EFFECT OF ARSENIC ON THE GROWTH, YIELD AND ARSENIC CONTENT IN LEAF AND GRAIN OF MAIZE

(*Zea mays*)

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CERTIFICATE

*This is to certify that the thesis entitled, **"EFFECT OF ARSENIC ON THE GROWTH, YIELD AND ARSENIC CONTENT IN LEAF AND GRAIN OF MAIZE (Zea mays)"** submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in the partial fulfillment of the requirements for the degree of MASTER OF SCIENCE (M.S.) IN AGRICULTURAL CHEMISTRY, embodies the result of a piece of bona fide research work carried out by **MD. TANZIM ISLAM, Registration No. 11-04604** 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 source of information, as has been availed of during the course of this investigation has duly been acknowledged.

Dated: June, 2017
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Dedicated to

My

Beloved Parents

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ABSTRACT

The experiment was performed in pot at the net house and laboratory, Department of Agcultural Chemistry, Sher-e-Bangla Agricultural University, Dhaka during November 2016 to March 2017 and to assess the effect of arsenic on growth, yield and arsenic content in leaf and grain of maize (*Zea mays*). The experiment was conducted using two varieties viz. V_1=BARI maize 7, V_2= Pacific 339 and five arsenic levels viz. T_0 = 0 mg As/kg soil, T_1 = 20 mg As/kg soil, T_2 = 40 mg As/kg soil, T_3 =60 mg As/kg soil and T_4 =80 mg As/kg soil.Arsenic was added as sodium arsenate ($Na_2HAsO_4.7H_2O$).The experiment was set in completely randomized design **(**CRD) having two factors with three replications. Results showed that variety had produced plant height, number of leaves per plant, total dry mater, number of cob per plant, number of grain per plant, grain yield per plant.The taller plant was found in Pacific 339. BARI maize 7 achieved maximum number of cob per plant, number of grain per cob, grain yield per plant. Among the two varieties the higher grain yield per plant (31.79 g) was found in BARI maize 7. Different arsenic dose significantly affected all growth characters.The maximum plant height, number of leaf per plant, total dry matter , number of cob per plant, number of grain per cob were recorded at 0 mg As/kg of soil levels of arsenic. The highest grain yield (58.54g) per plant was also recorded at 0 mg As/kg of soil level of arsenic. Among the combined effects of varieties and arsenic, highest grain yield per plant (63.75 g) was observed from the combination of BARI maize 7 at 0 mg As/kg of soil arsenic level. Among the two varieties, the N content in leaf was highest (0.87 %) in BARI maize 7. The highest K content, P content and As content in leaf was found in Pacific 339. The highest N, P and K content recorded in shoot of BARI maize 7. The As content in stem was highest in Pacific 339. The highest N, P and K content recorded in grain of BARI maize 7. The As content in grain was highest in Pacific 339. Amount of arsenic concentration was increased by both varieties significantly with the increasing arsenic doses in soil.

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LIST OF ABBREVIATIONS AND ACRONYMS

AEZ	=	Agro- Ecological Zone
BARC	=	Bangladesh Agricultural Research Council
BBS	=	Bangladesh Bureau of Statistics
FAO	=	Food and Agricultural Organigation
BRRI	=	Bangladesh Rice Research Institute
cm	=	Centi-meter
cv.	=	Cultivar
DAS	=	Days after transplanting
DAS	=	Days after sowing
DF	=	Degree of freedom
^{0} C	=	Degree centigrade
DMRT	=	Duncan Multiple Range Test
et al.	=	and others
etc.	=	Etcetera
LSD	=	Least Signicant Difference
ns	=	Non significant
%	=	Percent
CV %	=	Percentage of Coefficient of Variance
As	=	Arsenic
N	=	Nitrogen
P	=	Phosphorus
K	=	Potassium
ppm	=	Parts per million
ppb	=	Parts per billion
SAU	=	Sher-e- Bangla Agricultural University
M	=	Mollar
pH	=	Hydrogen ion concentration
TSP	=	Triple Super Phosphate
MOP	=	Muriate of Potash
t ha^{-1}	=	Tons per hectare
Zn	=	Zinc

DI	=	De-Ionized
TDM	=	Total Dry Matter
BARI	=	Bangladesh Agricultural Research Institute
Se	=	Selenium

CHAPTER I

INTRODUCTION

Maize (*Zea mays* L.) belongs to the family gramineae is one of the most leading cereal in the world next to rice and wheat (Aldrich *et al.*, 1975). Central America or Mexico is the most likely center of origin of this crop and south America is the possible secondary origin (Martin and Leonard, 1975). Africa, Asia and some central and south American countries use maize as an important staple food but it is mostly used as animal feed. Bangladesh has good potentiality to adopt it as a cereal crop due to its low cost of production, wide adaptability and diversified uses. Maize is now a popular crop because of its high yield potential. Kharif is main season, although it can be cultivated in both rabi and kharif season. Maize kernels have high nutritive value contains 66.2 % starch, 11.1 % protein, 7.1 % oil and 1.5 % minerals (Hulse *et al.*, 1980). It also contains 90 mg carotene, 1.8 mg niacin, 0.8 mg thiamin and 0.1 mg riboflavin per 100 g grains (Chowdhury and Islam, 1993). Hybrid variety is chosen due to some important points- better yield, improved color, greater uniformity, disease resistance . Maize acreage and production have an increasing tendency in Bangladesh. After introduction of hybrid since 1993 area, production and yield of maize have increased by 17 %, 33 %, and 16 % respectively which reflect the effect of adopting improved technology (Mohiuddin, 2003). The population growth in Bangladesh is high which puts great pressure on the country's food production. Cereal is still staple one for Bangladeshi people. Now maize is the third position next to rice and wheat in the country in terms of human consumption. Average yield of maize in Bangladesh is considerably low. The national average yield is only 11.24 t/ha (BBS, 2010) whereas the newly released hybrid varieties have the potential to produce more than 8.0 t/ha.

There is increasing concern worldwide regarding the contamination of soil with arsenic (As) and the potential risk to human and environmental health arising from such contamination (Smith *et al.*,1998). Arsenic is a toxic and carcinogenic element that occurs widely in soil environments around the world. Soil contamination with As occurs through both natural and anthropogenic pathways. In recent years, As pollution has become a major public concern in many countries (Smith and Naidu, 1998). Remediation of As contaminated soil and water is necessary for protecting both human life and agricultural production. There are a variety of physical-chemical technologies for remediation of As contaminated sites (U.S. Environmental Protection Agency, 1992.)

Arsenic (As), which is a highly toxic metalloid and is found ubiquitously in the environment, poses a serious risk to plants, animals and humans (Rosas-Castor *et al.*, 2014). Arsenic content in soils has increased substantially in recent years because of irrigation with As-rich water or from anthropogenic activities, such as ore mining, smelting, burning of coal, use of As pesticides and the application of wastes (Smith *et al*.,1998; Acharyya, 1999; Lambkin and Alloway, 2003). Excess of As in soil can inhibit seed germination and plant growth (Joinal Abedin and Meharg, 2003; Azizur Rahman, 2007; Shri,2009), disturb plant metabolism(Dixon, 1997) and cause plant death (Baker *et al.*, 1976 and Marin *et al.*, 1992). Arsenic can be taken up by plants and vegetables from the soil and irrigated water, and subsequently enter the food chain (Meharg, and Hartley-Whitaker, 2002). Thus, humans and other animals may consume As. Arsenic exposure can cause human diseases such as skin lesions, neurological defects, atherosclerosis and even cancer (Watts *et al.*, 2010). In recent years, the most serious As pollution problems have occurred in Bangladesh and West Bengal, India(Chowdhury *et al.*, 2001 and Bundschuh, 2012). Therefore, there is an urgent need to find out suitable methods to reduce the transmission of As to humans. One option is to remove As from the soil. However, traditional methods and phytoremediation are limited by their own shortcomings (Krämer, 2005; Pilon-Smits and Freeman, 2006). Yu *et al.*,(2006) have described the concept of the pollution-safe cultivar. This concept refers to the use of cultivars that accumulate a very low level of a specific pollutant, which ensures the crop remains safe for human consumption, even when grown in contaminated soil. The application of pollution-safe cultivar selection and breeding is considered a practical and cost-effective approach to minimize the entry of heavy metals into the human food chain, and has received widespread attention (Chen *et al.*, 2012 and Grant *et al*., 2008).

Human exposure to As occurs commonly by transfer from the crop–soil–water system (Rosas-Castor *et al.*, 2014). Recently, the accumulation and distribution of As have been studied in different crops. Abedin *et al.*, (2002) found that rice roots accumulated much more As than the straw and grain. The trend of As concentration in different rice tissues was as follows: grain <husk <straw <root (Smith *et al.*,2008). In maize,the total As content in different tissues was in the order: grain <shoot <root(Baig *at al*., 2010). Other studies have reported that the trend of As concentration in four different maize tissues was: kernels <bracts <stems <leaves (Ding, 2011 and Liu, 2012).

In maize, many studies have focused on the physiological and biochemical responses to As accumulation. The majority of these studies demonstrated a trend of decreasing As content from the roots to the aerial parts, including leaves, stems and seeds (Rosas-Castor *et al.*, 2014). Maize takes up the arsenic naturally present in the soil or arsenic that is added through groundwater irrigation or by soil additives contaminated with arsenic. Several studies have been described a significant relationship between the As concentration in the irrigation water or soil and the total As content accumulated by maize plants (Prabpai, 2009). Gulz*et al.*,(2005) observed that the correlation between the total accumulated As in maize plants and the water-soluble As fraction in the soil was higher than the total As content in the soil. Several factors, including pH, redox potential, organic matter content, interaction/competition with other elements and chemical forms of the pollutant, can affect As solubility in soils (Marwa *et al.*, 2012).

Maize is the most cultivated cereal in the world and is used as an important animal feed or a staple food crop for humans in many developing countries in Africa, Asia and Latin America (Rosas-Castor *et al.*, 2014). Hence, maize grown on Ascontaminated land could accumulate As and pose a risk to human health and other animals health. Thus, methods to reduce As accumulation in maize are urgently needed.Thus this study was under taken with the following objectives.

1. To determine the effect of different As concentrations on growth and yield of maize.
2. To make comparative study on arsenic accumulation from soil by different maize varieties.
3. To evaluate the amount of N, P, K and As content in maize grain and leaf.

CHAPTER II

REVIEW OF LITERATURE

Maize is the third important cereal crop which has received much attention of researchers throughout the world. Various investigators at different contries of the world worked with different maize varieties and arsenic effect. The information available on this subject from different studies by various workers at home and abroad has been reviewed in this chapter with following heading :

2.1Effect of arsenic onmaize

2.1.1Arsenic - a symbol for poison and crime

The word arsenic (As) has made its way through history on the strength of its killing properties. Currently it belongs to the general vocabulary, surrounded by mystery and myth, as a synonym for »toxic«. Since white arsenic (As_2O_3) is odorless and tasteless, it has remained the »king of poisons« for people with evil intentions. In France, the jocose name »poudre de succession«, or inheritance powder, for white arsenic was no laughing matter to the heads of the great families who were inclined to regard all relatives and friends with extreme suspicion (Azcue and Nriagu, 1994). White arsenic sublimes on heating, and it has been claimed that candles with poisoned wicks were used to kill Leopold I of Austria in 1670 (Bagachi, 1969). Until the nineteenth century, white arsenic was the preferred poison of most homicidal practitioners, to the point where laws were passed against the possession of it (Emsley, 1985).

