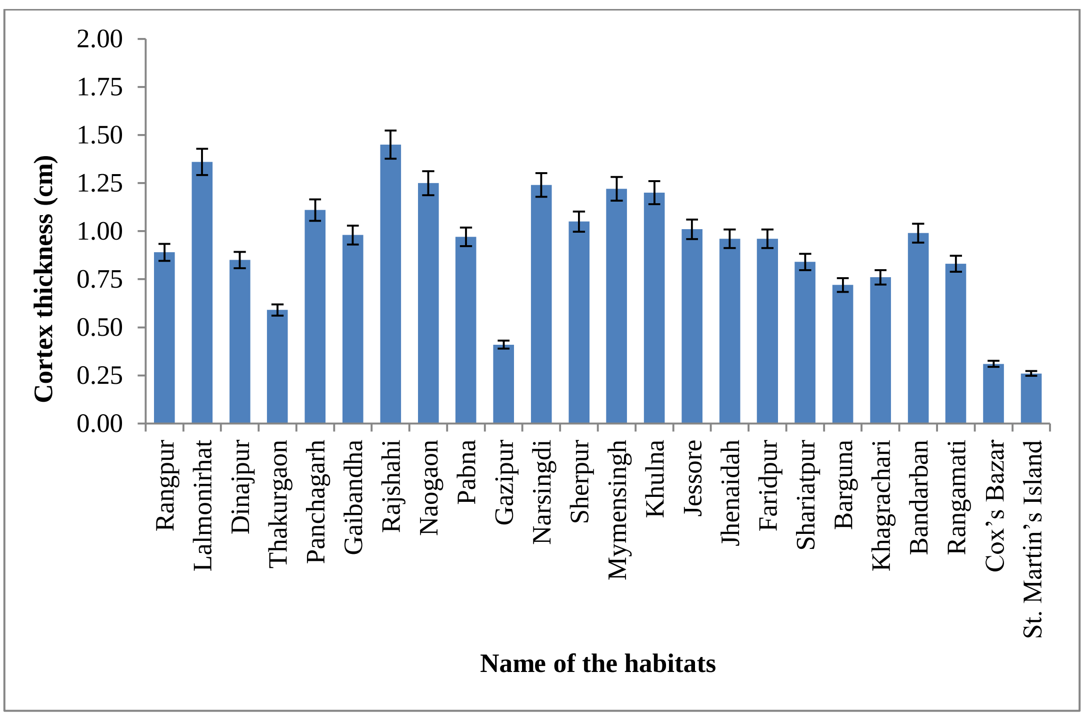


Fig. 80: Cortex thickness in stem of *C. dactylon* accessions collected from different habitats.

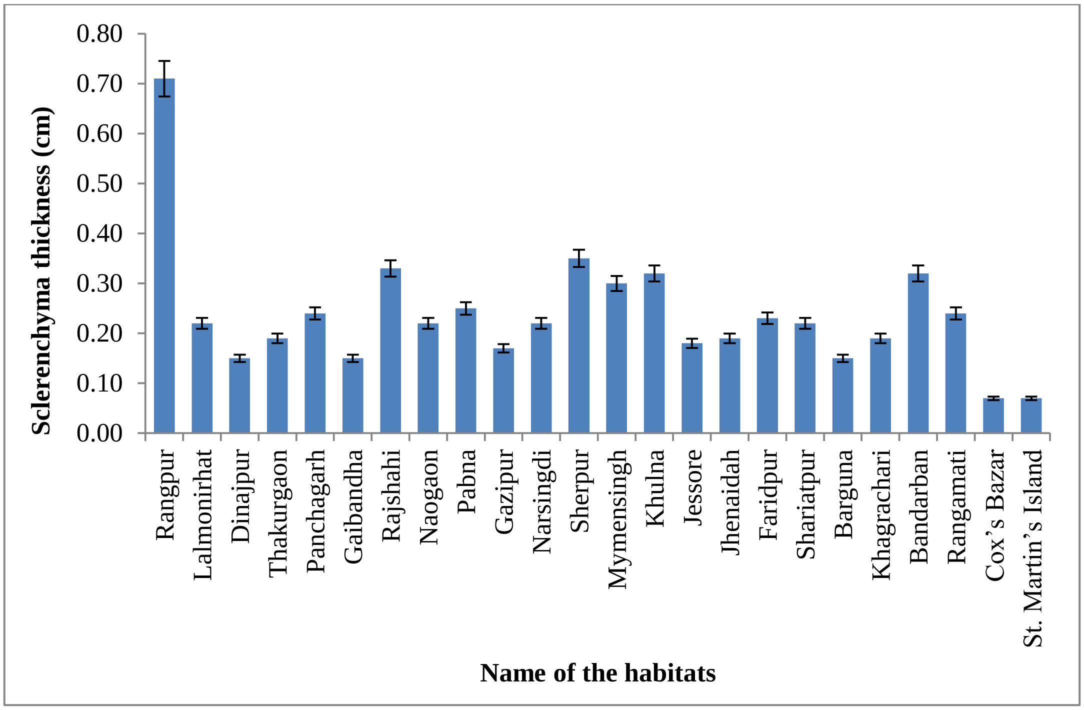


Fig. 81: Sclerenchyma thickness in stem of *C. dactylon* accessions collected from different habitats.

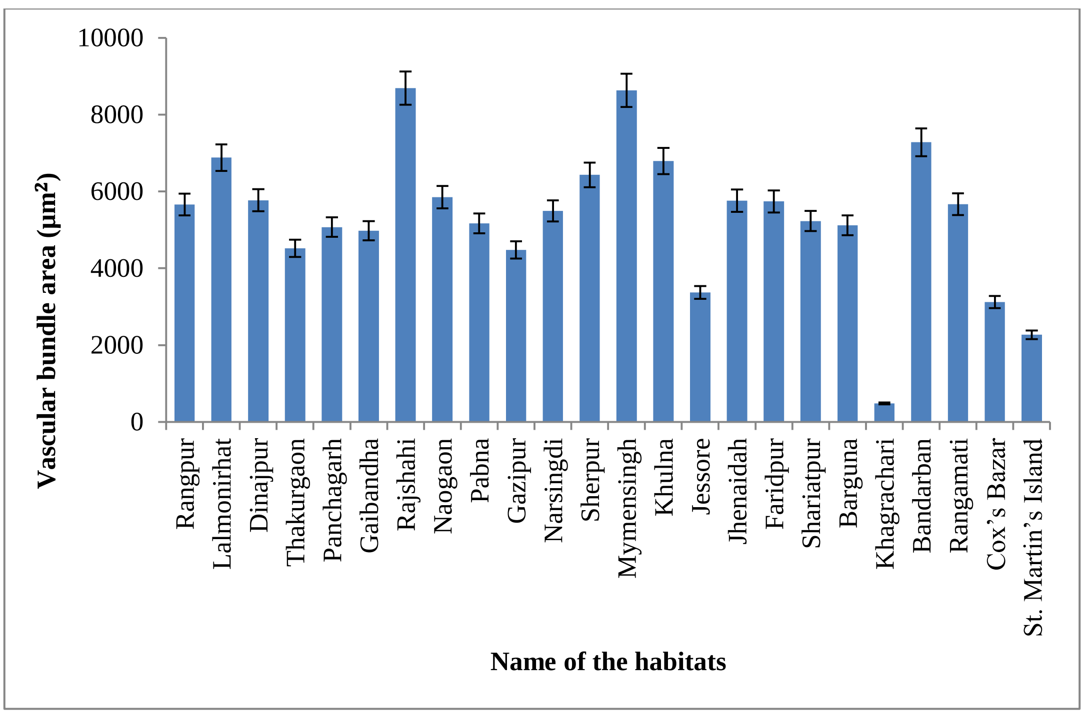


Fig. 82: Vascular bundle area in stem of *C. dactylon* accessions collected from different habitats.

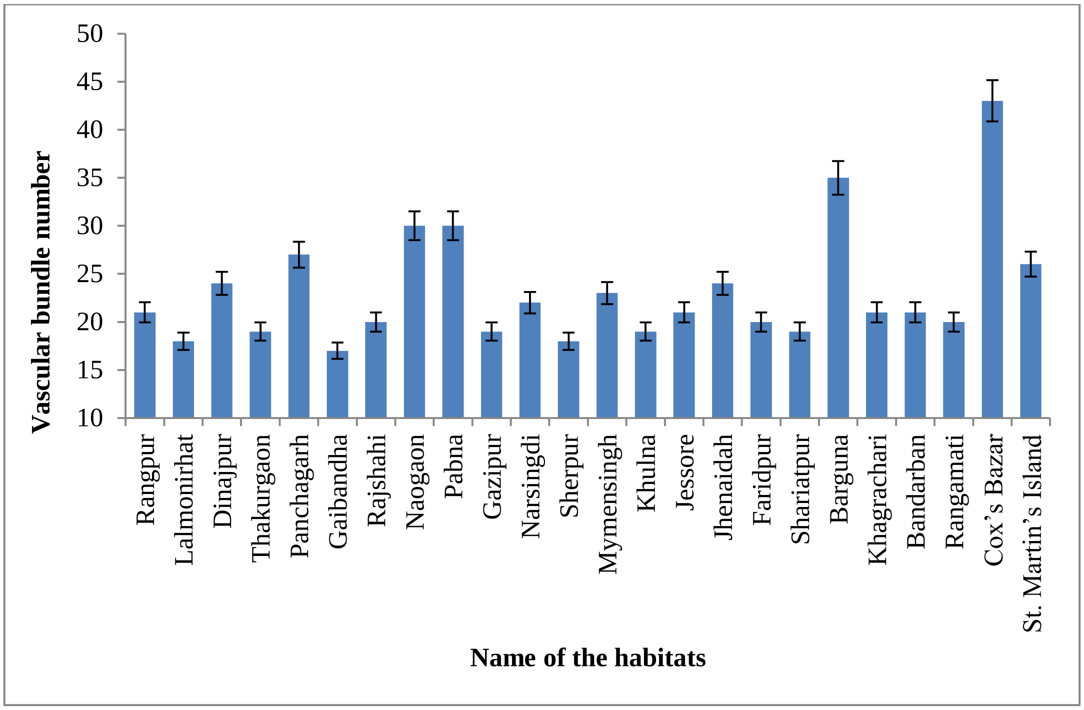


Fig. 83: Vascular bundle number in stem of *C. dactylon* accessions collected from different habitats.

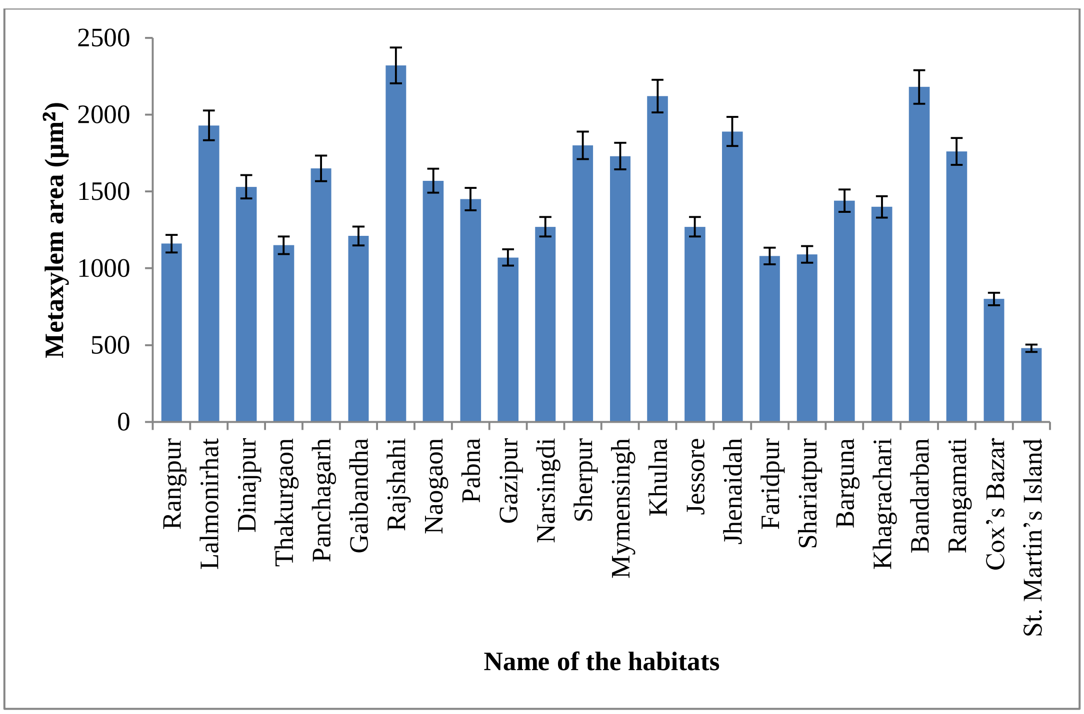


Fig. 84: Metaxylem area in stem of *C. dactylon* accessions collected from different habitats.

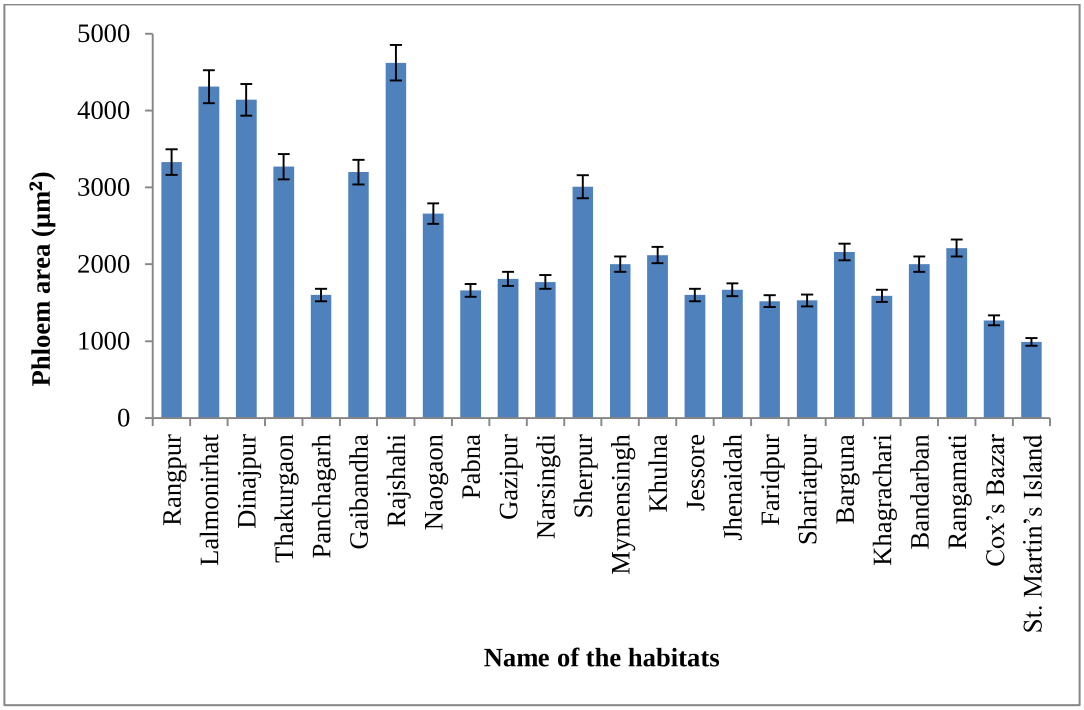


Fig. 85: Phloem area in stem of *C. dactylon* accessions collected from different habitats.

Analysis of variance (Table 31) reveals that all the anatomical features of stem showed significant differences at 0.1% level among the 24 accessions of *Cynodon dactylon* collected from different habitats of Bangladesh.

Table 31: Analysis of variance subjected to variation of different anatomical attributes of stem of *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Stem	Epidermis thickness	Habitat	23	600486.194	2.646***
		Error	48	0.227	
	Cortex thickness	Habitat	23	0.286	31.715***
		Error	48	0.009	
	Sclerenchyma thickness	Habitat	23	0.047	4.493***
		Error	48	0.010	
	Vascular bundle area	Habitat	23	9918495.109	3.153***
		Error	48	0.315	
	Vascular bundle number	Habitat	23	112.429	331.756***
		Error	48	0.339	
	Metaxylem area	Habitat	23	597560.326	1.998***
		Error	48	0.299	
	Phloem area	Habitat	23	3033156.522	9.848***
		Error	48	0.308	

*** = significant at $p \leq 0.001$.

4.4.2 Root anatomy

In this case root showed the features of epidermis, cortex, endodermis, pericycle, vascular bundle, conjunctive tissues and pith (Figs. 86-87 and Table 32).

4.4.2.1 Epidermis

Transverse section of mature root showed epidermis composed of a single layer of thin walled, tangentially elongated to irregular shaped cells. Highest mean value (4500 µm) of epidermis thickness of root was found in accession collected from Rajshahi and lowest mean value (1150 µm) was in accession of Barguna. But exceptional result was found in accessions of Cox's Bazar and Saint Martin's Island, where the epidermis thickness of root was high (3240 µm and 3400 µm, respectively) compared to that of other twenty-one accessions.

4.4.2.2 Cortex

Cortex was found to be differentiated into two zones, 1 or 2 layers of smaller, thin-walled, polygonal, lignified sclerenchymatous, and 4 to 6 layers of larger thin walled, elongated parenchymatous cells. Lysigenous aerenchyma was observed in roots of *Cyodon dactylon* in the present study. Highest mean value (2.48 cm) for cortex thickness of root was found in accession collected from Cox's Bazar and 2nd highest value (1.93 cm) in accession of Saint Martin's Island and lowest mean value (0.38 cm) was found in Gaibandha.

4.4.2.3 Endodermis

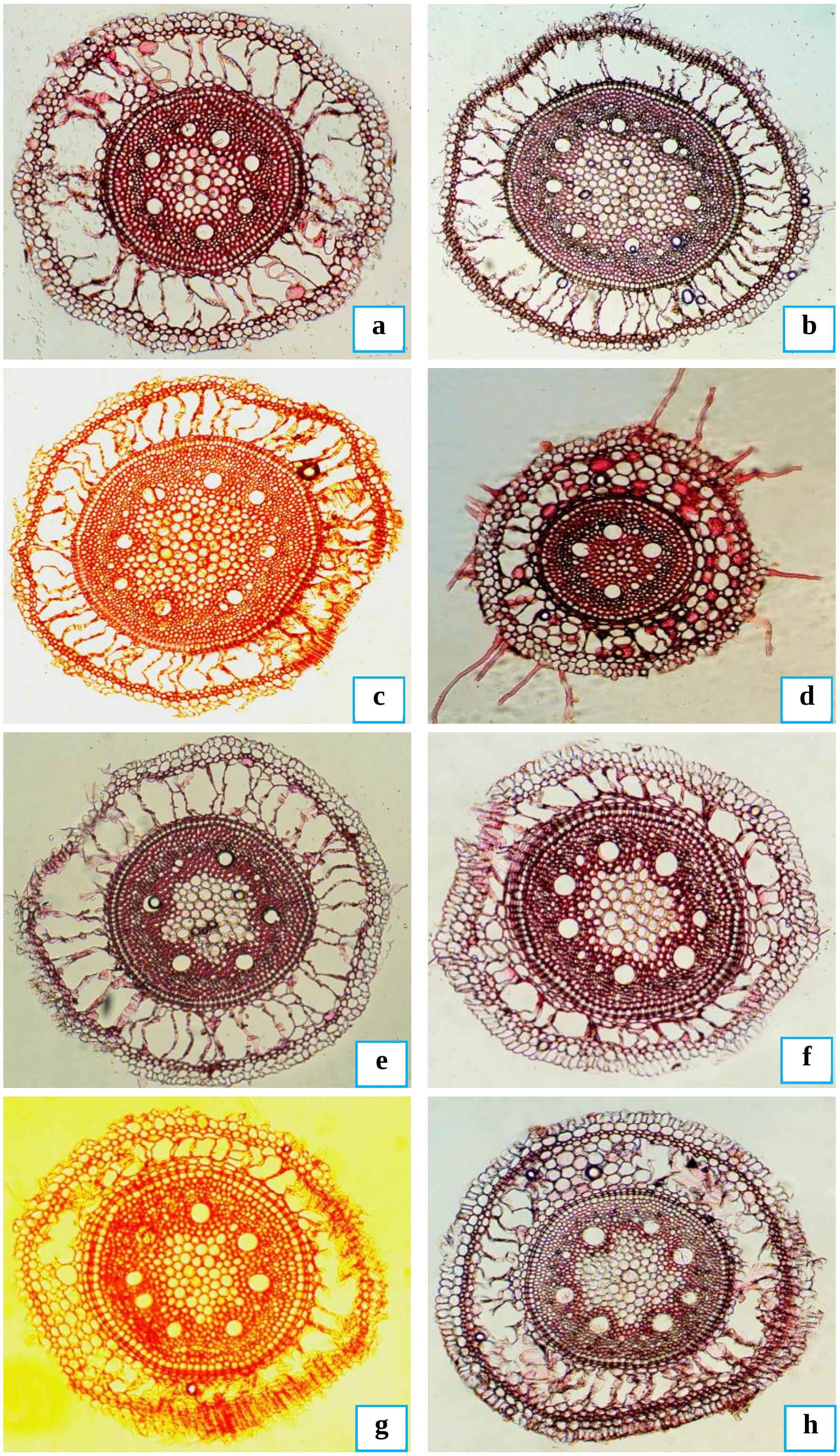
Endodermis was quite distinct, single layered, thick walled with tangentially elongated cells. Endodermis thickness of root was highest (6390 µm) in accession of Saint Martin's Island, 2nd highest value (4243 µm) in accession of Rajshahi, 3rd highest value (3920 µm) in accession of Cox's Bazar and lowest (1140 µm) in accession of Jhenaidah.

4.4.2.4 Sclerenchyma

Sclerenchyma thickness of root was highest (0.24 cm) in accession of Rajshahi and also high (0.21 cm, 0.20 cm, 0.14 cm) values were found in accessions of Cox's Bazar, Saint Martin's Island and Barguna, respectively and lowest (0.07 cm) in accession of Jhenaidah. Intensive sclerification in outer cortex, vascular region and endodermis layer were observed in root of Barguna, Cox's Bazar and Saint Martin's Island. In roots of others accessions low sclerification was noticed.

4.4.2.5 Vascular bundles

Vascular bundles consisting of xylem and phloem were found to be arranged in a ring on different radials; xylem exarch, having usual elements; center occupied by wide pith, composed of oval to rounded thick walled parenchymatous cells containing numerous simple, round to oval or angular starch grains. Highest mean value (5148 μm^2) of metaxylem area of root was found in accession collected from Rajshahi and lowest value (1800 μm^2) was in accession of Cox's Bazar. Highest mean value (3350 μm^2) for phloem area of root was also found in accession collected from Rajshahi and lowest value (830 μm^2) found in Barguna. Pith thickness of root was highest (2.74 cm) in accession of Mymensingh and lowest (0.27 cm) in case of Barguna. Pith cell area of root was highest (3300 μm^2) in accession of Rajshahi and lowest (240 μm^2) in accession of Cox's Bazar.



Figs. 86 (a-h): Root anatomy of *Cynodon dactylon* accessions collected from different habitats; a) Rangpur, b) Lalmonirhat, c) Dinajpur, d) Thakurgaon, e) Panchagarh, f) Gaibandha, g) Rajshahi, h) Naogaon.

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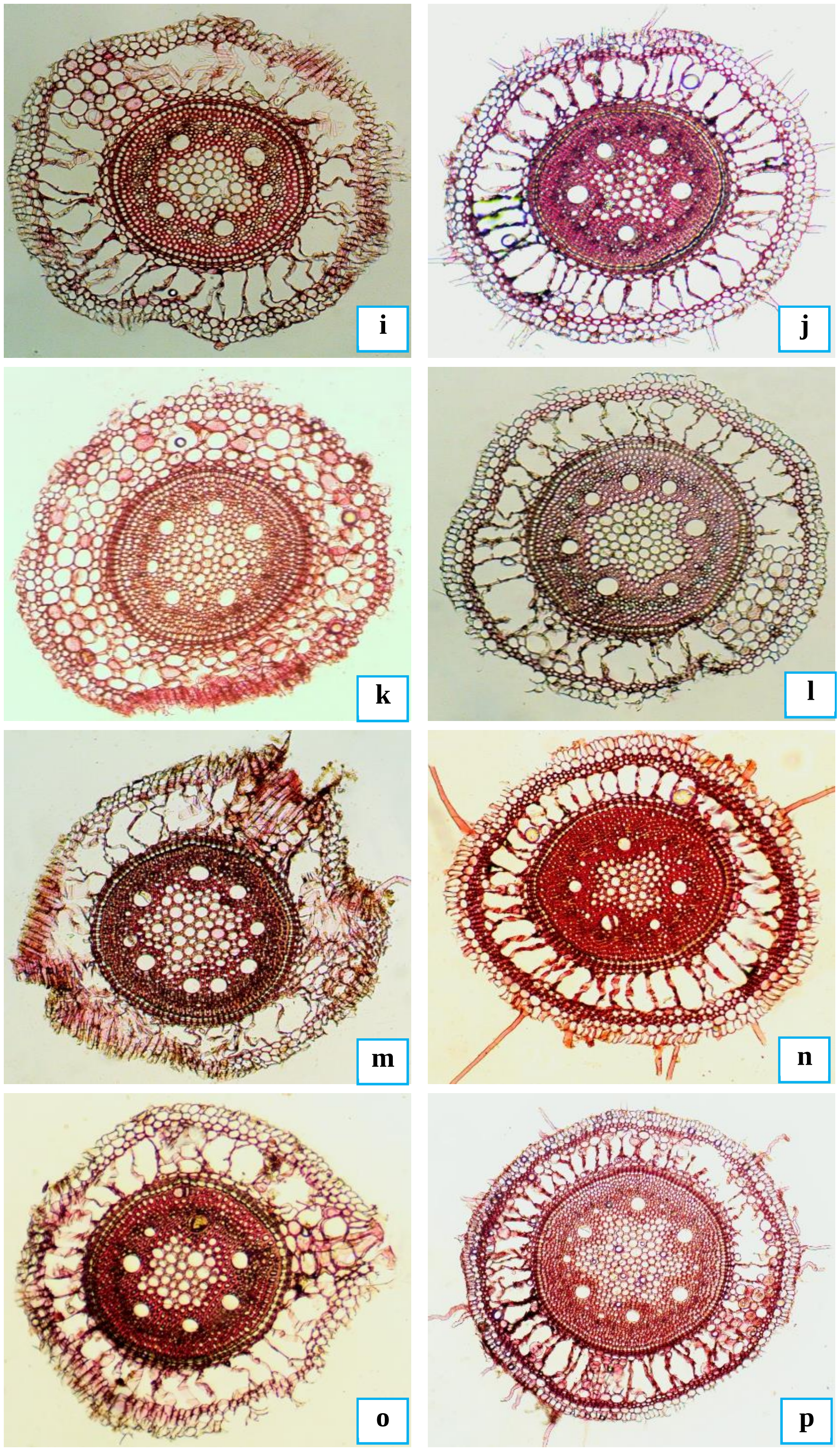

Figs. 86 (i-p): Root anatomy of *Cynodon dactylon* accessions collected from different habitats; i) Pabna, j) Gazipur, k) Narsingdi, l) Sherpur, m) Mymensingh, n) Khulna, o) Jessore, p) Jhenaidah.

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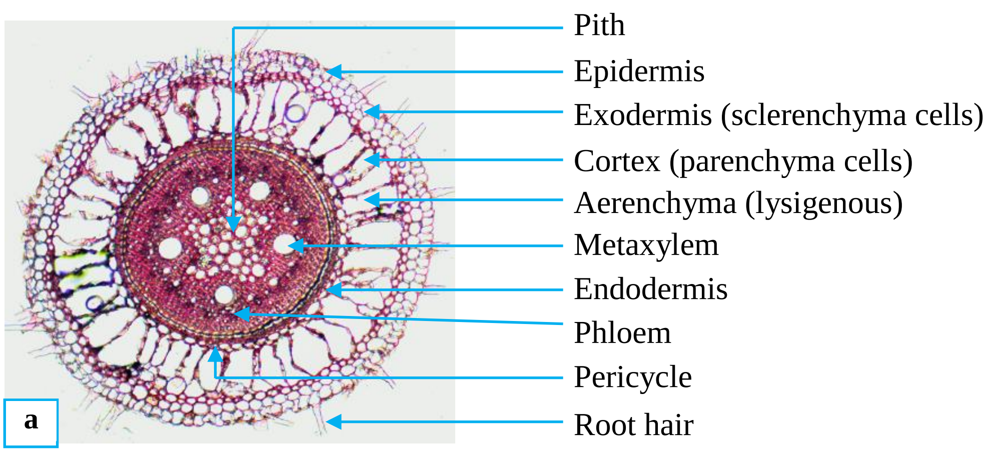
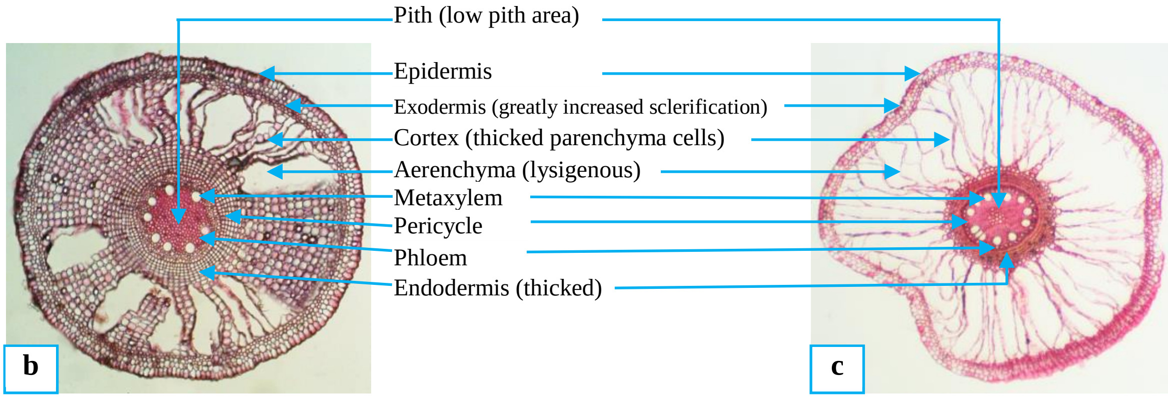
Figs. 86 (q-x): Root anatomy of *Cynodon dactylon* accessions collected from different habitats; q) Faridpur, r) Shariatpur, s) Barguna, t) Khagrachari, u) Bandarban, v) Rangamati, w) Cox's Bazar, x) Saint Martin's Island.





