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Khan, M.S. 1991. Angiosperms. In: *Two Centuries of Plant Sciences in Bangladesh and Adjacent Regions* (ed. A.K.M. Nurul Islam). pp. 175-194, Asiatic Soc. Bangladesh. Dhaka. Bhuiyan, A.L. 1964. *Fishes of Dacca.* Asiatic Society of Bangladesh, Dhaka. 148 pp.

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EFFICACY OF FRUIT PULP SOLVENT EXTRACTS OF CASSIA FISTULA LINN. AGAINST THE FOURTH INSTAR LARVAE OF THE MOSQUITO CULEX QUINQUEFASCIATUS SAY

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Abstract

The efficacy of the fruit pulp extracts of *Cassia fistula* Linn (Caesalpiniodae: Leguminosae) extracted with three solvents (viz. water, acetone and n- hexanes) was studied against the 4th instar larvae of *Culex quinquefasciatus* Say (Diptera: Culicidae) in the laboratory. Larval mortality was observed after 36 hours. The sun-dried extracts showed 18.67, 30.67, 41.33, 54.67 and 70.67 percent larval mortalities, respectively while the shade-dried extracts showed 6.67, 21.33, 42.67, 68.00 and 90.67 percent larval mortalities, respectively. The acetone based sun five extracts showed 18.67, 41.33, 60.00, 82.67 and 98.67 percent larval mortalities, respectively and the shade-dried extracts showed 17.33, 41.33, 73.33, 89.33 and 97.33 percent larval mortalities, respectively. The n-hexane based sun-dried extracts showed 21.33, 41.33, 59.67, 74.67 and 93.33 larval mortalities, respectively and the shade-dried extracts showed 33.33, 60.00, 68.00, 77.33 and 97.33 percent larval mortalities, respectively. The LC_{50} values for n-hexane, acetone and water based sun-dried extracts were 1.087, 1.097 and 3.211 mg/ml, respectively and the shade-dried extracts were 0.808, 1.054 and 3.048 mg/ml, respectively. No mortality was observed in control treatment.

Key words: Toxicity, *Culex quinquefasciatus, Cassia fistula*, Mosquito, Larvae, LC_{50}

Introduction

Mosquitoes are dominant insects of tropical areas causing a great concern for the public health and are common nuisance pests throughout the urban areas of all over the world. There are over 3500 species of mosquitoes on the globe under 41 genera (Leisnham 2007). There are about 117 species of mosquitoes recorded in Bangladesh (Ahmed *et al*. 2009). So far 79 Culicines and 36 Anophelines have been recorded (Bashar *et al*. 2013) of which about 25 species are under the genus *Culex,* most common and predominant one being the species *Culex quinquefasciatus* (Ahmed 1987). In Dhaka city more than 90 per cent of the mosquitoes belong to *Cx. quinquefasciatus* (Ameen *et al.* 1994). Recently, (Khan *et al*. 2014 and 2015) reported 13 species of mosquitoes in five wards of Dhaka metropolitan city of which *Cx. quinquefasciatus* was the predominant one.

The mosquitoes serve as vectors in the transmission of important pathogens to mankind that cause fatal diseases. About 300 species of mosquitoes are responsible for the transmission of the human and animal diseases. They transmit the pathogens of malaria, filaria, yellow fever, dengue fever, dengue hemorrhagic fever, encephalitis, dermatobiasis etc. Among them *Culex* mosquitoes carry encephalitis, filariasis, and the West Nile virus etc. Mosquito borne diseases attack about100 million people per year and more than one million of them die (Hill 1997).

That's why the mosquitoes create global health problem, particularly in the tropics (WHO 1999). So, the mosquito *Cx. quinquefasciatus* is a serious threat to the human health. As its population is increasing in an alarming rate it requires to be controlled.

Now-a-days synthetic pyrithroid, carbamate, hydrochlorine and other organophosphorus compounds are used to control mosquitoes. These are harmful to human health and to other non-target populations because of their non biodegradable nature and higher rate of biological magnification through ecosystem. So, an alternative way should be found out which will be effective in the mosquito management strategies, along with it will focus on public health, monitoring and surveillance, source reduction and environmentally least-toxic larval control. These factors have resulted in an approach to look for environment friendly, cost-effective, biodegradable and target specific insecticides against mosquito species. Plant materials used as potential pesticides were first time studied in the country by Ameen *et al*. (1983a & b and 1985). They studied the solvent based root extracts of *Derris elliptica* plant on the larvae of two mosquito species of the genera *Aedes* and *Culex*; they also isolated rotenone, the principal toxicant ingredient of *D. elliptica* and the chemical was bioassayed as insecticide on the larvae of the mosquito species. The plant *Cassia fistula* is a semi wild slender tree. It is known as "Golden Shower Tree" or "Sonalu" which is well known in Bangladesh as an ornamental tree for its beautiful bunches of yellow flowers. This plant is also well known for its traditional use as medicine. There is little information regarding the use of *C. fistula* as the source of insecticides and application to mosquito control in the country. However some information are available in literature in which different parts of *C. fistula* have been reported to be used as crude extracts with solvents as well as individual toxic ingredients for the control of different species of mosquitoes and other insect species (Govindarajan *et al*. 2008, Govindarajan 2009, 2013 a & 2013 b, Barakat *et al*. 2004, Danish *et al*. 2011, Duraipandiyan *et al*. 2011, Kumar *et al*. 2014, and Misra *et al*. 1997). The chemical components of *C. fistula* vary with in different parts of the plant. The pulp of pod of *C. fistula* contains anthraquinone glycosides, sennosides A and B, rhein and its glucoside, barbaloin, aloin, formic acid, butyric acid and their ethyl esters, oxalic acid, pectin, tannin, emodin, sennidin, aloe-emodin (Agarwal and Paridhavi 2005, Dave and ledwani 2012, Liptak and Szentagali 1937 and Misra *et al*. 1997). Some of these chemicals have larvicidal properties among which rhein was found to be a very effective larvicide (Duraipandiyan *et al.* 2011). Ethanol and hexane crude extracts of *C. fistula* reduce pupation, egg production, hatchability and increased per cent sterility in the cotton leaf worm, *Spodoptera littoralis* (Barakat *et al.*2004). Stearic acid has larvicidal properties and methanolic extracts of C. *fistula* are promising as larvicidal and ovicidal agents against mosquito (Danish *et al.* 2011).

The present study was conducted in the laboratory to determine the efficacy of three solvent based extracts of the fruit pulp of the plant *Cassia fistula* on the mosquito larvae of *Cx. quinquefasciatus.*

Materials and Methods

Test insect: The larvae of the mosquito *Cx. quinquefasciatus* Say (Diptera: Culicidae) were collected from the drains of Curzon Hall premises, Dhaka University in October 2015 and were carried to the Entomological Laboratory of the Zoology department, Dhaka University where these were reared and bioassay tests were conducted in the ambient environment of the laboratory at 27 ± 2° C and 75-85% RH. The larvae belonging to *Cx. quinquefasciatus* were identified by following the identifying key suggested by Bram (1967).

As the larvae of *Cx. quinquefasciatus* were required for the bioassay test with the plant extracts, a continuous supply of the larvae was maintained in the rearing room placed in the Animal Garden of the Department of Zoology, Dhaka University. The larvae were served with yeast powder, while the adults were provided with 10% glucose solutions as their food. After 3-4 days of emergence, the adult female mosquitoes were given a blood meal from a pigeon, *Columba livia* for egg maturation.

*Test botanical***:** The fruits of the plant *C. fistula* were collected from the Curzon Hall campus area, Dhaka University and "Osmani Milonayoton" premises, Dhaka in November 2015 and carried to the Entomology laboratory. The extraction procedure was conducted at the Entomological laboratory of the Department of Zoology and CARS (Centre for Advanced Research Science) of the University of Dhaka. The collected plant parts were taken to the Botany Department of the University for the Species Identification and the species was confirmed as *Cassia fistula*.

Plant extraction: The fruits of *C. fistula* were collected and washed with tap water. Then one half of the collected materials was placed under the sun and the other half in the shade for drying. After drying, the shade-dried sample was found slightly sticky in comparison to the sun-dried sample. Then, the fruit pulps, excepting seeds and outer covering, were taken into an electric blender machine and ground into powder and stored.

Six conical flasks (500 ml) were taken and rinsed with respective solvents for the preparation of different solvent based extract solutions. Then, 50 g of sun and 50 g of shade-dried fruit pulp powders of *C. fistula* were taken separately in each of the six flasks (three for sun-dried and three for shade dried extracts). With these 300 ml fresh distilled water, Acetone, and n-Hexane were added separately and kept for 24 hours with periodic shaking in an Orbital Shaker Machine at 100 rpm and 30^0C. The solutions were then collected and stored at temperature 4^0C in an air tight glass bottle for dose preparation in bioassay tests.

Dose preparation: Several doses of the extracts were prepared for the experiment using a process of serial dilution. The plant extracts were taken into a screw capped vial and then weighed in a weighing machine. As the organic solvents based extracts were insoluble in water media, Di-methyl Sulfoxide (DMSO) was used to make them soluble in water as per the suggestion of Nour *et al.* (2012). The concentrations of the five doses for water based extracts were 2.0, 2.5, 3.0, 3.5 and 4.0 mg/ml, and for acetone based extracts were 0.5, 1.0, 1.5, 2.0 and 2.5 mg/ml. But, in case of n-hexane solvent based extract, which was insoluble in DMSO solutions, were prepared by following the method proposed by Ravichandran *et al.* (2014). To prepare the stock solution, one gram of plant extract residue was dissolved in 5 ml of acetone, mixed well and then it was dissolved in 95 ml of distilled water. Each ml of this stock solution contains 10 mg of plant residue. By diluting this stock solution, a series of doses of different concentrations were prepared for bioassay tests and the concentrations of the prepared doses were 0.5, 1.0, 1.5, 2.0 and 2.5 mg/ml.

Bioassay tests: A larviciadal bioassay method, suggested by Dua *et al*. (2009) was followed. The method was conducted with slight modification. Twenty five actively swimming mosquito larvae of 4th instar, were taken into a conical flask (250 ml) containing 100 ml water along with one of the five doses of the plant extracts. The flasks were stored at room 29 ± 2°C, 80 ± 5% RH and at 14L: 10D (photoperiod). The mortality of the larvae was recorded after 36 hours of exposures and the moribund larvae were counted as dead. A set of Control using 2.0% DMSO was taken as control 1, a set of Control using 5% Acetone was taken as

control 2 and an untreated set of larvae in water (tap) as Control 3. These three sets of control were run for calculating the corrected mortality.

The toxicity of the extracts was calculated as LC_{50} and LC_{90} representing the 50% and 90% of the test larvae died, respectively; both LC_{50} and LC_{90} values were calculated for 36 hours of exposures. The number of larvae died at each of the dose concentrations at the end of the stipulated exposure periods was recorded and the mortality percentage values were calculated by using the formula-

$$\text{Percentage of mortality} = \frac{\check{R}n\ LC\ 牛\ A\ 狩\ C\ 懦狩\ L\acute{G}RL\acute{G}}{\check{R}n\ LC\ 牛\ SL\ 虐\ SA\ 狩\ C\ 懦狩\ L} \times 100$$

When the mortality in control was more than 5%, the percentage mortality was corrected by using Abbott's (1925) formula-

$$\text{Corrected mortality} = \frac{捻狩\ C\ 懦狩\ ACS\ 狩\ ARS\ 哦\ RaSCL\ 狩\ SLaS - 捻狩\ C\ 懦狩\ ACS\ 狩\ ARS\ 哦\ Ra\ni aSCA}{400 - \ni aSCACS\ 狩\ ARS\ 哦} \times 100$$

Statistical analysis: Larval mortality data were observed and corrected mortality was obtained by applying Abbott's formula (Abbott 1925). LC_{50} and LC_{90} at 95% confidence intervals, lower and upper confidence limits were determined by the probit analysis method suggested by Finney (1971). Other statistics like chi-square values, regression at 95% confidence intervals of upper confidence limits and lower confidence limits and ttests were calculated using the IBM SPSS statistics 20 (Statistical Package of Social Science) software; here significance level were set at $p < 0.05$.

Results and Discussion

The larvicidal efficacy of different solvent based sun-dried and shade-dried extracts of the fruit pulp of *C. fistula* was tested against the 4th instar larvae of *Cx. quinquefasciatus*. Data were recorded and mean percentage mortality was calculated. Five different dose concentrations (viz. 2.0, 2.5, 3.0, 3.5 and 4.0 mg / ml) of water based sun-dried extracts showed 18.67, 30.67, 41.33, 54.67 and 70.67% larval mortality, respectively; while the shade-dried extracts showed 6.67, 21.33, 42.67, 68.00 and 90.67% larval mortality, respectively. Five different dose concentrations (viz. 0.5, 1.0, 1.5, 2.0 and 2.5 mg / ml) of acetone based sun-dried extracts showed 18.67, 41.33, 60.00, 82.67 and 98.67% larval mortality, respectively; the shade-dried extracts showed 17.33, 41.33, 73.33, 89.33 and 97.33% larval mortality, respectively. And five different dose concentrations (viz. 0.5, 1.0, 1.5, 2.0 and 2.5 mg / ml) of n-hexane based sun-dried extracts showed 21.33, 41.33, 59.67, 74.67 and 93.33% larval mortality, respectively and the shade-dried extracts showed 33.33, 60.00, 68.00, 77.33 and 97.33% larval mortality, respectively. Based on the above larval mortalities, values of LC_{50}, LC_{90}, Chi-square, parameter estimation and 95% confidence limits were calculated and the results are presented in Table 1.

Table 1. Efficacy of different solvent based sun-dried and shade-dried extracts of the fruit pulp of *Cassia fistula* against the 4th instar larvae of *Cx. quinquefasciatus*.

Types	Used Solvents	LC_{50} value (LCL-UCL)	LC_{90} value (LCL-UCL)	Parameter estimation Concentration (LCL-UCL)	Intercept (LCL-UCL)	X^2 (df-13) value (significance)
Sun dried extracts	Water	3.211 (3.005-3.478)	6.074 (5.134-8.138)	4.630 (3.320-5.940)	-2.346 (-2.669- -2.023)	5.31 (0.968[a])
	Acetone	1.054 (0.945-1.163)	2.467 (2.144-2.982)	3.471 (2.823-4.120)	-0.08 (-o.161- -0.001)	14.995 (0.308[a])
	n-Hexane	1.087 (0.957-1.217)	3.041 (2.534-3.954)	2.869 (2.266-3.427)	-0.104 (-0.182- -0.026)	9.715 (0.717[a])
Shade dried extracts	Water	3.048 (2.937-3.166)	4.205 (3.956-4.570)	9.167 (7.520-10.814)	-4.436 (-4.847- -4.026)	9.124 (0.764[a])
	Acetone	1.097 (0.962-1.236)	3.187 (2.595-4.333)	2.768 (2.143-3.394)	-0.112 (-0.190- -0.033)	8.311 (0.760[a])
	n-Hexane	0.808 (0.663-0.938)	2.793 (2.271-3.823)	2.38 (1.800-2.960)	0.22 (0.145-0.295)	14.406 (0.346[a])

Here, LC= Lethal concentration, LCL= Lower confidence limit and UCL= Upper confidence limit of 95% confidence limits; df= Degree of freedom. a=Since the significance level is greater than 0.15, no heterogeneity factor is used in the calculation of confidence limits.

Comparison of the toxicity of different solvent based extracts of the sun-dried fruit pulp of C. fistula: The estimated LC_{50} for water, acetone and n-hexane based sun-dried extracts were 3.211 mg / ml, 1.097 mg / ml and 1.087 mg / ml, respectively; whereas LC_{90} values of these extracts were 6.074 mg / ml, 2.467 mg / ml and 3.041 mg / ml, respectively (Table 1). Here, the lowest LC_{50} and LC_{90} values belong to the n-hexane based extracts and the highest of these values belong to the water based extracts. This indicates that among all these three, the n-hexane based extracts showed highest toxicity and the water based extract showed lowest toxicity; On the contrary the acetone based extracts showed larval toxicity laying in-between the former two extracts (i.e. n-hexane and water based extracts). The relative potency of three types of sun dried fruit extracts of *C. fistula* against the larvae of *Cx. quinquefasciatus* on the basis of LC_{50} values in decreasing order were as follows: n-hexane (1.087 mg / ml) > acetone (1.097 mg / ml) > water (3.211 mg / ml).

Comparison of the toxicity of different solvent based extracts of the shade-dried fruit pulp of C. fistula: The estimated LC_{50} for water, acetone and n-hexane based extracts were 3.048, 1.054 and 0.808 mg / ml, respectively; whereas, the LC_{90}, values were 4.205, 3.187 and 2.793 mg / ml, respectively (Table 1). Thus, the n-hexen based extracts showed lowest LC_{50} and LC_{90} values and the water based extracts showed highest LC_{50} and LC_{90} values indicating that the shade-dried extracts showed the same results as that of the sun-dried extracts, i.e. among all these three, n-hexane based extract was highest in toxicity level and the water based extract was lowest and acetone based extract was moderate. The relative potency of three types of sun-dried fruit extracts of *C. fistula* against the 4th instar larvae of *Cx. quinquefasciatus,* on the basis of LC_{50} values in decreasing order were as follows: n-hexane (0.808 mg / ml) > acetone (1.054 mg / ml) > water (3.048 mg / ml).

Comparison of toxicity between sun-dried and shade-dried fruit pulps of C. fistula: Among all six types of solvent extracts, the n-hexane based shade-dried extract was found to be most effective (LC_{50}=0.808 mg/ml) larvicide against the tested larvae of *Cx. quinquefasciatus;* the acetone based sun-dried extract ranked 2nd in position of toxicity (LC_{50}=1.054 mg/ml); with the LC_{50} values of the n-hexane based sun-dried extract (LC_{50}= 1.087 mg/ml) and acetone based sun-dried extract (LC_{50}=1.097 mg/ml) were in the 3rd and 4th position, respectively. The water based extracts were found to be less effective than the acetone and n-hexane based extracts and the LC_{50}values were 3.048 mg/ml and 3.211 mg/ml for shade-dried and sun-dried extract, respectively. The relative potency of six types of extracts on the basis of the decreasing order of LC_{50} values were as follows: n-hexane shade (0.808 mg / ml) > acetone shade (1.054 mg / ml) > n-hexane sun (1.087 mg / ml)> acetone sun (1.097 mg / ml) > water shade (3.048 mg / ml) > water sun (3.211 mg / ml). So, for the same solvent based extract, effectiveness of the shadedried extracts was found better than the sun-dried extracts.

Comparison between sun and shade-dried extracts: Paired t-test was performed to show the comparative results between sun-dried and shade-dried extracts at all three solvents (Table 2). The water based shade-dried and sun-dried extracts, acetone based sun-dried and shade-dried extracts and n-hexane based sun-dried and the shade-dried extracts of the fruit pulp of *C. fistula* were found to be significant as shown in Table 2. The differences of the estimated means were 2.60, 1.2 and 2.8, respectively. The paired sample t-test values were 3.833, 1.39 and 4.221, respectively. And paired sample correlation values were 0.982, 9.74 and 9.83, respectively. As all the values were less than 0.005, the results were significant.

Table 2. Differences between the larvicidal efficacy of different solvent based sun dried and shade dried extracts of the fruit pulp of *C. fistula* against the 4th instar larvae of *Culex quinquefasciatus*.

Solvents	Type	Dead at each level Mean	Standard deviation	Standard error of mean	Paired t-test	Significance (2-tailed)	Degree of freedom	Paired Sample Correlation	Significance
	Sun	9.800	6.181	2.764					
Water	Shade	12.40	7.057	3.156	3.833	0.019	4	0.982	0.003
	Sun	14.60	7.603	3.400					
Acetone	Shade	15.80	8.258	3.693	1.395	0.235	4	0.974	0.005
	Sun	14.20	7.120	3.184					
n-Hexane	Shade	17.00	6.285	2.811	4.221	0.013	4	0.983	0.003

In the present study, the crude extracts of the fruit pulp of *C. fistula* were found to have effective larvicide against *Cx. quiquefasciatus*. The resulting larvicidal activities of the fruit pulp extracts of *C. fistula* may be comparable with some of the earlier reports. Govindarajan *et al*. (2008) studied the ovicidal and larvicidal efficacy of methanolic leaf extracts of *C. fistula* against two mosquitoes, viz. *Anopheles stephensi* and *Culex quinquefasciatus* and reported the LC_{50} values of 17.95 and 20.57 mg / L (=0.02057 mg / ml), respectively. Thus it seems that the methonolic extracts of the leaves of *C. fistula* are more toxic than all three solvent extracts of the fruit pulp of the same plant in the present study. Later Govindarajan

(2013) studied the efficacy of crude methanolic extracts of the flower of *C. fistula* against three mosquito species, viz. *Culex tritaeniorhynchus*, *Aedes albopictus* and *Anopheles subpitus,* and reported that the flower extracts of *C. fistula* were excellent larvicidal potential against the tested mosquito species showing the LC_{50} values of 136.59, 118.64 and 96.51 ppm, respectively, and with the LC_{90} values of 243.67, 231.79 and 174.39 ppm, respectively. In another earlier study, Govindarajan *et al*. (2009) reported the bioefficacy of *C. fistula* leaf extracts with different solvents like benzene, acetone and methanol against dengue vector *Aedes aegypti* and estimated LC_{50} values of the above extracts which were 10.69, 18.27 and 23.95 mg / L, respectively; these values indicate that the benzene leaf extracts of *C. fistula* had highest efficacy on the larvae of *Ae. Aegypti*.

Duraipandiyan *et al*. (2011) reported the antifeedant and larvicidal activities of the chemical rhein isolated from *C. fistula* and the LC_{50} value were 606.50 ppm (0.607 mg / ml) for *Heliothis armigera* and 1192.55 ppm (1.193 mg / ml) for *Spodoptera littoralis* and the larvae survived showed malformed adults. Kumar *et al*. (2014) reported the LC_{50} of *Cassia occidentalis* leaf petroleum ether and butanol extract against *Cx. quinquefasciatus* 3rd instar larvae were 98.4 and 161.6µg / ml, respectively. Barakat *et al*. (2004) reported that the ethanol and hexane crude extracts of *Cassia fistula* reduced pupation, egg production and hatchability, and increased per cent sterility; the dominant constituents were fatty acids, linoleic acid, hexadecanoic acid, and octadecanoic acid and their alkyl esters.

From the present findings and the relevant information available from literature it may be said that the plant *C. fistula* is a potential resource for the extraction of toxic ingredients which may be used as insecticides for the control of mosquito. Further research may be initiated to isolate the toxic ingredients from different plant parts of this species and biassayed on mosquito species.

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THREATS AND CONSERVATION PROBLEMS OF NON-HUMAN PRIMATES IN MOIST DECIDUOUS FOREST OF BANGLADESH

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Abstract

A study was conducted at the Madhupur deciduous forest, Tangail from April 2015 to October 2015. It was carried out. Formal and informal questionnaire survey was used to interview the forest staffs and local people of the peripheral villages of Madhupur forest. Habitat destruction due to expansion of agricultural land and illegal logging and firewood collection was the prime threats of primates' conservation especially Langur endurance. Hunting by ethnic people for using food was another threat for primates' survival. Diseases and road accident were additional cause of threats. Crop raid was the main factor for human primates' conflict. Paddy, pineapple and jackfruits were severely damaged by primates as they spoiled more crops than they actually eat. Time and economic loss was due to engagement of people guarding the crop field till harvesting.

Key words: Capped langur, Rhesus macaque, Habitat destruction

Introduction

The non-human primates are represented with 63 genera and about 600 species and subspecies in some 92 countries (Chopra *et al.* 2013). Among these 25 species are recorded from the Indian sub-continent (*op. cit.*) and 10 species occur in Bangladesh (IUCN Bangladesh 2015). All of these primates are threatened in Bangladesh, except Stumptail Macaque which is data deficient (*op. cit.*). Distributions of all these primates are habitat specific except Rhesus Macaque which is found in all forest habitat types as well as in and around human settlements (*op. cit.*). At present, about 6% of the land area of the country is covered by forests (Gain 2002) representing three types of forest, such as semi-evergreen and evergreen forest in the northeast and southeast hill tracts, moist deciduous forest in the central region and the Sundarbans mangrove forest in the southwest (Hasan *et al.* 2013). The Madhupur forest (340 km^2) is the largest deciduous forest of Bangladesh (NFA 2007). It is a fragmented and disturbed forest in Bangladesh. It is under high pressure for fuel wood, fallen dry leaves for cooking, grazing, illicit felling and fire hazards (Khan 2010). This forest is enriched with different wild fauna like 115 birds (Khan and Ahsan 2011) and many species of mammals including two non-human primates. Logging, fragmentation, urbanization, encroachment, habitat loss, shrinkage of forests, development activities, habitat destruction, jhum cultivation, deforestation, illegal tree falling, over exploitation and potential habitat declining are several factors for mammalian species declination of the country (IUCN Bangladesh 2015). Several works have been done on different aspects of primates in India (Biswas *et*

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al. 2009, Kumar and Solanki 2004, 2008 and Solanki *et al.* 2008) and in Bangladesh (Green 1978, Gittins 1980, Khan and Ahsan 1981, Feeroz *et al.* 1995 and Feeroz 2001). But data on threats and their conservation in this forest or throughout the country is scanty. The present study aims to describe the existing threats on primates for which their population is being declining and their conservation problems in Madhupur forest. Based on this study it would be possible to formulate effective conservation measures for primate conservation in this forest as well as in this country.

Materials and Methods

A preliminary field survey was done at first to find out the sites where the primates are found within the Park. Garo tribes, local peoples and forest guards were interviewed informally to know the distribution of Rhesus Macaque and Capped Langur in the park area. The area was visited on foot or by unmotorized vehicle or rickshaw van, animals were observed using 10 x 42mm Bushnell binocular, photograph was taken by using Canon 60D DSLR camera. Fortnightly two days long study was done. A total of 28 days, 280 hours was spent to carry out research at the study area. Surveys were carried out from dawn to dusk to record any disturbances due to human activities such as settlements, grazing, logging, agriculture, hunting and poaching. In association with direct observation a questionnaire survey was done to record information on the histories and present status of the primates', threats, human and primates' conflicts and attitudes of the people towards the primates' conservation of the study area. Both formal and informal questionnaires were used to interview the forest staffs and local people of the peripheral villages. The investigation was done on formally 25 local people (included both male and female) of different occupations like farmer, wood cutter, fire wood collector, cow boy, pineapple garden owner, forest guard, driver, student etc.

Study area: The study was carried out at the Madhupur deciduous forest, Tangail during April 2015 to October 2015. It is situated in the northern part of Bhawal-Madhupur *Shal* (*Shorea robusta*) forest tract, somewhat 50 km south of the Garo Hills of the Meghalaya State of India, and about 110 km north of Dhaka, the capital of Bangladesh. Geographically, it lies in 24°41.323′N and 90°.8.275′E. The Madhupur forests, commonly known as 'Madhupur Garh', forms a slightly elevated tract of approximately 1-2 m in height over the surrounding plains. There are numerous depressions with gentle slope intercepting the ridges (Ismail and Mia 1973). Flat ridges run north to south forming the irregular masses of high lands with gentle slopes which are then intercepted by numerous depressions in the form of long and narrow ditches. The forest is partly dense, partly thin and there are scrub jungles also. Two types of tribe (*Garos* and *Koch*) are used to live inside the Madhupur forest area and Bengali in the fringe areas. In general, this forest is dominated by *Shal* trees associated with other tree species like *Grewia laevigata*, *Zizyphus oenoplia*, *Phyllanthus embelica*, *Terminalia belerica*, etc (Adhikari 2008).

Results and Discussion

Threats

1. Habitat destruction: As the human population is increasing and the need for material, space, and land use is expanding at an alarming rate, people are rapidly encroaching and destroying the primate habitat for their own need. Non-human primates are just one of the any creatures which are threatened by human sprawl and resource exploitation. The followings are distinctive ways that humans are directly or indirectly responsible for the loss of habitat essential for primates' endurance in the study area:

Expansion of agricultural land and crop production: It was observed that local people at the forest edges were expanding their cultivated land regularly. By destroying forest habitat they preferred to cultivate paddy (*Oryza* spp.), banana (*Musa* spp.), pineapple (*Ananas sativus*), zinger (*Zingiber officinale*), turmeric (*Curcuma longa*), lemon (*Citrus aurantifolia*), seasonal vegetables etc. (Plate 1). Even after clearing the forest area the rubber garden was established. Even, inside the deep forest some landmass were cleared up and these crops were planted. Thus forest became fragmented and it was very tiresome for primates' movement especially for langurs' because they do not prefer to use the forest floor during movement from one place to another.

Logging and firewood collection: It has already mentioned that the forest areas were cleared up regularly and rapidly. Due to increase of human populations and rapid urbanization, the existing forest areas are still facing continuous threats of degradation. In addition to this, human population pressure has accelerated the fragmentation of wildlife habitats. Illegal logging (Plate 2) caused the destruction of both large and undergrowth canopies. The fire wood collectors caused massive habitat destruction throughout the year for their livelihood. They took not only the dried plants but also they broke or cut down live branches (young and thin branches) of the plants, kept down on the forest floor and collected after drying. The wood collectors regularly entered into the forest for wood collection either it was live or dry. It was known from the wood collector that the undergrowth collection was about 80 pounds in a day and sold in local market in 400-500 BDT. It was reported from the collectors that in total, 25 to 30 collectors regularly used to collect the woods from the area of Rasulpur, Beribaid, Lohoria and Dokhola of the National Park. Several groups (consisting of 3 to 8 people in each group) are responsible for illegal logging in this forest. Recently forest department has taken initiative to stop illegal logging and habitat destruction by implementing community participation management programme. Local people informed that due to this initiative, illegal logging and habitat destruction are becoming less in recent past years than the previous years. Moreover, the Rubber Garden area was seen quite well protected by local government branch office of related ministry and guards strictly patrolling the garden area. As a result of cutting down the large fruit plants for various purposes both primates were facing serious problems and invaded the local peoples' garden or entered into the human habitation for food. It was found that the Hindura (*Mellotus philippensis*) and Ziga (*Lannea coromandelica*) were highly cut down by both illegal logger and fire wood collectors.

Mammalian habitats have been destroyed and fragmented in an alarming rate because of accelerating human population pressure and different anthropogenic factors (IUCN Bangladesh 2015). Illegal timber collectors collect both large sized hard wood plants which eventually affect upper canopy used by arboreal mammals like langurs and other primates, and soft wood plants which provide food for them, creates uneven gaps in the forest and eventually affects both feeding and foraging of arboreal mammals (*op. cit.*). Habitat destruction was recorded as the principal threats of Capped langur in Northeast India (Biswas *et al.* 2009, Srivastava 2006 and Kumar and Solanki 2004). Illegal wood and fire wood collectors are also responsible for habitat destruction in Lantang National Park of Nepal (Regmi and Kandel 2008). Habitat loss and degradation are the causes of population declination of Capped Langur (Kumar and Solanki 2004). Due to the collection of non-timber forest the depletion of food plants important to the langurs were found to be most serious threats in Arunachal Pradesh in India (Kumar and Solanki 2008). Biswas *et al.* (2009) recorded rapid habitat loss, fragmentation, alteration of habitat, jhoom cultivation, hydro electrical dam construction etc. were severe problems of macaques conservation in the protected areas of Assam. The extraction of plant species including food plants by the local people inhabiting the surrounding area of the sanctuary is another threat of Capped Langur and other primate species depletion (Kumar and Solanki 2003, Kumar 2006). Kumar and Solanki (2008) mentioned the clear felling of mature forests for agriculture and settlements in the area which were bordering the Nameri National Park in Assam and Pakke Wildlife Sanctuary in Arunachal Pradesh in India. In Madhupur forest, it was found that the Capped langur is confined to a fragmented area. Kumar and Solanki (2008) also reported that the Capped langur is confined to a fragmented area in Arunachal Pradesh of India.

2. Hunting: It was reported that, local people used several methods to hunt Rhesus Macaque and Capped Langur. From the questionnaire survey it was noted that, ethnic people use their pet dog and fruits bait trap for hunting primates. Fruits bait traps usually used seasonal fruits (banana, pineapples, jackfruits and litchi) when these were available. Rhesus Macaque often trapped for pet or for using as performance monkey. During the study period one infant was trapped in the Harinatala village and later used as pet animal and the local people informed that three adult macaques were kept for pet.

In this forest, Capped langurs are under high hunting pressure due to their small group composition. Hunting was less reported in forest edges and other human dominated areas where forest guards or local people or visitor were regularly seen to visit. Moreover, the hunter preferred adult to hunt comparatively which supplied more flesh than younger one.

From the questionnaire survey it was revealed that the Capped langur was hunted for food during special occasion (Christmas day) of the ethnic people or opportunistically. It was also noted that ethnic people of 15-25 ages, mostly illiterate were known to be active hunter. Ethnic people were found to be engaged in group hunting (Plate 3).

Hunting and poaching are the major threats of mammals (IUCN Bangladesh 2015). Santals and Garo community of northern part of the country go out for group hunting in winter (*op. cit.*). Rhesus Macaques have been wiped out from the Sal forest of the north Bengal due to over hunting (*op. cit.*). Ethnic people hunt the primates for food which was

a cause of threat in Assam (Biswas *et al.* 2009) and Arunachal Pradesh in India (Kumar and Solanki 2008), Northeast India (Srivastava 2006 and Kumar and Solanki 2004). Hunting for medicinal purposes and artifacts for socio-cultural practices and religious and cult ceremonies are the causes of population declination (Kumar and Solanki 2004, 2008 and Biswas 2009). Hunting, poaching and habitat destruction are frequent in the adjacent forest areas of Pakke Wildlife Sancturay (Kumar and Solanki 2004).

3. Diseases: Local people reported that diarrhoea was known to occur at the child age of Rhesus Macaque and two were known to die. Another disease was found during study period at Beribaid beat area where the tail of Macaque at first was infected, wounded and finally fell off (Plates 4a,b) and gradually legs and other body parts were affected, fell off and eventually the animal died. During this time the infected macaque suffered from nausea and gradually became weak and thin. It was reported from the forest guards that two juveniles died due to this disease. In the study period one was died and two juveniles were seen to be infected.

4. Accident: The construction of roads inside the forest creates a severe disturbance which forced primates to invade the gardens of different fruits and agricultural fields near human habitation. Moreover, Mymensingh-Tangail highway with numerous bends in different areas has crossed and fragmented the potential habitat of the primates in forest area. During the study period, 4 accidents were reported; causing 4 spot dead and 2 injured (Table 1). All these accidents happened during crossing the road except Monarbaid beat while one adult male was jumped from one tree to other over the road and fell down on the running bus and injured severely. Many mammalian species die on roads through the crash with vehicles as many forest areas in the country have been bisected by roads, highways and railway roads (IUCN Bangladesh 2015). Road accident is recorded as a threat in Langtang National Park of Nepal (Regmi and Kandel 2008, Kumar and Solanki 2008 and Minhas *et al.* 2010).

Table 1. Consequence of road accident occurred in Madhupur National Park.

Location	Rhesus Macaque			Capped Langur		
	Adult Male	Juvenile	Consequence	Adult female	Infant	Consequence
Road near the Jaloi cottage	1	-	Dead	1	1	Dead
At the middle of the road passing through the Jaloi and Rosulpur beat	1	-	Dead	-	-	-
Monarbaid beat	1	-	Injured	-	-	-
At the middle of the road passing through the Jaloi and rosulpur beat		1	Injured	-	-	-

5. Tourist activities and others: Uncontrolled tourist activities were recorded in Madhupur Forest especially in winter and during any religious festival such as, Eid festival, Puja etc. Several areas (Shontoshpur, Dokhola, Beribaid beat) are being used as picnic spots or for tourist activities (Plate 5). Visitors were seen screaming, shouting and laughing high in forest trails. People were seen carrying out loud speakers and cooking food at the forest edges. At Beribaid Beat, Capped Langur was seen quite irregular near the picnic spot or any other event spot. Uncontrolled tourist activities severely interrupt normal daily activity of diurnal mammals (IUCN Bangladesh 2015). Loud sound, curious activities and non-ecofriendly activities by the visitors in the protected areas force the mammalian species to leave their home ranges (*op. cit.*). But large numbers of Rhesus Macaques were recorded in Shontoshpur, Dokhola and Beribaid beat where tourists and visitors came regularly and provided foods to them. Rhesus Macaques are being habituated with provisioning for tourist attraction (IUCN Bangladesh 2015). Tourists visiting these areas get a chance to feed these monkeys (*op. cit.*). The pesticides and insecticides used in the agricultural fields were known to get mixed with local ponds, drain water, causing serious problems to primates which usually drink this water.

Conflicts between human and non-human Primates
During the study period, following two types of conflicts were observed.

Conflicts between human and Rhesus Macaque: The Rhesus Macaque which raided the crops just before harvesting severely damaged crops like paddy and pineapples. When the pineapple fruits became matured, Rhesus Macaques entered into the forest-fringed gardens and caused massive damage, as each individual got a hold of at least one fruit during each raid. The macaques also raided ripe jackfruit (*Artocarpus heterophyllus*) (Plate 6).

