

**EFFECT OF WATERLOGGING AND SEEDLING AGE ON
MORPHOLOGICAL AND PHOTOSYNTHETIC
CHARACTERS OF EGGPLANT**

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BY

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This is to certify that the thesis entitled, “**Effect of waterlogging and seedling age on morphological and photosynthetic characters of eggplant**” submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE** in **HORTICULTURE**, embodies the result of a piece of *bona fide* research work carried out by **Md. Arafat Ali**, Reg. No. **05-01806** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or sources of information, as has been availed of during the course of this investigation has duly acknowledged.

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*DEDICATED
TO
MY BELOVED MOTHER AND
FATHER*

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ABSTRACT

An experiment was conducted at Bangladesh Agricultural Research Institute, Gazipur during the period of May to June 2011. Effect of waterlogging and seedling age on morphological and photosynthetic characters of eggplant was studied. Waterlogging was imposed for 144 hours on 30 and 40 days old seedlings with the water level 8 cm above the soil surface. Morphological and photosynthetic characters were affected drastically after 48 hours and 72 hours of waterlogging in 30 and 40 days old seedlings respectively and photosynthetic characters, chlorophyll content, photosynthesis rate and transpiration rate were decreased with reduction of stomatal conductance of CO₂. Conversely sub-stomatal CO₂ concentration increased for restricted assimilation of CO₂ in the leaves in both cases. The quantum efficiency of photosystem II (quantum yield) (F_v/F_m) of waterlogged seedlings observed lower

compared to that of seedlings grown under normal soil moisture condition as control and the ratio were rapidly decreased after 48 hours of waterlogging irrespective of seedlings age. Waterlogging effects became stronger with increasing the durations of waterlogging and differ with the age of the plant.

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LIST OF ACRONYMS

ABBREVIATIONS	ELABORATIONS
AEZ	Agro-Ecological Zone
ANOVA	Analysis of Variance
BARI	Bangladesh Agricultural Research Institute
BBS	Bangladesh Bureau of Statistics
HRC	Horticulture Research Centre
F_v/F_0	Initial fluorescence
kDa	Kilodalton (Atomic mass unit)
	Leaf water potential

F_v/F_m	Maximum quantum efficiency of the photosystem II
mV	Millivolts
Pn	Net photosynthesis
q_P	Photochemical quenching
PSII	Photosystem II
g_s	Stomatal conductance
E	transpiration

CHAPTER I

INTRODUCTION

Eggplant (*Solanum melongena* L) belongs to the family Solanaceae. It is also known as brinjal, aubergine and begun in Bengali. It is a major vegetable crop throughout the tropic and subtropics (Bose and Som, 1986) and thought to be originated in Indian sub-continent (Zeven and Zhukovesky, 1975). Eggplant has much importance in the warm areas of Far East, being grown extensively in Bangladesh, India, Pakistan,

Nepal, China, Japan and Philippines and also popular in France, Italy, USA, the Mediterranean and Balkan areas (Bose and Som, 1986).

Eggplant is a year round vegetable in Bangladesh (Rashid, 1993) and is grown in homestead and as a field crop in both winter and rainy seasons though bulk of its production is obtained during winter season. The vegetable production in summer is scanty and eggplant plays an important role to meet up the shortage of vegetable in this lean period. It is the second most important vegetable crop next to potato in Bangladesh in respect of acreage and production. The total area of eggplant cultivation in Bangladesh is 60,100 hectare where 22,500 ha in Kharif and 37500 ha in Rabi season with a total annual production of 338 thousand metric tons (BBS, 2010). It contains 92.7% moisture, 1.4g protein, 0.3 fat, 0.3g minerals, 4g carbohydrates, 18mg calcium, 0.9mg iron, 44 mg Sulphur, 16 mg magnesium, 18 mg oxalic acid, 47mg phosphorus, 124 I.U. vitamin A, 0.04 mg Thiamine, 0.11 mg Riboflavin, 0.09 mg Nicotinic acid, 12.0 mg Vitamin C, vitamin K, antioxidants etc. (Krishi diary, 2012)

Waterlogging is an environmental stress that severely limits crop growth and productivity. Waterlogging is common in rain-fed ecosystems, especially in soils with poor drainage. It can seriously reduce yield (Dennis *et al.*, 2000) up to 10% (Bange *et al.*, 2004) and 40% in severe cases (Hodgson and Chan, 1982). As a consequence of disturbed physiological functioning, vegetative and reproductive growth of plants is negatively affected by waterlogging (Kozlowski, 1984; Gibbs and Greenway, 2003).

Waterlogging is a condition caused when air in the soil is replaced with water. Gas exchange between the soil and the air is decreased, because gas diffusion in water is 10,000 times less than in air (Marschner, 1995). Under waterlogged conditions, most plants suffer from an oxygen deficiency which can also reduce the capacity of the

roots to take up and supply water and nutrients from the soil to the rest of the plant (De Simone *et al.*, 2002). A variety of plants showed reduced stomatal conductance (Smith and Moss, 1998) in response to root-zone saturation and stomatal closure under waterlogged conditions. This reduction was often accompanied by reduced CO₂ assimilation and water transpiration. Due to waterlogging photosynthesis rapidly decreased, with a subsequent decrease in carbohydrate production and consequently the growth rate was reduced (Musgrave and Ding, 1998). Inhibited nutrient uptake led to a nutrient deficiency in the shoots that reduced the chlorophyll content of the leaves, causing early leaf senescence, abscission and chlorosis (Huang *et al.*, 1994) and reduced leaf area (Bradford, 1982). If the plants were grown under these conditions for a long period, the plants switched from an aerobic-respiration pathway to an anaerobic pathway (Kozlowski, 1984). The anaerobic respiration induced a fermentation pathway and ATP synthesis decreased. Under such conditions, plants reduced their photosynthesis and their growth rate (Bray *et al.*, 2000).

Most physiological researches on waterlogging were focused on Timber and fruit crops but only some were reported on vegetable crops. During summer production, seedlings of eggplant face waterlogging due to excessive rain fall and yearly flooding. For this reason it is very important to study the physiology of eggplant seedling under waterlogging condition.

Common stress measurement techniques are based on destructive state. Those are time consuming and difficult to collect data from mass population. Leaf-gas exchange parameters are used to study the photosynthetic capacity of plants under stress condition. Fluorescence measurements can provide an indication of the photosynthetic performance as a means of measuring the stress tolerance of a plant. Leaf gas exchange and chlorophyll fluorescence measurement are relatively new technology that in recent years is become rapid performing and widely used non destructive and

non invasive powerful tool in stress measurement and plant screening for environmental stress.

Considering the above facts the present research work was carried out to measure waterlogging stress on eggplant seedlings by non destructive method and to achieve the following objectives:

- i) To study the morphological and photosynthetic characters of eggplant seedlings under waterlogged condition.
- ii) To determine the tolerant level of eggplant seedlings at different age and durations of waterlogging.

CHAPTER II

REVIEW OF LITERATURE

Waterlogging is a world-wide phenomenon that affects both the distribution of plants in their natural ecosystems as well as crop yield in agricultural regions. Waterlogging causes an immediate reduction in the exchange of gases between the plant and its environment. At the physiological level, waterlogging could greatly affect plant water relations. Most cases researches are going on to maximize the yield. Research works related to waterlogging on eggplant are limited. However, some of the important and informative works and research findings related to this crop on waterlogging so far been done at home and abroad have been reviewed in this chapter.

Aloni *et al.* (2006) found that flooding of the root system of tomato plants (*Lycopersicon esculentum*) caused cessation of leaf elongation, leaf epinasty, formation of adventitious roots, and increase in diffusive resistance associated with the wilting of leaves at the first stage of the stress. Upon development of adventitious roots, the wilted leaves regained their turgor and the diffusive resistance slowly decreased at a rate slower than that at which water potential increased. In the course of flooding, proline accumulated but after 11 days dropped back to the control level. The extent of proline accumulation in various tomato cultivars was positively correlated with the extent to which their leaf water potential dropped, but was not correlated with the changes in their diffusive resistance. Cultivars which accumulated the highest proline levels were those which showed the most severe injury, with only one cultivar as an exception. However, only in the cultivars producing high levels of

proline was the return of leaf turgor followed by resumption of leaf elongation. In cv. 'Hosen', which was severely injured by the stress, but accumulated a low level of proline, leaf elongation was not resumed. The results suggest that proline accumulation is an indicator of the cultivar's sensitivity to dehydration associated with the flooding stress, and confirm the notion that proline may play a role in the post-stress recovery process.

Anella and Whitlow (2000) conducts a study to investigate Photosynthetic Response to Flooding of *Acer rubrum* Seedlings from Wet and Dry Sites. Flooding reduced net photosynthesis, growth and chlorophyll levels in seedlings from both sites, but survival of both wet and dry site seedlings was near 100%. After 1 month of flooding net photosynthesis of wet and dry site seedlings were similar, but after 3 month, flooded wet site seedlings had higher photosynthetic rates than did dry site seedlings. Control wet site seedlings were significantly larger than dry site seedlings and had significantly higher photosynthetic rates and chlorophyll levels. These differences suggest either genetic variation between seedlings from the two habitats in response to the growing conditions or the influence of seed size differences and confound the spring flooding study results. Flooded wet site seedlings had higher, final, net photosynthetic rates than did dry site seedlings, but the response to flooding was greater for wet site seedlings than it was for dry site seedlings. In a second study (summer flooding study), 1-year old seedlings in full leaf were flooded for 22 days and then drained to determine if recovery from flooding stress differed for wet and dry site seedlings. Again, flooding decreased net photosynthesis for seedlings from both habitats but, when the trees were drained, net photosynthesis for wet site seedlings recovered more quickly and to a higher level than it did for dry site

seedlings. Flooding also caused a drop in chlorophyll levels for seedlings from both habitats, but chlorophyll levels of seedlings from neither habitat recovered when the seedlings were drained. There were no significant differences between wet site and dry site control seedlings for net photosynthesis; therefore, the quicker and larger recovery of photosynthetic potential in wet site seedlings in the summer flooding study suggests that ecotypic differentiation has occurred and those genetic differences, in part, account for red maple's occurrence on contrasting edaphic sites.

