

**MORPHO-PHYSIOLOGICAL AND BIOCHEMICAL  
RESPONSES, FODDER YIELD AND QUALITY OF  
NAPIER GRASS UNDER WATER STRESS**

**FARIA JANNAT**



**DEPARTMENT OF AGRONOMY  
SHER-E-BANGLA AGRICULTURAL UNIVERISITY  
DHAKA-1207**

**DECEMBER, 2021**

**MORPHO-PHYSIOLOGICAL AND BIOCHEMICAL  
RESPONSES, FODDER YIELD AND QUALITY OF NAPIER  
GRASS UNDER WATER STRESS**

**BY**

**FARIA JANNAT**

**REGISTRATION NO. 15-06589**  
**Email: [fariasnigdha15@gmail.com](mailto:fariasnigdha15@gmail.com)**  
**Contact no.: 01960599847**

*A Thesis Submitted to  
The Department of Agronomy, Faculty of Agriculture,  
Sher-e-Bangla Agricultural University, Dhaka,  
in partial fulfilment of the requirements  
for the degree of*

**MASTERS OF SCIENCE (MS)  
IN  
AGRONOMY  
SEMESTER: JULY-DECEMBER, 2021**

**APPROVED BY:**

---

**Dr. Mirza Hasanuzzaman**  
**Professor**  
**Department of Agronomy**  
**Supervisor**

---

**Dr. Md. Mahabub Alam**  
**Assistant Professor**  
**Department of Agronomy**  
**Co-Supervisor**

---

**Prof. Dr. Tuhin Suvra Roy**  
**Chairman**  
**Examination Committee**

***DEDICATED***  
***TO***  
***MY BELOVED PARENTS***



**DEPARTMENT OF AGRONOMY**  
Sher-e-Bangla Agricultural University  
Sher-e-Bangla Nagar  
Dhaka-1207

---

***CERTIFICATE***

*This is to certify that the thesis entitled “MORPHO-PHYSICAL AND BIOCHEMICAL RESPONSES, FODDER YIELD AND QUALITY OF NAPIER GRASS UNDER WATER STRESS” submitted to the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfilment of the requirements for the degree of MASTER OF SCIENCE (MS) in AGRONOMY, embodies the result of a piece of bonafide research work carried out by FARIA JANNAT, Registration No. 15-06589 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

*I further certify that any help or source of information, received during the course of this investigation has been duly acknowledged.*

**Dated:**  
**Place: Dhaka, Bangladesh**

---

**Prof. Dr. Mirza Hasanuzzaman**  
**Supervisor**

## **ACKNOWLEDGEMENTS**

*The author gives all praise and heartiest thanks to Almighty Allah for giving her the knowledge and capacity to complete her research successfully for the degree of Masters of Science (MS) in Agronomy. It gives her great pleasure to offer author's sincere gratitude.*

*The author would like to express her deepest respect, sincere appreciation, and immense debt of gratitude to her reverend supervisor, **Dr. Mirza Hasanuzzaman**, Professor, Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, for his academic guidance, helpful comments, and constant inspiration during the research process and thesis preparation.*

*The author is grateful to **Dr. Md. Mahabub Alam**, Assistant Professor, Department of agronomy, Sher-e-Bangla Agricultural University, Dhaka, her co-supervisor, for his insightful comments, constructive criticism, on the thesis writing. Particularly during the pandemic, he greatly aided the author in measuring data parameter and doing other things.*

*She also owed a debt of gratitude to all of her professors at the Department of Agronomy at Sher-e-Bangla Agricultural University, who served as a continual source of inspiration and enthusiasm during her MS program.*

*The author would like to express her sincere gratitude to **Prof. Dr. Kamrun Nahar**, Professor, Department of Agricultural Botany, Sher-e-Bangla Agricultural University, Dhaka, for sharing her insightful knowledge on conducting research and writing thesis throughout the entire study period.*

*The author also wants to express her gratitude to **Taufika Islam Anee**, Assistant Professor in the Department of Agronomy, for all of her consistent and thorough assistance, as well as for her skillful advice, and encouragement in the development of the thesis.*

*The author likes to acknowledge the Government of Bangladesh through its Ministry of Science and Technology for providing financial support (**NST fellowship**) to carry out this research work.*

*The author also wishes to special thanks to **Abdul Awal Chowdhury Masud**, Assistant Professor, Department of Agronomy for his unconditional help during the research work in the pandemic condition. She wants to thanks **Farzana Nowroz**, Assistant Professor, Department of Agronomy for her support and suggestions. Last but not the least thanks all of her fellow lab mates specially Mst. Mahbuba Khatun for being there in all works and sharing her joys and sorrows.*

*The author extends her sincere gratitude to her cherished parents, sweet sister & loving brother, friends for their all-time prayers, support, inspiration, and moral assistance in helping her pursue further education and lastly her dearest roommates for their continuous mental support.*

**The Author**

# **MORPHO-PHYSIOLOGICAL AND BIOCHEMICAL RESPONSES, FODDER YIELD AND QUALITY OF NAPIER GRASS UNDER WATER STRESS**

## **ABSTRACT**

Napier grass (*Pennisetum purpureum* Schumach) is grown in tropical and temperate regions well adopted as a fodder crop because of its high forage productivity and rapid regeneration. It is considered as drought tolerant and shows high water use efficiency. This experiment was done in the shed house of Sher-e-Bangla Agricultural University during the period of April-July, 2021. The study was to investigate the water stress-induced morphological, physiological and oxidative damages in napier grass and to differentiate between the responses of napier grass to drought and waterlogging conditions. Napier grass was grown for up to 21 days and then exposed to water stress: viz. drought and waterlogging for different durations (7, 14 and 21 days) and also there was maintained control condition. After each stress period, morphological, physiological, and biochemical data were measured following standard procedures. At 50 days after sowing the same parameters were recorded for all treatments and considered as recover data. Plant height, SPAD value, fresh weight, dry weight, relative water content, and fodder yield decreased under both waterlogging and drought stress conditions compared to control. The reduction was higher under drought conditions than in waterlogging. Root length, root shoot ratio, proline, malondialdehyde (MDA) and H<sub>2</sub>O<sub>2</sub> content were higher under stress conditions compared to control. Drought-stressed plants were more severely affected than waterlogged one. At 50 days after sowing, plant height, shoot fresh weight, shoot dry weight and fodder yield were decreased in plants stressed for longer periods. Root fresh weight, root dry weight, root length and root branch were decreased in plants stressed for 21 days, whereas increased under waterlogging. Proline, MDA and H<sub>2</sub>O<sub>2</sub> content were increased upon exposure to the long duration of stress. Quality parameters such as neutral detergent fibre and acid detergent fibre has been reduced under both stress condition but in case of drought, the reduction was significant. Dry matter, crude fibre, moisture content, hemicellulose and organic matter percentage were reduced under drought and waterlogging stress and whereas crude protein and ash content increased with increase of stress duration. As plants water stressed for 7 days got the highest days for recovery, so it showed better performance and even better than control. Our experiment concludes that napier grass is more sensitive to drought than waterlogging in case of morphology and plants also recovered more efficiently in case of waterlogging than drought. In case of oxidative damage, drought-exposed plants showed more tolerant capacity compared to waterlogged plants.

## TABLE OF CONTENTS

Chapter No.	Title	Page No.
	<b>ACKNOWLEDGEMENTS</b>	i
	<b>ABSTRACT</b>	ii
	<b>TABLE OF CONTENTS</b>	iii-vii
	<b>LIST OF FIGURES</b>	viii-ix
	<b>LIST OF APPENDICES</b>	x- xi
	<b>ABBREVIATIONS AND ACCRONYMS</b>	xii-xiii
<b>I</b>	<b>INTRODUCTION</b>	1-5
<b>II</b>	<b>REVIEW OF LITERATURE</b>	6-24
2.1	Napier grass	6
2.1.1	Botany	6
2.1.2	Importance	6-7
2.2	Abiotic stress	7-8
2.2.1	Plant response to abiotic stress	8-9
2.3	Drought	9-15
2.3.1	Plant response to drought stress	10
2.3.1.1	Effect of drought on crop growth	10-11
2.3.1.2	Effect of drought stress on crop physiology	11-13
2.3.1.3	Effect on yield and yield attributes	13-14
2.3.2	Drought-induced oxidative stress and antioxidant defense system	14-15
2.4	Waterlogging	15-16
2.4.1	Plant responses to waterlogging stress	16
2.4.1.1	Effect of waterlogging on crop growth	16-17
2.4.1.2	Effect of waterlogging on crop physiology and metabolism	17-19
2.4.1.3	Effect of waterlogging on yield and yield contributors	19-20
2.4.2	Waterlogging-induced oxidative stress and antioxidant defense system	20-21

## TABLE OF CONTENTS (Cont'd)

<b>Chapter No.</b>	<b>Title</b>	<b>Page No.</b>
2.5	Effect of water stress on napier grass	21
2.5.1	Effect of water stress on growth of napier	21-22
2.5.2	Effect of water stress on the physiology of napier	22
2.5.3	Effect of water stress on napier yield	22
2.5.4	Effect of water stress on the quality of napier	23-24
<b>III</b>	<b>MATERIALS AND METHODS</b>	<b>25-39</b>
3.1	Location	25
3.2	Soil	25
3.3	Weather	25
3.4	Materials	26
3.4.1	Plant materials	26
3.4.2	Plastic pot	26
3.5	Treatments	26
3.6	Design and layout of the experiment	27
3.7	Seed collection	27
3.8	Pot preparation	27
3.9	Fertilizers application	27
3.10	Seed sowing technique	28
3.11	Intercultural operations	28
3.11.1	Gap filling and thinning	28
3.11.2	Weeding and irrigation	28
3.11.3	Plant protection measure	28
3.12	General observation of the experimental pots	28
3.13	Data collection	29



## TABLE OF CONTENTS (Cont'd)

Chapter No.	Title	Page No.
3.13.1	Crop growth parameters	29
3.13.2	Physiological parameters	29
3.13.3	Oxidative stress indicators	29
3.13.4	Qualitative parameters	30
3.14	Procedure of sampling for growth study during the crop growth period	30
3.14.1	Plant height	30
3.14.2	Root length	30
3.14.3	Shoot fresh weight	31
3.14.4	Shoot dry weight	31
3.14.5	Root fresh weight	31
3.14.6	Root dry weight	31
3.14.7	Root-shoot ratio	32
3.14.8	Root branch	32
3.15	Procedure of sampling for physiological parameters	32
3.15.1	SPAD value	32
3.15.2	Relative water content	33
3.16	Procedure for measuring biochemical parameters	33
3.16.1	Measurement of proline content	33-34
3.16.2	Measurement of lipid peroxidation	34
3.16.3	Determination of hydrogen peroxide content	34
3.17	Procedure of sampling and measuring the fodder yield	35
3.17.1	Fodder yield	35

## TABLE OF CONTENTS (Cont'd)

Chapter No.	Title	Page No.
3.18	Procedure of sampling and measuring the qualitative parameter	35
3.18.1	Acid detergent fibre	35
3.18.2	Neutral detergent fibre	36
3.18.3	Dry matter	36
3.18.4	Crude protein	36
3.18.5	Crude fibre	37
3.18.6	Ash	37
3.18.7	Organic matter percentage	37
3.18.8	Hemicellulose	38
3.19	Statistical Analysis	38
<b>IV</b>	<b>RESULTS AND DISCUSSION</b>	39-70
4.1	Growth parameters	39
4.1.1	Plant height	39-40
4.1.2	Shoot fresh weight	40-41
4.1.3	Root fresh weight	41-42
4.1.4	Shoot dry weight	42-43
4.1.5	Root dry weight	43-44
4.1.6	Root length	45-46
4.1.7	Root-shoot ratio	46-47
4.1.8	Root branch	47-48
4.2	Physiological parameters	48
4.2.1	SPAD value	48-49
4.2.1	Relative water content	49-50
4.3	Biochemical parameters	51
4.3.1	Proline content	51-52
4.3.2	Malondialdehyde content	52-53
4.3.3	Hydrogen peroxide (H <sub>2</sub> O <sub>2</sub> ) content	53-54
4.4	Yield parameter	55
4.4.1	Fodder yield	55
4.5	Qualitative parameters	57

## TABLE OF CONTENTS (Cont'd)

<b>Chapter No.</b>	<b>Title</b>	<b>Page No.</b>
4.5.1	Acid detergent fibre	56-57
4.5.2	Neutral detergent fibre	57-58
4.5.3	Dry matter	58-59
4.5.4	Crude protein	59-60
4.5.5	Crude fibre	61
4.5.6	Ash	62
4.5.7	Organic matter percentage	63
4.5.8	Hemicellulose	64
<b>V</b>	<b>SUMMARY AND CONCLUSION</b>	65-69
	<b>REFERENCES</b>	70-86
	<b>APPENDICES</b>	87-95

## LIST OF FIGURES

<b>Figure No.</b>	<b>Title</b>	<b>Page No.</b>
1	Plant height of napier grass at completion of different stress duration and at recovery	39
2	Shoot fresh weight plant <sup>-1</sup> of napier grass at completion of different stress duration and at recovery	40
3	Root fresh weight plant <sup>-1</sup> of napier grass at completion of different stress duration and at recovery	42
4	Shoot dry weight plant <sup>-1</sup> of napier grass at completion of different stress duration and at recovery	43
5	Root dry weight plant <sup>-1</sup> of napier grass at completion of different stress duration and at recovery	44
6	Root length of napier grass at completion of different stress duration and at recovery	45
7	Root-shoot ratio of napier grass at completion of different stress duration and at recovery	46
8	Root branch number of napier grass at completion of different stress duration and at recovery	48
9	SPAD value of napier grass at completion of different stress duration and at recovery	49
10	Relative water content of napier grass at completion of different stress duration and at recovery	51
11	Proline content of napier grass at completion of different stress duration and at recovery	52
12	Malondialdehyde content of napier grass at completion of different stress duration and at recovery	53
13	Hydrogen peroxide content of napier grass at completion of different stress duration and at recovery	55

## LIST OF FIGURES (Cont'd)

<b>Figure No.</b>	<b>Title</b>	<b>Page No.</b>
14	Fodder yield of napier grass at recovery	56
15	Acid detergent fiber of napier grass at recovery	57
16	Neutral detergent fibre of napier grass at recovery	58
17	Dry matter content of napier grass at recovery	60
18	Crude protein of napier grass at recovery	61
19	Crude fibre of napier grass at recovery	62
20	Ash content of napier grass at recovery	63
21	Organic matter percentage of napier grass at recovery	64
22	Hemicellulose content of napier grass at recovery	65

## LIST OF APPENDICES

.Appendix No.	Title	Page No.
I	Phenotypic pictures of napier grass under different water stress treatment and recovery	88-90
II	Phenotypic pictures of napier grass at 50 days after sowing	91
III	Map showing the location of the experiment	92
IV	Monthly average air temperature, rainfall, and relative humidity of the experiment site during the period from March 2021 to July 2021	93
V	Mean square values and degree of freedom (DF) of plant height, shoot fresh weight, root fresh weight, shoot dry weight, and root dry weight at different stress duration (drought and waterlogging) of napier grass	93
VI	Mean square values and degree of freedom (DF) of plant height, shoot fresh weight, root fresh weight, shoot dry weight, and root dry weight at recovery of napier grass	93
VII	Mean square values and degree of freedom (DF) of root length, root-shoot ratio, and root branch at different stress duration (drought and waterlogging) of napier grass	94
VIII	Mean square values and degree of freedom (DF) of Root length, root-shoot ratio, and root branch at recovery of napier grass	94
IX	Mean square values and degree of freedom (DF) of SPAD value, relative water content, and proline at different stress duration (drought and waterlogging) of napier grass	94
X	Mean square values and degree of freedom (DF) of SPAD value, relative water content, and proline at recovery of napier grass	94
XI	Mean square values and degree of freedom (DF) of malondialdehyde content and hydrogen peroxide at different stress duration (drought and waterlogging) of napier grass	95

## LIST OF APPENDICES (Cont'd)

<b>Appendix No.</b>	<b>Title</b>	<b>Page No.</b>
XII	Mean square values and degree of freedom (DF) of malondialdehyde content and hydrogen peroxide and at recovery of napier grass	95
XIII	Mean square values and degree of freedom (DF) of fodder yield, acid detergent fiber, neutral detergent fibre, and dry matter content at recovery of napier grass	95
XIV	Mean square values and degree of freedom (DF) of crude protein, crude fibre and ash content at recovery of napier grass	95
XV	Mean square values and degree of freedom (DF) of moisture percentage, organic matter content and hemicellulose at recovery of napier grass	95

## ABBREVIATIONS AND ACRONYMS

ADF	Acid Detergent Fibre
ANOVA	Analysis of variance
AsA	Ascorbic acid (ascorbate)
BARI	Bangladesh Agricultural Research Institute
BBS	Bangladesh Bureau of Statistics
CF	Crude Fibre
Chl	Chlorophyll
CP	Crude Protein
CRD	Completely Randomized Design
CV	Coefficient of Variance
cv.	Cultivar
D	Drought
DAS	Days after sowing
DM	Dry matter
DW	Dry Weight
EL	Electrolyte Leakage
<i>et al.</i>	<i>et alibi</i> (and others)
ETC	Electron Transport Chain
FAO	Food and Agriculture Organization
FC	Field capacity
FW	Fresh Weight
GDP	Gross Domestic Product
GHGs	Green House Gases
i.e.	id est (That is)
LSD	Least significant difference
MDA	Malondialdehyde
Pro	Proline
ROS	Reactive oxygen species
RSR	Root-shoot Ratio
OM	Organic Matter



°C	Degree Celsius
µg	Microgram
Cm	Centimeter
G	Gram
Ha	Hectare
Kg	Kilogram
M	Meter
Mg	Milligram
MO	Moisture
mM	Millimole
RWC	Relative water content
viz.	Namely
WL	Waterlogging
WUE	Water Use Efficiency

## CHAPTER I

### INTRODUCTION

In this post-industrial area, fossil fuels burning and an increase in the amount of damaging greenhouse gases (GHGs) in the atmosphere are the major causes of climate change. The industrial revolution is considered as main reason of the world's climate change (Dutta *et al.*, 2020). Variations in temperature, rainfall, and air conditions due to climate change have exposed plants to extreme and severe climatic conditions that have an adverse influence on plant morphological, developmental, cellular, and molecular processes (Raza *et al.*, 2019).

The world population is expected to 11.2 billion people by 2100, according to the United Nations (2019). But agricultural areas won't be able to produce at the same rate. In addition, 83 million individuals are added to the world's population each year (UN, 2019). The only way to feed the one in nine people who are already hungry in the globe is to sustainably double food production (OECD/FAO, 2019). By 2050, the amount of food needed will have to increase by 70% to meet the challenge of a growing population (Hasanuzzaman *et al.*, 2018a). Bangladesh, an agricultural nation, is struggling to adapt to climate change and feed an expanding population to attain food and feed security (Islam *et al.*, 2017).

Abiotic stress has become more prevalent and has a greater negative impact on plants due to climate change, as seen throughout a large geographic area. According to FAO (2019), 96.5% of the world's agriculture area is subjected to some sort of stress. Due to abiotic stresses which include heat shock, chilling/freezing, water shortage, waterlogging, salt, nutritional imbalance, heavy metals, and xenobiotic stress, 50% of production loss is occurred (Saini *et al.*, 2018). Water stress can be two types, viz. drought/water deficit and waterlogging/flooding. Extremes of drought and flooding are encouraged by the condition in various places of the world (Feng *et al.*, 2013).

Since the 1980s, droughts have been increasing in frequency and intensity due to climatic change (Liu *et al.*, 2011; Trenberth *et al.*, 2014; Saud *et al.*, 2017), especially

in semi-arid regions of the Northern Hemisphere. When crop demand exceeds the amount of water in the soil layers, climate change and agriculture intensification could result in severe soil drought (Leng *et al.*, 2015). Water scarcity, constrained plant development, and reduced yield are all effects of drought-induced water deficit stress (Misra *et al.*, 2016). Significant relative and absolute water content reduction, unbalanced osmotic pressure, and loss of turgidity were all caused by the stress of drought (Nahar *et al.*, 2017). Water shortage stress causes a decrease in the plant's water potential and turgor to the point that it interferes with cells' ability to function normally. Reduced stomatal opening, slowed CO<sub>2</sub> fixation, accelerated O<sub>2</sub> photo reduction in the chloroplast, and increased photorespiration caused by water deficiency stress finally cause reactive oxygen species (ROS) buildup and oxidative damage in plants (Jalil *et al.*, 2017). According to Bhargava and Sawant (2013), severe drought circumstances promoted lipid peroxidation, which damaged proteins, the photosynthetic system, and cell membranes, and ultimately led to programmed cell death. Through poor water, ion, and nutrient uptake from soil matrix by roots, altered carbon and nitrogen cycle, stomatal closure, photosynthesis inhibition, decreased carbohydrate synthesis, increased respiration, decreased cell division, and elongation, mild to moderate drought stress frequently resulted in impairment of these physiological and biochemical processes (Hasanuzzaman *et al.*, 2018b; Bhuiyan *et al.*, 2019).

Soil flooding or waterlogging is brought on by an excessive buildup of water in the soil as a result of prolonged, heavy precipitation, inadequate drainage, etc. (Hossain and Uddin, 2011). Waterlogging harms around 10% of the world's land. Floods were responsible for two-thirds of all crop losses and damage globally between 2006 and 2016. Waterlogging is characterized by poor light, hampered gaseous exchange, hypoxia, and anoxia, and it covers plant roots (Fukao *et al.*, 2019). By 10,000 times lessening O<sub>2</sub> diffusion than air, it inhibits aerobic activity, including soil root respiration (Yamauchi *et al.*, 2019). Because the anoxic situation prevents chloroplast and mitochondrial ETC, ROS are therefore produced (Sasidharan *et al.*, 2018). Due to the change in soil pH, flooding also removes important nutrients from the soil, accumulates salts, and increases the availability of heavy metals. Plants eventually experience nutritional deficiencies and other stresses (salinity, heavy metals) as a result of these negative alterations (Steffens, 2014).

In Bangladesh, the livestock industry, which provides nutritious and affordable food, is crucial to the development of the rural economy. By providing nutritious, affordable, and balanced food in the form of milk and other animal products, the livestock subsector is important. In order to reduce rural poverty as well as maintain sustainable agricultural expansion, it is therefore ideal for the livestock sector to grow quickly. Agriculture contributes 18.70 percent of the country's GDP, and the livestock subsector contributes 2.45 percent. It employs roughly 25% of the labor force (BER, 2013). Additionally, keeping livestock provides possibilities to use collective grazing pastures, and diversifies income (Faruque, 2003). In addition to the animals' low genetic potential, hunger, undernutrition, or both are major contributors to our livestock's low output. Lack of a sufficient amount of high-quality feed will prevent animals from growing as expected. According to the study by Sayeed *et al.* (2008), the average area under cultivation for fodder was growing, which is encouraging for the livestock subsector. The production of nutrient-rich, high-yielding varieties of fodder is inevitable since fodder constitutes a significant portion of the daily protein intake of dairy animals. Ample and reasonably priced feed availability is crucial for profitable livestock operations. The practice of producing and preserving fodder is quite recent in Bangladesh. Farmers are anxious to grow fodder for their cattle as nourishing feed despite a number of obstacles. Raising improved breeds of cattle requires the cultivation of high-quality fodders. The worry of not being able to supply enough feed and fodder grows more pressing as animal populations rise over time. In most emerging nations, including Bangladesh, the demand for fodder is becoming a difficult problem. For the livestock sub-sector, a reliable supply of high-quality feed and fodder is essential.

Napier Grass (*Pennisetum purpureum*) under the family Poaceae is a perennial fodder crop native to Africa and is now induced and grown in many temperate as well as tropical countries. It is a tall grass that forms stems, has recently gained interest as a potential bioenergy crop due to its robust biomass output (Waramit and Chaugool, 2014). This thermophilic plant is a highly productive crop due to its tropical origin. such as those found in Thailand. Napier grass compared to many other plants, can produce more dry matter (DM) per unit area. if just clipped once a year (El Bassam, 2010). Along with its many features that make them suitable for use as a crop drought tolerance to various soil types (de Morais *et al.*, 2012). It is simple to grow napier grass by placing cuttings in the field (Lounglawan *et al.*, 2014).

Bangladesh is regarded as the most susceptible country to stress in the world due to its socioeconomic situation, geographic location, and the negative effects of climate change and climate variability (Akter and Rahman, 2012; Ali *et al.*, 2019; Islam and Nursey-Bray, 2017; Shahin *et al.*, 2014). As a result of its high population density, small land size, fragile economy, socioeconomic inequality, and limited capacity for adaptation, our nation is less able to deal with the effects of climate change. The primary source of income, accounting for 40.62% of employment and 14.23% of GDP, is agriculture (Finance Division, 2018). However, climate change is anticipated to have a substantial impact on agriculture and reduce agricultural GDP by 3.1% each year (Delaporte and Maurel, 2018). In the end, this will mostly have an impact on subsistence farming and the nation's food security. The farming communities are among the populations most at risk from climate change (Misra, 2017; Rahman *et al.*, 2017). Due to their high rates of poverty, reliance on agriculture, lack of adaptive capacity, and high variability in annual and seasonal rainfall, the Barind tract and the Tista floodplain regions of the country's northern and northwestern parts (collectively known as North Bengal) are more severely affected by drought than other regions are (Habiba *et al.*, 2014; Shahid and Behrawan, 2008). On the other hand, Bangladesh is highly vulnerable to waterlogging and two-thirds of its land is less than five meters above sea level (Islam and Das, 2014; Dasgupta *et al.*, 2011). According to estimates, the GDP volume could expand by 0.02% annually if there had not been such an economic loss during that time (Islam, 2016). In Bangladesh, waterlogging, which often results in smaller inundation depths and slower flow rates than floods, severely disrupts daily city life by causing infrastructure damage, traffic gridlock, health issues, and environmental issues (Subrina and Chowdhury, 2018).

In this regard, Napier grass can be a good selection. This grass has a high growth rate, high productivity, and good nutritive value. It is considered as a short-term drought tolerant forage which is a useful trait in areas with low soil moisture during the dry season, although it requires rainfall >1000 mm, so the species is adaptable to drought conditions (Nagasuga, 2003). Though it is tolerant to drought, under water stress conditions, the persistence and regeneration of this species are constrained. Maintenance of tissue moisture is critical to survival and productivity in dry environments. This is because it is often linked with the opening of stomata for the assimilation of CO<sub>2</sub> (Cardoso *et al.* 2015). Stomatal closure can conserve tissue

moisture and thereby stabilize tissue water potential when soil available water is low. It has been reported that stomatal conductance in grass species growing in the open tropical savannah is less responsive to changing soil water conditions, unlike the same species under the shade because they are more efficient in extracting water from dry soil. In case of excess water conditions, napier grass is sensitive to waterlogging. Under waterlogged conditions, the Napier grass doesn't show any tolerance mechanism and it is sensitive and as a result, it can't survive for long. Considering these facts this study has been designed with the following objectives:

- To investigate the water stress-induced morphological, physiological, biochemical, and oxidative damages in napier grass.
- To know the fodder yield under drought and waterlogging conditions.
- To study the quality attributes under different duration of drought and waterlogging conditions.
- To find the recovery performance of napier grass under drought and waterlogging conditions.

## CHAPTER II

### REVIEW OF LITERATURE

#### 2.1 Napier grass

Napier grass (*Pennisetum purpureum* Schumach) is a perennial fodder crop. It is a grass crop under the Poaceae family. It is also known as elephant grass, uganda grass. It is a multipurpose forage grass and grown in intensive or semi-intensive agriculture (Mkhutche, 2020). Napier grass is native to sub-Saharan Africa and extensively grown in the tropical and sub-tropical areas. Out of total global area of 10 million hectares, 35% of the area is used for fodder purpose (Joshi, 2015).

##### 2.1.1 Botany

Napier grass is a monocotyledonous, C<sub>4</sub> plant. It belongs to the ethnic group Paniceae in the subfamily Panicoideae of the Poaceae family (Bogdan, 1977). It is profusely tillering, tall growing (1.5-2.0 m). There are 150 varieties of napier grass introduced in the tropical and sub-tropical regions (Tudsri, 2005). Napier propagation mainly happens through cutting as the seeds produce weak seedlings (Negawo *et al.*, 2017). Seeds are highly heterozygous as it is open-pollinated. Green biomass is the main desirable part for using as fodder for livestock. It can produce 60-150 tons of green matter ha<sup>-1</sup> per year and can be repeated cutting for 4-6 times per year. It can tolerate high temperature, drought stress, low fertility status etc. (Dokbua *et al.*, 2021; Rusdy, 2016; Yan *et al.*, 2021).

##### 2.1.2 Importance

Napier is a quick-growing perennial grass that may thrive in dry and droughty environments (Sawasdee and Pisutpaisal, 2014). It is a promising crop for energy production due to its high biomass yields and year-round harvest ability in subtropical areas (Takara and Khanal, 2015). Plant biomass can be burned to produce hot gas (at temperatures between 800 and 1000 °C) as biofuel which is used in steam turbine power

generator boiler systems. Two primary types of processes, namely thermo-chemical and biochemical processes, may convert biomass into biofuel as a renewable and clean energy source (e.g., charcoal, bio-oils, ethanol, methane, hydrogen, etc.) (Zhang *et al.*, 2010).