Conflicts between human and Capped Langur: While different foods were available near human habitation, the Capped Langur started to move to human habitation area from forest area. They raided home gardens, fruits and vegetables garden destroying paddy (*Oryza* spp.), Papaya (*Carica papaya*), Banana (*Musa* spp.), Black berry (*Syzygium* spp.), Dewa (*Artocarpus lacucha*), Mango (*Mangifera indica*), Lichee (*Litchi chinensis*), Sajna (*Moringa oleifera*), Nut (*Cocos nucifera*), Jackfruit (*Artocarpus heterophyllus*), Guava (*Psidium guajava*), bamboo (*Bambusa* spp.). Capped Langur was also recorded to destroy vegetables, fresh leaves, plant shoots, buds, flowers, and fruits. The farmers informed that about 15-25% crops were destroyed during a season. Crop raiding by primates was also reported by several authors in different countries (Priston 2005 and Siex and Struhsaker 1999). It was reported that both Capped Langur and Rhesus Macaque spoiled more crops than they actually eat; local people informed that the Rhesus Macaque was the most frequent crop raiders than Langur. Similar observations were also reported by Chalise (2000).

Crop preference: From the questionnaire survey it was found that the primates severely caused paddy field destruction (Fig. 1). Besides the direct loss, they caused loss of crops by feeding upon the flowering and fruiting trees which reduce fruit production considerably. Rhesus Macaque highly raided the paddy fields. Cereals, fruits and tubers

are the most preferred and became vulnerable for raiding by macaques (Chalise 2000). Khatry (2006) reported that maize is the prominently vulnerable crops for raiding by primates.

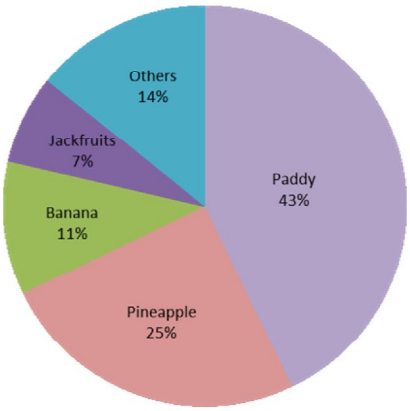


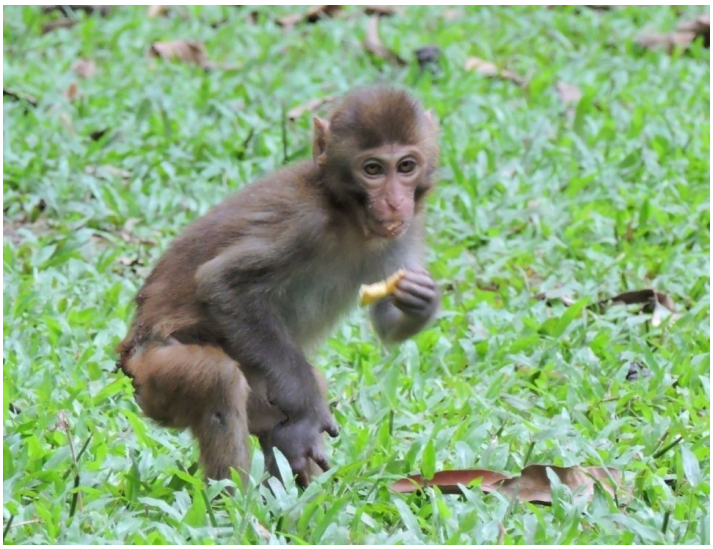
Fig.1. Crop damaged by primates.

Crop protection strategies: To protect crop fields from primates, farmers used many methods. The methods included patrolling and guarding the fields by farmers including their children and chasing of primates by using bamboo stick, catapult and branch of tree. Local people informed that people had to guard the paddy field with bamboo sticks, catapult and other tools to protect crops. Pineapple field owners had to keep permanent guard (Plate 7) to protect crops till harvesting. Regmi and Kandel (2008) reported that guarding of crop fields, scarecrows, tin-box, stones and catapults, keeping dogs, fencing with thorny twigs etc. are the methods used to protect the crops in Langtang National Park of Nepal.

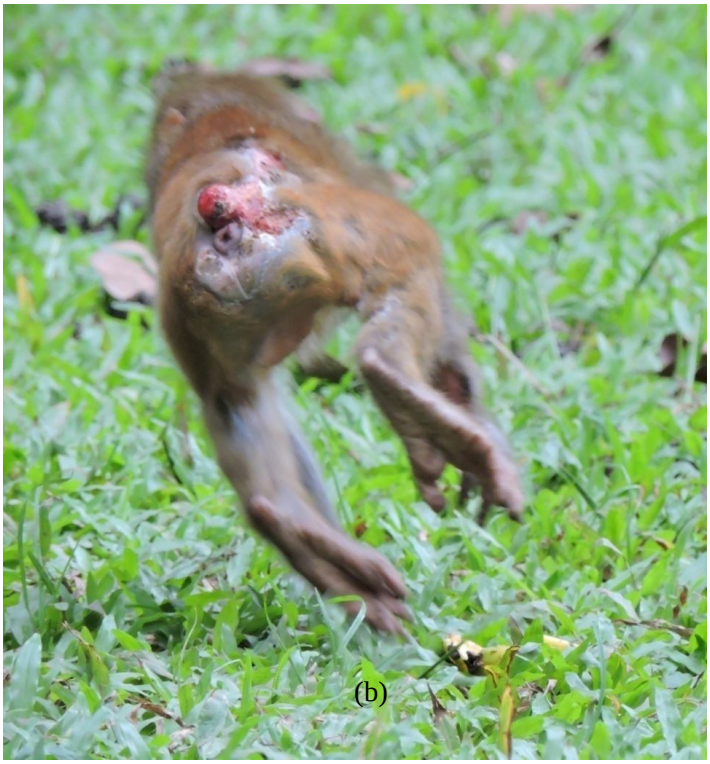
Plate 1. Cultivation of pineapple by clearing the primate habitat.

Plate 2. Regular practice of wood logging activities in MadhupurNational Park.

Plate 3. Ethnic group go out for group hunting in Madhupur National Park.



(a)



(b)

Plate 4. A young Rhesus Macaque having right ill hand (a) and dropped tail (b).

Plate 5. Tourist activities in Madhupur National Park.

Plate 6. Rhesus Macaque raided the ripe jackfruit.

The present study has given an idea about the existing threats of primates' conservation in Madhupur forest of Bangladesh. More research should be conducted to formulate a management plan and to aware people to conserve the primates' population, especially the Capped Langur in this forest.

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ANALYSIS OF WATER QUALITY OF THE MEGHNA RIVER USING MULTIVARIATE ANALYSES AND RPI

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Abstract

The present study was carried out to measure the physico-chemical variables of surface water of the Meghna River from September 2015 to March 2016. Air temperature, water temperature, soil pH, water pH, DO, BOD_5, COD, TSS, TDS, PO_4-P, NO_3-N and EC concentrations in water samples were found to range from 14-29°C, 11-28°C, 5.9-6.9, 7.2-7.5, 4.2-6.71 mg/L, 0.67-3.71 mg/L, 2.5-5.9 mg/L, 8-20 mg/L, 72-130 mg/L, 0.11-0.90 µg/l, 0.115-0.90 µg/l and 115.8-220 μScm^{-1} respectively. River Pollution Index (RPI) indicated that the water of river at impacted site was less to moderately polluted. Unimpacted site was also less polluted as per RPI and mostly used for agriculture. Correlation Matrix (CM) and Multivariate statistical analyses namely Cluster Analysis (CA), Principal Component Analysis (PCA) and Factors Analysis (FA) indicated that river water was polluted. The principal causes of the pollution of the river were haphazard industrialization, domestic sewage and agricultural inputs. Using of river water can pose serious problems to human health by biological food chain. This research suggests to proper management of the river with eco-friendly industrialization and urbanization for sustainable development of the country.

Key words: Water quality, Physico-chemical parameters, River pollution index, Industrialization, Meghna River

Introduction

Water, the most essential and important compound for all living creatures that form ecosystems (Manjare *et al*. 2010 and Kataria *et al*. 2011). Water quality can be assessed by its physical, chemical and biological properties (Manjare *et al*. 2010). This water delivers multiple uses for innumerable rural and urban communities and livestock, fish culture, recharge of ground water, control of floods etc. (Gurunathan and Shanmugam 2006). The quality of water is being degraded continuously due to haphazard industrialization (Manjare *et al*. 2010). Principally, the term industrialization is related with socio-economic activities (Thanoon *et al*. 2003, Richard 2005 and Jaillon and Poon 2009) that are basically responsible for the modification of the society setup (Abdullah *et al*. 2009) through the enormous production (Thanoon *et al*. 2003 and Abdullah *et al*. 2009). Most probably the effects of industrialization is in the state of beggar description on the aquatic biota. These may affect man unswervingly or through the provisions of water, agricultural ingredient and other biological products, the most common source of pollutants frequently discharged from domestic and industrial sectors. Chemicals produced from industries are a major source of water pollution that carried through

geological materials may cause problems (Kataria *et al*. 2011). Most of the industries are major contributor of trace metals. According to the WHO up to 80% of all illness and ailment in the world is triggered by poor sanitation and contaminated water (Budhathoki 2010). Least developed countries like Bangladesh, facing serious problems with water contaminations from different industries, domestic wastes and agrochemicals (Venugopal *et al.* 2009 and Islam *et al*. 2015a and b).

The main aim of this study was the assessment of water quality to report the baseline data of the area that will be useful in future to measure any anthropogenic contamination level.

Materials and Methods

Sampling Sites: Meghna River near Narsingdi Sadar is used as a river *Ghat* to move in various directions from Narsingdi District by cluster of wood made engine boats awaiting for passengers. The launch terminal is just steps away from this *Ghat*. There are a lot of textile mills and textile-related industries nearby. Moreover, dying industries and jute industries are also occupying in the catchment area. Therefore, the sources of pollution are various. Phosphorus in wastewater comes from food, pesticides, and industries (Site-1). Nitrates in wastewater originate from ammonia being converted to nitrates by bacteria in the treatment process. Chemicals that are known human endocrine disruptors include diethylstilbesterol (the drug DES), dioxin, PCBs, DDT, and some other pesticides. Annual rainfall in the area is about 1,329 mm. The surrounding is mostly used for agriculture where various types of pesticides e.g. DDT, Algin, Organophosphates are widely used (Site-2).

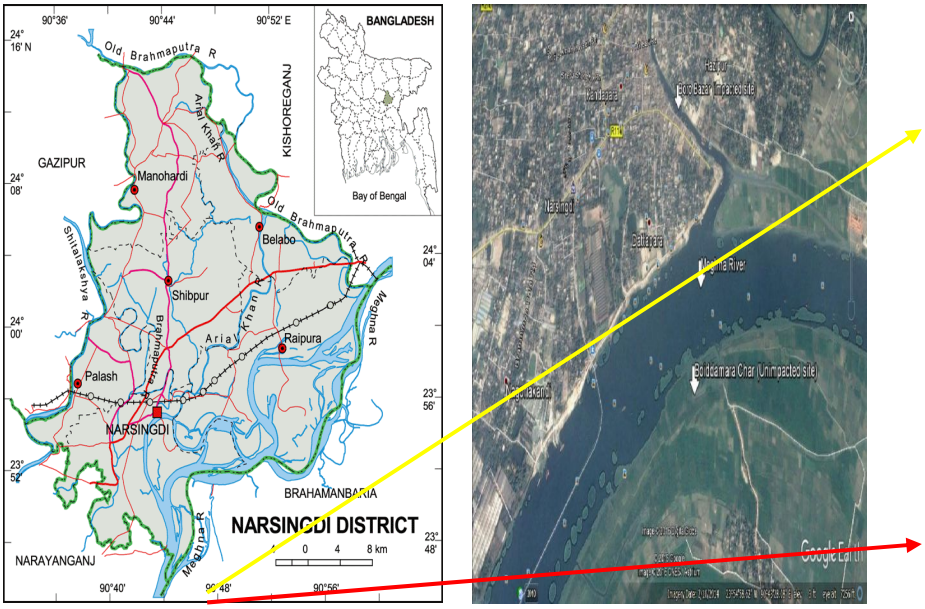

Fig. 1. Map showing sampling points of the Meghna River.

Water samples were collected from two points: 1. Effluent discharge area (Boro Bazar) and 2. far away from the discharged area (Boiddamar Char) of the Meghna River near Narsingdi District (Fig. 1). Sampling procedures were performed in three phases: firstly, September, 2015 (Rainy season); secondly, January, 2016 (Winter season) and thirdly, March, 2016 (Pre-monsoon).

Sample Collection and Preservation: After selection of sampling points, a total of 12 water samples was collected. 6 water samples were collected from industrial impacted site and rest 6 samples were collected from the pristine area (3 km far from industrial zone). 2 liters of surface water sample were collected in the morning hours between 10 to 11am, in polythene bottle regularly for every seasons. Immediately after collection, water sample were transferred to the laboratory of Bangladesh Council of Scientific and Industrial Research (BCSIR), Chittagong. pH, temperature and dissolved oxygen were measured from studied sites during sampling of water.

Sample Analysis: Water temperature was measured by a glass Celsius thermometer. Total suspended solid analyzed by the method stated by EPA (1979). The value of Hydrogen-ion-Concentration (pH) of water was determined by using pH paper (color pH indicators strips, Cat.9585, made in Germany). To determine the Dissolve Oxygen (DO) the collected samples in BOD bottles were fixed inside according to Azide Modification of Winkler (1988) and analyses were made by the same method. Bio-chemical Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Dissolved Solid (TDS), Nitrate-N (NO_3-N), Phosphate-P (PO_4-P) were analyzed by the method stated by APHA (2005).

Statistical Analysis: One Way Analysis of Variance ((Post-hoc LSD test) was done to show the variations in concentration of water parameters in terms of seasons and sites using SPSS (v.22). According to Dreher (2003), Principal Component Analysis (PCA) was performed on the original data set (without any weighting or standardization). Component Analysis (CA) is an effective tool to find out the similarity and variation with the influencing factors on different data sets (Wang *et al*. 2014). Moreover, CA is an important tool for the characterization and simplification of data sets with the behavior they possess. PCA was executed to sort out the principle features of variations in dataset with simplification and classification of raw data. According to Singh *et al*. (2004), PCA delivers strategies on spatial and temporal distribution of resultant factors. Pearson's product moment correlation matrix was done to identify the relation among parameters to make the result strong obtained from multivariate analysis. Cluster analysis (Dendogram) was performed to show the similarity among variables and to identify their sources of origin using PRIMER (v.6).

Results and Discussion

Water Temperature: Water temperature plays a significant role that influences the chemical, bio-chemical features of water body. The highest temperature was recorded 28^0C during pre-monsoon and lowest was recorded 11^0C during winter. The results found

to be similar with Simpi *et al*. (2011). Salve and Hiware (2008) reported that water temperature in pre-monsoon was high due to low water level, high temperature and clear atmosphere.

Table 1. Physico-chemical parameters of water at two sampling points during 3 seasons.

Parameters	Seasons	Air temperature (°C)	Water temperature (°C)	Soil pH	Water pH	DO (mg/L)	BOD_5 (mg/L)	COD (mg/L)	TSS (mg/L)	TDS (mg/L)	PO_4-P(µg/l)	NO_3-N (µg/l)	EC µScm^{-1})
Impacted site	Rainy	29	25	5.9	7.5	4.7	3.23	5.9	20	130	0.11	0.135	210
Unimpacted site		21	23	6.1	7.2	6.71	0.75	2.5	12	80	0.90	0.115	127.3
Impacted site	Winter	18	11	6.9	7.2	4.2	3.67	5.6	23	120	0.75	0.125	189.7
Unimpacted site		14	12	6.9	7.5	4.71	0.67	2.65	8	72	0.78	0.90	115.8
Impacted site	Pre-monsoon	31	28	6.8	7.3	4.3	3.71	5.4	25	122	0.78	0.124	220
Unimpacted site		29	26	6.9	7.4	4.56	3.61	3.01	10	75	0.80	0.122	135.9

Air Temperature: The temperature in 3 consecutive seasons of different sampling points fluctuated between (14-31)°C due to the influence of environmental temperature. Maximum temperature (31°C) was recorded from impacted site during pre-monsoon and minimum (14°C) was found in unimpacted site during winter. Dalal *et al*. (2013) recorded 26°C at the Dham River and Manjare *et al*. (2010) found 25°C at Tamdalge tank, Kolhapur district, Maharashtra.

Hydrogen-ion-Concentration (pH): Roy (1955), Moore (1972), APHA (2005), Mahmood and Bhuyian (1988), Sarma *et al*. (1982) and Campbell (1978) stated that the industrial or municipal waste materials had a significant role in increasing or decreasing pH of the adjacent water body where the waste materials were dumped. Air temperature is the prime responsible factor for changing the pH of water. Furthermore, bio-chemical and chemical reactions are influenced by the pH (Manjare *et al*. 2010). In the present study, the results indicate that the water of the river slightly alkaline (7.2-7.5) in nature. This report strictly agreed with the Farshad and Venkataramana (2012).

Dissolve Oxygen (DO): In the present study, the dissolve oxygen was recorded to be between (4.2-6.71) mg/L in sampling site. The highest DO recorded was 6.71 mg/L during rainy season. More or less similar observations were also recorded by Khan *et al*. (1976), Bhuyian (1979), Hossain *et al*. (1988), Mahmood and Bhuyian (1988), Hossain and Khan (1992), Jashimuddin and Khan (1993), Islam and Khan (1993), Alam *et al*. (1996) and Gasim *et al*. (2007).

Bio-chemical Oxygen Demand (BOD): Paul (1999) mentioned that, river water having BOD more than 10mg/L is considered to be moderately and more than 20 mg/L as to be highly contaminated water. In the present study, the BOD was found to range from 0.67-3.71 mg/L. More or less similar result was observed by Kataria *et al*. (2011) at Bhopal city water whereas higher BOD was recorded by Sikder *et al*. (2016) at the Turag River.

Chemical Oxygen Demand (COD): COD varied from (2.5-5.9) mg/L in the present study. Those results are far below than the results found by Sikder *et al*. (2016), Ahmed and Nizamuddin (2012) and Miah (2012). Higher BOD harmful to all aquatic life and can cause a substantial damage to submersed plants (Nian *et al*. 2007). Like BOD, higher COD is also harmful to all aquatic life.

Total Dissolved Solid (TDS): In the present study, the total dissolved solids fluctuated from (72-130) mg/L. The maximum value (130 mg/L) was recorded during rainy season at industrial polluted site. It is due to heavy rainfall of the season (Manjare *et al*. 2010) and minimum value (72 mg/L) was found during winter at unimpacted site.

Nitrate-N (NO_3-N): The values of nitrate ranges from 0.115 µg/l to 0.90 µg/l. The highest value (0.90 µg/l) was observed during winter at unimpacted site and lowest (0.115 µg/l) during rainy season at unimpacted site. Hasan *et al*. (2009) found 0.18 mg/L and 0.02 mg/L nitrite-nitrogen at the Buriganga and Panguchi River.

Phosphate-P (PO_4-P): The value of phosphate fluctuates from (0.11-0.90) µg/l. the maximum value (0.90 µg/l) was recorded during rainy season at unimpacted site and minimum value (0.11 µg/l) was recorded during rainy season at impacted site. This results fully acquiesced with results found by Manjare *et al*. (2010). The high values of phosphate recorded during monsoon might be due to rain, surface water runoff and agriculture run off (Manjare *et al*. 2010).

Spatial and Temporal Changes in Water Quality Parameters: Significant variations were found for air temperature, water temperature and soil pH with seasons as the significance level ($p \leq 0.05$) but water pH, DO, BOD, COD, TSS, TDS, PO_4-P, NO_3-N and EC showed no significant variations with seasons ($p \geq 0.05$). Furthermore, prevalent variations were found for COD, TSS, TDS and EC with sites as the alpha level ($p \leq 0.05$). No significant variations was found for air temperature, water temperature, soil pH, water pH, DO, BOD, PO_4-P and NO_3-N ($p \leq 0.05$) with sites (Fig. 2).

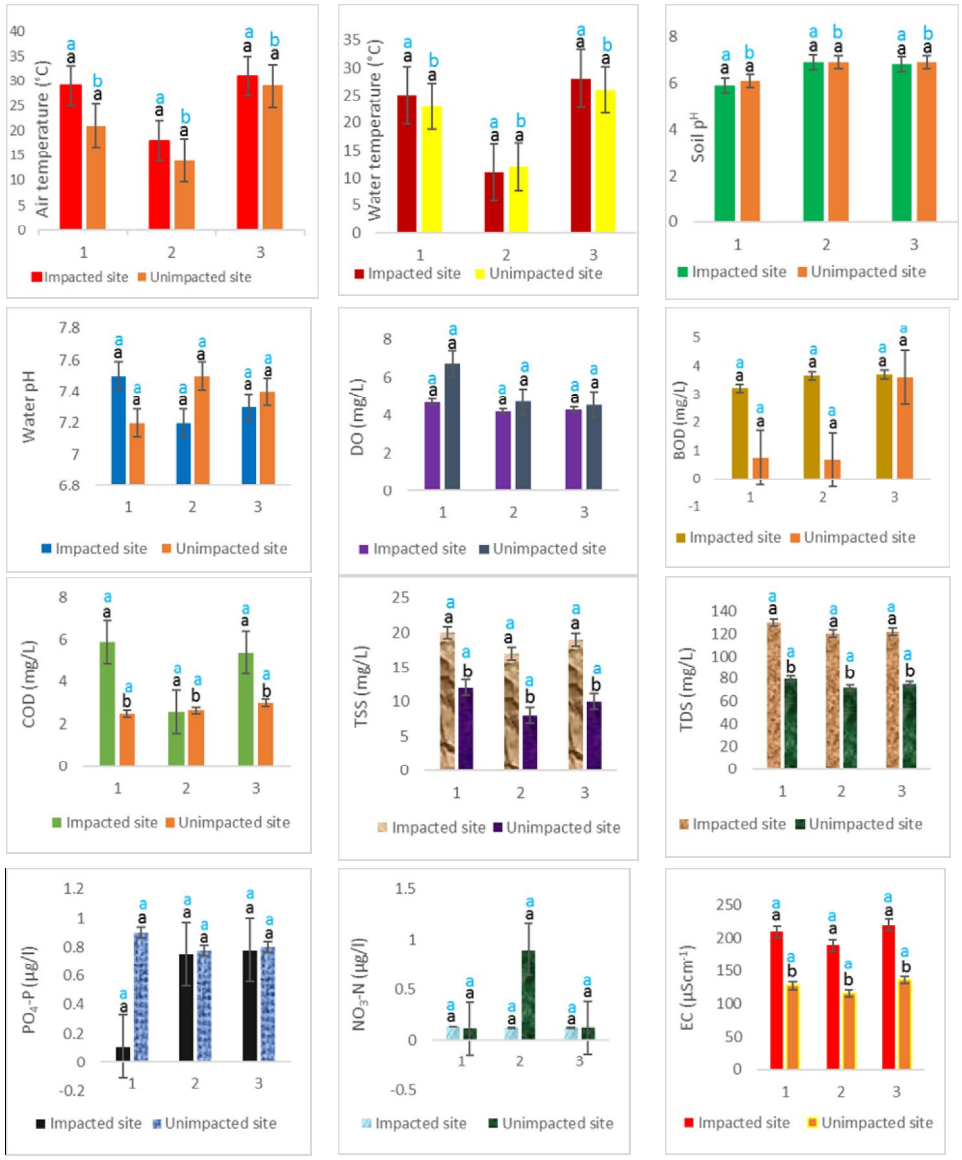

Fig. 2. Variations (Mean±SD) of water quality variables. Bars with the same letter are statistically indifferent.

1= Rainy season, 2= Winter season, 3= Pre-monsoon.

Black a & b= Variation with sites, Blue a & b= Variation with seasons.

Cluster Analysis: Different multivariate statistical analyses viz: Cluster Analysis (CA), Principal Component Analysis (PCA) and Factors Analysis (FA) act as fruitful guide for eloquent explanation of spatio-temporal parametric data. Many scholars have used these methods to evaluate and categorized the water quality. Wang *et al*. (2014) used these statistical analyses for the depict interpretation of the water chemistry. Furthermore, Talukder *et al*. (2016); Wang *et al*. (2012), Jiang-Qu *et al*. (2013), Venkastesharaju *et al*. (2010), Qadir *et al*. (2007) and Kowlkowski *et al*. (2006) stated that multivariate statistical methods (PCA, CA, FA) can be very effective tools for easy and clear interpretation of the complex data sets, recognizing pollution factors and assessing water quality parameters with spatio-temporal deviation.

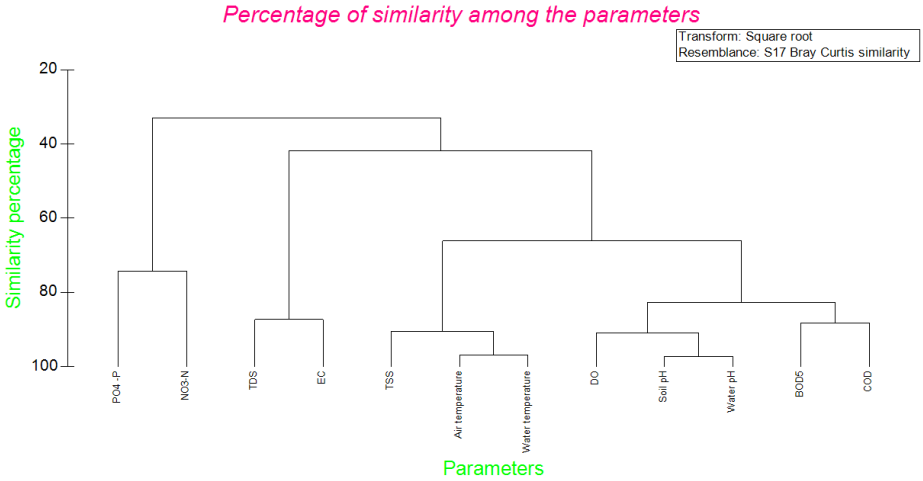

Fig. 3. Dendogram showing the percentage of similarity among parameters during different seasons at impacted site. (TSS=Total Dissolve Solid; BOD=Bio-Chemical Oxygen Demand; COD= Chemical Oxygen Demand; EC=Electrical Conductivity; DO=Dissolve Oxygen; TDS=Total Dissolved Solid).

Cluster analyses (CA) were executed using square root and Bray Curtis Similarity to show the similarity among the parameters that contribute hugely in water pollution. From the output of the cluster analysis total three clusters were found at impacted site during different seasons. Cluster 1 include: PO_4-P and NO_3-N; Cluster 2: TDS and EC; cluster 3: TSS, Air temperature and Water temperature; cluster 4: DO, Soil pH and Water pH; cluster 5: BOD_5 and COD (Fig. 3).

PO_4-P and NO_3-N represent strong linkage with minimum cluster distance that indicate those parameters have influencing power during seasonal variations. Parameters grouped together in less distance have higher affinity with similar identical behavior during temporal variations and also exert a probable effect to each other. Furthermore TDS and EC have also strong linkage but lesser than cluster 1 but contribute largely in environmental process. TSS, Air temperature and Water temperature were under the group of cluster 3 with minimum distance than cluster 1 and cluster 2 but have effects on environment. DO, Soil pH and Water pH formed cluster 4 but exert less influence than

cluster 1, 2 and cluster 3. BOD_5 and COD formed cluster 5 and have less effects on environment than cluster 1, 2, 3 and 4. Impacted site is the effluents discharged area of the river which highly affected by untreated industrial effluents, agricultural inputs and domestic wastes. Unimpacted area is located far from the effluents discharged area that can be treated as less polluted area.

Correlation Matrix: In river water environment, the inter linkage among water parameters deliver noteworthy information sources and pathways of parameters. The results of correlation between water parameters fully consented with the results obtained by PCA and CA that approve some new associations between variables. Very strong positive linear relationships were found between COD vs TDS (0.984), TSS vs TDS (0.981), TDS vs EC (0.971), TSS vs EC (0.969), COD vs EC (0.961), COD vs TSS (0.939), air temperature vs water temperature (0.919) (Table 3). Strong positive correlations were recorded between BOD vs Ec (0.747), BOD vs COD (0.741), air temperature vs BOD (0.667), BOD vs TDS (0.658), BOD vs TSS (0.642) and PO_4-P vs Soil pH (0.603) (Table 3). Moreover moderate positive linear relations were found between air temperature vs EC (0.583), Water pH vs NO_3-N (0.546), air temperature vs TSS (0.536), air temperature vs TDS (0.426) and air temperature vs COD (0.411) (Table 3). The very strong, strong and moderate correlations indicate that the parameters were originated from similar sources particularly from industrial effluents, domestic wastes and agricultural inputs. Besides, strong negative correlations were found between DO vs BOD (-0.708), NO_3-N vs Air temperature (-0.673), BOD vs NO_3-N (-0.632), TDS vs PO_4-P (-0.616), COD vs PO_4-P (-0.614) and TSS vs NO_3-N (-0.608) in river water.

Table 3. Correlation matrix of physico-chemical parameters in river water.

		Air temperature	Water temperature	Soil pH	Water pH	DO	BOD	COD	TSS	TDS	PO_4-P	NO_3-N	EC
Correlation	Air temperature	1.000											
	Water temperature	**0.919**	1.000										
	Soil pH	-0.246	-0.390	1.000									
	Water pH	0.104	0.068	-0.079	1.000								
	DO	-0.195	0.163	-0.592	-0.331	1.000							
	BOD	**0.667**	0.334	0.222	-0.081	-0.708	1.000						
	COD	**0.411**	0.097	-0.137	-0.047	-0.591	**0.741**	1.000					
	TSS	**0.536**	0.320	-0.372	-0.174	-0.316	**0.642**	**0.939**	1.000				
	TDS	**0.426**	0.162	-0.269	-0.111	-0.447	**0.658**	**0.984**	**0.981**	1.000			
	PO_4-P	-0.361	-0.200	**0.603**	-0.571	0.259	-0.307	-0.61	-0.59	-0.616	1.000		
	NO_3-N	-0.673	-0.583	0.334	**0.546**	-0.093	-0.632	-0.45	-0.61	-0.495	0.141	1.000	
	EC	**0.583**	0.321	-0.169	-0.092	-0.512	**0.747**	**0.961**	**0.969**	**0.971**	-0.539	-0.533	1.000

(At the significance level of 0.05).
Green color indicates very strong positive correlation.
Red color indicates strong positive correlation.
Blue color indicates moderate positive correlation.

Principal Component Analysis: The extraction method was used in PCA analysis that was Eigen values. The components were regarded as principal components whose Eigen values were greater than 0.6. Principal component highlighted the most vital factors that affect the water quality of the study area. PC1 had a highest initial Eigen value 6.06 and total variance 50.5%, with strong positive loading of EC, TSS, TDS, COD, BOD and air temperature, moderate positive loading of water temperature and moderate negative loading of NO_3-N, PO_4-P and DO resembled the loading of pollution mainly caused by untreated organic load with crucial anthropogenic effect (Table 4). PC 2 had Eigen value 2.32 and explained 19.3% of total variance, with strong positive loading of soil pH, high negative loading of DO and water temperature can be represented as effect of geological changes on environmental parameters. PC 3 explained 15.5 percent of total variance, with strong positive loading of PO_4-P. Strong negative loading of water pH and NO_3-N. The total variance of the PC4 was 12.1%. PC4 strongly correlated with Water temperature and Air temperature. From the present PCA study, it may be concluded that the source of PC 1 and PC 2 can be mixed source from anthropogenic inputs particularly from industrial wastes and agricultural actions in the study area. While the source of PC 3 and PC 4 can be considered as different source like lithogenic and anthropogenic inputs.

Table 4. Component matrix of four factors model with strong to moderate loadings in river water.

	Component			
	1	2	3	4
EC	0.961	0.140		-0.122
TSS	0.948			-0.287
TDS	0.927	0.156		-0.325
COD	0.923	0.298		-0.244
BOD	0.817	0.260	0.315	0.322
Air temperature	0.720	-0.381		0.576
NO_3-N	-0.670	0.510	-0.454	
DO	-0.458	-0.829		-0.317
Soil pH	-0.276	0.719	0.524	0.357
Water temperature	0.461	-0.657		0.563
Water pH		0.271	-0.850	0.451
PO_4-P	-0.627		0.743	
Eigen value	6.06	2.32	1.86	1.45
% Total variance	50.5	19.3	15.5	12.1
Cumulative %	50.5	69.8	85.8	97.4

River Pollution Index (RPI): In recent time, River pollution index (RPI) simple method is used concurrently by different organization like Taiwan EPA to assess the surface water quality. This method comprising with concentration level of four parameters: DO, BOD, SS, and NH_3-N. Pollution status is calculated using four-state of each parameter. The RPI is computed using following equation (Liou *et al.* 2004):

$$RPI = 1/4 \sum_{i=1}^{4} Si$$

Table 5. River Pollution Index (RPI) Chart (Chen *et al*. 2012 and Liou *et al*. 2004).

Items/ ranks	Good	Less polluted	Moderately polluted	Highly polluted
DO (mg/L)	>6.5	4.6-6.5	2.0-4.5	<2.0
BOD_5 (mg/L)	<3.5	3.0-4.9	5.0-15	>15
SS (mg/L)	<2.0	20-49	50-100	>100
NH_3-N (mg/L)	<0.5	0.5-0.9	91.0-3.0	>3.0
Index scores (Si)	1	3	6	10
Sub-index	<2	2.0-3.0	3.1-6.0	>6.0

In present study, the concentrations of DO, BOD_5 and TSS were compared with concentrations of RPI table to weigh the status of particular water variables (Table 1). Average DO in the impacted site was found to be 4.7 mg/L during rainy season that indicate the water is less polluted in comparison with RPI but the water of unimpacted site contained 4.2 mg/L DO that is the of good water. Averages DO in the impacted site was recorded 4.7 mg/L during winter season which can be treated as moderately polluted zone according to RPI index while 4.71 mg/L DO was found from the unimpacted site that direct the area is less polluted. During pre-monsoon the average DO concentration was recorded 4.3 mg/L, which designate moderate pollution in this area according to RPI table. Where 4.56 mg/L was found from unimpacted area that also renders the area characterized with less pollution. Average amount of BOD_5 were found between (3.23-3.71) mg/L for all seasons at the impacted site indicating that the water of the river is less polluted according to (Table 5). The mean concentration was recorded for BOD_5 3.61 mg/L during pre-monsoon at unimpacted area indicating the water is less polluted but the other two season's concentrations indicate that the water of unimpacted area is good. Average TSS for every seasons were found between (20-49) mg/L at impacted area that is a clue of less pollution according to RPI. At unimpacted site the average concentrations of TSS were recorded below (20-49) mg/L that indicate the water is good.

From the present findings it may be concluded that the water of the river is being polluted day by day due to unplanned industrialization, urbanization and agricultural activities. Using of river water for domestic, irrigation and pisciculture purposes might cause great harm to human being. Moreover, the risky concentrations of some water parameters can pose great risk to fish and human community dwelling in and adjacent to the Meghna River. Lately, the present research is giving preference on the better management of the river water to protect the health of riverine ecosystem.

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BACTERIAL LOADS IN SHRIMP AND FISH HATCHERY ENVIRONMENTS OF BANGLADESH

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Abstract

Artemia hatching tank of Cox's Bazar hatchery had similar total bacterial build up (2.59 ± 0.10×10^7cfu/g) in the water sampled and in the shrimp post larvae (PL) sampled at stage 10 and 12 (2.37 ± 0.11×10^7cfu/g and 2.42 ± 0.10×10^7cfu/g, respectively). In MA plate, no significant differences was observed in the bacterial count of these samples. Similar result was observed for the total presumptive vibrio count in TCBS plates ranging from 3.8 ± 0.60×10^3cfu/g to1.62 ± 0.50×10^3cfu/g.Total bacterial load (7.5 ± 0.11×10^7) measured in the water sampled from 25 day old fry rearing pond of tilapia from Mymensingh Hatchery, was similar to that of 33 day old fry (8.6 ± $.66\times10^7$). The bacterial density found in the 25 (1.6 ± 0.50×10^7), 28 (3.12 ± 0.14×10^7) and 40 day old fry (6.46 ± 1.52×10^6) samples was similar but significantly different from the sample of 33 day old fry and the water sample of the pond of 25 day old fry. In TCBS plate, bacterial abundance detected in the samples across all four age groups was similar (25 day old fry: 4.21 ± 3.79×10^3; 28 day old fry: 4.90 ± 3.50×10^3; 33 day old fry: 1.08 ± 0.12×10^3; 40 day old fry: 7.04 ± 2.08×10^3). In finfish hatchery ofBogra, the overall bacterial build up (2.03 ± 0.31×10^8) found in the samples of zeol fish fry in NA plate was significantly higher than that of thecorresponding rearing pond water (2.11 ± 0.459×10^7) and the water of the live food rearing tank (8.43 ± 0.57×10^6). Similar to that, TCBS plates had 2.3-, and 5.09-folds higher bacterial load (1.08± 0.25×10^3) in the samples of fish fry than in the samples of the corresponding water samples and water samples of the live food rearing tank, respectively (4.70 ±w 1.67×10^2 and 2.12 ± 0.28×10^2).

