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Editorial

Self-directed Learning in Medical Education

Mohammad Ali

Self-directed learning (SDL) is acquiring knowledge through one's own initiative. So that medical professionals can equip and adapt themselves to the rapidly changing, converging scientific informations. Self-directed learning (SDL) is a vital educational principle in higher education that has been promoted by various institutions due to its value in developing professionals to become lifelong learners. Medical education systems worldwide have embraced SDL so that medical students gain SDL skills to continuously equip themselves with relevant knowledge and skills in the ever evolving world of medicine. SDL is generally defined as learning on one's own initiative, with the learner having primary responsibility for planning, implementing, and evaluating the effort. In medical education, SDL is the process in which medical students take the initiative, with or without the help of others (e.g. instructors and colleagues), determine their learning needs, set learning goals, identify resources for learning, choose and implement learning strategies to acquire knowledge and finally evaluate learning outcomes. Hence, the medical student is responsible for his or her own learning. In a constantly changing environment, SDL is essential to enable medical students to develop independent learning skills, increased responsibility, assertiveness and accountability which are key attributes to a medical professional's career. Medical educators similarly seek to adopt SDL with the primary aim of producing learners who can manage their own learning in their careers and have a continuous quest for knowledge through critical thinking that will enhance retention and recall of information to promote better decision making.

Thus, health professionals need to be self-directed so as to increase independence, self-confidence in practice, motivation, self-discipline and goal orientation due to information explosion and the continuously evolving medical knowledge during their careers .SDL is consist of 7 key components as described by Malcolm Knowles.

1. The educator as a facilitator
2. Identification of learning needs
3. Development of learning objectives
4. Identification of appropriate resources
5. Implementation of process
6. Commitment to a learning contract
7. Evaluation of learning process

In recently updated MBBS curriculum Bangladesh Medical and Dental council has embraced and put emphasis on SDL It is a big step and a way forward creating impetus among medical professionals for lifelong learning.

Dr. Mohammad Ali
Associate Professor of Medicine
Comilla Medical College

Original Article

Effect of Ethanol Extract of *Musa sapientum* Flowers on Alloxan induced Type 2 Diabetes Mellitus in the Rat

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Abstract:

Background: Diabetes mellitus is a major risk factor for substantial morbidity, mortality and long- term complications occurring from untreated or uncontrolled hyperglycemia such as retinopathy, neuropathy and nephropathy in the human being. To maintain blood glucose concentrations at normal or near normal levels is the main objective of the treatment of the disease which may be obtained through administration of oral hypoglycemic agents or insulin. However, every medication possesses the risk of associated undesirable side effects. Plant sources were sought for as they are considered to have fewer side effects, cheaper possibly and of low cost in comparison to synthetic ones. *Musa sapientum*, commonly known as banana is well grown in South India, Bangladesh and Thailand and is well known for the food values of its flowers, fruits and stems. The juice of *Musa sapientum* flowers is claimed to possess anti-hyperglycemic potentials and so has been in use in Indian folk medicine as a remedy for diabetes mellitus from ancient times. Different parts of *Musa sapientum* have been used for various medicinal purposes including the treatment of diabetes mellitus in different parts of the world, although its exact status remains yet unexplored. **Objectives:** To evaluate anti-hyperglycemic and pancreatic islets protective effects of ethanol extract of flowers of *Musa sapientum* (MSFE) in comparison to metformin in alloxan-induced diabetic rats. **Methods:** This experiment was carried out with a total number of thirty rats which were divided into five groups of six rats each. Excepting the Control and MSFE group (Group III), other groups were induced type 2 DM by the dose of alloxan obtained from similar experiment of previous study (i.e.150 mg/kg body weight). The control group was (Group-I) (C) of normal untreated rats, diabetes mellitus was induced in rats treated with alloxan (Group-II), rats were treated with MSFE at a dose of 250 mg/kg body weight orally daily through an intra-gastric tube from 4th – 21st days (Group-III), another group of rats were induced type 2 DM with alloxan and treated with MSFE 250 mg/kg body weight orally daily through an intra-gastric tube from 4th – 21st days (Group-IV) and the last group of rats were induced type 2 DM with alloxan and treated with metformin (20 mg/kg body weight) orally daily through an intra-gastric tube from 4th – 21st days (Group-V). Fasting blood glucose (FBG), HbA1c and histology of the pancreas from all groups were performed. Statistical evaluation of the different groups were compared with those of the control group by student's't' test. Results were presented in tabulated forms. **Results:** There was significant ($p<0.001$) increase of FBG and HbA1c in the diabetic rat group (Group II). Histological parameters in Group II rats in comparison to those of Group I showed significant ($p<0.001$) reduction but no significant ($p>0.05$) difference was observed between control group (Group I) and (Group III) (MSFE) rats. Significant ($p<0.001$) reduction of FBG and HbA1c with significant ($p<0.001$) recovery of islets of

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Langerhans were observed in Group IV and Group V rats in comparison to diabetic rats (Group II). The statistical analyses could not establish any significant ($p>0.05$) difference in FBG and HbA1c between Groups IV and V rats but surprisingly there were significant ($p<0.001$) improvement in numbers and diameters of islets of Langerhans in Group IV in comparison to those of Group V. **Conclusion:** Results of the present study suggest that administration of MSFE and metformin in alloxan-induced diabetic rats had shown alleviation regarding hyperglycemia in Long-Evans male rats. However, histological observation has suggested better architectural conservation regarding number and diameters of islets of Langerhans in the MSFE-treated diabetic rats.

Key words: Diabetes mellitus, *Musa sapientum* flowers, alloxan.

(J Com Med Col Teachers Asso July 2021; 25(2): 51-57)

Introduction:

Diabetes mellitus (DM) is a group of metabolic diseases characterized by chronic hyperglycemia resulting from defects in insulin secretion, insulin action or both. Chronic hyperglycemia may be associated with impairment of growth and susceptibility to infection leading to life threatening consequences of ketoacidosis, nonketotic hyperosmolar syndromes, failure of different organs specially the eyes, kidneys, nerves, heart and blood vessels. The severity of the metabolic abnormalities in diabetes mellitus is reflected by the degree of hyperglycemia[1].

Globally, according to an estimate in the year 2017, about 425 million people were suffering from diabetes and by the year 2045, this figure is supposed to reach to about 629 million. Diabetes with its grievous consequences and complications remains a major burden upon health care systems of all countries. Research should be carried out for early interventions, improved screening and timely and more effective management to reduce the impact of diabetes mellitus on the individual and society.[2]

Diabetes mellitus, being a major endocrine disorder, is considered to be one of the growing health problems in Bangladesh. Population growth, ageing of population and urbanization with lifestyle changes play key roles for increased prevalence of the disease, specially type 2 in Bangladesh[3]. About 8.5 million people suffered from diabetes mellitus in the year 2014.[4] The total annual per capita expenditure on medical care was estimated to be 6.1 times higher for DMs than non-DMs which was 635 dollars and 104 dollars respectively.[5] It is projected that in the year 2045,Bangladesh will be the 9th among top ten countries for number of people with diabetes.[2]

Experimental animal models are one of the best strategies for the understanding of the pathophysiology of any disease in order to design and develop drugs for its treatment. One suitable method to study diabetes mellitus is to induce experimental diabetes mellitus in animals such as rats. Available reports suggest that alloxan induced diabetes model was the best known drug-induced diabetes model.[6,7]

The management of diabetes mellitus is a global problem until now and complete recovery from this grievous systemic metabolic disease is not yet discovered. The modern anti-diabetic therapeutic regimens can control the disease but they produce undesirable side effects. So, it is relevant that further scientific efforts are made to discover drugs or agents or plants with potential euglycemic or anti-hyperglycemic properties that would control progression of the disease and its complications even if cure is not possible. Therefore, the search for new, effective and safer drugs remains a top priority till today. Recently, some medicinal plants have been reported to possess anti-hyperglycemic potentials and have been used empirically as anti-diabetic remedies.[8]

One such plant is *Musa sapientum* of family *Musaceae*. The present study aims to obtain effects of ethanol extract of *Musa sapientum* (banana) flowers in alloxan induced diabetes mellitus (DM) in rats. A few recent reports have suggested ameliorating effects of *Musa sapientum* flower extract (MSFE) upon DM in rats.[9,10,11] The present study was such an attempt to investigate into the effects of *Musa sapientum* flowers on diabetes mellitus in the rat model. Diabetes mellitus was induced in rats by alloxan as it is well known to produce dose dependent depletion of the β-cells of the pancreas followed by repopulation by the β-cells.[6] The optimum dose of alloxan to produce partial obliteration of the β-cells was selected at 150mg/kg body weight of rats.[6,10,12,13] A diabetes mellitus type2 model was produced through such administration of alloxan.[6,7,13] Ethanolextract of *Musa sapientum* flowers was administered orally at the dose of 250mg/kg body weight[10] for 18 days to the alloxan-induced diabetic rats. Metformin (20mg/kg body weight) was used as the reference standard.[6]

Musa sapientum, also known as 'saba' is a triploid cultivated species of banana(genome is ABB or BBB) and so their flowers are rich in red anthocyanin pigment[14]. This anthocyanin provides high antioxidant capacity.[15,16] It may henceforth be assumed that ethanol extract of *Musa sapientum* flower has potent anti-hyperglycemic and antioxidant effects. Research may be conducted to utilize

these beneficial effects of Musa sapientum flower extract (MSFE) in alloxan-induced diabetic rats. The findings of this type of research might be helpful to explore new molecules for the treatment of type 2 DM in humans.

Methods

This experimental study was carried out at Pharmacology department of Bangabandhu Sheikh Mujib Medical University (BSMMU), Shahbagh, Dhaka from September 2016 to July 2018 after obtaining approval from Institutional Review Board (IRB). The flowers of *Musa sapientum* were collected from the banana gardens of Chaklapunji Tea Estate, Chunarughat, Hobiganj and were identified by the experts of Bangladesh National Herbarium, Mirpur, Dhaka. About 17.208 gram freeze-dried ethanol extract of *Musa sapientum* flowers was obtained from 7.23 kg of fresh flowers following standard operating procedures. This extract was reconstituted with 0.9% normal saline to obtain necessary concentration of *Musa sapientum* flower extract and was administered to rats by intra-gastric tubing.

The alloxan powder was collected from Loba Chemie Pvt. Ltd., Mumbai, India and the solution was freshly prepared by dissolving 1500 mg alloxan powder in 20 ml normal saline and mixed properly by vortex mixing for intraperitoneal (i.p.) injection.[12]

Metformin hydrochloride (500mg tablet) was collected from Beximco Pharmaceuticals Limited, Dhaka, Bangladesh. The tablets were finely powdered and suspended in normal saline at a concentration of 3.25 mg/ml to obtain concentration of metformin at a dose of 20mg/kg body weight for oral administration[6] by intra-gastric tubing.

Animals: A total of 43 adult male Long Evans rats weighing between 200g - 280g and aged 22-25 weeks were collected from the animal house, BSMMU, Shahbagh, Dhaka. Out of them 30 rats (12 normal and 18 alloxan-survived diabetic) could be included in this study. Diabetic animals that died during the post induction period or at follow-up stages were replaced to avoid compromising the final number of rats in the study.[17]

The rats were grouped into 5 groups and all groups of rats were housed in compartmentalized rectangular plastic cages (38×23×15 cubic centimeter) covered with stainless steel wire mesh cover with not more than four rats per cage. Each cage was properly labeled for identification of different groups. The animals were maintained at standard laboratory conditions in an air conditioned room with 12/12 hour natural dark/light cycle and 20 ± 2^{0}C temperature. They were given standard rat diet and water ad libitum. A layer of wood shavings was placed on the floor of the cages as bedding[18] and a water-filled glass bottle was kept in each cage at a height that the rats were able to reach easily to drink. Rat diet was also placed in the cages. The cages were cleaned regularly and proper hygienic and sanitary measures were maintained during the experimental period. Waste products were removed regularly. Rats were given treatments as described in the treatment schedule given in table I.

Table-I : Schedule of treatment

Serial No. of Groups	Groups	N	Drug/ Diet/ Musa extract with duration			Day of sacrifice
			Day 1	Day 1- 21	Day 4 – 21	
I (C)	Control (C)	6	Distilled water ad libitum and standard rat diet	Distilled water ad libitum and standard rat diet	Distilled water ad libitum and standard rat diet	22nd day
II (A)	Alloxan (A) (Diabetic control)	6	Alloxani.p.	Distilled water ad libitum and standard rat diet	Distilled water ad libitum and standard rat diet	22nd day
III (MSFE)	MSFE	6	Distilled water ad libitum and standard rat diet	Distilled water ad libitum and standard rat diet	MSFE (250 mg/kg b.wt/rat/day)	22nd day
IV (A+ MSFE)	A + MSFE	6	Alloxani.p.	Distilled water ad libitum and standard rat diet	MSFE (250 mg/kg b.wt/rat/day)	22nd day
V (A+ Metfor.)	A + Metfor.	6	Alloxani.p.	Distilled water ad libitum and standard rat diet	Metformin (20mg/kg b.wt/rat/day)	22nd day

n = number of rats in each group, MSFE = *Musa sapientum* flower extract, b.wt = body weight, kg = kilogram, A = Alloxan, Metfor. = Metformin, MSFE = Ethanol extract of *Musa sapientum* at a dose of 250 mg/kg b.wt.