Napier grass is one of the fodder crops that is planted most often in small-holder agricultural communities and also for use as fish food (Negawo *et al.*, 2017). It is a versatile fodder crop since it can be grazed directly or turned into hay or silage (Orodho, 2006). Young shoots of napier can be used as vegetable (Akah and Onweluzo, 2014).

As a crop, napier grass may serve as a windbreak, a living marker for the delineation of river buffer zones, and a source of fuel when dried out (Orodho, 2006). In the management of cropland systems, it serves as a mulch to prevent weed growth and soil erosion, and as a trap plant for pest control (Kabirizi *et al.*, 2013). It keeps the maize crop free from maize stem borer and popularly called pull-push crop (attract and repel the pests) (Joshi, 2015). Large biomass-producing plants like napier are being promoted more as a result of the effects of change, and it is called next-generation or second-generation biofuel crops (Phaenark *et al.*, 2009; Mahar *et al.*, 2016).

In Bangladesh, napier grass (*Pennisetum purpureum*) is grown mostly by farmers for the purpose of feeding dairy cows. It includes more crude protein compared to other forages. In the Barind region there is scarcity of water and napier grass is drought tolerant, they grow napier grass for the purpose of livestock feed. Moreover, napier grass is being popular in our country for being used as biofuel (Asaduzzaman, 2019).

## **2.2 Abiotic stress**

With the ever-increasing population, the security of food and feed has turned into a great concern nowadays. To assure this food and feed security, boosting production is very urgent. But due to climate change, a larger portion of the world's arable lands are facing various abiotic stress conditions (Gong *et al.*, 2020). The very common and significant environmental/abiotic stresses for crop productivity are drought, waterlogging, salinity, temperature extremes (high/low), high/low light intensity,



radiation (UVR, infrared, X-ray, etc.) stress, ozone, metal or metalloid toxicity, and other organic or inorganic pollutants (Hasanuzzaman *et al.*, 2019). These stresses hamper plant growth and when the stress is induced for longer period or in high intensity it may cause death to the plant which results in 70% loss of global crop production (Nahar *et al.*, 2017; Hasanuzzaman *et al.*, 2018a).

### **2.2.1 Plant responses to abiotic stress**

One of the main responses of abiotic stress is negative impact on crop growth and yield (Sehgal *et al.*, 2019; Rasheed *et al.*, 2020; Mehmood *et al.*, 2021). Additionally, it increases respiratory rates, reactive oxygen species (ROS), and electrolyte leakage (EL) (El-Banna and Abdelaal, 2018;). Abiotic stress conditions also interfere with the uptake of nutrients and water, assimilates synthesis, and partitioning, which significantly reduces yield (Abdelaal, 2015; Hassan *et al.*, 2020). Also causes lipid peroxidation, which damages proteins, the photosynthetic apparatus, cell membranes, and ultimately lead to cell death (Bhargava and Sawant, 2013).

The growth and morphology are hampered when plant exposed to various abiotic stress. When common bean exposed to salt stress, it was found that root and shoot biomass reduction was by 30 and 59% under 200 mM NaCl treatment compared to control (Taibi *et al.*, 2016). Leaf mortality increased with the increase of salt concentration at an early stage, critical decrease in plant biomass production along with shoot length, root length and number of roots occurred (Jiang *et al.*, 2016; Hussain *et al.*, 2017). Morphological parameters such as shoot length, ear length, and leaf area index (LAI) decreased with the increase of waterlogging duration and dry matter accumulation also declined under waterlogging in summer maize (Ren *et al.*, 2014). Shoot biomass of the cultivars under drought stress decreased by 19.77, 32.44, and 48.37% at 15, 30, and 45 DAF (days after flowering), respectively.

Physiology also hampered through abiotic stress. Chlorophyll content had been reduced by 52% under salt stress (Taibi *et al.*, 2016). Sesame (*Sesamum indicum* cv. BARI til-4) exposed to 2, 4, 6, and 8 days of waterlogging and showed that maximum reduction of RWC (70%) and proline (Pro) content (20%) of leaves were observed under a

prolonged waterlogging (8 days) (Anee *et al.*, 2019). Compared with the control, the relative water content (RWC) of three soybean leaves was reduced by under drought stress (Du *et al.*, 2020).

Biochemical responses under abiotic stress has seen in a great extent. Malondialdehyde (MDA) by 44% and at the same time, enzymatic and non-enzymatic activity was increased when exposed to salt stress (Taibi *et al.*, 2016.) Salinity-induced (150 mM NaCl) oxidative stress in faba bean is manifested by increased ROS production, especially H<sub>2</sub>O<sub>2</sub>, and higher activities of MDA and EL (Alzahrani *et al.*, 2019). Similarly, H<sub>2</sub>O<sub>2</sub>, MDA, EL, and O<sub>2</sub><sup>•-</sup> levels were two times higher than control plants in mung beans under salt stress (100 mM NaCl) (Ahmad *et al.*, 2019). Excessive ion accumulation and ROS production disturb plants' redox regulation under salinity stress (Tariq and Shahbaz, 2020).

At a molecular level, it was revealed that plants produce abiotic stress signalling chemical compounds called ROS such as singlet oxygen (<sup>1</sup>O<sub>2</sub>), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), superoxide (O<sub>2</sub><sup>•-</sup>) hydroxyl radical (HO•), etc. These compounds may be generated at a lower level under controlled conditions as the usually plant utilizes only 1-2% O<sub>2</sub> which favors normal growth and development without doing any damage (Noctor *et al.*, 2018). But when plants are exposed to any of the abiotic stress conditions, it generates an uncontrolled amount of ROS which causes rapid ROS burst and the system towards death due to the absence of ROS scavengers in sensitive crops. Tolerant genotypes have ROS scavenging genes and can endure oxidative stress through the synthesis of different non-enzymatic and enzymatic antioxidants (Mhamdi and Breusegem, 2018).

### **2.3 Drought**

The effects of the drought on plants will worsen with time. Drought alone lowers agricultural productivity annually, compared to all diseases (biotic stress) combined. Plants change their physiology, alter root development and architecture, and close stomata on their aboveground segments to respond to moisture gradients in soil (Gupta *et al.*, 2020). Drought stress has a complex impact on plant water relation traits such as

leaf water potential, RWC, stomatal conductance, temperature of the canopy, and the transpiration rate of different plant species (Fàbregas and Fernie, 2019).

### **2.3.1 Plant responses to drought stress**

According to plant genotypes and growth phases including germination, vegetative growth stage, reproductive growth stage, and maturity stage, the degree of drought-induced damage to crops and their responses vary, which reduces production. Drought induces a variety of morphological and physiological changes in plants, such as changed plant water relations, slower growth, lessened stem elongation, decreased leaf size, stomatal movement, and ion transport. photosynthesis, nutritional imbalance, increase water use efficiency, reducing transpiration and oxidation of membrane lipids, photosynthetic pigments (Engelbrecht *et al.*, 2007). Plant damage is mostly caused by proteins and nucleic acids, which due to the increased reactive oxygen species caused by dryness (ROS) generation that affects the redox state of cells (Nahar *et al.*, 2017).

#### **2.3.1.1 Effect of drought on crop growth**

Bhuiyan *et al.* (2019), conducted experiment with rapeseed under drought stress (20% PEG), rapeseed seedling height declined significantly (by 20% compared to control) along with the fresh weight (FW) and dry weight (DW) of the seedlings. Two genotypes of rapeseed were exposed to five levels of osmotic potentials ( $-0.1$ ,  $-0.2$ ,  $-0.3$ ,  $-0.4$ , and  $-0.5$  MPa) in an experiment, conducted by Khan *et al.* (2019). Root & shoot length, root-shoot length ratio, shoot fresh and dry weight and root fresh and dry weight was reduced significantly with the increase of osmotic potential in the case of both varieties. Of the two varieties, ZY 36 was more sensitive to drought stress than SG 127.

Qaseem *et al.* (2019) did an experiment with drought and heat stress in wheat plants. In case of drought stress (30% field capacity)- awn length, dry weight, leaf area, plant height, and the tiller number were reduced by 35, 21, 31, 7, and 18%., respectively compared to the control plants.

Ahmed *et al.* (2021) worked with two varieties of chickpea (*Cicer arietinum* L.) cv. BARI Chola-7 and BARI Chola-9 and induced 4 levels of drought stress. Shoot FW

and DW significantly decreased in both chickpea cultivars under drought stress. Shoot FW decreased by 36, 55, and 68%, respectively, in BARI Chola-7 and 41, 55, and 63% in BARI Chola-9 after exposure to D1(30% moisture content), D2, (20% moisture content) and D3 (10% moisture content) treatments. In comparison to control conditions, shoot DW decreased by 38, 49, and 61% in BARI Chola-7 and by 46, 50, and 70% in BARI Chola-9. In comparison to drought-stressed plants alone, Thiourea foliar application significantly raised shoot DW in D1 and D2-treated plants of BARI Chola-7 by 46 and 19%, respectively, and in BARI Chola-9 by 51 and 18%.

Rahman *et al.* (2021) conducted an experiment with different abiotic stress on the jute plant. In the case of drought stress, plant height decreased by 13 and 16% under mild (10 days of water deficit) and severe (15 days of water deficit) drought, respectively. In the case of other parameters like above-ground fresh weight and above-ground dry weight had decreased by 22, 30, and 16% over the control.

Saha *et al.* (2019) evaluated the detrimental effects of water stress (drought for 10 days) on the growth of five rice genotypes and found that root lengths were reduced by 24 to 45% in all genotypes after 10 days of treatment exposure, although BRRI Dhan-56 showed the smallest root. In addition, all five genotypes showed a decline in the ratio of shoots to roots, the length of the shoots, and fresh and dry weights. Similarly, Nasrin *et al.* (2020) observed that rice plants (BBRI dhan24) subjected to drought stress for 12, 15, 18 and 21 days decreased root length by 49, 71, 64.15 and 68%, respectively, , shoot length (28 to 47%) root fresh weight (95 to 98%), shoot fresh weight (84 to 93%), root dry matter (90 to 94%), and shoot dry matter (47 to 82%), compared to control. Additionally, drought stress reduced leaf area by reducing leaf length and width by 31 to 36% and 22 to 56%, respectively.

### **2.3.1.2 Effect of drought stress on crop physiology**

Upreti *et al.* (2021) conducted an experiment with two varieties of cluster bean (RGC-936, drought sensitive and RGC-1002, drought tolerant variety). With the drought stress progressed, RWC values decreased to 62.7 and 72.7% in RGC-936 and RGC-1002, respectively on the 10th day of water stress. Along with the RWC, the osmotic potential

was also reduced with drought stress and contributes towards maintenance of leaf turgor, at low RWC values. In variety RGC-936, osmotic potential decreased by 1.87 fold while in the variety RGC-1002 the decrease was by 2.08 fold on 10th day of drought.

An experiment was done by Tani *et al.* (2019) with forage grass (*M. arborea*, *M. sativa*, and Alborea). Under drought stress conditions, RWC and the gas-exchange parameters of net photosynthetic rate (Pn), stomatal conductance (gs), and transpiration rate (E) significantly decreased while the water use efficiency (WUE) rose. After three weeks of drought stress, the RWC of Alborea's leaves was around 29% lower than the control, whereas *M. sativa* and *M. arborea* showed corresponding reductions of about 19 and 15%, respectively. While having the lowest gs and E in both well-watered and water-deficient situations, the population of *M. arborea* had the greatest WUE. After two weeks of drought stress, gs and E progressively declined, but their WUE steadily increased.

Physiology was also hindered by the drought stress. In a study by Qaseem *et al.* (2019), chlorophyll content was decreased during and after anthesis and all stress treatments caused a significant reduction in chlorophyll content. During anthesis, chlorophyll content was reduced by 24% and after anthesis, it was reduced by 31% under drought (30% field capacity) compared to control. The relative water content was reduced by 55% compared to control.

In comparison to controls, the exposure to drought stress resulted in a significant reduction in chl a and chl b, as well as the overall quantity of chl content, which is also connected with carotenoid (Car), by 71, 60, 75, and 83%, respectively (Nawaz *et al.*, 2016). According to Hasanuzzaman *et al.* (2018c), the crop plants exposed to acute water stress (20% PEG), resulted in 52 and 43% reduction of chl a and total chl content, respectively, in the leaves of rapeseed plants compared to the control. However, coupled heat and drought stress (50% FC) decreased the levels of chl a, chl b, and total chl (Hussain *et al.*, 2019).

Under drought stress, the RWC of the leaves in both types of chickpea in BARI Chola-7 and BARI Chola-9, respectively, plants had the lowest RWC under extreme drought stress by 36 and 30% compared to control (Ahmed *et al.*, 2021).

### **2.3.1.3 Effect on yield and yield attributes**

Drought causes substantial physiological changes that delay or even stop growth and threaten the yield stability of the crop (Anjum *et al.*, 2011). As a result of the drought, field yield losses normally between 30 and 90% (Hussain *et al.*, 2019). The yield loss varies depending on the crop species, water sensitivity of the crop. Drought may have particularly significant effects on yield during various stages of crop development (Dietz *et al.*, 2021).

Fariaszewska *et al.* (2020) worked with nine varieties of forage grass under drought stress for two years and fodder yield had decreased with the increasing drought. The two *L. multiflorum* varieties ('Melmia' and 'Meldiva') had the highest total dry matter yield (DMY) in the control and drought circumstances, 20.2 and 22.8 t ha<sup>-1</sup>, respectively. In contrast, the *F. pratensis* variety had the lowest total dry matter yield for irrigated and non-irrigated plots. The biggest loss in total DMY under drought stress circumstances in 2014 was recorded for "Barolex" (10.1%), while "Meltador" even exhibited a very tiny rise in total DMY (6.7%). In the control conditions, "Barolex" had the highest total DMY, while "Felopa" had the lowest in 2015. 'Barolex' had the lowest drop in total DMY of 2015 following the drought season in cut III (7.8%), while 'Merifest Tp.' had the highest reduction (35.2%).

In comparison to well-watered controls, Anjum *et al.* (2017) found that under severe drought stress (40% FC), the kernels ear<sup>-1</sup>, weight of 100-grain, grain yield plant<sup>-1</sup>, and biological yield plant<sup>-1</sup> of three maize hybrids decreased by 2, 10, 13, and 6% in Dong Dan 80; 5, 14, 22, and 7% in Wan Dan 13; and 19, 24, 43, and 16% in Run Nong 35. Thus, Dong Dan 80 (6%), Wan Dan 13 (7%), and Run Nong 35 (16%) showed a substantial reduction in yield when all maize hybrids were subjected to drought stress.

Three genotypes of soybean, Shennong17 (CV.SN17), Shennong8 (CV.SN8), and Shennong12 (CV. SN12) were exposed to different duration of drought stress 15, 30, and 45 DAF (days of flowering). Seed weight decreased by 41.65% under drought stress compared to the control during the middle and late seed development stages (30–45 DAF).

### **2.3.2 Drought-induced oxidative stress and antioxidant defense system**

Because of drought, electron leakage, reactive oxygen species (ROS) generation, and disruption of solute transport is happened to create oxidative damage. These ROS interact with several cellular components, including Plants initiate various mechanisms to maintain normal homeostasis of cells, such as enzymatic and no enzymatic scavenging systems to protect cells from oxidative damage (Kabiri *et al.*, 2014). These enzymatic and no enzymatic scavenging systems are mediated by plants to detoxify the detrimental effect of drought stress. the nucleus, proteins, and membranes, affecting the integrity of those components.

According to Du *et al.* (2020), MDA content had been increased under drought stress compared to control. MDA concentration increment during drought stress was 52.86 to 90.11% in CV.SN17, 22.46 to 45.45% in CV.SN8 and 22.22 to 28.76% in CV.SN12 as compared to control during 15–45 DAF. Under drought conditions, proline concentrations were increased. Compared with the control, proline concentration had been increased by 33.23, 75.12, and 65.97% in CV.SN17, CV.SN8 and CV.SN12, respectively.

Amoah and Seo (2021) observed that wheat had the greatest increases in EL (64%), H<sub>2</sub>O<sub>2</sub> concentration (25%), and MDA (54%). Liu *et al.* (2019) found that the MDA contents rose considerably by 16% and led to ROS over-generation and cell lipid peroxidation in the imbibing seeds when drought-stressed (20% PEG, 5 d) plants were compared to control plants. According to Bhuiyan *et al.* (2019), compared to the control, rapeseed seedlings exposure to water stress had significantly enhanced MDA and H<sub>2</sub>O<sub>2</sub> contents by 82 and 131%, respectively. On the contrary, MDA and H<sub>2</sub>O<sub>2</sub>

levels in Xida 319 and Xida 889 maize hybrids were reduced by drought stress treatments compared to controls (Hussain *et al.*, 2019).

An experiment conducted by Khan *et al.* (2019) with six rapeseed genotypes (Zhongshuang 10, Huashuang 5, Zhongyou 36, Huaza 62, Shengguang 127, and Huaza 9) and five levels of water potential (-0.1, -0.2, -0.3, -0.4 and -0.5 MPa). Under the drought stress condition (-0.3 MPa) a sharp rise in H<sub>2</sub>O<sub>2</sub> content by 92%, MDA content by 181%, and EL by 112% for ZY 36 variety compared with SG 127 variety. When it considered for -0.5 MPa, 42, 35 and 47% increase in H<sub>2</sub>O<sub>2</sub>, MDA content and EL, respectively, was observed.

Gunes *et al.* (2007) observed that drought stress (40% FC) increased MDA content (25-68.7%) and H<sub>2</sub>O<sub>2</sub> content (55-133%) compared to control plants (60% FC). Patel *et al.* (2012) exposed four chickpea cultivars (Tyson, ICC 4958, JG 315, DCP 92-3) to early drought stress (EDS) and late drought stress (LDS) to study the chickpea genotypes' most responsive development stage to drought stress. They found that H<sub>2</sub>O<sub>2</sub> and MDA contents increased compared to water as usual. However, the difference between EDS and LDS was more significant, indicating that the pre-anthesis stage was more vulnerable to oxidative stress than the post-anthesis period.

## **2.4 Waterlogging**

The definition of waterlogging is “the condition of land where the subsoil water table is located at or near the surface with the result that the yield of crops typically grown on the land is reduced well below for the land, or, if the land is not cultivated, it cannot be put to its normal use due to high subsoil water” (FAO, 2015). Flooding and waterlogging have negative consequences for global agriculture production. Waterlogging can be two types, viz; submergence and flooding. Flooding can create hypoxia (lack of oxygen) and anoxia (absence of oxygen). Where there is heavy precipitation, inadequate soil drainage (Sundgren *et al.*, 2018), and considerable variations in the high groundwater level, waterlogging (WL) or flooding frequently occurs (Ren *et al.*, 2016a). Reduced plant growth potential and productivity under waterlogging stress are caused by changes in root hydraulic conductivity, light



interception, stomatal conductance, CO<sub>2</sub> assimilation, drastically reduced photosynthesis, altered respiration, and production of a variety of secondary metabolites (Ashraf, 2012). The metabolic activities of plants growing under waterlogged conditions may be hampered by inhibitions of respiration and the production of toxic compounds. Plants grown in waterlogged environments have been found to have stunted growth and lower biomass.

### **2.4.1 Plant responses to waterlogging stress**

Additionally, waterlogging is known to have negative effects on plant physiological processes, such as decreased cell permeability (Zhang *et al.*, 2021), decreased root activity and respiration (Kaur *et al.*, 2020), and antioxidant inhibition, which causes stomata to close, lower leaf chlorophyll concentrations, and a decrease in the net photosynthetic rate (Pn) (Tian *et al.*, 2021). Lower soil oxygen levels, which restrict both carbon and nitrogen metabolism and negatively impact the availability of soil nutrients, especially nitrogen, may be the cause of the decline in plant growth following waterlogging (Herzog *et al.*, 2016; Zhang *et al.*, 2021). Particularly waterlogged plants experience N shortage as a result of increased denitrification and mobile nitrogen leaching as well as slowed rates of N mineralization brought on by the excessive soil moisture (Kaur *et al.*, 2020).

#### **2.4.1.1 Effect of waterlogging on crop growth**

Zeng *et al.* (2020) did an experiment with 39 peanut ecotypes. At various growth phases, such as the S stage (four-leaf stage), the F stage (50% flowering and needling), and the P stage (pod-filling stage), waterlogging treatments were applied for 5, 10, or 15 days (from the beginning of the seed to the full seed). At the S and F phases, waterlogging for 5 and 10 days enhanced the stem dry weight (SDW) and leaf dry weight (LDW) of the three ecotypes. The maximum decline in total dry weight (TDW), SDW, LDW, and pod dry weight (PDW) for all ecotypes also occurred at the P stage, where maximum declines in TDW, SDW, LDW, and PDW for Zhanhong 2 were 47.64, 19.94, 41.21, and 69.78%, respectively, while for Zhongkiahua 1 they were 35.06, 7.47, 43.50, and 43.60%, and for Huayu 39 they were 49. The TDW and LDW of Zhanhong

2 grew by 4.76 and 23.20% after 5 days of therapy at the P stage, whereas those of Zhongkaihua 1 climbed by 14.22 and 13.49%, and those of Huayu 39 declined by 49.14 and 57.04%, respectively.

Ren *et al.* (2014) investigated several phases of summer maize under conditions of waterlogging. At the three-leaf stage (V3), six-leaf stage (V6), and the tenth day after the tasseling stage (10VT) of maize, the caused waterlogging for varying lengths (3 and 6 days). After two years of research, the findings showed that waterlogging severely impacted summer maize's overall growth and development.

Rahman *et al.* (2021) showed that plant height had decreased by 38 and 41% under mild (waterlogged for 5 days) and severe (waterlogged for 15 days) waterlogging, respectively. Above-ground fresh weight and above-ground dry weight decreased by 35 & 38%, and 45 & 48% over the control.

Prasanna and Rao (2014) demonstrated a significant variance in the growth parameters of mung bean cultivars in waterlogged condition. Throughout the whole life cycle, waterlogging significantly affected plant height, leaf area, leaf number, and total dry matter. The impact of a 4-day waterlogging was more severe than a 2-day waterlogging treatment above the control. Waterlogging caused a reduction in plant height, branch number, leaf number, and total dry matter by 33, 34, 31, and 31%, respectively.

Pais *et al.* (2021) conducted an experiment with 4 different genotypes of bread wheat and waterlogging stress was imposed at the tillering stage for 14 days. In the case of G3 and G4 didn't show any significant decrease under waterlogging but the SPAD value was decreased by 29 and 80% in G1 and G2, respectively.

#### **2.4.1.2 Effect of waterlogging on crop physiology and metabolism**

Waterlogging conditions cause disruptions in plant physiology and metabolism. Under conditions of waterlogging, plants exhibit a variety of reactions, such as reduced stomatal conductance, net CO<sub>2</sub>-assimilation rate, and root hydraulic conductivity (Ashraf, 2012), as well as decreased photosynthetic rate (Akhtar and Nazir, 2013).

Bajpai and Chandra (2015) conducted an experiment with sugarcane in two varieties for 0, 48, and 96 hours of waterlogging and one for recovery. Relative water content rose in both varieties as waterlogging time increased; it varied from 84.7 to 90.2% in variety V1 and 86.7 to 89.6% in variety V2. RWC content after recovery therapy seemed normal. Chl a, Chl b, and carotenoids concentration in leaf tissues were lowered by waterlogging. Maximum reduction in varieties V1(early maturing variety) and V2 (late maturing variety) was noticed at 96 hours. Chl a & b, and carotenoid concentrations in the Recovery treatment variety V1 did not demonstrate additional improvement.

At various developmental phases, the SPAD values of Zhanhong 2 and Zhongkaihua 1 dropped as the duration of waterlogging increased. A significant difference from CK under waterlogging at the S (seedling stage) and P (pod-filling stage) stages lowered the SPAD value for Huayu 39 but waterlogging at the F stage (flowering stage) for 5 days and 10 days enhanced the SPAD value. Waterlogging during 10 days and 15 days during the P stage decreased the SPAD value in Zhanhong 2, Zhongkaihua 1, and Huayu 39 by 21.08, 24.22, 20.25, and 12.22%, respectively (Zeng *et al.*, 2020).

To know the effect of waterlogging on different genotypes of soybean Sathi *et al.* (2022) conducted an experiment. When exposed to waterlogging, plants exhibited a reduction in leaf RWC. The lowest reduction in leaf RWC was observed in waterlogged BARI Soybean-5 (17%) and the highest reduction was in waterlogged BINA soybean-5 (42%) in comparison with the control condition. The reduction ranged between 18 and 41% in other genotypes when compared with their respective control plants.

Kumar *et al.* (2013) worked with four genotypes of mung bean varying in their waterlogging tolerance. Tolerant genotypes (T-44 and MH-96-1) maintained significantly greater RWC and membrane stability index (MSI) than sensitive genotypes (MH-1K-24 and Pusa Baisakhi) under waterlogging conditions. Under waterlogging stress, the rate of photosynthesis reduced in all genotypes that were studied, and these inhibitions became worse with time. Stomatal conductance showed the same trend as the photosynthetic rate. After 48 hours of waterlogging, leaf respiration grew more than in control plants. The leaf respiration rates of MH-96-1 and Pusa Baisakhi were higher than those of the other genotypes. On the sixth and ninth

days of waterlogging, a little reduction in respiratory rate was noted. However, during the waterlogging treatment phase, the rate of leaf respiration in T-44 did not change. Therefore, they came to the conclusion that no studied length of waterlogging resulted in a reduction in leaf respiration.

By the closure of stomata and lowering of PSII efficiency, flooding stress increased the number of leaves but lowered photosynthetic activity in the *Cichorium intybus* plant. Compared to control plants, the roots of wet plants shrank. However, there were no appreciable variations in FW and DW. While the leaves of the flooded plant gathered organic acids and reducing sugars, the roots accumulated glucose, fructose, sucrose, and 1-kestotriose. Due to flood stress, invertase and sucrose synthase activities increased in both leaf and root whereas sucrose-phosphate synthase activity did not alter. Because fructan: fructan 1-fructosyltransferase was suppressed, inulin synthesis was delayed in the roots of flooded plants, and its mean degree of polymerization was reduced (Vandoorne *et al.*, 2014).

#### **2.4.1.3 Effect of waterlogging on yield and yield contributing parameters**

Six wheat genotypes were subjected to flooding stress for 28 days, and all had significant grain production reductions. When compared to rainfed circumstances (control), waterlogging caused a grain yield reduction of 56% on average, with cultivars Ariana and Vaga seeing the largest reduction (74%) and Salammbô and Utique experiencing the smallest reductions (39%). The two cultivars, FxA and Hadra, had a mixed pattern of behavior with corresponding declines of 60% and 48% (Amri *et al.*, 2014).

In an experiment with four genotypes of mung bean viz., two tolerant (T 44, and MH-96-1), and two sensitive (Pusa Baisakhi, and MH-1K-24) mung bean genotypes under waterlogging stress, Kumar *et al.* (2013) discovered that yield was impacted by waterlogging in all the genotypes. The average grain yield losses in all mung bean cultivars were 20.01, 33.79, and 52% owing to 3, 6, and 9 days of waterlogging, respectively. At the vegetative stage, yield loss increased in a duration-dependent

manner. The grain production losses brought on by three days of waterlogging might be recovered by the tolerant genotypes. However, even three days of waterlogging reduced the yield (by up to 20%) for sensitive genotypes. After 9 days of waterlogging, sensitive genotypes had grain yield reductions estimated to range from 70.0 (Pusa Baisakhi) to 85% (MH-1K-24) compared to control plants.

In wheat, Rasaei *et al.* (2012) found a significant difference among all the waterlogging stress periods (10, 20, and 30 days). Even though 30 days of waterlogging had the worst effects on grain output, 10 and 20 days of waterlogging significantly differ from a non-waterlogged state. After 10, 20, and 30 days of waterlogging stress, when plants allowed to recover, the yield of wheat was 7518.4 kg/ha, 6815.5 kg/ha, 5587 kg/ha, and 4138.6 kg/ha, respectively.

To investigate the effects of waterlogging on summer maize development and yield at the three-leaf stage (V3), six-leaf stage (V6), and the tenth day after the tasseling stage, Ren *et al.* (2014) conducted an experiment in the field (10VT). The findings showed that the degree of stress (intensity and duration) and the various growth phases affected maize development and grain production responses to waterlogging. During V3 and V6, yield drastically declined as waterlogging duration increased. In treatments V3-3, V3-6, V6-3, V6-6, 10VT-3, and 10VT-6, the yields of the maize hybrid Denghai 605 (DH605) were, correspondingly, 23, 32, 20, 24, 8, and 18% lower than those of the control; the yields of Zhengdan 958 were decreased by 21, 35, 15, 33, 7, and 12%, respectively, compared to control.

#### **2.4.2 Waterlogging-induced oxidative stress and antioxidant defense system**

Reactive oxygen species are produced in various forms and subcellular compartments due to waterlogging (Jaspers and Kangasjärvi, 2010). Reactive oxygen species are created in transition when a plant or plant component either enters hypoxia/anoxia from normoxic conditions or returns to an aerobic environment (Irfan *et al.*, 2010).

