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Editorial**Use of Corrosive for Treatment of Haemorrhoid- A Rising Concern**

Haemorrhoid is recognized as one of the most common medical conditions in general population. People with haemorrhoids and those wrongly thought to have haemorrhoids, have a tendency to use self-medication rather than to seek proper medical attention. Treatment options mainly depend on the type and severity of haemorrhoids, patient's preference and the expertise of physicians. Haemorrhoids are effectively treated with dietary and lifestyle modification, medical treatment and/or office based procedures such as rubber band ligation and sclerotherapy. An operation is usually indicated in haemorrhoids refractory to nonsurgical treatment, high graded haemorrhoids and strangulated haemorrhoids. Dietary and lifestyle modification like increasing the intake of dietary fiber and oral fluid, having regular exercise, refraining from straining and reading on the toilet, and avoiding drug causing constipation or diarrhoea.¹

There are several modern drugs and traditional medicine used which are available in a variety of format including pill, suppository, cream and wipes. However, the published literature lacks strong evidence supporting the true efficacy of topical treatment for symptomatic haemorrhoids. For an oral preparation, flavonoids are the most common phlebotonic agent used for treating haemorrhoids.

Many office based procedures (such as rubber band ligation, injection sclerotherapy, infrared coagulation, cryotherapy, radio frequency ablation) are effectively performed for grade I & II haemorrhoids and some cases of grade III haemorrhoids with or without local anaesthesia.

Excisional haemorrhoidectomy is the mainstay operation for grade III and IV haemorrhoids and complicated haemorrhoids. Non excisional operation for haemorrhoids includes Doppler guided haemorrhoidal artery ligation (DGHAL) or known as trans-anal haemorrhoidal de-arterialization (THD), and plication of haemorrhoids (or known as ligation anopexy or mucopexy).¹

Stapled haemorrhoidopexy, also known as a procedure for prolapse and haemorrhoids (PPH), is an alternative and popular operation for treating advanced internal haemorrhoids.

Laser haemorrhoidoplasty is a minimally invasive alternative for the treatment of haemorrhoidal disease, especially for grade 2 and 3 haemorrhoids without major prolapse. Postoperative pain is minimal, and the risk of stenosis or incontinence is almost non-existent.² There is no treatment option of corrosive application for haemorrhoid both in the textbook book and literature.

In our country many traditional healer are using corrosive for treating haemorrhoid. They are advertising of nonsurgical treatment of haemorrhoid. Patients who are scared of surgery often go to them and become deceived. They are applying corrosive in the anal canal and burning the haemorrhoid leading to ulceration, pain and bleeding. In the long run when the wound heals it causes incomplete or sometimes complete anal stenosis resulting in difficulty in defecation and intestinal obstruction. Sometimes faecal diversion is needed for obstruction. Treatment of anal stenosis will result in anal incontinence which becomes a very distressful condition for the patient.

These traditional healer has no licence or registration for practice. Day after day they are practicing openly with advertisement. Most of the time we are overlooking or not giving much attention to them. Although registered physicians are often getting accused of wrong treatment, these traditional healer are out of accusation. That's why they are continuing this practice for long without any obstacle.

There should be rules and regulations to prevent them from applying corrosive for haemorrhoid treatment. Health authority, local government and law enforcing agency should work in collaboration to bring them under punishment. We should make the patients aware about the consequences of corrosive treatment. Media can play an important role for awareness.

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Original Article

Trend of Mode of Delivery in a Rural Government Primary Health Care Facility of Bangladesh

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Abstract

Background: Despite the dramatic rise of Cesarean Section in Bangladesh from 12% in 2010 to 31% in 2016, well above the recommended rate by WHO (5%-15%), decline of Maternal Mortality Ratio has stalled during the period. Evidences from regional and global studies suggest that unjustified preference of Cesarean Sections over vaginal deliveries increases anesthesia related hazards, per and post-operative complications, probability of getting nosocomial infections, delayed recovery and financial burden over family and state.

Aim: The aim of this study was to explore the trend of modes of deliveries and their determinants and outcomes in Upazila Health Complex, Fakirhat, Bagerhat, a rural, government, primary health care facility of Bangladesh.

Methodology: The study was conducted in Upazila Health Complex, Fakirhat, Bagerhat. Data of deliveries occurring in the facility from July 2019 to October 2019 were extracted from Emergency Obstetric Care (EmOC) segment of District Health Information Software, DHIS 2. Data were analyzed with Microsoft Excel 2010 software.

Results: Among the co-morbidities in women undergoing CS, PIH (8.8%) and GDM (6.6%) occurred most, followed by Br. Asthma (5.1%), Hypothyroidism (3.75%), HTN (3.7%) and DM (1.5%). 38.2% Cesarean Sections were done on emergency basis while 61.8% were elective. History of previous CS was the most common indication for CS (24.3%), followed by fetal distress (13.2%), patient's choice (10.2%), PROM (9.5%), postdated pregnancy (8.8%) and prolonged/obstructed labour (8.0%). Primary CS had accounted for 103(75.7%) while 33 (24.3%) were secondary CS. All women with history of previous CS went for secondary-elective CS. 17.6%, 11% and 8% of the patients undergone CS had spinal headache, wound infection and puerperal sepsis respectively. 63.2% of the babies weighing >3000 gram were born through VD.

Conclusion: Cesarean Section saves millions of maternal and neonatal lives around the globe in justified scenarios while its unindicted use leads to unfavorable outcomes involving mother and baby, family and society. To ensure its judicious use in the facility, recommendations were made regarding strict monitoring of partograph, increasing efficiency and awareness among the caregivers to reduce number of primary-elective CS and undertaking community based awareness programme to eliminate the apprehensions of VD and biased motives for CS.

Key words: Cesarean Section, Vaginal deliveries

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Introduction

Bangladesh, despite being a country with limited resources, has achieved substantial success in achieving the millennium development goals (MDGs), although the nation of sustainable development goals (SDGs) has replaced MDGs with the aim to reduce maternal mortality ratio (MMR) to less than 70 per 100,000 live births by 2030.¹ In Bangladesh, the fourth Health, Population & Nutrition Sector Programme (4 HPNSP) for 2017-2022 aims at an MMR target of 105 per 100,000 live births by 2022.

Between Bangladesh Maternal Mortality & Health Care Survey, BMMS 2001 and BMMS 2010, MMR declined significantly from 322 to 194 per thousand live births.² The remarkable progress was linked to fertility reduction, increased access to maternal health care, increased use of maternal health services in the antenatal, delivery and postpartum periods and socioeconomic improvements. Although Bangladeshi women are increasingly seeking maternal care from health facilities, decline of MMR has

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stalled in the current decade being 196 per 100,000 live births in 2016, almost identical to that of the estimate of BMMS 2010.² This particular period has witnessed the dramatic increase in delivery by Cesarean Section (CS) in the country, from 12% in 2010 to 31% in 2016.

Data from both developed and developing countries showed the overall rate of CS to be 27% in 2013.^{3,4} With this rising trend of CS globally, concern is also growing about unnecessary CSs which can elevate maternal and neonatal morbidity and mortality.

The reasons for this increase are complex, multifactorial and not often properly conceived. Professional practice styles, malpractice in the for-profit facilities, change in the maternal obstetric characteristics, economic, social, political and cultural factors have been attributed upon the phenomenon by researchers.^{6,7} The ongoing Demand-side Financing (DSF) Maternal Voucher Scheme has been considered to be another cause of CS rise, although such schemes are prevalent only in a few selected areas.⁸

Despite all these studies, surveys and analyses, lack of studies are there regarding the trend of deliveries in rural areas of Bangladesh. Fakirhat is an Upazila under the district of Bagerhat, located in the south-western part of Bangladesh having an area of 160.68 sq Km and a population of 146,813. This particular study was conducted with the objectives to explore the trend of modes of delivery Upazila Health Complex, Fakirhat, Bagerhat which is a rural, government, primary healthcare facility of Bangladesh as well as to see the determinants and outcomes of the delivery modes patients had undergone.

Methodology

The retrospective study was conducted in Upazila Health Complex (UHC), Fakirhat. Bagerhat under the division of Khulna. Data of the deliveries occurred in Upazila Health Complex, Fakirhat from July 2019 to October 2019 were collected from the Emergency Obstetric Care (EmOC) segment of District Health Information Software, DHIS 2.

The outcome variable was the mode of delivery the

patients had undergone, Cesarean Section (CS) and Vaginal Delivery (VD). Technically, Cesarean Section was defined as a surgical procedure in which incisions are made over the abdomen and uterus of a pregnant woman to deliver her baby" taking place among the admitted pregnant patients in UHC, Fakirhat between July 2019 to October 2019. Vaginal Delivery was defined as an event involving "expulsion of fetus through the vagina which may be spontaneous or assisted" taking place among admitted pregnant patients in UHC, Fakirhat from July 2019 to October, 2019. Elective CS meant the CS done on elective-ly by mutual agreement of the patient and the caregiver/-surgeon while Emergency CS means the CS done on emergency basis to save the life of mother and baby from July 2019 to October 2019 in UHC, Fakirhat. Primary CS implied the procedure occurring for the first time in a patient while Secondary CS implied the repetition of the procedure in a women undergone CS previously. Data were analyzed using Microsoft Excel 2010 software.

Results

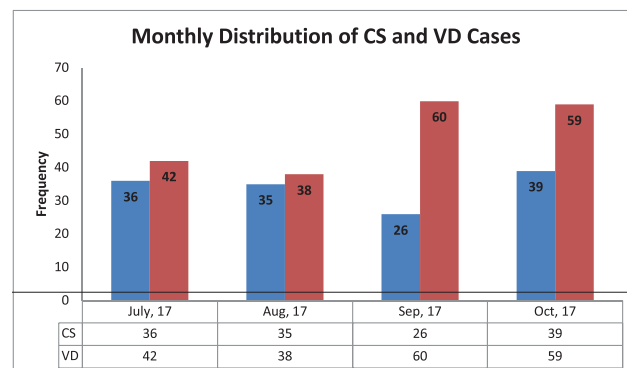
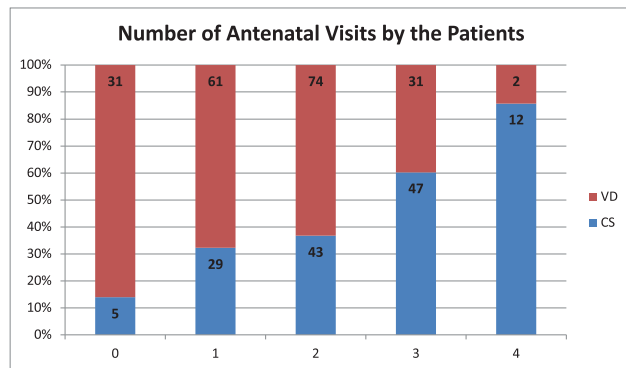


Fig-1: Monthly Distribution of CS and VD Cases

Table I: Age distribution of the patients (n=335)

Age in Years	CS Frequency (%)	VD Frequency (%)	Total
<20	26(33.3)	52(66.7)	78
20-24	49(31.8)	105(68.2)	154
25-29	31(48.4)	33(51.6)	64
30-34	22(75.9)	7(24.1)	29
≥35	8(80.0)	2(10.0)	10
Total	136	195	335

**Fig-2:** Number of Antenatal Visits by the Patients

Average ante natal visit by the patients were 2.2 in CS and 1.56 in VD cases. Patient with higher visits had undergone CS more in comparison to VD; 60% and 85% patients with number of ante natal visits of 03 and ≥ 4 respectively had undergone CS.