In a field experiment of Bingru *et al.* (1993), in order to investigate the responses of plant genotypes to waterlogging and nutrient supply, physiological, morphological, and anatomical features were determined for two genotypes of winter wheat (*Triticum aestivum*), 'Bayles' and "Savannah". Waterlogging reduced leaf water potential, stomatal conductance, photosynthesis, chlorophyll content, shoot nitrogen content, shoot and root growth for both Bayles and Savannah; however, there were more adverse effects on Bayles than on Savannah. Doubling the concentration of nutrients supplied to the water logged rooting medium reduced the rate of decline in photosynthesis, chlorophyll content, number of nodal roots, and improved shoot nitrogen status and growth, especially for Bayles. Waterlogging reduced the diameter of metaxylem and protoxylem vessels of the nodal roots for both genotypes. Waterlogging enhanced the formation of aerenchyma in the cortical tissue of both seminal and nodal roots, but to a greater extent for Savannah. Additionally, aerenchyma formed continuously along the seminal roots in Savannah. The results indicated that Bayles was more sensitive to waterlogging than Savannah and that increases in nutrient supply to waterlogged plants could alleviate some of the adverse effects of waterlogging.

The effects of waterlogging the soil on leaf water potential, leaf epidermal conductance, transpiration, root conductance to water flow, and petiole epinasty have been examined in the tomato (*Lycopersicon esculentum* Mill.) by Bradford and Hsiao (1982). Stomatal conductance and transpiration are reduced by 30% to 40% after approximately 24 hours of soil flooding. This is not due to a transient water deficit, as leaf water potential is unchanged, even though root conductance is decreased by the stress. The stomatal response apparently prevents any reduction in leaf water potential. Experiments with varied time of flooding, root excision, and stem girdling provide indirect evidence for an influence of roots in maintaining stomatal opening potential. This root-effect cannot be entirely accounted for by alterations in source-sink relationships. Although 1-aminocyclopropane-1-carboxylic acid, the immediate precursor of ethylene, is transported from the roots to the shoots of waterlogged tomato plants, it has no direct effect on stomatal conductance. Ethylene-induced petiole epinasty develops coincident with partial stomatal closure in waterlogged plants. Leaf epinasty may have beneficial effects on plant water balance by reducing light interception.

Bradford (1983) conducted an experiment to investigate the effects of soil flooding on leaf gas exchange of tomato (*Lycopersicon esculentum* Mill. cv Rheinlands Ruhm) plants. Carbon dioxide and water vapor exchange of tomato leaves were measured before and after 24 h of soil flooding to characterize both stomatal and nonstomatal responses to the stress. Leaf epidermal conductance to water vapor decreased by 47% after flooding, accompanied by an increase in the sensitivity of stomata to changes in CO₂ concentration. Assimilation rates under ambient conditions fell by 27%, and the inhibition could not be overcome by elevated CO₂ partial pressures. Stomatal

conductance limited the assimilation rate to approximately the same degree both before and after flooding. The reduction in photosynthetic capacity was not due to a decrease in apparent quantum yield or to an increase in photorespiration.

In a study, Devy (2002) observed that at seed and seedling phases, the electrolyte leakage and the activity of alcohol dehydrogenase (ADH) enzyme were significantly affected by anaerobiosis treatments. The treatment decreased 60-80% of the leakage, but increased enzyme activity by 120%. The ADH activity of petridish-grown seedlings was higher than the nursery-grown. The banding pattern of total soluble proteins in dry and treated seeds had more intense bands than treated seeds, especially at MW67 and 14.4 kDa. After 1 wk of waterlogging, the 36 and 48-h anaerobiosis-treated seeds exhibited a modification of the protein profile. In terms of seed germination, the treated seeds under waterlogged conditions decreased by about 95% after 1 wk. At the growth phase, root ADH activity increased by 10, 30, and 60% during 1, 5, and 10 days of waterlogging, respectively. At the same time, plant relative water content (RWC) was reduced significantly by 8.4% over the control at the end of the treatment. Some changes occurred in the root protein profile under waterlogged conditions. Low molecular weight proteins below 14 kDa were attenuated, while proteins with sizes ranging from 20 to 45 kDa were more pronounced in treated samples than in the normal. Waterlogging induced ethylene production but decreased plant respiration rate and plant dry weight. At the productive phase, both total production and number of fruits/plant were not affected by all treatments. Total production and fruit number/plant were 404.6 and 351.1 g and 6.6 and 6.1 in plants from normal and waterlogged conditions, respectively.

Dias-Filho (2002) compared some physiological and morphological responses of five *Brachiaria brizantha* accessions (BRA000591 cultivar Marandu, BRA003441, BRA002844, BRA004308 and BRA004391) for plants grown in pots under flooding and well-drained conditions for 14 days. Flooding caused a significant reduction in leaf dry mass production in all accessions, but, for root biomass, no differences between treatments could be detected in BRA003441 and BRA004391. No adventitious root production was observed in flooded BRA003441; all other accessions produced adventitious roots when flooded. Relative growth rate was reduced by flooding only in BRA000591 and BRA004308. Leaf elongation rate was reduced by flooding in all accessions, however, more severely in BRA003441. Net photosynthesis was reduced by flooding in all accessions, but with less intensity in BRA004391. For all accessions, there was a close relationship between net photosynthesis and stomatal conductance under flooding. The five accessions tested differed in tolerance to flooding. BRA004391 was the most tolerant. Accession BRA003441 was the most sensitive, followed by BRA000591 cultivar Marandu. Accessions BRA002844 and BRA004308 were classified as intermediate in flooding tolerance.

A greenhouse experiment was conducted by Ezin *et al.* (2010) to investigate the effect of flooding on two tomato cultivars and two wild related species. Fourty days old tomato plants were subjected to a continuous flooding stress of different durations: 0, 2, 4 and 8 days. Plant pots were placed inside larger plastic pots; they were irrigated with excessive quantity of tap water at 25°C so that the level of water above the surface of soil was 15 cm throughout the flooding period. At the bottom of each plastic pot a drilled hole allowed complete drainage of the pot after flooding.

Parameters studied include plant height, number of leaves, leaf length, chlorophyll fluorescence, chlorophyll content, wilting, leaf senescence, adventitious root formation, number of flowers and fruits from cluster 2 to 6, average weight per fruit, fruit length and width, total fruit weight from cluster 2 to 6, total yield per plant. LA1579 genotype was more subjected to the deleterious effect of flooding on almost all the parameters studied. Therefore LA1579 genotype is flooding sensitive. Genotypes CLN2498E, and CA4 showed high tolerance to flooding while LA1421 genotype was tolerant to some extent. This experiment provides information that could help in the identification of physiological and agronomical parameters associated with flood-tolerance in vegetables.

Ewa *et al.* (2008) conducted an experiment to investigate the effects of root flooding and stage of development on the growth and photosynthesis of field bean. Field bean plants were subjected to flooding stress for 7 days, during two stages of development: at the vegetative phase (4-week-old seedlings) and at the generative phase (8-week-old plants). The height of plants, total area of leaves, the number of undamaged leaves, dry plant matter, chlorophyll content, ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBPCO) activity, the maximum quantum yield of PSII photochemistry (Fv/Fm ratio), the photosynthesis rate (P_N) and stomatal conductance (g_s) were determined. A strong reduction in stem elongation and leaf area as well as in dry matter production was observed as a result of flooding. The responses from vegetative plants were greater than in generative plants. Waterlogging decreased chlorophyll a and b in leaves, notably at the vegetative stage, and persisted after cessation of flooding. After flooding, photosynthesis was strongly reduced and positively correlated with decreased stomatal conductance. Damage to the

photosynthetic apparatus resulted in a lower Fv/Fm especially in young seedlings. In vegetative plants Fv/Fm quickly returned to the control levels after the soil was drained. The results show that an excess of water in the soil limits growth and injures the photosynthetic apparatus in field beans, but that the extent of the injury is strongly age dependent.

In an experiment of Fernandez *et al.* (1997), photosynthesis and ¹⁴C-photosynthate distribution, root growth dynamics, shoot extension, leaf number and TCAI were monitored for 'Jonnee' apple on M.9 EMLA, M.26 EMLA and Mark rootstocks that were subjected to 2, 4, 8, 16 and 32 days of flooding. Stress was imposed for all flooding durations on the same date. Recovery was monitored until 45 days after the 32 day flooding stress. Photosynthetic response to flooding varies by rootstock and flooding duration. Distribution of ¹⁴C-photosynthates between the root and shoot system was affected for M.26 EMLA and Mark rootstocks. No differences were seen in root dynamics until 32 days of continuous flooding. From day 32 until termination of the experiment, fewer roots were produced for 32 day flooded M.9 EMLA and M.26 EMLA. There were fewer roots produced for 32 day flooded Mark on day 32 but root production was similar to controls during recovery. Shoot growth and TCAI were reduced substantially for all flooded rootstocks. The number of leaves decreased as flooding duration increased.

In a study, Hossain and Uddin (2011) stated that, waterlogging is a widespread limiting factor for wheat production throughout the world specially irrigated and high rainfall environments. The important biological consequence of waterlogging is the deficiency (hypoxia) or complete absence (anoxia) of oxygen in soil environment

which restricts the growth, development and finally yield in wheat. The tolerant genotypes of wheat can adapt to transient waterlogging by developing mechanisms related to morphology and metabolism to cope with the stress. The morphological mechanisms include the development of adventitious roots with well formed aerenchyma and sometimes with a barrier for ROL. Aerenchyma is a continuous gas filled channel, which provides a low resistance internal pathway for the movement of O₂ from the aerobic shoots to anaerobic roots to respire aerobically under hypoxia or anoxia. However, lack of oxygen induces the anaerobic roots to shift the energy metabolism from aerobic to anaerobic mode. Greater activities of glycolytic and fermentative enzymes, increased availability of soluble sugars, and involvement of antioxidant defense mechanism against post-stress oxidative damages are the main metabolic mechanisms for waterlogging tolerance in wheat.

Hongjun *et al.* (2010) examined the growth, biomass, photosynthesis, and chlorophyll fluorescence of *Typha domingensis* (southern cattail) in response to a six-week period of three flooding depths: 40, 91, and 137 cm above the soil surface and a subsequent four-week period of recovery at a 40-cm flooding depth. The 91- and 137-cm flooding significantly decreased the following: number of new shoots, number of live leaves, leaf, belowground and total biomass, and belowground biomass/leaf ratio. Plants flooded to a 91- or 137-cm depth had significantly lower chlorophyll concentration, photosynthesis, and fluorescence (Fv/Fm) as well as higher dark respiration and light compensation points than those exposed to the 40-cm flooding. Following the recovery period, most adverse impacts caused by the 91-cm flooding depth were reversed in terms of biomass, photosynthesis, and chlorophyll fluorescence, while the effect of the 137-cm flooding stresses exhibited by *T. domingensis* were not reversed.