Figs. 87 (a-c): Root anatomy of *C. dactylon* accessions collected from different habitats; a) Gazipur (non-saline habitat), b) Saint Martin's Island (saline habitat), c) Barguna (saline habitat).

Table 32: Root anatomical attributes of different accessions of *Cynodon dactylon* collected from different habitats.

Sl. No.	Name of the habitats	Epidermis thickness (µm)	Cortex thickness (cm)	Sclerenchyma thickness (cm)	Endodermis thickness (µm)	Metaxylem area (µm^2)	Phloem area (µm^2)	Pith thickness (cm)	Pith cell area (µm^2)
		Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE	Mean±SE
1.	Rangpur	2790±0.12l	0.89±0.04efg	0.13±0.03abc	2240±0.14f	2710±0.53k	2080±0.28i	1.36±0.08efgh	1660±0.14m
2.	Lalmonirhat	3430±0.53c	0.93±0.01efg	0.19±0.02abc	1960±0.40l	2950±0.50g	2620±0.23d	2.49±0.08bc	2130±0.23e
3.	Dinajpur	2880±0.21k	0.96±0.10def	0.18±0.01abc	2210±0.47g	3090±0.08d	3310±0.14b	2.64±0.04ab	2040±0.14f
4.	Thakurgaon	2660±0.48p	0.60±0.03jk	0.16±0.03abc	1970±0.53k	2880±0.15i	2470±0.50f	1.06±0.04ij	1100±0.37s
5.	Panchagarh	2757±0.09m	1.00±0.01cdef	0.15±0.01abc	1640±0.37t	2480±0.33o	2060±0.24j	1.48±0.05e	2333±0.45c
6.	Gaibandha	2720±0.39o	0.38±0.04l	0.11±0.05bc	1680±0.14s	2730±0.42j	2500±0.35e	1.22±0.02ghi	1960±0.42g
7.	Rajshahi	4500±0.05a	1.16±0.03c	0.24±0.05a	4243±0.10b	5148±0.38a	3350±0.32a	2.43±0.01cd	3300±0.49a
8.	Naogaon	2980±0.16i	0.71±0.03hijk	0.13±0.03abc	1480±0.38u	2580±0.27m	1620±0.35s	1.29±0.02efgh	1820±0.15i
9.	Pabna	2630±0.30q	0.88±0.07efg	0.18±0.05abc	2600±0.14e	3150±0.46c	2160±0.38h	1.46±0.03ef	1720±0.38l
10.	Gazipur	2610±0.27s	0.60±0.10jk	0.11±0.06bc	1690±0.37r	2440±0.23r	1750±0.44n	1.05±0.03ij	1590±0.39q
11.	Narsingdi	2620±0.48r	0.84±0.04fgh	0.12±0.02abc	1890±0.51o	2470±0.18p	1740±0.39o	1.39±0.05efg	1790±0.32j
12.	Sherpur	2400±0.11u	0.69±0.09higk	0.13±0.07abc	1930±0.16n	2560±0.26n	1890±0.12l	1.33±0.06efgh	1820±0.36i
13.	Mymensingh	3640±0.45b	1.13±0.07cd	0.19±0.01abc	2610±0.50d	3980±0.34b	2770±0.19c	2.74±0.03a	3110±0.32b
14.	Khulna	3110±0.48h	0.67±0.02hijk	0.16±0.02abc	1980±0.09j	2170±0.10t	1580±0.41t	1.07±0.11ij	1600±0.52p
15.	Jessore	2290±0.33v	0.57±0.05k	0.14±0.04abc	1970±0.36k	2160±0.42u	1750±0.41n	1.05±0.09	1880±0.24h
16.	Jhenaidah	2950±0.19j	0.99±0.01cdef	0.07±0.03c	1140±0.52v	3050±0.05e	2260±0.14g	2.28±0.05d	1770±0.17k
17.	Faridpur	3200±0.47f	0.78±0.06ghi	0.17±0.02abc	1940±0.35m	2320±0.16s	1680±0.40q	0.91±0.07j	1550±0.38r
18.	Shariatpur	2750±0.34n	0.66±0.04ijk	0.13±0.03abc	1970±0.39k	2890±0.25h	1650±0.29r	1.33±0.03efgh	1770±0.49k
19.	Barguna	1150±0.35x	0.99±0.06cdef	0.14±0.02abc	1707±0.24q	2640±0.46i	830±0.29v	0.27±0.05k	260±0.41t
20.	Khagrachari	1790±0.32w	1.02±0.01cde	0.13±0.03abc	2100±0.22h	2970±0.10f	2050±0.36k	1.34±0.17efgh	2210±0.41d
21.	Bandarban	2450±0.36t	0.66±0.07ijk	0.12±0.04abc	2020±0.12i	2460±0.25q	1820±0.44m	1.17±0.06hi	1620±0.35o
22.	Rangamati	3120±0.36g	0.77±0.02ghij	0.19±0.03abc	1840±0.45p	2950±0.23g	1740±0.40o	1.20±0.06ghi	1630±0.27n
23.	Cox's Bazar	3240±0.24e	2.48±0.06a	0.21±0.01ab	3920±0.40c	1800±0.14w	1710±0.14p	0.89±0.06j	240±0.41u
24.	St. Martin's Island	3400±0.49d	1.93±0.04b	0.20±0.04ab	6390±0.14a	2070±0.17v	1430±0.40u	1.25±0.05fghi	630±0.47s

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

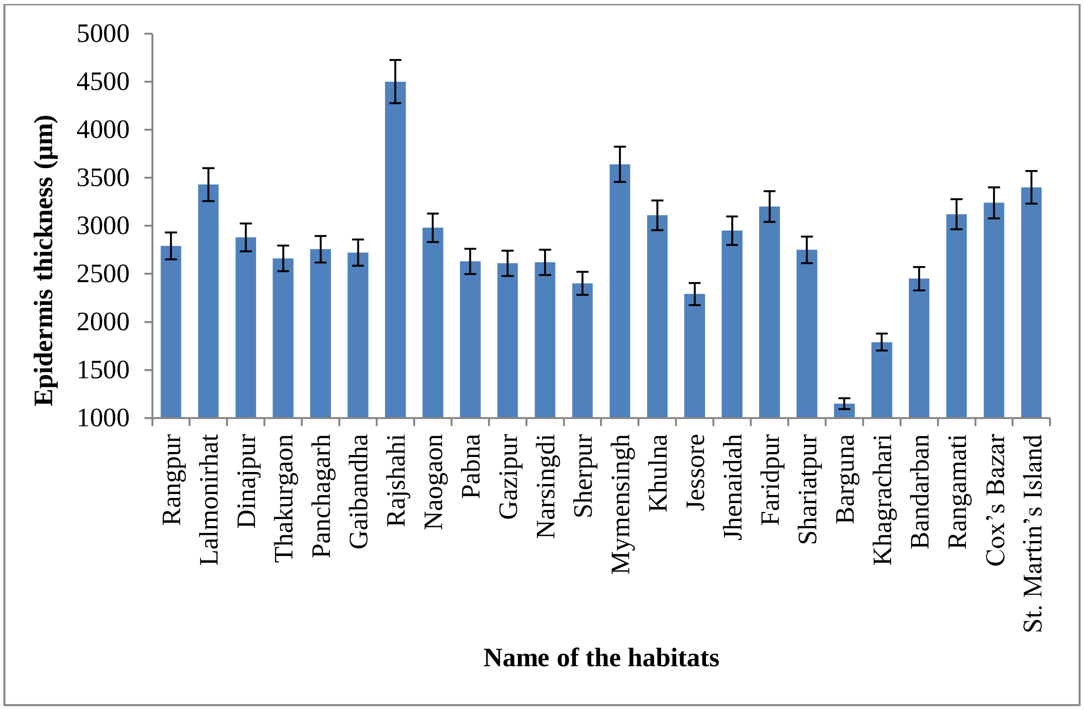


Fig. 88: Epidermis thickness in root of *C. dactylon* accessions collected from different habitats.

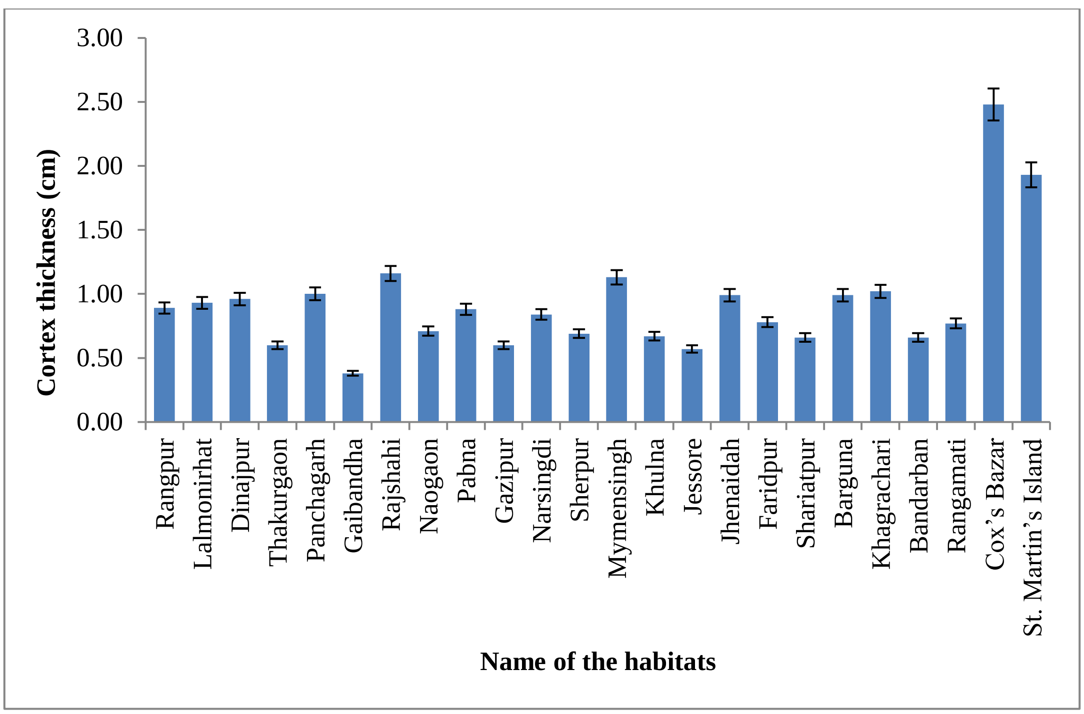


Fig. 89: Cortex thickness in root of *C. dactylon* accessions collected from different habitats.

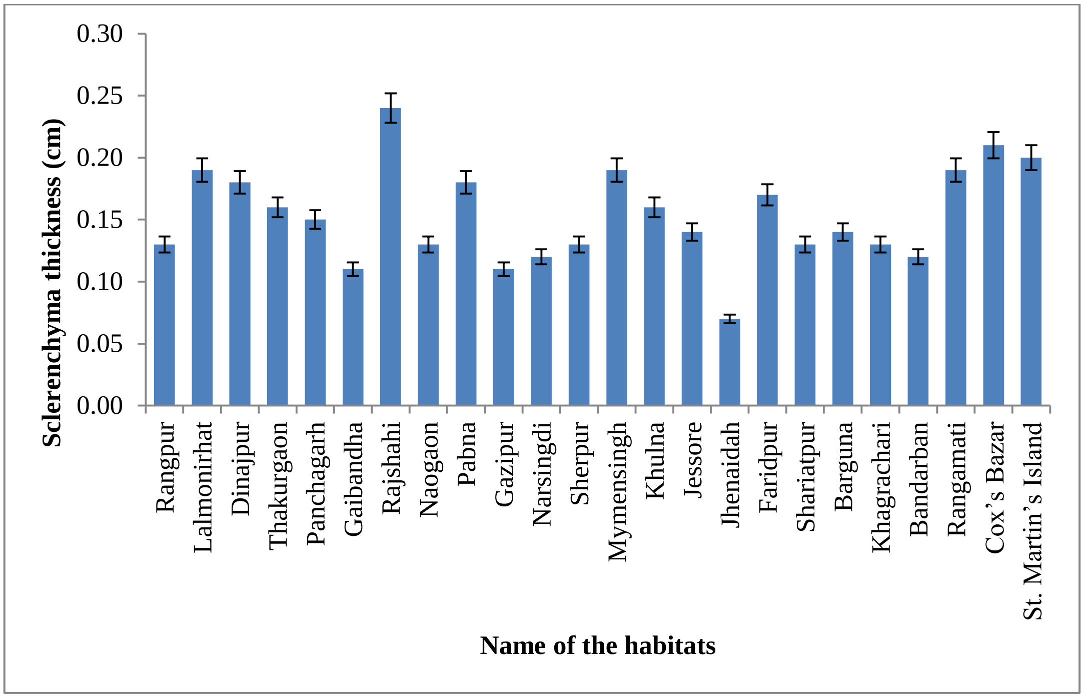


Fig. 90: Sclerenchyma thickness in root of *C. dactylon* accessions collected from different habitats.

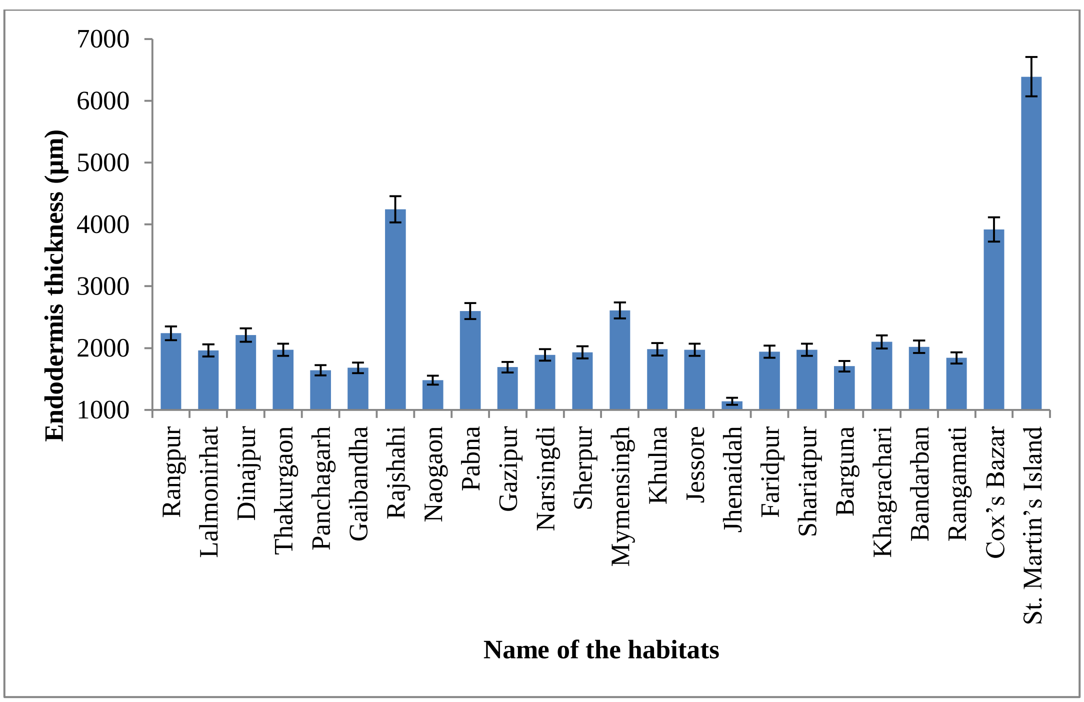


Fig. 91: Endodermis thickness in root of *C. dactylon* accessions collected from different habitats.

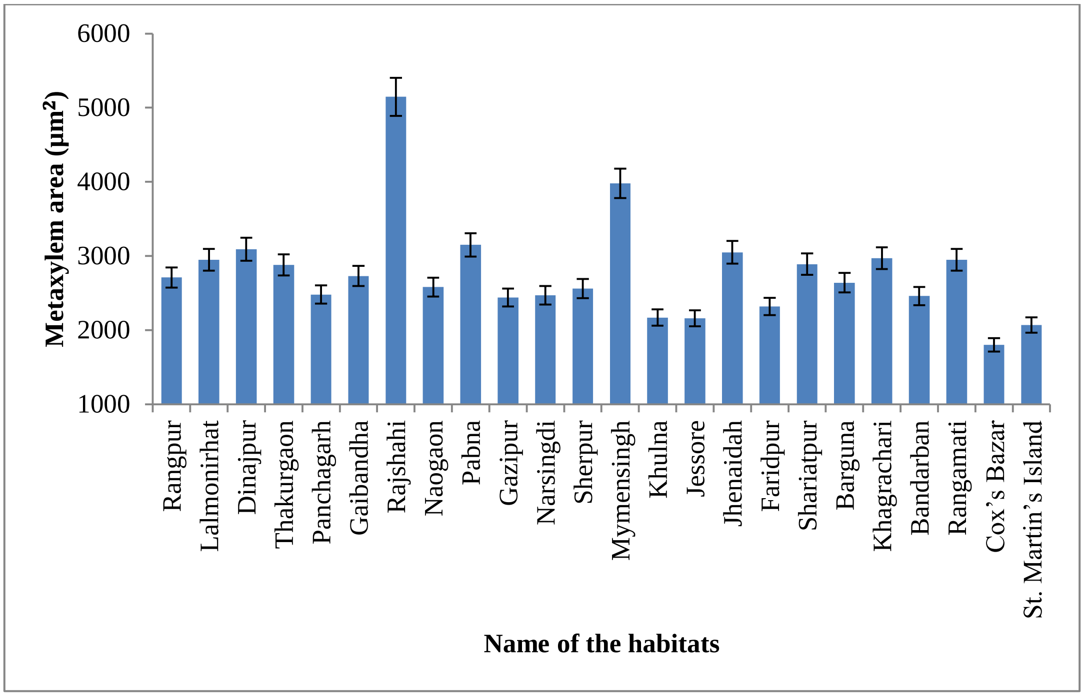


Fig. 92: Metaxylem area in root of *C. dactylon* accessions collected from different habitats.

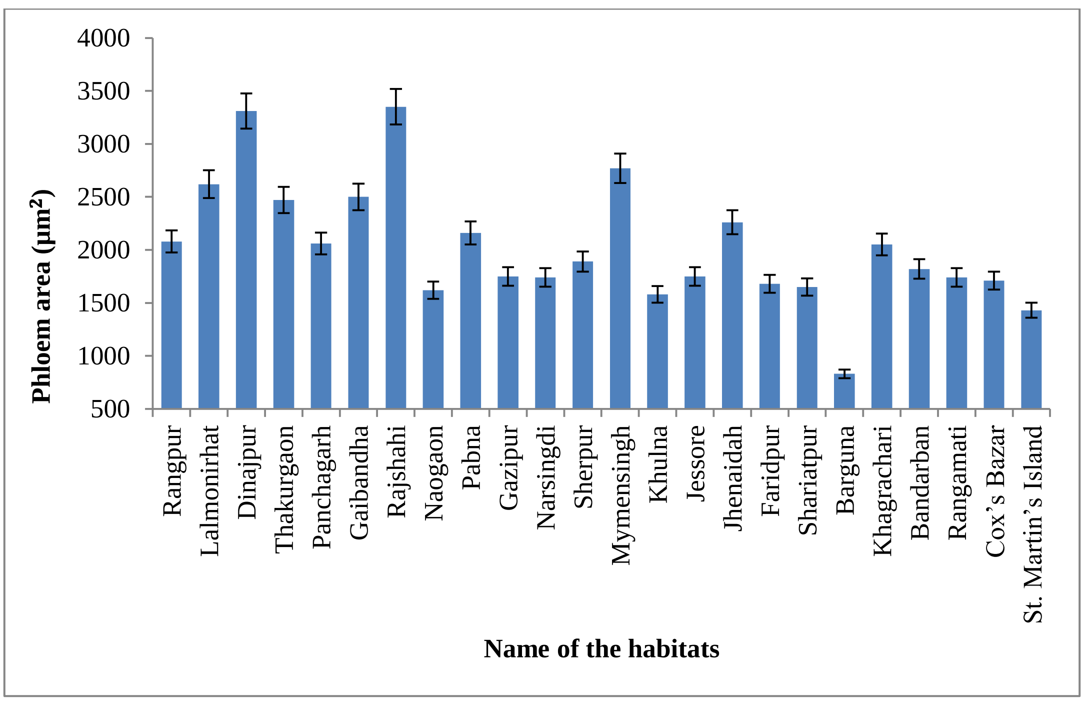


Fig. 93: Phloem area in root of *C. dactylon* accessions collected from different habitats.

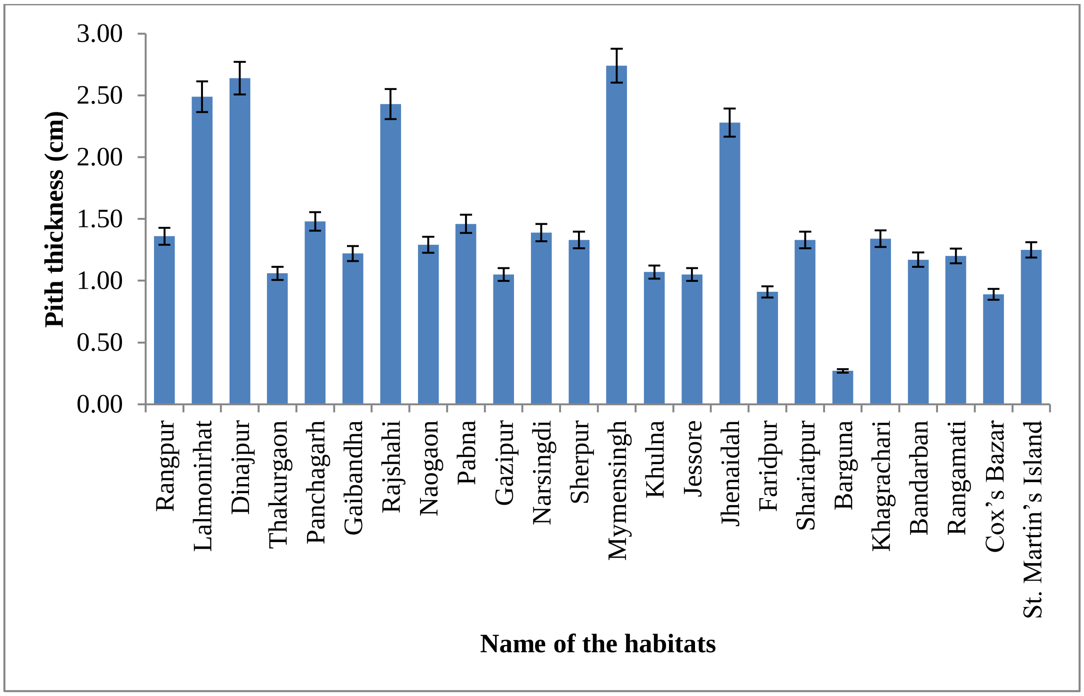


Fig. 94: Pith thickness in root of *C. dactylon* accessions collected from different habitats.

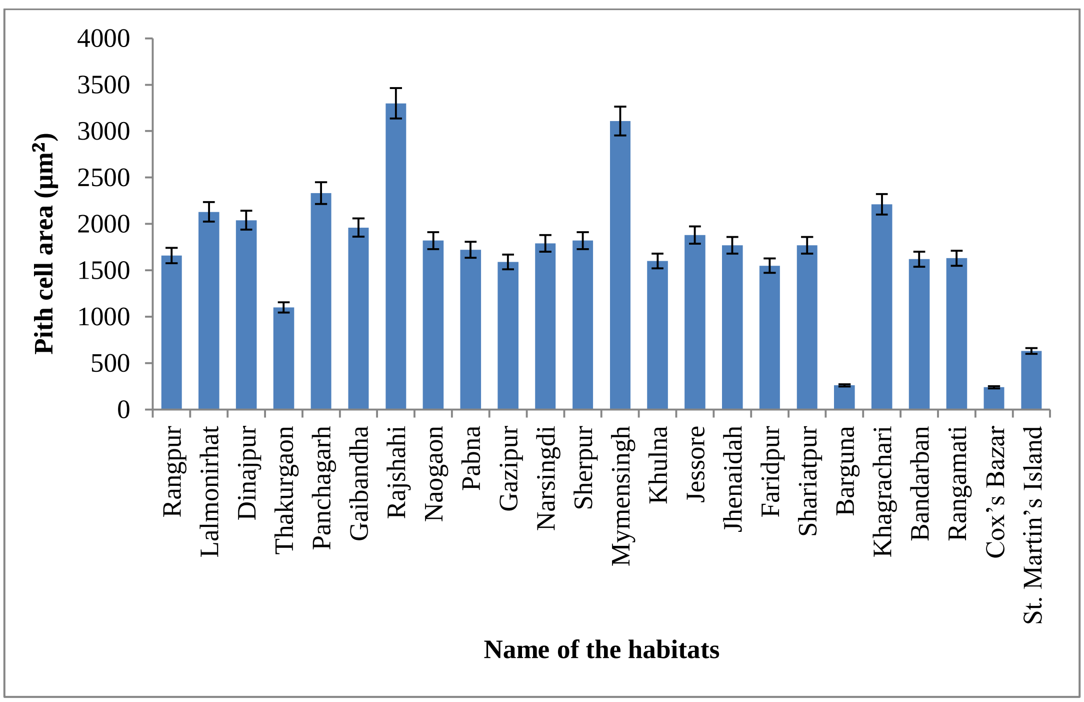


Fig. 95: Pith cell area in root of *C. dactylon* accessions collected from different habitats.

Statistical analysis (Table 33) reveals that except sclerenchyma thickness, all the root anatomical characters showed significant differences at 0.1 % level among the accessions of *Cynodon dactylon* collected from different habitats of Bangladesh.

Table 33: Analysis of variance subjected to variation of different anatomical attributes of root of *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

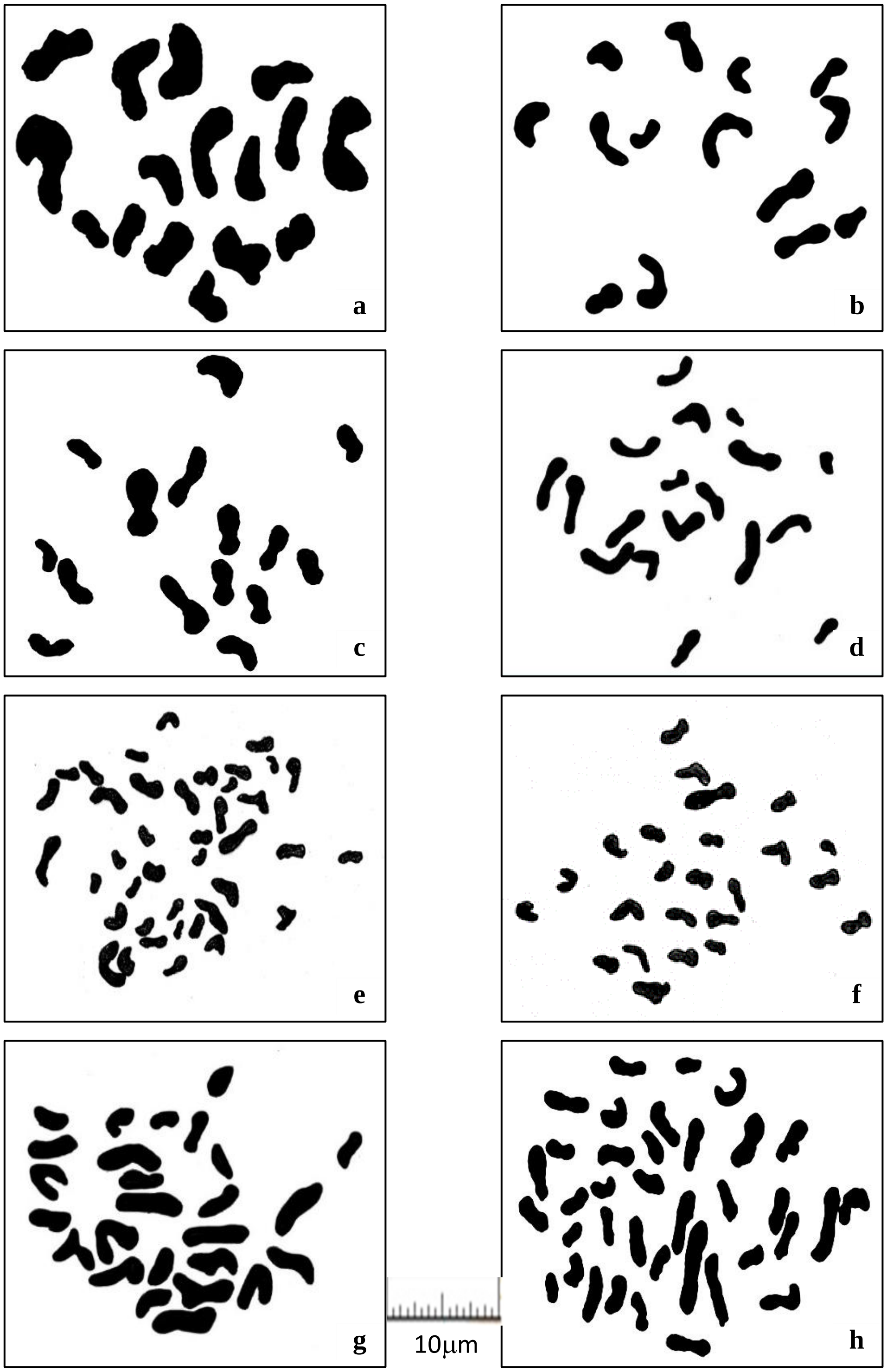
Subject	Data source	Source of variation	DF	Mean sum of square	F value
Root	Epidermis thickness	Habitat	23	1219255.908	3.439***
		Error	48	0.355	
	Cortex thickness	Habitat	23	0.593	70.328***
		Error	48	0.008	
	Sclerenchyma thickness	Habitat	23	0.005	1.229^{ns}
		Error	48	0.004	
	Endodermis thickness	Habitat	23	3677782.348	1.031***
		Error	48	0.357	
	Metaxylem area	Habitat	23	1359288.000	4.960***
		Error	48	0.274	
	Phloem area	Habitat	23	1003406.522	3.021***
		Error	48	0.332	
	Pith thickness	Habitat	23	1.140	91.943***
		Error	48	0.012	
	Pith cell area	Habitat	23	1487178.516	3.751***
		Error	48	0.396	