Beside the criminal use of white arsenic, also many cases of accidental arsenic poisoning due to the use of arsenical pigments for coloring artificial flowers, toys, wallpapers, soaps and wrapping paper have been reported. A vast literature discusses the hypothesis that Napoleon's death was due to arsenic containing pigments in his bedroom wallpaper (Bagachi, 1969). Also the death of Clare Boothe Luce, the U.S. Ambassador to Italy might have been due to arsenic poisoning. Supposedly lead arsenate-containing flakes of green paint, falling from the ceiling of the bedroom in the seventeenth-century embassy that she used as a private office, have possibly caused her death (Lenihan, 1988).

2.1.2 Historical and modern use of arsenic compounds

The knowledge about and the first uses of arsenic in antiquity remains a controversial topic. Some authors believe it was not known in antiquity, but there is a strong tendency to support the theory that arsenic was deliberately added to copper alloys in prehistoric times (Brown, 1948). Because of the prevalence of arsenical copper in some countries, Coghlan (1975) suggested an Arsenical-Copper age, rather than the accepted Copper age. The properties of the copper-arsenic alloys were valued by metal smiths in many parts of the world, from those of the Tape Yahya in Iran (fourth millennium BC) to the pre-Columbian Chimu artisans of the Central Andes (Lechtman, 1980). Arsenic was also used in the third millennium BC to produce a silvery surface effect on mirrors and animal statuettes and as one of the fluxing ingredients in the manufacture of glass (Coghlan, 1975). The bright red and yellow colours of the arsenic minerals, mainly realgar (AsS) and orpiment (AS_2S_3), continued to fascinate chemists during antiquity. Mainly Egyptian and Arabic alchemists have studied and tested the recipes previously described by Greek philosophers. Arsenic also played an important role in alchemical operations because of its capability to turn copper white (when red copper oxide is heated with white arsenic oxides it acquires the whiteness of silver); indeed this contributed substantially to the belief that copper can be transmuted into silver (Meyer, 1975).

Although arsenic has widely been used for alloying ores, the main uses of arsenic compounds in antiquity were pharmaceutical and medicinal. The medical use of arsenic dates back to the time of Hippocrates (469-377 BC) who recommended the use of a realgar paste as a treatment for ulcers. At the beginning of the sixteenth century, the revolutionary Paracelsus (1493-1541 AD) designated arsenic as part of the modern pharmacopoeia (Hunter, 1978), following his philosophy that only the dosage makes the poison. In 1786 »Fowler's« solution (1% potassium arsenite) was discovered and became the most widely used medication for a variety of illnesses over the next 150 years. Like »Donovan's« solution (arsenic iodide) and »de Valagin's« solution (arsenic trichloride), it was used to treat rheumatism, arthritis, asthma, malaria, tuberculosis, and diabetes (Leonard, 1991). Finally, the discovery of »Salvarsan« (arsphenamine) in 1909 by Paul Ehrlich, the founder of modern chemotherapy, made it the main medicine against syphilis until the discovery of antibiotics in the early 1940s (Azcue and Nriagu, 1994).

With the findings of other useful properties the use of arsenic increased exponentially in the last 150 years. Being an inexpensive by-product of the smelting of copper, iron, silver, cobalt, lead, gold, manganese, and tin (Leonard, 1991), arsenic became widely used as a pesticide in the wake of the industrial revolution. This usage reached a maximum in the 1950s. Then it was progressively and largely replaced by organochlorine pesticides (Azcue and Nriagu, 1994). Nevertheless, the major use of arsenic today is still in the agricultural field. For industrial purposes, arsenic is primarily used in the form of As trioxide. Industrial uses include the manufacture of ceramics and glass, electronics, pigments and antifouling agents, cosmetics and fireworks (Leonard, 1991). As in the ancient times, arsenic is also still added as a minor constituent to Cu and Cu-based alloys to raise the corrosion resistance of the metals (Azcue and Nriagu, 1994). In agriculture, arsenic remains to be used as a desiccant, rodenticide, and herbicide (Bhumbla and Keefer, 1994).

2.1.3 Agricultural arsenic inputs into soils

Pesticides are the major sources of As in agricultural soils (Jiang and Singh, 1994). Numerous cases of As contamination of agricultural soils due to arsenic containing pesticides have been reported (Merry *et al*., 1986; Peterson *et al*., 1981; Woolson *et al*., 1971a). From the late 1800s until the introduction of dichlorodiphenyltrichloroethane (DDT), lead arsenate ($PbAsO_4$), calcium arsenate ($CaAsO_4$), magnesium arsenate ($MgAsO_4$), zinc arsenate ($ZnAsO_4$), and Paris green [$Cu(CH_3COO)_2$*$3Cu(AsO_2)_2$] were used extensively as pesticides in agriculture

(Anastasia and Kender, 1973; Merry *et al*., 1983). Soil pollution by As pesticides has been extensively reported by Woolson (1975), Merry *et al*., (1983) and Nriagu (1994).

With the introduction of organochlorine pesticides there has been a shift from the inorganic to the organic pesticides (monosodium methylarsonate (MSMA), disodium methylarsonate (DSMA), dimethylarsinic acid (cacodylic acid), and arsenic acid). Due to the essential role of As in animal nutrition, organic arsenicals play an important role as food additives to promote the growth of farm animals (Christen, 2001). In addition they are used as desiccants and defoliants in the cotton industry and for weed control (Woolson, 1975). Despite immense controversy, also arsenic acid is still used as an ingredient of wood preservatives, while sodium arsenite solutions are used for debarking trees, in cattle and sheep dips and in aquatic weed control (Azcue and Nriagu, 1994).

2.1.4 Chronic arsenic poisoning in Bangladesh

The first case of a large-scale health problem caused by arsenic in drinking water was identified 1968 in Taiwan. Scientific interest was initially attracted by the results of an epidemiological study which clearly showed a relationship between high As concentrations in drinking water and the occurrence of skin cancer, keratosis, blackfoot disease as well as cancers of the excretory organs (Chakraborti, 1997; Dhar, 1997). Ever since, many cases of arsenic intoxication have been documented also in India, Vietnam, Inner Mongolia, Greece, Hungary, USA, Thailand, Ghana, Chile, Argentina and Mexico (Smedley and Kinniburgh, 2002). At present, the biggest calamity of arsenic poisoning takes place in Bangladesh.

In the early 1970s, insufficient access to adequate sanitation for a rapidly increasing population in Bangladesh had led to severe microbial contamination of surface water, resulting in high levels of morbidity and mortality. Diarrhoea accounted for 30 % of death in children under five years (Black, 1990). With financial support of the UNICEF and the Government of Bangladesh more than 4 million tubewells were installed between 1980 and 1990, so that the access to pure drinking water from ground water increased from 37 % to 96 % in the rural areas (Kränzlin, 2000). Only when an increasing number of As poisoning cases were reported in the late 1980s, high concentrations of As in a large number of wells were detected. At the time when the wells were installed, arsenic was not known as a problem in drinking water and therefore standard water testing procedures did not include tests for arsenic. Although the release mechanism is not yet fully understood, it appears that the concentration of As in the aquifers are controlled by the reduction of As bearing oxyhydroxide minerals under reducing conditions due to high amounts of organic matter in the aquifer (DPHE/BGS/MML, 1999; Hug *et al.*, 2001).

2.1.5 Effect of arsenic on corp

Naser *et al.,* (2015) carried out in the net house of Soil Science Division of Bangladesh Agricultural Research Institute (BARI), Joydebpur Gazipur on 16 March 2010 and 12 January 2011 with a view of study the effect of P addition to As-contaminated soils and the consequences on As uptake of maize (*Zea mays* L.)

plants. Experiments were conducted in consecutive two years. Arsenic was added to the pots at the rates of 0, 20 and 30 mg kg^{-1}, and P at 0, 30 and 60 mg kg-1. Thus there were seven treatment combinations, i.e., As_0P_0, $As_{20}P_0$, $As_{30}P_0$, $As_{20}P_{30}$, $As_{20}P_{60}$, $As_{30}P_{30}$, and $As_{30}P_{60}$. Phosphorus fertilization increased total As uptake, but the increase was restricted to the root. As concentration of root was much higher than that of shoot. As concentrations in shoot and root were positively correlated (r= 0.913, r = 0.975; P<0.01) in 2010 and 2011, respectively and plant As was positively correlated to the plant P in shoot (r = 0.883 and 0.875; P<0.01) and in root (r = 0.829, P<0.05 and 0.917; P<0.01). The plants took up much greater amounts of P than As. Although it is well known that phosphate inhibits arsenate uptake (Wang *et al*., 2002), but it is highlighted the role of P fertilization to increase As uptake in maize plants. The results presented here indicate P supply may effect in higher As allocation to the plant parts, which has practical application in soil-crop systems. These findings could have important implications for human health and agricultural systems, since it may reduce As contamination through the consumption of crops (phytoextraction) grown on contaminated soils.

This paper examines the influence of Pb^{+2}, Cd^{+2} and As^{+3} on growth of roots in legumes (broad bean, soybean, pea) and cereals (barley, maize) by Piršelová, *et al*., (2015). Roots of germinating plants were exposed to two different levels of Pb^{+2} (300 and 500 $mg^{-1}L^{-1}$), Cd^{+2} (100 and 300 $mg^{-1}L^{-1}$) and As^{+3} (50 and 100$mg^{-1}L^{-1}$) during four day experiment. During this time, length of roots was daily measured. Toxicity of metal treatment on plant roots was calculated as phytotoxicity index

(IP). In all cases, a moderate effect of lead treatment was observed (IP up to 56.67 %) while higher doses of cadmium and arsenic resulted in increase of IP above 50 %. In cases of barley and maize, the toxic effect of almost all test doses of the heavy metals was observed as soon as 24 hours after their application. Generally, a higher tolerance to tested metals showed roots of both bean cultivars (IP 16.27-69.53 %), while the most sensitive reactions had roots of barley and soybean (IP > 50 %, excluding dose Pb 300$mg^{-1}L^{-1}$).

Mehmood*et al.*, (2017) to study explores the role of compost addition (0, 1 and 2.5%) on morphological and gas exchange attributes and photosynthetic pigments (chlorophyll contents) of maize plants under As stress (0, 40, 80, 120 mg kg^{-1}), as well as soil As immobilization/mobilization in a pot experiment, using two contrasting soils. Results revealed that, in Narwala (sandy loam) soil, the addition of compost decreased shoot As concentration of maize plants ($p < 0.05$; 4.01–13.7 mg kg^{-1} dry weight (DW), notably at $C_{2.5}$ treatment, with significant improvement in shoot dry biomass, gas exchange attributes and chlorophyll (*a* and *b*) contents, i.e., 1.33-1.82, 1.20-2.65 and 1.34-1.66 times higher, respectively, over C_0 at all As levels. Contrastingly, in Shahkot (clay loam) soil, $C_{2.5}$ treatment increased shoot As concentration ($p < 0.05$; 7.02–17.3 mg kg^{-1} DW), and as such reduced the shoot dry biomass, gas exchange attributes and chlorophyll contents, compared to the control rather C_1 treatment was more effective and exhibited positive effect than $C_{2.5}$. Considerably, at $C_{2.5}$ treatment, phosphate extractable (bioavailable) soil As concentration was also found to be greater in the (postexperiment) Shahkot soil than that of Narwala soil (0.40-3.82 vs. 0.19-1.51 mg kg^{-1}, respectively).