Flooding to a 137-cm depth is detrimental to *T. domingensis* and needs to be avoided. Our results indicate that depth-duration thresholds for management of *T. domingensis* dominated marshes are necessary to minimize deepwater impacts to vegetation communities.

An experiment was conducted by Kuan-Hung *et al.* (2004) to study the response of antioxidative enzymes, and antioxidants of tomato and eggplant roots to waterlogged conditions. The roots of four entries, eggplants EG117 and EG203, and tomatoes TNVEG 6 and L4422 (*Lycopersicon pimpinellifolium* Mill), were subjected to seven flooding treatments. The activity of APX in roots significantly increased during the period of continuous waterlogging. Slight increases in total ASA, reduced ASA, GSH, and total glutathione contents in the roots were also observed throughout the entire waterlogging period. However, the activities of CAT, SOD and GR, and the contents of ASA, GSSG and α -tocopherol in the roots were unaffected by waterlogging. Entries responded differently to oxidative injury according to their various antioxidative systems. The results indicate that total ASA could be involved in flooding damage to tomato roots. Overall, following the waterlogging treatments, APX activity in the eggplants was generally higher than in the tomatoes. Their work suggested that the brownish roots of tomatoes induced by flooding may be the consequence of H_2O_2 scavenging possibly controlled by APX activity. The H_2O_2 scavenging system as represented by APX was clearly limiting or less efficient in the tomatoes, leading to an accumulation of H_2O_2 . The ability to maintain a balance between the formation and detoxification of activated oxygen species appeared likely to increase the survival potential and the tolerance of the roots against varying

oxidative stress. On the basis of their observations, they concluded that increased APX activity provides plant roots with increased waterlogged stress tolerance.

Kuo and Chen (1980) conducted an experiment to investigate physiological responses of tomato cultivars to flooding. One-month-old seedlings of 20 entries of tomato (*Lycopersicon esculentum*) were subjected to flooding for 3 days, or drenched with ethanol or (2-chloroethyl) phosphonic acid (ethephon) solution. Flooding reduced stem growth and leaf chlorophyll content of the lower leaves and promoted epinastic curvature of leaf petiole and adventitious roots.

Li *et al.* (2004) conduct an experiment to study the effects of soil moisture regimes on photosynthesis and growth in cattail (*Typha latifolia*). The study quantified the photosynthetic and growth responses of cattail (*Typha latifolia*), an important species of freshwater wetlands, to a wide range of soil moisture regimes. Results indicated that dark fluorescence yield was increased in response to periodic drought, while it showed decreases under continuous flooding. Net photosynthesis and stomatal conductance were enhanced by continuous flooding and periodic flooding. Recovery of net photosynthesis was noted, along with enhanced height growth, in both continuously and periodically flooded plants. Therefore, the combined photosynthetic performance and growth responses of cattail are likely to contribute to the ability of this species to thrive in flooded condition.

Lopez and Rosario (1983) investigated the effects of morphological, physiological, biochemical and anatomical characteristics of three varieties of tomato at three growth stages in relation to growth and yield. Waterlogging significantly reduced plant

height, leaf area, leaf chlorophyll content, respiration rate, percent survival, fruit set, total yield, seed set and dry weight of the whole plants and root free proline content. It increased epinastic curvature, stomatal resistance, leaf free proline content and the cross-sectional area of the intercellular spaces and cortical cells.

A study was undertaken by Musgrave (1994) to estimate the yield loss for wheat that is attributable to soil waterlogging and to compare physiological performance under waterlogging-stressed conditions by cultivars grown in Louisiana. In a 3-yr pot study conducted in a greenhouse, waterlogging stress was imposed by raising the water level to the soil surface. This treatment reduced the soil redox potential in the pots from an average of 409 to 149 mV, indicating an absence of free oxygen in the root zone. Compared with a well-drained control treatment, grain weight was decreased 37 to 45% by waterlogging in the eight cultivars tested. In a field experiment with 'Coker 9877', grain weight was depressed 51% in poorly drained plots compared with well-drained plots. Yield depression was due to reduced kernel number and kernel weight rather than to an effect on stand establishment. In the greenhouse experiments, flag-leaf photosynthesis correlated well with grain weight in the cultivars tested. Waterlogging caused only a small suppression of flag-leaf photosynthesis and leaf conductance, and there were no significant interactions between treatment and cultivar. These commercially available cultivars showed an equally poor tolerance of waterlogging stress.

Effects of soil flooding on photosynthesis and growth of *Genipa americana* L. seedlings, a neotropical fruit-tree species used in gallery forest restoration programs, were studied by Marcelo *et al.* (2003) under glasshouse conditions. Using leaf gas

exchange and chlorophyll fluorescence measurements, they tested the hypothesis that stomatal limitation of photosynthesis is the main factor that reduces carbon uptake and growth rates of *G. americana* seedlings. Throughout a 63-day flooding period, the survival rates were 100%. The maximum values of the net photosynthetic rate (A) and stomatal conductance to water vapor (g_s) of control seedlings were $9.86 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ and $0.525 \text{ mol H}_2\text{O m}^{-2} \text{ s}^{-1}$, respectively. The earliest effects of flooding were significant decreases in g_s and A , development of hypertrophied lenticels and decrease in the dry weight of roots. A strong effect of the leaf-to-air vapor pressure deficit (LAVPD) on g_s and A were observed that was enhanced under flooded conditions. Between 14 and 63 days after flooding, significant reductions in g_s (31.7% of control) and A (52.9% of control) were observed followed by significant increments in non-photochemical quenching (q_N) (187.5% of control). During the same period, there were no differences among treatments for the ratio between variable to initial fluorescence (F_v/F_0), the maximum quantum efficiency of the photosystem II (F_v/F_m) and photochemical quenching (q_P), indicating that there was no damage to the photosynthetic apparatus. Based on the results, they concluded that decreases in stomatal opening and stomatal limitation of photosynthesis, followed by decrease in individual leaf area are the main causes of reductions in carbon uptake and whole plant biomass of flooded seedlings.

Moldau (1973) examined the effect of flooding on bean plants (*Phaseolus vulgaris* L.) and concluded that an insufficient supply of metabolites from the roots in addition to stomatal closure were responsible for reduced photosynthesis. Root excision, which may be similar to the effect of waterlogging, reduces photosynthesis but is not related

to stomatal closure and further, suggests a broader role of the roots in maintaining photosynthesis.

Phung and Knipling *et al.* (1976) conducted an experiment to investigate photosynthesis and transpiration of citrus seedlings under flooded conditions. In their experiment photosynthesis and transpiration rates of seedlings of 4 citrus rootstocks under flooded conditions were measured over a 10-day period. For all rootstocks photosynthesis and transpiration decreased, but photosynthesis decreased relatively less than transpiration. Stomatal closure is inferred to account in part for the reductions observed. Flooding did not increase ethanol concentration in either tops or roots, suggesting that ethanol is not an end-product of anaerobic respiration in citrus seedlings. Only in the neutral soil was rough lemon (*Citrus limon* L. Burm. f.) found to be more tolerant to short-term flooding than 'Cleopatra' mandarin (*Citrus reticulata* Blanco) and trifoliolate orange (*Poncirus trifoliata* L. Raf.). Tolerance to flood injury was greater at a soil pH of 7 than 4.5.

Robert *et al.* (1983) in an examination of the effect of flooding on sunflowers, found a reduction in photosynthesis after 4 days without a major reduction in leaf conductance further suggesting factors other than stomatal conductance were influencing photosynthesis. It seemed possible that loss of chloroplast integrity was responsible.

A study was carried out by Suh *et al.* (1987) to investigate the influence of flooding on the metabolic activity, growth and yield of hot pepper in the field condition and artificial water bath for two years from 1985. Survival ratio, in the field condition, as

affected by waterlogging (10-20 cm from the soil surface) was not significantly decreased within 24 hours, but the fruit yield was significantly decreased at the same waterlogging treatment. 48 hours of waterlogging showed about 90 % of withering 1621 to death in hot pepper plant and all the plant could not survive more than 4 days of waterlogging treatment. In the pot trial, higher survival ratio was observed at the same period of waterlogging treatment with the field condition considering not only due to the experimental difference of drainage condition after waterlogging treatment but also the difference of fruit-set burden from the growth stage between pot and field. Leaf chlorosis and falling was linearly increased with the elongation and height of waterlogging treatment showing more than 90 % of withering to death in 7 days of waterlogging.

A study was conducted by Singh *et al.* (1991) to determine the effect of various flooding durations on the growth, water relations, and photosynthesis of the Snap bean (*Phaseolus vulgaris* L.). Greenhouse-grown plants of cv. Blue Lake 274 were flooded for 0 (control), 1, 3, 5, or 7 days. Leaf water potential (), stomatal conductance (g_s), transpiration (E), and net photosynthesis (Pn) were measured at the completion of the flooding period and after recovery for 7 days. Root, stem, and leaf dry weights were recorded after plants were allowed to recover from the flooding stress for 7 days. The values for , g_s , E, and Pn decreased quadratically with the increase in the duration of flooding. The Pn of plants flooded for 1 day was 17% lower than that of the control and it reached near zero in plants flooded for 7 days. The decrease in Pn after 1 day of flooding was not associated with or g_s ; however, for longer duration of flooding, Pn decline coincided with the decline in g_s . A week after the cessation of flooding, the level of recovery in , E, and Pn was linear and

that in g_s quadratic to the duration of prior stress experienced by the plant. However, after recovering for 7 days, none of the flooded plants regained gas exchange activities at par with the control. The relationship of stem dry weight to duration of flooding was linear, while a quadratic model provided the best fit for the regression of root and leaf dry weight on the number of days of flooding. Overall, even 1 day of flooding reduces photosynthesis in snap bean and causes a decrease in dry weight of the plant. The extent of decrease in both cases increase with the duration of flooding.