Rats of all groups were allowed normal rat diet and water ad libitum from day 1 to day 22. Group I (control group, C) was allowed only normal rat diet and water ad libitum. Diabetes was induced in rats of Group II (A) by a single i.p. injection of alloxan (150 mg/kg, freshly dissolved in normal saline) on day 1. Rats of Group III (MSFE) were given *Musa sapientum* flower extract (250 mg/kg body weight/day) orally suspended in normal saline through intra-gastric tubing from day 4 - day 21. Rats of Group IV(A+MSFE) were administered *Musa sapientum* flower extract (250 mg/kg body weight/day) orally suspended in normal saline through intra-gastric tubing from day 4 - day 21 concomitant with alloxani.p. (150 mg/kg body wt)(single injection). Rats of Group V (A+Metfor.) were orally administered metformin at a dose of 20mg/kg b.wt/rat/day from day 4 - day 21 concomitant with alloxani.p. (150 mg/kg body wt) (single injection). Rats of all groups were sacrificed on day 22.

Biochemical procedure: Blood samples were collected from the tip of the tail vein of each rat as a drop after depriving food for 12 hours with free access of drinking water. The drop was immediately placed on the strip of the Accu-check active Glucometer, Roche, Germany to find the glucose level quickly. Rats with FBG 11-19 mmol/L were considered having moderate hyperglycemia.[7,18,32] HbA1c of rats were measured from the blood of tip of the tail vein by Bio Hermes Glycohemoglobin Analysis

Histological Procedure:On the 22nd day animals were sacrificed by decapitation under light chloroform anesthesia. The abdomen of each rat was opened by midline incision and extended up to the thorax. The pancreas was identified and separated from the surrounding viscera, cut into small pieces and were fixed in 10% formalin with proper labeling. They were embedded in paraffin and were cut into 5µm thickness in microtome. These sections were collected in slides and then stained with Hematoxylin and Eosin. The stained sections were examined under low and high power by using an Olympus microscope.

Statistical analysis: The data were expressed as mean ± SD. Students unpaired t-test was used to compare between the control and the treatment groups. The level of significance was considered at $p<0.05$, $p<0.01$ and $p<0.001$. Statistical analysis was performed using SPSS (Statistical Package for the Social Sciences) for Windows (version 14).

Result:

Results are expressed in tabulated form. In all tables data are expressed as Mean ± SD, Number of rats in each group, n = 6, C = Control, A = Alloxan, MSFE = *Musa sapientum* flower extract, Metfor. = Metformin, FBG = Fasting blood glucose, * = $p>0.05$ (not significant),** = $p<0.05$ (significant),*** = $p<0.001$(highly significant), a = comparison between I and II, b= comparison between I and III, c=comparison between II and IV, d= comparison between II and V and e= comparison between IV and V.

Table-II: Fasting blood glucose levels from control and diabetic rats treated with MSFE and Metformin

Groups of rats (n=6)	FBG (mmol/L)	
	Mean ± SD	p value
I (C)	5.8±0.68	–
II (A)	23.55±3.48	0.00***a
III (MSFE)	6.06±0.85	0.603*b
IV (A+MSFE)	7.9±1.35	0.00***c
V (A+Metfor.)	10.4±3.21	0.00***d, 0.109*e

Table-III: HbA1c Levels (%) in control and diabetic rats treated with MSFE and Metformin

Groups of rats n=6	HbA1c (%)	
	Mean ±SD	p value
I (C)	5.48±0.12	–
II (A)	12.86±1.33	0.00***a
III (MSFE)	5.5±0.18	0.852 *b
IV (A+MSFE)	8.9±0.65	0.00***c
V (A+Metfor.)	10.03±1.36	0.005**d, 0.096*e

Histology of the pancreas in control and diabetic rats treated with MSFE and Metformin.

Transverse sections of 5µm thickness from pancreas of each rat were chosen. Therefore from each experimental group, 3 x 6 = 18 sections were selected from which 30 slides were prepared for each group. They were observed under the high power objective (HPF, x 400) of the Olympus microscope. Total number of islets of Langerhans were counted from a total of 50 fields examined under high power objective of the microscope in each group. It was expected to aid in qualitative and quantitative assessment of histological changes. The results are described below and are presented in Table IV, V and VI.

Table-IV: Microscopic appearance of Islets of Langerhans

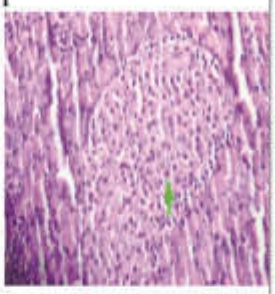
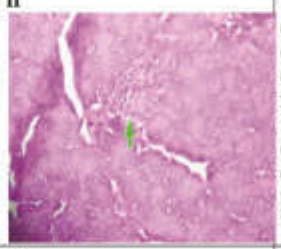
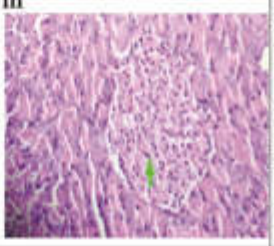
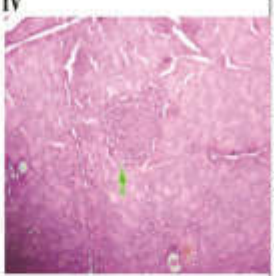
Groups	Plate number and Photomicrograph	Qualitative histological changes
I (C)	I 	Islets of Langerhans were distributed as pale colonies of densely packed cells here and there in the pancreatic parenchyma. They were mostly of similar sizes and oval in shape having a definite architectural pattern. Plenty of β-cells were arranged in regular manner with intact cell membrane, reddish nuclei and pale blue-green cytoplasm within the islets. In the picture, arrow indicates healthy islet of Langerhans and normal distribution of β-cells within the islet (x 400).
II (A)	II 	The islets appeared shrunken and contracted. There was loss of demarcation of islet boundary from the surrounding exocrine pancreas. The reduced β-cell mass created empty spaces within the islets. Islet numbers appeared lesser than control group. Arrow indicates disruption of normal architecture, atrophy of pancreatic islet and empty spaces within the islets (x400).
III (MSFE)	III 	No morphological changes of islets could be observed. Apparent features under the microscope were almost similar to those of control group (Group I). Arrow indicates normal architecture of islet with normal distribution of β-cells within the islet (x400). This bears resemblance to those of Control group (Group I)
IV (A+ MSFE)	IV 	Morphological appearance of the islets appeared to improve in respect to size and number, the β-cell mass appeared to improve in comparison to those of Group II (A) but their size and number appeared to diminish compared to those of control group (Group I). Arrow indicates near normal architecture of islet of Langerhans with possible repopulation of β-cells within the islet (x400).
V (A+ Metfor.)	V 	The islets appeared smaller, contracted and decreased in number compared to those of control group (Group I). However, β-cells appeared to increase in number and cell membranes appeared crenated having better outlines compared to those in the alloxan treated rat group (Group II).Arrow indicates disoriented architecture of islet with crenated membrane and empty spaces are still present within the islet (x400).

Table-V: Number of Islets of Langerhansin control and diabetic rats treated with MSFE and Metformin

Groups of rats (n=6)	No. of Islets	
	Mean ± SD	p-value
I (C)	13.67±1.75	–
II (A)	4.5±1.04	0.00***a
III (MSFE)	15.33±3.44	0. 316*b
IV(A+MSFE)	10.17±1.47	0.00***c
V (A+Metfor.)	06±1.09	0.036**d, 0.00*e

Average diameter of islets of Langerhans (µm) of control and diabetic rats treated with MSFE and Metformin. (Table VI.)

As has been described earliar18 sections of pancreas from each control and experimental group were selected. From each group 50 fields were selected from which diameters of the islets of Langerhans were measured through a scale attached to the high power objective of the microscope.

Table-VI: Average diameter of islets of Langerhans (µm) of control and diabetic rats treated with MSFE and Metformin

Groups of rats (n=6)	Average diameter of Islet (µm)	
	Mean ± SD	p-value
I(C)	23.5 ± 1.64	–
II(A)	05.83 ± 0.75	0.00***a
III(MSFE)	22.67 ± 2.25	0.48*b
IV(A+MSFE)	16.33 ± 2.42	0.00***c
V (A+Metfor.)	08.67 ± 1.50	0.002**d, 0.00***e

Discussion:

The present study was conducted to find out the effects of ethanol extract of *Musa sapientum* flower on alloxan-induced diabetic rats. It was found in very recent researches that oral administration of ethanol extract of *Musa sapientum* flower had caused significant reduction in fasting blood glucose levels and significant increase in serum insulin levels at a dose of 250mg/kg body weight in diabetic rats[10,19]. Thus it was assumed that the phytochemical components of *Musa sapientum* flowers possess insulin secretion stimulating properties[20,21]. The alleviating effect of *Musa sapientum* flower extract was evaluated by biochemical and histological observations at the current research.

In the control group of rats (Group I), the fasting blood glucose and HbA1c coincided with those recorded in previous studies[9,22,23,24,25]. The normal pancreatic architecture of rats was fully preserved in this group (Plate I) which was in accordance with the findings described in previous studies[26,27,28]. In alloxan-induced diabetic group of rats (Group II), biochemical observations had revealed highly significant ($p<0.001$) increase in both fasting blood glucose (FBG) and HbA1c in comparison to those of control group (Group I) (Table II and Table III). This would indicate that this group of rats was suffering from hyperglycemia. Histological studies of transverse sections of pancreas of this group of rats indicate highly significant ($p<0.001$) structural damage to the islets of Langerhans which was evident by shrunken islets with reduction of diameter and number β-cells with loss of demarcation from surrounding parenchyma and innumerable empty spaces within it (Plate II). These observations indicate that alloxan had caused architectural disorientation of rat pancreas leading to destruction and necrosis of β-cells to some extent. These findings appear similar to those of previous works reported earlier [26,27,28] and suggest that total loss of β-cells did not occur creating type 2 diabetic rat model[6,7] .

In Group III (MSFE) the rats showed no significant ($p>0.05$) difference in FBG and HbA1c levels (Table II and III) when compared to those of the control rats(Group I). Pancreatic histology of this group suggested no significant ($p>0.05$) difference both qualitatively and quantitatively from those of the control (Table IV, V, VI and Plate III). Such similarity indicate that the ethanol extract of the flowers of *Musa sapientum* was certainly non-toxic to the rats which also supports earlier observations about the non-toxicity of MSFE in rats[19,29].

In the diabetic rats treated with MSFE (Group IV), significant ($p<0.001$) reduction of FBG and HbA1c levels were observed in comparison to the diabetic control group (Group II) (Table II and III). Observations from the islets of Langerhans had suggested qualitatively lesser damage and a remarkable number of healthy β-cells could be visible in comparison to those of the diabetic group (II) though the sizes of the islets and the number of β-cells were still diminished when compared to control group of rats(Group I) (Plate IV). Thus MSFE could be held responsible for improvement of biochemical and histological parameters in diabetic rats which support the previous works reported by Talubmook and Buddhakala, 2013[19] who claimed hyperinsulinemic effect of MSFE. Perhaps due to concomitant administration of MSFE in the diabetic rats , gradual recovery of pancreatic function had taken place leading to significantly ($p<0.001$) higher number of islets with increased diameter as well as better number of β-cells in comparison to diabetic rats (Group II) (Table IV and V). These findings are also in support of previous researchers[19,26,27,28,30].

The effects of concomitant administration of metformin in diabetic rats (Group V) had caused significant ($p<0.001$ and<0.05) reduction of FBG and HbA1c levels compared to those of alloxan-induced diabetic rats (Group II) (Tables II and III). Increased number and diameter of islets of Langerhans ($p<0.05$) were observed in comparison to Group II rats (Tables V and VI). Histological study in metformin treated diabetic rats had revealed smaller appearance of islets, increased number of β-cells with better outlines of cell membranes (Plate V) in comparison to diabetic rats (Plate II). Although statistical analysis could not establish any significant ($p>0.05$) difference in FBG and HbA1c in MSFE and metformin treated diabetic rats (Group IV and Group V), but surprisingly MSFE treated diabetic rats (Group IV) had shown highly significant ($p<0.001$) increase in number and diameter of islets of Langerhans in comparison to those of metformin treated diabetic rats of Group V(Tables V and VI). These observations suggested ameliorating effects of Musa sapientum flower extract by its insulinotropic activity that was mentioned by previous researchers[9,10,19,24] due to presence of a red pigment (anthocyanin) which boost insulin production by regeneration of β-cells[20,27,31]. It is assumed that MSFE can induce pancreatic β-cell regeneration by stimulating their growth.

Conclusion:

The present study demonstrated the potential ability of ethanol extract of *Musa sapientum* flower to reduce fasting blood glucose (FBG), HbA1c and improvement of number and diameter of pancreatic islets in alloxan-induced diabetic rats. The most encouraging findings of the present study is better recovery of number and diameter of islets of Langerhans in MSFE treated diabetic rats in comparison to metformin treated diabetic rats. It is being greatly expected that complete amelioration might be possible.

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Original Article

Risk Factors, Clinical Profile and Immediate Outcome of Drowning in Children

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Abstract:

Background: Drowning in children is one of the leading causes of child mortality and morbidity. It is the second leading cause of death in western world. In Bangladesh, 46 children die from drowning every day. It has been frequently found among the paediatric patients admitted in Comilla Medical College Hospital. So for successful prevention it needs a regional study among the particular population. **Objectives:** To find out risk factors, clinical profile and immediate outcome of drowning in children and to suggest relevant preventive strategies. **Methodology:** This is a case control study to identify risk factors related to childhood drowning and cross sectional analysis was done for clinical features and immediate outcome. Here total 65 children with drowning were enrolled consecutively from January to December'2020 in Paediatric ward of Comilla Medical College Hospital. The data was collected through interviewing the attendant with a pre-designed questionnaire. Data analysis was performed using SPSS V.12.on. **Results:** Children below 5 years were found to be the commonest age group (61%) with slight male preponderance. Incidence of drowning was more in rural areas (92%) and in the month of June-July (38%). Ponds (63%) were the commonest sites of drowning. In 100% cases, the ponds lack surrounding protective fence. None of the caregivers know how to perform CPR. For children aged 1-4 years' significant risk factors included lack of close supervision (OR16.80; 95% CI 4.66 to 60.50) and poor health of the caregiver (OR 28; 95% CI 5.41 to 144.72). The CNS manifestations like unconsciousness and convulsion were the commonest presenting features. The outcome of children with drowning was good as majority (82%) recovered without morbidity. Seven cases suffered from aspiration pneumonia. There were two cases of death amounting 3% mortality in this study. **Conclusion:** In this study male child less than 5 years, lack of supervision, poor health of the caregiver and absence of fence around ponds are found to be the commonest risk factors. So to prevent drowning in children and its morbidity and mortality there should be adequate supervision of child at risk, proper swimming lesson and provision of fencing around ponds.