The effects of different levels of kinetin (KT) application on the growth, biomass, contents of chlorophyll (Chl a, Chl b, and carotenoid), arsenic uptake, and activities of antioxidant enzymes in maize seedlings under arsenic (As) stress were investigated by a hydroponic experiment. The results showed that KT supplementation increased the biomass in terms of root length, root number, fresh weight, and seedling length, and KT treatments also improved the contents of Chl a, As uptake, and Chl a : b ratio compared to cases with As treatment alone. However, no significant changes were observed in carotenoid content, and a reduction was found in Chl b content of seedlings. KT also increased the activities of catalase (CAT), peroxidase (POD), and superoxide dismutase (SOD) in the leaves of maize seedlings when 0.1 mgL^{-1}KT and As were applied, which decreased the content of malondialdehyde (MDA). These results suggested that KT could alleviate the toxicity of As to maize seedlings by keeping the stability of chlorophyll, enhancing the activities of antioxidant enzymes, and inhibiting the lipid peroxidation (Wang *et al.*, 2015).

Silva *et al.*, (2015) studied to investigate the effects of Si in alleviating As stress in maize plants grown in a nutrient solution and evaluate the potential of the spectral emission parameters and the red fluorescence (Fr) and far-red fluorescence (FFr) ratio obtained in analysis of chlorophyll fluorescence in determination of this interaction. An experiment was carried out in a nutrient solution containing a toxic rate of As (68 µmolL^{-1}) and six increasing rates of Si (0, 0.25, 0.5, 1.0, 1.5, and 2.0 mmolL^{-1}). Dry matter production and concentrations of As, Si, and photosynthetic pigments were then evaluated. Chlorophyll fluorescence was also measured thorough out plant growth. Si has positive effects in alleviating As stress in maize plants, evidenced by the increase in photosynthetic pigments. Silicon application resulted in higher As levels in plant tissue; therefore, using Si for soil phytoremediation may be a promising choice. Chlorophyll fluorescence analysis proved to be a sensitive tool, and it can be successfully used in the study of the ameliorating effects of Si in plant protection, with the Fr/FFr ratio as the variable recommended for identification of temporal changes in plants.

2.2 Effect of variety on the growth and yield of maize

Biswas *et al.,* (2014) to study the effect of planting geometry on yield and yield attributes of maize hybrids an experiment was conducted at the Regional Agricultural Research Station, Jamalpur during rabi 2009-2010 and 2010-2011. The results revealed that there was significant variation among the planting geometry during both years. There was no significant difference among the varieties in the first year but in the second year. The highest grain yield was obtained from the planting geometry 60cm × 20cm (83,333 plants m^{-2}) which was statistically similar to that of 75cm × 20cm (66,666 plants m^{-2}). The lowest grain yield was obtained from the planting geometry 75cm × 25cm (53,333 plants m^{-2}). Pacific-11 showed better performance during both years than the varieties BARI hybrid bhutta-5 and BARI hybrid bhutta-7. Significant variation was not found due to interaction effect of variety and planting geometry.

Enujeke (2013a) carried out in Teaching and Research Farm of Delta State University, Asaba Campus from March, 2008 to June, 2010 to evaluate the effects of variety and spacing on growth characters of hybrid maize. Three hybrid maize varieties were evaluated under three different plant spacing for such growth characters as plant height, number of leaves, leaf area and stem girth. The results obtained during the 8th week after sowing indicated that hybrid variety 9022-13 which had mean plant height of 170.0cm number of leaves of 13.2, leaf area of 673.2cm^2 and stem girth of 99.4mm was superior to other varieties investigated. Based on the findings of this study, it is recommended that (i) hybrid variety 9022-13 be grown in the study area of enhanced growth characters which interplay to improve grain yield of maize (ii) spacing of 75 cm x 35 cm be used to enhance increased stem girth and leaf area whose photosynthetic activities could positively influence maize yield.

Enujeke (2013b) carried out in the Teaching and Research Farm of Delta State University, Asaba Campus (Nigeria) from March to December in 2008 and replicated between March and December 2009, to evaluate the effects of variety and spacing on yield indices of Open-pollinated maize. It was a factorial experiment carried out in a Randomized Complete Block Design (RCBD) with four replicates. The results obtained indicated that variety BR9922-DMRSF2 was outstanding with number of cobs/plant of 1.7 in both 2008 and 2009, cob length of

27.7 cm and 26.7 cm in 2008 and 2009 respectively. Its grain weight was 4.7 t/ha in 2008 and 4.9 t/ha in 2009 and its number of grain/cob was 467.7 in 2008, and 463.9 in 2009. Their grain weights (t/ha) were 5.0 in 2008 and 5.2 in 2009, their cob lengths were 18.6cm in 2008 and 20.1cm in 2009, while their number of grains/cob were 363.0 in 2008 and 369.0 in 2009. The results of interaction showed that except variety x space, the parameters investigated were all significantly ($P < 0.05$) different and affected yield indices of open-pollinated maize. Based on the findings of the study, it was recommended that (i) open-pollinated variety BR9922-DMRF2 be grown in the study area for increased grain yield indices of maize. Plant spacing of 75 cm x 15 cm which resulted in higher number of cobs/plant and higher grain weight be adopted in maize production.

Shinggu *et al.*, (2009) conducted at the Institute for Agricultural Research farm, Samaru (11011 N; 07038' E and 686m above sea level) in the Northern Guinea Savanna ecological Zone of Nigeria during the wet season of 2000 and 2001. Extra early maize TZEE-W was used as test crop, two cowpea varieties (Kanannado and Sampea 7): two crop arrangements (alternate row and alternate stand arrangements) and ten periods of weed interference (weed free till 3, 6, 9, 12 weeks after sowing (WAS) and harvest and a corresponding set that were kept initially weed infested till 3,6,9,12 WAS and harvest. Two treatments were left weed free or weed infested throughout the crop life cycle. The treatments were evaluated in a split-plot design with varieties and crop arrangements allotted to main plot and period of weed interference to sub-plot. Varieties, crops arrangement and period of weed interference had significant effect on weed growth and yield parameters of maize. Maize grown in mixture with Kanannado gave lower weed dry matter (WDM), higher crop vigour score (CVS), higher grain yield and 100-grain weight. Maize in alternate row arrangement performs better than maize in alternate stand arrangement. Keeping the crop weed free till 6 WAS and beyond gave better crop performance.

Iptas and Acar (2006) conducted to determine the effect of row spacing (40, 60 and 80 cm) on forage dry matter (DM) yieldand quality of four hybrids grown in the years 2001 and 2002. The highest DM yield was obtained from the Arifiye(24.1 and 22.4 t/ha) while the lowest DM yield was obtained from Pioneer 3163 (19.9 and 19.8 t/ha) in the years 2001 and 2002 respectively. As row spacing increased, DM yield as an average of two years decreased from 27.2 to 16.6 t/ha. No differences were found among row spacing for DM content, harvest index (HI) and ear content. Asrow spacing increased, whole-plant acid detergent fiber (ADF) and neutral detergent fiber (NDF) content increasedfrom 214 to 227 g/kg and from 420 to 451 g/kg during the year 2001 respectively. However, ADF content decree-sed from 281 to 267 g/kg and NDF contents decreased from 530 to 515 g/kg with increasing row spacing duringthe year 2002. In this study, hybrids showed distinct differences for crude protein, ADF and NDF contents in bothyears. Forage quality parameter including ADF and NDF of Pioneer 3163, TTM 8119 and Karadeniz Yildizi were higher than Arifiye hybrid.

CHAPTER III

MATERIALS AND METHODS

The experiment was undertaken in November 2016 to March 2017 in the net house and laboratory at the department of Agricultural Chemistry,Sher-e-Bangla Agricultural University, Dhaka, Bangladesh to determine the effect of arsenic on growth, yield and arsenic content in leaf and grain of maize (*Zea mays*). The materials and methods followed during entire period of the experiment are described in this chapter.

3.1 Site of the experiment

The experiment was conducted at the nethouseand laboratory department of Agricultural Chemistry ,Sher-e-Bangla Agricultural University, Dhaka.

3.2 Experimental period

The experiment was conducted in pots during 15th November 2016 to 15th March 2017.

3.3 Materials

3.3.1 Seed

Two varieties were used in the study. Two varieties ‘BARI maize -7’ and Pacific 339 were used as plant material which were collected from Bangladesh Agricultural Research Institute (BARI); Joydebpur, Gazipur and different local market respectively.

3.3.2 Fertilizers

The amount of urea , TSP, MOP, Gypsum and compost required for each pot were calculated as per as their rates of application. This recommended dose were given as necessary for per pot. The whole amount of TSP, MOP, Gypsum and $1/4^{th}$ of urea were applied before the final preparation of the pots. There after the pots containing soil were moistened with water. One-half of urea were given at knee high stage (25-30 days after germination) and rest of the urea were given before emergence of tassel.

3.4 Methods

3.4.1 Treatments

Two factors were used as combination for 10 treatments. Two varieties and five levels of arsenic were used for the combination of ten (10) treatments of the present experiment.

Factor A: variety

Treatments : 2

(i) V_1 = BARI maize 7
(ii) V_2 = Pacific 339

Factor B:level of arsenic

Treatments : 4

(i) T_0 = 0 mg As/kg soil
(ii) T_1 = 20 mg As/kg soil
(iii) T_2 = 40mg As/kg soil
(iv) T_3 = 60mg As/kg soil
(V) T_4 = 80mg As/kg soil

3.4.2 Experimental design and layout

The experiment was set in Completely Randomized Design (CRD) having two factors with three replications. The two varieties in combination with five arsenic levels were randomly assigned to 30(5×2×3) experimental units per pot.

3.4.3 Collection and preparation of soil

The soils of the experiment were collected from Sher-e-Bangla Agricultural University (SAU) farm. The soil was non-calcarious red brown terrace soil with loamy texture belonging to the AEZ 28 (Madhupur Tract). The collected soil was pulverized and inert materials, visible insect pest and plant propagules were removed. The soil was dried in the sun, crushed carefully and thoroughly mixed.

3.4.4 Pot preparation

An amount of 8 kg soil was taken in a series of pots. The required number of plastic pots having 24 cm top, 18 cm bottom diameter and 22 cm depth were collected from the local market and cleaned before use. There were altogether 30 pots comprising 5 different treatments to two maize varieties with 3 replications. Water was added to the pot to bring the soil up to saturation.

3.4.5 Sowing of seeds

Sowing was done on 15th November 2016. One seed was sown per pot .

3.4.6 Irrigation

Water given when necessary to maintained the soil moisture at zoe condition .

3.5 Crop sampling and data collection

One plants was selected in each plot and marked with tag for recording plant characters.

3.6 Harvesting and threshing

Crops were harvested when 90% of the cob became golden in color. The matured cob was harvested and the harvested cobs were carried to the threshing floor. The cob was sun dried by spreading on the threshing floor. Seeds were then separated from the cobs.

3.7 Drying and weighing

Seeds and stovers thus collected were dried in the sun for a couple of days. Dried seeds and stovers of each plot was weighed.

3.8 Data collection

At harvesting, each plant was selected from all pot to record the following data.

i. Plant height (cm)

ii. Number of leaves per plant

iv. Total dry mater

v. Number of cob per plant

vi. Number of grains per cob

vii. Weight of grains per plant

3.9 Procedure of recording data

3.9.1 Plant height (cm)

Height of each plant was measured from ground level (stem base) to the tip of the plant. Mean plant height was calculated and expressed in cm.

