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EDITORIAL

Indoor smoke and prenatal and childhood growth: The role of (gestational) age

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Abstract

Growth at birth and during infancy predicts several outcomes in the immediate future as well as in the long term. Weight and height are commonly used surrogates of growth, however, infants and young children are constantly growing unlike adults. Hence, weight and height alone are insufficient measures of growth if the time component is not associated with them. Recent studies have investigated the relationship between indoor air pollution and growth using height and weight. In this commentary, I have argued using a directed acyclic graph, that a causal association between indoor pollution exposure and growth at birth cannot be established unless birth weight is adjusted for gestational age. Furthermore, to make any causal inference between growth during the first few years of life and indoor exposure, in addition to age standardization, studies must also account for fetal growth to discount any continuation of prenatal effects, which may be in the causal pathway. A careful consideration is warranted from future studies investigating these relationships.

Key words: Biofuel; Coal smoke; Wood smoke; Birth weight; Fetal growth; Height

Core tip: Prenatal and early childhood estimators of growth, such as birth weight, height *etc.* by themselves are inadequate measures for inter-individual comparison, unless accompanied by gestational or chronologic age. The existing evidence points toward an association between indoor air pollution and growth, however few considered age. In order to establish a causal relationship it is imperative to consider age adjusted growth.

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INTRODUCTION

Reduced growth prenatally and after birth, which is associated with childhood mortality and morbidity[1] and also with chronic diseases during adulthood[2], is considered a major problem in many developing countries. Coincidentally, indoor air pollution is also high in these countries[3]. More than half of the world population, predominantly from developing countries use some form of biofuel. A few studies have presented evidence that indoor air pollution may be one of the hitherto unknown factors associated with reduced prenatal and early childhood growth. This is a commentary on the studies mostly from the last decade, focusing on residual confounding that may arise if (gestational) age is not accounted for. There are many other sources of bias that pose similar threats to internal validity, which are not the focus of this commentary.

PRENATAL GROWTH

Prenatal exposure to indoor biofuel smoke and birth weight has now been investigated in several populations.

A Zimbabwean study[4] reported maternal exposure to combustion smoke from wood, dung or straw was associated with infants born with 175 g ($P < 0.01$) lower mean birth weight than those who resided in homes that used gas or electricity for cooking. In spite of the significant association, owing to the cross-sectional nature, it is difficult to establish if exposure preceded outcome. Furthermore, the association with birth weight may not be construed to be with prenatal growth, as it was not adjusted for gestational age.

A study on Guatemalan infants[5] reported 63 g (95%CI: 1-126) reduction in birth weight for those exposed to wood smoke compared to infants from households that used electricity or gas. This cross-sectional study accounted for some important variables but not gestational age and the possibility of exposure misclassification exists. This was one of the earliest studies and the magnitude of the point estimate is on the lower side compared to the estimates of other studies conducted subsequently.

Another Guatemalan study[6] reported 89 g (95%CI: -27-+204) higher birth weight and the association with low birth weight (LBW) was 0.74 (95%CI: 0.33-1.66) for infants born in families that used a chimney stove compared to infants born in families that used open fire. The study, though not sufficiently powered, provides higher quality of evidence owing to the longitudinal design but did not adjust for gestational age.

A population-based longitudinal study[7] from India, designed from a randomized trail reported a 105 g (95%CI: 140-69) reduction of birth weight and a RR of 1.49 (95%CI: 1.25-1.77) for LBW amongst those prenatally exposed to wood or dung smoke compared to those unexposed. The estimate was adjusted for a range of covariates including secondhand tobacco smoke (SHS) and a surrogate of socioeconomic status (SES). This study provides stronger evidence because of the design, availability of information on and adjustment for potential confounders as well as for the power to detect a significant difference in the exposed and unexposed group.

Another population based retrospective cohort study[8] from Pakistan reported adjusted OR of 1.64 (95%CI: 1.10-2.35) for LBW and -82 g (95%CI: -170-+9) reduction in birth weight (continuous) among those exposed to wood smoke compared to natural gas combustion exposure. The study accounted for a wide range of potential confounders directly, including tobacco use, maternal BMI *etc.* and others indirectly, using propensity scores. The association with LBW was significant, while the association with birth weight was not, even though, birth weight as a continuous variable would have more variations (than the dichotomy of LBW), and hence lend more power to the model, assuming both models had same number of observations. There was a degree of misclassification in growth when LBW (without gestational age) was considered because all LBW infants were not due to growth retardation. This dichotomy and hence the misclassification was absent in the continuous birth weight model and may have been the cause for the statistically non-significant result. Interestingly, the study from India[7] reported a RR of 1.21 (95%CI: 1.11-1.31) for small for gestational age (SGA, < 10 percentile of birth weight-for-gestational age), which was less than half of the LBW estimate (RR = 1.49) from the same study. SGA reflects retarded prenatal growth more appropriately than LBW or birth weight because it takes into account gestational age. It is appreciated that accurate gestational age measurement is a challenge in settings in which these studies were conducted.

A meta-analysis[9] pooled all the estimates from the above studies (and one more), to summarize information and increase power. However, is it prudent to pool together estimates from cross-sectional and longitudinal studies? Perhaps, a non-significant test of heterogeneity is an argument in favor. If one digs deeper, even the two longitudinal studies, the models from which the LBW/birth weight estimates were used for pooling[6,7], did not have a single covariate in common. Furthermore, it included two estimates from the Indian cohort study[7] one for term LBW and the other for preterm LBW. The original article did not anywhere associate gestational age with the LBW estimate (RR = 1.49); presumably the authors performed additional stratified analyses but the estimates (term LBW in the meta-analysis and all LBW in the original paper) are strikingly similar. The comparison in the meta-analysis is essentially between term and preterm LBW-while the former is certainly due to intrauterine growth retardation, the latter is due to early parturition and may or may not be from growth retardation? A careful approach with particular attention to study characteristics and their differences, beyond any statistical test, should be considered before choosing the meta-analytic approach, to avoid spurious results.

POSTNATAL GROWTH

A few studies also investigated exposure to indoor smoke and early childhood growth. About two decades ago the first study[10] provided evidence of an adverse association between exposure to smoke produced from gas burning during cooking and height at 10 years, a 3.3 cm reduction ($P < 0.03$) in the exposed compared to the unexposed. The cross-sectional study on Kuwaiti and European children was a modest attempt to explore the relationship by accounting for important determinants like SHS, SES, ethnicity *etc.* It acknowledged that there may have been selection bias because fewer Kuwaiti families participated and amongst those who did, a larger proportion used gas. The authors did not standardize height, which made the comparison amongst participants untenable unless they were all of 10 years when height was measured.

A seven country study[11] using data from national surveys investigated exposure to biofuel smoke (wood/straw/dung *vs* electricity/gas/biogas/kerosene) and child's height-

for-age and reported -0.13 (95%CI: -0.19--0.07) SD units reduction in a multilevel analyses. The analytical strategy and the wide range of covariates (including population differences) make this important evidence except the fact that owing to the cross-sectional nature temporal precedence of exposure cannot be established. In a categorical analysis it reported an OR of 1.27 (95%CI: 1.02-1.59) for severe stunting, defined as -3 SD $\leqslant Z <$ -2 SD, amongst exposed *vs* unexposed. Another study[12] using data from Indian national family health survey reported larger association for severe stunting, OR of 1.90 (95%CI: 1.49-2.42) for those using biofuels compared to those who used cleaner fuels (definition similar to the previous one). Amongst the strength was the large number of covariates used but it did not account for multistage clustered sampling and there may be some unaccounted variance in the models affecting the significance.

Two longitudinal studies provide evidence of the association between exposure to biofuel generated smoke and reduced height-for-age. The study from India[7] investigated growth at 6 mo and reported RR of 1.45 (95%CI: 1.20-1.75) for underweight (< -2 SD, weight-for-age) for those exposed to wood and dung smoke compared to those unexposed and RR of 1.30 (95%CI: 1.06-1.60) for stunting (< -2 SD, height-for-age) in the same comparison groups. As stated above, this is one of the high quality evidence we have so far. A second longitudinal study[13] measured continuous height-for-age of children 36 mo old in Caucasian children from the Czech Republic and reported 1.3 cm reduction in height of 36 mo old children exposed to coal smoke compared to those who were unexposed. Retarded growth (from biofuels exposure or something else) may be initiated prenatally; such a condition needs to be accounted if we are to estimate the magnitude of a true causal association with early childhood growth. The Czech study adjusted for growth at birth using birth weight-for-gestational age-and-sex thus discounting for any such conditions initiated prenatally.

The currently available studies are heterogeneous-one longitudinal study[13] reported continuous *Z*-scores while another[7] reported both continuous *Z*-scores and dichotomous stunting. Furthermore, the definition of stunting was different from those used in the cross-sectional studies.

CONCLUSION

The underlying question is - can exposure to indoor biofuel smoke impede normal growth before and after birth? Residual confounding is an important issue that needs to be addressed to answer this question. In case of prenatal growth without the adjustment for gestational age none of the evidence can be concluded as causal. Additionally, maternal tobacco smoking[14] and household ETS[15] exposure are also established confounders. ETS adjustment may appear to be conservative because some of the constituents of tobacco smoke and indoor biofuel smoke are similar (*e.g.,* PAH, nitrogen oxides, $PM_{2.5}$), it is still a robust approach to eliminate the possibility of type 1 error and to estimate the true magnitude of the effect. The converse argument against the idea could be that this may lead to over-adjustment, masking or attenuating a true association.

Childhood height and weight change relatively rapidly with inherent age and sex differences. Standardization for age and sex using reference populations is therefore necessary to make the results generalizable. Additionally, in the developing countries malnourishment is an important factor for retarded childhood growth and studies investigating indoor air pollution and postnatal growth in these settings should also account for factors (*viz.* SES and morbidity) that cause malnourishment. Interestingly, all but one childhood growth study used age-standardized measures[10], which is pertinent. If standardization by age is imperative for growth after birth, why is it not for prenatal growth, when it is well known that a week or even days of gestational age in the third trimester make a difference to birth weight? It will be inappropriate to establish causality between indoor air pollution exposure and prenatal growth, using birth weight (or LBW) without gestational age, which, at best, is a crude surrogate of growth.

Another key issue for studies on childhood growth is accounting for growth at birth. It is critical to differentiate reduction in growth due to the exogenous exposures (secondhand smoke or indoor air pollution) during childhood from that which is simply a continuation of a retarded trajectory initiated prenatally. A retarded trajectory initiated prenatally may be due to similar exposures accrued over the prenatal period or may be from other causes, *e.g.,* malnourished mother. Either way, it is important to make this distinction for an appropriate assessment of the magnitude of the association between indoor air pollution exposure and postnatal growth.

The key issues presented in the two paragraphs above can also be described using a Directed Acyclic Graph (DAG) shown in Figure 1. Age and gestational age are determinants of pre and postnatal growth, respectively and according to the definition of confounder these two variables need to be connected to exposure and outcome either through a directed or a backdoor path. Aside from the fact that higher age prolongs exposure, age or gestational age is not connected to indoor air pollution and the outcomes through any directed or unblocked backdoor paths in the DAG (Figure 1). However, age is absolutely necessary to measure growth when weight or height is used as the outcome. For example, an infant with 2800 g birth weight born at 42 wk does not have the same growth as another infant with the same weight but born at 37 wk. Therefore, it should not be mistaken that absence of any directed or backdoor path between age and exposure/outcome justifies it's exclusion from consideration in the relationship. On the contrary, weight or height as an outcome is an insufficient measure of growth without age. Additionally, the DAG shows that postnatal growth has a backdoor path connecting prenatal growth and indoor air pollution, which suggests adjustment of prenatal growth is imperative in any model

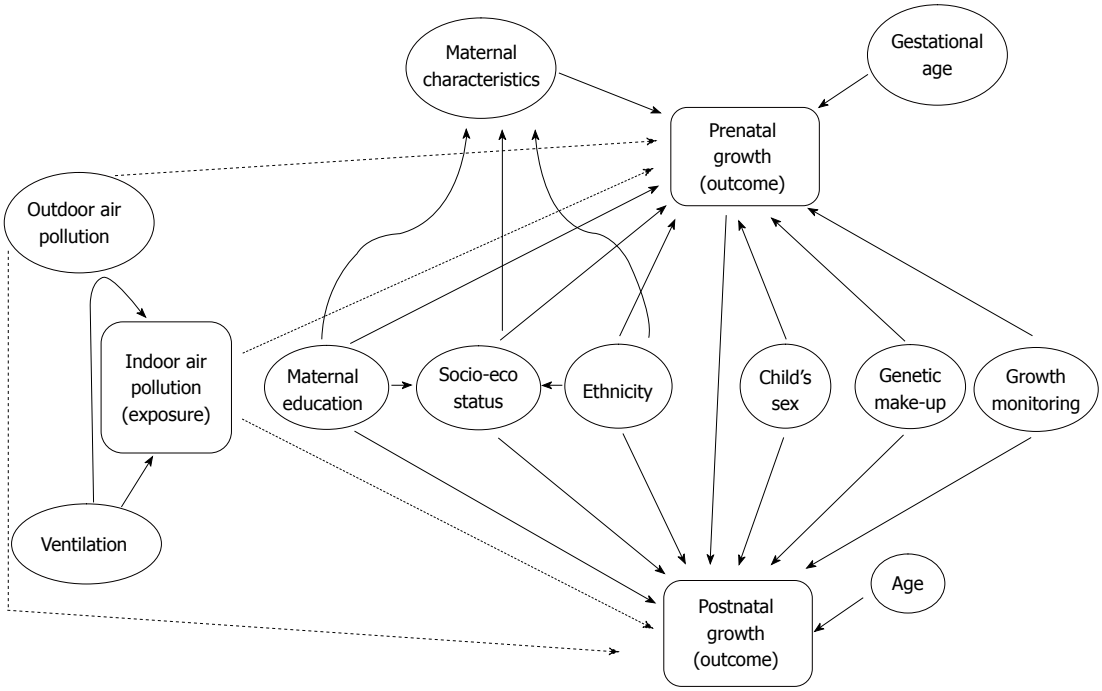

Figure 1 A directed acyclic graph depicting the relationship between indoor air and prenatal and early childhood growth.

associating indoor air pollution with postnatal growth.

The other variable that deserves a mention from the DAG (Figure 1) is outdoor air pollution. The causality of the relationship between outdoor air pollution and growth is yet to be ascertained. Meanwhile, outdoor air pollution will influence indoor air and vice versa if the windows and doors are kept open for long duration. This would apparently suggest that outdoor air pollution should be adjusted while investigating the relationship between indoor air pollution and growth. However, if outdoor and indoor air has the same pollutants and under the assumption of causal relationships, adjustment for one to investigate the relationship with the other, would be taking out the very association one is interested to find.

To conclude, the evidence point towards a potential adverse association between indoor air pollution exposure and growth, and encourages further well-designed investigation with adequate power, addressing the limitations of the current ones, to estimate the true magnitude. The last trimester and early childhood is marked by steady growth, it is therefore important to adjust for gestational age or chronologic age, respectively, to eliminate any differences in growth due to age before an adverse impact can be assessed. An exercise of procuring these datasets to perform a standardized analysis can be the next step forward.

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REVIEW

Echocardiography in children with Down syndrome

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Abstract

Congenital heart disease is a common problem in children with Down syndrome (DS). Echocardiography plays an important role in the detection of both structural and functional abnormalities in this group of patients. Fetal echocardiography can help in the early recognition of DS by detecting soft markers of DS, but its main role is to define the exact nature of the suspected cardiac problem in the fetus. Postnatal echocardiography is mandatory in the first month of life for all neonates with DS. It is also indicated before any cardiac surgery and for serial follow-up after cardiac surgery. In this article, we discuss the types and mechanism of cardiac abnormalities in DS children and the role of both fetal and postnatal echocardiography in the detection of these abnormalities.

Key words: Fetal; Echocardiography; Congenital heart disease; Down syndrome

Core tip: Cardiac affection is a common issue in the Down syndrome (DS) population, in the form of both congenital and acquired heart disorders. Echocardiography plays an important role in the detection of such disorders. Fetal echocardiography can detect cardiac disorders as early as 10-12 wk of gestation. Echocardiographic examination is recommended for all neonates with DS in the first month of life, before any cardiac surgery, to follow up after cardiac surgery and for serial evaluation of pulmonary hypertension. It is also indicated before involvement in non-cardiac major surgery and before involvement in physical exercise.

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INTRODUCTION

Down syndrome (DS) is an autosomal trisomy 21 and is one of the most frequently occurring chromosomal abnormalities. DS occurs once in every 600 to 800 live births and is frequently associated with congenital heart disease (CHD). The incidence of CHDs increases from 0.8% in the general population to approximately 40%-65% in patients with DS. At the same time, children with DS comprise approximately 10% of all children with CHD. Such malformations include all structural and functional cardiac defects present at birth, even if discovered later in life. These malformations can be single or multiple and usually lead to significant implications for the children and their families. These children may develop congestive heart failure, pulmonary vascular disease, pneumonia, or failure to thrive. CHDs are the most common cause of death in children with DS during the first two years of life[1,2].

Atrioventricular septal defects (AVSD; with or without other CHD) and ventricular septal defects (VSD; with or without other CHD) have both been reported as the most common congenital heart defects and make up approximately 45% and 35% of CHD associated with DS, respectively. Additionally, isolated secundum atrial septal defect (ASD), isolated persistent patent ductus arteriosus (PDA), and an isolated Fallout of Tetralogy

(TOF) individually comprise approximately 8%, 7%, and 4% of CHD associated with DS, respectively. The remaining 1% of CHDs found in patients with DS includes arch abnormalities (aortic coarctation, right aortic arch, aberrant right subclavian artery). DS tends to be associated with the more severe forms of endocardial cushion defect, while the inlet VSD is common in trisomy 21. Several cardiac lesions seen in the non-DS population are rarely if ever found in individuals with DS, *e.g.*, heterotaxy, aortic coarctation or transposition of the great arteries. These differences between the types of CHD in DS and non-DS populations could suggest the impact of a third copy of a gene or genes on chromosome 21 on only specific developmental points[3,4].

