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Editorial

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Contents

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EDITORIAL

Neonatal Jaundice - Do We Address It Appropriately ?

MAK Azad Chowdhury

Introduction

Major causes of neonatal mortality and morbidity in Bangladesh are preterm with its complications, perinatal asphyxia and septicemia.

Besides these, neonatal jaundice is also important as about 80% of term infants do have it in their neonatal period. The majority of neonatal jaundice is benign, need no intervention. But parental anxiety needs to be addressed. Few cases may be fatal causing death or long term neurological deficit. So the importance and impact of neonatal jaundice on heath system is more than its apparently benign nature.

As blood grouping is not presently done at the routine antenatal check-up, identification of ABO, Rh incompatibility, sometimes causing severe jaundice, cannot be anticipated and taken care of. Very clear idea about distinguishing physiological or benign jaundice from pathological or harmful jaundice is needed. Failure of which may result in undue prolonged morbidity, mortality and delayed referral. Careful history taking, physical examination and simple tests, can solve this problem.

Physiological Jaundice

In utero the foetus has to derive its total nutrition and oxygen from maternal blood perfusing the placental villae This blood is about 60-70% saturated with oxygen, that is it contains less oxygen than that we inhale in our lungs. So to maintain proper oxygenation of the very rapidly growing tissues of the foetus the foetal blood has special characteristics to extract as much as possible oxygen from the maternal blood.

Foetal haemoglobin is different from adult haemoglobin and has higher affinity for oxygen and also the amount of haemoglobin in the foetus is about 60% more (18-20 gms /dl of blood) than in adult. After birth, when the newborn starts breathing and receives oxygen directly from the air through his/her lungs, he/she no more requires those especially designed and excess haemoglobin to maintain oxygenation and metabolism of his/her tissues. At this time the redundant haemoglobin tends to rapidly breakdown and be cleared from the circulation. The life span of foetal RBC is also less (80-90 days) than that of adult RBC so they breakdown rapidly and in large amounts. During this period the bone marrow finding no necessity to produce much RBC, takes some rest and functions minimally for several weeks. The breakdown of this excess red cells & their haemoglobin produces excessive amounts of bilirubin pigments (35 mg of bilirubin is liberated from 1 gm of haemoglobin) and is presented to the yet to fully mature liver (structurally and enzymatically) to handle this bilirubin load for conjugation and excretion. As the load of bilirubin gradually increases to beyond capacity of the liver to conjugate, clinically apparent jaundice appears. In a normal full term baby by day 7 to 10 there is usually balance between production and excretion of bilirubin and the jaundice gradually disappears.

In a pre-term baby this process of maturity takes some longer time and the jaundice usually disappears between 15-20 days of life unless some other complication arises.

So, Physiological Jaundice will have clinically be evident after 2-3 days of birth, gradually increases for 4-5 days, decline spontaneously after one week of age and be almost unnoticeable after 2 weeks of age. Stool colour of the baby will be normal (yellowish), the baby's activity including sucking, swallowing will remain normal.

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Pathological Jaundice

About ten percent of the clinically apparent neonatal jaundice may be outside the physiological domain and require investigations and active management. The principal reasons for this abnormal neonatal jaundice are: Excessive haemolysis, eg. Blood group incompatibility (Rh , ABO and minor blood group incompatibility), enzyme deficiencies, eg. G-6-PD deficiencyand pyruvate kinase deficiency, Haemoglobinopathis, eg. ±-thalassaemia, from sequestrated blood, eg. cephalhaematoma, multiple bruises, septicaemia, impaired enterohepatic circulation, e.g.. Intestinal obstruction, hypertrophic pyloric stenosis, obstruction to biliary flow and Immaturity or congenital absence of hepatic enzymes. Jaundice appearing within day one or two of birth, Increases very rapidly (>0.5mg/kg/hour), the Jaundice appears to be very high at any time (the lower limbs and soles being prominently yellow) , the jaundice don't start decreasing clinically after 7-10 days of age, the baby is sick otherwise with less activity, poor feeding, fever, vomiting, unhealthy umbilicus etc.The stool color of the baby is pale. Before sending blood for investigations the baby should be very carefully examined for signs of infection, injury, haematoma, hepatosplenomagaly, rashes, etc.

The simple investigations will give clue to the diagnosis in more than ninety percent cases of neonatal jaundice:cMother's Blood group, Baby's Blood group, Hb%, Direct Coomb's tes Peripheral blood film, Reticulocyte count. S. bilirubin-Total (Direct and indirect). If the direct bilirubin is predominantly high (>20% 0f total) the following possibilities need to be considered: Sepsis, Congenital metabolic problem like galactosaemia, Neonatal Hepatitis syndrome, Biliary atresia, Rubella, Toxoplasma, CMV infection, antitrypsin deficiency Choledochal cyst etc.

If the indirect or unconjugated bilirubin is predominantly high the following flow chart will help in reaching diagnosis. After the initial tests further investigations should be done depending on the possibility e.g-for suspected biliary atresia ultrasonogram of hepatobiliary system and HIDA scan, for congenital infection- TORCH screening etc. If there is strong suspicion of biliary atresia on the basis of high direct bilirubinaemia, pale stools, the baby should urgently be referred to a place where surgical intervention is possible. Because early diagnosis and surgery has high degree of success and good prognosis and delay of only six to eight weeks will cause irreversible damage to the liver

Clinical Assessment

So, whenever a neonate is brought with jaundice we should carefully take the history including the day it was first noticed, other associated features, mother's blood group, and very careful examination of the baby from head to toe. Clinical Assessment of jaundice should made in open day light. Gentle pressure over the skin of face, trunk, limbs will give idea about the degree of jaundice. If the jaundice is mainly limited to the face, chest and abdomen- it is likely to be mild to moderate (bilirubin usually bellow 10-12 mgm/dl). If the lower limbs soles and palms are yellow prominently it is usually moderate to high jaundice (bilirubin usually above 15mgm/dl). Our clinical experience correlates with this observation. So at places where serum bilirubin estimation is not easily available, this guideline may be used to decide whether a neonate with jaundice should be referred to a centre where proper investigations and management are available.

Breastfeeding jaundice

Jaundice can occur when a breastfeeding baby is not getting enough breast milk because of difficulty with breastfeeding or because the mother's milk is not in yet. This is not caused by a problem with the breast milk itself, but by the baby not getting enough to drink.

Breast Milk Jaundice

Some mother's milk contain excessive amount of a progestational hormone (ß -pregnendiole) which competes with bilirubin in the hepatocellular conjugation process, so indirect bilirubin remains high for prolonged period. The baby is otherwise healthy, sucks well and grows well. Blood investigations will show no abnormality. If one highly suspects breast milk jaundice and wishes to confirm it for parents satisfaction, estimation of S. bilirubin before and after temporary

withholding of breast milk for about 24 hours will show a significant decline in bilirubin level..

Management

Physiological jaundice-requires reassurance to parents, encouragement for frequent breast feeding to avoid dehydration. Early morning sun-bath for 15-30 minutes will accelerate the disappearance of jaundice and parents may like to do this but is not essential. In pathological jaundice treatment is according to causes. Sometimes needs phototherapy and exchange transfusion. Besides some drugs, IVIG can decrease the degree of jaundice.

Management of breast milk jaundice is only explanation and reassurance to parents. Breast feeding should never be stopped for this. Explanation of the disease to the parents is very very important and should never be ignored.

So, by doing antenatal test of ABO, Rh (D) blood types of mother and appropriate follow up in case of presence of risk factors, giving maternal anti-D, increasing facility in peripheral center and simple but stepwise judicious approach to a case of neonatal jaundice for its proper and effective management should be the proper way of addressing this disease.

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SPECIAL ARTICLE

Methotrexate - The Gold Standard Drug in Pediatric Rheumatology

Probir Kumar Sarkar

Introduction

Methotrexate (MTX), formerly known as amethopterin, is a chemotherapy agent and immune system suppressant. It is used to treat cancer, autoimmune diseases, ectopic pregnancy, and for medical abortions. Low-dose weekly MTX has emerged as one of the most useful agents in the treatment of rheumatic diseases in children, and it has become the first-choice second-line agent in childhood arthritis, and in some cases arguably a first-line agent. It is also used in many other chronic inflammatory disorders.[1]

Methotrexate was made in 1947 and initially came into medical use to treat cancer, as it was less toxic than the current treatments. In 1956 it provided the first cures of a metastatic cancer. It is on the World Health Organization's List of Essential Medicines, the most effective and safe medicines needed in a health system.[2] Methotrexate is available as a generic medication. Nowadays MTX, an antimetabolite, is a cornerstone of therapy in pediatric rheumatology.[3]

During the past few years, remarkable advances in the treatment of rheumatic diseases have been made with the advent of new disease modifying anti-rheumatic drugs (DMARDs). They work to decrease pain and inflammation, reduce or prevent joint damage and preserve the structure and function of the joints.[4]Though traditional DMARDs exert their beneficial effects a few weeks after initiation of therapy, there are clinical experiences and data from different studies, suggesting that early use of DMARDs may induce long term disease suppression. So the concept of a therapeutic pyramid, starting with nonsteroidal anti-inflammatory drugs (NSAIDs) with gradual addition of more active drugs have been reversed.[5,]

Commonly used DMARDs are methotrexate, hydroxychloroquine, leflunomide, and sulfasalazine. Of them MTX has been recognized as the most effective disease modifying anti-rheumatic drug.

Mechanism of action of MTX

Methotrexate is an antimetabolite of the antifolate type. It is thought to affect cancer and rheumatoid arthritis by two different pathways. For cancer, methotrexate competitively inhibits dihydrofolate reductase (DHFR), an enzyme that participates in the tetrahydrofolate synthesis. The affinity of methotrexate for DHFR is about 1000-fold that of folate. Folic acid is needed for the de novo synthesis of the nucleoside thymidine, required for DNA synthesis. Also, folate is essential for purine and pyrimidine base biosynthesis, so synthesis will be inhibited. Methotrexate, therefore, inhibits the synthesis of DNA, RNA, thymidylates, and proteins.[6, 7]

MTX polyglutamates, also interfere with de novo purine biosynthesis by inhibition of an enzyme in the purine biosynthetic pathway 5-aminoimidazole 4-carboxamide ribonucleotide (AICAR) trans-formylase leading to an increase of extracellular adenosine and consequently, cyclic adenosine monophosphate, which inhibits the production of proinfalmmatory cytokines including tumor necrosis factor TNF-± and interleukin-1² and their downstream effects on lymphocyte activation and proliferation.

Additionally, MTX inhibits thymidylate synthetase (TS) directly and indirectly via depletion of THF, leading to inhibition of pyrimidine (thymidylate) biosynthesis with a resultant antiproliferative effect.

MTX also modulates the function of many of the cells involved in inflammation and affects the production

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of various cytokines including the reduction of TNF-±, interferon-³ (INF-³), IL-1, IL-6 and IL-8 production, thereby acting as a potent inhibitor of cell-mediated immunity.

By reducing the expression of adhesion molecules on endothelial cells, it may reduce the permeability of vascular endothelium, adenosine inhibits adherence of stimulated neutrophils to endothelial cells thereby protecting the vascular endothelium from neutrophil-induced damage.

MTX may also have more direct effects in inflamed joints by inhibiting the proliferation of synovial cells and synovial collagenase enzymes.

Pharmacology

MTX has multiple actions such as anti-inflammatory, immunomodulatory, and as antimetabolite. It should be administered on an empty stomach with water or clear beverages. Bioavailability with intramuscular injection is 15% better than oral bioavailability. Bioavailability of the subcutaneous and intramuscular route of drug administration is similar with the former being more acceptable for children requiring parenteral therapy. After administration, MTX has a relatively short half-life in plasma before it is redistributed to the tissues. The predominant route of elimination is renal, with more than80% of the drug eliminated unchanged via glomerular filtration and tubular secretion within 8 to 48 hours. A smaller but significant route of elimination is the biliary tract. Any decrease in glomerular filtration rate can prolong tissue exposure to MTX and increase the risk of toxicity.

Methotrexate rapidly enters into the cells and is polyglutamatedby hepatocytes, red cells, fibroblast, bone marrow myeloid precursors and possibly other cells. Although MTX is cleared rapidly by the kidneys, polyglutamated MTX accumulates intracellularly, is more stable and reliable biomarkers of the effect of MTX which is the basis of single weekly dose.[3, 8,9, 10]

Dosage, route of administration and duration of methotrexate therapy. Standard effective dosing regimens of MTX in children with JIA are 10 to 15 mg/m^2/week or 0.3 to 0.6 mg/kg/week. Improvement is generally seen by about 6 to 8 weeks on effective doses, but may take up to 6 months to see the full effect.Usually children can tolerate much higher doses than adults and some series have described using 20 to 25 mg/m^2/week or 1.1 mg/kg/week in children with resistant disease, with relative safety in the short term.[9, 11]

Oral route is satisfactory in most patients as a single weekly dose on an empty stomach. For dose more than 15 mg/m^2/week, the parenteral route is preferred because of the reduced oral bioavailability of the drug at high doses. Subcutaneous MTX has a 10 to 12% increased absorption compared with oral preparation. Studies in adult patients with rheumatoid arthritis suggest that oral absorption of MTX is considerably reduced at doses of 15 mg or more. [10,12] Parenteral MTX at initiation of treatment may ensure complete absorption and early disease remission. Subcutaneous MTX is administered by using an insulin syringe and it is very easy and safe to administer. So parents/caretakers and older children themselves can take it at home if they are taught the technique properly. Till date no local side effects related to SC MTX is reported.

The issue of when, how, and by what criteria to consider withdrawing MTX therapy in JIA remains unclear. However, the criteria for remission or relapse have usually not been well defined or standardized among various studies, and the assessment of outcomes has not been the subject of blind studies. It is well recognized that MTX should be continued for many months to years after a remission have been achieved. MTX withdrawal may result in disease flare in more than 50% of patients; this rate may be even higher in young children. Cellular biomarkers such as myeloid-related protein (MRP) 8 and MRP 14 heterocomplex (calprotectin, or MRP8/14) secreted by activated phagocytes at local sites of inflammation may be viable biomarkers to determine the appropriate time to discontinue MTX. So it is the residual synovial inflammation which seems to influence the rate of relapses after discontinuation of MTX. Recent studies recommended that patients maintaining remission for one year can gradually discontinue MTX to reduce potential long term toxicities.[9,10,13]

Side effects of MTX

Although MTX is associated with many potential toxicities, the documented overall frequency and severity of adverse effects in children with arthritis have been low. Most side effects are mild and reversible and can be treated conservatively. Although the precise mechanism of all MTX-related toxicities is not clearly understood, at least some of

MTX's adverse effects are directly related to its folate antagonism and its cytostatic effect. This relationship is especially evident in tissues with a high cell turnover rate, such as the GI tract and bone marrow, that have a high requirement for purines, thymidine, and methionine, which may explain why supplementation with folic or folinic acid may diminish these symptoms.

Gastrointestinal toxicity is the most common adverse effect and occurs in 13% of the patients. Additional side effects include hepatotoxicity, oral mucosal ulcerations, immunosuppression, pancytopenia, pulmonary disease and increased risk of malignancies. The toxicity of MTX is related more to the length of exposure than to the dose given. Liver enzyme (ALT) level may be increased about two to three times of upper limit than normal, but often it is transient and usually becomes normal within two to three weeks.

The issue of greatest concern with the long term use of low-dose MTX in children has been the potential for significant liver fibrosis or cirrhosis. But fortunately, cirrhosis has not been reported in children using MTX for rheumatic disease, except in adults who have comorbidities that may include heavy alcohol consumption, preexisting liver disease, obesity, insulin-dependent diabetes mellitus and renal insufficiency. Several small studies did not reveal any significant liver biopsy abnormalities in children with JIA after 2.3 to 6 years of treatment with MTX.[7, 14]

Gonadal function and reproduction are not altered by MTX. However, both males and females should wait for 3 months after discontinuing MTX before conception as MTX is a powerful teratogen.[7]

Contraindications of MTX

Therapy must be withheld if the ALT is more than two times above upper level of the normal range, hemoglobin <8 gm/dl, platelets <50000/mm^3, white cell count <3000/mm^3, and creatinine clearance <30ml/min. Hepatitis B or C virus infection, child coming in contact with chicken pox or develops chicken pox, immunodeficiency state or renal and liver failure are also contraindications for MTX therapy.[5, 10, 16]

Monitoring of treatment

Before starting treatment with MTX, base line investigations like complete blood count, liver enzyme (ALT), serum creatinine, urine for routine examination and sometimes bone marrow examination should be done. After the introduction of MTX patients should be monitored initially every 4 weeks for the first three months of treatment, then every 8-12 weeks, with more frequent intervals after dosing adjustment or in response to abnormal values.[3, 16]

Folic acid supplementation

As folate deficiency has been thought to play an important role in the development of MTX related side effects, supplementation of folic acid has been shown to lessen gastrointestinal and mucocutaneous side effects without altering the therapeutic effects of MTX. All patients on oral or subcutaneous MTX should take 1 mg folic acid once daily and skip the drug on the day before or the day of MTX administration.[15, 16]There are no clinical trials in paediatrics to support any specific regimen, and the least frequent dose is often preferred for reasons of patient compliance. Folic acid deficiency is rare in children as they have an adequate intake of folic acid in their diet.

Conclusion

Despite availability of new molecules, MTX remains a drug to recon with in treatment of rheumatological diseases in children due to its cost, ease of administration, long track record of safety and efficacy.

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ORIGINAL ARTICLE

Clinical Response to Levamisole Versus Low Dose Prednisolone in Frequently Relapsing Steroid Sensitive Nephrotic Syndrome

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Abstract

Background: *Levamisole is an immunomodulating drug has shown high response rate, reducing relapse rate and significant steroid sparing effects in frequent relapse nephrotic syndrome (FRNS).*

Objective: *To evaluate the effectiveness of levamisole versus low dose prednisolone in maintain remission in children with steroid sensitive nephrotic syndrome (SSNS) who had a frequent relapsing and/or steroid dependent course.*

Methodology: *This prospective randomized controlled trial was conducted during the period from July 2003 to August 2005. Children in relapse were randomly assigned to either the levamisole group or low dose prednisolone group. A total of 40 patients were enrolled. Remission was firstly induced by steroid then levamisole was added and steroids were completely withdrawn within 3 months. Low dose prednisolone (LDP) group was also induced remission with standard dose prednisolone. In both groups treatment was continued for 1 year.*

Results: *Median age was 6 years (2-13 years), 29 were male and 11 were female. Mean number of relapse/patient/year was reduced from 4.40±0.75 to 0.75±0.20 in levamisole group ($p<0.001$) and from 4.40±0.82 to 1.70±0.98 in LDP group ($p<0.001$). Eight months after stopping treatment mean number of relapse is significantly less in levamisole group than LDP group (0.90±0.55 and 1.40±0.60 respectively, $p< 0.05$). No significant side effects were reported in any group.*

Conclusion: *Levamisole is more effective in reducing relapse rate than low dose prednisolone. It is safe and less costly than low dose prednisolone.*

Key word: *Nephrotic syndrome, frequent relapse, levamisole, low dose prednisolone.*

Introduction

The nephrotic syndrome, which occurs mainly in children is manifested by edema, albuminuria, hyperlipemia and hypoproteinemia usually with relatively normal level of non protein nitrogen in the blood and relatively normal blood pressure and is characterized by a chronic course with exacerbations, remissions and, in many instances, eventual recovery.[1] Most frequent type (77%) of idiopathic nephrotic syndrome (INS) is minimal change nephrotic syndrome[2] and more than 90% of MCNS well responded to steroid therapy.[3] Three quarters of responders will have a subsequent relapse and one third will suffer from frequent relapse.[4]

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Childhood nephrotic syndrome (NS) remains a major challenge for pediatric nephrologists because of its complex evaluation, chronicity and long term management. Levamisole is an immunomodulating drug. In the therapeutic concentrations levamisole restores to normal, both in vivo and in vitro, many of the effectors functions of T-cells and phagocytes, and induce the maturation of immature T-cells. However, these effects are reproducible only when the immune system is depressed. Levamisole was used sporadically with variable success and minimal side effects in the 80's. This was followed by randomized studies, which showed the high response rate and significant steroid sparing effects effective in reducing relapse of levamisole. In view of the result of these studies, levamisole had been recommended as the treatment of frequent relapsing SSNS and these had led to successful reports from different parts of the world.[5] However, to date there is no report of its use in Bangladeshi children in whom environmental, genetic and social factor may altered their pattern of response to therapy. Management of mild, but frequently relapsing, NS using low dose, alternate day prednisolone therapy appears to successfully minimize relapse and avoid the undesirable steroid effects of obesity, retardation of growth velocity, and infections susceptibility.[6]

At our institution, the 2 most frequently used drugs are levamisole (L) and low dose prednisolone (LDP). Very limited study has been reported in the literature.[6,7,8] Therefore, we conducted this prospective trial in frequently relapsing and/or steroid dependant nephrotic syndrome Bangladeshi children, who had not received any form of adjunctive therapy, to study their pattern of response to a fixed and longer period of (12 months) of L or LDP treatment, following them up for a further 8 months after stopping the drug. So this study was carried out to compare the effectiveness of levamisole versus low dose prednisolone in maintaining remission in children with steroid-sensitive nephrotic syndrome who had a frequently relapsing and/or steroid dependant course.

Materials and Methods

This was a randomized control trial conducted from 1st July 2003 to 31st August 2005 at follow up clinic and nephrology indoor of Dhaka Shishu (Children) Hospital. First 6 months were spent to enroll patients; then 1 year of intervention and 8 months post intervention follow up. Forty children of steroid sensitive frequently relapsing nephrotic syndrome or steroid dependent nephrotic syndrome, who's remission was induced by steroid and none received any type of adjunctive therapy for their nephrotic states before the study, visited in Nephrology follow up clinic or admitted in nephrology indoor having normal renal function, liver function and complete blood count were enrolled and followed up for 20 months from date of inclusion. Steroid resistant nephrotic syndrome, any secondary cause of nephrotic syndrome or any associated disease eg. TB, hepatitis were excluded from the study. All patients were in relapse at the start of the study. Relapse was defined as edema and early morning urine for protein (by heat test) of at least 3^+ for a minimum of 3 consecutive days. Every alternate child in relapse was randomly assigned to either the levamisole (L) group or the low dose prednisolone (LDP) group. Induction of remission was instituted by prednisolone 60mg/m^2/day until urine protein free (by heat lest) for 3 consecutive days. The same dose of prednisolone was then given on alternate days and tapered by 5mg every 2 weeks interval until complete withdrawal within three months.