3.9.2 Number of leaves per plant

Number of leaves of each selected plant was counted and recorded.

3.9.3 Total dry matter

Total dry mater of plant at harvest was calculated by aggregating the dry matter weight of leaves, stems, roots, cob and other immature reproductive parts.

3.9.4 Number of cobs per plant

Number of cobs per plant was counted from the eachselected plant and then the average cob number was calculated.

3.9.5 Number of grain per plant

Number of grainper plant was counted from 2/3randomly selected cobs and then the average grain number was calculatedin each cob thereafter per plant.

3.9.6 Grain yield per plant

The grain of each plant was weighed with a digital electric balance. The grain weight was recorded in gram.

3.10 Chemical analysis

3.10.1 Collection and preparation of plant samples

Leaf, stem and grain and straw samples were collected after threshing for N, P, K and As analyses. The plant samples were dried in an oven at 70^0C for 72 hours and then ground by a grinding machine (wiley-mill) to pass through a 6-mesh sieve. The samples were stored in plastic vial for analyses of N, P, K and As. The leaf, stem and grain samples were analyzed for determination of N, P, K and As concentrations. The methods were as follows:

3.10.2 Digestion of plant samples for N determination

For the determination of nitrogen exactly 0.5 g oven dry, ground sample were taken in a macrokjeldahl flask. 1.1 g catalyst mixture (K_2SO_4: $CuSO_4$. $5H_2O$: Se in the ratio of 100: 10: 1), and 10 ml conc. H_2SO_4 were added. The flasks were heated at 160^0C and added 2 ml 30% H_2O_2 then heating was continued at 360^0C until the digests become clear and colorless. After cooling, the content was taken into a 100 ml volumetric flask and the volume was made up to the mark with distilled water. A reagent blank was prepared in a similar manner. Nitrogen in the digest was estimated by distilling the digest with 40 %NaOH followed by titration of the distillate trapped in H_3BO_3 indicator solution with 0.01N H_2SO_4.

3.10.3 Digestionof plant sample for P and K determination

Exactly 1g oven-dried samples of maize plant were taken in digestion tube. About 10 mL of Di-acid mixture (conc. HNO_3 and 60% $HClO_4$) in a digestion tube and left to stand for 20 minutes and then transferred to a digestion block and continued heating at 100 ^{0}C. The temperature was increased to 365 ^{0}C gradually to prevent frothing (50 ^{0}C steps) and left to digest until yellowish color of the solution turned to whitish color. Then the digestion tubes were removed from the heating source and allowed to cool to room temperature. About 40 mL of distilled water was carefully added to the digestion tubes and the contents filtered through Whatman no. 40 filter paper into a 100 mL volumetric flask and the volume was made up to the mark with distilled water. The samples were stored at room temperature in clearly marked containers.

3.10.4 Determination of potassium

The amount of potassium (K) was estimated from the sample with the help of flame photometer .

3.10.5 Determination of phosphorus

The amount of phosphoru**s** (P) was estimated from the sample with the help of spectrophotometer.

3.10.6 Determination of arsenic (As)

Total arsenic concentration was determined from the digest by flame atomic absorption spectrophotometer with HVG (Hydride Vapour Generator).

Sample Information: Digest Sample, pH<2 with HCl 5mL/L

Sample Storage: Refrigerator, temperature <4^{o}C

Method detection Limit:0.1 ppb

Instrument: Flame Atomic Absorption Spectrophotometer with HVG. Ar gas (purity 99.999%) as carrier of sample. HCl 5M and 0.4% $NaBH_4$ as reagent for HVG. Sample flow rate 5 mL/min.

Reagent used:

(i) KI (ii) Conc. HNO_3 (iii) Conc. HCl (iv) De-Ionized Water (DI Water) (v) 1000 ppm Standard Solution of As (vi) $NaBH_4$. (vii) 5MHCl

Preparation of Reagents:

Preparation of $NaBH_4$ Solution:

2.5 g Sodium Hydroxide (Merck, Germany) and 2.0 g Sodium Borohydrate (Sigma-Alorich, USA) were dissolved in 500 mL vol. flask and marked up to volume with DI water.

Preparation of KI

20g KI (Merck, Germany) was taken in 100 mL water then dissolved in waterandmarked up to volume.

Preparation of 5MHCl:

Around 200 mL of DI water was taken in a 500 mL volumetric flask then added 208 mL ofHCl (37%) then marked up to volume with DI water.

Treatment and Preparation of Sample:

0.5 to 1.0 g of well-mixed sample was transferred to a beaker. 10 mL Conc. HNO_3 was added . The sample was covered with a watch glass and heated on hot plate at 90^{0} to 95^{0}C until the volume reduced to 15-20 mL. The beaker was removed and allowed cooling. The beaker walls was washed down and watch glass with DI water when necessary filter or centrifuge the sample to remove silicates or other insoluble material. Then it was made final volume to 50 mL with the diluent. After that was taken 40 mL of this in 50 mL volumetrics flask and added 4 mL of 37 % HCl and 2 mL of freshly prepared 20% (w/v) KI to it and left to dark for 15 minutes.

3.11 Statistical analysis

The data obtained for different parameters were statistically analyzed to find out the significant difference between the results of growth, yield and yield contributing characters of maize. The mean values of all the characters were calculated and analysis of variance was performed by the 'F' (variance ratio) test. The significance of the difference among the treatment means was estimated by the Duncan's Multiple Range Test (DMRT) at 5% level of probability (Gomez and Gomez, 1984).

CHAPTER IV
RESULTS AND DISCUSSION

The experiment was conducted to determine effect of arsenic growth, yield and arsenic content in leaf and grain of maize. Data on different parameters were analyzed statistically. The result of the present study have been presented and discussed in this chapter under the following headings.

4.1 Plant height

Plant height of the varieties was measured at maturity. It was evident from figure 1 that the height of the plant was influenced by variety. The taller plant (144.07 cm) was found in Pacific 339 (V_2) and the shorter (137.383 cm) plant was in BARI maize 7 (V_1) at harvest probably the genetic makeup of varieties was responsible for the variation in plant height. This confirms the reports of Shamsuddin *et al.,* (1988) that plant height differed due to varietal variation.

The height of the plant was significantly influenced by arsenic. At harvest, the highest (147.3 cm) plant height were observed in 0 mg As/kg soil and the lowest (136.7 cm) values were found in 80mg As/kg soil (T_4), which was statistically similar with (T_2) and (T_3) (Fig. 2). All the growth parameters tested in their experiment viz. plant height were affected by the application of As.

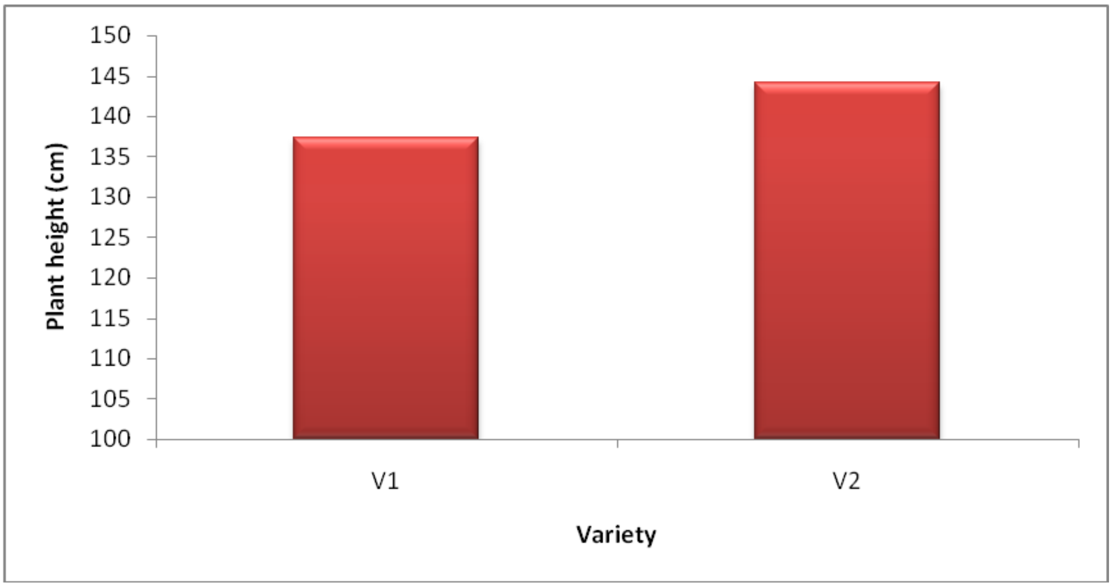

Fig. 1. Effect of variety on the plant height of maize

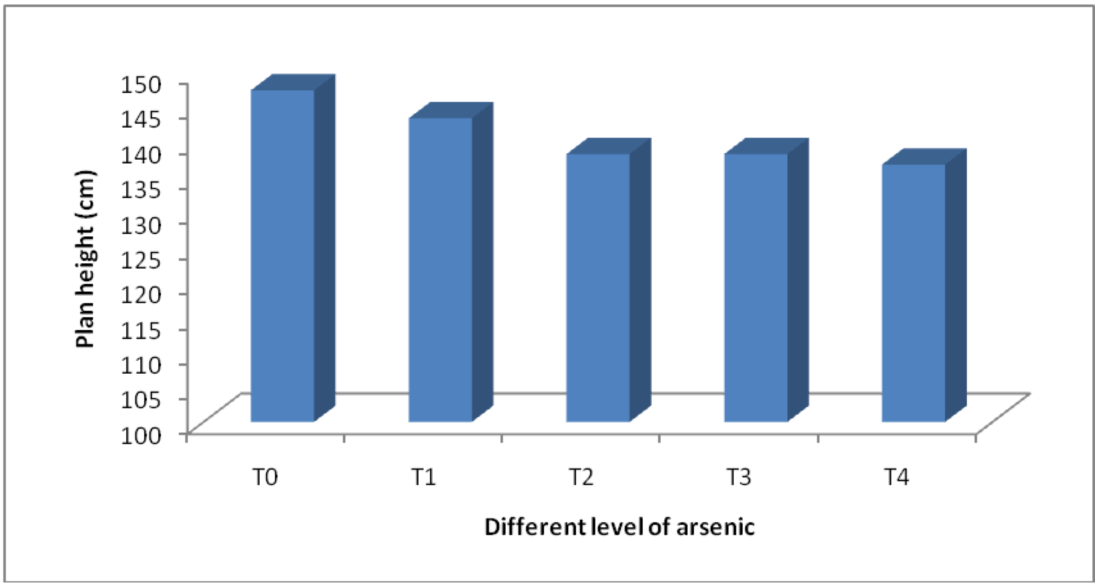

Fig. 2. Effect of different levels of arsenic on the plant height of maize

Table 1. Combined effect of variety and different arsenic levels on the growth character of maize

Treatment	Plant height (cm)		Number of leaf per plant		Total dry mater (g)	
V_1T_0	140.20	bc	15.77	a	230.10	b
V_1T_1	141.20	bc	13.67	ab	219.30	bc
V_1T_2	136.20	c	14.33	ab	207.20	bcd
V_1T_3	135.10	c	13.33	bc	158.70	bcd
V_1T_4	134.10	c	11.67	cd	144.90	d
V_2T_0	153.40	a	15.67	a	302.50	a
V_2T_1	150.40	ab	13.67	b	219.60	bc
V_2T_2	142.20	bc	14.33	ab	209.50	bcd
V_2T_3	138.20	c	13.00	bcd	193.70	bcd
V_2T_4	136.10	c	11.33	d	154.30	cd
$LSD_{(0.05)}$	9.61		1.79		64.85	
CV(%)	6.02		7.67		8.79	

LSD = Least of significant difference, CV = Coefficient of varience

In combination effect of variety and arsenic levels was significantly influenced on plant height. The plant height of different maize varieties significantly decreased with increasing in different arsenic levels (Table 1). The highest (153.40 cm) plant height was found in Pacific 339 with 0 mg As/kg of soil (V_2T_0) and the lowest (134.10 cm) plant height was found in BARI maize 7with 80mg As/kg of soil(V_1T_4) level of arsenic.