There is an increased prevalence of persistent pulmonary hypertension of the neonate (PPHN) in children with DS. These children have an increased risk of developing PPHN even in the absence of structural heart disease and should be followed until resolution of the pulmonary hypertension[5]. Weijerman *et al*[6], 2010 observed a significantly increased and elevated incidence of PPHN in neonates with DS (5.2%) compared to the general population. Another rare presentation in neonates with DS is the association of hypertrophic cardiomyopathy with complete atrioventricular canal defect in an infant with trisomy 21[7].

While a defect of the atrioventricular canal remains the most common heart malformation in children with DS, the type of associated CHD may be affected by various factors. For example, the parents' consanguinity status could affect the pattern of CHDs. Al-Jarallah[8], 2009 documented a slightly higher frequency of CHD in a sample of DS children from a Saudi population with a high consanguineous marriage rate. That study found that VSD was the most frequently detected CHD with the predominance of left-right shunt lesions and the relative rarity of cyanotic and complex CHD in this DS population. However, Chéhab *et al*[9] previously showed that the risk of congenital cardiac anomalies in children with DS was not associated with the parents' consanguinity; instead, having a maternal age above 32 years was more associated with a lesser occurrence of congenital cardiac anomalies in children with DS.

Ethnicity and sex are additional factors that appear related to the type and frequency of CHD in the DS population. In a study conducted in the United States of America, Freeman *et al*[10] showed that atrioventricular septal defects had the most significant sex and ethnic differences, with twice as many females affected and with twice as many blacks and half as many Hispanics affected compared to whites. In the Saudi population with DS, VSD was the most common (33.3%) followed by AVSD (22.8%), ASD (21.1%), patent ductus arteriosus (14%) and tetralogy of Fallout (11%). In a Turkish sample, the most common single defect was AVSD (34.2%), followed by second ASD (16.7%) and VSD (16.5%)[11,12]. On the other hand, PDA was the most common cardiac malformation observed in Guatemalan children with DS, followed by VSD, ASD and then AVSD[13]. The most common cardiac malformations in Mexican children with DS were ASD, VSD and PDA, while the AVSDs were less common than the other malformations[14].

Children with DS may also have dysfunctional autonomic cardiac regulation even in the absence of concomitant congenital heart disease, which may be manifested mainly in a reduced heart rate response to excitatory stimuli, including arousal from sleep. This is especially noted if accompanied by sleep-disordered breathing (SDB). O'Driscoll *et al*[15] described a compromised acute cardio-respiratory response and dampened sympathetic response to SDB in children with DS and SDB compared to typically developed children with SDB. This altered response may be due to inadequate sympathetic activation or blunted vagal withdrawal and could reflect autonomic dysfunction in children with DS that may place them at increased risk for cardiovascular complications, such as pulmonary hypertension.

DS children with CHD have a greater predisposition to develop irreversible pulmonary arterial hypertension (PAH). The increased incidence of pulmonary hypertension in DS children could be a result of additional related problems, such as an upper airway obstruction, pulmonary hypoplasia, structural lung disease, thinner media of the pulmonary arterioles, abnormal pulmonary vasculature growth, alveolar hypoventilation, recurrent pulmonary infection or gastro-esophageal reflux[16]. It has been suggested that PAH may develop earlier and may have a more violent course in patients with Down's syndrome, carrying with it a high risk of morbidity in a relatively young patient[17,18].

MECHANISM OF CHD IN CHILDREN WITH DS

Heart development in humans is complex and starts very early, from the third to eight weeks of gestation. Development begins with a primitive tube that beats at 25 d of gestation and ends in the four-chamber heart. Many steps occur after the formation of the primitive heart tube, including looping, cell migration, cell transition, and septation events[19]. The development of CHD is a multifactorial condition and is affected by a series of molecular signaling pathways and morphological events. In children with DS, it is postulated that variations in gene dosage of chromosome 21, environmental factors and genetic modifications not linked to chromosome 21 contribute to the development of CHD[20].

Down's syndrome is most commonly caused by the presence of an extra copy of all or part of human chromosome 21 (Hsa21). The extra set of the approximately 200-300 genes on the chromosome leads to the many abnormalities associated with this condition. Due to triplication of Hsa21, there is a 1.5-fold increase in the expression of some, if not all, of these genes present in the

extra copy. However, all of these genes do not necessarily exhibit a straightforward 1.5-fold increase in expression, and only 30% of *Hsa21* genes are significantly overexpressed. Gene expression may be regulated by dosage compensation, which means that only a subset of these genes will exhibit the expected 50% increase in expression[21]. Genetic imbalance caused by the presence of an extra copy of chromosome 21 will seriously disrupt one or more developmental pathways. This imbalance could also induce also an interaction between *Hsa21* genes and other disomic genes with relatively subtle or massively disruptive effects on genes located on chromosomes other than 21[22]. These effects could be mediated through modulation of transcription factors, chromatin remodeling proteins, and related molecules or other targets. Thus, the imbalance-induced dysregulation of pathways involved in heart development may cause the cardiac defects observed in DS[23].

The DS critical region (DSCR) is located on the long (q) arm of chromosome 21 and contains a number of genes that are thought to be responsible for some, if not all, of the features of DS. These genes may include the DS critical region 1 gene, or DSCR1, on chromosome 21q11.2; DSCR2 on chromosome 21q22.3; DSCR3 on chromosome 21q22.2; DSCR4 on chromosome 21q22.2; and DSCR5 in the chromosome region 21q22.1-q22.2. DS critical region 1, also known as Calcipressin-1, Adapt78, myocyte-enriched calcineurin-interacting protein 1 or regulator of calcineurin 1, is a 252 amino acid protein that belongs to the RCAN family (regulators of calcineurin) and exists as 4 alternatively spliced isoforms. DSCR1 is abundantly expressed in skeletal muscle, as well as in the brain and heart, and is thought to influence cardiac and nervous system development[24]. DSCR1 is possibly part of a signal transduction pathway involving both the heart and brain and is implicated in cardiac valve formation and in the inhibition of cardiac hypertrophy[25]. Overexpression of DSCR1 may be involved in the pathogenesis of DS, in particular mental retardation or cardiac defects[26]. Barlow *et al*[27] had previously mapped the DS-CHD region in humans to a 5.27-Mb chromosomal segment containing 82 genes, and Korbel *et al*[28] had narrowed this segment to a 2.82-Mb critical region likely involved in a DS-CHD endocardial cushion defect.

The presence of specific gene variants or modifiers could, in addition to trisomy 21, further increase the susceptibility to cardiac defects. One such genetic modifier is cysteine-rich with epidermal growth factor (EGF)-like domains (CRELD1), initially identified as a candidate for the AVSD2 locus. CRELD1 belongs to an epidermal growth factor-like family and encodes a cell surface protein that likely functions as a cell adhesion molecule. CRELD1 encodes a novel cell adhesion molecule that is expressed during cardiac cushion development. Missense mutations in CRELD1 have been found in DS patients with AVSD. CRELD1 (3p25.1) is one of the well-studied non-chromosome 21 loci variations that may predispose one to a heart defect. Other genetic modifiers have also been shown to affect heart development. For example, somatic mutations in basic helix-loop-helix (bHLH) transcription factor *HEY2* (gridlock) have been identified in CHD in people with DS but not in euploid populations with heart defects[29-32].

Environmental factors interact with the trisomic genome and may modify the occurrence of associated CHD in children with DS. Maternal cigarette smoking, for instance, has been associated with ASVD, TOF, and ASD but not with VSD[33]. However, Khoury and Erickson observed an inverse relationship between maternal alcohol use and the presence of VSD in children with DS. Maternal folic acid supplementation may be associated with a reduced risk for CHD. Individuals with DS are at a high risk for CHD and have been shown to have abnormal folate metabolism[34]. Bean *et al*[35] found that a lack of maternal folic acid supplementation was more frequent among infants with DS and AVSD or ASD than among infants with DS and no heart defect or with VSD.

FETAL ECHOCARDIOGRAPHY

Fetal echocardiography is considered in cases of suspected DS. Such instances include the observation of a fetal nuchal translucency measurement of 3.5 mm or greater in the first trimester, the presence of a single umbilical artery or following an abnormal or incomplete cardiac evaluation on an anatomic scan, 4-chamber study. Fetal echocardiography can identify fetal cardiac structures as early as 10-12 wk of gestation using vaginal probes with high-resolution transducers, while conventional trans-abdominal echocardiography can detect fetal cardiac anomalies by 16-18 wk of gestation. The optimum period in which to perform a screening examination is at 20-22 wk because, at that time, the fetal cardiac structures can be defined more clearly with ultrasound screening in more than 90% of cases. Fetal echocardiographic examination can be difficult because of fetal physiology, the effect on flow across defects and valves, the inability to see the fetus for orientation reference, and an inability to examine the fetus for clinical findings. Likewise, a detailed heart evaluation can be very difficult during the third trimester because of acoustic shadowing, as in cases of maternal obesity or prone fetal position[36].

Fetal echocardiography can assist in the early recognition of DS by detecting soft syndrome markers, but its main role is to define the exact nature of the cardiac anomaly suspected in the fetus. Such an assessment helps both the parents and the treating physician make the most informed decisions. Fetal echocardiography can provide the possibility of pregnancy termination in cases of severe malformations and of treating in utero the potentially treatable and less severe disorders, *e.g.*, fetal supraventricular tachycardia[37]. In addition, fetal echocardiography can identify babies with complex congenital heart diseases that need delivery in a special tertiary care level center equipped with a Neonatal Intensive Care Unit so that during the transition from pre- to post-natal

life, the baby does not face periods of hypoxia or acidosis and can be given immediate care[38].

Soft syndrome markers are ultrasound findings that are considered abnormal and whose presence increases the risk for underlying fetal aneuploidy and congenital heart diseases. These markers are nonspecific and could also present in fetuses without abnormalities. They are often transient and can be readily detected during the second trimester[39].

Echogenic intracardiac foci (EICF) are examples of these soft markers that have been associated with DS, as well as with trisomy 13. They are a common finding seen in approximately 4% of obstetric sonograms, and their incidence can vary with ethnicity. The lowest rates of EICF are seen in black populations, while the highest rates occur among Asian patients. These foci are of no hemodynamic or other short or long term clinical or functional significance, but their importance arises from being a possible marker of a chromosomal abnormality[40]. These foci are discrete areas of increased echogenicity in the region of papillary muscles. They may be single or multiple and usually present in the left ventricle (88% of cases) but occasionally present in the right ventricle (5%) and may be bilateral in approximately 7% of cases. The right-sided, biventricular, multiple, or significantly obvious EICF are associated with a higher risk for fetal aneuploidy compared with the more common single, left ventricular EICF[41,42].

These foci may result from the aggregation of chordal tissues that have failed to fenestrate completely, the enhancement of abnormal tissue, or from a collection of fibrous tissue with increased echogenicity. They may also represent true microcalcifications within the cardiac muscle[43]. The larger size of the left ventricular papillary muscle and the larger masses of chordae tissue, the more likely is to see echogenic foci in this area[44].

A finding of EICF is subjective. Its detection depends on a variety of factors, including the resolution of the sonographic equipment, technique, thoroughness of the examination, and the sonographer's experience[45]. For proper visualization and grading of EICF, an appropriate transducer frequency ($\leqslant$ 5 MHz) and an appropriate gain setting should be used. These foci can be diagnosed on the standard 4-chamber view of the fetal heart. Fetal position is also important because intracardiac foci are best visualized when the cardiac apex is oriented toward the transducer. The foci echogenicities are graded according to their comparison to the surrounding bones, especially the thoracic spine. In grade 1, the echogenicity is less than that of the thoracic spine and the EICF image will be lost before that of the thoracic spine. Grade 2 suggests that the echogenicity is representative of bone and that the EICF and thoracic spine images disappear at the same gain setting. In grade 3, the echogenicity of the EICF is greater than that of bone, and the thoracic spine image will be lost before that of the EICF[46].

The foci should be differentiated from other cardiac conditions that can be misdiagnosed with these foci. These conditions include intra-cardiac tumors (rhabdomyomas, teratomas, fibromas, hemangiomas), ventricular thrombi (especially if adherent to the papillary muscles in the left ventricle), dystrophic valves, air in the chambers from fetal demise, endocardial fibroelastosis that is usually multiple and along the endocardial surface, idiopathic infantile arterial calcification, viral infections or metabolic disorders[47].

The presence of an aberrant right subclavian artery (ARSA) is another potential new soft marker that is more commonly seen in fetuses with trisomy 21 and other chromosomal defects than in normal fetuses. An ARSA has been found postnatally in 1%-2% of normal individuals (from neonates to adulthood) at autopsy, but its incidence is increased in cases of DS, with figures ranging between 2.9% and 37.0%[48]. Although it can be considered a weak marker, the second trimester diagnosis of an ARSA should prompt a detailed search for additional "soft markers" and potential structural defects[49].

Once DS is suspected, fetal echocardiography should be performed to detect associated structural cardiac abnormalities. A cardiac anomaly can be classified according to its detectability by fetal echocardiography and its severity. With regard to detectability, the structural cardiac abnormalities are classified as detectable or undetectable. Detectable cardiac anomalies are those that can be identified during routine antenatal assessment incorporating a four-chamber view of the heart at approximately eighteen week' gestation. These abnormalities usually produce marked cardiomegaly, an obvious abnormality of the atrioventricular connection, or a disparity between the sizes of the atria or ventricles or both. In undetectable abnormalities, the four-chamber view fails to identify them as types of major malformations affecting the left or right heart outflows. Ventricular septal defect, pulmonary stenosis, aortic stenosis, and coarctation of the aorta are considered among the possible undetectable abnormalities[37].

With regard to severity, structural cardiac abnormalities are classified into "complex," "significant," and "minor" heart disease. Complex heart disease occurs when there is an atretic or hypoplastic valve or cardiac chamber, which may include a hypoplastic left heart, Truncus arteriosus, pulmonary atresia with ventricular septal defect, or a double outlet ventricle. Heart diseases are considered significant when four chambers of normal or increased size and four valves are present but require intervention, *e.g.*, Ebstein/tricuspid valve dysplasia, significant complete AVSD, large VSD, partial AVSD, ASD, PDA, severe aortic stenosis, severe pulmonary stenosis (PS), transposition of the great arteries (TGA), coarctation of the aorta, total anomalous pulmonary venous connection, or TOF. Cardiac abnormalities are considered minor when no treatment is required, such as in cases with small VSD or relatively mild aortic or pulmonary valve stenosis. If there are multiple cardiac abnormalities, the disorder is

classified according to the most severe diagnosis. There are two exceptions from this classification: the endocardial fibroelastosis, which is classified as "complex", and complete AVSD, which is classified as "significant"[50].

There is a further classification according to severity suggested by Choi *et al*[51], who identified 5 classes of fetal echocardiography results: normal, minor abnormalities, simple cardiac anomalies, moderate cardiac anomalies, and complex cardiac anomalies. Minor abnormalities are those that do not require any interference, such as PFO, isolate peripheral PS, abnormally looking aortic arch, simple right aortic arch, tortuous ductus without obstruction, and mild right ventricular dilation with or without tricuspid regurgitation (at most mild regurgitation). Simple cardiac anomalies are defined as a simple defect or a defect completely correctable by medical treatment, such as VSD, ASD, and possibly CoA. Moderate cardiac anomalies include defects that are surgically correctable with a low risk for reoperation, such as TOF, CoA, AVSD, and complete TGA. Complex cardiac anomalies are defined as defects correctable with surgery but that carry a high risk for sequelae or defects potentially suitable for a Fontan procedure, such as a double outlet right ventricle, TGA with PS, critical PS, and other Fontan candidates (pulmonary atresia with intact ventricular septum, functional single ventricle, hypoplastic left heart syndrome). There is an additional classification according to surgical risk that has the disadvantages of having great variability between institutions and the need to change the risk stratification in accordance with future advancements in surgical treatment[51,52].

For optimal views of the heart, it is best to direct the fetal cardiac apex toward the anterior maternal wall. Optimization of the sonographic images could be achieved by appropriate adjustment of technical settings, such as acoustic focus, frequency selection, signal gain, image magnification, temporal resolution, harmonic imaging, and Doppler-related parameters. Accurate interpretation of obtained echocardiographic images will depend on a firm understanding of the anatomy of the fetal heart, either to diagnose a congenital abnormality or to confirm normality. According to guidelines from the American Institute for Ultrasound in Medicine, fetal echocardiography should include imaging of the aortic arch, ductal arch, four-chamber view, inferior vena cava, left ventricular outflow tract (LVOT), right ventricular outflow tract (RVOT), short-axis views ("low" for ventricles and "high" for outflow tracts), superior vena cava, and three-vessel and trachea views[53].

For fetuses with major CHD, a full diagnosis requires a sequential segmental approach, similar to postnatal practice[54]. The first step taken in studying the fetal heart is to definitely recognize the "Situs." *In situs* solitus, the inferior vena cava (IVC) is anterior and to the right of the aorta, the abdominal aorta is posterior and to the left of the spinal cord, the gastric air bubble is on the left side and the liver is on the right. In situs inversus, there is a mirror image pattern, with the aorta to the right of the IVC, and in situs ambiguous, the aorta and IVC are located on the same side of the spine in right isomerism, while the aorta is centrally located with an interrupted IVC in left isomerism[55]. For determination of the cardiac position and axis, the heart is normally located within the left chest, with the apex pointing to the left (levocardia). In dextrocardia, the heart is located within the right chest with the apex pointing to the right, while in mesocardia the heart is centrally located with the apex pointing anteriorly. Dextroposition should be distinguished from dextrocardia by the normal left-sided axis of the heart despite being displaced to the right due to extracardiac reasons (for example, diaphragmatic hernia, congenital pulmonary cystic adenomatoid malformation, pleural effusion, *etc.*)[38,56].