Levamisole group was treated with Levamisole 3 mg/kg body weight on alternate days[5] was instituted immediately after induction of remission by daily prednisolone and continued for 1 year. Low dose prednisolone group received prednisolone 0.5 mg/kg body weight on alternate days after induction of remission by prednisolone followed by tapering 5mg every 2 weeks interval until the prednisolone dose reached to 0.5mg/kg/48 hours. Then prednisolone 0.5mg/kg/48 hours continued for 1 year. Patients were followed up monthly during the 12 months treatment period and for a further 8 months after stopping treatment.

A detailed history with special emphasize on history of atopy, precipitating factor and associated complication with detailed physical examination including height, weight, blood pressure, ocular findings and presence of any signs of infection were recorded before entry into trial. Counseling of long term use of levamisole was done by adequate explanation regarding the history of the medicine and that it was originally used as an antihelmenhic but found to be useful in cases of SSNS and misconception regarding its treatment was removed. Also baseline investigation as urine routine and

microscopic examination, complete blood count. alkaline phosphatase, serum creatinine, serum albumin was also done on entry into trial.

First follow up was arranged just after remission i.e. the patients were advised to come after disappearance of edema to note the weight and to adjust dose of drugs. Next visit were at monthly internal or any time if relapse occurred or if any complication arise. At each visit any new complications, height, weight, temperature, blood pressure, pallor, jaundice, edema, side effect of levamisole or prednisolone was recorded. Follow up investigations were routine urinanalysis on every month, complete blood count on every 3 months. Serum creatinine with Alanine aminotransferase (ALT) on every 6 months. Relapse that occurred during administration of levamisole or LDP were treated with prednisolone regimen of Arbeitsgemeinschaft fu¨r Pa¨diatrische Nephrologie (APN protocol) while levamisole was continued and in case of LDP group at the end of standard therapy with prednisolone, LDP was resumed. Informed consent was obtained from the parent before the study and the study was approved by scientific and ethical committee of the hospital. All the data were collected using questionnaire and were analyzed using computers based programme windows version SPSS 11.5.

Results

Forty two patients were enrolled in the present series. Two patients, one from each group lost to follow up. Among them 32 (80%) were frequently relapsing without steroid dependent NS and 8(20%) were frequently relapsing with steroid dependent NS (Table-I).

Table I
Distribution of nephrotic children depending on relapse (n= 40)

Type of relapse	Number	Percentage
FRNS (Without SDNS)	32	80%
FRNS +SDNS	8	20%

The age at onset and treatment, body weight, height, mean number of relapses prior trial of children studied were similar in the two groups. Three fourth of the patients in both groups were male. There is statistically significant difference between WBC counts which is not clinically significant. The duration of follow up was similar in both groups. One patient from each group was lost to follow up group (Table-II).

Table II
Comparison of baseline patient characteristics between groups

Baseline characteristics#	Group		p values
	Levamisole (n = 20)	Low-dose prednisolone (n = 20)	
Age at treatment (yrs)	6.83 ± 0.59	6.02 ± 0.63	0.355
Sex (male/female)	14/6	15/5	0.500
Age at onset (yrs)	3.17 ± 0.35	3.22± 0.26	0.911
Body-weight (kgs)	21.38 ± 1.35	21.13± 1.84	0.910
Height (cms)	111.70 ± 3.51	106.27 ± 5.08	0.385
WBC (/cu-mm of blood)	10795 ± 298	9675 ± 230	0.005
No. of relapse (last 1 yr)	4.40 ± 0.17	4.40 ± 0.18	0.999
Side effect (%)	35	40	0.769

All the variables, except sex and side-effect are expressed as mean ± SEM; sex and side-effect are presented as male-female ratio and percentages respectively.

** The data, except sex and side-effect, were analysed using Student's t-test; sex and side-effect were analysed with help of Chi-square (χ^2) test; level of significance was 0.05.

Clinical presentation manifests that 100% patients present with facial puffiness and ankle edema, majority with ascites and scanty urine and a few with scrotal swelling. Other manifestations are fever, cough, abdominal pain, diarrhoea, anorexia/vomiting transient hypertension, leg cramps, respiratory distress, wheeze. There is no statistically significant difference in clinical presentation in two groups (Table-III).

At the end of 1 year of treatment in levamisole group mean number of relapse per patient per year has fallen to 0.075±0.20 ($p<0.001$) and 8 months after stopping the drug it is 0.90±0.12 ($p<0.001$) and is statistically significant. Steroid side effects have also resolved significantly after treatment with levamisole ($p=0.026$). WBC count also decreased but does not reach to the level of leucopenia (Table-IV).

Table III

Comparison of clinical presentation before intervention between groups

Clinical presentation	Group		p values
	Levamisole (n = 20) No (%)	Low-dose prednisolone (n = 20) No (%)	
Facial puffiness	20 (100.0)*	20 (100.0)	(——)**
Oedema	20 (100.0)	20 (100.0)	(——)**
Ascitis	16 (80.0)	11 (55.0)	0.091CS
Scanty urine	11 (55.0)	15 (75.0)	0.185CS
Scrotal swelling	4 (20.0)	5 (25.0)	0.500FE
Fever	6 (30.0)	5 (25.0)	0.723CS
Cough	4 (20.0)	6 (30.0)	0.465CS
Abdominal pain	4 (20.0)	1 (5.0)	0.171FE
Diarrhoea	3 (15.0)	00	0.115FE
Anorexia/Vomiting	2 (10.0)	1 (5.0)	0.500FE
Transient hypertension	2 (10.0)	1 (5.0)	0.500FE
Leg cramps	3 (15.0)	4 (20.0)	0.500FE
Respiratory distress	00	2 (10.0)	0.244FE
Wheeze	1 (5.0)	1 (5.0)	0.756FE

* Figures in the parentheses indicate corresponding percentage.
** No statistics can be computed, for all the patients of both the groups had puffiness and oedema.
CS = Chi-square (χ^2) test was done.
FE = Fisher's Exact test as 2 cells had expected count < 5.

Table IV

Evaluation of outcome before and after intervention (Levamisole Group)

Outcome variables#	Levamisole Group		**p values
	Before intervention (n = 20)	After intervention (n = 20)	
No. of relapse	4.40 ± 0.17	0.90 ± 0.12	<0.001
WBC (/cu-mm of blood)	10795 ± 298	8340 ± 225	<0.001
Side effect (%)	35	20	0.026

All the variables, except side-effect are expressed as mean ±SEM; side-effect is presented as %.

** The data, except side-effect, were analysed using Paired sample "t" Test; side-effect was analysed with help of Chi-square (χ^2) test; level of significance was 0.05.

Table V
Evaluation of outcome before and after intervention (in Low-dose prednisolone)

Outcome variables#	Low-dose prednisolone Group		**p values
	Before intervention(n = 20)	After intervention(n = 20)	
No. of relapse	4.40 ± 0.18	1.40 ± 0.13	<0.001
WBC (/cu-mm of blood)	9675 ± 230	9510 ± 163	0.350
Side effect (%)	40	35	0.769

All the variables, except side-effect are expressed as mean ±SEM;side-effect is presented as %.
** The data, except side-effect, were analysed using Paired sample "t" test; side-effect was analysed with help of Chi-square (χ^2) test; level of significance was 0.05.

In LDP group also shows significant reduction in mean number of relapses per patient per year (1.70±0.22 $p<0.05$) comparing before intervention and also after stopping the drug (1.40±0.13, $p<0.005$). There is no significant difference in WBC count or steroid side effect before and after intervention (Table-V)

Fifty percent patient maintaining remission (no relapse) during treatment whereas twenty percent patient maintaining remission (no relapse) i.e. continued suppression of relapses 8 months after stopping the levamisole. Fourteen patients i.e. 70% patients having only one relapse after stopping the drugs. In LDP group maintaining remission (no relapse) is only 10% during treatment and no patient after stopping the drug (Fig 1 & 2).

In levamisole group among 20 patients, side effect of steroid before trial was found in 7 (35%) patients. The side effects were chushingoid facies (3), obesity (2), short stature (3), hypertension (2) had dropped to 4 patients (20%) that is 1 patient with cushingoid facies and hypertension resolved 1 regresses obesity and 1 improved height. There is statistically significant difference (p=.026) in resolving steroid side effects before and after intervention with levamisole (Table-VIa). Whereas in LDP group 8 patients (40%) had steroid side effects, among them only 1 patients (5%) having short stature improved height after intervention with LDP (Table-VIb).

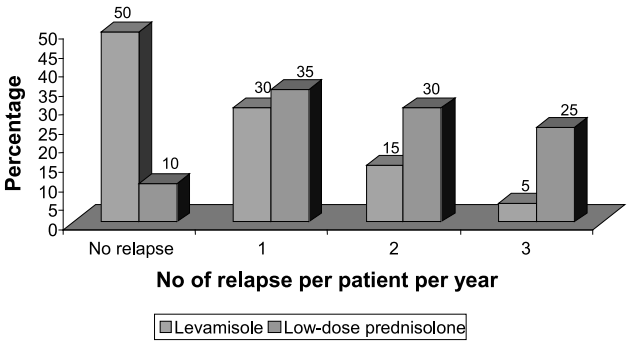

Fig 1 *Compares the no of relapses during 1 yr intervention*

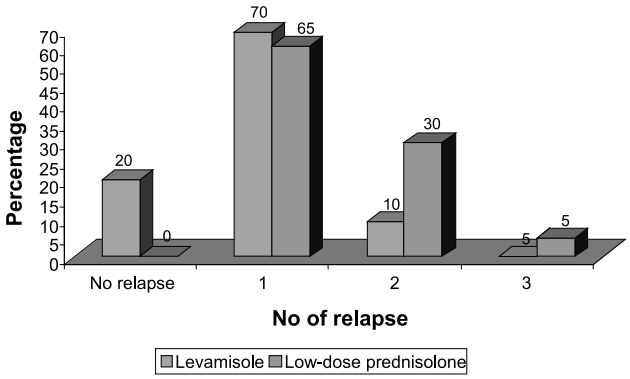

Fig 2 *Compares relapses during 8 months follow up*

Table VIa
Steroid side-effects of levamisole before and after intervention

Side effects	Levamisole	
	Before interventio n (No)	After intervention (No)
Cushingoid facies and short stature	1	1
Cushingoid facies & obesity	1	1
Cushingoid facies & hypertension	1	0
Short-stature	2	1
Hypertension	1	1
Obesity	1	0
Total	7 (35%)	4 (20%)

Table VIb
Steroid side-effects of low dose prednisolone before and after intervention

Side effects	Low dose prednisolone	
	Before intervention (No)	After intervention (No)
Cushingoid facies and short stature	1	1
Cushingoid facies & short stature & hypertension	1	1
Cushingoid facies & hypertension	1	1
Cushingoid facies	1	0
Short-stature	1	0
Obesity	2	2
Overweight & gynaecomastia	1	1
Total	8 (40%)	6 (35%)

Table VII
Comparison of outcome after intervention between groups

Outcome variables	Group		p values
	Levamisole (n = 20)	Low-dose Prednisolone (n = 20)	
Body-weight (kgs)	23.72 ± 1.43	23.65 ± 2.07	0.698
Height (cms)	119.17 ± 3.34	117.05 ± 3.13	0.758
WBC (/cu-mm of blood)	8340 ± 225	9510 ± 163	<0.001
No. of relapse during 1 yr of intervention	0.75 ± 0.20	1.70 ± 0.22	0.004
No. of relapse during 8 months follow up	0.90 ± 0.12	1.40 ± 0.13	0.038
Side effect (%)	20	35	0.026
Cost of treatment (Tks/patient/yr)	165.6 ± 39.7	272.7 ± 25.1	<0.001

All the variables, except side-effect are expressed as mean ±SEM;side-effect is presented as %.
** The data, except side-effect, were analysed using Mann Whitney test; side-effect was analysed with help of Chi-square (χ^2) test; level of significance was 0.05.

There was no significant difference between the groups with respect to body weight and height. But mean number of relapses during and after intervention was observed to be significantly less in levamisole group compare to low dose prednisolone group. There was also significant difference in resolving steroid side effect in levamisole group comparing LDP group ($p < 0.05$). None of our 20 patients treated with L had serious side effect. Only 4 patients (20%) developed nausea/vomiting, headache, anorexia and abdominal pain which were not serious enough to get admit to hospital. None of our patient developed neutropenia. However levamisole treatment did result in a reduction in leucocyte count. The comparative cost analysis based on local drug prices revealed that 1 year levamisole treatment cost was much lower than 1 year LDP (165±39.7 taka vs 272.7±25.1 taka). The difference is statistically significant ($p < 0.001$) [Table VII].

Discussion

A number of children suffering from idiopathic nephrotic syndrome (INS) are steroid dependent or have frequent relapses that impair their quality of life. They generally need long term alternate day steroid therapy. However, this schedule does not affect the frequency of relapses after withdrawal of therapy. In addition some patients need high doses of steroids to be kept in remission. They are, therefore, at risk of steroid toxicity.[7]

Immunosuppressive agents often cause a remission of the disease which lasts until sometimes after withdrawal therapy. Nevertheless the administration of these drugs is hampered by short and long term toxicity (cyclosphosphamide, chlorambucil) or by the lack of a wide experience of therapeutic results and of side effects (cyclosporin).[9]

There are few reports of successful use of LDP by Elzouki et al[6], Wingen et al [7], Srivastava et al [8]. The use of levamisole in SSNS was 1st describe by Tanpaichitr et al[10] is an uncontrolled study in 1980. This was followed by several reports (Bagga et al[11], British Association for Paediatric Nephrology[12], Mongeau et al[13]) showing the successful treatment of FR/SD nephrotic syndrome with L. L was originally developed for use as an anti-helminthic agent, but subsequently was shown to have immunomodulating properties. Despite almost 3000 reports about experimental and clinical effect of levamisole, there is no acceptable explanation of the mode of action of this drug. Since levamisole inhibits the in vitro formation of the oxidized and active form of immunosuppressive lymphokine it is said to restore many of the effector functions of T cells and phagocytes to normal, and to induce the maturation of immature T cells.

Fifty percent patients are maintaining remission (no relapse) after one year of levamisole treatment which is similar to the results obtained by Donia et al[14], Mongeau et al[13] and Alshaya et al[5]. There are several reports of beneficial results using small daily dose of prednisolone but none of the study has similar dose frequency and duration comparable to our study.

A students't' test comparison of the number of relapses during and after L/LDP therapy revealed that L was associated with greater reduction of number of relapses ($p<0.005$ and $p< 0.05$ respectively).

In levamisole group among 20 patients, side effect of steroid before trial was found in 7 (35%) patients. The side effects were cushingoid facies (3), obesity (2), short stature (3), hypertension (2) had dropped to 4 patients (20%) that is 1 patient with cushingoid facies and hypertension resolved 1 regresses obesity and 1 improved height. There is statistically significant difference ($p=.026$) in resolving steroid side effects before and after intervention with levamisole. Bagga et al[11] also shows disappearance of steroid side effect after treatment with levamisole. This is because relapse rate were significantly decreased that results in overall reduction in steroid requirement thus steroid toxicity. Whereas in LDP group 8 patients (40%) had steroid side effects, among them only 1 patient (5%) having short stature improved height after intervention with LDP. This observation is different from Elzouki et al[6] study. None of our 20 patients treated with L had serious side effect. Only 4 patients (20%) developed nausea/ vomiting, headache, anorexia and abdominal pain which were not serious enough to get admit to hospital. This agrees well with the safety of levamisole reported by British Association for Paediatric Nephrology.[12] None of our patient develops neutropenia. However levamisole treatment did result in a reduction in leucocyte count. Since the occurrence of agranulocytosis is not dose dependent and cannot be predicted, regular blood counts are necessary. The comparative cost analysis based on local drug prices revealed that 1 year levamisole treatment cost was much lower than 1 year LDP (165±39.7 taka versus 272.7±25.1 taka). The difference is statistically significant ($p <0.001$).

Conclusion

Levamisole therapy appears to be effective in about 50% of steroid dependent and/or frequently relapsing NS patients. Long term therapy with levamisole reduces the overall relapse rate in Bangladeshi Children with SSNS. It is cheaper than LDP and its extended use is usually not associated with significant side effects and beneficial effect may continue even after stoppage.

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DS (Child) H J 2016; 32(1) : 17-21

ORIGINAL ARTICLE

Comparison of Clinical Features and Hematological Parameters Between Culture Positive and Negative Neonatal Sepsis

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Introduction

Neonatal sepsis is the most important cause of morbidity and mortality especially among low birth weight and preterm babies in developing countries. Neonatal sepsis is diagnosed when generalized systemic features are associated with pure growth of bacteria from one or more sites. When clinical and laboratory findings are consistent with bacterial infection but blood culture is sterile, infant is labeled to have "probable sepsis". According to pooled hospital data based on NNPD survey, the incidence of neonatal sepsis is around 30 per 1000 live births.[1] The burden of neonatal septicaemia has remained high worldwide and even more severe in the developing countries like ours. Clinical manifestation is variable and non-specific thereby resulting in delay in diagnosis. Blood culture which is the gold standard for diagnosis of neonatal septicaemia (NNS) has many drawbacks due to long waiting time for culture process, low yield, improper inoculation adding to the problem of late diagnosis. Haematological parameters have been utilized in rapid and early diagnosis of NNS and prompt treatment thus circumventing problems associated with drawbacks in blood culture.[2] Sepsis in neonates may be difficult to differentiate from other conditions because the clinical signs are non specific. The hematological response to inflammation in neonates includes many changes in the form of an abnormal total count, morphological changes, thrombocytopenia and various inflammatory markers in serum.[3]

Early recognition, diagnosis and treatment of serious infection in the neonate are essential because of the risk of permanent morbidity and mortality. Neonatologists have a critical need for laboratory tests that aid in the early diagnosis of neonatal sepsis. Various studies have shown that hematological parameters are simple, quick and cost effective tool in the early diagnosis of neonatal sepsis. Present study is undertaken to evaluate the role of haematological profile in early diagnosis of neonatal sepsis because this is simple test which can be done within a short time before putting neonate on antibiotic therapy.

Material and Methods

This was a consecutive prospective study conducted at the Department of Neonatology, Dhaka Shishu (Children) Hospital over a period of eighteen months from 1st April 2013 to 31st September 2014. The study was carried on 110 neonates who met the inclusion criteria and detailed history, examination findings were recorded onto study proform.All study patients were grouped into GroupA(culture positive) and Group B (culture negative). Inclusion criteria were preterm/term neonate with features suggestive of neonatal septicaemia such as fever,hypothermia, poor sucking, respiratory distress, lethargy, abdominal distention, Maternal risk factors such as peripartum pyrexia, prolong rupture of membrane >18hours and chorio amnionitis, History of delivery in unhygienic setting. Exclusion criteria include: History of peripartum antibiotic use in the mother or in the neonate prior to presentation Presence of congenital defect in the neonate. History of surgery of surgery prior to presentation and parental decline to consent to included in the research. The patients had blood culture done, and also other sepsis screening was done when it was indicated. Blood for

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culture was taken on admission from a peripheral vein (antecubital fossa or dorsum of the hand) using a 24 gauge scalp vein needle after thorough skin preparation with application of 2% iodine tincture and followed by cleansing with 70% alcohol solution. Two culture bottles, containing 12 ml of culture media- Brain Heart Infusion (BHI) was inoculated aseptically with at least 1 ml of blood each and immediately incubated at 37ÚC for 18 - 24 hrs. Each specimen was examined twice a day for growth. Turbidity in the culture bottles and/or excess production of gas with signs of haemolysis indicates bacterial growth. It was sub-cultured on blood agar using sheep blood and/or MacConkey agar and this was incubated at 37ÚC for 18 - 24 hrs. Other specimen that was cultured including urine, umbilical swab and cerebrospinal fluid when it was indicated. When the culture was positive, sensitivity pattern was carried out against the common antibiotic discs provided by the use of disc diffusion method according to clinical and laboratory standard institute. Another 2 ml of blood was also collected in an EDTA container for a complete haematological profile by direct counting using an improved Neubaur's Chamber. The packed cell volume (PCV), total white blood cell count (TWBC), with differential white cell count, leucocytes morphology and absolute neutrophil count, platelet count (PC) as well as immature to total neutrophil ratio (I/TNR) was estimated on peripheral blood smear (PBS) stained with Leishmann's stain and value > 0.2 was considered abnormal.

Results

The mean age found 8.33±7.36 days in group A and 9.71±7.02 days in group B. The mean weight was 2816.25±288.6 grams in group A and 2894.58±296.7 grams in group B. Male were predominant in both groups, which was 36(75.0%) in group A and 30(62.5%) in group B. The difference was not statistically significant (p>0.05) between two groups (Table-I). Regarding previous history of the patients. It was observed that delayed cry was found 20(41.7%) in group A and 16(33.3%) in group B. Stopped feeding well was 46(95.8%) in group A and 40(83.3%) in group B. Convulsion was 18(37.5%) and 16(33.3%) in group A and group B respectively. Excessive cry was found 4(8.3%) in group A and 10(20.8%) in group B (TableII). Regarding clinical features it was observed that irritability was found 22(45.8%) in group A and 12(25.0%) in group B. Pyrexia (>98.4°F) was found 26(54.2%) in group A and 14(29.2%) in group B. Which was statistically significant (p<0.05) between two groups. Other clinical features were not statistically significant (p>0.05) between two groups (Table -III). Regarding hematological parameters in neonatal septicaemia. It was observed that mean PCV was found 43.75±7.06% in group A and 46.91±7.42% in group B. Mean neutrophil was 68.75±7.39 % in group A and 64.38±6.12% in group B. Mean lymphocyte was 27.58±6.61% and 30.81±7.38% in group A and group B respectively. Mean micro-ESR was 8.13±4.17 mm/hr in group A and 5.79±3.24 mm/hr in group B. Mean absolute platelet count was 147.97 ±33.67 10^9/L and197.02±45.82 10^9/L in group A and group B respectively. Mean immature to mature neutrophil was 0.28±12.32 in group A and 0.18±0.07 in group B. Mean immature to total neutrophil was 0.25±0.06 and 0.14±0.09 in group A and group B respectively. Which were statistically significant (p<0.05) between two groups (Table -IV).