In a study of Smethurst and Shabala (2003), a comparative analysis of waterlogging effects on leaf photosynthesis, pigment composition, PSII photochemistry, and plant growth characteristics was undertaken using four different lucerne cultivars (Aurora, Hunter River, L153 and Sequel HR). Two-month-old plants, grown in half-strength Hoagland nutrient solution, were waterlogged for 16 days, and plant physiological characteristics were monitored at regular intervals (every few days). All cultivars had significantly reduced fresh and dry weight for both shoots and roots after 16 days of waterlogging. Root biomass showed a greater percentage of reduction than did shoot biomass. As waterlogging stress developed, chlorophyll content, CO_2 assimilation rate, transpiration rate, stomatal conductance and maximal quantum efficiency of PSII (F_v/F_m) decreased significantly. Chlorophyll *a* and *b* content gradually decreased over the time of the experiment in the stressed cultivars, and leaf chlorosis became increasingly evident. Although most of the parameters showed significant changes as waterlogging progressed, limitations render some of them inapplicable for screening. A significant difference between control and waterlogged plants became evident as early as day 7.

The effect of waterlogging on growth, development and yield of cotton was quantified in a furrow irrigated field experiment conducted on cracking clay by Thongbai *et al.* (2001). He observed that soil O₂ decreased rapidly in the waterlogged treatment, which became anoxic within 48 hours of inundation. At 24 hrs after waterlogging, photosynthesis decreased by 30% relative to non-waterlogged plants, while reduction in the leaf greenness occurred slower and biomass production did not decrease until after the 3rd waterlogging event. Yield was not different between varieties, but waterlogging significantly lowered yield in the low ridge but not in the high ridge plots.

A field experiment was conducted by Youngsukying and Nakasathien (2008) to study the Physiological Responses of Four *Eucalyptus camaldulensis* Clones to Waterlogging in a Hydroponic System. They found that waterlogging has a detrimental effect on environmental stress in *Eucalyptus camaldulensis* (Dehn.), as does calcareous soil, while together they act synergistically to induce stress in the plants. These stresses can reduce the biomass and yield of eucalypts at all stages of growth. The net photosynthetic rate (A_{max}), stomatal conductance (gs) and transpiration rate (E) rapidly reduced in C2 and C4 after 16 days of waterlogging. The maximum quantum yield (Fv/Fm), chlorophyll content and biomass of the four clones decreased in all cultivars under waterlogging at day 16. From measured physiological parameters, C2 and C4 suffered more severe stress under waterlogging than C1 and C3. Overall, a difference between the control and treated plants could be observed at 16 days after treatment. The physiological parameters of gas exchange and the Fv/Fm ratio were proven to be suitable indicators of waterlogging-tolerant traits.

Yinghua *et al.* (2006) investigated the effect of waterlogging on morphological and physiological characteristics of Cork oak (*Quercus variabilis*) and China wingnut (*Pterocarya stenoptera*). They measured the maximum net photosynthesis rate $P(\max)$, stomatal conductance $G(s)$, fluorescence maximum quantum efficiency (F_v/F_m) , chlorophyll content and water potential on different days during a period of waterlogging. Throughout a 70-day waterlogged period, the survival rates of seedlings of the two species were 100%. The earliest effects of waterlogging were significantly decreased $P(\max)$, $G(s)$ and F_v/F_m . On the 5th day of waterlogging, F_v/F_m of cork oak and China wingnut decreased to 0.694 and 0.757, respectively. On the 7th day, significant reduction of $P(\max)$ (cork oak, 39% of control; China wingnut, 42%) and $G(s)$ (cork oak, 38.8% of control; China wingnut, 71.9%) were observed. With prolonged waterlogging, $P(\max)$, $G(s)$ and F_v/F_m recovered gradually to control values in China wingnut, but not in cork oak where values decreased continuously. On the 70th day, $P(\max)$ of cork oak was reduced by 94.1% of control and F_v/F_m was only 0.537. Waterlogged China wingnut seedlings developed hypertrophied lenticels and adventitious roots at their stem base and exhibited no visible symptoms of injury (neither mortality, leaf chlorosis, leaf necrosis, leaf abscission nor reduced chlorophyll content). However, cork oak showed leaf necrosis with waterlogging. The chlorophyll content of cork oak steadily declined from the 33rd day and the Chl_a/Chl_b ratio also decreased. Predawn leaf water potential was higher in waterlogged cork oak seedlings compared to the control on the 10th and 70th days, but lower in waterlogged China wingnut seedlings. This indicated that water potential changes under waterlogging are species specific. All results show that cork oak exhibits damage to its photosynthetic apparatus under waterlogging. In contrast, although the photosynthetic apparatus of China wingnut is initially affected, it can recover,

indicating that China wingnut seedlings can tolerate prolonged soil waterlogging better than cork oak seedlings. They conclude that it is more suitable to plant China wingnut in the drawdown areas of reservoirs and ponds than cork oak.

CHAPTER III

MATERIALS AND METHODS

This chapter describes the materials used and methods of the experiment done in the laboratory to study the effect of waterlogging on eggplant seedlings. The materials and methods that were used and followed for conducting the experiment presented under the following headings:

3.1 Experimental site and duration

The experiment was conducted at the Plant Physiology Research Laboratory of Horticulture Research Center (HRC), Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur-1701, under the agro-ecological zone of Modhupur Tract (AEZ No. 28) from the First of May to the last of June 2011, in a screened enclosure with a clear polyethylene roof. The location of the site was about 35 km North of Dhaka city with 24.9⁰ N latitude and 90.26⁰ E longitude and elevation of 8.40 m from the sea level (Khan, 2009).

3.2 Climatic condition of the experimental site

The climate of the experimental site was under the subtropical climate, characterized by three distinct seasons, winter season from November to February and the pre-monsoon or hot season from March to April and the monsoon period from May to October (Edris *et al.* 1979). Details of the meteorological data during the period (May and June) of the experiment were collected from the Metrological Centre, Bangladesh Agricultural Research Institute, Gazipur and presented in Appendix I.

3.3 Plant Material

Eggplant cv. BARI Begun- 10 was selected for evaluation and the seeds of eggplant were collected from Olericulture Division, HRC, BARI, Gazipur.

3.4 Treatment of the experiment

The experiment consisted of two factors. Details were presented below:

Factor A: Seedling age

- i. A₁: 30 Days from emergence
- ii. A₂: 40 Days from emergence

Factor B: Durations of waterlogging

- i. B₀: 0 hours
- ii. B₁: 24 hours
- iii. B₂: 48 hours
- iv. B₃: 72 hours
- v. B₄: 96 hours
- vi. B₅: 120 hours
- vii. B₆: 144 hours

The seedlings were subjected to waterlogging stress of different durations (0, 24, 48, 72, 96, 120 and 144 hours) by placing poly bags with plant inside the larger plastic tubs, then irrigating with an excessive quantity of tap water at 27.5°C, so that the level of water above the surface of the soil was 8 cm throughout the waterlogged periods. The seedlings which were growing under normal conditions serve as the corresponding control. Seedlings in the control treatment were irrigated at one day interval and excess water was allowed to drain.

3.5 Design and layout of the experiment

Two factors experiment was laid out and each treatment was replicated three (3) times in a Completely Randomized Design (CRD). An experimental unit consisted of 5 seedlings per tub (each seedlings with more than 5 fully expanded leaves), among them 3 seedlings were evaluated. The data were collected from selected 3 seedlings per replication per treatment.

3.6 Potting media

Seedlings were in 15 cm x 10.5 cm polyethylene bags filled with 450 g of soil composed of 70% sandy loam soil and 30% well decomposed cowdung. The soil was air-dried, grind and passed through 2 mm sieve and analyzed for important physical and chemical parameters in the Soil Science Division, BARI, Gazipur and have been presented in Appendix II. The potting media were fertilized by 10g Urea, 10g TSP and 10g MOP per cubic feet soil at 15 days before the seed sowing.

3.7 Raising of seedlings

Three seeds of eggplant were sown directly in polyethylene bag. The emergence of the seedlings took place with 5 to 6 days after sowing. Then they were thinned to one plant per bag.

3.8 Data collection

Before initiating treatments, plants were irrigated one day interval to the pot capacity for one week in order to improve root development. Excess water drained through the

holes in the bases of the pot. The data were collected at 11.30 am to 13.30 pm during the experimental period.

3.8.1 Morphological parameters

Everyday within 11 am to 13 pm Plant height (cm), Stem girth (mm), No. of leaf, Leaf length (cm), Leaf breadth (cm) and Petiole Length (cm) were recorded.

3.8.1.1 Plant height: Height of selected eggplants were measured from the base of the plant up to tip of the plant and expressed in centimeter (cm).

3.8.1.2 Stem girth: Girth of selected plant was recorded at the base of the third leaf and expressed in centimeter (mm).

3.8.1.3 Retention of leaves per plant: Retention of leaves per plant were counted and recorded.

3.8.1.4 Leaf length: Length of growing first leaf was measured from base of leaf and expressed in centimeter (cm).

3.8.1.5 Leaf breadth: Breadth of growing first leaf was recorded from the leaf which was selected for measuring length and expressed in centimeter (cm).

3.8.1.6 Petiole Length: Length of petiole of growing first leaf was measured from petiole base to initiation point of frond and expressed in centimeter (cm).

3.8.2 Photosynthetic characters

Photosynthetic characters were measured from the youngest fully-expanded leaf in the third position from the tip.

3.8.2.1 Gas exchange characters

Gas exchange attributes viz. photosynthesis rate (A), sub-stomatal CO₂ (ci), stomatal conductance (gs) and transpiration rate (E) were measured by using an ADC LCpro+4 portable Photosynthesis System (ADC BioScientific Limited, Hoddesdon, UK).

3.8.2.2 Chlorophyll fluorescence

The polyphasic rise of chlorophyll fluorescence transients was measured by an ADC Infrared Gas Analysis Plant Efficiency Analyzer (PEA, Handsatech Instruments Ltd., King's Lynn, UK). Chlorophyll fluorescence measurements were conducted under dark conditions during the period from 11.30 am to 13.30 pm using a dark leaf clip (DLC-8) to protect the readings from the effect of temperature. The initial fluorescence (F₀) value was measured under a low irradiance and the maximum fluorescence (F_m) value was measured under a high irradiance. The quantum efficiency of open photosystem II centres (quantum yield) was then calculated as F_v/F_m.

$$F_v/F_m = (F_m - F_0)/F_m$$

3.8.2.3 Chlorophyll content

Leaf chlorophyll content was measured from the youngest fully-expanded leaf in the third position from the tip by Chlorophyll Content Index (CCI) meter (Chlorophyll Content meter, CCM-200, OPTI-SCIENCES, USA). CCI is a linear scale. These instruments and scales have been shown to be correlated to chlorophyll chemical tests for chlorophyll content except at very high levels.