The four-chamber view (Figure 1A and B) is a key view of the fetal heart. It is effective in prenatal cardiac screening and can detect 43%-96% of fetal anomalies. It has the advantage of using fetal ribs as external reference points to ensure that the sonographer has "cut" the thorax in the appropriate plane. In a correct four-chamber view, there should be the appearance of a single rib around the fetal thorax. The four-chamber view is situated in a more horizontal plane than in the postnatal period because of the large liver. It can show the two atria and ventricles along with atrioventricular (AV) valves (mitral and tricuspid) and septa (interventricular and interatrial)[57]. The detectability of CHDs by fetal echocardiography increases if the outflow tracts are examined as well as the four-chamber view, but appreciation of abnormalities in the outflow tracts is more challenging than in the four-chamber view[58]. "Extended basic views" of LVOT and RVOT increase the sensitivity of anomaly detection. Alternatively, a comprehensive set of five short-axis projections can be acquired. The LVOT view can be obtained by tilting the transducer 45° from the four-chamber view perpendicular to the septum. This view can show the aorta originating from the LVOT with its main great vessels of the head and neck appearing distally. The RVOT view can be obtained by further rotation in the same direction and by gentle rocking the transducer from the LVOT view. In this view, the pulmonary artery can be seen rising from the RV and dividing into its main branches. The left pulmonary artery and ductus arteriosus can be observed, and the ascending aorta is seen centrally, wrapped by the RV and PA. A comprehensive set of five short-axis projections can also be obtained by cranial or caudal angulation of the ultrasound probe from the four-chamber view with the ventricular septum parallel to the ultrasound beam and the fetal spine[54]. The pulmonary artery and RVOT can be visualized in the most cephalad short-axis view as they course around the aorta. Despite being small, the fetal pulmonary branches bifurcation can still be identified. The ductus arteriosus can likewise be identified and traced into the descending aorta. With caudal and more horizontal angulation from this plane, the ventricles and their respective AV valves can be observed. These views are better for the detection of conotruncal

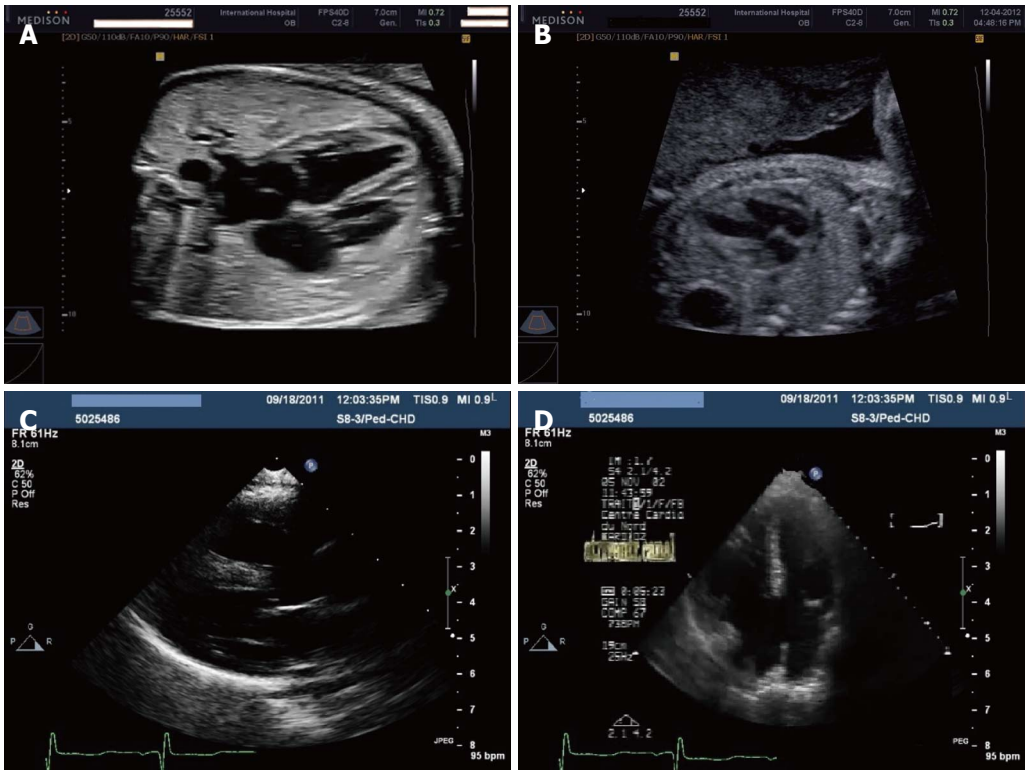

Figure 1 Ultrasonography. A: Normal 4-chamber view by fetal echocardiography at 26 wk gestation; B: Four-chamber view by fetal echocardiography at 22 wk gestation showing ventricular septal defects; C: Left-parasternal long-axis view in an infant with Down syndrome and complete atrioventricular (AV) canal and pulmonary hypertension; D: Apical 4-chamber view in an infant with Down syndrome and complete AV canal and pulmonary hypertension.

abnormalities that could otherwise be missed in more routine views. However, a specific diagnosis should not be made from a single plane.

The left and right fetal ventricles are nearly the same size, and the diameter of the pulmonary artery is typically larger than the aorta by approximately 10%. Ventricular volumes can be measured in 2-D mode using the Simpson rule. Other important measures include the RV/LV ratio, LV wall thickness, septal wall thickness, left atrial dimension, PA diameter, and aortic root diameter. These measures should be plotted against gestational age, which can be determined by measurement of the biparietal diameter or fetal length. These measures are helpful in the diagnosis of ventricular hypoplasia (either left or right) and cardiomegaly due to various congenital abnormalities, including pericardial effusion, aneurysms, cardiomyopathies, or tumors. Fetal heart motion, heart rate, wall thickness, chamber size, and motion of the valves or myocardium can be easily traced by M-mode, which can provide information on wall thickness and ventricular shortening fraction. The fetal long axis function may provide additional insight into endocardial function, which is most usefully in the detection of early ischemic changes before the development of sonographically detectable endocardial fibroelastosis. Moreover, color Doppler can be used to detect vascular flow through cardiac chambers, vascular structures, and septal defects[59-63].

Clur *et al*[64] showed that cardiac functions in trisomy 21 fetuses were abnormal irrespective of the presence of CHD. Evidence for cardiac loading (increased preload and afterload) and LV systolic (in the first trimester) and later diastolic dysfunction was observed. The authors showed significant reduction of tricuspid valve (TV) A-wave velocity and aortic valve peak velocity in trisomy 21 compared with normal fetuses. In addition, they also identified significant increases in the TV-E/A ratio and the ductus venosus-pulsatility index for veins and decreased pulmonary valve peak velocity. Moreover, the authors observed some evidence of left ventricular (LV) systolic dysfunction, such as a reduction of stroke volume (SV) and an increased myocardial performance index. They also found significant reduction of the mitral valve A-wave peak velocity and E/TVI ratio after 14 wk of gestation in the trisomy 21 fetuses with normal hearts compared to the controls with increased nuchal translucency thickness.

A complete AV canal can be more easily visualized in a 4-chamber view than an ASD primum, so that diagnosis of ASD primum type should be considered whenever a defect is noted in the portion of the atrial septum near the AV valves (septum primum). Opening the AV valve during diastole makes a large complete AV canal more obvious on the 4-chamber view, while during systole, the atrial and ventricular septal defects can be clearly seen above and below the closed AV valves, rendering its diagnosis possible as early as the late first trimester. Color

flow Doppler can show mixing of flow in the area of the septum primum defect, the dysplastic AV valves, and the ventricular septal defect. It can also show a lack of separation between the right and left ventricular inflow tracts in diastole, producing a classic "H" configuration. Color flow Doppler is also helpful in detecting and evaluating the degree of dysplastic AV insufficiency that may be severe enough to produce fetal heart failure with ascites[65].

TRANSTHORACIC ECHOCARDIOGRAPHY

Although it has been recommended that infants with trisomy should have an echocardiogram in the first month of life, the value of routine neonatal echocardiography in this population is still in debate. Nevertheless, physical examination alone is not sufficient to identify cardiovascular anomalies in neonates with DS. In the newborn with DS, the potential benefits of early diagnosis, in the context of physical examination findings, should be considered in determining whether an echocardiogram should be performed during the neonatal period. Echocardiography should be obtained in all children with DS before proceeding with surgery[66,67].

Echocardiographic examination can provide thorough real-time, non-invasive anatomic and functional information at relatively low cost. In neonates, the echocardiographic windows are more easily obtained and clearer than at any other age because of the reduced interference by lung tissue, which is impenetrable to ultrasound, and because the heart and the great vessels are nearer the probe. Echocardiographic examination should be conducted systematically with the classic standard views [left parasternal, apical, subcostal and suprasternal (Figure 1C and D)] and completed with Doppler ultrasound (color Doppler, pulsed Doppler and continuous wave Doppler). Trans-thoracic echocardiographic examination can usually detect cardiac defects in most cases. It can also determine the level of intra-cardiac shunting, along with its degree and direction, which can be confirmed by saline contrast injection. One mL of saline/blood mixture (which provides a greater intensity and more prolonged effect than the use of agitated saline only) is rapidly injected into a peripheral vein while capturing a four-chamber view of the heart. The simultaneous appearance of bright echoes from air bubbles in the fluid in the right ventricle and left atrium documents right-to-left atrial shunting as the air bubbles produced by hand agitation are too large to cross the pulmonary vascular bed, thereby predominantly aiding visualization of the right heart. The injection of the fluids into a vein that drains into the inferior vena cava could produce better results[68,69].

Echocardiography can also evaluate cardiac functions. For example, left ventricular systolic function can be evaluated by measuring left ventricular ejection fraction, fraction shortening, SV, stroke index, cardiac output and cardiac index. Left ventricular diastolic function can be evaluated by measuring the left ventricular inflow velocity pattern or trans-mitral flow velocity pattern [the early diastolic filling velocity (E-wave) is normally higher than the peak atrial filling velocity (A-wave)], the pulmonary venous flow velocity pattern (pulmonary venous flow velocity pattern usually consists of the antegrade flow during ventricular systole (S-waves: S and S), antegrade flow during early ventricular diastole (D wave), and retrograde flow), the flow propagation velocity (Vp) during the rapid filling period and the peak early diastolic velocity of the mitral annulus (Ea, E'). The global function of LV can be measured using the Tei index. The Tei index is the first comprehensive index of cardiac functions that covers both systolic and diastolic functions, and it deteriorates and improves with either systolic or diastolic dysfunction[70].

The quantitative assessment of RV size and function is often difficult because of the complex geometric anatomy, anatomical position under the sternum and the heavily trabeculated chamber with poor endocardial definition. However, 2-D echocardiography can easily obtain valuable information about RV size and function. Right ventricular dilatation, compared to the LV size, is a sign of RV dysfunction. Additionally, abnormal motion of the interventricular septum and the eccentricity index estimate RV pressure overload. The Tei index and the tricuspid annular plane systolic excursion both correlate well with RV function[71].

Echocardiography can also evaluate the presence and severity of pulmonary arterial hypertension, which is relatively more common in DS neonates than in non-DS neonates. Doppler echocardiography allows estimation of both systolic and diastolic pulmonary artery pressure (PAP). The systolic PAP can be estimated by measuring the tricuspid regurgitation jet that represents the right ventricular-right atrial pressure gradient. Both PAP and RV systolic pressure are nearly equal in the absence of stenosis of the RV outflow tract, which is the reason that systolic PAP is commonly estimated by techniques that measure RV systolic pressure[72].

Additionally, echocardiography is a useful tool for following up cases and for evaluating treatment outcome. Likewise, it has a further role even in the absence of congenital heart disease. Echocardiography can detect the presence of pericardial effusion in children with DS and hypothyroidism. A number of case studies have described the presence of pericardial effusion in this group of patients. Anah *et al*[73], for instance, described the presence of a complex association of DS-hypothyroidism-pericardial effusion.

Echocardiography may also reveal impaired cardiac function even in the absence of congenital or structural cardiac defects. A number of studies have documented impaired cardiac functions in patients with DS. They recommended echocardiographic examination before involving patients with DS in surgery or physical exercise, even in the absence of structural cardiac diseases[74,75].

Recommendation

Echocardiographic examination is recommended in chil-

dren with DS in the following situations: (1) in the first month of life for all neonates with DS; (2) before any cardiac surgery; (3) follow-up after cardiac surgery; (4) serial evaluation of pulmonary hypertension; (5) before involvement in major non-cardiac surgery; and (6) before involvement in physical exercise.

CONCLUSION

Echocardiography plays an important role in the detection of both structural and functional abnormalities in children with DS. Fetal echocardiography can help in the early recognition of DS by detecting soft markers of DS; however, its main role is to define the exact nature of the cardiac problem suspected in the fetus. Postnatal echocardiography is recommended in the first month of life for all neonates with DS. It is also indicated before any cardiac surgery and for serial follow-up after cardiac surgery.

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REVIEW

Bacterial colonization and intestinal mucosal barrier development

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Abstract

The intestinal tract is colonized soon after birth with a variety of ingested environmental and maternal microflora. This process is influenced by many factors including mode of delivery, diet, environment, and the use of antibiotics. Normal intestinal microflora provides protection against infection, ensures tolerance to foods, and contributes to nutrient digestion and energy harvest. In addition, enteral feeding and colonization with the normal commensal flora are necessary for the maintenance of intestinal barrier function and play a vital role in the regulation of intestinal barrier function. Intestinal commensal microorganisms also provide signals that foster normal immune system development and influence the ensuing immune responses. There is increasingly recognition that alterations of the microbial gut flora and associated changes in intestinal barrier function may be related to certain diseases of the gastrointestinal tract. This review summarizes recent advances in understanding the complex ecosystem of intestinal microbiota and its role in regulating intestinal barrier function and a few common pediatric diseases. Disruption in the establishment of a stable normal gut microflora may contribute to the pathogenesis of diseases including inflammatory bowel disease, nosocomial infection, and neonatal necrotizing enterocolitis.

Key words: Bacterial colonization; Intestinal barrier; Intestinal microflora; Microbiota; Neonatal necrotizing enterocolitis; Nosocomial infection; Premature infants; Short chain fatty acids

Core tip: This review summarizes recent advances in understanding the complex ecosystem of intestinal microbiota and its role in regulating intestinal barrier function and a few common pediatric diseases. There is increasingly recognition that the stimulation of initial intestinal microbial colonization is important for proper maturation of the innate immune system and continued regulation and maintenance of intestinal barrier function. Disruption of the establishment of a stable normal gut microflora may contribute to the pathogenesis of diseases including inflammatory bowel disease, nosocomial infection, and neonatal necrotizing enterocolitis in premature infants.

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INTRODUCTION

The human gut is home to a large collection of micro-

organisms, the composition of which varies along the intestinal tract. The important role of normal microbial intestinal colonization in human health has gained increased recognition over the past several decades. The gut microflora, which is composed of approximately 10^{14} bacteria, or approximately 10 times the number of body cells, is now considered as a functional human organ[1]. New studies such as the Human Microbiome Project are revealing how the gut microflora manipulates and complements physiology in ways that are important for the host[2-4]. The normal gut microflora provides protection against infection, educates the immune system, ensures tolerance to foods, and contributes to nutrient digestion and energy harvest. In addition to these important functions, normal microbial colonization of the intestine is important in the induction of the host innate response and plays a vital role in the regulation and maintenance of intestinal barrier function. Disruption in the establishment of a stable normal gut microflora may be associated with or even contribute to the pathogenesis of diseases including inflammatory bowel disease (IBD), nosocomial infection, and neonatal necrotizing enterocolitis (NEC)[5]. This review summarizes recent advances in understanding the complex ecosystem of the gut microflora and the roles of gut microflora in regulating intestinal barrier function as well as a few common pediatric diseases which may be related to an altered gut microbiota.

ESTABLISHMENT OF NORMAL INTESTINAL MICROBIAL COLONIZATION IS ESSENTIAL FOR THE POSTNATAL INTESTINAL BARRIER MATURATION

Before birth, the intestine is sterile. The intestinal tract becomes colonized soon after birth with a variety of ingested environmental and maternal microorganisms. This process is influenced by many factors including mode of delivery, diet, environment, and the use of antibiotics[6,7]. For example, a breast-fed full-term infant normally has an intestinal microbiota in which bifidobacteria predominate over potentially harmful bacteria, whereas in formula-fed infants, enterococci, bacteroides and clostridia predominate[8]. In premature infants, the immature intestinal mucosa is even more sensitive to gut colonizing bacteria. Host defenses can be improved by feeding the breast milk which helps the immature intestinal mucosal immune system to develop and respond appropriately to the highly variable bacterial colonization[8].

The dense communities of bacteria in the intestine are separated from body tissues by a monolayer of intestinal epithelial cells. Therefore, normal intestinal function depends on the establishment and maintenance of the mucosal epithelial barrier which prevents the invasion of host tissues by resident bacteria. The assembly of the multiple components of the intestinal barrier is initiated during fetal development and continues during early postnatal life; thus the intestinal barrier has not completely developed soon after birth, particularly in preterm infants[9]. Several studies indicate that the normal bacterial colonization process may be important for postnatal intestinal barrier development. By using a newborn piglet model, Kansagra *et al*[10] demonstrated that intestinal barrier function was significantly less developed in full term newborn piglets receiving total parental nutrition compared to those receiving enteral nutrition. Even in the mature intestine, lack of enteral nutrition is associated with loss of intestinal epithelial barrier function which can lead to bacterial translocation and subsequent sepsis[11]. In a rodent model, replacing enteral nutrition with parenteral nutrition can lead to bacterial translocation from the gut[11,12]. Total parenteral nutrition significantly increases intestinal permeability, which can be ameliorated by enteral feeding and especially with a fiber enhanced diet[13]. All of these results suggest that enteral feeding and colonization with the normal commensal flora are necessary for the maintenance of intestinal barrier function.