Table II: Co-Morbidities/Pre-Existing conditions of the patients (n=77)

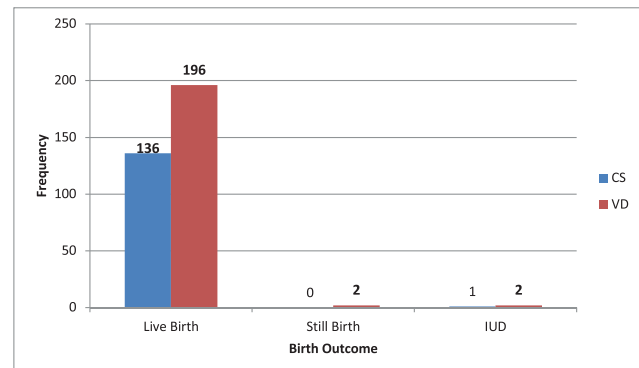
Co-Morbidities Pre Existing Conditions	CS Frequency (%)	VD Frequency (%)	Total
HTN	05(6.8)	01(0.5)	06
PIH	12(8.8)	0(0.0)	12
DM	02(1.5)	0(0.0)	202
GDM	09(6.6)	0(0.0)	09
Br. Asthma	07(5.1)	03(1.5)	10
Hypothyroidism	05(3.7)	0(0.0)	05
Others[Prev. C/S]	33(24.3)	0(0.0)	33
Total	73(out of 136 cases)	4(out of 199 cases)	77 (out of 335 cases)

Table III: Indications for CS (n=136)

Indications for CS	Frequency (%)
Previous CS	33(24.3)
PROM	13(9.5)
Postdated Pregnancy	12(8.8)
APH	06(4.4)
Oligobdramnios	08(5.9)
Polyhydramnin	04(2.9)
Fetal Distress	18(13.2)
CPD	07(5.1)
PE	05(3.7)
Prolonged/Obstructed Labour	11(8.0)
Mal-presentation	05(3.7)
Patient's Choice	14(10.2)
Total	136

Table IV: Complications during/ following CS and VD (n=83)

Complications	CS Frequency (%)	VD Frequency (%)	Total
Retained Placenta	0(0.0)	03(1.5)	03
Postpartum Hemorrhage	05(3.7)	03(1.5)	08
Shock	04(2.9)	0(0.0)	04
Spinal Headache	24(17.6)	N/A	24
Wound Infection [abdominal/episiotomy]	15(11.0)	04(2.0)	19
Vulval Hematomas	0(0.0)	04(2.0)	04
Perineal Tear	0(0.0)	05(2.5)	05
Postpartum Eclampsia	01(.7)	0(0.0)	01
Puerperal Sepsis	11(8.0)	06(3.0)	17
Others	01(.7)	0(0.0)	01
Total	61 (out of 136 cases)	22 (out of 199 cases)	83 (out of 335 cases)

**Fig-3:** Birth Outcome**Table V:** Duration of Hospital Stay (n=335)

Hospital stay in Days	CS Frequency (%)	VD Frequency (%)	Total
1-2	0(0.0)	182(100.0)	182
3-4	0(0.0)	9(100.0)	09
5-6	112(95.8)	5(4.2)	117
≥ 07	24(88.9)	3(11.1)	27
Total	136	199	335

Table VI: DSF Enlisting of Patients (n=335)

DSF Enlisting	CS Frequency (%)	VD Frequency (%)	Total
DSF Enlisted	92(46.9)	104(53.1)	196
DSF Not Enlisted	44(31.7)	95(68.3)	139
Total	136	199	335

Discussion

From July 2019 to October 2019, total 335 deliveries had occurred in UHC Fakirhat. CS contributed 40.6% while NVD accounted for 59.4% of the deliveries. The rate was well above the recommended rate of the WHO.⁵ The study found VD predominating the <20 years and 20-24 years age group, accounting for 66.7% and 68.2% of the deliveries respectively, CS and VD rate was similar in 25-29 years age group (48.4% 51.6%). Advanced maternal age was associated with higher rates of CS. 75.9% and 80% in 30-34 and 35 years age group respectively. Higher parity was also associated with Increased incidence of CS CS was responsible 39.7% and 37.7% deliveries in women with birth order of 1 and 2, while the rate jumped to 70.8% in birth order of 03. These findings were similar to other studies.⁹ Increased maternal age has a relation with increased medical disorders in pregnancy and higher parity is associated with increased mal-presentation and the likelihood of already having one or more CS.

It was found that 73 (53.6%) out of 136 patients undergoing CS had some form of co-orbidities/ pre-existing conditions 40 (29.4%) of these patients had medical disorders and 33(24.3%) had history of one or more previous CS Co-morbidities were remarkably low 04(2%) among patinets with VD. Results showed that of all the CS, 52 (38.25%) were done on emergency basis and 84 (61.8%) were elective CS. Again, primary CS had accounted for 103 (75.7%) of all the CS while 33 (24.3%) were secondary CS. All women with history of previous CS went for secondary. elective CS. History of previous CS accounted for the highest number of the CS (24.39%), followed by fetal distress (13.2%), patient's choice (10.2%), PROM (9.5%), postdated pregnancy (8.89%) and prolonged/obstructed labour (8.0%).

Total 83 patients (24.7%) had developed some form of complications following delivery, the number being 61 and 22 among the patients having undergone CS and VD respectively. Spinal headache (post dural puncture headache) occurred among 17.6% patients undergoing CS similar to study findings when a 250 Quincke needle was used¹⁰ Postpartum hemorrhage occurred in 3.7% and 1.5% CS and

VD patients respectively. Retained placenta was found in 03 VD cases while 01 CS patient had developed postpartum eclampsia. Wound infection in CS patients were 11% whereas 029% of VD patients had episiotomy wound infections, the global estimated range of surgical site infection being 5%-15%, 02% had valval hematoma and 2.5% had perennial tear.¹¹ CS patients had more incidents of puerperal sepsis in contrast to VD patients (08% vs. 0.3%). One patient undergoing CS had developed ARDS and was referred to higher facility. All these results suggest the complications associated with CS are more than those with VD; they raise morbidity and mortality. 46.9% of the patients enlisted under the DSF Maternal Voucher Scheme underwent CS while the rate was 31.7% in those who were not enlisted in the project. The study has a number of limitations. Important determinants like level of education and economic status could not be evaluated due to lack of availability of related data in both DHIS2 and the delivery register. Direct communication with the patients would have verified certain data found on the registers. Maternal height, weight, BMI and other gynecological factor that have association with the mode of delivery could not be assessed. No comparison of results was done with those of from other Upazila Health Complexes with no ongoing DSF Maternal Health Voucher Scheme project.

Conclusion

No wonder of medical science is without adverse effects. Cesarean Section saves millions of maternal and neonatal lives around the globe in justified scenarios while its unindicted use leads to undesirable outcomes involving mother and baby, family and society. There it is high time we thought about judicious use of it as its rising trend has not reduce MMR in Bangladesh in the current decade. Though rationalizing a multimodal approach, extensive researches, facility and adoptable policy making generating political commitment, cooperation and eliminating inequalities to approach, access, and quality healthcare services can lead to a gradual and sustained reduction in unjustified Cesarean Sections.

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Original Article

Comparison of Body Composition Parameters Among Different Groups of Female Medical Students

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Abstract

Background: Body composition refers to the percentages of fat, bone, water and muscle mass in human bodies. Body composition improves physical activity. Regular physical activity is required to maintain overall health. The more weight they gain, the lower physical performance they have. With decline in physical performance, they tend to have functional limitation, to be less physically active and consequently gain more weight. Individuals who are overweight and obese suffer more frequently from joint pain and low back pain.

Objective: The present study aimed at comparing the body composition parameters among different groups of female medical students which might be used as base-line findings for other similar sedentary professionals.

Methods: A cross-sectional analytical type of study was conducted in the department of Anatomy, Dhaka Medical College, Dhaka, from January 2017 to December 2017. The study was performed on 118 female Bangladeshi medical students. It was conducted on three groups depending on body mass index- normal group, overweight group and obese group.

Results: Abdominal skin fold, supra iliac skin fold and calf skin fold were found to be significantly lower ($P<0.05$) in normal group than overweight group but higher in obese group. Body fat percentage, body fat mass were significantly lower ($P<0.05$) in normal group than overweight group but higher in obese group. The study findings suggest that overweight and obese subjects should be careful about regular physical activity for controlling body weight.

Conclusion: The results of the present study demonstrate that body composition parameters were significantly lower in normal group than overweight group and higher in obese group.

Key words: Body composition, Suprailiac, Body fat percentage, Body fat mass

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Introduction

Body composition has two components- a) body fat which is literally fat located within the body b) fat free mass includes bone, water, muscle and tissues.¹ Body composition can be assessed from the various anthropometric measurements such as skin fold thickness, body fat percentage, body fat mass.² The imperfect body composition may

lead to unfavorable physiological consequences in the future. Overweight and obesity causes unhealthy body composition. Lack of physical activity increases the loss of lean muscle tissue and makes the daily activities much more difficult to perform.³

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A sedentary lifestyle is a type of lifestyle with no or irregular physical activity. Sedentary activities include sitting, reading for much of the day with little or no vigorous physical exercise.⁴ Many medical students experience changes in their life patterns during their college years. They spend maximum hours in everyday on studying. Most of them are physically inactive. So, they lead a sedentary lifestyle.⁵ Before entering into the sedentary life style, some of them have normal body weight, some of them are overweight and some of them are obese. Those students who are conscious about their body weight, their weight usually remain in normal. Those students who are not conscious about their body weight; they gain more weight. There is evidence that excessive participation in sedentary behaviours, such as sitting for work/study purposes, is associated with higher risk of overweight and obesity.⁶ Sedentary activities have been associated with a range of adverse health outcomes independently from physical activity.⁷ An inverse relationship between obesity and physical activity leads to muscle weakness which causes risk of injury, back pain, leg pain and joint pain.⁸

To avoid these clinical problems, every student should have knowledge about the importance of body composition that can help them to take measures for a healthy life. The present study aims at comparing body composition parameters of female medical students.

The lifestyles of female medical students established during college years have a significant influence on the health. Their lifestyle is accompanied by decreasing physical activities and increasing sedentary behaviour due to prolonged study hours every day. Physical activity and sedentary behaviour are two distinct modes of behaviour influencing weight and health independently. The findings of the study might be useful in providing data for the anatomist, nutritionist, physiatrist and physiotherapist.

Materials and Methods

This was a cross-sectional analytical type of study. One hundred eighteen female medical college students of 18-25 years of age were selected from different medical colleges of Dhaka city. Medical students usually get admitted in the medical college at or above the age of 18 years and

completed their MBBS course at or above the age of 23 years. Hence, the medical students between the ages of 18-25 years were chosen as subjects.

Subjects were selected according to their availability and willingness. None of their weaknesses were exploited, nor was any undue pressure created to make them participate in the study. The subject who were done regular physical exercise like instrumental exercise, swimming and cycling and any active disease were excluded. Information regarding inclusion and exclusion criteria of female medical students was obtained directly by questionnaire and by physical observation as far as possible. Their ages were determined by the national ID cards. Each participant was given an ID number to avoid repetition. Grouping of participants (normal group, overweight group and obese group) were done according to their body mass index. Grouping mentioned in Table I

All skin folds were measured by skin fold caliper in mm. The skin of measuring sites was wiped with clothes to make it dry and oil-free. To measure the skin fold, the appropriate site for each skin fold (abdominal, suprailiac, calf) was marked with a skin marker on the right side of the body. The skin fold was lifted by placing the thumb and index finger around 3 inches apart on a line that was perpendicular to the long axis of the skin. Skin fold and subcutaneous tissue was firmly raised between thumb and index or middle finger of the left hand and away from the underlying muscle at the marked site. The fold was lifted 1 cm above the site to be measured. The jaws of the caliper were placed perpendicular to the fold, approximately 1 cm below the thumb and index finger. All skin folds were taken while the subject was stand straight except calf skin fold which was taken at sitting position.

Abdominal skin fold was measured 2 cm to the right side of the umbilicus. At first, umbilicus was identified. A point was marked 2 cm to the right side of the umbilicus. The fold was lifted on the marked point and parallel to the long axis of the body. The jaws of the caliper were placed perpendicular to the fold and the measurement was recorded.¹⁰ (Figure: 1a, 1b)

Suprailiac skin fold was measured about 20mm above the iliac crest and 20mm toward the medial line. At first, iliac crest was identified on palpation and was marked by a point. Another point was marked on about 20mm above the iliac crest and 20mm toward the medial line of the body. Then it was measured by raising the fold at the second marked point.¹¹ (Figure: 2a, 2b)

Maximum circumference of the calf was determined on the medial side of the leg. Then medial calf skin fold was taken by raising a vertical skin fold at the maximum circumference of the calf. The medial calf skin fold was measured with the subject seated.¹¹ (Figure: 3a, 3b)

Body fat percentage was calculated by following formula²

$$\text{Body fat percentage} = (1.2 \times \text{Body mass index}) - (10.8 \times \text{Sex}) + (0.23 \times \text{Age}) - 5.4$$

Where sex entered as male 1, female 0

Body fat mass was calculated by following formula³

$$\text{Body fat mass} = \text{Body weight} \times \text{body fat percentage} / 100.$$

All data were checked and edited after collection. Later the data were put into computer and were analyzed with the help of SPSS version 19.0 for windows. Statistical analyses were done by ANOVA test.