*** = significant at p≤ 0.001, ns = non-significant.

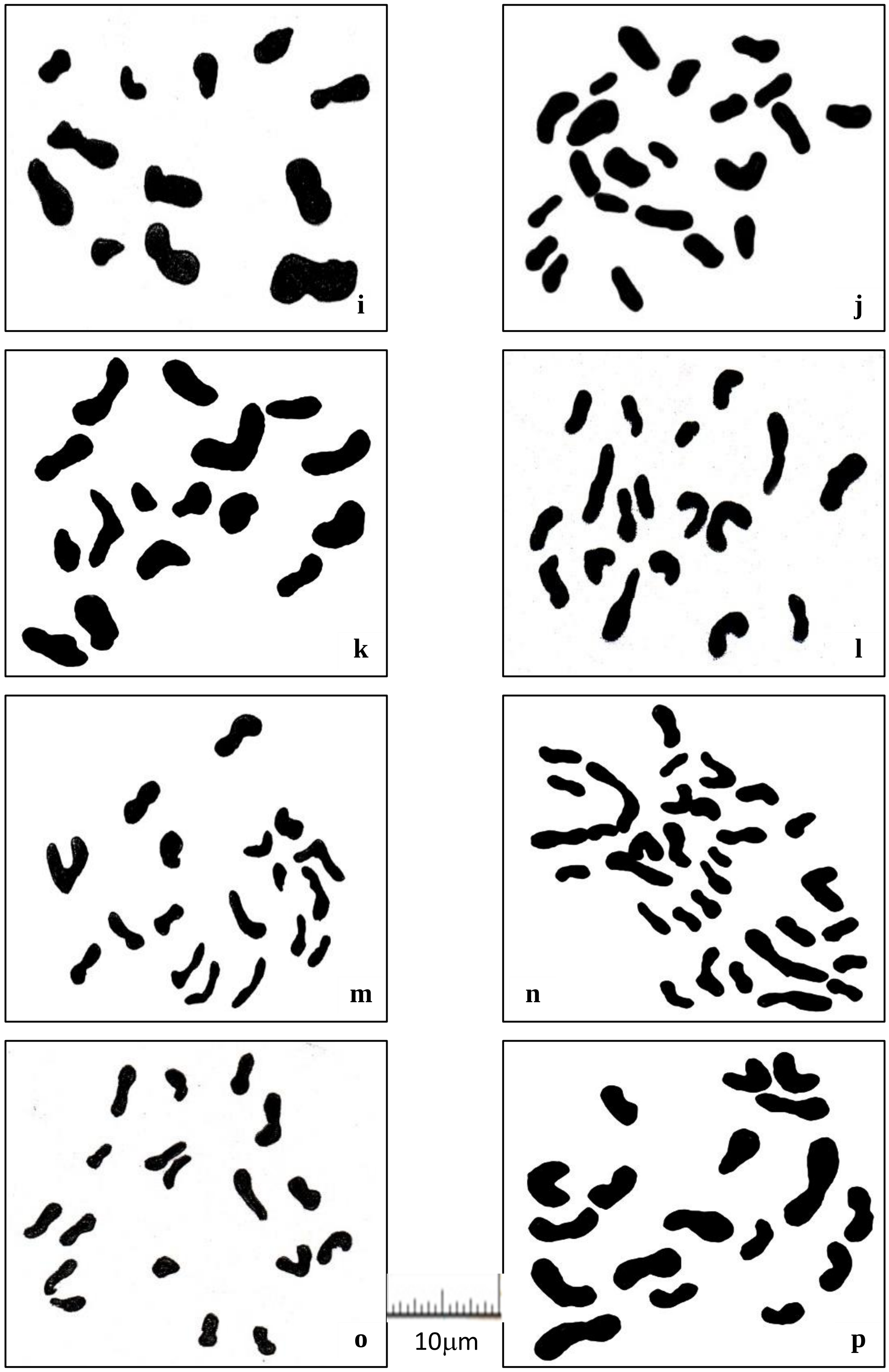
4.5 Cytotypes

All the 24 accessions of *Cynodon dactylon* in present study exhibited chromocentric nuclear organization. The chromocenters were seen distinctly on a very light background of the microscope. Few of the representative plates of interphase showing chromocentric structure are shown in Figs. 97(a-i). The number of chromocenters of each accession alongwith their chromosome numbers are given in Table 34. This table also reveals heterochromatin percentage per nuclear area, nuclear volume and interphase chromosome volume. Chromocenter numbers of 24 accessions were found to range from 11.4 (Pabna) to 17.6 (Sherpur, Faridpur, Khagrachari) and the values for each accession were not found to correspond with their respective somatic chromosome number. In this study, out of 24 accessions nine were diploid with 2n = 18 chromosomes. Only one collected from Naogaon was found to be tetraploid with 2n = 4x = 36 and rest of them were aneuploid showing the chromosome numbers 12, 14, 14, 14, 16, 16, 20, 22, 22, 22, 24, 26, 32, 40 subsequently. The number of chromocenters for each accession were not found to be corresponding with their somatic chromosome number. Sometimes it was found to be significantly less and definitely not more than their respective chromosome number.

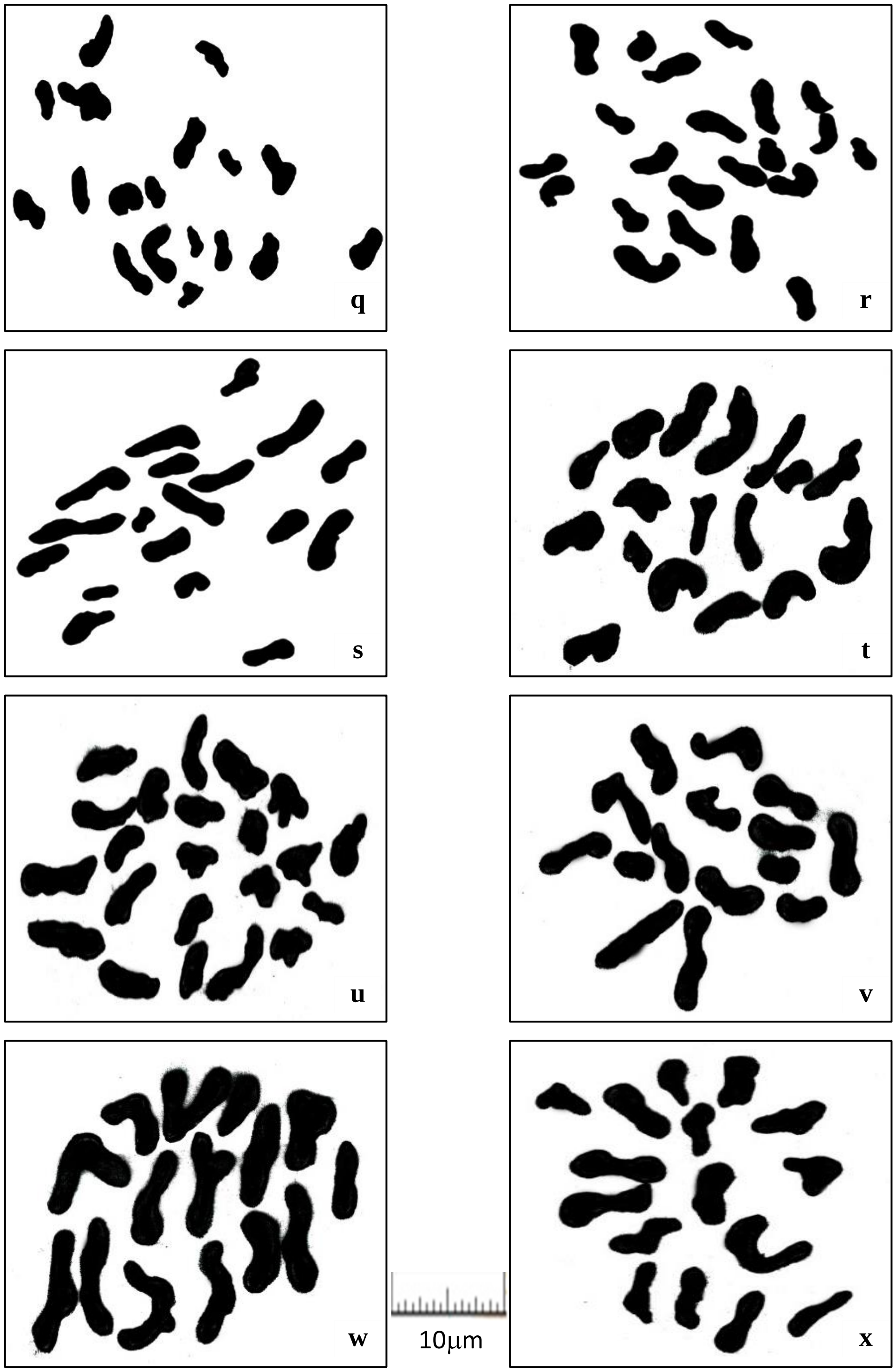
Percentage of heterochromatin values were recorded by determining the area of the interphase nucleus and of chromocenters by planimetry and then the heterochromatin values were expressed as percent per nuclear area. The heterochromatin values were found to range from 19.759% (Jhenaidah) to 66.022% (Shariatpur). The values for heterochromatin percentage per nuclear area obtained in this study were very much variable. Nuclear volume as well as interphase chromosome volume were determined. The values for nuclear volume were found to range from 0.674 μm^3 (Gaibandha) to 41.921 μm^3 (Gazipur). In case of this parameter the obtained values were not found to depend upon the number of chromosomes. Similarly, the values for interphase chromosome volume were found to range from 0.028 μm^3 (Gaibandha) to 2.507 μm^3 (Rangamati). Here also apparently no relation was observed between chromosome number and interphase chromosome volume. For finding the results regarding chromosome number in different accessions collected from 24 locations of Bangladesh, root tip cells were used. The chromosome count was performed from photo-micrographs but for clear visualization camera lucida drawings were made (Fig. 96). Among 24 accessions aneuploid with different chromosome numbers were preponderant.



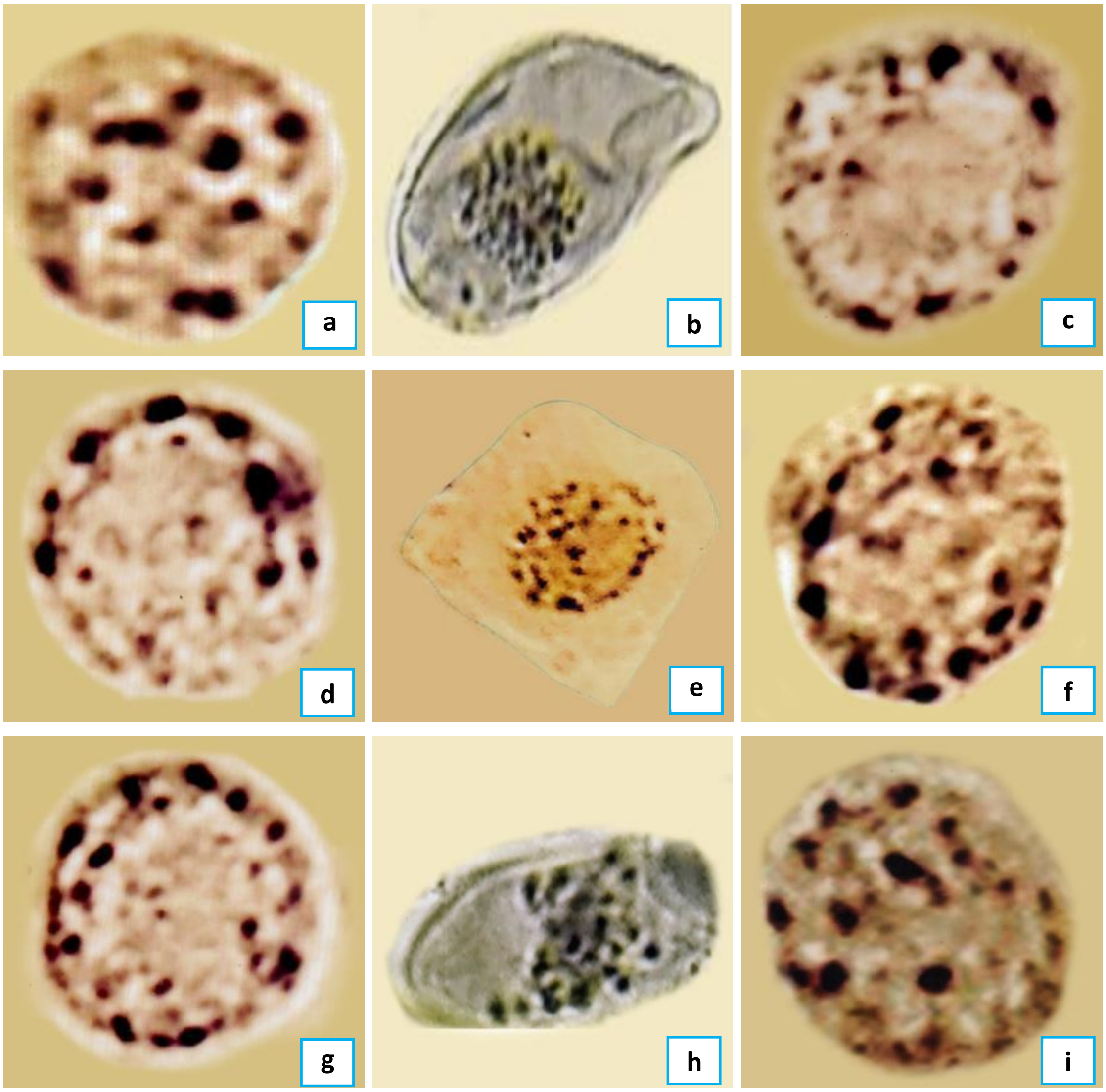
Figs. 96 (a-h): Camera-lucida drawings of respective chromosome numbers in *Cynodon dactylon* accessions collected from eight habitats*;* a) Rangpur (2n = 18), b) Lalmonirhat (aneuploidy, 14), c) Dinajpur (2n = 18), d) Thakurgaon (2n = 18), e) Panchagarh (aneuploidy, 40), f) Gaibandha (aneuploidy, 24), g) Rajshahi (aneuploidy, 26), and h) Naogaon (2n=4x=36).



Figs. 96 (i-p): Camera-lucida drawings of respective chromosome numbers in *Cynodon dactylon* accessions collected from eight habitats*;* i) Pabna (aneuploidy, 12), j) Gazipur (aneuploidy, 22), k) Narshingdi (aneuploidy, 16), l) Sherpur (2n = 18), m) Mymensingh (aneuploidy, 32), n) Khulna (aneuploidy, 32), o) Jessore (2n = 18), and p) Jhenaidah (2n = 18).



Figs. 96 (q-x): Camera-lucida drawings of respective chromosome numbers in *Cynodon dactylon* accessions collected from eight habitats*;* q) Faridpur (2n = 18), r) Shariatpur (aneuploidy, 22), s) Barguna (2n = 18), t) Khagrachari (aneuploidy, 22), u) Bandarban (2n = 18), v) Rangamati (aneuploidy, 14), w) Cox's Bazar (aneuploidy, 14), and x) Saint Martin’s Island (aneuploidy, 16).



Figs. 97 (a-i): Representative plates of interphase showing chromocentric structure of nucleus and chromocenter numbers in *Cynodon dactylon* accessions collected from different habitats.

Table 34: Number of chromosomes, nuclear volume, interphase chromosome volume, number of chromocenters and heterochromatin percentage per nuclear area in different accessions of *Cynodon dactylon* collected from different habitats of Bangladesh.

Sl. No.	Habitats	Number of chromosme	Nuclear volume (μm^3)	Interphase chromosome volume (μm^3)	Number of chromocentres	Heterochromatin percentage per nuclear area
			Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
1.	Rangpur	18 *	2.807±0.102m	0.156±0.006j	14±0.566fg	28.675±1.627ghi
2.	Lalmonirhat	14**	12.077±0.093fg	0.863±0.007e	14±0.632fg	51.037±5.982bcd
3.	Dinajpur	18 *	14.354±0.089d	0.797±0.005e	16.4±0.669abcde	25.548±1.272hi
4.	Thakurgaon	18*	2.706±0.117m	0.150±0.006j	15.2±1.073bcdefg	46.661±3.738bcde
5.	Panchagarh	40**	9.134±0.152i	0.228±0.004i	16.4±0.877abcde	57.955±6.537ab
6.	Gaibandha	24**	0.674±0.084o	0.028±0.003k	16.6±0.358abcde	29.461±3.547fghi
7.	Rajshahi	26**	13.996±0.154d	0.538±0.006g	17±0.283abc	29.455±2.044fghi
8.	Naogaon	36***	3.578±0.066l	0.099±0.002jk	16.8±0.438abcd	24.389±1.124i
9.	Pabna	12**	4.620±0.088k	0.385±0.007h	11.4±.219h	45.185±7.983bcde
10.	Gazipur	22**	41.921±0.180a	1.905±0.008b	17.6±0.358a	39.793±2.242defg
11.	Narsingdi	16**	17.238±0.295c	1.077±0.018d	15.2±.715bcdefg	33.82±6.588efghi
12.	Sherpur	18*	5.209±0.093j	0.289±0.005i	17.6±0.358a	38.706±5.132defgh
13.	Mymensingh	20**	0.862±0.084o	0.043±0.004k	13±0.707gh	41.782±1.033cdefg
14.	Khulna	32**	5.057±0.09j	0.158±0.003j	16±0.80abcdef	30.048±5.094fghi
15.	Jessore	18*	5.058±0.171j	0.281±0.004i	16.4±1.043abcde	47.653±1.411bcde
16.	Jhenaidah	18*	11.811±0.150h	0.656±0.008f	17.40±.358ab	19.759±2.714i
17.	Faridpur	18*	1.348±0.105n	0.075±0.006k	17.6±0.358a	30.304±5.303fghi
18.	Shariatpur	22**	13.183±0.089e	0.599±0.004fg	14.4±1.431efg	66.022±5.481a
19.	Barguna	18*	29.886±0.097b	1.660±0.005c	14.4±1.189efg	65.286±7.938a
20.	Khagrachari	18*	11.549±0.081h	0.642±0.041f	17.6± 0.219a	20.555± 2.274i
21.	Bandarban	22**	12.348±0.091f	0.561±0.045g	17.4± 0.237ab	54.404± 1.568abc
22.	Rangamati	14**	30.083±0.114b	2.507±0.057a	14.6± 0.438defg	55.336± 1.788abc
23.	Cox's Bazar	14**	14.234±0.116d	1.017±0.058d	14.4± 0.219efg	43.316± 1.540cdef
24.	St. Martin's Island	16**	4.468±0.087k	0.291±0.044i	15.0± 0.400cdefg	38.37± 1.280defgh

* = Diploid chromosome number, ** = Aneuploid, *** = Tetraploid

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

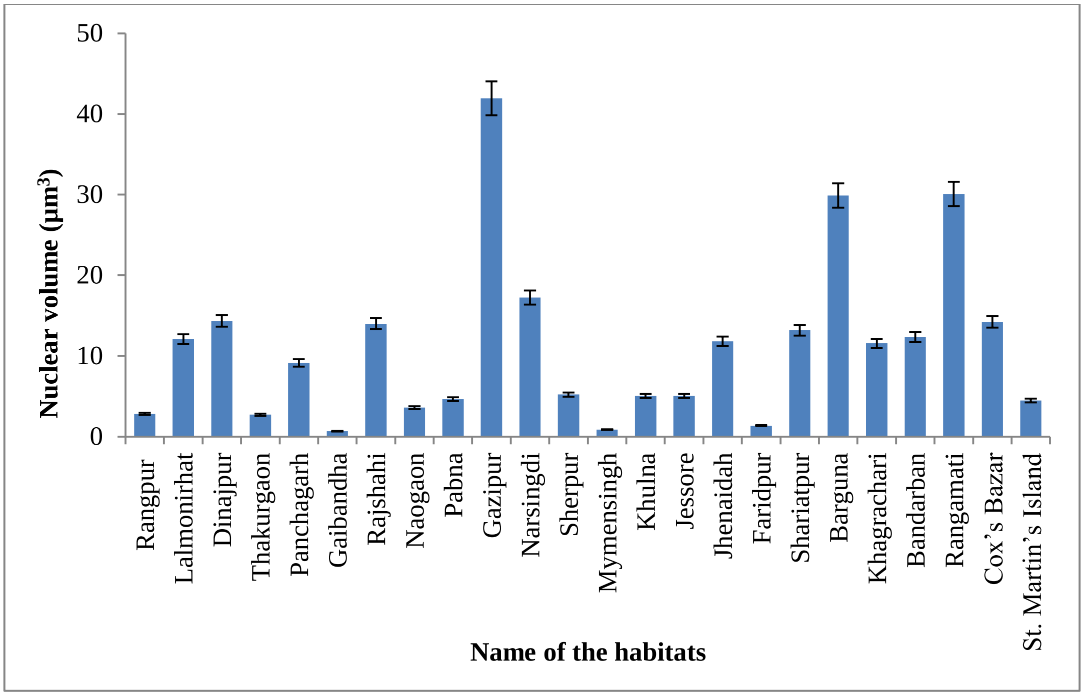


Fig. 98: Nuclear volume at interphase of *C. dactylon* accessions collected from different habitats.

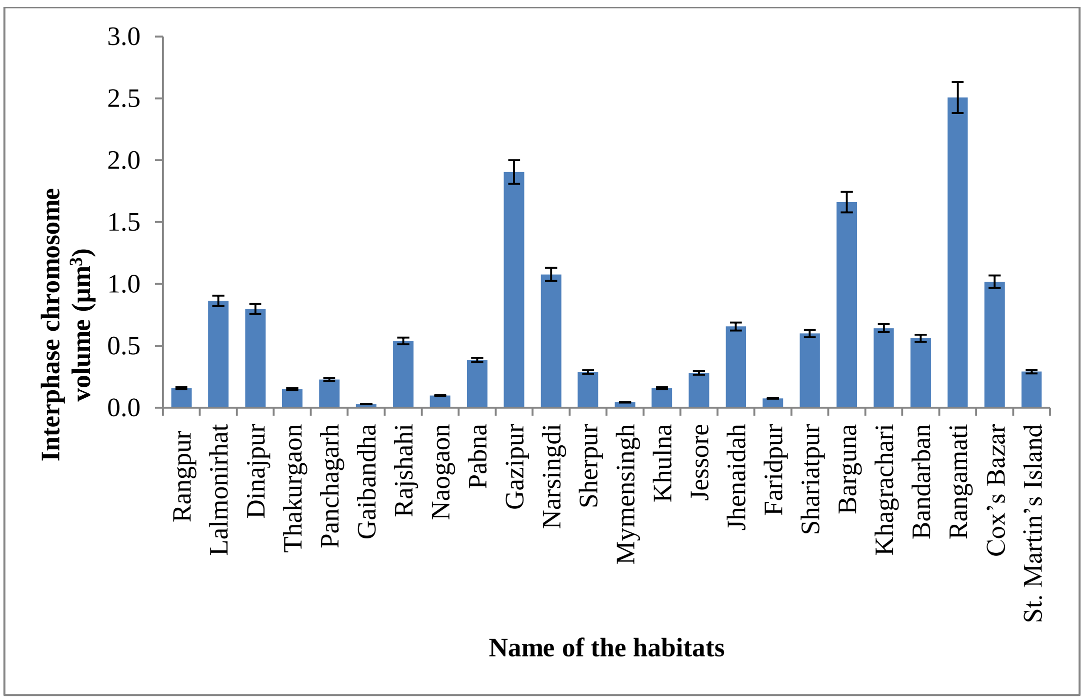


Fig. 99: Interphase chromosome volume in root meristem of *C. dactylon* accessions collected from different habitats.

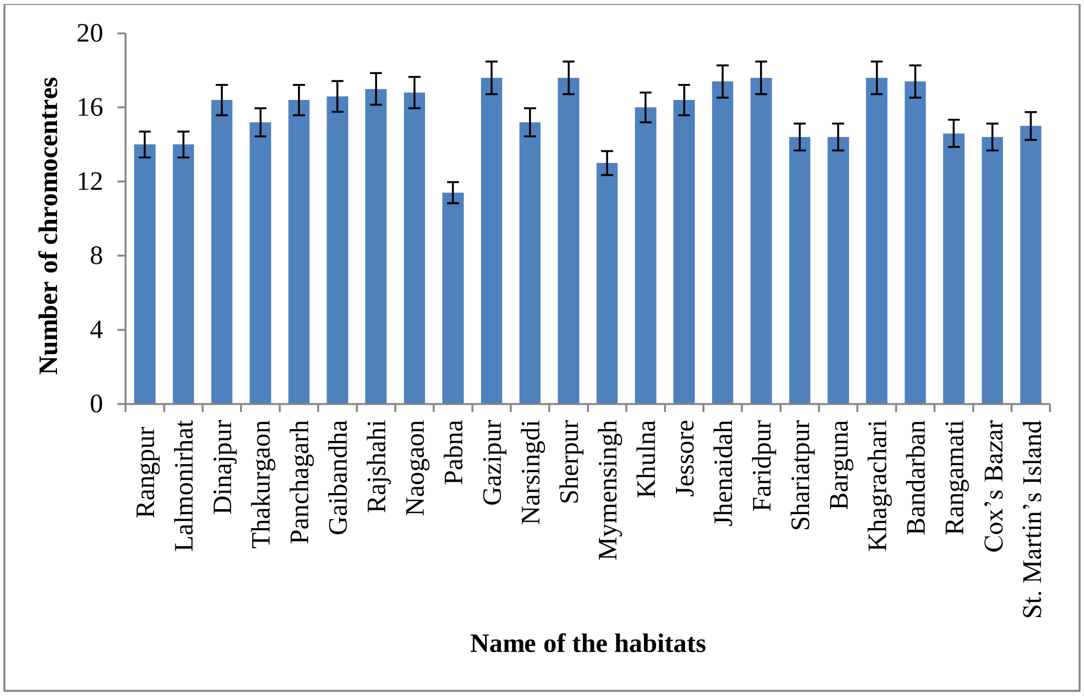


Fig. 100: Number of chromocentres in interphase nucleus of *C. dactylon* accessions collected from different habitats.

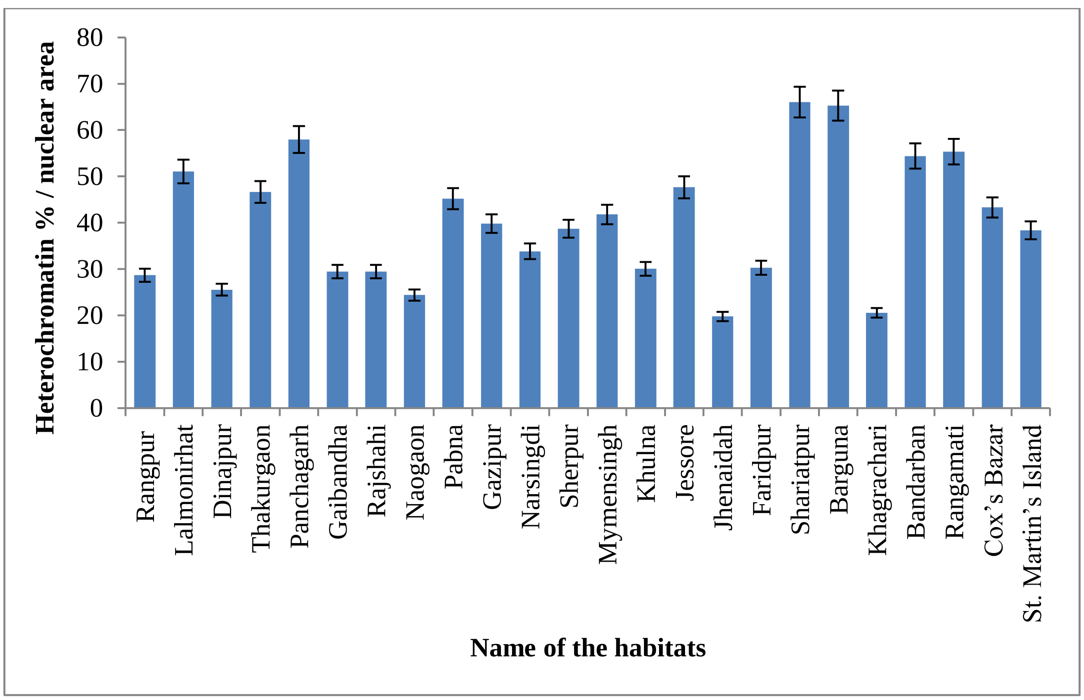


Fig. 101: Heterochromatin percentage per nuclear area at interphase of *C. dactylon* accessions collected from different habitats.

The ANOVA (Table 35) reveals that parameters of interphase nuclear structure were highly significant from each other.

Table 35: Analysis of variance subjected to variation of different parameters of interphase nuclear structure of *Cynodon dactylon* on the basis of 24 habitats of Bangladesh.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Interphase nuclear structure	Nuclear volume	Habitat	23	318.143	6.709 ***
		Error	48	0.047	
	Interphase chromosome volume	Habitat	23	1.201	733.827 ***
		Error	48	0.002	
	Number of chromocentries	Habitat	23	8.389	6.241 ***
		Error	48	1.344	
	Heterochromatin percentage / nuclear area	Habitat	23	544.354	10.231 ***
		Error	48	53.205	

*** = significant at $p \leq 0.001$.

Since there is a general trend in plants for stomatal density to decrease and stomatal size to increase as ploidy level increases or decreases, one tetraploid and 14 aneuploid obtained in the present study were taken into consideration to find out their effect on stomatal characters such as stomatal frequency and stomatal index on abaxial and adaxial surface of the leaves of *C. dactylon* (Table 36). But in this study no such effect was found apparently, when stomatal frequency and stomatal index were predicted both in case of euploid (tetraploid) and aneuploid.

Table 36: Effect of euploid and aneuploid accessions of *Cynadon dactylon* on stomatal frequency and stomatal index on abaxial and adaxial surfaces of leaves in different habitats.