Recent studies have demonstrated that certain commensal bacteria increase intestinal epithelial cell survival by inhibiting the activation of the epithelial cell pro-apoptotic pathway associated with pathogenic bacteria[14]. The intestinal commensal flora is also involved in maintenance of barrier function by inducing increased epithelial cell proliferation and enhancing intestinal epithelial integrity, through translocation of the tight junction proteins and up-regulation of genes involved in desmosome maintenance[15,16]. Fermentation products of commensal bacterial have been shown to enhance the intestinal barrier function by facilitating the assembly of tight junctions through the activation of AMP-activated protein kinase[17]. On the other hand, the deletion of all detectable commensal flora in the gut by a four-week course of orally administration of vancomycin, neomycin, metronidazole, and ampicillin leads to a more severe intestinal mucosal injury in a dextran sulfate sodium induced mouse colitis model[18]. The importance of normal bacterial colonization in the development and maintenance of the intestinal barrier is further supported by the observations that the gastrointestinal tract gene expression profile and intestinal barrier development are altered by early treatment with broad-spectrum antibiotics or withholding enteral feeding[19].

INTESTINAL MICROFLORA STIMULATES THE MATURATION OF THE MUCOSAL INNATE IMMUNE SYSTEM

The colonization with normal gut microflora protects against infection from pathogenic bacteria. This long-known but poorly understood protection provided by commensal flora against pathogens is commonly referred to as colonization resistance[20-24]. Several mechanisms have been proposed to explain colonization resistance including a direct competition for nutrients, prevention of access to adherence sites, limitation of pathogen proliferation

through production of inhibitory substances or conditions, and stimulation of host natural immune defenses[21,25].

Before birth, the fetus is protected from microbial exposure, and postnatal stimulation by initial intestinal microbial colonization is important for the proper maturation of the innate immune system. Exposure to immunostimulatory microbial constituents may trigger activation of the infant's mucosal innate immune system as shown recently in the gnotobiotic (germ-free) mouse model[26-28]. For example, when compared to mice with normal intestinal microbial colonization, gnotobiotic mice need 30% more calories to maintain their body weight, exhibit an enhanced susceptibility to infection with enteropathogenic bacteria, and have an immature immune system[28].

Remarkably, intestinal mucosal homeostasis is maintained despite the large surface area continuously being exposed to different bacterial species[29,30]. To face the menace represented by intimate contact with a huge concentration of bacteria, the intestinal epithelium has evolved into a highly regulated barrier that can recruit immune cells of hematopoietic origin and produce mucus and a diverse arsenal of antimicrobial proteins that directly kill or inhibit the growth of microorganisms[30,31]. The intestinal epithelium comprises several cell lineages. Enterocytes constitute the most abundant epithelial cell type, and secrete several antimicrobial proteins. Paneth cells are unique to the small intestine and secrete abundant quantities of antimicrobial proteins, such as α-defensins. Finally, goblet cells secrete mucin glycoproteins and trefoil factors that assemble to form a thick mucus layer overlying the epithelium[32].

The development of the gut immune system is initiated before birth by a genetic program that drives the formation of Peyer's patches and mesenteric lymph nodes, but its postnatal maturation depends on the establishment of a balanced indigenous microbiota[28]. Intestinal commensal microorganisms provide signals that foster normal immune system development and influence the ensuing immune responses[33]. Signals delivered by these commensal microorganisms drive the development of isolated lymphoid follicles, stimulate maturation of Peyer's patches and initiate the migration of IgA producing plasma cells and mature T lymphocytes into the mucosa[33-35]. Among the first colonizing organisms evident in the intestine of newborn infants are strains of *Escherichia coli* (*E. coli*) derived from the maternal gastrointestinal tract[36-38]. It has been demonstrated in animal models that commensal *E. coli* strains can inhibit invasive *E. coli* O157:H7 growth in the intestine[39]. These studies prove that the gut commensal microflora clearly have important effects on the development of normal immunity.

INTERACTION BETWEEN INTESTINAL MICROFLORA AND IMMUNE HOMEOSTASIS

The intestinal microflora regulates immune homeostasis through many different mechanisms. Gut epithelia actively sense enteric bacteria and play an essential role in maintaining host-microbial homeostasis at the mucosal interface. Many innate immune responses are regulated by Toll-like receptors (TLRs), a conserved family of innate immune receptors that recognize microbial-derived molecules, including lipopolysaccharide, lipoprotein, RNA, and methylated DNA. Experiments in mice demonstrate that the beneficial effects of commensal bacteria are mediated *via* TLRs[40]. Intestinal epithelial cells and mucosal immune cells express pattern-recognition-receptors such as TLRs, enabling them to respond to distinctive microbial-associated molecular patterns[41]. TLRs are therefore critical for the specific detection of microbe-associated patterns, allowing differentially regulated responses to commensal versus pathogenic flora. The recognition of commensal bacterial-derived molecules by TLRs represents a critical component of the symbiosis between the host and indigenous microflora and is important for protection against gut injury and associated mortality[42]. Deregulated interactions between commensal bacteria and TLRs have been reported to promote chronic inflammation and tissue damage, such as that seen in IBD[43]. TLRs may also direct expression of the MyD88-dependent antimicrobial response. Paneth cell-intrinsic MyD88 activation limits translocation and dissemination of microbes across the mucosal barrier, while having little impact on luminal microbial numbers[44]. These results highlight the essential role of TLR-dependent pathways in compartmentalization of enteric commensal bacteria[45].

TLR-dependent signals mediate important regenerative signals to maintain intestinal mucosal integrity[46]. TLR-mediated protection may works through both constitutive and damage-induced production of protective factors by TLRs expressed on colonic epithelium[40]. Stimulation of TLRs results in activation of multiple signaling cascades that control expression of a wide range of innate immune response genes[47,48]. Recent evidence indicates a role for CpG DNA, the bacterial agonist of TLR9, in mediating some of the beneficial effects of probiotics in the intestine, and more generally, to modulate the immuno-physiological status of the gut[49]. Furthermore, TLR2 has been demonstrated to be responsible for the effects of bacteroides fragilis. This bacterium possesses an unusual capsular polysaccharide A that exerts potent immunoregulatory effects and can dampen intestinal inflammation in several models of colitis[50,51]. Systemic administration of flagellin, a bacterial protein that stimulates TLR5 protects mice from infection with vancomycin-resistant enterococcus (VRE)[52].

Another immune regulatory effect of commensal bacteria involves the inhibition of the nuclear factor-kappaB (NF-κB) pathway through the stabilization of IγBα[53]. IγBα is a central inhibitor of the NF-κB pathway, which acts by retaining the inactive NF-κB dimers in the cytosol. Most pro-inflammatory signals trigger the NF-κB pathway by inducing the phosphorylation of IγBα, which targets the molecule for degradation by the

ubiquitin/proteasome system[54]. Incubation of epithelial cells with nonpathogenic Salmonella has been shown to induce the accumulation of IγBα through the down-regulation of the protein's ubiquitination[53]. Similarly, it has been recently reported that the probiotic bacteria Lactobacillus casei also inhibits the NF-κB pathway by targeting the degradation of IγBα[55].

The specialized intestinal epithelial cells are capable of secreting proteins into the lumen of the intestinal tract which enhance epithelial barrier function and/or interact with the bacterial flora resident in the intestine. Goblet cells are highly polarized exocrine epithelia that secrete proteins apically into the lumen of the small and large intestine through the release of secretory granules. One particular class of glycoproteins produced by goblet cells, known as mucins, forms a viscoelastic protective gel that covers the intestinal epithelium[56]. Another class of secretory peptides, now designated as trefoil factors, is also normally produced by goblet cells and is important for the maintenance and repair of the gut mucosal barrier[57]. Mucins interact with trefoil factors and perform a defensive role during establishment of the intestinal barrier. Mucin oligosaccharides influence the bacterial milieu of the intestinal tract by enhancing the ability of certain bacteria to colonize the intestinal tract while inhibiting the adherence of others[58]. Mucins are also a direct source of carbohydrates and peptides that can promote the growth of bacteria[59]. To further enhance the symbiotic relationship between gut bacteria on the host, bacteria can alter mucus synthesis, secretion, and chemical composition[60]. Changes in mucin profile in response to bacterial colonization suggest a potential role as a protective mechanism against pathogenic invasion of the intestinal mucosa during early development[61].

The mucosal barrier is also reinforced by secretory immunoglobulin A (sIgA) and sIgM generated through external translocation of locally produced dimeric IgA and pentameric IgM. The dimeric IgA and pentameric IgM, containing a small polypeptide called joining chain to form sIgA and sIgM, can be actively exported by secretory epithelia. This external transport is mediated by the polymeric Ig receptor (pIgR), also known as membrane secretory component (SC)[62]. Notably, pIgR/SC knock-out mice that lack secretory IgA and IgM antibodies exhibit reduced epithelial barrier function with aberrant mucosal leakiness[63]. sIgA and sIgM form the first line of defense against pathogens as well as other potentially dangerous agents. Therefore, secretory immunity is of great importance for the intestinal epithelial barrier. In newborn infants, only a few IgA-secreting cells circulate in the blood. However this number is remarkably increased after 1 mo of age mainly due to the progressive microbial stimulation of gut-associated lymphoid tissues[64]. A much faster establishment of secretory immunity is often seen in infants from developing countries where there is exposure to a heavy microbial load, and an associated lower incidence of atopy[65]. Altogether, the secretory immune system is critical for the mucosal barrier function and the intestinal epithelial barrier maturation depends on exposure to microbial factors from the environment and the indigenous microbiota.

DISEASES ASSOCIATED WITH AN ALTERED INTESTINAL MICROFLORA AND ABNORMAL BARRIER FUNCTION

Alterations of the microbial gut flora and changes in intestinal barrier function are associated with certain diseases of the gastrointestinal tract. There is growing evidence that changes of the intestinal flora composition may play a pathogenic role in IBD, nosocomial infection, and NEC[66-68]. It has been proposed that a genetic predisposition causes IBD patients to have a deregulated immune response against harmless antigens derived from intestinal commensal bacteria and changes of the intestinal flora composition have been described in patients with IBD[66]. Several studies found an enhanced number of Proteobacteria and Actinobacteria but decreased numbers of Firmicutes (particularly the species Faecalibacterium prausnitzii) in stool samples of IBD patients as compared to healthy controls[69,70]. In biopsies of pediatric IBD patients, higher numbers of mucosa-associated aerobic and facultative-anaerobic bacteria were found, whereas bacterial species of the normal anaerobic intestinal flora such as Bifidobacteria were reduced[71,72].

It is well recognized that nosocomial infection is frequently a consequence of gut derived organisms. The infections with highly antibiotic-resistant bacteria are usually acquired during hospitalizations. Destruction of the normal flora by antibiotic administration disinhibits antibiotic-resistant members of these bacterial families, leading to their expansion to very high densities[73]. Reintroduction of a diverse intestinal microbiota to densely VRE-colonized mice eliminates VRE from the intestinal tract[74]. Characterization of the fecal microbiota of patients undergoing allogeneic hematopoietic stem cell transplantation demonstrated that intestinal colonization with Barnesiella confers resistance to intestinal domination and bloodstream infections with VRE[74]. Furthermore, there is an increased incidence of septic complications in patients receiving parenteral as opposed to enteral nutrition and this, in some cases, is due to alterations in intestinal barrier function predisposing to bacterial translocation[75].

In premature infants, colonization with abnormal gut flora increases the risk of hospital acquired infection or late-onset sepsis (LOS)[76]. Prolonged use of broad spectrum antibiotics, reduced bowel motility, immature epithelial host defenses, lack of enteral feeding, and parenteral nutrition are common risk factors for an altered microbial gut flora and abnormal mucosal barrier function. The possible subsequent bacterial translocation from the gastrointestinal tract may be an important pathway initiating LOS in premature infants[76]. Mai *et al*[77] analyzed stool samples that had been prospectively

collected from ten preterm infants with LOS and from 18 matched controls. A 16S rRNA based approach was utilized to compare microbiota diversity and identify specific bacterial signatures that differed in their prevalence between cases and controls. They found that the types and distributions of bacteria that initially colonize the intestine in premature infants differ in those with LOS compared to uninfected control babies. Therefore, it was proposed that a distortion in normal microbiota composition, and not an enrichment of potential pathogens, is associated with LOS in preterm infants. This may suggests that administration of probiotics may protect high-risk neonates and infants from developing sepsis. However, currently there is no clinical evidence regarding the usefulness of probiotics or prebiotics for the prevention of nosocomial sepsis in preterm infants[78].

Failure of the postnatal developmental of the intestinal barrier in the immature intestine plays an important role in the pathogenesis of NEC, a devastating disease seen mainly in preterm infants[68,79,80]. A major component of the pathophysiology of NEC is the nature of the interaction of bacteria with the premature gut. The pattern of bacterial colonization in the intestine of the premature neonate is quite different from that of the healthy full-term infant. Infants requiring intensive care acquire intestinal organisms slowly, and the establishment of bifidobacteria flora is retarded. A delayed bacterial colonization of the gut with a limited number of bacterial species tends to be virulent. Indeed, several clinical observational studies have shown that the duration of antibiotic exposure including prenatal exposure to antibiotics is associated with an increased risk of NEC in preterm infants[81-83]. Therefore, an aberrant colonization of the bowel of the premature infant has been proposed to contribute to the development of NEC[84]. By using non-culture-based microbial analyses of feces, Wang *et al*[85] studied fecal samples of ten preterm infants with NEC and ten matched controls and found that patients with NEC had less bacterial diversity and an increased abundance of γ-proteobacteria in the stools. Similar findings were presented in another study by Mai *et al*[86], in which one of the bacterial signatures detected more frequently in NEC patients matched closest to γ-proteobacteria. These observations suggest that abnormal patterns of microbiota contribute to the cause of NEC. However, a study by Mshilddazde *et al*[87] using the same technology demonstrated that the overall microbial profiles in patients with NEC were not different from those of control infants. Thus to date, molecular methods have not clarified the bacterial pathogenesis of NEC.

None of the clinical studies to date has been able to fulfill Koch's postulate linking NEC to a particular pathogen. Nevertheless, we proposed a hypothesis that excessive production/accumulation of short chain fatty acids (SCFAs) due to bacterial fermentation of undigested formula or abnormal bacterial colonization contributes to the pathogenesis of NEC[88,89]. There is substantial indirect clinical evidence to support the theory that bacterial fermentation is involved in the development of NEC in premature infants[90]. Further, in two separate studies, increased breath hydrogen excretion (an indicator of bacterial fermentation and an indirect measurement of SCFAs production) was found in NEC patients even prior to the onset of clinical symptoms[91,92]. The well-known finding of pneumatosis intestinalis (gas in the bowel wall) in NEC patients is also thought to be secondary to hydrogen gas produced by bacterial fermentation[93]. Recent reports of several cases of premature infants who developed NEC after they were fed SimplyThick®, a xanthan gum-based thickener used in the management of dysphagia, is another example[94,95]. Increased production of hydrogen and SCFAs as the consequence of accumulation of luminal carbohydrates and fecal bacteria fermentation of xanthan gum were proposed as the main mechanisms for NEC[94]. Both probiotics and prebiotics have been proposed to promote a healthy gut microbiota in human, and oral probiotics have been proven to be effective in reducing the incidence of NEC in premature infants in several clinical trials[96-98]. On the other hand, there is no evidence showing that prebiotics can effectively reduce the incidence of infection or NEC in premature infants. Therefore, there is insufficient evidence to recommend the use of oligosaccharides as prebiotics in formula for premature infants since these may prove to be unsafe.

In summary, this review summarizes recent advances in understanding the complex ecosystem of intestinal microbiota and its role in regulating intestinal barrier function and a few common pediatric diseases. There is increasingly recognition that the stimulation of initial intestinal microbial colonization is important for proper maturation of the innate immune system and continued regulation and maintenance of intestinal barrier function. Disruption of the establishment of a stable normal gut microflora may contribute to the pathogenesis of diseases including IBD, nosocomial infection, and NEC in premature infants.

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Imaging evaluation of hemoptysis in children

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Abstract

Hemoptysis is an uncommon but distressing symptom in children. It poses a diagnostic challenge as it is difficult to elicit a clear history and perform thorough physical examination in a child. The cause of hemoptysis in children can vary with the child's age. It can range from infection, milk protein allergy and congenital heart disease in early childhood, to vasculitis, bronchial tumor and bronchiectasis in older children. Acute lower respiratory tract infections are the most common cause of pediatric hemoptysis. The objective of imaging is to identify the source of bleeding, underlying primary cause, and serve as a roadmap for invasive procedures. Hemoptysis originates primarily from the bronchial arteries. The imaging modalities available for the diagnostic evaluation of hemoptysis include chest radiography, multi-detector computed tomography (MDCT), magnetic resonance imaging (MRI) and catheter angiography. Chest radiography is the initial screening tool. It can help in lateralizing the bleeding with high degree of accuracy and can detect several parenchymal and pleural abnormalities. However, it may be normal in up to 30% cases. MDCT is a rapid, non-invasive multiplanar imaging modality. It aids in evaluation of hemoptysis by depiction of underlying disease, assessment of consequences of hemorrhage and provides panoramic view of the thoracic vasculature. The various structures which need to be assessed carefully include the pulmonary parenchyma, tracheobronchial tree, pulmonary arteries, bronchial arteries and non-bronchial systemic arteries. Since the use of MDCT entails radiation exposure, optimal low dose protocols should be used so as to keep radiation dose as low as reasonably achievable. MRI and catheter angiography have limited application.