4. 2 Number of leaf per plant

The number of leaf per plant was influenced by variety at all stages of crop growth. Varietal effects on the formation of total number of leaves are shown in Figure 3. BARI maize 7 was achieved maximum leaves per plant (13.73), where as the minimum leaves per plant (13.6) production was observed in Pacific 339 (V_2) during harvest. Variable effect of variety on number of leaf per plant was also reported by Hussain *et al.*, (1989) who noticed that number of leaf per plant differed among the varieties.

Number of leaves per plant was influenced by different arsenic levels (Fig.4). The maximum number of leaf per plant (15.67) was produced from 0 mg As/kg of soil(T_0) and the minimum total number of leaf per plant (11.50) was produced form 80 mg As/kg of soil(T_4) treatment.

The combined effect of varieties and different arsenic levels were statistically significant (Table 1). At harvest the maximum number of leaf per plant(15.77) was found from BARI maize 7 with 0 mg As/kg of soil (V_1T_0), which was statistically similar with(V_2T_0) and minimum number of leaf per plant(11.33) from Pacific 339 with 80 mg As/kg of soil (V_2T_4).

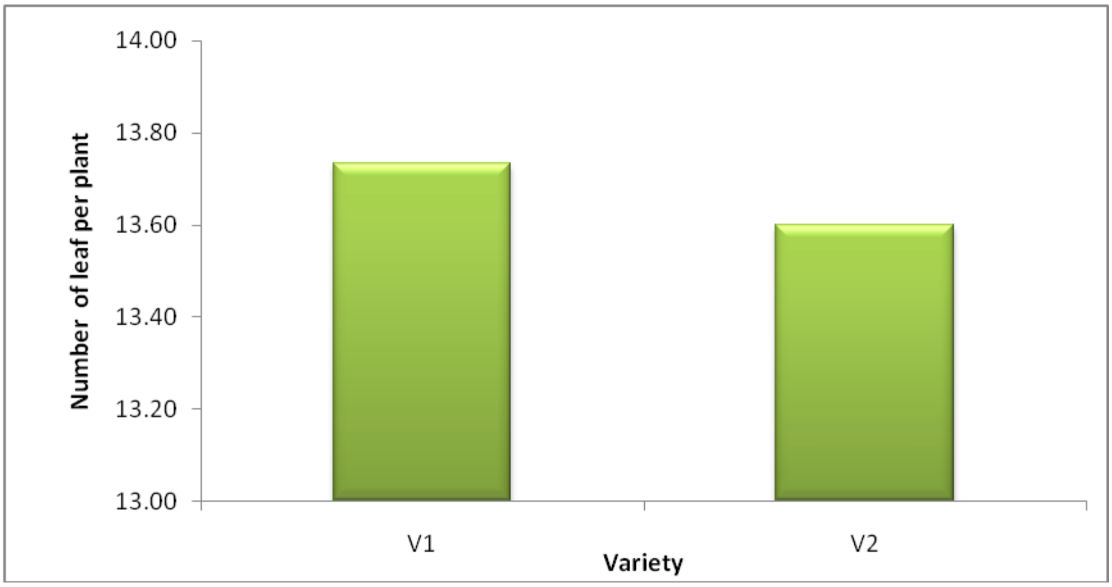

Fig. 3. Effect of variety on the number leaf per plant of maize

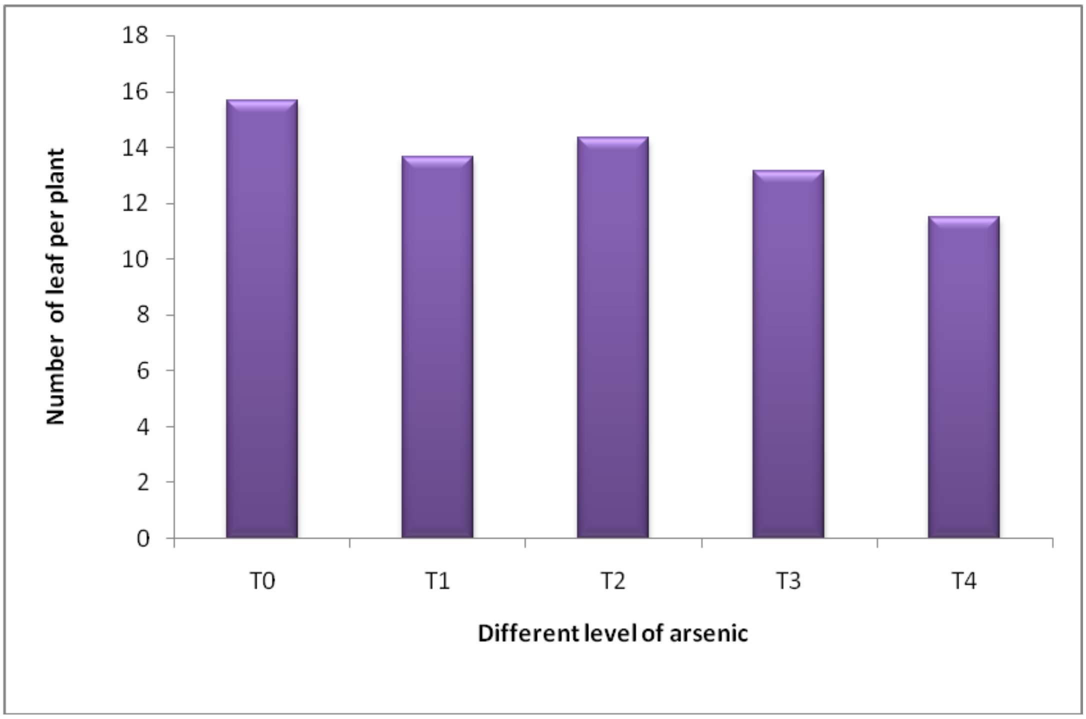

Fig. 4. Effect of different arsenic levels on the number of leaf per plant of maize

4.3 Total dry matter (g)

The Total dry matter was influenced by variety. Among the two maize varieties the highest total dry mater (TDM) (215.94 g plant^{-1}) was recorded in V_2, whereas the lowest TDM (192.03 g plant^{-1}) was in V_1 (Fig. 5).

The result presented in Fig.6 shows that the TDM significantly decreased with increasing the arsenic levels. The TDM was significantly influenced by the arsenic levels. The TDM was highest (254.94 g) at 0 mg As/kg of soil(T_0) and it was lowest (149.60 g) at 80 mg As/kg of soil level of soil arsenic.

The effect of different arsenic levels on TDM of selected maize varieties differed significantly. The highest TDM (302.5 g) was found in BARI maize 7 at 0 mg As/kg of soil and the lowest value (144.9 g) was in Pacific 339 with 80 mg As/kg of soil(Table 1).

4.4 Number of cob per plant

Different varieties show significant variation in number of cob per plant. The number ofcob per plant (0.87) was highest in BARI maize 7 and that was lowest (0.73) in Pacific 339(Table 2). BRRI (1994) found that number of cob per plant significantly differed due to variety.

The number ofcob per plantwas significantly influenced by different arsenic levels (Table 2). The highest number ofcob per plant (1.42) was recorded at 0 mg As/kg of soil and the lowest (0.42) was found at 80 mg As/kg of soil arsenic level, which was statistically similar with 60 mg As/kg of soil arsenic level.

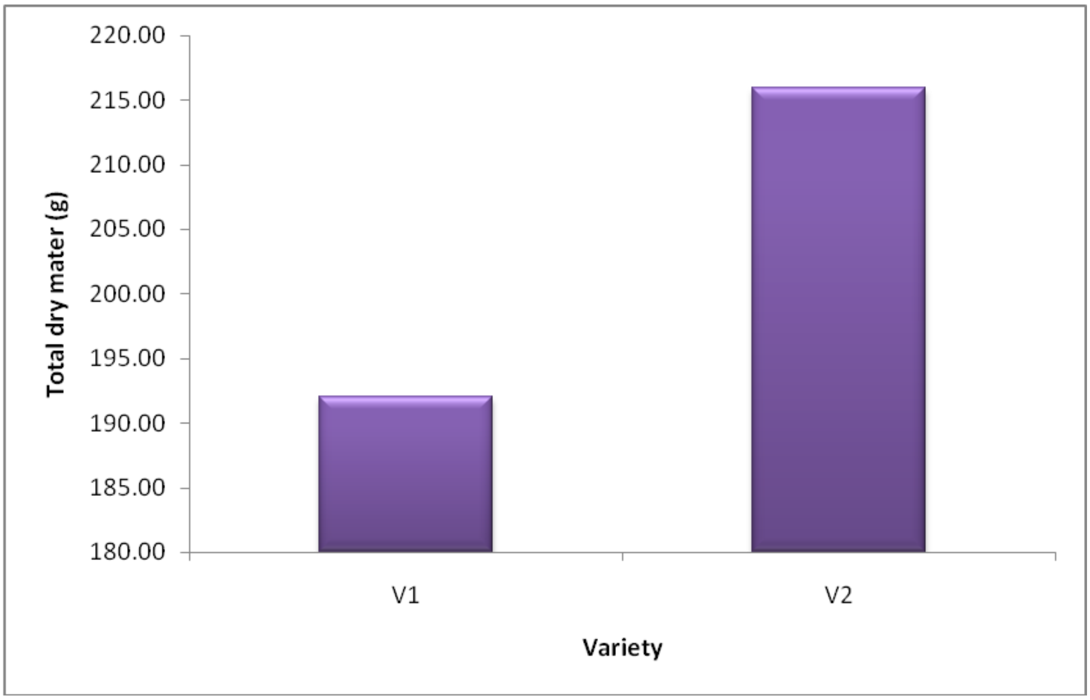

Fig. 5. Effect of variety on the total dry mater of maize

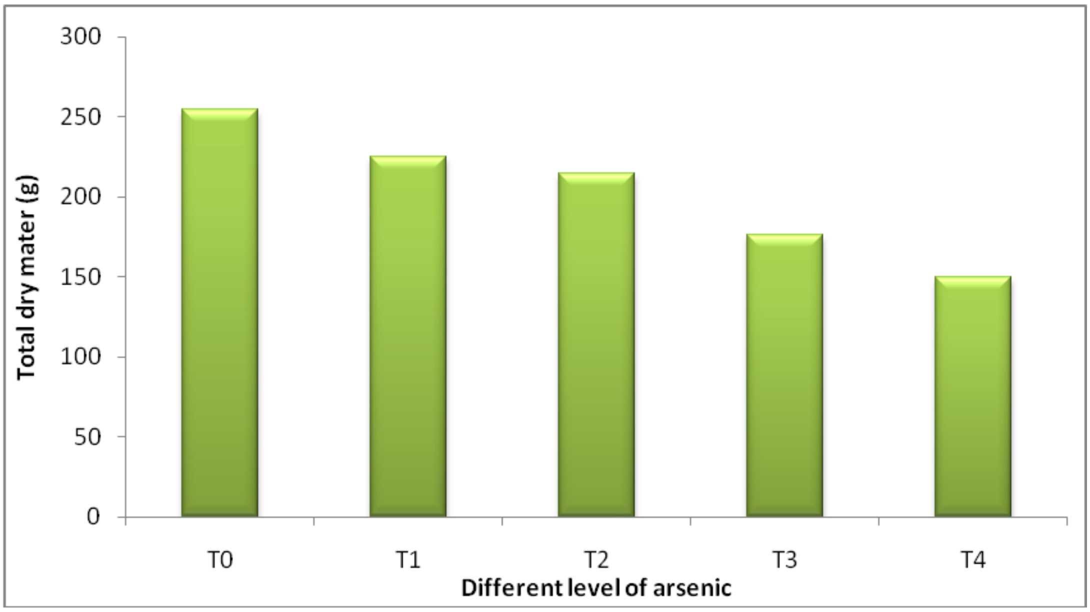

Fig. 6. Effect of different arsenic levels on the total dry mater of maize

Table2. Effect of variety, different level of arsenic and their interaction on the yield contributing characters of maize