3.9 Statistical analysis

The data obtained for different characters were statistically analyzed. The mean values of all the recorded characters were evaluated and analysis of variance was performed by the 'F' (variance ratio) test. The significance of the difference among the treatment of means were estimated by LSD Test at 5% level of probability (Gomez and Gomez, 1984) by using MSTAT software and t test was performed by Microsoft office excel 2007 software.

CHAPTER IV RESULTS AND DISCUSSION

The experiment was conducted to study the effect of waterlogging on morphological and photosynthetic characters at seedling stage of eggplant. The analyses of variance (ANOVA) of the data are presented in Appendix III-IV. The results have been presented by using table and graphs and discussed with possible interpretations under the following headings:

4.1 Effect of waterlogging on morphological characters

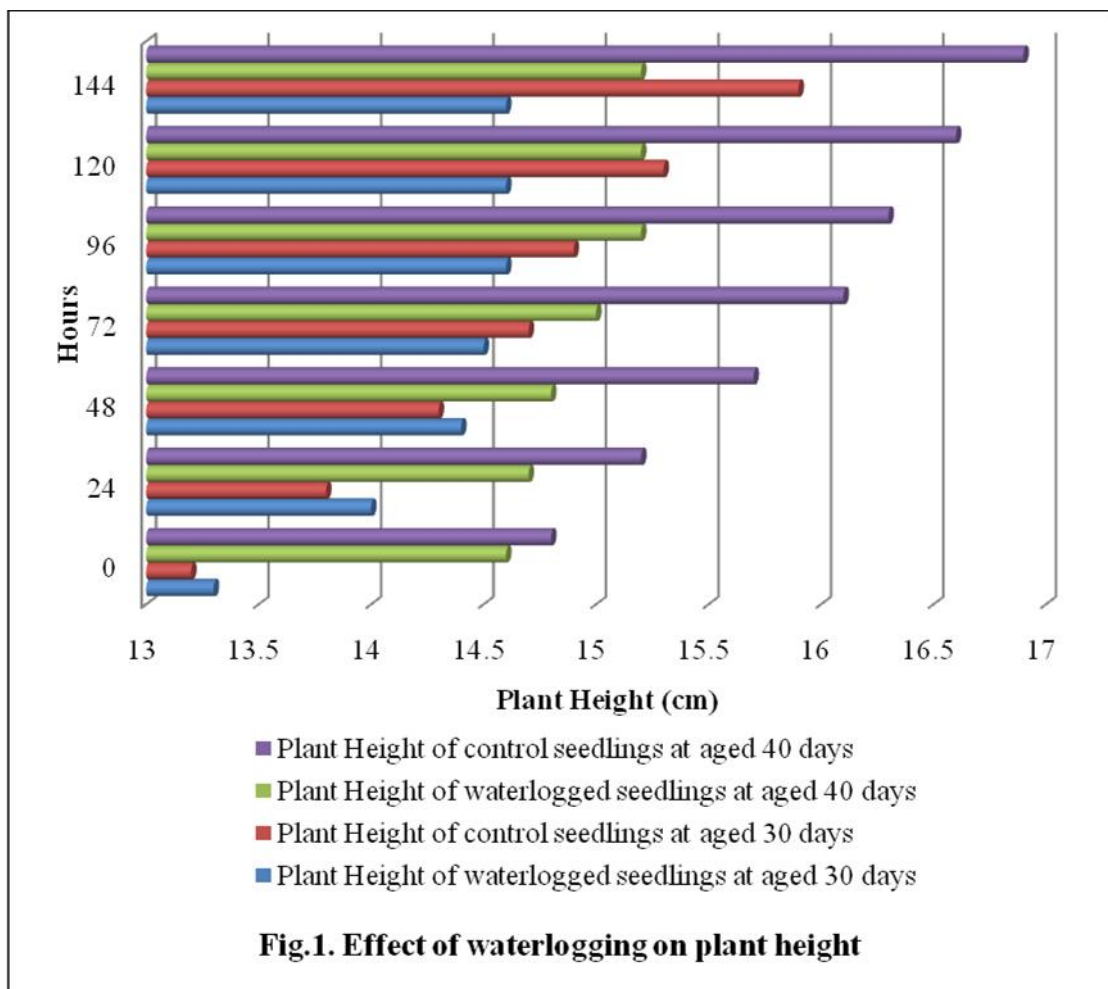
4.1.1 Plant Height

Plant height was affected by waterlogging. Significant variation was observed in different aged 30 and 40 days old seedlings compared to control. The variations between the age of seedlings and among the durations of waterlogging were not statistically significant. Plant height increased gradually in 30 and 40 days old control seedlings. On the other hand plant height increased up to 96 hours of waterlogging in both 30 and 40 days old seedlings and then the upward growth became stopped

(Figure 1). Similar effect was observed by Ezin *et al.* (2010) in tomato seedlings that plant height was decreased as the days of flooding increased.

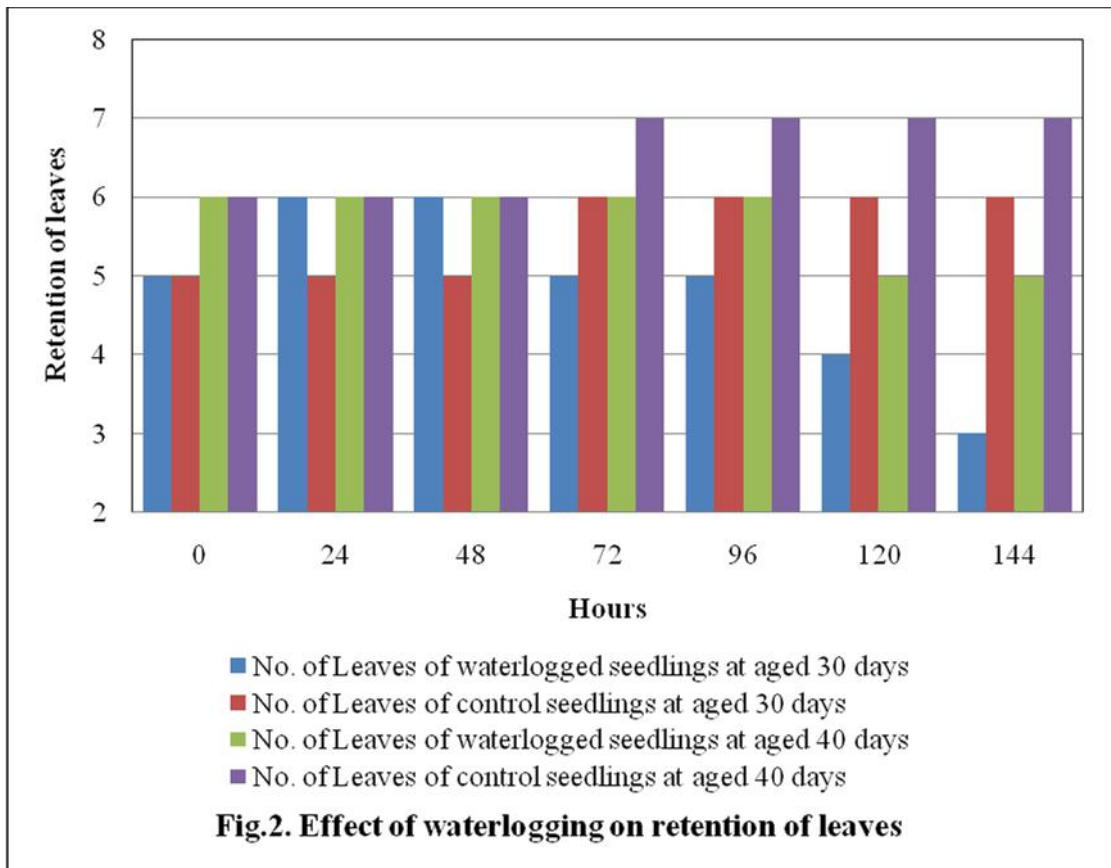
4.1.2 Stem Girth

Growth of stem girth significantly reduced under waterlogged condition compared with control in both 30 and 40 days old seedlings (Table 2 and 3). Normal growth of stem girth observed at the first 24 hours of waterlogging and then growth became slower to 72 hours. Growth of stem girth discontinued after 96 hours in 30 days old and after 72 hours in 40 days old seedlings under waterlogging (Table 1). Similar effect was observed in the combination of seedling age and the durations of waterlogging. Bennett (2003) reported that when roots are partially submerged (waterlogged) or completely submerged (flooded) shoot growth become reduced.