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Introduction:

Drowning, an unintentional childhood injury is a global child health problem and is one of the important leading causes of child death and disability. Children in the developed world are thought to be affected most by this event. A Unicef report found that, in 26 of the world's richest nations, drowning was the second leading cause of the injury related death.[1] With one-third of the survivors sustaining neurological damage.[2,3] However more recent descriptive epidemiological studies have demonstrated that childhood drowning rates are higher in the developing countries including Bangladesh and that it is a major public health issue in these settings.[4,5] After submersion in a liquid medium, suffocation and asphyxia may occur, with or without pulmonary aspiration. Within a few minutes, hypoxia and ischemia can rapidly lead to irreversible multisystem injury and often to death. Neurological injury from hypoxemia and ischemia is the primary cause of mortality and long-term morbidity.[6] Drowning death represents the geographical features of the country. Villages are surrounded and intersected by canals and rivers and there are numerous ponds surrounding the households which are used for washing and bathing purposes throughout the year. Children go to these ponds and rivers for the bathing and playing. Most of the villages are

inundated for several months in the monsoon. So it is easy to say the peak of these drowning deaths is in monsoon season. This provided the rationale for considering drowning prevention as a key child survival strategy in Bangladesh. While reductions in infectious disease have resulted in impressive declines in under five child mortality in Bangladeshdrowning is becoming proportionately more important .[7] In Matlab study mothers ranked drowning as the 4th most common perceived cause of childhood deaths behind infectious disease which constitute 20% of deaths in children between 1 to 4 years of age.[6,8] In the present situation as drowning is going to be an important cause of child death in Bangladesh, considerable research and programmatic work is required to understand the nature of the problem and develop appropriate interventions for prevention of drowning.

Highest global rates of drowning in 1yr old male children in Bangladesh (328/100000).[11,15,16] China and India together constitute 43% of all drowning deaths worldwide.[6] It is also the leading cause of death among children of same age group in several European countries.[17] The death rate was 13.1 per 100000 population in Africa[14] and 7.6 in Mexico.[18] In Vietnam, seven of nine drowning cases were of children <15 years[19] and in Thailand, 35% of deaths from injury among school children were due to drowning.[16]

Materials and Methods:

Case control study was done to identify the risk factors. Clinical features and outcome analysis was done by cross sectional study done in Pediatrics ward, Comilla Medical College Hospital, Comilla. Duration of study was one year from January 2020 to December 2020. Children with drowning from 0-12yrs of age admitted in the Pediatrics ward were enrolled. Children beyond 12yrs of age was excluded in this study because of the admission criteria of pediatrics ward. Total 65 cases and 65 suitably matched controls were enrolled in this study. Cases were enrolled randomly-Systematic random sampling was employed. Every alternate case was taken. Control was taken from patients admitted in paediatric ward of Comilla Medical College Hospital during study period reasons other than drowning who were matched precisely for age and gender and did not have any drowning history previously.

Data collection procedure:

The data of both cases and controls were collected by face to face interview. The study protocol was approved by the ethical committee of Comilla Medical College Hospital. All respondents signed an informed consent form before they participate in the survey. After admission from emergency department, every alternate case was taken and each patient was assessed with a careful history and a thorough clinical examination. All parents were interviewed, when available, and grandparents or uncles, when parents were not available. The relevant information's were recorded in a preset questionnaire. The patients were given ABC life support management at the beginning. The unconsciousness and convulsion was considered as CNS manifestation. These patients were monitored with GCS scale. Patients having respiratory problems like fast breathing, respiratory distress was managed with 02administration, airway cleaning and supportive treatment. Children with drowning but without any clinical features of any complication were discharged with counseling after a day of hospital observation. The outcome was categorized as death, survival with morbidity and survival without morbidity.

Data analysis:

x2- test was used to test the differences between cases and controls. Multivariate logistic regression was used to identify factors associated with drowning. All analysis were performed using SPSS V.12.on

Result

Total 65 cases of children with drowning and 65 controls were enrolled in this study. Cases and controls were similar with respect to sociodemographic characteristics. Drowning was more common among boys and among children aged 1-4 years (Table-I)

Table-I: difference in sociodemographic characteristics between cases and Controls.

Sociodemographic characteristics	Case (n=65) n (%)	Control (n=65) n (%)	P value
Age group (years)			
1-4	40 (61.5%)	41 (63.1%)	0.977[ns]
5-9	16 (24.6%)	15 (23.1%)	
10-12	09 (13.8%)	09 (13.8%)	
Sex			
Male	37 (56.9%)	35 (53.8%)	
Female	28 (43.1%)	30 (46.2%)	0.724[ns]

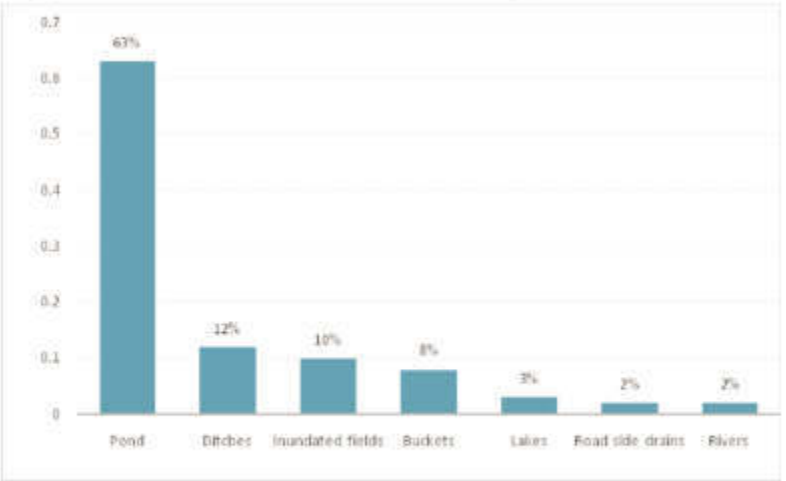
ns=not significant

P value reached from chi square test.

Marked regional variation of drowning was observed. In the present study, total 86% (n=56) drowning events happened in rural areas while only 14% in urban areas.

Children most frequently drowned in rainy season (n=25, 38%) and summer (n=19, 29%). It had been observed that the ponds were very close to the living house and had no surrounding fence in all cases. Total 60% of the drowning events occurred during day time between 10am to1pm whereas 20% between 4pm to 6pm.

Figure- 1: Different sites of drowning



Majority (90.7%) of the drowning incidence occurs due to lack of supervision. In majority of the cases caregivers were busy with meal preparation or other household activities. One child drowned during swimming and another during fishing.

Table II shows the differences in characteristics between cases and controls as well as between their caregivers.Drowning in groups of children was encountered in one occasion where children boarding on a bus felt to a road side ditch following a road accident. A slightly higher proportion of children in the case group were looked after by their grandparents (55% vs 37%), although the difference was marginally non significant. More care givers in the case group had poor health (15% vs 5%, p value<.05). More children in the control group reported that they had provided constant and close supervision of the child when the child was playing in or near water (78% vs 58%,p<0.01).None of the caregivers knew how to perform CPR.

Table-II: difference in baseline characteristics between cases and controls and their caregivers (whole group).

	Case (n=65) n (%)	Control (n=65) n (%)	P value
How often does the child play near water?			
Regularly	57 (87.7%)	58 (89.2%)	0.783^{ns}
Not often	08 (12.3%)	07 (10.8%)	
Did the child have proper swimming lessons?			
No	59 (90.8%)	53 (81.5%)	0.127^{ns}
Yes	06 (9.2%)	12 (18.5%)	
Childs personal character			0.011^{s}
Active	06 (9.2%)	18 (27.7%)	
Inactive	23 (35.4%)	24 (36.9%)	
In between.	36 (55.4%)	23 (35.4%)	
Child's main caregiver			
Mother	22 (33.8%)	35(53.8%)	0.065^{ns}
Grandparents	36 (55.4%)	24 (36.9%)	
Others	07 (10.8%)	06 (9.2%)	
General health of child's caregiver			
Good	55 (84.6%)	62 (95.4%)	0.040^{s}
Poor	10 (15.4%)	03 (4.6%)	
Can the child's caregiver swim?			
No	47 (72.3%)	48 (73.8%)	0.843^{ns}
Yes	18 (27.7%)	17 (26.2%)	
Was there close and constant supervision by the childs caregiver?			
No	27 (41.5%)	14 (21.5%)	0.014^{s}
Yes	38 (58.5%)	51 (78.5%)	
Does the child's caregiver know how to perform CPR?			
No	65 (100.0%)	65 (100.0%)	
Yes	0 (0.0%)	0 (0.0%)	

s=significant; ns=not significant
P value reached from chi square test

Table-III: Differences in health behaviors between cases and controls and their caregivers by age group

Heath behaviors	Case (n=40) n (%)	Control (n=41) n (%)	P value
Aged 1-4years			
Was there close and constant supervision by the child's caregiver?			
No	24 (60.0%)	10 (24.4%)	0.001^{s}
Yes	16 (40.0%)	31 (75.6%)	
General health of child's caregiver.			
Good	28 (70.0%)	38 (92.7%)	0.008^{s}
Poor	12 (30.0%)	03 (7.3%)	

Aged 5-12 years	n=25, n(%)	n=24, n(%)	
Did the child have proper swimming lessons?			
No	24 (96.0%)	20 (83.3%)	0.143[ns]
Yes	01 (4.0%)	04 (16.7%)	
How often did the child play near water?			
Regularly	16 (64.0%)	22 (91.7%)	0.020[s]
Not often	09 (36.0%)	02 (8.3%)	
Childs personal character			
Inactive	9 (36.0%)	14 (58.3%)	0.004[s]
Active	02 (8.0%)	07 (29.2%)	
In between	14 (56.0%)	03 (12.5%)	

s=significant; ns=not significant P value reached from chi square test

In Table III the most noticeable difference was that the risk factors were completely different betweentwo age groups. For the age group of 1-4 years constant and close supervision was reported by 40% of the case group and 76% of the controls which is statistically significant ($p<.05$). In the 5-12years age group those children who did not have a habit of playing near water regularly (36% vs 8% $p<.05$) were drowned more.
In this study 33% of caregivers were estimated illiterate and on the other hand 38%had only primary education.78% victims came from poor socio economic status.

Multivariate logistic regressions stratified by age groups, were fitted to examine factors associated with drowning. Risk factors identified for children aged 1-4 yrs were poor health of the caregivers and lack of close and constant supervision.

Table-IV: Multivariate logistic regression results showing factors associated with rural drowning(n=130)

	Adjusted	95% CI		PValue
	OR	Lower	Upper	
Aged 1-4 years				
Was there close and constan supervision by the child's caregiver?	16.80	4.66	60.50	0.001[s]
General health of child's caregiver	28.00	5.41	144.72	0.001[s]
Aged 5-12 years				
Did the child have proper swimming lessons?	0.794	0.00	0.00	0.999[ns]
How often did the child play near water?	0.74	0.00	0.00	0.999[ns]

s=significant

The approximate submersion times were 5 to 10 minutes in 37 (56.9%) cases though accurate determinations of submersion times were not available. The times that elapsed from the event of submersion to the emergency department arrival were more than 60 minutes in 49 cases, within 30 to 60 minutes in 14 cases and only two cases came within 30 minutes. Some of them were brought directly from the scene of the event to the emergency department. Twenty nine cases came after receiving some traditional practice of resuscitation. 19 children with drowning received preliminary treatment at local hospital before attending the emergency rescuing the child from water andthe most frequently reported measure was rocking the child overhead (25%).Others were spinning and rubbing the body with ash. Mouth to mouth breathing was given in only two cases.

The clinical manifestation has been portrayed in figure 2.Here the commonest manifestation was unconsciousness (84.6%). Other manifestations in decreasing frequency were respiratory distress, convulsion, tachypnoea, stridor, abdominal distension, vomiting, shivering, aspiration syndrome, febrile response.

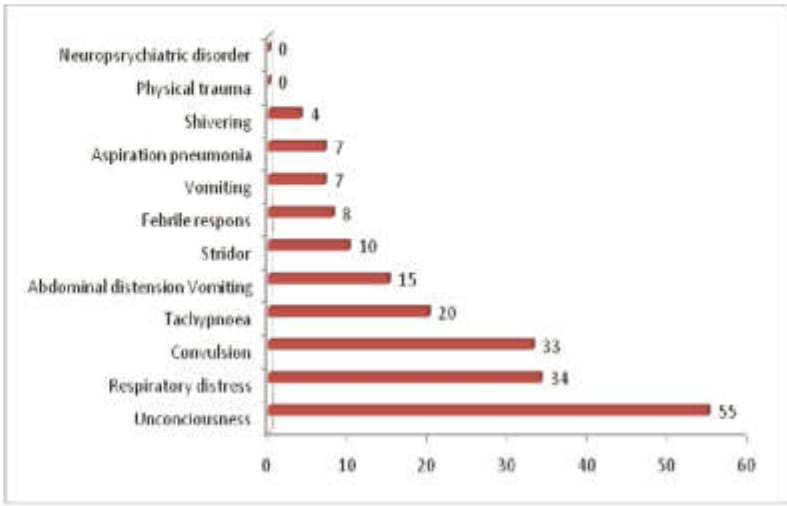


Figure-2: Clinical manifestation of children with drowning (n=65)

None of the cases showed any major sign of physical trauma or previous history of neuro-psychiatric disorder e.g.-epilepsy. Among sixty-five cases those who had received resuscitation at various levels 59(90.7%) of them showed good response, 5 of them showed poor response and one had no response.