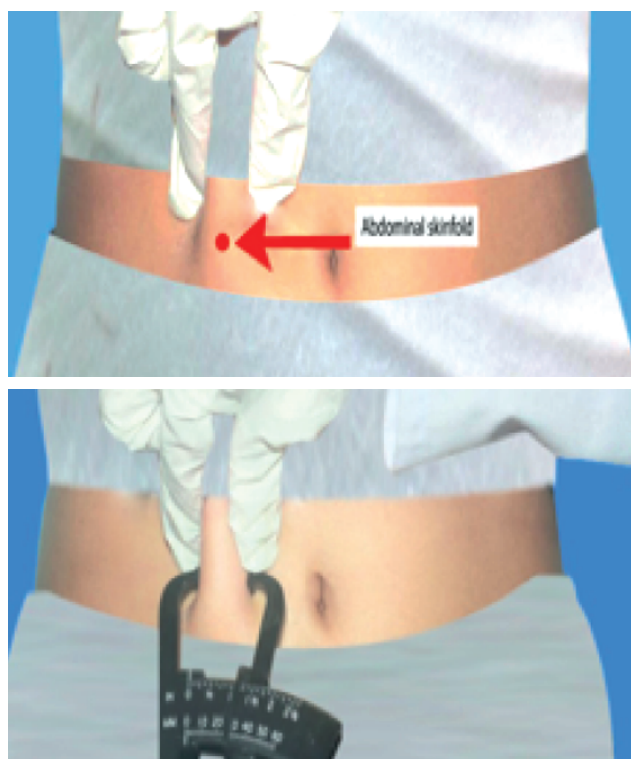


Fig. 1 (a, b) : Photograph showing measurement of abdominal skin fold

- Red dot indicating 2 cm to the right side of the umbilicus
- Measurement of abdominal skin fold



Fig. 2 (a, b): Photograph showing measurement of suprailiac skin fold

- Red dot indicating 20mm above the iliac crest and 20mm toward the medial line
- Measurement of suprailiac skin fold

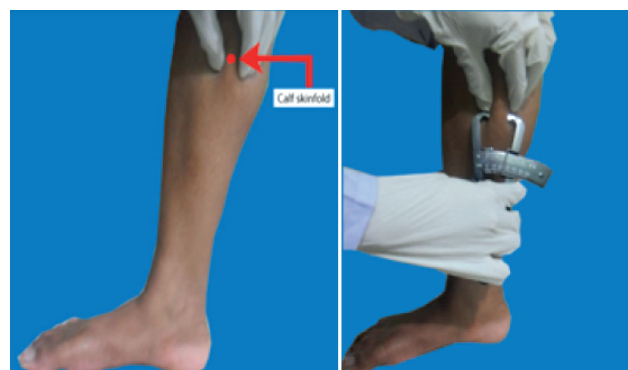


Fig. 3 (a, b): Photograph showing measurement of calf skin fold

- Red dot indicating medial side of the leg
- Measurement of calf skin fold

Results

The present study was carried out on 118 female medical students (18-25 years of age) of different medical colleges of Dhaka city. They were grouped into three categories according to body mass index. (Table I) They were normal group, overweight group and obese group. After collection of data, the findings of this study were analyzed by SPSS (Statistical Package for Social Sciences) version 19.0.

Table I: Grouping of participants

Group	BMI (kg/ m2)	Number (n)
Normal	18.5 - 24.9	48
Overweight	25.0 - 29.9	40
Obese	≥30	30

In normal group, abdominal skin fold was ranged from 14.00-30.00 mm and the mean ($\pm$ SD) abdominal skin fold was 21.96 ± 4.71 mm. In overweight group, abdominal skin fold was ranged from 18.00-32.00 mm and the mean ($\pm$ SD) abdominal skin fold was 25.30 ± 3.61 mm. In obese group, abdominal skin fold was ranged from 22.00-34.00 mm and the mean ($\pm$ SD) abdominal skin fold was 27.93 ± 2.85 mm (Fig 4).

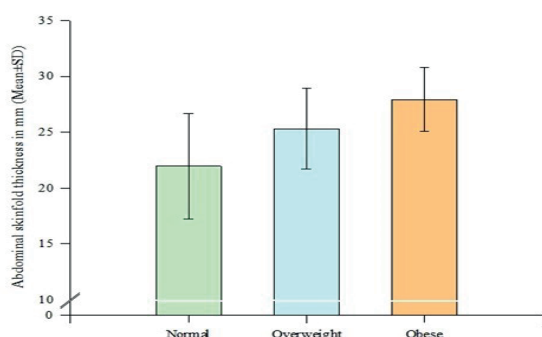


Figure 4: Comparison of abdominal skinfold thickness among normal, overweight and obese students

Difference between mean abdominal skin fold of normal and overweight groups was significant ($p < 0.001$). Mean abdominal skin fold of normal group was lower than overweight group. Significant difference was observed between normal and obese groups ($p < 0.001$) in mean abdominal skin fold. Mean abdominal skin fold of normal group was lower than obese group. There was significant difference observed between mean abdominal skin fold of overweight and obese groups ($p < 0.01$). Mean abdominal skin fold of overweight group was lower than obese group.

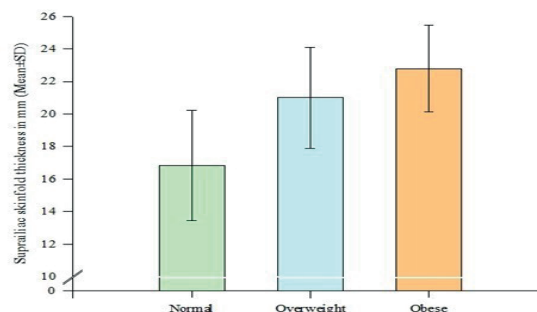


Figure 5: Comparison of suprailiac skinfold thickness among normal, overweight and obese students

In normal group, suprailiac skin fold was ranged from 12.00-24.00 mm and the mean ($\pm$ SD) suprailiac skin fold was 16.83 ± 3.37 mm. In overweight group, suprailiac skin fold was ranged from 16.00-26.00 mm and the mean ($\pm$ SD) suprailiac skin fold was 21.00 ± 3.11 mm. In obese group, suprailiac skin fold was ranged from 20.00-28.00 mm and the mean ($\pm$ SD) suprailiac skin fold was 22.80 ± 2.66 mm (Fig 5).

Difference between mean suprailiac skin fold of normal and overweight groups was significant ($p < 0.001$). Mean suprailiac skin fold was lower in normal group than overweight group. Significant difference was observed between normal and obese groups in mean suprailiac skin fold ($p < 0.001$). Mean suprailiac skin fold of normal group was lower than obese group. There was significant difference observed between mean suprailiac skin fold of overweight and obese groups ($p < 0.05$). Mean suprailiac skin fold of overweight group was lower than obese group.

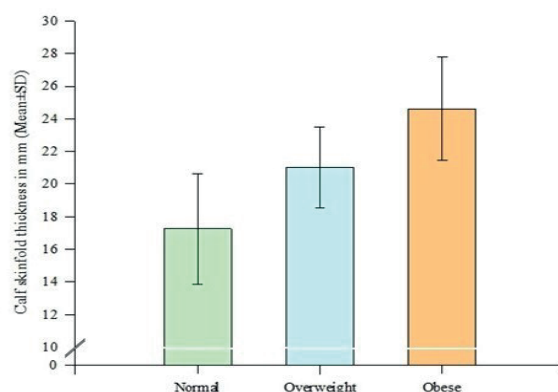


Figure 6: Comparison of calf skinfold thickness among normal, overweight and obese students

In normal group, calf skin fold was ranged from 12.00-24.00 mm and the mean ($\pm$ SD) calf skin fold was 17.25 ± 3.37 mm. In overweight group, calf skin fold was ranged from 18.00-26.00 mm and the mean ($\pm$ SD) calf skin fold was 21.00 ± 2.48 mm. In obese group, calf skin fold was ranged from 20.00-32.00 mm and the mean ($\pm$ SD) calf skin fold was 24.60 ± 3.16 mm. Significant difference was observed among normal, overweight and obese groups in the mean calf skin fold ($p < 0.001$). Mean calf skin fold was lower in normal group than overweight group but higher in obese group (Fig 6).

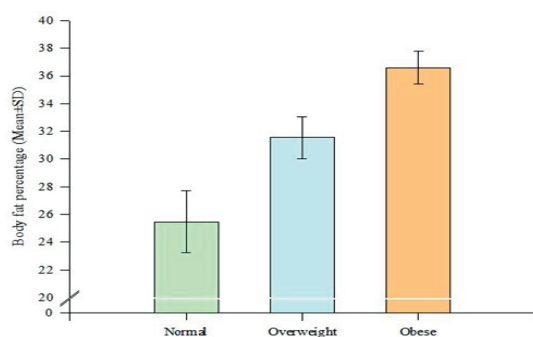


Figure 7: Comparison of body fat percentage among normal, overweight and obese students

In normal group, body fat percentage was ranged from 21.70-28.70 and the mean ($\pm$ SD) body fat percentage was 25.46 ± 2.23 . In overweight group, body fat percentage was ranged from 29.30-34.60 and the mean ($\pm$ SD) body fat percentage was 31.54 ± 1.52 . In obese group, body fat percentage was ranged from 35.20-39.80 and the mean ($\pm$ SD) body fat percentage was 36.58 ± 1.18 . Significant difference was observed among normal, overweight and obese groups ($p < 0.001$) in mean body fat percentage. Mean body fat percentage of normal group was lower than overweight group and obese group; lower in overweight group than obese group (Fig 7).

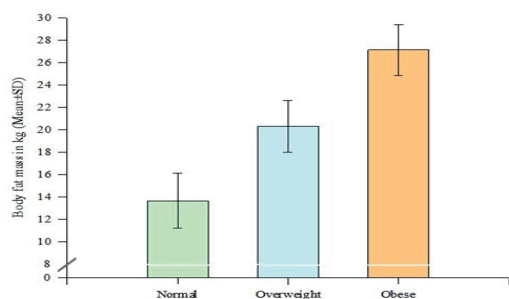


Figure 8: Comparison of body fat mass among normal, overweight and obese students

In normal group, body fat mass was ranged from 8.70-17.50 kg and the mean ($\pm$ SD) body fat mass was 13.66 ± 2.43 kg. In overweight group, body fat mass was ranged from 16.60-25.10 kg and the mean ($\pm$ SD) body fat mass was 20.29 ± 2.30 kg. In obese group, body fat mass was ranged from 22.90-32.60 kg and the mean ($\pm$ SD) body fat mass was 27.11 ± 2.26 kg. Significant difference was observed among normal, overweight and obese groups in the mean body fat mass ($p < 0.001$). Mean body fat mass was lower in normal group than overweight group and obese group; lower in overweight group than obese group (Fig 8).

Discussion

Body mass index or fat mass can influence the performance measures. Regular physical activity improves body composition. The present study was carried out on adult Bangladeshi females comprising of three groups depending on body mass index. The findings of the study were statistically analyzed and revealed important information about body composition parameters. In this discussion chapter, all the variables used for body composition parameters are discussed separately.

Pietraszewska, et al. (2014) found 9.2 ± 3.72 mm, 10.4 ± 5.33 mm and 7.6 ± 2.99 mm, 8.7 ± 3.83 mm mean abdominal and suprailiac skin folds in judo and ju-jitsu athletes respectively. When compared with the present study, mean abdominal and suprailiac skin folds of normal and overweight groups reported by Pietraszewska, et al., were significantly lower ($P < 0.001$).

A study was conducted on suprailiac skin fold by Duerenberg, et al. They reported 22.8 ± 7.3 mm mean suprailiac skin fold in Singaporean Chinese. The mean suprailiac skin fold of normal group of the present study was significantly lower ($P < 0.001$) than the findings of Duerenberg, et al. Leite, et al. (2014) reported 18.52 ± 6.67 mm mean calf skin fold. When compared with the present study, mean calf skin folds of overweight group reported by Leite, et al., were significantly lower ($P < 0.001$).

Pietraszewska, et al. (2014) conducted a study on calf skin fold. They found 4.6 ± 1.72 mm and 4.9 ± 1.81 mm mean calf skin fold in judo and ju-jitsu athletes respectively. When compared with the present study, mean calf skin fold of normal and overweight groups reported by Pietraszewska, et al., was significantly lower ($P < 0.001$).

Rolland, et al. (2003, pp. 552-557) reported 30.2 ± 7.5 , 39.2 ± 5.0 and 45.2 ± 4.4 mean body fat percentage of normal, overweight and obese groups respectively and 30.2 ± 7.5 kg, 39.2 ± 5.0 kg and 45.2 ± 4.4 kg mean body fat mass of normal, overweight and obese groups respectively. The mean body fat percentage and body fat mass of normal, overweight and obese groups of the present study were significantly lower ($P < 0.001$) than the findings of Rolland, et al. A study was conducted on body fat percentage by Ngam, Chayanupatkul and Kittisuparnongkon. They reported 10.27 ± 0.43 , 12.99 ± 1.17 and 16.88 ± 1.27 mean body fat percentage of normal, overweight and obese groups respectively. When compared with the present study, mean body fat percentage of normal, overweight and obese groups reported by Ngam, Chayanupatkul and Kittisuparnongkon was significantly lower ($P < 0.01$). Another study was conducted on body fat mass by Pietraszewska, et al. (2014). They found 12.1 ± 4.26 kg and 15.6 ± 7.78 kg mean body fat mass in judo and ju-jitsu athletes respectively. When compared with the present study, mean body fat mass of normal and overweight groups reported by Pietraszewska, et al., was significantly lower. Hulens, et al. (2000) also conducted a study on body fat mass. They found 16.6 ± 4.1 kg and 48.0 ± 11.4 kg mean body fat mass of normal and obese groups respectively. The mean body fat mass of the normal and obese groups of the present study was significantly lower ($P < 0.001$) than the findings of Hulens, et al. The present study was conducted on a small sample size. Only 118 female students were included in this study. So the students of the present study do not represent all the female medical students of Bangladesh. Some variables could not be measured in proper exposure due to privacy. So there might be error in measurements. Few numbers of publications by other researchers with similar study were available to compare with the findings of the present study. Data was collected from only two medical colleges (Dhaka Medical College Hospital and Delta medical College) in Dhaka city. So the data obtained from the female medical students of the present study cannot be generalized for the whole female medical students of Bangladesh. To get more

precise data on anthropometric measurements and body composition parameters, the use of newer advanced technology like CT scans, X-ray, electrical impedance and hydrostatic weighing etc. are suggested.