Table I
Comparison of age and weight of neonates between the study groups

Parameters	Group A Mean±SD	Group B Mean±SD	p value
Age (days)	8.33±7.36	9.71±7.02	0.351^{ns}
Weight(g)	2816.25±288.6	2894.58±296.7	0.193^{ns}
Sex			
Male	36 (75%)	30(62.5%)	$>0.05^{ns}$
Female	12 (25%)	18(37.5%)	

Table II
Previous history of study patients

History	Group A n(%)	Group B n(%)	p value
Delayed cry	20(41.7)	16(33.3)	
Stopped feeding well	46(95.8)	40(83.3)	
Convulsion	18(37.5)	16(33.3)	
Excessive cry	04(8.3)	10(20.8)	

Table III
Distribution of the patients by clinical features

Clinical feature	Group A n (%)	Group B n (%)	p value
Lethargy	46(95.8)	44(91.7)	0.67^{ns}
Abdominal distension	8(16.7)	6(12.5)	0.56^{ns}
Severe respiratory distress	32(66.7)	30(62.5)	0.67^{ns}
Apnoea	12(25.0)	8(16.7)	0.31^{ns}
Severe chest in drawing	28(58.3)	26(54.2)	0.68^{ns}
Cyanosis	24(50.0)	22(45.8)	0.68^{ns}
Irritability	22(45.8)	12(25.0)	0.03*
Diminished reflexes	46(95.8)	42(87.5)	0.26^{ns}
Bleeding	2(4.2)	0	0.49^{ns}
Jaundice	10(20.8)	8(16.7)	0.60^{ns}
Raised respiratory rate (>60 b/min)	26(54.2)	24(50.0)	0.68^{ns}
Raised heart rate (>160 b/min)	2(4.2)	2(4.2)	1.000^{ns}
Pyrexia (>98.4^{o}F)	26(54.2)	14(29.2)	0.013*

Table IV
Shows hematological parameters in neonatal septicaemia.

Hematological parameters in neonatal septicaemia.	Group A (n=48) Mean±SD	Group B (n=48) Mean±SD	p value
PCV (%)	43.75±7.06	46.91±7.42	0.035
TWBC (109/L)	10.12±2.21	9.67±3.11	0.415
Neutrophil (%)	68.75±7.39	64.38±6.12	0.002
Lymphocyte (%)	27.58±6.61	30.81±7.38	0.026
Platelet count (109/L)	147.97±33.67	197.02±45.82	0.001
Immature to total Neutrophil	0.25±0.06	0.14±0.09	0.001

Discussion

The early diagnosis of neonatal septicemia is primarily based on clinical evaluation but laboratory diagnosis requires a microbiologic-clinical correlation. Blood culture is considered as gold standard for diagnosis of sepsis.[4] A drawback of culture based diagnosis is the assay time of up to 36 hours.[4] In order to diagnose septicemia early, several rapid diagnostic tests have been described, which are easily performed and have the benefit of quick availability of reports. The need for early recognition, diagnosis of neonatal septicaemia and prompt institution of treatment is very much needed so as to prevent unwanted death and complication associated with neonatal septicaemia, but it is quiet often difficult and delayed because of the variable and non-specific clinical manifestation of neonatal septicaemia.[5] In our study considering all other parameters to identify the sepsis diagnosis. The mean age was found 8.33±7.36 days in group A and 9.71±7.02 days in group B. The mean weight was 2816.25±288.6 grams in group A and 2894.58±296.7 grams in group B. Male were predominant in both groups, which was 36(75.0%) in group A and 30(62.5%) in group B. The difference was not statistically significant (p>0.05). Compared with the Saleem et al[6] study showed the mean age of participants was 12.4 ± 7.3 days, of which 54.1% were male. Most neonates (52.4%) were in the age group up to 10 days. In Khair et al[7] study, among the sepsis group male (58.33%) was more common than female newborns. Regarding clinical features it was observed that irritability was found 22(45.8%) in group A and 12(25.0%) in group B. Pyrexia (>98.4^{o}F) was found 26(54.2%) in group A and 14(29.2%) in group B. Which was statistically significant (p<0.05) between two groups. Other clinical features were not statistically significant (p>0.05) between two groups. Some of the clinical features that are presumably described to neonatal infections can also occur in other neonatal conditions like hypoglycaemia, perinatal asphyxia among others.[8,9] The clinical symptoms were fever, poor feeding, excessive cry, difficulty in breathing, yellowish skin discoloration, skin rashes, jitteriness and irritability; they were the common features at admission in the study subjects. Hyperthermia, hypothermia, tachypnoea, septic umbilical stump hepatomegally, skin lesion and convulsions were the common physical signs observed, this was similar to those that were reported by some workers.[10,11] It was also observed that features like weakness, hypothermia and apnoea were more common among preterm septic neonates, while excessive cry, pyrexia and convulsions were more in the term septicaemic neonates and these observation was similar to the finding in Asia and America.[10-12]

Regarding hematological parameters in neonatal septicaemia. It was observed that mean PCV found 43.75±7.06% in group A and 46.91±7.42% in group B. Mean neutrophil was 68.75±7.39 % in group A and 64.38±6.12% in group B. Mean lymphocyte was

27.58±6.61% and 30.81±7.38% in group A and group B respectively. Mean micro-ESR was 8.13±4.17 mm/hr in group A and 5.79±3.24 mm/hr in group B. Mean absolute platelet count was 147.97 ±33.67 10^9/L and197.02±45.82 10^9/L in group A and group B respectively. Mean immature to mature neutrophil was 0.28±12.32 in group A and 0.18±0.07 in group B. Mean immature to total neutrophil was 0.25±0.06 and 0.14±0.09 in group A and group B respectively. Which were statistically significant ($p<0.05$) between two groups. This study evaluated these simple and common haematological indices that can be done in laboratory of most developing countries like ours, it was found that these parameters had a good correlation with blood culture positivity and was statistically significant when it was subjected to Student-T tests ($p < 0.05$). Such an observation was also reported by Monroe et al. [13], and Ottolini et al. [14]. However, it was not so by other workers who did not find any relationship between haematological indices and blood culture positive neonatal septicaemia [15]. In the this study, TWBC <5000 or >18,000 had low sensitivity, specificity, positive and predictive value in determining neonatal septicaemia and this was in agreement with the report from Pakistan,[16] but disagree with report by Srilonka and co-workers who observed a higher values in these range.[17]

On evaluating the parameters of HS, the association of sepsis was found to be significant ($p<0.05$) with I: T ratio, I: M ratio, Immature PMN count, Total WBC count and thrombocytopenia as also in other studies[18-20]. Whereas, degenerative changes observed in neutrophils and total PMN count were nonsignificant findings, which is supported by various studies [18,19]. Thrombocytopenia was frequently associated with sepsis and indicated poor prognosis. This is thought to be due to increased platelet destruction, sequestration secondary to infections, failure in platelet production due to reduced megakaryocytes or damaging effects of endotoxin.[19] This correlated well with various other studies done by Speer *et al*., Rodwell *et al*., Philip *et al*., and Basu *et al.* [21-24] The feasibility and the cost effectiveness of the system increase the usefulness of this test. This helps the clinicians to reach a probable diagnosis, decreasing the death toll and institute a rational approach towards the patient medication.

Conclusion

Treatment of Neonatal Sepsis should not be delayed for culture report when suspected, as the clinical features and hematological parameters are similar in both the culture positive and negative cases.

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ORIGINAL ARTICLE

Etiology of Neonatal Cholestasis: An Experience in a Tertiary Centre of Bangladesh

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Abstract

Background*: Neonatal cholestasis is a major cause of morbidity & mortality in young infants. It has a varied etiology and difficult diagnostic problem. The disorder has rarely been studied in centers from Bangladesh.*

Objective: *To determine the etiology of both extra-hepatic & intra-hepatic cholestasis at a tertiary referral center in Bangladesh.*

Materials & Methods: *A prospective, descriptive study was done in the department of Pediatric Gastroenterology, Hepatology & Nutrition, Dhaka Shishu (Children) Hospital, Dhaka, Bangladesh from January 2014 to June 2016 among 80 infants who presented with cholestatic jaundice in <1 year of age.*

Results: *Out of 80 children, 48 (60%) were intra-hepatic causes & 32 (40%) were extra-hepatic causes. Among all cholestasis, biliary atresia (BA) 30 (37.5%), idiopathic neonatal hepatitis (INH) 20 (25%) and TORCH infections 19 (23.7%) were the commonest ones. Among 32 extra-hepatic cholestasis, predominant causes were biliary atresia 30 (93.7%) followed by choledochal cyst 2 (5.3%). In 48 Intra-hepatic cholestasis, idiopathic neonatal hepatitis (INH) 20 (41.6%) and TORCH infections 19 (39.5%) were the dominant ones. Two (4.1%) cases of hypothyroidism, galactosemia and intra-hepatic bile duct paucity (non-syndromic) each. Down's syndrome with hypothyroidism, urinary tract infection (UTI) with sepsis and progressive familial intra-hepatic cholestasis (PFIC) were evaluated in 1 (2%) case of each. Among 19 TORCH infections, Cytomegalovirus (CMV) 13 (68.4%) was the commonest followed by CMV with herpes-simplex virus (HSV) co-infection 4 (21.1%) and Toxoplasmosis 2 (10.5%).*

Conclusion: *The more frequent etiological factors were biliary atresia (BA), idiopathic neonatal hepatitis (INH) & TORCH infections. CMV were predominant cause of TORCH infections.*

Key words: *Neonatal cholestasis, cholestatic jaundice, biliary atresia, Idiopathic neonatal hepatitis, Conjugated hyperbilirubinemia.*

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Introduction

Cholestatic jaundice in infancy is an uncommon but potentially serious problem that indicates hepatobiliary dysfunction. [1] Neonatal cholestasis can be defined as conjugated hyperbilirubinemia in the 1st 90 days of extra-uterine life that occurs when conjugated bilirubin is higher than 1 mg/dl, if the total serum bilirubin is d"5 mg/dl, or >20% of total serum bilirubin when it is >5 mg/dl. [2,3] Conjugated hyperbilirubinemia at any age in a newborn is pathological and requires evaluation. Any new born with jaundice and dark yellow urine staining the diaper with or without pale stools should be strongly suspected to have neonatal cholestasis.[4]

Idiopathic neonatal hepatitis (INH) and biliary atresia are the most frequent causes of cholestatic jaundice in the first months of life.[5,6] Syndrome of neonatal hepatitis has diverse causes. Idiopathic form, sepsis/ urinary tract infection, genetic diseases of metabolism and congenital infections are relatively common causes compared to toxic causes, post-haemolytic states, neonatal acute hepatic necrosis, parenteral nutrition, chromosomal anomalies, familial syndromes etc.[7]

The aim of this prospective, descriptive study is to determine common etiological factors of cholestasis in infants attending a tertiary care hospital of Bangladesh.

Materials and methods

A prospective, descriptive study was conducted in the department of Pediatric Gastroenterology, Hepatology & Nutrition, Dhaka Shishu (Children) Hospital, Dhaka, Bangladesh from January 2014 to June 2016. A total of 80 admitted cases of neonatal cholestasis defined by conjugated bilirubin is higher than 1 mg/dl, if the total serum bilirubin is d"5 mg/dl, or >20% of total serum bilirubin when it is >5 mg/dl, age <1 year, both sexes were enrolled in this study. Patients e"1 year, jaundice due to other causes were excluded from the study.

Complete blood count with culture and liver function tests (S. bilirubin total, direct, indirect, Alanine aminotransferase (ALT), Alkaline phosphatase (ALP), Gamma-glutamyl transpeptidase, (GGT) Prothrombine time (PT) and albumin were done. Urine was tested for non-glucose reducing substances (NGRS) and for bacterial culture. Ultrasonography of hepatobiliary system (fasting & after feed) was done in all cases. Hepatobiliary scintigraphy (HIDA scan) was done from Institute of Nuclear Medicine, Bangabandhu Sheikh Mujib Medical University after administration of phenobarbitone (5 mg/kg/day orally in two divided doses for at least 5 days). At scintigraphy, absence of radioactivity in the small bowel after 24 hours was taken as absent tracer excretion. The biopsy was done with the help of Automated Biopsy Gun (18G×16 cm, PD 22 mm) under local anesthetics (2% lidocaine) and intravenous midazolam (0.1 mg/kg) with 3 hours fasting. The biopsy specimens were immediately fixed in 10% formal-saline. Biopsy was not done in patients with huge ascites, Hemoglobin <10 gm/dl, platelet count <80,000/cu mm, increased prothrombin time (INR >1.3), prolong bleeding and/or clotting time and lack of parental consent.

Biliary atresia were diagnosed on the basis of clinical (full term with good birth weight and persistent pale stool), biochemical (moderate elevation of serum bilirubin, ALT but high elevation of ALP & GGT), ultrasonographic (non-visualized/contracted/small gall bladder), scintigraphc (absent tracer excretion) and liver biopsy (bile ductular proliferation, portal tract fibrosis & bile plugs in portal triads).

To identify the intra-hepatic causes of cholestasis, TORCH screening, thyroid function test and HBsAg were done. If Cytomegalovirus (CMV) IgM positive or IgG >10 fold than normal then urinary CMV PCR was done. Katyotyping was done in one suspected case of Down's syndrome and Galactose-1-phosphate uridylyltransferase (GALT) assay only in NGRS positive cases. Intra-hepatic bile duct paucity was diagnosed by characteristic biopsy finding and PFIC (Type I/II) was diagnosed by low or normal GGT as DNA testing not available here. When all test results were insignificant for any condition, then termed as idiopathic neonatal hepatitis.

A preformed semi structured data collecting form was used as a data collection instrument. Data were collected by the researcher and analyzed by Statistical Package of Social Science (SPSS) version 11.5 programme.

Results

All the children were <1 year of age. Out of 80 children, 48 (60%) were intra-hepatic cholestasis & 32 (40%) were extra-hepatic cholestasis (Fig-1).

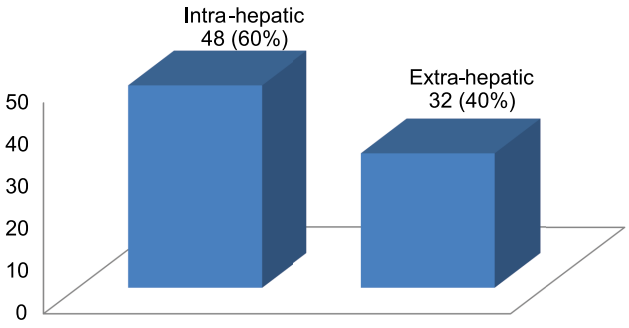

Fig 1 *Incidence of Intra-hepatic & Extra-hepatic cholestasis*

Among all children, biliary atresia (BA) 30 (37.5%) were the cause of cholestasis for most of the cases followed by idiopathic neonatal hepatitis (INH) 20 (25%) and TORCH infections 19 (23.7%) (Fig-2).

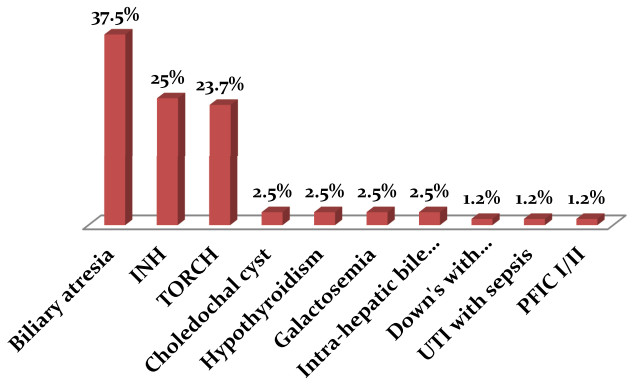

Fig 2 *Etiology of neonatal cholestasis*

Among 32 extra-hepatic cholestasis, biliary atresia (BA) 30 (93.7%) were the most common cause followed by choledochal cyst 2 (6.3%) (Fig-3).

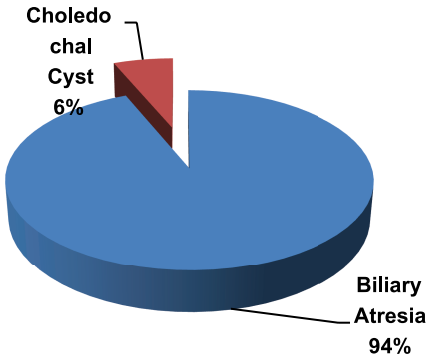

Fig 3 *Causes of Extra-hepatic cholestasis*

Among 48 intra-hepatic cholestasis, most common were idiopathic neonatal hepatitis 20 (41.6%) & TORCH infections 19 (39.5%). Among others, hypothyroidism, galactosemia and intra-hepatic bile duct paucity were found in 2 (4.1%) cases of each. Down's syndrome with hypothyroidism, UTI with sepsis and PFIC were found in 1 (2.0%) case of each (Fig-4)

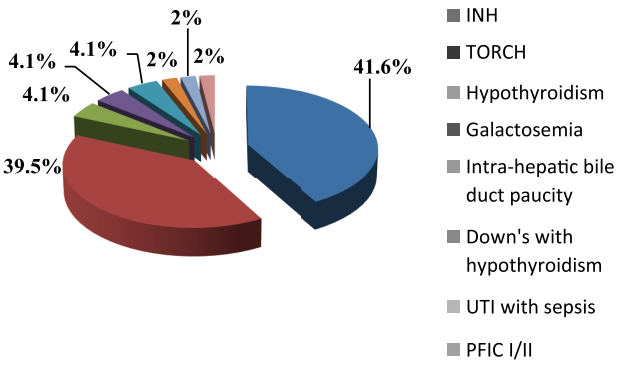

Fig 4 *Causes of Intra-hepatic cholestasis*

Among 19 TORCH infections, Cytomegalovirus (CMV) 13 (68.4%) were the most commonest followed by CMV with herpes-simplex virus (HSV) co-infection 4 (21.1%) and Toxoplasmosis 2 (10.5%) (Fig-5).

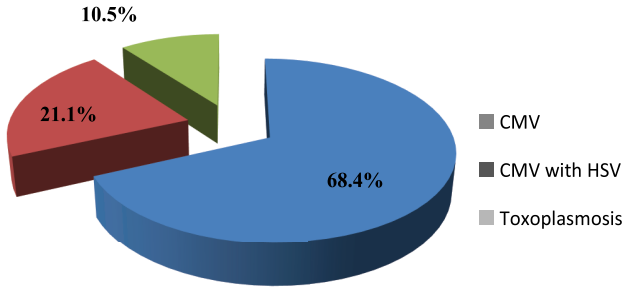

Fig 5 *Responsible viruses in TORCH infection*

Discussion

Diagnosis of cholestatic disorder is difficult because of lack of specificity of available diagnostic tests. On the other hand, early diagnosis & referral to the experienced centers is very important for proper management.[8] Many causes have been identified to cause injury to the newborn liver. These may include intrahepatic causes, which may be either sporadic or familial, and extrahepatic causes.[9]

In the present study from 80 infants, biliary atresia (BA) 30 (37.5%) were the most common cause of

neonatal cholestasis (NCS) followed by idiopathic neonatal hepatitis (INH) 20 (25%) & TORCH infections 19 (23.7%). Nahid et al[10] [BA-19 (31.6%), INH-19 (31.6%) & TORCH 13 (21.6%)], Karim et al[8] [(BA 19 (26.6%), INH 15 (24.2%) & TORCH 17 (27.4%)] from Bangladesh and Jain et al[11] [BA 41 (41%), INH-18 (18%) & neonatal hepatitis 20 (20%)] from India had similar findings. However, Hamid et al[12] were observed a different result where Neonatal hepatitis 18 (60%) were the most common followed by BA 12 (40%).

Among 32 cases of Extra-hepatic cholestasis, most of the infants suffering from BA 30 (93.7%) followed by choledochal cyst (CC) 2 (6.3%). From the desk of extra-hepatic cholestasis, Nahid et al.[10] [(BA 19 (90.5%) & CC 2 (9.5%)] also stated the similar result. BA is characterised by progressive fibrosing obliteration of both intra-hepatic and extrahepatic bile ducts. It is the most important cause of neonatal cholestasis worldwide, including Malaysia. It is also the most important indication for childhood liver transplantation the world over.[9]

Among 48 cases of Intra-hepatic cholestasis, INH 20 (41.6%) was the most frequent followed by TORCH infection 19 (39.5%). From 39 cases of intra-hepatic cholestasis, Nahid et al.[10] were observed INH 19 (48.7%) and TORCH infections 13 (33.3%). It has an incidence of 1:5000 births and constitutes approximately 50% of prolong neonatal jaundice.[13] Most patients in whom no aetiology was found were considered to have INH/transient neonatal cholestasis (TNC) by some authors. TNC was characterized by early-onset cholestasis, absence of a known cause of neonatal cholestasis, normalization of clinical and biochemical parameters during follow up and no history of some neonatal injurious events such as asphyxia, sepsis, total parenteral nutrition. As this study also had shown INH/TNC as one of the major reasons for cholestasis in infants.[7]

In 19 cases of TORCH infections, Cytomegalovirus (CMV) 13 (68.4%) were the commonest followed by CMV with herpes-simplex virus (HSV) co-infection 4 (21.1%) and Toxoplasmosis 2 (10.5%). Nahid et al[10]. [CMV 9 (69.2%, HSV 1 (7.1%), Toxoplasmosis 2 (14.2%)] and Hamid et al.[12] [(CMV 17 (73.9%, HSV 6 (26.1%)] of this country were stated the near similar result. A study in Srilanka, out of eight cases of intra-hepatic cholestasis 5 (62.5%) of them became positive for CMV infection.[7] Chang et al[13] also reported that most of their patients with neonatal hepatitis were due to CMV infection. It is important to have a well-organized and structured approach to detect TORCH related cholestasis, in particular CMV hepatitis. Because these were under.[7]

There are some endocrine, metabolic & genetic disorders may present with intra-hepatic cholestasis[13]. In our study among 48 cases of intra-hepatic cholestasis, isolated hypothyroidism in 2 (4.1%) and hypothyroidism with Down's syndrome in 1 (2%) case. Nahid et al.[10] were reported the near similar results among 39 intra-hepatic cases. Isolated hypothyroidism in 4 (10.2%) and hypothyroidism with Down's syndrome in 1 (2.5%) case. In another study on Bangladesh, Karim et al[8]. also observed hypothyroidism with down's syndrome and hypothyroidism with CMV infection in each 1 (2.3%) of cholestatic infant. In the present study, 2 (4.1%) cases of galactosemia among intra-hepatic cholestasis were diagnosed which were unlike in Nahid et al[10]. & Karim et al.[8] observation. This may due to using recent investigational approach like enzyme assay. Two (4.1%) cases of Intra-hepatic bile duct paucity (non-syndromic) were evaluated in the present study which were also evident in Karim et al.[8] [1 (1.6%)] observation. Luthufdeen et al[7]. were stated 2 (4.7%) infants of Alagille syndrome which was unlike our study. It may due to lack of genetic testing facility of our country. One (2%) case of Progressive Familial Intra-hepatic Cholestasis (PFIC) was diagnosed in our study which was like Nahid et al.[10] 1 (2.5%) study. Karim et al[8]. were reported 5 (11.9%) cases among 42 cases of neonatal cholestasis due to urinary tract infection (UTI) but we observed only 1 (2%) case of UTI with sepsis. It may due to increase awareness of parents as well as primary health care workers regarding early diagnosis and antibiotic use of any kind of infection. Tiker et al.[14] were reported that important cause of neonatal cholestasis was perinatal hypoxic-ischemia. Other studies have also identified the same factor of transient neonatal cholestasis. Bile secretion processes, which are already underdeveloped in neonates, can be further impaired by hepatic hypoxia-ischemia. No infants in our study having cholestasis with hypoxic-ischemic insult.