Total 82% cases were discharged without any sign of complication (Figure 3) and were in hospital only for < 24hrs, 5% were for 48 hours for lethargy, 10% of the cases were for about one week due to aspiration pneumonia, gastroenteritis etc.

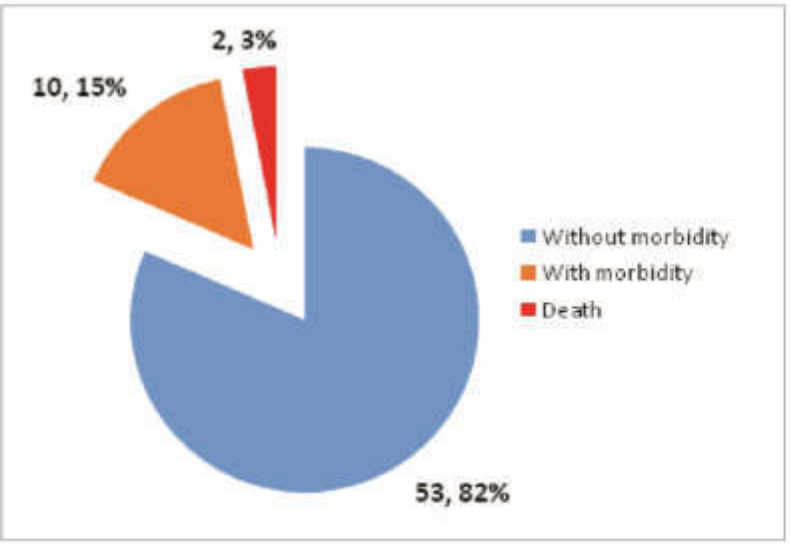


Figure-3: Immediate outcome of childhood drowning

Two cases suffered prolong unconsciousness with repeated convulsion and had prolonged hospital stay. One case suffered from shock. He ultimately died 2hours after admission. Another case was brought dead.

Discussion:

This is a case control study to identify the risk factors related to childhood drowning. Clinical profile and outcome analysis amongst children hospitalized with drowning was done by cross sectional study.

In many studies it is found that there are two peaks in the age specific drowning incidence. The first peak occurs in children 1 to 4 year old and the second peak in adolescent group.[6,18] In this study, the majority of drowning events occurred in children below five years of age (61%) showing a similarity in age predilection. Second peak was not found as admission criteria may have excluded those. Another study published from Bangladesh recently found that the incidence of drowning among children aged 1 to 4 years old was 156 per 100000 children per year.[11] Although causes are multi-factorial; the age specific risk factors exhibit the inherent characteristics of the young children. A child of this age has limited strength, judgment and physical coordination, but a highly developed sense of curiosity with an absent or limited concept of danger.[6, 21, 22]

In this study amongst sixty-five patients, 56% were male and 43% were female. Male childs are mostly affected in all age group. Another study in comilla medical college hospital showed similar result.[10] Similarity also found in Western Australian study.[9] A study on 2007 reported one of the highest global rates of drowning in 1 yr old male children in Bangladesh (328 per 100000).[14,15,21]

Marked regional variation of drowning was observed in this study showing 86% cases in rural areas whereas only 14% in urban areas. This again shows similarity with another study in Comilla Medical College Hospital.[10] A study among Western Australian population showed that 45% of all drowning cases occurred in rural areas, while only 25% in metropolitan areas.[9] Several studies in Matlab, Bangladesh also shows higher rates of drowning events in rural areas that are surrounded by canals, rivers and ponds which are used for bathing and washing.[11, 23, 24]

In this study majority (80%) of the drowning events occurred due to lack of supervision by the caring person mostly in under five children confronting to the results of other reports.[6,10, 18] In several studies the lack of adequate adult supervision has been identified as an important causal factor in childhood immersions. Many instances involved brief lapse in supervision for no more than two to five minutes.[22,25,26] The Brisbane Drowning Study identified lack of parental supervision as a contributing factor in 71% of all serious childhood immersions.[27] In this study 55% of the caregivers were grandparents and their poor health status significantly influence the close and constant supervision . This simulates another study in rural China.[19]

Previous research reported that children especially boys who are more active and bold are at higher risk of injury related mortality and morbidity.[6] However in this study more active children and those who often played near water network were found to have a lower risk. This could imply that as long as precautionary measures such as swimming lessons are taken a more active person might be able to react to sudden changes of water and other condition in a timely manner.

Seasonal variations were observed as majority (38%) of drowning incidence in children happened in the month of June and July which is the period of rainy season whereas 29% in April and May that is in summer. Study in Matlab and Western Australia also showed that most of deaths from drowning occur in summer.[8,9] This is probably due to the reason that during rainy season ponds and other water bodies are full of water. Probably in summer due to hot environment children's are more fascinated to water and this may be responsible for increasing drowning incidence in this season.

Studies both nationally and internationally have identified ponds, ditches, inundated fields, rivers, lakes and canals as a common drowning site for children.[21, 25] In this study majority (63%) of drowning events occurred in ponds whereas in western countries more than 50% of all submersion occurred in private swimming pool.[6,20] The easy access and availability of the drowning sites have direct relation to the incidence of submersion injury in children. Ponds were used for bathing and washing purposes in the rural community and most of the time child's were accompanied by their mother during working in the ponds. They have no fence around them and most of

them are very close to the living house. All these factors testify the validity that ponds emerge as the leading environmental risk factor for drowning in children. Any body of water can cause a hazard to a child. 12% drowning incidences had occurred in ditch whereas 10% in inundated fields in this study group. Bucket drowning is common constituting up to 24% of all toddlers drowning in some region of USA.[6] But in this study drowning in bucket were found only in 8% of all drowning sites showing some dissimilarity where most of drowning events occurred in open bodies of water. A Western Australian study also showed children's are more likely to drown in open bodies of water whereas a small percentage in bucket. Young children with relatively large heads and high centre of gravity can fall in without up-ending the bucket and drown in even a small volume of water.[9]

Most of the drowning events (60%) occurred during day time between 10 am to1pm whereas 20% between 4-6 pm in this study. Similarity showed in Western Australian study. The higher incidence during this specified time period may be due to more engagement of mothers in their household activities. [9, 10]

In this study 78% victims came from poor socio economic status. Other studies also support that drowning rates are higher among children from poor families compared with the non-poor.[9, 10, 11]

Lack of maternal or other caregivers' formal education could influence unintentional drowning associated deaths among these high-risk age groups.[13] In this study 33% caregivers were estimated illiterate and 38% had only primary education. A case- control study in rural areas of China showed that providing an educational program on drowning to parents could help to reduce the childhood drowning rate by 40%.28 Lack of effective formal education and poor income are probably closely related to awareness of parents.

For good recovery resuscitation should be instituted at the scene of drowning.[6] In this study none of the child's caregivers knew how to perform CPR. There had been improper and even harmful traditional practices of resuscitation applied over the victim in 48% cases methods that have no known benefit. Only 3% of families tried proper resuscitation that is mouth to mouth breathing. A case study in Matlab Bangladesh shows that traditional rescue methods applied over 55% children and the most frequently reported measure was spinning the child over head (35%).[29]

For a better outcome it is also important to recover the child from water as early as possible and to start resuscitation if needed. In this study it was found that though most of (52.30%) mothers had first identified her child in water, they refrain from rescuing the child from water due to a belief that if a parent touches a drowning child, the child will die. The Matlab study also revealed similar report. They also found other causal explanations that are primarily associated with "evil spirits" believed to entice young children to water or bewitch mothers so that they forget about the child.[29,30] This believes also make delay in starting resuscitation. CNS injury is the most frequent cause of mortality and long term morbidity.[6] In this study the commonest clinical features were unconsciousness (84%) and convulsion (50%) and this due to hypoxic injury of brain of drowning children. Other reports also support this finding.[10, 17,31] So preventive measures should focus on early resuscitation practices to reduce neurological insult. The respiratory distress (52%) was the second commonest feature. This could be due to aspiration of water in the lung or due to laryngospasm.The vomiting (10%) and abdominal distension (23%) were due to ingestion of water.

The majorities (81%) of drowning patients recovered without morbidity and were in hospital for less than 24hrs. This could be due to short time period of submersion. Another study showed similar findings.[10,31] So if we take effective preventive measures we will be able to reduce child mortality from drowning. Among othersthree cases were suffered from aspiration pneumonia, one from shock who ultimately died. Another two cases suffered prolonged unconsciousness with repeated convulsion and had developed severe neurological dysfunction. They were referred for better management.

There were two cases of death amounting 3% mortality in this study. One case was suffered from shock. He died 2hours after admission. Another case was brought dead. Another study from Bangladesh focused 43% of drowning deaths in children less than 5 yrs and in a national survey 20% of deaths in 1-4 years old children.[23,32,33] Fatality rate may be high because of the delayed rescue and resuscitation efforts. However, this may not the actual data of drowning death probably due to under reporting.

Limitation of the study:

The present study had the following limitations. These should be kept in mind while deciding on the implications of the findings of the study.

a) The sample size was relatively small.

c) The duration of this study was short.

e) Children beyond 12 yrs of age could not be included in this study due to hospital admission criteria.

Conclusion:

This study has provided an insight into the following factors associated with childhood drowning incidents: Majority of drowning incidents found in children below five years of age due to lack of adequate adult supervision.

Children living in rural areas are more likely to drown than those in urban areas. Drowning occur throughout the year with the majority occurring during rainy season. Most of the drowning events occur between 10am-1pm when mothers remain busy with household works. Ponds are the commonest site of drowning which have no surrounding fence. CNS injury is the most frequent cause of mortality and long term morbidity. The majorities of drowning patients recovered without morbidity and were in hospital for less than 24hrs.

Recommendation:

Because most of the drowning events are due to accidents, preventive measures are the first priorities to be considered. Preventive measures should be focused on adequate adult supervision for risk group, swimming training, life jacket in river journey, appropriate fencing of ponds or other open bodies of water.Pre- hospital care and community training programs on Basic resuscitation skill should also be learned by somebody in the rural community. Parental awareness should be generated regarding the potential risk of household containers particularly bucket. A paediatric ICU should be established in every tertiary level hospital.

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Original Article

Assessment of the Utility of the Serum CA 19-9, CEA and CA 125 in Between Benign and Malignant Pancreatic Disease - A Study of 50 Cases.

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Abstract:

Background: : Pancreatic cancer is considered to be one of the leading causes of cancer deaths in advanced countries and it is on the rise in developing countries like India and Bangladesh. A large number of patients with pancreatic cancer receive only palliative therapy because of advanced stage at presentation. Pancreatic cancer is the fifth leading cause of cancer related death in the US with an annual incidence rate of 9 cases per 100,000 people. Approximately 28,000 to 30,000 cases of pancreatic cancer (ductal adenocarcinoma being the most common form) are diagnosed per year. Men are at a slightly greater risk compared to women. **Objectives:** To assess the utility of the serum CA 19-9, CEA and CA 125 in between benign and malignant pancreatic disease. **Methods:** A Cross sectional Observational Studywas conducted over a period of one year July 2016 to June 2017 in Department of Surgery, Dhaka Medical College hospital, Dhaka. A Total of 50 patients were included in this study. All patients with history, sign-symptoms and clinical examination suggesting pancreatic disease attended in Dhaka Medical College Hospital for treatment. Data were collected from these patients by preformed questionnaire and finally data were analyzed Analysis was done by SPSS 22.0 for windows software. **Results:** The CA 19-9, CEA and CA 125 marker levels are useful in the preoperative differential diagnosis of malignant and benign pancreatic disease. The major limitations of the methods are false-negative and false-positive results affecting the diagnostic reliability of the tests. Further clinical investigation is necessary to define more reliable markers and to analyze several markers concomitantly with modern imaging techniques.

Conclusion: Having increased levels of the CEA, CA 19-9 and CA 125 markers in patients with pancreatic pathology usually indicates a malignant nature of the lesion.