Key words: Hemoptysis; Lower respiratory tract infection; Bronchiectasis; Cystic fibrosis; Foreign body

Core tip: Hemoptysis is a cause of immense concern to the child, the family and the pediatrician. Thorough history and physical examination is necessary to ascertain its presence, which is particularly challenging in the pediatric population. Imaging has an important role in identifying the source of bleeding and its underlying cause. Acute lower respiratory tract infections are the most common cause of pediatric hemoptysis. The imaging modalities include chest radiography, multi-detector computed tomography (MDCT), magnetic resonance imaging (MRI) and catheter angiography. MDCT is a rapid multiplanar imaging modality which should be used judiciously to keep radiation dose to a minimum. MRI and catheter angiography have selected application.

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INTRODUCTION

Hemoptysis is defined as expectoration of blood or blood tinged-sputum due to bleeding from the respiratory tract[1]. Massive hemoptysis is termed as blood loss > 8

Table 1 Causes of hemoptysis in children

Acute lower respiratory tract infections
Bacterial
Viral
Fungal
Parasitic
Bronchiectasis
Aspiration
Cystic fibrosis
Ciliary dyskinesia
Post-infectious
Congenital heart diseases
Eisenmenger syndrome
Aplasia/hypoplasia of pulmonary artery or veins
Primary pulmonary hypertension
Pulmonary artery narrowing
Infectious
Inflammatory
Pulmonary thromboembolism
Pulmonary arteriovenous malformation
Alveolar hemorrhage syndrome
Idiopathic
Associated with rheumatologic disease
Pulmonary-renal syndrome
Neoplasms
Bronchial carcinoid
Bronchial adenoma
Metastatic
Foreign body
Trauma
Cryptogenic

mL/kg in 24 h[2]. The life-threatening element in massive hemoptysis is asphyxiation due to flooding of the airways by blood. Hence, securing the airway needs immediate attention. Hemoptysis in lesser amounts poses a diagnostic challenge in pediatric patients as it may initially remain unnoticed because children tend to swallow their sputum and are unable to provide a clear history or undergo thorough physical examination. It is a cause of immense concern to the child, the parents and the pediatrician. After confirming the presence of hemoptysis, the next step is to establish the cause, so that an appropriate treatment regimen can be adopted. The spectrum of causes of hemoptysis in children is considerably different from that of the adults. Imaging has a pivotal role in evaluation of hemoptysis. There are various modalities which can be resorted to, namely, conventional radiography, multidetector computed tomography (MDCT), magnetic resonance imaging (MRI) and in certain cases, catheter angiography which can also fulfill a therapeutic purpose. The advent of MDCT has paved the way for non-invasive multi-dimensional visualization of the thoracic vasculature, tracheobronchial tree and pulmonary parenchyma. This is of tremendous value as it can obviate the need for invasive bronchoscopic procedure with its attendant complications. The following sections illustrate the etiology, pathogenesis and role of imaging in hemoptysis.

ETIOPATHOGENESIS

There are several causes of hemoptysis in children (Table 1). The common causes are acute lower respiratory tract infections, bronchiectasis (due to cystic fibrosis, aspiration, ciliary dyskinesias, post infectious), congenital heart disease and foreign body aspiration. Of these, acute lower respiratory tract infections may constitute upto 40% of cases[3]. The etiology also varies with the child's age. Sim *et al*[4] observed that infection, Heiner syndrome (milk protein allergy) and congenital heart disease were the major causes in early childhood; while during late childhood, vasculitis, bronchial tumor and bronchiectasis were far more prevalent.

The lungs receive dual blood supply; one from the high pressure bronchial arteries, the other from the pulmonary arteries with relatively lower pressure. The pulmonary arteries account for 99% of the arterial blood supply to the lungs and take part in gas exchange while the bronchial arteries provide nourishment to the supporting structures of the airways and form the vasa vasora of the pulmonary arteries. The bronchial vasculature is in close proximity to the pulmonary arteries at the level of the vasa vasorum where the two systems are connected by thin-walled anastomoses between the systemic and pulmonary capillaries[5,6]. Pulmonary vascular obstructive disorders (congenital heart disease, vasculitis, embolism) open up these anastomoses in regions of the lung that are deprived of their pulmonary arterial blood flow. This subjects these fragile vessels to increased systemic arterial pressure and can cause hemoptysis by rupturing into the alveoli or bronchial airways.

In the setting of tracheobronchial infection, there is inflammation of the airways. As a result, they become congested and friable, which increases their susceptibility to bleed. Chronic inflammation (as in bronchiectasis) can lead to increase in systemic arterial flow due to release of angiogenic growth factors which lead to neovascularization and formation of "leaky" vessels prone to rupture. Approximately 5% of patients with cystic fibrosis can present with massive hemoptysis[7]. This is due to hypertrophy of bronchial arteries along with the presence of multiple bronchopulmonary anastomoses. Foreign body aspiration causes hemoptysis due to mechanical trauma or due to associated intense inflammation incited by organic matter. Pulmonary hemosiderosis is an uncommon but significant cause of hemoptysis in children. It is mostly idiopathic; however, it may be associated with an allergy to cow's milk (Heiner syndrome)[8]. Although rare in children, endobronchial or pulmonary parenchymal tumors (carcinoid, bronchial adenoma) may cause significant hemorrhage.

Imaging evaluation of hemoptysis

The aim of initial diagnostic evaluation is to identify the immediate source of bleeding along with determination of the primary cause of hemoptysis. Traditionally, the diagnostic algorithms in acute setting have been based on various combinations of conventional radiography, chest computed tomography (CT) and thoracic aortography. MDCT has now made comprehensive visualization of

thoracic anatomy possible. It provides high-resolution images of the thoracic and upper abdominal vasculature which aids in diagnosis and also provides a roadmap prior to any intervention. CT findings can forewarn the endoscopist about the presence of peribronchial or endoluminal aneurysms. MRI does not have a role in the acute setting. However, it may serve as a problem-solving tool in certain situations.

Conventional radiography

Chest radiography serves as a valuable screening modality. It can help in lateralizing the bleeding with high degree of accuracy and can detect several parenchymal and pleural abnormalities. The commonly used views include frontal and, in some cases, lateral. The utility of lateral radiographs is in case of presence of a radio-opaque foreign body on frontal view, when it can determine if it is in the trachea or esophagus. Lynch *et al*[9] observed that addition of a lateral radiograph in children with pneumonia did not improve diagnostic accuracy. Common findings include presence of focal infiltrates which may suggest infection. Unilateral air-trapping with hyperinflation can give a clue towards presence of an unsuspected tracheobronchial foreign body. A radio-opaque foreign body may be seen. Ancillary findings include "tram-track" appearance of bronchiectasis; pulmonary nodules, lymphadenopathy, pleural effusion; cardiomegaly; and vascular redistribution due to pulmonary venous obstruction.Approximately one-third of chest radiographs may be normal in children with hemoptysis. A tracheobronchial source of bleed may eventually be identified in about half of these cases[10]. Therefore, additional follow-up testing is recommended in patients with hemoptysis in whom the underlying cause is not detected by initial radiography.

MDCT

The role of MDCT in evaluation of hemoptysis includes: (1) depiction of underlying disease; (2) assessment of consequences of hemorrhage which may be a cause of clinical concern or may conceal the underlying abnormalities; and (3) panoramic visualization of the thoracic vasculature by various reconstruction techniques.

Technique

CT technique involves acquisition of multiple sections from the base of the neck to the level of the renal arteries (L2 level). Optimal enhancement of the pulmonary and systemic arteries is achieved by administration of 2 mL/kg body weight of iodinated non-ionic contrast media containing 300 mg I/mL at a rate of 4 mL/s *via* a wide gauge cannula. The scan should be started during the phase of peak systemic arterial enhancement (scanning delay of 18 s or a threshold of 100 HU in the descending aorta with bolus tracking). Images should be acquired with thin collimation and with the table movement adjusted to allow wide volume coverage during a single breath-hold. Radiation exposure is a significant concern in the pediatric population. Hence, the exposure parameters and kilovoltage need to be adjusted according to the patient's weight so as to minimize radiation dose with optimal image quality.

Data processing and interpretation

Since a large volume of data is acquired, the images are best interpreted at the scanner console or remote workstation. The soft-tissue structures and lung parenchyma can be assessed adequately in axial sections of 5 mm thickness with mediastinal and lung window settings, respectively (Figure 1A and B). High resolution CT images allow detailed evaluation of the pulmonary parenchyma. The tracheobronchial airway can be evaluated on thinner sections and reformatted images.

Two-dimensional maximum intensity projection images (Figure 1C) in the coronal/oblique and sagittal planes can demonstrate the tortuous course of the bronchial arteries from the descending thoracic aorta to the lungs. Intercostal and internal mammary arteries are best visualized in the coronal planes while the inferior phrenic arteries and celiac axis branches are demonstrated well in axial images. Three-dimensional volumetric and shaded-surface-display images not only depict the abnormal vessel, but also illustrate its relationship with the surrounding structures, thus providing a preview of the internal anatomy.

Minimum Intensity Projection can be used to generate images of the central airways (Figure 1D) and demonstrate areas of air-trapping within the lungs. These can provide valuable perspective in defining a lesion prior to any intervention. Therefore, a host of reconstructed images need to be analyzed for a thorough CT assessment of hemoptysis.

STEPWISE STRUCTURAL ASSESSMENT

Lung parenchyma

The pulmonary parenchyma should be evaluated for presence of bronchiectasis, consolidation and ground-glass opacity. The site of hemorrhage can be localized on the basis of presence of fluid density material in the segmental and lobar bronchi and ground-glass opacity with hazy consolidation which represents alveolar hemorrhage. Acute hemorrhage can mask the underlying pathology. Blood clots can also simulate more ominous entities like masses.

Tracheobronchial tree

This should be evaluated for presence of any stenosis which may be due to intraluminal (foreign body, neoplasm) or extraluminal (lymphadenopathy, fibrosing mediastinitis) causes. Multiplanar reformatted (MPR) images are accurate in detection of lesions, depiction of degree of narrowing, distal visualization and calculation of distance of the lesion from the carina in selected locations[11].

Pulmonary arteries

The pulmonary arteries should be analyzed for any narrowing due to extrinsic or intrinsic causes. The presence

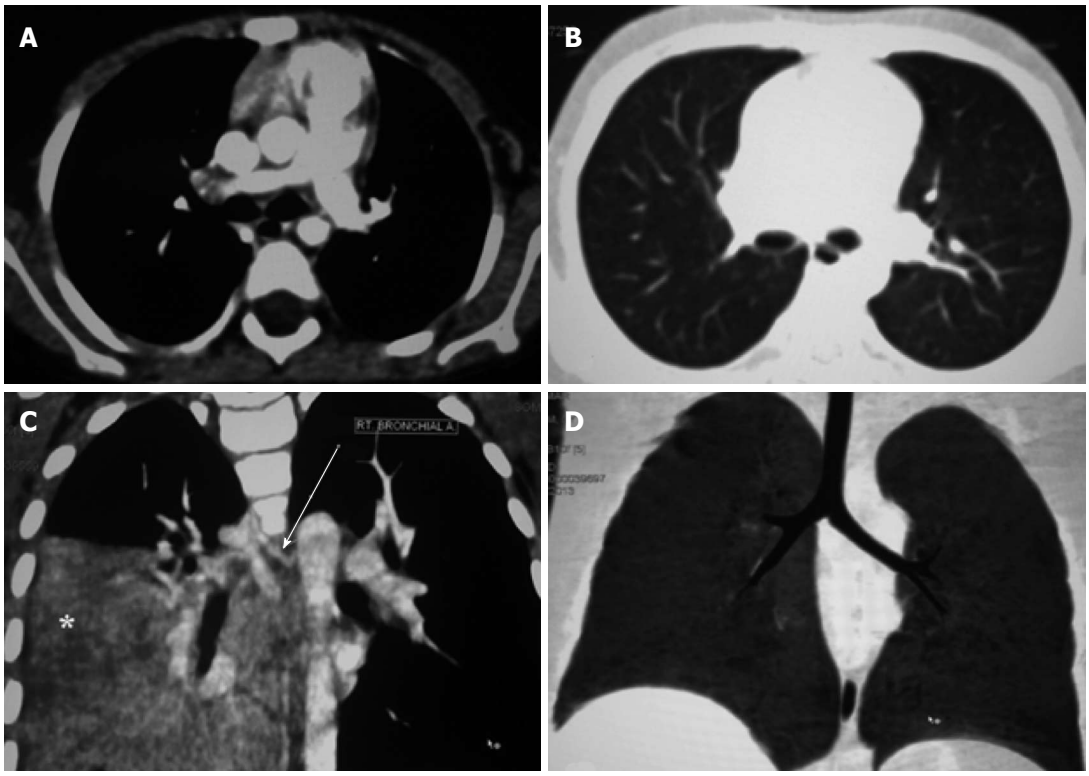

Figure 1 Multi-detector computed tomography image interpretation. Axial computed tomography image showing mediastinal window (A) and lung window (B). Coronal maximum intensity projection image (C) demonstrating the origin and proximal portion of the right bronchial artery (arrow). There is consolidation in the right lower lobe (asterisk). Coronal Minimum Intensity Projection image (D) delineating the central airways.

of accompanying subpleural areas of enhancement can represent lung infarcts. The pulmonary arteries can also show dilatation (Rasmussen aneurysm) and pulmonary arteriovenous malformations.

Bronchial arteries

Hemoptysis originates from bronchial arteries in 95% of cases[12]. A bronchial artery diameter of more than 2 mm is considered abnormal[13]. In 70% of individuals, bronchial arteries arise from the descending thoracic aorta between T5 and T6 levels. There are usually one or two bronchial arteries supplying each lung, arising independently or from a common trunk. They are visualized as a cluster of enhancing nodules in the posterior mediastinum just below the level of the aortic arch on axial images. Active bleeding can rarely be detected on CT. Anomalous bronchial arteries are defined as arteries which originate outside the T5-T6 level. Their most common site of origin is the concavity of the aortic arch[14].

Non-bronchial systemic arteries

Non-bronchial systemic arteries can arise from the branches of brachiocephalic arteries, subclavian arteries, axillary, internal mammary and infradiaphragmatic branches from the inferior phrenic artery and celiac axis[15,16]. On CT, these are seen as dilated tortuous arteries that are not parallel to the bronchi. The presence of pleural thickening greater than 3 mm with enhancing arteries within the extrapleural fat is a pointer of presence of these vessels[17].

MRI

MRI does not have any utility in imaging evaluation of acute hemoptysis. Since it has superior soft-tissue resolution, it is excellent in the evaluation of the mediastinum and hilum in the non-emergent setting. It provides less information about the lung parenchyma. It may be used to demonstrate arteriovenous malformations and congenital anomalies of the pulmonary arteries and delineate the nature of mediastinal soft-tissue in fibrosing mediastinitis. With the introduction of hyperpolarized nuclei like ^{3}He and ^{129}Xe, the horizon of MRI is likely to expand from limited utility in evaluating the pulmonary parenchyma to evaluation of pulmonary structure, function and metabolism with a high sensitivity[18]. Ventilation and dynamic imaging in patients with asthma and cystic fibrosis have shown regional patterns of obstruction and ventilation defects in these individuals. Further knowledge can go a long way in the early diagnosis, monitoring disease progression and evaluation of response to treatment[19-21].

COMMON CAUSES OF PEDIATRIC HEMOPTYSIS

Acute lower respiratory tract infections

Tracheobronchitis, pneumonia and lung abscess can lead to hemoptysis. The infective process may be bacterial, viral, fungal or parasitic in origin. Although tuberculosis is a significant cause of adult hemoptysis, very few cases have been reported in the pediatric literature[22]. Chest radio-

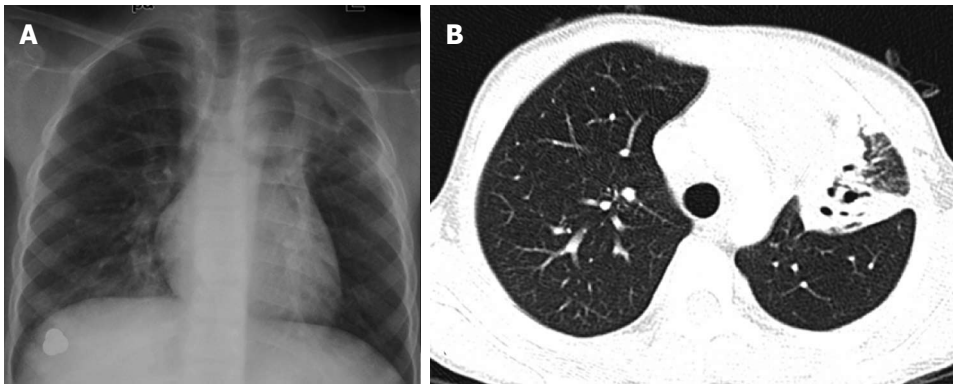

Figure 2 Chest radiograph (A) and axial computed tomography image (B) showing consolidation with cavitation in the left upper lobe.