Key words: Bacterial loads, *Vibrio*, Shrimp hatchery, Fish hatchery

Introduction

Bacteriology is one of the most important areas determining the pond/hatchery dynamics and health and hygiene of fish farming system. The present day fish farming is based on nutritive feeds in addition to other management practices. Consequently, the bacteriology of cultured fishes in the tropics is receiving greater attention since some species of bacteria associated with fish cause diseases under stress condition. Fish is in direct contact with microflora in the environment and the opportunistic pathogens already present in the water invade the host under stress (Rekhari *et al.* 2014).

Recent interest on microbial study of aquaculture products also increases the importance of knowledge of microflora associated with fish (Reilly and Kaferstein 1997). Bacterial

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load and bacterial type in shrimp and fish ponds have received attention of researchers (Otta *et al*. 1999, Al-Harvi and Uddin 2007) but little literature is available on the bacterial flora in cultivable fish (Cahill 1990 and Sugita 2006). There is limited literature available on microbiological studies in fresh water fish and the culture environment. The information will be of great value in determining whether there is need to control bacteriological parameters in farming system.

Bacterial flora on fish reflects the aquatic environment which affects the quality and storage life of fishery products (Shewan 1976). It has been repeatedly suggested that the bacterial flora of fish might reflect the bacteriological conditions of the water and a potential indicator of pollution. Therefore, to understand the vulnerability and quality of the hatchery environments and to detect the prevalence of *Vibrio* spp., both total bacterial count and total presumptive *Vibrio* count was taken for the samples using Nutrient Agar (NA) and Thiosulfate Citrate Bile Salts Sucrose (TCBS) media, where the latter is selective for *Vibrio* like species. Marine Agar (MA) is used only for the shrimp hatchery samples to understand the overall density of the marine micro flora.

According to a World Bank report that estimated the global losses due to shrimp diseases are around US$ 3 billion (Lundin *et al.* 2006). Fisheries in both saline water and freshwater are becoming increasingly vulnerable to bacterial infection due to the ease with which pathogens are transmitted in aquaculture. Nevertheless disease outbreaks are being increasingly recognized as a noteworthy impediment on aquaculture production and trade, affecting the economic development of Bangladesh like many other countries. Various infectious diseases caused by bacteria, virus and protozoa are now a primary concern in aquaculture (Rahman *et al.* 2014). Disease outbreak is often directly related with the bacterial density in a particular environment. However, there is no bacteriologicalstudy in the hatchery environment of Bangladesh.

In the present study, three different types of hatcheries (shrimp, tilapia and finfish) were investigated with equal emphasis considering their pervasiveness in Bangladesh. The objective of the present study was to assess the bacteriological status of the sampled shrimp and fish hatchery environments of Bangladesh, in order to comment on the possibility of future disease outbreak.

Materials and Methods

Sampling: A total of 42 samples, 16 of which were from coastal shrimp hatcheries, 14 from tilapia hatchery and 12were from freshwater fish hatcheries, was randomly collected and examined (Table 1). The sampling was done during the period of June 2015 to August 2015. The samples usedwere shrimp (*Penaeus monodon*) post-larvae (PL), brine shrimp nauplii- *Artemia* spp. (live feed), fry of tilapia (*Oreochromis niloticus*), Asisan stinging catfish (*Heteropneustes fossilis*), walking catfish (*Clarias batrachus*) and striped catfish (*Pangasianodon hypophthalmus*), and the culture-water. Samples were collected from three different districts of Bangladesh- Cox's Bazar, Mymensingh and Bogra. All the laboratory investigations were carried out in the Aquatic Laboratory of Department of Fisheries, University of Dhaka. The samples were kept in icebox maintaining temperature at 4^0C and then transferred to the laboratory. All samples were collected following the

method of American Public Health Association (APHA 1998). Fry and PL samples were aseptically grinded in a mortar and blended with physiological saline (0.85% NaCl). All blended samples were kept in a distance to reduce cross contamination.

Bacterial enumeration: Serial dilution technique (APHA 1998) was used for counting the bacterial colonies. 100 µL blended suspension was mixed with 900 µL of sterile saline water in an eppendorf using vortex machine. This process was repeated three more times to get the final 4th dilution from which 100 µL suspension was spread in NA plate and then the plates were kept at 37^{0}C for 24 hours in the incubator. Bacterial colonies grown in the NA media were counted.The same procedure was followed for total bacterial count in MA Plates.

Table 1. List of samples randomly collected from the hatcheries of Cox's Bazar, Mymensingh and Bogra districts of Bangladesh

Shrimp hatchery of Cox's Bazar		Tilapia hatchery of Mymensingh		Finfish hatchery of Bogra	
Sample ID	Sample	Sample ID	Sample	Sample ID	Sample
C1	Artemianauplii from Tank 1	M1	Tilapia fry, 40 days, Big size	B1	Walking catfish fry, 5 days
C2	Artemianauplii from Tank 2	M2	Tilapia fry, 40 days, Medium	B2	Water from the tank of walking catfish, 5 days
C3	Shrimp PL of 10 days, Tank 1	M3	Tilapia fry, 40 days, Small size	B3	Water from live feed pond
C4	Water from PL(10) Tank 1	M4	Tilapia fry, 25 days	B4	Stinging catfish fry, 6 days
C5	Shrimp PL of 10 days, Tank 2	M5	Tilapia fry, 25 days	B5	Water from the tank of stinging catfish, 6days
C6	Water from PL(10) Tank 2	M6	Tilapia fry, 25days	B6	Walking catfish fry, 6 days
C7	Shrimp PL of 12 days	M7	Tilapia fry, 28days	B7	Water from the tank of walking catfish, 6 days
C8	Water from PL(12) Tank	M8	Tilapia fry, 28days	B8	Striped catfish fry, 5 days
C9	Artemianauplii from Tank 1	M9	Tilapia fry, 28days	B9	Water from the tank of striped catfish, 5 days
C10	Artemianauplii from Tank 2	M10	Tilapia fry, 33days	B10	Striped catfish fry, 1 day
C11	Shrimp PL of 8 days from PL (8) Tank	M11	Tilapia fry, 33days	B11	Water from the tank of striped catfish, 1 day
C12	Water from PL(8) Tank	M12	Tilapia fry, 33days	B12	Water from live feed pond
C13	Shrimp PL of 12 daysTank2	M13	Water from Tilapia hapa		
C14	Water from PL(12) Tank 2	M14	Water from Tilapia hapa		
C15	Shrimp PL of 10 days, Tank 3				
C16	Water from PL(10) Tank 3				

100 µL blended raw suspension from each sample was spread in TCBS plate and were kept at 37^0C for 24 hours. Then the *Vibrio* colony count was done.

Statistical analysis: Bacterial density data were transformed into natural log before statistical analysis. The means of bacterial load were compared using ANOVA followed by Tukey's post hoc for multiple comparisons. Statistical software SPSS version 20.0 was used to analyze the data with the level of significance at $p < 0.05$. For plotting the graphs Microsoft Excel (2010) was used.

Results and Discussion

Bacterial density (cfu/g) found in shrimp hatchery of Cox's Bazar in Nutrient Agar (NA) Plate:The bacterial load ($2.59 \pm 0.10\times10^7$) detected in the water sampled from Artemia hatching tank of shrimp hatcheryis similar to the density observed in the shrimp post-larvae (PL) sampled at stage 10 and 12 ($2.37 \pm 0.11\times10^7$ and $2.42 \pm 0.10\times10^7$ respectively; (Fig. 1**)**. However, bacterial load determined from the samples of water corresponding to the stages of PL were similar but different from the samples of Artemia tank and PL stages of 10 and 12. But the bacterial load ($1.38 \pm 0.19\times10^7$) found in the PL 8 stage was different from the load sampled from PL tank water and other PL stages.

Similar bacterial density between Artemiatank and shrimp PL as observed in this study in NA plate could be due to the use of *Artemia nauplii* and shrimp PL as the host organisms. Lower density of total bacteria in the water of PL rearing tank in comparison with the PL, supports the findings of Raoand Surendran(2013) and denotes the absence of host organisms. Progressively higher density of total bacterial abundance in the shrimp PL of stages from 8 to 12 could be due to size variation.

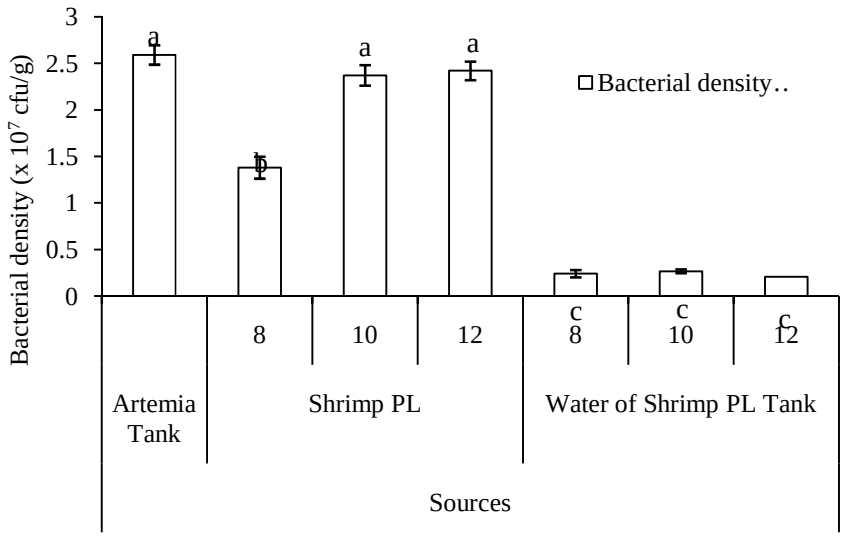


Fig. 1. Bacterial density (cfu/g) found in shrimp hatchery of Cox's Bazar in Nutrient Agar (NA) Plate. Bars (mean ± 1 SEM) with different letters are significantly different (ANOVA, HSD; $P < 0.05$).

Bacterial density (cfu/g) found in shrimp hatchery of Cox's Bazar in Marine Agar (MA) Plate: In MA plate, no significant differences was observed in the bacterial count detected in water sampled from *Artemia* tank and in the PL and corresponding water of PL rearing tank water samples (Fig. 2).

Only few thousands (<5000 cfu/g) bacterial abundance in MA plate across all samples of Artemiatank, shrimp PL and the water of the PL rearing tank could have resulted since the growth was observed in marine selective media. Therefore, the growth was similar across all samples.

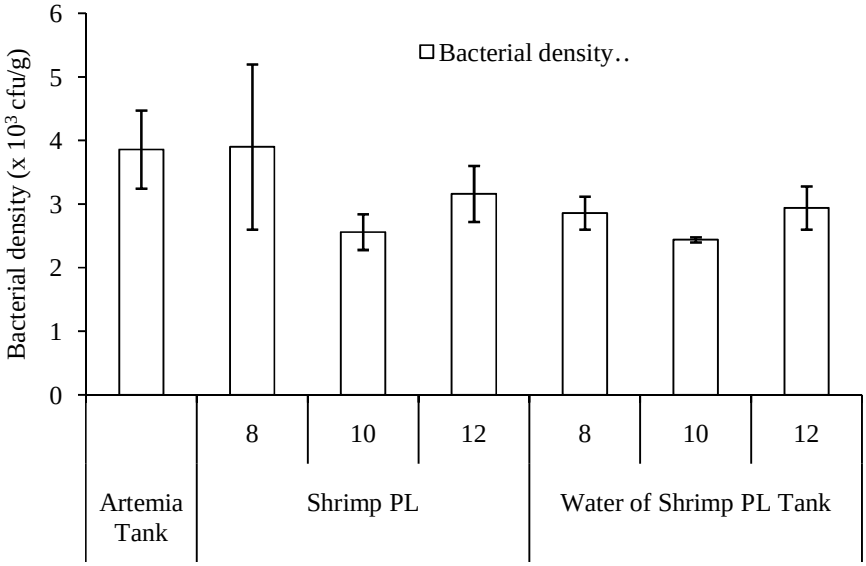


Fig. 2. Bacterial density (cfu/g) found in Shrimp hatchery of Cox's Bazar in Marine Agar (MA) Plate. Bars (mean ± 1 SEM) with no letters indicate no significant difference (ANOVA, HSD; P<0.05).

Bacterial density (cfu/g) found in shrimp hatchery of Cox's Bazar in TCBS Agar Plate: Similar to MA plate, TCBS plates also did not result in any significantly different bacterial density in the water sampled from *Artemia* tank, in the PL and corresponding water sampled from different PL rearing tanks (Fig. 3).

In TCBS plate, bacterial density was found to range between 1620 and 3800 cfu/g which also indicates selective growth of *Vibrio* spp. But shrimp PL had greater mean loads of presumptive *Vibrio* than in their surrounding water body which is similar with the previous reports (Otta *et al.* 2001).

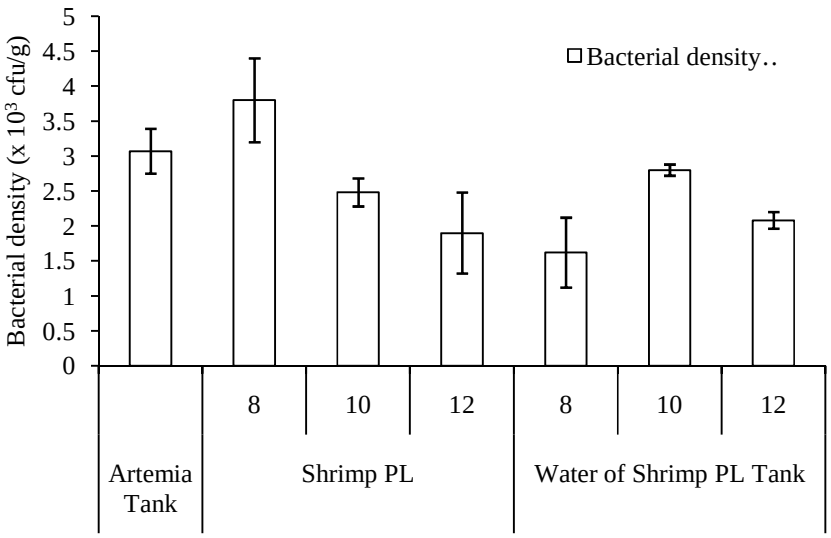


Fig. 3. Bacterial density (cfu/g) found in Shrimp hatchery of Cox's Bazar in TCBS Agar Plate. Bars (mean ± 1 SEM) with no letters denote no significant difference (ANOVA, HSD; P<0.05).

Bacterial density (cfu/g) found in Tilapia Hatchery of Mymensingh in Nutrient Agar (NA) Plate: In NA plate, the bacterial load ($7.5 \pm 0.11\times10^7$) measured in the water sampled from 25 day old fry rearing pond of tilapia was similar to that of 33 day old fry ($8.6 \pm .66\times10^7$; Fig. 4a). The bacterial density found in the 25 ($1.6 \pm 0.50\times10^7$), 28 ($3.12 \pm 0.14\times10^7$) and 40 day old fry ($6.46 \pm 1.52\times10^6$) samples were similar but significantly different from the sample of 33 day old fry and the water sample of the pond of 25 day old fry.

Very high bacterial abundance ($7.55 \pm 0.11\times10^7$) as found in the water of the 25 day old tilapia fry rearing pond could have resulted due to anthropogenic contamination. Interestingly, 40 day old tilapia fry had 10 times lower bacterial build up than in the 33 day old tilapia fry. The reason responsible behind this variation in the total count of bacteria in NA plate is unknown. Typically smaller fish should have lower bacterial load compared to the bigger ones.

Bacterial density (cfu/g) found in Tilapia Hatchery of Mymensingh in TCBS Plate: In TCBS plate, bacterial abundance detected in the samples across all four age groups was similar (25 day old fry: $4.21 \pm 3.79\times10^3$; 28 day old fry: $4.90 \pm 3.50\times10^3$; 33 day old fry: $1.08 \pm 0.12\times10^3$; 40 day old fry: $7.04 \pm 2.08\times10^3$; Fig. 4b). No bacterial count was found in the water sampled from 25 day old fry rearing pond.

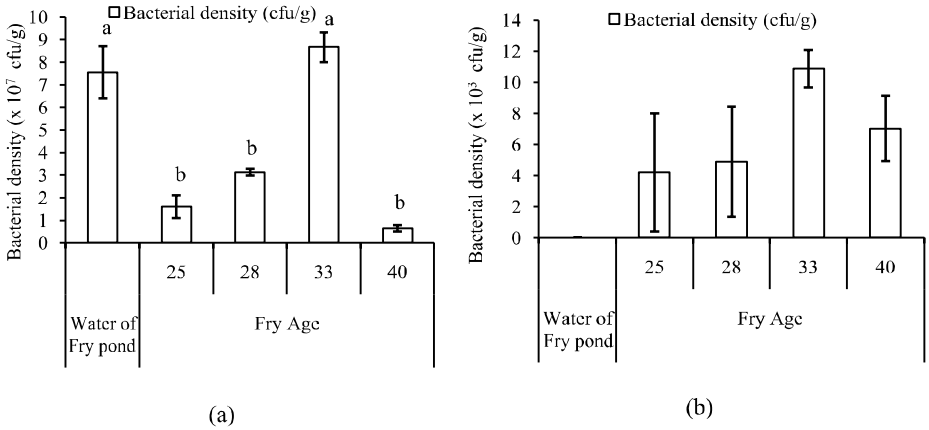


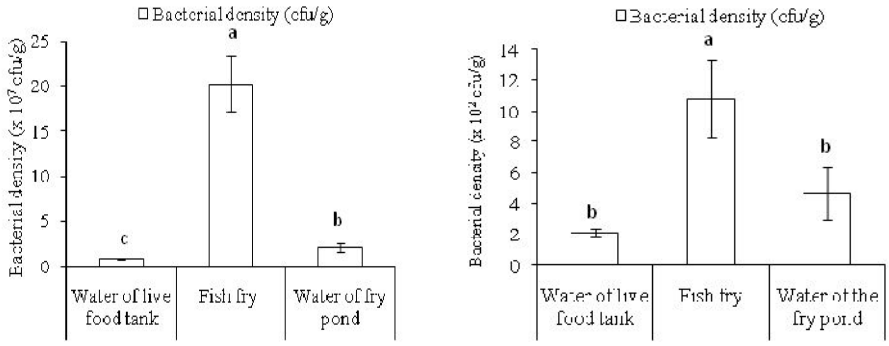
Fig. 4. Bacterial density (cfu/g) found in the tilapia fry rearing pond water and in the fries of 25, 28, 33 and 40 days old in Tilapia Hatchery, Mymensingh in (a) NA plate and (b) TCBS plate. Bars (mean ± 1 SEM) with different letters are significantly different (ANOVA, HSD; P<0.05).

In TCBS plate, no growth of any bacteria denotes absence of *Vibrio* spp. in 25 day old tilapia fry rearing pond water. This absence of *Vibrio* in the fry rearing pond water indicates no contamination from anthropogenic sources as well as denotes the quality level of the fry feed. However, tilapia fry aged from 25-40 day old had similar *Vibrio* growth that indicates the possibility of later contamination from unknown sources. Even environmental parameters such as temperature, salinity, pH and dissolved oxygen play a foremost part in the distribution of bacteria (Palaniappan 1982 and Parvez *et al*. 2015).

Overall Bacterial density (cfu/g) found in finfish hatchery of Bogra in Nutrient Agar (NA) Plate:

The overall bacterial build up ($2.03 \pm 0.31\times10^8$) found in the samples of fish fry in NA plate was significantly higher than that of the corresponding rearing pond water ($2.11 \pm 0.459\times10^7$) and the water of the live food rearing tank ($8.43 \pm 0.57\times10^6$; Fig. 5a).

Nearly 10 and 20 times higher total bacterial abundance in the fry of striped catfish, walking catfish and stinging catfish compared to that of live food rearing tank water and the water of fry rearing ponds in NA plate could also be responsible for the established fact that fish body carries higher microbial organisms than that of surrounding water body. This might be due to the high organic load in the incoming water (Otta *et al.* 2001). The same reasons could also be responsible for the presumptive *Vibrio* spp. growth in TCBS plates of the corresponding samples.



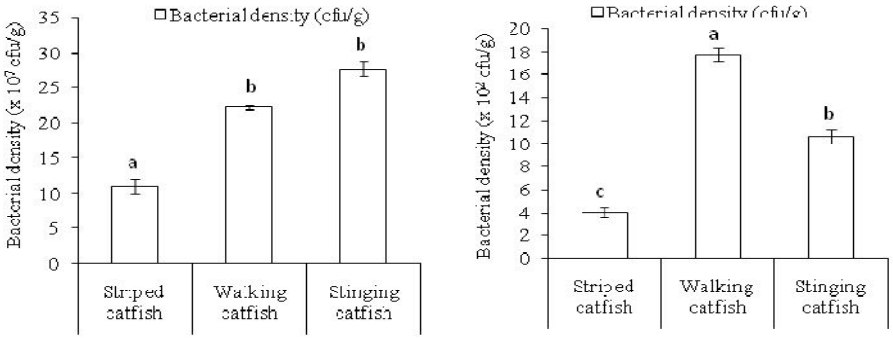
(a) (b)

Fig. 5. Overall bacterial load (cfu/g) detected in (a) NA plate and (b) TCBS plate from the samples of water of the live food tank, fry of fishes (stinging catfish, walking catfish, and striped catfish) and water of the fry rearing pond of finfish hatchery of Bogra. Bars (mean ± 1 SEM) with different letters indicate significant difference (ANOVA, HSD; P<0.05).

Overall Bacterial density (cfu/g) found in finfish hatchery ofBogra in TCBS plate: Similar to the overall bacterial density found in NA plate, TCBS plates had 2.3-, and 5.09-folds higher bacterial load ($1.08\pm 0.25\times10^3$) in the samples of fish fry than in the samples of the corresponding water samples and water samples of the live food rearing tank, respectively ($4.70 \pm 1.67\times10^2$ and $2.12 \pm 0.28\times10^2$; Fig. 5b).

Bacterial density (cfu/g) found in fish fry offinfish hatcheryofBograin NA Plate: In NA plate, the bacterial load found in stinging catfish fry ($2.77 \pm 0.11\times10^8$) samples was similar to the density of walking catfish fry ($2.23 \pm 0.03\times10^8$) but significantly higher than in the sample of striped catfish fry ($1.10 \pm 0.10\times10^8$; Fig.6a).

Particularly the reason of significant differences in total bacterial load in NA plate between the striped catfish fry and the near similar walking catfish and stinging catfish fry could be the discrepancy in their hardiness. The hardier the fish species the more bacterial load it may contain.



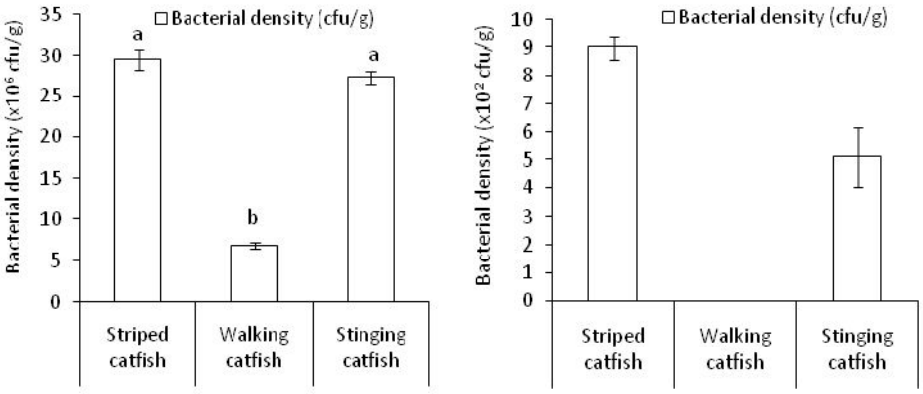
(a) (b)

Fig. 6. Bacterial load detected in (a) NA plate and (b) TCBS plate in the samples of striped catfish, walking catfish and stinging catfish fry of finfish hatchery of Bogra. Bars (mean ± 1 SEM) with different letters are significantly different (ANOVA, HSD; P<0.05).

Bacterial density (cfu/g) found in finfish hatchery of Bogra in TCBS Plate: In TCBS plate, while walking catfish fry had the highest density of bacteria ($1.78 \pm 0.06\times10^3$) the lowest density ($4.05 \pm 0.45\times10^2$) was detected in the striped catfish fry (Fig.6b). However, stinging catfish fry resulted in the bacterial build up ($1.60 \pm 0.06\times10^3$) that was significantly lower than that of walking catfish fry but higher than did the striped catfish fry.

The statistical significant difference in the amount of *Vibrio* like bacteria of the striped catfish, walking catfish and stinging catfish fry samples in TCBS plate also advocates for the previously stated reason.

Bacterial density (cfu/g) found in finfish hatchery ofBogra in NA Plate: In NA plate, while water of striped catfish and stinging catfish fry rearing pond water had similar (striped catfish: $2.95\times10^7 \pm 0.13\times10^7$; stinging catfish: $2.72\times10^7 \pm 0.07\times10^7$) but significantly higher bacterial load that did walking catfish fry rearing pond water ($6.76\times10^6 \pm 0.34\times10^6$; Fig.7a).



(a) (b)

Fig. 7. Bacterial load (cfu/g) counted in (a) NA plate and (b) TCBS plate from the water samples of the corresponding striped catfish, walking catfish and stinging catfish fry rearing pond in finfish hatchery, Bogra. Bars (mean ± 1 SEM) with different letters are significantly different (ANOVA, HSD; $P<0.05$) and bars with no letters denote no significant difference

Bacterial density (cfu/g) found in finfish hatchery of Bogra in TCBS Plate: In TCBS plate, while the water of walking catfish fry rearing pond had no bacterial load, the stinging catfish and striped catfish fry rearing pond water had $5.10 \pm 1.10\times10^2$ and $9 \pm 0.04\times10^2$, respectively (Fig. 7b).

The bacterial load in NA plate of the water samples of walking catfish fry rearing pond is significantly different from the water samples striped catfish and stinging catfish fry rearing pond. Water of walking catfish fry rearing pond has lowest both TBC and TVC in NA and TCBS plates, respectively. The reason behind this variation perhaps lies on the

treatments applied and the quality of the water of the corresponding fish fry rearing ponds. However, the mean total bacterial density in the fry rearing ponds exceeded the reported range of Otta *et al.* 2001; while the mean total presumptive *Vibrio* count was a bit lower than their recommended values. Better water management systems adopted by hatcheries might play important role in this respect (Rao *et al.* 2013).

Under stressful conditions, bacteria may become opportunistic and attack the body tissue and produce disease. The need is thus felt to monitor and regulate the bacterial parameters in the present aquaculture system where lot of management is done to enhance production. The high density of total bacteria in the fresh water hatcheries demands molecular analysis of these species to investigate the presence of potential probiotics that may use in shrimp/finfish aquaculture and of opportunistic fish or shrimp pathogens in that community.

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EFFECTS OF GIBBERELLIC ACID (GA3) ON GROWTH AND YIELD PARAMETERS OF FRENCH BEAN (PHASEOLUS VULGARIS L.)

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Abstract

A field study was conducted during the Rabi season of 2009-2010 in the research field of Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhaka. Six levels of GA_3, *viz.* 0, 30, 50, 70, 90 and 110 ppm were sprayed at 18 days after sowing (DAS). GA_3 treatments significantly increased plant height than the control plants. GA_3 with 30 to 90 ppm significantly increased number of branches and leaves, leaf area, leaf area index (LAI), leaf dry matter and total dry matter at different growth stages. GA_3 at 30 to 70 ppm gradually increased crop growth rate (CGR), net assimilation rate (NAR) and relative growth rate (RGR) and declined advanced growth stages. Number of dry pods /plant, number of seeds /pod, 1000 seed weight, fresh fodder, fresh pod, dry seed yield and harvest index also significantly increased. Positive significant correlations were found among growth parameters and as well as yield contributing characters.

Key words: French bean, GA_3, LAI, NAR, yield, HI

Introduction

French bean is best known and widely cultivated as field crop. Its green pods and grains are consumed as vegetables and dry seeds as a pulse. Fresh leaves along with shoot also have fodder value. It is widely cultivated in tropical and subtropical region. In Bangladesh, it is grown in Sylhet, Chittagong, Chittagong Hill Tracts, Cox's Bazar and Comilla (Rashid 1999).

French bean plays a key role in crop rotation due to their ability to fix nitrogen through symbiotic association with bacteria, *Rhizobium* forms nitrogen fixing root nodules, which are agronomically significant (Burns and Hardy 1975). Some Kharif crops like Aus paddy HYV (mid March to August), jute, sun hemp, Lady's finger, bitter gourd, snake gourd, white gourd and sesame can easily cultivated as alternate crops (BBS 2011). It is a new crop in Bangladesh. The production quality of it is low in this country due to lack of modern agricultural practices.

Gibberellic acid (GA_3) is the most widely used plant growth regulator which increases stem elongation along with plant height, growth, dry matter accumulation as well as yield in various crops (Harrington *et al.* 1996, Akter *et al*. 2007 and Emongor 2007). However, very few works have been done on the application of GA_3 on French bean in Bangladesh (Noor 2014). Therefore, the present investigation was undertaken to study the effect of GA_3 on plant growth, dry matter and yield parameters of French bean.

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Materials and Methods

A field experiment was conducted in the Rabi season during the period from November 2009 to February 2010 at the research field of Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhaka. The field is located at latitude 23°44′23.3″ N and longitude 90°23′03.8″ E at altitude of 4 m above the sea level. The soil sample was tested at Soil Research and Development Institute (SRDI), Dhaka. The soil was a silty loam having pH 7.3, and low amount of chemical composition, *i*. e., 1.05% organic matter, including 0.056% total N, 10.17, 3.1and 0.25 meq/100g soil Ca, Mg and K respectively. Other elements like P, S B, Cu, Fe, Mn and Zn were 20.9, 10.3, 0.27, 5.7 ,147.6, 76.1and 3.1 µ.g/g soil respectively. Based on the organic and elemental condition N, P, K, S and B were applied at the rate of 90, 35, 30, 10 and 1Kg /ha in the form of urea, triple super phosphate, muriate of potash, gypsum and borax, respectively as per the recommendation of Fertilizer Recommendation Guide (2005). Among these fertilizers, P, B and S were applied entirely as basal doses. One third of N and K were applied as basal doses, rest two third were added as top dressing at 20 and 40 days after sowing (DAS), respectively. Furrow irrigation was given at an interval of 7 days. Thinning and weeding were done at an interval of 10 days.

The experiment was laid out in a randomized complete block design (RCBD) with three replications. The unit plot size was 2.0 m × 1.5 m. Seeds of French bean cv. BARI bush bean-1 were sown on 15 November, 2009. The distance between line and seed were 30 and 15 cm, respectively. The six treatments applied were as follows; Control (distilled water spray) and 30, 50, 70, 90 and 110 ppm GA_3. The treatments were applied as foliar spray at 18 DAS.

Ten plants selected at random were cut at the above ground level. Data on growth parameters were collected at 5 different growth stages, *i.e*. 18, 28, 38, 48 and 58 DAS for 4 leaves, budding, flowering, pod setting and pod filling, respectively. Leaf area index (LAI) was calculated by the formula of Hunt (1981). Whole aerial parts of plants were oven dried at 70°C till constant weight and then the dry weights were taken and used as leaf dry matter and total dry matter (TDM). CGR, NAR and RGR were calculated according to Brown (1984) and Radford (1967), respectively. Young pods from 10 sampled plants of each treatment was collected from 48 to 68 DAS at an interval of 7 days for measurement of fresh pod yield/plant. After harvesting of pods at 68 DAS, the remaining whole green shoot along with leaves were harvested for fresh fodder yield/plant. Number of dry pods/plant, number of seeds/pod, seed yield/plant and harvest index (HI) were measured from the demarcated areas according to Ullah (2006) and Noor (2014). Data were statistically analyzed by Duncan's Multiple Range Test (DMRT) and LSD test at 5% level of significance (Gomez and Gomez 1984).

Results and Discussion

The height of French bean (cv. BARI bush bean 1) plant treated with GA_3 significantly increased. The magnitude of increase of plant height was found to be more pronounced in treatment with 50 ppm GA_3, followed by 70 to 110 ppm of the same (Table 1). At 58 DAS, the highest plant height, 59.40 cm was recorded for 50 ppm GA_3, whereas the lowest plant height 38.67 cm was found for the control.GA_3 enhances growth activities of plant, stimulates the rate of cell division, cell elongation, and thus, also contributes to internode and stem elongation (Taiz and Zeiger 2002). The results of the present study are in agreement with those of mustard plant reported by Akter *et al*. (2007). They recorded the highest plant height (95.77 cm) with the application of 50 ppm GA_3, which was statistically similar to 75 ppm GA_3 treatment and, the lowest (77.63 cm) was found for the control.

Table 1. Effects of GA_3 on plant height (cm) and number of leaves/plant of French bean at different growth stages.

Treatments	Plant height (cm)					Number of leaves/plant				
	Days after sowing (DAS)					Days after sowing (DAS)				
	18	28	38	48	58	18	28	38	48	58
Control	9.97c	25.30c	34.63c	36.73c	38.67d	4.000	9.00b	10.33c	11.33c	10.33c
30 ppm GA_3	10.05b	33.10b	43.63b	48.63b	51.17c	4.000	10.00a	11.67ab	12.67b	12.00b
50 ppm GA_3	10.06a	45.43a	56.40a	57.90a	59.40a	4.000	10.33a	12.00a	14.00a	13.00a
70 ppm GA_3	10.05b	45.47a	56.33a	57.62a	59.04a	4.000	10.00a	11.67ab	13.00ab	12.00b
90 ppm GA_3	10.05b	45.47a	56.30a	57.50a	58.93a	4.000	9.67ab	11.67ab	12.67b	11.67b
110 ppm GA_3	10.05b	45.50a	56.27a	57.23a	57.80b	4.000	9.67ab	11.00bc	12.33bc	11.67b
LSD (0.05)	0.00	0.53	1.88	0.92	0.34	0.00	0.24	0.32	0.56	0.24
CV (%)	0.31	20.58	17.41	15.34	14.24	0.00	5.61	6.13	7.66	7.46

*Mean in a vertical column followed by same letter do not differ significantly at 5% level.

A gradual increase in number of leaves was observed from 18 to 48 DAS and thereafter it was found to decline for all treatments (Table 1). The maximum number of leaves per plant was recorded for 50 ppm GA_3, followed by 70 and 30 ppm. 50 ppm GA_3 treated plants produced 14.81, 16.13, 23.53 and 25.81% more leaves per plant compared to those of the controls at 28, 38, 48 and 58 DAS, respectively. Similarly, Sarkar *et al*. (2002) observed that GA_3 at 100 ppm concentration in treated soybean plants produced higher number of leaves at the later stages of 60 and 80 DAS. It might be due to GA_3 promote cell enlargement and cell division that enhance plant height, number of branches and number of leaves.

Number of branches /plant gradually increased from 18 to 48 DAS and reached a steady condition (Fig. 1). GA_3 with 30 to 110 ppm concentrations significantly increased number branches per plant at 28 to 48 DAS. The highest number of branches per plant was obtained for 50 ppm GA_3 treated plants, which was 21.43 to 23.08 % more at 28 to 48 DAS over the control. The results were in consonance with the works of Abdul *et al*.

(1988) who noticed significantly increased number of branches per plant by increasing the concentration of GA_3 (50 to 100 ppm) in pepper.

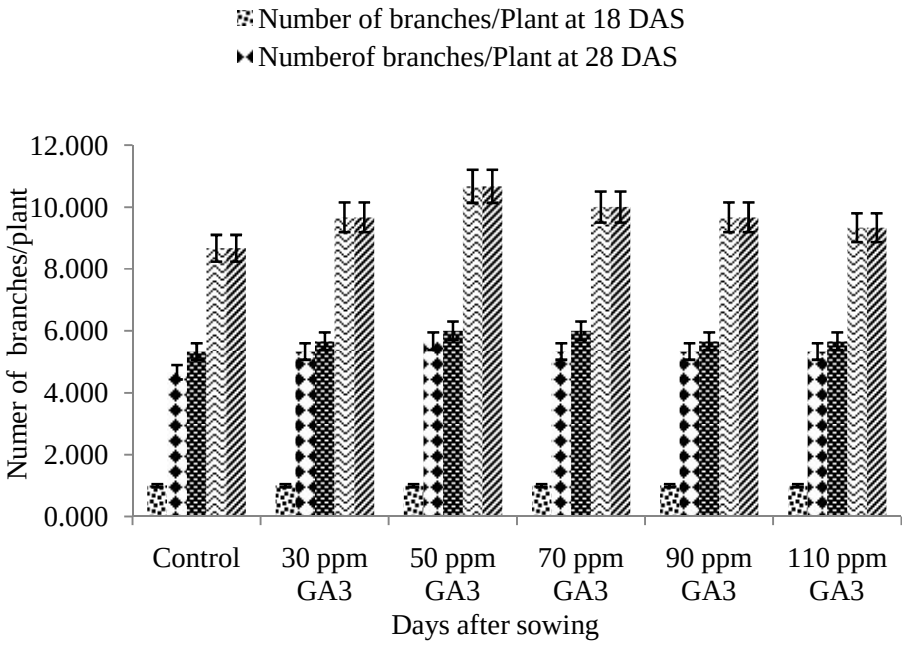

Fig. 1. Effects of GA_3 on number of branches /plant of French bean at different growth stages.