Sl. No.	Habitats	Number of chromosomes	Abaxial surface of leaves (+)		Adaxial surface of leaves (++)	
			Stomatal frequency	Stomatal index	Stomatal frequency	Stomatal index
			Mean±SE	Mean±SE	Mean±SE	Mean±SE
1.	Lalmonirhat	14**	4.59±0.08efgh	26.34±0.62ab	4.59±0.42defg	26.67±0.36b
2.	Panchagarh	40**	4.26±0.15hij	28.00±0.91ab	5.44±0.22abcde	26.81±0.64b
3.	Gaibandha	24**	4.29±0.13hij	25.40±0.82ab	6.31±0.56ab	25.73±0.56bc
4.	Rajshahi	26**	4.03±0.08jk	25.61±0.88ab	5.29±0.38bcde	25.93±0.64bc
5.	Naogaon	36***	4.93±0.13cde	26.40±0.61ab	5.76±0.28abcd	25.60±0.46bc
6.	Pabna	12**	4.82±0.15cdef	25.78±0.52ab	4.61±0.38cdefg	26.07±0.28bc
7.	Gazipur	22**	5.56±0.10a	25.80±0.61ab	3.85±0.24fghi	25.80±0.61bc
8.	Narshingdi	16**	4.19±0.08ij	26.25±0.30ab	5.92±0.22abc	25.87±0.33bc
9.	Mymensingh	20**	4.77±0.13cdef	27.42±1.40ab	4.86±0.26cdefg	25.78±0.43bc
10.	Khulna	32**	5.40±0.10ab	27.78±1.31ab	5.23±0.20bcde	28.76±0.43a
11.	Shariatpur	22**	4.69±0.13cdefg	27.36±1.78ab	6.61±0.38a	25.40±0.31bc
12	Bandarban	22**	5.07±0.08bc	26.13±0.71ab	5.58±0.32abcde	26.27±0.61bc
13.	Rangamati	14**	4.52±0.10fghi	26.17±0.56ab	5.57±0.60abcde	26.37±0.41bc
14.	Cox's Bazar	14**	2.13±0.13l	26.31±0.30ab	3.34±0.40hi	26.26±0.33bc
15.	St. Martin's Island	16**	1.78±0.15m	26.59±0.78ab	3.70±0.32ghi	27.04±0.41b

** = Aneuploid, *** = Tetraploid

+, ++ = Data used from the Tables 22 & 23 respectively.

In a column, means followed by the same letter(s) are not significantly different at the 5% level by DMRT.

Chapter V

Discussion

5. DISCUSSION

5.1 Distribution

Cynodon dactylon, commonly known as 'bermudagrass' and as 'durba' in Bangladesh probably originated in tropical Africa, and the Bengal region of India, and Bangladesh have been considered as its home. It is also believed that this grass species started in India and most of the improved strains of *C. dactylon* were developed from African stock (Kingsbury 1964). Everist (1979) stated that *C. dactylon* seems to be native in Australia, though some evidence suggests that this grass might have been introduced. So, it can be said that durba grass is an indigenous herb and growing in different bio-ecological zones of Bangladesh. The present findings on distribution of this herbaceous grass species indicates its cosmopolitan behaviour throughout the country. This grass was found to grow everywhere of the studied area. Bermudagrass may grow on any well-drained soil and that may be either acid or alkaline, provided moisture and plant food nutrients are adequate (Philips Petroleum Company 1958). In present study, accessions of *C. dactylon* were found to grow nicely on alkaline soil. Bermudagrass is tolerant to alkali and it was recorded that patches of bermudagrass in submerged condition in the Salton Sea for over two years, were still alive and made new growth from the stems when that body of water finally evaporated to a lower level (Robins *et al.* 1970). Besides they stated that bermudagrass is introduced in many ways into new areas when the seeds are common impurity in commercial seeds and plants bearing seeds are carried in hay, in packing, in bedding for livestock and in feedstuffs, the seeds are carried by wind and irrigation water. In Bangladesh spreading of this grass species all over the country might be mainly due to wind, irrigation water and flood. Thus, it has become cosmopolitant at each and every part of the country. This grass species is common also in grasslands, lawns and pastures. FAO (2012) reported similar results. It grows dominantly in uncultivated areas: roadsides, sea coast sandy dunes, or along rivers and irrigated land (Ecoport 2012). It grows well on overgrazed and trampled areas (Cook *et al.* 2005). It grows in area where an average annual temperature range within 6-28°C and it does better where daily temperatures are in the range of 17-35°C. In Bangladesh this sort of temperature is usual and helps the durba grass very enormously. Though bermudagrass prefers deep and well-drained fertile soils, it can adapt to a broad range of soils including those that are relatively infertile, with a pH ranging from 4.3 to 8.4 (optimum >5.5). This type of soil pH fully supports the present findings, where soil pH ranged from 4.9 to 8.5 in the studied areas. Three accessions of *C. dactylon* were found to be saline tolerant in this study collected from Barguna, Cox's Bazar and Saint Martin's Island. This grass species has some saline soil tolerance, hence its ability to be grown on coastal areas (Cook *et al.* 2005, Ecoport 2012, FAO 2012).

Three accessions were found to be saline tolerant and were transplanted them in experimentation field of Rajshahi, University. But same sort growth habit was observed in both original and transplanted area, although Wu *et al.* (2009) suggested that genetic variation and diversity of common bermudagrass may be associated with their geographic origins, and stress response of this grass species is inheritability one to another progeny (Schwartz *et al.* 2009). Roots of salt tolerance bermudagrass are reported to have efficient osmotic system to avoid negative osmosis in saline soil (Hameed and Ashraf 2008). Water and soil salinity are normal hazards in many parts of the coastal area in Bangladesh, affecting different uses of water including irrigation, drinking, household, fisheries and functioning of the ecosystem (Khanom and Salehin 2012). In reference to this statement Mahtab and Zahid (2018) conducted a study on coastal surface water suitability analysis for irrigation in Bangladesh. Their analysis showed that coastal surface water in Bangladesh is overall suitable for irrigation during wet period, while it needs treatment for using in case of irrigation during dry period. But in case of durba grass in the present study particularly the accessions collected from Barguna and Saint Martin's Island showed their growth high and medium, respectively. Most striking behaviour of the accession collected from Cox's Bazar was found about its soil pH, which was highly acidic and its growth was low. It might be due to distantly located hilly area from sea shore and its low growth because of high acidic soil.

Cynodon dactylon is found abundantly as weed in crop fields and can readily take position of any uncultivated area. During winter, this grass species remains dormant and turns brown in colour. Growth is found to be promoted by full sun shine and retarded by full shade. Plants are readily propagated by cuttings and rooting. It can spread very quickly from the rooted runners, which grow more than 7.5 cm / day (Nagori and Solanki 2011). Planting is best done in wet weather to ensure quick sprouting. It gives a complete ground cover in 4-8 weeks when sprigged 30-45 cm apart (Huxley 1992). In present study, plant heights and stand density were found to vary for all accessions in their habitat as well as in their experimentation field. Mature leaf blades of different bermudagrass accessions from different locations were also varied. These variations in shoot height, stand density, leaf width etc. were observed with naked eye in their habitat and also in their experimentation field. The variations in growth habits of bermudagrass observed in this study may be due to the variations in edaphic, topographic and climatic factors of the regions and even the locations from where the accessions of bermudagrass were collected. Distribution of durba/ bermudagrass seeds over great distances increase the possibility of rapid development of ecotypes that are better adapted to the new environment (Dickens 1974). The differences that occur in growth and development of durba grass collected from 24 locations of Bangladesh and grown under same environmental conditions in experimentation field, give indication that the eco-, bio-/cytotypes represent various stages of evolutionary differentiation more than just local population which differ genetically (Al-Juboory and Hassawy 1980). Glausen (1967) stated that the differences in the species, such as growth habit, date of flowering, dormancy etc. are controlled by certain genes, each with a certain affect, and when a group of genes share in their effect at the same time and in the same direction, they result in a new characteristic.

The ability of a plant to survive an unfavourable external water deficit is termed drought resistance (Beard 1973). Green cover defined as the percentage of green leaves in a turf plot and leaf relative water content during the drought period have been used as a criteria to selected drought resistant genotype in turfgrass research (Huang *et al.* 1997b, Richardson *et al.* 2008). Based on these traits, there is a considerable genetic variation of drought resistance among genotypes within certain turfgrass species. Significant variability exists among genotypes within couch grasses (*Cynodon dactylon* spp.) as demonstrated by Zhou *et al.* (2009). In the present study four accessions of *C. dactylon* were found to be drought tolerance / resistant and it was determined based on annual temperature, rainfall, humidity, soil moisture etc. shown in previous chapter. All these four accessions were found from North-West zone of Bangladesh and their growth were found to be high and medium. It is noticeable that in all cases soil was found to be somewhat alkaline and pH ranged from 7.1 to 8.1, and their growth were found to be round the year. For *C. dactylon*, morphological traits such as internode length and branching habit may influence evapo-transpiration through their effects on leaf density and leaf area (Ebdon and Petrovic 1998). Low evapotranspiration is an important mechanism responsible for drought resistance (Huang 2008). The present findings, however, suggest that there is enormous scope to select highly drought resistant genotypes with good turf characteristics among the existing accessions in Bangladesh. Now-a-days *C. dactylon* is widely used as warm season turf and forage species in temperate and tropical region (Li and Qu 2004) and the population of this grass species harbour many useful physiological traits such as tolerance to high temperature and drought (Bethel 2005). Regarding distribution of *C. dactylon* from ecological point of view, it may be said that this grass species tolerates a wide range of soil type and conditions, growth is greater on heavy clay soil than on light sandy soils of dry regions. It grows on soils with a wide range of pH and alkaline soils are tolerated more than acidic alongwith high temperature and moderate soil moisture for proper growth.

5.2 Morphological characteristics

Grasslands are one of the Earth's major biomass and the native vegetation of up to 48% of Earth's terrestrial surface. Grasslands occur on every continent except Antartica, are ecologically and economically important, and provide critical ecosystem goods and services at local, regional, and global scales (Blair *et al.* 2014). With a variety of lifeforms (annual, biennial and perennial species) grasslands become species rich ecosystem. The grasses are found to be adapted to many sorts of the climatic forces and their morphology and developmental patterns make them well suited to different environment. Their protective meristematic tissue belowground helps grasses to survive through regrowing when above ground tissues are destructed by different environmental causes. Their aboveground part called shoots grow from the basal part of the plant may be vegetative or reproductive, and consists of node and internode with an axillary bud, cylindrical sheath, and leaf blade.

The present study was conducted in an experimentation field of the Institute of Biological Sciences, University of Rajshahi, Bangladesh to evaluate the morphological (both botanic and agronomic) behaviour of 24 collected native populations of *Cynodon dactylon* from different habitat of Bangladesh. Somewhat similar type of works on *C. dactylon* was carried out by Viggiani *et al.* (2015) in Italy. During the first step of research, they identified 11 sites from six regions of Southern and Central Italy and from these sites they collected 24 ecotypes for determining their habitus and phenology plus some biometrical parameters. Plants are greatly restricted in growth field under stress conditions, and have evolved many mechanisms for rapidly adapt to drought stress condition to keep growth and productivity (Zhu 2002). The experimentation field of the present study belongs to semi-drought condition and the soil is highly alkaline, while the organic matter, potassium, total nitrogen and phosphorus are very low. In response to these conditions *C. dactylon* accessions collected from different regions of Bangladesh were found to be adapted very soon and nicely. This might be due to such mechanism as physiological, biochemical, molecular and cellular changes to cope with limited water supply (Xu and Huang 2010, Luo *et al.* 2011). Infact after transplanting the different accessions of this grass species each block was irrigated in limited way. Comparatively *C. dactylon* is one of the most drought tolerant turfgrasses (Kim *et al.* 2009, Zhao *et al.* 2011). Some studies have suggested that drought tolerance of *C. dactylon* might be correlated with plant development such as leaf firing, root and shoot systems, and mass production (Carrow 1996, Huang *et al.* 1997a & b, Qian *et al.* 1997), accumulation of dehydrin (Hu *et al.* 2010a, Cambell and Close 1997), evapotranspiration (Carrow 1995), leaf water content, chlorophyll content, proline content and antioxidant enzyme activities (Lu *et al.* 2007 & 2009, Hu *et al.* 2010b).

Gobilik *et al.* (2013) studied five ecotypes of *C. dactylon* and in his general assessment, the Beaufort ecotype was found with shorter shoot length and the Sipitang ecotype had a higher shoot number. In addition, the Beaufort ecotype had higher shoot dry weight in salt treatment and on the contrary, Sipitang ecotype had higher shoot fresh weight. Salt causes roots to experience negative osmosis, distorts root nutrients uptake and impedes plant growth (Hajibagheri 1989). The soil of the experimentation field was highly alkaline and that might be the cause of nonsignificant variation of root length, root fresh weight and root dry weight. Gobilik *et al.* (2013) found also all the five ecotypes with similar root fresh weight. Another important character of root i.e., number of rhizomes showed significant variation among the accessions. Dong and de Kroon (1994) stated that *C. dactylon* does not form rhizomes under shaded conditions. The present experimentation field was very much ambient and under high intensity of sun light. Even then this character showed wide variation. However, Van de Wouw *et al.* (2009) stated that the presence of rhizomes in *C. dactylon* is the only identifying character compared to that of other *Cynodon* species.

This also might be due to the salt accumulation in mature leaves which damages leaf tissues and decreases leaf photosynthetic activity (Gobilik *et al.* 2013). Hussain *et al.* (2012) stated that negative osmosis causes mesophyll turgidity loss and partial stomata opening reducing total leaf CO_2 assimination per day. In saline soil, plants may die when new leaf formation can not match leaf death or produce sufficient energy for plants (Munns and Termaat 1986). However, it is not expected in case of *C. dactylon* always, because this grass species can increase its root mass when salt concentration rises until it reaches a peak before starts to decline (Dudek *et al.* 1983). Probably same sorts of mechanism worked for maximum plant height, which was found with significant variation among all the plant accessions. Van de Wouw *et al.* (2009) found considerable diversity of plant height in a large group of accessions of *C. dactylon* and *C. nlemfuensis*. The horizontaly growing aboveground part of *C. dactylon* stems, called stolons were found to show nonsignificant variations in the present study. Van de Wouw *et al.* (2009) found 98 accessions of *Cynodon* belonging four different species with stolons, with considerable variation in number and length, and they stated that this might be due to very plastic response of stolons to light intensity and nutrient availability. Their findings support the present findings in case of all the 24 accessions of *C. dactylon* .

In *C. dactylon* , flowering stems were upright and beared a terminal group of 3-7 spike like branches, originating in a single whorl on the tip of the stem. The colour of the spikelet were found to show somewhat variation of the main colour brown. In the same accession the spikelets were not purely brown, rather it was found with light green or green. Nasiri *et al.* (2012) described inflorescence of this grass species to be raceme like panicle and spikes to be glabrous, elongated and green to purple green, or green with purple spots and yellowish at maturity. However, the ANOVA made in the present study showed no significant variation in case of inflorescence number and length of racemes. But the seed characters like seeds number per shoot and seeds number per plant were found with statistical variation. There are two types of *C. dactylon* and they are namely common and giant. Both these types are seeded varieties, they correspond to a wide range of genotypes and they are selected for their adaptability to different cultivation conditions (Ball and Pinkerton 2002). In some cases, common type has become synonymous with any seed propagated Bermuda grass (Busey 1989). Seeded varieties are outstanding tolerant to drought and heavy grazing. All the 24 accessions of *C. dactylon* in the present study were found to be seeded and since they are showing significant variation in respect of their number per shoot and per plant, their seeds may be commonly mixed for making commercial blend. Juska and Hanson (1964) stated that Bermuda grass (*C. dactylon*) vary with respect to this characteristics of seed heads, when they appear and how long they persist. Some accessions have seed heads from early in the growing season throughout much of the summer, whereas with others seed heads may persist for a relatively short time. They also mentioned that genetic factors and day length affect seed head formation. Seed characters found in 24 accessions of the present study might be due to these two regions in relation to their different habitats McWhorter (1971) found extreme variability between ecotypes of Jhonson grass (*Sorghum halepense* (L.) Pers.) in seed production, shattering characteristics and in size and appearance of panicles. Seed of all ecotypes occurred in all spikelets clustered on short racemose branchlets. This type of mechanism may also be applicable in case of bermudagrass.

However, Al-Juboory and Hassaway (1980) stated that the differences that occur in the morphological development of Cogongrass (*Imperata cylindrica*) collected throughout Iraq and grown under the same condition, give another indication that the ecotypes represent various stages of evolutionary differentiation more than just local population. They further said that this may be due to the process of sorting and controlling environmental factors (habitat factors) on species population which differ genetically. Their statement fully supports the finding in case of morphological variation of 24 accessions of *C. dactylon* in the present study.

5.3 Physico-chemical properties of the soil of different habitats

In the present study, an experiment was conducted on 24 accessions of *Cynodon dactylon* in the experimentation field of Rajshahi university, Bangladesh. These 24 accessions along with the soil of their habitats were collected from different habitat of Bangladesh. The physicochemical properties of the soil of experimentation field as well as that of the 24 different habitats were determined. Adaptibility of plants to different environment by acquisition of physico-chemical and climatic conditions is an important subject matter. The plants change their growth habit and become more flexible in that unfavourable habitat. This is not applicable for all the plant species. There are more grass species among which *Cynodon dactylon* is enormously adaptable and has become cosmopolitan particularly in temperate, tropical and sub-tropical regions of the world. Tropical Africa is considered to be the center of origin of this grass species. Its growth is reduced with the onset of lower temperature and shows best growth with temperature. This grass species grows on a wide range of soils, from heavy clays to deep sands provided soil fertility is not a limiting factor (Juska and Hanson 1964). It grows well on both acidic and calcareous soils and very much tolerant to saline conditions. This grass species also can tolerates floodings but can not thrive on water logged soil. It is also drought resistant but can not be grown in arid regions without supplementary water. Their growth under shade is not satisfactory and thus, its growth reaction to such condition is poor.

However, the soil of the present experimentation field was found to be alkaline and the organic matter was low. Total nitrogen, potassium and phosphorus were also low. Other nutrients were somewhat favourable. The soil texture of the experimentation field was found to be clay loam under agroecological zone 26. In such a condition 24 accessions from different habitats were planted to find out their morphological variations alongwith adaptation. Almost under different conditions Ramakishnan and Gupta (1973) conducted an experiment with differential response of nitrogen, phosphorus and potassium. In conclusion they have said that besides calcium which was shown to be partially responsible for the restriction of three ecotypes of *C. dactylon* to their respective habitats, the availability of nitrogen, potassium and also phosphorus may play an important role. But striking by all the accessions of *C. dactylon* in the present study showed no such poor performance comparatively among them and they were found to be well adapted.

In most of the soils of different habitats in the present study organic matters and nitrogen were very low or low. At the same time Calcium and Magnesium were very high and high, respectively in the experimentation field. Calcium was found to be available in soils of different habitats. Only magnesium was low in the soil of Narshingdi. It is evident now that the macronutrients such as N, P, K^+ and Ca^{2+} are essentially required for regulation of enzyme activities, protein synthesis, integrity of cell wall and plasma membrane, and also as components of proteins, photosynthetic protein complexes, photosynthetic pigments, RNA and DNA (Taiz and Zeiger 2002, Akram *et al.* 2008). Potassium in the form of cation (K+) plays a vital role in regulation of osmotic adjustment by lowering osmotic potential of cells (Akram *et al.* 2008). It has been reported by Premachandra *et al.* (1990) that water stress generally increase the K^+ concentration especially under low phosphorus levels. Similar situation might be occurred in the present study where water supply was very much limited and phosphorus levels were low or medium in 12 soils of the habitat. In the present study, phosphorus concentrations were high in most of the soils of different habitats and that might be the cause of reduction in shoot and root characters. Very high and optimum level of Ca caused promising shoot character but low level of nitrogen was not favourable for shoot characters, particularly in case of minimum water supply in all the accessions of *C. dactylon*. In of few other micronutrients, sulfur was found to be low, while availability of Zinc was somewhat better in soils of different habitats in the present study. On the contrary, the amount of copper, iron and manganese were sufficient almost in all the habitats. It is now evident that the most reliable micronutrients for healthy plants are zinc, boron, copper and manganese. These are used by plants in small quantities. In spite of the low requirement, critical plant functions are limited if micronutrients are unavailable resulting many plant abnormalities.

However, sandy soils with low organic matter may contain deficient micronutrients. High pH soils may make some nutrients low. Cations of copper, iron, manganese and zinc may help to the soil properties, solubility becomes high under acid condition and becomes deficient on calcareous soils or becomes high in organic matter where strong chelation decreases availability (Meugel 1990, Bennett 1993, Stevans *et al.* 2002, Kelling 2005). All the 24 accessions of *C. dactylon* collected from different habitats and transplanted in an alkaline soil of the experimentation field were found to be adapted nicely in spite of their different physico-chemical and environmental conditions. That might due to many physiological and biochemical mechanisms to any environmental conditions keeping their growth and productivity more or less properly.

According to Wu *et al.* (2009) several studies reported nutrient data for warm season turfgrass including bermudagrass (*C. dactylon*) in many transition zones in the world. Mineral composition of bermudagrass was studied by McCrimmon (2001), Snyder and Cisar (2000) and Walworth and Kopec (2004) reported different results depending on the cultivar and different nutrient sources and rates. Volterrani *et al.* (2012) stated that use of plug plants shows several advantages, since the use of fully developed root systems and actively growing shoots enhances their colonization potential. These features also might be the appropriate reason for proper growth and adaptation of different accession of 24 different habitats in this experiment.

5.4 Qualitative leaf epidermal characters

The findings regarding qualitative leaf epidermal characters are discussed here. The leaves were found to be amphistomatic. The stomata were paracytic type, dumbbell shaped with two subsidiary cells placed parallel to the pore. Two guard cells were found with two subsidiary cells lateral to the guard cells. Subsidiary cells in *Cynodon dactylon* were found to be dome shaped at both abaxial and adaxial surface of the leaves. Hepworth *et al.* (2018) reported, amphistomatic leaves with dumbbell-like and aligned stomata as usual in grass species. Like almost all grasses stomata in *Cynodon dactylon* are paracytic type earlier proved by Abid *et al.* (2007). Due to disposition of the subsidiary cells, the stomata were markedly paracytic, which is also typical character of Poaceae family (Rudall *et al.* 2017). Stomatal shape was more responsive to salt stress in the salt range population where elliptic stomatal complex transform to rhomboid and smaller ones under high salt stress (Hameed *et al.* 2014), supports the present findings in case of the accessions collected from three coastal areas (Barguna, Cox’s Bazar and Saint Martin’s Island) of Bangladesh. The epidermal cells were elongated and arranged in vertical rows parallel to the long axis of the leaf. All epidermal cells in *Cynodon dactylon* were found to be sinuous. In present study, almost all epidermal cells in *Cynodon dactylon* were found to be sinuous or wavy similar to the findings of Ahmed *et al.* (2010). *Cynodon dactylon* accessions had epidermal cells with sinuous cell walls, a common feature among species belonging to the Poaceae family (Khan *et al.* 2017) and which is related to the increase of the surface for higher light uptake (De Castro *et al.* 2009).

Length and width of long cells are significant parameters which help in identification and classification of grasses (Elahi and Ashraf 2002). The long cell margins were found to show sinuous almost in all the accessions. But, in case of the accessions of Barguna, Cox’s Bazar and Saint Martin’s Island, the long cell margins were found to be slightly sinuous. Silica bodies were saddle shaped and found in the materials collected from Rangpur, Lalmonirhat, Dinajpur, Thakurgaon, Panchagarh, Gaibandha, Rajshahi, Naogaon, Gazipur and Jhenaidah. Cross shaped silica bodies were found in materials collected from Pabna, Sherpur, Mymensingh, Khulna, Shariatpur, Khagrachari, Bandarban and Rangamati. Both saddle and cross shaped silica bodies were found in case of Narsingdi, Jessore and Faridpur. Horizontally elongated silica bodies were found in accessions of Barguna, Cox's Bazar and Saint Martin's Island. Silica bodies are a type of phytolith in specialized epidermal cells of grass leaves. Various workers have considered silica bodies to be diagnostic feature for the family Poaceae (Twiss *et al.* 1969, Brown 1984, Mulholland 1989). Piperno and Peasall (1998) studied the silica bodies of Tropical American grasses and discussed their taxonomic implications. Thomasson (1986) noted that micro-morphological characters of the leaf provided information on the fossils phylogeny and taxonomic relationships. According to Metclafe (1960), Chaudhary *et al.* (2001b), and Ahmed (2009) silica bodies in *Cynodon dactylon* are saddle shaped, which were again confirmed by the present research.

Prickles angular were pointed at the tip and they were present in almost all the accessions except those from Barguna, Cox's Bazar and Saint Martin's Island. Hooks were also present in almost all the accessions except that of Barguna, Cox's Bazar and Saint Martin's Island. Macro-hairs were present in all accessions. No micro-hair was found in adaxially or abaxially in this present study. Chaudhary *et al.* (2001b) found that in *Cynodon dactylon* stomata were with triangular subsidiary cells, silica bodies were saddle shaped, and micro-hairs with hemispherical distal cells were present, while macro-hairs were absent. Freire *et al.* (2005) observed the presence of micro hairs in *Cynodon dactylon*. In the current study, an opposite result has been observes, macro hair was present but micro hair was absent. These features were similar to the results of Prat (1934, 1961), Metcalfe (1960), Ahmad (2009), and Khan *et al.* (2017). It may be due to environmental variations as *C. dactylon* is a wide spreading grass, which varies considerably in different habitat. Ishtiaq *et al.* (2018) also found that micro-hair was absent in *Cynodon dactylon*. In the present investigation, *Cynodon dactylon* showed dome shaped subsidiary cells at both abaxial and adaxial surface of the leaves.

5.5 Quantitative leaf epidermal characters

The findings on quantitative leaf epidermal characters are discussed here. For long cell numbers per mm^2 highest mean value (7.72) was found in accession collected from Khulna, while the lowest mean value (2.14) was found in case of Saint Martin's Island in abaxial surface of leaves. In adaxial surface of the leaves highest mean value (9.65) for long cell numbers per mm^2 was found in accession collected from Shariatpur and lowest mean value (3.31) was found in Barguna. In abaxial surface of the leaves long cell length was highest (110.26 µm) in case of Saint Martin's Island and lowest (32.20 µm) in case of Cox's Bazar. On the contrary, long cell length was highest (90.76 µm) in case of Barguna and it was lowest (31.39 µm) in case of Shariatpur in adaxial surface of the leaves. Long cell width was found to be highest (8.82 µm) in case of Saint Martin's Island and lowest (4.03 µm) in case of Cox's Bazar in abaxial surface of the leaves. But in adaxial surface of the leaves long cell width was highest (12.33 µm) in the accessions of Cox's Bazar and lowest (3.40 µm) in case of Rangamati. The variations regarding different parameters in case of long cells might be due to variations of environmental behaviour such as water stress and changes in temperature.

Stomatal frequency was found to be highest (5.56 mm^{-2}) in the accession of Gazipur and lowest (1.78 mm^{-2}) in case of Saint Martin's Island in abaxial surfaces of the leaves. In adaxial surfaces of the leaves stomatal frequency was highest (6.61 mm^{-2}) in case of Shariatpur and lowest (3.02 mm^{-2}) in case of Barguna. Leaf morphological characters like stomatal frequency, their distribution, and few other epidermal features may affect gas exchange quite remarkably and their relationships with key environmental factors such as light, water status, and CO_2 levels which have been found to respond to changing environmental variables of temperature, rainfall, irradiance and CO_2 (Royer 2001a). Therefore, they mainly can contribute to the ability of plants to control their water relations and to gain carbon (Hetherington and Woodward 2003). It has been shown that environmental signals such as light intensity, carbon dioxide concentration and water availability may affect stomatal development by modifying their size and frequency (Knapp *et al.* 1994, Dyki *et al.* 1998). Therefore, it is possible that variations in stomatal characteristics may influence plant growth and productivity (Kundu and Tigerstedt 1998). Reduction in stomatal frequency and size might also be an efficient feature of checking under water loss via transpiration during limited water availability and under high salinities as reported by different researchers (Walsh 1990, Bray and Reid 2002). The salt range ecotype showed decreased stomatal area and frequency under saline conditions on the adaxial leaf surface, so due to this it can be regarded as the best adapted ecotype against highly saline environments.

Inherent variation is a factor that may obscure the potential use of stomatal frequency as a paleoclimatalogical tool (Wagner *et al.* 2005). In terms of stomatal frequency this congenital variation is the variability in stomatal distribution across the leaf surfaces (Poole and Kurschner 1999) and has the potential to be large in angiosperms (Uhl and Kerp 2005). The reality of reduced stomatal conductance as a response to increased CO_2 has been inferred from measurements of transpiration rates during CO_2 -doubling experiments with agricultural species as well as tree seedlings. An increasing CO_2 concentration often leads to a significant decrease in leaf conductance corresponding with an increase in water-use efficiency (Eamus 1991). These parameters are helpful to differentiate the species. However, the stomatal features may prove to be a little taxonomic value unless the developments of different stomata types were studied. A greater number of information on taxa will be helpful to understand the taxonomic value of stomata type and distribution. Beerling (1995), and McElwain and Chaloner (1995) provided evidence that stomata frequency declined in response to increasing CO_2 and might have occurred over geological time. Stomatal frequency in present day can be estimated by growing them at different CO_2 concentration (Vesque 1989).

Epidermal cell number per mm^2 was found to be highest (23.89) in accession of Gaibandha and lowest (4.5) in case of Saint Martin’s Island in abaxial surface of the leaves. On the other hand, epidermal cell number per mm^2 was highest (19.99) in case of Shariatpur and that was lowest (9.28) in case of Barguna in adaxial surface of the leaves. Fernandez and Mujica (1973) determined that an increase in light intensity decreased the epidermal cell number and increased the stomatal index and size. Schoch *et al.* (1984) reported that blue and far-red light reduced the stomatal index while red light increased this stomatal index. Kim *et al.* (2004) showed that blue and red light increased the stomata size and decreased the stomata number. Lee *et al.* (2007) found that white light increased the stomatal number and size, while blue light reduced the mentioned parameters.