Under typical aerobic circumstances, lipid peroxidation—another potential generator of ROS and other radicals—is a normal metabolic process. Moreover, it is one of the

most thoroughly investigated effects of ROS activity on membrane composition and function. Several researchers have found higher H<sub>2</sub>O<sub>2</sub> buildup and enhanced lipid peroxidation in anaerobic environments (Hasanuzzaman *et al.*, 2012a, b; Sairam *et al.*, 2011). The primary cause of oxidative stress is the inability of the scavenging system to metabolize the hazardous active oxygen due to either increased ROS production or reduced activity of the scavenging enzymes (Yordanova *et al.*, 2004).

Asha *et al.* (2021) conducted an experiment with two genotypes of maize for 10 days of waterlogging. Both the sensitive genotypes Popcorn and BM-6 saw a significant rise in H<sub>2</sub>O<sub>2</sub> as a marker of oxidative stress, but the tolerant genotype BHM-9 experienced a drop and the tolerant genotype BHM-13 experienced a minor increase. Under waterlogging stress, the susceptible genotype BM-6 showed the largest rise in H<sub>2</sub>O<sub>2</sub> concentration (9.05 times) as compared to the control. Under waterlogging stress, all tolerant and susceptible genotypes had higher levels of APX and POD as compared to the control, although two tolerant genotypes, BHM-9 and BHM-13, had relatively high levels than the two susceptible genotypes, Popcorn, and BM-6. Under waterlogging treatment, the moderately tolerant genotype BHM-9's content of APX increased by 3.67 times, that of POD by 2.33 folds, and that of POD by 2.46 folds in the tolerant genotype BHM-13 compared to control.

## **2.5 Effect of water stress on napier grass**

### **2.5.1 Effect of water stress on napier growth**

Three napier cultivars were used in an experiment by Sarker *et al.* (2021) at two distinct sites. Cultivar and location showed a highly significant impact on plant height. Regardless of location, BN-1 (171.2 cm) had the maximum plant height. However, regardless of cultivar, the non-drought site (174.6 cm) had the highest plant height when compared to the drought location. The stem height of four napier cultivars described in a recent study by Maleko *et al.* (2019) ranged from 145.44 to 210.81 cm with substantial differences among them. While there were no significant differences in the number of tillers per hill between cultivars the number of tillers at the non-drought site (28.1

no/hill) was substantially greater than the number at the drought location. (Maleko *et al.*, 2019).

The same type of effect was also studied by Sarker *et al.* (2021) and showed that the differences in leaf stem ratio (LSR) across cultivars are caused by features of the stem and leaves. While some types have stout stems and few leaves, others have narrow stems and many leaves. The maximum LSR, regardless of cultivar, was found in BN-3 (0.86) and at the drought site (0.95).

### **2.5.2 Effect of water stress on the physiology of napier**

Cardoso *et al.*, (2015) conducted an experiment with two forage grasses under drought stress. Similar to other C<sub>4</sub> grasses, napier grass and Mulato II the effects of the drought on growth were unaffected conditions. This implies that the gas exchange in leaves, stomatal control, rather than other mechanisms, primarily regulates water loss and plant growth during drought conditions both types of grass' stomatal density decreased. Moreover, the stomata density was 1.4 times higher. However, levels of leaf gas were higher in Mulato II than in napier grass after the trial. In non-stressed plants, the interchange was comparable. Stomata size in the leaves of napier grass was more likely to be greater than Mulato II.

### **2.5.3 Effect of water stress on napier yield**

The herbage mass of plants is affected by all growth factors of plant. The result of this study showed that the interaction of drought stress and kind grasses have not affected herbage mass. Furthermore, the kind of grasses treatment has a significant effect on herbage mass. Decreasing napier grass herbage mass have no significant difference with all treatment of period stress, while in guinea grass herbage mass significance decreased by DS258 treatment. Forage yield decreased the effect of drought stress. Guinea grass decreased 25.9% after three times stress compared to control. Napier decreased 22.20% forage yield compared to the control (Purbajanti *et al.*, 2012)

#### **2.5.4 Effect of water stress on the quality of napier**

The experiment of Sarker *et al.* (2021) showed that location had no significant ( $P > 0.05$ ) effect on DM contents in all botanical fractions of napier cultivars. Maleko *et al.* (2019) reported leaf, stem, and total DM contents in four varieties of napier to be ranged from 17.44% to 22.87%, 8.29% to 14.7%, and 12.87% to 18.78%, respectively which are agreed with our investigation. DM content largely depends on plant maturity and moisture in the soil. The importance of soil moisture to plant growth was highlighted by the marked reduction of dry matter yields.

The crude protein (CP) contents in whole plant and stem were found to have significant variation for the effect of cultivar, while it was not varied significantly in leaf. The highest CP contents in the whole plant (10.69%) and stem (6.21%) were obtained in BN-1 and BN-3, respectively. On the other hand, the location had a highly significant effect on CP contents in all botanical fractions. The highest CP contents in all botanical fractions were estimated at the non-drought location. Regardless of cultivar and location, the least squares mean of CP contents in the whole plant, leaf and stem were 9.96, 12.26, and 5.89%, respectively (Sarker *et al.*, 2021). According to Ishrath *et al.*, (2018), the napier hybrid in general contains about 10.2% CP, which closely agrees with our study. In another study, CP contents in 3 napier hybrid cultivars to be 16.5 to 17.2% in the leaf, 3.6 to 5.6% in the stem, and 10.4 to 11.2% in the whole plant, which mostly agrees with this study, except that of CP content in leaf.

The ash contents in all botanical fractions of the plant were found to have no significant ( $P > 0.05$ ) effects for cultivars, while those varied significantly ( $P < 0.001$ ) for the direct effect of location. Regardless of cultivar and location, the least squares mean of ash contents in the whole plant, leaf and stem were 13.81, 12.67, and 14.54%, respectively. Rengsirikul *et al.* (2013) reported ash concentration to be 10.9 to 15.9% in napier harvested in dry seasons. Ash contents in different hybrid Napier cultivars ranged from 11.62 to 13.07% in leaves and 12.30 to 13.90% in the stem. In contrast, Maleko *et al.* (2019) reported comparatively lower estimates of ash contents in four varieties of napier cultivar ranging from 7.96 to 9.38%.



The acid detergent fiber (ADF) contents in all botanical fractions were found to have highly significant effects on the cultivar. The lowest ADF contents in all botanical fractions were found in BN-3. On the other hand, the location had no significant effect on ADF contents in all botanical fractions, which agrees well with. Regardless of cultivar and location, the least squares mean of ADF contents in the whole plant, leaf and stem were 43.77, 42.31, and 45.67%, respectively. Earlier, Elanchezhian and Reddy (2009) obtained 39.57% ADF in the hybrid Napier cultivar (CO3) at the green stage.

The lowest NDF contents in all botanical fractions were found in BN-3. On the other hand, the location had a significant effect on ADF contents in the whole plant and stem, being lowest at the non-drought location, while ADF content in the leaf was not varied significantly. However, NDF values as found in our study followed within the range of 45 to 65% which is regarded as roughage feed of moderate quality (Turano *et al.*, 2016)

## CHAPTER III

### MATERIALS AND METHODS

This chapter shows a short description of the experimental period, site description, climatic condition, crop or planting materials, treatments, experimental design and layout, crop growing procedure, fertilizer application, uprooting of seedlings, intercultural operations, data collection, and statistical analysis.

#### 3.1 Location

The experiment was conducted at the experimental shed of the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka ( $90^{\circ} 77'$  E longitude and  $23^{\circ} 77'$  N latitude), Bangladesh, during the period from April 2021 to July 2021. The location of the experimental site has been shown in Appendix I.

#### 3.2 Soil

The soil of the experimental pot belonged to the Modhupur tract (AEZ No. 28). It was a medium-high land with non-calcareous dark grey soil. The pH value of the soil was 5.7. The physical and chemical properties of the experimental soil have been shown in Appendix II.

#### 3.3 Weather

The experimental area was under the subtropical climate and characterized by high temperature, high humidity, and heavy precipitation with a chance of a blast of winds during the period from April to July. The detailed meteorological data in respect of air temperature, relative humidity, rainfall, and sunshine hour recorded by the meteorology center, Dhaka for the period of experimentation have been presented in Appendix III.

## **3.4 Materials**

### **3.4.1 Plant materials**

Super napier seed was used in this experiment. It is the hybrid napier grass obtained by crossing elephant grass and pearl millet. It can be grown in Kharif-I and Kharif-II seasons. Super napier can be up to 400-500 cm in height. It has long leaves in size and leaves are also broader (6-8 cm). High leaf bearing capacity with 400 to 450 leaves per pitch. Fodder can be harvested eight times a year. The yield is 150-200 tons per annum.

### **3.4.2 Plastic pot**

14 liters' plastic pots with 18-inch depth and 14-inch diameter were used for this experiment. Thirteen kilograms of sun-dried and sterilized soils along with organic manures (cow dung, vermicompost) and fertilizers (nitrogen, phosphorus, potassium, sulfur) were put in each pot according to their doses. After that, the all components were mixed properly and pots were prepared for seed sowing.

## **3.5 Treatments**

1. Control
2. D1 (Drought for 7 days; at 21-28 DAS)
3. D2 (Drought for 14 days; at 21-35 DAS)
4. D3 (Drought for 21 days; at 21-42 DAS)
5. WL1 (Waterlogging for 7 days; at 21-28 DAS)
6. WL2 (Waterlogging for 14 days; at 21-35 DAS)
7. WL3 (Waterlogging for 21 days; at 21-42 DAS)

Treatments were applied at the 21 days after sowing (DAS) for different durations. Plants were grown under field capacity condition up to 21 days after sowing. Then both drought and waterlogging stress was imposed to the plants. For drought stress, Moisture content was 20, 10, 5 % for 7, 14 and 21 days durated drought, respectively. For waterlogging, water level was 6-8 inches higher than root level of plants and maintained for 7, 14 and 21 days, respectively.

### 3.6 Design and layout of the experiment

The experiment was laid out in a Completely Randomized Design (CRD) with four replications. So, the total number of pots was 28.

### 3.7 Seed collection

The seed was collected from the local seed market. The variety used in this experiment was Bakshi Bran (Super Napier Grass).

### 3.8 Pot preparation

The collected soil was sun-dried, crushed, and sterilized properly. Then the soil, organic manure, and fertilizers were mixed properly, and then placed the soil into the pot. Each pot was filled with 13 kg of mixed soil. Pots were placed at the shed house of Sher-e-Bangla Agricultural University. Leveling was done to the pot for each treatment. Finally, water was added to bring the soil water level to the field capacity.

### 3.9 Fertilizer application

Fertilizers used in the experimental pots were organic manure, urea, triple super phosphate, muriate of potash, and gypsum at the recommended dose. The whole amount of fertilizers was incorporated with soil at final pot preparation before sowing. Fertilizer doses are as follows:

Fertilizers	Doses (kg ha <sup>-1</sup> )	Actual amount per pot (g)
Urea	45	0.978
Triple super phosphate	12	0.60
Muriate of potash	15	0.30
Gypsum	5	0.28

### **3.10 Seed sowing technique**

Sixteen healthy seeds were sown in each pot. Before sowing seeds were soaked in water for 24 hours. After germination, 10 plants were allowed to grow in each pot.

### **3.11 Intercultural operations**

#### **3.11.1 Gap filling and thinning**

After sowing seeds continuous observation was kept. It was observed that some seeds failed to germinate. So, there was a need for gap filling. Thinning was done to maintain 10 seedlings per pot. Thinning was done to maintain the required spacing of the plants.

#### **3.11.2 Weeding and irrigation**

Occasionally, there were some weeds in pots that were uprooted manually. Irrigation was given to maintain field capacity moisture level.

#### **3.11.3 Plant protection measure**

There was infestation through leaf roller at a very early stage. So, insecticide had been sprayed 3 times at 3 days' intervals.

### **3.12 General observation of the experimental pots**

The experimental pots were regularly observed and they looked normal green. The growth was not uniform in case of drought and waterlogged plants compared to control.

### **3.13 Data collection**

Data were collected after the completion of each treatment. As there were three different duration of treatment, data were also taken. All types of data were taken after each stress duration and also at 50 days after sowing. Qualitative parameters were taken at 50 days after sowing only.

#### **3.13.1 Crop growth parameters**

- Plant height
- Root length
- Root-shoot ratio
- Shoot fresh weight
- Root fresh weight
- Shoot dry weight
- Root dry weight

#### **3.13.2 Physiological parameters**

- SPAD value
- Relative water content

#### **3.13.3 Oxidative stress indicators**

- Proline content
- Malondialdehyde (MDA)
- Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) content

### **3.13.4 Qualitative parameters**

- Acid Detergent Fibre
- Neutral Detergent Fibre
- Ash content
- Dry matter
- Crude Protein
- Crude fiber
- Moisture
- Organic matter content
- Hemicellulose

### **3.14 Procedure of sampling for growth study during the crop growth period**

#### **3.14.1 Plant height**

The height of the napier plants was recorded after the completion of the duration of treatment. Five plants were sampled randomly and height from the above ground up to the peak of the plants was recorded. The average of these 5 five plants had been considered as the plant height of the plants. Similarly, plant height data was recorded at 50 days after sowing for all together treatments.

#### **3.14.2 Root length**

The root length of the napier plants was recorded after the completion of the duration of treatment. Three plants were uprooted from each pot randomly and washed properly with clean water. Then the roots were cut from the plant and measured with a scale. Averaged the value and considered root length. Similarly, plant height data was recorded at 50 days after sowing for all together treatments.

### **3.14.3 Shoot fresh weight**

The shoot fresh weight of the napier plants was recorded after completion of the duration of treatment. Three plants were uprooted to measure the root length. After cutting the root from the uprooted plants, shoots were weighed through balance. The value of shoot fresh weight was averaged to have the shoot fresh weight per plant. Similarly, shoot fresh weight was recorded at 50 days after sowing for all treatments.

### **3.14.4 Shoot dry weight**

The shoot dry weight of the napier plants was recorded after completion of the duration of treatment. After measuring the shoot's fresh weight, the samples were dried in an electric oven at 80 °C for 72 hours. Then the samples were weighed through electric balance and averaged the values which were considered as shoot dry weight per plant. Similarly, shoot dry weight were recorded at 50 days after sowing for all treatments.

### **3.14.5 Root fresh weight**

The root fresh weight of the napier plants was recorded after completion of the duration of treatment. The root from the uprooted plants was measured through balance to have root fresh weight. Then the value was averaged and considered as root fresh weight per plant. Similarly, root fresh weight data were recorded at 50 days after sowing for all together treatments.

### **3.14.6 Root dry weight**

The root dry weight of the napier plants was recorded after the completion of the duration of treatment. After measuring the root's fresh weight, the samples were dried in an electric oven at 80 °C for 72 hours. Then the samples were weighed through electric balance and averaged the values which were considered as root dry weight per plant. Similarly, root dry weight data were recorded at 50 days after sowing for all together treatments.



### **3.14.7 Root-shoot ratio**

The root-shoot ratio of the napier plants was recorded after the completion of the duration of treatment. The proportion of shoot length and root length is considered as the root-shoot ratio. The following formula was used to calculate the value:

$$\text{Root-shoot ratio} = \frac{\text{Root length}}{\text{Shoot length}}$$

Similarly, the root-shoot ratio was recorded at 50 days after sowing for all treatments.

### **3.14.8 Root branch**

The root branch of the napier plants was recorded after the completion of the duration of treatment. Root branches were counted from the uprooted plant of three samples from each treatment. Then, the number was averaged and considered as a root branch number. Similarly, the root branch was recorded at 50 days after sowing for all together treatments.

## **3.15 Procedure of sampling for physiological parameter**

### **3.15.1 SPAD value**

The SPAD value of the napier plants was recorded after the completion of the duration of treatment. Five leaves were randomly selected from each pot. The top and bottom of each leaf were measured with atLEAF (FT Green LLC, USA) as atLEAF value. Then it was averaged and the total chlorophyll content was measured by the conversion of the atLEAF value into SPAD units and then total chlorophyll content was measured. Similarly, the SPAD value was recorded at 50 days after sowing for all together treatments.

### 3.15.2 Relative water content

The relative water content of the napier plants was recorded after the completion of the duration of treatment. Relative water content (RWC) was calculated according to Barrs and Weatherly (1962). Three fully expanded leaves were collected from each treatment and weighed the leaves. That weight was considered fresh weight (FW). Then the leaves were floated in distilled water in Petri dishes and kept for 24 hours in a dark place. After 24 hours, the leaves were weighted again removing the excess water with soft tissue and that weight was turgid weight (TW). The leaves were dried in an electric oven at 80 °C for 24 hours and weighed to measure dry weight (DW). Then RWC was calculated as follows-

$$\text{RWC (\%)} = \frac{\text{FW}-\text{DW}}{\text{TW}-\text{DW}} \times 100$$

Similarly, relative water content was recorded at 50 days after sowing for all treatments.

### 3.16 Procedure of sampling for biochemical parameter

#### 3.16.1 Measurement of proline content

Proline content in leaf tissue was calculated by following the protocol of Bates *et al.* (1973). Fresh leaf tissue (0.5 g) was collected from each treatment and kept in ice-cold condition. Then the leaf tissue was homogenized well in 5 mL of 3% sulfosalicylic acid also in ice-cold condition and the homogenate was centrifuged at 11,500×g for 15 minutes. One mL of the filtrate was mixed with 1 mL of acid ninhydrin (1.25 g ninhydrin in 30 mL glacial acetic acid and 20 mL 6 M phosphoric acid) and 1mL of glacial acetic acid. Then the total mixture was incubated in a water bath maintaining 100 °C for 1 hour. The incubated mixture was transferred into a test tube and kept in ice for cooling. After that 4 mL toluene was added to the cooled mixture and mixed thoroughly by vortex mixture. After some time, the chromophore containing toluene was read spectrophotometrically at 520 nm wavelength by transferring it to the upper aqueous layer. To measure blank, toluene was used. The proline content was

determined by comparing it with the standard curve of known concentration of proline and it was expressed through  $\mu\text{m proline g}^{-1}\text{ FW}$ .

### **3.16.2 Measurement of lipid peroxidation**

Lipid peroxidation was calculated by measuring the Malondialdehyde (MDA) content in fresh leaf tissue using the method of Heath and Packer (1968) with little modifications as described by Hasanuzzaman *et al.* (2012b). A fresh leaf sample (0.5 g) was collected from each treatment and homogenized in 3 mL of 5% (w/v) trichloroacetic acid (TCA) in ice-cold condition and then the homogenate was centrifuged at  $11,500\times g$  for 15 minutes. After centrifuging the supernatant (1 mL) was mixed with the thiobarbituric acid (4 mL, TBA) reagent. Then the mixture was heated at  $95\text{ }^{\circ}\text{C}$  for 30 min in a water bath machine and then cooled in an ice bath again centrifuging was repeated for 10 minutes. The absorbance reading of the colored supernatant was read spectrophotometrically at 520 nm and corrected by submitting the absorbance reading which was read at 600 nm. MDA content was calculated by using extinction coefficient  $155\text{ mM}^{-1}\text{ cm}^{-1}$  and expressed as  $\text{nmol g}^{-1}\text{ FW}$ .

### **3.16.3 Determination of hydrogen peroxide content**

Hydrogen peroxide determination was done by following the method given by Yu *et al.* (2003). Fresh leaf tissue (0.5 g) was collected at kept in ice-cold condition. Then the leaf tissue was homogenized properly with 3 mL of 50 mM potassium– phosphate (K–P) buffer (pH 6.5) at  $4\text{ }^{\circ}\text{C}$ . The homogenate was centrifuged at  $11,500\times g$  for 15 minutes. The centrifuged 2 mL supernatant was mixed with  $666.4\text{ }\mu\text{L}$  of 0.1%  $\text{TiCl}_4$  in 20%  $\text{H}_2\text{SO}_4$  (v/v) and was kept at room temperature for 10 minutes. After that, the mixture was again centrifuged at  $11,500\times g$  for 12 min. Then spectrophotometrically the supernatant was read at 410 nm to determine  $\text{H}_2\text{O}_2$  content using extinction coefficient  $0.28\text{ }\mu\text{M}^{-1}\text{ cm}^{-1}$  and was expressed as  $\text{nmol g}^{-1}\text{ FW}$ .

### **3.17 Procedure of sampling and measuring the fodder yield**

#### **3.17.1 Fodder yield**

The fodder yield of the napier plants was measured at 50 days after sowing for all together treatments. Three plants were randomly selected from each pot. Then the value is made average to get fodder yield per pot.

### **3.18 Procedure of sampling and measuring the qualitative parameter**

#### **3.18.1 Acid detergent fibre**

According to the method of Van soest *et al.*, (1991), one gram (1 g) of the air-dried sample was ground and passed to a 20 to 30 mesh (1 mm) screen. Then, 100 mL cold (room temperature) acid detergent solution and 2 mL decahydronaphthalene was added and heated for 5 to 10 minutes. To avoid foaming, the heat was reduced when boiling began; refluxed for 60 minutes from the onset of boiling, and then adjusted boiling to a slow, even level. Then the solution was filtered on a previously tared Gooch crucible (set on the filter manifold) and used the light suction. The filtered mat broke up with a rod and washed two times with hot water (90°-100°) and rinsed the sides of the crucible in the same manner. Repeated wash was done with acetone until it removes no more color; broke up all lumps so that the solvent comes into contact with all particles of fiber. Another optional wash was done with hexane. Hexane has been added while the crucible still contains some acetone. (Hexane can be omitted if lumping is not a problem in lignin analysis.) Sucked the acid-detergent fiber free of hexane and dry at 100° C. for 8 hours or overnight and weigh. The acid-detergent fiber was calculated by following-

$$\text{Acid-detergent fiber} = (W_o - W_t) (100) / S$$

Here,  $W_o$ =weight of oven-dry crucible including fiber;

$W_t$ =tared weight of oven-dry crucible;  $iS$  = oven-dry sample weight

### **3.18.2 Neutral detergent fibre**

According to the method of Van soest *et al.*, (1991). In a beaker with 1 0 0 ml of ND and 50 p1 of heat-stable amylase (dietary fiber kit; Sigma catalog number A3306) added before the beaker is put on heat, a 0.5-g sample is heated to boiling. If necessary, sodium sulfite (.5 g) is now added. The sample is boiled for one hour and then filtered using Whatman 54 paper (Whatman, Clifton, NJ) or a prepared coarse sintered glass crucible. The ash content should be recorded or left out of the NDF due to the different soil compositions in forages and feeds. The starch-specific enzyme is AOAC-approved, stable to boiling, and insensitive to EDTA. Samples must be ground through a 1-mm screen, but not any further because doing so might exacerbate filtering problems.

### **3.18.3 Dry matter**

At the time of silo filling and after ensiling, samples were collected and subjected to physical and proximate analysis. After about 1.5 months, the ensiled materials went a physical test to assess the smell/odor, texture, color, and percentage of deterioration. To calculate the dry matter and moisture contents, samples of various weights were oven- and sun-dried. Each treatment's remaining ensiled samples from each treatment were dried at 70 °C until they reached a consistent dry weight. Following the AOAC (2005) guidelines, dried samples were crushed to a 40 mesh size before being evaluated for dry matter.

### **3.18.4 Crude protein**

At the time of silo filling and after ensiling, samples were collected and subjected to physical and proximate analysis. After about 1.5 months, the ensiled materials went a physical test to assess the smell/odor, texture, color, and percentage of deterioration. To calculate the dry matter and moisture contents, samples of various weights were oven- and sun-dried. For each treatment's remaining ensiled samples the remaining ensiled samples from each treatment were dried at 70 °C until they reached a consistent dry weight. Following the AOAC (2005) guidelines, dried samples were crushed to a 40 mesh size before being evaluated for crude protein (CP).

### **3.18.5 Crude fiber**

Crude fiber (CF) was also determined by AOAC (2005) method. At the time of silo filling and after ensiling, samples were collected and subjected to physical and proximate analysis. After about 1.5 months, the ensiled materials went a physical test to assess the smell/odor, texture, color, and percentage of deterioration. To calculate the dry matter and moisture contents, samples of various weights were oven- and sun-dried. For each treatment's remaining ensiled samples the remaining ensiled samples from each treatment were dried at 70 °C until they reached a consistent dry weight. Following the AOAC (2005) guidelines, dried samples were crushed to a 40 mesh size before being evaluated for crude fiber (CF).

### **3.18.6 Ash**

At the time of silo filling and after ensiling, samples were collected and subjected to physical and proximate analysis. After about 1.5 months, the ensiled materials went a physical test to assess the smell/odor, texture, color, and percentage of deterioration. To calculate the dry matter and moisture contents, samples of various weights were oven- and sun-dried. For each treatment's remaining ensiled samples the remaining ensiled samples from each treatment were dried at 70 °C until they reached a consistent dry weight. Following the AOAC (2005) guidelines, dried samples were crushed to a 40 mesh size before being evaluated for Ash.

### **3.18.7 Organic matter percentage**

Organic matter percent was calculated from the sample of 50 days after sowing and was derived from the ash content of each treatment. To calculate the organic matter percentage following formula was used:

$$\text{OM (\%)} = 100 - \text{Ash content}$$

### **3.18.8 Hemicellulose**

Hemicellulose was measured at 50 days after sowing. This data was also derived data and converted from acid detergent fiber and neutral detergent fiber. The calculation was done with the following formula:

$$\text{Hemicellulose} = \text{Acid detergent fiber} - \text{Neutral detergent fiber}$$

### **3.19 Statistical analysis**

Data accumulated from different parameters were subjected to analysis of variance (ANOVA) using CoStat v.6.400 (CoStat, 2008) a computer based software. Correlation analysis was done considering Least Significant Difference (LSD) test at 5% level of significance.

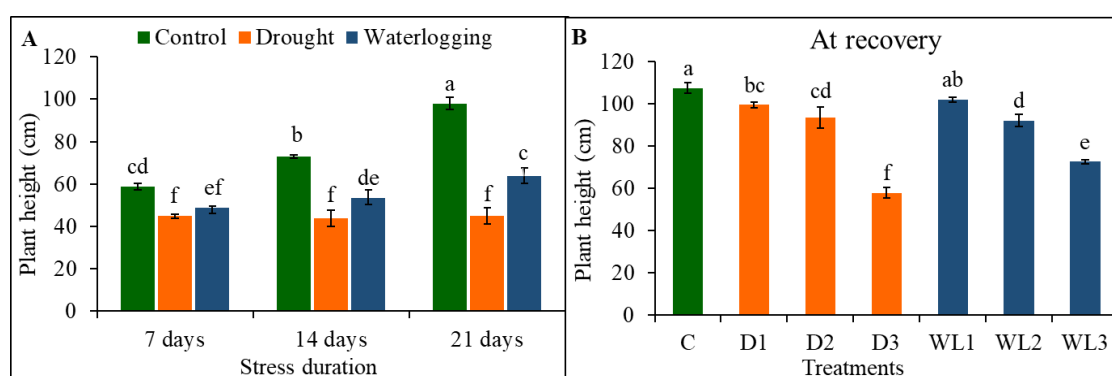
## CHAPTER IV

### RESULTS AND DISCUSSION

#### 4.1 Growth parameter

##### 4.1.1 Plant height

Plant height has been significantly decreased under the different duration of stress conditions compared to control one. The plant height has decreased by 23 and 17% under drought and waterlogging conditions for 7 days, respectively, 40 and 27% under drought and waterlogging for 14 days, respectively and 54 and 35% under drought and waterlogging conditions for 21 days, respectively compared to control condition (Fig. 1A). Plant height reduction was severe in case of drought stress compared to waterlogging conditions.



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 1.** Plant height of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

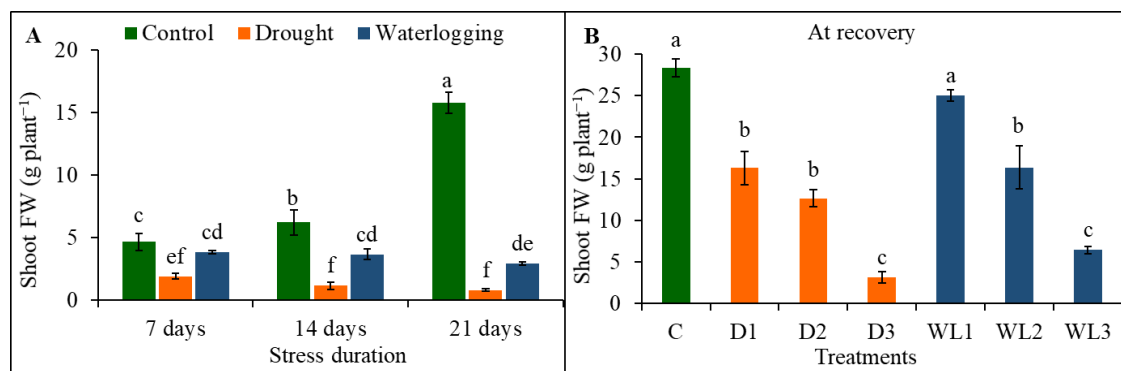
At 50 days after sowing, the highest plant height (107.38 cm) was observed from the control plant and the lowest (57.67 cm) was from drought for 21 days. Plant height was reduced significantly by 10, 18 and 47% under D1, D2, D3 and by 7, 16 and 34% under WL1, WL2, WL3, respectively (Fig. 1B).