Conclusion

The present study was carried out to compare the body composition parameters among normal, overweight, obese students. Skin folds (abdominal, suprailiac and calf), body fat percentage, body fat mass were significantly lower in normal group than overweight group and obese group and all parameters were lower in overweight group than obese group.

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Original Article

Cytology of Voided Urine and Estimation of Serum CA 19-9 Level in the Diagnosis of Bladder Cancer and Correlation with Histopathology

Rahman TS¹, Badruddoza SM², Asafudullah SM³, Rahman TR⁴, Rahman MA⁵

Abstract

Background: Urinary bladder cancer (UBC) is a disease of significant morbidity and mortality. Many tumor markers have been tested for its early diagnosis. But none was proved to be of optimum accuracy.

Aim: This study was intended to evaluate the role of serum CA19-9 and voided-urine cytology in diagnosis of UBC.

Methods: This cross-sectional study was conducted in Department of Pathology, Department of Urology, Rajshahi Medical College & Hospital (RMCH) over a period of two years between January 2016 to December 2017. A total of 56 clinically suspected cases of UBC patients admitted in the RMCH or attended at outpatient department (OPD) of Urology were included in the study. History of the patients and salient clinical findings were obtained from the hospital records. CA 19-9 level was determined by ELISA method. For voided samples, a mid-stream urine was collected and slide was prepared and stained by the hematoxyline and eosin stain and was examined under a light microscope for malignant cells. Tissue biopsy from the site of lesion or operated specimen of tissue was fixed with 40% formalin and was processed stained with haematoxyline and eosin stain and was examined. Of the 56 cases 32 were histologically confirmed as UBC and the rest 24 were other lesions of bladder which served as control in this study.

Result: In the present study, cytology of the voided-urine revealed 2(3.6%) cases to be positive for transitional cell carcinoma (TCC) and CA19-9 was found to be raised in 17(30.4%) cases. Of the 56 cases, 32(57.1%) were histologically confirmed TCC and the rest 24(42.9%) were malignancies or non-specific findings. The sensitivity of CA 19-9 was found to be poor (40.6%) and its specificity was high (83.3%) with high yield of false positive (23.5%) and false negative (48.7%). The overall diagnostic accuracy of the test is 59%. The sensitivity of voided-urine cytology in detecting TCC was extremely low (6.3%), but its specificity was 100% in excluding those who did not have TCC with overall diagnostic accuracy being 46.4%. In only 4.4% cases the two diagnostic modalities had an agreement in the detection of TCC ($k\text{-value} = 0.044$, $p = 0.538$).

Conclusion: The study concluded that CA 19-9 has poor sensitivity in early detection TCC, although its specificity is high. Voided urine cytology has an unwantedly low sensitivity, although its specificity is appreciably high. The two screening markers are in a poor agreement in the detection of TCC.

Keywords: Urinary bladder cancer, urine cytology, serum CA 19-9

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Introduction

Bladder cancer is the ninth most common cancer when combining both sexes and it is more common in industrial countries than in developing countries. This frequency, coupled with the relapsing nature of urinary bladder cancer

(UBC), poses an enormous burden on health care systems.¹ Over three-quarters (77%) of the tumors occur in men and smokers are four times more likely to develop bladder cancer than the non-smokers.^{2,3,4}

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Most patients with bladder cancer were diagnosed after they presented with gross or microscopic hematuria. Bladder cancer is a heterogeneous disease with a variable natural history. Most patients (70%) present with superficial tumors (stages Ta, T1, or carcinoma in situ). However, 3 out of 10 patients present with muscle-invasive disease (T2-4) with a high risk of death from distant metastases. Moreover, roughly between 50% and 70% of superficial tumors do recur, and approximately 10% to 20% of them progress to muscle-invasive disease.^{5,6}

High rate of recurrence and inability to define population at risk, monitoring measures such as periodic cystoscopic examination and urine-based diagnostic tests have been studied extensively for bladder cancers by various investigative groups.^{7, 8} Early detection is essential for effective management and various urinary, serological and molecular markers have been illustrated for TCC. Several tumor markers including the bladder tumor antigen (BTA) series of markers, nuclear matrix proteins (NMP22), and fibrinogen degradation products have been approved for clinical use. However, due to low screening sensitivity/specificity and immense screening costs, none of these markers are used routinely.

Since no reliable markers predicting TCC presence, TCC muscle invasiveness or metastatic TCC have been established to date. Cystoscopy, though a relatively short procedure to be performed with local urethral anesthesia and identifies nearly all papillary and sessile lesions, it is invasive and causes discomfort and distress to the patient. In addition, cystoscopy may be inconclusive at times because of the grossly abnormal appearance of the bladder mucosa, especially in patients with an indwelling catheter or active inflammation. Although considered gold standard for diagnosis, cystoscopy has a false-negative rate either from operator's error or from small areas of carcinoma in situ (CIS), which may be difficult to detect.^{9, 10, 11}

Cytology of voided-urine or bladder washes which involves microscopic identification of exfoliated tumor cells based on cytologic criteria is the most established noninvasive method in the work-up of hematuria and follow-up in patients with a history of bladder cancer and is used as an adjunct to cystoscopy. The method has a high

specificity but relatively low sensitivity, particularly in well-differentiated bladder tumors.¹² A meta-analysis that included data on 18 published series with 1,255 patients reported a sensitivity of 34% and specificity of 99% (95% confidence interval 20-53% and 83-99.7%, respectively).¹³

In the recent years, few studies have demonstrated the clinical significance of measuring urinary CA19-9 levels in bladder cancer patients^{14, 15} and also high serum CA19-9 level in cases of urothelial carcinoma.^{15,16,17} They have shown that this marker may be helpful in predicting extravesical disease and presence of lymph node metastases in TCC.¹⁸ Still it comes into view that the role of CA19-9 as a serum marker in urothelial cancers has not yet been well defined.

Therefore, the present study attempts to address the usefulness of serum CA19-9 for the diagnosis of urothelial carcinoma. The aim of this study was to see the sensitivity and specificity of serum CA 19-9 and urine cytology in diagnosis of bladder cancer as noninvasive diagnostic tools.

Materials and Methods

This cross-sectional study was conducted in Department of Pathology, Department of Urology, Rajshahi Medical College & Hospital (RMCH) over a period of two years between January 2016 to December 2017. A total of 56 clinically suspected cases of UBC patients admitted in the RMCH or attended at outpatient department (OPD) of Urology were included in the study. Written informed consent was taken. History of the patients and salient clinical findings were obtained from the hospital records. Patients having other carcinoma were excluded, for they may be positive for CA 19-9. Of them 32 were histologically confirmed as UBC and the rest 24 were other lesions of bladder which served as control in this study.

CA 19-9 level was determined by ELISA method. The value of CA 19-9 was expressed in U/ml and value upto 37U/l was taken cut-off upper value for normal. For voided samples, a mid-stream urine was collected. No fixatives were required. A glass slide was prepared with Mayer's albumin. Laboratory specimen preparation methods included centrifugation of the specimen for 10 minutes and

then transfer of the sediment to a glass slide before staining. The slide containing the cytospecimen was air-dried for 10-30 minutes, rinsed with 95% ethanol for 10 minutes, stained by the hematoxyline and eosin stain and was examined under a light microscope for malignant cells. The Paris System of reporting urinary cytology described in review chapter was followed for diagnosing and categorizing UCB. Category 5, High grade urothelial carcinoma (HGUC) and 6, Low grade urothelial neoplasm(LGUN) were categorized as positive while others as negative.

Tissue biopsy from the site of lesion or operated specimen of tissue was fixed with 10% formalin and was processed stained with haematoxyline and eosin stain and was examined. Histopathology specimen obtained were of transurethral resection biopsy (TURBT) specimen. So UBC staging was done according to American Joint Commission on Cancer/Union Internationale Contre le Cancer (AJCC/UICC) cancer staging system. Of the 56 cases 32 were histologically confirmed as UBC and the rest 24 were non-specific lesions which served as control in this study.

Results

Over 40% of the patients were 70 years old or more with mean age of the patients being 63.5 ± 14.2 years. Majority of the patients was male (85.7%) and poor (98.2%). The farmers comprised over 40% of the patients. Majority (83.9%) of the patients was smoker and more than one-third (37.5%) had family history of cancer.

Table I: Grading and Staging of UBC

Grading	Frequency
Grade I	4
Grade II	23
Grade III	5
AJCC/UICC staging	
T1	28
T2	04

Of the 32 TCC cases confirmed by histopathology, 23(71.8%) belonged to grade II, 5(15.6%) grade III and only 4(12.5%) Grade I. In terms of AJCC/UICC staging, more than 87.5%(28) were in stage T1, 12.5%(04) in stage T2 and no case of stage T3. (Table I)

Table II: Accuracy of CA19-9 in predicting transitional cell carcinoma

CA19-9 (U/ml)	Histopathological Examination		Frequency (n = 24)
	Urothelial carcinoma (n = 32)	Others	
≥ 37	13	4	17
< 37	19	20	39
Total	32	24	56

Table II shows the sensitivity of the test, serum CA19-9 in differentiating TCC from non-specific findings is 40.6% and specificity of the test in correctly detecting those who do not have TCC is 83.3%. The positive and negative predictive values of the test are 76.5% and 51.3% respectively, while the percentages of false positive and false negatives are 23.5% and 48.7% respectively. The overall diagnostic accuracy of the test is 58.9%.

Table III: Accuracy of voided urine cytology in predicting Urothelial carcinoma

Voided urine cytology	Histopathological Examination		Grading of Urothelial carcinoma	
	Urothelial carcinoma (n = 32)	Others (n = 24)		
Positive for Urothelial carcinom	2	0	Grade III	2
Negative for Urothelial carcinoma	30	24	Grade I, II, III	54
Total	32	24		56

The sensitivity of urine cytology in differentiating TCC from non-specific findings was 6.3% and specificity of the test in correctly diagnosing those who did not have TCC was 100.0%. The positive and negative predictive values of the test were 100.0% and 44.4% respectively, while the percentages of false positive and false negatives are 0% and 55.6% respectively. The overall diagnostic accuracy of the test is 46.4%. (Table III)

Table IV: Test of agreement between CA 19-9 and voided urine cytology

Components of Accuracy Test	Screening test findings		Measures of agreement	
	CA 19-9 (%)	Voided Urine cytology (%)	Kappa statistics (p-value)	Status of agreement
Sensitivity	40.6	6.3	0.044 (p = 0.538)	Poor
Specificity	83.3	100.0		
PPV	76.5	100.0		
NPV	51.3	44.4		
Diagnostic accuracy	58.9	46.4		

In only 4.4% cases the two diagnostic modalities had an agreement in the detection of TCC (k-value = 0.044, p = 0.538).(Table IV)

Discussion

Successful management of transitional cell carcinoma of urinary bladder is largely dependent upon regular surveillance and early detection of persistent or recurrent carcinoma. But the greatest challenge is its early diagnosis. The standard follow-up of patients with a history of bladder cancer is based on cystoscopic examination, an invasive procedure that causes discomfort to the patients. Urine cytology has an excellent specificity with few false-positive cases, but its overall sensitivity is poor, especially for those patients with well-differentiated low-grade transitional cell carcinoma.¹⁹ The ideal assay for bladder cancer should be noninvasive, sensitive, specific, easy, and cost-effective. In recent years, cases of urothelial carcinoma displaying high serum level of CA 19-9 have been reported ^{16, 20, 21} and the present study was intended to investigate the suitability of CA 19-9 and urine cytology as screening tests in the diagnosis of urothelial cancer.

In this study an attempt was made to assess the role of serum CA19-9 and voided urine cytology as screening tests to differentiate transitional cell carcinoma from other neoplasm. In the present study the sensitivity of CA 19-9 was found to be poor (40.6%) and its specificity was high (83.3%) with high yield of false positive (23.5%) and false negative (48.7%). The overall diagnostic accuracy of the test was 58.9%. Voided-urine cytology revealed only two positive cases both of which were high-grade tumour. The

sensitivity of voided-urine cytology in detecting UBC was observed to be extremely low (6.3%), but its specificity was 100% in ruling out those who did not have UBC with overall diagnostic accuracy being 46.4%. In only 4.4% cases the two diagnostic modalities had an agreement in the detection of UBC (k-value = 0.044, p = 0.538).