In abaxial surface of the leaves stomatal index was highest (28.43 %) in the accession of Jessore and lowest (25.36 %) in case of Jhenaidah. Stomatal index was found to be highest (28.76 %) in case of Khulna and lowest (24.97 %) in case of Jhenaidah in adaxial surface of the leaves. Stomatal length with guard cell was highest (21.50 µm) in accession of Cox’s Bazar and lowest (11.50 µm) in accession of Mymensingh in abaxial surface of the leaves. In adaxial surface of the leaves stomatal length with guard cell was highest (17.59 µm) in case of Saint Martin’s Island and lowest (11.56 µm) in case of Mymensingh. In adaxial surface of the leaves stomatal breadth with guard cell was found to be highest (13.71 µm) in case of Saint Martin’s Island and lowest (8.11 µm) in case of Khulna in abaxial surface of the leaves. In adaxial surface of the leaves stomatal breadth with guard cell was highest (15.62 µm) in case of Barguna and lowest (8.11 µm) in case of Khulna. Tufail *et al.* (2017) observed stomata with small dimensions in ecotype of *Cynodon dactylon*, which could be related to a more efficient physiological regulation since less turgor is required for the opening and closing of the ostiole. It reinforces what had been previously reported in fossils of species belonging to several families, including Poaceae (Franks and Beerling 2009) along with different plant species in which higher stomatal densities mediated by small sized stomata provide enhanced conductivity and higher photosynthesis rates (Franks *et al.* 2009 a & b, Drake *et al.* 2013, Vrablova *et al.* 2017). Nevertheless, Hetherington and Woodward (2003) pointed out that when the environmental alterations are unfavourable the conductance of small stomata is quickly reduced. Carpenter and Smith (1975) had been able to establish such a relationship involving stomata size and growth habit. Xerophytic species have much smaller stomata than mesophytic species. It may be compensating for the presence of larger stomata and it may be associated with adaptive success of polyploids (Van de Peer *et al.* 2017), mainly regarding water stress and changes in temperature (Simonneau *et al.* 2017).

Epidermal cell length was found to be highest (80.06 µm) in accession of Saint Martin's Island and lowest (23.07 µm) in accession of Faridpur in abaxial surface of the leaves. The values for epidermal cell length were highest (72.06 µm) in case of Barguna and lowest (28.76 µm) in accession of Khulna in adaxial surface of the leaves. Epidermal cell breadth was found to be highest (21.74 µm) in case of Cox's Bazar and that was lowest (7.31 µm) in case of Khulna in abaxial surface of the leaves. Epidermal cell breadth was highest (12.20 µm) in accession of Cox's Bazar and lowest (7.10 µm) in accession of Faridpur in adaxial surface of the leaves. The epidermis is comprised of various types of functionally specialized cells which play may vital role in restricting water loss, regulate gaseous exchange, defense, attract pollinators, photosynthesis, transpiration, respiration, mechanical strength and flexibility. Palmer and Tucker (1981) also observed that foliar epidermal features were useful in the systematics and characterization within sub families and tribes. Many leaf epidermal characters such as length and shape of epidermal cells, stomata, stomatal type, papillae, prickle angular, macro and micro hair, hooks, margins and silica bodies are taxonomically informative and can be used as an important tool in the delimitation of grasses (Prat 1932, Metcalfe 1960, Ellis 1979, Petronela and Nevana 2010). Watson and Dallwitz (1992) reported detailed description of the leaf epidermis in numerous taxa, pointing out the significance of these characters in the systematics of the Poaceae.

The values for number of silica bodies per mm^2 was found to be highest (10.26) in case of Bandarban, while it was lowest (2.13) in case of Saint Martin's Island in abaxial surface of the leaves. In adaxial surface of the leaves number of silica bodies per mm^2 was highest (7.95) in case of Naogaon and lowest (2.00) in case of Cox's Bazar. Prickles angular number per mm^2 was highest (0.32) in case of Gaibandha and lowest (0.19) in accession of Mymensingh and no prickles angular was found in case of Barguna, Cox's Bazar and Saint Martin's Island in abaxial surface of the leaves. Prickles angular number per mm^2 was found to be highest (0.42) in case of Thakurgaon and lowest (0.18) in case of both Pabna and Rangamati and no prickles angular was found in case of Barguna, Cox's Bazar and Saint Martin's Island in adaxial surface of the leaves. Number of hooks per mm^2 was highest (0.96) in accession of Rajshahi and lowest (0.02) in accession of Thakurgaon, Sherpur and Saint Martin's Island in abaxial surface of the leaves. Number of hooks per mm^2 was highest (1.69) in case of Lalmonirhat and it was lowest (0.02) in case of Barguna in adaxial surface of the leaves. In accession of Cox's Bazar, hook was not found in both abaxial and adaxial surfaces of the leaves. Macro hair number per mm^2 was found to be highest (0.26) in case of Cox's Bazar and lowest (0.02) in case of Khagrachari in abaxial surface of the leaves. In adaxial surface of the leaves macro hair number per mm^2 was highest (0.52) in case of Cox's Bazar and lowest (0.02) in case of Gazipur. No macro hair was found in case of Pabna. However, all these characters were found to be highest and lowest both abaxially and adaxially, which indicates the taxonomic importance of the foliar characters. The foliar epidermis provide a number of significant taxonomic characters. The biosystematic and taxonomic studies of a number of families established the importance of leaf epidermis (Baranova 1972, Raju 1981, Stace 1984). Although the taxonomists realized lately the importance of micromorphology of the epidermis and thus, the taxonomic monographs are now being considered incomplete without it (Rejdali 1991). The diversity and distributional pattern of the above mentioned characters can be viewed from different perspectives and used as a model system for investigations into developmental biology, ecology, physiology, morphology and evolution.

5.6 Stem and root anatomical characters

Monocot stems have scattered vascular bundles and most of them are found on the outside edge of the stem. They are found to be surrounded by large parenchyma in the cortex region. Pith region is found to be absent in monocot and as compared to the pith region parenchyma cells are found to make up the cortex region in between smaller bundles and epidermis. On the other hand, monocot roots exhibit vascular bundles arranged in a ring shows no secondary growth, except some cases like bamboo and palm tree. However, monocot secondary growth differs from dicot secondary growth in that new bundles are formed at the edge of the stem, which are close together and provides support for the stem. In monocot root, secondary growth is absent. In this case, the xylem and phloem are radial, around central pith xylem and phloem are arranged in circular fashion many xylem bundles, and exarch condition is found to occur.

Anatomical characteristics are salient features, which can be implicated in taxonomical diagnosis, as well as in describing ecological conditions. It is very much evident now that *Cynodon dactylon* grows well on a wide variety of soils from heavy clays to deep sands, provided fertility is not limiting. It tolerates both acid and alkaline soil conditions and is highly tolerant to saline conditions. This grass species survives some flooding but does best on well drained sites. They are also drought tolerant. Despite of many types of unfavourable conditions *C. dactylon* can survive changing their anatomical and physio characteristics. Considering this view point anatomical features of the stem and root of twentyfour accessions of *C. dactylon* collected from different habitats were studied in the present investigation.

Usually, moderate soil temperature and acidic clay soil do not have drastic effect on their morphological, anatomical and physiological properties. But natural population of *Cynodon dactylon* can have considerable genetic variation for tolerance to soil temperatures, salinity and drought (Speranza 1995). Few accessions of this grass species in the present study have been found to tolerate salinity successfully. However, growth of this grass species is stimulated by moderate salinities and it can tolerate relatively high salinities (Mass and Hoffman 1977). The accessions collected from Barguna, Cox's Bazar and Saint Martin's Island studied in the present investigation revealed somewhat different anatomical features both in case of shoot and root. In case of hypodermis these three accessions showed greatly increased sclerification outside the vascular bundles. In other accessions sclerification was low and this is why, sclerenchyma thickness values were high in all the accessions except that of Cox's Bazar and Saint Martin's Island. Similarly, epidermis thickness values in all the accessions were high compared to that of Cox's Bazar. Munns (2002) stated that this type of plant material can be directly used for revegetation of such affected areas and the introduction of new salt tolerant genes to increase tolerance of crop cultivars also. In spite of different sorts of anatomical and morphological modification in plant bodies this grass species was found to be competent to minimize the detrimental effects of salt stress. Salt tolerant plant species shows a wide range of anatomical adaptive features like increased succulence both in case of root and stem, thick cuticle and deposition of wax, salt secretory trichome and glands, thick and many layered epidermis and well developed water storing tissues in the cortex, widening of casparian band and enhanced developments of root endodermis (Akram *et al.* 2002, Wahid 2003). In present study cortex was parenchymatous having thin wall cells, but these cells surrounding the vascular bundle gradually sclerified into thick walled cells. Cortex thickness values were significantly higher in all the accessions of bermudagrass except that of Saint Martin's Island and cortex, where the values were remarkably low.

In case of stem of bermudagrass in the present study cambium was found to be absent as the whole procambium was consumed in the formation of vascular tissues. The vascular bundles were, therefore, conjoint, collateral but closed. Number of vascular bundles per transverse section of stem was highest in salt tolerant accession collected from Cox's Bazar and then 2nd highest value was found in case of Barguna, and lowest in that of Gaibandha. But Awasthi and Pathak (1999) reported increased vascular bundle area in relatively less salinity tolerant plants. The population growing in salt range area, however, showed slight decrease in this character along with increased its number and that may be better adaptation for efficient water uptake as reported by Yujing *et al.* (2000). Xylem vessels are generally reduced under salt stress (Gadallah and Ramadan 1997) and Ling *et al.* (2002) and this characteristic, however, remains very much stable in both salt range and non-saline area. This might be due to nature and type of plant species which varies from mesophyte to xerophyte. In population of *C. dactylon* from the salt range, the stem was found to be increased under saline regime. This increased succulence in stem may aid to store additional water and this is why this grass species shows better survival under unfavourable conditions. Hameed *et al.* (2010) reported that salt range population of *C. dactylon* showed specific root and stem anatomical adaptations for its better survival under harsh saline environment. Increased exodermis and sclerenchyma, endodermis, cortex and pith parenchyma in roots were critical for checking water loss and enhancing water storage capability. In stem, increased stem area, increased epidermis and sclerenchyma thickness, increased cortex thickness and increased number and area of vascular tissue seemed to be crucial for its better survival under harsh saline environment.

Xylem of stem in the present study was found to made up of vessel, tracheids, xylem parenchyma and a few xylem fibers. The phloem was found to made up of sieve elements and phloem parenchyma. Younis *et al.* (2014b) stated that salinity elevation in soil has contrary effects on xylem and phloem area of the stem and root as their area tended to decrease under salt stress condition. It was due to the reduction of water uptake by the plants under high saline conditions and this reduction in water intake by the cells created osmotic condition. However, plants growing under saline conditions undergo different anatomical and cytological changes (Winter 1988, Huang and Van Steveninck 1990). The modifications are different in organ to organ at different levels of organization (Mills 1989). Salt tolerance of natural populations depends on the existence of available genetic variability and evolutionary history of the habitats (Ashraf 2004). Exploration of genetic variation in natural populations, identification of genetically based markers and finally the incorporation of stress tolerance traits into glycophytic plants have been emphasized recently (Munns 2011).

Roots of few plant species have been studied and few details have been reported about the adaptive anatomy of wet land grass rhizomes or stolons. In present study transverse section of bermudagrass roots showed epidermis was composed of a single thin layered wall, tangentially elongated to irregular shaped cells and highest value for epidermis thickness were higher in the accessions of non-saline area compared to that of saline area. On the other hand, cortex thickness was found to be higher in case of the accessions collected from saline belt. Endodermis was quite distinct single layered, thick walled with tangentially elongated cells. Yang *et al.* (2011) reported that in *C. dactylon*, the endodermis had lignified casparian bands and almost complete suberin lamellae with 5 mm of the root tip; a few passage cells were present. The endodermis was found to be heavily suberized with thick outer tangential walls and with lignified secondary cell walls in old roots. Adjacent cell layers of the inner cortex developed thick walls.

In present study, lysigenous aerenchyma was observed in roots of *C. dactylon* in all the accessions but comparatively the aerenchyma area in accessions of saline range was higher. Arber (1920), Justin and Armstrong (1987), Jackson and Armstrong (1999) and Evans (2003) stated that lysigenous aerenchyma is formed by the creation of gas spaces as a result of death and subsequent lysis causing collapse of files of cells, e.g. root cortical cells. The stems of *C. dactylon* do not have selective barrier layers of endodermis and exodermis, but they have a thick and cuticular epidermis. While adventitious roots had extensive lysigenous aerenchyma (Yang *et al.* 2011). Air space tissues take two general forms, cavities (especially in pith but sometimes also in cortex) and aerenchyma (organized tissues, structured during their development) as per suggestion of Jung *et al.* (2008) and Seago *et al.* (2005). Jung *et al.* (2008) also suggested that in grass roots, lysigenous aerenchyma is widespread. Takahashi *et al.* (2014) stated that the formation of aerenchyma tissue is an anatomical adaptation to excess water stress. Aerenchyma consists of longitudinally interconnected gas spaces that enable the rapid transport of gases between, and within shoots and roots (Evans 2003). In many wet land and aquatic plants, aerenchyma is developed in the shoots as well as in the roots. In roots, primary parenchyma forms in the primary cortex and can be broadly classified into two types: schizogenous aerenchyma and lysigenous aerenchyma (Armstrong and Armstrong 1994, Seago *et al.* 2005, Jung *et al.* 2008). In present study the identified aerenchyma might be lysigenous, since it develops based on the excess water stress during flooding and waterlogging. Soil waterlogging or flooding of the soil occurs when soil is saturated with water. Owing to the soil diffusion of gases in water, gas exchange between the soil and the atmosphere is strongly hindered (Colmer 2003). Soil waterlogging reduces plant growth as O_2 availability in the root zones decreases (Armstrong 1979, Jackson and Drew 1984). To cope with waterlogging, plants usually develop new roots with aerenchyma (Laan *et al.* 1989, Visser *et al.* 1996, Huber *et al.* 2009). Aerenchyma refers to tissue with air spaces that provide an internal pathway for oxygen diffusion in organs under water logged / submerged conditions (Armstrong 1979). Apart from this, plants may display other adaptive strategies that might act together to improve root aeration and oxygen consumption within the root (Cardoso *et al*. 2013).

In present study, sclerenchyma thickness of the root of *C. dactylon* was found to vary from 0.07 cm to 0.24 cm and intensive sclerification in outer cortex, vascular region and endodermis layer were observed in roots of the accessions collected from Barguna, Cox's Bazar and Saint Martin's Island. In accessions of non-saline area sclerification in roots was found to be low. This might be due to differential tolerance of the three ecotypes of bermudagrass from salt range relates well to the soil physicochemical properties of the habitats. Maintenance of ion balance in plants subjected to saline conditions is vital for sustaining growth and productivity under such environmental adversities (Munns and Tester 2008). Anatomical modifications to overcome high salinities are very specific not only in the grass species but also in the ecotypes (Hameed *et al.* 2011).

Vascular bundles in the roots of *C. dactylon* in present study were found to be arranged in a ring of differential radials. The different characters of vascular bundles like metaxylem area, phloem area, pith area and pith thickness were found to differentiate among the accessions of non-saline and saline habitats in terms of their highest and lowest values. Ashraf (1994) and Hameed and Ashraf (2008) stated that plants growing on naturally salt affected soils must have evolved a multitude of morpho-anatomical and physiological adaptive characteristics in view of considerable length of time they have been exposed to high selection pressure of the habitat like high salinity and aridity. This view point might be the region of differential response of the same plant species like *C. dactylon* in the present study. Moreover, Hameed *et al.* (2011) stated that specific anatomical and physiological modifications in plants exposed to stressful environments may enable them to thrive well on such environment. Although plants use a variety of physiological phenomena to counter act salt stress, regulation of ion homeostatis is one of the premier physiological process operating in plants exposed to salt stress (Zhu 2003). This includes selective ion uptake (Flowers and Colmer 2008), accumulation of toxic ions, partially in terms of increased succulence (Hammed *et al.* 2009), and excretion of such unwanted toxic ions (Ramadan and Flowers 2004, Naz *et al.* 2009).

Twentyfour accessions of *C. dactylon* were studied in this investigation and a few of them were collected from that type of habitat which belongs to somewhat drought area as well as to different altitudes. Thus, the anatomical features of all these accessions were different to some extent. Altitudinal stress had a very little impact on stem anatomy. Epidermal cells were radially broader than tangential direction and no such differences were found in the length of epidermal cells of all the internodes. Hypodermis was found to be made up of thick walled lignified sclerenchyma cells. Down to hypodermis vascular bundle were found without cambium to show some pattern of arrangement. Ahmad *et al.* (2016) reported that epidermal cell was found to be decreased significantly along the elevation in ecotypes from foot hill up to top hill sites. In contrast, sclerification increased significantly along with increase of elevation and the maximum increase in sclerification was recorded in top hill ecotype. In their study, cortical cell area, vascular bundle area and numbers generally decreased alongwith increase in altitude. They also reported that root anatomical characteristics like epidermal cell area and cortical region thickness showed a consistent decrease from foot hill to top hill with increase in the elevation. They reported that cortical and endodermal cell area significantly increased similarly but further increase in altitude showed significantly decreased cortical cell area in top hill ecotype. Sclerenchymatous thickness increased invariably in all ecotypes with increase in the altitude. However, in the present study a very small type of variations in root and stem anatomy might be due to no such great differences in altitudinal variation from sea levels to northern part of Bangladesh. Nevertheless, a few remarkable anatomical features were noticed in those accessions, collected from Chittagong hill tracts and saline area.

On the contrary, root and shoot anatomical characteristics were found to show a little variation among the accessions in the present study and that might be due to seasonal drought conditions. Angiosperms that occupy continuously wet habitats have constitutive aerenchyma, but roots of many other angiosperms can respond to anaerobiosis by developing lysigenous or schizogenous cortical aerenchyma (Konings and Lambers 1991). In dry land plants, this response is mediated by increased ethylene levels, which are the result of reduced diffusion out of the roots under conditions of poor aeration or waterlogging as well as of increased ethylene production under low oxygen concentrations (Kawase 1981, Drew 1990, Konings and Lambers 1991). The most striking feature of the grass species *C. dactylon* under study was the positive response of root aerenchyma in both dryland and saline habitat. This is one of the adaptive responses of this grass species. This grass species had a well differentiated hypodermis including exodermis with a few isodiametric cell layers. However, it has been reported that a compact exodermis may play a role in preventing the collapse of the cortex. Simultaneously, a thickend exodermis may act as a barrier excluding toxic reduced ions and also reduce the outward radial fusion of oxygen (Moog and Janiesch 1990). The cell wall thickenings in exo- and endodermis under drought may function in protecting the stele and cortex from dessication (Stasovski and Peterson 1991). Baruch and Merida (1995) stated that the effect of drought on root anatomy is less understood. Roots are exposed to drying soil may exhibit a pronounced suberization of the exodermis and endodermis, which is believed to protect the death of cortical cells.

5.7 Interphase nuclear phenotype and chromosome count

In the present experiment, interphase nuclei of different accessions of *C. dactylon* were found to be chromocentric. Joshi and Ranjekar (1983), and Patankar and Ranjekar (1984) found similar type of chromocentric nuclear organization in *Phaseolus* species. Lafontaine (1974) stated that structural organization in plant cell nuclei are two types and they are namely chromocentric and reticulate. The number of chromocentres was always less compared to that of their somatic chromosome numbers. The values ranged from 11.4 (Pabna) to 17.60 (Gazipur, Sherpur and Faridpur). The chromocentric number did not correlate with the somatic chromosome numbers of the diploid, tetraploid or aneuploid accession. Kabir (1993) reported more or less same number of chromocenter in six *Crotalaria* species. The reduction in number of chromocenters might be due to fusion or overlapping of chromocenters indicating the somatic association of chromosomes (Kabir and Singh 1989). Dayal and Prasad (1983) stated that the number of chromocenters is considered to be controlled genetically and is therefore a species specific character.

In case of heterochromatin percentage per nuclear area the values obtained in this study were highly variable. The values ranged from 19.759% (Jhinaidah) to 66.022 (Shariatpur). In this study higher percentage of heterochromatin might be due to the presence of different classes of heterochromatin (Vosa 1970, La Cour 1978). HCl-Giemse banding is also known to visualize most of the types of heterochromatin on chromosome (Joshi and Ranjekar 1982). The variable result in the present investigation might be due to the fact that HCl-haematoxylin banding revealed more than one class of heterochromatin in interphase chromosomes. Lafontaine (1974), and Nagl and Fusenig (1979) stated that chromocentric nuclear organization is assumed to be governed by small size of chromosomes and low DNA content. This holds partially true for all the accessions of *C. dactylon* which showed very small size of chromosomes.

Yamakawa and Sparrow (1965 and 1966) studied interphase chromosome volume as a reliable index of radio sensitivity in plants and their work established a positive linear correlation between radiation sensitivity and chromosome volume in plant cells. In present study this parameter (interphase chromosome volume) was considered since all the accessions of *C. dactylon* were collected from different ecological conditions, where the temperature as well as radiation index may vary from one zone to another. In this study, the highest value (2.507 μm^3) for interphase chromosome volume was found in the accession collected from Rangamati and the lowest value (0.028 μm^3) was found in the accession collected from Gaibandha. This character was not correlated with chromosome number. Thus, it may be said that increase or decrease of interphase chromosome volume may be caused due to alteration of cell membrane configurations, modification of chromosomal volume and changes in sensitivity to physical or chemical factors, functioning always in the environment. Yamakawa and Sparow (1966) reported increase in mutation rate per roentgen in higher plants and that was highly correlated with increase in both ICV (interphase chromosome volume) and DNA content per chromosome.

In present study, the most striking feature of *C. dactylon* was found in case of chromosome number. Range of chromosome number was really surprising when the basic number is considered to be x = 9 (Forbes and Burton 1963, Clayton and Harlan 1970). In this experiment only nine accessions out of 24 were found to be diploid (2n = 18). Only one was found to be tetraploid and rest of them were aneuploid. De Silva and Snaydon (1995) stated that most populations contained only tetraploid plants (2n = 36), but populations from roadsides and lawns in the wet region and from forest in the hill country contained only diploids plants (2n = 18). They also said that, soil acidity seems to be the main factor determining the distributions of the cytotypes. They did not report any triploid or any aneuploid. On the contrary Dhaliwal *et al.* (2018) reported *Cynodon dactylon* with the basic chromosome number of n = 8, while few other reports on five different cytotypes with basic chromosome number n = 8, n = 9 and n = 18 have been reported in his literature based on works of several investigators (Malik and Tripathi 1968, Gupta and Srivastava 1970, Sachdeva and Bhatia 1980, Kumar and Sachdeva 1988, Sinha *et al.* 1990). However, the present findings may be supported by Federov (1974), Christopher and Abraham (1974), Brown (1950) and Hurcombe (1947). Gupta and Shrivastava (1970) reported one diploid (2n = 18) and three triploid (2n = 27) collections of *C. dactylon* . Chiavegatto *et al.* (2016) aimed to determine the karyotype asymmetry index among accessions of *Cynodon* to discriminate them. They ultimately reported two diploid, one triploid, four tetraploid and one pentaploid accession. Their findings indicated predominant level of ploidy for the accessions was tetraploid accessions. Along with their findings on chromosome counts it may be said that there is a wide genetic variability which is very important for origin, evolution and development of plant species. The preponderance of aneuploidy observed in the present study throws light on the evolutionary tendencies which might go hand to hand in the phylogenetic evolution due their different adaptive values. When one of the characters might put a type or accession of *C. dactylon* at highest level of advancement then the others may be still in original stage. However, aneuploids play important role in the breeding programme and thus, the main objective of forage breeding programme may be an important way to obtain those genotypes with desirable agronomic traits.

5.8 Effects of polyploidy on the stomatal characters of *Cynodon dactylon*

In the present study 15 accessions of *Cynodon dactylon* were found to be polyploid in nature, where chromosome numbers indicated both euploid and aneuploid behaviour. Only one accession collected from Naogaon was tetraploid (2n = 4x = 36) i.e; euploid and rest of them were aneuploid. Generally, the size of stomata is used as criteria for the detection of polyploid. The size of stomata can be compared in 2x and 4x plants of the same species. But in this study no such remarkable difference was found when stomatal frequency and stomatal index were predictedly both in case of aneuploid and tetraploid accessions. Similarly, the fourteen aneuploid accessions showed no such variations regarding the above said parameters when compared apparently to that of tetraploid accession. This might be due to the statement made by Gupta (1999) that since the size of organs and organisms depends on cell elongation and the number of cells, the increase on original size may not always lead to increase on the size of the organ and the organism, because the number of cell division is reduced in polyploids. However, Chaves *et al.* (2018) studied the effect of polyploidy on the leaf epidermis structure of *Cynodon dactylon* and reported that as polyploidy level increased, there was an increase in stomata dimension and a decrease in stomatal density. They also mentioned that the variables stomatal density and stomatal index were not efficient to discriminate the accessions. Thus, it may be said that the present study is being supported by the findings of Chaves *et al.* (2018) particularly in case of stomatal frequency and stomatal index.

Chapter VI

Summary

6. SUMMARY

The objectives of the present study on *Cynodon dactylon* were to examine role of some ecological factors on intraspecific variation based on distribution and adaptation of this grass species, to make an analysis of morphological variations of the ecotypes from botanic and agronomic point of view, to determine the physico-chemical properties of the soil of original habitat and experimentation field, to focus on the qualitative and quantitative leaf epidermal characteristics with special reference to stomatal features, to study anatomy of root and stem in relation to distribution and arrangement of different tissue and tissue system qualitative and quantitatively, and to determine the nuclear phenotype along with the estimation of ploidy level.

To accomplish the above mentioned objectives a good numbers of literature were reviewed and based on the findings and discussion of several research workers different methods were adopted, which helped to arrange the style and contents of the present study. The literature reviewed in this study were mainly before 1995 but a good number of recent articles were also reviewed which reflected the distribution, adaptation and cytotypic diversity of *C. dactylon* very effectively. As a whole, these research articles helped also to narrate the findings in the present study very fruitfully. A total of 24 accessions of *C. dactylon* growing in different bio-ecological zones of Bangladesh along with their habitat soils were studied in the present investigation. These were used as the experimental materials. All the 24 accessions were also transplanted plot wise in research field of the Institute of Biological Sciences, University of Rajshahi, Bangladesh. No fertilizers were used in the experimentation field and the accessions were found to be established from root splits in 1.5 m × 1.4 m plots.

The morphological growth and development were observed within the first four months after cutting before most accessions had started flowering. Six randomly selected areas of 25 cm × 25 cm in the plot were chosen for rhizome observation and six randomly areas of 200 cm × 50 cm at the end of plot were chosen for stolon observation. From this experimentation field data were recorded on twentysix different quantitative characters from each accession and their mean values were tabulated.

Soils from rhizosphere at 16 cm depth were taken for each habitat to analyze their physico-chemical characteristics. The soil extract was used to determine the pH, organic matters, K^{+}, Ca^{2+}, Mg^{2+}, total nitrogen, P, S, B, Cu, Fe, Mn and Zn. The soil test values were interpreted based on critical limits following Fertilization Recommendation Guide.

For leaf epidermal anatomy in relation to ecotypic adaptation the features considered were stomata type, shape of subsidiary cells and silica bodies, types of long cell margins, presence of macro- and micro hairs, prickles angular and hooks, number of epidermal cells, long cells, silica bodies, prickles angular, length and width of long cells, breadth of stomata and epidermal cells, stomatal frequency and index on both abaxial and adaxial surface of the leaves of *C. dactylon*.

For studying stem anatomy of all the 24 accessions the anatomical characters like epidermis thickness, cortex thickness, sclerenchyma thickness, vascular bundle area and number, metaxylem and phloem area from transverse sections of stem were recorded. The anatomical characters recorded for roots were epidermis thickness, cortex thickness, endodermis thickness, sclerenchyma thickness, metaxylem and phloem area, pith thickness and pith cell area.

For nuclear phenotype and chromosome counts root tips of all the accessions of *C. dactylon* were used. Stained meristematic zones of the root tips revealed their chromosome numbers. Along with nuclear and interphase chromosome volume chromocenter numbers were counted, which revealed also values for heterochromatin in case of all the accessions.

At present Bangladesh comprises sixty-four districts of them twentythree districts plus one location of high salinity were considered as the study area. Accessions of *C. dactylon* from all these twentyfour habitats were collected along with their habitat soils. In almost all the locations *C. dactylon* were found to creep extensively by means of scaly rhizomes or by flat stolons. Some of them were found to produce seeds side by side vegetatively. It was also observed that in case of plain land they were growing moderately on well drained soil, either acidic or alkaline. They need mainly soil moisture along with nutrients and sunlight. Accessions of *C. dactylon* used in the present study were collected mainly from roadside or in the vicinity of lawn or homestead. Besides they were found to grow in the crop lands. Findings reveal growth intensity of this grass species from low to high. Nine accessions showed low intensity, six showed medium and rest of them showed high intensity of growth. According to the opinion of the local peoples all these accessions of *C. dactylon* were found to show their duration/period/season of production round the year. High intensity of growth was found in those locations where the soils were highly alkaline and those particular habitats were at Rajshahi, Pabna, Narsingdi, Mymensingh, Jessore, Jhenaidah, Faridpur, Shariatpur and Barguna. On the contrary, low intensity of growth was observed in case of Lalmonirhat, Thakurgaon, Panchagarh, Gaibandha, Gazipur, Khagrachari, Bandarban, Rangamati and Cox's Bazar where the soils were found to be acidic. Among twenty-four study area soil pH was found to range from 4.9 to 8.5 and almost all the alkaline soils showed the growth of this grass species as medium or high. Among all these accessions four were found to be drought tolerant and three were salt tolerant very prominently. Present findings indicate that in less than 50% locations of Bangladesh *C. dactylon* is growing very enormously as perennial grass. Thus, it may be said that this grass has become truly cosmopolitan all over the country and exhibits a wide ecological tolerance due to its great plasticity and capability of growing in different soil types under varied climatic conditions.

The accessions of *C. dactylon* showed variation in many of the morphological characters of taxonomic importance. Shoot number and length, node number, internode length, fresh and dry weight of shoot were found with higher values in the accessions collected from Cox's Bazar, Khagrachari, Rangamati and for the last three characters in Bandarban, subsequently. On the contrary, lower values of shoot length and internode length were found in the accessions of Panchagarh and Dinajpur, for shoot number and number of nodes in Panchagarh and for last two characters in Shariatpur, respectively. In the same way highest and lowest values for root number and length, number of inflorescence, number and length of raceme, seeds per shoot, number of seeds per plant, leaf characters like number of leaves, total leaf area, total leaves fresh and dry weight, leaf length and leaf width alongwith maximum and minimum plant height, number and length of stolons were evaluated from each accession for the respective values. Growth habit of all the twenty-four accessions showed erect and prostrate nature based on angle of stem to ground. Among the accessions, only eight showed their both the growth habit in the ratio of 1:1 or 2:1 and rest of them were either erect or prostated.