Previous research has shown that water stress decreased seedling growth because it reduced carbon acclimation and partitioning and inhibited cell growth (Jabbari *et al.*, 2013) and as a result, plant height decreased. Misra *et al.* (2020) also found a similar result in case of sugarcane plant height. Khalid *et al.* (2018) similarly found comparable outcomes in sugarcanes under drought stress. In case of waterlogging stress, Promkhambut *et al.* (2010) found the same result of us. With the increase in the duration of the waterlogging condition, the height was decreased up to 30%. This finding is also agreed with the findings in wheat (*Triticum aestivum*) (Malik *et al.*, 2001), ryegrass (*Lolium perenne* L.) (Mcfarlane *et al.*, 2004), maize (*Zea mays* L.) (Zaidi *et al.*, 2004).

#### 4.1.2 Shoot fresh weight

A noticeable change had occurred in case of shoot fresh weight compared to control. Shoot fresh weight has been decreased by 59 and 18% under 7 days' drought and waterlogging, respectively, 81 and 41% under 14 days' drought and waterlogging condition, 95 and 81% under 21 days' drought and waterlogging respectively compared to control condition (Fig. 2A). Between both water stress, drought stress hamper severely shoot fresh weight compared to waterlogging conditions.



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 2.** Shoot fresh weight of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

At 50 days after sowing, the highest shoot fresh weight (28.32 g plant<sup>-1</sup>) was observed from the control plant and the lowest (3.12 g plant<sup>-1</sup>) was from 21 days' drought-

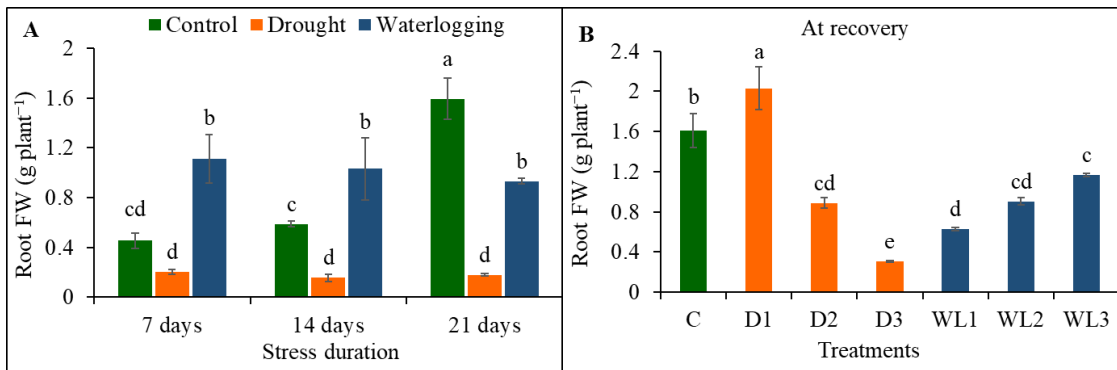
stressed plant. Shoot fresh weight was significantly reduced by 42, 54 and 89% under D1, D2, D3 and 12, 42 and 77% under WL1, WL2, WL3, respectively compared to control (Fig. 2B).

In case of the wheat plant, the same result was observed by Stallmann *et al.* (2020). Above ground shoot fresh weight had decreased under drought conditions compared to control conditions. This happens due to lower stomatal conductance and low carbon dioxide assimilation (Catola *et al.*, 2016). Reduced nutrient mobility within the soil, nitrogen uptake by roots, and nutrient translocation to shoots during a water scarcity may all be contributing factors. The coordination of growth-related processes during drought is controlled by complicated signaling networks that include abscisic acid. Wheat plants exposed to drought stress may have spent more on root development and/or altered root architecture relative to shoot biomass, which is the typical response to water scarcity to enhance water and nutrient intake from the soil (Boyle *et al.*, 2016). When there is waterlogging, the synthesis of carbohydrates and the absorption of nutrients will be significantly reduced (Pampana *et al.*, 2016). So, shoot fresh weight decreased significantly.

#### **4.1.3 Root fresh weight**

Under water stress conditions, plant root weight has been reduced under drought conditions and increased under waterlogging conditions compared to control condition. In case of drought stress, root FW (fresh weight) has been decreased by 55% under 7 days' stress, 73% under 14 days' stress, and 89% under 21 days' stress compared to their respective control one. Root fresh weight has been increased by 145, 76 and 41% under 7, 14 and 21 days of waterlogging stress compared to their respective control plant, respectively (Fig. 3A).

At 50 days after sowing, the highest root FW ( $1.609 \text{ g plant}^{-1}$ ) was found from the control plant, and the lowest root FW ( $0.305 \text{ g plant}^{-1}$ ). Root FW has been decreased significantly by 26, 45 and 81% under D1, D2, D3 and 61, 44 and 27% under WL1, WL2, WL3, respectively compared to control (Fig. 3B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 3.** Root fresh weight  $\text{plant}^{-1}$  of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

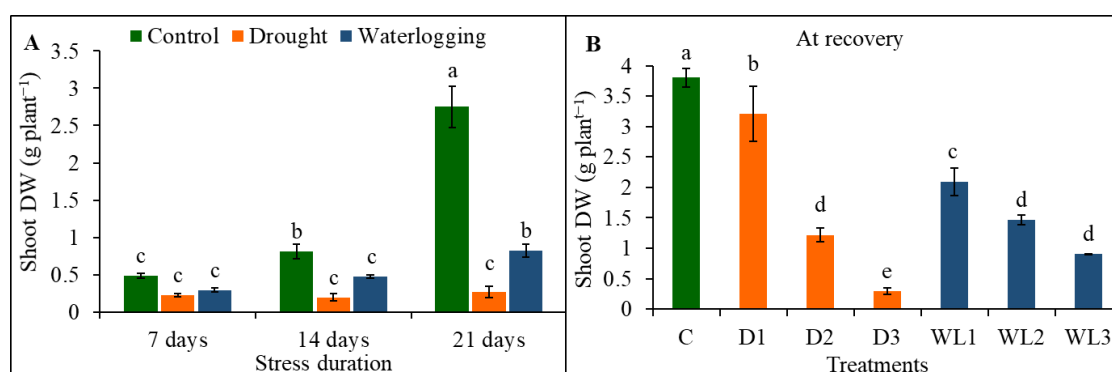
A similar result was observed under drought stress. Root fresh weight decreased sharply with the increase of drought stress duration. Under mild and moderate waterlogging conditions (7 & 14 days) root biomass has been increased though under severe waterlogging (21 days) fresh weight of root was decreased compared to control. A similar result was observed by Ploschuk *et al.* (2017). In another experiment with maize root weight increased by 235.42% under waterlogging than control (Bajpai and Chandra, 2015). The increasing fresh weight because of high root porosity allows amazing internal tissue aeration through aerenchyma (Colmer and Voisenek, 2009) for which root growth was unaffected during waterlogging.

#### 4.1.4 Shoot dry weight

Shoot dry weight has been reduced to a great extent under both drought and waterlogging conditions. After 7 days of water stress, shoot DW (dry weight) has been reduced by 54 and 39% under drought and waterlogging respectively compared to control but data were statistically similar. Shoot DW has been reduced by 76 and 41% under 14 days' drought & waterlogging and 90 and 70% under 21 days' drought & waterlogging respectively compared to control condition (Fig. 4A).

At 50 days after sowing, the highest shoot dry weight ( $3.80 \text{ g plant}^{-1}$ ) was observed from the control plant and the lowest ( $0.294 \text{ g plant}^{-1}$ ) was from drought for 21 days.

Shoot dry weight was reduced by 16, 68 and 92% under D1, D2, D3 and 45, 62 and 76% under WL1, WL2, WL3, respectively compared to control (Fig. 4B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 4.** Shoot dry weight plant<sup>-1</sup> at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

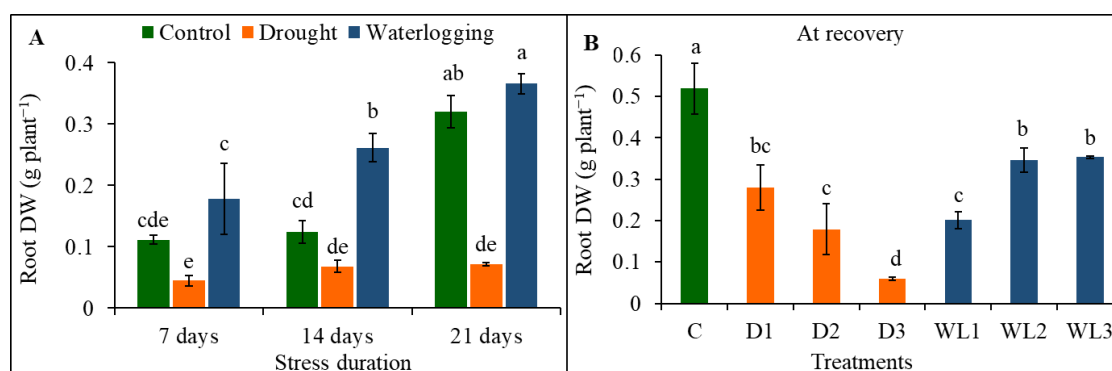
When Ahmed *et al.* (2021) revealed the same results under 3 levels of drought stress. Under severe drought stress, 65% & 68% shoot DW had decreased in case of BARI chola-7 and BARI chola-9, respectively. A similar result was found that shoot DW decreased with the increase of drought stress which agreed with previous studies (Nahar *et al.*, 2016; Hasanuzzaman *et al.*, 2018c; Bhuiyan *et al.*, 2019). Shoot dry weight was decreased significantly under the waterlogging condition. In another study, shoot dry weight of barley and wheat showed a significant decrease under waterlogging in comparison with control Xu *et al.*, 2022) being consistent with the results in previous works (Liu *et al.*, 2017; Xie and Shen, 2021).

#### 4.1.5 Root dry weight

Root dry weight has been decreased under drought stress and increased under waterlogging condition compared to control condition. Root DW (dry weight) has been decreased by 60% under drought and increased by 60% under waterlogging conditions after 7 days of water stress compared to the control one. Similarly, root DW has decreased by 45% under drought & increased by 111% under waterlogging conditions after 14 days of water stress and decreased by 78% under drought and increased by

14% under waterlogging conditions after 21 days of water stress compared to the respective control plant (Fig. 5A).

At 50 days after sowing root DW has been reduced and the highest dry weight ( $0.519 \text{ g plant}^{-1}$ ) was found in the control plant and the lowest dry weight ( $0.06 \text{ g plant}^{-1}$ ) was found from drought for 21 days. Root DW was reduced by 47, 65 and 88% under D1, D2, D3 and 61, 33 and 32% under WL1, WL2, WL3, respectively compared to control (Fig. 5B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 5.** Root dry weight plant<sup>-1</sup> at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

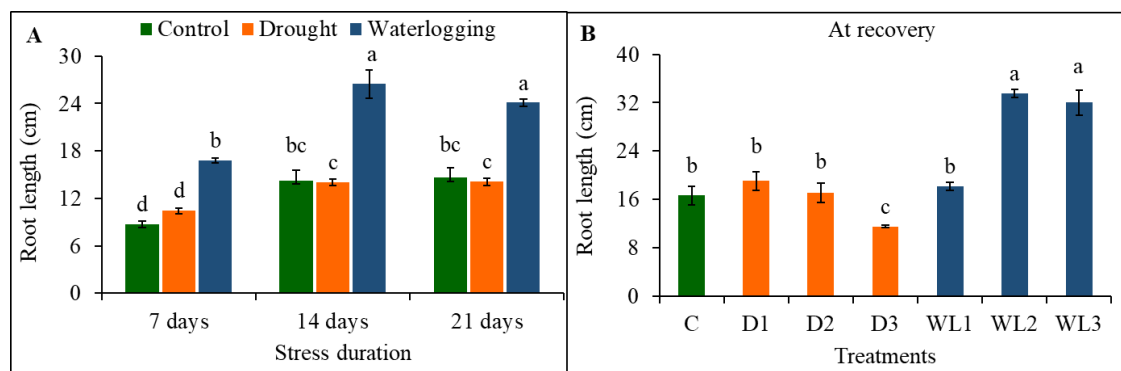
Root dry weight was decreased significantly due to the reduction of carbohydrate synthesis under drought conditions. In a study with barley and wheat, shoot dry weight showed a significant decrease under drought in comparison with control (Xu *et al.*, 2022) being consistent with the results in previous works (Liu *et al.*, 2017; Xie *et al.*, 2021). Under waterlogging, root dry weight had been increased by 4.02% in the case of sugarcane (Misra *et al.*, 2020).

#### 4.1.6. Root length

Root length has been increased under both drought and waterlogging for 7 days, but when stress duration was longer, the length has been increased under waterlogging but decreased under drought. Root length has been increased by 20 and 93% under drought

and waterlogging respectively compared to the control condition. In the case of 14 days' stress, root length has been decreased by 2% and increased by 86% under drought and waterlogging respectively compared to control. Root length has been decreased by 4 and increased by 65% under 21 days of drought and waterlogging respectively (Fig. 6A).

After 50 days of sowing, the highest root length (33.52 cm) was found from 14 days of the waterlogged plant, and the lowest root length (11.50 cm) from 21 days of the drought-stressed plant. Root length was increased by 14, 3 and decreased by 31% under D1, D2, and D3, respectively compared to control. Root length has been increased by 9, 101 and 92% under WL1, WL2, and WL3, respectively compared to control (Fig. 6B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

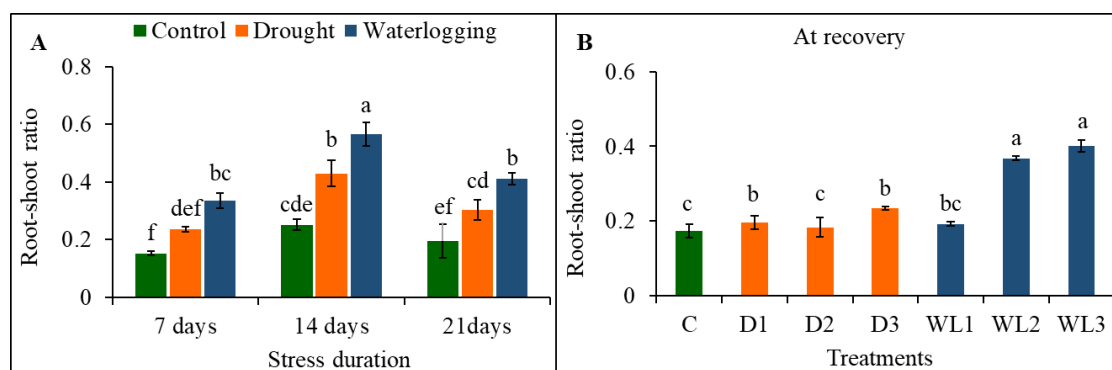
**Figure 6.** Root length of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

With the intensity of the water stress, root length dramatically increased, although root fresh weight and root dry weight decreased. Under moderately stressed plants of both types, drought stress produced the longest roots. In drought-stressed plants, maximum extraction of the soil moisture reserves is required to make it available for transpiration, and this might be accomplished through a root structure-related adaptation process. Root fresh and dry weight decreased but root length grew in alfalfa during PEG-induced drought stress (Zeid and Shedeed, 2006). Accordingly, Alghabari and Ihsan (2018)

found that at 50% FC, barley had the longest roots but the smallest roots in terms of fresh and dry weight due to a shortage of water availability.

#### 4.1.7 Root-shoot ratio

The root-shoot ratio has been increased under both drought and waterlogging conditions. Root-shoot ratio has been increased by 55 and 120% under 7 days of drought and waterlogging conditions, respectively, 71 and 125% under 14 days of drought and waterlogging conditions, respectively and by 56 and 111% under 21 days of drought and waterlogging conditions, respectively compared to control condition. The increase was significant under waterlogging conditions compared to drought (Fig. 7A).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 7.** Root-shoot ratio of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

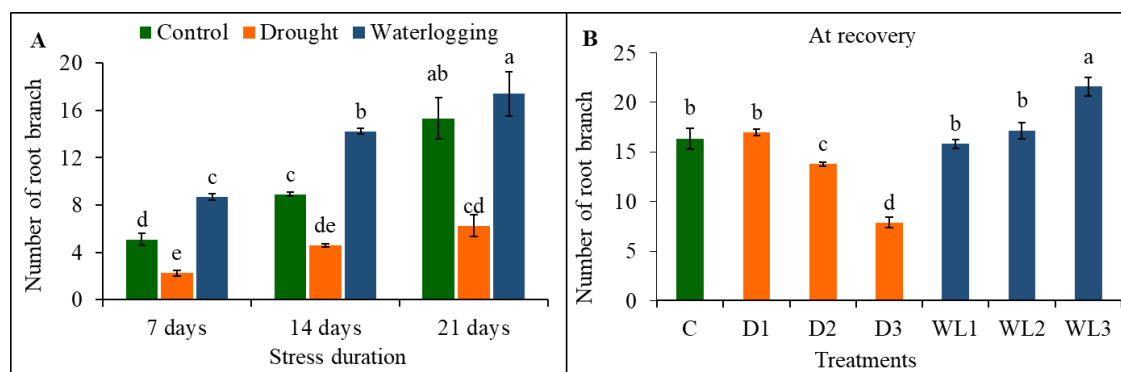
At 50 days after sowing, the root-shoot ratio has been increased in case of all treatments compared to the control. Highest RSR (root-shoot ratio) (0.4) was found from waterlogged for 21 days and the lowest RSR (0.17) was found from the control plant. Root-shoot ratio was increased by 13, 6 and 35% under D1, D2, D3 and 11, 112 and 131% under WL1, WL2, WL3, respectively compared to control (Fig. 7B).

Root-shoot ratios (R/S) that are higher in Napier grass early stages of Mulato II during a drought imply that Napier grass spends more assimilation in root formation at these

times than in Mulato II. conditions. Resource allocation for root growth is increased is believed to enhance water intake and, as a result, drought adaptation (Chimungu *et al.*, 2014). However, under a drought, greater root development could not offer much if a bigger shoot, which requires more water, is also advantageous (Palta *et al.*, 2011; Vadez *et al.*, 2013). It's possible functionally defined water absorption vs. water loss by the root length-to-leaf area ratio (RL/LA) (Comas *et al.*, 2013).

#### 4.1.6. Root branch

Root branches have been decreased under drought stress and increased under waterlogging condition compared to control condition. Root branches have decreased by 56% under drought and increased by 70% under waterlogging conditions after 7 days of water stress compared to the control one. Similarly, the root branch has decreased by 49% under drought & increased by 60% under waterlogging conditions after 14 days of water stress and decreased by 59% under drought and increased by 14% under waterlogging conditions after 21 days of water stress compared to the respective control plant (Fig. 8A).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 8.** Root branch number of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

At 50 days after sowing, the root branch increased under waterlogging conditions and the highest root branch number (21.61) was found from waterlogging for 21 days and the lowest root branch number (7.88) was found from drought for 21 days. Root branch

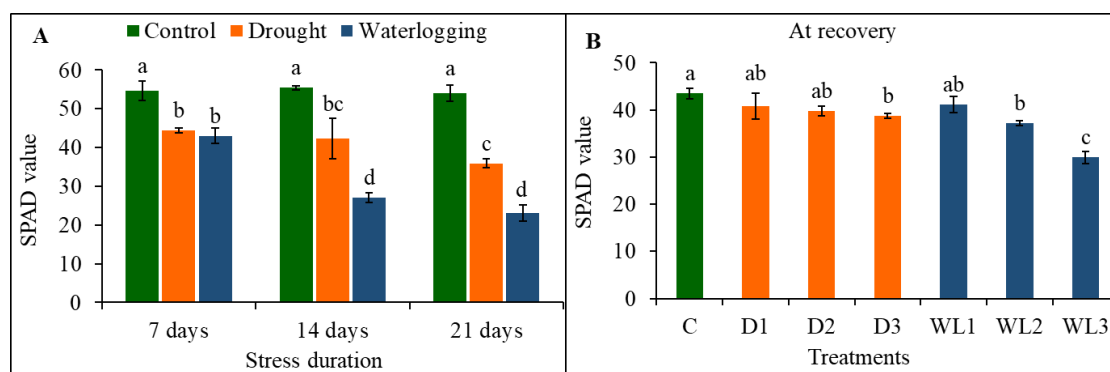


was increased by 4% and reduced by 16 and 52% under D1, D2, D3 respectively and reduced by 3%, and increased by 5 and 32% under WL1, WL2, WL3, respectively compared to control (Fig. 8B).

## 4.2 Physiological parameter

### 4.2.1 SPAD value

SPAD value of leaf has been reduced under drought and waterlogging at different durations compared to control. At 7 days durated stress, the SPAD value has been reduced by 19% & 21% under drought & waterlogging conditions compared to control and the SPAD value is more or similar under both stress. SPAD value has been reduced by 24 and 51% under 14 days durated drought & waterlogging conditions, respectively and by 34 and 57% under 21 days durated drought & waterlogging conditions, respectively. The reduction was more in case of waterlogging conditions than drought stress (Fig. 9A).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 9.** SPAD value of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

At 50 days after sowing, the highest SPAD value (43.49) was observed from the control (well-watered) condition and the lowest (29.92) was from 21 days of waterlogged plants. SPAD value has been reduced by 6, 9 and 11% under D1, D2, and D3, respectively, and reduced by 5 and 14 and 31% under WL1, WL2, WL3, respectively

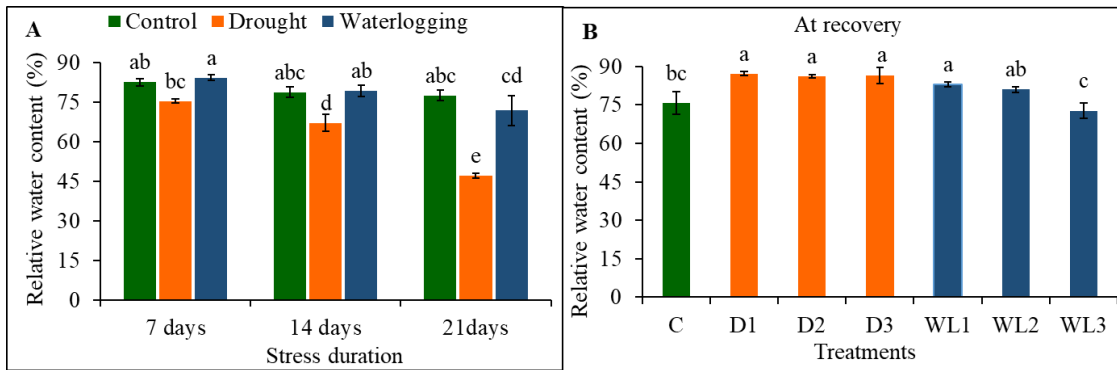
compared to control. The SPAD value was similar in the case of control and drought & waterlogging except for WL3 (Fig. 9B).

Under waterlogging, the SPAD value decreased by 10.49% compared to the control (Bajpai and Chandra, 2015). Chlorophylls are essential for photosynthesis and photo assimilation and can be utilized to detect the senescence of leaves (Anee *et al.*, 2019; Manik *et al.*, 2019). Owing to waterlogging, chlorophyll is degraded to allow nitrogen to be remobilized to younger leaves (Herzog *et al.*, 2016; Fukao *et al.*, 2019). One of the most obvious symptoms of leaf senescence is the degradation of chlorophyll (Brickman *et al.*, 2019). The fall in SPAD values, which are highly connected with chlorophyll content, previous research has shown that waterlogging accelerates leaf senescence and reduces photosynthetic capability (Tian *et al.*, 2019; Ren *et al.*, 2016b). In our experiment, waterlogging caused decreases in the SPAD to varying degrees, indicating that waterlogging caused the chlorophyll in the functional leaves on the main peanut stem to deteriorate and reduced the photosynthetic capacity, which supported earlier research findings (Ahmed *et al.*, 2006).

#### **4.2.2 Relative water content**

Relative water content (RWC) has been reduced under drought and increased under waterlogging when stress was imposed for 7 and 14 days' duration but reduced under both stress conditions when stress was imposed for 21 days. RWC has been reduced by 7% under drought and increased by 2% under waterlogging of 7 days and reduced by 12% under drought of 14 days. When water stress was imposed for 21 days, RWC has been reduced by 30 and 6% under drought & waterlogging conditions respectively compared to control (Fig. 10A).

At 50 days after sowing, the highest RWC (87.23%) was observed from D1 (7 days' drought stress) condition and the lowest (72.69%) was from 21 days of waterlogged plants. Relative water conditions have been increased by 11, 10 and 11% under D1, D2, and D3 respectively, and increased by 7, 5 and reduced by 3% under WL1, WL2, WL3, respectively compared to control (Fig. 10B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 10.** Relative water content of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

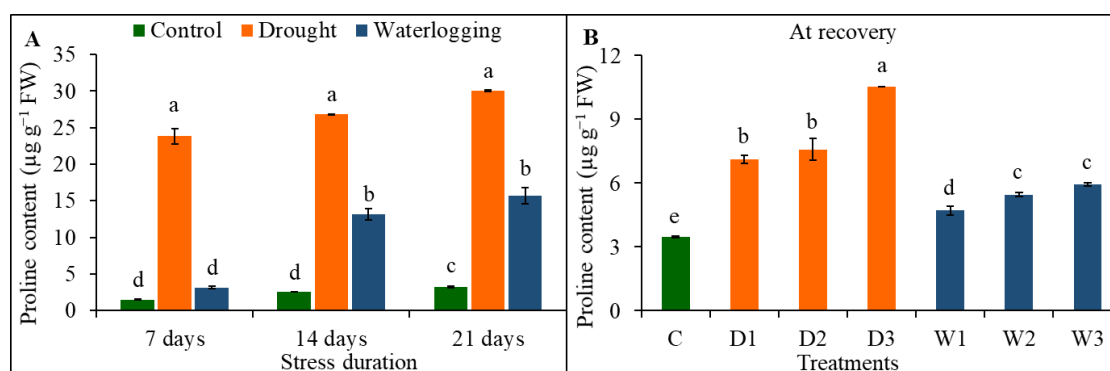
Under drought stress, the relative water content of the leaves in both types of chickpea such as BARI Chola-7 and BARI Chola-9, plants had the lowest RWC under extreme drought stress by 36 and 30%, respectively compared to control (Ahmed *et al.*, 2021). Shariatmadari *et al.* (2017) stated that leaf RWC decreased with the increase of drought stress levels at 70, 50, and 30% FC in *C. arietinum*. Plant tolerant to osmotic stress brought on by waterlogging has been discovered to be significantly influenced by relative leaf water content. In our investigation, waterlogging causes the RWC content in napier to be significantly decreased (Figure 10). Reduced leaf RWC is a sign that there is not enough water available for cell growth (Katerji *et al.*, 1997). Even though there was plenty of water available in the wet circumstances, RWC decreased. Withering is induced which impeded the root's permeability (Ashraf, 2012). A similar drop in RWC owing to waterlogging was also noted in mung beans (Kumar *et al.*, 2013) and sesame (Anee *et al.*, 2019).

## 4.3 Biochemical parameter

### 4.3.1 Proline content

Proline content is an indicator of antioxidant defence element and generally, increase when a plant faces stress condition. Proline content has been increased by 5669 and 282% under drought & waterlogging of 7 days, 937 and 396% under 14 days of drought and waterlogging and 302 and 99% under drought and waterlogging condition for 21 days, respectively compared to their control condition. In the case of drought stress, proline was higher than in waterlogging conditions (Fig. 11A).

At 50 days after sowing, proline content has been increased in case of all treatments compared to control. The highest proline content ( $3.34 \mu\text{g g}^{-1} \text{FW}$ ) was found in control plants and the lowest proline content ( $1.07 \mu\text{g g}^{-1} \text{FW}$ ) was found from 7 days waterlogged plant. Proline content has been decreased by 48, 25 and 21% under D1, D2, D3 and 68, 30 and 22% under WL1, WL2, WL3, respectively compared to control. But the values were statistically similar (Fig. 11B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

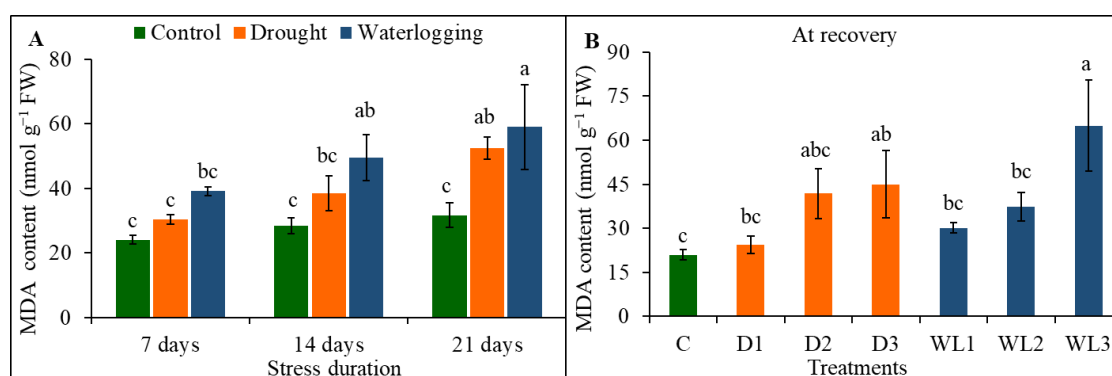
**Figure 11.** Proline content of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Other results also showed that proline concentration has been increased in all grass species in response to drought. According to Bandurska and Józwiak (2010), the accumulation of proline depends on many factors such as plant age, which may explain

a higher concentration of proline was characterized by the highest increase of this osmolyte. Proline seems to be associated with a better drought tolerance grass species, which was also suggested by Perlikowski *et al.* (2014) and Fariaszewska *et al.* (2016). Under waterlogging, proline was also increased with the increase of waterlogging duration (Bajpai and Chandra, 2015).