Limitations of study

The main limitation of the present study is that was a single-centre study with a limited sample size.

Conclusions

Biliary atresia, idiopathic neonatal hepatitis and TORCH infections were the commonest causes of neonatal cholestasis. Among extra-hepatic cholestasis biliary atresia and intra-hepatic cholestasis INH & TORCH infections were the commonest. In TORCH infections, CMV were the prominent one.

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ORIGINAL ARTICLE

Growth Pattern of Exclusively Breast-Fed Preterm Babies

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Abstract

Background: *Breast milk is uniquely adopted most appropriate natural, preferred food for all infants including premature newborns. Preterm infants have greater nutritional need to achieve optimal growth in the neonatal period than any other time of their life. To achieve this optimal growth of preterm, human milk is the gold standard nutrient, though there are some reports showing that growth of mixed fed preterm babies, are better than exclusively breast–fed preterm babies.*

Objective: *To assess the growth of both exclusive and mixed breast-fed preterm babies.*

Methods: *A prospective observational study was carried out in the special care baby unit of Dhaka Shishu Hospital from April 2009- May 2010. Growth pattern of exclusively breast–fed preterm and mixed fed preterm babies were recorded at 2 and 3 months.*

Results: *A total 94 preterm babies were enrolled at discharge and followed up. They were non-randomly selected. Mean weight of exclusively breast–fed preterm and mixed fed preterm babies respectively were (4676.53±653.45gm vs 4020.00±701.41gm), Length were (54.78±3.09cm vs52.96±4.50cm) and OFC were (37.84±2.07cm vs 37.09±1.14cm) at 3 months of age. Growth (weight, length and OFC) of exclusive breast fed preterm babies were also comparable with the recommended CDC growth chart findings.*

Conclusion: *Length, weight and OFC were increased significantly in 32-34 weekers exclusively breast-fed preterm babies in comparison to mixed fed babies.*

Key words: *Growth, exclusively breast-fed, preterm babies.*

Introduction

Prematurity and low birth weights are common global problems. The magnitude of preterm low birth weight infants in developing world is enormous. In 2004, about 8.1% of babies born in United States are LBW (<2500gm) and about 1.47% of babies were VLBW (<1500gm).[1] Globally about 15.5% of all births are born with LBW.[2] In Bangladesh prevalence of PT LBW is 22%.[3] In our country neonatal mortality rate is 28 per thousand live births and these deaths comprise 61% of all under-5 deaths. The neonatal mortality rate is nearly three times greater than post neonatal mortality.[4] In Bangladesh over the last 20 years reveals the neonatal mortality declined at a slower pace than infant and child mortality, with the result that neonatal deaths have changed from 60 percent of all infant deaths in 1993-1994 to 74 percent in 2010-2014.[4] Among them 31% is due to preterm births.[5] Premature mortality can be reduced by giving proper thermal care, nutrition, prevention of infection and other feasible short term interventions under the ENC and related programs of the country. Since the dawn of civilization, human milk has been regarded as the best and complete nourishment for a neonate. Nature has not produced any food as nutritiously appropriate and unique as human milk. Preterm infants have

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greater nutritional need to achieve optimal growth in the neonatal period than any other time of their life.[6] For preterm baby human milk is the gold standard nutrient.[7]The committee on Nutrition of the American Academy of Pediatrics recommends an energy intake of approximately 120cal/kg /day to enable most premature infants to achieve satisfactory growth rates.[8] A higher concentration of calories, proteins, fat and lower concentration of lactose has been reported in preterm milk as compared to term. The amount and composition of preterm milk is ideal to cover the needs of newborn infants.[9] Several studies conducted both in India and abroad have recommended the use of mother's milk for adequate growth and development of a preterm neonate.[10]

However, other study found that medium chain triglyceride content formula promoting short term growth of preterm infants.[11] Another study showed nutrient enriched post discharge formula in preterm infants was associated with improved short-term growth and potentially, with improved linear growth.[12] Infant mortality rate, Under 5 mortality rate in Pakistan and India is higher and Population growth rate and exclusive breast feeding rate is lower than Bangladesh. It indicates that probably Bangladesh have succeeded in controlling Infant mortality rate, Under 5 mortality rate and Population growth rate due to much higher rates of exclusive breast feeding.[13] Given the economic advantages, metabolic efficiency, immunological benefit and demonstration of higher protein and mineral content in the milk produced by mothers of preterm infants, breastfeeding from their own mothers appears to be the best option for these infants.[14] So this study was conducted to see the impact of growth of preterm baby who were on exclusive breast feed up to 3 months of age and compare growth parameters of them with mixed fed preterm babies with standard CDC growth chart.

Materials and Methods

This was a prospective observational study conducted at Special Care Baby Unit (SCABU) of Dhaka Shishu (Children) Hospital from May 2009 to April 2010. Purposive sampling procedure was done. Less than twenty day's old babies whose gestational age was 32 -34 weeks and in exclusive breast feeding were enrolled in this study. Preterm babies who were severely sick (congenital anomaly, perinatal asphyxia, any heart disease, septicemia etc.) were excluded from the study. Selection criteria of control were 32-34 weeker babies within 20 days who were in mixed feeding.

Data were collected in a pre formed questionnaires. Gestational age of all the babies were determined by maternal record (Maternal recall of LMP or available Ultra-sonogram reports) and by New Ballard Score System. Birth weight was recorded from the accompanied document. Weight was taken with accuracy and precision by keeping the baby undressed and before feeding by the registered neonatal nurse. Crown-heel length was measured by using an infantometer to the nearest 1mm.Occipito-frontal circumference (OFC) was measured with an inelastic standard plastic measuring tape to the nearest 1mm. Sixty exclusive breast fed babies and fifty mixed fed babies were enrolled. Among the exclusive breast fed group 1 baby did not come and 6 babies changed their feeding pattern. So on 1st visit at the age of 2 months of their age 53 babies were in exclusive breast fed group. During 2nd visit at the age of 3 months they were 49 in number. Two babies did not come for 2nd visit and 2 babies started mixed feeding. In mixed fed group 47 babies came on the 1st visit, as 3 babies did not come and on the 2nd visit another 2 babies did not come. So 45 babies were enrolled in mixed fed group. Finally 49 babies in exclusive breast fed group and 45 babies in mixed fed group who fulfill the criteria were enrolled. On admission and at 2 months and 3 months baseline parameter (weight, length, OFC) was assessed. Growths of exclusive breast fed preterm babies were compared with the standard CDC growth chart. Statistical analysis were done by SPSS program (12 versions).The results were presented in tables. For significant differences unpaired student t' tests were done. A probability 'p' value < 0.05 was considered as significant.

Results

Hundred ten preterm babies were enrolled for study and follow up. Sixteen neonates were dropped out from the study. Finally 94 preterm neonates completed the total follow up schedule. Forty nine were in exclusive breast fed group and forty five were in mixed fed group.

Table I

Anthropometry of patient at enrollment (n =94)

Group	Length(cm) Mean ± SD	Weight(gm) Mean ± SD	OFC(cm) Mean ± SD	p value
Exclusive Breast fed (n=49)	42.86 ± 3.35	1748.57 ±419.67	30.63 ± 2.05	
Mixed fed (n=45)	41.78 ± 3.11	1613.56 ±282.50	30.52 ± 1.36	

Table II

Distribution & comparison of length, weight and OFC between exclusive & mixed fed infants at 1st follow up

Parameter	Group	Mean ± SD	t-value	p value
Length (cm)	Exclusive(n=49)	49.94±2.88	2.49	<0.05
	Mixed(n=45)	48.53±2.56		
Weight (gm)	Exclusive(n=49)	3443.88±685.69	3.75	<0.001
	Mixed(n=45)	2944.89±596.54		
OFC(cm)	Exclusive (n=49)	35.02±1.85	-.075	>0.05
	Mixed (n=45)	35.04±1.21		

Table III

Distribution & comparison of length, weight and OFC between exclusive & mixed fed infants at 2nd follow up

Parameter	Group	Mean ± SD	t-value	p value
Length (cm)	Exclusive(n=49)	54.78±3.09	2.27	<0.05
	Mixed(n=45)	52.96±4.50		
Weight (gm)	Exclusive(n=49)	4676.53±653.45	4.70	<0.001
	Mixed(n=45)	4020.00±701.41		
OFC(cm)	Exclusive (n=49)	37.84±2.07	2.19	<0.05
	Mixed (n=45)	37.09±1.14		

Table IV

Distribution of study samples (Weight. length, OFC) of exclusive fed preterm at last visit in relation to recommended growth chart findings (50th percentile)

Samples (Mean)	Exclusive (n=49)		Mixed (n=45)	
	Study finding	Recommended growth chart findings (50th percentile)	Study finding	Recommended growth chart findings (50th percentile)
Length of infants (cm)	54.78	49.42	52.96	49.42
Weight of infants (gm)	4676.53	3266.67	4020.00	3266.67
OFC of infants (cm)	37.84	34.50	37.09	34.50

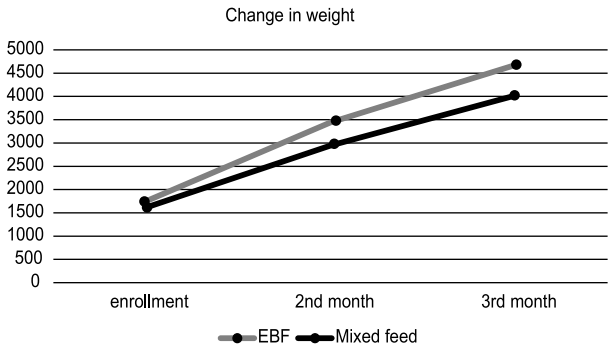

Fig 1 *Comparison of weight between exclusive & mixed fed infants*

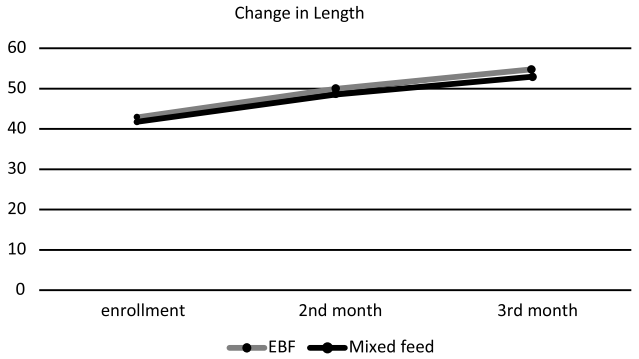

Fig 2 *Comparison of length between exclusive & mixed fed infants*

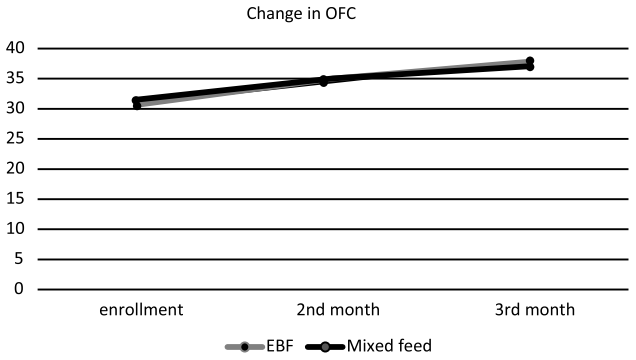

Fig 3 *Comparison of OFC between exclusive & mixed fed infants*

Discussion

This study showed male preponderance. Among 94 patients 54 (57.4%) were male and 40 (42.6%) were female. In this study growth pattern of preterm were seen. Mean weights were seen between exclusive breast-fed and mixed fed infants at 1st and 2nd follow up. Here mean weight of exclusive breast-fed infants at 1st and 2nd follow up was highly significant ($p<0.001$). This finding partially supported by a randomized study where they found that there were no significant growth advantages with fortification; although, when breast milk exceeded 50% of intake, fortification promoted faster weight gain.[14] Length was higher in exclusive breast-fed infants than mixed fed infants with statistical significant ($p<0.05$) in both follow up. OFC was also statically significant ($p<0.05$) in exclusive fed infants in 2nd follow up. This result was almost similar to the results of Lucas *et* al.[14] Similar study conducted by Chan et al[15] noted weight, length and OFC were similar for human milk and formula feeder at discharge from hospital. However RÕnnholm et al[16] said weight and length growth velocity was high in the fortifier fed infant. Exclusive breast-fed preterm babies gained their weight, length and OFC much higher than recommended growth chart findings in 50th percentile. All of them were above 50th percentile in their weight, length and OFC. LBW preterm infants on being exclusively breast-fed by their own mother's, gained weight and had an increase in their OFC and length to the levels almost comparable to the standard fetal infant growth norms said Sing et al.[17] Muhudhia et al[18] noticed that the weight gain occurred significantly in preterm after initial loss of weight in exclusively breast fed preterm and more in 1000-1250gm preterm. Lubetzky et al[19] found preterm infants have lower energy expenditure when they are fed breast milk than when they are fed preterm infant formula.

Howtever Lucas et al[20] found that breast milk fortifiers can improve short-term growth (when breast milk intakes are high) but beneficial effects on long term development remained unproven. In 2009 Evely et al[14] found similar result. Berseth et al[21] mentioned that human milk fortifiers are safe, well tolerated and facilate comparable good growth. Arslanoglu et al[22] also found premature infants with the new adjustable fortification regimen had significantly higher weight and head circumference gains than infants managed with standard fortification. However O Connor et al[23] found that despite a slower early growth rate, human milk fed LBW infants have development at least comparable to that of infants fed nutrient enriched formula. Breast feeding protects the newborn from infectious diseases. This is especially important for very LBW preterm infants, whose immune systems are immature. Tarcan et al[24] found the mean number of CD3-CD16/56 cells in the formula fed infants was significantly lower than the corresponding means in the groups fed human milk alone. So, chance of complication developed more in mixed fed infants. Bier et al[25] findings were showed ingestion of human milk post discharge is associated with a reduction of upper respiratory symptoms in infants during their 1st year of life. Some authors highlighted the higher risk of NEC in newborns who are fed fortified milk due to the increase in osmolality may result in the reduction in nutrient intake which can explain lower weight gain.[26]

It is evident from this study that growths of the exclusive breast-fed preterm babies are fairly satisfactory. The findings could have important implication on a reduction of prematurely related morbidity and mortality in developing countries like Bangladesh. Hopefully, breast feeding will play an important role for ideal growth and development of a preterm baby. Here non breast-fed group was not considered as control group.

Conclusion

Length, weight and OFC were increased significantly in 32-34 weekers exclusively breast-fed preterm babies in comparison to mixed fed babies.

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ORIGINAL ARTICLE

Causes of Hypertension Among Adolescents in A Tertiary Hospital of Bangladesh

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Abstract

Background: *High blood pressure in children and adolescents is a growing health problem that is often overlooked by physicians.The current prevalence of hypertension in children is estimated to be about 1-5%, with higher rates among minority adolescents. Primary or essential hypertension is more common in adolescents and has multiple risk factors, including obesity and a family history of hypertension. Early detection and intervention in children and adolescents with hypertension are potentially beneficial in preventing long-term complications of hypertension.*

Methods: *This retrospective study was carried out among the patients admitted under the department of adolescent pediatric medicine, Dhaka Shishu (Children) Hospital, over a period of 1 year & 6 months from 01st October 2015 to 31st March 2017.*

Results: *Among 54 study population, male were 40(74.07%) and female 14(25.93%)with male female ratio 2.9:1.Among the adolescents 61.11% were from urban area and 38.89% were from rural. Most of the adolescent presented with AGN 25(46.30 %) followed by Nephritic Nephrotic Syndrome 9(16.67%), Obesity related HTN 7(12.97%), HSP with nephritis 5(9.26%), Nephrotic Syndrome 3(5.56%), GBS with HTN 2(3.7%), Renal artery stenosis 2(3.7%), Essential Hypertension 1(3.7%).*

Conclusion: *Hypertension is a growing health problem among adolescents. Much higher incidence of secondary hypertension is seen among adolescents. Renal parenchymal diseases and obesity accounts for most of the secondary hypertension.*

Key words: *Hypertension, adolescents.*

Introduction

It is well established that high BP can be identified in children and adolescents.[1-3] High blood pressure in children and adolescents is a growing health problem that is often overlooked by physicians. The current prevalence of hypertension in children is estimated to be about 1-5%, with higher rates among minority adolescents.[4-6] In persons 3 to 18 years of age, the prevalence of prehypertension is 3.4 percent and the prevalence of hypertension is 3.6 percent.[7] The combined prevalence of prehypertension and hypertension in adolescents who are obese is greater than 30 percent in boys and is 23 to 30 percent in girls.[5] The prevalence and

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rate of diagnosis of hypertension in children and adolescents appear to be increasing.[4] In a cross-sectional study limited to the adolescence agein a cohort of 6,790 high school students (11-17 years), the prevalence of hypertension was found 3.2% and the prevalence of pre-hypertension was 15.7% in adolescence.[8] Among adolescents with high risk BP values, including those designated from a single measurement as having pre-hypertension and hypertension combined, 68% of boys and 43% of girls had developed pre-hypertension or hypertension 2 years later.[9]In the study on high school students by McNiece et al[5] the prevalence of hypertension and pre-hypertension combined was over 30% in obese boys and from 23-30% in obese girls, depending on ethnicity.Within the National Childhood Blood Pressure database, a segment of adolescents underwent BP measurement at intervals of 2 years and 4 years. An analysis of these data found that, among adolescents with pre-hypertension, 14% had developed hypertension 2 years later, which yielded an approximate incidence rate of 7% per year.[10] Primary or essential hypertension is more common in adolescents and has multiple risk factors, including obesity and a family history of hypertension. Overweight and obesity are strongly correlated with primary hypertension in children.[11] Secondary hypertension is more common in preadolescent children, with most cases caused by renal disease. There is evidence that childhood hypertension can lead to adult hypertension.[12] Based on these observations, early detection of and intervention in children with hypertension are potentially beneficial in preventing long-term complications of hypertension.

Material and Methods

This retrospective study was carried out among the patients admitted under the department of adolescent pediatric medicine, Dhaka Shishu (Children) Hospital, over a period of one year & six months from 01st October 2015 to 31st March 2017. During this period 1080 patient was admitted in adolescent unit. B.P. of each patients were recorded routinely as apart of general examination. Hypertension was defined as systolic and/ or diastolic BP greater than the 95th percentile for age, gender and height on three or more separate occasions. Based on this definition, out of 1080 adolescents 54 patient had hypertension. These 54 hypertensive adolescents were included in this study. Each patient was evaluated carefully. A detailed history including presenting complaints, duration of symptoms, socio-economic status, demographic profile, developmental, nutritional, personal & family history were recorded. Specialist consultation like nephrologist, cardiologist, nutritionist, were taken whenever it was required. Necessary routine laboratory investigations and relevant special investigations like renal function test, CXR, ECG, ECHO, Lipid profile, droplller studies of cardiac and renal vessels , etc. were performed when indicated. The data were recorded on a semi-structured manner and statistically analyzed with the one of the statistical package of social science program (SPSS version 15.0 for windows, Chicago, IL).

Results

Out of 1080 total 54(5%) adolescents were admitted with Hypertension in Adolescent Medicine Unit of Dhaka Shishu(Children) Hospital from 1st October 2015 to 31st March 2017. Among these 40(74.07%) were male and 14(25.93%) were female with male female ratio 2.9:1 (Fig. 1).

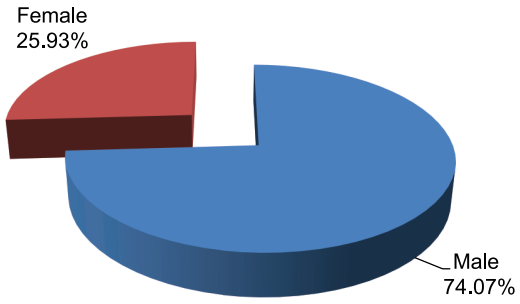

Fig 1 *Pie diagram of the respondents by sex (n=54)*

Out of 54 cases 61.11% Adolescent were from urban area and 38.89% were from rural area (Fig. 2).

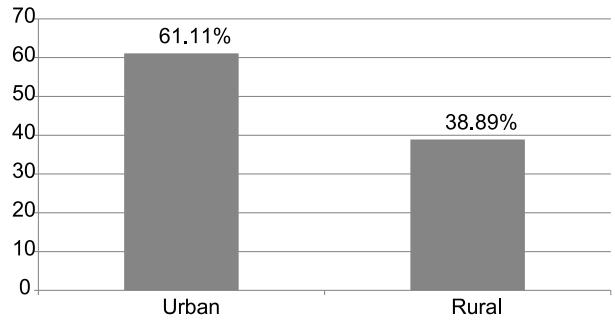

Fig 2 *Bar diagram of respondents according to residential area (n=54)*

Among hypertensive adolescents, 25(46.30 %) were diagnosed as AGNfollowed by Nephritic Nephrotic Syndrome 9(16.67%), Obesity related HTN 7(12.97%), HSP with nephritis 5(9.26%), Nephrotic Syndrome 3(5.56%), GBS with HTN 2(3.7%), Renal artery stenosis 2(3.7%) and Essential Hypertension 1(3.7%) [Fig. 3].

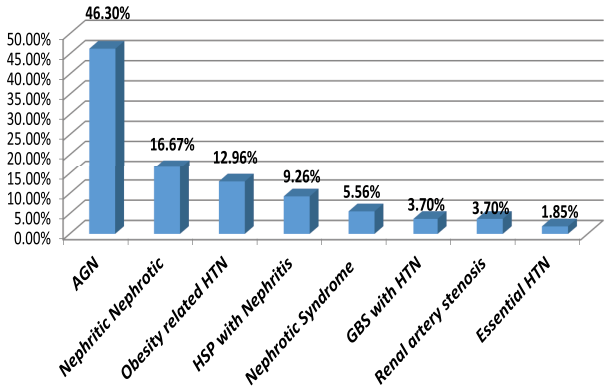

Fig 3 *Bar diagram of Diagnostic pattern Hypertensive adolescent. (n=54)*

Among male adolescent,15(37.5%)were diagnosed as AGN followed by Nephritic Nephrotic Syndrome 9(22.5%), Obesity related HTN 7(17.5%), HSP with nephritis 3(7.5%), Nephrotic Syndrome 3(7.5%), Renal artery stenosis 2(5.0%) and Essential Hypertension 1(2.5%)[Fig. 4].