As the findings of the study were compared and contrasted with those of other studies conducted around the world, a similar picture is evident. Kumar and associates (2017) in prospective analysis of patients with painless hematuria and follow up patients of bladder cancer demonstrated the sensitivity of urinary cytology to be 13.33%, compared to cystoscopy as a gold standard, whereas the specificity of urinary cytology was 100%.²² Cytology was positive only in high grade cases and as such the investigators recommended to omit voided urinary cytology as a screening test. Pall (2012) in an attempt to determine the screening value of serum CA19-9 in detecting urothelial carcinoma found that the marker ranges from 2 to 122 U/ml in patients of urothelial carcinoma with a mean of 26.33 ± 29.28 , while in control, it was 8.48 ± 5.01 U/ml.²⁰ The sensitivity of CA19-9 was 29%. Serum CA19-9 was significantly elevated in invasive disease in comparison with superficial disease (47.17 ± 34.43 vs 16.53 ± 20.13 , $p < 0.001$). A significantly high proportion of patients (58.3%) with invasive disease had value

≥ 37 U/ml compared to superficial disease (15.7%) ($p < 0.001$). Patients with high-grade tumor and metastatic disease also had raised value of CA 19-9 indicating that serum CA19-9 is a marker of aggressiveness of urothelial carcinoma and is almost invariably raised in patients with metastatic disease.

CA 19-9 has been studied alone as well as with other markers, but the results have not been consistent. Serum CA 19-9 has been found to serve as a significant marker for advanced cancer and for tumors with highly malignant potential and is useful for predicting the prognosis of the disease.^{20, 23, 24}

The most important modality for detection of bladder tumor in a new patient with hematuria or a known patient of bladder cancer is cystoscopic evaluation followed by

histopathological examination of resected tumor, if present. Cystoscopy has been proven quite successful in surveillance and follow-up of patients with treated bladder cancer. The drawback of cystoscopy is that it is quite expensive, invasive and uncomfortable.¹⁰ To overcome this, voided urine cytology is used as an adjunctive test in identifying occult cancers that may be missed on cystoscopy. Investigators found sensitivity of urine cytology to vary widely from as low as 16% to as high as 85%. Urine cytology should be performed at the same time as cystoscopy, although its routine use for screening is controversial.²⁵ Urine cytology is associated with a significant false-negative rate, especially for low-grade carcinoma (10 – 50% accuracy rate).

Summarizing the findings of the present study and those of other investigators compared and contrasted, it is evident that neither of the two screening procedures for early detection of UBC is suitable. CA 19-9 has poor sensitivity, although its specificity is high. Voided urine cytology has an extremely low sensitivity, although its specificity is commendably high. The two screening tests were in a poor agreement in the detection of UBC.

Conclusion

From the findings of the study it can be concluded that CA 19-9 has poor sensitivity in early detection of UBC, although its specificity in excluding those who do not have UBC is high. Voided urine cytology has an unwantedly low sensitivity, although its specificity is appreciably high. The two screening markers are in a poor agreement in the detection of UBC. So, neither of the two markers could be dependably employed as a screening tool for early detection of UBC. However, CA 19-9 may be used as prognostic marker and the two diagnostic modalities in conjunction may improve the diagnostic accuracy.

Conflicts of interest: None

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Original Article

Procalcitonin Test More Effective for Early Diagnosis of Neonatal Sepsis

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Abstract

Background: Globally, sepsis is still one of the major causes of morbidity and mortality among neonate. Blood culture is still considered as gold standard for diagnosis of sepsis. Due to low sensitivity and delay in diagnosis by blood culture, Procalcitonin (PCT) level used for rapid diagnosis of neonatal sepsis and to take appropriate measure for early treatment in neonatal sepsis. PCT is more specific than other markers and is detected faster than CRP.

Objective: Therefore, this study has been designed for evaluation of procalcitonin for early diagnosis of neonatal sepsis.

Methods: It was a cross sectional and analytical study place was Department of Microbiology, Sir Salimullah Medical College, Dhaka. The study was conducted from January 2018 to December 2018. Study population included neonates admitted to Neonatal intensive care unit (NICU) with sepsis of Sir Salimullah Medical College and Mitford Hospital. In my study 58 samples were taken.

Result: It was observed that maximum 62.1% patients had early onset (0-3 days) of neonatal sepsis and rest of the patient had late onset ($\geq 3-28$ days) of neonatal sepsis 37.9%. In this study we found that Procalcitonin level was raised in most cases (84.5%) of neonatal sepsis. PCT is a sensitive, independent and useful biomarker of neonatal sepsis. Its persistence correlates with the severity of sepsis.

Conclusion: Continuous monitoring of PCT level could be suggested for proper management of neonatal sepsis.

Key words: Neonatal sepsis, EONS, LONS, Procalcitonin, CRP.

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Introduction

Neonatal sepsis is still contributing significantly to neonatal morbidity and mortality in Neonatal intensive care unit NICU, despite the advances in neonatal care having reduced complications and improved survival in neonates.¹ The leading cause of neonatal death globally are pre-term (35.7%), intrapartum complication (23.4%) and sepsis 15.6%).² The immature immune system and poorly

developed skin barrier of the neonates make them highly susceptible to infections.³

According to World health organization estimate, there are about 5 million neonatal deaths each year, 98% of death occurring in developing countries.⁴ In a study assessing causes of neonatal death in Bangladesh it is shown that sepsis constituted 12% of neonatal deaths.⁵

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In another study in Bangladesh in 2010, severe infections, sepsis, pneumonia and tetanus contributed 20% of neonatal deaths.⁶

Globally, sepsis is still one of the major causes of morbidity and mortality among neonate.

Blood culture is the gold standard test for defining the septicemia.⁷ But 24 to 72 hrs is needed for the culture report to be available. Therefore, antibiotics are administered before culture reports are obtained to reduce morbidity and mortality rates and this in turn increases antibiotic resistance.⁸ Also, The CRP value does not rise significantly until almost 14-48 hrs, after the onset of infection.⁹ PCT is more specific than other markers and is detected faster than CRP. Therefore, PCT is believed to be a new marker of infection, however its use as the only marker for early diagnosis is controversial.¹⁰

Therefore, using accurate, fast and reliable diagnostic methods including specific laboratory markers could be beneficial for early diagnosis of neonatal sepsis. Therefore, this study has been designed for evaluation of procalcitonin for early diagnosis of neonatal sepsis.

Materials & Methods

It was a cross sectional and analytical study placed in Department of Microbiology, Sir Salimullah Medical College, Dhaka. The study was conducted from January 2018 to December 2018. Study population included neonates admitted to Neonatal intensive care unit (NICU) with sepsis of Sir Salimullah Medical College and Mitford Hospital. In my study 58 samples were taken. Procalcitonin level as a diagnostic marker of clinically suspected neonatal sepsis was assessed.

Inclusion criteria: Neonates presenting with one or more of the common features(hypothermia, unable to suck, hypotonia, tachycardia, respiratory distress, purpura, vomiting, abscess etc.) of sepsis (Management Protocol of Newborn, BSMMU 2016).¹¹

Exclusion criteria: Neonates with congenital anomalies and Perinatal asphyxia were excluded.

Technical procedure: With all aseptic precaution, venipuncture site (antecubital vein, radial vein, popliteal vein) (Management Protocol of Newborn, BSMMU 2016) with

10% povidone iodine followed by 70% alcohol, a single sample of blood was collected by using sterile disposable syringe tagged with butterfly needle. Total 3 ml of blood was collected. 1ml of blood was collected for conventional blood culture method. The rest 2 ml blood was for CRP and Procalcitonin test.¹¹

PCT was done by rapid quantitative determination of procalcitonin in serum. It was immunofluorescence Assay.

The protocol was approved by Ethical Review Committee of Sir Salimullah Medical College. After proper explanation regarding the nature of the study written consent were taken from legal guardian of neonates. In this study SPSS-22 used for data analysis. Chi-square test and Unpaired student t-test were done to see the relationship. Validity of PCT as diagnostic marker was determined by comparing with blood culture which takes as gold standard in the diagnosis of neonatal sepsis. A probability 'p' value of <0.05 was considered statistically significant.

Result

It was observed that maximum 62.1% patients had early onset (0-3 days) of neonatal sepsis and rest of the patient had late onset ($\geq 3-28$ days) of neonatal sepsis 37.9% (Figure 1).

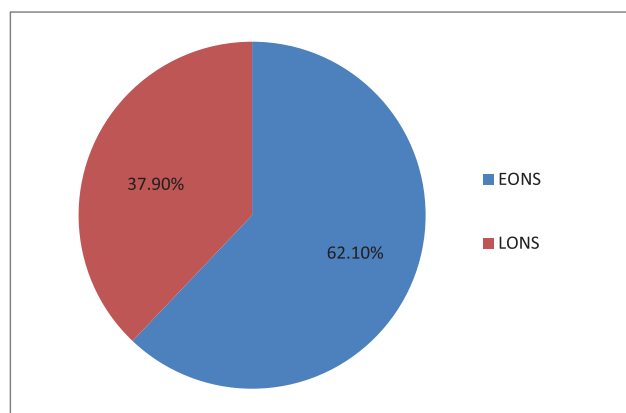


Figure 1 : Distribution of neonates according to type of neonatal sepsis (n=58)

EONS = Early onset neonatal sepsis

LONS = Late onset neonatal sepsis

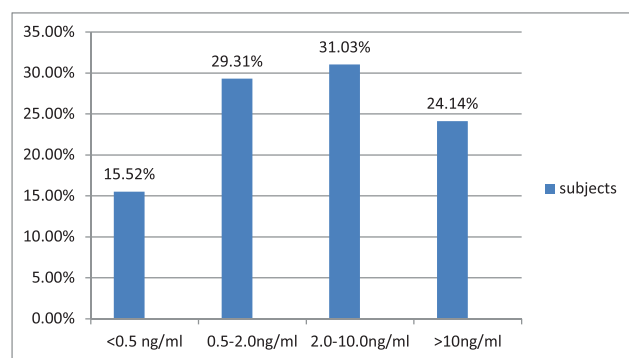


Figure 2: Procalcitonin (PCT) level in blood of study population.

PCT normal range <0.5 ng/ml

PCT Raised ≥ 0.5 ng/ml

In this study we found that Procalcitonin level was raised in most cases (84.5%) of neonatal sepsis.

Table I: Comparison of PCT and CRP value between blood culture positive and negative cases (n=58)

Parameters	Blood culture		p-value
	Positive	Negative	
	(n=14)	(n=44)	
	Mean $\pm$ SD	Mean $\pm$ SD	
PCT	19.34 $\pm$ 23.25	6.76 $\pm$ 4.50	0.001*
CRP	24.86 $\pm$ 16.45	13.73 $\pm$ 13.14	0.012*

P-value reached from Unpaired student t-test, *significant
From this study it was showed that the mean PCT and CRP level significantly higher in blood culture positive cases compare to blood culture negative cases (Table-I).

Table II: Association of PCT with blood culture (n=58)

PCT	Blood culture		p-value
	Positive	Negative	
	(n=14)	(n=44)	
	n (%)	n (%)	
Positive	13	36	0.320ns
Negative	1	8	
Total	14	44	

Note: Gold standard blood culture

Chi-square test was done, ns= not significant

Among 58 patients it was found that PCT was positive in 13 patients out of 14 blood culture positive cases. Total 49(84.5%) were PCT positive and 9(15.5%) were PCT

negative. 36(81.8%) case were PCT positive but blood culture negative. The association statistically not significant ($p > 0.05$) shown in (Table-II).

Table III: Comparison of results of PCT with blood culture (n=58)

PCT	Blood culture		Total
	Positive (n=14)	Negative (n=44)	n (%)
Early onset (0-3 days)			
Positive	9	22	31(83.3%)
Negative	0	5	5(13.9%)
Total	9	27	36(100.0%)
Late onset (>3-28)			
Positive	4	14	18(81.8%)
Negative	1	3	4(18.2%)
Total	5	17	22(100.0%)

Parenthesis indicates the percentage

Note: Gold standard blood culture

The study showed early onset group PCT was positive in 9 patients out of 9 blood culture positive cases. Total 31(83.3%) were PCT positive and 5(13.9%) were PCT negative. In late onset group, PCT was positive in 4 patients out of 18 blood culture positive cases. Among 4 negative patient's blood culture positive in 1 case (Table-III).

Table IV: Performance of PCT and CRP in comparison to blood culture (n=58)

Tests	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy
PCT	92.9	18.2	26.5	88.8	36.2
CRP	85.7	65.9	44.4	93.5	70.69

On comparing the diagnostic performance of sepsis screen following observations were noted: PCT are found to be highly sensitive in diagnosis of neonatal sepsis. The sensitivity, specificity is 92.9%, 18.2% respectively, PPV 26.5% and NPV 88.9%. In comparison of blood culture CRP performance was sensitivity 85.7%, specificity 65.9%, PPV 44.4% and NPV 93.5 % (Table-IV).

Table V: Association of CRP with blood culture (n=58)

CRP	Blood culture		p-value
	Positive (n=14)	Negative (n=44)	
Positive	12	15	0.001s
Negative	2	29	
Total	14	44	

Out of 14 cases of blood culture positive cases, CRP positive in 12(85.7%) cases and negative in 2(14.3%). This finding statistically more significant ($p<.05$) shown in (Table-V).

Discussion

Neonatal sepsis is a major cause of neonatal mortality and morbidity. Blood culture is still considered as gold standard for diagnosis of sepsis. Due to low sensitivity and delay in diagnosis by blood culture, we wanted Procalcitonin (PCT) level for rapid diagnosis of neonatal sepsis and to take appropriate measure for early treatment in neonatal sepsis. Based on the timing of the infection neonatal sepsis has been classified into early onset sepsis (EONS) and lateonset sepsis (LONS).¹² This classification helps to guide antibiotic therapy as it implies differences in the presumed mode of transmission and predominant organisms.