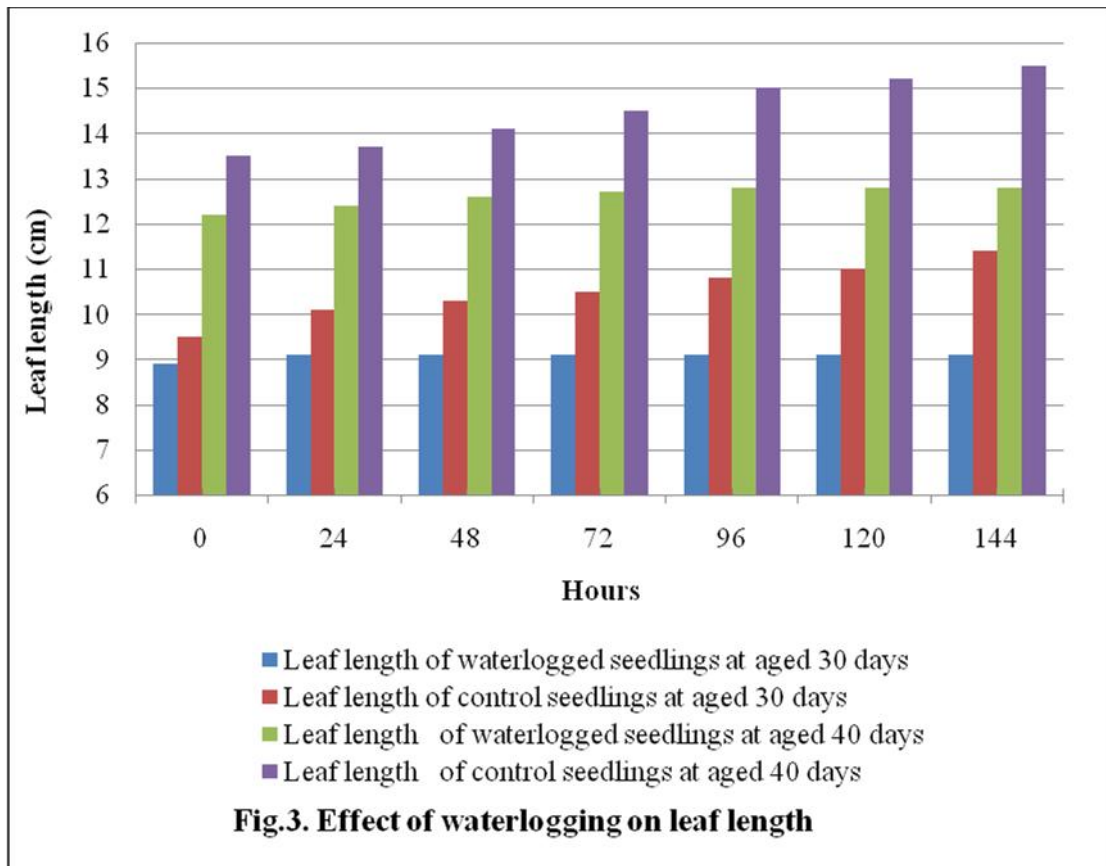
4.1.3 Retention of Leaves

Waterlogging caused early senescence of leaves. Number of leaves significantly reduced with increasing the duration of waterlogging. Senescence of leaves started from 48 hours later in 30 days old seedlings and 96 hours later in 40 days old seedling (Figure 2). Farther more leaf fall was not observed in control seedlings within the period. Kramer (1951) stated that the symptom of flooding injury is yellowing and death of the leaves, from the lower ones to the stem. Drew and Sisworo (1977) reported that the early senescence of leaves in flooded plants are caused by the inhibition of nitrogen (N) uptake, and the consequent redistribution of nitrogen within the shoot.



4.1.4 Leaf Length

Waterlogging induce negative effect on leaf growth. Leaf length of waterlogged and control seedlings significantly differed with duration of waterlogging. Furthermore, the variation among the duration of waterlogging and combination of durations and seedling age showed similar effect. In case of leaf length sever effect was observed in 30 days old seedlings, leaf growth became restricted after 24 hours of waterlogging and in 40 days old seedlings it was 72 hours later (Fig. 3). Drew (2000) stated that during waterlogging hypoxia or anoxia of the soil causes inhibition of the vegetative growth of plants.



4.1.5 Leaf Breadth

Different duration of waterlogging significantly affects on the leaf breadth in 30 and 40 days old waterlogged seedlings compared to control (Table 2 and 3). The effect within the duration of waterlogging in the same aged seedlings and combination of age and duration were not varied. Growth became seized after 24 hours in 30 days old seedlings and 48 hours in 40 days old seedlings (Table 1). According to Bray *et al.* (2000) under waterlogged condition, plants reduce their growth rate.

4.1.6 Petiole Length

Petiole length differed significantly in different age of seedlings and durations of waterlogging with the control seedlings (Table 2 and 3). Moreover, variation among the durations of waterlogging in the same aged seedlings and combination of seedling age and durations of waterlogging were statistically insignificant (Table 1). Growth of

petiole length was stopped after 72 hours of waterlogging in both 30 and 40 days old waterlogged seedlings.

Table 1. Effect of waterlogging on morphological characters at 30 and 40 days old eggplant seedling

Seedling Age	Waterlogged period (Hours)	Parameters		
		Stem Girth (mm)	Leaf Breadth (cm)	Petiole Length (cm)
A ₁	B ₀	3.7	5.9	3.8
	B ₁	4.4	6.0	4.0
	B ₂	4.5	6.1	4.1
	B ₃	4.5	6.1	4.3
	B ₄	4.6	6.1	4.3
	B ₅	4.6	6.1	4.3
	B ₆	4.6	6.1	4.4
A ₂	B ₀	5.4	8.1	4.9
	B ₁	6.2	8.2	5.1
	B ₂	6.5	8.3	5.2
	B ₃	6.7	8.3	5.3
	B ₄	6.7	8.3	5.3
	B ₅	6.7	8.3	5.3

	B ₆	6.7	8.3	5.3
	CV%	6.09	14.89	14.59
	LSD (.05)	0.5522	1.790	1.159

A₁: 30 days old eggplant seedling

A₂: 40 days old eggplant seedling

B₀: No waterlogging (control)

B₄: 96 hours waterlogging

B₁: 24 hours waterlogging

B₅: 120 hours waterlogging

B₂: 48 hours waterlogging

B₆: 144 hours waterlogging

B₃: 72 hours waterlogging

Table 2. Effects of waterlogging on morphological characters of 30 days old waterlogged and control eggplant seedlings

Waterlogged period (Hour)	Parameters					
	Leaf Breadth(cm)		Stem Girth(mm)		Petiole Length(cm)	
	Waterlogged	Control	Waterlogged	Control	Waterlogged	Control
B ₀	5.9	5.8	3.7	3.7	3.8	4.3
B ₁	6.0	6.3	4.4	3.9	4.0	4.6
B ₂	6.1	6.5	4.5	4.1	4.1	5.0
B ₃	6.1	6.8	4.5	4.2	4.3	5.1
B ₄	6.1	7.1	4.6	4.4	4.3	5.4
B ₅	6.1	7.4	4.6	4.4	4.3	5.6
B ₆	6.1	7.8	4.6	4.5	4.4	5.8
Calculated t	0.02121		0.009628		1.00	
Level of significant	**		**		**	

*indicates significant at 5% level

**indicates significant at 1% level

B₀: No waterlogging (control)

B₄: 96 hours waterlogging

B₁: 24 hours waterlogging

B₅: 120 hours waterlogging

B₂: 48 hours waterlogging

B₆: 144 hours waterlogging

B₃: 72 hours waterlogging

Table 3. Effects of waterlogging on morphological characters of 40 days old waterlogged and control eggplant seedlings

Waterlogged period (Hour)	Parameters					
	Leaf Breath(cm)		Stem Girth(mm)		Petiole Length(cm)	
	Waterlogged	Control	Waterlogged	Control	Waterlogged	Control
B ₀	8.1	7.2	5.4	5.7	4.9	4.7
B ₁	8.2	7.3	6.2	5.8	5.1	4.9
B ₂	8.3	7.8	6.5	6.3	5.2	5.0
B ₃	8.3	8.1	6.7	6.7	5.3	5.2
B ₄	8.3	8.4	6.7	6.7	5.3	5.4
B ₅	8.3	8.6	6.7	6.8	5.3	5.5
B ₆	8.3	8.8	6.7	6.9	5.3	5.7
Calculated t	0.323164		1.00		0.000332	
Level of significant	**		**		**	

*indicates significant at 5% level

**indicates significant at 1% level

B₀: No waterlogging (control)

B₄: 96 hours waterlogging

B₁: 24 hours waterlogging

B₅: 120 hours waterlogging

B₂: 48 hours waterlogging

B₆: 144 hours waterlogging

B₃: 72 hours waterlogging

4.2 Effect of waterlogging on Photosynthetic characters

4.2.1 Chlorophyll fluorescence

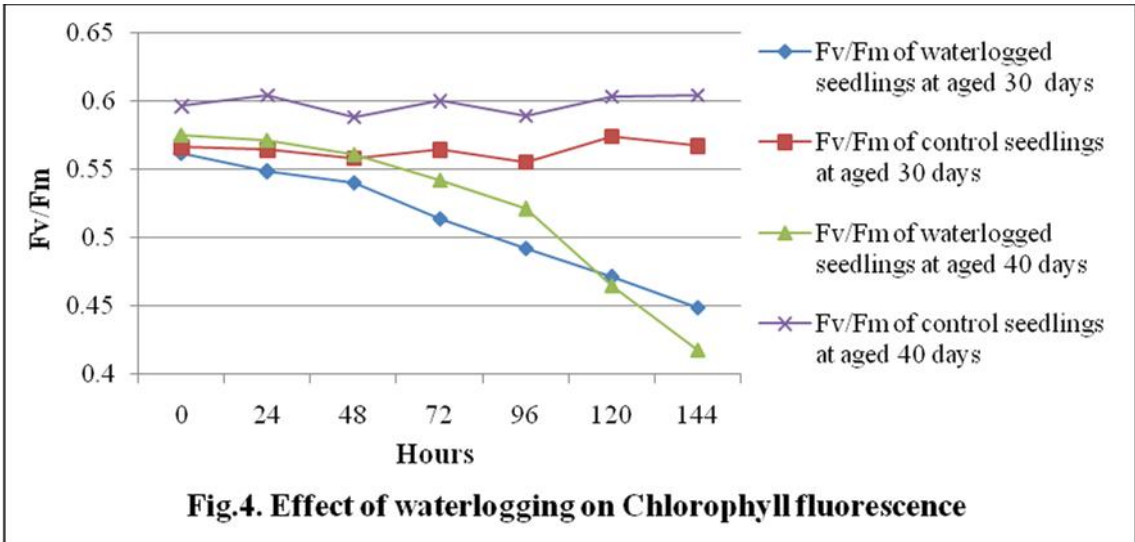
Light- adapted F_v/F_m estimated the efficiency of excitation energy transfer to open PSII centers. The Chlorophyll fluorescence values (F_v/F_m ratio) of waterlogged eggplant seedlings were decreased significantly. Under environmental stress, the plant disperses excess light in the form of heat and fluorescence. In the waterlogged treatment, aged seedlings had a lower F_v/F_m than in the control plants at the end of 144 hours. In this experiment, as the measurements were taken in the dark, there was no effect of temperature. In case of F_v/F_m ratio slight change was observed up to 48 hours of waterlogging and then dramatic reduction took place in both cases. After 48 hours of waterlogging gradual reduction was observed in 30 days old seedlings. On the other hand gradual fall down continued after 48 hours of waterlogging upto 96 hours in 40 days old seedlings then after drastic fall down observed in the rest of the period (Figure 4). Light- adapted F_v/F_m ratio observed lower in waterlogged seedlings compared to control. Baker (1993) and Cornic (1994) reported that under severe water stress, electron transport to O_2 and increased quenching of excitation energy in the photo system II (PSII) antennae may be unable to dissipate the excess excitation energy in the PSII antennae and photo damage of PSII will result, with a possible net loss of D_1 protein of PSII reaction centers. Such effects can have significant consequences for the photosynthetic productivity of plants (Long *et al.*, 1994). A decrease in the ratio of variable to maximal chlorophyll fluorescence (i.e., dark-adapted F_v/F_m) has been employed as an indicator of the degree of photoinhibition (Gravett 1992; Groom *et al.* 1992; Moseki 1997; Kreslavski *et al.* 2008).