Treatment	Cob per plant		Number of grain per plant		Grain weight per plant (g)	
Effect of variety						
V_1	0.87		111.27		31.77	
V_2	0.73		91.47		26.32	
CV(%)	5.72		12.45		5.77	
Effect of arsenic						
T_0	1.42	a	207.00	a	58.54	a
T_1	0.83	ab	113.30	b	32.76	b
T_2	0.83	ab	91.33	b	28.13	bc
T_3	0.50	b	51.17	c	12.88	c
T_4	0.42	b	44.00	c	12.17	c
$LSD_{(0.05)}$	0.89		38.08		19.15	
CV(%)	5.72		12.45		5.77	
Interaction effect of variety and arsenic						
V_1T_0	1.50	a	226.00	a	63.75	a
V_1T_1	0.83	ab	117.00	b	33.89	b
V_1T_2	0.67	ab	74.00	bc	24.43	bc
V_1T_3	0.67	ab	70.67	bc	19.43	bc
V_1T_4	0.67	ab	68.67	bc	17.33	bc
V_2T_0	1.33	a	188.00	a	53.33	a
V_2T_1	0.83	ab	109.70	b	31.63	b
V_2T_2	1.00	ab	108.70	b	31.83	b
V_2T_3	0.47	b	31.67	c	8.45	c
V_2T_4	0.33	b	19.33	c	6.33	c
$LSD_{(0.05)}$	0.75		56.03		17.20	
CV(%)	5.72		12.45		5.77	

LSD = Least of significant difference, CV = Coefficient of varience

The cob per plantof two selected maize genotypes was significantly influenced by different arsenic levels (Table 2). At 0 mg As/kg of soil level of arsenic, the maximum cob per plant (1.50) were found in BARI maize 7, which was statistically similar with V_2T_0 (Pacific 339at 0 mg As/kg of soil) and that was minimum (0.33) also in Pacific 339 at 80 mg As/kg of soil, which was statistically similar with V_2T_3 (Pacific 339at 60 mg As/kg of soil).

4.5 Number of grain per plant

Results showed that variety had significant effect in respect of the number of grain per plant (Table 2). BARI maize 7 produced maximum number (111.27) of grain per plant and Pacific 339produced minimum number (91.47) of grain per plant.This variation might be due to genetic characteristics. BRRI (1994) found that number of filled grains panicle^{-1} significantly differed due to variety.

It is revealed from the results that the number of grainplant^{-1} differed significantly due to different arsenic levels. Statistically the highest number of grain plant^{-1}(207.00)was recorded at 0 mg As/kg of soil levels of arsenic and it was the least at 80 mg As/kg of soil levels of arsenic (Table 2).

Interaction effect of varieties and different arsenic levels showed significant response on grain per plant (Table 2). The maximum number of grain plant^{-1} (226.00) was found in BARI maize 7 at 0 mg As/kg of soil and the minimum number of grainplant^{-1}(19.33) was recorded in Pacific 339 at 80 mg As/kg of soil level of arsenic (Table 6). The sterility and significant reduction in seed setting in maize were assumed to be not merely due to reduction or inhibition of different biochemical constituents and physiological functions, but were also due to limitation of soluble carbohydrate translocation in cob, accumulation of more As and less K^+ in all the floral parts, and highly significant inhibition of specific activity of starch synthetase in developing maize grains.

4.6 Grain yield per plant

Grain yield is a function of interplay of various yield components such as number of grainplant^{-1} and 1000grain weight (Hassan *et al.*, 2003). The grain yield plant^{-1} of two selected maize varieties differed due to the mean effect of different arsenic treatments (Table 2). The highest grain yield plant^{-1} (31.77 g) was found in varietyBARI maize 7 and the lowest yield (26.32 g) was recorded in Pacific 339. Grain yield differences due to varieties were reported by Suprithatno and Sutaryo (1992), Alam (1998) and IRRI (1978) who recorded variable grain yield among tested varieties.

A highly significant variation in grain yield plant^{-1} of maize varieties was observed due to the different arsenic levels (Table 2)**.** The highest grain yield plant^{-1}(58.54 g) was recorded at control treatment and it was lowest (12.17 g) at 80 mgAs/kg of soil level of arsenic. Grain yield decreased with increased in arsenic levels. It clearly indicates the poisonous and detrimental effect of arsenic on plant. This result agreed with Hossain*et al.*, (2005) who found that yield reductions on more than 40 and 60 % for two popular rice varieties (BRRI dhan-28 and Iratom-24) when 20 mg/kg of arsenic was added to soils, compared to the control.

It was evident from the table 2 that interaction of variety and different arsenic levels significantly affected the grain yield. The highest grain yield plant^{-1} (63.75 g) was found in BARI maize 7 at 0 mg As/kg of soil arsenic level, which was statistically similar with Pacific 339 at 0 mg As/kg of soil arsenic leveland the lowest yield (6.33 g) was obtained in Pacific 339 at 80 mg As/kg of soil arsenic level, which was statistically similar with Pacific 339 at 60 mg As/kg of soil arsenic level. Grain yield is the function of number of cobplant^{-1}, number of grain plant^{1}. All the yield contributing characters contributed for the yield reduction perplant under arsenic conditions; contribution of the seriously affected number of cob per plant was the highest.

4.7 Nitrogen content in leaf

The percent content of nitrogen (N) in leaf of the entire two selected maize varieties varied. The N content in leaf washighest (0.87 %) in V_1 and lowest (0.86 %) in V_2 (Table 3).

The nitrogen (N) content in leaf of maize significantly varied due to the effect of different arsenic levels; wherethe N content in leaf decreased with the increasing level of arsenic in soil.The highest N content (0.99%) in leaf was recorded in 0 mg As/kg of soil level of arsenic which is statistically identical with 20 mg As/kg of soil level of arsenic and then was lowest (0.70%) in 80 mg As/kg of soil(Table 3).

The combined effect of arsenic and variety on N (%) in leaf was found significant. The N content decreased with the increasing levels of arsenic in both leaf of all varieties (Table 3). The highest N content (1.00 %) in leaf was found in BARI maize 7 at 0 mg As/kg of soil and it was lowest (0.68 %) in the varietyPacific 339 at the 80 mg As/kg of soil arsenic level, which was statistically similar with V_2T_3.

Table3. Effect of variety, different level of arsenic and their interaction on the N, P, K and As content in leaf of maize

Treatment	N (%)		K (%)		P (%)		As (ppm)	
Effect of variety								
V_1	0.87		1.88		0.20		2.40	
V_2	0.86		1.84		0.14		2.41	
CV(%)	7.33		8.73		8.48		6.59	
Effect of arsenic								
T_0	0.99	a	2.22	a	0.24	a	0.00	e
T_1	0.97	a	2.14	ab	0.22	a	1.14	d
T_2	0.89	b	1.93	ab	0.17	a	1.85	c
T_3	0.77	c	1.61	ab	0.13	a	3.18	b
T_4	0.70	d	1.42	b	0.10	a	5.87	a
$LSD_{(0.05)}$	0.07		0.72		0.34		0.10	
CV(%)	7.33		8.73		8.48		6.59	
Interaction effect of variety and arsenic								
V_1T_0	1.00	a	2.25	a	0.36	a	0.00	e
V_1T_1	0.98	ab	2.18	ab	0.22	bc	1.13	d
V_1T_2	0.88	bc	1.90	c	0.18	bcd	1.88	c
V_1T_3	0.78	cd	1.63	d	0.14	cd	3.16	b
V_1T_4	0.71	d	1.45	e	0.10	d	5.85	a
V_2T_0	0.99	ab	2.19	ab	0.26	b	0.00	e
V_2T_1	0.96	ab	2.10	b	0.15	cd	1.14	d
V_2T_2	0.91	ab	1.96	c	0.12	cd	1.83	c
V_2T_3	0.76	d	1.58	d	0.10	d	3.19	b
V_2T_4	0.68	d	1.39	e	0.09	d	5.89	a
$LSD_{(0.05)}$	0.11		0.12		0.09		0.27	
CV(%)	7.33		8.73		8.48		6.59	

LSD = Least of significant difference, CV = Coefficient of varience

Potassium content in leaf

It appears from the results presented in Table 3 that there was a significant variation in K content intwo selected maize varieties under mean effect of different arsenic levels. The highest K content in leaf was found in Pacific 339 (1.88%) and that was lowest (1.84%) in BARI maize 7.

The K contents in leaf of maize also significantly varied due to the effect of different arsenic levels; wherethe K content decreased with the increasing level of arsenic in leaf (Table 3).The highest K content in leaf(2.22%) was recorded in 0 mg As/kg of soil and it was lowest (1.42%) in 80 mg As/kg of soil arsenic level.

The combined effects of arsenic and variety on K (%) in leaf were differed significantly. The content of K in leaf of all the selected varieties progressively decreased with increasing the arsenic levels. The highest K content (2.25%) in leaf was found in Pacific 339 at 0 mg As/kg of soil and it was lowest (1.39%) in the BARI maize 7 variety at the 80 mg As/kg of soil arsenic level, which was statistically identical with V_2T_4 (Table 3).

4.9 Phosphorous content in leaf

The results presented in Table 3 show that the percent phosphorous (P) content in maize leaf of the two selected varieties had affected due to the mean effect ofdifferent arsenic levels.The highest P content (0.20 %) recorded in leaf of V_1 and it was lowest (0.0.14 %) inV_2.

The effect of different arsenic levels on % P content in leaf of maize plant differed insignificantly. The P content recorded in maize leaf was highest (0.24%) in 0 mg As/kg of soil and it was lowest (0.10 %) in 80 mg As/kg of soil (Table 3).

The combined effect of variety and arsenic was found significant in case of percent P content in leaf of maize plant (Table 3); where the highest P content (0.36 %) found in leaf of BARI maize 7 at 0 levels of arsenic and it was lowest (0.9 %) in Pacific 339 at 80 mg As/kg of soil, which was statistically similar with V_2T_3 and V_1T_4.

4.10 Arsenic content in leaf

The content of arsenic (As) in leaf of the entire two selected maize varieties varied at different levels of arsenic. Its content in leaf washighest (2.41 ppm) in Pacific 339 and lowest (2.40 ppm) in BARI maize 7 (Table 3).