### 4.3.2 Malondialdehyde content

Malondialdehyde (MDA) content is also known as a lipid peroxidation indicator and is increased under stress conditions. Malondialdehyde content has been increased by 26 and 62% under drought & waterlogging for 7 days, 35 and 75% under 14 days of drought & waterlogging and 66 and 86% under drought and waterlogging condition for 21 days respectively compared to their control condition. In case of the waterlogging condition, MDA content was higher than in drought conditions (Fig. 12A).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 12.** Malondialdehyde content of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

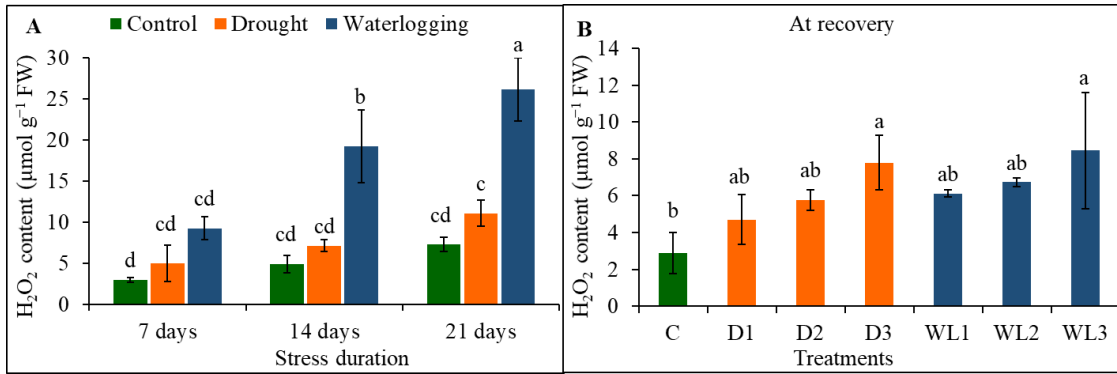
At 50 days after sowing, MDA content has been increased in case of all treatments compared to control. The highest MDA content ( $64.95 \text{ nmol g}^{-1} \text{ FW}$ ) was found from 21 days waterlogged plant and the lowest MDA content ( $20.99 \text{ nmol g}^{-1} \text{ FW}$ ) was found from the control plant. Malondialdehyde content has been increased by 16, 99 and 114% under D1, D2, D3 and 43, 78 and 209% under WL1, WL2, WL3, respectively compared to control (Fig. 12B).

Several experiments showed that drought-induced ROS generation, including H<sub>2</sub>O<sub>2</sub>, O<sub>2</sub>, and OH<sup>-</sup>, among many others, protein and lipid has severe damage and, as a result, stress affects membrane function (Miller *et al.*, 2010; Liu *et al.*, 2013; Ye *et al.*, 2016). The rapeseed seedlings in a study demonstrated enhanced ROS generation, and lipid peroxidation (increased MDA level) indicating membrane damage as a result of drought. In another study, after waterlogging stress was applied, black gram genotypes showed an instantaneous increase in lipid peroxidation and cell membrane damage (Bansal *et al.*, 2019). It exhibits decreased membrane stability, as well as Proteins injury resulting from ROS produced during waterlogging. Earlier research suggested that free cell surface peroxidation brought on by radicals shows cellular damage brought on by stress. MDA increased significantly as a result of waterlogging (Jain *et al.*, 2011). A similar result was found by Singh *et al.* (2017) when working with mung beans.

#### **4.3.2 Hydrogen peroxide content**

Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) content has been increased under both drought and waterlogging conditions compared to the control condition. Hydrogen peroxide has been increased by 68 and 211% under drought and waterlogging for 7 days, 45 and 292% under 14 days of drought & waterlogging and 53 and 260% under drought and waterlogging condition for 21 days respectively compared to their control condition. In case of the waterlogging condition, H<sub>2</sub>O<sub>2</sub> was higher than in drought conditions (Fig. 13A).

At 50 days after sowing, H<sub>2</sub>O<sub>2</sub> content has been increased in case of all treatments compared to control. The highest H<sub>2</sub>O<sub>2</sub> content (8.46 μmol g<sup>-1</sup> FW) was found in 21 days waterlogged plant and the lowest H<sub>2</sub>O<sub>2</sub> content (2.80 μmol g<sup>-1</sup> FW) was found in the control plant. Malondialdehyde content has been increased by 64, 100 and 171% under D1, D2, D3 and 113, 134 and 195% under WL1, WL2, WL3, respectively compared to control (Fig. 13B).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

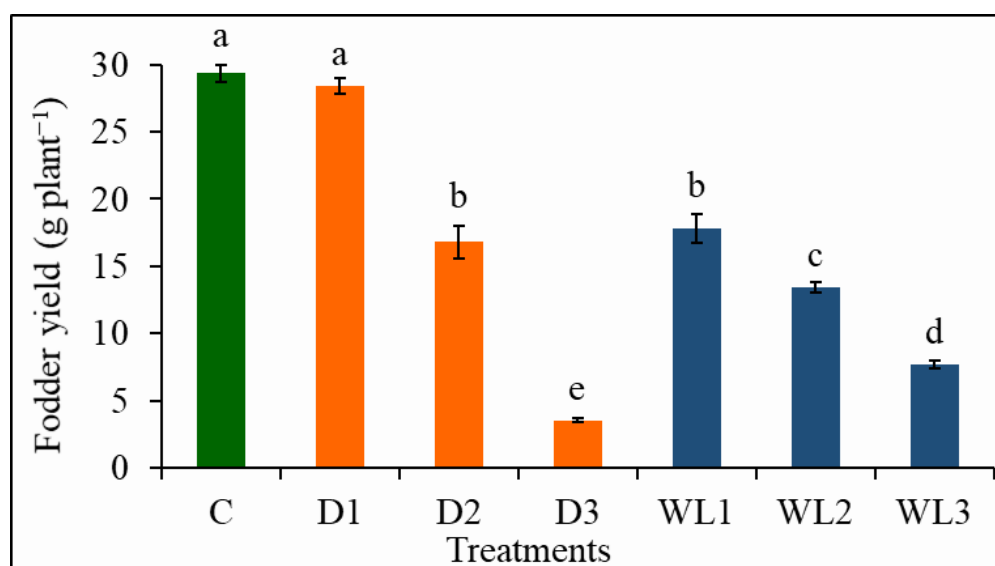
**Figure 13.** Hydrogen peroxide content of napier grass at completion of different stress duration (A) and at recovery (B). Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Under any stress condition, ROS production is increased. So, the H<sub>2</sub>O<sub>2</sub> increase is due to this reason. Compared with seedling growth under normal conditions (0.00 MPa), under the drought stress conditions (- 0.3 MPa) a sharp rise in H<sub>2</sub>O<sub>2</sub> content by 92%, ZY 36 compared with SG 127 for which a 42% increase in H<sub>2</sub>O<sub>2</sub>. The ROS produced due to impairment of the photosynthesis apparatus are very reactive and the oxidation of nucleic acids, lipids, carbohydrates, and proteins occurs and as well as damages to the cell membrane (Sabra *et al.*, 2012). A similar result was observed where, H<sub>2</sub>O<sub>2</sub> has been increased by 9 folds compared with control in maize under waterlogging stress (Asha *et al.*, 2021). Numerous biological molecules and metabolites are harmed by these ROS because of their high reactivity (Ashraf, 2009). Hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) levels increased in both resistant and sensitive maize genotypes following waterlogging stress, although the sensitive genotype showed considerably greater H<sub>2</sub>O<sub>2</sub> content under waterlogging circumstances, according to Yadav and Srivastava (2017).

## 4.4 Yield parameter

### 4.4.1 Fodder yield

Fodder yield decreases with the increase of stress duration. The reduction was severe under drought. Fodder yield has been reduced under drought and waterlogging compared to control. At recovery, the highest fodder yield (29.35 g plant<sup>-1</sup>) was observed from the control condition and the lowest (3.52 g plant<sup>-1</sup>) was from 21 days waterlogged plants. Fodder yield has been reduced by 3, 43 and 88% under D1, D2, and D3, respectively, and reduced by 35, 54 and 74% under WL1, WL2, WL3, respectively compared to control (Fig. 14).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 14.** Fodder yield of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

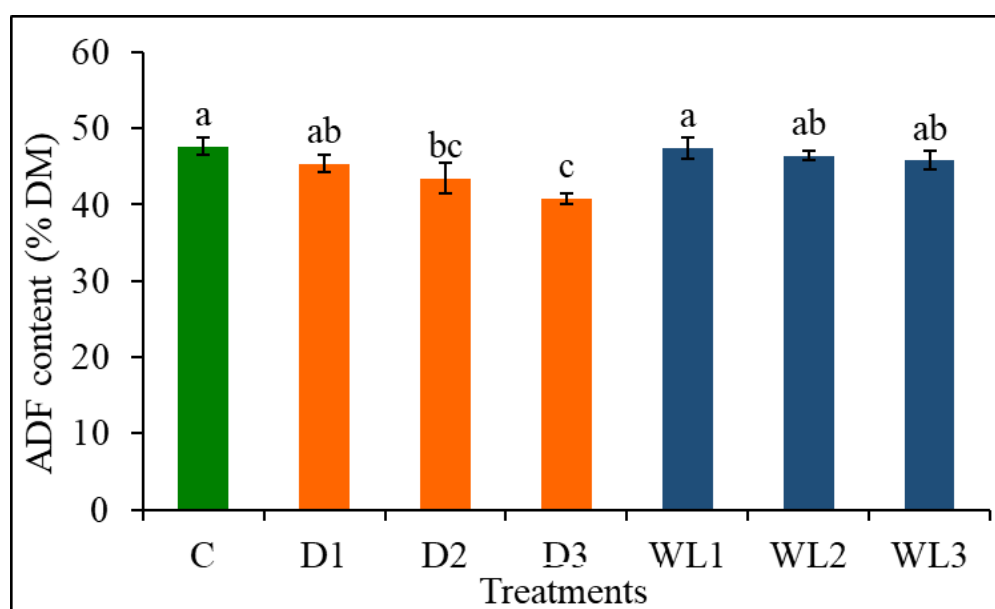
A similar result was found by Purbajanti *et al.* (2012). Guinea grass decreased 25.9% after three times stress compared to control and napier decreased 22.20% forage yield compared to control. Somegowda *et al.* (2021) experimented with sorghum and observed the same result. Water stress, however, has been shown to lower the fodder output and growth parameters in (Perrier *et al.*, 2017). Furthermore, Nouri *et al.* (2020) showed that reduced vegetative traits were influenced by the field's decreasing soil moisture level.



## 4.5 Qualitative parameter

### 4.5.1 Acid detergent fibre

Acid detergent fibre (ADF) content was calculated only at 50 days after sowing. The ADF content has been reduced under both drought and waterlogging stress condition. Highest ADF (47.67%) was found in the control plant and the lowest ADF (40.75%) in 21 days' drought-stressed plant. Acid detergent fibre was reduced by 5, 9 and 5% under D1, D2, D3 and 1, 3 and 4% under WL1, WL2, WL3, respectively compared to control. Under waterlogging conditions, ADF contents were statistically similar (Fig. 15).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

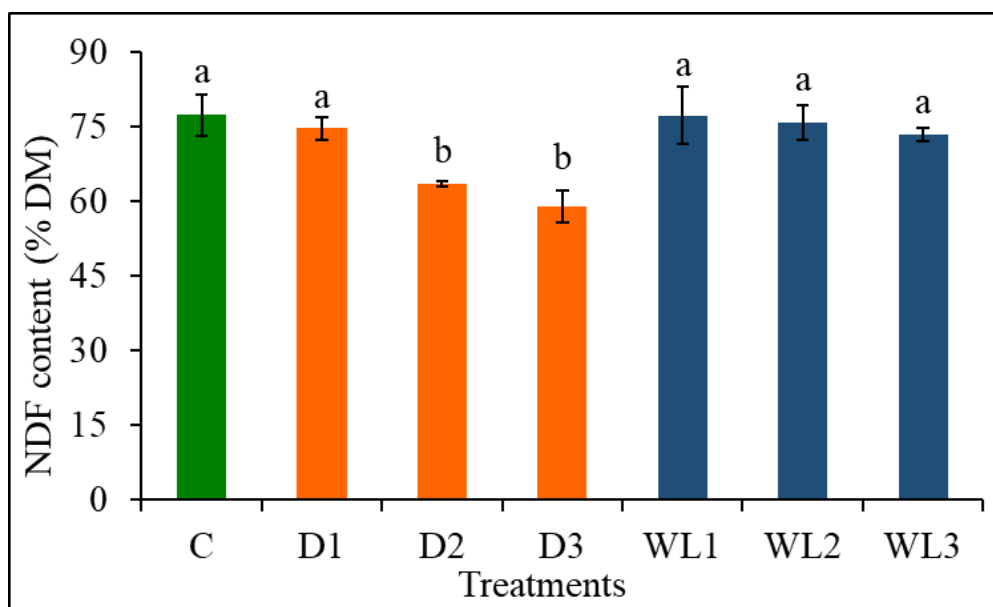
**Figure 15.** Acid detergent fibre of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Küchenmeister *et al.* (2013) also found a similar result when they worked with forage. ADF decreased when exposed to water stress. ADF content offers a forecast of cellulose and lignin, which have a negative impact on forage digestibility. ADF concentrations that are lower might cause an improvement in the forage's digestibility and higher use of plants. Moore *et al.* (2008) reported that longer water stress exposure makes it harder for plants to change their cell membranes to maintain growth in conditions with lowered

water potential. These findings were published in accord with other research findings indicating that plants' ADF content dropped as a result of water stress. Küchenmeister *et al.* (2013) found a decrease in the ADF % of perennial forage legumes under intense stress. Abid *et al.* (2016) reported Because, in extreme water conditions, alfalfa's ADF levels decreased stress.

#### 4.5.2 Neutral detergent fibre

Neutral detergent fibre (NDF) content was calculated only at 50 days after sowing. The NDF content has been reduced under both drought and waterlogging stress condition. Highest NDF (77.47%) was found in the control plant and the lowest NDF (58.98%) from 21 days stressed plant. Acid detergent fibre was reduced by 9, 18 and 24% under D1, D2, D3 whereas D2 and D3 are statistically similar and 1, 2 and 5% under WL1, WL2, WL3, respectively compared to control. Under drought conditions, drought for 14 days and 21 were similar and under waterlogging conditions, NDF contents were statistically similar (Fig. 16).



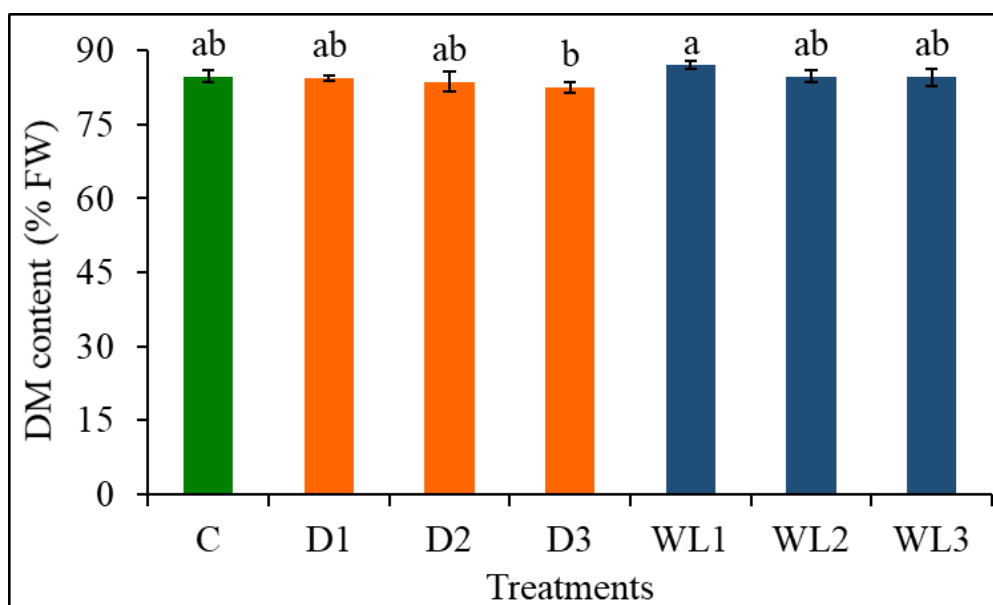
Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 16.** Neutral detergent fibre of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

NDF serves as a marker for cell wall elements. (containing lignin, cellulose, and hemicellulose) and is negatively associated with forage quality. Many variables commonly affect the fibre concentration of the plant's stage of development, the leaf-to-stem ratio, and climate factors (drought, temperature). According to Fulkerson *et al.* (2007), lower NDF and ADF concentrations are related to delayed maturity in plants under drought conditions. Research by Küchenmeister *et al.* (2013) revealed that NDF levels drop while under a lot of stress. Abid *et al.* (2016) studied how various irrigation amounts affected three populations of alfalfa and their nutritional quality. They reported that fibres are significantly affected by drought. The amount of NDF dropped from 41.33 to 25% of the field under control circumstances to 35.73% capacity.

### **4.5.3 Dry matter content**

The DM content has been reduced under both drought and waterlogging stress conditions with the increase in the duration of stress. Highest DM (86.99%) was found from 7 days waterlogged plant and the lowest DM (82.48%) from 21 days of a drought-stressed plant. Dry matter content was reduced by 9, 18 and 24% under D1, D2, D3 whereas D2 & D3 are statistically similar and 1, 2 and 5% under WL1, WL2, WL3, respectively compared to control. Under both drought and waterlogging conditions, there was no difference from the control in DM content (Fig. 17).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

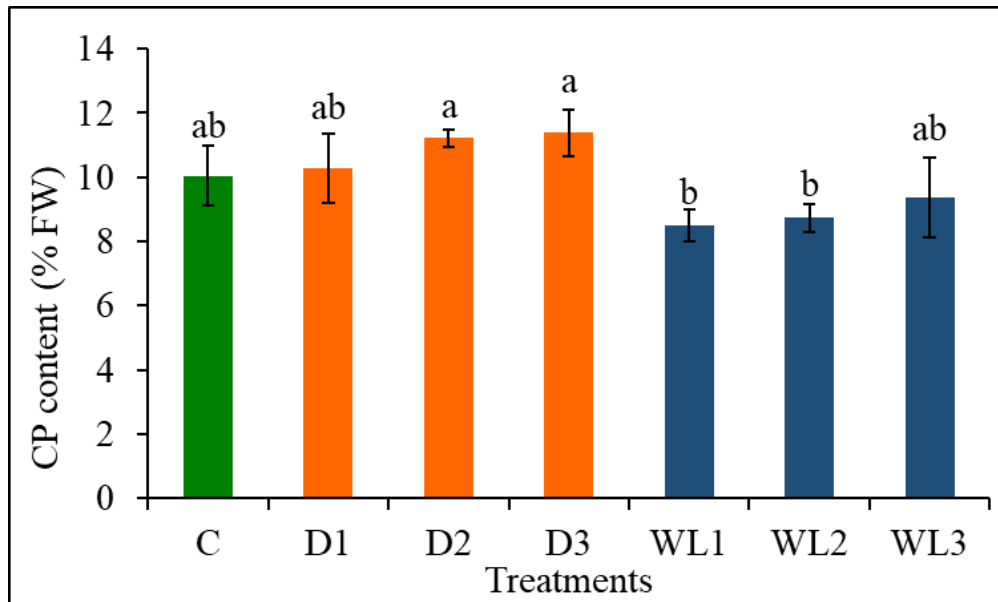
**Figure 17.** Dry matter content of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Balazadeh *et al.* (2021) also found the same result in case of dry matter under drought stress. Reduced soil water content makes it more difficult for roots to absorb nutrients like nitrogen and phosphorus, which limits growth, development, and yield. Reduced transpiration also lessens the soil's ability to transmit nutrients to plants, decreasing soil moisture affects roots and growth from root to shoot. Cellular photosynthesis and Under water stress, growth would be the first function to be constrained (Munns *et al.*, 2006). Stomatal conductivity declines under mild drought stress, reducing CO<sub>2</sub> availability and reducing photosynthesis in the process. Under drought stress, DM yield decreased additional investigations (Jahanzad *et al.*, 2013; Marsalis *et al.*, 2009; Vasilakoglou *et al.*, 2011) have also reported on these situations.

#### 4.5.4 Crude protein

Crude protein (CP) content was calculated only at 50 days after sowing. The CP has increased with the increase of duration of stress under both drought and waterlogging conditions. Highest CP (11.39%) was found from 21 days' of drought-stressed plant and the lowest CP (8.485%) from 7 days' of waterlogged plant. Crude protein was

increased by 9, 18 and 24% under D1, D2, D3 whereas D2 and D3 are statistically similar and 1, 2 and 5% under WL1, WL2, WL3, respectively compared to control. Under both drought and waterlogging conditions, there was no difference in CP content between the treatments (Fig. 18).



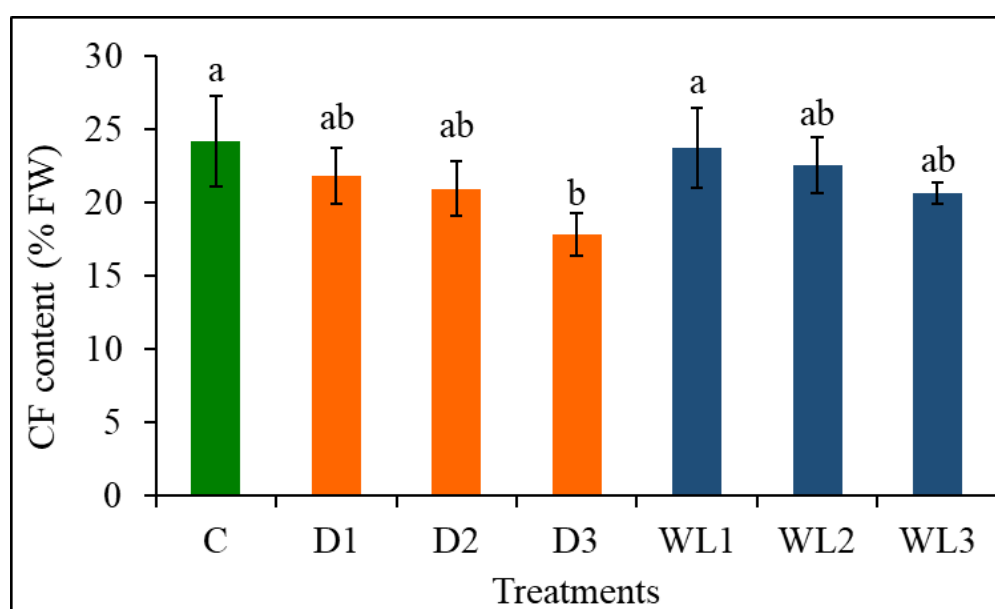
Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 18.** Crude protein of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Crude protein content increased by 8.36 % and 18.62 % under moderate and severe water stresses, respectively. (Taiz and Zeiger, 2010). Crude protein content has a significant part in raising the quality of fodder crops (Sun *et al.*, 2018). Nitrogen uptake, which is the cause of the CP concentration, and the availability of water have a big impact. They explain how plants expand their root and surface area. length density that is conducive to mild water stress absorption of nutrients by Li *et al.* (2013). Similar findings have been reported by researchers on how water stress affects the protein amount of plant life. According to Rostamza *et al.* (2011), As a result of stress, pearl millet's CP% rose. Bibi *et al.* (2012) demonstrated that rising moisture stress elevated crude protein content in sorghum-sudangrass hybrids; Meisser *et al.* (2016) and Fariaszewska *et al.* (2017) found that minor water stress drastically reduced the fodder grasses' protein content was raised. Khalil *et al.* (2018) claimed that raising crude protein percentage fell under water stress.

### 4.5.5 Crude fibre

Crude fibre (CF) content was calculated only at 50 days after sowing. The CF has decreased with the increase in the duration of stress under both drought and waterlogging conditions. Highest CF (24.23%) was found in the control plant and the lowest CF (17.83%) in 21 days' drought-stressed plant. Crude fibre was reduced by 10, 13 and 26% under D1, D2, D3 whereas D1, D2 and D3 are statistically similar and 2, 7 and 15% under WL1, WL2, WL3 respectively compared to control. Under both drought and waterlogging conditions, there was no difference in CF content between the treatments (Fig. 19).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

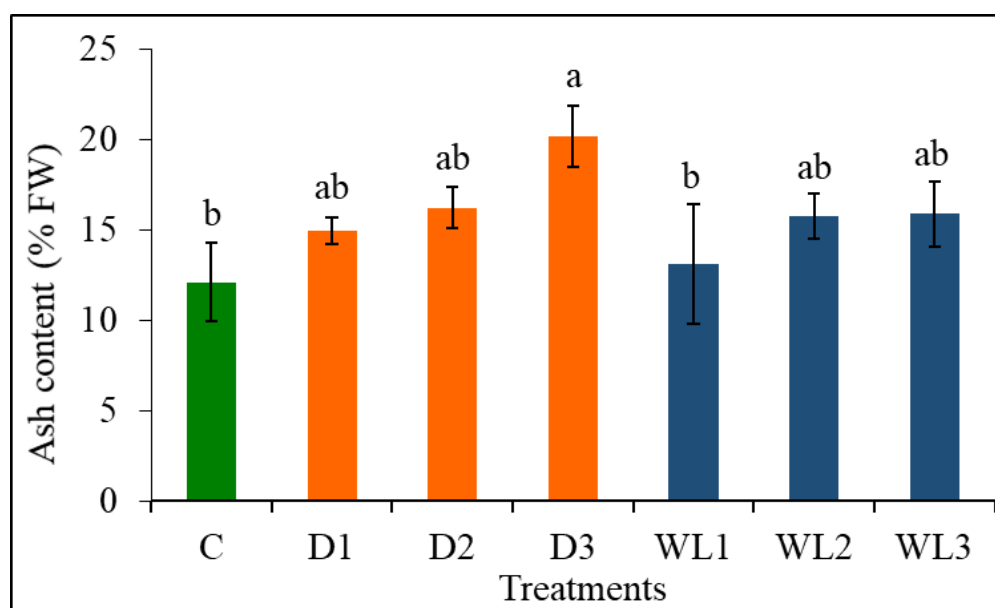
**Figure 19.** Crude fibre of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Crude fibre content was reduced by increasing water stress severity. The highest CF (55.5 g/kg DM) was observed in non-stress conditions. This trait decreased by 4.86 % and 8.83%, respectively under moderate and severe water stress. According to Onwugbuta-Enyi (2004), water-stressed plants had very low crude fibre concentrations in their cowpea seedlings. According to research by Bibi *et al.* (2012), sorghum-sudangrass hybrids' crude fibre is reduced during water stress as opposed to under

regular watering. African basil (*Ocimum gratissimum* L.) and Bushbuck leaves have significantly less crude fibre content when exposed to water stress, according to research by Osuagwu and Edeoga (2013).

#### 4.5.6 Ash content

Ash content was calculated only at 50 days after sowing. The ash content has increased with the increase in the duration of stress under both drought and waterlogging conditions. The highest ash (21.1%) was found from the control plant and the lowest ash (13.08%) from 7 days' waterlogged plant. Ash content has been increased by 23, 34 and 67% under D1, D2, D3 and 8, 30 and 31% under WL1, WL2, WL3, respectively compared to control. Under both drought and waterlogging conditions, there was no difference in ash content between the treatments (Fig. 20).