As mentioned earlier regarding the physico-chemical properties of the habitat soil, the soil of the experimentation field was also analyzed physico-chemically and their magnitude of organic and inorganic matters were determined. The soil of this experimentation field was found to be loamy and pH value was found to be 8.1. The values for organic matter, K, Ca, Mg, total nitrogen, P, S, B, Cu, Fe, Mn and Zn in experimentation field of Rajshahi were found to be 1.33, 0.17, 42.90, 1.01, 0.08, 10.0, 9.10, 0.70, 2.67, 15.6, 6.0 and 2.95, respectively. The soil test values based on critical limits magnitude of physico-chemical properties of the soil of different habitats considered in the present study was interpreted.

Findings on leaf epidermal anatomical characteristics in relation to ecotypic adaptation of *C. dactylon* were illustrated under two sub-heads as qualitative and quantitative leaf epidermal characters. Qualitatively the leaves were found to be amphistomatic. The stomata were paracytic and dumbbell shaped with two subsidiary cells. The epidermal cells were elongated and arranged in vertical rows parallel to the long axis of the leaf. The long cell margins were found to show sinuous in all the accessions except that of Barguna, Cox’s Bazar and Saint Martin’s Island. Silica bodies were saddle shaped in most of the collections but saddle and cross shaped silica bodies were found in case of Narsingdi, Jessore and Faridpur. On the contrary, horizontally elongated silica bodies were found in the accessions collected from Barguna, Cox’s Bazar and Saint Martin’s Island. Macrohairs were present in all the accessions but microhairs were not found in any of the accessions abaxially or adaxially. Prickles angular and hook were found to be present in all the accessions except that from Barguna, Cox’s Bazar and Saint Martin’s Island. Quantitative leaf epidermal characters showed variable values for different characteristics on both abaxial and adaxial surface of leaves. The characteristics considered for quantitative measurement were long cell number per mm^2, length and width of long cells, epidermal cell number per mm^2, length and breadth of epidermal cell, frequency and index of stomata, stomata length with guard cell, stomata breadth with guard cell, number of silica bodies per mm^2, prickles angular number per mm^2, hooks number per mm^2 and macrohair number per mm^2. The diversity and distribution of the above mentioned characters can be viewed in different perspectives and used as a model system for investigation into ecological morphology.

Anatomical features of stem and root of twenty-four accessions of *C. dactylon* were illustrated through transverse section, which reveals the structure of epidermis to be comprised of a single layer with thick cuticle apparently. The outermost layer of stem was found to be made up of compactly arranged transparent, elongated and rectangular barrel shaped parenchyma cells. No visual differences were found in the length of epidermal cells of all the internodes. Highest value for epidermis thickness of stem was found in the accession collected from Lalmonirhat and lowest value was found in that of Cox's Bazar. Hypodermis was found to be made up of thick walled lignified sclerenchyma cells. Greatly increased sclerification outside vascular bundles were observed in stem of Barguna, Cox's Bazar and Saint Martin's Island. Sclerenchyma thickness was highest in accession of Rangpur and that was lowest in case of Cox's Bazar and Saint Martin's Island. In *C. dactylon* third tissue system was the ground tissue which did not show distinctive differences into cortex, endodermis, pericycle, pith and pith rays. The ground tissue system was found to be made up of parenchyma cells, which occupied the whole stem interior. Outer cells were small and angular towards the hypodermis but became large and oval in the inner region. Pith region was large and consisted of parenchyma cells. Abundant intercellular spaces were present in the ground tissue. Down to the hypodermis, vascular bundles were found to show some pattern of arrangement. Outer cycle vascular bundles were small and arranged in a complete circle. The vascular bundles of center were much larger than those next to hypodermis. Hypodermal vascular bundles were of two different sizes, large and small, arranged almost alternately with equal spacing. Cortex was parenchymatous usually having thin cell walls and these cells surrounding a vascular bundle gradually sclerified into thick walled cells. The vascular bundles were rounded in outline and they contained both phloem and xylem cambium was absent as the whole pro-cambium was consumed in the formation of vascular tissues. The vascular bundles were, therefore, conjoint, collateral but closed. Vascular bundle area of stem was highest in accession collected from Rajshahi and the lowest in accession of Khagrachari. Number of vascular bundles per stem was highest in accession from Cox’s Bazar, second highest in case of Barguna and lowest in accessions collected from Gaibandha. There were many vascular bundles scattered in the heavily sclerified area of the accessions collected from Barguna, Cox’s Bazar and Saint Martin’s Island Island. Metaxylem area of stem was highest in case of Rajshahi and that was lowest in case of Saint Martin’s Island. In case of phloem area the highest value was found in case of Rajshahi and lowest in accession of Saint Martin’s Island. Phloem and xylem were found to be separated from each other either by fibrous cells or by trachiary elements xylem parenchyma and phloem parenchyma with sclerified areas were observed in most of the bundles.

Root anatomy showed the features as epidermis, cortex, endodermis, pericycle, vascular bundle, conjunctive tissues and pith. Transverse section of mature root showed epidermis to be composed of a single layer of thin walled, tangentially elongated to irregular shaped cells. Highest value of epidermis thickness of root was found in accession collected from Rajshahi and lowest value was found in accession of Barguna. But exceptional results were found in accessions of Cox’s Bazar and Saint Martin’s Island, where the values for epidermis thickness of root were high compared to that of twentytwo accessions. Cortex was found to be differentiated into two zones, 1 or 2 layers of smaller, thin-walled, polygonal, lignified sclerenchymatous, and 4 to 6 layers of larger thin walled, elongated parenchymatous cells. Lysigenous aerenchyma was observed in roots of *C. dactylon* accessions in the present study. The value of cortex thickness was found to be highest in accession of Cox’s Bazar and lowest in case of Gaibandha. Endodermis thickness was highest in case of Saint Martin’s Island and lowest in case of Jhenaidah. Highest value of sclerenchyma thickness was found in accession of Rajshahi and that was lowest in case of Jhenaidah. Intensive sclerification in outer cortex, vascular region and endodermis layer were observed in roots collected from Barguna, Cox's Bazar and Saint Martin's Island. Vascular bundles consisting of xylem and phloem were found to be arranged in a ring on different radials. The highest and lowest values for metaxylem, phloem area, pith thickness and pith area were found in accession collected from different habitats not always on the basis of saline and non-saline area. Although the accessions grown in alkaline soil showed almost highest values for the above mentioned characters.

All the twentyfour accessions of *C. dactylon* in present study exhibited chromocentric nuclear organization. The chromocenters were seen distinctly on a very light background of the microscope. The number of chromocenters for each accession were not found to be corresponding with their somatic chromosome number. Sometimes, it was found to be significantly less and definitely not more than their respective chromosome number. In this study, out of twentyfour accessions only nine were found to be diploid with 2n = 18 chromosomes. Only one accession collected from Naogaon was found to be tetraploid with 2n = 4x = 36 chromosomes. Rest of the accessions collected from different habitats were found aneuploid with chromosome numbers of 12, 14, 16, 22, 24, 26, 32, 40 etc. Percentage of heterochromatin values were recorded by determining the area of the interphase nucleus and of chromocentres by planimetry and then the heterochromatin values were expressed as percent per nuclear area. The values for heterochromatin percentage per nuclear area obtained in this study were very much variable. Nuclear volume as well as interphase chromosome volume were determined. The obtained values for these two nuclear characters were not found to depend upon the number of chromosomes. Apparently no such relation was found between chromosome number and interphase chromosome volume. Since there is a general trend in plants for stomatal density and stomatal size to increase as ploidy level increase or decreases, one tetraploid and fourteen aneuploids recorded in the present study did not show such significant effect when stomatal frequency and index were estimated both in case of euploid and aneuploid.

Except distribution all the data recorded for adaptation of *C. dactylon* in terms of morphological variation, leaf epidermal anatomy, and stem and root anatomical attributes, and also cytotypic diversity were analyzed statistically. Both DMRT and ANOVA were applied for analyzing the data regarding adaptation and cytotypic diversity of *Cynodon dactylon* in twentyfour habitats of Bangladesh.

Chapter VII
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7. REFERENCES

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INTERPHASE NUCLEAR PHENOTYPE AND CHROMOSOMAL VARIATION IN *CYNODON DACTYLON* (L.) PERS

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Abstract

Interphase nuclear phenotype in different accessions (Acc.) of *Cynodon dactylon* studied in the present experiment showed chromocentric nuclear organization and the chromocenters were found to be visible clearly. The chromocenter numbers were not same and sometimes it was found to be significantly less and never more than total number of chromosomes. Percentages of heterochromatin values were expressed per nuclear area and the values range from 19.759% (Acc. 16) to 66.022% (Acc.18). Nuclear volume as well as interphase chromosome volume was found to vary 0.674 μm^3 (Acc.6) to 41.921 μm^3 (Acc.10) and from 0.028 μm^3 (Acc. 6) to 1.905 μm^3 (Acc. 10), respectively. The somatic chromosome number found to vary from 12 to 40. 2n = 18 chromosomes were found in eight accessions of *C. dactylon.* Only one accession was found to be tetraploid and rest of them aneuploid whose chromosome numbers were 12, 14, 16, 22, 24, 26, 32, 40 etc. The availability of aneuploid shows great aspects of forage breeding programme.

Key words: Aneuploidy, Camera-lucida, Chromosome number, *Cynodon dactylon,* Interphase, Nuclear volume

Introduction

Cynodon dactylon (L.) Pers. (commonly known as Bermuda grass and locally as Durba) is a typical warm-season turfgrass belonging to Poaceae. This species is widely distributed in warm, humid and semi-arid areas around the Himalayas with excellent heat and drought tolerance capacity (Harlan et al. 1970, Dunne et al. 2019). This grass species is widely adapted to various environments of tropical and sub-tropical regions of the world and there are some ways for genetic diversity and population structure analysis of Bermudagrass (Zheng et al. 2017). Bermuda grass has attracted interest of plant scientists because of their ease of cultivation, high forage production and high nutritional value (Pedreira 1996). This species shows high growth rate, grazing resistance, high carrying capacity and rapid establishment (De Lima and Vilela 2005). Cytogenetic information like interphase nuclear phenotype and chromosomal characterization are very useful parameters in distinguishing the cytotypes, accessions (Acc.) and even germplasm of a plant species (Huang et al. 2014). The information on chromosome number of *C. dactylon* is limited to the determination of ploidy level. On the basis of previous research work, this species has the basic chromosome number x = 9 (Dhaliwal and Gupta 2011, Zhi-Yun et al. 2013). However, this species has been reported with different

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ploidy level such as triploid (2n = 3x = 27), tetraploid (2n = 4x = 36), few pentaploid (2n = 5x = 45), and hexaploid (2n = 6x = 54) according to Harlan et al. (1970) and Wu and Taliaferro (2009).

In contrast Hunter (1934), Hurcombe (1947), Moffett and Hurcombe (1948) and Rochecouste (1962) recorded different chromosome number such as 2n = 40 for this species with probable basic number x = 10. However, information on aneuploidy of this species is insufficient and it seems like that real differences in chromosome number along with variation might be related to ecological condition. Parvin (2002) reported aneuploidy in *C. dactylon*. Thus, the goal of this study was to discriminate the different accessions of *C. dactylon* collected from different ecological zones of Bangladesh.

There are two types of structural organization in plant cell nuclei and they are namely chromocentric and reticulate (Lafontaine 1974). Interphase nuclear phenotype of plants is species specific and indicates several important features regarding chromosomal arrangement (Patankar and Raujekar 1984). Its reasons are unknown although the role of nuclear DNA content and repetitive DNA sequence has been suggested to be associated with those features (Lafontaine 1974, Nagl and Fusenig 1979). They also suggested that chromocentric nuclear organization is assumed to be governed by small size of chromosomes and low DNA content. As chromocenters correspond to heterochromatin (Nagl and Fusenig 1979), percentage of heterochromatin values may be obtained by determining the area of nucleus and of chromocenters by planimetry (Kabir and Singh 1989). Keeping these points in mind interphase nuclear phenotype of *C. dactylon* accessions have been determined in this study along with interphase chromosome volume.

Materials and Methods

Cynodon dactylon were used as experimental material in the present study and at least ten plants of each accession these were collected from 19 different districts of Bangladesh. The materials were grouped into four different groups based on their growing area (habitat) as North, West, East and South zones and these were transplanted in the experimentation field of the Institute of Biological Science, University of Rajshahi, Bangladesh.

When these were grown well in the experimental field, their root tips (approximately 1-1.5 cm in length) were treated with saturated solution of paradichlorobenzene at 10°C in the refrigerator for 4 - 4.5 hrs. Then the root tips were washed with distilled water and fixed in 1:3 aceto-alchohol for 48 hrs at room temperature. After fixation they were washed again with distilled water, preserved in 70% ethanol and kept in the refrigerator for further uses. For cytological study, the root tip cells were stained with 0.5% haematoxylin according to the method of Haque et al. (1976). Stained meristematic zone was taken on a clean slide, squashed with a drop of 0.5% acetocarmine and a gentle pressure was applied by thumb over the cover glass. For chromosome counting, photomicrography was made from metaphase plate using 40 × 10 magnification and at the same time for clear visualization camera lucida drawings were made. For measuring Nuclear Volume (NV) from interphase, occulometer was used and the values were converted in micron (μ) with the help of stage micrometer, using the formula, $NV = 4/3\ \pi r^3$ (Nayar et al. 1971). These values divided by somatic chromosome number gave the interphase chromosome volume. Chromocenters were counted and heterochromocentric values were determined by planimetry (add reference and mention the equation).

Results

All the accessions of *C. dactylon* studied in the present experiment showed chromocentric nuclear organization. The chromocenters were seen distinctly on a very light background of the microscope and a few of the representative plates of interphase showing chromocentric structure are presented in Fig. A (1-9). The numbers of chromocenters for each accession are not corresponding with their somatic chromosome number. Sometimes it was found to be significantly less and obviously not more than their respective chromosome number. Percentage of heterochromatin values were recorded by determining the area of the interphase nucleus and of chromocenters by planimetry and the heterochromatin values were expressed as percent per nuclear area. Nuclear volume as well as interphase chromosome volume along with their somatic chromosome numbers, number of chromocenters and heterochromatin percentage is given in Table 1. The somatic chromosome numbers were found to vary from 12 to 40. For confirmation, camera lucida drawings of metaphase chromosomes of all the accessions of *C. dactylon* collected from different districts were made very keenly and these are shown in Fig. B (10-28). In the present study, eight among 19 accessions were diploid with 2n=18 chromosomes. Only one accession collected from Naogaon was found to be tetraploid with 2n = 4x = 36 and rest of these were aneuploid showing the chromosome numbers 12, 14, 16, 22, 24, 26, 32, 40, etc.

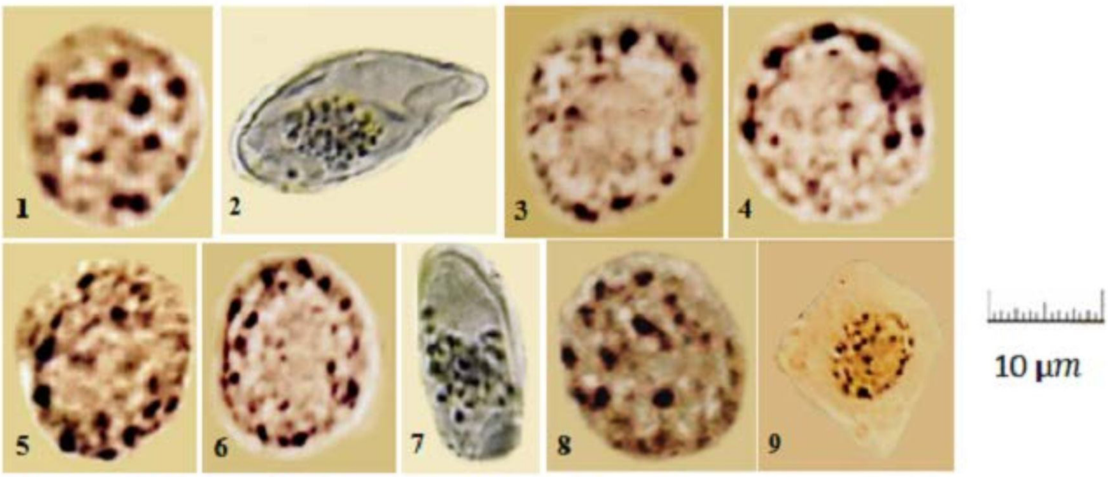

Fig. A (1-9): Representative figures of interphase showing chromocentric structure of nucleus and chromocenters in nine accessions of *Cynodon dactylon* collected from different habitats. 1 = Lalmonirhat, 2 = Gazipur, 3 = Thakurgaon, 4 = Shariatpur, 5 = Barguna, 6 = Naogaon, 7 = Jhenaidah, 8 = Narshingdi, 9 = Maymensingh.

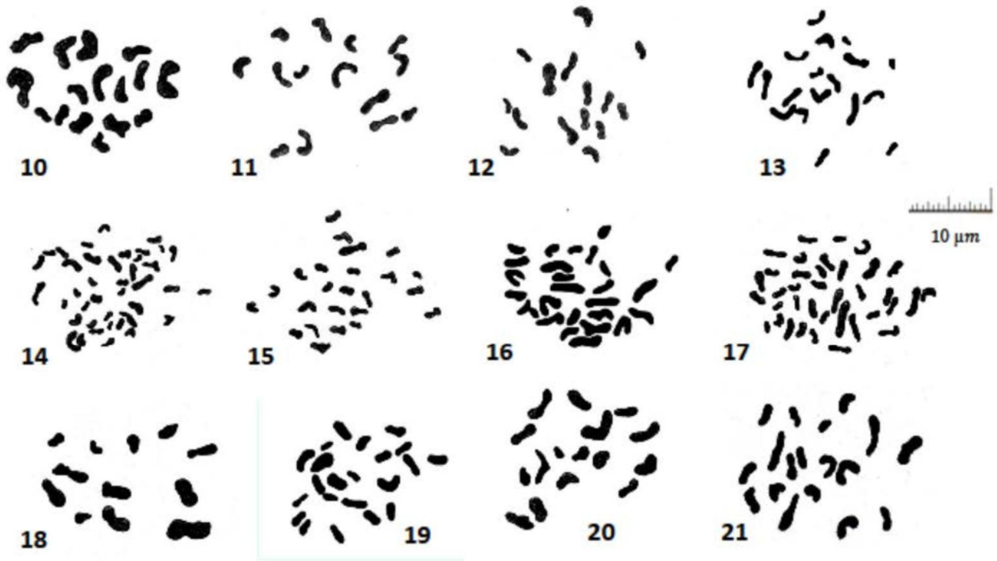

Fig. B (10-21): Camera-lucida drawings of respective chromosome number in 12 accessions of *Cynodon dactylon* collected from four different zones of Bangladesh. 10 = Rangpur (2n = 18), 11 = Lalmonirhat, Aneuploidy (14), 12 = Dinajpur (2n = 18), 13 = Thakurgaon (2n = 18), 14 = Panchagarh, Aneuploidy (40), 15 = Gaibandha, Aneuploidy (24), 16 = Rajshahi, Aneuploidy (26), 17 = Naogaon (2n = 4x = 36), 18 = Pabna, Aneuploidy (12), 19 = Gazipur, Aneuploidy (22), 20 = Narshingdi, Aneuploidy (16), 21 = Sherpur (2n = 18).

Contd. Fig. B

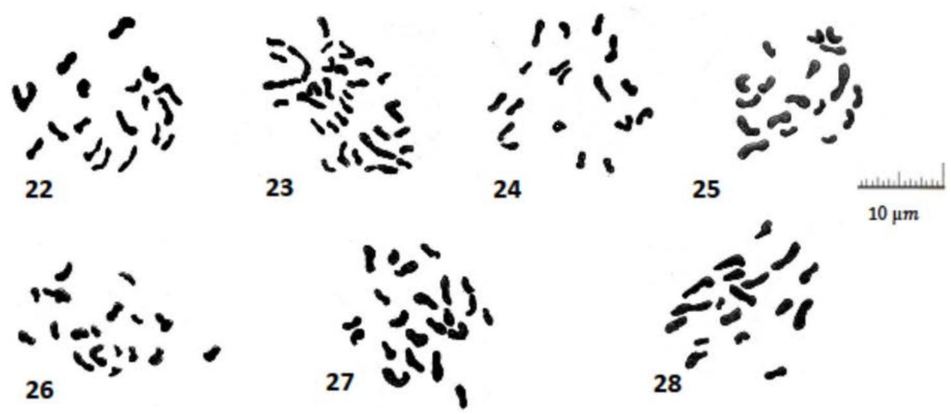

Fig. B (22-28): Camera-lucida drawings of respective chromosome number in 07 accessions of *Cynodon dactylon* collected from four different zones of Bangladesh. 22 = Maymensingh, Aneuploidy (32), 23 = Khulna, Aneuploidy (32), 24 = Jessore (2n = 18), 25 = Jhenaidah (2n = 18), 26 = Faridpur (2n = 18), 27 = Shariaetpur, Aneuploidy (22), 28 = Barguna (2n = 18)

Table 1. Number of chromosome, nuclear volume, interphase chromosome volume, number of chromocentres and heterochromatin percentage per nuclear area in different accessions of *Cynodon dactylon* collected from different districts under different zones of Bangladesh

Zone	Accessions number	Name of the districts	Number of Chromosome	Nuclear Volume (INV) M ± SE (μm^3)	Interphase Chromosome Volume (ICV) M ± SE (μm^3)	Number of Chromocentres M ± SE	Heterochromatin percentage per nuclear area M ± SE
North	1.	Rangpur	18*	2.807±0.102	0.156±0.006	14±0.566	28.675±1.627
	2.	Lalmonirhat	14**	12.077±0.093	0.863±0.007	14±0.632	51.037±5.982
	3.	Dinajpur	18 *	14.354±0.089	0.797±0.005	16.4±0.669	25.548±1.272
	4.	Thakurgaon	18*	2.706±0.117	0.150±0.006	15.2±1.073	46.661±3.738
	5.	Panchagarh	40**	9.134±0.152	0.228±0.004	16.4±0.877	57.955±6.537
	6.	Gaibandha	24**	0.674±0.084	0.028±0.003	16.6±.358	29.461±3.547
West	7.	Rajshahi	26**	13.996±0.154	0.538±0.006	17±0.283	29.455±2.044
	8.	Naogaon	36***	3.578±0.066	0.099±0.002	16.8±0.438	24.389±1.124
	9.	Pabna	12**	4.620±0.088	0.385±0.007	11.4±.219	45.185±7.983
East	10.	Gazipur	22**	41.921±0.180	1.905±0.008	17.6±0.3578	39.793±2.242
	11.	Narshingdi	16**	17.238±0.295	1.077±0.018	15.2±.715	33.82±6.588
	12.	Sherpur	18*	5.209±0.093	0.289±0.005	17.6±0.358	38.706±5.132
	13.	Mymensingh	20**	0.862±0.084	0.043±0.004	13±0.707	41.782±1.033
South	14.	Khulna	32**	5.057±0.09	0.158±0.003	16±0.80	30.048±5.094
	15.	Jessore	18*	5.058±0.171	0.281±0.004	16.4±1.043	47.653±1.411
	16.	Jhenaidah	18*	11.811±0.150	0.656±0.008	17.40±.358	19.759±2.714
	17.	Faridpur	18*	1.348±0.105	0.075±0.006	17.6±0.358	30.304±5.303
	18.	Shariatpur	22**	13.183±0.089	0.599±0.004	14.4±1.431	66.022±5.481
	19.	Barguna	18*	29.886±0.097	1.660±0.005	14.4±1.189	65.286±7.938

*= Diploid chromosome number, **= Aneuploid, ***= Tetraploid

Discussion

In the present experiment, interphase nuclei of different accessions of *C. dactylon* were found to be chromocentric. Joshi and Ranjekar (1983) and Patankar and Ranjekar (1984) found similar type of chromocentric nuclear organization in *Phaseolus* species. The number of chromocenters was always less compared to that of their chromosome numbers. The values ranged from 11.4 (Acc.9) to 17.60 (Acc.10, Acc.12 and Acc.17). The chromocentric number did not correlate with the somatic chromosome numbers of the diploid, tetraploid or aneuploid accession. Kabir (1993) reported more or less same number of chromocenter in six *Crotalaria* species. The reduction in number of chromocenters might be due to fusion or overlapping of chromocenters indicating the somatic association of chromosomes (Kabir and Singh 1989). Dayal and Prasad (1983) stated that the number of chromocenters is considered to be controlled genetically and is, therefore a species specific character.

In case of heterochromatin percentage per nuclear area the values obtained in this study were highly variable. The values ranged from 19.759% (Acc.16) to 66.022 (Acc.18). In this study, higher percentage of heterochromatin might be due to the presence of different classes of heterochromatin (Vosa1970 and La Cour 1978). HCl-Giemsa banding is also known to visualize most of the types of heterochromatin on chromosome (Joshi and Ranjekar 1982). The variable result in the present investigation might be due to the fact that HCl-haematoxylin banding revealed more than one class of heterochromatin in interphase chromosomes. Lafontaine (1974), Nagl and Fusenig (1979) stated that chromocentric nuclear organization is assumed to be governed by small size of chromosomes and low DNA content. This holds partially true for all the accessions of *C. dactylon* which showed very small size of chromosomes.

Yamakawa and Sparrow (1965 and 1966) studied interphase chromosome volume as a reliable index of radio sensitivity in plants and their work established a positive linear correlation between radiation sensitivity and chromosome volume in plant cells. In present study this parameter (interphase chromosome volume) was considered since all the accessions of *C. dactylon* were collect from different ecological conditions, where the temperature as well as radiation index may vary from one zone to another. In this study, the interphase chromosome volume (1.905 μm^3) was found in the accession collected from Gazipur and the lowest value (0.028 μm^3) was found in the accession collected from Gaibandha. This character was not correlated with chromosome number. Thus, it may be said that increase or decrease of interphase chromosome volume may be caused due to alteration of cell membrane configurations, modification of chromosomal volume and changes in sensitivity to physical or chemical factors, functioning always in the environment. Yamakawa and Sparow (1966) reported increase in mutation rate per roentgen in higher plants and that was highly correlated with increase in both ICV (interphase chromosome volume) and DNA content per chromosome.

In present study the most striking feature of *C. dactylon* was found in case of chromosome number. Range of chromosome number was really surprising when the basic chromosome number is considered to be x = 9 (Forbes and Burton 1963, Clayton and Harlan 1970). In this experiment only 08 (eight) accessions out of 19 were found to be diploid (2n = 18). Only one was found to be tetraploid and rest of them were aneuploid. De

Silva and Snaydon (1995) stated that most populations contained only tetraploid plants (2n = 36), but populations from roadsides and lawns in the wet region and from forest in the hill country contained only diploids plants (2n = 18). They also said that soil acidity seems to be the main factor determining the distributions of the cytotypes. They did not report any triploid or any aneuploid. On the contrary Dhaliwal et al. (2018) reported *Cynodon dactylon* with the basic chromosome number of n = 8, while few other reports on five different cytotypes with basic chromosome number n = 8, n = 9 and n = 18 have been reported in his literature based on works of several investigators (Gupta and Srivastava 1970, Kumar and Sachdeva 1988, Malik and Tripathi 1968, Sachdeva and Bhatia 1980, Sinha et al. 1990). However, the present findings may be supported by Federov (1974), Christopher and Abraham (1974), Brown (1950) and Hurcombe (1947). Gupta and Shrivastava (1970) reported one diploid (2n = 18) and three triploid (2n = 27) collections of *C. dactylon*. Chiavegatto et al. (2016) aimed to determine the karyotype asymmetry index among accessions of *Cynodon* to discriminate them. They ultimately reported two diploid, one triploid, four tetraploid and one pentaploid accession. Their findings indicated predominant level of ploidy for the tetraploid accessions. Along with their findings on chromosome counts, it may be said that there is a wide genetic variability which is very important for origin, evolution and development of plant species. The preponderance of aneuploidy observed in the present study throws light on the evolutionary tendencies which might go hand to hand in the phylogenetic evolution due their different adaptive values. When one of the characters might put a type or accession of *C. dactylon* at highest level of advancement then the others may be still in original stage. However, aneuploids play an important role in the breeding programme and thus, the main objective of forage breeding programme may be an important way to obtain those genotypes with desirable agronomic traits.

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Morphological characteristics of different accessions of *Cynodon dactylon* (L.) Pers. and physico-chemical properties of soil of their growing region in Bangladesh

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Abstract

Distribution as well as adaptation of *Cynodon dactylon* to different ecological zones of Bangladesh is mainly dependant on weather and adaphic factors of their respective habitats. Generally, it is a warm season perennial grass species that initiates growth in the vernal season and its growth continues rapidly when moisture is adequate and they find the alkaline clay soil as their habitat. From that point of view, this study was aimed at analysing the morphological variations of *Cynodon dactylon* ecotypes along with determining the physicochemical properties of soils from their particular habitats. A total of 19 ecotypes /accessions from four different zones of Bangladesh were collected along with the habitat soil and all of them were transplanted in experimentation field of Institute of Biological Sciences, University of Rajshahi, Bangladesh. All the accessions were established in 1.5 × 1.4 m plots separately. At maturity just after started flowering, the morphological data on 26 characters were recorded quantitatively and those were analysed statistically. Rhizomatous and stoloniferous nature was observed from randomly selected areas within the plot. Both significant and non-significant variations were found among the morphological characters. Most of the vegetative and reproductive characters were found to show significant variations among the accessions. In addition, the values obtained on physio-chemical properties of soil were tabulated and their magnitude were determined and interpreted following Fertilization Recommendation Guide. In respect of soil properties variation on morphological parameters of almost of all the accessions were observed. In this study, all the accessions were found to be adapted nicely in alkaline soil of the experimentation field and that might be due to their many physiological and biochemical mechanisms.

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Introduction

Cynodon dactylon (L.) Pers is a prostrate and perennial grass. It spreads by scaly rhizomes and flat stolons, and widely naturalized in tropical and subtropical regions of the world. This plant species is a C_4 grass included in the Global Compendium of Weeds (Randall, 2012) and it is listed as one of the most serious agricultural and environmental weeds in the world (Holm *et al.*, 1977).

This grass species has different vernacular names. In Bengali, it is known as Durva, Dub, Dubla, Durba, Doorva, Neel Doorva (Asthana *et al.* 2012). In English it is termed as Couch grass, Bahama grass, Bermuda grass, Dun grass, Doab grass etc. The common name for all the East African rhizomatous species of *Cynodon* is Bermuda grass (Harlan, 1970; Burton and Hanna, 1985). *Cynodon dactylon* however, has become a ubiquitous cosmopolitan weed (Harlan and de Wet, 1969).