Results presented in Table 2 revealed that leaf area gradually increased from 18 to 48 DAS for all the treatments. Leaf area significantly increased over the control following all the GA_3 treatments at different growth stages. 50 ppm GA_3 increased the leaf area by 22.66, 31.17, 22.88 and 22.67% over those of the control at 28, 38, 48 and 58 DAS, respectively.

Results presented in Table 2 revealed that leaf area gradually increased from 18 to 48 DAS for all the treatments. Leaf area significantly increased over the control following all the GA_3 treatments at different growth stages. 50 ppm GA_3 increased the leaf area by 22.66, 31.17, 22.88 and 22.67% over those of the control at 28, 38, 48 and 58 DAS, respectively.

Leaf area index (LAI) varied from 1.27 to 1.56, 2.52 to 3.31, 2.81 to 3.45 and 2.68 to 3.28 at 28, 38, 48 and 58 DAS, respectively (Table 2). Among the treatments, 50 ppm GA_3 showed significantly highest LAI, whereas the lowest was found for the control. GA_3 induced higher leaf area and LAI were reported in tomato plants (Khan *et al*. 2006) and in rice plants (Liu *et al*. 2012).

Table 2. Effects of GA_3 on Leaf Area (cm^2/plant) and Leaf Area Index (LAI) of French bean at different growth stages.

Treatments	Leaf Area (cm^2/plant)					Leaf Area Index (LAI)				
	Days after sowing (DAS)					Days after sowing (DAS)				
	18	28	38	48	58	18	28	38	48	58
Control	81.80	572.33f	1134.89f	1263.67f	1204.67f	0.18	1.27f	2.52e	2.81f	2.68f
30 ppm GA_3	81.81	632.19c	1277.57c	1442.79c	1403.46c	0.18	1.41c	2.84c	3.21c	3.12c
50 ppm GA_3	81.82	702.00a	1488.62a	1552.75a	1477.75a	0.18	1.56a	3.31a	3.45a	3.28a
70 ppm GA_3	81.81	657.44b	1356.59b	1480.82b	1417.82b	0.18	1.46b	3.02b	3.29b	3.15b
90 ppm GA_3	81.81	618.86d	1266.66d	1393.19d	1334.19d	0.18	1.38d	2.82c	3.10d	2.97d
110 ppm GA_3	81.81	607.52e	1255.00e	1334.52e	1295.19e	0.18	1.35e	2.79d	2.97e	2.88e
LSD (0.05)	0.00	26.30	53.50	43.17	41.64	0.00	0.0001	0.0003	0.0002	0.0002
CV (%)	0.01	6.63	8.56	6.91	6.80	0.00	6.63	8.56	6.91	6.80

*Mean in a vertical column followed by same letter do not differ significantly at 5% level.

Results presented in Table 3 showed that leaf dry matter gradually increased from 18 to 58 DAS. GA_3 significantly increased leaf dry matter from 17.61 to 40.65% at 58 DAS in comparison to the control. Among the treatments, GA_3 at 50 ppm level produced the highest leaf dry matter. For 50 ppm GA_3 treatment, the leaf dry matter was found to increase by 36.24, 50.50, 44.00, and 40.65 % over those of the control at 28, 38, 48 and 58 DAS, respectively. These findings are in agreement with those reported by Ali *et al*. (2012) for *Hibiscus sabdariffa* L. where application of GA_3 increased the shoot and root dry weight compared to other treatments.

Total dry matter was found to increase gradually by GA_3 from 18 to 58 DAS (Table 3). All the GA_3 treatments significantly increased it at 28, 38, 48 and 58 DAS. The highest total dry matter of 36.15 g/plant was obtained for 50 ppm GA_3, whereas, the lowest of 21.19 g/plant was found for the control at 58 DAS. GA_3 at 50 ppm increased the total dry matter by 35.71, 51.37, 60.99 and 70.61 % over the control at 28, 38, 48 and 58 DAS, respectively and it was found to be significantly different from all other treatments. These results are more or less similar to the findings of Akter *et al*. (2007), who observed a significant variation in total dry matter due to the application of different levels of GA_3 in mustard. The exogenous application of GA_3 increased growth parameters like plant height, number of branches, number of leaves, leaf area, leaf dry matter, and along with total dry matter.

Table 3. Effects of GA_3 on Leaf dry matter (g/plant) and total dry matter (g/plant) of French bean at different growth stages.

Treatments	Leaf dry matter (g/plant)					Total dry matter (g/plant)				
	Days after sowing (DAS)					Days after sowing (DAS)				
	18	28	38	48	58	18	28	38	48	58
Control	0.289b	3.132f	6.418e	7.624e	5.746d	0.765	4.21d	9.38f	16.37f	21.19f
30 ppm GA_3	0.292a	3.347c	7.051c	8.375c	7.570b	0.769	4.53c	10.52c	19.09c	25.46c
50 ppm GA_3	0.292a	4.267a	9.659a	10.979a	8.082a	0.771	5.71a	14.20a	26.36a	36.15a
70 ppm GA_3	0.292a	3.486b	7.446b	8.781b	7.599b	0.770	4.73b	11.19b	20.31b	27.52b
90 ppm GA_3	0.292a	3.298d	6.932d	7.901d	7.066c	0.768	4.44c	10.32d	18.38d	24.47d
110 ppm GA_3	0.292a	3.261e	6.929d	7.897d	6.758c	0.768	4.39c	10.19e	17.91e	23.74e
LSD (0.05)	0.000	0.000	0.000	0.002	0.065	0.000	0.01	0.00	0.00	0.01
CV (%)	0.394	11.098	14.608	13.551	11.099	0.260	10.96	14.46	16.63	18.48

*Mean in a vertical column followed by same letter do not differ significantly at 5% level.

Data from Table 4 showed that Crop growth rate (CGR) gradually increased from 18 – 28 to 38- 48 DAS and thereafter decreased in advanced growth stages. The highest CGR was registered for 50 ppm GA_3, followed by 70 ppm GA_3, and these were significantly different from all other treatments. However, the lowest was found for the control. It was found to significantly increased by 43.46, 64.10, 73.92 and 103.31 % for 50 ppm GA_3 in comparison with the control at 18 - 28, 28 - 38, 38 - 48 and 48 - 58 DAS, respectively. The data on net assimilation rate (NAR) was highest at 18 – 28 DAS, and thereafter started to decrease in advanced growth stages (Table 4). NAR was found to be highest for 50 ppm GA_3 among all the treatments and increased significantly by 25.38, 28.86, 37.06 and 65.60 % over the control.

Table 4. Effects of GA_3 on crop growth rate ($g/m^2/day$), net assimilation rate ($g/m^2/day$) and relative growth rate (g/g/day) of French bean at different growth stages.

Treatments	Crop growth rate ($g/m^2/day$)				Net assimilation rate ($g/m^2/day$)				Relative growth rate (g/g/day)			
	Days after sowing (DAS)				Days after sowing (DAS)				Days after sowing (DAS)			
	18-28	28-38	38-48	48-58	18-28	28-38	38-48	48-58	18-28	28-38	38-48	48-58
Control	7.65d	11.50e	15.53f	10.70f	13.65c	6.30c	5.83f	3.90d	0.170d	0.080c	0.056d	0.026e
30 ppm GA_3	8.36c	13.30c	19.06c	14.14c	13.97bc	6.53b	6.32c	4.47c	0.177c	0.084b	0.060b	0.029c
50 ppm GA_3	10.98a	18.87a	27.00a	21.76a	17.12a	8.12a	7.99a	6.46a	0.200a	0.091a	0.062a	0.032a
70 ppm GA_3	8.80b	14.36b	20.27b	16.01b	14.33b	6.70b	6.43b	4.97b	0.181b	0.086b	0.060b	0.030b
90 ppm GA_3	8.16c	13.07cd	17.91d	13.54d	13.83bc	6.51bc	6.06d	4.47c	0.176c	0.084b	0.058c	0.028d
110 ppm GA_3	8.06c	12.89d	17.15e	12.96e	13.83bc	6.50bc	5.96e	4.44c	0.174c	0.084b	0.056d	0.028d
LSD (0.05)	0.06	0.06	0.004	0.03	0.16	0.02	0.001	0.003	0.000	0.000	0.000	0.000
CV (%)	13.08	17.20	19.40	24.02	8.82	9.39	11.63	17.46	5.584	4.443	3.932	6.599

*Mean in a vertical column followed by same letter do not differ significantly at 5% level.

Relative growth rate (RGR) was found to range from 0.170 - 0.200, 0.080 - 0.091, 0.056 - 0.062 and 0.026 - 0.032 g/g/day at 18 - 28, 28 - 38, 38 - 48 and 48 - 58 DAS, respectively (Table 4). The highest RGR were observed for 50 ppm GA_3 treated French bean plants at all the growth stages, which were significantly different from all other treatments. GA_3 at 50 ppm increased it by 17.42, 13.75, 10.71 and 23.08 % over the control at 18 - 28, 28 - 38, 38 - 48 and 48 - 58 DAS, respectively. These were found to be in agreement with the results in a number of crops where CGR, NAR and RGR are increased in early growth and juvenile stages and thereafter began to decline. Maske *et al*. (1998) reported that for soybean GA_3 effectively increased CGR from 30 - 45 to 45 - 60 DAS and enhanced the yield contributing components. They also reported that the foliar application of GA_3 at 30 DAS had the most regulatory effect in increasing root, stem, leaf and total dry matter, LAI, CGR, RGR and NAR in soybean (cv. PB - 1). Sarkar *et al*. (2002) reported that soybean plants treated with GA_3 significantly increased CGR, RGR and NAR at 60 DAS compared to the control. Similarly, GA_3 treatments (50 - 200 ppm) on either seeds or leaves increased TDM, LAI, RGR and NAR (Rahman *et al*. 2004).

Fresh fodder yield /plant varied from 56.20 to 73.40 g (Table 5). Significantly highest fodder yield/plant was recorded for 50 ppm GA_3 and the increase was 30.61% over the control. On the other hand, the lowest value was found for the control plants. GA_3 increased vegetative growth which can be used as fodder (Emongor 2007, Sarkar *et al*. 2002, Rahman *et al*. 2004, Azizi *et al*. 2012 and Liu *et al*. 2012).

Table 5. Effects of GA_3 on yield attributes of French bean .

Treatments	Fresh fodder yield/plant (g)	Number of dry pods/plant	Number of seeds/ pod	1000 seed weight (g)	Fresh pod yield/plant (g)	Seed yield/plant (g)	Harvest index (%)
Control	56.20f	12.40d	5.81b	213.50d	69.26e	13.22d	54.18c
30 ppm GA_3	66.67c	14.00c	6.00a	232.27b	123.35c	17.60bc	60.55ab
50 ppm GA_3	73.40a	16.23a	6.10a	235.47a	158.37a	20.87a	63.76a
70 ppm GA_3	70.60b	14.62b	6.02a	232.58b	130.41b	18.60b	62.00a
90 ppm GA_3	62.67d	13.90c	6.00a	227.15c	117.63d	16.86c	59.82b
110 ppm GA_3	58.47e	13.90c	5.97a	216.37c	115.53d	16.65c	59.77b
LSD (0.05)	0.02	0.10	0.10	2.7	6.89	0.841	1.96
CV (%)	9.84	7.35	1.53	3.78	22.88	10.98	4.11

*Mean in a vertical column followed by same letter do not differ significantly at 5% level.

It was observed that number of dry pods per plant ranged from 12.40 to 16.23 (Table 5). It was significantly higher for 50 ppm GA_3 (16.23), followed by 70 ppm GA_3 (14.62) and 30 ppm GA_3 (14.00). Number of dry pods per plant increased by 12.07 to 30.88 % over the control following different GA_3 treatments. Enhanced number of fruits /plant due to GA application were reported by Sarkar *et al*. (2002) in soybean and Khan *et al*. (2006) in tomato.

Number of seeds per pod was highest for 50 ppm GA_3 which was statistically at par with 70, 30, 90 and 110 ppm GA_3 (Table 5). These results are in agreement with those of Akter *et al*. (2007), who reported that number of seeds per siliqua was significantly influenced by different levels of GA_3 treatments and, the highest number of seeds/siliqua was obtained for 50 ppm GA_3.

Thousand seed weight ranged from 213.50 to 235.47 g (Table 5). Significantly maximum 1000 seed weight was registered for 50 ppm GA_3 which was 10.29% higher compared to the control. It was found that all the GA_3 treatments significantly increased 1000 seed weight, which is in agreement with the studies of Emongor (2007) for cowpea and Tiwari *et al*. (2011) for rice.

Data from Table 5 showed that fresh pod yield /plant ranged between 69.26 and 158.37g and the maximum was recorded due to 50 ppm GA_3. All the GA_3 treatments significantly increased fresh pod yield/plant by 66.82 to 128.67% compared to the control. Significant increase of fruit yield in many crops due to the application of different concentrations of GA_3 was reported by many investigators *viz.* Hye *et al*. (2002) in onion and Khan *et al*. (2006) in tomato. This might be due to an inhibition of vegetative growth and thus making available the food reserves for developing fruits, which was evident from the significantly increased number of pods per plant and fresh pod yield of French bean.

Seed yield /plant ranged from 13.22 to 20.87 g (Table 5). Significantly the highest seed yield/plant was obtained from 50 ppm GA_3 followed by 70 ppm. That GA_3 with different concentrations significantly enhanced the seed yield in many crops were reported by a number of researchers *viz.* Sarkar *et al*. (2002) for soybean and Tiwari *et al*. (2011) for rice. GA_3 may induce the development of xylem and phloem and in turn, increase the flow and deposition of assimilation products in seeds (Secer 1989).

Harvest index (HI) varied from 54.18 to 63.76 % (Table 5). All the GA_3 treatments significantly increased HI by 10.31 to 17.68% where 50 ppm GA_3 treated plants produced maximum over the control. Positive influences of GA_3 on HI was reported by Emongor (2007) for cowpea .

The results of correlation study showed that there were positive and significant correlations among growth and yield parameters of French bean (Table 6). The ‘r’ values of LAI, CGR, NAR, fresh pod yield/plant and fresh fodder yield/plant with TDM were 0.902**, 0.999**, 0.948**, 0.881**and 0.876**, respectively. And the ‘r’ values of LAI, TDM, CGR, NAR and fresh fodder yield/plant with fresh pod yield/plant were 0.939**, 0.881**, 0.875**, 0.741** and 0.881**, respectively.

Positive correlation among yield parameters like number of seeds/pod, 1000 seed weight, seed weight /plant and seed yield/ha were reported by Ullah *et al*. (2007) in cowpea and Azizi *et al*. (2012) in soybean. Hasanuzzaman *et al*. (2007) found positive relationship among plant height, number of branches /plant, number of pods /plant, 1000 - seed weight and yield of chickpea.

Table 6. Correlation among leaf area index (LAI), total dry matter (TDM), crop growth rate (CGR), net assimilation rate (NAR), fresh pod yield/plant, fresh Fodder yield/plant.

Parameters	Leaf Area Index at 48 DAS	Total Dry Matter at 58 DAS	Crop Growth Rate at 38-48 DAS	Net Assimilation Rate at 18- 28 DAS	Fresh pod yield/plant	Fresh fodder yield/plant at 68 DAS
Leaf Area Index at 48 DAS	1	0.902 **	0.908 **	0.753 **	0.939 **	0.986 **
Total Dry Matter at 58 DAS		1	0.999 **	0.948 **	0.881 **	0.876 **
Crop Growth Rate at 38-48 DAS			1	0.945 **	0.875 **	0.885 **
Net Assimilation Rate at 18- 28 DAS				1	0.741 **	0.730 **
					**	**
Fresh Pod yield/plant					1	0.881 **
Fresh fodder yield/plant at 68 DAS						1

** Significant at 1 % level.

The regression value of R^2= 0.9628, showed that there was a significant, positive and linear relationship between number of dry pods/plant and seed yield/plant (Fig. 2). Moreover, significant, positive and linear relationship (R^2= 0.9685) was found between harvest index (HI) and seed yield/plant (Fig. 3). Similar trend was reported by Hasanuzzaman *et al*. (2007) in chickpea. Linear regression with positive significant relationship for yield and GA_3 concentrations in cowpea was reported by Emongor (2007).

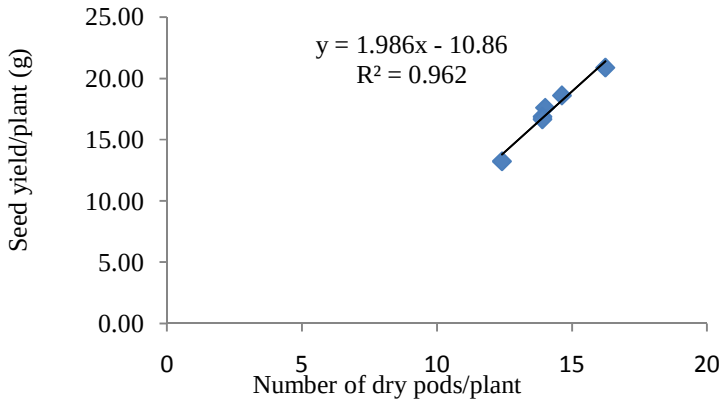

Fig. 2. Relationship between seed yield /plant and seed yield/plant of French bean.

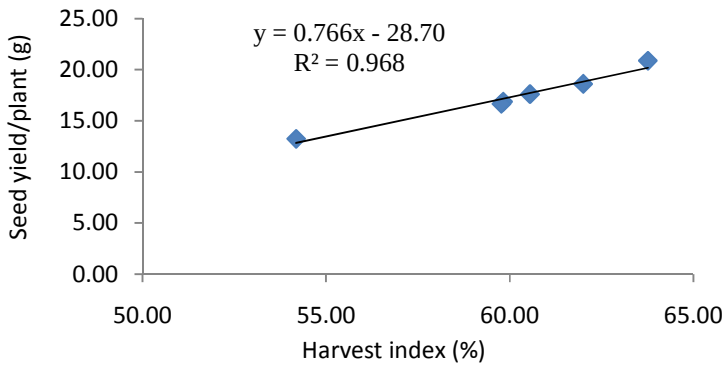

Fig. 3. Relationship between seed yield /plant and harvest index (%) of French bean.

From the results of the study it is apparent that GA_3 had significant effect on growth and yield parameters of French bean. GA_3 at 50 ppm was found to be optimum concentration for the highest growth and yields (fresh pod, dry seed and fodder yield /plant) of French bean.

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WATER QUALITY ASSESSMENT ALONG WITH POLLUTION SOURCES OF THE HALDA RIVER

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Abstract

In the present study water quality assessment and land based sources of pollutants discharged into the Halda River through four major canals were investigated. The ranges of pH, DO, BOD, NO_3-N, PO_4-P, chloride and total alkalinity of the collected samples varied from 7.1-8.8, 3.35-4.70 mg/L, 0.055-5.0 mg/L, 0.12-3.1 mg/L, 0.06-0.16 mg/L, 8.4-69.30 mg/L and 73-220 mg/L, respectively. pH, DO, BOD showed significant changes at Mondakini Canal and Chengkhali Canal for monsoon, post-monsoon and winter ($p < 0.05$) while chloride at Chengkali Canal for all seasons ($p \leq 0.05$). Moreover, TA, NO_3-N and PO_4-P exhibited significant variation in the concentration at Mondakini Canal, Madari Canal and Chengkali Canal for three seasons ($p \leq 0.05$). River Pollution Index indicated that the water of river at Mondakini Canal, Madari Canal, Chengkhali Canal and Khondakia Canal varied from low to moderate pollution. According to the interviewers, the Halda River is being polluted due to industrial waste (53%), sewage contamination (20%), tobacco farming (13%), rubber dam (8%) and sand extraction (6%). Therefore, necessary preventive measures should be taken in order to control the unwise and unauthorized discharge of harmful pollutants into the Halda River for the better management and conservation of natural resources.

Key words: Water Quality, Assessment, Pollution Sources, Parameters, Halda River

Introduction

Halda is the third main river of Chittagong district and has become the effecters of development providing fresh water supply, fish production, transportation and waste assimilation provision along with a wide array of recreation and tourism options (Kabir *et al*. 2013). The water of the Halda River is used for irrigation, agriculture, fish farming and livestock rearing, drinking and bathing. Halda River has a unique feature since it is the only natural breeding ground of major carps in Bangladesh (Kibria *et al.* 2009). Spawning area of this river is extended from Garduara to Maduna Ghat where a total of 1100 egg collectors and 2000 fisherman catch fish throughout the year (Islam 2009). Various species of fish namely *Catla catla*, *Labeo rohita*, *Cirrhinus mrigala* and other fresh water fish species chose to breed in the breeding ground of the Halda River (Tsai *et al.* 1981 and Patra and Azadi 1985).

River water uses for innumerable rural and urban communities and livestock, fish culture, recharge of ground water, control of floods etc. (Gurunathan and Shanmugam 2006). But the water quality is being degraded unceasingly due to haphazard industrialization

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(Manjare *et al.* 2010). Most of the industries discharged their insufficiently treated waste into the rivers or streams, which makes serious problem to aquatic flora and fauna (Kesalkar *et al.* 2012). Polluted water reduces fish production and availability of fish and also has an impact on other aquatic resources in the water (Mallick 2011). The dark color of the waste water exhibits the toxic effects on the biota and inhibits the photosynthetic activity by reducing the sunlight (Swamy 2011).

River pollution is a matter of concern all over the world (May *et al*. 2006, Noori *et al*. 2010 and Ouyang *et al*. 2006). Bangladesh is facing serious problems with water contaminations from different industries, domestic wastes and agrochemicals (Venugopal *et al*. 2009 and Islam *et al*. 2015a and b). Due to lack of waste management and sanitation facilities, raw and partially decomposed industrial and urban wastes and various hazardous chemicals substances from Raozan, Fatickchari, Hathazari and Biazid Thana find their way to the Halda River. Halda River is not focused well yet in terms of pollution status. Therefore, the present study was aimed to assess the water quality of the Halda River and to find out major pollution sources of the River.

Materials and Methods

Sampling Sites: The study was conducted in the Halda river which lies between 22°25'13"-22º48'51.37"N and 91º45'00"-91°52'33"E (Fig. 1). It is the sole natural breeding ground for carps (ruhi, katol, mrigal etc) due to the unique biochemical properties of its water. Industrial wastes, sewage discharge, tobacco farming, rubber dam and sand extraction are the main polluters of the river which are carried by canals. Four canals namely Mondakini (Station-1), Madari (Station-2), Chengkhali (Station-3) and Khondakia (Station-4) located from Nazirhat to Baluchara area along the Halda river bank were selected as the main discharge routes responsible for the Halda river pollution (Fig. 1).

Sample Collection and Preservation: After selection of sampling points, a total of 24 water samples was collected. Two water samples were collected for each seasons from the Mondakini, Madari, Cheng khali and Khondakia. Two liters of surface water sample were collected in the morning hours between 10 to 11am, in polythene bottle regularly during every sampling. Immediately after collection, water sample were transferred to the laboratory of Institute of Marine Science and Fisheries, University of Chittagong. pH and DO were measured from studied sites during sampling of water. To find out the pollution sources; data were collected data using surveys, focus groups, key informant interviews and direct observations. Questionnaire survey was done for this study and interviewees were selected randomly (Henry 1990). The survey design was based on expert advice from local NGO staff and guidelines for identifying the major land based pollution sources of the Halda River (Kronen *et al*. 2007).

Sample Analysis: pH of water was determined by using Hanna pH meter. Chloride was measured by Volhard Method, alkalinity was determined by Titrimatric, DO was determined by using DO meter (Hanna DO meter, HI-9146), Bio-chemical oxygen demand (BOD) by modified Winkler method (1988). Moreover, NO_3-N and PO_4-P were measured following standard methods recommended by the APHA (2005) for waste water.

Fig. 1. Map showing sampling sites and pollution sources of the Halda River.

Statistical Analysis: The variations in water quality parameters of the studied canals measured in three different seasons were analyzed by using Two-Way ANOVA (SPSS v.22).

River Pollution Index (RPI): Each water quality variable used to determine RPI is changed to one of four index scores (S_i = 1, 3, 6, or 10). Particularly, RPI refers to the arithmetic average of these index scores with respect to the water quality (Table 1).

RPI is computed using following equation (Liou *et al*. 2004).

$$RPI = 1/4 \sum_{i=1}^{4} Si$$

where S_i represents the index scores and the RPI value ranges from 1 to 10.

Table 1. River Pollution Index (RPI) Chart (Chen *et al*. 2012 and Liou *et al*. 2004).

Items/ ranks	Good	Less polluted	Moderately polluted	Highly polluted
DO (mg/L)	>6.5	4.6-6.5	2.0-4.5	<2.0
BOD_5 (mg/L)	<3.5	3.0-4.9	5.0-15	>15
SS (mg/L)	<2.0	20-49	50-100	>100
NH_3-N (mg/L)	<0.5	0.5-0.9	91.0-3.0	>3.0
Index scores (Si)	1	3	6	10
Sub-index	<2	2.0-3.0	3.1-6.0	>6.0

Results and Discussion

pH, DO, BOD showed significant changes in station-1 and station-3 for monsoon, post-monsoon and winter ($p < 0.05$) while chloride at station-3 for all seasons ($p \leq 0.05$). Additionally, total alkalinity, NO_3-N and PO_4-P exhibited significant variation in the concentration in station-1, station-2 and station-3 for three seasons ($p \leq 0.05$).

There was no significant change in BOD level at different stations but significant shift was noticed in terms of total alkalinity at station-1 (73 mg/L) and station-2 (210 mg/L). This changing pattern followed by chloride at station-1 (8.5 mg/L) and station-2 (65.6 mg/L) during monsoon period (Table-3). In station-1, water pH was 7.1 followed by station-2 (8.7), station-3 (7.9) and station-4 (8.1) during post Monsoon. DO and BOD also varied according to stations but not significantly. Significant change was observed in case of chloride in different stations. Following monsoon and post Monsoon, water quality also changed in winter but not significantly. Significant variation was observed in terms of chloride (station-1 and station-4) and total alkalinity (station-1 and station-2). The concentration of DO and BOD found almost equal in station-3 and station-4 which is shown in Table-3. The water quality variables of the Halda River compared with other rivers and with international standard shown in Table 2.

Dissolved Oxygen (DO): DO is an important ecological factor that decides environmental health of water bodies and support a well-balanced aquatic living organisms (Chang 2005 and George *et al*. 2012). The highest amount of DO recorded was 4.70 mg/L at Mondakini canal during winter. The lowest concentration recorded was 3.35 mg/L at Khondakia canal during monsoon (Table 3). More or less similar observations were also recorded by Jashim Uddin and Khan (1993), Islam and Khan (1993), Hossain (1998), Gasim *et al*. (2007) and Effendi *et al*. (2015). The low concentration of DO might be due to large amount of wastes discharged by canal and drainage system which utilized significant amount of DO for bio-chemical degradation. In the present investigation, the depletion of DO was found particularly due to the effluent discharged from Asian paper mill and Hathazari Picking power plant.

Table 2. Water quality parameters of the Halda River comparing with international standard and other Bangladeshi study.

Parameters	Present study	Previous study (Ahmed *et al.* 2010)	Previous study (Majid and Sharma 1999)	WHO (1982)	ECR (1997)
DO (mg/L)	3.35-4.70	3.02-9.90	ND	4-6	6
pH	7.1-8.8	7.03-8.60	5.65-7.34	NYS	6.5 – 8.5
Chloride (mg/L)	8.4-69.30	2.41-73.50	3.00-7.00	600	150 – 600
Total Alkalinity	73-220	6.28-90.78	22-72	-	-
BOD_5 (mg/L)	0.055-5.0	0.70-5.08	ND	6	0.2
NO_3-N (mg/L)	0.12-3.1	0.00	ND	NYS	-
PO_4-P (mg/L)	0.06-0.14	0.73-4.28	0.09-0.40	0.8	-
BDL= Below Detection Level, ND= Not Done					

Table 3. Seasonal variation of water quality indicators of the Halda River.

Parameters	Season	Station-1	Station-2	Station-3	Station-4
DO (mg/L)	Monsoon	3.9	3.5	3.45	3.35
	Post-monsoon	4.2	3.6	4.35	4.30
	Winter	4.7	4.1	4.31	4.28
pH	Monsoon	7.3	8.8	7.8	8.0
	Post-monsoon	7.1	8.7	7.9	8.1
	Winter	7.2	8.6	8.0	7.9
Chloride (mg/L)	Monsoon	8.5	65.60	56.30	60.15
	Post-monsoon	8.9	69.30	59.10	62.30
	Winter	8.4	59.20	52.60	61.15
Total Alkalinity (mg/L)	Monsoon	73	210	198	183
	Post-monsoon	77	218	207	192
	Winter	81	220	210	195
BOD_5 (mg/L)	Monsoon	0.28	0.057	0.055	0.056
	Post-monsoon	4.3	3.7	4.2	3.5
	Winter	4.1	2.5	5.0	3.4
NO_3-N (mg /L)	Monsoon	0.18	0.42	0.46	3.1
	Post-monsoon	0.15	0.37	0.39	0.27
	Winter	0.12	0.35	0.29	0.22
PO_4-P (mg/L)	Monsoon	0.08	0.14	0.14	0.13
	Post-monsoon	0.07	0.13	0.13	0.12
	Winter	0.06	0.12	0.12	0.1

pH: pH is commonly known as the controlling variable in water since many properties, processes and reaction are pH dependent (Millero 1986). Alkaline water stimulates high primary production (Kumar and Prabhahar 2012). In the present study the value of pH showed slightly alkaline in nature at investigated areas which showed similarity with Roy (1955), Campbell (1978), Mahmood and Sarma *et al.* (1982), Bhuyian (1988) and Bhuyan and Bakar (2017).

Chloride (Cl): Chlorides are not detrimental to public's health. Although the sodium part of NaCl salt connected to heart and kidney disease (Florescu *et al*. 2010). Sodium chloride (NaCl) may render a salty taste at 250 mg/L. Higher concentrations of dissolved salts in water compromise its use for domestic or agricultural purposes. Excessive amount of Cl^- in inland water is usually considered as an index of pollution and can be provided across the hygienic and industrial waters (Florescu *et al*. 2010). In the present study the concentrations of chlorides recorded were between 8.4-69.30 mg/L. Maximum amount (69.30 mg/L) was found at station-2 during post-monsoon. Minimum value (8.4 mg/L) was recorded at station-1 during winter (Table 3).

Total Alkalinity: Total alkalinity was found to range between 73-220 mg/L (Table 3). The amount recorded in the present investigation was higher than Akter (2012) and Hossain (2004). Hoque *et al*. (2012) reported that the value of alkalinity in monsoon season was 50.4 mg/L and in winter season it was 146.5 mg/L in the Bansi River. The total alkalinity at Madari Canal was higher than that of the other stations during the period of investigation. These high values might be due to the highest alkaline effluent discharged by the Asian paper mill. More or less similar results were found by Hossain (1992) and Hossain (2004). Bhuyian (1979), Andrews (1984) and APHA (2005) stated that if the industrial effluents contain high concentration of chloride that could increase the chloride of water body where the effluent was being dumped.

Biochemical Oxygen Demand: BOD is the amount of oxygen used by microbes to decay carbon-based materials in water within five days period (APHA 2005). In the present study BOD varied between 0.055-5.0 mg/L (Table 3). According to Paul (1999), when river water contains BOD more than 10 mg/L it is considered to be moderately polluted and more than 20 mg/L to be highly polluted water but the present result was lower than these values. The acceptable value of BOD for fishing water is ≤ 6 ppm. Hossain (1988) and Hossain (1992) observed higher values of BOD in the disposal zone due to consumption of oxygen for the oxidation of large amount of wastes discharged from the municipal sewerage and surface runoff. These observations are in agreement with the present investigation.

Nitrate Nitrogen: The concentration of NO_3-N was recorded between 0.12-3.1 mg/L (Table 3). The maximum values 3.1 mg/L was observed at Khondakia canal in the monsoon season and minimum value 0.12 mg/L at Mondakini canal in winter. The value fluctuated from monsoon to winter. Ahmed *et al*. (2010) found (BDL-0.87 mg/L) nitrate values in the Halda river and 0.00-1.63 mg/L from the Karnafuli river water.

Phosphate Phosphorus: The value of PO_4-P was found to range between 0.06-0.14 mg/L (Table 3). The maximum value 0.14 mg/L was found at Madari canal and Chengkhali canal during monsoon. Majid and Sharma (1999) studied the Karnafuli River and found very low concentration of Phosphate. Ahmed *et al*. (2010) recorded 0.12-4.94 mg/L PO_4-P from the Karnafuli river water. Alam *et al*. (2007) worked on the river water quality and stated that the effluent and discharge area have a low concentration of phosphate.

River Pollution Index (RPI): In recent time, River pollution index (RPI) method is used concurrently by different organization like Taiwan EPA to assess the surface water

quality. This method comprising with concentration level of four parameters namely DO, BOD, SS, and NH_3-N. Pollution status is calculated using four-state of each parameter.

In present study, the concentrations of DO and BOD_5 were compared with concentrations of RPI table to weigh the status of particular water variables (Table 1). Average DO in the Mondakini Canal was found 3.9 mg/L during monsoon season that indicate the water is moderately polluted in comparison with RPI. The water of Madari canal contained 4.1 mg/L DO that indicate the water is moderately polluted. Chengkhali Canal and Khondakia canal are also moderately polluted according to RPI Table.

During post-monsoon, average DO in the Mondakini Canal was recorded 4.2 mg/L which can be treated as moderately polluted zone according to RPI index while 4.35 mg/L and 4.30 mg/L DO were found from Chengkhali Canal and Khondakia canal that direct the area is moderately polluted. Where 3.6 mg/L was found from Madari canal area that also renders the area characterized with moderately polluted.

During winter the average DO concentration recorded was 4.7 mg/L at Mondakini canal, which designate less pollution in this area according to RPI table. Madari Canal, Chengkhali Canal and Khondakia canal are considered moderately polluted zone according to RPI index.

BOD_5 were found between 3.0-4.9 mg/L for post-monsoon and winter at all the sampling sites indicating that the water of the river is less polluted according to (Table 3). During monsoon the water quality was found good. During winter the amount of BOD_5 at Chengkhali canal was recorded 5.0 mg/L during winter season that indicate the water of the river canal is moderately polluted.

Sources of pollution: Structured questionnaire survey and direct observation were used to identify pollution sources of the river. According to the interviewers, the Halda River (the country's largest natural breeding ground for carps) is being polluted due to industrial waste (53%), sewage contamination (20%), tobacco farming (13%), rubber dam (8%) and sand extraction (6%) (Fig. 2).

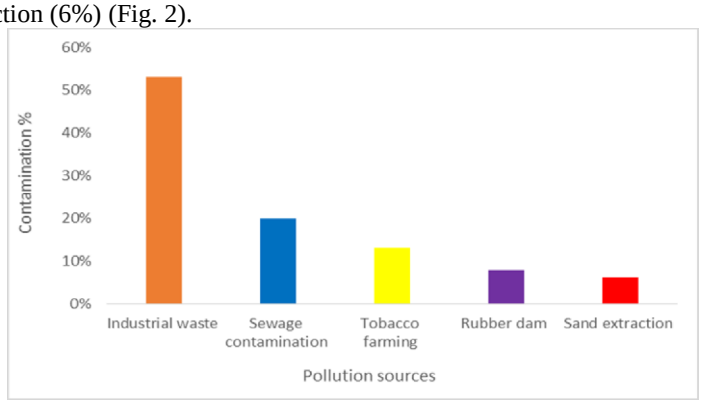

Fig. 2. Pollution contributors of the Halda River.

The present study shows that the river Halda is being polluted from industrial and commercial sources at an alarming rate. Therefore, it is recommended that industrial and other effluents should be discharged into the river after proper treatment (ETP).

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EFFECTS OF SLUMS ON ECOSYSTEM COMPONENTS

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Abstract

An *in vitro* study was conducted to assess the effects of slums on ecosystem services particularly soil, plant and water. The research findings indicated that, samples of soil, plant and water were contaminated with heavy metals, many of which were above permissible limits. The heavy metals exceeding the permissible limits pose serious threats to the surrounding plant and animal lives. Hence, there is a health risk associated with the consumption of the vegetables grown in the slums due to the fact that the heavy metals can bio-accumulate into human body through the food chain.