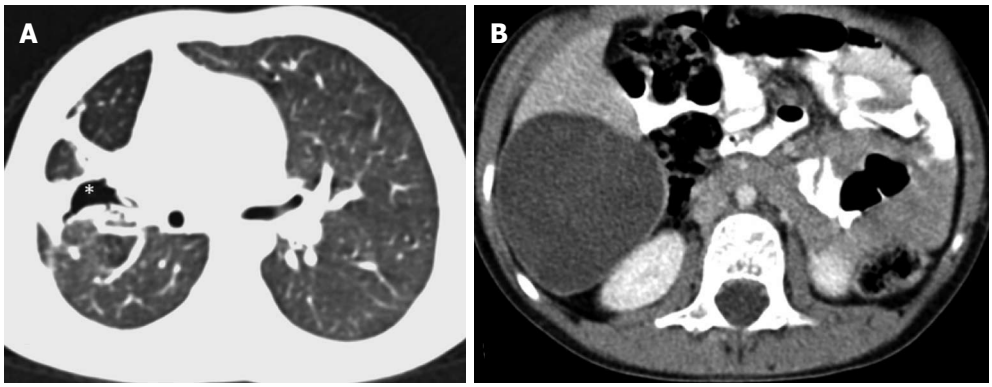

Figure 3 A seven-year-old girl with ruptured pulmonary hydatid cyst. Axial computed tomography image (A) showing the ruptured cyst with air (asterisk) in the right upper lobe along with surrounding consolidation. Axial section of the abdomen (B) shows an unruptured cyst in the segment VI of the liver.

graphs can show pulmonary infiltrates, hyperinflation and cavity with or without air-fluid level (Figure 2A). There may be associated pleural effusion and lymphadenopathy. CT findings can be in the form of consolidation, ground-glass opacity, interstitial thickening, air-trapping, cavity with shaggy walls and air-fluid level, pleural effusion and mediastinal or hilar lymphadenopathy (Figure 2B). CT can also demonstrate complications like empyema (thick enhancing visceral and parietal pleura, "split pleura" sign), bronchopleural fistula, *etc*.

Parasitic cysts (Echinococcus) can cause hemoptysis by rupturing into the airway. These may be seen as fluid density lesions with a smooth wall and air foci due to communication with the adjacent bronchus (Figure 3A). Detached membranes and daughter cysts can be visualized within the cyst. Concomitant cysts may be seen in other organs, most commonly in the liver (Figure 3B).

The most commonly implicated fungus is Aspergillus. It can have a varied spectrum of presentation, namely aspergilloma, allergic bronchopulmonary aspergillosis (ABPA), semi-invasive aspergillosis, airway or angioinvasive aspergillosis[23,24]. Aspergilloma is the saprophytic colonization of a pre-existing cavity by the fungus and is typically seen as an opacity within a cavity producing the "air-crescent" sign. It is mobile and can show postural change in position. ABPA is a manifestation of type I and Ⅲ hypersensitivity reaction to the organism and presents as central bronchiectasis with mucous plugged bronchi producing 'finger-in-glove' appearance with upper lobe preponderance on radiograph. The mucous plugs have a high-density on CT (Figure 4). There may be centrilobular nodules with "tree-in-bud"appearance. Some patients can also have associated allergic fungal sinonasal disease. Invasive aspergillosis is encountered in immunocompromised patients. Invasive airway disease presents as peribronchial areas of consolidation and multiple branching centrilobular nodules on CT[25]. Nodules with surrounding ground-glass opacity (halo sign) or pleural-based, wedge shaped areas of consolidation (Figure 5) are the hallmark of angioinvasive aspergillosis[26].

Bronchiectasis

Bronchiectasis can occur secondary to aspiration, infections, cystic fibrosis and ciliary dyskinesias. On chest radiographs, it manifests as "tram-track", parallel line opacities, ring opacities and tubular structures (Figure 6A). However, chest radiographs are insensitive for detecting mild to moderate disease. CT (Figure 6B) has a higher sensitivity and on CT imaging, bronchiectasis is characterized by the absence of normal bronchial tapering, the presence of visible bronchi in the peripheral 1 cm of the lung and a bronchoarterial ratio more than 1 (signet ring sign). The etiology can be narrowed by considering the anatomic location and distribution of pathology.

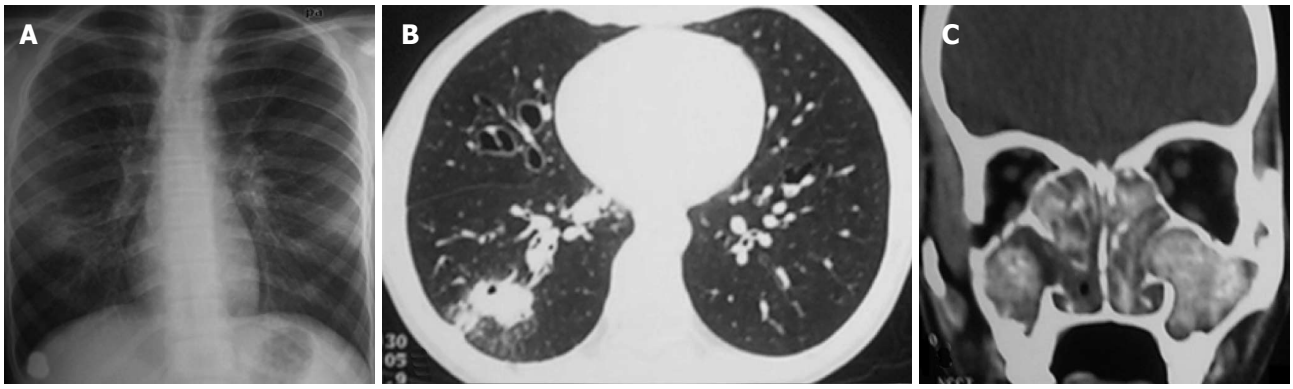

Figure 4 A 12-year-old girl with allergic bronchopulmonary aspergillosis. Frontal chest radiograph (A) and axial computed tomography (CT) image (B) showing tubular opacities with consolidation in the right lung suggestive of mucoceles along with cystic bronchiectasis in bilateral lungs. Coronal CT image (C) of the patient showing evidence of bilateral allergic fungal rhinosinusitis.

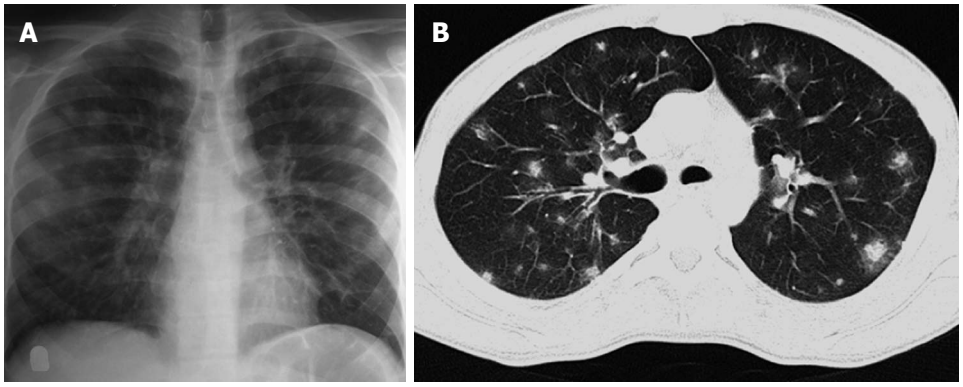

Figure 5 A 17-year-old boy with acute lymphocytic leukemia along with angioinvasive aspergillosis. Chest radiograph (A) showing multiple fluffy nodules in bilateral lung fields. High resolution CT image (B) of the same patient shows multiple nodules with surrounding ground glass opacity (halo sign).

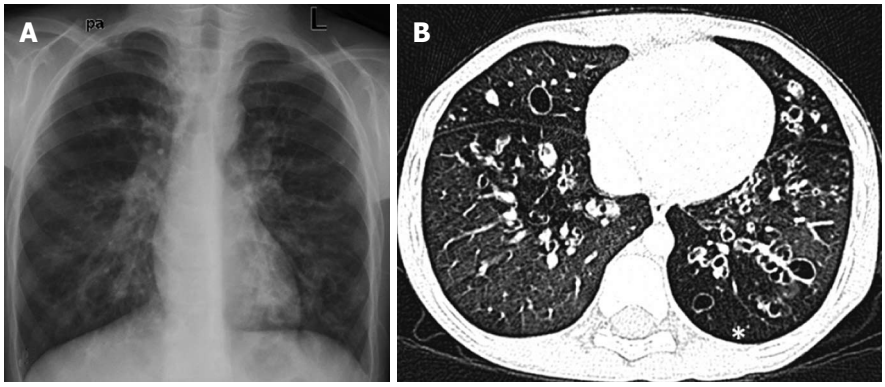

Figure 6 A 10-year-old boy with post-infectious bronchiectasis. Frontal chest radiograph (A) showing multiple cystic lucencies and tubular opacities in both lungs. Chest high resolution CT image (B) shows multiple areas of cystic bronchiectasis with associated air trapping (asterisk).

Aspiration tends to involve the lower lobes (right > left). Cystic fibrosis shows lung hyperinflation and interstitial infiltrates with upper lobe preponderance (Figure 7). Bronchiectasis due to ciliary dyskinesias has a lower lobe predisposition[27].

Congenital heart disease

Hemoptysis can occur in patients with congenital heart diseases associated with pulmonary artery or venous stenosis or atresia. This is attributed to hemorrhage from enlarged, tortuous aorto-pulmonary collateral arteries and thrombotic lesions in the small pulmonary arteries[28]. Chest radiography may show cardiomegaly with abnormality in cardiac silhouette and a small hilum. An abnormal vascular channel parallel to the right cardiac border (scimitar vein) can be seen in pulmonary venolobar hypoplasia[29]. There may be associated pulmonary volume loss. MDCT is the modality of choice to demonstrate the site and extent of pulmonary artery narrowing anddelineate anomalous pulmonary venous drainage (Figure 8). It exquisitely depicts the various aorto-pulmonary collaterals. Other associated cardiac anomalies can also be evaluated[30].

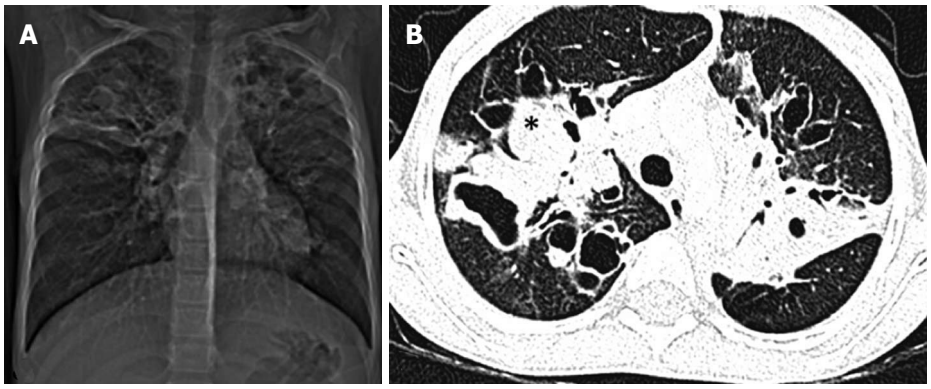

Figure 7 A 7-year-old boy with cystic fibrosis. Computed tomography (CT) scout image (A) and axial CT chest image (B) showing bilateral upper lobe bronchiectasis with bronchocele formation (asterisk) due to mucous plugging and sparing of lower zones.

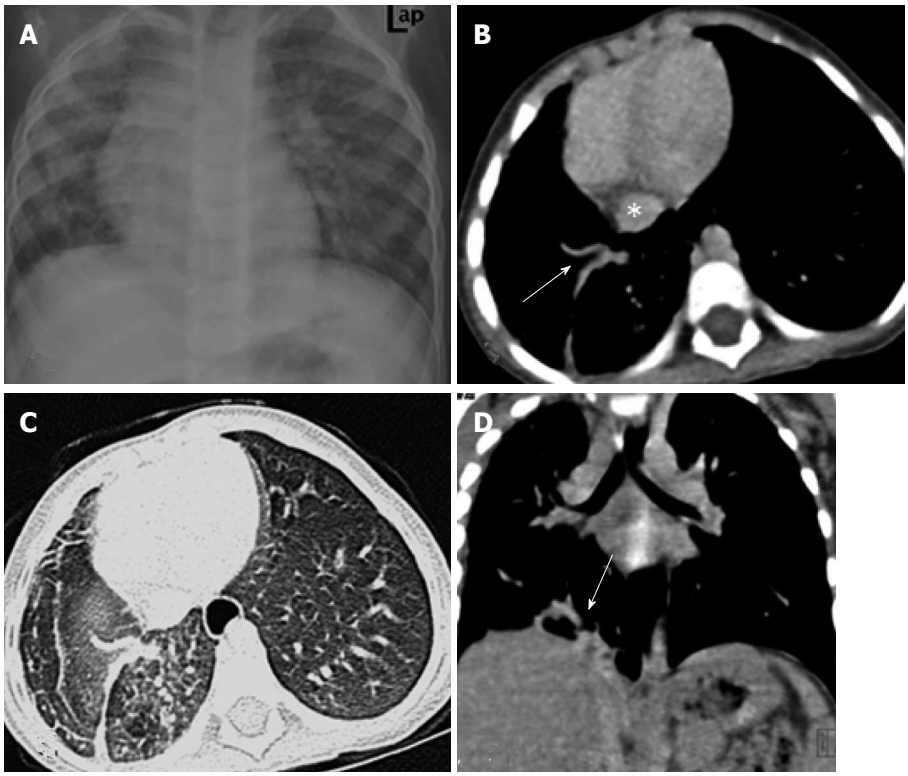

Figure 8 Pulmonary venolobar hypoplasia. Chest radiograph (A) shows volume loss of the right hemithorax with ipsilateral mediastinal shift. Contrast-enhanced computed tomography images (B-D) showing anomalous right inferior pulmonary vein (arrows) coursing inferiorly towards the inferior vena cava (asterisk).

Pulmonary artery narrowing

Chronic pulmonary artery narrowing can occur due to a variety of causes like infections, inflammation and thromboembolism[31]. Infections are the most common cause. Narrowing of the pulmonary artery can be caused in the setting of infection by mediastinal lymphadenopathy or fibrosis. Fibrosis may be focal or diffuse. CT finding of focal fibrosis is a calcified soft-tissue mass in the paratracheal and hilar location. It can occur secondary to tuberculosis in the developing countries and histoplasmosis in United States. The diffuse form manifests as an infiltrative, non-calcified soft-tissue mass extending into multiple mediastinal compartments. It can be associated with autoimmune disorders, drugs, or be idiopathic[32]. Pulmonary artery narrowing in these cases leads to pulmonary hypoperfusion and consequent bronchial artery hypertrophy while can lead to hemoptysis of varying severity. CT pulmonary angiography is the investigation of choice in this condition as it elucidates pulmonary artery narrowing and bronchial/systemic artery hypertrophy[33].

Pulmonary arteriovenous malformations

Pulmonary arteriovenous malformations (PAVM) are direct communication between the branches of the pulmonary artery and veins without capillary bed. There is a strong association between PAVM and hereditary hemorrhagic telangiectasia[34]. Chest radiography is an important tool for diagnosis and follow-up. Classic findings of PAVM are a round or oval well-defined mass, frequently lobulated, ranging in size from 1-5 cm. Two-thirds of these are located in the lower lobe. A connecting vessel may be seen radiating from the hilum. MDCT can identify the PAVM and connecting vessels more accurately (Figure 9). PAVMs have rapid blood flow and hence produce a low intensity signal on MRI. Catheter angiography remains the gold standard in diagnosis of PAVM as it defines the angio-architecture which is necessary before therapeutic embolization or surgical resection.

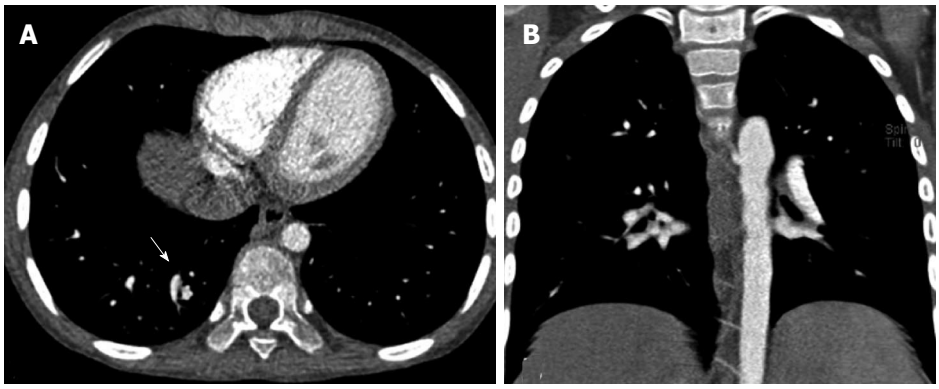

Figure 9 Pulmonary arteriovenous malformation. Axial (A) and coronal (B) computed tomography images showing abnormal communication between branches of the pulmonary artery and vein in right lower lobe (arrows).

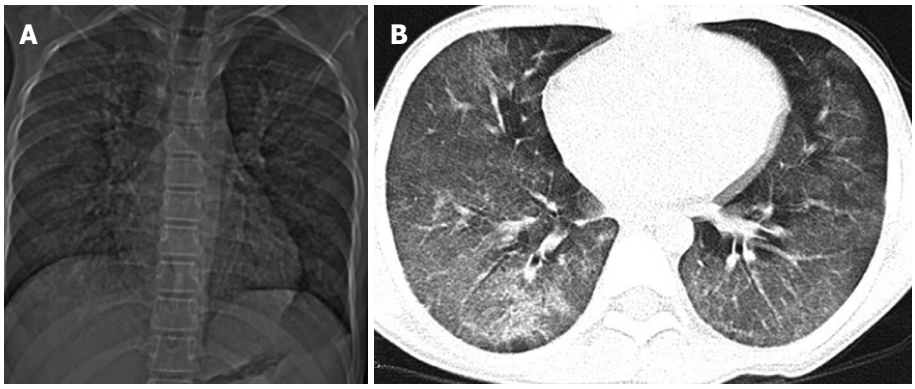

Figure 10 A 15-year-old boy with idiopathic pulmonary hemosiderosis. Scout computed tomography (CT) image (A) and axial CT image (B) showing diffuse ground glass opacity in bilateral lungs.