The arsenic (As) content in leaf of maize significantly varied due to the effect of different arsenic levels; where the As content in leaf increased with the increasing level of arsenic in leaf.The highest As content (5.872 ppm) in leaf was recorded in 80 mg As/kg of soil level of arsenic and then was lowest (0.00 ppm) in 0 mg As/kg of soil(Table 3).

The combined effect of arsenic and variety on content of As (ppm) in leaf was found significant. The As content increased with the increasing levels of arsenic in both leaf of all varieties (Table 3). The highest As content (5.89 ppm) in leafwas found in Pacific 339 at 80 mg As/kg of soil and it was lowest (0.00ppm) in the variety BARI maize 7 at the 0 mg As/kg of soil arsenic level and Pacific 339 at the 0 mg As/kg of soil arsenic level.

4.11 Nitrogen content in stem

The results presented in Table 4 show that the percent nitrogen (N) content in maize stem of the two selected varieties had affected due to the mean effect of different arsenic levels.The highest N content (0.58%) recorded in stem of BARI maize 7 and it was lowest (0.56 %) in BARI maize 7.

The effect of different arsenic levels on % N content in stem of maize plant differed significantly. The N content recorded in maize stem was highest (0.69%) in 0 mg As/kg of soil and it was lowest (0.45 %) in 80 mg As/kg of soil (Table 4).

The combined effect of variety and arsenic was found significant in case of percent N content in stem of maize plant (Table 4); where the highest N content (0.69 %) found in stem of BARI maize 7 at 0 levels of arsenic, which was statistically similar with Pacific 339 at 0 mg As/kg of soil and it was lowest (0.43 %) in Pacific 339 at 80 mg As/kg of soil.

Table 4. Effect of variety, different level of arsenic and their interaction on the N, P, K and As content in stem of maize

Treatment	N (%)		K (%)		P (%)		As (ppm)	
Effect of variety								
V_1	0.58		1.89		0.15		1.58	
V_2	0.56		1.77		0.14		1.61	
CV(%)	5.60		4.75		6.20		10.11	
Effect of arsenic								
T_0	0.69	a	2.19	a	0.26	a	0.00	e
T_1	0.62	b	2.01	ab	0.18	b	0.85	d
T_2	0.57	c	1.86	bc	0.11	bc	1.15	c
T_3	0.51	d	1.64	cd	0.10	c	2.14	b
T_4	0.45	e	1.45	d	0.09	c	3.85	a
$LSD_{(0.05)}$	0.02		0.25		0.07		0.20	
CV(%)	5.60		4.75		6.20		10.11	
Interaction effect of variety and arsenic								
V_1T_0	0.69	a	2.27	a	0.28	a	0.00	e
V_1T_1	0.63	b	2.08	b	0.19	b	0.83	d
V_1T_2	0.58	bcd	1.96	c	0.09	d	1.08	cd
V_1T_3	0.53	de	1.68	de	0.10	d	2.12	b
V_1T_4	0.46	fg	1.48	f	0.09	d	3.82	a
V_2T_0	0.69	a	2.11	b	0.25	a	0.00	e
V_2T_1	0.61	bc	1.93	c	0.16	bc	0.86	d
V_2T_2	0.56	cd	1.76	d	0.12	cd	1.22	c
V_2T_3	0.49	ef	1.60	e	0.10	d	2.16	b
V_2T_4	0.43	g	1.42	f	0.09	d	3.88	a
$LSD_{(0.05)}$	0.05		0.11		0.05		0.27	
CV(%)	5.60		4.75		6.20		10.11	

LSD = Least of significant difference, CV = Coefficient of varience

4.12 Potassium content in stem

It appears from the results presented in Table 4 that there was variation in potassium (K) content in two selected maize varieties under mean effect of different arsenic levels. The highest K content in stem was found in BARI maize 7(1.89%) and that was lowest (1.77%) in Pacific 339.

The Potassium (K) contents in stem of maize also significantly varied due to the effect of different arsenic levels; wherethe K content decreased with the increasing level of arsenic in stem (Table 4).The highest K content in stem K (2.19%) was recorded in 0 mg As/kg of soil and it was lowest (1.45%) in 80 mg As/kg of soil arsenic level respectively.

The combined effects of arsenic and variety on content of K (%) in stem were differed significantly. The content of K in stem of all the selected varieties progressively decreased with increasing the arsenic levels. The highest K content (2.27%) in stem was found in BARI maize 7 at 0 mg As/kg of soil, which was statistically similar with Pacific 339 at 0 mg As/kg of soil and it was lowest (1.42%) in the Pacific 339 variety at the 80 mg As/kg of soil arsenic level (Table 4).

4.13 Phosphorous content in stem

The results presented in Table 4 show that the percent phosphorous (P) content in maize stem of the two selected varieties had significantly affected due to the mean effect of different arsenic levels.The highest P content (0.15%) recorded in stem of BARI maize 7 and it was lowest (0.14 %) in Pacific 339.

The effect of different arsenic levels on % P content in stem of maize plant differed significantly. The P content recorded in maize stem was highest (0.26 %) in 0 mg As/kg of soil and it was lowest (0.09 %) in 80 mg As/kg of soil (Table 4).

The combined effect of variety and arsenic was found significant in case of percent P content in stem of maize plant (Table 4); where the highest P content (0.28 %) found in stem of BARI maize 7at 0 levels of arsenic, which was statistically similar with Pacific 339 at 0 mg As/kg of soiland it was lowest (0.09 %) in Pacific 339 at 80 mg As/kg of soil.

4.14 Arsenic content in stem

The percent content of arsenic (As) in stem of the entire two selected maize varieties varied significantly grown at different levels of arsenic. The As content in stem was highest (1.61 %) in Pacific 339 and lowest (1.58 %) in BARI maize 7 (Table 4).

The arsenic (As) content in stem of maize significantly varied due to the effect of different arsenic levels; where the As content in stem increased with the increasing level of arsenic in stem.The highest As content (3.85%) in stem was recorded in 80 mg As/kg of soil level of arsenic and then was lowest (0 %) in 0 mg As/kg of soil respectively (Table 4).

The combined effect of arsenic and variety on content of As (ppm) in stem was found significant. The As content increased with the increasing levels of arsenic in stem of two varieties (Table 4). The highest As content (3.88ppm) in stem was found in Pacific 339 at 80 mg As/kg of soil, which was statistically similar with BARI maize 7 at the 80 mg As/kg of soil arsenic level and it was lowest (0.00%) in the variety BARI maize 7 at the 0 mg As/kg of soil arsenic level and Pacific 339 at 0 mg As/kg of soil.

4.15 Nitrogen content in grain

The results presented in Table 5 show that the percent nitrogen (N) content in maize grain of the two selected varieties had affected due to the mean effect of different arsenic levels.The highest N content (1.13 %) recorded in grain of BARI maize 7 and it was lowest (0.93 %) in Pacific 339.

The effect of different arsenic levels on % N content in grain of maize plant differed significantly. The N content recorded in maize grain was highest (1.13 %) in 0 mg As/kg of soil and it was lowest (0.56 %) in 80 mg As/kg of soil (Table 5).

The combined effect of variety and arsenic was found significant in case of percent N content in grain of maize plant (Table 5); where the highest N content (1.25 %) found in grain of BARI maize 7 at 0 levels of arsenic and it was lowest (0.10 %) in Pacific 339 at 80 mg As/kg of soil.

Table 5. Effect of variet, different level of arsenic and theirinteraction on the N,P,K and As content in grain of maize

Treatment	N (%)		K (%)		P (%)		As (ppm)	
Effect of variety								
V_1	1.13		2.37		0.46		0.70	
V_2	0.93		2.35		0.44		0.75	
CV(%)	5.37		10.25		4.40		5.48	
Effect of arsenic								
T_0	1.23	a	2.93	a	0.63	a	0.00	c
T_1	1.17	a	2.72	b	0.54	b	0.66	b
T_2	1.11	a	2.40	c	0.44	c	0.77	b
T_3	1.08	a	1.93	d	0.37	d	0.92	ab
T_4	0.56	b	1.81	d	0.29	e	1.28	a
$LSD_{(0.05)}$	0.34		0.14		0.07		0.39	
CV(%)	5.37		10.25		4.40		5.48	
Interaction effect of variety and arsenic								
V_1T_0	1.25	a	2.95	a	0.65	a	0.00	d
V_1T_1	1.18	ab	2.74	b	0.51	bcd	0.67	bc
V_1T_2	1.12	abc	2.43	c	0.42	de	0.75	bc
V_1T_3	1.09	bc	1.90	f	0.38	ef	0.91	bc
V_1T_4	1.02	c	1.82	g	0.30	f	1.12	ab
V_2T_0	1.20	ab	2.90	a	0.60	ab	0.00	d
V_2T_1	1.15	abc	2.71	b	0.56	abc	0.65	c
V_2T_2	1.10	bc	2.36	d	0.46	cde	0.78	bc
V_2T_3	1.08	bc	1.96	e	0.35	ef	0.92	bc
V_2T_4	0.10	d	1.79	g	0.27	f	1.43	a
$LSD_{(0.05)}$	0.12		0.05		0.11		0.41	
CV(%)	5.37		10.25		4.40		5.48	

LSD = Least of significant difference, CV = Coefficient of varience

4.16 Potassium content in grain

It appears from the results presented in Table 5 that there was variation in potassium (K) content in two selected maize varieties under mean effect of different arsenic levels. The highest K content in grain was found in BARI maize 7 (2.37%) and that was lowest (2.35%) in Pacific 339.

The Potassium (K) contents in grain of maize also significantly varied due to the effect of different arsenic levels; wherethe K content decreased with the increasing level of arsenic in grain (Table 5).The highest K content in grain(2.93%) was recorded in 0 mg As/kg of soil and it was lowest (1.81%) in 80 mg As/kg of soil arsenic level respectively.

The combined effects of arsenic and variety on content of K (%) in grain were differed significantly. The content of K in grain of two selected varieties progressively decreased with increasing the arsenic levels. The highest K content (2.95%) in grain was found in BARI maize 7 at 0 mg As/kg of soil, which was statistically similar with Pacific 339 at 0 mg As/kg of soil and it was lowest (1.79%) in the Pacific 339 variety at the 80 mg As/kg of soil arsenic level, which was statistically similar with Pacific 339 variety at the 60 mg As/kg of soil arsenic level (Table 5).

4.17 Phosphorous content in grain

The results presented in Table 5 show that the percent phosphorous (P) content in maize grain of the two selected varieties had significantly affected due to the mean effect of different arsenic levels.The highest P content (0.46 %) recorded in grain of BARI maize 7 and it was lowest (0.44 %) in Pacific 339.

The effect of different arsenic levels on % P content in grain of maize plant differed significantly. The P content recorded in maize grain was highest (0.63 %) in 0 mg As/kg of soil and it was lowest (0.29 %) in 80 mg As/kg of soil (Table 5).

The combined effect of variety and arsenic was found significant in case of percent P content in grain of maize plant (Table 5); where the highest P content (0.65 %) found in grain of BARI maize 7 at 0 levels of arsenicand it was lowest (0.27%) in Pacific 339 at 80 mg As/kg of soil.

4.18 Arsenic content in grain

The percent content of arsenic (As) in grain of the two selected maize varieties varied significantly grown at different levels of arsenic. The As content in grain washighest (0.75ppm) in Pacific 339 and lowest (0.70 ppm) in BARI maize 7 (Table 5).