- Early onset neonatal sepsis
- Late onset neonatal sepsis

Early onset neonatal sepsis (EONS)

EONS is caused by organisms prevalent in the maternal genital tract or in the delivery area. It can occur due to ascending infection following rupture of membrane or during passage through birth canal and at the time of resuscitation.¹³

Late onset neonatal sepsis (LONS)

The onset of symptoms is usually delayed beyond 72 hours after birth and the presentation is that of septicemia, pneumonia or meningitis. LONS is a common complication of prolonged admission to the NICU following preterm birth.

In recent years, measurement of procalcitonin and other inflammatory mediators have been reported as a sensitive parameter for the early diagnosis of sepsis and evaluating its outcome.¹⁴

Out of the 58 clinically suspected cases of neonatal sepsis,¹⁴ were culture positive which accounts to 24.14 % in our

study. This is comparable to a study conducted by Kapoor et al. (2005) and Tsering et al. (2011) who reported 20% and 22% respectively of culture proven cases of neonatal sepsis^{15,16}

In this study EONS 62.06% and LONS were 38%. A higher rate of Eons were reported from other developing countries eg., in Iran (77.5% vs 22.5%).¹⁷ The possible explanation for a higher frequency of EOS in their study might be due to PROM and maternal fever.

In this study early onset group PCT was positive in 9 patients out of 9 blood culture positive cases. Total 31 (83.3%) were PCT positive and 5 (13.9%) were PCT negative. In late onset group, PCT was positive in 4 patients out of 14 blood culture positive cases. Among 4 negative patients blood culture positive in 1 case. This study was similar to another study.¹⁸

In this study early onset group was positive in 7 patients out of 9 blood culture positive cases. Out of 13 (41.7%) were CRP positive 2 cases were negative by blood culture. In late onset group, CRP was positive in 5 patients out of 5 blood culture positive cases. Among 10 CRP negative patients, no patients were found to be blood culture positive. This is consistent with another study.¹⁹

In Current study the diagnostic performance of sepsis screen, following observations were noted. PCT are found to be highly sensitive in diagnosis of neonatal sepsis. The sensitivity and specificity were 93%, 18.2% respectively, PPV 26.5% and NPV 88.9%. In comparison of blood culture CRP performance was sensitivity 85.7%, Specificity 65.9%, PPV 44.4% and 93.5%. These findings are similar to another study.²⁰

Specificity of PCT was less in this study because blood culture positivity rate was only around 24%. Quantity of blood taken for conventional blood culture was small as. On the other hand in case of CRP, specificity was more than PCT because sensitivity of CRP was 85.7% and PCT was 92.9% in comparison to blood culture. That's why false positive was more in PCT. Limitations of this study were small sample size. Study population of this study was neonate. Quantity of blood was insufficient for proper

isolation of bacteria. Only 0.5 to 1 ml blood was taken for conventional blood culture because collection of blood was difficult from neonates.

Conclusion

Neonatal septicaemia continues to be a major problem for neonates in neonatal intensive care units around the world. If diagnosed early and treated aggressively it is possible to save most cases of neonatal sepsis. PCT is a sensitive, independent and useful biomarker of neonatal sepsis. Its persistence correlates with the severity of sepsis. Additional measurement of CRP may increase the specificity. Continuous monitoring of PCT level could be suggested for proper management of neonatal sepsis.

Conflicts of interest: None

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Original Article

A Histomorphological Study of Urinary Bladder Biopsies: A Retrospective Study in a Tertiary Care Hospital

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Abstract

Introduction: Wide spectrums of noneoplastic and neoplastic lesions are observed in urinary bladder. Malignancies of the bladder are becoming more common and are regarded as a major contributor to the morbidity and death associated with cancer. Histopathology is still the gold standard study for precise diagnosis and therapy.

Objective: To study the histomorphology of urinary bladder lesions obtained through Trans Urethral Resection of Bladder Tumour (TURBT) and cystoscopic biopsies.

Methods: The present study was a two years retrospective study conducted from July 2021 to June 2023 and data were collected from records carried out in the Department of Pathology and Rajshahi Medical College, Rajshahi. Record of all patients with lower urinary tract symptoms and obstructive bladder symptoms and subjected to cystoscopy and TURBT were included in this study.

Results: The data for the present study was collected over a period of 2 years; during which a total of 70 patients were histopathologically evaluated. The majority of participants fell within the age range of 61-70 years, constituting 35.7% of the total sample. Papillary Urothelial Carcinoma (Low Grade) was the most common lesion category, accounting to 19 cases (27% of the sample as a whole). Subsequently, 14 cases (20%) had Papillary Urothelial Carcinoma (High Grade), whereas 13 cases (18.6%) had Urothelial Dysplasia and 6 cases (8.6%) had Urothelial Carcinoma in Situ. There were fewer incidences of papilloma (10%) and chronic non-specific inflammation (14.3%) respectively.

Conclusion: The prevalence of Papillary Urothelial Carcinoma (Low Grade) emphasizes the significance of early detection and management in this age group. Additionally, the presence of high-grade lesions and precancerous conditions necessitate vigilant monitoring and appropriate interventions.

Keywords: Urinary bladder, histopathology, papillary urothelial carcinoma, biopsy

J Khulna Med Coll. 2024; 3(1) : 26-31

Introduction

Urinary bladder biopsies play a crucial role in diagnosing various pathological conditions affecting the bladder. Histomorphological examination of these biopsies provides valuable insights into the nature, extent, and characteristics of bladder diseases, aiding clinicians in accurate diagnosis and appropriate management.

The most frequent urologic cancer with the highest recurrence rate is bladder cancer. Urothelial carcinoma is the most prevalent form.¹ Adenocarcinomas and squamous cell carcinoma are two other kinds.² Urinary bladder tumors are more common in urban than rural areas and in men compared to women, with a ratio of 3-5:1.³ In affluent countries compared to poor ones, bladder cancers are six

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times more common. This trends suggest a role for industrial substance in development of bladder cancer.⁴ Several risk factors like cigarette smoking, numerous occupations are associated with diesel exhaust, petroleum products, plumbing and schistosoma haematobium infection are associated with increased prevalence of bladder cancer.⁴

Urinary bladder is a sac that holds urine, it is situated in the extraperitoneal area of the pelvis behind the pubic bones.⁵ There are two primary sections of the bladder, each with unique morphological, histological, and functional characteristics.⁶ The most prevalent inflammatory urinary bladder disorder is cystitis, which can manifest in both acute and chronic forms. It is further divided into different categories based on the origin, length of time, and histological appearance.⁷ Data from the radiology, urologist, and surgical pathology labs must be combined in order to accurately diagnose urinary bladder lesions.⁸

Cystoscopy enables direct visualization of the bladder mucosa and biopsies of the suspected lesions. It is the primary diagnostic method for individuals who are suspected of having bladder tumors.⁹ Tumor differentiation, depth of infiltration, and total removal of non-invasive bladder papillary tumors are all assessed by transurethral resection of bladder tumors (TURBT), a therapeutic and diagnostic method.¹⁰

The condition known as urinary bladder cancer is diverse and multifaceted, exhibiting a wide range of histological characteristics and possibly fatal symptoms. Patients diagnosed with muscle-invasive carcinoma continue to face disease progression, recurrence, and, ultimately, death despite advancements in surgical procedures and intravesical and systemic therapy.¹¹

For a definitive diagnosis, histomorphological study is mandatory.¹² The role of the pathologist is not just limited to giving a diagnostic label, but also to giving additional information that can have an impact on the treatment.¹³

This histomorphological study of urinary bladder biopsies in a Tertiary Care Hospital seeks to contribute to the existing body of knowledge, offering a comprehensive understanding of the histopathological spectrum of bladder disorders and informing clinical practice for improved patient care.

Materials and Methods

This cross sectional retrospective study was conducted in the Department of Pathology of Rajshahi Medical Collage, Rajshahi over two years beginning July 2021 to June 2023, following the acquisition of institutional ethical clearance. The study comprised 70 cases in total.

Inclusion and exclusion criteria: There was a record of every patient who visit to the Department of Urology with symptoms related to the lower urinary tract and obstructive urinary symptoms and subjected to TURBT and cystoscopy biopsy were included in this study the biopsy after taken by urologist sent to the Department of Pathology.

Biopsy specimens were fixed in 10% formalin for 24 hours before the tissue was processed for paraffin blocks. Five micron sections were cuts and the prepared slides were stained with Hematoxylin and Eosin (H&E) stain. The Histopathological features were studied relevant finding were noted. The lesions were categorized into papillary urothelial carcinoma (low grade), papillary urothelial carcinoma (high grade), urothelial carcinoma in situ, urothelial dysplasia, papilloma and chronic non-specific inflammation.

Excel sheets with data were filled up and examined. Percentages were used to summarize categorical variables.

Results

A total of 70 bladder biopsy cases were studied, age range of the cases in the study is from 31-80 years with most common age group of 61-70 years and sex ratio of female to male was 1:1.18.

Table I: Distribution of Participants by Age Group (n=70)

Age group	Frequency (%)
31-40	2 (2.9)
41-50	12 (17.1)
51-60	16 (22.9)
61-70	25 (35.7)
71-80	15 (21.4)

The majority of participants fell within the age range of 61-70 years, constituting 35.7% of the total sample.

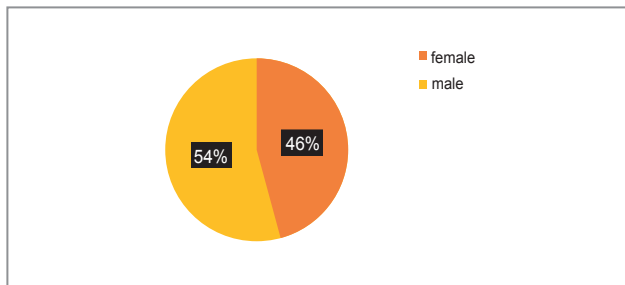


Figure 1: Pie chart according to sex

The study included 32 female (45.7%) and 38 male (54.3%) participants, indicating a relatively balanced representation of both genders in the sample.

Table II: A Histopathological overview of benign and malignant lesion according to sex

Gender	Inflammatory and Benign lesions (23)			Malignant lesions (47)		
	Chronic Non-specific Inflammation	Urothelial Dysplasia	Urothelial Papilloma	Urothelial Carcinoma In Situ	Papillary Urothelial Carcinoma (Low Grade)	Papillary Urothelial Carcinoma (High Grade)
Male	6 (35%)	4 (8%)	4 (23%)	5 (9%)	11 (21%)	7 (13%)
Female	4 (24%)	2 (4%)	3 (18%)	7 (13%)	8 (15%)	9 (17%)

Table II presents a breakdown of histological findings categorized by gender and pathology type, distinguishing between benign and malignant conditions. In total, there were 23 cases of inflammatory and benign findings and 47 cases of malignant findings. Among males, the most prevalent malignant condition was Papillary Urothelial Carcinoma (High Grade), comprising 21% of cases, followed by Chronic Non-specific Inflammation and Papillary Urothelial Carcinoma (Low Grade). In females, the distribution was somewhat similar, with Papillary Urothelial Carcinoma (High Grade) also being the most frequent malignant condition at 15%, followed by Urothelial Carcinoma in Situ and Chronic Non-specific Inflammation. Notably, Urothelial Dysplasia was less common in both genders. These findings underscore the importance of gender-specific considerations in the evaluation and management of urothelial pathology, while also highlighting the prevalence of malignant conditions, particularly high-grade carcinomas, warranting vigilant monitoring and appropriate intervention strategies.

Table III: Age distribution according to type of lesion

Lesions	Age group (Years)						Total
	31-40	41-50	51-60	61-70	71-80	n	
Urothelial Papilloma	1	1	-	4	1	7	10%
Chronic Non-specific Inflammation	-	2	2	5	1	10	14%
Papillary Urothelial Carcinoma (Low Grade)	1	4	5	6	3	19	27%
Papillary Urothelial Carcinoma (High Grade)	-	2	4	6	4	16	23%
Urothelial Carcinoma In Situ	-	2	3	3	4	12	17%
Urothelial Dysplasia	-	1	2	1	2	6	9%
Total	2	12	16	25	15	70	100.0%

Papillary urothelial carcinoma, both low and high grade, shows a tendency to increase with age, peaking in the 61-70 age range. Conversely, urothelial papilloma and chronic non-specific inflammation appear more evenly distributed across age groups. Urothelial dysplasia displays a slight increase in the older age groups. Urothelial carcinoma in situ shows a relatively consistent occurrence across age categories. Overall, the data suggests age-related trends in the prevalence of various urothelial lesions, with carcinomas being more prevalent in older individuals (Table III).