Idiopathic pulmonary hemosiderosis

Idiopathic pulmonary hemosiderosis (IPH) is a rare pulmonary disorder which manifests as a triad of hemoptysis, anemia and diffuse parenchymal infiltrates on chest radiographs[35]. Diagnosis is confirmed by detection of hemosiderin-laden macrophages in broncho-alveolar lavage fluid, sputum or gastric aspirate. Secondary hemosiderosis is associated with systemic vasculitis, bleeding disorders and cardiac disease. Imaging findings are non-specific and need to be correlated with clinical and laboratory data to arrive at a diagnosis of IPH. Chest radiographs may reveal symmetric diffuse or patchy alveolar shadows sparing lung apices,which can clear on follow-up imaging. CT can show diffuse or patchy ground-glass opacity (Figure 10). There can be interstitial thickening in some cases[36].

Foreign body

Foreign body aspiration can be a cause of hemoptysis primarily in patients less than 3 years of age. The aspirated foreign body can be visualized on a radiograph if it is radio-opaque. Associated radiographic findings include non-specific infiltrates, atelectasis, areas of hyperinflation, parenchymal consolidation or bronchiectasis (Figure 11A). Chest radiographs can be normal in 30% cases. MPR and endoluminal virtual bronchoscopic images derived from MDCT can delineate the shape, location and volume of a foreign body. It can reveal associated pulmonary parenchymal changes (Figure 11B and C). Thus, imaging can help the surgeon plan the bronchoscopy for safe removal of foreign body[37].

Neoplasm

Bronchial neoplasms are a rare cause of hemoptysis (Figure 12). Bronchial carcinoid tumors are the most frequent primary pulmonary neoplasms of childhood. The lesion can be central or peripheral. Radiological findings include hilar or perihilar masses with lobulated margins and associated obstructive changes (atelectasis, consolidation, bronchocele or hyperinflation)[38]. On CT, carcinoid is seen as a well-defined, centrally located mass that narrows or deforms the airway and contains diffuse or punctuate calcification. It shows intense homogenous contrast enhancement. However, all carcinoids do not enhance. There can be associated pulmonary obstructive changes and mediastinal/hilar lymphadenopathy[39].

CONCLUSION

Hemoptysis is a distressing symptom for the child, the

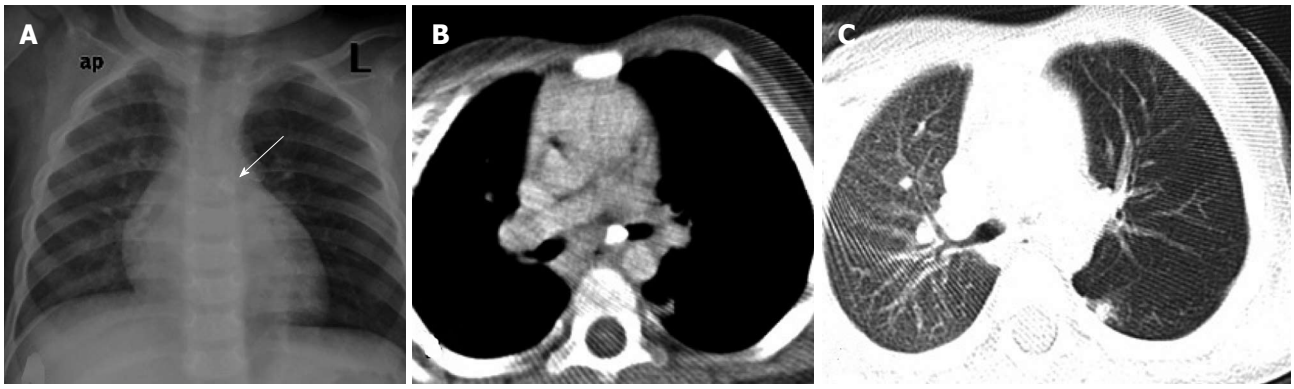

Figure 11 Foreign body aspiration. Chest radiograph (A) shows a radio-opaque foreign body in the left main bronchus (arrow) with hyperinflation of the left lung. Axial computed tomography images (B, C) delineate the morphology of the foreign body in the left main bronchus causing luminal compromise. There is associated air trapping in the left lung with patchy consolidation in the apical segment of the lower lobe.

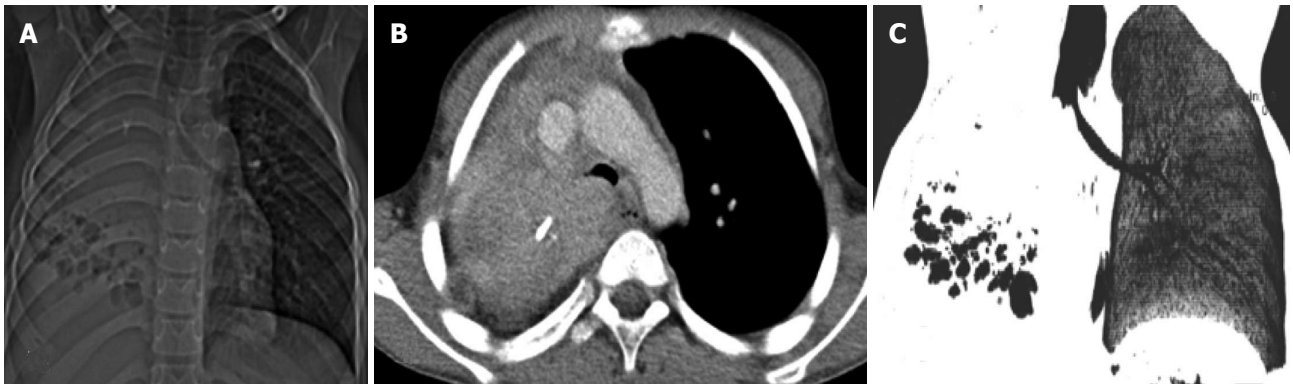

Figure 12 Bronchial carcinoid. Scout computed tomography (CT) image (A) revealing non-visualization of the right main bronchus with volume loss and opacification of the right hemithorax along with bronchiectasis in right lower zone. Axial CT image (B) shows a mass in the right lung with mediastinal infiltration. Coronal Minimum Intensity Projection image (C) shows the outline of the mass projecting in the right main bronchus along with bronchiectasis in the right lower lobe.

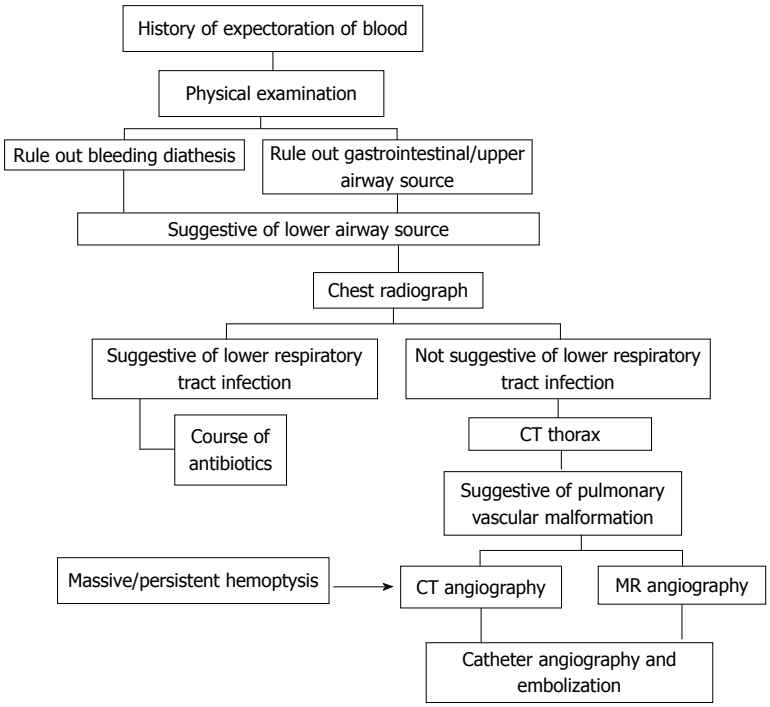

Figure 13 Flowchart depicting approach to a child presenting with hemoptysis. MR: Magnetic resonance; CT: Computed tomography.

family and the pediatrician. It poses a diagnostic challenge. Once the presence of hemoptysis has been ascertained, one needs to identify the source of bleeding and primary underlying cause. Acute lower respiratory tract infections are the most common cause of pediatric hemoptysis. The imaging modalities available for the work-up of hemoptysis include chest radiography, MDCT, MRI and catheter angiography. Chest radiographs may be normal in 30% cases. MDCT is a rapid, non-invasive multiplanar imaging modality which should be tailored to keep radiation dose to a minimum for optimal evaluation of hemoptysis in pediatric patients. MRI and catheter angiography have selected application. The use of the various imaging tools available is determined by the clinical presentation and the possible etiology (Figure 13). Maximum diagnostic and therapeutic benefit can be attained by the judicious use of imaging modalities in a child presenting with hemoptysis.

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BRIEF ARTICLE

Tramadol use in pediatric sickle cell disease patients with vaso-occlusive crisis

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Abstract

AIM: To evaluate whether the addition of scheduled oral tramadol to intravenous morphine and intravenous ketorolac reduces morphine requirements.

METHODS: This single-centered, Institutional Review Board-approved, retrospective study at Moses Cone Memorial Hospital included pediatric patients who were ⩾ 2 years old with vaso-occlusive crisis (VOC) caused by sickle cell disease (SCD), were on morphine patient-controlled analgesia (PCA), and had scheduled oral tramadol added to their standard pain regimen. The study population was admitted between March 2008 and March 2011. The data was collected from electronic records and included age, weight, morphine use, tramadol use, hemoglobin, pain scores, number of days on PCA, length of hospital stay, respiratory rate, and polyethylene glycol use. Thirty patients were analyzed as independent admissions and seven patients as paired admissions.

RESULTS: Eighteen pediatric SCD patients with VOC received morphine PCA and intravenous ketorolac and twelve patients received morphine PCA and intravenous ketorolac and scheduled oral tramadol. Baseline characteristics were similar between both groups with the exception of the average weight, which was greater in the tramadol group than in the morphine group. The average morphine requirements in patients with and without the use of tramadol were similar, both for the independent admissions [0.58 mg/kg per day *vs* 0.65 mg/kg per day (P = 0.31)] and the paired admissions [0.71 mg/kg per day *vs* 0.77 mg/kg per day (P = 0.5)]. The daily polyethylene glycol requirement was less in the tramadol group for both the independent [0.5 g/kg per day *vs* 0.6 g/kg per day (P = 0.64)] and paired admissions analyses [and 0.41 g/kg per day *vs* 0.55 g/kg per day (P = 0.67)].

CONCLUSION: The addition of scheduled tramadol in patients receiving concomitant morphine and ketorolac demonstrates a trend toward decreased morphine and polyethylene glycol use.

Key words: Pediatrics; Sickle cell; Tramadol; Morphine; Vaso-occlusive crisis

Core tip: A small clinical study has shown that balanced analgesia using intravenous morphine, intravenous ketorolac, and intravenous tramadol followed by erythrocytapheresis was effective, as shown by pain relief and significant improvement in mood and sleep, in seven sickle cell disease patients aged three to twenty-eight years who presented with vaso-occlusive crisis. The objective of this study is to evaluate whether the addition of scheduled oral tramadol to intravenous morphine plus intravenous ketorolac provides adequate pain relief, and reduces morphine requirements, adverse effects, length of patient-controlled analgesia therapy, and length of hospital stay.

Borgerding MP, Absher RK, So TY. Tramadol use in pediatric

sickle cell disease patients with vaso-occlusive crisis. *World J Clin Pediatr* 2013; 2(4): 65-69 Available from: URL: http://www.wjgnet.com/2219-2808/full/v2/i4/65.htm DOI: http://dx.doi.org/10.5409/wjcp.v2.i4.65

INTRODUCTION

One of the major causes of hospitalization for patients with sickle cell disease (SCD) is vaso-occlusive crises (VOC). The hallmark characteristics of VOC include organ damage and pain due to the presence of dense red blood cells. The pain is generated through multiple pathways including somatic, neuropathic, and vascular mechanisms[1-4].

Balanced analgesia is a strategy based on the co-administration of drugs with different pharmacological mechanisms in order to control pain at different origins, improve the efficacy of treatment, and reduce adverse effects of each drug[5]. The administration of ketorolac and tramadol has been proven as a form of effective balanced analgesia, particularly for post-operative pain and pain caused by trauma[6]. Tramadol and its active metabolite (M1) work by binding to the mu-opioid receptors in the central nervous system (CNS) and inhibiting the reuptake of norepinephrine and serotonin, causing inhibition of the ascending pain pathway and altering the perception of and response to pain[7]. Tramadol is clinically known to have a better safety profile than the other major opioids, causing less respiratory depression and constipation.

In 2005, de Franceschi *et al*[8] published a study evaluating balanced analgesia using intravenous (*iv*) morphine, *iv* ketorolac, and *iv* tramadol followed by erythrocytapheresis in seven SCD patients aged three to twenty-eight years who presented with VOC. The co-administration of tramadol and ketorolac was effective in all VOC, as shown by pain relief and significant improvement in mood and sleep. The use of erythrocytapheresis, which is not available at our hospital, Moses Cone Memorial Hospital (MCMH), likely contributed to pain relief in this study.

At MCMH pediatric patients who presented prior to August 2010 with VOC caused by SCD were routinely prescribed *iv* morphine and *iv* ketorolac for pain control. However, because of the high morphine requirement in this patient population, severe respiratory depression and constipation can occur. After August 2010, pediatric physicians began adding scheduled oral (*po*) tramadol to the standard regimen of *iv* morphine patient-controlled analgesia (PCA) and *iv* ketorolac in an attempt to reduce narcotic-induced side effects. The objective of this retrospective study is to evaluate whether the addition of scheduled *po* tramadol to *iv* morphine and *iv* ketorolac reduces morphine requirements, provides adequate pain relief, decreases length of hospital stay, and reduces severe respiratory depression, severe constipation, and length of PCA therapy.

MATERIALS AND METHODS

This single-centered, Institutional Review Board (IRB)-approved, retrospective study included pediatric patients who were ⩾ 2 years old with VOC caused by SCD, were on PCA morphine and *iv* ketorolac and had scheduled *po* tramadol added to their regimen. Tramadol was dosed at 1-2 mg/kg per dose *po* every four to 6 h (max: 400 mg/d and 100 mg/dose) and ketorolac at 0.5 mg/kg per dose *iv* every 6 h (Max: 30 mg/dose). Morphine PCA orders included a basal rate, intermittent dose, lockout interval, and a 1-hour and 4-hour limit. Using the International Classification of Diseases (ICD)-9 code for SCD, all patients < 21 years old who were admitted between March 2008 and March 2011 were included in this retrospective review. Patients were excluded from the review if they did not have a diagnosis of VOC or did not receive morphine PCA. The data was collected from electronic records and included age, weight, morphine use, tramadol use, hemoglobin, pain scores, number of days on PCA, length of hospital stay, respiratory rate, and polyethylene glycol (PEG) use. All patients were analyzed as independent admissions. Additionally, patients with multiple admissions during the study period (at least one with morphine only and one with both morphine and tramadol) were analyzed as paired admissions, acting as their own controls.

The primary outcome of this study was average daily morphine requirement. Secondary outcomes included average pain scores, respiratory rate, PEG dose, length of PCA therapy and number of days in the hospital.

All patients were analyzed as independent admissions using the Wilcoxon Rank Sum test. Patients who had multiple admissions, one with tramadol use and one without were also analyzed as paired admissions using the Wilcoxon Signed Rank test. The statistical analysis was completed using Stata, Version 10.1 (Cary, NC).

RESULTS

Between March 2008 and March 2011 eighteen pediatric SCD patients with VOC received morphine PCA and *iv* ketorolac and twelve patients received morphine PCA plus *iv* ketorolac and scheduled *po* tramadol. Baseline characteristics were similar between both groups with the exception of the average weight, which was greater in the tramadol group than in the morphine group because the latter group had a younger sample (Table 1).

Average morphine requirements with and without tramadol were similar in the independent admission analysis [0.58 mg/kg per day and 0.65 mg/kg per day, respectively (P = 0.31)]. Average morphine requirements with and without tramadol were also similar in the paired admissions analysis [0.71 mg/kg per day and 0.77 mg/kg per day, respectively (P = 0.5)]. Contradictory to what was expected, pain scores were higher when tramadol was added to the pain regimen for both the independent admissions (6.75 *vs* 5) and the paired admissions (6.5 *vs* 5.5). The daily

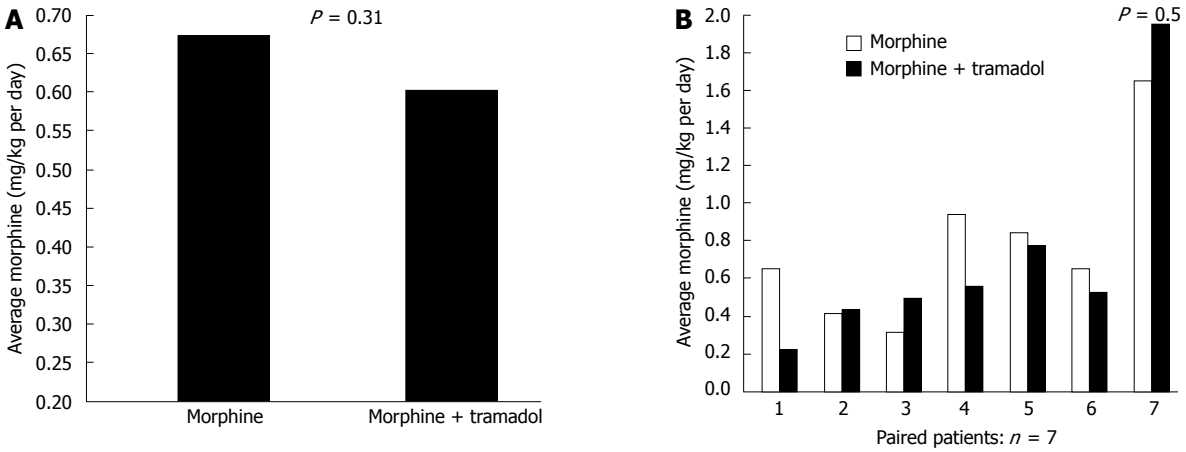

Figure 1 Average morphine requirement for independent admissions (A) and average morphine requirement for paired admissions (B).