Key words: Slums, Ecosystem, Heavy metal, Bio-accumulation, Food chain

Introduction

Bangladesh is one of the most densely populated countries in the world with more than 160 million people living in a territory of around 56,977 sq. miles. One quarter of the population lives in urban areas, where population density is 200 times greater than the national figure and population growth is twice the national average (BBS 2011). Millions of slum-dwellers in Bangladesh live stressful lives among teaming rubbish because there is no or little waste disposal system. In these areas where wastes have been dumped without coverage, the soil will contain high amounts of pollutants. Industrial wastes originating from mining industries, chemical industries, metal processing industries etc. are major sources of soil contamination. These wastes include a variety of chemicals like heavy metals, phenols etc. (Babich and Stotzky 1977 and Mueller *et al*. 1989). Above certain concentrations and over a narrow range, the heavy metals turn into toxins (Babich *et al.* 1982). Moreover, the heavy metals deposited in the soil are bound preferentially to inter-aggregate soil materials and accumulation preferentially occurs in parts of the soil where plant roots are concentrated and in the forms that are easily accessible to plants (Wagner 1993). However, plants can absorb heavy metals in larger amounts than needed and the excessive amount will be stored in leaves and other edible parts. Through food consumption theses metals are then transferred to humans and animals. A high intake of heavy metals can damage organs and increase the risk of human cancer (Martin and Griswold 2009). Thus, the present study was conducted to assess the effects of slum areas on ecosystem components *viz.*, soil, plant and water.

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Materials and Methods

Soil, plant and water samples were collected from different slums in Dhaka city and were analyzed in the laboratory to find out the contents of nutrients and heavy metals.

Collection, preparation and processing of soil, plant and water samples

Soil Sampling: A total of eight soil samples was collected from four different slum areas of Dhaka city namely Kalyanpur, Gabtoli, Baunia and Sadek Khan. Table 1 shows the geographical coordinates of the sampling sites. The soils belong to Tejgaon series, which is mostly red, sticky, slightly acidic, clayey-loam having low to moderately low pH values (Hussain 1992). Soil samples representing 0–15 cm depth were collected by composite sampling method as suggested by the Soil Survey Staff of the USDA (1951). The collected soil samples were oven dried for 5 days. Visible roots and debris were removed. The samples were exposed to sunlight for speeding up the drying process. Ground samples were passed through a 2 mm sieve and mixed thoroughly. A portion ofthe soil samples (2 mm sieved) were further ground and screened to pass through a 0.5 mm sieve. The homogenized samples were used for physico-chemical analyses.

Table 1. Geographical coordinates of the sampling sites.

Sampling Site	Symbol	Latitude	Longitude
Kalyanpur	K	23º 47’ 032’’N - 23º47' 033"N	90º 21’432’’E - 90º 21' 448"E
Gabtoli	G	23º 46’815”N - 23º 46’ 921”N	90º 20’ 405”E- 90º 20’ 474’’E
Baunia	B	23º 49’ 093”N - 23º 49’ 161”N	90º 22’ 976”E - 90º 23’ 000”E
Sadek Khan	SK	23º 44' 952"N - 23º 44’ 969” N	90º 21’ 516”E - 90º 21' 543"E

Plant samples: A total of 28 plant samples was collected from three different slum areas namely Kalyanpur, Gabtoli, Baunia. The plant was uprooted and processed for laboratory analyses as suggested by Imamul Huq and Alam (2005). After collection, the whole plant was washed in tap water. Different plantparts were separated and were rewashed thrice with distilled water. These plant samples were first air-dried and then oven-dried (80 – 85º C), grounded and finally sieved through a 0.25mm sieve.

Water samples: The water samples were collected by Grab and Catch method from different sources of wells and ponds of the slum areas (Imamul Huq and Alam 2005).Few drops of concentrated HCl were added to the bottle in order to acidify the samples. Then the bottle was filled with water samples for chemical analyses following filtration.

Laboratory analysis of soil and plant samples: The quality control and quality assurance (QC/QA) of the analyses were done as described in Imamul Huq *et al.* (2008). Nitrogen in bothsoil and plant was determined by alkali distillation of the Kjeldahl digests (Imamul Huq and Alam 2005). Phosphorous was determined following colorimetric method using a spectrophotometer and potassium by a flame photometer (Jackson 1962). Organic carbon was determined by Walkley and Black’s wet oxidation method (Imamul Huq and

Alam 2005) The heavy metals were analyzed by using Atomic Absorption Spectrophotometer (Varian AA 240) after digesting the samples with aqua-regia (for soil) and ternary acid mixture (for plants).

Data analysis: All data were statistically analyzed by using Microsoft Excel and MINITAB (version 17) package.

Results and Discussion

Nutritional status of soil and plant samples: Contents of nitrogen, phosphorous, potassium and organic matter in soil were determined (Figs. 1 to 3). The nitrogen of the soils was found to be insignificant as compared to the standard value (Fig. 1a). However, the available nitrogen was found to be higher compared to that of the standard value (Fig. 1b). The available phosphorous in the soil (Fig. 2b) was not significant except one sample (Sadek Khan) although the total phosphorous (Fig. 2a) was higher than the standard limit set for soil. The soils contained less potassium than the standard value for both soil and plant (Figs. 3a and 3b). The organic matter of the soils in the slums were mostly around 1% except the soil of Sadek Khan sampling site which possessed around 3.1% organic matter.

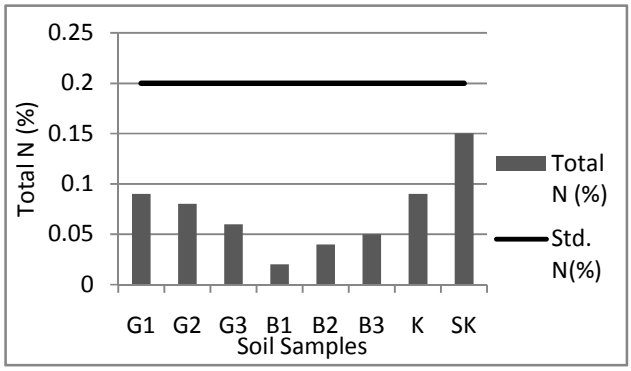

Fig. 1(a). Total Nin soils.

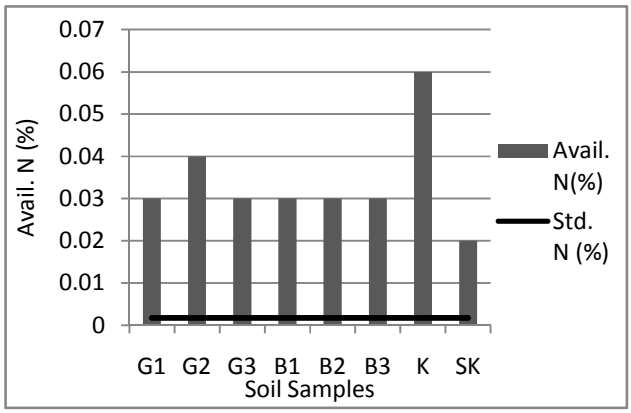

Fig. 1(b). Available N in soils.

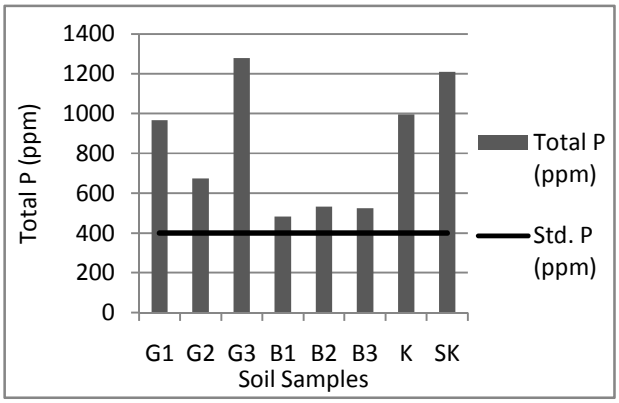

Fig. 2(a). Total P in soils.

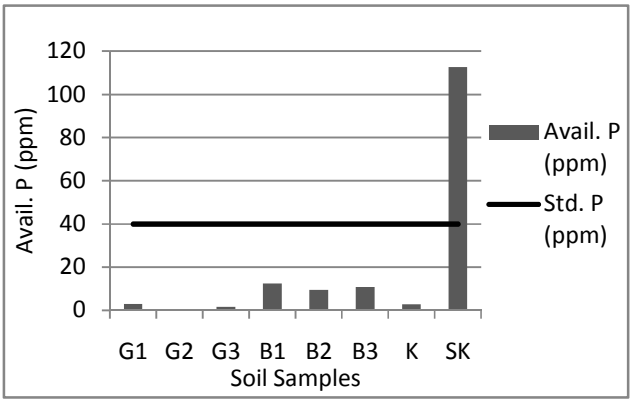

Fig. 2(b). Available P in soils.

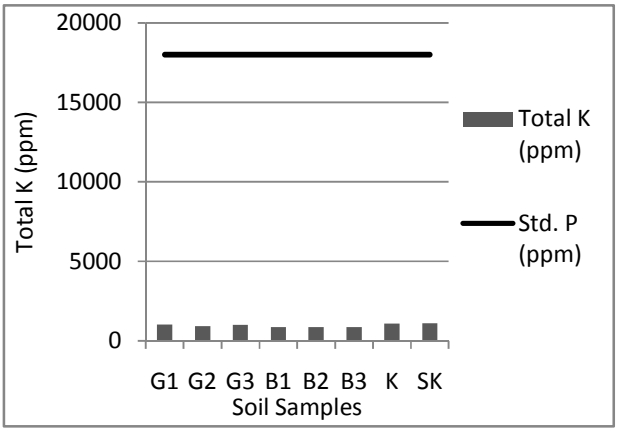

Fig. 3(a). Total K in soils.

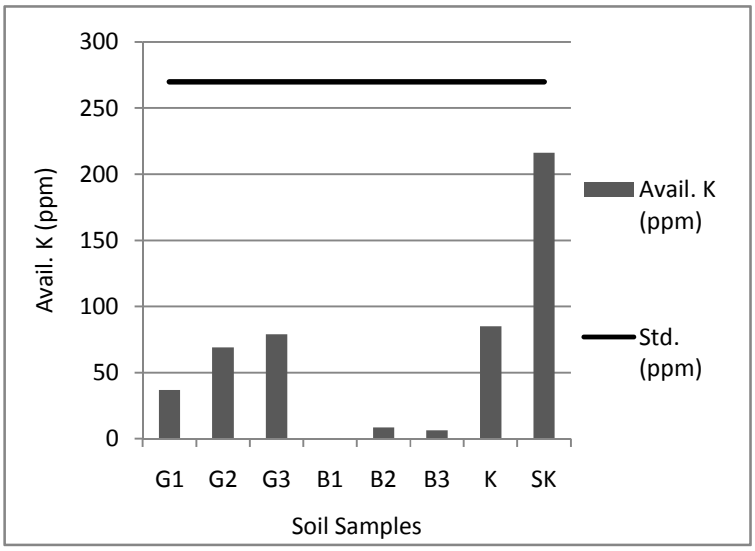

Fig. 3(b). Available K in soils.

Heavy metals in soils and plants: Contents of heavy metals (Pb, Cr, As, Zn, Mn and Al) recorded in soils and plants are presented in Figs. 4 to 9.

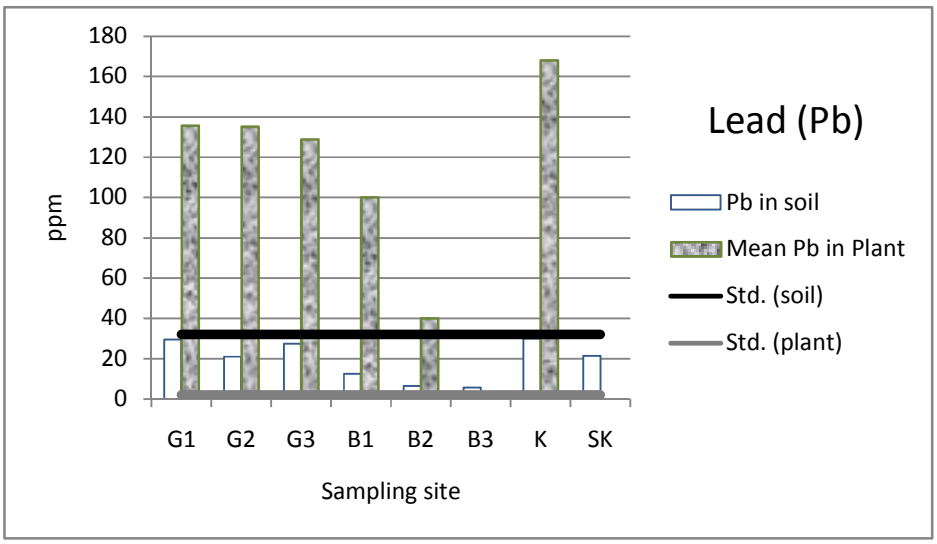

Fig. 4. Concentration of lead (Pb) in soils and plants.

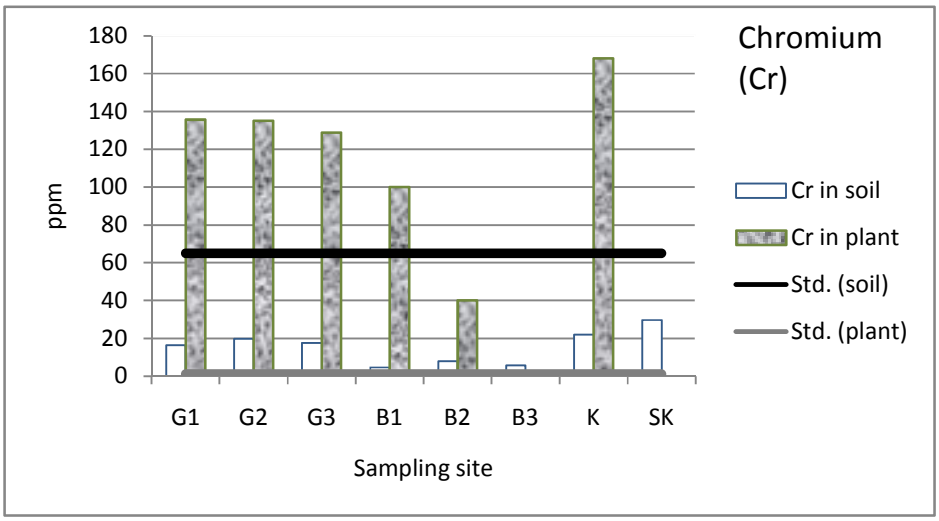

Fig. 5. Concentration of chromium (Cr) in soils and plants.

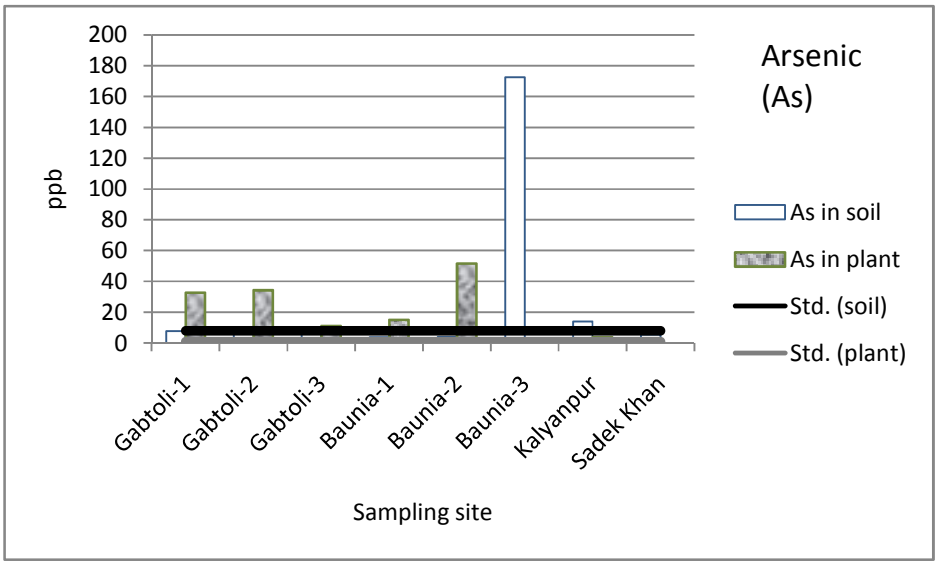

Fig. 6. Concentration of arsenic (As) in soils and plants.

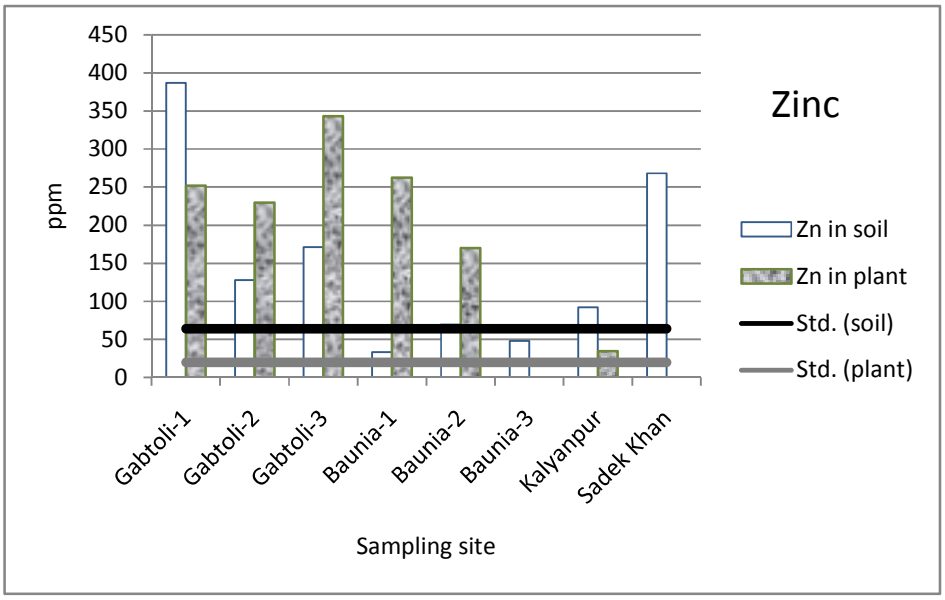

Fig.7. Concentration of zinc (Zn) in soils and plants.

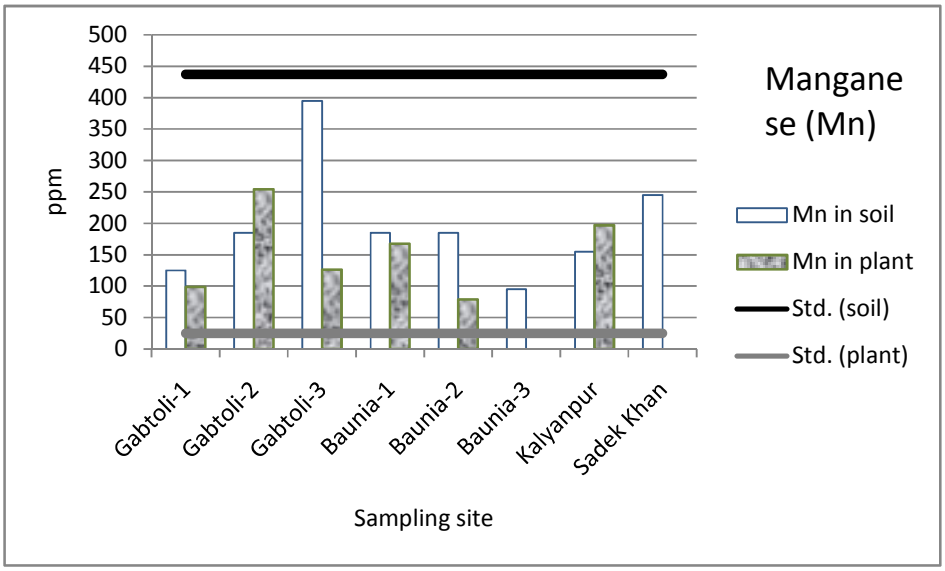

Fig. 8. Concentration of manganese (Mn) in soils and plants.

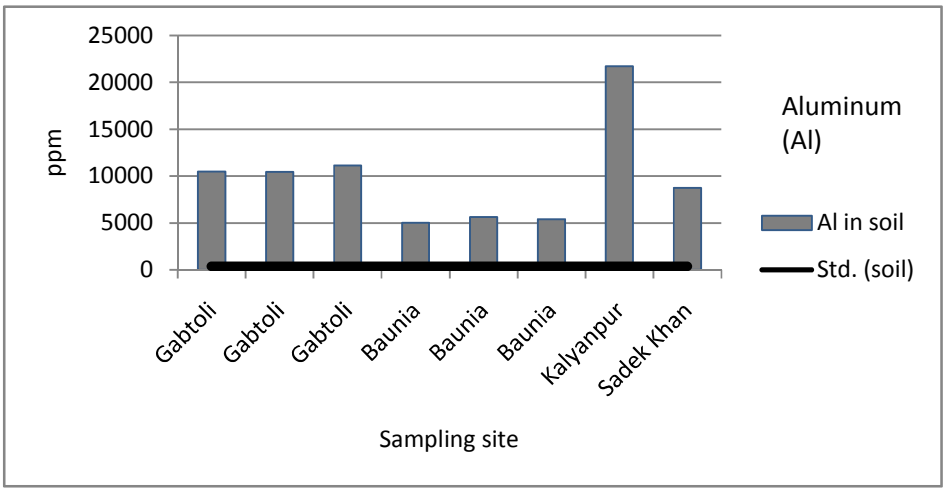

Fig. 9. Concentration of aluminum (Al) in soils and plants.

Table 3 shows that heavy metals *viz.*, Cd, Al, As and Fe in water bodies were present in excessive amounts as compared to the standard limits for irrigation purpose. Manganese (Mn) was present in moderate level. However, Cr and Zn were found below the standard limits. This indicates that there is a possibility for plants to uptake heavy metals from the water used for irrigation purpose, which would further aggravate the situation.

Table 3. Total heavy metals contents in irrigation water.

Water samples	Zn*	Mn*	Pb*	Cd*	Cr*	Al*	As*	Fe*
Gabtoliwater sample	BDL	0.25	0.42	0.07	0.07	33.79	0.00614	126.84
Bauniawater sample	BDL	0.22	0.35	0.09	0.06	34.34	0.00581	79.63
Kalyanpurwater sample	BDL	0.17	0.73	0.06	0.09	35.69	0.00536	78.27

*values are in mg/L; BDL= Below Detection Level

It is apparent from the Figs. 4 to 9 that plants grown in slum areas contained more heavy metals compared to that of the corresponding soils. However, there was an excessive amount of aluminum (Al) in soils which might initiate soil acidity if the pH drops below 5.2 (Brady and Weil 2002).Almost all of the plant biomasses contained very high amounts of lead (Pb), which was present in the edible parts (fruit, leaf and shoot) and thus could be transferred through food chain in human body. The highest concentrations were found in soils and plants of Kalyanpur area (Fig. 4).

All of the plants contained chromium (Cr) much higher than the Maximum Allowable Concentration (MAC) and cannot be considered safe. The highest concentration of Cr was found in soils of Kalyanpur site and plants of Sadek Khan site (Fig. 5).Soils of

Baunia-3 site showed extremely high concentration of As and no plants were found to grow in this site. However, plants growing in Gabtoli-1, Gabtoli-2 and Baunia-2 contained As in amounts exceeding the upper limit, making it impossible to consume these plants (Fig. 6).

Nutritional status of plant samples: The plants contained sufficient amount of N and K (Table 2). But the phosphorous content of the plants was found to be below the standard level i.e. 2000 ppm (Imamul Huq and Alam 2005). This may happen partly because the soils contained lower amounts of available phosphorous.

Table 2. Total N, P, K contents in plants.

Sampling Site/ Plant Sample	TotalN (%)	Total P (ppm)	Total K (ppm)
Kalyanpur site			
Papaya (*Carica papaya*) (Leaf)	2.39	1468.15	17840.0
Sweet potato (*Ipomoea batatas*)	1.74	1567.92	14140.0
Gourd (*Lagenaria siceraria*)	3.36	1640.83	15940.0
Bean (*Phaseolus vulgaris*)	2.58	1721.41	13540.0
Tomato (*Solanum lycopersicum*)	1.21	1429.78	15440.0
Arum	1.43	1778.97	16840.0
Gabtoli site-1			
Red amaranth (*Amaranthus gangeticus*) (Leaf)	1.71	1295.47	13140.0
Red amaranth (*Amaranthus gangeticus*) (Shoot)		1541.06	25940.0
Tomato (*Solanum lycopersicum*) (Leaf)	3.13	1729.09	14540.0
Tomato (*Solanum lycopersicum*) (Root)		1587.11	9740.0
Tomato (*Solanum lycopersicum*) (Shoot + Fruit)		1706.06	18940.0
Bean (*Phaseolus vulgaris*) (Shoot + Leaf)	3.00	1276.29	11440.0
Gourd (*Lagenaria siceraria*) (Shoot)	4.27	1495.01	22940.0
Gourd (*Lagenaria siceraria*) (Leaf)		1345.36	14940.0
Gabtoli site-2			
Red amaranth (*Amaranthus gangeticus*) (Leaf + Root)	2.85	1552.57	25640.0
Green amaranth (*Amaranthus viridis*) (Leaf + Shoot)	2.70	1775.13	15040.0
Tomato (*Solanum lycopersicum*) (Leaf + Root)	1.92	1175.13	8640.0
Gabtoli site-3			
Indian spinach (*Basella alba*)(Leaf + Shoot)	0.57	1844.21	9340.0
Gourd (*Lagenaria siceraria*) (Shoot + Leaf)	2.28	1717.58	18040.0
Green amaranth (*Amaranthus viridis*) (Shoot + Root)	3.84	2055.23	13340.0
Green amaranth (*Amaranthus viridis*) (Leaf)		1544.90	15540.0
Baunia site-1			
Indian spinach (*Basella alba*)	1.21	1782.81	17240.0
Gourd (*Lagenaria siceraria*) (Leaf)	1.71	1890.25	11440.0
Gourd (*Lagenaria siceraria*) (Shoot)		1778.97	17140.0
Bean (*Phaseolus vulgaris*) (Leaf + Shoot)	2.56	1932.46	11240.0
Baunia site-2			
Red amaranth (*Amaranthus gangeticus*) (Leaf + Root)	4.00	1610.13	16040.0
Red amaranth (*Amaranthus gangeticus*) (Stem)		1425.94	20840.0
Green amaranth(*Amaranthus viridis*) (Shoot + Root)	3.84	2055.23	13340.0
Green amaranth (*Amaranthus viridis*) (Leaf)		1544.90	15540.0

The amounts of zinc (Zn) and manganese (Mn) in plants were found in amounts higher than the standard value. Fig. 8 shows that the highest concentration of Mn was found in the soils of Gabtoli-3 and plants of Gabtoli 2. However, the maximum concentration of Zn was found in the soils of Gabtoli 1 and plants of Gabtoli 3.

The pH values of the top soils were between 5.22 and 6.70, which might make the heavy metals available for the growing plants. Figs. 4 to 9 illustrate that the soils contained smaller concentration of heavy metals than plants alone did. This indicates that, there might be another source of heavy metals from which plants are taking them up. It could be water with which the soils were irrigated or the fertilizers which were applied to the soils to meet the nutritional deficiencies. However, if the pH decreases in soil, more metals are expected to be released and become available for plant uptake as the soil buffers the acidity.

It is observed from Figs. 4 to 9 that the soils in the slum areas contained heavy metals in varying concentrations; some of them were present at toxic levels. In case of plants, the scenario was quite different. These figures illustrate that almost all the plants grown in slums contained excessive amounts of heavy metals. Pb, As, Cr, Zn and Mn were present in all kinds of plant biomass (edible parts) at a toxic concentration which,if consumed for long period may contribute to carcinogenic diseases in human body.

Adverse effects of slums on the ecosystem are one of the burning questions of today's modern world. The present study provided a precise idea about the contents of nutrient elements and heavy metals present in soil, plant and water of different slum areas. The results confirmed that both the soils and plants in all slum areas contained high concentration of heavy metals exceeding the permissible limits that ultimately led to decreased agricultural crop production. The high concentration of heavy metals in soil might become readily available to the plants that could aggravate health risks of the slum dwellers. In addition, nitrogen, phosphorous and potassium contents were found to be less compared to those of the standard values. Water collected from various sources of slum areas did not meet the acceptable limit for irrigation. Thus, it is pertinent to assess the pollution level of soil, plant and water present in and around the slum areas. Further detailed study is needed regarding the type and magnitude of the treatments for these polluted components of ecosystem.

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WOMEN HEALTH AND MENSTRUAL HYGIENE MANAGEMENT IN NATURAL DISASTERS: A STUDY IN ISLAMPUR UPAZILA OF JAMALPUR DISTRICT

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Abstract

The study examines health and menstrual hygiene management of disaster affected women at Kulkandi Village under Islampur *Upazila* of Jamalpur District and explores their needs and problems regarding menstrual hygiene issues during disasters. This present study based mainly on primary field investigation including questionnaire survey with women and girls, focus group discussions (FGDs) and key informant interviews (KIIs) reveals that unhygienic sanitation and unclean water during disaster leads them to severe health issues including gynaecological problems. A significant number of respondents (21%) experienced urinary tract infections and 24% faced rashes on sensitive private parts of their bodies. Managing their menstruation and maintaining a good standard of menstrual hygiene is difficult for them because of the factors established in this study, such as inadequate knowledge, culture and traditions, inadequate facilities and services, poverty and lack of access to health care. Effective cooperation from both the government agencies and NGOs is needed to promote good menstrual hygiene management (health care issues, sanitary pads in relief materials, workshops) in the study area.

Key words: Menstrual Hygiene, Natural disasters, Vulnerability of women and girls, Kulkandi, Jamalpur

Introduction

Bangladesh is a low lying delta with very gentle slopes and is located at the lowest end of the Ganges, Brahmaputra and Meghna Basin (Roy *et al.* 2009). The geographical location of Bangladesh, its low relief of the land, seasonality of monsoon precipitation, intricate networks of rivers and streams, and the funnel shape of the coastline at the head of the Bay of Bengal make it highly vulnerable to natural hazards. The problem is further aggravated by the high concentration of a very large population in a small area, exposing the region to disasters causing loss of lives and assets. The typical geographical features have significant impact on the climate system of Bangladesh that makes it one of the worst victimized countries on earth causing significant loss of lives and properties (Nizamuddin 2001). The consequences of natural disasters not only affect the overall population but also it unequally affects the rich and the poor and it is unequal in terms of

gender. Natural disasters reinforce, perpetuate and increase gender inequality, making situations from bad to worse for women in Bangladesh. A study of 4,605 natural disasters found that disasters shortened women's life expectancy significantly more than men's (encouragingly, this association was reduced where women's status is more equal) (Rahman 2013). Many women are made vulnerable not only due to lack of access to sources of emergency information, health, hygiene and sanitation opportunities but also lack of decision-making power in disaster prevention and preparedness programs. Moreover, they are also often excluded from disaster recovery operations and from planning at the national level. This caused a reason for the intensive search for women specific health problems, risks and adaptation strategies.

Globally about 52% of the female population is of reproductive age, meaning menstruation is part of their normal life and menstrual hygiene is therefore an essential part of basic hygienic practices (House *et al.* 2012). Though menstruation is a natural process, it has been and still is a matter of secrecy in most of the developing countries including Bangladesh. It is a neglected issue when it comes to research or relief or recovery activities during or after disaster. Women health, hygiene especially menstrual hygiene is not given much importance in the context of Bangladesh. At the same time literature review indicates that not enough academic and research study were undertaken to analyze the impact of disasters on women’s health and menstrual hygiene and their coping strategies adopted at the northern part of Bangladesh. The present study was conducted at Kulkandi Union of Islampur *Upazila*, Jamalpur district, one of the remotest and disaster prone areas in Bangladesh, with the aim to explore women's health and hygiene condition especially menstrual hygiene situation, preparedness, risk and loss, cultural and conditional behaviour, adaptability and recovery capacity from the natural disasters. The study was conducted to analyze the impact of disasters on women physical and mental health and menstrual hygiene condition including their needs and problems to survive throughout the disaster; to explore their own preparedness techniques and recovery strategies; to assess health care services available for women in the study area and to identify the drawback programs or activities taken by the government and other organizations for disaster affected women.

Materials and Methods

Both qualitative (focus group discussion and key informant interview) and quantitative (household survey) techniques have been applied in this study. Households were selected through random sampling technique. Primary data were collected by triangulation of different data collection techniques of household survey, focus group discussion (FGD) and key informant interviews (KIIs). As Kulkandi is a small village with homogenous characteristics in terms of livelihood pattern, civic facilities and environmental conditions, 100 household surveys, 3 FGDs and 5 KIIs were conducted using semi-structured questionnaires. Adolescent girls, middle aged women, aged women having menopause were chosen as respondents for household survey. On the other hand, FGD groups consisted of female participants of three groups such as adolescent school going girls, adolescent girls who are not going to school and middle aged women. Both informal contact and cold-calling approaches were utilized to reach participants for FGD. KII was done with the local CDMC women president, Bangladesh Red Crescent Society's girl volunteer, FWC (Family Welfare Centre), female doctor, Jamalpur Sadar Hospital gynaecologist and nurse of the hospital. The surveyed data were then processed and analyzed using Statistical Package for Social Science (SPSS) IBM 20 and Microsoft Office Excel software.

Study Site: Kulkandi Village (Fig. 1) under Kulkandi Union is located in Islampur *Upazila* of Jamalpur District, the northern part of Bangladesh which is badly affected by floods every year and therefore, it has been purposively selected for the present study area. It has an area of 7,024 acres and 400 households (Hasan 2014). This is one of the most vulnerable villages in the union as every year the inhabitants of this village suffer due to severe monsoon floods. Kulkandi is frequently affected by flash floods and water logging for its geographical location as it is surrounded by the Garo hills to the north and the major hydrological regime of the district is influenced by the Jamuna River and its distributaries. The Jamuna brings in huge quantum of water during monsoon inundating vast low-lying areas along the Jamuna flood plains. Flood generally occurs along the Jamuna system, while flash flood generally occurs along the smaller rivulets adjacent to the Garo Hills. Flash floods are short lived phenomenon, while only a few hours of intense rainfall can effectively make the area inaccessible for over a day or two. However, the quickly on-setting nature of the hazard washes away almost everything on the way of rushing runoff. Not only standing crops are devastated, physical infrastructure (especially roads and poorly constructed culverts, hospitals) are often devastated. This condition creates vulnerable condition for the women health in this region as they extremely lack of knowledge on menstrual hygiene and sanitation.

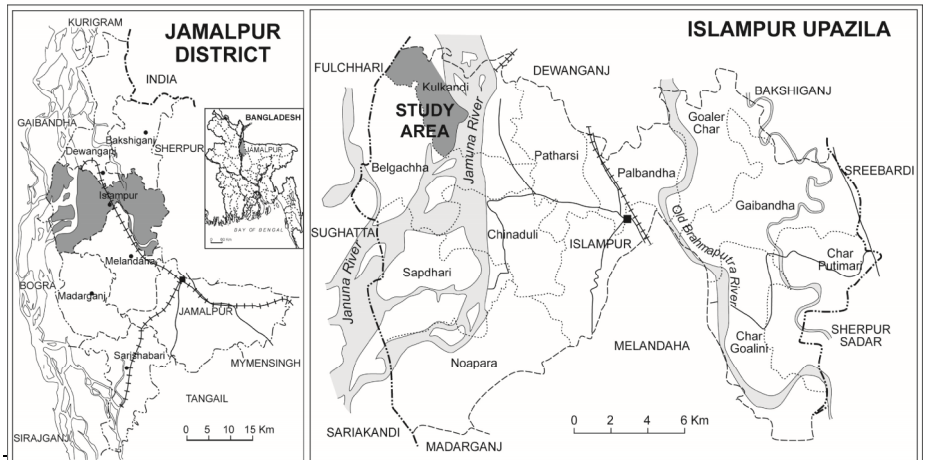

Fig. 1. Location of the study area.

Results and Discussion

Demographic and Socio-Economic Background: Age, marital status, religion, education, occupation and income are important socio-economic factors in menstrual hygiene management situation analysis. In order to know the situation of women and girls in the study area, age group selection was very important. Adolescent girls group aged from 11 to 20 years (24% of the respondents), youth group aged from 21 to 30 years (34% of the respondents), middle aged women group aged from 31 to 40 years (23% of the respondents) and elder women having their menopause aged from 41 to 50 years (19% of the respondents) were selected for the household survey.

Menstruation is referred as the girl is ready to be married when she has her first menstruation. In the study area, percentage of married respondents is high (73%), whereas only 19% of them are unmarried. Around 2% women were separated while 6% of these women are widows. Even adolescent girls were married at the age of 12 or 13 after their first menstruation. Being married at such an early age is not an uncommon scene in the rural areas of Bangladesh.