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Introduction:

A large number of patients with pancreatic cancer receive only palliative therapy because of advanced stage at presentation. Pancreatic cancer is the fifth leading cause of cancer related death in the US with an annual incidence rate of 9 cases per 100,000 people. Approximately 28,000 to 30,000 cases of pancreatic cancer (ductal adenocarcinoma being the most common form) are diagnosed per year. Men are at a slightly greater risk compared to women[12]. It is 11th most common cancer in the UK (2014), accounting for 3% of all new cases. In males, it is the 12th most common cancer, and is the ninth in females. The incidence of pancreatic cancer in India is low (0.5-2.4 per 100,000 men and 0.2-1.8 per 100,000 women). The incidence of pancreatic cancer is higher in the urban male populations of western and northern parts of India but time trends show that this figure is increasing for both carcinoma of the pancreas and also periampullary tumors[30]. The annual pancreatic cancer load of India in 2001 was approximately 14,230 patients: the estimated current figure is approximately 17,000 and is likely to increase in the near future. The death rate of pancreatic cancer in India is 2.6%. The prevalence of it is 0.68% in Bangladesh[32]. The annual mortality rate per 100,000 people regarding pancreatic cancer in Bangladesh has increased by 12.7% since 1990, an average of 0.6% per year. The chances of survival for untreated patients are very poor. Considering the average of all stages of the disease, 1-year survival rate is 19% and the 5-year survival rate is 4%. At the time of diagnosis

approximately 80% of all pancreatic cancers are metastatic, leaving complete resection as the choice of treatment for only 20% of patients. The 5-year survival of operated patients approaches 40% when performed at specialized medical hospitals[11]. As the pancreas is the retroperitoneal organ, the diagnosis of pancreatic carcinoma has got some difficulties and always the patient presented as a late case. However, a number of continually evolving imaging modalities are available to help diagnose pancreatic carcinoma in patients in whom the disease is suggested clinically. These include the following: Computed tomography (CT), Transcutaneous ultrasonography (TUS), Endoscopic ultrasonography (EUS), Magnetic resonance imaging (MRI), Endoscopic retrograde cholangiopancreatography (ERCP), Positron emission tomography (PET) which has a good role for diagnosis and staging of pancreatic carcinoma. Despite the use of advanced imaging methods and image-guided biopsy procedures in differentiating the pancreatic carcinoma from benign pancreatic diseases, diagnostic limitations still exist. Besides, these procedures have drawbacks since they can be performed only in experienced centers, are invasive and expensive. The people of our country are poor and there are still lack of awareness. In addition there is no medical insurance facility available in Bangladesh.

Different serum tumor markers are being investigated as simple and inexpensive methods in the diagnosis of pancreatic carcinoma. Among these, the value of carcinoembryonic antigen (CEA) is generally reported to be low –between 30 to 35%%[2, 6, 31, 23 and 14] The CA 19-9 is reported to have sensitivity ranging between 68 and 92%[25; 20; 26, 24]. It is also stated that the value of these two tumor markers decrease in differentiating the benign and malignant pancreatic diseases [15; 22] and [16]. There are very few studies investigating the importance of serum CA 125 level in the diagnosis of pancreatic carcinoma [6; 28; 8]. In our study, the value of the serum tumor markers CEA, CA 19-9, and CA 125 alone or in combination, in benign and malignant pancreatic diseases was assessed. A total of 50 patients of pancreatic disease were admitted between July 2016 to June 2017, was enrolled in the study. The association of CEA, CA 19-9, and CA 125 was assessed by determining P values for trends. The multivariate linear regression analysis was used to adjust for clinico-pathological confounding factors to analyze the main outcome measures.

Materials and Methods:

Results: The study was carried out during the period of July 2016 to June 2017. Total 50 patients with history, sign-symptoms and clinical examination suggesting pancreatic disease (both benign and malignant) attended in Dhaka Medical College Hospital for treatment were included for the study.

Table-I: Age distribution of the patients (n=50)

Age (years)	Number of patients	Percentage (%)
30-45	15	30.0
46-60	17	34.0
≥ 61	18	36.0
Total	50	100.0
Mean±SD	53.4±13.1	
Range	(30 – 77) years	

Table-I shows the age distribution of the study patients, age range between 30-77 years. Mean±SD age of the patients was 53.4±13.1 years. 36.0% patients were aged above 61 years and 34.0% patients age was within 46-60 years.

Table-II: Distribution of the patients by sex (n=50)

Sex	Number of patients	Percentage (%)
Male	30	60.0
Female	20	40.0
Total	50	100.0

Table-II shows the distribution of by sex. Maximum patients were male 60.0% and rest 40.0% patient were female. Male: Female ratio was 1.5:1.

Table-III: Distribution of the patients by presence of jaundice (n=50)

Sex	Benign	Malignant
Icteric	13.6%	100.00%
Non-icteric	86.4%	00.0%

Table-III shows the distribution by presence of jaundice. 100% patients of malignant group were ecteric and 13.6% patient of benign group were ecteric.

Table-IV: Distribution of the study patients by type of pancreatic disease (n=50)

Type of disease	Benign Pancreatic Disease (n=22) No. (%)	Malignant Pancreatic Disease (n=28) No. (%)	P value
Benign	22(100.0%)	0(0.0%)	
Acute pancreatic disease	6(27.3%)	0(0.0%)	
Chronic pancreatitis	10(45.5%)	0(0.0%)	0.001[s]
Pancreatic pseudocyt	6(27.3%)	0(0.0%)	
Malignant	0(0.0%)	28(100.0%)	

s= significant

P value reached from Fisher's exact test

Table-IV showed 27.3% patients had acute pancreatic disease, 45.5% patients had chronic pancreatitis, 27.3% patients had pancreatic pseudocyst in benign group.

Table-V: Comparison between histopathological finding and CEA evaluation for pancreatic disease (n=50)

CEA	No	Benign Pancreatic Disease No. (%) (n=22)	Malignant Pancreatic Disease No. (%) (n=28)	P value
Positive	30	2(9.1%)	28(100.0%)	
Negative	20	20(90.9%)	0(0.0%)	<0.001[S]

Data were expressed as frequency and percentage
Fisher's exact test was performed to see the association between benign and malignant pancreatic disease.In case of CEA, 100% of malignant patients found it is significantly high. On the other hand in benign group only 2 patients (9.1%) it is positive.

Table-VI: Comparison between histopathological finding and CA 19-9 evaluation for pancreatic disease (n=50)

CA 19-9	No	Benign Pancreatic Disease No. (%) (n=22)	Malignant Pancreatic Disease No. (%) (n=28)	P value
Positive	31	3(13.6%)	28(100.0%)	
Negative	19	19(86.4%)	0(0.0%)	<0.001[S]

Data were expressed as frequency and percentage
Fisher's exact test was performed to see the association between benign and malignant pancreatic disease. In CA19-9, in malignant group 100% patients found that the test is significantly high but only 3 patients (13.6%) the test was positive.

Table-VII: Comparison between histopathological finding and CA 19-9 evaluation for pancreatic disease (n=50)

CA 125	No	Benign Pancreatic Disease No. (%) (n=22)	Malignant Pancreatic Disease No. (%) (n=28)	P value
Positive	33	9(22.7%)	24(85.7%)	
Negative	17	13(77.3%)	4(14.3%)	<0.001[S]

Data were expressed as frequency and percentage
Fisher's exact test was performed to see the association between benign and malignant pancreatic disease.In CA-125 cases, 85.7% patients shows significant high level in malignant group but only 9 (27.7%) patients were found having high level of this marker.

Table-VIII: Assessment of tumor marker in the diagnosis of pancreatic disease (n=50)

	Diagnostic validity test with 95% CI			
Tumor marker	Sensitivity (%)	Specificity (%)	PPV	NPV
CEA	93.33%	100.0%	100.0%	90.91%
	(77.9% to 98.9%)	(83.0% to 100.0%)		
CA 19-9	90.32%	100.0%	100.0%	86.36%
	(74.2% to 97.8%)	(82.2% to 100.0%)		
CA 125	85.71%	77.27%	82.76%	80.95%
	(67.32% to 95.88%)	(54.62% to 92.09%)		
CEA+CA19-9+CA125	75.0%	100.0%	100.0%	75.86%
	(55.12% to 89.26%)	(84.4% to 100.0%)		

As seen in Table VIII, The markers were assessed individually, CEA had the highest (93.3%) sensitivity and its specificity was 100.0%. CA 19-9 had sensitivity of 90.32% and specificity was 100.0%. The marker CA 125 showed 85.71% sensitivity and 77.27% specificity. In combination of three markers, sensitivity was found 75.0% and specificity was 100.0%.

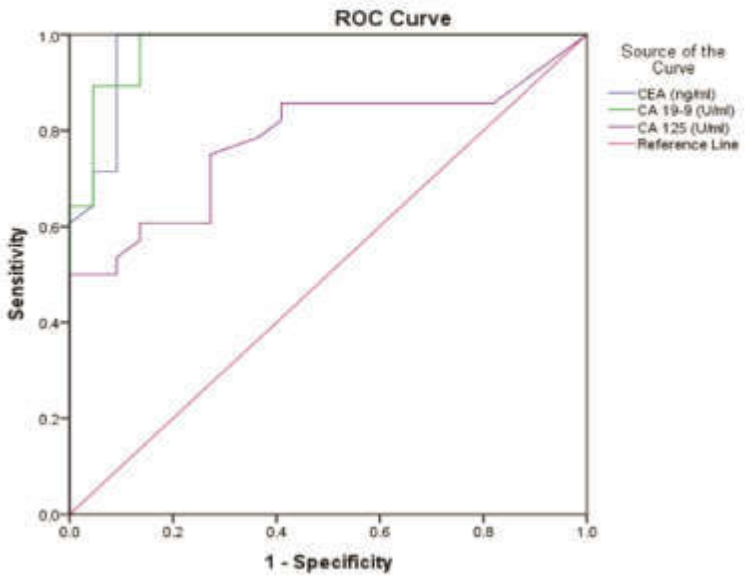

Figure 1: Receiver-operator characteristic curves of CEA, CA 19-9 and CA 125.
Receiver-operator characteristic (ROC) curve of CEA, CA 19-9 and CA 125 for prediction of pancreatic disease

Receiver-operator characteristic (ROC) were constructed using CEA, CA 19-9 and CA 125of the patients with pancreatic disease, which gave a CA 19-9cut off value of (≥ 38.0 U/ml) as the value with a best combination of sensitivity and specificity for pancreatic disease. At this cut-off value the sensitivity and specificity of CEA in diagnosing pancreatic disease were found to be 96.4% and 90.9%, respectively. At this cut-off value the sensitivity and specificity of CA 19-9 in diagnosingpancreatic disease were found to be 96.4% and 86.4%, respectively. At this cut-off value the sensitivity and specificity of CA 125 in diagnosing pancreatic disease were found to be 85.7% and 59.1%, respectively.

Table-IX: Receiver-operator characteristic (ROC) curve of CEA, CA 19-9 and CA 125 for prediction of pancreatic disease

	Cut of value	Sensitivity	Specificity	Area under the ROC curve	95% Confidence interval (CI)	
					Lower bound	Upper bound
CEA (ng/ml)	≥5.3	96.4	90.9	0.970	0.926	1.000
CA 19-9 (U/ml)	≥38.0	96.4	86.4	0.974	0.936	1.000
CA 125 (U/ml)	≥35.0	85.7	59.1	0.779	0.648	0.910

Discussion:

This is cross sectional study carried out in the Department of Surgery, Dhaka Medical College Hospital, Dhaka during the period July 2016 to June 2017 included 50 patients with history, sign-symptoms and clinical examination suggesting pancreatic disease. The aim of the study is to assess the relation between the level of serum CA 19-9, CEA and CA 125 in benign and malignant pancreatic disease.

In this study the age range was between 30-77 years. Mean±SD age of the patients was 53.4±13.1 years. Maximum patients were male 60.0% and 40.0% patient was female. Male: Female ratio was 1.5:1.[1] reported in a study with the aim of 2 neoplastic markers, cancer antigen (CA) 19-9 and CA 125, in the differential diagnosis of pancreatic tumors, ages ranged between 26 and 78 years.

In our study, the sensitivity of CEA in pancreatic carcinoma and benign pancreatic diseases (acute pancreatitis, chronic pancreatitis and pancreatic pseudocyst) was the highest (93.3%) among the three tumor markers we used in our study, which had a specificity of 100% regarding the recommended cut-off level. Many studies shows that sensitivity was reported to vary in a wide range between 28 and 79.4% and specificity between 61.5 and 87.3% in different series [14; 8. 3] found the sensitivity of CEA in the differential diagnosis of pancreatic carcinoma and benign pancreatic diseases was the lowest (39%) among the three tumor markers used in their study, which had a specificity of 91.4%.

In this study, CA 19-9 had the second highest sensitivity (90.32%) for the pancreatic carcinoma, with a specificity of 100%. Sensitivity was reported to range between (74.2% to 97.8%) and specificity between (82.2% to 100.0%).[3] reported CA 19-9 had the highest sensitivity (81.3%) for the pancreatic carcinoma, with a specificity of 75.9%. Many studies demonstrate sensitivity was range between 68 and 92% [6; 28] and specificity between 78 and 94.7% [25, 26]

On the other hand in our study only 9.1% patients were positive in benign cases, regarding CA19-9.[3] showed CA 19-9 positivity in benign pancreatic diseases with jaundice was as high as 64.7% for the recommended cut-off levels and 41.2% for the high cut-off levels. Also in some other studies, high positivity rates of CA 19-9 in benign obstructive jaundice were found, which returned to normal or so after the relief of the jaundice; on the other hand CA 19-9 levels usually remained high in malignant obstructive jaundice after the palliative biliary by-pass [6; 20; 22] and [16].

CA 19-9 antigen is synthesized both by the epithelial cells of the normal biliary tract and by the tumor cells and excreted within the bile [16]. It is suggested that the CA 19-9 antigen, which is high in concentration in the bile of the patients with benign and malignant obstructive jaundice, refluxes into the bloodstream due to the increase in the permeability between bile and blood, secondary to the bile stasis; moreover, it is stated that there can be an inability to degrade the antigen in the liver due to a hepatic dysfunction [20]. Therefore, remeasurement of CA 19-9 after the jaundice subsides can be useful in differential diagnosis of some CA 19-9-positive patients with obstructive jaundice, and if the concentration is still high, then the malignancy potential is high.[9]

Suggested that cutoff levels of 120 IE/liter for CA 19-9 and 100 kU/liter for CA-50 assist in differentiating between malignant and benign diseases of the pancreas, the gallbladder, and the bile duct. No false-positive findings were then seen in benign disease. With CA 19-9 the sensitivity for pancreatic cancer was 82% and with CA-50 the sensitivity for pancreatic cancer was 81%. With sensitivity rates of 83 to 100% the same markers seem to be of value also for gallbladder carcinoma.