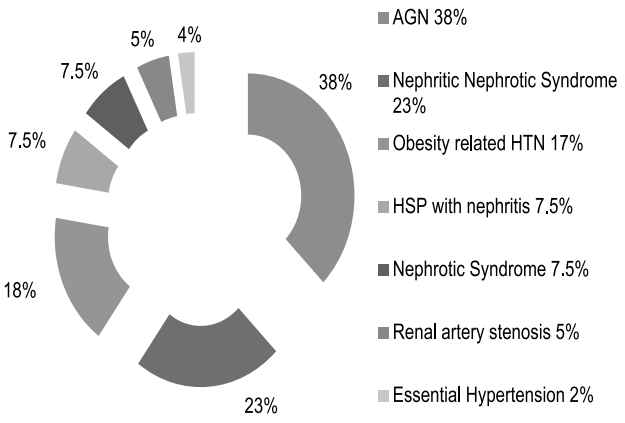

Fig 4 *Pie diagram of Diagnostic pattern Hypertension in male adolescent (n=40)*

Among female adolescents10 (71.40%) were diagnosed as AGN, 2(14.28%) GBS with HTN and 2(14.28%) HSP with Nephritis (Fig. 5).

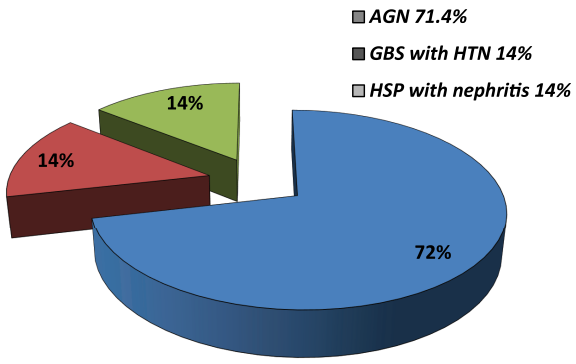

Fig 5 *Pie diagram of Diagnostic pattern Hypertension in female adolescent (n=14)*

Discussion

In children, the prevalence of hypertension is estimated to be between 2% and 5%, and the prevalence of prehypertension between 4% and 15%,[4,5] but studies in both the United States and Europe have found that only 13% to 26% of childhood hypertension is properly diagnosed.[13-17] Children with primary hypertension may be asymptomatic or the symptoms may be mild and seemingly unrelated, such as changes in behavior or school performance, nosebleeds, headaches, or shortness of breath.[18]The current prevalence of hypertension in children is estimated to be about 1-5%, with higher rates among minority adolescents.[4-6] In this present study 5% adolescents were admitted with hypertension. In the study by Anochie et al[19] among 123 adolescent, male and female ratio was 1.1:1. In another study by Malla et al[20], the proportion of male was higher with the male female ratio 1.6:1.Like the above studies**,** this study shows male preponderance with male 74.07%. In children with hypertension, 30% to 60% have secondary hypertension, while 40% to 70% have primary hypertension.[21] Children and adolescents have a much higher incidence of secondary hypertension compared with adults.[1,4,22-25] Younger children or children with stage 2 hypertension are more likely to have secondary hypertension, whereas primary hypertension becomes more prevalent in adolescence and young adulthood.[22]Renal parenchymal disease and renal vascular diseases account for most cases of secondary hypertension.[26-27]In this study most of the adolescents with hypertension were diagnosed to have renal disorders, AGN 46.30 %, Nephritic Nephrotic Syndrome 16.67%, HSP with nephritis 9.26%, Nephrotic Syndrome 5.56%, Renal artery stenosis

3.7%. Acute glomerulonephritis is a common renal disease in children and adolescents. Yuniarchan et al[28] study found 101 pediatric AGN cases admitted over a period of 4 years. In a study by Elzouki et al[29] in eastern Libya, the major renal disease were post streptococcal glomerulonephritis. Similarly in our study 25 AGN cases were admitted over a period of 1 year and 6 month. Primary hypertension (PH) also referred to as essential hypertension, previously considered a disease of adulthood, has now become increasingly common in the pediatric population largely due to the obesity epidemic.[30,31]According to Bernstein et al[32] essential hypertension is the most common form of hypertension in adults and it is recognized more often in adolescents than in younger children. In the present study,3.7% adolescent was diagnosed to have essential hypertension. In the study by Boyedetal[24] hypertension was found in 2.1% cases in HSP with nephritis. In our study, HSP with nephritis was 3.7%.Studies found that obese children are three times more likely to develop hypertension than their non-obese counterparts.[33,34] The relationship between obesity and hypertension has been clearly defined in multiple studies across different ethnic and gender groups.[32-39] The etiology of obesity related hypertension has been linked to sympathetic hyperactivity, insulin resistance and vascular structure changes.[40-41]Sorof et al[34] demonstrated the presence of sympathetic nervous system hyperactivity in obese school age children, evidenced by increased heart rate and blood pressure variability which contributed to the pathogenesis of isolated systolic hypertension. Increased sodium content of the cerebrospinal fluid has been shown to increase sympathetic nervous system activity through activation of the renin-angiotensin-aldosterone pathway in the brain.[40,41] Obese individuals have selective insulin resistance, which leads to increased sympathetic activity and alteration of vascular reactivity and resultant sodium retention as evidenced by decreased urinary sodium excretion.[42] The present study reveals that 12.97% adolescents had Obesity related HTN. Stapletonet al[43] report a case of a child with GBS and hypertension that appeared to be related to increased renin-angiotensin activity and Propranolol therapy successfully controlled the hypertension. In another study by Mitchel et al[44] the mechanism of hypertension in a nineteen year old girl with a polyradiculitis (Guillain-Barré syndrome) was investigated.They found an increase in circulating catecholamines, and the results of the studies suggested a pathological alteration of the sympathetic nervous system, whereby high levels of pressor amines were produced, as the mechanism of hypertension. Minami et al[45] report a case of hypertension associated with Guillain-Barré syndrome where the circadian variation of blood pressure was interrupted and examination of neurohumoral factors revealed a hyperactive sympathetic nervous system and an increase in plasma renin activity.These observations suggest that autonomous hyperactivity of the efferent pathway of the sympathetic nervous system may cause the sustained hypertension throughout the day in case of Guillain-Barré syndrome. In the current study 3.7% adolescents hadGBS with HTN.

Several challenges regarding hypertension now confront clinicians who care for children and adolescents. These include detecting hypertension, distinguishing secondary hypertension from primary hypertension, examining patients for hypertension-related risk factors and target organ damage, applying interventions to control blood pressure, and encouraging preventive lifestyle. Better BP monitoring in children and adolescents in an attempt to prevent the long-term complications of hypertension is of utmost important. More research efforts in pediatric and adolescent population with hypertension are needed in a developing country like Bangladesh.

Conclusion

Hypertension is a growing health problem among adolescents. Much higher incidence of secondary hypertension is seen among adolescents. Renal parenchymal diseases and obesity accounts for most of the secondary hypertension.

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DS (Child) H J 2016; 32(1) : 38-41

ORIGINAL ARTICLE

Outcome of T Tube Ileostomy with N Acetyl Cysteine Irrigation For The Management of Uncomplicated Meconium Ileus

Md. Samiul Hasan[1], M Kabirul Islam[2], Ayub Ali[3], Sabbir Karim[3]

Abstract

Background: *Meconium ileus is a common cause of neonatal intestinal obstruction. Various surgical procedures are in practice for uncomplicated meconium ileus. T tube ileostomy seems to offer important advantages over surgical techniques previously described.*

Objective: *Objective of this prospective interventional study was to evaluate the outcome of T-tube ileostomy for the treatment of uncomplicated meconium ileus.*

Methods: *It was a prospective interventional study, held in department of surgery, Dhaka Shishu (Children) Hospital from January 2015 to December 2016. Total 21 neonates were included after fulfilling selection criteria. After T tube ileostomy all patients were followed up for 6 weeks post-operatively. Data were collected in a predesigned, semi structured questionnaire and outcome of all patients were noted.*

Result: *The age range of the patients was 1 to 6 days where majority were male. Mean operation time was 60.76±5.81 minutes. After operation, mean time of bowel movement, establishing oral feeding and tube removal was 4.90±1.41days, 6.35±1.27 days and 8.10±1.45 days respectively. One neonate had intraperitoneal leakage and died of sepsis. Mean hospital stay was 9.45 1.31 days. Stomas closed spontaneously in all survived neonates.*

Conclusion: *T-tube ileostomy with N acetyl cysteine irrigation is a safe and effective procedure for uncomplicated meconium ileus.*

Key Words: *Meconium ileus, T-tube ileostomy.*

Introduction

Meconium ileus is a common cause of neonatal intestinal obstruction. Here obstruction occur secondary to the intraluminal accumulation of inspissated and desiccated meconium. About 50% patients present with complication like volvulus, atresia, perforation and meconium cyst.[1] In the past, meconium ileus was considered to be closely associated with cystic fibrosis (CF). However, recent studies demonstrate that meconium ileus occurs frequently in the absence of CF as well.[2] Uncomplicated meconium ileus can be treated with therapeutic contrast enemas as described by Noblett et al.[3] Several complications have been reported following Gastrografin enemas which are perforation, necrotizing enterocolitis, shock and occasional death. Therefore, Noblett et al[3] set some criteria to be fulfilled before performing this procedure. Copeland et al[4] reported effectiveness of this procedure decreased and role of contrast enema is diminishing. In our setup fluoroscopy and neonatal surgical ICU care are not available, so all patients with meconium ileus undergo surgical management.

Options for surgical management of uncomplicated meconium ileus include resection of dilated ileum and Bishop-Koop ileostomy, Santulli procedure or

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Mikulicz procedure. These are extensive operation, associated with gut reduction and high stoma output. A second surgery also required to close the stoma.[1] Tube enterostomy without gut resection was performed by O'Neill which was modified by Harberg et al using T tube.[5] T-tube ileostomy has several advantages. This procedure do not require gut resection and there is no intraperitoneal anastomosis. After extraction of T-tube the wound heal spontaneously and second operation to close the stoma is not required.[6] The aim of this study was to evaluate the outcome of T-tube ileostomy for the treatment of uncomplicated meconium ileus.

Materials and Methods

This was a hospital based prospective interventional study, conducted in the division of pediatric surgery, Dhaka Shishu (Children) Hospital, Dhaka, from January 2015 to December 2016. Sample size was 21. Preterm, low birth weight and patients with multiple congenital anomalies were excluded. T tube ileostomy was performed in all patients. Patients were followed up for 6 weeks after operation. Data were collected in a pre-designed, semi-structured questionnaire, after taking consent from guardians in the consent form. Ethical clearance was taken from hospital ethical committee. After confirmation of diagnosis at laparotomy, an enterotomy was made on antimesenteric border of ileum, 3–4 cm proximal to distal narrow segment. Thick meconium and pellets were evacuated through the enterotomy with minimal bowel handling. A 14/16 Fr T-tube was inserted through the enterotomy and was secured with double purse string suture with catgut (Fig.-1).

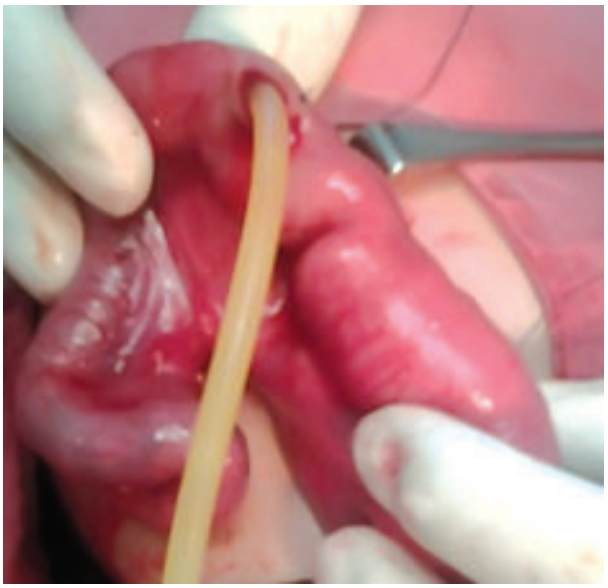

Fig 1 Insertion of T tube

Irrigation was given with saline and 2% N-acetyl cysteine. The T-tube was brought out through a stab incision in the right iliac fossa and enterostomy was secured to the anterior abdominal wall (Fig.-2).

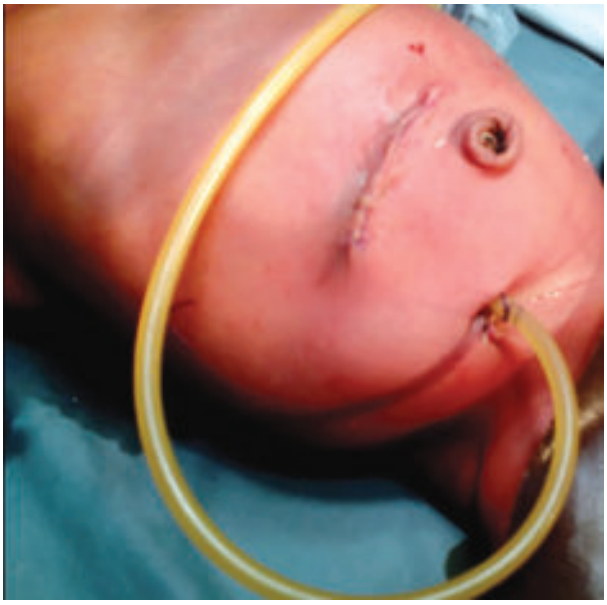

Fig 2 *Fixation of T tube*

T tube was irrigated with 10 ml of 2% N acetyl cysteine starting from 1st postoperative day until spontaneous bowel movement establishes.

Results

Out of 21 neonates 13 (61.9%) were male and 8 (38.1%) were female (Fig 1). Age range was 1 to 6 days and weight range was 2.5 to 2.9 kg (Table-I). Mean operation time was 60.76±5.81 min (range 53-68 min). Bowel movement started on an average 5th post operative day (range 4-7 day). Oral feeding was started after establishment of bowel movement. Tube removed on an average 8th post operative day. Hospital stay ranged from 7 to 12 days. One patient had intraperitoneal leakage on 4th post operative day. This patient died after second laparotomy. Stoma closed spontaneously (Photograph 3) in all patients (n=20) survived within 6 weeks (Table-II).

Table I
Age & weight distribution of study population (n=21)

Variable	Range	Mean ± SD
Age (day)	1-6	3.67±1.49
Weight (Kg)	2.5 – 2.9	2.68 ±0.12

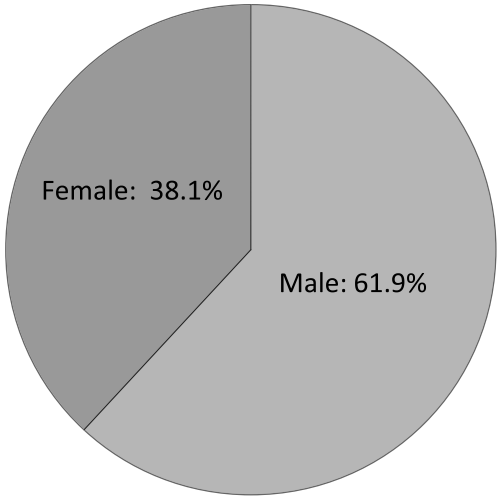

Fig 3 Sex distribution of study population (n=21)

Table II
Outcome of T tube ileostomy

Variables	Range	Mean ± SD
Operation Time	53-68 min	60.76±5.81 min
Time to start bowel movement	4-7 days	4.90±1.41 days
Time to start oral feeding	4-9 days	6.35±1.27 days
Tube removal	7-12 days	8.101.45 days
Hospital stay	7-13 days	9.45 1.31 days
Complication	1 (Intraperitoneal leakage)	
Spontaneous closure of stoma	All (within 6weeks)	

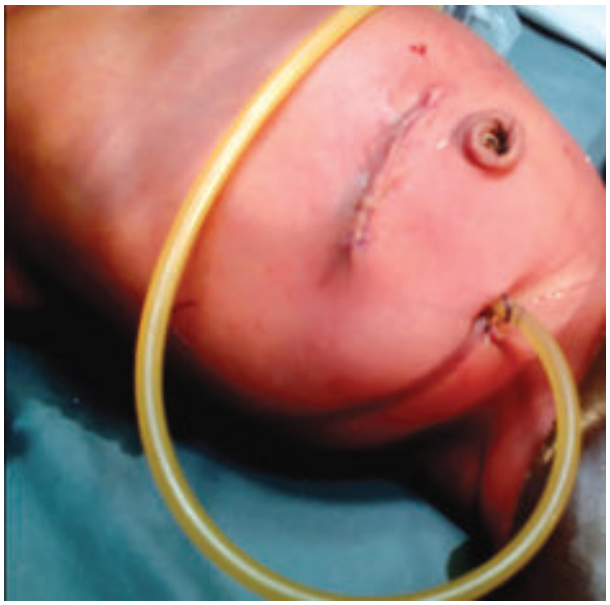

Fig 4 *Six weeks after removal of T tube*

Discussion

Percentage of male neonates was higher than female. Similar result was found by Venugopal et al[7] but Copeland et al[4] found opposite. No difference in the gender was described by Ciprandi et al.[8]The mean operation time was relatively shorter than other procedure used for uncomplicated meconium ileus. Bhattacharyaya et al. stated T tube ileostomy requires less time compared to the other methods.[9] Operations were performed by three different surgeons. Time may vary if performed by single surgeon.Tube irrigation was given with 2% N-acetyl cystine 2 times daily from 1st post operative day. Bhattacharyaya et al[9] also used 2% N-acetyl cysteine, whereas Mak et al[10] used N-acetyl cystine in 50% cases and pancreatic enzyme in 50% cases for tube irrigation. Venugopal et al[7] used 5% N acetyl cysteine for irrigation. Bhattacharyaya et al[9] reported bowel movement on approximately 7th (range 5th to 9th) post operative day and Mak et al[10] reported it on 5th (range 2 -12 days) day. These finding was almost similar with this study. Timing of oral feeding, tube removal and hospital stay matched with Harberg et al[5] Mak et al[10] and Bhattacharyaya et al. [9]

One neonate had intraperitoneal leakage and died after second laparotomy. No complication reported by Millar et al[6] and Bhattacharyaya et al.[9] Mak et al[10] reported persistent obstruction in 3 patients out of 23. Ziegler showed survival of neonates with meconium ileus is now approaching 95% to 100% and the credit goes to optimum surgical, pulmonary and nutritional care.[1] Preterm, low birth weight and neonates with other congenital anomalies were excluded from the study which might influence the morbidity and mortality.

Spontaneous closure of stoma occurred in all patients survived with T-tube ileostomy. This finding is similar with the study of Millar et al,[6] Bhattacharyaya et al,[9] and Mak et al.[10] This avoids a second laparotomy to close stoma in a compromised child.

Conclusion

T tube ileostomy with N acetyl cysteine irrigation is a safe and effective procedure for uncomplicated meconium ileus. After removal of tube wound heals spontaneously, which also reduce reoperation rate.

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REVIEW ARTICLE

Chikungunya Fever: A Vector Borne Morbid Disease

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Abstract

The term "Chikungunya" often refers to both the virus (CHIKV) and the illness or fever (CHIKF) caused by this virus. Chikungunya is an arthropod-borne self-remitting febrile viral illness viral disease caused by the chikungunya virus (CHIKV). First described during an outbreak in southern Tanzania in 1952, the virus derives its name from the Makonde language and means "to become contorted" or "that which bends up." The disease shares some clinical signs with dengue and zika, and can be misdiagnosed in areas where they are common. The proximity of mosquito breeding sites to human habitation is a significant risk factor for chikungunya. The disease mostly occurs in Africa, Asia and the Indian subcontinent. However a major outbreak in 2015 affected several countries of the Region of the Americas. The median incubation period is 2 to 4 days. Vertical transmission of disease from mother to child has also been documented. Clinical manifestations are very variable, from asymptomatic illness to severe debilitating disease. Children are among the group at maximum risk for severe manifestations of the disease and some clinical features in this group are distinct from those seen in adults. Common clinical features include: abrupt onset high grade fever, skin rashes, minor hemorrhagic manifestations, arthralgia/ arthritis, lymphadenopathy, conjunctival injection, swelling of eyelids and pharyngitis. Unusual clinical features include: neurological manifestations including seizures, altered level of consciousness, blindness due to retrobulbar neuritis and acute flaccid paralysis. Watery stools may be seen in infants. There is no cure for the disease. Treatment is focused on relieving the symptoms. Treatment is symptomatic. Generally non-steroidal anti-inflammatory drugs are avoided. Paracetamol may be used for pain and fever. However, NSAIDS may be required for relief of severe arthralgia during convalescent phase.