Religious status is another important determinant to know the menstrual practices in the study area as menstrual taboos and management varies from religion to religion. In the study area mainly two types of religion (Islam and Hinduism) are dominant. Among the respondents 88% are Muslims and other 12% are Hindu respondents.

Educational background is important as educated women and girls are aware of proper menstruation hygiene practice. More than half (56%) of the respondents are literate, while 46% of women have completed primary education but then they left school

because of poverty and getting married and 10% have completed high school education, while about 44% of the respondents are illiterate.

Employment status was measured in terms of the respondents own and the head of the household in the study area. Among the 100 respondents, majority of the respondents (65%) are housewives including the adolescent married girls, young girls, middle aged women and older women. Around 20% of the respondents were students and only 17% women are housemaid. Household head occupations of the respondents were found diversified including farmers (34%), day labourers (28%), fisherman (14%), small business (12%), service holders including govt. and NGO workers (9%) and auto driver (3%) in the study area. Economic condition of the people at Kulkandi is not well-off which is evident from their housing structure. More than 80% of the houses are *katcha* (floor, wall and roof of these houses are made of earth material, bamboo, thatch or wood). Besides, only 2% houses are brick-built, which can be considered as disaster resilient. Semi *pucca* houses are resilient to storm and flood. Overall, all of the houses, except very few, are physically highly vulnerable to any disasters.

Occurrence and Impacts of Disasters in the Study Area

Major Disasters: As mentioned earlier, the study area is subject to frequent floods. Although the district as a whole is prone to seasonal drought and riverbank erosion, the study area vulnerable to flash flood, river flood and water-logging. River flood (38.5%) associated with flash flood (28.3%) are the major disasters in the study area. Water-logging (24.8%) is the during-disaster and after-disaster effect of the floods. Riverbank erosion (8.4%) is also perceived as one of the threats increasing gradually in the study area.

Early Warning System and Shelters during Disasters: From the study, it has been found that 72% get early warning of disasters. From the FGDs it is found that they receive early warning from the *masjid* Imam or Red Crescent volunteers in the locality whereas other 28% don't receive early warning but they can identify the disaster time by observing the flow and color of the river water and full moon.

In the study area, 46.8% of the respondents mentioned that they take shelter on the dam during floods, whereas 23.7% take shelters in *madrasa* as their houses go under water. About 20% of the respondents stay at home using *macha* and they try to survive somehow. Respondents having strong brick-built house normally remain safe during floods. Another 10.9% said that they stay with their relatives.

Impacts of Disasters: Impact of disasters includes the socioeconomic problems created due to disasters, availability of the relief after the disasters and diseases that occur during and after the events causing huge problems and sufferings in the lives of the people. Their normal lifestyle becomes badly affected and they suffer from unemployment leading them to poverty. Health and sanitation practices are extremely unhygienic during and post disaster periods, which creates the outbreak of various diseases. Female are more vulnerable and neglected during disaster in the context of health, hygiene and sanitation

in the study area. Table 1 shows the problems created due to the impacts of disasters in the study area.

Table 1. Problems created in the study area due to disasters.

Problems	Number of responses (Multiple Responses, N)	Percentage
Food shortage	84	14.61
Scarcity of drinking water	91	15.83
Outbreaks of different types of diseases	75	13.04
Scarcity of fodder	39	6.78
Increasing food price and other commodities	77	13.39
Sanitation problems	86	14.95
Damage of houses	19	3.30
Poverty	73	12.69
Unemployment	26	4.52
Don't know	5	0.87
Total	N=575	100%

Relief Facilities: Among all the respondents, around half of the respondents who received relief in the past disaster claimed that sufficient relief was not supplied. Food such as *chal*, *cheera* and *gur* are given as food materials to the flood affected people. The family welfare centre in Islampur had given free medicine to the flood affected people. This free medicine included only food saline and normal painkillers.

Outbreak of Diseases due to Disasters: Unhealthy and unhygienic conditions are prevalent in the study area during and post disaster periods. During disaster, a majority of the respondents cannot afford to take boiled water and as a result diarrhoea, typhoid and dysentery are very common during this time. Skin diseases are also severe in this period as people have to live with unhygienic condition and continuous contact with contaminated water (Fig. 2).

Female are mostly affected due to these diseases severely as they feel neglected at the time of disaster. Moreover, they have to look after the family members. Skin diseases are common among the women and girls of all ages.

Water and Sanitation Condition: Unclean water and lack of access to hygiene and sanitation during disasters are sources of health hazards for women. Water is essential life element which is frequently used by women to care for children, the elderly and the sick, as well as to carry out many household tasks (Rahman 2013). During disasters, sanitation becomes a problem as women and girls carry the highest risk of getting in contact with polluted water and falling sick during and after disasters (Mehta 2007 and Dasgupta *et al*. 2010). Water-borne diseases might be expected to be more widespread among women, who are nutritionally disadvantaged (Rahman 2013).

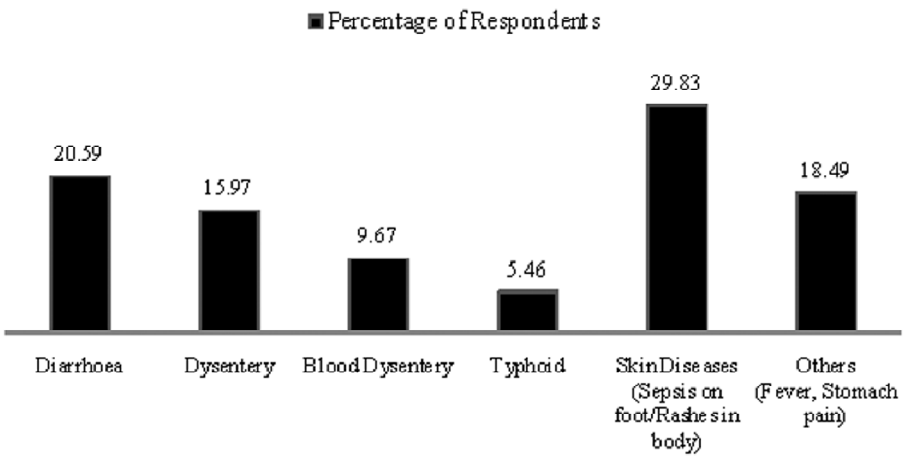

Fig 2. Outbreaks of diseases during and post disaster periods.

Drinking Water Sources during Disaster Period: Nearly half of the respondents collect water from the Jamuna River during normal period. Another around 40% of the respondents use deep tube-well water and other 17% of the respondents collect water from ponds in the study area. The situation gets changed during flooding events. Around one fourth of the respondents collect water from deep tube-well during disasters whereas a majority of the respondents (60.8%) collect water from river water as they have to live on the embankment. Therefore it is easy to collect the river water or flood water in that time. Only 13% of the respondents collect water from ponds which are not affected by flood water but these are located far away. It is beyond doubt that during disasters a majority of the respondents collects drinking water from river which is highly unhygienic. Drinking water sources can be a good indicator that helps to understand the reason behind the occurrences of different diseases in the study area. As water borne diseases become severe during disaster, women and children become mostly affected by this and also women have to be in contact with the unclean water for a long time to maintain their family member's need. Various kinds of skin diseases are also common in disaster period due to the continuous contact with the unclean water.

Purifying Techniques: Drinking unclean water without purification is the main cause of diarrhoea, dysentery and typhoid types of water borne diseases. During disaster 72% of the respondents drink water without boiling whereas only 28% uses purification techniques (e.g., tablets or *fitkiri* or drink tube-well water directly which are not affected by flood water).

Sanitation Facilities: During disasters, 70% of the respondents mentioned about their own modifying toilet facilities which is called *"kala gacher vela"* made of banana tree, mainly a floating toilet women and girls use for sanitation. This technique is undoubtedly very unhygienic and risky for women as there are no sanitation facilities, safety and security in this type of toilet. Fifteen percent of the respondents use sanitary latrines of school or latrines that are not inundated during flood time. They have reported that they can't use the toilets of *madrasas* because they are not allowed to go to the toilets which create sanitation problems. Other 15% of the women have said that they go to open place during nights in the flood time as there are scarcity of sanitation facilities for 400 families during floods. They face shyness and security problems so they use the toilets in nights and control their needs for going to toilets during day by not drinking enough food and water. Besides going to use toilets that are far way costs them to wet themselves on their way to toilets and shelter which makes them more careless about toilet facilities during disaster.

Women Health and Menstrual Hygiene Management in Disasters

Generally women spend around six to seven years of their lives menstruating so menstrual hygiene is a vital and very sensitive issue for women in reproductive ages. Every woman has the right of having safe, personal and cultural surroundings to manage their menstruation in healthy and hygienic ways as well as with dignity. However, the ability to enjoy this right is often far from reality because women health is often not only a neglected issue or less important issue but also menstrual hygiene management is often neglected in general health agendas. This can be seen in emergency situations like disaster periods such as in the time of flood, cyclones etc. most temporary or permanent shelters are not women friendly and therefore women's security, privacy and health needs are largely ignored. Especially in developing countries like Bangladesh, menstruation is often handled in secrecy. In emergency situations, the normal life style of victims has changed and they are under immense psychological pressure which aggravates problems. Providing basic needs such as food and medicine gets priority while the pressing need of securing menstrual hygiene is often neglected.

Knowledge on Menstruation: Menstrual hygiene knowledge and practices of an area or community are very important to assess the overall hygiene and health management of women of that area or community. Among 100 respondents, most of the girls and women (about 49%) have experienced their first "period" at the age from 10 to 12 years, while 46% of them have their first period in 13-15 years age and very few (only 5%) women and girls have their period in years between 16 to 18 years.

A girl's first menstruation is called menarche. Most of the respondents (72%) of the study area don't have the knowledge of menstruation before menarche. It has been explored from the study that most of the women and girls do not have adequate knowledge on menstrual hygiene management and they have got their first knowledge on menstruation after having their first "period"/menstruation. Recently some of the respondent (28%) especially adolescent girls (especially those are in secondary high schools) are getting this knowledge from school and friends but their percentage is low. From the FGD of adolescent school going girls, it is found that Menstruation education is not taught at primary level at all, and at secondary level only in biology and home economics, which are not compulsory for everyone. Just over 50% of girls and women got their first knowledge of menstruation from their sister and sister in law as they feel hesitation to talk about it with their mother whereas 31% of them get knowledge from friends and 15% get knowledge from their relatives.

Regularity of "period" is a good sign of menstrual hygiene maintenance. Irregular period is not a symptom of good hygiene. A majority (58%) of the women are having regular period which also include the girls (7%) having experience of the menstruation recently (less than 5 months). Around 30% are having irregular period and a little portion of them are taking medicine from Family Welfare Centre (*Ma Ebong Shishu Kalyan Kendra*) to continue their period. From the key respondents (Family Welfare Centre Doctor), it has been found that they have patients regarding irregular period who have come to them for medicine to regulate their menstruation. 14% of the women have experienced their menopause in the study area.

Menstrual practices: During menstruation, it is common for the Bangladeshi woman, especially from poor socio-economic background to use pieces of old clothing to absorb blood. A significant number (56%) of the women uses old cloths or rags in their menstruation time. Only a few girls (11%) use sanitary pads. Some of them (14%) use both sanitary pads and rags for menstruation. Just over 15% of the women especially elderly women use nothing as they said that they experience lower blood flow which is extremely unhygienic. Only 2% of female respondents use cotton as sanitary pads is too costly for them.

Menstrual hygiene can be maintained through changing the cloths/rags or pads of every 4 hours which is necessary for maintaining gynaecological health. But it is not possible for rural women and girls to change their cloths in that interval as they are not aware of proper menstrual hygiene maintenance. Over 50% women (13% adolescent and 41% women) change their menstrual cloths once per day whereas 37% (9% adolescent and 28% women) of the respondents change the menstruation cloths/pads twice per day. Only

9% (2% adolescent and 7% women including youth and middle aged group) of the respondents change their cloths three times per day.

Cleaning and Drying Techniques of Menstrual Cloths: Management of used cloths include washing and reusing of the cloths and also the disposal of cloths. Around 90% of the respondents wash and reuse their cloths. Some (11%) dispose their cloths after it is too old to use or they dispose their sanitary pad after one use and mainly bury them in soil. In the study area around three fourth of the respondents use soap and water in normal time while washing their menstrual rags. Only 26 % uses only water to clean which is extremely unhygienic. They mainly clean their cloths after the menstruation is over completely.

According to female respondents, drying of cloths is a matter of shame to them. They want to dry their menstruation cloth very secretly so that the male members in the house can't see it. More than 26.9% of the respondents dry their cloths on the roof of their house with other cloths. But most of them (31.4%) dry their cloths in the corner of their room with other cloths and in the monsoon time they use the dump cloths again though it is no completely dry. Just over 20% of the respondents dry their cloths beside the side of the toilet walls whereas only 18.59% of the respondents dry their menstruation cloths in sunlight but covered with other cloths.

Menstrual Hygiene Management Scenario during Disasters: The menstrual hygiene management during normal time is different than the time of disaster as the situation is not friendly and cooperative enough during disaster. Only 18% women and girls store their menstrual cloths (clean cloths and sanitary pads) if they have the probability of having menstruation in that time. Majority of the respondents (82%) have no preparation regarding menstruation hygiene before disaster. There is a difference in the time of disaster and in normal time in regards to using menstrual cloths. It is not possible always to buy the sanitary pads before the disaster so majority such as 62% of the respondents use menstruation rags during flood time. Only 6% of the respondents use sanitary pads and other 9 % use the both. 23% of the respondents use nothing as they have nothing to use and it is difficult for them to clean the cloths during the flood time. As it is difficult to clean and reuse the cloths during disaster period, the disposal rates of menstruation cloths increases in this time. 19% of respondents especially the adolescent girls dispose their cloths or rags after menstruation as they feel lazy to clean it. But 81% of the respondents clean it in the flood water and reuse it which is very unhygienic.

Among them about 56% of the respondents only use soap and water for cleaning their menstrual cloths during disaster but they clean their cloths not only with the cleaning soap but also with the soap they use to clean body which is not hygienic whereas 44% of

the respondents use only flood water to clean their cloths as it is difficult to manage soap all the time. Middle aged and older women are careless about cleaning their cloths. At Kulkandi, the respondents mentioned that it becomes more difficult to dry their cloths during the time of floods as they have to take their shelter in riverbank dam. They have to clean and dry their menstruation cloths in home coming from shelter by walking in the flood water and they become wet as it is shameful for them to clean or dry the cloths before male members. 62% of the women dry their cloths on the roof of the houses during this time as the roof are not inundated this time. Those who live in house or shelters dry their cloths with other cloths in the corner of the shelter or house. Other 12% of them dry their cloths in sunlight with other cloths.

Health Problems of Women and Girls: The respondents in the study area face many health problems regarding their menstruation. More than 32% of the female respondents have abdominal pain with weakness during menstruation. 24.48% of the respondents have rashes in their private parts during flood time or normal time as they don't clean the cloths or maintain any hygiene practices during or before disaster. 21.99% of the respondents have reported about having urinary tract infection and this is because they eat less and don't drink enough water and using one cloth for too long time during menstruation. 19.09% of the respondents reported about vaginal infection which increases after the flood according to them. A very tiny fraction of the respondents (0.02%) of the respondents are having ovary cyst and irregular period because of that.

Social Problems: Health problems are also associated with social problems in the study area. Women and girls in the study area face many social problems during disaster. Physical assaults become common while living in the shelter or riverbank as there is no privacy or safety situation. Adolescent girls and young women become mostly victimized by physical assault by the young boys. Family members try to save their girls from the evil boys and men. Women have reported about domestic violence from their husband and male children. During the time of disaster the earning person of the family became unemployed that creates frustration so they become angry and often beat their wives and girl children without any reason. 15% of the women have reported about sexual harassment such as being inappropriately touched by other men while staying in embankments when they go to collect water or washrooms. Privacy becomes a severe problem during the disaster period. Fig. 3 shows the social problems faced by the women during disasters in the study area.

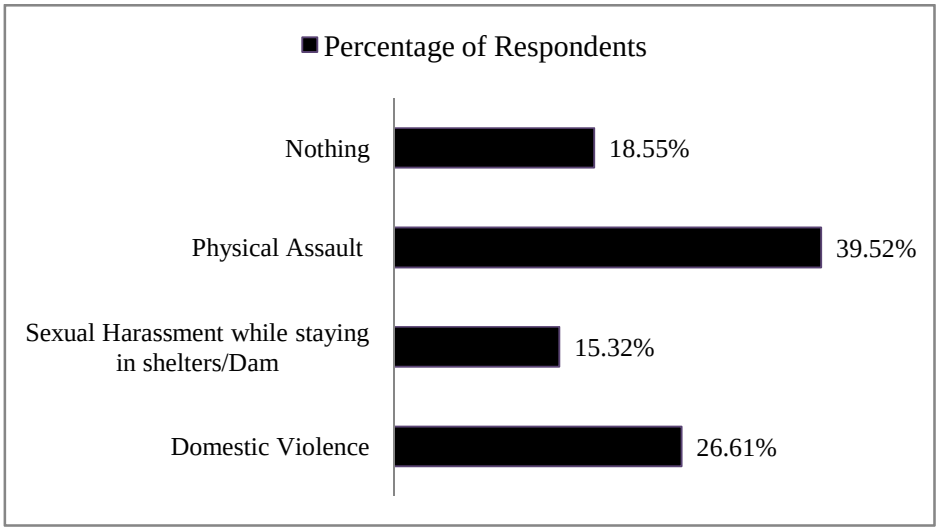

Fig. 3. Social problems faced by the respondents during disasters.

Availability of Menstrual Health Care Facilities: All of the respondents have reported that they have no health care facilities regarding menstrual hygiene in the study area Kulkandi. They have to go to *upazila* hospitals or Jamalpur Sadar hospital or family welfare clinics for getting help for gynaecological problems. For their gynaecological problems women feel shy to go for hospitals or clinics. Among all the respondents, 47% of the respondents go to MBBS doctors in the *upazila* hospitals or other private hospitals for their gynaecolocial health problems in the study area. Twenty eight percent still go to *kabiraz* or *jharfuk* for their health related problems like menstruation problems, pregnancy, typhoid, jaundice etc. and 25% of the respondents go to local quacks as they are too poor to see a MBBS doctor.

Availability of Normal Health Care Facilities: According to most of the respondents (22.96%), Sadar hospitals have all the facilities so they go there for their treatment. Other female respondents about 19.07% go to Dewanganj and 20.62% of the respondents go to Islampur *Upazila* Hospital as these two are not more distant from Kulkandi. As Kulkandi clinic has very few facilities, only 17.90% of the respondents go there for their normal problems like fever or diarrhoea treatment. Only 5.05% of the respondents are able to go to private clinics because these clinics are costly (Table 2).

Table 2. Respondents visiting different health care centres.

Types of health care centres	Frequency (Multiple responses, N)	% respondents visit these places
Kulkandi Health Clinic	46	17.90
Islampur *Upazila* Hospital	53	20.62
Dewanganj *Upazila* Hospital	49	19.07
Jamalpur Sadar Hospital	59	22.96
Family Welfare Centres	37	14.40
Other Private Clinics	13	5.05
Total	N=257	100%

During the time of disaster only 44% get their health care facilities from the health clinic of BDRCS in Kulkandi Union. The male doctor visits the study area every three months. But women is deprived of that regarding menstruation as the clinic has only one male doctor so they feel hesitation and cannot tell their problems regarding menstruation. Only 25% of the female respondents get the health care facilities such as saline and painkillers for fever during disaster from Family Welfare Centre but nothing for their gynaecological problems. Other 31% of the respondents don't get any facilities during disaster.

Existence of Awareness or Training Programs: Awareness on MHM (Menstrual Hygiene Management) is very poor in the study area. Though Bangladesh Red Crescent Society (BDRCS) has organized trainings on MHM in 2015, still they are not aware enough to maintain proper hygiene during their menstruation. Only the adolescent girls are aware and try to maintain their menstruation practices hygienic. Adolescent girls also have the menstruation education in school in class eight but the education is not compulsory for every student. Only 23% of the respondents are aware of MHM where most of the respondents (77%) are not aware of any training or awareness program of MHM.

Work Management of the respondents during disaster: It is found that during the disaster, 36.28% of the respondents were busy with their family maintenance and management. 30.23% of the female respondents did the cooking for their family members and 20% were responsible for collecting drinking water from shelter or tube wells and 13.49% of the respondents are responsible for collecting relief facilities as men become too lazy and frustrated during this time.

Types of Decision Made by Women before and after Disasters: Decision making power before the time of disaster and after disaster is the indicator of women's position and empowerment in a society. Women are mainly responsible for family management and even their house management. Tables 3 and 4 show the types of activities including decision making by the women during and after disasters.

Table 3. Types of decision taken by women before disasters.

Types of decision	Frequency (Multiple responses, N)	Percentage
Storing Menstrual Cloths/pads	18	7.32
Storing dry foods	44	17.89
Storing fuels	38	15.45
Be aware and create awareness	19	7.72
Store necessary things	52	21.14
Savings	46	18.70
Creating sanitation facilities for women	29	11.79
Total	N=246	100%

Table 4. Types of decision taken by women after disasters.

Types of Decision	Frequency (Multiple Responses, N)	Percentage
Reconstruction or cleaning houses	79	32.24
Cleaning debris	84	34.28
Helps in earning	18	7.36
Help others	64	26.12
Total	N=245	100%

Findings related to Menstrual Hygiene Management of Adolescent Girls and Women: Menstrual Hygiene management problems are age specific. From the Qualitative research method (FGD and KII survey), it has been identified that the needs and problems of adolescent girls and women are different. Findings from the study reveal that adolescent girls miss their school 2 to 3 days every month. Many girls do not learn about menstruation until they menstruate themselves and, when they do, they have many unanswered question. It is also reported that menstruation is a topic that should be kept secret and it cannot be discussed openly. Most of the girls do not prepare for their period to arrive and go home from school when it happens and they become less active and participate less in daily life when menstruating. They receive most of their information and practices regarding menstruation from sister, friends, sister-in laws and mother. They desire to have access to disposable pads but they have no dispose dustbin in the school for sanitary pads that makes them uncomfortable to dispose and they carry it to home and then dispose it by burying them in soil. Some schools have separated washrooms for girls and boys but not with hygiene facilities like soap for hand wash. Finally, the price of the sanitary pads is very costly. In Kulkandi Village, the price of the sanitary pads were found very high (such as 120 Taka) for each sanitary pad packet whereas in Sadar the price is only 70-80 Taka per packet.

Findings from adolescent girls who are not going to school: They mainly use rags but don't change them much (once a day) when menstruating. They do not eat much this time. During flood they have only a little meal in this time. They believe that it will decrease their blood flow which is good. They feel uncomfortable in using cloths as they have to clean it for a long time and bloods is not fully soaked in the rags. During disaster they don't have the facilities to clean their menstruation rags properly and they don't have the place to dry. As a result they often use the dump, unclean rags. They feel abdominal pain and weakness during menstruation and they do not have the energy to do the housework during their menstruation time. They have to go to washroom less during flood time so they eat and drink less food and water. They do not go out much during this time.

Findings from middle aged women: Women are careless about the changing of their rags. They mainly change their rags once in a day. They are careless about hygiene too. They often don't use any cloths during disaster and don't use soap to clean the rags during floods. They clean their rags with unclean flood water and they don't get any health care facilities regarding menstruation hygiene. They don't get any relief facilities for menstruation practices like cloths or pads. They have no female doctor in the health care clinic in Kulkandi. Very often they use dump cloths when they don't find enough chance to dry their cloths

Myths and Taboos: From FGD, It was found that myths and taboos were not that much strict in the area but still those exists such as for Muslim women praying and touching religious books or going to *Masjid* is strictly prohibited whereas for non- Muslims such as for Hindu women participating in *pujas* are prohibited. There are others taboos such as:

- "Eat less amount of food so that blood flow will be low".
- "Don't drink water too many times so that there will be no need to go to washroom several times".
- "Don't go outside after sunset, otherwise there will be evil influence *(kharap batash)* on you!"
- "Don't eat fish and meat (No fishy smell from blood)"

The present study reveals that women are unfairly treated when they extremely require satisfying their needs particularly in terms of menstrual hygiene before and during disaster. Though menstruation is a natural process, it has been and still is a matter of secrecy in most of the developing countries including Bangladesh. Women and girls are more vulnerable than men and during disasters there is no special relief facility regarding health and menstrual hygiene for them in the study area. Unhygienic sanitation and unclean water during disaster leads them to severe health issues including gynaecological

problems. The study identified that menstruating girls and women face tremendous challenges during disasters. Managing their menstruation and maintaining a good standard of menstrual hygiene is difficult for the women and girls because of the factors existing in the study area, such as inadequate knowledge, culture and traditions, inadequate facilities and services, poverty and lack of access to health care services. In order to satisfy the needs uncovered by the present study, it is needed to establish a health care clinic specialized for women and provide female gynaecologist and nurses as early as possible. Govt organizations and NGOs should be co-operative in terms of giving reliefs to the flood affected areas considering the menstrual hygiene management. Clean cloths and sanitary pads with free supply of medicine for skin diseases should be added in the relief materials for women. Flood shelters should be established in Kulkandi with the facility of soaps and water to promote safe sanitation including separated toilets for Male and Female population to ensure their privacy and safety during disasters. Effective cooperation between local NGOs and Government is needed to promote increased amount of food and medicine relief materials to all flood affected people specially women and children in order to maintain women and children nutrition and health care facilities. Govt. should arrange workshops and trainings on menstrual health and hygiene knowledge and management in order to create awareness among women and adolescent girls. This topic should be added as educational curriculum and made compulsory for both male and female students in order to create awareness and understanding situation. More volunteers should be trained by Govt. And local NGOs to facilitate the women and children during disaster (flood time) in community level. Menstrual hygiene management (MHM) should be included in the national level policy.

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KNOWLEDGE OF ETHNOMEDICAL PLANTS AND INFORMANT CONSENSUS IN AND AROUND LAWACHARA NATIONAL PARK

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Abstract

Consensus of the people's healthcare knowledge of ethno-medicinal plants in and around Lawachara national park was conducted from December 2014 to November 2015. The main aim of the study was to record and document plants species used for the treatment of various ailments and to find out level of consensus and agreement between informants regarding uses of plant for particular ailment categories. Data of medicinal uses of plants were recorded through semi-structured interviews, key informant discussions and informal conversations with local and ethnic people including herbal practitioners. A total of 124 medicinal plant species with 245 formularies to treat 53 ailments were recorded. For each species scientific name, local name, family, part used, ailments to be treated and mode of treatment are presented. Leaf is the dominant part used followed by fruit, root and rhizome, whole plant, seed, stem, bark, petiole, bulb, peduncle, latex and flower. In the documented 124 species, herbs were represented by 43%, trees by 31%, shrubs by 15% and climbers by 11% species. Oral consumption is the main mode of treatment in the study area and followed by external application. Maximum formularies were found in the six ailment groups including gastrointestinal complain, diarrhoea and dysentery, fever and cough, dermatitis, jaundice and impotence. According to the local people most of the plants (59%) were harvested from the park vegetation and minimum (41%) harvested from cultivated source. Maximum consensus values (Factor Informants Consensus (Fic) values more than 0.80) were obtained in case of cut and wounds and followed by Jaundice, Respiratory related ailments, Diabetes, Diarrhoea and dysentery, Anthelmintic, Gastrointestinal complain, Impotence, Dermatitis and High blood pressure. *Chromolaena odoratum, Cericoides campanulata, Oroxylum indicum, Cuscuta reflexa, Averrhoa carambola, Cajanus cajan, Justicia adhatoda* and *Citrus aurantifolia* showed 100% Fidelity values (Fl). *Litsea glutinosa, Mikania cordata, Ocimum sanctum* and *Azadirachta indica* were scored maximum Percent of Respondents knowledge values (PRK). It is recommended that species which showed high Fic, Fl and PRK values could be used for further ethno-lead phytochemical analysis to investigate active compounds to discover drugs from plants. Finally, a number of threats to medicinal plants were identified and some measures for conservation are also suggested.

Key words: Consensus, Healthcare, Ethno-medicinal, Lawachara National Park

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Introduction

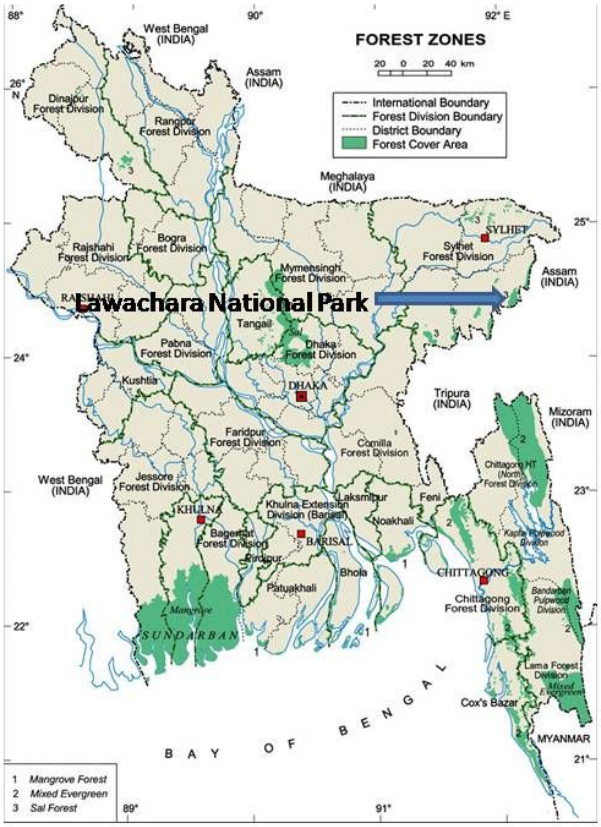
Consensus of the people's in the use of healthcare scientific knowledge of ethno-medicinal plants is the gateway in identifying new plant products of potential and commercial values. It is estimated that there are 250,000 to 500,000 species of plants on Earth (Borris 1999). A relatively small percentage (1 to 10%) of these is used as food by both humans and other animal species. It is possible that even more are used for medicinal purposes (Moerman 1996). Hippocrates (in the late fifth century B.C.) mentioned 300 to 400 medicinal plants (Schultes 1978). In the first century A.D., Dioscorides wrote *De Materia Medica*, a medicinal plant catalog which became the prototype for modern pharmacopoeias. Documented medicinal plants with high degree of consensus can serve as a basis for future investigation of modern drug (Khan *et al*. 2014). Plant based traditional medicine plays a key role in the development of novelties in drug discovery (Wright 2005). Recent studies showed that over 80% rural people of the world rely on herbal medicines (Setzer *et al*. 2006). The world market for herbal medicines based on traditional knowledge is now estimated at US$ 60 billion (Breevot 1998). World leaders met in Rio de Janeiro during 1992 to formulate biodiversity conservation policy including agenda 21 which also gave emphasis on the documentation and sustainable utilization of traditional knowledge of medicinal plants.

Currently, Ethnomedicinal knowledge of plants has been eroding at alarming rate from the nature before proper documentation and evaluation. In order to protect such knowledge, documentation of ethnomedicinal plants has already been started in Bangladesh. A number of articles was published in this field including Mia and Huq (1988), Hassan and Khan (1986, 1996), Alam (1992), Alam *et al.* (1996), Uddin (2006), Uddin *et al.* (2001), Khan *et al.* (2002), Yusuf *et al.* (2002), Uddin *et al.* (2004), Uddin *et al.* (2006), Yusuf *et al.* (2006), Uddin and Roy (2007), Uddin *et al*. (2008), Uddin *et al.* 2012, Haque *et al.* (2014) and Uddin and Hassan (2014). All such articles were listed a good number of medicinal plants of particular community or particular diseases or particular areas of Bangladesh. But there are still more medicinal plants used as sources of herbal drugs by the ethnic and local people of Bangladesh yet to be discovered. Unfortunately no such works have covered the quantitative documentation of ethnomedicinal plants of rural people living in and around Lawachara national park. The park supports a large number of plant species. Among them many species are medicinal those need to be documented and conserved. Local people in and around the park had been using such plants in their primary health care. Currently medicinal plants and traditional knowledge have been eroding due to globalization and climate change. Some of such knowledge is going to eliminate before documentation which is alarming to sustain cultural heritage. In order to save the healthcare knowledge and medicinal plants, in the present study an attempt was made to achieve the following objectives: to record, integrate and document all scattered distribution of traditional healthcare knowledge of medicinal plants; to determine ethno-medically potential and culturally important and

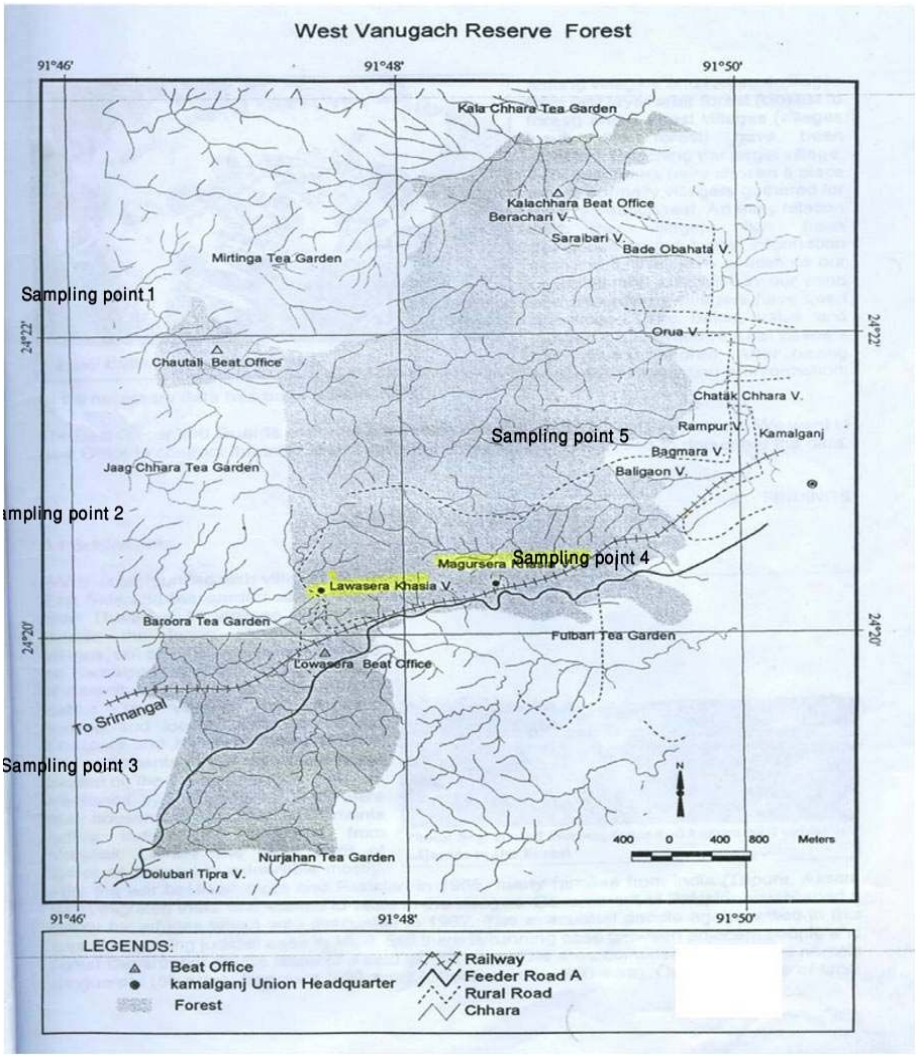
most cited medicinal plants using statistical models; to find out the threats to medicinal plants in the natural habitats and to suggest measures for the conservation

Materials and Methods

Lawachara national park, under Kamalganj upazila of Maulvi Bazar district, is a part of West Bhanugach reserve forest, which was declared reserve in early nineteenth century as per the Forest Act 1878, the Assam Forest Manual 1898 and the Forest Act 1927 (USAID 2006). The park is located nearly 160 km northeast of Dhaka and approximately 60 km south of Sylhet city (Map 1).It lies between 24˚30´-24˚32´ N and 91˚37´-91˚39´ E. The forest was declared as a national Park in 1996 having a total area of 1250 ha and with a plan to extend this area further to include 281 additional ha of the Reserve Forest (Green 1990, Canonizado and Rahman 1998, Riadh 2007 and Ahsan 2007).



Map 1 (a). Bangladesh Map showing Lawachara National Park.



Map 1. (b) Enlarged part showing different sampling points of Lawachara National Park. (Source: Collected from Web).

Present forest types of Lawachara are a combination of planted exotic species and mixed forest with a deciduous canopy and an evergreen understory (Ahsan 2000). The forest originally supported an indigenous vegetation cover of mixed tropical evergreen type (Alam 1998). The topography of Lawachara National Park is undulating, with slopes and hillocks that range from 10 to 50 m in elevation (Rizvi 1970 and Riadh 2007). These hillocks are scattered and interspersed with numerous streams that flow through the forest. The hills are composed of upper tertiary rocks in which sand stone largely

predominates (Ahmad 1970 and Stevens 1986) along with siltstones and mudstones, locally altered to slates and shales. The significant soils in the hills of Maulovi Bazar belong to Ramgarh and Rangamati series on Dupitila formation (Stevens 1986). Soils of the park are generally sandy loam and the rest are mostly clayey loam (Ahmad 1970). The area enjoys a moist tropical climate characterized by a period of high precipitation from April to September and five months of relatively dry period from November to March.