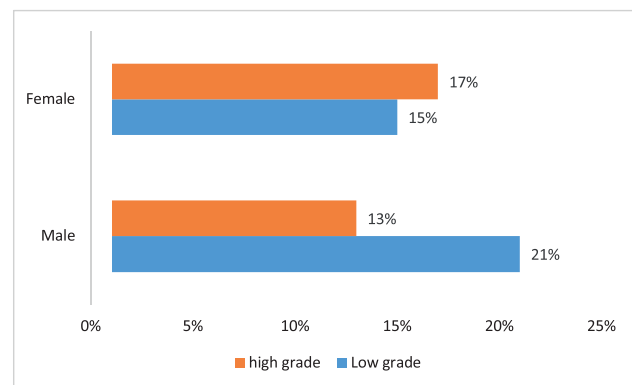


Figure 2: Bar chart according to grading of malignant tumor



Figure 3: Gross photograph showing multiple grayish white fragmented pieces of tissue about 8 ml in volume (TURBT biopsy-case 04)

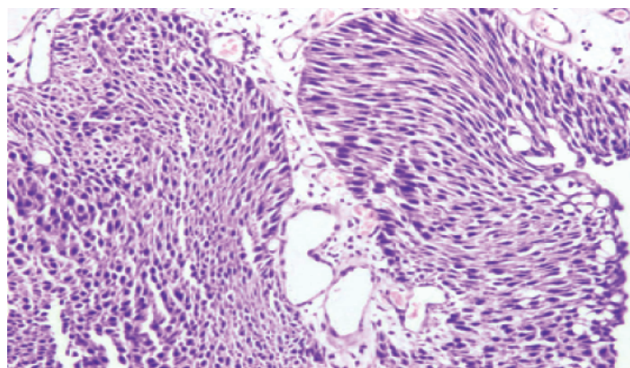


Figure 4: Photomicrograph showing a case of inverted urothelial papilloma in which normal urothelial cells are arranged in trabeculae, Tumor cells have no mitotic activity (case-4, H & E stain, 400x)

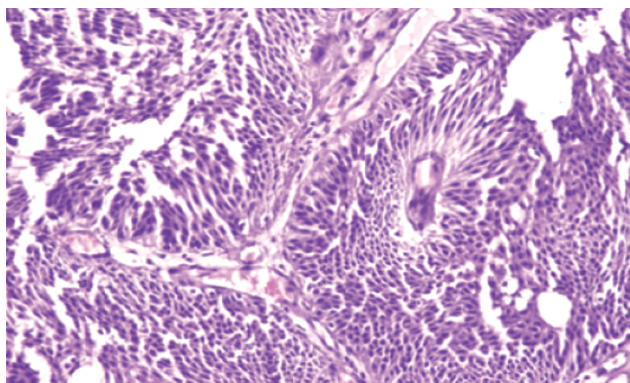


Figure 5: Photomicrograph showing a low grade papillary urothelial carcinoma in which neoplastic proliferation of urothelium in papillary configuration invading the lamina propria with no invasion of the basement membrane (case 18, H & E stain, 400x)

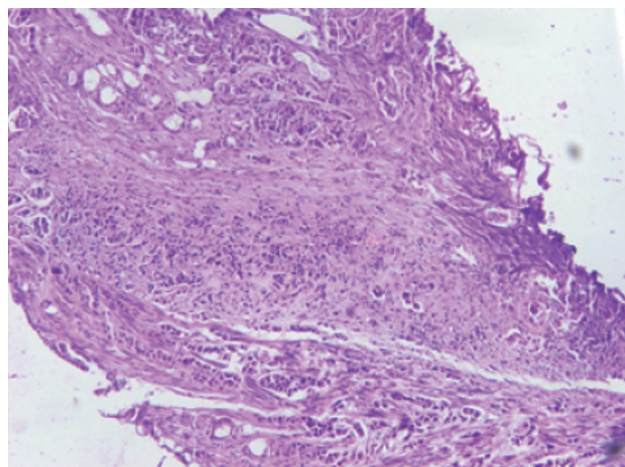


Figure 6: Photomicrograph showing a high grade papillary urothelial carcinoma in which anaplastic urothelial cells infiltrating up to muscle layer (case 24, H&E stain, 400x)

Discussion

A total of 70 urinary bladder biopsy specimens were evaluated in our study. The common age group affected according to our study was between 61-70 years constituting 57.81% of the cases and was in concordance with Vaidya S et al. (2013) and Matalka I et al. (2008) study.¹³⁻¹⁴ The older age can play as a risk factor in the development of bladder cancer due to loss of elasticity and exposure to numerous carcinogens. In our study 54.3% participants are male and 45.7% are female. Our results is consistent with the study of Shah PY et al. (2016) and Srikousthubha et al. (2013).¹⁵⁻¹⁶

The histomorphological diagnosis of urothelial lesions, as presented in our study, reveals a spectrum of pathological entities encountered in urinary bladder specimens. Notably, Papillary Urothelial Carcinoma (Low Grade) constituted 29% of the cases and Papillary Urothelial Carcinoma (High Grade) at 20%, corroborating with prior studies that highlight the aggressive nature and potential for progression associated with high-grade lesions. A 21 cases (14.58%) invasive papillary urothelial carcinomas showed squamous differentiation and 1 case (0.69%) with glandular differentiation which was similar to studies done by Shruthi HP et al. (2015) and Goyal VK et al. (2015).¹⁷⁻¹⁸

In our study, the identification of Urothelial Carcinoma in Situ in 18% of cases underscores the importance of recognizing early-stage malignancies. Among different malignant lesions, urothelial carcinomas were followed by 2 cases each of (0.29%) urachal carcinoma and neuroendocrine tumours and 1 case (0.14%) of pure squamous cell carcinoma which is in accordance with Goyal VK et al., (93%) and discordance with Muhammed M et al. (2014) and Mahesh K et al. (2012).¹⁸⁻²⁰

The presence of Urothelial Dysplasia in 9% of cases raises interesting considerations, as this entity is recognized as a potential precursor to malignancy which is similar to the study of Ranadip et al.(1983).²¹ Papillomas, comprising 10% of cases, exhibit a lower prevalence compared to the malignant counterparts consistent with the study of Vaidya S et al. (2013). Chronic Non-specific Inflammation has a direct effect on bladder function. In this study Chronic Non-specific Inflammation is identified in 14% of cases, which is accordance with the study of Srikousthubha et al. (2013). In the present study, authors concentrated mainly on the histomorphological features and hence could not contribute much to the natural history and pathogenesis of the disease in the patients.

Conclusion

In this study, 70 urinary bladder biopsies were studied. There were Neoplastic lesions are more common. Both low-grade and high-grade papillary urothelial carcinomas exhibit an increasing trend with age. Urothelial dysplasia and urothelial papilloma, are a precursor to malignancy, demonstrate same pattern with a notable prevalence in the 51-60 and 61-70 age groups. Histopathological evaluations of bladder lesions are crucial to provide guidance for early detection of disease and proper treatment.

Conflicts of interest: None

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Case Report

A Case Report of Mixed Connective Tissue Disease with Pulmonary Hypertension

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Abstract

Introduction: Mixed connective tissue disease (MCTD) is a multisystem autoimmune disease that was rarely reported in Bangladesh. It is a disease of overlapping features of systemic sclerosis, myositis & systemic lupus erythematosus (SLE), all occur in the same patient. It commonly presents with indolent puffiness of the fingers with raynaud's phenomenon and myalgia. Most of the patient have anti-U1RNP antibody. We present a case of 20 years female who was admitted into medicine ward of Khulna Medical College Hospital, Khulna. She came with initial complaint of low grade intermittent fever since four years; exertional dyspnoea which is progressive and limiting her daily activities, occasional palpitations, gradual tightening of skin at face & extremities, painful bluish discolouration of fingers & toes on cold exposure, symmetrical small joint arthritis since two years; weight loss, anorexia and constipation since eight months as well as infertility since 1 ½ (one and half) years. Detailed history & physical examination revealed bluish discolouration of fingers on exposure to low temperature, difficulty in opening the mouth, sclerodactyly and proximal myopathy. Laboratory work up and radiography concluded the diagnosis of Mixed connective tissue disease. Pulmonary function test suggested restrictive lung disease and 2D & M-mode Echocardiography showed pulmonary arterial hypertension (PASP -57mmHg), tricuspid regurgitation with dilatation of pulmonary artery & right ventricle. Our patient followed both the Alarcon-Segovia criteria and Kasukawa diagnostic criteria for mixed connective tissue disease.

Keywords: Mixed connective tissue disease, systemic sclerosis, myositis, systemic lupus erythematosus, Anti-U1RNP antibodies.

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Introduction

Mixed connective tissue disease is an autoimmune disease which was first described in 1972 by Sharp et al. as a disease syndrome with overlapping features of Systemic sclerosis, Systemic lupus erythematosus and Polymyositis.¹

Therefore, mixed connective tissue disease(MCTD) is sometimes referred to as an overlap disease- the initial presentation of the patients usually comprises non-specific

signs such as swollen digits, arthralgia, myalgia or muscle weakness, acid reflux or dysphagia, Raynaud's phenomenon, shortness of breath on activity, general malaise, fatigue & weight loss. Over a period of time the symptoms are dominated by symptoms of either of one of the three illnesses along with high titre of anti-U1RNP antibody.

The aetiology of the MCTD is unknown but being an autoimmune disease MCTD can run within the families

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and is known to affect women more than men. No causal associations and the varied presentations make the diagnosis of this rare condition difficult.

We encountered one such case with low grade intermittent fever, gradual tightening of skin at face & extremities, Raynaud's phenomenon, symmetrical small joint arthritis and weight loss as the initial presentation that eventually lead to the diagnosis of MCTD with restrictive lung disease and pulmonary arterial hypertension.

Case report

A 20 years old female was admitted into KMCH with the complains of occasional fever since last 04 years, fever was low grade intermittent in nature, there is no chills & rigor, responded to anti-pyretics since 02 years. There is skin tightening of face & extremities, symmetrical inflammatory small joint pain of hands with feet as well as exertional dyspnoea and day by day it worsened progressively thereby limiting her daily activities. On enquiry, she gave history of painful bluish discolouration of fingers & toes while washing hands and feet with water or in cold temperature since last two years (Figure-1). Difficulty in opening mouth for last one year and dyspeptic symptoms along with weight loss, constipation and generalized weakness with myalgia since eight months. There is secondary infertility since one and half years.

On examination, there was mild pallor, mild pedal edema and temperature of 99°F, pulse 110 beats/min with regular rhythm, Blood pressure 110/70 mmHg, Respiratory rate 22 breaths/min and JVP was raised. Facial skin was smooth, shiny, taut, immobile with pinched up nose and hyperpigmented area around eyes & at tip of nose. There is little puckering of skin around the lips with small mouth orifice and tightening of skin around the mouth with difficulty in opening.

Musculoskeletal system examination revealed that skin of fingers and toes was smooth, shiny, taut, immobile with peripheral cyanosis & sclerodactyly. There are hyperpigmented area around elbows & knees, swelling of fingers and toes with G-2 tenderness of PIP, DIP, MCP, MTP, IP joints and active-passive movement are slightly restricted.

Physical examination of precordium revealed left parasternal heave, palpable P2 and loud S2 and pansystolic murmur at tricuspid area with no radiation, accentuates during holding breath at the end of inspiration.

Respiratory system exam. revealed sign of bilateral pleural effusion at lower region of chest and there was no other abnormalities.

However, Per abdominal and CNS examination provided no physical findings.

Investigating Haemoglobin level was found 10.0 g/dl, ESR- 65 mm in 1st hour, TLC-11800/mm³, Differential count- Neutrophil-72%, Lymphocytes: 25%, Eosinophils -2%, Monocytes: 1%, Basophil-0%, CRP- 12mg/l (N-6mg/l), Platelet count -254000/mm³ of blood, RBS- 5.2 mmol/l, Serum TSH-2.38 μ IU/ml, USG of abdomen revealed normal study, chest X-ray showed bilateral pleural effusion(Fig.-2), ECG showed sinus tachycardia, Echocardiography showed dilated RA, RV, PA with TR along with PASP of 57mmHg and EF-62%. Urine analysis(Routine and microscopy) was normal, renal and liver function tests are normal and serum electrolytes level are within normal limit. However, Serum CPK level was raised(842 IU/L), pulmonary function test revealed restrictive lung disease, endoscopy of upper GIT was normal.

Our patient followed the Alarcon-Segovia diagnostic criteria (Table-I) with positive serology and three of the five clinical criteria's namely Raynaud's phenomenon, swelling of hands & feet and myositis depicted by high serum CPK value.

Our Patient also followed the Kasukawa diagnostic criteria (Table-II) with both the common symptoms of Raynaud's phenomenon, generalized weakness, myalgia, swollen and thickened hands and feet being present associated with positive serology and mixed findings of sclerodactyly, proximal myopathy & high CPK.

Serological examination showed anti-RNP Ab, Anti-Sm Ab, Anti-Scl Ab-70, ANA are positive.

However, Anti-Ro Ab, Anti-La Ab, Anti-Jo-1 Ab, Anti-dsDNA are negative. (Table-III)

A diagnosis of Mixed connective tissue disease with pulmonary hypertension was made.