In our study, the sensitivity of CA 125 was lowest, 85.71% and specificity 77.27% for the pancreatic carcinoma. In some other studies, the sensitivity was reported to be 45% [6] and 63% [28], and specificity 75%[6]. [3] reported the sensitivity of CA 125 was 56.9% and specificity 77.6% for the pancreatic carcinoma.[8] found the sensitivity of CA 125 to be 24%, referring to the cut-off level that will provide 95% specificity. Also in the study of [6], the positivity rate of CA 125 in benign extrahepatic cholestasis is found to be low. Since CA 125 positivity in the jaundiced subgroup was 55.1% for pancreatic carcinoma and 23.5% for benign pancreatic diseases in the study of [3].

In our study the combination of these all three marker, sensitivity was 75.0% and specificity was 100.0%. [3]reported combination of three markers was examined in the differential diagnosis of pancreatic carcinoma from benign pancreatic diseases, the combination with the highest sensitivity (91.9%) was the one which demanded the positivity of either CA 19-9 or CA 125; nevertheless, the specificity of this combination was 60.3%. So, as CA 125 increased the sensitivity of CA 19- 9 by 10.6%, reduced its specificity by 15.6%. On the other hand, sensitivity reduced to unacceptable values for the combinations which increased the specificity. For this reason, CEA and CA 125 have no contribution to the diagnostic value of CA 19-9 alone in pancreatic carcinoma.[28] suggested that CA 19-9/CA 125 combination provided 97% sensitivity and 96% specificity in differential diagnosis between

pancreatic carcinoma and chronic pancreatitis, thus it had a higher predictive value than using each marker alone.[2] and [6] stated that CEA and CA 19-9 combination is not useful in the diagnosis of pancreatic carcinoma.[3] reported, the ratio of unresectable tumors was significantly higher in the marker-positive patients when compared with marker-negative patients, referring the recommended cut-off levels for each marker. Regarding the high cut-off levels, the ratio of unresectable tumors was significantly higher only in CA 19-9-positive patients. Depending on these findings, it should be taken into account that the probability of unresectability is higher in marker-positive patients. [22] stated that CA 19-9 has no value in predicting the resectability. [29] suggested that CA 19-9 is useful in predicting resectability, while CEA is not. [27] found that CA 19-9 levels were significantly lower in patients with resectable tumors than in those with unresectable tumors. pancreatic carcinoma and chronic pancreatitis, thus it had a higher predictive value than using each marker alone. [2]and[6] stated that CEA and CA 19-9 combination is not useful in the diagnosis of pancreatic carcinoma. [3] reported, the ratio of unresectable tumors was significantly higher in the marker-positive patients when compared with marker-negative patients, referring the recommended cut-off levels for each marker. Regarding the high cut-off levels, the ratio of unresectable tumors was significantly higher only in CA 19-9-positive patients. Depending on these findings, it should be taken into account that the probability of unresectability is higher in marker-positive patients. [22] stated that CA 19-9 has no value in predicting the resectability. [29] suggested that CA 19-9 is useful in predicting resectability, while CEA is not. [27] found that CA 19-9 levels were significantly lower in patients with resectable tumors than in those with unresectable tumors.

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Original Article

The Perception and Attitude of Antenatal Women Demanding Mode of Delivery

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Abstract:

Background: Caesarean section is one of the most commonly performed operations worldwide. Caesarean section on maternal request or demand in the absence of clinical indication seems to be the reason for increasing its rate. **Objectives:** The purpose of this study was to determine the perception and attitude of the antenatal women toward mode of delivery and caesarean section on maternal demand. **Methods:** The present cross sectional study was done in the department of Gynaecology and Obstetrics of District Sadar Hospital, Laxmipur during the period of March 2018 to August 2018. The information about age, parity, education, occupation, previous mode of delivery (where applicable), knowledge and preference of mode of delivery with its reasons and attitude toward caesarean section on maternal demand were collected in the structured questionnaire from the 218 pregnant women after 36 completed weeks of gestation. Finally data from 214 women (nulliparous 109 and multiparous 105) were analyzed. **Result:** Vaginal delivery was the preferred mode in 88% of nulliparous and 94% of multiparous women, and 12% of nulliparous and 6% of multiparous (average 9%) women preferred caesarean section. Those who preferred vaginal delivery they thought that vaginal delivery were the natural process, safer, rapid recovery possible, good for maternal health and cost effective. Fear of labour pain, perineal injury, risk to the baby and maternal convenience were the reasons for choosing caesarean section by nulliparous women and avoidance of perineal injury and previous unpleasant birth experience were the main reasons by multiparous women. About 35.5% of study women believed that they should have the right to demand for caesarean delivery. **Conclusion:** Only a minority of study women expressed their preference for caesarean section in absence of clinical indication. However one third of women still felt that they should have to demand for caesarean delivery.

Key words: Caesarean section, maternal demand.

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Introduction:

There has been a gradual increase in the caesarean section (CS) rate during the last few decades both in the developed and developing countries[1,2]. In the USA, CS rate reached near to 30% in the year 2004[3]. The reasons for the rising CS rate has been attributed at least partly to non-obstetric factors like maternal demand or request for the operation [4,5].

The American College of Obstetricians and Gynecologist (ACOG) argues patient choice should be respected and supported so long as it does not negate ethical medical practices[6]. This view of maternal request caesareans is contrary to recommendations of the international Federation of Obstetrics and Gynecology (FIGO). FIGO states that "Performing cesarean section for non-medical reasons is ethically not justified"[7]. Despite this ethical debate, CS on maternal demand or request is still existing around the globe. Many studies have assessed maternal preferences with respect to mode of delivery. Only 2% of Irish mother[8] (all of them had previous CS) and 4% of British midwives[9] preferred caesarean operation. In Italy, elective CS on maternal request had been practiced by obstetricians since 1996. After implementation of maternal request caesareans, a retrospective study from 1996 to 2000 revealed 7% of caesarean operation were done due to maternal request[10]. In contrast to these low figures, 14% of women in Wales attending antenatal clinics at 39 weeks of

gestation wanted caesarean delivery[11]. A study in England among female obstetricians with an uncomplicated singletone pregnancy shown that 31% of them desired elective CS for themselves[12].

The present study was conducted to determine the knowledge and attitude of Bangladeshi antenatal women toward mode of delivery and on demand caesarean delivery.

Methods:

This cross sectional study was conducted in the Department of Obstetrics and Gynaecology of District Sadar Hospital, Laxmipur during the period of 1st March 2018 to 31st August 2018. The written permission for the study was taken from the hospital authority. Antenatal attendees with previous CS were excluded from the study. A total of 227 women after 36 completed weeks of gestation were invited to participate in the study. Nine women refused to participate because of personal affair. The remaining 218 women were interviewed after obtaining written consent. The structured questionnaire were filled up with information collected from the study subjects. Information about age, parity, education, occupation and previous mode of delivery (where applicable) were recorded. The knowledge of the participants about mode of delivery, their preference of mode of delivery with its reasons and attitude towards CS on maternal demand were also included in the questionnaire.

The quantitative data were tested by unpaired t-test and the qualitative data by Chi-square test. The p- value <0.05 was considered statistically significant. The collected data were entered and analyzed using the Statistical Package for Social Sciences (SPSS) version- 26.

Results:

A total of 218 questionnaires were filled up, of which 4 were discarded due to incomplete information. The remaining 214 (109 nulliparous and 105 multiparous) were included for final analysis. Most of the women (nulliparous 71% and multiparous 78%) were of 20-29 years of age. At least secondary level of education was found in 90% nulliparous and 89% of multiparous women. Majority of the women (83% nulliparous and 79% multiparous) were housewives (Table-I).

Table-I: Sociodemographic parameters of the participants

Parameters	Nulliparous (n=109)	Multiparous (n=105)	p-value
Age (Years)			
<20	27 (25%)	02(2%)	
20-29	78 (71%)	82(78%)	
≥30	04 (4%)	21(20%)	
Mean± SD	23.38±4.49	26.78+4.19	0.001
Educational status			
Primary	11 (10%)	12(11%)	0.933
Secondary	52 (48%)	48(46%)	
Tertiary	46 (42%)	45(43%)	
Occupation			
Housewife	91 (83%)	83(79%)	0.405
Service	18 (17%)	22(21%)	

Vaginal delivery was the preferred mode in 88% of nulliparous and 94% of multiparous women. As an average, 9% of study subjects (12% nulliparous and 6% multiparous) preferred caesarean delivery (Table - II).

Table- II: Personal preference of mode of delivery

Personal preference	Nulliparous (n= 109)	Multiparous (n=105)	p-value
Vaginal Delivery	96(88%)	99(94%)	0.110
Caesarean Section	13(12%)	06 (06%)	

The reasons for choosing vaginal delivery were as it seems to be natural process, safer, rapid recovery, good for maternal health and cost effective (Table-III)

Table-III: Factors responsible for the preference of vaginal delivery.

Factors	Nulliparous (n= 96)	Multiparous (n= 99)	p-value
Natural process	40(42%)	45(45%)	0.594
Safer	81(84%)	82(83%)	0.771
Good for maternal health	58(60%)	60(61%)	0.978
Rapid recovery	31(32%)	29(29%)	0.650
Less cost	13(14%)	16(16%)	0.607

Among the study subjects only 13 (12%) nulliparous and 6 (6%) multiparous women preferred caesarean delivery. Fear of labour pain, perineal injury, risk to the baby and maternal convenience were the reasons for choosing CS by nulliparous women. Avoidance of perineal injury and previous unpleasant birth experience were the main reasons behind the preference of CS by multiparous women (Table - IV)

Table-IV: Factors responsible for the preference of caesarean section

Factors	Nulliparous (n= 13)	Multiparous (n= 6)	p-value
Fear of labour pain	12(92%)	02(33%)	0.007
Fear of perineal or pelvic floor injury	11(85%)	05(83%)	0.944
Risk to baby from vaginal delivery	12(92%)	02(33%)	0.007
Convenience	06(46%)	03(50%)	0.875
Previous birth experience	00(0%)	05(83%)	0.001

Only 36% of nulliparous and 35% of multiparous participants believed that they should have the right to demand for CS (Table-V).

Table-V: Women should have the right to demand for caesarean section

Right to demand caesarean section	Nulliparous (n= 109)	Multiparous (n= 105)	p-value
Yes	39(36%)	37(35%)	
No	70(64%)	68(65%)	
Total	109(100%)	105(100%)	0.934

Discussion:

A steady increase in CS rate globally brings the question in front, which mode of delivery is preferable to our pregnant women. Since 1985 the World Health Organization (WHO) remains within the same stand that CS rate should be within the 10%-15% [13].

The global rising CS rate has been attributed to maternal positive attitude towards caesarean delivery. But in the present study only 8.8% of women expressed their preference toward CS. This probably indicates that our women culturally and socially ready for vaginal delivery. A systematic review of 17 different studies done by McCourt et al [14] evaluating maternal preferences or requests for CS, concluded that their preferences varied from 0.3% to 14%. Another review article written by Kingdon et al[15] evaluating the nulliparous women's views on planned caesarean birth. Their study showed that rate of CS on maternal request without clear medical indication was about 9.0%. In a study on Chilean women regarding their preference toward mode of delivery in the setting where CS rate was about 40%, only 9.4% of study women preferred caesarean delivery[16].

In our study among the total 214 study population (nulliparous women 109 and multiparous women 105), 12% of nulliparous and 6% of multiparous women preferred CS and on an average of 8.8% preferred it. The women's fear about labour pain, risk to the perineal or pelvic floor damage and safety of their babies were the major concern for caesarean delivery among both nulliparous and multiparous women. In multiparous women, a previous negative birth experience was also the important factor for preference of CS over vaginal delivery. These factors for preference of CS is supported by many other international studies [17-22].

About 35.5% of our study population believed that women should have the right to choose the CS. The result is almost similar to the study on Napalese women, where 35% of women thought that they should have the right to demand CS[23]. In contrast to the result of our study, 71% of Singaporean women thought that they should have the right to demand caesarean delivery[24]. The difference may indicate the difference in mental makeup, educational status and socioeconomic condition of developed and developing countries.

Conclusion:

This study concluded that only a few women expressed their preference for caesarean delivery in absence of medical indications. The women's fear and perceptions of genital tract injuries, risk to their babies and previous negative birth experience were the important factors for choosing CS. Proper counseling with psychological support during antenatal visit may help to remove the fear and anxiety. However one third of women, both nulliparous and multiparous, still felt that they should have the right to demand for CS.

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Original Article

Maternal and Foetal Outcome of Prolonged Labour in Dhaka Medical College Hospital

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Abstract:

Background: Prolonged labour is a vivid example of poor maternal and reproductive health care and unfortunate ending of motherhood leading to unacceptably high maternal death and disability. This study was performed with the aim to evaluate the women who presented with prolonged labour in Dhaka Medical College Hospital. **Method:** This prospective study was done in the department of Obstetrics and Gynaecology of Dhaka Medical College Hospital during the period of July 2015 to December 2015 among 105 patients with prolonged labour with h/o trial of labour at home. **Result:** Among 105 patients 66.6% delivered by LSCS, 12.5% had instrumental delivery and20.9% had vaginal delivery. Foetal outcome was 95.3% live birth,3.8% stillbirth and 0.9%neonatal death. **Conclusion:** Prolonged labour has shown a significant impact on maternal and perinatal morbidity and mortality.

Key wards: Prolonged labour, Caesarean section, Maternal morbidity.