Methods of data collection

The study area has been visited four times in different seasons of the year of 2015 including summer, rain, autumn and winter. Each field trip lasted for five days. The data of medicinal uses were recorded through semi-structured interviews, key informant discussions and informal conversations with local people including herbal practitioners (Alexiades 1996, Chambers 1994 and Martin 1995). A total of 163 informants was interviewed using questionnaire. Among them 117 male and rest 46 are female. Age ranges from 15 to 95 years old. Education levels of the informants were from illiterate up to Bachelor degree. Professionally they were mostly farmer, day labor, house wife, small shopkeepers and medicine men. During the field survey, information on uses of plants to treat human, parts used, mode of preparation and administration was documented. The vernacular names were collected with the help of local people. Voucher specimens for each medicinal plant were collected and processed using standard herbarium techniques (Hyland 1972 and Alexiades 1996). The specimens were identified consulting different Floras viz., Hooker 1872-1897, Prain 1903, Uddin and Hassan 2004, Siddiqui *et al.* 2007c and Ahmed *et al*. 2008a, 2008b, 2009b, 2009c, 2009d and 2009e. Specimens available at Dhaka University Salar Khan Herbarium (DUSH) were consulted in identifying the collected plant specimens. The updated nomenclature of the species followed Siddiqui *et al.* 2007c and Ahmed *et al*. 2008a, 2008b, 2009b, 2009c, 2009d, and 2009e. Voucher specimens are preserved at DUSH.

Factor of informant consensus

In order to estimate use diversity of the medicinal plants and to determine which plants are particularly interesting in the search for bioactive compounds, factor of informant consensus (Fic) was calculate (Trotter and Logan 1986 and Heinrich *et al*. 1998).

Fic is thus calculated applying the following equation: Fic= Nur – Ntaxa/Nur-1

Where Nur is the number of use reports in each category, Ntaxa is the number of species in each category. The relative importance of a species is evaluated by the proportion of

respondents who cited it. The Fic provides a range of 0–1, where high values (close to 1) are obtained when only one or a few plant species are reported to be used by a high proportion of informants to treat a particular ailment. High Fic thus means that there is a narrow well-defined group of species used to cure a particular ailment category and/or that information is exchanged between informants. On the other hand, low Fic values (close to zero) indicate that informants disagree over which plant to use due to random choosing or lack of exchange of information about use among informant.

Fidelity level (Fl)

The Fl value is useful for identifying the informants most preferred species in use for treating certain ailments (Friedman *et al.*1986). the Fl index, $Fl=I_p/I_u \times 100$, where I_p is number of informants who indicate use of a species for the same major ailment, I_u is the total number of informants who mentioned the same plant for any other use. The Fl values range from 0 to 100%. Medicinal plants that are widely used by the local people for certain ailment have higher Fl values those that are less popular.

Percentage of Respondent Knowledge of medicinal plants (PRK%)

PRK values are useful to determine most common medicinal plants in the study area. PRK values of medicinal plants were estimated using the formula: (number of people interviewed citing species/the total number of people interviewed) x100 (Friedman *et al*. 1986).

Results and Discussion

A total of 124 medicinal plants from in and around Lawachara national park for the treatment 53 ailment through 245 formularies was recorded. These species belong to 65 families. For each species scientific name, local name, family, parts used, ailments to be treated, application mode treatment and citation frequency are presented (Table 1). Most cited medicinal plants families in the study area are Rutaceae, Lamiaceae, Mimosaceae, Meliaceae, Combretaceae, and Asteraceae. Diversity in parts used for medicines was recorded in the study area. Leaf is the dominant parts used followed by Fruits, root and rhizome, whole plants, seeds, stem, bark, leaf and bark, petiole, bulb, peduncle, latex and flowers (Fig. 1). Dominant parts used leaf indicated that sustainable used of medicinal plants exist in the study area. In case roots and bark used may promote extinction process of species from nature. Part used fruits and seeds also created problem in natural regeneration of plants, if they do not collect properly.

Table 1. Ethno botanical data on medicinal plants and uses in the study area (S=Shrub, C=Climber, H=Herb, T=Tree)

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
Abroma augusta (L.) L. f., Z-223	Ulatkombol	Sterculiaceae	s	Impotence	Stem	Juice is taken
Abrus precatorious L., Z-224	Jostimodhu	Fabaceae	c	Cough	Stem	Juice is taken
*Achyranthes aspera*L., Z-88	Upathlenga	Amaranthaceae	h	Jaundice	Leaf	Juice is taken
Acorus calamus L,. Z- 225	Bach	Araceae	h	Appetizer	Rhizome	Cooked rhizome is taken
Justicia adhatoda L.,Z-22	Bashak	Acanthaceae	h	cough	Leaf	Juice is taken
Aegle marmelos (L.) Corr., Z-226	Bel	Rutaceae	t	Dysentery	Green fruits	Raw fruits is taken
				Diarrhoea	Green fruits	Green fruit is taken
				Constipation	Fruits	Pulp is taken
Allium cepa L., Z-227	Peaj	Liliaceae	h	Flue	Bulb	Juice is taken
*Allium sativum*L., Z -228	Roshun	Liliaceae	h	Gastric	Bulb	Juice is taken
				Cold, cough	Bulb	Juice is taken
Alocasia cuculata L., Z- 142	Bishkachu	Araceae	h	Body ache	Rhizome	Cooked rhizome is taken
				Rheumatic pain	Root	Juice is taken
Alocasia macrorrhizos (L.) G. Don, Z-229	Mankachu	Araceae	h	Rheumatic pain	Rhizome	Cooked rhizome is taken
Aloe vera(L.) Burm. f. , Z- 230	Alovera	Aloaceae	h	Impotence	Leaf	Juice is taken
Alstonia scholaris (L.) R. Br., Z-141	Chatim	Apocynaceae	t	Ringworm	Latex	Latex is applied
				Pimple	Latex	Latex is applied
				Dysentery	Bark	Juice is taken
				Diarrhoea	Bark	Juice is taken
				Abscess	Latex	Latex is applied
Alternenthera sessilis (L.) R. Br. Ex Roem. & Schult., Z- 12	Helencha	Amaranthaceae	h	Pox	Leaf	Juice is taken
Amaranthus tricolor L., Z-275	Laoshak	Amaranthaceae	c	Reduced pressure	Leaf	Cooked leaf taken
Amaranthus spinosus L., Z-89	Kanta dugi	Amaranthaceae	h	Urinary problem	Stem	Decoction is taken
				Rheumatic pain	Root	Juice is taken
				Dropsy	Root	Juice is taken
				Diabetes	WP	Cooked plant is taken
Amorphophallus bulbifer (Roxb.) Blume, Z-58	Olkachu	Araceae	h	Rheumatic pain	Rhizome	Cooked rhizome is taken
Andrographis paniculata (Burm. f.) Wall. *ex* Nees., Z- 31	Kalomegh	Acanthaceae	h	Malaria	WP	Juice is taken
				Diabetes	WP	Juice is taken
				Dermatitis	Leaf	Paste is applied
				Anthlemintic	Leaf	Juice is taken
Annona squamosa L., Z-231	Anaros	Annonaceae	h	Anthlemintic	Fruits, root	Juice is taken
Breonia chinensis (Lamk.) Capuron, Z-276	Kadam	Rubiaceae	t	Rheumatic pain	Leaf	Heated leaf is applied
Aphanamixis polystachya(Wall.) R. N. Parker, Z-277	Roina	Meliaceae	t	Dermatitis	Leaf	Tablet is taken
Aristolochia indica L., Z-278	Ishwarmul	Orchidaceae	c	Dysentery	Root	Juice is taken
Asparagus racemosus Willd., Z-279	Shatamuli	Liliaceae	c	Impotence	Root	Juice is taken
*Averrhoa carambola*L., Z-132	Kamranga	Averrhoaceae	t	Jaundice	Fruits	Fruit is taken

Contd.

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
Azadirachta indica A. Juss., Z-38	Neem	Meliaceae	t	Toothache	Leaf	Decoction used for Gargling
				Malaria	Leaf	Tablet is taken
				Gastric	leaf	Juice is taken
				Fever	Leaf	Juice is taken
				Diabetes	Leaf	Juice is taken
				Dermatitis	Leaf	Paste is applied
				Anthlemintic	Leaf	Tablet is taken
				Pox	Leaf	Paste is applied
Baccaurea ramiflora Lour., Z- 117	Bhubi	Euphorbiaceae	t	Appetizer	Fruits	Fruit is taken
Bambusa tulda Roxb., Z-171	Bamboo	Poaceae	s	Impotence	stem	Cooked stem taken
				Cut	Stem	Powder is applied
Bauhinia acuminata L., Z- 248	Shetkanson	Ceasalpiniaceae	t	Tears of Eye	Leaf	Juice is applied
Blumea lacera (Burm. f.) DC, Z-70	Shialmutra	Asteraceae	h	Diarrhoea	Leaf	Fried leaf is taken
*Bombax ceiba*L., Z-107	Shimul	Bombacaceae	t	Impotence	Root	Juice is taken
Bulbophyllum lilacinum Ridl., Z-247	Ishwarmul	Orchidaceae	h	Impotence	leaf	Juice is taken
				Dysentery,	leaf	Juice is taken
				Diabetes	leaf	Juice is taken
				Heart pain	leaf	Juice is taken
Bursera serrata Wall.*ex* colebr., Z-246	Neur	Burseraceae	t	Appetizer	Fruits	Fruit is taken
Cajanus cajan (L.) Millsp.,Z-245	Orhor	Fabaceae	s	Jaundice	Leaf	Juice is taken
Calotropis procera (Ait.) R. Br.,Z-04	Akanda	Asclepiadaceae	s	Ringworm	Leaf	Paste is applied
				Rheumatic pain	Leaf	Heated leaf is applied
Canabis sativa L., Z-249	Gaza	Malvaceae	s	Reduced pressure	Leaf	Juice is taken
Carica papaya L., Z- 17	Pepe	Caricaceae	s	Stomach ache	Fruits	Cooked fruit is taken
				Jaundice	Fruits	Cooked fruit is taken
				Gastric	Fruits	Cooked fruit is taken
Careya arborea Roxb., Z-244	Bidipata	Lecythidaceae	t	Dysentery	Leaf	Juice is taken
Cassia alata L., Z-250	Daudgash	Caesalpiniaceae	s	Ringworm	Leaf	Juice is applied
Cassinopsis ilicifolia (Hochst.) Sleumer., Z- 100	Kantalebu	Rutaceae	s	Fever	Fruits	Juice is taken
Centella asiatica (L.) Urban, Z- 52	Tunimakuni	Apiaceae	h	Urinary problem	WP	Cooked plant is taken
				Jaundice	Wp	Juice is taken
				Gastric	WP	Cooked plant is taken
				Dysentery	WP	Paste is taken
				Diarrhoea	WP	Cooked plant is taken
				Cataract eye	Leaf	Juice is applied
				Brain tonic	WP	Juice is taken
Ceriscoides campanulata (Roxb.) Tirveng., Z-239	Behlom	Rubiaceae	s	Jaundice	Fruits	Cooked fruit is taken

Contd.

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
Phyllanthus acidus (L.) Merr., Z-251	Leboi	Euphorbiaceae	t	Fever	Fruits	Fruit is taken
Citrus aurantifolia (Christm. & Panzer) Swingle, Z- 40	Lebu	Rutaceae	s	Jaundice	Fruits	Juice is taken
Clerodendrum viscosum Pers., Z-238	Bhait	Verbenaceae	h	Stomach ache	Leaf	Juice is taken
				Dysentery	Leaf	Juice is taken
				Cut	Leaf	Juice is applied
				Cough	Leaf	Juice is taken
				Anthlemintic	Leaf	Juice is taken
Coccinia cordifolia Congn., Z- 13	Telakucha	Cucurbitaceae	c	Diabetes	Leaf	Cooked leaf taken
Cocos nucifera L., Z-240	Narikel	Arecaceae	t	Jaundice	Green fruits	Water is taken
				Diarrhoea	Green fruits	Water is taken
Colocasia esculenta (L.) Schott., Z-08	Kachu	Araceae	h	Iron tonic	WP	Cooked plant is taken
				Cut	Leaf	Juice is applied
Crinum asiaticum Roxb.,Z-209	Crinum	Liliaceae	h	Cow gastric	Fruits	Paste is taken
Curcuma zedoaria (Christm.) Rosc., Z-281	Shathi	Zingiberaceae	h	Diarrhoea	Rhizome	Powder is taken
Cuscuta reflexa Roxb.,Z-280	Cuscuta	Cuscutaceae	h	Jaundice	Stem	Juice is taken
Cynodon dactylon (L.) Pers., Z-158	Durba	Poaceae	h	Cut	WP	Juice is applied
				Cut and wound	Leaf	Paste is applied
Dalbergia sissoo Roxb.,Z-274	Sissue	Fabaceae	t	Jaundice	Leaf	Juice is taken
Datura metel L.,Z-273	Dutra	Solanaceae	s	Dermatitis	seed	Paste is applied
Dillenia indica L., Z- 143	Chalta	Dilleniaceae	t	Reduced pressure	Fruits	Fruit is taken
Dillenia pentagyna Roxb., *Z-242*	Harganja	Dilleniaceae	t	Fractured bone	Leaf, Bark	Paste is applied
Eclipta prostrata (L.) Mant, Z-11	Kesharaj	Asteraceae	h	Impotence	WP	Juice is taken
				Hair tonic	Wp	Juice is applied
Entada scandens auct. *non* Benth., *Z-241*	Gila	Mimosaceae	c	Rheumatic pain	Seed	Seed is taken
Erythrina indica Lamk., Z-14	Mandar	Fabaceae	t	Jaundice	Leaf, Bark	Juice is taken
Chromolaena odoratum(L.) King and Robinson , Z-272	Pisais	Asteraceae	h	Cut	Leaf	Paste is applied
Paederia foetida L. Z-272	padra pata	Rubiaceae	c	Diarrhoea	Leaf	Cooked leaf taken
Ficus benghalensis L., Z- 77	Bot	Moraceae	t	Diabetes	Fruits	Cooked fruit taken
Ficus racemosa L.,Z-271	Jogdumur	Moraceae	t	Diabetes	Fruits	Cooked leaf taken
Cyperus rotundus L., Z-168	Gandhavadlu	Cyperaceae	c	Diarrhoea	Leaf	Cooked leaf taken
Garcinia cawa Roxb. *ex* DC., Z-270	Kao	Clusiaceae	t	Appetizer	Fruits	Fruit is taken
Garcinia xanthochymous Hook. f. *ex* T. Anders, Z-269	Dayphal	Clusiaceae	t	Appetizer	Fruits	Fruit is taken

Contd.

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
Glycosmis arborea (Roxb.) A. DC., Z-20	Awapata	Rutaceae	s	Stomach ache	Leaf	Juice is taken
				Jaundice	Leaf	Juice is taken
				Heart pain	Leaf	Juice is taken
				Head ache	Leaf	Leaf paste is applied
				Fever,	Leaf	Juice is taken
				Dysentery	Leaf	Juice is taken
				Cough	Leaf	Juice is taken
				Appetizer	Leaf	Juice is taken
				Anthlemin tic	Leaf	Juice is taken
Hibiscus sabdariffa L., Z-60	Amila	Malvaceae	h	Jaundice	Leaf	Cooked leaf is taken
Hygrophila spinosa T. Anders., Z-268	Talmakhna	Acanthace ae	h	Eye complain	Seed	Juice is applied
Hydnocarpus kurzii (King)Warb.,Z-87	Chalmugra	Archariace ae	t	Leprosy	Fruits	Oil is applied
				Dysentery	Root	Juice is taken
Hyptis suaveolens (L.) Poit., Z-267	Tokma	Lamiaceae	h	Reduced pressure	seed	Seed is taken
			h	Dysentery	Seed	Juice is taken
			h	Constipati on	seed	Seed is taken
Ipomoea fistulosa Mart. *ex* Choisy, Z-203	Khulum	Convolvul aceae	h	Cut	Latex	Latex is applied
Ipomoea mauritiana Jacq., Z-265	Bhuikumra	Convolvul aceae	c	Miscarriag e	Root	Juice is taken
Jatropha curcas L., *Z-266*	Keke	Euphorbia ceae	s	Toothache	Leaf	Juice is applied
Kalanchoe pinnata (Lamk.), Z-46	Pathorkuch	Crassulaceae	h	Urinary problem	Leaf	Juice is taken
				Dysentery	Leaf	Juice is taken
				Cough	Leaf	Juice is taken
Leucas lavandulaefolia Smith., Z-28	Dandakala sh	Lamiaceae	h	Stomach ache	Leaf	Juice is taken
				Diarrhoea	Leaf	Fried leaf is taken
				Cough	Leaf	Fried leaf is taken
Litsea glutinosa (Lour.) Robinson, Z- 94	Chengpisla	Lauraceae	t	Stomach ache	Leaf, Bark	Juice is taken
				Impotence	Leaf, Bark	Juice is taken
				Dysentery	Leaf, Bark	Juice is taken
				Constipati on	Leaf, Bark	Juice is taken
Lawsonia inermis L., Z- 35	Mehedi	Lythraceae	s	Impotence	leaf	Juice is taken
				Impotence	leaf	Juice is taken
				Hair tonic	Leaf	Paste is applied
				Gastric	Leaf	Juice is taken
				Birth control	Leaf	Juice is taken

Contd.

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
Mangifera indica L., Z-243	Aam	Anacardiaceae	t	Toothache	Leaf	Juice is applied
				Dysentery	Leaf	Paste is taken
Melia sempervirens (L.) Sw., Z-34	Bela	Meliaceae	t	Dermatitis	Leaf	Paste is applied
Mentha arvensis L., Z-264	Pudina	Lamiaceae	h	Stomach ache	Leaf	Juice is taken
Mikania cordata (Burm. f.) B.L. Rob., Z-261	Refujeelata	Asteraceae	c	Dysentery	Leaf	Juice is taken
				Cut	Leaf	Juice is applied
Mimosa pudica L., Z-15	Chaitamara	Mimosaceae	h	Waist pain	root	Juice is taken
				Toothache	Root	Juice is applied
				Measles	Stem	Juice is applied
				Malaria	Root	Juice is taken
				Jaundice	Leaf	Juice is taken
				Fever	Leaf	Juice is taken
				Anthlemintic	Leaf	Juice is taken
				Abscess	Root	Paste is applied
				During birth	Root	Paste is applied
Acacia intisia (L.) Willd., *Z-262*	Sadachaitamara	Mimosaceae	h	Snakebite	Root	Juice is applied
				Impotence	root	Juice is taken
				Allergy	Root	Juice is taken
Moringa oleifera Lamk.,Z-252	Sajna	Moringaceae	t	Stomach ache	Fruits	Cooked fruit is taken
				Rheumatic pain	Leaf, Bark	Cooked leaf is taken
				Diarrhoea	Leaf	Juice is taken
Murraya paniculata (L.) Jack., Z-25	Kamini	Rutaceae	t	Toothache	Leaf	Juice is applied
				Toothache	Leaf	Juice is applied
Murrya koenigii (L.) Spreng., Z-254	Norshing	Rutaceae	s	Diarrhoea,	Leaf	Cooked leaf is taken
				Cough	Leaf	Cooked leaf is taken
Musa paradisiaca L, Z-253.	Kola	Musaceae	h	Dysentery	Fruits	Fruit is taken
				Cut	Peduncle	Juice is applied
				Constipation	Peduncle	Cooked peduncle is taken
Nicotiana plumbaginifolia Viv., Z-255	Tamak	Solanaceae	h	Cut	Leaf	Juice is applied
Nigella sativa L., Z-256	Kalojira	Ranunculaceae	h	Impotence	seed	Oil is taken
				Diabetes	seed	Water extract is taken
Nyctanthes arbor-tristis L., Z-257	Shephaliful	Nyctaginaceae	s	Fever	Leaf	Juice is taken

Contd.

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
				Anthlemintic	Leaf	Juice is taken
Ocimum americanum L. Z-237	Rossetpata	Lamiaceae	h	Stomach ache	Leaf	Cooked leaf is taken
				Gastric	Leaf	Cooked leaf is taken
				Diarrhoea	Leaf	Juice is taken
				Cough	Leaf	Juice is taken
*Ocimum basilicum*L., Z-02	Kalotulsi	Lamiaceae	h	Reduced pressure	Leaf	Paste is applied
				fever	leaf	Juice is taken
				Cough	Leaf	Juice is taken
Ocimum sanctum L., Z-153	Sada tulsi	Lamiaceae	h	Reduced pressure	Leaf	Paste is applied
				Cough	Leaf	Juice is taken
				cough	Leaf	Juice is taken
Oroxylum indicum (L.) Kurz., Z-110	Kanaidingi	Bignoniaceae	t	Jaundice	Bark	Juice is applied
Oryza sativa L., Z-236	Dhan	Poaceae	H	Diarrhoea	Seed	Powder is taken
Oxalis corniculata L., Z-23	Zinzil	Oxalidaceae	h	Stomach ache	Leaf	Juice is taken
				Appetizer	Leaf	Juice is taken
Pandanus foetidus Roxb., Z--235	Keya	Pandanaceae	H	Cough	Rhizome	Juice is taken
*Phyllanthus emblica*L., Z-44	Aola	Euphorbiaceae	t	Impotence	Fruits	Fruit is taken
				Appetizer	Fruits	Fruit is taken
Piper betel L., Z- 187	Pan	Piperaceae	c	Cut	Leaf	Juice is applied
				cut	Leaf	Juice is applied
Plumbago zeylanica L., Z-261	Chita	Plumbaginaceae	h	Dermatitis	Root	Juice is applied
Cnesmone javanica Blume, Z-260	Chutra	Euphorbiaceae	h	Dermatitis	Leaf	Juice is applied
Persicaria hydropiper (L.), Spach Z-138	Bishkatali	Polygonaceae	h	Dermatitis	Leaf	Juice is applied
				Jaundice	Leaf	Juice is taken
Chylocalyx perfoliatus (L.) Hassk. *ex* Miq.L., Z- 48	Kantaamrai	Polygonaceae	h	Diabetes	Leaf	Paste is taken
				Cough	Leaf	Fried leaf is taken
*Psidium guajava*L., Z- 43	Peara	Myrtaceae	t	Toothache	Leaf	Decoction is used for gargling
				Dysentery	Leaf	Juice is taken
				Diarrhoea	Leaf	Juice is taken
Ricinus communis L., Z-258	Verenda	Euphorbiaceae	s	Constipation	Seed	Oil is taken
				Vomiting	Bark	Juice is applied
Saccharum officinarum L., Z-234	Akh	Poaceae	h	Jaundice	Stem	Juice is taken
Scoparia dulcis L., Z-16	Chinipata	Scrophulariaceae	h	Eye complain	Leaf	Juice is applied
				Diarrhoea	Leaf	Juice is taken
				Diabetes	Leaf	Juice is taken

Contd.

Scientific name, Voucher number	Local name	Family	Habit	Ailments	Part used	Treatment mode
Sesamum indicum L. Z-259	Til	Pedaliaceae	h	Eczema	Seed	Oil is applied
Smilax macrophylla Roxb., Z 233	Kumarilata	Smilaceae	c	Impotence	Leaf	Leaf is taken internally
Solanum violaceum Ortega, Z-129	Boroibegun	Solanaceae	s	Stomach ache	Fruits	Paste is taken
Spilanthes acmella (L). L., Z- 19	Piperman	Asteraceae	h	Toothache	Flowers	Juice is applied
				Ear and mouth rot	Flowers	Juice is applied
Sterculia villosa Roxb. *ex* Smith, Z- 125	Udal	Sterculiaceae	t	Jaundice	Petiole	Juice is taken
				impotence	Petiole	Juice is taken
				Gastric	Petiole	Juice is taken
				Dysentery	Petiole	Juice is taken
				Diabetes	Petiole	Juice is taken
				Constipation	Petiole	Juice is taken
Streblus asper Lour., Z-84	Sheora	Moraceae	t	Dysentery	Leaf	Juice is taken
Swietenia mahagoni Jacq., Z-42	Mehogoni	Meliaceae	t	Diabetes	Seed	Juice is taken
Syzygium cumini (L.) Skeels, Z-39	Jam	Myrtaceae	t	Toothache	Leaf	Juice is applied
				Diabetes	Seed	Seed is taken
*Tagetes erecta*L., Z- 79	Gada	Asteraceae	h	Fire injury	leaf	juice is applied
				Cut	Leaf	Juice is applied
Tamarindus indica Willd., Z- 116	Tentul	Caesalpiniaceae	t	Sensitize	Fruits	Fruit is taken
				Reduced pressure	Fruits	Fruit is taken
				Diarrhoea	Fruits	Fruit is taken
				Appetizer	Fruits	Fruit is taken
Terminalia arjuna (Roxb. *ex* DC.) Wight & Arn., Z- 130	Arjun	Combretaceae	t	Heart pain	Bark	Juice is taken
				Gastric	Bark	Juice is taken
				Dysentery	Bark	Juice is taken
Terminalia bellirica (Gaertn.) Roxb., Z- 108	Bohera	Combretaceae	t	Stomach ache	Fruits	Fruit is taken
				Impotence	Fruits	Fruit is taken
				Dysentery	Fruits	Fruit is taken
				Appetizer	Fruits	Fruit is taken
Terminalia chebula Retz., Z- 55	Horitaki	Combretaceae	t	Stomach ache	Fruits	Fruit is taken
				Jaundice	Bark	Decoction is taken
				Impotence	Fruits	Fruit is taken
				Appetizer	Fruits	Fruit is taken
Tinospora crispa (L.) Hook. f. & Thoms, Z-232	Gulancha	Menispermaceae	c	Anthlemintic	Stem	Juice is taken
Vitex negundo L., Z- 131	Nishinda	Verbenaceae	s	cough	Leaf	Juice is taken
Vitis quadrangualris Wall. *ex* Wight & Arn., Z- 136	Harjora	Vitaceae	c	Fractured bone	Stem	Paste is applied
Zingiber officinale Rosc., Z-114	Ada	Zingiberaceae	h	Neck pain	Rhizome	Decoction is taken
Ziziphus mauritiana Lamk., Z-05	Boroi	Rhamnaceae	t	Wound	Leaf	Boiled water applied

Of 124 species documented in the study area, herbs have been represented by 43%, trees by 31%, shrubs by 15% and climbers by 11%of total species (Fig. 2). The result reflected that herbs are the most dominant life form among the medicinal plants in the study area. In order to maintain medicinal diversity in nature, herbaceous plants can be cultivated easily as a major source of crude drugs because of short life cycle. Usually people take medicines in different ways including oral application, external application and adjunct therapy. Oral application is the main mode of treatment in the study area and followed by external application (Fig. 3).

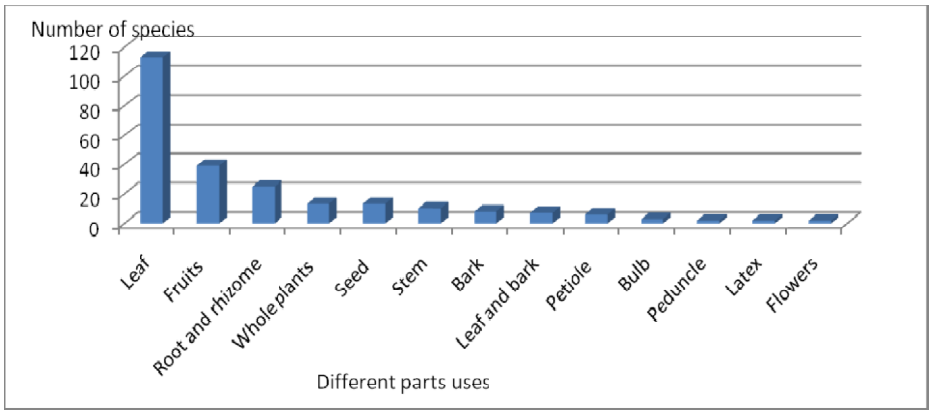


Fig. 1. Diversity in parts used of medicinal plants.

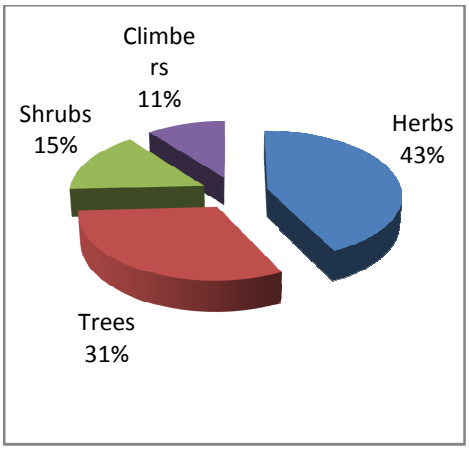


Fig. 2. Different life forms of species.

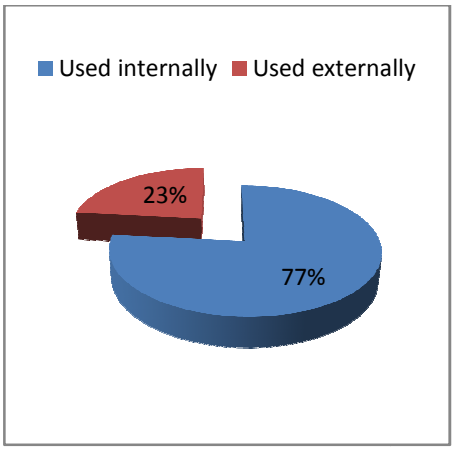
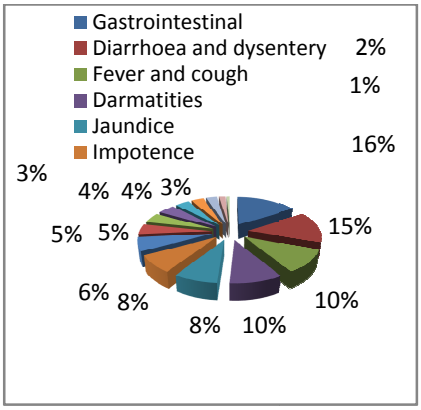


Fig. 3. Application modes of medicines.



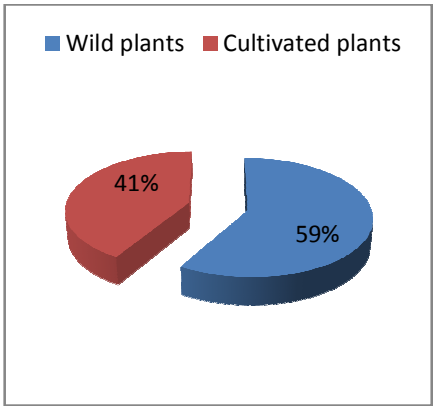


Fig. 4. Fomularies.

Fig. 5. Sources of medicinal plants.

Number of formularies for major ailment group showed variation. Maximum formularies were found in the six ailment groups including gastrointestinal complain, diarrhea and dysentery, fever and cough, dermatitis, jaundice and impotence (Fig. 4). It is proved that such complains are very common in the study area. According to the local people most plants (59%) were harvested from the wild vegetation and minimum (41%) harvested from cultivated plants (Fig.5). Cultivation of medicinal plants is still in low because of availability of wild vegetation in national park. It is observed in the field that harvesting of medicinal plants from the wild is not sustainable. Local people are not aware of sustainable use of medicinal plants. They like to earn some cash money for their livelihood by selling illegal plant parts from the wild. As far literature there is no appropriate policy and guide line for medicinal plants harvesting from the wild in Bangladesh. Currently some cultivation program of medicinal plants has been started here in Bangladesh. But herbal industries are yet depending on foreign supply for raw drug materials (Personal communication).

Table 2. Consensus of agreement in the uses of medicinal plants among the informants.

Major ailments group	Nur	Nt	Fic
Cut and wound	188	15	0.93
Jaundice	196	21	0.90
Respiratory related (malaria, fever and cough)	235	26	0.90
Diabetes	90	12	0.88
Diarrhoea and dysentery	258	36	0.87
Anthelmintic	50	8	0.86
Gastrointestinal complain	263	39	0.86
Impotence	118	18	0.86
Dermatitis	155	24	0.85
High blood pressure	62	12	0.82
Urinary disorder	32	7	0.81
Rheumatic pain	49	11	0.79
Toothache	34	11	0.70
Eye complain	9	4	0.63

Based on the information obtained from the informants, the recorded ailments were grouped into 14 categories (Table 2). The results could be useful in prioritizing medicinal plants for further scientific validation of plant products, as pharmacologically effective remedies with higher Fic values. Fic values range from 0.00 to 1.00. High values are obtained when only one or few plants species are reported to be used by a high proportion of informants to treat a particular ailment, whereas low Fic values indicate that the informants disagree over which plants to use. Higher Fic values can thus be used to pinpoint particularly interesting species for the search of bioactive compounds. In our analysis Fic values for major ailment categories showed variation (Table 2). The average Fic value for all ailment categories obtained was0.78. Such value indicated that maximum people in the study area were well informed about the medicinal knowledge of plants and also showed agreement on the use of medicinal plants for such categories of ailments. Top Fic values found in case of ailment cut and wound. The most cited species used to treat such ailment are *Mikania cordata, Chromolaena odoratum* and *Cynodon dactylon*. Among them *Cynodon dactylon* and *Chromolaena odoratum* showed maximum Fl values.These species can be used for further phytochemical analysis to find active compounds for treatment of cut and wound. Jaundice and respiratory related (malaria, fever, cold and cough) treatment scored second highest Fic values. Most cited species to treat such categories of ailmentwere *Ceriscoides campanulata, Oroxylum indicum, Cuscuta reflexa, Averrhoa carambola, Citrus aurantifolia, Cajanus cajan, Ocimum sanctum, Andrographis paniculata, Ocimum basilicum and Justicia adhatoda* . Such species can be used for further phytochemical analysis to find active compounds for the treatment of Jaundice and respiratory related ailments.

Fidelity level is useful for identifying the key informants most preferred species used for treating certain ailments. The medicinal plants that are widely used by the local people have higher FL values than those that are less used. On the other hand, medicinal plants that are known as remedies of a single aliment have 100% fidelity level than those that are used as remedies for more than one type of aliment. In our analysis Fl values showed variations. Among the most cited species 9 scored Fl values of 100%. The maximum Fl for plants indicated 100% choice of informants for treating specific ailments. Such species are *Chromolaena odoratum, Ceriscoides campanulata, Oroxylum indicum, Cuscuta reflexa, Averrhoa carambola, Cajanus cajan, Justicia adhatoda* and *Citrus aurantifolia.* These are cultural bounded species and indicated their healing potential.

Table 3. Fidelity level (Fl) values of the frequently reported plants and their major uses.