Key Words: *Chikungunya; virus; arthropod-borne; self-remitting*

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History/Background

The word 'chikungunya' is believed to have been derived from a description in the Makonde language, meaning "that which bends up", of the contorted posture of people affected with the severe joint pain and arthritic symptoms associated with this disease.[1] The disease was first described by Marion Robinson[2] and W.H.R. Lumsden[3] in 1955, following an outbreak in 1952 on the Makonde Plateau, along the border between Mozambique and Tanganyika (the mainland part of modern-day Tanzania). According to the initial 1955 report about the epidemiology of the disease, the term 'chikungunya' is derived from the Makonde root verb kungunyala, meaning to dry up or become contorted. In concurrent research, Robinson glossed the Makonde term more specifically as "that which bends up."[4]

Since its discovery in Tanganyika, Africa, in 1952, chikungunya virus outbreaks have occurred occasionally in Africa, South Asia, and Southeast Asia, but recent outbreaks have spread the disease over a wider range. The first recorded outbreak of this disease may have been in 1779.[5] This is in agreement with the molecular genetics evidence that suggests it evolved around the year 1700.[6]

Though known for many decades, the disease has got renewed attention because of widespread occurrence of outbreaks across India. Between the outbreaks of chikungunya fever the virus may be senescent for several years to a few decades. This variability in the disease occurrence has been attributed to many factors including – the occurrence of a sylvatic cycle, varying susceptibility to the virus among its human, animal and insect hosts and, changes in breeding conditions and vector density. Globalization also facilitates the introduction of the virus from endemic areas to new localities.[7]

This typical pattern of variability is seen in India as well. The disease was first reported in India in 1963-64 in Kolkata and in the later half of 1964 in Madras. Over 3 lakh people were affected during this outbreak. In 1973, an epidemic of chikungunya occurred in Barsi in Maharashtra. After this there was a period of respite till 2005 as there were no reported cases for the next 32 years.[8]

The present outbreak in India began in end 2005 along the coasts of Andhra Pradesh. It subsequently affected Tamil Nadu, Karnataka, Kerala and north India as far as Delhi. The disease was reported in a total of 16 states of the country in 2006 and within a period of 10 months of onset of the epidemic there were over 1.25 million suspected cases on chikungunya (the figure rising to 1.39 million by the end of the year). In 2007, till October there were 37,683 reported cases.[9]

The currently ciculating strain of the virus is of the east/central African genotype and Aedes aegypti continues to be the major vector.[10]

Epidemiology

Historically, chikungunya has been present mostly in the developing world. The disease causes an estimated 3 million infections each year.[11] Epidemics in the Indian Ocean, Pacific Islands, and in the Americas, continue to change the distribution of the disease[12].In Africa, chikungunya is spread by a sylvatic cycle in which the virus largely cycles between other non-human primates, small mammals, and mosquitos between human outbreaks. During outbreaks, due to the high concentration of virus in the blood of those in the acute phase of infection, the virus can circulate from humans to mosquitoes and back to humans.[13]

The transmission of the pathogen between humans and mosquitoes that exist in urban environments was established on multiple occasions from strains occurring on the eastern half of Africa in non-human primate hosts. This emergence and spread beyond Africa may have started as early as the 18th century. Currently, available data does not indicate whether the introduction of chikungunya into Asia occurred in the 19th century or more recently, but this epidemic Asian strain causes outbreaks in India and continues to circulate in Southeast Asia.[14]

In Africa, outbreaks were typically tied to heavy rainfall causing increased mosquito population. In recent outbreaks in urban centers, the virus has spread by circulating between humans and mosquitoes.[15]

Global rates of chikungunya infection are variable, depending on outbreaks. When chikungunya was first identified in 1952, it had a low-level circulation in West Africa, with infection rates linked to rainfall. Beginning in the 1960s, periodic outbreaks were documented in Asia and Africa.However, since 2005, following several decades of relative inactivity, chikungunya has re-emerged and caused large outbreaks in Africa, Asia, and the Americas. In India, for instance, chikungunya re-appeared following 32 years of absence of viral activity.[16]

Outbreaks have occurred in Europe, the Caribbean, and South America, areas in which chikungunya was not previously transmitted. Local transmission has also occurred in the United States and Australia, countries in which the virus was previously unknown.[15]

An analysis of the chikungunya virus's genetic code suggests that the increased severity of the 2005–present outbreak may be due to a change in the genetic sequence which altered the E1 segment of the virus' viral coat protein, a variant called E1-A226V. This mutation potentially allows the virus to multiply more easily in mosquito cells.[17] The change allows the virus to use the Asian tiger mosquito (an invasive species) as a vector in addition to the more strictly tropical main vector, Aedes aegypti.[18]

After the detection of zika virus in Brazil in April 2015, the first ever in the Western Hemisphere, it is now thought some chikungunya and dengue cases could in fact be zika virus cases or coinfections.[19]

Etiology

Chikungunya virus (CHIKV) is a member of the alphavirus genus, and Togaviridae family. It was first isolated in 1953 in Tanzania and is an RNA virus with a positive-sense single-stranded genome of about 11.6kb.[20] It is a member of the Semliki Forest virus complex and is closely related to Ross River virus, O'nyong'nyong virus, and Semliki Forest virus.[21]

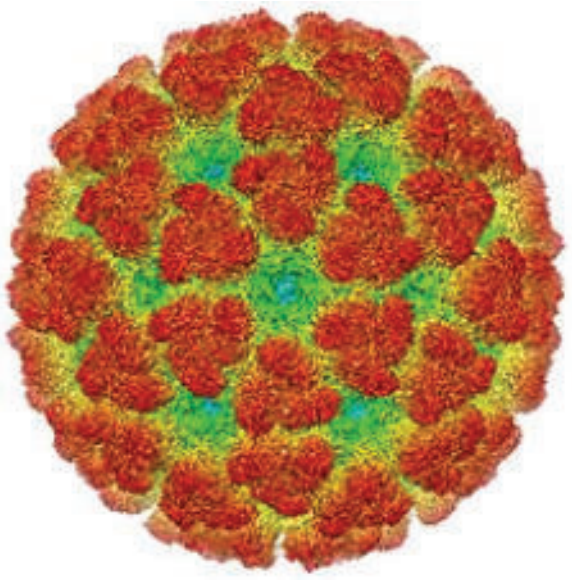

Fig 1 *Cryoelectron microscopy reconstruction of chikungunya virus.*

Because it is transmitted by arthropods, namely mosquitoes, it can also be referred to as an arbovirus (*ar*thropod-*bo*rne virus). In the United States, it is classified as a category C priority pathogen and work requires biosafety level III precautions.[22]

Transmission

Chikungunya is generally transmitted from mosquitoes to humans. Mosquitoes become infected when they feed on a person already infected with the virus. Infected mosquitoes can then spread the virus to other people through bites. They bite during the day and at night. Less common modes of transmission include vertical transmission, which is transmission from mother to child during pregnancy or at birth. Transmission via infected blood products and through organ donation is also theoretically possible during times of outbreak, though no cases have yet been documented.[15]

Chikungunya is related to mosquitoes, their environments, and human behavior. The adaptation of mosquitoes to the changing climate of North Africa around 5,000 years ago made them seek out environments where humans stored water. Human habitation and the mosquitoes' environments were then very closely connected. During periods of epidemics humans are the reservoir of the virus. Because high amounts of virus are present in the blood in the beginning of acute infection, the virus can be spread from a viremic human to a mosquito, and back to a human. In theory, the virus could be spread through a blood transfusion. To date, there are no known reports of this happening.[23] During other times, monkeys, birds and other vertebrates have served as reservoirs.[24]

Three genotypes of this virus have been described, each with a distinct genotype and antigenic character: West African, East/Central/South African, and Asian genotypes.[25]

Fig 2 *Chikungunya virus is transmitted to people through mosquito bites.*

Chikungunya is spread through bites from Aedes mosquitoes, and the species A. aegypti was identified as the most common vector, though the virus has recently been associated with many other species, including A. albopictus.[15]

Pathophysiology

The chikungunya virus is passed to humans when a bite from an infected mosquito breaks the skin and introduces the virus into the body. The pathogenesis of chikungunya infection in humans is still poorly understood, despite recent outbreaks. It appears that in vitro, chikungunya virus is able to replicate in human epithelial and endothelial cells, primary fibroblasts, and monocyte-derived macrophages. Viral replication is highly cytopathic, but susceptible to type-I and -II interferon. In vivo, in studies using living cells, chikungunya virus appears to replicate in fibroblasts, skeletal muscle progenitor cells,and myofibers.[26-28]

The type-1 interferon response seems to play an important role in the host's response to chikungunya infection. Upon infection with chikungunya,the host's fibroblasts produce type-1 alpha and beta interferon (IFN-± and IFN-²).[29] In mouse studies, deficiencies in INF-1 in mice exposed to the virus cause increased morbidity and mortality.[29-31] The chikungunya-specific upstream components of the type-1 interferon pathway involved in the host's response to chikungunya infection are still unknown.[32]

In the acute phase of chikungunya, the virus is typically present in the areas where symptoms present, specifically skeletal muscles, and joints. In the chronic phase, it is suggested that viral persistence (the inability of the body to entirely rid itself of the virus), lack of clearance of the antigen, or both, contribute to joint pain. The inflammation response during both the acute and chronic phase of the disease results in part from interactions between the virus and monocytes and macrophages.[14]

Chikungunya virus disease in humans is associated with elevated serum levels of specific cytokines and chemokines. High levels of specific cytokines have been linked to more severe acute disease: interleukin-6 (IL-6), IL-1², RANTES, monocyte chemo-attractant protein 1 (MCP-1), monokine induced by gamma interferon (MIG), and interferon gamma-induced protein 10 (IP-10). Cytokines may also contribute to chronic chikungunya virus disease, as persistent joint pain has been associated with elevated levels of IL-6 and granulocyte-macrophage colony-stimulating factor (GM-CSF).In those with chronic symptoms, a mild elevation of C-reactive protein (CRP) has been observed, suggesting ongoing chronic inflammation. However,there is little evidence linking chronic chikungunya virus disease and the development of autoimmunity.[23]

Following the bite of an infected Aedes mosquito, CHIKV is injected into the dermis and locally targets connective tissue, epithelial cells and fibroblasts where viral replication takes place. In addition, during the viremic phase (d"5–7 days), circulating monocytes are responsible for dissemination into the bloodstream.Secondary infection sites include muscles and joints where fibroblasts are the main target cells. CHIKV may also be identified in the epithelial and endothelial cells of many organs particularly in the liver, spleen and brain. The acute phase of infection is characterized by a strong type I interferon response by infected fibroblasts and other cell types. This response is usually short-lived, being primarily limited to the viremic phase, and it can be more pronounced in infants. The type I interferon response seems to be prolonged in adults with chronic arthritis. Adaptive immunity against CHIKV is less well understood and only develops after the first week when viral replication has been limited by innate immunity. CHIKV-specific immunoglobulins protect against infection, but both B and T cells may contribute to pathogenesis and long-term joint disease.[33]

Clinical Features

The incubation period of the chikungunya virus ranges from one to twelve days, and is most typically three to seven.[34] The disease may be asymptomatic, but generally is not, as 72% to 97% of those infected will develop symptoms. Characteristic symptoms include sudden onset with high fever, joint pain, and rash. Other symptoms may occur, including headache, fatigue, digestive complaints, and conjunctivitis.[13]

Chikungunya fever may result in a chronic phase as well as the phase of acute illness.[15] Within the acute phase, two stages have been identified: a viral stage during the first five to seven days, during which viremia occurs[14] followed by a convalescent stage lasting approximately ten days, during which symptoms improve and the virus cannot be detected in the blood.[35]

Typically, the disease begins with a sudden high fever that lasts from a few days to a week, and sometimes up to ten days. The fever is usually above 39 °C (102 °F) and sometimes reaching 40 °C (104 °F) and may be biphasic-lasting several days, breaking, and then returning. Fever occurs with the onset of viremia, and the level of virus in the blood correlates with the intensity of symptoms in the acute phase. Fever is typically high-grade in both children and adults. In children, febrile seizures frequently are described and commonly occur beyond the typical age range of 6 months to 6 years. Typically, these seizures last for 3–5 days, with a maximum of 10 days.[36]

When IgM, an antibody that is a response to the initial exposure to an antigen, appears in the blood, viremia begins to diminish. However, headache, insomnia and an extreme degree of exhaustion remain, usually about five to seven days.[37]

Following the fever, strong joint pain or stiffness occurs; it usually lasts weeks or months, but may last for years. The joint pain can be debilitating, often resulting in near immobility of the affected joints.[38]

Myalgia, arthralgia and arthritis often are present in adults with chikungunya but typically less so in children (between 30% and 50% of affected children).Joint pain is reported in 87–98% of cases, and nearly always occurs in more than one joint, though joint swelling is uncommon.[34]

Typically the affected joints are located in both arms and legs, and are affected symmetrically. Joints are more likely to be affected if they have previously been damaged by disorders such as arthritis[15]. Pain most commonly occurs in peripheral joints, such as the wrists, ankles, and joints of the hands and feet as well as some of the larger joints, typically the shoulders, elbows and knees. Pain may also occur in the muscles or ligaments .Swelling without other signs of synovitis typically is reported with a symmetric, distal, polyarticular pattern. Permanent destruction of affected joints is rarely reported. Other rheumatic manifestations include tenosynovitis, tendinitis or bursitis at the acute and subacute stages (<day 90). It is now widely recognized that in adults arthralgia may persist for years.[15,34] Rash occurs in 40-50% of cases, generally as a maculopapular rash occurring two to five days after onset of symptoms.[35] The skin lesions most frequently reported are pigmentary changes in the centrofacial area, maculopapular rash and intertriginous aphthous-like ulcers. The rash usually is present for 5 days, with hyperpigmentation sometimes following the rash. Infants younger than 6 months of age may exhibit extensive bullous skin lesions with blistering covering up to 35% of the body surface area. Hemorrhagic manifestations including epistaxis, gingival bleeding and purpura are also observed in approximately 10% of pediatric cases.[39]

Digestive symptoms, including abdominal pain, nausea, vomiting or diarrhea, may also occur.[40] In more than half of cases, normal activity is limited by significant fatigue and pain.[34] Infrequently, inflammation of the eyes may occur in the form of iridocyclitis, or uveitis, and retinal lesions may occur.[41]Temporary damage to the liver may occur.[42]

Rarely, neurological disorders have been reported in association with chikungunya virus, including Guillain–Barré syndrome, palsies, meningo-encephalitis, flaccid paralysis and neuropathy.[39] In contrast to dengue fever, chikungunya fever very rarely causes hemorrhagic complications. Symptoms of bleeding should lead to consideration of alternative diagnoses or co-infection with dengue fever or coexisting congestive hepatopathy.[14]

Perinatal Infection

Perinatal CHIKV infection first was described during the La Réunion outbreak in 2005.[13] Although intrauterine transmission of CHIKV was absent or exceptionally rare in early pregnancy, it rose to nearly 50% when mothers were viremic in the week just preceding delivery. Infected neonates developed symptoms around day 4 (range:3-7) of life. Signs most commonly included fever, rash and edema. Other frequent observations were petechiae, thrombocytopenia and lymphopenia. Complications included intracerebral hemorrhages, status epilepticus and multiorgan failure, which led to mechanical ventilation in one quarter of the neonates.[43]

The long-term outcome of survivors was poor: half the children exhibited diminished neurocognitive performance at 2 years of age.[44]

Chronic disease

Observations during recent epidemics have suggested chikungunya may cause long-term symptoms following acute infection[45,46]. This

condition has been termed chronic chikungunya virus-induced arthralgia.[47] Common predictors of prolonged symptoms are advanced age and prior rheumatological disease.[48]

Currently, the cause of these chronic symptoms is not fully known. Markers of autoimmune or rheumatoid disease have not been found in people reporting chronic symptoms. However, some evidence from humans and animal models suggests chikungunya may be able to establish chronic infections within the host. Viral antigen was detected in a muscle biopsy of a person suffering a recurrent episode of disease three months after initial onset.[49]

Diagnosis

Chikungunya is diagnosed on the basis of clinical, epidemiological, and laboratory criteria. However, it is important to keep in mind that cases may appear in places where chikungunya is not endemic. Clinically, acute onset of high fever and severe joint pain would lead to suspicion of chikungunya. Epidemiological criteria consist of whether the individual has traveled to or spent time in an area in which chikungunya is present within the last twelve days (i.e. the potential incubation period). For laboratory confirmation of chikungunya, virological and serological tests are necessary. During the first 5 days of infection, the virus can be found in the blood by reverse transcriptase polymerase chain reaction. In samples obtained later, enzyme-linked immunosorbent assays may confirm the presence of IgM and/or IgG anti-chikungunya antibodies. IgM antibodies appear between days 2 and 7 after onset of disease, whereas IgG antibodies frequently are detected after the first week of illness. The World Health Organization, therefore, recommends both serological and virological testing of samples collected during the first week after onset of symptoms. IgM antibodies peak at 3-5 weeks after onset of symptoms and then decline 2 months later but still may persist for years. IgG antibodies are believed to be detectable lifelong.[55]

Diagnostic Criteria for Chikungunya Fever[56]

The case definition of Chikungunya fever as proposed by the World Health Organization (WHO) Regional Office for Southeast Asia is discussed below:

Suspected case

A suspected case involves a patient presenting with acute onset of fever, usually with chills/rigors, that lasts for 3-5 days with pain in multiple Joints/swelling of extremities that may continue for weeks to months.

Probable case

A probable case is characterized by conditions that support a suspected case (as above) along with one of the following conditions:

- History of travel or residence in areas reporting outbreaks
- Ability to exclude malaria, dengue, and any other known cause of fever with joint pains

Confirmed case

Chikungunya fever is confirmed in the patient meets one or more of the following findings irrespective of the clinical presentation:

- Virus isolation in cell culture or animal inoculations from acute-phase sera
- Presence of viral ribonucleic acid (RNA) in acute-phase sera as determined with RT-PCR
- Presence of virus-specific IgM antibodies in single serum sample in acute phase or 4-fold increase in virus-specific IgG antibody titer in samples collected at least 3 weeks apart

Presently, there is no specific way to test for chronic signs and symptoms associated with Chikungunya fever although nonspecific laboratory findings such as C reactive protein and elevated cytokines can correlate with disease activity.[57]

Differential Diagnosis

The differential diagnosis of febrile patient with recent travel to, or residency in, tropical areas is broad. It should include malaria, dengue, typhoid fever, influenza, hepatitis, leptospirosis and rickettsial infection. Among the diseases listed, dengue is the infection most capable of mimicking chikungunya. In this regard, clinical signs including arthralgia and rash cannot reliably be used to distinguish between dengue and chikungunya.[15] Overall, however, rash appears earlier in the course of chikungunya than it does with dengue. Furthermore, thrombocytopenia more frequently is seen in patients with dengue; however, up to 50% of children with chikungunya also have mild thrombocytopenia.[58]

Management

Currently, no specific treatment for chikungunya is available. Supportive care is recommended, and

symptomatic treatment of fever and joint swelling includes the use of nonsteroidal anti-inflammatory drugs such as naproxen, non-aspirin analgesics such as paracetamol (acetaminophen) and fluids.[47] Aspirin is not recommended due to the increased risk of bleeding.[59] Despite anti-inflammatory effects, corticosteroids are not recommended during the acute phase of disease, as they may cause immunosuppression and worsen infection.[15]

There is no specific treatment for chikungunya. Management, therefore, focuses on adequate hydration, antipyretics and analgesics. Some experts recommend withholding salicylates and nonsteroidal anti-inflammatory drugs (NSAIDs), as these may precipitate bleeding manifestations.[60]Others recommend excluding dengue fever before prescribing NSAIDs. Although the symptoms of patients with persistent joint pain may be challenging to manage, NSAIDs together with corticosteroids or methotrexate successfully have been used in adults. Ribavirin also has been shown to improve chronic arthralgia/arthritis in some adult patients, but its benefit in acute pediatric infection remains unknown.[15, 47]

Passive immunotherapy has potential benefit in treatment of chikungunya. Studies in animals using passive immunotherapy have been effective, and clinical studies using passive immunotherapy in those particularly vulnerable to severe infection are currently in progress[61]. Passive immunotherapy involves administration of anti-CHIKV hyperimmune human intravenous antibodies (immunoglobulins) to those exposed to a high risk of chikungunya infection. No antiviral treatment for chikungunya virus is currently available, though testing has shown several medications to be effective in vitro.[62]

Long-Term Monitoring

Arthralgias resolve spontaneously within 3 weeks in about 70% of patients. However, they can persist for 3-6 months in 30% of patients, for 20 months in 15%, and for 3-5 years in 12%. Elderly patients and patients with prior rheumatologic conditions are at higher risk for chronic polyarthritis, tenosynovitis, and bursitis. Bouquillard et al have reported the possible unmasking or occurrence of rheumatoid arthritis in patients infected with Chikungunya virus. Patients with chronic arthritis may need long-term follow-up with both infectious disease and rheumatology experts.[63]

Prognosis

Although Chikungunya fever is a self-remitting illness, rare cases of complications have been reported in during major outbreaks among patients with comorbidities (cardiovascular, respiratory, neurological), neonates, elderlypatients, immunocompromised patients.[50] Neonates are vulnerable as it is possible to vertically transmit chikungunya from mother to infant during delivery, which results in high rates of morbidity, as infants lack fully developed immune systems.[51]

The likelihood of prolonged symptoms or chronic joint pain is increased with increased age and prior rheumatological disease [48].Persistent severe arthralgias could lead to long-term disability and loss of work days. Thus, the burden on the economy in terms of loss of productivity and income is estimated to be significant.[52] Intrauterine infection in pregnant women with vertical transmission has also been reported.[53]

The mortality rate of chikungunya is slightly less than 1 in 1000.[54] Those over the age of 65, neonates, and those with underlying chronic medical problems are most likely to have severe complications.[23]

Prevention

Because neither specific treatment nor any approved vaccine exist, the most effective means of prevention are protection against contact with the disease-carrying mosquitoes and controlling mosquito populations by limiting their habitat.[7] Mosquito control focuses on eliminating the standing water where mosquitos lay eggs and develop as larva;

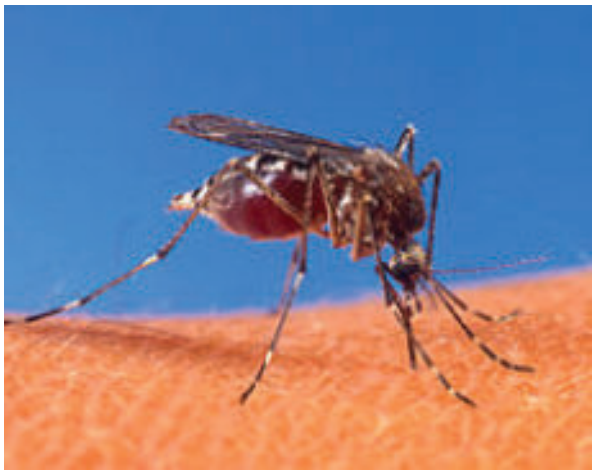

Fig 3 *A aegyptti mosquito biting a person*

if elimination of the standing water is not possible, insecticides or biological control agents can be added. Methods of protection against contact with mosquitos include using insect repellents with substances such as DEET, icaridin, PMD (p-menthane-3,8-diol, a substance derived from the lemon eucalyptus tree), or IR3535. However, increasing insecticide resistance presents a challenge to chemical control methods. Wearing bite-proof long sleeves and trousers also offers protection, and garments can be treated with pyrethroids, a class of insecticides that often has repellent properties. Vaporized pyrethroids (for example in mosquito coils) are also insect repellents. As infected mosquitos often feed and rest inside homes, securing screens on windows and doors will help to keep mosquitoes out of the house. In the case of the day-active A. aegypti and A. albopictus, however, this will have only a limited effect, since many contacts between the mosquitoes and humans occur outdoors.[14]

Vaccine

As of 2017, no approved vaccines are available. A phase-II vaccine trial used a live, attenuated virus, to develop viral resistance in 98% of those tested after 28 days and 85% still showed resistance after one year[64]. Even with a vaccine, mosquito population control and bite prevention will be necessary to control chikungunya disease.[65]

WHO response[66]

WHO responds to chikungunya by**:**

- formulating evidence-based outbreak management plans;
- providing technical support and guidance to countries for the effective management of cases and outbreaks;
- supporting countries to improve their reporting systems;
- providing training on clinical management, diagnosis and vector control at the regional level with some of its collaborating centres; and
- publishing guidelines and handbooks on case management and vector control for Member States.