Cynodon dactylon can tolerate wide range of soil types and their growth becomes healthy on heavy clay soil than on light sandy soil in dry regions and this might be due to water holding capacity of clay (Burton and Hanna ,1985). However, alkaline soils are tolerated nicely by this grass species than acidic ones. Drought conditions, high temperature, intensive sunlight and alkaline soils are found to be best requirements for healthy growth of this grass. *Cynodon dactylon* shows direct competition for space and nutrients by its rapid growing ability and adaptive capacity.

Cynodon dactylon is found with underground rhizomes and on ground runners (Cabrera, 1968; Covas and Salavi,1970).These rhizomes penetrate to the soil of a depth of above 30 cm(Perez and Labrada,1985; Philips and Moaisi,1993). The rhizomes may be twice as wide as the runners and this is found to be one of the variable characters in their populations. Runner or rhizome nodes generally bear up to three viable buds. Leaves show alternate distal pattern of distribution along the runners. Leaf blades are unhaired but the ligule is found with a conspicuous fringe of white hairs. Leaf blades are green to dull green, lanceolate, finely parallel-ribbed on both the surfaces, without midrib (Rosengurt *et al.*, 1960). The inflorescence is supported on a culm and consists of a single whorl of 3-7 racemes. Spikelets are closely appressed to the rachis. Glumes are one nerved, lemma is silky pubscent on the keel and palea is glabrous. Caryopses are sub-eliptical, compressed and brownish in colour (Kissmann, 1991).

Cynodon dactylon has some sort of social impact, as its pollen has been found to cause allergic symtoms in asthmatics in Malaysia (Sam-Choonkook *et al.*, 1998) and Brazil (Kissmann, 1991). The use of *Cynodon dactylon* in different religious ceremonies has been studied by Dubey *et al.* (2000).This grass species is used medicinal plant and antioxidant of *C. dactylon* used for management of neurodegenerative diseases has been studied extensively by Auddy *et al.* (2003). This plant species is used as pasture grass in many countries, since it contains many soils of chemical components like proteins, carbohydrates, mineral constituents phosphorous, calcium, potassium, vitamin C, carotene, fats, palmitic acid etc. Paranjpe (2001) reported a good amount of crude protein,fiber and total ash.

Cynodon species have been classed as noxious weeds, especially *C. dactylon*, which is very invasive (Fernaudez, 2003). This species is suitable for erosion control and soil stabilization. But mechanical tillage of weed-infested fields makes an effect on fragmentation and dispersal of stolon and rhizome propagules, and thereafter infestation is increased.

However, Van De Wouw *et al.* (2009) stated that, the International livestock Research Institute (ILRD) collected some commercial cultivars and accessions of *Cynodon* species from wild habitats. These collections cover four different species of *Cynodonviz.C. aethiopicus*, *C. dactylon*, *C. incompletus* and *C. nlemfuensis* along with hybrids of *C. dactylon* and *C. transvaalensis*. The concerned investigators determined the amount of morphological variation present in the collections, based on what the users can select the appropriate accessions for their further experiment. The objective of the present study was to make an analysis of morphological variation of *C. dactylon* ecotypes from botanic as well as agronomic points of view, and their determination of physico-chemical properties of their habitat soils.

Material and methods

Study sites

A total of 19 ecotypes of *Cynodon dactylon* were collected from 04(four) different zones of Bangladesh. Selection was based on environmental conditions of the collection site. The selective districts were presented in Table 1.

The study was carried out at the research field of IBSc, Institute of Rajshahi University. The site has an annual average rainfall of 661.2 mm of which the majority falls during the main rainy season (July-September). No fertilizers were used in the experimentation field. The soil at the research field is loamy soil. All (19) the accessions of *C. dactylon* were established from root splits in 1.5m ×1.4m plots, collected from 19 habitats belonging to 04(four) zones. Data were recordedon 26 characters. The characters were selected for their agronomic/taxonomic relevance and expected variation among accessions. The morphological characters were observed within the first 4 months just after cutting before most accessions had started flowering. Owing to the rhizomatous and stoloniferous nature of *Cynodon dactylon* it was difficult to identify individual plants. So, observations were made randomly within the plot. Six randomly selected areas of 25cm ×25 cm in the plot were chosen for rhizome observations and six randomly selected areas of 200cm×50cm at the end of the plot for stolon observations. Plants were removed from three areas at the time of cutting and after 7 weeks the numbers of rhizomes or stolon in the selected areas were counted. The collected ecotypes were grown under same environmental conditions at research field of IBSc Institute of Rajshahi University, till their acclimatization to evaluate genetically fixed characteristics during long evolutionary history. Plants were uprooted carefully and washed with distilled water. For dry weight, plant samples were oven-dried at 65°c for 48 hours and constant dry weight was achieved. Data were recorded of the following plant parts and their mean values were tabulated (Table 2).

For determination of the leaf area single segment of leaf was cut off with the help of a sharp knife, weighted after oven -drying and then it was calculated by using the following formula:

$$\text{Leaf area} = \frac{\text{Area of segment} \times \text{weight of leaves}}{\text{Weight of segment}}$$

Analysis of soil samples

Soil from rhizosphere was taken from each habitat to analyze the physico-chemical characteristics at 16 cm depth. The soil extract was used to determine the pH, organic matters %, potassium(K^+), Calcium (Ca^{2+}), Magnesium (Mg^{2+}) total nitrogen, phosphorus(P), Sulfur (S), Boron(B), Copper (Cu), Iron (Fe), Manganese (Mn) and Zink (Zn) of soil.The19 different soil samples were collected from 19 different habitats. Then collected soil was air-dried for 7 days and then sieved using a 2mm mesh to remove debris. Air-dried and sieved soil (250gm) was used for soil analysis. Soil pH was measured by HANNA HI 92210N ATC pH Meter. Total organic matter was determined following the wet oxidation method as described by Walkley and Black (1934) and the micro Kjeldahi procedure was used for the determination of total nitrogen (Bremnerv and Mulvaney, 1982). Available phosphorus, sulfur and boron were measured by using spectrophotometer at 890nm.Magnesium, Calcium, Manganese, Copper, Iron and Zinc were obtained using atomic absorption spectrophotometer (Perkin- Elmer Model 403, Shelton, Connecticut, USA).

Available Potassium was measured by using a flame photometer. Phosphorus was determined following the Olsen method [pH = 6.6 – 7.3 (Neutral),6.5>, (Acidic)7.4<(Alkaline) and Organic matter (OM) =below 1- Very low, 1-1.7 –Low, 1.8-3.4 -Medium, 3.4<High].

A change of pH and oxidation state can increase or decrease the potential bioavailability of metals in soil. Metals such as Zn, Cu, and Mn are essential for living organisms at low concentrations, but became toxic of increasing concentrations. The soil test values were interpreted based on critical limits following Fertilization Recommendation Guide (Table 3).

Statistical analysis

A statistical comparison of means of different accessions and their characters was carried out using analysis of variance (ANOVA) followed by Dunkan's Multiple Range Test (DMRT). Significance level was set at $p<0.05$. The data analysis was done busing SPSS version 20.0 for Windows. Graphs were drawn by Excel software.

Results

The accessions showed variation in many of the morphological characters (Tables 4-8). Shoot number, shoot length, node number, internode length and fresh and dry weight of shoot were found with higher values in the accessions collected from Lalmonirhat, Rajshahi, Gazipur and Thakurgaon, respectively (Table 4). On the contrary, lower values of these characters were found in the accessions collected from Panchagarh, Dinajpur, Panchagarh and Dinajpur again, Dinajpur and Shariatpur, respectively. Regarding shoot characters the variations are also shown based on geographical zones (Fig. 1).

Table 1. Nineteen districts considered as habitat of the accessions of *C. dactylon* under four different zones of Bangladesh.

North zone	West zone	East zone	South zone
Rangpur	Rajshahi	Gazipur	Khulna
Lalmonirhut	Naogaon	Narshingdi	Jessore
Dinajpur	Pabna	Sherpur	Jhenaidah
Thakurgaon		Mymensingh	Faridpur
Panchagarh			Shariatpur
Gaibandha			Barguna

Table 2. Twenty six different quantitative characters of each accession considered for recording both vegetative and reproductive attributes.

1	Number of shoots/plant	14	Total leaves fresh weight/ plant(g)
2	Shoot length(cm)	15	Shoot dry weight/plant(g)
3	Number of nodes/plant	16	Root dry weight /plant(g)
4	Internode length(cm)	17	Total leaves dry weight /plant(g)
5	Root length	18	Inflorescence number /plant
6	Number of roots/plant (cm)	19	Length of racemes (cm)
7	Number of leaves/plant	20	No. of racemes /plant
8	Single leaf length(cm)	21	Seed number /shoot
9	Single leaf width(mm)	22	Seed number /plant
10	Total leaf area/plant(cm2)	23	Maximum height(cm)
11	Number of rhizomes /plant	24	Minimum height(cm)
12	Shoot fresh weight /plant (g)	25	Number of stolons
13	Root fresh weight/plant(g)	26	Stolon length

Table 5 reveals higher values for root number, root length, fresh and dry weight of root and rhizome number in case of the accessions collected from Khulna, Faridpur, Rajshahi and Khulna, respectively. Lower values were obtained for root number in accessions of Gaibandha, for root length in case of Rajshahi, fresh weight in case of Shariatpur, dry weight in case of Gazipur and for rhizome number in the accession collected from Sherpur.

The variations in root characters are shown in Fig. 2 based on zone (Fig. 2).

Table 2. Morphological variation of 19 accessions of *Cynodon dactylon* shoot collected from different habitats of Bangladesh.

Zone (Bangladesh)	Name of District	Parameters of shoot (Mean)						
		Number	Length (cm)	No. of node	Internode length (cm)	Colour	Fresh weight (gm)	Dry weight (gm)
North	Rangpur	8.50abcdef	14.72cd	6.00ab	2.52a	Green	4.31bcd	1.4b
	Lalmonirhat	*12.00a*	16.13bc	4.50b	2.73a	Green	4.83bcd	1.82ab
	Dinajpur	7.67cdef	10.66f	4.17b	1.44a	Green	2.95d	1.05b
	Thakurgaon	6.83def	19.45ab	4.83b	2.80a	Green	8.11a	3.48a
	Panchagarh	5.67f	13.92cdef	3.83b	3.23a	Green	3.27cd	1.12b
	Gaibandha	8.17bcdef	14.36cde	4.50b	2.72a	Green, Gray	5.89abc	1.91ab
West	Rajshahi	11.83ab	20.02a	9.17a	2.77a	Green	3.99bcd	1.91ab
	Naogaon	9.33abcdef	14.66cde	4.17b	2.28a	Green	4.14bcd	1.60b
	Pabna	9.50abcde	17.11abc	5.67b	2.12a	Green	5.30bcd	2.01ab
East	Gazipur	6.50def	16.41bc	4.83b	4.13a	Green	4.20bcd	1.29b
	Narshingdi	8.50abcdef	12.48def	4.17b	2.23a	Green	5.86abc	1.94ab
	Sherpur	8.50abcdef	17.42abc	6.67ab	3.48a	Green	5.10cd	1.69ab
	Mymensingh	9.00abcdef	16.12bc	6.17ab	2.73a	Green, Gray	6.64ab	2.65ab
South	Khulna	7.67cdef	15.72cd	5.50b	2.42a	Green	4.64bcd	1.75ab
	Jessore	6.67def	15.56cd	4.67b	2.08a	Brown	3.79bcd	1.65b
	Jhenaidah	5.83ef	16.15bc	5.33b	2.78a	Green	4.58bcd	1.44b
	Faridpur	10.67abc	16.33bc	4.17b	2.15a	Green	3.11cd	1.1b
	Shariatpur	8.50abcdef	12.03ef	5.23b	2.50a	Green	2.57d	0.81b
	Barguna	9.67abcd	15.88cd	5.83ab	3.32a	Green	6.17ab	2.13ab

In case of flower characters, spikelet colour was found to vary (Table 6). Highest value for number of inflorescences was found in accession collected from Mymensingh and lowest in that of Panchagarh. Highest and lowest values for number of racemes were found in the accessions collected from Rajshahi and Lalmonirhat respectively. For length of racemes highest and lowest values were found in the accessions of Gazipur, Khulna and Rajshahi respectively. Number of seeds per shoot and per plant was found to be highest and lowest in the accessions collected from Jessore and Gaibandha, and from Faridpur and Barguna, subsequently. Fig. 3 reveals that the number of seeds per shoot and per plant were highest in accessions collected from east and south zone. Figure 4 also shows similar results with a little variation in case of inflorescence and raceme characters.

In case of leaf characters (Table 7) number of leaves, leaf width, total leaf area and leaves dry weight were found with highest values in the accession collected from Rajshahi. On the other hand, lowest values for these characters were found in that case of Faridpur, Sherpur, Jessore and Jessore again, successively.

Values for leaf length and leaves fresh weight were highest for the accession of Faridpur and Panchagarh, lowest values were found for these characters in case of Pabna and Jessore, respectively.

Table 3. Morphological variation of 19 accessions of *Cynodon dactylon* root collected from different habitats of Bangladesh.

Zone (Bangladesh)	Name of District	Parameters of Root (Mean)				
		Number of Root	Length (cm)	Fresh weight (gm)	Dry weight (gm)	Number of Rhizome
North	Rangpur	14.17abcd	10.77ab	1.93b	0.71b	4.00def
	Lalmonirhat	15.00abc	10.25ab	1.60b	0.76b	4.33def
	Dinajpur	9.50de	8.50abc	1.28b	0.58b	9.00ab
	Thakurgaon	14.17abcd	11.10ab	2.29b	0.85b	9.17ab
	Panchagarh	9.67de	8.92abc	1.71b	0.72b	3.33ef
	Gaibandha	8.00e	6.65bc	2.08b	0.82b	5.20bcdef
	Rajshahi	14.83abc	4.92c	4.61a	2.37a	7.50bcd
West	Naogaon	11.50bcde	11.05ab	1.73b	0.63b	7.33bcde
	Pabna	11.00cde	8.50abc	1.69b	0.73b	8.83ab
East	Gazipur	13.50abcd	10.25ab	1.22b	0.47b	4.00def
	Narshingdi	15.00abc	6.98bc	1.30b	0.62b	6.33bcdef
	Sherpur	16.00ab	9.30abc	1.63b	0.67b	3.00f
	Mymensingh	11.50bcde	10.68ab	1.48b	0.64b	4.17def
South	Khulna	16.67a	8.92abc	1.61b	0.59b	11.50a
	Jessore	13.50abcd	6.88bc	1.25b	0.53b	5.33bcdef
	Jhenaidah	12.83abcde	10.00abc	1.80b	0.49b	4.50cdef
	Faridpur	11.33bcde	12.77a	1.49b	0.61b	8.33abc
	Shariatpur	11.17bcde	11.48ab	0.91b	0.61b	5.50bcdef
	Barguna	10.67cde	10.53ab	2.15b	0.90b	5.33bcdef

Table 8 reveals plant height (maximum and minimum), and number of stolons per plant and stolon length. Plant height was found with maximum and minimum values in accession of Thakurgaon. Value for stolon per plant was found to be highest in accession collected from Mymensingh and lowest value was found for stolon length in the accession of Khulna. Growth habit of all the 19 accessions showed erect and prostrated nature based an angle of stem to ground.

Table 4. Morphological variation of 19 accessions of *Cynodon dactylon* flower and seed collected from different habitats of Bangladesh.

Zone (Bangladesh)	Name of District	Parameters of Flower (Mean)				Parameters of Seed (Mean)	
		No. of Inflorescence /plant	No. of racemes	Length of racemes (cm)	Spikelet colour	No. of Seeds/ Shoot	No. of Seeds/ plant
North	Rangpur	3.6ab	4.02a	4.00a	Brown	90.40de	325.60f
	Lalmonirhat	5.0ab	3.73a	3.42a	Green, Brown, Dark Brown	73.60hi	338.80e
	Dinajpur	3.6ab	3.88a	3.07a	Light Green, Brown	73.40hi	264.00i
	Thakurgaon	4.8ab	4.18a	3.54a	Brown, Light Green	77.48h	319.80g
	Panchagarh	2.0b	4.10a	4.27a	Brown	88.90ef	177.80l
	Gaibandha	2.8b	3.80a	3.60a	Brown	66.10k	182.20l
West	Rajshahi	3.8ab	5.95a	2.77a	Green, Brown	88.28efg	318.60g
	Naogaon	3.0b	4.05a	3.67a	Brown	85.44efg	244.00j
	Pabna	4.0ab	3.92a	3.17a	Light Green, Brown	86.39efg	325.00f
East	Gazipur	2.8b	3.93a	4.47a	Brown	83.70fg	214.60k
	Narshingdi	3.0b	4.25a	3.84a	Brown, Greenish Brown, Dark Brown	99.06c	290.60h
	Sherpur	5.6ab	4.41a	3.30a	Brown, Light Green, Dark Brown	83.51fg	470.80c
	Mymensingh	7.2a	4.12a	3.03a	Light Green, Brown	71.53i	525.40b
South	Khulna	2.6b	4.00a	4.47a	Brown	95.28cd	247.20j
	Jessore	3.6ab	4.01a	3.93a	Brown, Light Brown, Light Green	113.72a	392.00d
	Jhenaidah	2.8b	4.40a	4.24a	Brown, Dark Brown, Light Green	105.70b	259.00i
	Faridpur	5.0ab	4.51a	3.90a	Brown, Light Green	109.32ab	537.80a
	Shariatpur	4.8ab	4.04a	2.98a	Brown, Light Green	82.96g	393.80d
	Barguna	2.6b	4.27a	3.26a	Brown, Green, Light Green	68.10ij	172.40m

Among all the accessions only 08(eight) accessions showedtheir both the growth habit in the ratio of 1:1 or 2:1, and rest of them were either erect or prostrated. Variation in plant height and stolon characters are visualized in Fig. 7.

The ANOVA analysis for shoot and root characters showed the significant variations (Table 9) for number of shoots, shoot length, fresh weight of shoot, number of root and number of rhizomes.

Table 5. Morphological variation of 19 accessions of *Cynodon dactylon* leaf collected from different habitats of Bangladesh.

Zone (Bangladesh)	Name of Dist.	Parameters of Leaf (Mean)						
		No. of Leaves/ plant	Leaf length (cm) (single)	Leaf width (mm) (single)	Total Leaf Area (cm^2)	Total leaves fresh weight /plant(gm)	Total leaves dry weight/ plant(gm)	Leaf Nature (soft/ stiff)
North	Rangpur	22.67fg	4.78a	1.82a	224.25g	1.73a	0.39a	Soft
	Lalmonirhat	39.50b	4.90a	2.00a	242.45e	2.24a	0.37a	Soft
	Dinajpur	20.00gh	4.77a	2.03a	200.10i	1.53a	0.31a	Stiff
	Thakurgaon	23.50defg	5.68a	2.07a	154.30m	1.61a	0.24a	Soft
	Panchagarh	27.83cd	4.73a	2.30a	346.60b	2.49a	0.48a	Soft
	Gaibandha	27.67cde	4.33a	2.02a	161.12l	1.89a	0.31a	Soft
West	Rajshahi	80.33a	5.40a	2.67a	414.70a	2.39a	0.51a	Soft
	Naogaon	13.50j	4.82a	2.28a	345.40b	2.33a	0.49a	Soft
	Pabna	15.67ij	3.75a	2.02a	249.50d	1.79a	0.45a	Soft
East	Gazipur	25.17cdef	4.72a	2.02a	188.50k	1.69a	0.30a	Soft
	Narshingdi	19.33ghi	4.88a	2.03a	238.35f	1.61a	0.38a	Soft
	Sherpur	25.33cdef	5.23a	1.58a	211.15h	1.70a	0.42a	Soft
	Mymensingh	17.17hij	5.05a	2.20a	196.20j	1.52a	0.33a	Soft
South	Khulna	27.33cde	5.37a	2.00a	280.65c	1 .70a	0.46a	Soft
	Jessore	23.17efg	4.48a	1.77a	73.02p	1.09a	0.16a	Soft
	Jhenaidah	25.67cdef	5.13a	1.88a	118.05n	1.17a	0.23a	Soft
	Faridpur	9.50k	6.38a	2.17a	189.55k	2.00a	0.29a	Soft
	Shariatpur	24.67def	5.53a	1.78a	91.35o	1.37a	0.16a	Soft
	Barguna	29.50c	5.47a	2.18a	223.60g	1.91a	0.34a	Soft

Rest of the characters showed non-significant variation among all the accessions collected from different habitats. Again, the ANOVA analysis for leaf plant height and stolon characters (Table 10) revealed significant variation for number of leaves, total leaf area and maximum plant height. Other seven characters showed non-significant variation. In case of inflorescence and seed characters only number of seeds per shoot and plant were found to show significant variation (Table 11). Tables 12a and 12b reveals physico-chemical properties of the soil of 19 original habitats along with the experimentation habitat with values and their magnitude of organic and inorganic matters, respectively. The soil of the experimentation field was found to be loamy. Gangetic alluvium comprises the districts of Jessore, Kushtia, Rajshahi, Faridpur, Khulna, Barisal and Dhaka. In this areas soil texture varies from clay loam to sandy loam and pH ranges from 7.0 to 8.5. The pH value of all the habitats were also determined and its highest value 8.5(alkaline) was found in the soil of Narshingdi, which was somewhat highest than that of experimentation habitat, Rajshahi, Jessore, Jhenaidah and Shariatpur.

Table 6. Morphological variation of 19 accessions of *C. dactylon* plant height, stolon and growth habit collected from different habitats of Bangladesh.

Zone (Bangladesh)	Name of dist.	Parameters of plant height (cm) (Mean)		Parameters of stolon (Mean)		Growth habit: Angle of stem to ground (prostrated/erect)
		Maximum height(cm)	Minimum height(cm)	No. of Stolon/plant	Stolon Length	
North	Rangpur	24.03bcd	16.47ab	7.00a	2.94a	Erect
	Lalmonirhat	26.40abc	16.28ab	5.00a	2.72a	Prostrated
	Dinajpur	17.97f	13.35abc	4.67a	2.77a	Erect
	Thakurgaon	29.13a	17.17a	6.83a	3.30a	Erect : Prostrated 2:1
	Panchagarh	22.23cdef	14.53abc	6.83a	3.21 a	Erect
	Gaibandha	21.33def	10.50c	6.20a	2.54a	Erect : Prostrated 1:1
West	Rajshahi	27.02ab	16.75a	7.17a	3.33a	Prostrated
	Naogaon	24.18bcd	16.12ab	4.33a	2.88a	Erect
	Pabna	23.35bcde	12.63abc	6.00a	2.83a	Erect : Prostrated 1:1
East	Gazipur	26.17abc	13.88abc	6.72 a	3.12a	Erect
	Narshingdi	19.02ef	13.43abc	6.5a	3.28a	Erect
	Sherpur	25.87abcd	13.57abc	4.50a	2.90a	Erect : Prostrated 1:2
	Mymensingh	26.05abcd	16.42ab	9.67a	2.83a	Prostrated
South	Khulna	23.62bcd	15.07abc	4.83a	4.41a	Erect : Prostrated 1:1
	Jessore	21.93cef	11.37bc	6.71a	2.70a	Erect : Prostrated 2:1
	Jhenaidah	25.70abcd	16.83a	4.50a	2.85a	Erect
	Faridpur	25.60abcd	13.83abc	7.00a	2.66a	Erect : Prostrated 1:1
	Shariatpur	22.48bcde	17.15a	6.33a	3.32a	Erect : Prostrated 2:1
	Barguna	23.73bcd	14.93abc	6.33a	3.55a	Erect

Table 7. Analysis of variance (ANOVA) subjected to variation of different parameters of shoot and root in *Cynodon dactylon*on the basis of 19 habitats.

Subject	Data source	Source of variation	DF	Mean SS	F value
Shoot	No of Shoot	Habitat	18	9.677	2.627**
		Error	38	3.684	
	Shoot Length	Habitat	18	15.808	4.683***
		Error	38	3.376	
	No. of Nodes	Habitat	18	4.649	1.350^{NS}
		Error	38	3.444	
	Internode Length	Habitat	18	1.075	0.451^{NS}
		Error	38	2.382	
	Shoot Fresh Weight	Habitat	18	5.840	2.719**
		Error	38	2.148	
	Shoot Dry Weight	Habitat	18	1.125	1.311^{NS}
		Error	38	0.858	
Root	No of Root	Habitat	18	17.150	2.712**
		Error	38	6.323	
	Root Length	Habitat	57	11.639	1.643^{NS}
		Error	18	7.085	
	Root Fresh Weight	Habitat	38	1.771	1.492^{NS}
		Error	57	1.187	
	Root Dry Weight	Habitat	18	0.500	1.522^{NS}
		Error	38	0.329	
	No. of Rhizomes	Habitat	57	16.806	3.967***
		Error	18	4.237	

= significant at $p \le 0.01$, *= significant at $p \le 0.001$ and NS= non-significant.

The acidic habitats were found in case of Lalmonirhat, Thakurgaon, Panchagarh, Gaibandha, Gazipur and Sherpur.Rest of the habitats were somewhat alkaline. The highest values for organic matter, Potassium, Calcium and Magnesium were found in the soil of Faridpur, Dinajpur, Rajshahi and Dinajpur again, respectively. The total nitrogen was found to highest in the soil of Gazipur, somewhat similar to that of Pabna and Faridpur. The highest values for Phosphorus Sulfur, Boron,Copper and Zinc were found in the soil of Pabna. On the other hand, highest amount of Iron and Manganise were found in the soil of Rangpur and Dinajpur, respectively (Fig. 8).

Table 8. Analysis of variance (ANOVA) subjected to variation of different parameters of leaf, plant height and stolon in *Cynodon dactylon*on the basis of 19 habitats.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Leaf	No of Leaf	Habitat	18	641.721	110.598***
		Error	38	5.802	
	Leaf Length	Habitat	18	0.972	0.236NS
		Error	38	4.111	
	Leaf Width	Habitat	18	0.170	0.114NS
		Error	38	1.493	
	Total Leaf Area	Habitat	18	22387.091	4716.439***
		Error	38	4.747	
	Total Leaves FW W/plant	Habitat	18	0.450	0.357NS
		Error	38	1.259	
	Total Leaves DW/plant	Habitat	18	0.033	0.551NS
		Error	38	0.060	
Plant Height	Maximum Height	Habitat	18	22.950	3.938***
		Error	38	5.828	
	Minimum Height	Habitat	18	11.804	1.639NS
		Error	38	7.202	
Stolon	No. of Stolon	Habitat	18	5.110	0.611NS
		Error	38	8.370	
	Stolon Length	Habitat	18	0.553	0.117NS
		Error	38	4.723	

***= significant at p ≤ 0.001 and NS= non-significant.

Table 9. Analysis of variance (ANOVA) subjected to variation of different parameters of inflorescence and seeding *Cynodon dactylon* on the basis of 19 habitats.

Subject	Data source	Source of variation	DF	Mean sum of square	F value
Inflorescence	No. of Inflorescence	Habitat	18	5.038	1.474NS
		Error	38	3.417	
		Total	57		
	No. of Racemes	Habitat	18	0.677	0.136 NS
		Error	38	4.974	
		Total	57		
	Length of Racemes	Habitat	18	0.809	0.192 NS
		Error	38	4.221	
		Total	57		
Seed	Seeds no./ shoot	Habitat	18	552.996	58.247***
		Error	38	9.494	
		Total	57		
	Seeds no./plant	Habitat	18	35661.319	3668.259***
		Error	38	9.722	
		Total	57		

***= significant at p ≤ 0.001 and NS= non-significant.

Considering interpretation of soil test values based on critical limits (Table 3) magnitude of physico-chemical properties (12b) the soil of different habitats considered in the present study were estimated.

Discussion

The present study was conducted to evaluate the morphological (both botanic and agronomic) behaviour of 19 collected native populations of *Cynodon dactylon* from 04 (four) different zones of the country regarding the variations among them. Somewhat similar type of works on *C. dactylon* was carried out by Viggiani *et al.* (2015) in Italy. During the first step of research, they identified 11 sites from six regions of Southern and Central Italy and from these sites they collected 24 ecotypes for determining their habitus and phenology plus some biometrical parameters.

Table 10a. Physicochemical properties of soil obtained through soil analysis of habitats in Bangladesh.

Soil of different habitats	P^H	OM (%)	K	Ca	Mg	Total nitrogen (%)	P	S	B	Cu	Fe	Mn	Zn
			C mol (+)/kg				Microgram/gm						
IBSc Field	8.2	1.77	0.13	34.60	1.70	0.10	11.70	1.50	0.54	1.06	18.0	14.8	1.26
Rangpur	7.2	1.09	0.19	4.37	1.10	0.06	60.0	2.00	0.73	1.55	248.6	12.7	0.71
Lalmonirhat	5.4	2.11	0.34	0.92	0.25	0.12	62.9	3.33	0.28	0.36	137.3	9.6	2.86
Dinajpur	7.1	1.77	1.88	39.10	7.10	0.10	68.6	1.80	0.55	0.86	163.6	45.6	4.84
Thakurgaon	6.5	0.85	0.61	3.20	1.03	0.05	10.3	16.20	0.41	1.25	45.2	17.7	0.52
Panchagarh	6.6	1.71	0.57	33.70	1.83	0.10	20.0	1.50	0.53	0.44	53.4	4.0	0.61
Gaibandha	6.5	1.62	0.11	8.07	2.42	0.09	55.8	7.00	0.43	1.78	190.9	30.6	2.71
Rajshahi	8.1	1.33	0.17	42.90	1.01	0.08	10.0	9.10	0.70	2.67	15.6	6.0	2.95
Naogaon	7.0	1.29	0.62	5.15	1.65	0.08	26.6	1.90	0.58	1.18	91.0	18.6	1.85
Pabna	7.8	2.42	0.40	5.04	1.57	0.14	116.2	57.40	1.07	4.06	86.8	36.9	10.81
Gazipur	6.4	2.73	0.11	7.78	1.89	0.16	13.6	1.61	0.39	1.04	146.7	36.6	8.31
Narshingdi	8.5	0.52	0.10	12.10	0.66	0.03	28.4	28.67	0.52	1.98	24.2	6.8	0.87
Sherpur	6.8	1.55	0.29	5.00	1.98	0.09	67.4	1.44	0.88	1.88	130.2	12.4	1.48
Mymensingh	7.8	1.33	0.24	10.11	2.90	0.08	27.4	23.43	0.41	0.26	87.1	16.9	2.19
Khulna	7.5	2.70	0.53	17.99	5.50	0.16	17.8	33.85	0.52	0.80	59.9	30.0	0.94
Jessore	8.2	0.49	0.10	26.70	4.20	0.03	12.1	5.20	0.73	0.48	12.3	5.6	1.03
Jhenaidah	8.0	1.20	0.30	17.84	2.98	0.07	26.8	2.45	0.38	1.94	18.9	14.0	1.00
Faridpur	7.9	2.43	0.12	5.33	2.34	0.14	21.3	2.00	0.95	1.78	41.6	9.2	10.60
Shariatpur	8.4	1.25	0.28	22.37	2.76	0.07	16.4	5.26	0.29	2.68	31.0	7.5	0.43
Barguna	7.8	0.69	0.62	5.07	1.62	0.04	17.0	41.40	0.70	1.39	40.4	12.6	0.55

OM = Organic matter, K= Potassium, Ca= Calcium, Mg = Magnesium, P = Phosphorus, S = Sulfur, B = Boron, Cu = Copper, Fe = Iron, Mn = Manganese, Zn = Zinc.