4.2.2 Photosynthesis

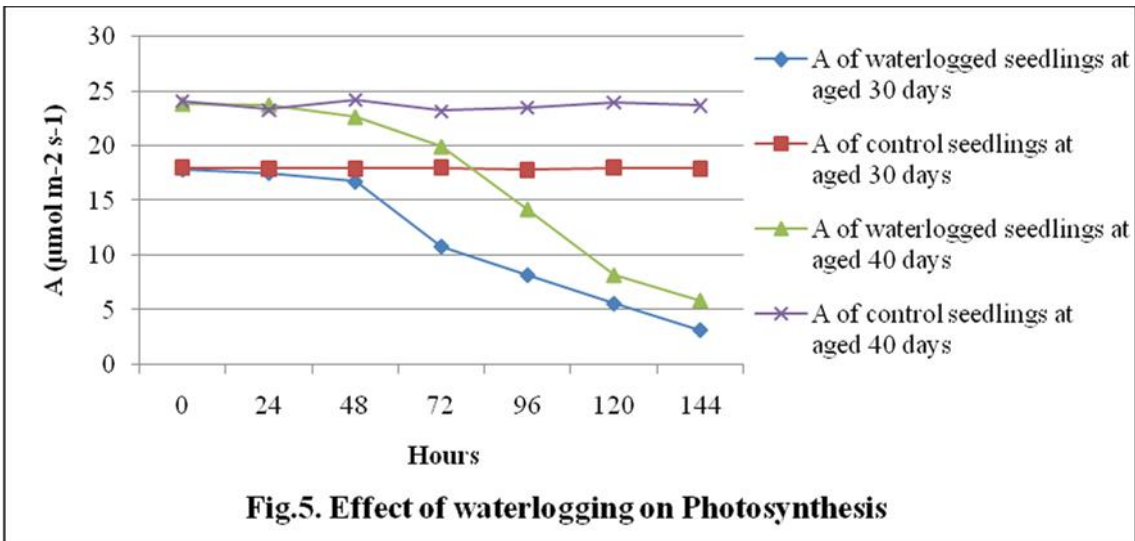
Photosynthetic rate (A) of different aged waterlogged seedlings and among the durations of waterlogging were significantly decreased (Figure 5). Gradual reduction of photosynthetic rates (A) of waterlogged 30 and 40 days old seedlings were started after 48 hours of waterlogging was imposed. According to Musgrave and Ding (1998) photosynthesis rapidly decreased, with a subsequent decrease in carbohydrate production and consequently the growth rate was reduced. Kozłowski and Pallardy (1984) reported that the reduction in net photosynthesis may have resulted from effects of waterlogging on carboxylation enzymes and the loss of chlorophyll.

4.2.3 Stomatal conductance of CO₂

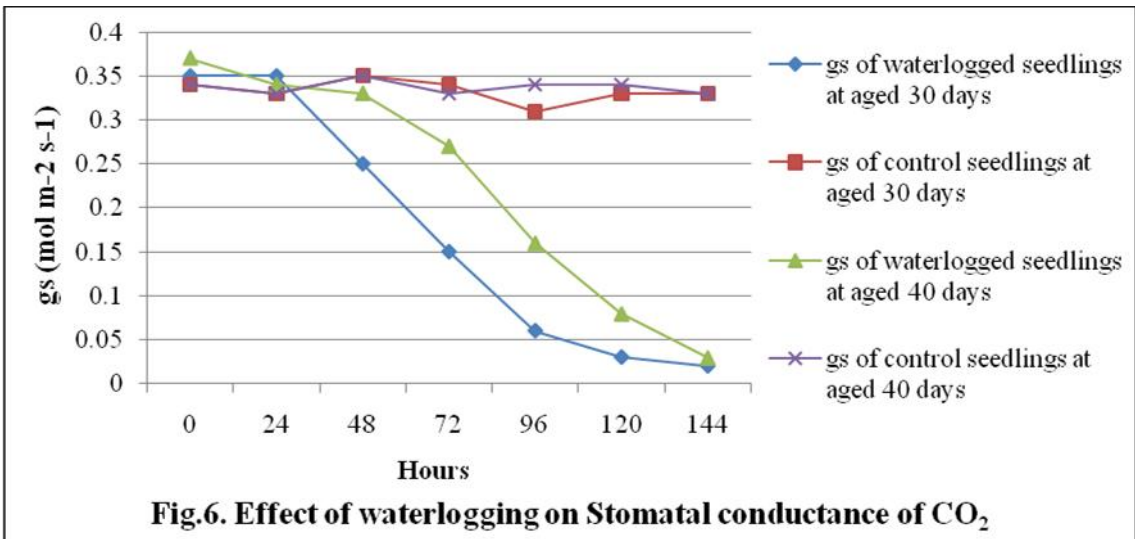
Waterlogging severely affect stomatal conductance. Stomatal conductance of CO₂ (gs) significantly declined in both 30 and 40 days old waterlogged seedlings. Stomatal conductance of CO₂ decreased rapidly after 24 hours in 30 days and 48 hours of waterlogging in 40 days old waterlogged seedlings (Figure 6). Smith and Moss (1998) stated that waterlogging affects some major physiological functions. Root-zone saturation and stomatal closure lead to reduced stomatal conductance. Bradford *et al.* (1982) observed in tomato leaves that waterlogging had no significant effect on stomatal conductance of CO₂ during the first day. On the second and subsequent days of waterlogging, stomatal conductance of CO₂ of waterlogged plants averaged only 72% of the control value.



*Fv/Fm: Chlorophyll fluorescence



*A: Photosynthesis



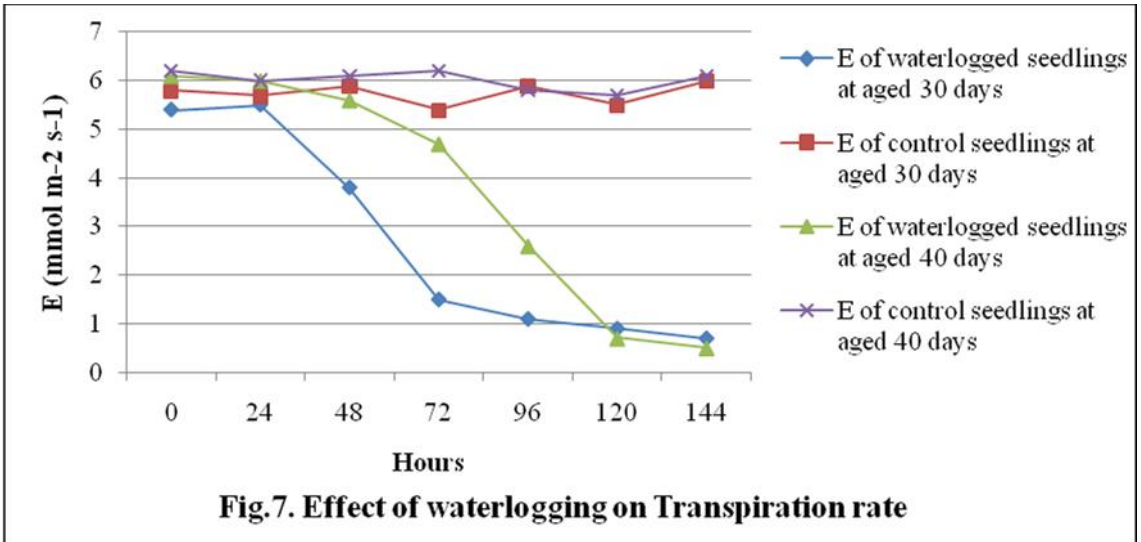
* g_s : Stomatal conductance of CO_2

4.2.4 Transpiration rate

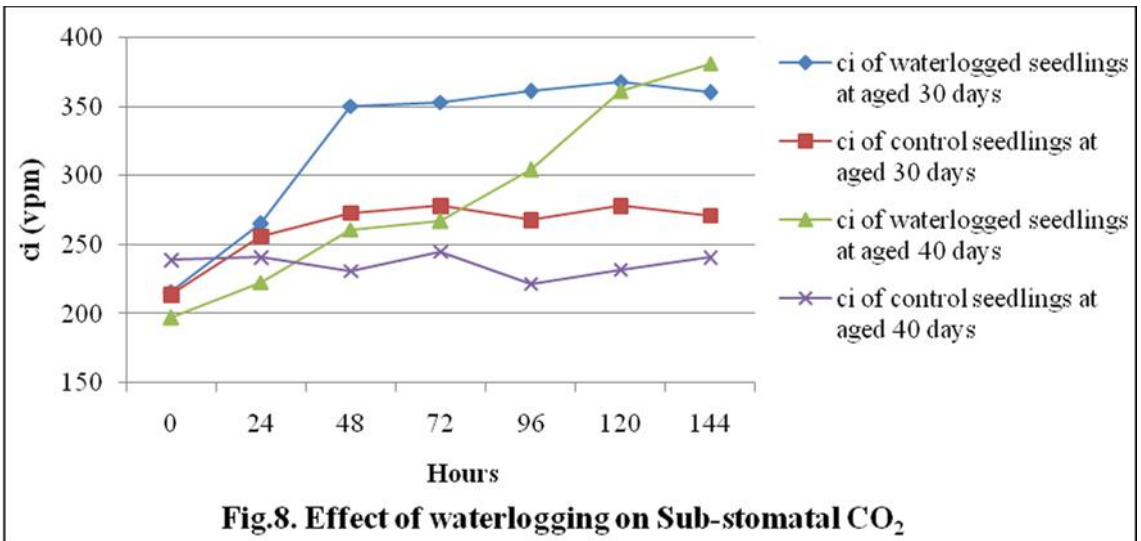
Transpiration rate (E) closely related to stomatal closure and responded linearly with stomatal conductance (g_s) and also with photosynthesis rate (A). It declined when stomatal conductance (g_s) was decreased. Insignificant variation was observed in first 24 hours and the subsequent time it was significant compared to control. Progressive decline of transpiration rate started after 24 hours of waterlogging in 30 days old seedlings. In case of 40 days old seedlings a sharp decrease of transpiration rate was seen between hours 24 and 72 followed by a dramatic decrease (Figure 7). Bradford *et al.* (1982) studied waterlogged tomato plants and concluded that transpiration was reduced in close proportion to the changes in stomatal conductance, showing no effect on the first day of flooding and averaging 68% of the control values on subsequent days.