Future Perspectives

Chikungunya fever is an emerging global disease with several intriguing and unanswered questions such as the reason for sudden major rapid outbreaks with disease-free intervals, mode of survival or maintenance of the virus in nature between epidemics, factors that trigger the outbreaks, and strain replacements during outbreaks. More research is needed to understand the epidemiology and natural history of this disease. Until then, prevention and vector control at personal and community level should be implemented [67].Research into development of a live-virus and attenuated-virus vaccine against Chikungunya virus is ongoing. However, no vaccines are available at this time.[68]

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DS (Child) H J 2016; 32(1) : 52-56

CASE SERIES

Aseptic Meningitis and Acute Mumps Associated Parotitis

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Abstract

Introduction*: The classic "Mumps" is known as a viral parotitis caused by mumps virus belonging to the genus Rubula virus in the Paramyxoviridae family.*

Case Presentation*: We reviewed six patients diagnosed with aseptic meningitis due to acute mumps associated parotitis based on clinical scenario and minimum laboratory finding(e.g. complete blood count) as specific investigations such as Polymerase Chain Reaction(PCR) or detection of Mumps specific IgG/IgM facilities are still not commonly available in Bangladesh. While there are many similarities between the clinical characteristics of classic mumps and acute non-mumps associated parotitis, some significant differences exist. In addition, there are some important differences between aseptic meningitis associated with mumps parotitis and aseptic meningitis associated with non-mumps parotitis.*

Discussion*: Appropriate testing for the most likely causative pathogens including PCR or ELISA of serum or CSF is indispensable for accurate diagnosis. Therefore in the developing countries like us where standard laboratory resources are mostly unavailable regarding virus isolation through PCR or virus specific investigation tools, careful history taking and meticulous physical examinations along with viral infection specificcomplete blood counts can guide us to diagnose the mumps with aseptic meningitis cases.*

Conclusion*: MR vaccine has already been introduced in the EPI schedule by the government since 2012 but MMR vaccine instead would be more appropriate for the prevention of mumps virus related complications.*

Keywords*: Mumps, Parotitis, Meningitis, Aseptic*

Introduction

The classic "Mumps" is known as a viral parotitis caused by mumps virus belonging to the genus Rubula virus in the Paramyxoviridae family, but various viral pathogens have been identified as causes of acute viral infection of the salivary glands.[1,2] These include viruses such as Parainfluenza (types 1, 2 and 3) virus, Influenza, Coxsackie virus, ECHO (enteric cytopathic human orphan) virus and Lymphocytic choriomeningitis virus.[3-6] Moreover, Cytomegalovirus and Adenovirus have been reported as causative pathogens of acute parotitis in patients with AIDS. Direct HIV (human immunodeficiency virus) infection of the parotid glands is rare, but is characterized by chronic, cystic parotid enlargement.[7-9]

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The World Health Organization (WHO) recommended clinical case definition states, "acute onset of unilateral or bilateral tender, self-limited swelling of the parotid or other salivary gland, lasting fortwo or more days without other apparent cause". The complications regarding classic mumps syndrome have been widely documented as orchitis followed by infertility later on, oophoritis, mastitis, sensorineural hearing loss, pancreatitis, aseptic meningitis and encephalitis. Of these complications, aseptic meningitis is the most common neurologic manifestation which occurs in 1-10% of patients infected with mumps[10]. On the other hand, other viruses responsible for acute parotitis are less common, and understanding of their associated complications is more limited. We reported six pediatric patients with clinical manifestations of aseptic meningitis as a complication of mumps associated parotitis, presented at the High Dependency and Isolation Unit of Dhaka Shishu (Children) Hospital,Dhaka, Bangladesh.

Cases Presentation

All of the six cases were referred to our department between January 2016 and December 2016, with conclusive diagnosis of "secondary aseptic meningitis due to acute mumps associated parotitis" based on the clinical and laboratory findings. Among the six cases, four were male and two were female with ages ranging from 4 to 12 years. The characteristics of individual cases are given in Table 1. There were prodromal symptoms including headache, myalgias, arthralgias, anorexia and malaise prior to the development of parotitis. The initial symptoms were unilateral parotid gland swelling followed by bilateral parotid swelling in 4 cases along with headache and high grade continuous fever up to 100 ~ 101°F in six cases respectively. Some of them had the left parotid gland swelling initially and some developed initial right parotid gland swelling followed by bilateral parotid gland swelling in all of the cases and had headache and fever a few days later after the development of parotid swelling. All of them had an alert mental state with signs of meningeal irritation positive in physical examination. Due to unavailability of confirmatory serological investigations for Mumps in Bangladesh, complete blood count has been considered as a supportive tool for confirming viral parotitis. All of them responded to conservative management and showed recovery without sequelae within 11 days.

Table I
Clinical Characteristics of Six Cases

Patient	Age (yr)/ Sex	Chief Complaints	Onset	Location	Associated symptoms	Signs of Meningeal irritation	Hospital Stay
1.	4/M	Both sided neck swelling	5 days ago	Initially left followed by bilateral parotid swelling	Headache, fever upto 100^0 F, nausea, malaise	Positive	10 days
2.	7/F	Both sided neck swelling	10 days	Initially right followed by bilateral parotid swelling	Headache, fever upto 100^0 F, nausea, arthralgia	Positive	11 days
3.	5/M	Both sided neck swelling	7 days	Initially right followed by bilateral parotid swelling	Headache, fever upto 101^0 F, nausea, vomiting	Positive	9 days
4.	9/M	Both sided neck swelling	10 days	Initially left followed by bilateral parotid swelling	Headache, fever upto 101^0 F, nausea, vomiting, alert mental status	Positive	10days
5.	11/F	Both sided neck swelling	7 days	Initially left followed by bilateral parotid swelling	Headache, fever upto 101^0 F, nausea, vomiting, myalgia	Positive	11days
6.	9/M	Both sided neck swelling	5 days	Initially right followed by bilateral parotid swelling	Headache, fever upto 101^0 F, nausea, vomiting, myalgia, alert mental status	Positive	10 days

On laboratory tests, the highest levels of serum amylase were found in all cases at initial work-up (Table II). On the second day of hospitalization, spinal tapping was performed in all cases. Analysis of cerebrospinal fluid (CSF) was shown in the Table III. Biochemical analysis and cytology of CSF supported the diagnosis of viral meningitis.

Table II
Laboratory Test Results

Patient (Serum)	Amylase, Highest, IU/L	Complete Blood Count						
		Total Count (per mm^3)	Dif. Count	Neutrophil (%)	Lymphocyte (%)	Eosinophil (%)	Basophil (%)	Platelet (per mm^3)
1.	530	3,000		28	65	4	3	1,80,000
2.	480	7,200		30	60	5	5	2,50,000
3.	1120	8,500		40	55	3	2	1,05,000
4.	760	11,560		35	55	4	1	1,89,000
5.	799	13,870		25	72	3	0	1,24,000
6..	837	4,900		30	70	0	0	2,00,000

Table III
Cerebrospinal fluid analysis

Patient (CSF)	Colour	Total Leukocyte Count (per mm^3)	PMN (%)	Lymphocyte (%)	RBC (permm^3)	Sugar (mg/dL)	Protein (mg/dL)	Staining	Lactate (mmol/L)
1.	Clear	220	0	100	0	45	150	No bacteria	1.5
2.	Clear	100	5	95	0	59	120	No bacteria	2.0
3.	Clear	346	0	100	0	52	110	No bacteria	2.2
4.	Clear	120	2	98	0	44	105	No bacteria	1.2
5.	Clear	105	8	92	0	50	95	No bacteria	2.4
6.	Clear	85	0	100	0	65	125	No bacteria	1.9

Discussion

Mumps virus can be easily detected from saliva, cerebrospinal fluid, urine, or seminal fluid within the first week of onset of parotitis.[11, 12]If viral detection fails, a definitive diagnosis can be performed by serological markers. Serological confirmative diagnosis is mainly based on detection of virus-specific IgM and IgG antibodies, measured by direct or indirect ELISA.[13] In our case series, we did not perform any Mumps virus specific serological tests due to unavailability of laboratory facilities.None of the cases had any prior history of infection. A licensed vaccine to prevent measles first became available in 1963, and an improved one in 1968. Vaccines for mumps and rubella became available in 1967 and 1969, respectively. The three vaccines (for mumps, measles, and rubella) were combined in 1971 to become the measles-mumps-rubella (MMR) vaccine.[14] However The mumps vaccination is not included in our expanded programme of immunization (EPI) schedule till date but available in the private health sectors approximately since 2000 and none of our six cases were immunized with MMR vaccine. So, we considered mumps virus as the causative agent of acute parotitis in our case series.

Acute non-mumps associated parotitis caused by parainfluenza viruses or non-paramyxoviruseshas a low incidence rate and therefore its clinical characteristics and complications including aseptic meningitis have rarely been reported. Of the non-mumps viruses mentioned earlier, parainfluenza virus (types 2 and 3) has been the only virus reported representing acute parotitis and aseptic meningitis simultaneously.[4, 6, 10, 16, 17]While there are some similarities between the clinical characteristics of classic mumps and non-mumps associated parotitis, some significant differences exist. First of all, approximately two thirds of patients have short prodromal symptoms before the development of parotitis presenting low-grade fever, headache,

myalgias, arthralgias, anorexia, and malaise in classic mumps.[13] There were recognized prodromal symptoms in all of our cases. Whereas in non-mumps associated parotits, there is usually no prodromal symptoms prior to the onset of parotits. Second, in classic mumps, swelling occurs in both parotid glands in 90% of cases. Glandular swelling generally begins on one side, followed by contralateral involvement within 1 to 5 days.[13] These findings were very much similar to our cases. However, only unilateral swelling of the parotid gland is usually found in non-mumps associated parotitis cases. Third, in classic mumps, 85% of patients occur in children younger than 15 years and all of our patients were below 12.[5] On the contrary, non-mumps associated parotitis has a rather high developmental age involving older children.

Aseptic meningitis due to mumps infection is the most common extra-salivary manifestation which is a benign entity without essential risk of mortality or long-term sequelae.[13] Typical symptoms include high fever, headache, vomiting, neck stiffness, and lethargy.[17] The diagnosis of CNS complications is relatively easy if there is salivary gland involvement, but in up to 50% of cases without salivary gland involvement, an accurate diagnosis can be made only by serologic tests.[18, 19] Furthermore, in patients with mumps meningitis, virus-specific IgM and IgG can be detected in CSF study.[20]In our case series, there are some similar characteristics with classic mumps meningitis. First, aseptic meningitis occurred mostly in male patients. Second, our patients admitted with high fever and headache lasting for 72 ~ 96 hours. Third, aseptic meningitis was a self-limited disease which showed spontaneous recovery without sequelae within 7 to 10 days with conservative management. However, there are following important differences. First, there is difference in the developmental stage of meningitis. In cases of aseptic meningitis due to mumps, it can manifest about 5 days after the onset of mumps parotitis or it can precede mumps parotitis by a week.[21]In our cases meningitis was diagnosed with 5-10days of onset of parotits, similar to classic mumps. However, aseptic meningitis occurs with the onset of parotitis in non-mumps associated parotits. Second, in meningitis due to mumps, meningeal irritation signs were reported in 43-93% of casesand appear much higher in older children, adolescents, and adults, but all of our patients showed positive meningeal irritation signs in physical examination.[17] Third, in aseptic meningitis due to mumps, it occurs without salivary gland involvement in 50% of cases, and all of our patients showed bilateral parotid gland involvement and vice versa with non-mumps associated meningitis.

We presented six patients as mumps associated parotitis with aseptic meningitis. However, in the reported patients, important specific viral testing was not performed. First, testing for mumps virus by PCR (polymerase chain reaction) from serum and CSF was not performed. Furthermore, an early infection might have potentially been missed. Second, we discussed parainfluenza virus, influenza virus, coxsackie virus, echovirus and lymphocytic choriomeningitis virus as possible causative pathogens of salivary gland infections. However, testing for these pathogens (serology, PCR) was not performed. Furthermore, lack of this data leaves the definite cause of the disease unresolved. If acute parotitis accompanies with clinical manifestations different from classic mumps and associated with aseptic meningitis in early stage of the disease with initially negative serological test for mumps, acute parotitis with aseptic meningitis caused by non-mumps virus should be considered and various serological tests should be performed to identify the causative virus.

Conclusion

Appropriate testing for the most likely causative pathogens including PCR or ELISA of serum or CSF is indispensable for accurate diagnosis. Therefore in the developing countries like us where standard laboratory resources are mostly unavailable regarding virus isolation through PCR or virus specific investigation tools, careful history taking and meticulous physical examinations along with viral infection specific complete blood counts can guide us to diagnose the mumps with aseptic meningitis cases. Though MR vaccine has already been introduced in the EPI schedule by the government but MMR vaccine instead would be more appropriate for the prevention of mumps virus related complications.

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CASE REPORT

Milroy Disease: A Report of Rare Disease

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Introduction

Milroy disease is named after William Milroy, who described 97 members of a family, of whom 26 had leg edema. In the family described by Milroy, the edema was painless, non-progressive, and confined to the lower limbs. Hereditary lymphedema of the legs was also described by Nonne 1891; hence, the term Nonne-Milroy disease has been used in the past. The prevalence of Milroy disease is not known but it appears to be one of the more common causes of primary lymphedema and occurs in all ethnic groups.[1,2]

Milroy disease is characterized by lower-limb lymphedema, present as pedal edema at (or before) birth or developing soon after. Occasionally it develops later in life. Swelling is usually bilateral but can be asymmetric. The degree of edema can progress but in some instances can improve, particularly in early years. Other features sometimes associated with Milroy disease include hydrocele (37% of males), prominent veins (23%), upslanting toenails (14%), papillomatosis (10%), and urethral abnormalities in males (4%). Cellulitis, which can damage the lymphatic vessels, occurs in approximately 20% of affected individuals, with infection significantly more likely in males than female.[3-4] Other name of Milroy disease, congenital familial lymphedema, hereditary lymphedema type I.[5]

Milroy disease is inherited in an autosomal dominant pattern, which means one copy of the altered gene in each cell is sufficient to cause the disorder. In many cases, an affected person inherits the mutation from one affected parent. Other cases may result from new mutations in the FLT4 gene. These cases occur in people with no history of the disorder in their family. About 10 percent to 15 percent of people with a mutation in the FLT4 gene do not develop the features of Milroy disease.[6,7] FLT4 gene provides instructions for producing a protein called vascular endothelial growth factor receptor 3 (VEGFR-3), which regulates the development and maintenance of the lymphatic system. Mutations in the FLT4 gene interfere with the growth, movement, and survival of cells that line the lymphatic vessels (lymphatic endothelial cells). These mutations lead to the development of small or absent lymphatic vessels. If lymph fluid is not properly transported, it builds up in the body's tissues and causes lymphedema. It is not known how mutations in the FLT4 gene lead to the other features of this disorder. Many individuals with Milroy disease do not have a mutation in the FLT4 gene. In these individuals, the cause of the disorder is unknown.[8]

Case report

Abdul Nabi, a 5years old boy, immunized as per EPI schedule, first issue of his consanguineous parents coming from Brammonbaria got admitted with the complaints of assymmetrical swelling of one side of the body since birth. He was delivered normally with average birth weight at term without any significant perinatal complication. Following birth parents noticed assymmetrical enlargement of right upper

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limb and face followed by lower limb which was gradually increasing. For seven days patient developed abdominal distension and scrotal swelling which was deteriorating for three days. For this problem, they consulted several physicians and took medications but as the condition did not improve they came to Dhaka Shishu Hospital for further evaluation and better management, There was no history of fever, pain, respiratory discomfort and none of his family member suffer from such type of illness. Birth history was uneventful, developmentally he was age appropriate. Now he is on normal family diet. Nabi is the first child of his family other sibling, a two year old girl is apparently healthy.

On Exam, Nabi was conscious, cooperative, mildly pale. Nonpitting oedema was present on the right hand and right lower limb. Circumferential difference at the level of mid upper arm of both arm is 6cm and mid thigh level is 2cm. Regarding vital signs, pulse 80/min, BP 90/60mm Hg, RR 24/min, Temp 98F, BSUA 1plus, Anthropometrically he was age appropriate. On skin exam, skin over the enlarged limb normal, BCG mark present.ENT normal. There was no lymphadenopathy. Alimentary system exam revealed abdomen was distended, flanks were full, umbilicus was centrally placed, everted with transverse slit. Huge ascitis present, scrotal swelling present. Other system revealed normal finding.

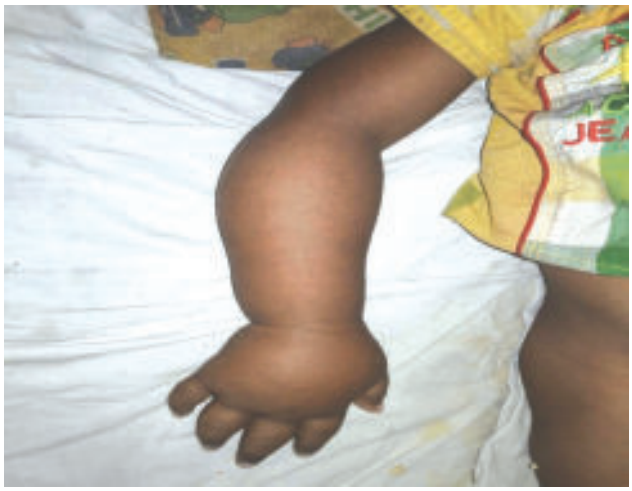

Fig 1 *Swelling of right upper limb*

Investigation: CBC with film Hb12.9gm/dl, WBC 13,400/cumm, Platelet 662000/cumm, N 77%, L19%, M 02%, E02% PBF Normocytic normochromic RBC, platelet adequate, WBC as distribution. Serum Creatinin 37.5 micromol/L, serum albumin10.0mg/dl, SGPT 23U/L,,BUN 3.2mmol /L,S Cholesterol 3.3mg/dl.ICT for Filaria Negative, USG of Abdomen revealed huge ascitis, Ascitic Fluid Study showed colour-milky white,total leukocyte 170/cumm, polymorph 30%, Lymphocyte 60%, other cell histiocyte10%, Ziel Nelson staining-AFB not found. Lymphoscintigraphy showed, Left upper limb-Normal lymphatic channels, Right upper limb-primary lympoedema. They could not do gene study as this test was not available in our country. For treatment purpose we send the child to a lymphadenema therapist and he advised some physical therapy, message and elevation of the limb.

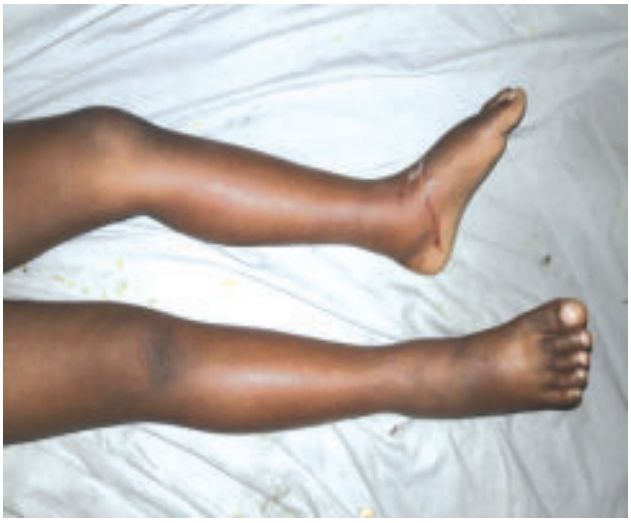

Fig 2 *Swelling of right lower limb*

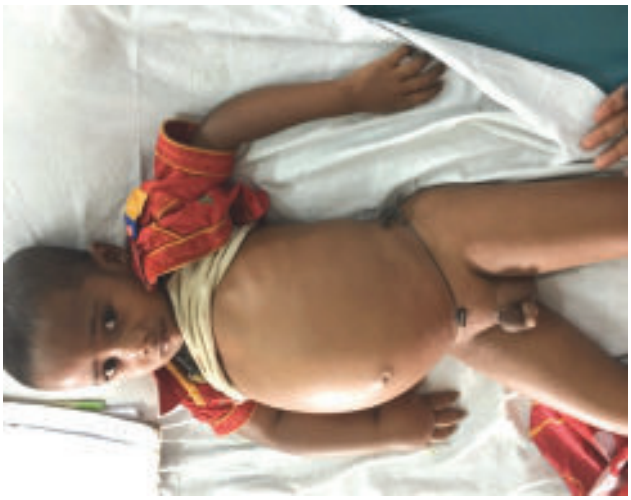

Fig 3 *Ascitis and scrotal swelling*

Discussion

The most common symptom of Milroy disease is build-up of fluids (lymphedema) in the lower limbs, which is usually present from birth or before birth. However, the degree and distribution of swelling varies among affected people.It sometimes progresses, but may improve in some cases. Other signs and symptoms may include hydrocele and/or urethral abnormalities in males; prominent veins; upslanting toenails; papillomatosis (development of wart-like growths); and cellulitis. Cellulitis may cause additional swelling.[9-10]

Milroy disease is diagnosed by clinical findings and confirmed by molecular genetic testing. Lympho-scintigraphy can be performed; the characteristic finding is lack of uptake of radioactive colloid in the ilioinguinal lymph nodes caused by a paucity of lymphatic vessels or abnormal function of the vessels in the lower limbs. Lymphoscintigraphy normally replaces lymphangiography (x-ray after direct injection of dye into the lymphatic vessels in the foot) as it is less invasive. Lymphangiography is also technically more problematic because of difficulties locating lymphatic vessels for cannulation. Lymphoscintigraphy is not essential to make the diagnosis and one can proceed directly to molecular testing. *FLT4 (VEGFR3*) is the only gene known to be associated with Milroy disease.[11-13]

Management include Genetic counselling

Treatment of manifestations*:* A lymphedema therapist may utilize fitted stockings and massage to improve the cosmetic appearance or decrease the size of the limb and reduce the risk of complications. Improvement in swelling is usually possible with use of properly fitted compression hosiery and/or bandaging and well-fitting, supportive shoes. Toe gloves may be of benefit and good skin care is essential.

Prevention of secondary complications: Frequency of cellulitis can be reduced through good skin hygiene, prompt treatment of infections with antibiotics, and prophylactic antibiotics for recurrent episodes.

Agents/circumstances to avoid: Wounds to limbs, long periods of immobility with the legs in a dependent position and medications that can cause increased leg swelling.