Table 10b. Magnitude of physicochemical properties of soil obtained through soil analysis of 19 habitats in Bangladesh.

Soil of different district	P^H	OM (%)	K	Ca	Mg	Total nitrogen (%)	P	S	B	Cu	Fe	Mn	Zn
			C mol (+)/kg				Microgram/gm						
IBSc Field	Alkaline	Low	Low	VH	High	Low	Low	VL	Opt.	VH	VH	VH	Med.
Rangpur	Neutral	Low	Med.	Med.	Med.	VL	VH	VL	High	VH	VH	VH	Low
Lalmonirhat	Acidic	Med.	Opt.	VL	VL	Low	VH	VL	Low	Med.	VH	VH	VH
Dinajpur	Neutral	Low	VH	VH	VH	Low	VH	VL	Opt.	VH	VH	VH	VH
Thakurgaon	Acidic	VL	VH	Med.	Med	VL	Low	Med.	Med.	VH	VH	VH	Low
Panchagarh	Neutral	Low	VH	VH	High	Low	Med.	VL	Opt.	Med.	VH	VH	Low
Gaibandha	Acidic	Low	Low	VH	VH	VL	VH	VL	Med.	VH	VH	VH	VH
Rajshahi	Alkaline	Low	Low	VH	Med	VL	Low	Low	High	VH	VH	VH	VH
Naogaon	Neutral	Low	VH	Opt.	High	VL	Opt.	VL	Opt.	VH	VH	VH	High
Pabna	Alkaline	Med.	High	Opt.	High	Low	VH	VH	VH	VH	VH	VH	VH
Gazipur	Acidic	Med.	Low	VH	VH	Low	Low	VL	Med.	VH	VH	VH	VH
Narshingdi	Alkaline	VL	Low	VH	Low	VL	Opt.	Opt.	Opt.	VH	VH	VH	Low
Sherpur	Neutral	Low	Opt.	Opt.	VH	VL	VH	VL	VH	VH	VH	VH	Opt.
Mymensingh	Alkaline	Low	Med.	VH	VH	VL	Opt.	Opt.	Med.	Low	VH	VH	High
Khulna	Alkaline	Med.	VH	VH	VH	Low	Med.	High	Opt.	VH	VH	VH	Med.
Jessore	Alkaline	VL	Low	VH	VH	VL	Low	VL	High	Opt.	High	VH	Med.
Jhenaidah	Alkaline	Low	Opt.	VH	VH	VL	Opt.	VL	Med.	VH	VH	VH	Med.
Faridpur	Alkaline	Med.	Low	Opt.	VH	Low	Med.	VL	VH	VH	VH	VH	VH
Shariatpur	Alkaline	Low	Opt.	VH	VH	VL	Med.	VL	Low	VH	VH	VH	VL
Barguna	Alkaline	V L	VH	Opt.	High	VL	Med.	VH	High	VH	VH	VH	Low

OM = Organic matter, K = Potassium, Ca = Calcium, Mg = Magnesium, P = Phosphorus, S = Sulfur, B = Boron, Cu = Copper, Fe = Iron, Mn = Manganese, Zn = Zinc, Opt.= Optimum, Med.= Medium, VH = Very High, VL = Very Low.

Plants are greatly restricted in growth field under stress conditions, and have evolved many mechanisms to rapidly adapt to drought stress condition to keep growth and productivity (Zhu, 2002). The experimentation field of the present study belongs to semi-drought condition and the soil is highly alkaline, while the organic matter, potassium, total nitrogen and phosphorus are very low. In response to these conditions *C. dactylon* accessions collected from different regions of Bangladesh were found to be adapted very soon and nicely.

Table 11. Interpretation of soil test values based on critical limits (loamy soil for upland crops).

Nutrient Element	Very low	Low	Medium	Optimum	High	Very high
N%	≤0.09	0.091-0.18	0.181-0.27	0.271-0.36	0.361-0.45	>0.45
P (μg/g soil) (Olsen method)	≤7.50	7.51-15.0	15.1-22.5	22.51-30	30.1-37.5	>37.5
S (μg/g) soil	≤7.50	7.51-15.0	15.1-22.5	22.51-30	30.1-37.5	>37.5
K (meq/100g)	≤0.09	0.091-0.18	0.181-0.27	0.271-0.36	0.361-0.45	>0.45
Ca (meq/100g)	≤1.50	1.51-3.0	3.1-4.5	4.51-6.0	6.1-7.5	>7.5
Mg (meq/100g)	≤0.375	0.376-0.75	0.751-1.125	1.126-1.5	1.51-1.875	>1.875
Cu (μg/g)	≤0.15	0.015-0.3	0.31-0.45	0.451-0.6	0.61-0.75	>0.75
Zn (μg/g)	≤0.45	0.451-0.9	0.91-1.35	1.351-1.8	1.81-2.25	>2.25
Fe (μg/g)	≤3.0	3.1-6.0	6.1-9.0	9.1-12.0	12.1-15.0	>15.0
Mn (μg/g)	≤0.75	0.756-1.5	1.51-2.252	2.256-3.0	3.1-3.75	>3.75
B (μg/g)	≤0.15	0.151-0.3	0.31-0.45	0.451-0.6	0.61-0.75	>0.75

Ref. Fertilization Recommendation Guide.

This might be due to such mechanism as physiological, biochemical, molecular and cellular changes to cope with limited water supply (Xu and Huang, 2010; Lue et. al., 2011). Infact after transplanting the different accessions of this grass species each block was irrigated in limited way. Comparatively *C. dactylon* is one of the most drought tolerant turfgrasses (Kim et. al., 2009; Zhao *et. al.,* 2011). Some studies have suggested that drought tolerance of *C.dactylon* might be correlated with plant development such as leaf firing, root and shoot systems, and mass production (Carrow, 1996; Huang et. al.1997a; 1997b; Qian *et al.*, 1997), accumulation of dehydrin (Hu *et al.*, 2010a; Cambell and Close, 1997), evapotranspiration (Carrow, 1995), leaf water content, chlorophyll content, proline content and antioxidant enzyme activities (Hu *et al.*, 2010b; Lu *et al.*, 2007, 2009).

In the present study, shoot character consisting of seven parameters showed variations among 19 accessions both significantly and nonsignificantly. Number of shoots, shoot length and shoot fresh weight revealed significant variation. Almost similar results were found in case of root characters. Here number of root and number of rhizomes were found to show significant variations among the said accessions. Gobilik *et al.* (2013) studied five ecotypes of *C. dactylon* and in his general assessment, the Beaufort ecotype was found with shorter shoot length and the Sipitang ecotype had a higher shoot number.In addition, the Beaufort ecotype had higher shoot dry weight in salt treatment and on the contrary, Sipitang ecotype had higher shoot fresh weight. Salt causes roots to experience negative osmosis, distorts root nutrients uptake and impedes plant growth (Hajibagheri, 1989). The soil of the experimentation field was highly alkaline and that might be the cause of non-significant variation of root length, root fresh weight and root dry weight. Gobilik *et al.* (2013) found also all the five ecotypes with similar root fresh weight. Another important character of root i.e., number of rhizomes showed significant variation among the accessions. Dong and de Kroon (1994) stated that *C. dactylon* does not form rhizomes under shaded conditions.

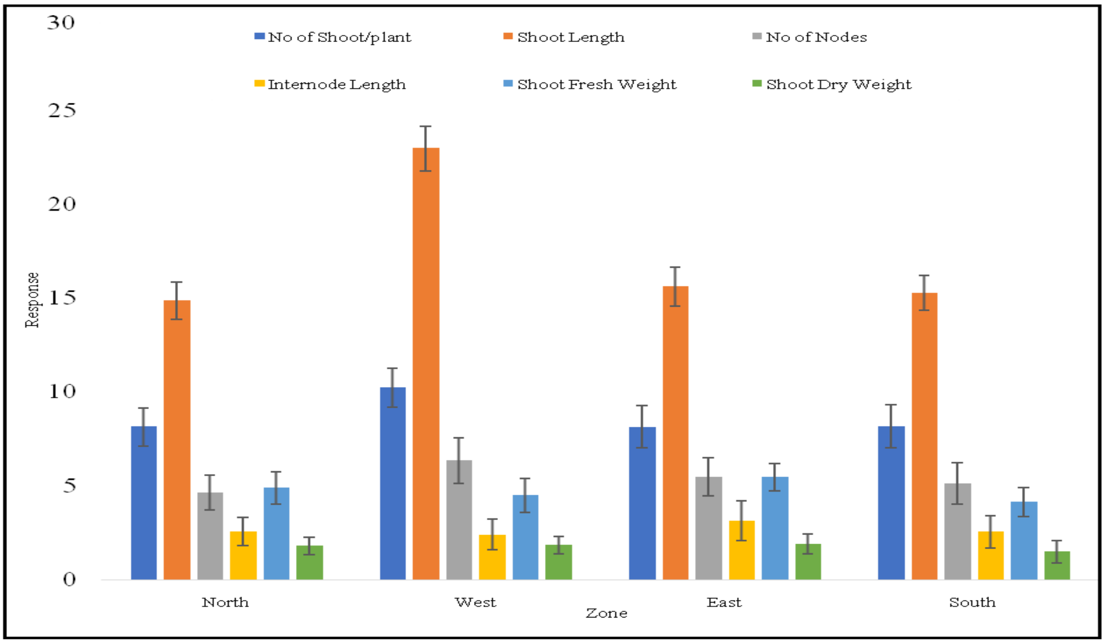

Fig. 1. Morphological variation of shoot of *Cynodon dactylon* in different zone of Bangladesh.

The present experimentation field was very much ambient and under high intensity of sun light. Even then this character showed wide variation.

However, Van De Wouw *et al.* (2009) stated that the presence of rhizomes in *C. dactylon* is the only identifying character compared to that of other *Cynodon* species. In case of leaf characters only number of leaf and total leaf area were found to show significant variations among the accessions studied in the present study. Other four characters showed no variations statistically.

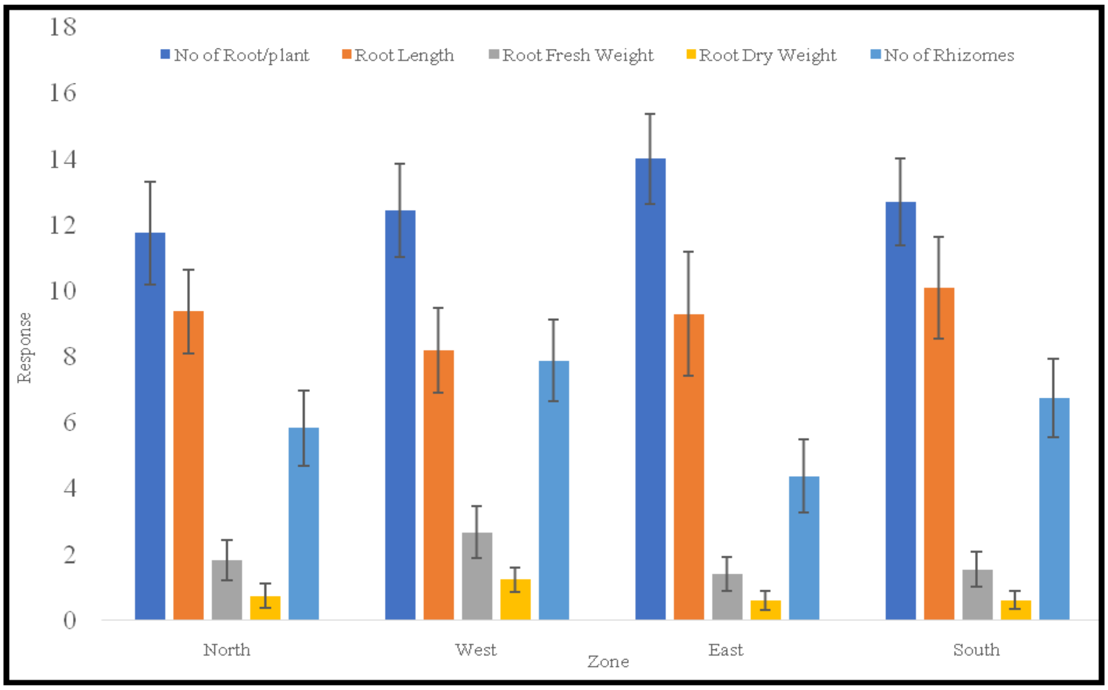

Fig. 2. Morphological variation of root of *Cynodon dactylon* in different zone of Bangladesh.

This also might be due to the salt accumulation in mature leaves which damages leaf tissues and decreases leaf photosynthetic activity (Gobilik *et al.* 2013). Hussain *et al.* (2012) stated that negative osmosis causes mesophyll turgidity loss and partial stomata opening reducing total leaf CO_2assimination per day. In saline soil, plants may die when new leaf formation cannot match leaf death or produce sufficient energy for plants (Munns and Termaat,1986).However, it is not expected in case of *C. dactylon* always, because this grass species can increase its root mass when salt concentration rises until it reaches a peak before starts to decline(Dudek *etal.*,1983). Probably same sorts of mechanism worked for maximum plant height, which was found with significant variation among all the plant accessions. Van De Wouw *et al.* (2009) found considerable diversity of plant height in a large group of accessions of *C. dactylon* and *C. nlemfuensis*.

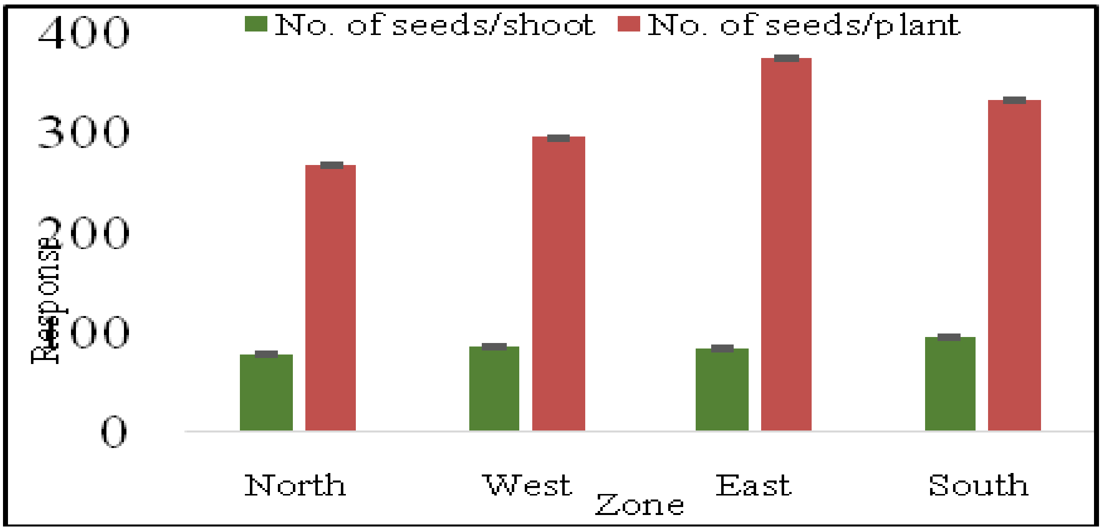

Fig. 3. Morphological variation of flower of *Cynodon dactylon* in different zone of Bangladesh.

The horizontally growing aboveground part of *C. dactylon* stems, called stolon were found to show non-significant variations in the present study. Van De Wouw *et al.* (2009) found 98 accessions of *Cynodon* belonging four different species with stolons, with considerable variation in number and length, and they stated that this might be due to very plastic response of stolons to light intensity and nutrient availability. Their findings support the present findings in all the 19 accessions of *C. dactylon*.

In *C. dactylon*, flowering stems are upright and bear a terminal group of 3-7 spikelike branches, originating in a single whorl on the tip of the stem. The colour of the spikelet was found to show somewhat variation of the main colour brown. In the same accession the spikelets were not purely brown, rather it was found with light green or green. Nasiri *et al.* (2012) described inflorescence of this grass species to be raceme like panicle and spikes to be glabrous, elongated and green to purple green, or green withpurple spots and yellowish at maturity. However, the ANOVA analysis in the present study showed no significant variation in case of inflorescence number and length of racemes. But the seed characters like seeds number per shoot and seeds number per plant were found with statistical variation. There are two types of *C. dactylon* and they are namely common and giant. Both these types are seeded varieties and they correspond to a wide range of genotypes and they are selected for their adaptability to different cultivation conditions (Ball and Pinkerton, 2002).

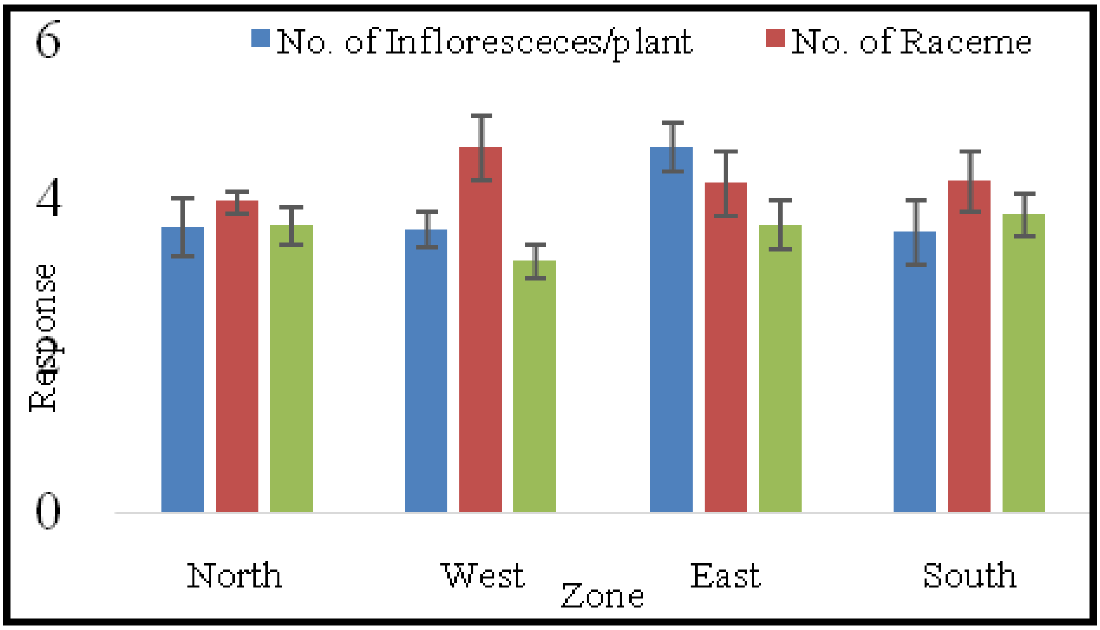

Fig. 4. Morphological variation of seed of Cynodon dactylon in different zone of Bangladesh.

In some were, "common" has become synonymous with any seed propagated Bermuda grass (Busey, 1989). Seeded varieties are outstanding tolerant of drought and heavy grazing. All the 19 accessions of *C. dactylon* in the present study were found to be seeded and since they are showing significant variation in respect of their number per shoot and per plant, their seeds may be commonly mixed for making commercial blend. Juska and Hanson (1964) stated that Bermuda grass (*C. dactylon*) vary with respect tothis characteristics of seed heads, when they appear and how long they persist. Some accessions have seed heads from early in the growing season throughout much of the summer, whereas with others seed heads may persist for a relatively short time.

They also mentioned that genetic factors and day length affect seed head formation. Seed characters found in 19 accessions of the present study might be due to these two regions in relation to their different habitats, Mc Whorter (1971) found extreme variability between ecotypes of Jhonson grass (*Sorghum halepense*(L.)Pers.) in seed production, shattering characteristics and in size and appearance of panicles. Seed of all ecotypes occurred in all spikelets clustered on short racemose branchlets. This type of mechanism may also be applicable in case of Bermuda grass. However, Al-Juboory and Hassaway (1980) stated that the differences that occur in the morphological development of Cogongrass (*Imperata cylindrica*) collected throughout Iraq and grown under the same condition, give another indication that the ecotypes represent various stages of evolutionary differentiation more than just local population.

They further said that this may be due to the process of sorting and controlling environmental factors (habitat factors) on species population which differ genetically. Their statement fully supports the finding in case of morphological variation of 19 accessions of *C. dactylon* in the present study.

The soil of the present experimentation field was found to be alkaline and the organic matter was low. Total nitrogen, potassium and phosphorus were also low. Other nutrients were somewhat favourable. The soil texture of the experimentation field was found to be clay loam under agroecological zone 26.In such a condition 19 accessions from different habitats belonging to four different zones were planted to find out their morphological variations along with adaptation. Almost under different conditions Ramakishnan and Gupta(1973) conducted an experiment with differential response of nitrogen, phosphorus and potassium. In conclusion they have said that besides calcium which was shown to be partially responsible for the restriction of three ecotypes of *C. dactylon* to their respective habitats, the availability of nitrogen, potassium and also phosphorus may play an important role. But striking all the accessions of *C. dactylon* in the present study showed no such poor performance comparatively among them and they were found to be well adapted.

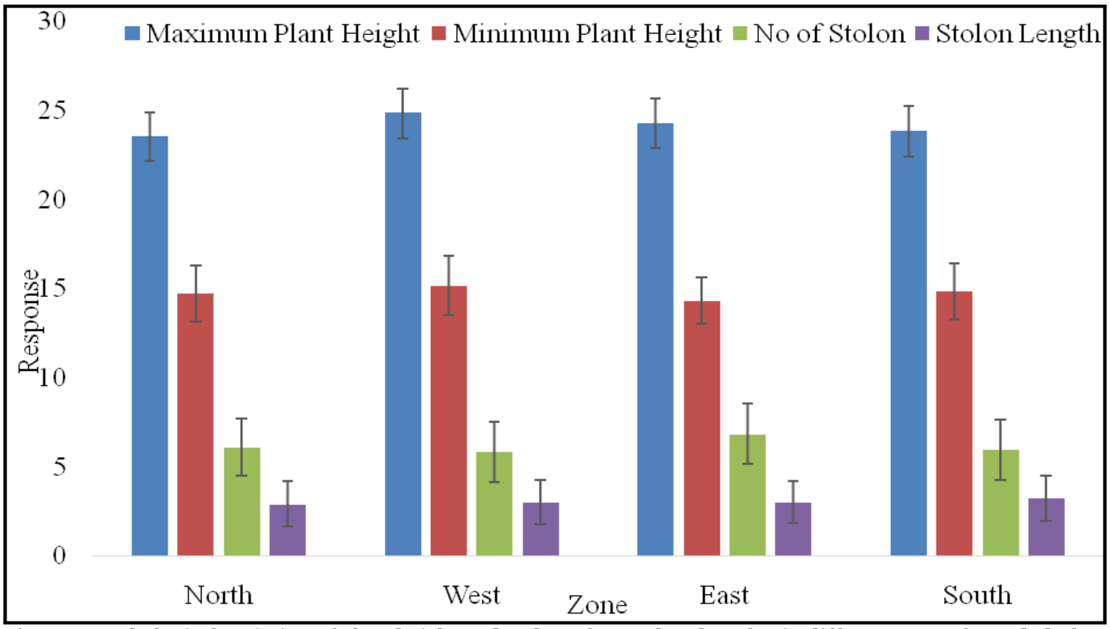

Fig. 5. Morphological variation of plant height and stolon of *Cynodon dactylon* in different zone of Bangladesh.

In most of the soils of different habitats in the present study organic matters and nitrogen were very low or low. At the same time Calcium and Magnesium were very high and high, respectively in the experimentation field. Calcium was found to be available in soils of different habitats. Only magnesium was low in the soil of Narshingdi. It is evident now that the macronutrients such as N,P,K^+ and Ca^{2+} are essentially required for regulation of a multitude of phenomena such as the activities of enzymes, protein synthesis, integrity of cell wall and plasma membrane, and as components of proteins, photosynthetic protein complexes, photosynthetic pigments, RNA and DNA(Taiz and Zeiger,2002; Akram *et al.*, 2008).Potassium in plants in the form of cation(K+) plays a vital role in regulation of osmotic adjustment by lowering osmotic potential of cells (Akram *et al.*,2008). It has been reported by Premachandra *etal.*(1990)that water stress generally increase the K^+ concentration especially under low phosphorus levels. Similar situation might be occurred in the present study where water supply was very much limited and phosphorus levels were low or medium in 7 soils of the habitat. In the present study, phosphorus concentrations were high in most of the soils of different habitats and that might be the cause of reduction in shoot and root characters. Very high and optimum level of Ca caused promising shoot character but low level of nitrogen was not favorable for shoot characters, particularly in case of minimum water supply in case of all the accessions of *C. dactylon*. In the case of few other micronutrients, sulfur was found to be low, while availability of Zinc was somewhat better in soils of different habitats in the present study. On the contrary the amount of copper, iron and manganese were sufficient almost in all the habitats.

Its now evident that the most reliable micronutrients for healthy plants are zinc, boron, copper and manganese. These are used by plants in small quantities. Inspite of the low requirement, critical plant functions are limited if micronutrients are unavailable resulting many plant abnormalities.

However, sandy soils with low organic matter may contain deficient micronutrients. High pH soils may make some nutrients low. Cations of copper, iron, manganese and zinc may hep to the soil properties, and solubility becomes high under acid condition and becomes deficient on calcareous soils or becomes high in organic matter where strong chelation decreases availability (Meugel, 1990; Bennett, 1993; Stevans *et al.*, 2002; Kelling, 2005).

All the 19 accessions of *C. dactylon* collected from different habitats and transplanted in an alkaline soil of the experimentation field were found to be adapted nicely inspite of their different physico-chemical and environmental conditions. That might due to many physiological and biochemical mechanisms to any environmental conditions keeping their growth and productivity more or less properly.

Several studies report nutrient data for warm season turfgrass including bermuda grass (*C. dactylon*) in many transition zones in the world (Wu *et al.*,2009).Mineral composition of bermudagrass was studied by McCrimmon (2001), Snyder and Cisar (2000) and Walworth and Kopec (2004) with different results depending on the cultivar and different nutrient sources and rates. Volterrani *et al.* (2012) stated that use of plug plants shows several advantages and this technique is useful for noninvasive putting green conversion, since the use of fully developed root systems and actively growing shoots enhances their colonization potential. These features also might be the appropriate reason for proper growth and adaptation of different accession of 19 different habitats in this experiment.

Conclusion

The present research study was of academic interest particularly in relation to adaptation of *Cynodon dactylon* accessions collected from four different zones of Bangladesh. A total of nineteen accessions were evaluated agro-morphologically and they showed variability as far as the quantitative parameters were concerned. Most of the collected germplasms were found to be susceptible to the weather and alkaline clay soil of the experimentation field. Observations from this study indicate both significant and non-significant variations among the accessions in terms of vegetative and reproductive characters. Rhizomatous and stoloniferous nature showed both maximum and minimum growth variation. Soil properties of their habitat were found to be somewhat different in many cases compared to that of the experimentation field. But at the same time all the accessions were found to be well adapted to the soil of experimentation field which indicates the functional qualities like rigidity, resiliency, elasticity, rooting capacity etc. of this grass species. All the end, it may be said that further studies are needed for better characterization of *C. dactylon* in order to assess the potential benefits compared that of other alike grass species.

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Dear **SK Nitu**,

I am pleased to inform you that your paper titled "**LEAF EPIDERMAL ANATOMY OF CYNODON DACTYLON (L.) PERS IN RELATION TO ECOTYPIC ADAPTATION**" by **SK Nitu, H Tarique and SMS Islam** has been accepted for the publication in the issue of Vol. 28(1), June 2021 of the Bangladesh Journal of Plant Taxonomy.

Regards,

Yours sincerely,

Dr. Mohammed Almujaddade Alfasane
Chief Editor
Bangladesh Journal of Plant Taxonomy

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C. List of Publications

1. **SK Nitu**, MM Ud-Deen and SA Haider **(2009)**. Effect of different *Rhizobium* strains on the growth attributes of black gram. J. Life Earth Sci., 3-4: 7-11.
2. **SK Nitu** and SA Haider **(2009)**. Response of growth indices in black gram due to *Rhizobium* inoculation. J. Life Earth Sci., 3-4: 51-54.
3. **SK Nitu**, MM Ud-Deen and SA Haider **(2009)**. Influence of different *Rhizobium* based biofertilizers on the yield and yield components of black gram (*Vigna mungo* L. Hepper). J. Bangladesh Soc. Agric. Sci. Technol., 6 (1&2): 99-104.
4. AHM Mahbubur Rahman, Z Ferdows, **SK Nitu** and AK M Rafiul Islam **(2015)**. Herbaceous plant species in and around Rajshahi metropolitan city, Bangladesh. International Journal of Advanced Research. 3(5): 1002-1018.
5. **SK Nitu**, SMS Islam and MH Tarique **(2019a)**. Interphase nuclear phenotype and chromosomal variation in *Cynodon dactylon* (L.) Pers. J. Bio-Sci., 27: 133-141.
6. **SK Nitu**, SMS Islam and MH Tarique **(2019b)**. Morphological characteristics of different accessions of *Cynodon dactylon* (L.) Pers. and physico-chemical properties of soil of their growing region in Bangladesh. Int. J. Biosci., 15(5): 350-369.
7. **SK Nitu**, H Tarique and SMS Islam **(2021).** Leaf epidermal anatomy of *Cynodon dactylon* (L.) Pers. in relation to ecotypic adaptation. Bangladesh Journal of Plant Taxonomy, Vol 28(1), June 2021. Accepted (Ref. No. BJPT/10-01/2020).