4.2.5 Sub-stomatal CO_2

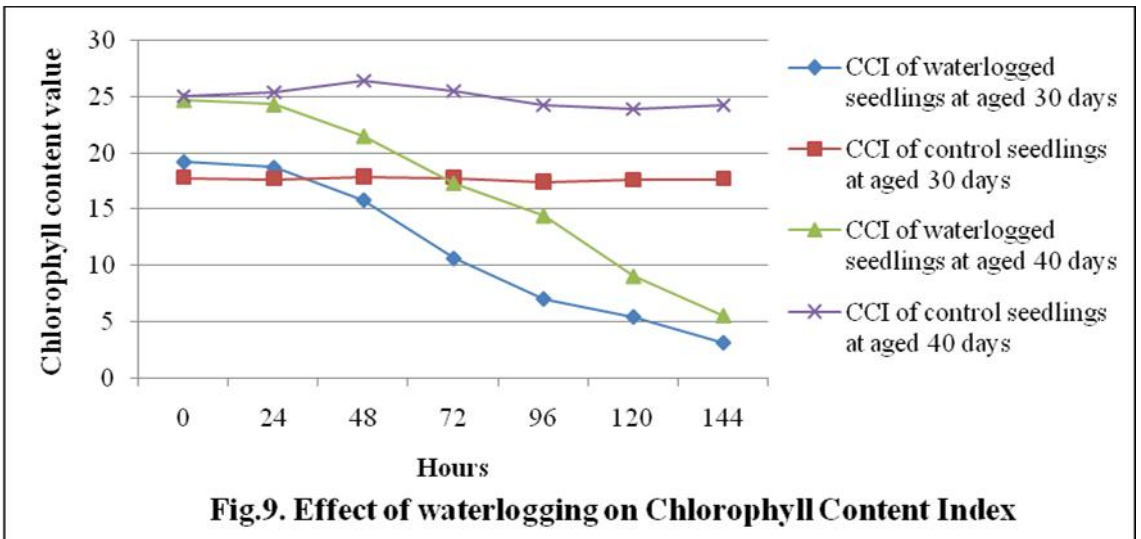
Sub-stomatal CO_2 showed negative relation with stomatal conductance of CO_2 (g_s) in waterlogged seedlings. Sub-stomatal CO_2 significantly varied with durations of waterlogging and the different age of seedlings. Sub-stomatal CO_2 rapidly increased between 24 and 48 hours of waterlogging and then became similar in 30 days old seedlings while it gradually increased in 40 days old waterlogged seedlings along the period (Figure 8). Sub-stomatal CO_2 (c_i) was increased because of decreasing CO_2 assimilation and inhibition of CO_2 metabolism. Two principal effects are invoked by Cornic (1994), restricted diffusion of CO_2 into the leaf which is caused by stomatal closure (Quick, 1992; Brestic *et al.*, 1995 and Ort *et al.*, 1994) and inhibition of CO_2 metabolism (Gimenez *et al.*, 1992).



*E: Transpiration rate



*ci: Sub-stomatal of CO₂



4.2.6 Chlorophyll Content Index

Chlorophyll content was strongly affected by waterlogging. Significant reduction was observed within the durations of waterlogging and different age of the seedlings (Figure 9). Chlorophyll content started to decline after 24 hours of waterlogging in both 30 and 40 days old waterlogged seedlings after 144 hours of waterlogging both aged seedlings had a lower level of chlorophyll compared with the control. Youngsukyng and Nakasathien (2008) found that, the total chlorophyll content declined more drastically in the waterlogged plants compared to the control.

CHAPTER V SUMMARY AND CONCLUSION

SUMMARY

An experiment was conducted at the Plant Physiology Research Laboratory of HRC, BARI, Joydebpur, Gazipur-1701, during the period of May to June 2011, to study the effect of waterlogging on morphological and photosynthetic characters of eggplant seedling. The experiment consisted of two factors. The experiment consisted of two factors. Factor A: Seedling age; A₁: 30 Days from emergence; A₂: 40 Days from emergence and Factor B: Durations of waterlogging; B₀: 0 hours, B₁: 24 hours, B₂: 48 hours, B₃: 72 hours, B₄: 96 hours, B₅: 120 hours and B₆: 144 hours. The experiment was laid out in a Completely Randomized Design with three replications. Seeds of BARI Begun-10 were selected for evaluation in this experiment.

Waterlogging, an abiotic stress of nature, takes place when air in the soil is replaced with water caused by excessive rainfall, flooding or poor drainage. The effects of waterlogging on morphological parameters were more or less similar between the ages of seedlings and among the durations of waterlogging. Plant height increased up to 96 hours of waterlogging in both seedling age and then upward growth became stopped. Normal growth of stem girth was observed at the first 24 hours and then slower to 72 hours. Moreover, growth of stem girth discontinued after 96 hours in 30 days old and 72 hours in 40 days old seedlings. Senescence of leaves started from 48 hours later in 30 days old seedlings and 96 hours later in 40 days old seedlings. Furthermore, growth of leaf length became restricted after 24 hours of waterlogging in 30 days and in 40 days old seedlings it was 72 hours later. Again the growth of leaf breadth became seized after 24 hours in 30 and 48 hours in 40 days old seedlings and the growth of petiole length was stopped after 72 hours of waterlogging in both seedling ages.

In case of F_v/F_m ratio slight change was observed up to 48 hours of waterlogging then dramatic reduction took place in both cases. After 48 hours of waterlogging gradual reduction was observed in 30 days old seedlings. On the other hand gradual decrease continued up to 96 hours, then after drastic fall down was observed in the rest of the period in 40 days old seedlings. Gradual reduction of photosynthetic rates of waterlogged seedlings were started from 48 hours of waterlogging in both cases and stomatal conductance of CO_2 decreased rapidly after 24 hours and 48 hours of waterlogging in 30 and 40 days old seedlings respectively. Rapid reduction of transpiration rate started after 24 hours of waterlogging in 30 days old seedlings and in 40 days old seedlings, a sharp decrease of transpiration rate was seen between hours 24 and 72 followed by a dramatic change. On the other hand sub-stomatal CO_2

rapidly increased between 24 and 48 hours of waterlogging and then became stable in 30 days old seedlings while gradually increased in 40 days old waterlogged seedlings and chlorophyll content gradually declined in both 30 and 40 days old waterlogged seedlings.

CONCLUSION

Waterlogging severely affects morphological and photosynthetic characters of eggplant seedlings. The negative impact increased with increasing the durations of waterlogging in both 30 and 40 days old seedlings. Drastic effect of waterlogging started after 48 hours of waterlogging in 30 days old seedlings and in 40 days old it was 72 hours. The results of this experiment revealed that the effect of waterlogging correlated with the duration of waterlogging and age of the seedlings. These results will be helpful to select waterlogging tolerant eggplant germplasm.

RECOMMENDATION

From the above discussion the following recommendations may be made-

1. Repetition of this experiment will be needed to confirm this result.
2. Further investigation need to be conducted including higher age of seedlings.

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APPENDICES

Appendix I. Monthly record of temperature, relative humidity, rainfall during May and June 2011

Year	Month	*Air temperature (°C)		*Relative Humidity (RH) (%)	*Rainfall (mm)
		Max.	Min.		
2011	May	32.90	24.50	76.00	339.4
2011	June	32.10	26.10	82.20	340.40

* Monthly average,

Source: Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur.

Appendix II. Analytical data of soil sample

Year	p ^H	Total N	OM	Ca	Mg	K
		%			meq/100g	
2011	6.1	0.077	1.46	4.76	1.97	0.15
Critical level				2.0	0.8	0.2

Appendix II. Cont'd.

Year	p ^H	P	S	B	Cu	Fe	Mn	Zn
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µg/g								
2011	6.1	15	38	0.32	6.0	232	10	3.30
Critical level		14	14	0.2	1.0	10.0	5.0	2.0

Source: Soil Science Division, Bangladesh Agricultural Research Institute (BARI),
Gazipur.

Appendix III. Analysis of variance of the data on morphological characters of eggplant as influenced by seedling age and different duration of waterlogging

Source of variation	Degrees of freedom	Mean square					
		Morphological characters					
		Plant height (cm)	Stem Girth (mm)	Retention of Leaves	Leaf Length (cm)	Leaf Breadth (cm)	Percentage of waterlogging
Seedling age (A)	2	4.496**	43.819**	4.667**	129.326**	50.381**	
Duration of waterlogging (B)	6	0.832**	0.969**	2.206NS	0.137NS	0.034NS	
Interaction (A×B)	6	0.129NS	0.050NS	0.444NS	0.038NS	0.001NS	
Error	28	0.923	0.109	0.048	1.445	1.146	

** : Significant at 0.01 level of significance

* : Significant at 0.05 level of significance

NS: Non significant

Appendix IV. Analysis of variance of the data on photosynthetic characters of eggplant as influenced by seedling age and different duration of waterlogging

Source of variation	Degrees of freedom	Mean square					
		Photosynthetic characters					
		Chlorophyll fluorescence (Fv/Fm)	Photosynthesis ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	Stomatal conductance of CO_2 ($\text{mol m}^{-2} \text{s}^{-1}$)	Transpiration rate ($\text{mmol m}^{-2} \text{s}^{-1}$)	Sub-stomatal CO_2 (vpm)	Percentage of waterlogging
Seedling age (A)	2	0.001**	317.845**	0.025**	8.979**	50.381**	
Duration of waterlogging (B)	6	0.015**	271.727**	0.121**	34.173**	0.034**	
Interaction (A×B)	6	0.001**	7.707**	0.005**	2.466**	0.001**	
Error	28	0.001	0.483	0.001	0.150	1.146	

** : Significant at 0.01 level of significance;

* : Significant at 0.05 level of significance

NS: Non significant