Evaluation of relatives at risk: Evaluating relatives at risk ensures identification of those who will benefit from treatment early in the disease course.[14-15]

The symptoms and severity of Milroy disease can vary among affected people (even within the same family), so the long-term effects of the condition may be difficult to predict. Swelling varies in degree and distribution, and can be disabling and disfiguring. For some people the outlook depends on how chronic the lymphedema is, as well as whether complications arise. However, Milroy disease is rarely associated with significant complications.[17-18]

The degree of edema sometimes progresses, but in some cases can improve (particularly in early years). Complications of lymphedema may include recurrent bouts of cellulitis and/or lymphangitis, bacterial and fungal infections, deep venous thrombosis, functional impairment, cosmetic embarrassment, and amputation. Complications following surgery are common. It has also been reported that people with chronic lymphedema for many years may have a significantly higher risk to develop lymphangiosarcoma (a type of angiosarcoma). This type of tumor is highly aggressive and has a very poor prognosis.[19-21]

Conclusion

Milroy disease is a rare clinical condition.However, proper counselling and psychological support to the patient and his family is the most important aspect of management of these patients.

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DS (Child) H J 2016; 32(1) : 61-63

CASE REPORT

GM1 Gangliosidosis: A Rare Neurometabolic Disease

Shayla Imam Kanta[1], Mir Mohamad Yousuf Pavel[2], Naila Z Khan[3], AKM Khairul Islam[4]

Introduction

GM1 gangliosidosis is an autosomal recessive lysosomal storage disorder characterized by the generalized accumulation of G_{M1} ganglioside, oligosaccharides, and the mucopolysaccharide keratan sulfate (and their derivatives). Deficiency of the lysosomal hydrolase,acid ²-galactosidase,causes G_{M1} gangliosidosis.[1] Three clinical subtypes of G_{M1} gangliosidosis are recognized, classified by age of onset, as follows:Infantile (type 1) Juvenile (type 2) and adult (type 3).We present the following case in a patient who is the first issue of a cosanguinous parents which have similar features with type1.Infantile G_{M1} gangliosidosis the most common infantile form with coarse facial features, hepatosplenomegaly, generalized skeletal dysplasia (dysostosis multiplex), macular cherry-red spots, and developmental delay/arrest (followed by progressive neurologic deterioration) usually occur within the first 6 months of life. Nonimmune hydrops has been reported. An increased incidence of Mongolian spots has also been reported. A wide spectrum of variability is observed in the appearance and progression of the typical dysmorphic features. As many as 50% of affected infants have a macular cherry-red spot.[1, 2, 3,12]

Case report

Abdullah a 10 months old boy only issue of a consanguineous parents was admitted in to Dhaka Shishu(Children) Hospital with the complaints of repeated Respiratory Tract Infection(RTI) causes hospitalization for 2 times, regression of acquired skills and seizure for 2 months. He has also been suffering from growth failure and developmental delay. On examination the child has coarse facies, disproportionate size of digits in both hands, having short trunk with protruded abdomen, Mongolian blue spots over the back and lower limbs, mildly pale, kyphosis. Anthropometrically the boy is severely stunted and wasted and his upper segment lower segment ratio:1:1.5, OFC- 47 cm and having wide open anterior fontanel. Neurologically the child is conscious having generalized muscular hypotonia with reduced muscle bulk and muscle power:3/5, no facial asymmetry, all the cranial nerves are intact, all the deep tendon reflexes are exaggerated and bilateral extensor planter reflexes, The child having protruded abdomen with hepatosplenomegaly. Developementaly the child is below 3 months. Fundoscopic examination revealed Cherry red spot in the retina.

Fig 1 *Child having coarse facies*

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Fig 2 *Mongolian blue spot*

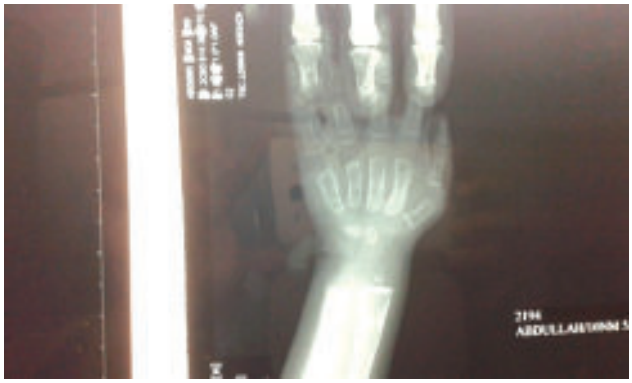

Fig 3 *Dysostosis multiplex*

Discussion

Three clinical subtypes of G_{M1} gangliosidosis are recognized, classified by age of onset, as follows:Infantile (type1) The classic infantile subtype combines the features of a neurolipidosis (ie, neurodegeneration, macular cherry-red spots) with those of a mucopolysaccharidosis (ie, visceromegaly, dysostosis multiplex, coarsened facial features)[2,3] This form of G_{M1} gangliosidosis most frequently presents in early infancy and may be evident at birth . In our case the features ae similar with type 1 or infantile form because in (type 2) the juvenile subtype is marked by a slightly later age of onset and clinical variability in the classic physical features.[4-6] In (type 3) the adult subtype is marked by normal early neurologic development with no physical stigmata and subsequent development of a slowly progressive dementia with parkinsonian features, extrapyramidal disease, and dystonia. Juvenile: The juvenile form is characterized by a later age of onset, less hepatosplenomegaly (if any), fewer cherry-red spots (if any), dysmorphic features, or skeletal changes (vertebral dysplasia may be detected radiographically).[7,8, 13] The adult form is characterized by normal early neurologic development, with variable age of clinical presentation. Slowly progressing dementia with parkinsonian features and extrapyramidal disease is common. Intellectual impairment may be initially absent or mild but progresses with time. Generalized dystonia with speech and gait disturbance is the most frequently reported early feature. Typically, no hepatosplenomegaly, cherry-red spots, dysmorphic features, or skeletal changes are present aside from scoliosis (mild vertebral changes may be revealed with radiography), but short stature is common.[9-10]

Acid ^{2} -galactosidase is a lysosomal hydrolase that catalyzes the removal of the terminal ^{2} -linked galactose from glycoconjugates (eg, G_{M1} ganglioside), generating G_{M2} ganglioside. It also functions to degrade other ^{2}-galactose–containing glycoconjugates, such as keratan sulfate. Enzyme activity is markedly reduced in patients with G_{M1} gangliosidosis. Deficiency of acid ^{2} -galactosidase results in the accumulation of glycoconjugates in body tissues and their excretion in urine. G_{M1} ganglioside and its derivative asialo-G_{M1} ganglioside (GA1), glycoprotein-derived oligosaccharides, and keratan sulfate are found at elevated intracellular concentrations.[1,6,7,8,9]

Diagnosis is mostly clinical diagnosis and confirmation by enzyme assay and genetic analysis[10, 11] currently, no effective medical treatment is available for the underlying disorder in patients with G_{M1} gangliosidosis. Bone marrow transplantation was successful in an individual with infantile/juvenile G_{M1} gangliosidosis; however, no long-term benefit was reported. Presymptomatic cord-blood hematopoietic stem-cell transplantation has been advocated by some as a possible treatment because of success in other lysosomal storage disorders. Symptomatic treatment for some neurologic sequelae is available but does not significantly alter the clinical course nutritional support does not change the disease course, and some families may choose to forgo invasive alimentation procedures. Neurologic and orthopedic sequelae may preclude adequate physical activity, and patients may benefit from physical and occupational therapy.[11-12]

Patients with G_{M1} gangliosidosis are at risk for aspiration pneumonia and recurrent respiratory infections resulting from neurologic compromise. Congestive heart failure may result secondary to cardiomyopathy. Atlantoaxial instability can develop

because of abnormally shaped cervical vertebrae. If this occurs, patients should be monitored, and they eventually should undergo surgical stabilization to avoid the risk of spinal cord injury[12-13]

Death usually occurs during the second year of life because of infection and cardiopulmonary failure juvenile (type 2) death usually occurs before the second decade of life Adult (type 3) Phenotypic variability is marked, but progressive development of neurologic sequelae usually leads to a shortened lifespan.[13]

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ABSTRACT FROM CURRENT LITERATURE

Diagnostic Accuracy of Indian Scale for Assessment of Autism (ISAA) in Children Aged 2-9 Years

Sharmila Banerjee Mukherjee, Manoj Kumar Malhotra, Satinder Aneja, *Satabdi Chakraborty, Smita Deshpande

Indian Pediatrics Volume 52 March 15, 2015:212-216

Department of Pediatrics, LHMC and Associated Hospitals, New Delhi; *Department of Psychiatry, Pt. BD Sharma PGIMS, Rohtak, Haryana; and #Department of Psychiatry, PGIMER – Dr. Ram Manohar Lohia Hospital, New Delhi, India. Correspondence to: DrSharmila B Mukherjee, Department of Pediatrics, Kalawati Saran Children's Hospital, Bangla Sahib Road, New Delhi 110 001, India. theshormi@gmail.com Received: July 07, 2014; Initial review: September 01, 2014; Accepted: January 09, 2015.

Objective: To determine the diagnostic accuracy of Indian Scale for Assessment of Autism (ISAA) in children aged 2-9 year at high risk of autism, and to ascertain the level of agreement with Childhood Autism Rating Scale (CARS).

Design: Diagnostic Accuracy study

Setting: Tertiary-level hospital.

Participants: Children aged between 2 and 9 year and considered to be at a high risk for autism (delayed development, and age-inappropriate cognition, speech, social interaction, behavior or play) were recruited. Those with diagnosed Hearing impairment, Cerebral palsy, Attention deficit hyperactivity disorder or Pervasive developmental disorders (PDD) were excluded.

Methods: Eligible children underwent a comprehensive assessment by an expert. The study group comprising of PDD, Global developmental delay (GDD) or Intellectual disability was administered ISAA by an investigator after one week. Both evaluators were blinded. ISAA results were compared to the Expert's diagnosis and CARS scores.

Results: Out of 102 eligible children, 90 formed the study group (63 males, mean age 4.5y). ISAA had a sensitivity 93.3, specificity of 97.4, positive and negative likelihood ratios 85.7 and 98.7 and positive and negative predictive values of 35.5 and 0.08, respectively. Reliability was good and validity sub-optimal (r low, in 4/6 domains). The optimal threshold point demarcating Autism from 'No autism' according to Receiver Operating Characteristic curve was ISAA score of 70. Level of agreement with CARS measured by Kappa coefficient was low (0.14).

Conclusions: The role of ISAA in 3-9 year old children at high risk for Autism is limited to identifying and certifying Autism at ISAA score of 70. It requires re-examination in 2-3 year olds.

Kawasaki Disease with Autoimmune Hemolytic Anemia

Dhwanee Thakkar, Nita Radhakrishnan, *Pk Pruthi And Anupam Sachdeva

Indian Pediatrics Volume 52, March 15, 2015:245-246

Background: Association of autoimmune haemolytic anaemia has been seldom reported with Kawasaki disease. Case characteristics: A 7-month-old boy, presented with prolonged fever, erythematous rash, severe pallor and hepatosplenomegaly.

Observations: Positive Direct Coombs test and coronary artery aneurysm on echocardiography. He was managed with steroids along with intravenous immunoglobulins and aspirin.

Outcome: Early identification of the condition helped in the management.

Message: Patients of autoimmune hemolytic anemia with unusual features such as prolonged fever, skin rash, and mixed antibody response in Coombs test should be evaluated for underlying Kawasaki disease as a possible etiology.

Nocturnal Enuresis among Nigerian Children and its Association with Sleep, Behavior and School Performance

Ou anyanwu, Rc Ibekwe, Ml Orji

Indian pediatrics volume 52__july 15, 2015:587-589

Objective: To study the association of nocturnal enuresis with sleep, behavior and school performance.
Methods: Hospital-based, cross-sectional descriptive study of 216 children (e"6-year-old) using structured questionnaire and behavioral tools.

Results: Prevalence of enuresis was 37.0%. Nocturnal enuresis was significantly associated with abnormal behaviour (P=0.049) and poor sleep hygiene (P<0.05). School performance was not associated with enuresis.

Conclusion: Children with nocturnal enuresis were at an increased risk of behavioral problems and poor sleep hygiene.

DSH NEWS

Mrs. Shaheen Siddique is donating a Haemodialysis Machine to Honorable Chairman of the Management Board, Dhaka Shishu (Children) Hospital, National Professor Dr. Shahla Khatun and Honorable Director Professor Manzoor Hussain for the better management of acute kidney failure patients on: 25th April 2016.

One week programme of free treatment is giving on the occasion of National Children's Day (17th March) by Honorable Academic Secretary of Bangladesh Institute of Child Health (BICH) and Honorable Deputy Director along with a group of senior paediatricians of Dhaka Shishu (Children) Hospital

BICH NEWS

BICH is the academic wing of Dhaka Shishu Hospital. It was established in 30th January, 1983. It is affiliated with Dhaka University, Bangabandhu Sheikh Mujib Medical University (BSMMU) and Bangladesh College of Physicians and Surgeons (BCPS). It has been conducting different courses e.g. DCH, FCPS, MD Paediatrics, MS Paediatric surgery & B.Sc in Health technology. It also conducts different sub-specialty courses e.g. FCPS Neonatology, FCPS Haemato-oncology, FCPS Nephrology, MD Neonatology, MD Haemato-oncology and MD Nephrology. It conducts 3 months certificate course in Paediatrics and 15 days Intensive course for MCPS. It organizes IMCI training and Palli Shishu Rural Health Training. Apart from this, the Institute also runs its regular academic activities. It has established Basic Science Department since 2006.

Diploma course of paediatric nursing has been started from 1st January 2012 and Diploma in paediatric physiotherapy under process.

Library facilities

The library of BICH has a rich collection of updated medical texts and reference books and reputed Medical Journals of home and abroad. BICH has introduced Broad Band facilities which are open to all students, teachers/ consultants of hospital for 24 hours. Facilities of library are also improved by HINARI. Students can download 2230 Medical Journals & more than 50 Paediatric Journals.

Present News

A newly formed classroom in BICH has been named as Prof. Sultan Ahmed Chowdhury as a tribute to First Honarary Director of Dhaka Shishu Hospital.

DS (Child) H J 2016; 32(1) : 67

Postgraduate courses/training in paediatrics and child health

1. FCPS in paediatrics : Twice in a year, in the months of January and July.
2. Recognized center by BCPS for training in FCPS (Paeditric surgery) .
3. Recognized centre for course and training in different subspeciality as: Neonatology, Pediatric Nephrology, Paediatric Haematology and Onchology, Paediatric Pulmonology and Paediatric Neuroscience.
3. MD/MS in paediatrics : Part I: In the month of January every year; 2nd and 3rd parts twice every year.
4. DCH course : Once in a year in the month of July.
5. Three months certificate course : The institute every year runs 3 months certificate course on paediatrics for general practitioners & other post graduate candidates e.g. MCPS.
   (Ist August – 31st October)
6. Training programme on IMCI (Integrated management of childhood illness), Essential Newborn Care for doctors and nurses, KMC (Kangaroo Mother Care) traing, ETAT (Emmergency Triage, Assessment and Treatment) training.

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DS (Child) H J 2016; 32(1) : 68

Students Qualified from Bangladesh Institute of Child Health

Undergoing Courses of BICH

Institution	Courses
Bangabandhu Sheikh Mujib Medical University	MD (Paediatrics) MD Paediatric Nephrology (sub-speciality) MD Neonatology (sub-speciality) DCH MS (Paediatric Surgery)
Bangladesh College of Physicians and Surgeons (BCPS)	FCPS Part II (Paediatrics) FCPS Neonatology FCPS Paediatric Nephrology FCPS Paediatric Surgery FCPS Paediatric Neurology & Development FCPS Paediatric Pulmonology
Dhaka University	B.Sc in Health Technology (Lab)

Students Qualified from BICH till- June-2016

Course	Number
DCH	329
MD (Paediatrics)	96
MS (Paediatric)	93
FCPS (Paediatric)	23
MD (Neonatology)	12
MD (Paediatrics Nephrology)	4
Total	**559**

Foreign Student Qualified from BICH Till January 2013

Country of origin	Course	Number
Nepal	DCH	23
	MS (Paed Surgery)	2
	MD (Paed)	1
India	MD (Paed)	1
Iran	DCH	1
Iraq	DCH	1
Somalia	DCH	1
Sudan	DCH	1
Total		**31**

Present Students from January 2016-June 2016

Name of Courses		Number of Students
DCH	-	11
MD (Paediatrics)	Phase-A	15
MD (Neonatology)	Phase-A	2
MD (Paediatric Nephrology)	Phase-A	2
MS (Paediatric Surgery)	Phase-A	9
FCPS (Paediatric)	Part-II	2
Total		41

DS (Child) H J 2016; 32(1) : 69

Seminar/Symposium & CME/CPD programs held at BICH (January to June, 2016)

Date	Topic	Presenter
23.01.2016	Vaccine Update	Department of Paediatric Cardiac Medicine (MU-1) Dhaka Shishu (Children) Hospital
31.01.2016	Epilepsy in Childhood	Department of Paediatric Neuroscience (MU-3) Dhaka Shishu (Children) Hospital
28.02.2016	Fluid & Electrolytes in Newborn	Department of Neonatal Medicine (MU-4) Dhaka Shishu (Children) Hospital
20.03.2016	Management of Bronchiolitis An Update	Department of Paediatric Respiratory Medicine (MU-5) Dhaka Shishu (Children) Hospital
24.04.2016	Hydrocephalus: Approach to Management	Department of Paediatric Neurosurgery and Paediatric Surgery Dhaka Shishu (Children) Hospital
22.05.2016	Primary Immunodeficiency in Children	Department of Neonatal Medicine (MU-4) Dhaka Shishu (Children) Hospital
05.06.2016	Zika Virus	Department of Paediatric Infectious Diseases & Community Paediatrics Dhaka Shishu (Children) Hospital

DS (Child) H J 2016; 32(1) : 70-71

INSTRUCTIONS FOR AUTHORS

Dhaka Shishu Hospital Journal is the official organ of BICH which is the academic wing of DSH. It is published twice a year since 1984. The present editorial board has decided that the cover design will be in accordance with the subject of editorial in each issue. The editor welcome articles to be published in the journal as leading article, original article, review article, case report, current issues of child health, short report and junior's page where trainee doctors are encouraged to publish their topic of interest.

Original papers written in *english* will be considered for publication provided these have not been published previously and are not under consideration for publication elsewhere.

Conditions for manuscript submission:

- All manuscripts will be subjected to peer and editorial review.
- Accepted manuscripts become the property of the *Dhaka Shishu Hospital Journal*. Any reproduction in whole or part will require written permission from the editorial board of the journal.
- The author should obtain written permission from appropriate authority if the manuscript contains any table; data or illustration from previously published in other journals. The letter of permission should be submitted with the manuscript.
- If the photographs are not disguised, permission from the patient or parents/ guardians to print should accompany the manuscript. Otherwise identity will be blackened out.
- Rejected manuscripts/electronic copies/ illustrations/photographs will not be returned to the authors.
- Editors are not responsible for courier/postal failure.

Manuscript preparation: The format of the Dhaka Shishu Hospital Journal complies with *"Uniform Requirements for Manuscripts Submitted to Biomedical Journals"* published by the International Committee of Medical Journal Editors in Vancouver, British Columbia in 1979 (the widely accepted ***"Vancouver style"***)' published in the Annals of Internal Medicine 1982; 96: 766-71. All scientific units should be expressed in *System International (SI) units*. Authors are referred to Annals of Internal Medicine 1987; 106: 114-29 for guidance in the use of SI units. All drugs should be mentioned in their generic form.

- Should be typed in english and on one side of A4 (290 x 210cm) size white paper, using *Times New Roman* font size 12, with single space.
- There should be one original and two paper copies and one IBM compatible electronic copy. (CD or Pen drive)
- There should be a margin of 2.5 cm at top and bottom, and 1.2 cm left and right.
- Pages should be numbered in english numerical at the upper right hand, consecutively, beginning with the title page.
- Manuscripts should be submitted in the following order:
  - Title : should not exceed100 characters (Font size 16, bold)
  - Name of authors, e.g. 1. Prof. Saiful Islam FCPS, FRCP, 2. Dr. Nurun Nahar MD, these two author's name will be written like this; S Islam[1], N Nahar[2], etc. (Font size 12) Author's designation and name of place of study will be written after the end of the abstract. (Font size 10).
  - ***Abstract with a specific format with five sections(about 350 words maximum): Background, Objective, Methodology, Results, Conclusion, address of correspondence. All these sections will be Times New Roman, Font size 12 and italic , bold but text will not be bold. No references are allowed in the abstract.***

- Text (Introduction, Materials & Methods, Results, Discussion, Conclusion).
- Acknowledgements
- References

- Photographs:
  - In CD/ Pen drive
  - With appropriate labeling (number in English numerical, title of photographs and title of manuscripts.) It should be placed in appropriate place of the article.
- Illustrations:
  - All illustrations should be cited in the text
  - Illustration should be numbered in English numerical and labeled properly, placed appropriately in relation to text of manuscript.
- Tables:
  - Should be appropriately titled.
  - Numbered with Roman numerical serially in order of text description.
  - Abbreviations if used, should be explained in footnotes.
  - Same table should not be repeated as chart.
- Figures:
  - Should be appropriately titled and title will be placed below the figure.
  - Numbered with English numerical serially in order of text description.
- Placement:
  - All photographs, illustrations, tables and figures should be placed in the text in their appropriate places where their description are given.
- References:
  - *References from journal* should be indicated by superscript numbers consecutively in the text and placed after full stop (e.g. ....has been reported.[1] or as shown by Akbar[2] .....) in the order in which they are first mentioned and should be listed in numerical order on a separate sheet at the end of the article.
  - References cited only in tables or legends or illustrations should be numbered in accordance with a sequence established by the first mention in the text.
  - Titles of journals should be abbreviated according to Index Medicus or given in full.
  - References must include: (i) all authors, surnames and initials (if there are 6 authors or fewer) or if there are more than 6 authors, the first six authors followed by et al; (ii) the full title of the paper; (iii) the abbreviated or full title of the journal in italic; (iv) the year of publication; (v) the volume no will be bold; (vi) the first and last page numbers followed by full stop.*Example*: Khan NZ. A study of mentally handicapped children: aetiology and associated factors. *Bangladesh Journal of Child Health* 1985; **9**(2):102-08.
  - *References from books* must include: (i) authors name, (ii) title of article, (iii) editors name/s, (iv) name of the chapter, (v) place of publication, (vi) name of publisher, (vii) year of publication and page numbers. *Example*:Razvani I. An approach to inborn errors of metabolism. In: Behrman RE, Kliegman RM, Jenson HB, editors. Nelson Textbook of Paediatrics.Philadelphia:Saunders,2004: p.397-98.
  - *Documents in electronic format* must include: (i) title, (ii) authors name,(iii) year of publication, (iv) web site address, (v) date of access. *Example*: United Nations Programme on HIV/AIDS. Children living in a world with AIDS. Geneva, 1978 (htpp://www.....), accessed on (dd/mm/year).

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