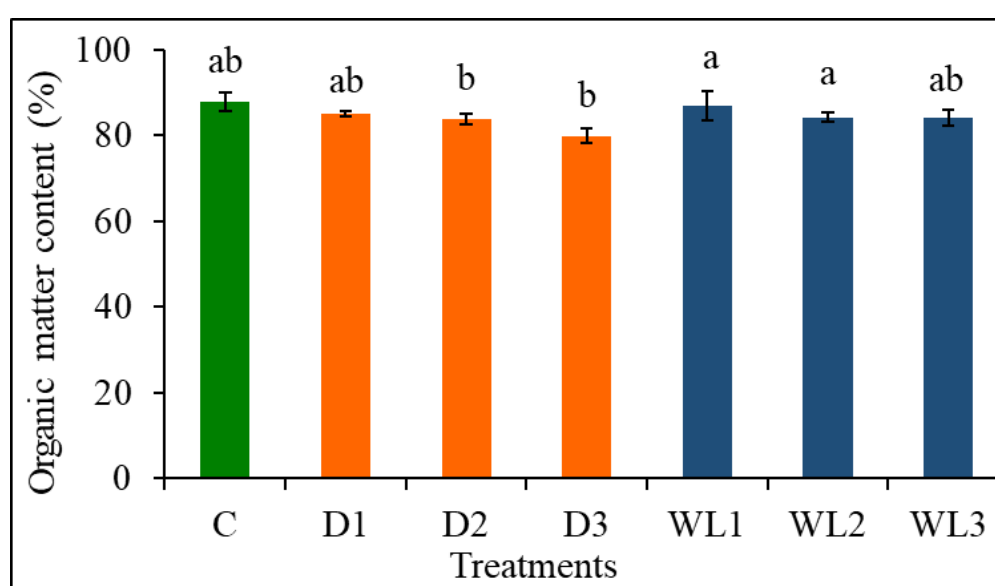
Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 20.** Ash content of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

Similarly, our results are agreed with Sarker *et al.* (2021) and Maleko *et al.* (2019). When working with napier variety, they found increasing ash content with the increase of drought stress.

#### 4.5.7 Organic matter percent

Organic matter percentage (OM%) was calculated only at 50 days after sowing. The OM% has decreased with the increase in the duration of stress under both drought and waterlogging conditions. Highest OM (87.9%) was found in the control plant and the lowest OM (89.84%) from 21 days' drought stressed plant. Organic matter percentage was reduced by 3, 5 and 9% under D1, D2, D3 and 1, 4 and 4% under WL1, WL2, WL3 respectively compared to control. Under both drought and waterlogging conditions, there was no difference in OM% between the treatments (Fig. 22).



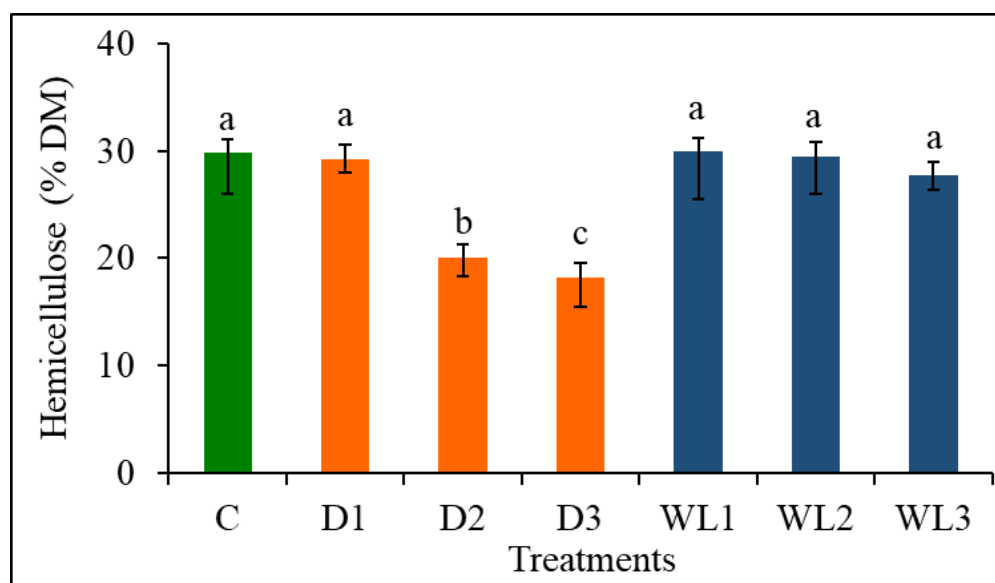
Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 21.** Organic matter percentage of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.



### 4.5.8 Hemicellulose

The hemicellulose has decreased with the increase in the duration of stress under both drought and waterlogging conditions. The highest hemicellulose (29.88%) was found from 7 days waterlogged plant and the lowest hemicellulose (18.23%) from 21 day's drought-stressed plant. Hemicellulose was reduced by 2, 33 and 39% under D1, D2, D3 and 1, 1 and 7% under WL1, WL2, WL3, respectively compared to control (Fig. 23).



Here, C= Control, D1= Drought for 7 days, D2= Drought for 14 days, D3= Drought for 21 days, WL1= Waterlogging for 7 days, WL2= Waterlogging for 14 days, WL3= Waterlogging for 21 days.

**Figure 22.** Hemicellulose content of napier grass at recovery. Mean ( $\pm$ SD) was calculated from three replicates for each treatment. Values in a column with different letters are significantly different at  $p \leq 0.05$  applying LSD test.

However, in other studies, drought-treated plants of all genotypes consistently had a significantly higher content of hemicellulosic polysaccharides than their respective control plants. Some of the differences may also be explained by differences in the duration of the applied drought (Jiang *et al.*, 2012; Emerson *et al.*, 2014; Rakszegi *et al.*, 2014). Long-term drought exposure, plants force to change the structure of their cell walls to maintain cell growth with lower water potential. Hemicelluloses reinforce the cell wall matrix by cross-linking to lignin and cellulose fibres, which increases the stiffness of the cell wall (Gall *et al.*, 2015).

## Chapter V

### SUMMARY AND CONCLUSION

The experiment was conducted at the experimental shed of the Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh, to investigate the morphological, physiological, biochemical, and qualitative responses of napier grass under different levels of water stress. The experiment was arranged in a completely randomized design (CRD) with three replications. Seedlings were grown in a controlled environment where drought and waterlogging stress were imposed on napier grass for 7, 14 and 21 days. The data were taken by sampling the leaves after the completion of each stress period. There were about 10 seedlings maintained in each pot. Different data on morphology (plant height, shoot FW plant<sup>-1</sup>, shoot DW plant<sup>-1</sup>, root FW plant<sup>-1</sup>, root DW plant<sup>-1</sup>, root length plant<sup>-1</sup>, root-shoot ratio, and root branch number plant<sup>-1</sup>), physiology (SPAD value, leaf RWC) and biochemical (Pro content, MDA content and H<sub>2</sub>O<sub>2</sub> content), yield (fodder yield), qualitative (ADF, NDF, DM content, ash, CP, CF, moisture, OM percentage, hemicellulose) were measured to investigate the responses of napier grass.

Plant height was reduced significantly under different duration of drought and waterlogging. Plant height was decreased by 23, 40 and 54% under 7, 14 and 21 days durated drought and 17, 27 and 35% under 7, 14, 21 days durated waterlogging stress, respectively. At the 50 days after sowing, the highest (107.38 cm) plant height was observed from control plant and lowest (57.67 cm) plant height was from 21 days durated drought stressed plant.

A prominent change was occurred in shoot FW and shoot DW. Shoot FW was decreased by 59, 81 and 95% under 7, 14 and 21 days durated drought and 18, 41 and 81% under 7, 14, 21 days durated waterlogging stress, respectively. At the 50 days after sowing, the highest (28.32 g plant<sup>-1</sup>) plant height was observed from control plant and lowest (3.12 g plant<sup>-1</sup>) plant height was from 21 days durated drought stressed plant. Shoot DW was decreased by 54, 76 and 90% under 7, 14 and 21 days durated drought and 39, 41 and 70% under 7, 14, 21 days durated waterlogging stress,

respectively. At the 50 days after sowing, the highest (3.80 g plant<sup>-1</sup>) plant height was observed from control plant and lowest (0.294 g plant<sup>-1</sup>) plant height was from 21 days durated drought stressed plant.

Root FW and root DW both also decreased under drought of different duration. In case of waterlogging stress, both parameters have increased compared to control. At 50 days after sowing the highest (1.609 g plant<sup>-1</sup>) root FW was observed from control plant and lowest (0.305 g plant<sup>-1</sup>) root FW was from 21 days durated drought stressed plant. At 50 days after sowing the highest (0.519 g plant<sup>-1</sup>) root DW was observed from control plant and lowest (0.06 g plant<sup>-1</sup>) root DW was from 21 days durated drought stressed plant.

Root length has been increased under both drought and waterlogging for 7 days, but when stress duration was longer, the length has been increased under waterlogging but decreased under drought. After 50 days of sowing, the highest root length (33.52 cm) was found from 14 days of the waterlogged plant, and the lowest root length (11.50 cm) from 21 days of the drought-stressed plant. The root-shoot ratio has been increased under both drought and waterlogging conditions. Root-shoot ratio was increased by 55, 71 and 56% % under 7, 14 and 21 days durated drought and 120, 125 and 111% under 7, 14, 21 days durated waterlogging stress, respectively. At 50 days after sowing, the root-shoot ratio has been increased in case of all treatments compared to the control. Highest RSR (root-shoot ratio) (0.4) was found from waterlogged for 21 days and the lowest RSR (0.17) was found from the control plant.

SPAD value decreased under drought and waterlogging stress. SPAD value was decreased by 19, 24 and 34% under 7, 14 and 21 days durated drought and 21, 51 and 57% under 7, 14, 21 days durated waterlogging stress, respectively. At 50 days after sowing, the highest SPAD value (43.49) was observed from the control (well-watered) condition and the lowest (29.92) was from 21 days of waterlogged plants.

Relative water content (RWC) has decreased under drought but increased under waterlogging stress. SPAD value was decreased by 7, 12 and 27% under 7, 14 and 21 days durated drought and 2% increased under 7 days waterlogging and 6% decreased under 21 days durated waterlogging stress, respectively. At 50 days after sowing, the

highest RWC (87.23%) was observed from D1 condition and the lowest (72.69%) was from 21 days of waterlogged plants.

Biochemical parameter, like proline content, MDA content and  $H_2O_2$  content has increased under both types of water stress. Proline content increased significantly. In the case of drought stress, proline was higher than in waterlogging conditions. At 50 days after sowing, proline content has been increased in case of all treatments compared to control. The highest proline content ( $3.34 \mu\text{g g}^{-1}$  FW) was found in control plants and the lowest proline content ( $1.07 \mu\text{g g}^{-1}$  FW) was found from 7 days waterlogged plant. Malondialdehyde content increased by 26, 35 and 66% under 7, 14 and 21 days durated drought and 62, 75 and 86% under 7, 14, 21 days durated waterlogging stress, respectively. At 50 days after sowing, MDA content has been increased in case of all treatments compared to control. The highest MDA content ( $64.95 \text{ nmol g}^{-1}$  FW) was found from 21 days waterlogged plant and the lowest MDA content ( $20.99 \text{ nmol g}^{-1}$  FW) was found from the control plant.

Hydrogen peroxide ( $H_2O_2$ ) content has been increased under both drought and waterlogging conditions compared to the control condition. Hydrogen peroxide has been increased by 68 and 211% under drought & waterlogging for 7 days, 45 and 292% under 14 days of drought & waterlogging and 53 and 260% under drought and waterlogging condition for 21 days respectively compared to their control condition. At 50 days after sowing,  $H_2O_2$  content has been increased in case of all treatments compared to control. The highest  $H_2O_2$  content ( $8.46 \mu\text{mol g}^{-1}$  FW) was found in 21 days waterlogged plant and the lowest  $H_2O_2$  content ( $2.80 \mu\text{mol g}^{-1}$  FW) was found in the control plant.

Fodder yield decreased by 58, 85 and 93% under 7, 14 and 21 days durated drought stress and 18, 26 and 73% under 7, 14 and 21 days durated waterlogging stress. At 50 days after sowing, the highest fodder yield ( $29.35 \text{ g plant}^{-1}$ ) was observed from the control (well-watered) condition and the lowest ( $3.52 \text{ g plant}^{-1}$ ) was from 21 days waterlogged plants.

Fodder quality parameters also showed the response to stress conditions. The ADF content has been reduced under both drought and waterlogging stress condition.

Highest ADF (47.67%) was found in the control plant and the lowest ADF (40.75%) in 21 days' drought-stressed plant. The NDF content has been reduced under both drought and waterlogging stress condition. Highest NDF (77.47%) was found in the control plant and the lowest NDF (58.98%) from 21 days stressed plant. The DM content has been reduced under both drought and waterlogging stress conditions with the increase in the duration of stress. Highest DM (86.99%) was found from 7 days waterlogged plant and the lowest DM (82.48%) from 21 days of a drought-stressed plant.

The CP has increased with the increase of duration of stress under both drought and waterlogging conditions. Highest CP (11.39%) was found from 21 days' of drought-stressed plant and the lowest CP (8.485%) from 7 days' of waterlogged plant. The CF has decreased with the increase in the duration of stress under both drought and waterlogging conditions. Highest CF (24.23%) was found in the control plant and the lowest CP (17.83%) in 21 days' drought-stressed plant. The ash content has increased with the increase in the duration of stress under both drought and waterlogging conditions. The highest ash (21.1%) was found from the control plant and the lowest ash (13.08%) from 7 days' waterlogged plant.

The OM% has decreased with the increase in the duration of stress under both drought and waterlogging conditions. Highest OM (87.9%) was found in the control plant and the lowest OM (89.84%) from 21 days' drought stressed plant. The hemicellulose has decreased with the increase in the duration of stress under both drought and waterlogging conditions. The highest hemicellulose (29.88%) was found from 7 days waterlogged plant and the lowest hemicellulose (18.23%) from 21 day's drought-stressed plant.

Considering these responses, we can conclude that the reduction was higher under drought conditions than in waterlogging. Root length, root shoot ratio, proline, malondialdehyde (MDA) and H<sub>2</sub>O<sub>2</sub> content were higher under stress conditions compared to control. Drought-stressed plants were more severely affected than waterlogged one. At 50 days after sowing, plant height, shoot fresh weight, shoot dry weight and fodder yield were decreased in plants stressed for longer periods. Root fresh weight, root dry weight, root length and root branch were decreased in plants stressed for 21 days, whereas increased under waterlogging. Proline, MDA and H<sub>2</sub>O<sub>2</sub> content

were increased upon exposure to the long duration of stress. As plants stressed for 7 days got the highest days for recovery, so it showed better performance and even better than control. Our experiment concludes that napier grass is more sensitive to drought than waterlogging in case of morphology and plants also recovered more efficiently in case of waterlogging than drought. In case of oxidative damage, drought-exposed plants showed more tolerant capacity compared to waterlogged plants.

## REFERENCES

- Abdelaal, K.A. (2015). Effect of salicylic acid and abscisic acid on morpho-physiological and anatomical characters of faba bean plants (*Vicia faba* L.) under drought stress. *J. Plant Prod.* **6**(11): 1771-1788.
- Abid, M., Mansour, E., Ben Yahia, L., Bachar, K.H., Ben Khaled, A. and Ferchichi, A. (2016). Alfalfa nutritive quality as influenced by drought in South-Eastern Oasis of Tunisia. *Ital. J. Anim Sci.* **15**(2): 334-342.
- Ahmad, P., Ahanger, M.A., Alam, P., Alyemeni, M.N., Wijaya, L. Ali, S. and Ashraf, M. (2019). Silicon (Si) supplementation alleviates NaCl toxicity in mung bean (*Vigna radiata* L. Wilczek) through the modifications of physio-biochemical attributes and key antioxidant enzymes. *J. Plant Growth Regul.* **38**: 70-82.
- Ahmed, N., Rahman, K., Rahman, M., Sathi, K.S., Alam, M.M., Nahar, K., Islam, M.S. and Hasanuzzaman, M. (2021). Insight into the thiourea-induced drought tolerance in two chickpea varieties: Regulation of osmoprotection, reactive oxygen species metabolism and glyoxalase system. *Plant Physiol Biochem.* **167**: 449-458.
- Ahmed, S., Nawata, E. and Sakuratani, T. (2006). Changes of endogenous ABA and ACC, and their correlations to photosynthesis and water relations in mungbean (*Vigna radiata* (L.) Wilczak cv. KPS1) during waterlogging. *Environ. Exp. Bot.* **57**: 278–284.
- Akah, N.P. and Onweluzo, J.C. (2014). Evaluation of Water-Soluble Vitamins and Optimum Cooking Time of Fresh Edible Portions of Elephant Grass (*Pennisetum purpureum* L. Schumach) Shoot. *Niger Food J.* **32**: 120-127.
- Akhtar, I. and Nazir, N. (2013). Effect of waterlogging and drought stress in plants. *Int. J. Water Resour. Environ. Sci.* **2**: 34-40.
- Akter, K.S. and Rahman, M.M. (2012). Spatio-Temporal Quantification and Characterization of Drought Patterns in Bangladesh. *J. Water Environ. Technol.* **10**(3): 277-288.
- Alghabari, F. and Ihsan, M.Z. (2018). Effects of drought stress on growth, grain filling duration, yield and quality attributes of barley (*Hordeum vulgare* L.). *Bangladesh J. Bot.* **47**(3): 421-428.
- Ali, M.A., Pan, G., Cheng, K., Williams, S.A., Ogle, S.M., Yeluripati, J.B., Begum, K., Kuhnert, M., Parton, W.J. and Smith, P. (2019). Modelling greenhouse gas emissions and mitigation potentials in fertilized paddy rice fields in Bangladesh. *Geoderma.* **341**: 206–215.
- Alzahrani, S.M., Alaraidh, I.A., Migdadi, H., Alghamdi, S., Khan, M.A. and Ahmad, P. (2019). Physiological, biochemical, and antioxidant properties of two genotypes of *Vicia faba* grown under salinity stress. *Pak. J. Bot.* **51**(3): 786-798.

- Amoah, J.N. and Seo, Y.W. (2021). Effect of progressive drought stress on physio-biochemical responses and gene expression patterns in wheat. *3 Biotech.* **11**(10): 1-18.
- Amri, M., El Ouni, M.H. and Salem, M.B. (2014). Waterlogging affect the development, yield and components, chlorophyll content and chlorophyll fluorescence of six bread wheat genotypes (*Triticum aestivum* L.). *Bulg. J. Agric. Sci.* **20**(3): 647-657.
- Anee, T.I., Nahar, K., Rahman, A., Mahmud, J.A., Bhuiyan, T.F., Alam, M.U., Fujita, M. and Hasanuzzaman, M. (2019). Oxidative damage and antioxidant defense in *Sesamum indicum* after different waterlogging durations. *Plants.* **8**: 196.
- Anjum, S.A., Ashraf, U., Tanveer, M., Khan, I., Hussain, S., Shahzad, B., Zohaib, A., Abbas, F., Saleem, M.F., Ali, I. and Wang, L.C. (2017). Drought induced changes in growth, osmolyte accumulation and antioxidant metabolism of three maize hybrids. *Front. plant sci.* **8**: 69.
- Anjum, S.A., Wang, L.C., Farooq, M., Hussain, M., Xue, L.L. and Zou, C.M. (2011). Brassinolide application improves the drought tolerance in maize through modulation of enzymatic antioxidants and leaf gas exchange. *J. Agron. crop sci.* **197**(3): 177-185.
- AOAC (Association of Official Analytical Chemist). 2005. Official Methods of Analysis of the Association of Official Analytical Chemist. Benjamin Franklin Station. Washington.
- Asaduzzaman, M. (2019). Effect of Napier Grass on Economic Turkey Production in Bangladesh. *Indian J. Sci. Technol.* **12**(24). DOI: 10.17485/ijst/2019/v12i24/144894.
- Asha, S.N., Sultana, N., Hassan, L., Akhter, S. and Robin, A.H.K. (2021). Response of morphological and biochemical traits of maize genotypes under waterlogging stress. *J. Phytol.* **13**: 0108-0121.
- Ashraf, M. (2009). Biotechnological approach of improving plant salt tolerance using antioxidants as markers. *Biotechnol. Adv.* **27**: 84–93.
- Ashraf, M.A. (2012). Waterlogging stress in plants: A review. *Afr. J. Agril. Res.* **7**(13): 1976–1981.
- Bajpai, S. and Chandra, R. (2015). Effect of Waterlogging Stress on Growth Characteristics and Sod Gene Expression in Sugarcane. *Int. J. Sci Res. Publ.* **5**(1).
- Balazadeh, M., Zamanian, M., Golzardi, F. and Torkashvand, A.M. (2021). Effects of Limited Irrigation on Forage Yield, Nutritive Value and Water Use Efficiency of Persian Clover (*Trifolium Resupinatum*) Compared to Berseem Clover (*Trifolium Alexandrinum*). *Commun. Soil Sci. Plant Anal.* **52**:16: 1927-1942.



- Bandurska, H. and Józwiak, W. (2010). A comparison of the effects of drought on proline accumulation and peroxidases activity in leaves of *Festuca rubra* L. and *Lolium perenne* L. *Acta Soc. Bot. Pol.* **79**: 111–116.
- Bansal, R., Sharma, S., Tripathi, K. and Kumar, A. (2019). Waterlogging tolerance in black gram [*Vigna mungo* (L.) Hepper] is associated with chlorophyll content and membrane integrity. *Ind. J. Biochem. Biophys.* **56**: 81-85.
- Barickman, T.C., Simpson, C.R. and Sams, C.E. (2019). Waterlogging causes early modification in the physiological performance, carotenoids, chlorophylls, proline, and soluble sugars of cucumber plants. *Plants.* **8**:160.
- Barrs, H.D. and Weatherley, P.E. (1962). A re-examination of the relative turgidity technique for estimating water deficits in leaves. *Aust. J. Biol. Sci.* **15**(3): 413-428.
- Bates, L.S., Waldren, R.P. and Teare, I.D. (1973). Rapid determination of free proline for water-stress studies. *Plant soil.* **39**(1): 205-207.
- BER (2013). Bangladesh Economic Review, Ministry of Finance, Government of the Peoples' Republic of Bangladesh, Dhaka.
- Bhargava, S. and Sawant, K. (2013). Drought stress adaptation: metabolic adjustment and regulation of gene expression. *Plant Breed.* **132**: 21–32.
- Bhuiyan, T.F., Ahamed, K.U., Nahar, K., Mahmud, J.A., Bhuyan, M.H.M.B., Anee, T.I., Fujita, M. and Hasanuzzaman, M. (2019). Mitigation of PEG-induced drought stress in rapeseed (*Brassica rapa* L.) by exogenous application of osmolytes. *Biocatal. Agric. Biotechnol.* **20**: 101197.
- Bibi, A., Sadaqat, H.A., Tahir, M.H.N. and Akram, H.M. (2012). Screening of sorghum (*Sorghum bicolor* var Moench) for drought tolerance at seedling stage in polyethylene glycol. *J. Anim. Plant Sci.* **22**(3): 671-678.
- Bogdan, A.V. (1977). Tropical pastures and fodder plants. Longman, London.
- Boyle, R.K.A., McAinsh, M. and Dodd, I.C. (2016). Stomatal closure of *Pelargonium x hortorum* in response to soil water deficit is associated with decreased leaf water potential only under rapid soil drying. *Physiol. Plant.* **156**: 84–96.
- Cardoso, J.A., Pineda, M., de la Cruz Jiménez, J., Vergara, M.F. and Rao, I.M. (2015). Contrasting strategies to cope with drought conditions by two tropical forage C<sub>4</sub> grasses. *AoB Plants.* **7**.
- Catola, S., Marino, G., Emiliani, G., Huseynova, T., Musayev, M., Akparov, Z. and Maserti, B.E. (2016). Physiological and metabolomic analysis of *Punica granatum* (L.) under drought stress. *Planta.* **243**(2): 441-449.
- Chimungu, J.G., Brown, K.M. and Lynch, J.P. (2014). Reduced root cortical cell file number improves drought tolerance in maize. *Plant Physiol.* **166**: 1943–1955.

- Colmer, T.D. and Voesenek, L.A.C.J. (2009). Flooding tolerance: suites of plant traits in variable environments. *Func. Plant Biol.* **36**(8): 665-681.
- Comas, L.H., Becker, S.R., Cruz, V.M.V., Byrne, P.F. and Dierig, D.A. (2013). Root traits contributing to plant productivity under drought. *Front. Plant Sci.* **4**:442.
- CoStat, (2008). CoStat-Statistics Software version 6.400. CoHort Software. 798 Lighthouse Ave, PMB 320, Monterey, CA, 93940, USA.
- Dasgupta, S., Huq, M., Khan, Z.H., Sohel Masud, M., Ahmed, M.M.Z., Mukherjee, N. and Pandey, K. (2011). Climate Proofing Infrastructure in Bangladesh: The Incremental Cost of Limiting Future Flood Damage. *J. Environ. Dev.* **20**: 167–190.
- de Morais, R.F., Quesada, D.M., Reis, V.M., Urquiaga, S., Alves, B.J. and Boddey, R.M. (2012). Contribution of biological nitrogen fixation to Elephant grass (*Pennisetum purpureum* Schum.). *Plant Soil.* **356**(1): 23-34.
- Delaporte, I. and Maurel, M., (2018). Adaptation to climate change in Bangladesh. *Clim. Policy.* **18**: 49–62.
- Dietz, K.J., Zörb, C. and Geilfus, C.M. (2021). Drought and crop yield. *Plant Biol.* **23**(6): 881-893.
- Dokbua, B., Waramit, N., Chaugool, J. and Thongjoo, C. (2021). Biomass Productivity, Developmental Morphology, and Nutrient Removal Rate of Hybrid Napier Grass (*Pennisetum purpureum* x *Pennisetum americanum*) in Response to Potassium and Nitrogen Fertilization in a Multiple-Harvest System. *Bioenerg. Res.* **14**: 1106–1117.
- Du, Y., Zhao, Q., Chen, L., Yao, X., Zhang, W., Zhang, B. and Xie, F. (2020). Effect of drought stress on sugar metabolism in leaves and roots of soybean seedlings. *Plant Physiol Biochem.* **146**: 1-12.
- Dutta, P., Chakraborti, S., Chaudhuri, K.M. and Mondal, S. (2020). Physiological responses and resilience of plants to climate change. **In**: New frontiers in stress management for durable agriculture. A. Rakshit, H.B. Singh, A.K. Singh, U.S. Singh, L. Fraceto, (eds.). Springer, Singapore. pp 3–20.
- El Bassam, N. (2010). Handbook of bioenergy crops: a complete reference to species, development and applications. Routledge.
- Elanchezhian, N. and Reddy, D.V. (2009) Nutritional Evaluation of Co 3 Grass in Goats. *Indian J. Anim. Sci.* **79**: 252-253.
- El-Banna, M.F. and Abdelaal, K.A. (2018). Response of strawberry plants grown in the hydroponic system to pretreatment with H<sub>2</sub>O<sub>2</sub> before exposure to salinity stress. *J. Plant Prod.* **9**(12): 989-1001.
- Emerson, R., Hoover, A., Ray, A., Lacey, J., Cortez, M., Payne, C., Karlen, D., Birrell, S., Laird, D., Kallenbach, R. and Voigt, T. (2014). Drought effects on

- composition and yield for corn stover, mixed grasses, and *Miscanthus* as bioenergy feedstocks. *Biofuels*. **5**(3): 275-291.
- Engelbrecht, B.M., Comita, L.S., Condit, R., Kursar, T.A., Tyree, M.T., Turner, B.L. and Hubbell, S.P. (2007). Drought sensitivity shapes species distribution patterns in tropical forests. *Nature*. **447**(7140): 80-82.
- Fàbregas, N. and Fernie, A.R. (2019). The metabolic response to drought. *J. Exp. Bot.* **70**(4): 1077-1085.
- FAO (2019). [http://www.fao.org/nr/water/topics\\_qual\\_waterlogging.html](http://www.fao.org/nr/water/topics_qual_waterlogging.html) (accessed on 22 August, 2022).
- Fariaszewska, A., Aper, J., Van Huylenbroeck, J., Baert, J., De Riek, J., Staniak, M. and Pecio, Ł. (2016). Mild drought stress-induced changes in yield, physiological processes and chemical composition in *Festuca*, *Lolium* and *Festulolium*. *J. Agric. Crop Res.* **203**: 103–116.
- Fariaszewska, A., Aper, J., Van Huylenbroeck, J., De Swaef, T., Baert, J. and Pecio, Ł. (2020). Physiological and biochemical responses of forage grass varieties to mild drought stress under field conditions. *Int. J. Plant Prod.* **14**(2): 335-353.
- Faruque, M.G. (2003). Adoption improved livestock production in practices by farmers. *Progress. Agric.* **14**(1&2): 151- 155.
- Feng, X., Porporato, A. and Rodriguez-Iturbe, I. (2013). Changes in rainfall seasonality in the tropics. *Nat. Clim. Chang.* **3**: 811–815.
- Finance Division. (2018). Bangladesh Economic review. Ministry of Finance, Government of the People’s Republic of Bangladesh, Dhaka.
- Fukao, T., Barrera-Figueroa, B.E., Juntawong, P. and Peña-Castro, J.M. (2019). Submergence and waterlogging stress in plants: A review. Highlighting research opportunities and understudied aspects. *Front. Plant Sci.* **10**: 340.
- Fulkerson, W.J., Neal, J.S., Clark, C.F., Horadagoda, A., Nandra, K.S. and Barchia, I. (2007). Nutritive value of forage species grown in the warm temperate climate of Australia for dairy cows, grasses and legumes. *Livest. Sci.* **107**: 253-264.
- Gall, H.L., Philippe, F., Domon, J.M., Gillet, F., Pelloux, J. and Rayon, C. (2015). Cell wall metabolism in response to abiotic stress. *Plants*. **4**(1): 112-166.
- Gong, Z., Xiong, L., Shi, H., Yang, S., Herrera-Estrella, L.R., Xu, G., Chao, D.Y., Li, J., Wang, P.Y., Qin, F., Li, J., Ding, Y., Shi, Y., Wang, Y., Yang, Y., Guo, Y. and Zhu, J.K. (2020). Plant abiotic stress response and nutrient use efficiency. *Sci. China Life Sci.* **63**(5), 635-674.
- Gunes, A., Pilbeam, D.J., Inal, A., Bagci, E.G. and Coban, S. (2007). Influence of silicon on antioxidant mechanisms and lipid peroxidation in chickpea (*Cicer arietinum* L.) cultivars under drought stress. *J. Plant Interact.* **2**(2): 105-113.