The management of the patient included:

- Cefuroxime+ clavulenic acid 250mg,PO, bid
- Naproxen sodium 250 mg, PO, bid
- Esomeprazole 20mg,PO,A/C, bid
- Diltiazem 6.25 mg,PO, ½ -0- ½
- Hydroxychloroquine (HCQ) 250mg bid
- Furosemide+ Spironolactone 40/50mg,PO, bid
- Calcium+ Vitamin-D, PO, 0-1-0
- Prednisolone 40mg,PO tapered every two weeks to 35mg,30mg,25mg,20mg,15mg,10mg,5mg ; 1-0-0
- Sildenafil citrate 25mg ,PO,OD
- Supplemental oxygen was given for dyspnoea.

The patient was discharged after 21st days of hospitalization and is currently on Sildenafil citrate , HCQ , Furosemide+ Spironolactone , Diltiazem , Prednisolone , Calcium+ Vitamin-D and Esomeprazole.

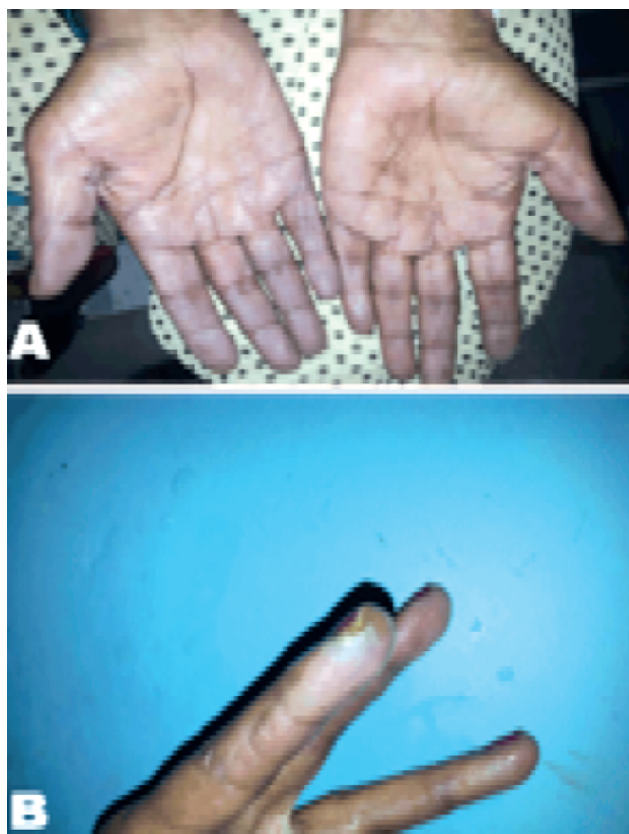


Figure 1: Bluish discolouration of fingers on exposure to cold A

Hand of the patient: Thickened fingers B
(Alarcon: Segovia, 1989)⁶



Figure 2: Chest X-ray P/A view: bilateral pleural effusion

Table I: Alarcon-Segovia diagnostic criteria for mixed connective tissue disease

Serologic Criteria :	Positive anti-U1RNP at hemagglutination titer >1:1600
Clinical Criteria :	Edema of hands Synovitis Myositis Raynaud's Acrosclerosis

Table II: Kasukawa diagnostic criterion for mixed connective tissue disease

1) Common Symptoms	Raynaud's Phenomenon Swollen fingers or hands
2) Presence of Anti U1 RNP	
3) Mixed findings	
A. Systemic lupus erythematosus (SLE) like	Polyarthritis Pericarditis/pleuritis Lymphadenopathy Facial erythema Leucopenia/thrombocytopenia
B. Scleroderma like	Sclerodactyly Pulmonary fibrosis Esophageal dysmotility
C. Polymyositis like	Muscle weakness High creatine phosphokinase (CPK) Myopathic electromyogram (EMG)
Requirement for diagnosis: At least one common symptom with positive UIRNP antibodies and one or more findings in at least two of the three categories A, B, and C.	

Table-III: Serological markers for autoantibodies

ENA PROFILE			
Autoantibody	Results	Interpretation	Reference range
Anti- RNP antibody (IgG)	>200 U/ml	Positive	Normal : < 15.00 U/ml Borderline : 15.00 – 25.00 U/ml Elevated : > 25.00 U/ml
Anti- S _m antibody (IgG)	81.00 U/ml	Positive	Normal : < 15.00 U/ml Borderline : 15.00 – 25.00 U/ml Elevated : > 25.00 U/ml
Anti- SS-B antibody/L _a antibody (IgG)	3.90 U/ml	Negative	Normal : < 15.00 U/ml Borderline : 15.00 – 25.00 U/ml Elevated : > 25.00 U/ml
Anti-Scl-70 antibody(IgG)	>200 U/ml	Positive	Normal : < 15.00 U/ml Borderline : 15.00 – 25.00 U/ml Elevated : > 25.00 U/ml
Anti- Jo-1 antibody(IgG)	7.60 U/ml	Negative	Normal : < 15.00 U/ml Borderline : 15.00 – 25.00 U/ml Elevated : > 25.00 U/ml
Anti-nuclear antibody Test			
ANA	11.60	Negative	Negative index: <1.00 Borderline index : 1.00- 1.20 Positive index : >1.20 Method : ELISA
Anti-double stranded DNA antibody Test			
Anti-dsDNA (IgG)	10.90 IU/ml	Negative	Negative: < 20.00 IU/ml Equivocal : 20.00 – 25.00 IU/ml Positive : > 25.00 IU/ml

Discussion

The initial history of low grade intermittent fever, Raynaud's phenomenon, skin tightening of face & extremities, symmetrical inflammatory small joint pain of hands with feet as well as exertional dyspnoea and generalized weakness. Physical examination findings of sclerodactyly and microstomia guided our laboratory and radiological workup & thereafter helped us to reach the diagnosis.

A MCTD is a rare disease with unknown etiology but few cases have been reported after occupational handling of Vinyl chloride.² and even after breast augmentation surgery.³ There are familial cases of MCTD with

increased instance of HLADR4 compared with controls. A 2011 Norwegian study showed a 3.8 per 100000 prevalence of MCTD among adults with an incidence of 2.1 per million per year.⁴

Mean age of onset was 31.9 years and more than three quarters of patients were females.⁵ This confirms the rarity of the disease and a predilection for female sex.

MCTD should be kept as a differential diagnosis when overlapping signs of autoimmune diseases are present such as swollen digits, arthralgia, myalgia, or muscle weakness, acid reflux or dysphagia, Raynaud's phenomenon, shortness of breath on activity, generalized malaise and fatigue.

Many criteria have been described to classify MCTD. Table-1 gives the Alarcon-Segovia's criteria and Table-2 gives the Kasukawa diagnostic criteria for MCTD. Alarcon-Segovia's criteria are simple & comprises five clinical manifestations in addition to the serological status.⁶

Our patient showed the Alarcon-Segovia's criteria with positive serology & three of five clinical criteria namely Raynaud's phenomenon, Swelling of hands & feet and myositis depicted by high serum CPK value of 842 IU/L. Our Patient also followed the Kasukawa diagnostic criteria (Table II) with both the common symptoms of Raynaud's, generalized weakness, myalgia, swollen and thickened hands and feet being present associated with positive serology and mixed findings of sclerodactyly, proximal myopathy & high CPK.

Myositis clinically presents as muscle weakness is more common than laboratory increase in muscle enzymes but in our patient a frank increase in CPK was seen. Lung function test specially the single breath diffusing capacity may be abnormal in many patients but only a small fraction of these are symptomatic. Meanwhile our patient had dyspnoea as the presenting complaint. Pulmonary arterial hypertension (PAH) is not common in early MCTD.⁷ PAH occurs secondary to ILD is an increasingly recognized complication and our patient was suffering from PAH as diagnosed by 2D & M-mode Echocardiography.

The therapy for MCTD would be combination of many drugs to suppress inflammation including NSAIDs like Naproxen, COX-2 inhibitors like coxib, steroids such as prednisolone, anti-malarials (HCQ) & immunosuppressants like Azathioprine. Nifedipine, Nitroglycerine, Losartan are used for Raynaud's phenomenon. TNF blockers like Etanercept, Infliximab, Adalimumab are used in inflammatory arthritis in some cases. So, two people with the same disease but different presentation would require an altogether different management.⁸

The use of therapeutic antibodies like Rituximab (anti-CD-20 antibody) in MCTD have shown benefit in cases of refractory polymyositis & lower CPK.⁹

Membranous GN and mesangial proliferative GN are main renal manifestations. Less common renal manifestation includes diffuse proliferative GN, Vascular or glomerular sclerosis. It is clinically manifested mainly with proteinuria, rarely with haematuria.⁸ Our patient had no haematuria on urine examination.

A case study showed that a patient of MCTD has distinctive dermatological manifestations of dispersed raised red scaling plaques with central atrophy on the extremities, some of them having resolved -causing hyperpigmentation, atrophy and scarring. Moreover; friable nails, brittle hair, clubbing and patches with excessive hair on the forearms and face were also seen. Intraoral findings include; fibrosis of the tongue with reduced mobility, loss of papillae, generalized pallor and blanching of the mucous membrane, and diffuse fibrosis of the buccal mucosa were also seen.¹¹ Our patient had no such dermatological and intra-oral involvement.

Our patient had developed secondary infertility during the course of her illness. Patient of MCTD may suffer from infertility and it is rarely reported in the published articles.

The absence of severe renal disease is a hallmark of MCTD; it is possible that high titers of anti-RNP

antibodies, which are characteristic of MCTD, may protect against the development of diffuse proliferative glomerulonephritis. But patients may have glomerulonephritis, nephritic syndrome or renal crisis of scleroderma.¹² Our patient had none of these manifestations.

Neurological manifestations are less common in MCTD, presenting as aseptic meningitis, trigeminal neuralgia, demyelination, transverse myelitis and peripheral neuropathy.^{12,14} Our patient had none of these manifestations.

Our patient developed characteristic gastrointestinal symptoms ranging from dyspepsia, heartburn, constipation and weight loss but disordered motility in the upper gastrointestinal tract, symptoms of malabsorption which are the commonest problems were absent.^{12, 13}

Pleuropulmonary involvement is common, it may be asymptomatic or the patient may present with pleurisy and effusion, interstitial lung disease, pulmonary arterial hypertension.

tension (PAH)^{12, 16}. Our patient had pulmonary arterial hypertension and pleural effusion but there was no clinical evidence of pleurisy and interstitial lung disease.

All three layers of the heart may be involved in MCTD with abnormal ECG findings.¹⁵ Our patient had none of this except sinus tachycardia.

In the early phases of the MCTD, many patients complain of easy fatigability, poorly defined myalgias, arthralgias and Raynaud's phenomenon. Systemic involvements such as: fever of unknown origin may be the presenting feature of MCTD. The most common skin change is the Raynaud's phenomenon (75%-100% of patients). Approximately 60 percent of patients develop an obvious arthritis. Myalgias and myositis can be seen in 30%-50% of patients. Others are malar rash, generalized erythematous rash.^{12,17} Our patient had all of these manifestations except malar rash, generalized erythematous rash.

No controlled trials have been performed to guide therapy. Instead, the management of patients with MCTD generally rests upon the effectiveness of specific therapies for similar problems seen in SLE, scleroderma, or polymyositis. By comparison, scleroderma-like features (eg, Raynaud phenomenon, pulmonary hypertension) are usually less responsive to therapy. NSAIDs are given for pain, calcium channel blockers for PAH, Immunosuppression (steroid and other agents) is particularly required in cases with severe arthritis, pulmonary hypertension and serositis involving pericardium or pleura.^{12,18-19}

Although MCTDs can generally be clinically and serologically defined as distinct and separate entities, many patients diagnosed with autoimmune rheumatic disease cannot be categorized easily into one of the established conditions.^{11, 20}

MCTD has relatively good prognosis in view of low prevalence of serious renal disease and life-threatening

neurologic problems. Morbidity is quite high in patients with MCTD due to multiple factors including recurrent musculoskeletal pain, fibromyalgia, gastroesophageal acid reflux etc. Mortality associated with MCTD ranges, in different studies, from 16 to 28 percent at 10 to 12 years.¹⁸

Those patients with more features of scleroderma and polymyositis had a worse prognosis. The major causes of death include progressive pulmonary hypertension and its cardiac complications.¹⁶

Conclusion

Pulmonary arterial hypertension presents late in the illness when other clinical signs are easily recognizable and should be treated aggressively as most death in Mixed connective tissue disease are due to heart failure caused by pulmonary arterial hypertension.

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- Standard abbreviation should be used. The full form for which an abbreviation stands should precede its first use in the text.

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A) **Original Articles** (length: 2000±500 words)

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A structured abstract should include headings such as Background, Objective, Methods, Results, and Conclusions. Title be added at the top and Key words (<5) at the bottom of the abstract.

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In this section study type, place and time, description of study subjects and grouping, sampling, selection criteria, approval of the study involving human subjects by ethical review committee and description of the ethical aspects in such study, description of study variable, description of procedure, methods, apparatus, drugs or chemicals as applicable should be given. Description of statistical procedure for obtaining all P values should be provided to allow verification. Statistical software, if used should be specified.

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A brief concluding paragraph presenting the implications of the study results and possible new research directions on the subject.

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