The arsenic (As) content in grain of maize significantly varied due to the effect of different arsenic levels; where the As content in grain increased with the increasing level of arsenic in grain.The highest As content (1.28 ppm) in grain was recorded in 80 mg As/kg of soil level of arsenic which is statistically identical with 60 mg

As/kg of soil level of arsenic and then was lowest (0 %) in 0 mg As/kg of soil respectively (Table 5).

The combined effect of arsenic and variety on content of As in grain was found significant (Table 5). The highest As content (1.43 ppm) in grain was found in Pacific 339 at 80 mg As/kg of soil and it was lowest (0.00 ppm) in the variety BARI maize 7 at the 0 mg As/kg of soil arsenic level and Pacific 339 at 0 mg As/kg of soil.

CHAPTER V

SUMMARY AND CONCLUSIONS

The experiment was conducted at the net house and laboratory of department of Agricultural Chemistry, Sher-e-Bangla Agricultural University, Dhaka under pot-culture to determine the effect of arsenic on growth, yield and arsenic content in leaf and grain of maize (*Zea mays*). The experiment was consisted using two varieties viz. V_1=BARI Maize 7, V_2= Pacific 339 and five arsenic levels viz. T_0 = 0 mg As/kg soil, T_1 = 20 mg As/kg soil, T_2 = 40 mg As/kg soil, T_3 = 60 mg As/kg soil and T_4 =80 mg As/kg soil. The experiment was set in Completely Randomized Design **(**CRD) having two factors with three replications.

The results on the effect of morphological characters indicated that plant height, number of leaves, total dry mater, number of cob per plant, number of grain per plant, grain yield per plant were influenced by the variety. The taller plant (144.07 cm) was found in Pacific 339 (V_2). BARI maize 7 was achieved maximum leaves per plant (13.73). The highest total dry mater (TDM) (215.94 gm plant^{-1}) was recorded in V_2. The number of cob per plant (0.87) was highest in BARI maize 7. BARI maize 7 produced maximum number (111.27) of grain per plant. The highest grain yield plant^{-1} (31.79 g) was found in variety BARI maize 7 and the lowest yield (26.32 g) was recorded in Pacific 339. The N content in leaf was highest (0.87 %) in V_1. The highest K content in leaf was found in Pacific 339 (1.88 %). The highest P content (0.198 %) recorded in leaf of Pacific 339. The highest N (0.58%), K (1.89%) and P content (0.149 %) recorded in stem of BARI maize 7. The As content in stem was highest (1.613 %) in Pacific 339. The highest N content (1.131

%), K (2.37%) and P content (0.463 %) recorded in grain of BARI maize 7. The As content in grain was highest (0.754 ppm) in Pacific 339.

All parameter was statistically influenced by different arsenic levels. The highest (147.3 cm) plant height, number of leaf per plant (15.67), TDM (254.94 g), number of cob per plant (1.42), number of grain plant^{-1} (207.00) were recorded at 0 mg As/kg of soil levels of arsenic. The highest grain yield plant^{-1} (58.54) was recorded at control treatment and it was lowest (12.17 g) at 80 mg As/kg of soil level of arsenic. The highest N content (0.99%), K content (2.22%), P content (0.24%) in leaf as recorded in 0 mg As/kg of soil level of arsenic. The highest As content (5.872 ppm) in leaf was recorded in 80 mg As/kg of soil level of arsenic N (0.687 %), K (2.19%) and P (0.263 %) was recorded in 0 mg As/kg of soil level of arsenic. The highest As content (3.853%) in stem was recorded in 80 mg As/kg of soil level of arsenic. The highest N (1.127 %), K (2.93%) and P (0.627 %) were recorded in maize grain in 0 mg As/kg of soil. The highest As content (1.278 ppm) in grain was recorded in 80 mg As/kg of soil.

In combination effect of cultivars and arsenic levels was significantly influenced on all parameter. The highest (153.40 cm) plant height was found in Pacific 339 with 0 mg As/kg of soil (V_2T_0). The maximum number of leaf per plant (15.77) was found from BARI maize 7 with 0 mg As/kg of soil (V_1T_0). The highest TDM (302.5 g) was found in BARI maize 7 at 0 mg As/kg of soil. At 0 mg As/kg of soil level of arsenic, the maximum cob per plant (1.50) were found in BARI maize 7. The maximum number of grain plant^{-1} (226.00) was found in BARI maize 7 at 0 mg As/kg of soil. The highest grain yield plant^{-1} (63.75 g) was found in BARI maize 7 at 0 mg As/kg of soil arsenic level and the lowest yield (6.33 g) was obtained in Pacific 339 at 80 mg As/kg of soil arsenic level. The highest N (1.00 %) and P (0.36%) in leaf were found in BARI maize 7 at 0 mg As/kg of soil. The highest K content (2.25 %) was found in leaf of Pacific 339 at 0 level of As. The highest As content (5.89 ppm) in leaf was found in Pacific 339 at 80 mg As/kg of soil, which was statistically similar with V_1T_4 treatment. The highest N content (0.69 %), K content (2.27%) and P content (0.28 %) found in stem of BARI maize 7 at 0 levels of arsenic. The highest As content (3.88 %) in stem was found in Pacific 339 at 80 mg As/kg of soil, which was statistically similar with V_1T_4 treatment. the highest N content (1.25 %), K content (2.95%) and P content (0.65 %) found in grain of BARI maize 7 at 0 levels of arsenic. The highest As content (1.43 ppm) in grain was found in Pacific 339 at 80 mg As/kg of soil.

Based on the above results following conclusions and recommendation may be made -

● The BARI maize 7 had better yield than Pacific 339.

● The content of N, P and K in leaf, stem and grain decreased and As content increased by increasing the level of Arsenic in soil.

● Based on the above conclusions plant breeder may adapt the technique of selection or screening the genotypes and develop As tolerant maize cultivars.

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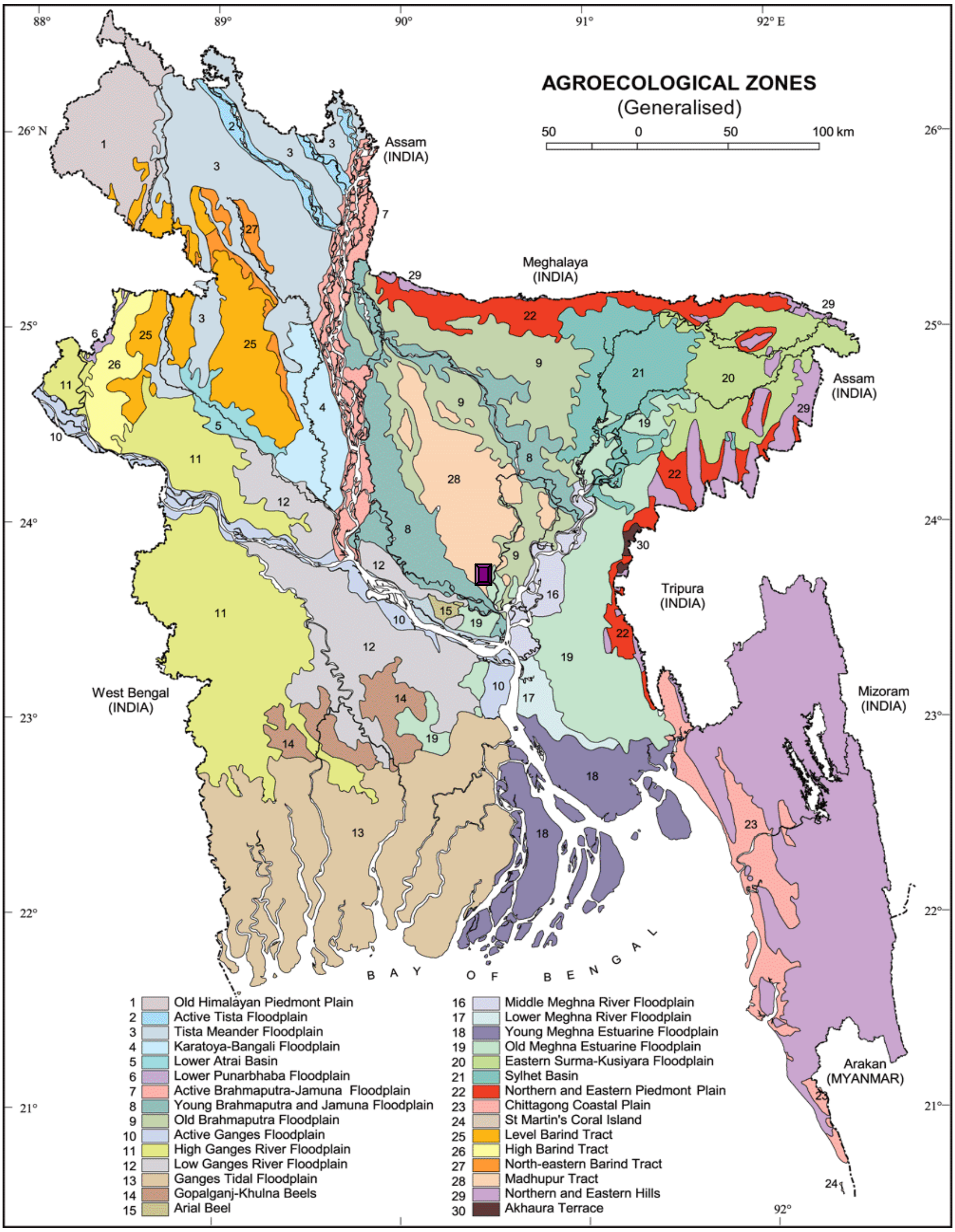
APPENDICES

Appendix I. Map showing the experimental sites under study



■ **The experimental site under study**

Appendix II. Analysis of variance of the data on the growth and yield of maize influenced by variety and arsenic level

Source of variation	Degrees of freedom	Mean square					
		Plant height	leaf per plant	TDM	Cob per plant	grain per plant	grain weight per plant
Factor A	1	335.2	0.133	4286.7	0.133	2940.3	223.04
Factor B	4	119.62*	14.083*	10293*	0.929*	25820*	2110.3*
AB	4	81.195*	0.05*	2908.4*	0.154*	1760.3*	101.34*
Error	20	71.818	1.1	3449.7	0.192	1082.1	101.96

* Significant at 0.05 level of probability

Appendix III. Analysis of variance of the data on N, P, K, As content in leaf of maize influenced by variety and arsenic level

Source of variation	Degrees of freedom	Mean square			
		N	K	P	As
Factor A	1	0.001	0.047	0.022	0
Factor B	4	0.1*	2.475*	0.023^{NS}	30.468*
AB	4	0.001*	0.009*	0.024*	0.002*
Error	20	0.004	0.101	0.023	0.025

* Significant at 0.05 level of probability

Appendix IV. Analysis of variance of the data on N, P, K, As content in stem of maize influenced by variety and arsenic level

Source of variation	Degrees of freedom	Mean square			
		N	K	P	As
Factor A	1	0.004	0.043	0	0.007
Factor B	4	0.053*	4.237*	0.033*	13.043*
AB	4	0.001*	0.041*	0.001*	0.008*
Error	20	0.001	0.1	0.001	0.026

* Significant at 0.05 level of probability NS: Non significant

Appendix V. Analysis of variance of the data on N, P, K, As content in grain of maize influenced by variety and arsenic level

Source of variation	Degrees of freedom	Mean square			
		N	K	P	As
Factor A	1	0.313	0.408	0.006	0.026
Factor B	4	0.432*	1.587*	0.109*	1.31*
AB	4	0.242*	0.249*	0.001*	0.03*
Error	20	0.025	0.1	0.004	0.059

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