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Introduction:

Pregnancy is a physiological event of life for a woman. Every year more than 200 million women become pregnant[1]. About 95 % of women can make a safe delivery and only 5% cases of labour gets prolonged and obstructed[2]. Labour is said to be prolonged when the combined duration of the first and second stage is more than the arbitrary time limit of 18 hours[3]. In Bangladeshab out 90% delivery occurs at home and conducted by untrained attendants[4]. The most recent demographic and health survey shows that only 35.7% of urban and 6.7 % of rural women take assistance at delivery from doctors, trained nursesor midwives[5]. In many cases labour become prolonged and neglected. In rural areas the women live far away from health facilities. They are seldom referred to proper health care facilities. So, in many cases labour are unduly prolonged and many of the patients are taken to hospital in a moribund condition. Under this circumstance the women who survive, suffer from serious morbidities like fistulae, perineal tear, cervical tear, vaginal stenosis and genital prolapse. Perinatal morbidity and mortality are even higher[6]. The effect of prolonged labour is reflected in the parameter of high maternal and foetal mortality and morbidity in our country. Maternal mortality is a serious public health concern in Bangladesh. With the current mortality rate of 194 per 10,0000 live births[7]. Mortalityrateis higher in hospital admitted patients than in the community, because most of the complicated cases of labour come to the hospital with a poor general condition. Major causes of death include hemorrhage, abortion, eclampsia, puerperal sepsis and obstructed labour[4,7]. Motherhood is a physiological transition period in a women's life. This may be a sweetest feeling of a mother. But there is a tragic correlation between pregnancy and death. The estimated number of maternal deaths in 2000 for

the world was 528,000. Every minute one woman dies as a result of pregnancy related complications[8]. We observed many patients with a prolonged and neglected labour admitted in labour ward of Dhaka Medical College Hospital (DMCH). The outcome of these pregnancies was far from the satisfaction. Maternal and neonatal morbidity and mortality were very high which inspired us to undertake a study on the background of prolonged labour and its effects on mother and foetus.

Method:

This prospective study based on observation was done on patients with prolonged labour conducted in the department of Obstetrics and Gynecology of DMCH, during the period of July 2015 to December 2015. Admitted patients were evaluated by history and clinical examination. Socio-demographic factor- age, socioeconomic condition, occupation, educational status was recorded. After obtaining informed written consent patients were enrolled consecutively. Excluded those who had other obstetric or medical complications like eclampsia, diabetes mellitus, hypertension, heart disease, renal disease andintra uterine foetal death. During admission, clinical status of patients (labour pain in hours, ruptures membranes in hours, temperature, dehydration, foetal lie, presentation, position and heart sound was recorded. Pelvic examination was carried out to assess the cervical dilatation, state of liquor amni, degree of caput, moulding and pelvic assessment. Pre partum condition, mode of previous delivery, associated complications and at post-partum period maternal outcome- postpartum hemorrhage, cervical and perineal tear, foul smelling discharge, fever, character of wound were recorded. Foetal condition was evaluated by nature of feeding, development of jaundice, umbilical condition and fever. The data was collected and recorded in a data collection sheet. The statistical analysis was done in SPSS version 2015.

Result:

A total 105 respondents were included in the study. Among them highest frequency of obstructed labour was found among the 21-30 years of age group(57.2%) associated with primi parity (64.7%). Mean age was 25.32+-2 years. Most of them 58.3% had no antenatal care. About 74.9%of the patients were immunized by Injection tetanus toxoid (Table-I).

Table-I: Associated medico-social factors of prolong labour

Factors		Patients No (%)
Maternal age	Below 20 years	27 (25.7%)
	21- 30 years	60 (57.2%)
	31- 40 years	18 (17.1%)
Parity	Primi	68 (64.7%)
	Multi	37 (35.3%)
ANC	Regular	30 (25.2%)
	Irregular	20 (16.5%)
	No ANC	65 (58.3%)
Immunization by T.T	Yes	78 (74.9%)
	No	27 (25.1%)

Prolonged labour was associated with lack of education of the Patients and low socio-economic status. Most of the patients were housewife and had no income of their own (Table-II).

Table-II: Associated socio-demographic factors

Factors		Patients No (%)
Education of the patients	No education	44 (39.2%)
	Primary	48 (42.4%)
	Secondary	18 (15.1%)
	Higher	05 (3.3%)
Occupation	Housewife	75 (71.6%)
	Service holder	30 (28.4%)
Socioeconomic condition	Poor	70 (68.4%)
	Middle class	30 (27.5%)
	Higher class	5 (4.1%)

Majority of the patients (67.6%) were need Caesarean section and 32.4% delivered per vaginally.The rate of CS was highest in our study. Treatment was aimed at relieving the obstruction without any delay in view of decreasing the morbidity (Table-III).

Table-III: Mode of delivery

Mode of Delivery		Patients No (%)
Vaginal		22 (20.7%)
Assisted Vaginal	Ventouse	10 (9.3%)
	Forceps	03 (2.4%)
Caesarean section		70 (67.6%)

Among the study group 51.4% of the patients had labour pain for 24 hours and 8.4% for more than 24 hours. Membranes were ruptured for >24 hours in 8.4% case. 25.28% patients had dehydration and 28.5% patients had raised temperature. 79.2% patients were attended by Dai and more than half (57.2%) of the foetus were in distress (Table-IV).

Table-IV: Clinicalstatusof patientson admission

Factors		PatientsNo (%)
Labour pain hours	Up to 12 hours	42(40.2%)
	12 – 24 hours	54(51.4%)
	>24 hours	9 (8.4)
Ruptured membranes in hours	Up to 12 hours	22 (48.88%)
	12 – 24 hours	15(33.35%)
	>24 hours	8 (17.77%)
Dehydration	Present	28 (25.28%)
	Absent	77 (74.72%)
Temperature	Normal	75 (71.5%)
	Raised	30 (28.5%)
Foetal condition	Distress	60 (57.2%)
	Normal	45 (42.8%)
Attended	By Dai	78 (79.2%)
	By relatives	27(20.8%)

In the current study it was found that major cause of obstructed labour was uterine Hypo tonicity (52.5%). Among them 40.3% primi and 12.2% multipara. Cephalopelvic disproportion (CPD) was found 15.2%, malposition (persistent Occipito-posterior) was present in 16.2% cases. Malpresentation mostly (shoulder and breech) was found with 3.8% casesand unknown cause was 12.3% (Table-V).

Table-V: Causes of prolonged labour

Cause		PatientsNo (%)
Uterine hypo tonicity	Primi	42 (40.3%)
	Multi	13 (12.2%)
Cephalopelvic disproportion (CPD)		16 (15.2%)
Malposition (Occipito-posterior)		17 (16.2%)
Malpresentation		04 (3.8%)
Unknown		13 (12.3%)

Out of 105 cases studied foetal outcome was 90.4% live birth. 3.8% still birth and neonatal death was0.95%.Among the live babies 14.2% cases develop fever, 11.4% develop infection and 23.8% cases develop neonatal jaundice (Table-VI) .

Table-VI: Foetal with Perinatal outcomes

Factors		Patients N (%)
Live birth		95 (95.3%)
Perinatal death	Stillbirth	4 (3.8%)
	Neonatal death	1 (0.9%)
Fever		15 (14.28%)
Umbilical sepsis		12 (11.42%)
Jaundice		25 (23.80%)
No complications		53 (50.50%)

Table VIIshows 57.2% of the total patients had normal puerperium and 42.8% had abnormal puerperium. Many patients had more than one complication. The most common complications were Puerperal sepsis (15.2%), PPH (11.6%), Perineal tear (8.5), Cervical tear (5.7%) and Wound dehiscence (1.8%) No maternal death in this study (Table-VII).

Table-VII: Maternal outcomes in prolonged labour in relation to duration of labour pain

Parameters	Duration of labour hours	(%)		
	Up to 12	13-24	>24	
Normal puerperium	35	25	00	(57.2)
Abnormal puerperium				
P.P.H	2	7	3	(11.6)
Cervical tear	1	4	1	(5.7%)
Perineal tear	2	6	1	(8.5%)
Puerperal sepsis	4	9	3	(15.2%)
Maternal death	0	0	0	(0.0%)
Wound dehiscence	0	1	1	(1.8%)

Discussion:

A small fraction of total population in Bangladesh attends hospital to deliver theirbabies. Those who are fortunate and mainly reside in urban area can enjoy the benefit of modern medical help from hospitals or maternity centers. Only around 6% deliveries in the country takes placein hospitals[9].

This study aims to find a profile of prolonged labour admitted in DMCH. High incidence of prolonged labour was found in primi gravid 64.7%.Age incidence of this study was comparable to other studies[10,11,13]. About 17.1% of the elderly patient (≥30 years) had prolonged labour. Donald[14] reported prolonged labour in about 25% of the elderly patient, where 35 years has been taken as a cut off age for elderly patient. Prolonged labour is said to be 4 times more common in primigravida than multi gravida as reported in fundamental obstetrics and Gynecology[1]. According to Holland and Brow[15], first labour is more prolonged than subsequent ones. Most of the prolonged labour comes from low socio-economic groups (68.4%). Kaniz Fatema's study[10] also showed high incidence of prolonged labour in low socioeconomic (67.85%) and Shanaj's study[12] shows high incidence (70.12%) considering the health facility utilization is inability to bear the cost.

These studyshows 58.3% had no h/o antenatal checkup, Habiba's study[16] showed 70% had no antenatal checkup. But 74.9% was immunized which was like Habiba's study[16] (73.5%) and Kaniz Fatema's study[10] (75.3%). These picture shows that the antenatal care is still low. The checkup during antepartum periodis important because this could find out the risk cases as well as there is an

opportunity to inform the mother and family about the danger signs of labour as well as duration of labour. As for the complication is concerned, in present study 25.28% of the patient were dehydrated and 28.5% had raised temperature. In Fatema's study1027.4% were dehydrated and 29.1% had raised temperature. Habiba's study[17] showed 30% were dehydrated and 32% had raised temp. Though result was not same, but complications were present in all the study e.g. cervical tear, perineal tear, and puerperal sepsis. Habiba's study[16] shows in 33% cases membrane was ruptured for more than12 hours at the time of admission.

In present study 33.35% patient had membrane ruptured for more than 12 hours which was very similar to [10,16]. Foetal outcome shows 95 foetuses were delivered live, 4 were stillbirth and 1 neonatal death. Perinatal mortality was 460/1000 live births in Rashid'sstudy[17], 220/1000in Paramenik's study11and 290/1000 in Ghosh's study13. It is estimated that about 7.3 million perinatal deaths occur annually in the world, most of these in the developing countries. Most developing countries have rates around 35-60/1000 live births[18]. In India the Perinatal mortality is about377/1000 live births[19]. The Perinatal mortality data from institutions are always higher due to the admissions of unbooked cases and emergencies. The rate ranges from 6.9/1000 in the national university hospital, Singapore, 53.6 in Aligarh, India and 103.6 in university hospital, Zaria, Nigeria. Risk factor for perinatal mortality in West Africa a population-based study of 20,326 pregnancies showed that 2.05% perinatal mortality was due to prolonged labour[20].

This study shows that neonates delivered after prolonged labour had poor outcome. Regarding the cause of prolonged labour, this study shows uterine hypotonicity was present 40.3% in primi and 12.2% in multi parity, Kanij Fatema's study[10] showed 51.78%, Shahnaj's study[12] showed 49% case which was less but Habiba's study[16] showed that it was most common factor 55% for prolonged labour. Beischear and Makay [21] their series showed 10% cases with Hypotonicity (23%) and majority had birth canal problem. Though Llewellyn-Jones[22] reported 20% cases having birth canal problem. Hypotonicity was the factor which could be managed well by proper intervention. Butthe present study shows in case of 12.3%, the cause is unknown which is muchlower from Shahnaj's study[12] which showed 37% cases cause is unknown. Further study might reveal the total factors concerning this problem specially the factors underlying the unknown cause.16.2% cases of the present study presented with occipitoposterior, however a study by Beischer and Makay [21] shows a slightly lower.
In this study the incidence of caesarean section is quite high (67.6%). This may be because rate of Forceps delivery and destructive operations are decreasing. Now a days Majority of the patient and no antenatal checkup due to which was not possible to diagnose CPD or Malpresentation and they only presented at the time of delivery with moribund condition and had to managed by caesarean section. A study done by Jeffcoate[23] in Mill Road Maternity Hospital, Liverpool shows incidence of spontaneous vaginal delivery 25%, Forceps 22% and caesarean section 24%. With proper management the high rate of LSCS could be curtailed in patients of prolonged labour. Another study done by Beischer and Mackay[22] shows spontaneous vaginal delivery 62.6%, forceps 26.1% and caesarean 12.2%. The difference shown in these studies are probably due to difference in circumstances particularly age, parity, antenatal care etc. High incidence of caesarean section in this series is due to the facts that this hospital is one of the major referral hospital in Bangladesh which deals with mostly high risk pregnancies and most of the patient in this series reported with labour pain which was already delayed. Pregnancy is a dynamic and changing state and some risk factors that seen important at its beginning (for example, multiparity) must be re-evaluated when new information become available during its process.

Conclusion:

Most of the patients came from poor socio-economic condition and majority did not any antenatal checkup. Labour was conducted at home and they arrived at the hospital only after being mishandled by untrained personnel. Prolonged labour were found to be due to uterine hypotonicity. Most of the patients were delivered by caesarean section and some developed life-threatening complication like PPH. Although overall perinatal mortality was not significant, but a majority of the live born neonates developed complications because of prolonged labour. Prolonged labour has shown a significant impact on maternal and perinatal morbidity and mortality.