- Gupta, A., Rico-Medina, A., and Caño-Delgado, A.I. (2020). The physiology of plant responses to drought. *Science*. **17**: 266-269.
- Habiba, U., Shaw, R. and Takeuchi, Y. (2014). Farmers' adaptive practices for drought risk reduction in the northwest region of Bangladesh. *Nat. Hazards*. **72**: 337–359.
- Hasanuzzaman M., Bhuyan M.H.M., Nahar, K., Hossain, M., Mahmud, J.A., Hossen, M. and Fujita, M. (2018c). Potassium: a vital regulator of plant responses and tolerance to abiotic stresses. *Agronomy*. **8**(3): 31.
- Hasanuzzaman, M., Al Mahmud, J., Anee, T.I., Nahar, K. and Islam, M.T. (2018a). Drought stress tolerance in wheat: omics approaches in understanding and enhancing antioxidant defense. **In**: Abiotic stress-mediated sensing and signaling in plants: an omics perspective. Springer, Singapore. pp. 267-307.
- Hasanuzzaman, M., Bhuyan, M.H.M.B., Anee, T.I., Parvin, K., Nahar, K., Mahmud, J.A., Fujita, M. (2019). Regulation of Ascorbate-Glutathione Pathway in Mitigating Oxidative Damage in Plants under Abiotic Stress. *Antioxidants*. **8**: 384.
- Hasanuzzaman, M., Hossain, M.A., Silva, J.A. and Fujita, M. (2012a). Plant response and tolerance to abiotic oxidative stress: antioxidant defense is a key factor. *In Crop stress and its management: perspectives and strategies* (pp. 261-315). Springer, Dordrecht.
- Hasanuzzaman, M., Nahar, K., Alam, M.M. and Fujita, M. (2012b). Exogenous nitric oxide alleviates high temperature induced oxidative stress in wheat (*Triticum aestivum* L.) seedlings by modulating the antioxidant defense and glyoxalase system. *Aust. J. Crop Sci.* **6**(8): 1314-1323.
- Hasanuzzaman, M., Nahar, K., Anee, T.I., Khan, M.I.R. and Fujita, M. (2018b). Silicon-mediated regulation of antioxidant defense and glyoxalase systems confers drought stress tolerance in *Brassica napus* L. *South Afr. J. Bot.* **115**: 50-57.
- Hassan, A., Ijaz, M., Sattar, A., Sher, A., Rasheed, I., Saleem, M.Z. and Hussain, I. (2020). Abiotic stress tolerance in cotton. **In**: Advances in Cotton Research. M.U. Rahman (ed.). IntechOpen, London. pp.25-42.
- Heath, R.L. and Packer, L. (1968). Photoperoxidation in isolated chloroplasts in kinetics and stoichiometry of fatty acid peroxidation. *Arch. Biochem. Biophys.* **125**(1): 189-198.
- Herzog, M., Striker, G.G., Colmer, T.D. and Pedersen, O. (2016). Mechanisms of waterlogging tolerance in wheat—A review of root and shoot physiology. *Plant Cell Environ.* **39**: 1068–1086.
- Hossain, M.A. and Uddin, S.N. (2011). Mechanisms of waterlogging tolerance in wheat: Morphological and metabolic adaptations under hypoxia or anoxia. *Aust. J. Crop Sci.* **5**: 1094–1101.

- Hussain, H.A., Men, S., Hussain, S., Chen, Y., Ali, S., Zhang, S. and Wang, L. (2019). Interactive effects of drought and heat stresses on morpho-physiological attributes, yield, nutrient uptake and oxidative status in maize hybrids. *Sci. Rep.* **9**(1): 1-12.
- Hussain, S., Zhang, J.H., Zhong, C., Zhu, L.F., Cao, X.C., Yu, S.M., Bohr, J.A., Hu, J.J. and Jin, Q.Y. (2017). Effects of salt stress on rice growth, development characteristics, and the regulating ways: A review. *J. Integr Agric.* **16**(11): 2357-2374.
- Irfan, M., Hayat, S., Hayat, Q., Afroz, S. and Ahmad, A. (2010). Physiological and biochemical changes in plants under waterlogging. *Protoplasma.* **241**(1): 3-17.
- Ishrath, P.K., Thomas, U.C. and Dhanya, G.A.N.E.S.H. (2018). Effect of cutting intervals on yield and quality fodder production in hybrid napier. *Forage Res.* **44**(2): 137-140.
- Islam, A.R.M., Shen, S., Hu, Z. and Rahman, M.A. (2017). Drought hazard evaluation in boro paddy cultivated areas of western Bangladesh at current and future climate change conditions. *Adv. Meteorol.* DOI: 10.1155/2017/3514381.
- Islam, M.T. and Nursey-Bray, M. (2017). Adaptation to climate change in agriculture in Bangladesh: The role of formal institutions. *J. Environ. Manage.* **200**: 347-358.
- Islam, M.R. and Das, S. (2014). Assessment of Waterlogging and Landslide Vulnerability Using CVAT Tool in Chittagong City Corporation Area; Chittagong University of Engineering and Technology: Chattogram, Bangladesh.
- Jabbari, H., Akbari, G.A., Sima, N.A.K.K., Rad, A.H.S., Alahdadi, I., Hamed, A. and Shariatpanahi, M.E. (2013). Relationships between seedling establishment and soil moisture content for winter and spring rapeseed genotypes. *Ind. Crops Prod.* **49**: 177-187.
- Jahanzad, E., Jorat, M., Moghadam, H., Sadeghpour, A., Chaichi, M.R. and Dashtaki, M. (2013). Response of a new and a commonly grown forage sorghum cultivar to limited irrigation and planting density. *Agric. Water Manag.* **117**: 62–69.
- Jain, M., Mathur, G, Koul, S. and Sarin, N.B. (2011). Ameliorative effects of proline on salt stress-induced lipid peroxidation in cell lines of groundnut (*Arachis hypogea* L). *Plant Cell Rep.* **20**: 463.
- Jalil, S.U., Ahmad, I. and Ansari, M.I. (2017). Functional loss of GABA transaminase (GABA-T) expressed early leaf senescence under various stress conditions in *Arabidopsis thaliana*. *Curr. Plant Biol.* **9**: 11–22.
- Jaspers, P. and Kangasjärvi, J. (2010). Reactive oxygen species in abiotic stress signaling. *Physiol. Plant.* **138**(4): 405-413.

- Jiang, C., Cui, Q., Feng, K., Xu, D., Li, C. and Zheng, Q. (2016). Melatonin improves antioxidant capacity and ion homeostasis and enhances salt tolerance in maize seedlings. *Acta physiol. Plant.* **38**(4): 1-9.
- Jiang, Y., Liang, G. and Yu, D. (2012). Activated expression of WRKY57 confers drought tolerance in Arabidopsis. *Mol. plant.* **5**(6): 1375-1388.
- Joshi, M. (2015). Napier Grass. In: Textbook of field crops. Ghosh, A.K. PHI Learning Private Limited, Rimjhim House, 111, Patpargay Industrial Estate, Delhi-110092.
- Kabiri, R., Nasibi, F. and Farahbakhsh, H. (2014). Effect of exogenous salicylic acid on some physiological parameters and alleviation of drought stress in *Nigella sativa* plant under hydroponic culture. *Plant Protec. Sci.* **50**(1): 43-51.
- Kabirizi, J., Ziiwa, E., Mugerwa, S., Ndikumana, J. and Nanyennya, W. (2013). Dry Season Forages for Improving Dairy Production in Smallholder Systems in Uganda. *Trop. Grassl. - Forrajes Trop.* **1**: 212-214.
- Katerji, N., Hoorn, J.W.V., Hamdy, A., Mastrorilli, V and Karzel, E.M. (1997). "Osmotic adjustment of sugar beets in response to soil salinity and its influence on stomatal conductance, growth and yield." *Agric. Water Manag.* **34**(1). Pp. 57-69.
- Kaur, G., Singh, G., Motavalli, P.P., Nelson, K.A., Orlowski, J.M. and Golden, B.R. (2020). Impacts and management strategies for crop production in waterlogged or flooded soils: A review. *Agron. J.* **112**(3): 1475-1501.
- Khalid, M., Rahman, H., Farhatullah, F., Rabbani, A., Lightfoot, D.A., Iqbal, M. and Khan, I. (2018). The Effect of Two Different Agro-Climatic Conditions on Growth and Yield Performance of Sugarcane Genotypes. *Plant Gene Trait.* **9**.
- Khalil, N., Fekry, M., Bishr, M., El-Zalabani, S. and Salama, O. (2018). Foliar spraying of salicylic acid induced accumulation of phenolics, increased radical scavenging activity and modified the composition of the essential oil of water stressed *Thymus vulgaris* L. *Plant Physiol. Biochem.* **123**: 65-74.
- Khan, M.N., Zhang, J., Luo, T., Liu, J., Ni, F., Rizwan, M., Fahad, S. and Hu, L. (2019). Morpho-physiological and biochemical responses of tolerant and sensitive rapeseed cultivars to drought stress during early seedling growth stage. *Acta Physiol. Plant.* **41**: 25.
- Küchenmeister, K., Küchenmeister, F., Kayser, M., Wrage-Mönnig, N., Isselstein, J. (2013). Influence of drought stress on nutritive value of perennial forage legumes. *Int. J. Plant Prod.* **7**(4): 693-710.
- Kumar, P., Pal, M., Joshi, R. and Sairam, R.K. (2013). "Yield, growth and physiological responses of mung bean [*Vigna radiata* (L.) Wilczek] genotypes to waterlogging at vegetative stage." *Physiol. Mol. Biol. Plants.* **19**(2): 209-220.

- Leng, G.Y., Tang, Q.H. and Rayburg, S. (2015). Climate change impacts on meteorological, agricultural and hydrological droughts in China. *Glob Planet Change*. **126**: 23–34.
- Li, F., Han, Y., Feng, Y., Xing, S., Zhao, M., Chen, Y. and Wang, W. (2013). Expression of wheat expansin driven by the RD29 promoter in tobacco confers water-stress tolerance without impacting growth and development. *J. Biotech*. **163**(3): 281-291.
- Liu, D., Pei, Z.F., Naeem, M.S., Ming, D.F., Liu, D.F., Khan, F. and Zhou, W.J. (2011). 5-Aminolevulinic acid activates antioxidative defence system and seedling growth in *Brassica napus* L. under water-deficit stress. *J. Agron. Crop Sci*. **197**(4): 284–295.
- Liu, D., Wu, L., Naeem, M.S., Liu, H., Deng, X., Xu, L., Zhang, F., Zhou, W. (2013). 5-Aminolevulinic acid enhances photosynthetic gas exchange, chlorophyll fluorescence and antioxidant system in oilseed rape under drought stress. *Acta Physiol. Plant*. **35**: 2747–2759.
- Liu, J., Hasanuzzaman, M., Wen, H., Zhang, J., Peng, T., Sun, H. and Zhao, Q. (2019). High temperature and drought stress cause abscisic acid and reactive oxygen species accumulation and suppress seed germination growth in rice. *Protoplasma*. **256**(5): 1217-1227.
- Liu, M., Hulting, A. and Mallory-Smith, C. (2017). Comparison of growth and physiological characteristics between roughstalk bluegrass and tall fescue in response to simulated waterlogging. *PLoS One*. **12**(7): e0182035.
- Lounglawan, P., Lounglawan, W. and Suksombat, W. (2014). Effect of cutting interval and cutting height on yield and chemical composition of King Napier grass (*Pennisetum purpureum* x *Pennisetum americanum*). *APCBEE procedia*. **8**: 27-31.
- Mahar, A., Wang, P., Ali, A., Awasthi, M.K., Lahori, A.H., Wang, Q., Li, R. and Zhang, Z. (2016). Challenges and opportunities in the phytoremediation of heavy metals contaminated soils: A review. *Ecotoxicol. Environ. Saf*. **126**: 111–121.
- Maleko, D., Mwilawa, A., Msalya, G., Pasape, L. and Mtei, K. (2019). Forage Growth, Yield and Nutritional Characteristics of Four Varieties of Napier Grass (*Pennisetum purpureum* Schumach) in the West Usambara Highlands, Tanzania. *Sci. Afr*. **6**: e00214.
- Malik, A.I., Colmer, D.T.D., Lambers, H. and Schortemeyer, M. (2001). Changes in physiological and morphological traits of roots and shoots of wheat in response to different depth of waterlogging. *Aust. J. Plant Physiol*. **28**: 1121-1131.
- Manik, S.M.N., Pengilley, G., Dean, G., Field, B., Shabala, S. and Zhou, M. (2019). Soil and crop management practices to minimize the impact of waterlogging on crop productivity. *Front. Plant Sci*. **10**: 140.

- Marsalis, M.A., Angadi, F., Contreras-Govea, S.F. and Kirksey, R.E. (2009). Harvest timing and byproduct addition effects on corn and forage sorghum silage grown under water stress. *Bull.* **799**: 1–16.
- Mcfarlane, N.M., Ciavarella, T.A. and Smith, K.F. (2004). The effects of waterlogging on growth, photosynthesis and biomass allocation in perennial ryegrass (*Lolium perenne* L.) *J. Agric. Sci.* **141**: 241-248.
- Mehmood, M., Khan, I., Chattha, M.U., Hussain, S., Ahmad, N., Aslam, M.T., Hafeez, M.B., Hussan, M., Hassan, M.U., Nawaz, M. and Hussain, F. (2021). Thiourea application protects maize from drought stress by regulating growth and physiological traits. *Pak. J. Sci.* **73**(2): 355.
- Meisser, M., Vitra, A., Deleglise, C., Dubois, S., Probo, M., Mosimann, E., Buttler, A. and Mariotte, P. (2019). Nutrient limitations induced by drought affect forage N and P differently in two permanent grasslands. *Agric. Ecosyst. Environ.* **280**: 85-94.
- Mhamdi, A. and Breusegem, F.V. (2018). Reactive oxygen species in plant development. *Development.* **145** (15): dev164376.
- Miller, G., Suzuki, N., Ciftci-Yilmaz, S. and Mittler, R. (2010). Reactive oxygen species homeostasis and signalling during drought and salinity stresses. *Plant Cell Environ.* **33**: 453–467.
- Misra, M. (2017). Smallholder agriculture and climate change adaptation in Bangladesh: questioning the technological optimism. *Clim. Dev.* **9**: 337–347.
- Misra, V., Solomon, S. and Ansari, M.I. (2016). Impact of drought on post-harvest quality of sugarcane crop. *Adv. Life Sci.* **20**: 9496–9505.
- Misra, V., Solomon, S., Mall, A.K., Prajapati, C.P., Hashem, A., Abd-Allah, E.F. and Ansari, M.I. (2020). Morphological assessment of water stressed sugarcane: A comparison of waterlogged and drought affected crop. *Saudi J. Biol. Sci.* **27**: 1228–1236.
- Mkhutche, C.D. (2020). Evaluation of feed resources for local goat production under traditional management systems in Golomoti EPA Dedza and on-station at Bunda Campus, LUANAR, Malawi. Phd Thesis. International Institute of Tropical Agriculture.
- Moore, J.P., Vire-Gibouin, M., Farrant, J.M. and Driouich, A. (2008). Adaptations of higher plant cell walls to water loss, drought vs desiccation. *Physiol. Plant.* **134**: 237-245.
- Munns, R., James, R.A. and Läuchli, A. (2006). Approaches to increasing the salt tolerance of wheat and other cereals. *J. Exp. Bot.* **57** (5): 1025–43.
- Nagasuga, K. (2003). Effect of growth light intensity on the water transport regulation and leaf photosynthesis in napier grass (*Pennisetum purpureum* Schumach.). *Bull. Inst. Trop. Agr. Kyushu Univ.* **26**: 895-905.



- Nahar, K., Hasanuzzaman, M., Alam, M., Rahman, A., Mahmud, J.A., Suzuki, T. and Fujita, M. (2017). Insights into spermine-induced combined high temperature and drought tolerance in mung bean: osmoregulation and roles of antioxidant and glyoxalase system. *Protoplasma*. **254**(1): 445-460.
- Nahar, K., Hasanuzzaman, M., Alam, M.M., Rahman, A., Suzuki, T. and Fujita, M. (2016). Polyamine and nitric oxide crosstalk: Antagonistic effects on cadmium toxicity in mung bean plants through upregulating the metal detoxification, antioxidant defense and methylglyoxal detoxification systems. *Ecotoxicol. Environ. Saf.* **126**: 245-255.
- Nasrin, S., Saha, S., Begum, H.H. and Samad, R. (2020). Impacts of drought stress on growth, protein, proline, pigment content and antioxidant enzyme activities in rice (*Oryza sativa* L. var. BRRI dhan-24). *Dhaka Univ. J. Biol. Sci.* **29**(1): 117-123.
- Negawo, A.T., Teshome, A., Kumar, A., Hanson, J. and Jones, C.S. (2017). Opportunities for Napier Grass (*Pennisetum purpureum*) Improvement Using Molecular Genetics. *Agronomy*. **7**: 28.
- Noctor, G., Reichheld, J.P., Christine, H. and Foyer. (2018). ROS-related redox regulation and signaling in plants. *Semin. Cell Dev. Biol.* **80**: 3-12.
- Nouri, E., Matinizadeh, M., Moshki, A.R., Zolfaghari, A.A., Rajaei, S. and Janoušková, M. (2020). Arbuscular mycorrhizal fungi benefit drought-stressed *Salsola laricina*. *Plant Ecol.* **221**: 683-694.
- OECD/FAO. (2019). [https://doi.org/10.1787/agr\\_outlook-2019-en](https://doi.org/10.1787/agr_outlook-2019-en). Accessed on 17 September 2020.
- Onwugbuta-Enyi, J. (2004). Water balance and proximate composition in cowpea (*Vigna unguiculata* (L.) Walps) seedlings exposed to drought and flooding stress. *J. Appl. Sci. Environ. Manag.* **8**: 55–57.
- Orodho, A.B. (2006). The Role and Importance of Napier Grass in the Smallholder Dairy Industry in Kenya. Food and Agriculture Organization, Rome. P. 2011.
- Osuagwu, G.G.E. and Edeoga, H.O. (2013). The effect of water stress (drought) on the proximate composition of the leaves of *Ocimum gratissimum* (L) and *Gongronema latifolium* (Benth). *Int. J. Med. Aromat. Plants.* **3**(2): 293-299.
- Pais, I.P., Moreira, R., Semedo, J.N., Reboredo, F.H., Lidon, F.C., Maças, B. and Scotti-Campos, P. (2021, November). Effects of Waterlogging on Growth and Development of Bread Wheat Genotypes. *Biol. Life Sci. Forum.* **11**(1): 38).
- Palta, J.A., Chen, X., Milroy, S.P., Rebeztke, G.J., Dreccer, M.F. and Watt, M. (2011). Large root systems: are they useful in adapting wheat to dry environments? *Funct. Plant Biol.* **38**: 347–354.
- Pampana, S., Masoni, A. and Arduini, I. (2016). Response of cool-season grain legumes to waterlogging at flowering. *Can. J. Plant Sci.* **96**(4): 597-603.

- Patel, P.K., Hemantaranjan, A. and Sarma, B.K. (2012). Effect of salicylic acid on growth and metabolism of chickpea (*Cicer arietinum* L.) under drought stress. *Indian J. Plant Physiol.* **17**(2): 151-157.
- Perlikowski, D., Kosmala, A., Rapacz, M., Kościelniak, J., Pawłowicz, I. and Zwierzykowski, Z. (2014). Influence of short-term drought conditions and subsequent re-watering on the physiology and proteome of *Lolium multiflorum*/*Festuca arundinacea* introgression forms, with contrasting levels of tolerance to long-term drought. *Plant Biol.* **16**: 385–394.
- Perrier, L., Rouan, L., Jaffuel, S., Clément-Vidal, A., Roques, S., Soutiras, A., Baptiste, C., Bastianelli, D., Fabre, D., Dubois, C., Pot, D. and Luquet, D. (2017). Plasticity of sorghum stem biomass accumulation in response to water deficit, a multiscale analysis from internode tissue to plant level. *Front. Plant Sci.* **8**: 1-14.
- Phaenark, C., Pokethitiyook, P., Kruatrachue, M. and Ngernsarsaruay, C. (2009). Cd and Zn accumulation in plants from the Padaeng zinc mine area. *Int. J. Phytoremediat.* **11**: 479–495
- Ploschuk, R.A., Grimoldi, A.A., Ploschuk, E.L. and Striker, G.G. (2017). Growth during recovery evidences the waterlogging tolerance of forage grasses. *Crop Pasture Sci.* **68**(6): 574-582.
- Prasanna, Y.L. and Rao, G.R. (2014). Effect of waterlogging on growth and seed yield in greengram genotypes. *Int. J. Food Agric. Vet. Sci.* **4**: 124-128.
- Promkhambut, A., Younger, A., Polthanee, A. and Akkasaeng, C. (2010). Morphological and physiological responses of sorghum (*Sorghum bicolor* L. Moench) to waterlogging. *Asian J. Plant Sci.* **9**(4): 183.
- Purbajanti, E.D., Anwar, S., Wydiati and Kusmiyati, F. (2012). Drought stress effect on morphology characters, water use efficiency, growth and yield of guinea and napier grasses. *Int. Res. J. Plant Sci.* **3**(4): 47-53.
- Qaseem, M.F., Qureshi, R. and Shaheen, H. (2019). Effects of pre-anthesis drought, heat and their combination on the growth, yield and physiology of diverse wheat (*Triticum aestivum* L.) genotypes varying in sensitivity to heat and drought stress. *Sci. Rep.* **9**(1): 1-12.
- Rahman, K., Rahman, M., Ahmed, N., Alam, M.M., Rahman, A., Islam, M.M. and Hasanuzzaman, M. (2021). Morphophysiological changes and reactive oxygen species metabolism in *Corchorus olitorius* L. under different abiotic stresses. *Open Agric.* **6**(1): 549-562.
- Rahman, M.A., Atiqur, M., Yunsheng, L. and Sultana, N. (2017). Analysis and prediction of rainfall trends over Bangladesh using Mann–Kendall, Spearman’s rho tests and ARIMA model. *Meteorol. Atmos. Phys.* **129**: 409–424.
- Rakszegi, M., Lovegrove, A., Balla, K., Láng, L., Bedő, Z., Veisz, O. and Shewry, P.R. (2014). Effect of heat and drought stress on the structure and composition of arabinoxylan and  $\beta$ -glucan in wheat grain. *Carbohydr. Polym.* **102**: 557-565.

- Rasaei, A., Ghobadi, M.E., Jalali-Honarmand, S., Ghobadi, M. and Saeidi, M. (2012). Impacts of waterlogging on shoot apex development and recovery effects of nitrogen on grain yield of wheat. *Eur. J. Exp. Biol.* **2**(4): 1000-1007.
- Rasheed, A., Hassan, M.U., Aamer, M., Batool, M., Sheng, F.A.N.G., Ziming, W.U. and Huijie, L.I. (2020). A critical review on the improvement of drought stress tolerance in rice (*Oryza sativa* L.). *Not. Bot. Horti Agrobot. Cluj. Napoca.* **48**(4).
- Raza, A., Razzaq, A., Mehmood, S.S., Zou, X., Zhang, X., Lv, Y. and Xu, J. (2019). Impact of climate change on crops adaptation and strategies to tackle its outcome: A review. *Plants.* **8**: 34.
- Ren, B., Dong, S., Liu, P., Zhao, B. and Zhang, J. (2016a). Ridge tillage improves plant growth and grain yield of waterlogged summer maize. *Agric. Water Manag.* **177**: 392–399.
- Ren, B., Zhang, J., Dong, S., Liu, P. and Zhao, B. (2016b). Root and shoot responses of summer maize to waterlogging at different stages. *Agron. J.* **108**(3): 1060-1069.
- Ren, B., Zhang, J., Li, X., Fan, X., Dong, S., Liu, P. and Zhao, B. (2014). Effects of waterlogging on the yield and growth of summer maize under field conditions. *Can. J. Plant Sci.* **94**: 23–31.
- Rengsirikul K, Ishii Y, Kangvansaichol K, Sripichitt P, Punsuvon V, Vaithanomsat P, Nakamanee G, Tudsri S. (2013). Biomass yield, chemical composition and potential ethanol yields of 8 cultivars of napier grass (*Pennisetum purpureum* Schumach) Harvested 3- monthly in Central Thailand. *J. Sustain. Bioenergy Syst.* **3**:107-112.
- Rostamza, M., Chaichi, M.R., Jahansouz, M.R. and Alimadadi, A. (2011). Forage quality, water use and nitrogen utilization efficiencies of pearl millet (*Pennisetum americanum* L.) grown under different soil moisture and nitrogen levels. *Agric. Water Manage.* **98**(10): 1607-1614.
- Rusdy, M. (2016). Elephant grass as forage for ruminant animals. *Livest. Res. Rural Dev.* **28**: 49.
- Sabra, A., Daayf, F. and Renault, S. (2012). Differential physiological and biochemical responses of three Echinacea species to salinity stress. *Scientia Hort.* **135**: 23–31
- Saha, S., Begum, H.H. and Nasrin, S. (2019). Effects of drought stress on growth and accumulation of proline in five rice varieties (*Oryza Sativa* L.). *J. Asiatic. Soc. Bangladesh Sci.* **45**(2): 241-247.
- Saini, P., Gani, M., Kaur, J.J., Godara, L.C., Singh, C., Chauhan, S.S., Francies, R.M., Bhardwaj, A., Kumar, N.B. and Ghosh, M.K. (2018). Reactive oxygen species (ROS): A way to stress survival in plants. **In:** Abiotic Stress-Mediated Sensing and Signaling in Plants: An Omics Perspective. S.M. Zargar, M.Y. Zargar, (Eds.) Springer, Singapore. pp. 127–153.

- Sairam, R.K., Dharmar, K., Lekshmy, S. and Chinnusamy, V. (2011). Expression of antioxidant defense genes in mung bean (*Vigna radiata* L.) roots under waterlogging is associated with hypoxia tolerance. *Acta Physiol. Plant.* **33**(3): 735-744.
- Sarker, N.R., Habib, M.A., Yeasmin, D., Tabassum, F. and Mohammed, R.A. (2021). Studies on Biomass Yield, Morphological Characteristics and Nutritive Quality of Napier Cultivars under Two Different Geo-Topographic Conditions of Bangladesh. *American J. Plant Sci.* **12**: 914-925.
- Sasidharan, R., Hartman, S., Liu, Z., Martopawiro, S., Sajeev, N., van Veen, H., Yeung, E. and Voesenek, L.A. (2018). Signal dynamics and interactions during flooding stress. *Plant Physiol.* **176**: 1106–1117.
- Sathi, K.S., Masud, A.A.C., Falguni, M.R., Ahmed, N., Rahman, K. and Hasanuzzaman, M. (2022). Screening of soybean genotypes for waterlogging stress tolerance and understanding the physiological mechanisms. *Adv. Agric.* 2022.
- Saud, S., Fahad, S., Yajun, C., Ihsan, M.Z., Hammad, H.M., Nasim, W., Amanullah, J., Arif, M. and Alharby, H. (2017). Effects of nitrogen supply on water stress and recovery mechanisms in kentucky bluegrass plants. *Front. Plant Sci.* **8**: 983.
- Sawasdee, V. and Pisutpaisal, N. (2014). Feasibility of Biogas Production from Napier Grass. *Energy Procedia.* **61**: 1229–1233.
- Sayeed, M.A., Rahman, S.M.A., Alam, J. and Sarker, N.R. (2008). An Economic study on cultivation of fodder and competing crops in some selected areas of Bangladesh. Annual Report of Annual Research Review Workshop. Pp78-79.
- Sehgal, D., Baliyan, N. and Kaur, P. (2019). Progress towards identification and validation of candidate genes for abiotic stress tolerance in wheat. *Cham.* **2**:31-48.
- Shahid, S. and Behrawan, H. (2008). Drought risk assessment in the western part of Bangladesh. *Nat. Hazards.* **46**: 391–413.
- Shahin, M.A., Ali, M.A. and Ali, A.B.M.S. (2014). Vector Autoregression (VAR) Modeling and Forecasting of Temperature, Humidity, and Cloud Coverage. In: T. Islam, P. Srivastava, M. Gupta, X. Zhu, S. Mukherjee. (eds.) Computational Intelligence Techniques in Earth and Environmental Sciences. Springer, Dordrecht.
- Shariatmadari, M.H.; Parsa, M., Nezami, A. and Kafi, M. (2017). The effects of hormonal priming on emergence, growth and yield of chickpea under drought stress in glasshouse and field. *Biosci. Res.* **14**: 34–41.
- Singh, V.P., Srivastava, J.P. and Bansal, R. (2017). Biochemical responses as stress indicator to waterlogging in pigeon pea (*Cajanus cajan* L.). *Indian J. Biochem. Biophys.* **54**: 300.