Scientific name	Ailments	Number of informants (Ip)	Total informants (Iu)	Fidelity level (%)
Chromolaena odoratum	Cut and wound	32	32	100
Ceriscoides campanulata	Jaundice	28	28	100
Oroxylum indicum	Jaundice	27	27	100
Cuscuta reflexa	Jaundice	25	25	100
Averrhoa carambola	Jaundice	22	22	100
Cajanus cajan	Jaundice	21	21	100
Justicia adhatoda	cough	20	20	100
Citrus aurantifolia	Jaundice	20	20	100
Cynodon dactylon	Cut and wound	37	37	100
Syzygium cumini	Diabetes	25	26	96.15
Mikania cordata	Cut and wound	58	61	95.08
Ocimum sanctum	Cough	53	60	88.33
Ocimum basilicum	Cough	30	34	88.24
Litsea glutinosa	Dysentery	64	79	81.01
Andrographis paniculata	Malaria	34	50	68
Clerodendrum viscosum	Stomach ache	30	48	62.5
Terminalia arjuna	Heart pain	44	81	54.32
Centella asiatica	Dysentery	40	76	52.63
Azadirahta indica	Dermatitis	50	115	43.48
Sterculia villosa	impotence,	27	68	39.71
Terminalia arjuna	Dysentery	20	81	24.69
Azadirahta indica	Anthlemintic	20	115	17.39

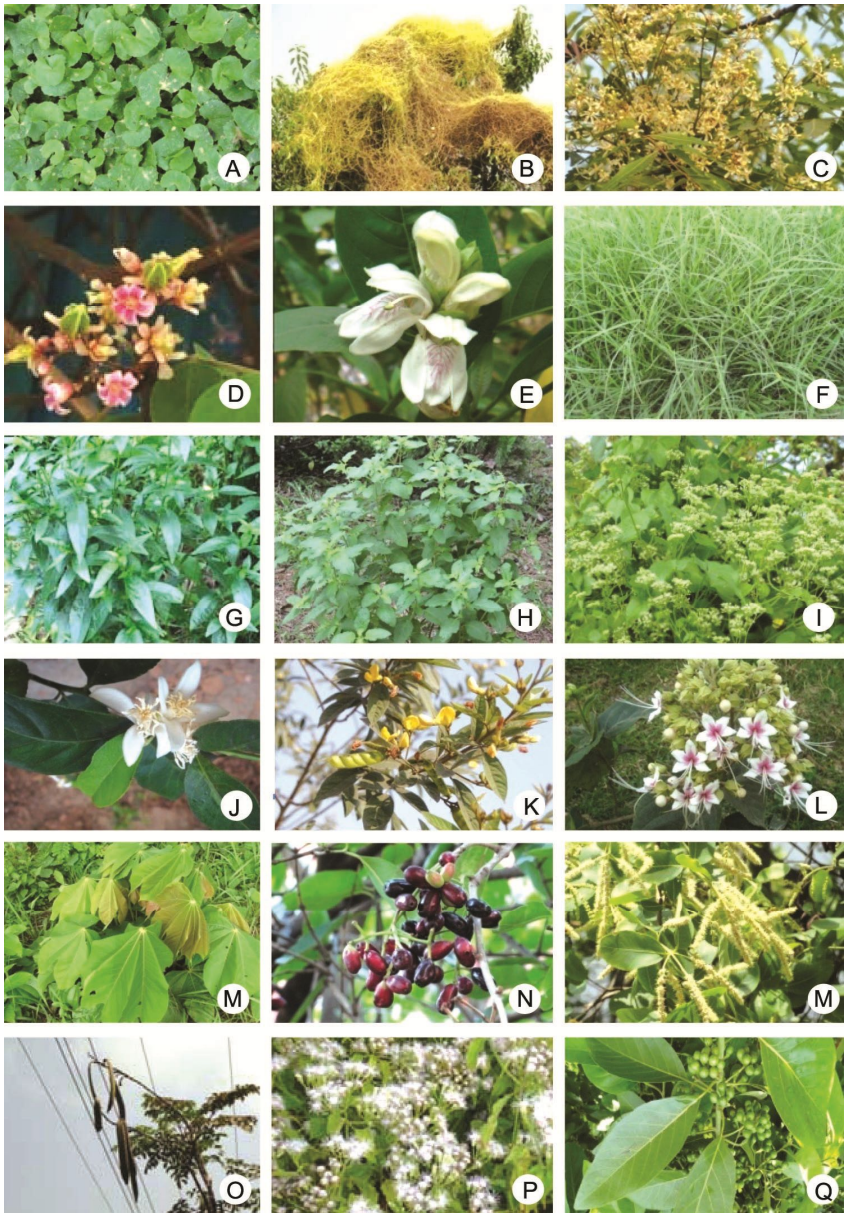


Plate 1. Images of most cited medicinal plants by the local people in the study area: *a. Centella asiatica*, b. *Cuscuta reflexa*, c. *Azadirachta indica*, d. *Averrhoa carambola*, e. *Justicia adhatoda* , f. *Cynodon dactylon*, g. *Andrographis paniculata*, h. *Ocimum sanctum*, i. *Mikania cordata*, j. *Citrus aurantifolia*, k. *Cajanus cajan*, l. *Clerodendrum viscosum*, m. *Sterculia villosa*, n. *Syzygium cumini*, o. *Terminalia arjuna*, p. *Oroxylum indicum*, q. *Chromolaena odoratum*, r. *Litsea glutinosa.*

PRK values varied from species to species as indicated in the Table 4. *Litsea glutinosa* scored top PRK value indicating very popular plant species in the study area and used for diarrhoea, dysentery and impotence. *Mikania cordata, Ocimum sanctum, Azadirachta indica, Terminalia arjuna, Centella asiatica, Andrographis paniculata* and *Cynodon dactylon* were also most cited species in the study area. These species are also well known medicinal plants in our country. The present analysis also confirmed their popularity among the local people of Lawachara national park. High PRK data of medicinal plants is the indication for further ethno-lead drug research to find active new drugs.

Table 4. Citation frequency of most cited medicinal plants.

Scientific name	Family	Ailments	Citation no.	Percentage of Respondents Knowledge (PRK%)
Litsea glutinosa	Lauraceae	Dysentery	64	39.26
Mikania cordata	Asteraceae	Cut and wound	58	35.58
Ocimum sanctum	Lamiaceae	Cough	53	32.52
Azadirachta indica	Meliacae	Dermatities	50	30.67
Terminalia arjuna	Caesalpiniacae	Heart pain	44	26.99
Centella asiatica	Clusiaceae	Dysentery	40	24.54
Andrographis paniculata	Acanthaceae	Malaria	34	20.86
Cynodon dactylon	Poaceae	Cut and wound	34	20.86
Chromolaena odoratum	Asteraceae	Cut wound	32	19.63
Clerodendrum viscosum	Verbenaceae	Stomach ache	30	18.40
Ocimum basilicum	Lamiaceae	Cough	30	18.40
Ceriscoides campanulata	Rubiaceae	Jaundice	28	17.18
Oroxylum indicum	Bignoniaceae	Jaundice	27	16.56
Sterculia villosa	Sterculiaceae	impotence,	27	16.56
Cuscuta reflexa	Cuscutaceae	Jaundice	25	15.34
Syzygium cumini	Myrtaceae	Diabetes	25	15.34
Averrhoa carambola	Averrhoaceae	Jaundice	22	13.50
Cajanus cajan	Fabaceae	Jaundice	21	12.88
Justicia adhatoda	Acanthaceae	cough	20	12.27
Azadirachta indica	Meliaceae	Anthlemintic	20	12.27
Citrus aurantifolia	Rutaceae	Jaundice	20	12.27
Terminalia arjuna	Combretaceae	Dysentery	20	12.27

Observations in the field, interviews and discussions with local people, a good number of threats to medicinal plants have been identified. The most serious threats are exotic timber species plantation in and around national park, fallow lands, homesteads, roadsides and even in the edges cultivated lands. *Acacia auriculiformis, Acacia mangium, Eucalyptus camadulensis, Dalbergia sissoo, Laeucaena leucocephala, Swietenia mahagoni and Cassia siamea* are most preferred plant species for plantation. According

to local people perception such species are very selfish plants and they do not support native species under their canopy. Illegal logging and over exploitation from the national park vegetation are other threats to medicinal plants in the study area. Lack of awareness among the local people about impact of exotics on native plants and environment is another threat to medicinal plants. Availability of the modern medicines which promotes the negligence of use of herbal medicines among the local people in the study area is also threats to medicinal plants. Senior people with herbal knowledge do not like to share their knowledge with juniors. Due to sudden death of such people, herbal knowledge of the area lost forever.

A list of conservation measures is made based on present survey results and observations. Distribution map of all culturally important medicinal plant species in the study area could be made. Population status of such species across the habitats could be determined. Current rate of exploitation by local people could be calculated. If it seems that medicinal plants are vulnerable in the natural habitats, necessary measures could be taken for *ex situ* conservation. Awareness programs among the local influential persons who can make change could be created.

The present endeavor in and around Lawachara national park is very preliminary. It would be better if we could take more interviews with local people in the study area. Due to budget and time constrains, the present study is not well enough to draw sound conclusion. It needs further long term survey to further validate all ethnomedically important data for ethno-lead drug exploration research. In future, joint research in ethnobotany with pharmacognosy and phytochemistry are essential to confirm traditional knowledge of medicinal plants. The present quantitative evaluation of ethnobotanical data from in and around Lawachara national park probably is the first effort. The data indicated that the study area has plenty of medicinal plants (124 species) and diversity health care uses (53 ailments with 244 formularies) of such plants. Using modern mathematical tools Fic values on the uses of medicinal plants have been determined. The average Fic value for all ailment categories obtained 0.78. Such value indicated that maximum people in the study area were well informed about the medicinal knowledge of plants and also showed agreement on the use of medicinal plants for such categories of ailments. Maximum Fic values (more than 0.80) obtained in case of Cut and wound, Jaundice, Respiratory related ailments, Diabetes, Diarrhoea and dysentery, Anthelmintic, Gastrointestinal complain, Impotence, Dermatitis and High blood pressure. *Chromolaena odoratum, Ceriscoides campanulata, Oroxylum indicum, Cuscuta reflexa, Averrhoa carambola, Cajanus cajan, Justicia adhatoda* and *Citrus aurantifolia* showed 100% Fl values. *Litsea glutinosa, Mikania cordata, Ocimum sanctum* and *Azadirahta indica* were scored maximum PRK value. It is recommended that species showed high Fic, Fl and PRK values could be used for further ethno-lead phytochemical analysis to find active compounds to discover drugs from plants. As the medicinal plants and traditional knowledge in and around the park are in threatened condition, appropriate measures should be taken for sustainable conservation. For the community development, resource conservation, primary health care and economic growth such plants and knowledge can play an important role.

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Short Communication

DEER POPULATION GROWTH IN THE BANGABANDHU SAFARI PARK IN BANGLADESH

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Deer are the most ancient of all typical ruminants (Prater 1980) and widely distributed across the Indian Peninsula, Burma, Sri Lanka, and Indo-Malayan countries (Blandford 1888-91). The Family Cervidae belongs to 17 genera where the entire Asia has 9 (2 extended to Europe) (Grubb and Gardner 1998). Four deer species, Spotted Deer (*Axis axis)*, Hog Deer (*Axis porcinus)*, Sambar (*Cervus unicolor)* and Barking Deer (*Muntiacus muntjak*) are kept in the Bangabandhu Safari Park (BSP). Presently, Spotted Deer has a restricted distribution in Bangladesh; Hog Deer is remarkably a rare species both in wild and captive conditions. The Barking Deer is comparatively widely distributed but Sambar is the rarest species in the wild.

A limited number of individuals of Sambar is surviving in captivity in our country, but a large number of them live in the semi-captivity like the BSP with a good breeding and growth rates. The BSP authority has been trying to provide a natural environment for establishing Sambar population since 1982. At present Sambar population in the DSP is the most important stock in Bangladesh. The breeding rate of the Spotted Deer is remarkably high in this semi-captive environment than any other species whilst reintroduction of Spotted Deer in the Chittagong Hill Tracts had failed (IUCN-Bangladesh 200 and Khan 1985). On the contrary, Spotted Deer and Barking Deer have been breeding successfully in all zoos of the country. In the BSP, habitats include a mixture of hardwoods, bush lands and pasturelands which provide the most suitable environment.

The Bangabandhu Safari Park is the first of its kind in Bangladesh. It is situated 107 km south of Chittagong City in 1982 under Chakoria upazila of Cox's Bazar District surrounded by the Fasiakhali Range of the Cox's Bazar North Forest Division. The geographic location is 21°40'6.7" to 21°40'6.9" N latitude and 92°4'68.44" to 92°4'68.48" E longitude. The altitude is about 9 meter from the mean sea level.

The current study was conducted between June 2007 and May 2008. A 3-day field visit in each fortnight interval was conducted to collect data. In addition, Safari Park related

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information and data were gathered from secondary sources including office and staffs of the park, Forest Department office records, published papers and unpublished reports.

The Spotted Deer. Twenty Spotted Deer caught from the Sundarbans of Bangladesh were later released in this park in 1982. The line transect surveys shown that the wild population of Spotted Deer in the park was 266 and later 34 were kept in an enclosure including some of confiscated as illegal collections. The population growth of Spotted Deer in this park was noted remarkably high. Of the deer, 75 (25%) adult males, 180 (60%) adult females, 30 (10 %) juveniles and 15 (5%) infants observed during the study. The adult male-female ratio was 1:2.4 and adult - young ratio was 1:0.17.

The adult male of Spotted Deer (wild population) is longer and heavier than the adult female. Their pinna and tail lengths are more or less same. Measures in wild population (n=8), showd that adult body length was 95-110 cm, weight 43-52 kg, shoulder height 47-55 cm, pinna 9-11 cm and tail 10-14 cm. Similarly, the body length of a captive adult is 90-105 cm, weight 38-50 kg, shoulder height 43-55 cm, pinna 9-11 cm and tail 10-13 cm (n=8). The observations indicated that the wild Spotted Deer in the park is slightly longer and heavier than that of captive ones. The weight of adult was measured by several workers, viz., adult male 59.8 kg and female 54.4 kg (Anonymous 1989); male 65.77 kg at Kanha National Park in India (Schaller 1967); and the large male may reach up to 86.2 kg (Brander 1923).

The Hog Deer. Wildlife biologists believed that Hog Deer had gone extinction from wild of Bangladesh (Khan 2004, Biswas and Mathur 2000 and Khan 1985) but an individual was caught from Rangamati in 2003 and kept it in the BSP. Later, 4 individuals brought to the park from a private collection of a person in Khagrachari. The Hog Deer also gave birth at the park and now there are 8 individuals of which 2 adult males, 3 adult females, 1 juvenile and 2 infants.

The average body length of an adult was 104 cm, weight 39.75 kg, and shoulder height 27.75 cm, pinna 9.75 cm, and tail 8.37 cm. The male is heavier than the female.

The Sambar Deer. Two adults (male and female) were released during the establishment of Bangabundhu Deer Breeding Centre in 1982. Later 5-6 adults were released in the enclosures. Currently, 30 deer (7 adult males, 15 adult females, 3 juveniles and 5 infants) are living in the enclosures with 1:2.1 adult male-female and 1:0.36 adult juvenile-infant ratios. On the basis of annual count (2006-2008), the growth rate of Sambar population is 7.14%.

The adult male is longer and heavier than an adult female. The pinna and tail lengths are more or less same where antler of the male is 30-50 cm long. The average body length of an adult is 393.25 cm, body weight 144 kg, pinna 17.1 cm and tail 20.6 cm (n=10). According to Slee (1984), considering the ambient environment and food availability body length and height even body weights have a relation.

The Barking Deer. In 1982, during the establishment of the Bangabandhu Deer Breeding Centre, 8 Barking Deer were found in the area. Later, 15 were brought from private collections of few persons in the Chittagong Hill Tracts in 2003 and released them in this park. Time to time confiscated individuals were released, but the number could not be ascertained from the park office. The line transects surveys have shown that the wild population size is 186 and 14 individuals were seen in an enclosure of this captivity. Of them, 45 (22.5 %) were adult males, 100 (50%) adult females, 30 (15%) juveniles, and 25 (7.5%) infants. The adult male-female ratio was 1:2.2 and adult-young ratio was 1:0.04.

Generally, the adult male of Barking Deer is longer and heavier than the adult female. Their pinna and tail lengths are more or less same. The wild adult body length is 92-115 cm, weight 35-47 kg, shoulder height 50-62 cm, pinna 8-10 cm and tail 10-12 cm (n=6). Similarly, captive adult body length is 96-115 cm, weight 53-61 kg, shoulder height 32-41 cm, pinna 8-10 cm and tail 10-12 cm (n=6). Therefore, the measurements of wild and captive individuals are same.

Beginning of the year 1982, the BSP authority had collected (able to collect) two individuals of Sambar male and female. After 26 years the population growth had reached with 30 individuals including 7 males, 15 females, 3 juveniles and 5 infants. The Hog Deer population was found to be with 8 individuals including 2 males, 3 females, 1 juvenile and 2 infants in the captivity with good environment. The Hog Deer and Sambar are critically endangered (CR) in the wild (IUCN-Bangladesh 2015) and they almost brink of extinction in Bangladesh.

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-Short Communication

EFFECTS OF SOIL AND WATER SALINITY ON pH, EC AND THE SELECTED ION CONTENTS IN DIFFERENT CROPS GROWN IN NON-SALINE AND SALINITY AFFECTED AREAS OF BANGLADESH

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Salinity intrusion is a growing problem in the coastal areas around the globe. Climate change associated hazards like sea level rise, cyclone and storm surge have been contributing to this problem. Currently, cyclones accompanied by storm surge and increased salinity intrusion into fresh water and soils are the most catastrophic phenomena for coastal communities of Bangladesh, especially in Satkhira, a vulnerable coastal district. In last decade, number of cyclonic events from Bay of Bengal increased. Cyclone Sidr in 2007, cyclones Nargis and Reshmi in 2008 and cyclone Aila in 2009 caused huge damage. The government of People's Republic of Bangladesh estimated that it directly affected about five million families and crops of about 0.7 million hectares (DMB 2010). According to Bangladesh Bureau of Statistics, Sidr caused damage to more than 0.1 million tons of rice crop in Khulna (BBS 2010). Cyclone and storm surges force saline water into agricultural lands along coast, which damages crops in year of cyclone hits and for several years afterwards. This happened in area Shyamnagar *Upazila* under Satkhira district, studied in this present research. Saline water intrusion caused by cyclone Aila in 2009 led to loss and damage to rice crops in this area. Some recent studies indicated that salinity intrusion in both soil and water might increase further because of escalating intensity of cyclone and storm surge (SRDI 2010 and Rabbani *et al.* 2010). Coastal rice crops in Asia, for instance, are frequently affected by exposure to sea water brought in by cyclones around the Indian Ocean (Sultana *et al.* 2001). Salinity in Bangladesh river networks is reasonably well understood, with empirical and physically based study. Sea-level rise is likely to play a significant role in increasing salinity in natural drinking water sources in the future (Aerts *et al*. 2000 and Khan *et al*. 2011). The present study was undertaken to calculate soil, water, crops and salinity balances for different regions of Bangladesh that differ widely in water availability and the salt contents of the irrigation water. The prime aim was to study the impact of salinity on some selected ion contents such as Na, K, Ca, Mg, and Cu present in soil, water and crop samples.

Selection of the sampling area and sample collection: The study was conducted in four villages under Unions at Shyamnagar, Tala and chitalmari *Upazila* in Satkhira and Bagerhat District (salinity affected); Tongbari under Munshiganj District (non-saline).

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Satkhira and Tala are exposed coastal area in South West region under Khulna division of Bangladesh. Samples were collected from the field and were carried to laboratory in plastic bag. In laboratory, they were washed with deionized water to remove dirt. Separated parts were dried in air and at lower oven drying temperature 80˚C (Jarrett 1983) for few hours until constant weight was attained. After cooling samples were grinded and sieved. They were then preserved for analyses in air tight plastic bag.

Sample preparation for laboratory analysis: Reagent grade $HClO_4$ and HNO_3 were procured from E. Merck, Germany. Certified standard stock solutions of nutrients were obtained from Varian, Australia for calibration purpose. All working solutions were also prepared in distilled water. Prior to analysis samples were digested with conc. HNO_3 and $HClO_4$ (2:1) mixture in acid digestion bomb (Model 4749A). Na and K were analyzed by flame photometer and other inorganic nutrients were measured by AAS. Salinity, pH and conductivity were measured by HACH Sension 156 portable multi meter.

The chemical parameters like pH, Electrical conductivity and salinity of water and soil samples are presented in Table 1. Concentrations of Na, K, Ca, Mg, Cu and Zn in different samples collected from various salinity and non-saline areas are presented in Tables 2–3. Concentrations in 12 crop samples varied within the range of Na (1796 - 8362 mg/kg), K (9710 - 48921 mg/kg), Mg (433 - 4660 mg/kg), Ca (415 - 7477 mg/kg), Cu (16 - 46 mg/kg) and Zn (5 - 11 mg/kg).

The pH of the collected water samples ranged from 6.75 to 7.35 with an average value of 7.05. The highest and the lowest pH values were detected respectively at Tala area (pH = 7.35) and Shymnagar (pH = 6.75). The pH values of the soil samples ranged from 6.50 to 7.50 with an average value of 7. The highest pH value was recorded at Tala (pH = 7.50) and the lowest pH value was found at Tangbari (pH = 6.50). According to the standard value set by the Guide to the Environmental Conservation Act 1995 and Rules 1997, for irrigation and drinking water purposes the value of pH is 6.5 - 8.4. At the studied area, pH values of water and soil samples were found within the permissible limit. The EC values of the collected water samples ranged from 965 to 4570 µS/cm with an average value of 2527 µS/cm. The highest and lowest EC values of water samples were recorded at Shymnagar and Tala, respectively. The EC value of the collected soil samples ranged from 150 to 3590 µS/cm with an average value of 1870 µS/cm. The highest EC value was found at Shymnagar (EC = 3590 µS/cm) and the lowest value was recorded at Tala *Upzila* (EC = 150 µS/cm). The highest EC value of the region indicated that the area is salinity affected. So there can be harmful effect by salt concentration or salinity hazard in respect to EC. The salinity of the water and soil samples ranged from 0.2 ppt to 20 ppt and from 0.2 ppt to 0.6 ppt, respectively. The highest salinity value was found at Shyamnagar in both water and soil samples. Result clearly showed Shyamnagar as more salinity affected area. The lowest values were found at Munshiganj (0.2) and Tala (0.4 ppt) for water and soil samples respectively evinced that soil contained less amount of salt than that of water sample.

Table 1. Chemical parameters of soil and water samples collected from sampling areas.

Sampling Locations	Sample types	pH	Conductivity(µS/cm)	Salinity(ppt)
Tala	Water	7.35	646	0.3
Shyamnagar		6.75	4575	20.0
Chitalmari		7.15	1250	0.6
Tangibari		7.04	484	0.2
Tala	Soil	7.50	150	0.2
Shyamnagar		6.90	3590	0.6
Chitalmari		7.12	318	0.3
Tangibari		6.50	780	0.4

Sodium concentration of the collected water and soil samples ranged from 2 to 480 mg/kg and 1389 to 11160 mg/kg respectively. The highest value of Na in water sample was found in Shyamnagar and the lowest value was found in Tangibari. The highest value of Na in soil was recorded at Shymnagar and the lowest value was recorded at Tala. Potassium and magnesium contents were highest (9438 and 13821 mg/kg) in Chitalmari soil samples and the lowest values were recorded (0.19 and 0.65 mg/kg) in Tala water samples. Calcium content was found highest (12578 mg/kg) in Tala soil samples and the lowest (1.25 mg/kg) in Tala water samples. Cu and Zn content (48 and 88 mg/kg) were highest in Chitalmari soil and the least (0.003 & 0.0013 mg/kg) in Shymnagar and Tala water samples respectively (Table 2).

Table 2. Concentrations of selected ion contents of soil and water samples collected from sampling areas.

Location	Sample Type	Concentration of ion contents(mg/kg)					
		Na	K	Mg	Ca	Cu	Zn
Tala	Soil (S-1)	1389	6932	9822	12578	26	66
	Water (W-1)	8	0.2	0.7	1	0.003	0.001
Shyamnagar	Soil(S-2)	11160	6776	10270	5578	12	72
	Water(W-2)	480	14	50	17	BQL	0.002
Chitalmari	Soil(S-3)	2352	9438	13821	11844	48	88
	Water(W-3)	10	0.8	0.9	4	BQL	0.004
Tangibari	Soil(S-4)	1873	6856	12159	7364	44	14
	Water(W-4)	2	1.5	0.7	2	BQL	0.04

BQL= below quantification limit

Among twelve different crop samples, bitter lemon of Tala (8363 mg/kg) followed by cucumber of Shyamnagar (8335 mg/kg) recorded for the maximum Na content and Tangibari potatoes were recorded for the minimum Na concentration (1796 mg/kg). Bitter lemon (Shymnagar) and cucumber (Tangibari) were found to contain the highest and least amounts (48921 and 9711 mg/kg) of K content (Table 3). Both the Mg and Ca

contents were recorded to be highest in cucumber of Shymnagar area (4660 and 7478 mg/kg). In Cucumber and Potato of Tala area minimum concentrations (434 and 416 mg/kg) of Mg and Ca respectively. On the other hand in Bitter lemon of Tala area maximum Zn and Cu contents (46.95 and 11.27 mg/kg) were recorded. Potatoes contained minimum concentrations of Zn and Cu (16.74 and 5.54 mg/kg). Sampling locations with the higher electrical conductivity leads to high salinity. Shyamnagar was found to be most salinity affected area. Studied ion concentrations were found higher in soil than those of water samples and the higher concentrations were recorded in salinity affected areas than those of non-salinity affected areas. When the salt concentration is higher in the soil, water moves from plant into soil. When salts accumulate in soils, problems arise for two main reasons: the soil becomes less permeable and salt damages. Especially high Na concentration affects soil and can lead to sodic soil condition. High Ca, Mg and Zn contents were recorded in samples. If Ca and Mg present in large quantities encounter the effect of Na and help to maintain good soil properties.

Table 3. Concentrations of ions in crop samples collected from sampling areas.

Samples	Concentration of ionic contents (mg/kg)					
	Na	K	Mg	Ca	Zn	Cu
S-1: Potato(Tala)	2925	14535	960	416	19.86	5.64
S-2: Potato(Shyamnagar)	3774	23799	1401	721	25.15	7.47
S-3: Potato(Chitalmari)	3549	25822	1460	425	16.02	5.54
S-4: Potato(Tangibari)	1796	11159	1095	463	16.74	5.73
S-5: Cucumber(Tala)	6026	38605	434	684	45.39	7.66
S-6: Cucumber(Shyamnagar)	8335	40693	4660	7478	43.68	8.01
S-7: Cucumber(Chitalmari)	8065	29385	3200	6055	37.23	6.79
S-8: Cucumber(Tangibari)	3512	9711	759	1186	22.59	6.32
S-9: Bitter Lemon (Tala)	8363	43434	3115	1840	46.95	11.27
S-10: Bitter Lemon (Shyamnagar)	4847	48921	2921	2838	34.76	7.67
S-11: Bitter Lemon (Chitalmari)	7347	34841	2283	1704	29.79	6.99
S-12: Bitter Lemon (Tangibari)	8024	34047	2644	3104	31.26	7.78

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Short Communication

FLIES FOR THE POLLINATION OF GREENHOUSE MANGO (MANGIFERA INDICA L., ANACARDIACEAE) IN THE SUBTROPICAL IRIOMOTE ISLAND, JAPAN

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Mango (*Mangifera indica* : Anacardiaceae) is a tropical fruit. De Candolle (1884) described its original home is in South Asia and Malay Archipelago. According to Mukherjee (1951b) mango originated in the Indo-Burma region and later it was spread to the countries of South Asia and also to other parts of the world. Mango is mainly grown throughout the tropical and subtropical areas of the world from nearly sea level to at about 4000 feet height. It is cultivated in open orchard, as well as in greenhouses. In the South East Asian countries some farmers use greenhouses for its cultivation, where they use pollinators. The necessity of pollination of mango depends on the situation, where it is cultivated and its cultivars. According to Mukherjee (1953) and Allard (1960) mango is a cross pollinated plant. When cultivated in greenhouses for commercial production, pollinators are obviously needed (Naik and Rao 1943). Ruehle and Ledin (1955) and Wolfe (1962) revealed that lack of pollination might attribute low yields of mangoes.

The inflorescence of mango is long and branched. It could be over 50 cm long, where several hundred to several thousand individual could have flowers and requires about a month to bloom completely. Naik and Rao (1943), Mukherjee (1951b) and Singh (1961) reported that 65-97% flowers are un-pollinated in nature. There are perfect and staminate flowers in the same panicle. Numbers of male flowers are 70-90%, and 65-85% bisexual flowers are un-pollinated in natural condition. This suggests that by using pollinators production can be increased (0.25-2.4%). Fraser (1927) stated that fruit bud formation and pollination were two big problems in growing mangoes. He pointed out that in some cases only 2 to 3 percent of the flowers on a panicle are perfect, in others 60 to 70 percent. The shape of perfect flower may be 5-8mm long that has globular ovary (rarely two or three) and a lateral style, which is absent in staminate flower. Both generally have one, but sometimes two even three functional stamens and several sterile staminodes. There are usually five greenish-yellow sepals, and three to nine but usually five cream-colored petals that take on a pinkish tinge before falling (Naik and Rao 1943). In the perfect or hermaphrodite flower, a nectar-secreting fleshy disk surrounds the ovary. The stamen is on the outer margin of this disk. The pistil and stamen are of the same length; therefore,

pollinating insects that feed on either nectar or pollen are likely to transfer pollen from the anther to the stigma (Juliano and Cuevas 1932 and Sturrock 1966). Maximum pollen shedding is from 8 am to noon. When the flowers open, considerable quantity of nectar is secreted which attract a large number of insects (Mukherjee 1953). However, relatively little pollen is produced on the anther (Popenoe 1917).

Popenoe (1920), Galang and Lazo (1937) and Sing and Sturrock (1969) reported that mango has entomophiber pollens. Singh (1960) described that honey bees do not visit mango flowers, but Singh (1954) listed this plant as a source of pollen and nectar for bees. Recently Fajardo *et al.* (2008) reported that 21 species of insects under 5 Orders, visit mango flowers in the Phillipines. They mentioned *Trigona biroi*, *Chrysomya* spp. *Eristalis* spp. and also honey bees (*A. cerana* and *A. mellifera*) as primary pollinators of mango. Fajardo *et al.* (2008) also reported that due to introduction of bee colonies, fruit set increased significantly (41%), whereas without bee pollination it was only 0.7%. However, in the present study necessity and rearing methods of flies for mango pollination were investigated.

The study was conducted on Iriomote Island (lat .24º20´N), southwestern archipelago of Japan in the greenhouse mango orchards during 2003 to 2004. Flies specially the carrion breeders were attracted with rotten fish meat placed on an open tray *ca.* 15 days before flowering initiated (Plate-1). To attract the outside syrphid flies and other insect pollinators to the mango flowers, doors and sidewalls of the greenhouse were kept open. The emergence of carrion breeding flies *e.g.* calliphorids were synchronized by calculating the flowering time and the developmental period of the flies, so that when they emerged as adults, flowers were also ready to be pollinated. The flies were collected on the flowers by using an insect nets and tube respirator. Special attention was taken so that the inflorescences were not harmed. The methodology of Mizuno *et al.* (2006) was followed for rearing desired flies.

Table 1. Fly species observed visiting flowers of mango (*Mangifera indica* L., Anacardiaceae) on Iriomote Island, Japan.

Family	Species
Syrphidae*	
1	*Eristalinus arvorum* (Fabricius)
Caliphoridae	
1	*Chrysomya megacephala* (Fabricius)
2	*Chrysomya rufifacies* (Macquart)

*Four more species of Syrphidae are yet to confirm their identification.

Seven promising pollinating species were recorded from Syrphidae and Calliphoridae on the greenhouse mango flowers (Table 1). Among them 1 syrphid was *Eristalinus arvorum* (Fabricius) (Plate-2), and 2 calliphorids were *Chrysomya rufifacies* (Macquart) (Plate-1) and *Chrysomya megacephala* (Fabricius) (Plate-3). All of them were also seen in the open atmosphere.

The main food from the mango flowers for the adult flies was nectar. During the licking of nectar they adhered some pollen grains in the bristle hairs of their mouth parts, especially on their glossae. When they visited other flowers afterward they transferred that to those flowers. In some subtropical islands of Japan *e.g.* Iriomote, Ishigaki, Miyako and Okinawa, commercial mango is cultivated in greenhouses (Plate-4). The orchard owners used pollinators for the pollination of the flowers in order to have good fruit sets, as well as good harvests.

In some South Asian countries like India, Bangladesh, Thailand growers keep honeybee colonies in the mango orchard for pollination, whereas in Japan, Taiwan, and the Philippines they use flies for this purpose. Honeybee is polylectic that goes to any flower attractive to them. Therefore, they are reluctant to stick to a single flower. In case of mango flower, it is open, small and sticky with its concentrated nectar. It is possible that honeybees are not attractive to this flower, but when there is no flower around them they are bound to go to mango flowers. In case of flies, they are regular visitors to the mango flowers. Their presence is much higher than the bees normally, and they also can transfer pollen from anther to stigma that helps mango flowers to be cross pollinated.

Plate 1. Fish meat to attract carrion breeders to lay eggs on it.

Plate 2. A syrphid fly foraging on mango flowers in the greenhouse.

Plate 3. A calliphorid fly foraging on mango flowers in the greenhouse of Iriomote Island Japan.

Plate 4. Mature ‘apple mango’ in the greenhouse Iriomote Island, Japan.

In Iriomote Island the flowering started from the month of February/March and fruit started from the March/April. Farmers kept only 1-3 mangoes per inflorescence. They

removed all the rest at different time intervals. They did not have any use of green mangoes, as a consequence they selected the best ones from one inflorescence, rest all were cut off and left on the floor of the orchard. Selling of ripe mango started from the months of July. Growers kept per tree about 50 (40-60) mangoes. On an average 3 mangoes made 1 kg. When the mangoes matured those were enclosed inside a white paper bag and tied with a hanging rope, so they did not fall down and got damaged. They collected mangoes before two to three days of ripening then boxed and sent to the market. The variety of mango produced on Iriomote is called “apple mango”.

Young (1942) studied pollination of 'Haden' mango in Florida and reported no significant difference between percentages of set in selfed and cross-pollinated flowers. Sturrock (1944) considered the flowers self- fertile. This self-fertility was supported by Popenoe (1917) who stated that the mango is self-fertile but cross-pollination is beneficial to increase fruit set. However, Singh *et al.* (1962) reported that crossed flowers set fruit whereas selfed ones did not, indicating a degree of self-sterility. Ruehle and Ledin (1955) considered that the lack of efficient pollination might be responsible in part for the low yields of some Florida cultivars. The studies indicated that the need for cross-pollination between mango cultivars was not critical, at least for most cultivars, but there was need for pollinating insects to transfer the pollen from anther to stigma within some cultivars to obtain satisfactory crop yields. The present study shows that within the cultivars there was a need for transferring pollen from anther to stigma by an outside agent, without what fruit set was not up to the level of commercially viable in maintaining the orchard. Mangoes outside greenhouse were not comparable with the within greenhouse mangoes on Iriomote Island in respect of quality and quantity, although the quantitative data was not collected from the present experiment. Because of this reason the orchard owner used flies in their greenhouses.

Burns and Prayag (1921) mentioned in Poona, India that flies of the genera *Psychonosoma* and *Pyrellia* are the chief pollinators of mango, but in Florida it was represented by insects of four Orders (Diptera, Hymenoptera, Lepidoptera, and Coleoptera). Singh (1954) also mentioned that flies were the pollinators of mango. In the present study 5 promising species of Syrphidae and 2 species of Calliphoridae were observed on the flowers of the greenhouse mango. Syrphid flies are promising pollinators but calliphorids are also good for mango pollination. Calliphorids were the major pollinators of mango in subtropical Asia (Bhatia *et. al.*, 1965 and Fajardo Jr. *et. al.*2008). Calliphorids also effectively pollinated many other flowering plants (Eardley and Mansell, 1996 and Stone *et al.* 1998). Number of flies in the orchard had very important role. It was necessary to have as many flies as possible per tree, reason being that fly did not move from flower to flower like bees. Flies started visiting flowers from early morning to noon till dehiscence of anthers continued. The flowers opened early in the morning, and the stigma was immediately receptive.

Bangladesh is a big mango producing country and almost all commercial orchards are in the districts of North Bengal (Rajshahi, Shibganj, Volahat, Chapai Nawabganj, Rangpur, Dinajpur). All those orchards are open. So all the natural pollination are held in this case. Only sporadically some commercial orchard owners use *A. cerana* or *A. mellifera* for pollination purposes along with goal to collect honey. In Rajshahi hundreds of *Apis* mostly *A. cerana* were observed on the mango flowers. Most interesting thing was visiting mango flowers by *Xylocopa* sp. The mango growers understood the benefits of using pollinators, but as they did not have easy access to them, consequently they did not use them. If available they are willing to use the pollinators in their orchard.

In Jezan, Saudi Arabia there are some mango orchards where several cultivars are used for the production of mango. No pollinator is used up until now but natural pollination is done by flies that are known from the officials of Agricultural Research Institute in Jezan. Mango growers do not keep *A. mellifera* colony even for honey production, as it is done in some other south Asian countries (Alqarni, unpubl.).

The main problems to use honey bees as pollinators of mango at open orchard are 1) flowers are not attractive to them, 2) they have choice of flowers, as many others bloom at the same time and 3) at over 35°C, which is normal during mango season, they cannot forage and mostly die in the greenhouse. There is no exact limit of using honey bee colonies for pollination of mangoes. Rather it is evident that concentration of colonies within the mango grove would result in increased floral visit and possibly more stabilized set of fruit. Mizuno *et.al*. (2006) tested 2 species of honeybees *A. cerana* and *A. mellifera* and 1 bumblebee (*Bombus ignitus*) for the pollination of "Irwin mango" in the greenhouse. They mentioned that pollination efficiency in superior order was *cerana*>*mellifera*>*ignitus*. Popenoe (1917) reported that honey bees were the most important hymenopteran insect visitors to the mango flowers, but the number present was variable.

It is apparent that there is need for pollinating insects within some cultivars to obtain satisfactory fruit harvest, although the need for cross-pollination between some mango cultivars is not critical, as they could be self fertile. From the present study on Iriomote Island it is understood that the fly visitors increased production both in quantity and quality for that cultivar of 'apple mango'. Therefore, the utilization of flies for mango pollination was an effective approach for economic profit. Efforts could be employed to use flies even for the open orchard. Calliphorid flies were regular visitor of the mango flowers and its method of propagation is mentioned, as well. Kobayaashi (1981) tried some baits to rear various syrphid flies in artificial condition. However, more organized research is needed to look for easier methods of rearing the syrphids in the greenhouse through future studies that could also be used for mango pollination.

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