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Original Article

Antimicrobial Effects of *Terminalia Chebula* Against Clinical Isolates of *SalmonellaTyphi* and *Escherichia Coli.*

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Abstract:

Background: The king of medicine "*Terminalia Chebula*" is always listed at the top of "Ayurvedic Materia Medica" because of its extraordinary power of healing. The increasing failure of chemotherapeutics and antibiotic resistant facing us the thread of superbugs pathogenic bacteria. This resistance problem needs a renewed effort, resulting in researching effective antibacterial agents against pathogenic microorganisms resistant to current antibiotics **Objective:** To find out the antimicrobial effects of *Terminalia chebula* against *S. typhi* and *E. coli*. **Methodology:** It was a Quasi experimental study. Aqueous and Ethanol extracts of *T. chebula* were prepared and preserved at 4°C temperature. Collected sample *S. Typhi* and *E. coli*. were preserved in skim milk media at 4°C and sub-cultured into Mac Conkey agar media. The bacterial isolates were inoculated in Mueller Hinton Agar media (Oxoid, England). Diameter of zone of inhibition and interpretation were done according to CLSI. **Result:** In case of S. typhi group Mean zone of inhibition produced by aqueous extract of T. chebula was 8.30± 5.40, Ethanol extract 15.80±4.00, Ceftriaxone 21.90±27.78. P-value is 0.001. So Ceftriaxone is clearly superior. In case of E. coli group Mean zone of inhibition produced by Ethanol extract of T. chebula was 18.40±2.83, Ceftriaxone 11.90±10.20. P-value is 0.006. So the result is highly significant means ethanol extract is better than Ceftriaxone against E. coli. Conclusion: Ethanol extract of T. chebula is better than aqueous extract. Ethanol extract producing better inhibitory effects then Ceftriaxone in case of E. coli group. These results support the beneficial effects of T. chebula fruit and would be a potential source for the development of alternative antimicrobial agent in order to treat microbial infections.

Key words: Terminalia chebula, Ceftriaxone, Salmonella typhi, Escherichia coli.

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Introduction:

Human has been encountering life threatening disease from very beginning of their life. They search different remedy from natural source from the ancient period. Today plant and natural sources contribute greatly to the commercial drug preparation. About 25% of the drugs prescribed worldwide are derived from plants.[1] Example includes antimicrobials penicillin (penicillium mould), cardiac stimulant digoxin (digitalis purpurea), salicylic acid (willow bark), atropine (Atropa belladona), theophylline (tea), antimalarial quinine (cinchona bark) and erythromycin (actinomycetes).[2] Antimicrobial resistance is a great concern in twenty first century. Rapid spread of multidrug resistance bacteria causing treatment failure with the conventional antimicrobial agents. According to WHO estimated death from antimicrobial resistance in 2050 is ever higher then cancer![3] Plant source getting popularized due to their wide biological and medicinal activities, higher safety margin and lesser costs. Plants are rich in a wide variety of secondary metabolites such as tannins, alkaloids, terpenoids and flavonoids having been found in vitro since they have antimicrobial properties and may serve as an alternative, effective, cheap and safe antimicrobial for the treatment of microbial infections.[3] Terminalia chebula (Family Combretaceae; local name, Haritaki) is one of the most commonly used plants in traditional medicinal systems in Indian subcontinent. Terminalia chebula fruits are used as antimicrobial, Antioxidant, anti

-inflammatory, wound healing, sore throat, eye and heart diseases.Terminalia chebula contains chebulinc acid, tannic acid, gallic acid, resin, anthroquinone and sennoside. It also contains glycosides, sugar, triterpenoids, steroids and small quantity of phosphoric acid. These compounds were proven to exhibit anti-bacterial, anti-fungal, anti-viral and anti-carcinogenic. The structure of terchebulin, an ellagitannin having a novel tetraphenyl carboxylic acid (terchebulic acid) moiety and biogenetically related.[4] Among the infectious diseases, urinary tract infections (UTIs) are the second most common type of infections in the body.[5] A high level of antibiotic resistance is very significant in uropathogenic bacteria especially Escherichia coli (E. coli), the main aetiological agent of UTIs. It has developed resistance to conventional antibiotics including extended spectrum cephalosporins, fluoroquinolones and carbapenem.[6] another common infectious disease typhoid caused by Salmonella typhi (S. typhi) is also increasing resistance to the conventional antibiotics. The present investigation was designed to study the potential antimicrobial activity of the fruits of Terminalia chebula over S. Typhi and E. Coli by extracting them on organic as well as aqueous solvents. Ceftriaxone is the commonly prescribed drug for infections about 30.19%. But its irrational use makes it resistant to many clinically important pathogens. The study was carried out by taking the extracts at a concentration of 100mg/ml and their activity was recorded by estimating zone of inhibition as produced by disc-diffusion method on Mueller–Hinton agar media.

Methods:

This Quasi experimental study was carried out in the department of Pharmacology and Therapeutics, Chittagong Medical College During the period of January 2016 to July 2017.

Preparation of plant: Collected Terminalia chebula Dried at room temperature and Makes fine powder then Immersion in D/W and Ethanol and kept for 3 days after Filtering by Whatman no. 1. Extract produced by rotatory evaporator and kept at 4°c and Prepare 100mg/ml by Dimethyl sulfoxide (DMSO).

Microorganism: The isolates of S. Typhi and E. coli were collected from microbiology department of Chittagong Medical College, Chittagong. Total 60 samples were collected 30 in each group and identified by a standard method.

Culture and Sensitivity: Collected clinical isolates were Preserved at skim milk media then Subculture at MacConkey agar and kept for 24 hours. Preparation of inoculum and standardization done at 0.5 McFarlands standard. Bacterial suspension kept in Mueller Hinton agar media (Oxoid, England), Aqueous, Ethanol Extract, DMSO and Ceftriaxone (CRO) added and Incubated at 37°c for 24 hour. Susceptibility tests were performed by a Kirby-Bauer disk diffusion method.

Interpretation of zone size: Diameter of zone of inhibition is measured after 24 hours. >13mm zone of inhibition is considered as resistant, 14-20mm as intermediate sensitive and 21 or more is sensitive for both S. Typhi and *E. coli*.[7] In case of extract greater the diameter more active the type tested on the colony of the organisms. Relative effectiveness was also done in relation with standard ceftriaxone.

Statistical analysis: Data was analyzed by SPSS software: Version 18.0. The statistical analysis was included mean, unpaired students T-test, standard deviation, two tailed P value, F Ratio. To examine the statistical significant T-test and ANOVA were done.

Results:

Zone of inhibition produced by Aqueous and Ethanol extract of T. chebula in relation with Ceftriaxone and DMSO

Table-I: Zone of inhibition in mm in Mueller-Hinton agar media.

Pathogen	Aqueous	Ethanol	Ceftriaxone	DMSO
S. typhi	8.87±1.05	16.13±0.68	22.60±1.78	0
E. coli	13.87±0.81	18.34±0.48	12.20±1.98	0

Results are express as mean ± SEM.

Table-II: Inhibitory response of T. chebula in relation to Ceftriaxone

aqueous extract	Pathogen	Zone of inhibition (Mean value)		Inhibitory response
		Aqueous	CRO	
	S. typhi	8.87	22.60	39.20%
	E. coli	13.87	12.20	113.6%
ethanol extract		Ethanol	CRO	
	S. typhi	16.13	22.60	71.3%
	E. coli	18.34	12.20	150.3%

Table-III: Relative effectiveness of aqueous and ethanol extracts of Terminalia chebula in comparison to Ceftriaxone

Standard	Aqueous		Ethanol	
	S. typhi	*E. coli*	*S. typhi*	*E. coli*
Ceftriaxone	39.24%	113.69%	71.30%	150.32%

Table-IV: Mean difference between of T. chebula and Ceftriaxone.

aqueous extract	Pathogen	Zone of inhibition (mean±SEM)	t-test sign. n=30	
		Aqueous	CRO	
	S. typhi	8.87±1.05	22.60± 1.78	**t=6.6437 P=0.0001****
	E. coli	13.87±0.81	12.20± 1.98	**t=0.7806 P=0.4382/ns**
ethanol extract		Ethanol	CRO	
	S. typhi	16.13±0.68	22.60± 1.78	**t=3.3955 P=0.0012****
	E. coli	18.34±0.48	12.20± 1.98	**t=3.0137 P=0.0038****

Table-V: Mean difference between aqueous extract and ethanol extract of T. chebula.

Pathogen	Zone of inhibition (mean±SEM)		t-test sign. n=30
	Aqueous	Ethanol	
S. typhi	8.87±1.05	16.13±0.68	**t=5.8035 P=0.0001****
E. coli	13.87±0.81	18.34±0.48	**t=4.7475 P=0.0001****

Table-VI: Association of mean diameter regarding antimicrobial effectiveness of aqueous extract, ethanol extract of T. chebula and Ceftriaxone.

	Mean	SD			SS	df	MS	F	P
S. typhi									
Aqueous	8.87	5.65	ANOVA	Between	2832.26	2	1416.41	890.08	0.0001**
Ethanol	16.13	3.70		Within	138.41	87	1.59		
Ceftriaxone	22.60	9.63		Total	2970.68	89			
E. coli									
Aqueous	13.87	4.38	ANOVA	Between	603.47	2	301.34	186.09	0.0001**
Ethanol	18.34	2.59		Within	141.07	87	1.62		
Ceftriaxone	12.20	10.72		total	744.54	89			

Discussion:

In case of S.typhi group Mean zone of inhibition produced by aqueous extract of T. chebula was 8.87±1.05, Ethanol extract of T. chebula 16.13±0.68, Ceftriaxone 22.60±1.78. Ethanol extract of T. chebula producing near double zone of inhibition then aqueous extract, which is well correlated with the finding of Mostafa, Rahman and Karim (2011)[8]. In case of E.coli group Mean zone of inhibition produced by aqueous extract of T. chebula was 13.87±0.81, Ethanol extract of T. chebula 18.34±0.48, Ceftriaxone 12.20±1.98. Ethanol extract of T. chebula producing more zone of inhibition then Ceftriaxone and aqueous extract. These findings are very close to the findings of Shankara, Ramachandra, Rajan, Preetham and Ganapathy (2012)[9]. Inhibitory response of ethanol extract of T. chebula in relation to ceftriaxone is 71.3%, 150.3% respectively against S. Typhi and E. Coli group. Ethanol extract of T. chebula is more batter than the ceftriaxone in E. coli group (table 4). These data are alike with Naqvi, Asif, Rehman, and Ahmed (2010)[10]. As the plant extracts has no definite guidelines about zone of inhibition interpretation, so we calculated the relative effectiveness of extracts with that of antibiotic. Relative effectiveness of aqueous and ethanol extracts of T. chebula is 39.24%, 71.30% in S. typhi group. Both extracts effectiveness is less than the ceftriaxone. Relative effectiveness of aqueous and ethanol extract of T. chebula is 113.69%, 150.32% in E. coli group. So the effectiveness is relatively more batter than ceftriaxone. These findings are interchangeable with Mostafa, Rahman and Karim (2011)[8]. In case of S. typhi, aqueous extract of T. chebula produce 8.87 mm (mean zone of inhibition in mm) and Ceftriaxone produces 22.60 mm in agar well diffusion method. P value 0.0001 indicate Ceftriaxone is clearly superior then aqueous extract of T. chebula. Findings are homogeneous with Mostafa, Rahman and Karim (2011)[8]. In case of E. coli Aqueous extract of T. chebula produce 13.87mm (mean zone of inhibition in mm) and Ceftriaxone produces 12.20mm. P value is 0.4382. So the result is not statistically significant though mean zone of inhibition is more in aqueous extract (table-6). In case of S. typhi, Ethanol extract of T. chebula produce 16.13mm (mean zone of inhibition in mm) and Ceftriaxone produces 22.60mm in agar well diffusion method. P value 0.0012, indicate the difference is statistically significant. Ceftriaxone producing significantly more zone of inhibition. In case of E. coli ethanol extract of T. chebula produce 18.34mm (mean zone of inhibition in mm) and Ceftriaxone produces 12.20mm. P value is 0.0038. The result is statistically significant. Ethanol extract of T. chebula shows significantly better zone of inhibition then Ceftriaxone against E. coli. In E. coli group aqueous and ethanol extracts of T. chebula producing more zone of inhibition then Ceftriaxone. These findings are due to resistance of Ceftriaxone against E. coli. Ceftriaxone is about 68% resistance to E. coli in our study. These findings are indistinguishable with Rahman and Huda (2014)[11]. In case of S. typhi ethanol extract of T. chebula producing near double zone of inhibition. P value is 0.0001. In caes of E. coli group aqueous and ethanol extract of T. chebula produce 13.87 and 18.34mm zone of inhibition. P value is 0.0001. In both case p is highly significant. Ethanol extract of T. chebula is significantly better then aqueous extract of T. chebula. These observations may be attributed to two reasons, firstly, due to the nature of biologically active components (alkaloids, flavonoids, sterols, quinine, tannins etc.) which might be enhanced in the presence of ethanol.

In the study of Tariq and Reyaz (2013)[12], It has been documented that alkaloids, flavonoids and tannins are plants metabolites well known for their antimicrobial activity. Secondly, the stronger extraction capacity of ethanol could have produced a greater number of active constituents responsible for antibacterial activity. Only one antibiotic is used to examine the efficacy and comparison with the extracts. So it also needs to evaluate the other antibiotic according to national guideline against the investigated pathogens. Different concentration can also be examined. Antibiotic resistant microbes can be reexamined by adding extracts of T. chebula with that of antibiotic.

Conclusion:

Ethanol extract of T. chebula is better than aqueous extract. Ethanol extract producing better inhibitory effects then Ceftriaxone in case of E. coli group. These results support the beneficial effects of T. chebula fruit and would be a potential source for the development of alternative antimicrobial agent in order to treat microbial infections.

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