- Somegowda, V.K., Vemula, A., Naravula, J., Prasad, G., Rayaprolu, L., Rathore, A., Blümmel, M. and Deshpande, S.P. (2021). Evaluation of fodder yield and fodder quality in sorghum and its interaction with grain yield under different water availability regimes. *Curr. Plant Biol.* **25**: 100191.
- Stallmann, J., Schweiger, R., Pons, C.A. and Müller, C. (2020). Wheat growth, applied water use efficiency and flag leaf metabolome under continuous and pulsed deficit irrigation. *Scientific Reports.* **10**(1): 1-13.
- Steffens, B. (2014). The role of ethylene and ROS in salinity, heavy metal, and flooding responses in rice. *Front. Plant Sci.* **5**: 685.
- Subrina, S. and Chowdhury, F.K. (2018). Urban Dynamics: An Undervalued Issue for Water Logging Disaster Risk Management in Case of Dhaka City, Bangladesh. In *Procedia Engineering*; Elsevier Ltd.: Amsterdam, The Netherlands. **212**. pp. 801–808.
- Sun, D., Yang, H., Guan, D., Yang, M., Wu, J., Yuan, F., Jin, C., Wang, A. and Zhang, Y (2018). The effects of land use change on soil infiltration capacity in China: a meta-analysis. *Sci. Total Environ.* **626**:1394–1401
- Sundgren, T.K., Uhlen, A.K., Lillemo, M., Briese, C. and Wojciechowski, T. (2018). Rapid seedling establishment and a narrow root stele promotes waterlogging tolerance in spring wheat. *J. plant physiol.* **227**: 45-55.
- Taïbi, H., Taïbi, F., Abderrahim, L.A., Ennajah, A., Belkhodja, M. and Mulet, J.M. (2016). Effect of salt stress on growth, chlorophyll content, lipid peroxidation and antioxidant defence systems in *Phaseolus vulgaris* L. *S. Afr.J. Bot.* **105**: 306-312.
- Taiz, L. and Zeiger, E. (2010) *Plant Physiology*. (5th Ed, pp. 782). Sinauer Associates Inc., Sunderland.
- Takara, D. and Khanal, S.K. (2015). Characterizing compositional changes of Napier grass at different stages of growth for biofuel and biobased products potential. *Bioresour. Technol.* **188**: 103–108.
- Tani, E., Chronopoulou, E.G., Labrou, N.E., Sarri, E., Goufa, M., Vaharidi, X., Tornesaki, A., Psychogiou, M., Bebeli, P.J. and Abraham, E.M. (2019). Growth, physiological, biochemical, and transcriptional responses to drought stress in seedlings of *Medicago sativa* L., *Medicago arborea* L. and their hybrid (Alborea). *Agronomy.* **9**(1): 38.
- Tariq, A. and Shahbaz, M. (2020). Glycinebetaine induced modulation in oxidative defense system and mineral nutrients sesame (*Sesamum indicum* L.) under saline regimes. *Pak. J. Bot.* **52**: 775-782.
- Tian, L., Li, J., Bi, W., Zuo, S., Li, L., Li, W. and Sun, L.E. (2019). Effects of waterlogging stress at different growth stages on the photosynthetic characteristics and grain yield of spring maize (*Zea mays* L.) under field conditions. *Agric. Water Manag.* **218**: 250–258.

- Tian, L.X., Zhang, Y.C., Chen, P.L., Zhang, F.F., Li, J., Yan, F., Dong, Y. and Feng, B.L. (2021). How does the waterlogging regime affect crop yield? A global meta-analysis. *Front. Plant Sci.* **12**: 634898.
- Trenberth, K.E., Dai, A., Schrier, G., Jones, P.D., Barichivich, J., Briffa, K.R. and Sheffield, J. (2014). Global warming and changes in drought. *Nat. Clim. Change.* **4**: 17–22.
- Tudsri, S. (2005). Tropical Forage. Kasetsart University Publishing, Bangkok, Thailand. Pp. 534.
- Turano, B., Tiwari, U.P. and Jha, R. (2016). Growth and nutritional evaluation of napier grass hybrids as forage for ruminants. *Trop. Grassl.* **4**(3): 168-178.
- United Nations, Department of Economic and Social Affairs, Population Division. (2019). World population prospects 2019: data booklet. Pp. 1-28.
- Upreti, P., Narayan, S., Khan, F., Tewari, L.M. and Shirke, P.A. (2021). Drought-induced responses on physiological performance in cluster bean [*Cyamopsis tetragonoloba* (L.) Taub.]. *Plant Physiol. Rep.* **26**(1): 49-63.
- Vadez, V., Kholova, J., Zaman-Allah, M. and Belko N. (2013). Water: the most important ‘molecular’ component of water stress tolerance research. *Funct. Plant Biol.* **40**: 1310–1322.
- Vandoorne, B., Descamps, C., Mathieu, A.S., Van den Ende, W., Vergauwen, R., Javaux, M. and Lutts, S. (2014). Long term intermittent flooding stress affects plant growth and inulin synthesis of *Cichorium intybus* (var. sativum). *Plant soil.* **376**(1): 291-305.
- Van soest, P.J., Robertson, J.B. and Lewis, B.A (1991). Methods for Dietary Fiber, Neutral Detergent Fiber, and Nonstarch Polysaccharides in Relation to Animal Nutrition. *J Dairy Sci.* **74**:3583-3597
- Vasilakoglou, I., Dhima, K., Karagiannidis, N. and Gatsis, T. (2011). Sweet sorghum productivity for biofuels under increased soil salinity and reduced irrigation. *Field Crops Res.* **120**(1):38–46.
- Waramit, N. and Chaugool, J. (2014). Napier grass: A novel energy crop development and the current status in Thailand. *J. Int. Soc. Southeast Asian Agric. Sci.* **20**(1): 139-150.
- Xie, X. and Shen, J. (2021). Waterlogging resistance evaluation index and photosynthesis characteristics selection: using machine learning methods to judge poplar’s waterlogging resistance. *Mathematics.* **9**(13): 1542.
- Xu, Z., Shen, Q. and Zhang, G. (2022). The mechanisms for the difference in waterlogging tolerance among sea barley, wheat and barley. *Plant Growth Regul.* **96**(3): 431-441.

- Yadav, D.K. and Srivastava, J.P. (2017). Temporal Changes in Biochemical and Antioxidant Enzymes Activities in Maize (*Zea mays* L.) under Waterlogging Stress during Early Growth Stage. *Int. J. Curr. Microbiol. Appl. Sci.* **6**: 351-362.
- Yamauchi, T., Abe, F., Tsutsumi, N. and Nakazono, M. (2019). Root cortex provides a venue for gas-space formation and is essential for plant adaptation to waterlogging. *Front. Plant Sci.* **10**: 259.
- Yan, Q., Li, J., Lu, L., Gao, L., Lai, D., Yao, N., Yi, X., Wu, Z., Lai, Z. and Zhang, J. (2021). Integrated analyses of phenotype, phytohormone, and transcriptome to elucidate the mechanism governing internode elongation in two contrasting elephant grass (*Cenchrus purpureus*) cultivars. *Ind Crops Prod.* **170**: 113693.
- Ye, J., Wang, S., Deng, X., Yin, L., Xiong, B. and Wang, X. (2016). Melatonin increased maize (*Zea mays* L.) seedling drought tolerance by alleviating drought-induced photosynthetic inhibition and oxidative damage. *Acta Physiol. Plant.* **38**: 1–13.
- Yordanova, R.Y., Christov, K.N. and Popova, L.P. (2004). Antioxidative enzymes in barley plants subjected to soil flooding. *Environ. Exp. Bot.* **51**(2): 93-101.
- Yu, C.W., Murphy, T.M. and Lin, C.H. (2003). Hydrogen peroxide induced chilling tolerance in mung beans mediated through ABA-independent glutathione accumulation. *Funct. Plant Biol.* **30**(9): 955–963.
- Zaidi, P.H., Rafique, S., Rai, P.K., Singh, N.N. and Srinivasan, G. (2004). Tolerance to excess moisture in maize (*Zea mays* L.): Susceptible crop growth stage and identification of tolerant genotypes. *Field Crops Res.* **90**: 189-202.
- Zeid, I.M. and Shedeed, Z.A. (2006). Response of alfalfa to putrescine treatment under drought stress. *Biol. Plant.* **50**(4): 635-640.
- Zeng, R., Chen, L., Wang, X., Cao, J., Li, X., Xu, X., Xia, Q., Chen, T. and Zhang, L. (2020). Effect of waterlogging stress on dry matter accumulation, photosynthesis characteristics, yield, and yield components in three different ecotypes of peanut (*Arachis hypogaea* L.). *Agronomy.* **10**(9): 1244.
- Zhang, L., Xu, C. and Champagne, P. (2010). Overview of recent advances in thermochemical conversion of biomass. *Energy Convers. Manage.* **51**: 969–982.
- Zhang, Y., Liu, G., Dong, H. and Li, C. (2021). Waterlogging stress in cotton: Damage, adaptability, alleviation strategies, and mechanisms. *Crop J.* **9**(2): 257-270.

## APPENDICES

### Appendix I. Phenotypic pictures of napier grass under different water stress treatment and recovery

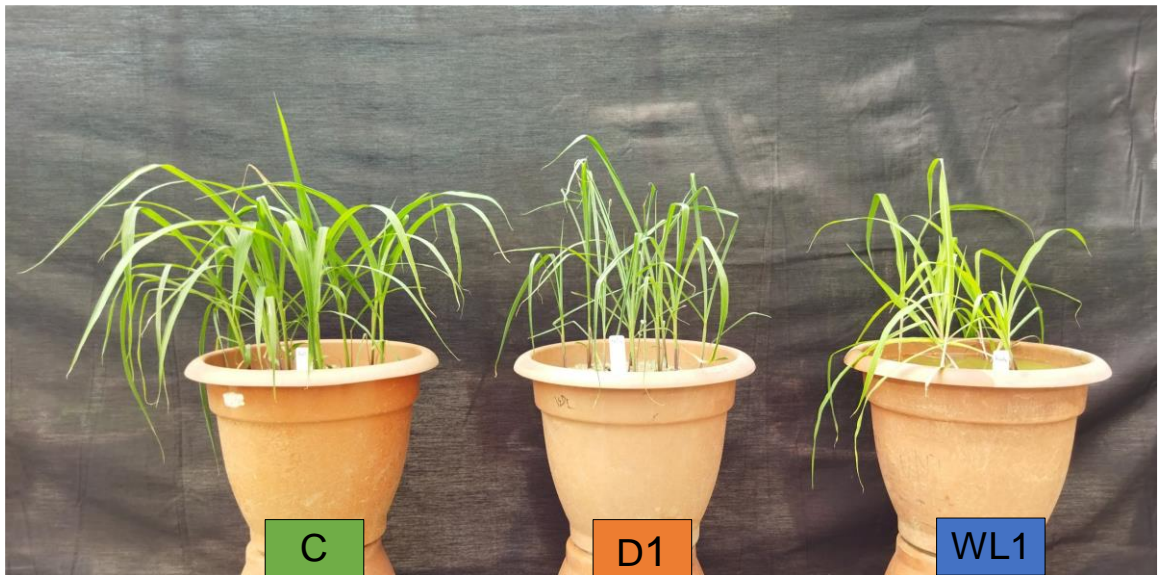


Plate 1. Napier grass after 7 days' stress treatments

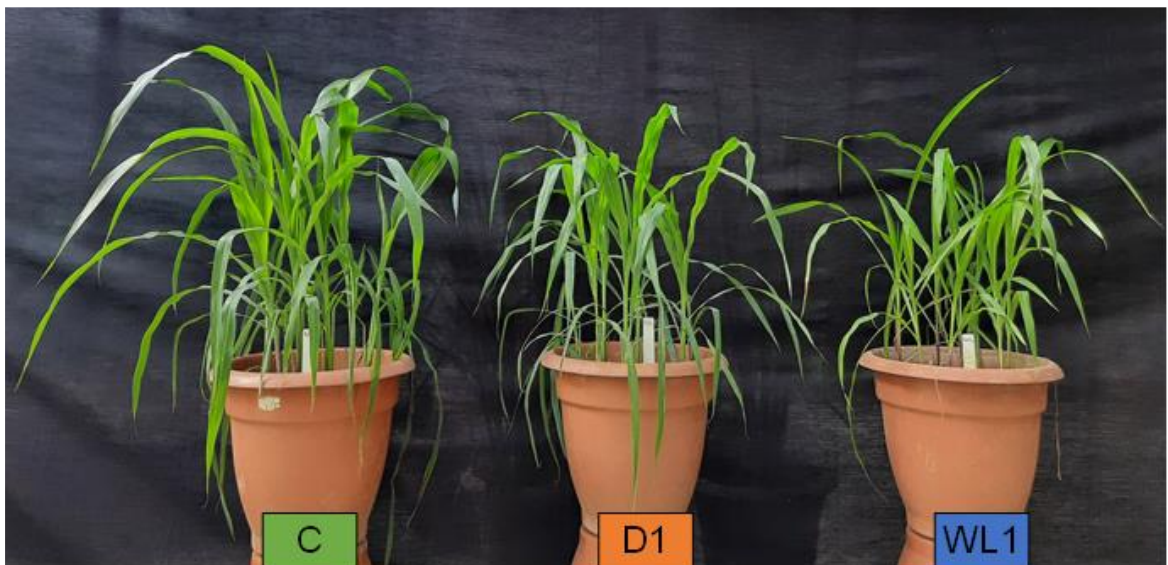


Plate 2. Napier grass after 7 days' recovery



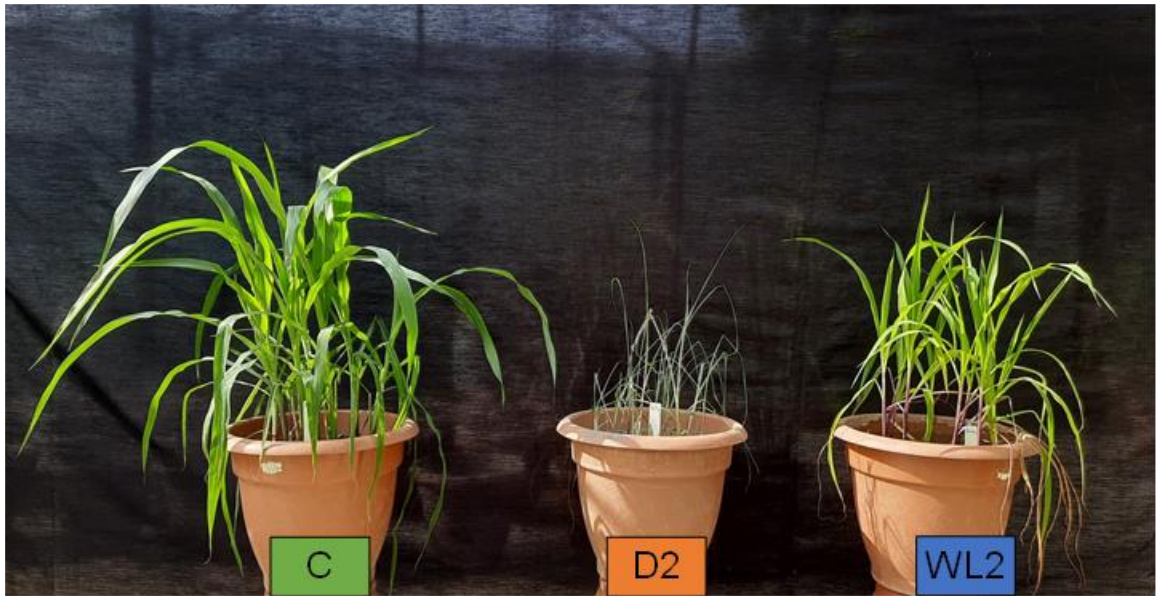


Plate 3. Napier grass after 14 days' stress treatments

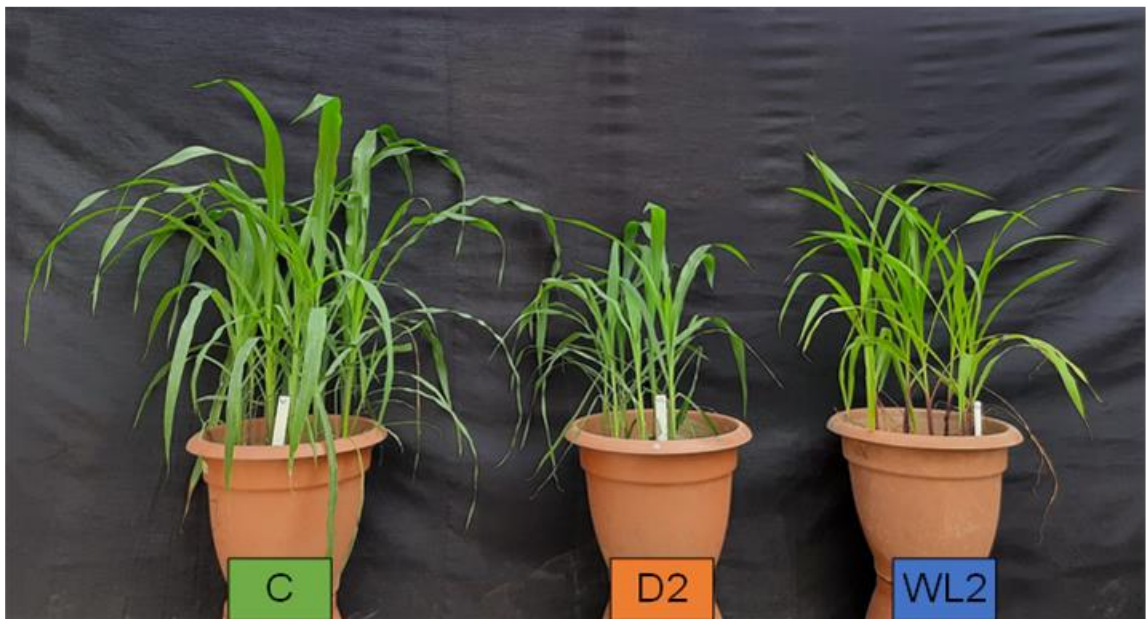


Plate 4. Napier grass after 14 days' recovery

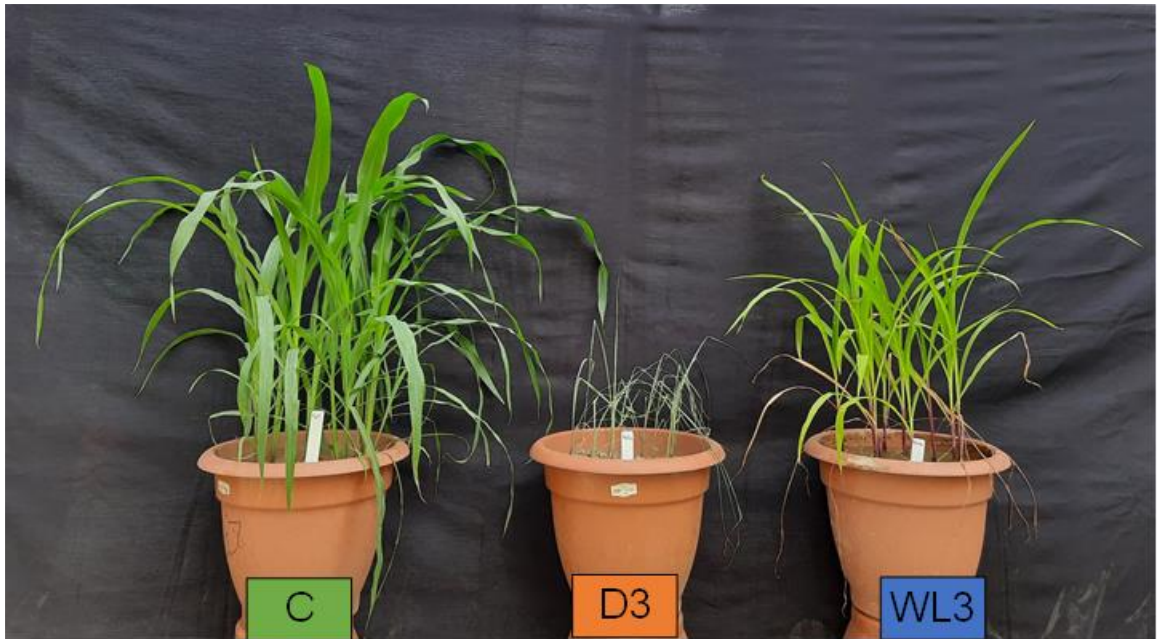


Plate 5. Napier grass after 21 days' stress treatments



Plate 6. Napier grass after 21 days' recovery



APPENDIX II. Phenotypic pictures of napier grass at 50 days after sowing

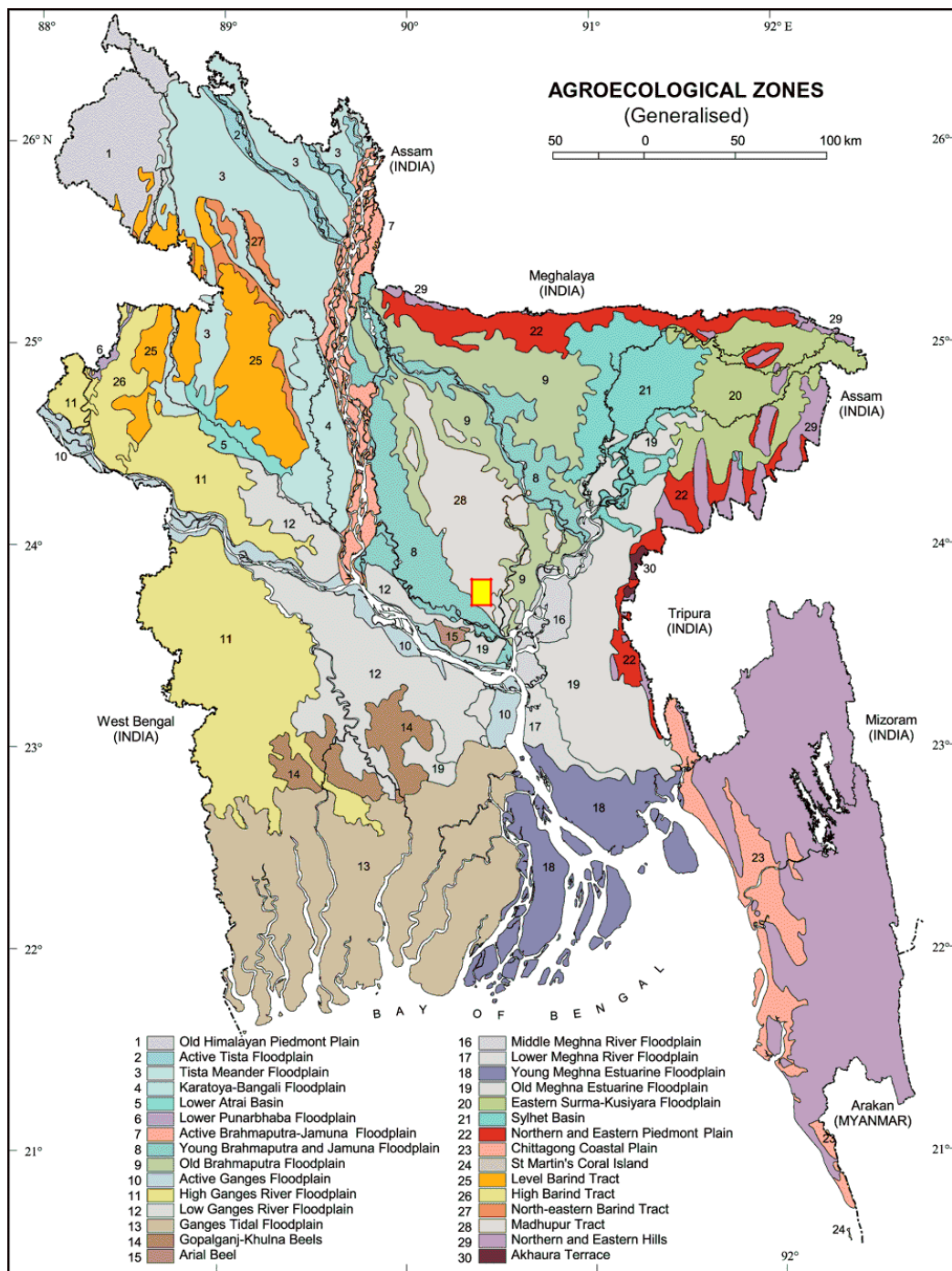


Plate 7. Napier grass at 50 days after sowing



Plate 8. Napier grass at 50 days after sowing

Appendix III. Map showing the location of the experiment



Appendix IV. Monthly average air temperature, rainfall, and relative humidity of the experiment site during the period from April 2021 to July 2021

Months	Air temperature (°C)		Relative humidity (%)	Total rainfall (mm)
	Maximum	Minimum		
April, 2021	33.7	23.6	71	156.3
May, 2021	32.9	24.5	76	339.0
June, 2021	32.1	26.1	82	340.4
July, 2021	31.4	26.2	83	373.3

Appendix V. Mean square values and degree of freedom (DF) of plant height, shoot and root fresh weight, shoot dry weight, and root dry weight at different stress duration (drought and waterlogging)

Source of variance	DF	Mean square values				
		Plant height	Shoot fresh weight	Root fresh weight	Shoot dry weight	Root dry weight
Treatments	8	946.860	61.766	0.756	1.941	0.041
Error	18	7.816	0.283	0.015	0.012	0.001

Appendix VI. Mean square values and degree of freedom (DF) of plant height, shoot and root fresh weight, shoot dry weight, and root dry weight at recovery of napier grass

Source of variance	DF	Mean square values				
		Plant height	Shoot fresh weight	Root fresh weight	Shoot dry weight	Root dry weight
Treatments	6	941.104	249.679	1.032	4.794	0.066
Error	14	6.976	2.034	0.011	0.043	0.002

Appendix VII. Mean square values and degree of freedom (DF) of root length, root-shoot ratio, and root branch at different stress duration (drought and waterlogging)

Source of variance	DF	Mean square values		
		Root length	Root-shoot ratio	Root branch
Treatments	8	102.175	0.051	84.849
Error	18	0.835	0.001	0.882

Appendix VIII. Mean square values and degree of freedom (DF) of root length, root-shoot ratio, and root branch at recovery of napier grass

Source of variance	DF	Mean square values		
		Root length	Root-shoot ratio	Root branch
Treatments	6	207.368	0.027	51.949
Error	14	1.778	0.001	0.468

Appendix IX. Mean square values and degree of freedom (DF) of SPAD value, relative water content, and proline at different stress duration (drought and waterlogging) of napier grass

Source of variance	DF	Mean square values		
		SPAD value	Relative water content	Proline
Treatments	8	416.108	380.501	86.639
Error	18	5.534	6.468	0.475

Appendix X. Mean square values and degree of freedom (DF) of SPAD value, relative water content, and proline at recovery of napier grass

Source of variance	DF	Mean square values		
		SPAD value	Relative water content	Proline
Treatments	6	57.003	95.710	1.569
Error	14	2.119	5.930	0.968

Appendix XI. Mean square values and degree of freedom (DF) of malondialdehyde content and hydrogen peroxide at different stress duration (drought and waterlogging) of napier grass

Source of variance	DF	Mean square values	
		Malondialdehyde	Hydrogen peroxide
Treatments	8	433.355	174.007
Error	18	32.119	5.174

Appendix XII. Mean square values and degree of freedom (DF) of malondialdehyde content and hydrogen peroxide at recovery of napier grass

Source of variance	DF	Mean square values	
		Malondialdehyde	Hydrogen peroxide
Treatments	6	664.991	10.655
Error	14	69.119	2.221

Appendix XIII. Mean square values and degree of freedom (DF) of fodder yield, acid detergent fiber, neutral detergent fibre, and dry matter content at recovery of napier grass

Source of variance	DF	Mean square values			
		Fodder yield	Acid detergent fiber	Neutral detergent fiber	Dry matter content
Treatments	6	294.556	17.778	161.169	5.583
Error	14	0.990	1.540	11.361	1.803

Appendix XIV. Mean square values and degree of freedom (DF) of crude protein, crude fibre and ash content at recovery of napier grass

Source of variance	DF	Mean square values		
		Crude protein	Crude fibre	Ash content
Treatments	6	3.875	14.011	20.074
Error	14	0.658	4.347	3.582

Appendix XV. Mean square values and degree of freedom (DF) of organic matter content and hemicellulose at recovery of napier grass

Source of variance	DF	Mean square values	
		Organic matter content	Hemicellulose
Treatments	6	3.875	74.914
Error	14	0.658	8.392