Table 1 Baseline characteristics

	Morphine	Morphine + tramadol
Sample size	18	12
Age (yr), Mean (range)	11.5 (3-20)	13.4 (7-20)
Gender		
Male	9	6
Female	9	6
Weight (kg), mean (range)	38.7 (11.6-77.4)	52 (27.0-77.4)[a]
Hemoglobin (mg/dL), mean (range)	9.0 (6.0- 11.9)	9.4 (6.5-12.0)

[a]$P < 0.05$ *vs* morphine.

polyethylene glycol requirement was less in the tramadol group for both the independent and paired admissions analyses (0.5 g/kg per day *vs* 0.6 g/kg per day and 0.41 g/kg per day *vs* 0.55 g/kg per day, respectively) but neither difference was statistically significant ($P = 0.64$ and 0.67, respectively). The paired admissions analysis demonstrated a greater difference in PEG requirements which, while not statistically significant, may provide a more accurate comparison. Furthermore, there was no difference in the length of stay, number of days on PCA, or respiratory rate between groups in either analysis (Tables 2 and 3).

DISCUSSION

The addition of scheduled oral tramadol in patients receiving concomitant morphine and ketorolac did result in numerically lower average daily morphine requirements (Figure 1A) and polyethylene glycol use; however, differences in these endpoints were not statistically significant (Table 2). In the paired admissions analysis, four of the six patients who received less than 1 mg/kg per day of morphine used less morphine when tramadol was added to their regimen (Figure 1B); however, pain scores did not correlate with the decreased morphine requirement.

The lack of correlation between pain scores and morphine requirement may be due to the subjectivity of pain. After completion of the study, a pediatric psychiatrist spoke with several patients in the study and found that many patients did not understand how to use the visual analogue pain scale or the numeric scale to rate their pain. When asked to rate their pain as red, yellow or green (red corresponding to the most pain and green the least), patients gave more accurate representations of their true pain level. Extensive education on rating pain would be required to provide a more precise representation of pain relief with and without the use of tramadol.

There were several limitations to this study. The retrospective nature of the study forced us to rely on electronic nursing documentation for all of our data collection, which may have resulted in some inaccurately charted data. Frequently, bowel movements were not documented. We therefore measured average daily polyethylene glycol use to assess constipation. Additionally, we were unable to stratify patients based on their basal morphine PCA rate and determine how much of their daily morphine requirement was due to demand dosing, as this information was not documented electronically. Patients with multiple admissions for VOC can develop tolerance to narcotics, resulting in an increased morphine basal rate requirement. Documentation of this may have provided a more accurate assessment of which analgesic regimen provided better pain control and fewer narcotic-induced side effects.

VOC most commonly involves the back, legs, knees, arms, chest and abdomen. The location of the vaso-occlusive crisis has a significant impact on the intensity of pain and the abilitiy to control that pain; however, this study did not stratify patients based on VOC location or disease severity[9]. Additionally, the disease severity has interpatient and intrapatient variability making it more difficult to compare patients. Also, one patient in this study had drug-seeking behavior, which may have skewed the results causing increased average morphine requirement and pain scores.

A larger, controlled study would be more likely to determine statistical difference in the primary and secondary endpoints. Pediatric physicans at MCMH no longer routinely prescribe tramadol in this population but con-

Table 2 Independent admissions statistical analysis

	Morphine (n = 18)	Morphine + tramadol (n = 12)	P-value[1]
Morphine requirement (mg/kg per day), mean ± SD	0.65 ± 0.35	0.58 ± 0.48	0.31
Pain score, median (range)	5 (0-8)	6.75 (3-9)	0.07
PCA duration (d), median (range)	4.5 (0-13)	5 (3-14)	0.33
LOS (d), median (range)	7 (4-14)	7.5 (3-17)	0.85
Respiratory rate, mean (range)	21 (14-29)	19 (16-23)	0.28
Polyethylene glycol dose (gm/kg per day), mean (range)	0.6 (0-1.66)	0.5 (0-1.12)	0.64

[1]*P*-value calculated using Wilcoxon Rank Sum test. PCA: Patient-controlled analgesia; LOS: Length of stay.

Table 3 Paired admissions statistical analysis (n = 7)

	Morphine	Morphine + tramadol	P-value[1]
Morphine requirement (mg/kg per day), mean ± SD	0.77 ± 0.44	0.71 ± 0.57	0.50
Pain score, median (range)	5.5 (2-8)	6.5 (3-8)	0.27
PCA duration (d), median (range)	6 (4-13)	7 (3-10)	0.35
LOS (d), median (range)	7 (5-14)	9 (3-12)	0.61
Respiratory rate, mean (range)	19 (14-20)	19 (17-23)	0.87
Polyethylene glycol dose (gm/kg per day), mean (range)	0.55 (0.28-1.1)	0.41 (0.28-0.98)	0.67

[1]*P*-value calculated using Wilcoxon Rank Sum test. PCA: Patient-controlled analgesia; LOS: Length of stay.

tinue to use it in patients who have clinically shown improved pain control with tramadol in the past. Tramadol use is appropriate in this population as it has proven safe, usually causing no additional side effects and potentially providing some benefit in controlling pain and reducing narcotic-induced constipation. If tramadol continues to be clinically beneficial for pain control in other patients in this population, it may be possible to review our primary and secondary endpoints on a larger scale to determine any true differences.

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COMMENTS

Background

Pain is a hallmark of vaso-occlusive crisis (VOC) caused by sickle-cell disease (SCD). Intravenous (*iv*) morphine plus *iv* ketorolac is generally the combination of choice for VOC in SCD patients at Moses Cone Memorial Hospital (MCMH); however, morphine can cause severe respiratory depression and constipation. The objective of this study is to evaluate whether the addition of scheduled oral tramadol to *iv* morphine plus *iv* ketorolac provides adequate pain relief, and reduces morphine requirements, adverse effects, length of PCA therapy, and length of hospital stay.

Research frontiers

In 2005, de Franceschi *et al* published a study evaluating balanced analgesia using *iv* morphine, *iv* ketorolac, and *iv* tramadol followed by erythrocytapheresis in seven SCD patients aged three to twenty-eight who presented with VOC. The co-administration of tramadol and ketorolac was effective in all VOC, as shown by pain relief and significant improvement in mood and sleep.

Innovations and breakthroughs

This is the first article evaluating the use of oral tramadol in addition to *iv* morphine and ketorolac on whether this combination provides adequate pain relief, and reduces morphine requirements, adverse effects, length of PCA therapy, and length of hospital stay.

Applications

The study results suggest that the addition of scheduled oral tramadol in patients receiving concomitant morphine and ketorolac demonstrates a trend toward decreased morphine and polyethylene glycol use.

Terminology

Erythrocytapheresis is a process in which red blood cells are extracted from the whole blood.

Peer review

This is a very good clinical study in which the authors analyzed the efficacy of adding oral tramadol to usual pain regimen used for sickle cell pain crisis.

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BRIEF ARTICLE

Pediatric *vs* adult pulmonary tuberculosis: A retrospective computed tomography study

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Abstract

AIM: To compare the manifestations of chest tuberculosis (TB) in pediatric and adult patients based on contrast enhanced computed tomography of chest.

METHODS: This was a retrospective study consisting of 152 patients of chest TB including 48 children and 104 adults who had undergone contrast enhanced computed tomography of chest prior to treatment. The patterns and severity of parenchymal, mediastinal and pleural manifestations were analyzed and compared among different age groups.

RESULTS: Parenchymal changes observed include consolidation, air space nodules, miliary TB, cavitation, bronchiectasis and fibrosis and these were noted in 60% of children, 71% of adolescents and 76.9% of adults. These changes were more common in right upper lobe in all age groups. There was no significant difference in the frequency of these changes (except nodules) in different age groups. Centrilobular nodules were seen less commonly in children less than 10 years (P = 0.028). Pleural effusion was noted in 28 (18.42%) patients and pericardial effusion in 8 (5.3%) patients. No significant difference in the serosal involvement is seen among children and adults. Mediastinal adenopathy was seen 70% of children, 76.3% adolescents and 76.9% of adults and paratracheal nodes were seen most frequently. Nodes had similar features (except matting) among all age groups. Matting of nodes was seen more commonly in children (P = 0.014).

CONCLUSION: Pediatric chest tuberculosis can have severe parenchymal lesions and nodal involvement similar to adults. The destructive lung changes observed in children needs immediate attention in view of the longer life span they have and hence in formulating optimal treatment strategies.

Key words: Tuberculosis; Pulmonary; Primary tuberculosis; Children; Computed tomography

Core tip: Primary tuberculosis in children was traditionally thought to be distinct from reactivation tuberculosis in terms of location, pattern and severity. On the contrary, aggressive forms of pulmonary tuberculosis akin to adult forms are increasingly seen in pediatric clinical practice especially in adolescents. Our study revealed that similar to older patients, children with tuberculosis are equally prone to develop significant destructive lung changes with severe sequelae. Having longer life expectancy the impact is much more severe in children.

Moreover, the cavitating lesions with high bacterial load make them highly infective and pose an important threat to community health.

Thotton Veedu P, Bhalla AS, Vishnubhatla S, Kabra SK, Arora A, Singh D, Gupta AK. Pediatric *vs* adult pulmonary tuberculosis; a retrospective CT study. *World J Clin Pediatr* 2013; 2(4): 70-76 Available from: URL: http://www.wjgnet.com/2219-2808/full/v2/i4/70.htm DOI: http://dx.doi.org/10.5409/wjcp.v2.i4.70

INTRODUCTION

Pulmonary tuberculosis (TB) is a common lung infection worldwide with higher prevalence in developing countries. It continues to be a major medical and social problem with high morbidity and mortality. TB is second only to human immunodeficiency virus (HIV)/acquired immune deficiency syndrome (AIDS) as the greatest killer worldwide due to a single infectious agent. In 2011, 8.7 million people contracted tuberculosis out of which 1.4 million died from TB. About half a million children (0-14 years) fell ill with TB, and 64000 children died from the disease in 2011[1]. The annual risk of tuberculous infection in children in developing countries is 2%-5%. About 8%-20 % of deaths due to tuberculosis occur in children[2,3].

Tuberculosis in children is mostly related to primary infection and earlier studies stated that it present with various forms of relatively less aggressive primary tuberculosis[4]. Traditionally it was thought that manifestations of primary tuberculosis are distinct from reactivation tuberculosis in terms of location and pattern[5,6]. The most common form of pediatric TB, the classical primary complex consists of a focal parenchymal lesion typically in mid-lower zones with enlarged draining hilar/paratracheal node. Other presentations of primary TB include miliary TB[7], exudative pleuritis and tracheo-bronchial TB[8,9]. However contrary to the common notion, aggressive forms of pulmonary tuberculosis akin to adult forms are increasingly seen in pediatric clinical practice especially in adolescents[10-13]. A thorough review of available literature did not reveal any comparative studies with computed tomography (CT) scan in pediatric and adult tuberculosis. Our retrospective study is aimed to compare the pulmonary manifestations of TB in pediatric and adult population.

MATERIALS AND METHODS

In this retrospective study we analyzed CT records of 152 patients of pulmonary tuberculosis who underwent CT scans from November 2010 to January 2013. The diagnosis of pulmonary tuberculosis was based on clinical/and radiographic/and pathologic criteria. Patients were presented with clinical symptoms such as cough for more than two weeks, fever, weight loss, hemoptysis or anorexia. Along with clinical features two out of four of the following criteria had to be met: (1) A positive mantoux test; (2) History of contact with a sputum positive patient of tuberculosis; (3) Radiographic findings of mycobacterium tuberculosis such as primary complex, miliary disease, cavitary lesion, or hilar adenopathy; and (4) Isolation of AFB from sputum, gastric aspirate or broncho-alveolar lavage, lymph node aspirate[14-19]. Immunocompromised patients were excluded from the study. Informed consent and clearance from the local ethical committee was not required due to the retrospective nature of the study.

CT scans were performed either on Somatom Sensation 40 (Siemens, Erlangen, Germany) or Somatom Definition Flash (Siemens healthcare, Forchheim, Germany). Images were acquired after administering intravenous nonionic iodinated contrast [Iomeron 400 (Iomeprol, Bracco, Milano, Italy), Iohexol 300 (Omnipaque, GE Health care, Ireland)] which was injected by hand with an average delay of 50-70 s and thus providing venous phase images. Adult patients were given 60 mL of contrast and pediatric dose was calculated according to the body weight not exceeding 2 mL/kg.

Scanning was performed in adults with a collimation of 1.2 mm, a pitch of 1.4:1, a 512 × 512 matrix, field view of 38 cm, 120 kVp, and 100 mAs. In children images were acquired with a smaller field of view with 120 kVp and variable mAs (on Somatom Sensation 40) and variable kVp and mAs (on Somatom Definition Flash) according to the body thickness by tube current modulation.

Image reconstruction

After acquisition images were reconstructed in lung and mediastinal windows. Lung window images were reconstructed using sharp kernel (B60f) and a wide window width of 1500 HU with centre at -600 HU. Mediastinal window images are reconstructed using smooth kernel (B30f) and window width of 400 HU with centre at 40 HU. For both lung and mediastinal windows images were reconstructed with a thickness of 5mm in adults and 3 mm in children. HRCT images were reconstructed in lung window settings using ultra-sharp kernel (U80f) with section thickness of 1.5 mm. For HRCT reconstruction interslice gap was 10 mm in adults and 8 mm in children.

Image analysis

Images were reviewed by two radiologists, with 15 years and 7 years of experience in thoracic imaging and interpretations made by consensus. Images were qualitatively analyzed for the presence of parenchymal changes and lymph nodal involvement. Parenchymal changes such as consolidation, centrilobular nodules, miliary nodules, bronchiectasis, cavitation and fibrosis were assessed. For analyzing the zonal predominance and bulkiness of the disease both lungs were divided into upper, mid and lower zones. Lung field from apex to carina as upper zone, carina to the level of inferior pulmonary veins as mid zone and below as lower zone. Distribution of

abnormalities and total number of zones involved as a measurement of bulkiness of the disease were assessed. Mediastinal and hilar lymph nodes were assessed for the size, necrosis, matting and calcification. Other findings like pleural and pericardial effusion were noted.

Statistical analysis

For descriptive statistics the study population was divided into three groups; group A: children(less than or equal to10 years), group B: adolescents (11-18 years) and Group C: adults (above 18 years). The incidence, pattern and severity of parenchymal changes and nodal involvement were compared among the groups. They were also divided as below and above 10 years; and below and above 18 years for the determination of statistical significance. Statistical analysis was done using stata statistical software (version 12.1). χ^2 test and Fisher's exact test were used for analysis. A P value of less than 0.05 was considered as significant.

RESULTS

The study group included 80 males and 72 females patients ranging in age from 3 mo to 96 years (mean 30 years). They were comprised of 10 children, 38 adolescents and 104 adults. Incidence and pattern of parenchymal changes and lymph nodal involvement were analyzed and compared among different groups (Figure 1).

We compared the parenchymal changes among pediatric, adolescent and adult patients. Parenchymal lesions were noted in 6 (60%) children, 27 (71%) adolescents and 80 (76.9%) adults (Table 1). Parenchymal changes were more common in right upper followed by right middle zone in all age groups. Higher incidence of changes noted in upper and middle zones than lower zones. Left lower zone is least commonly involved in patients older than 10 years. Among the patients with parenchymal disease, multiple zones (≥ 3) were seen to be involved in 3 (30%) children, 16 (42.1%) adolescents and 45 (56.25%) adult patients (Table 1). The average number of zones involved in children, adolescent and adult patients were 2.83, 3.33 and 3.32 respectively.

Among the 152 patients we studied, consolidation was found in 54 (35.53%), centrilobular nodules in 93 (61.18%), bronchiectasis in 26 (17.11%), miliary in 4 (2.63%), fibrosis in 22 (14.47%) and cavitation in 36 (23.84%) patients. There was no statistically significant difference in the incidence of consolidation, miliary nodules, bronchiectasis, cavitation and fibrosis among different age groups studied. Centrilobular nodules were seen less commonly in children ($P = 0.028$) (Table 1).

Mediastinal lymphadenopathy was seen in 7 (70%), 29 (76.3%) and 74 (71.2%) respectively in children, adolescents and adult patients (Table 1). Among the mediastinal lymph nodes right paratracheal is the most commonly involved followed by subcarinal in all age groups. Involvement of multiple nodal groups (≥ 2) was seen in 28 (58%) of younger patients (≤ 18 years) and 51 (49%) older patients. In less than 10 years category all the 7 (70%) children with significant lymphadenopathy had involvement of multiple nodal groups. Involvement of multiple nodes (≥ 2 groups) was more commonly seen in children less than 10 years. The average number of nodal groups involved in children, adolescent and adult patients were 3.57, 2.52 and 2.23 respectively. Lymph node matting was seen more commonly in children (Table 1).

In 4 (40%) patients below 10 years lymphadenopathy was the only finding. Similarly 11 (29 %) patients between 10 to 18 years and 24 (23%) patients above 18 years had only lymphadenopathy.

Pleural effusion was noted in 28 (18.42%) patients and 13 of them showed loculation. Pericardial effusion was present in 8 (5.3%) patients. No significant difference in the serosal involvement is seen among children and adults (Table 1).

DISCUSSION

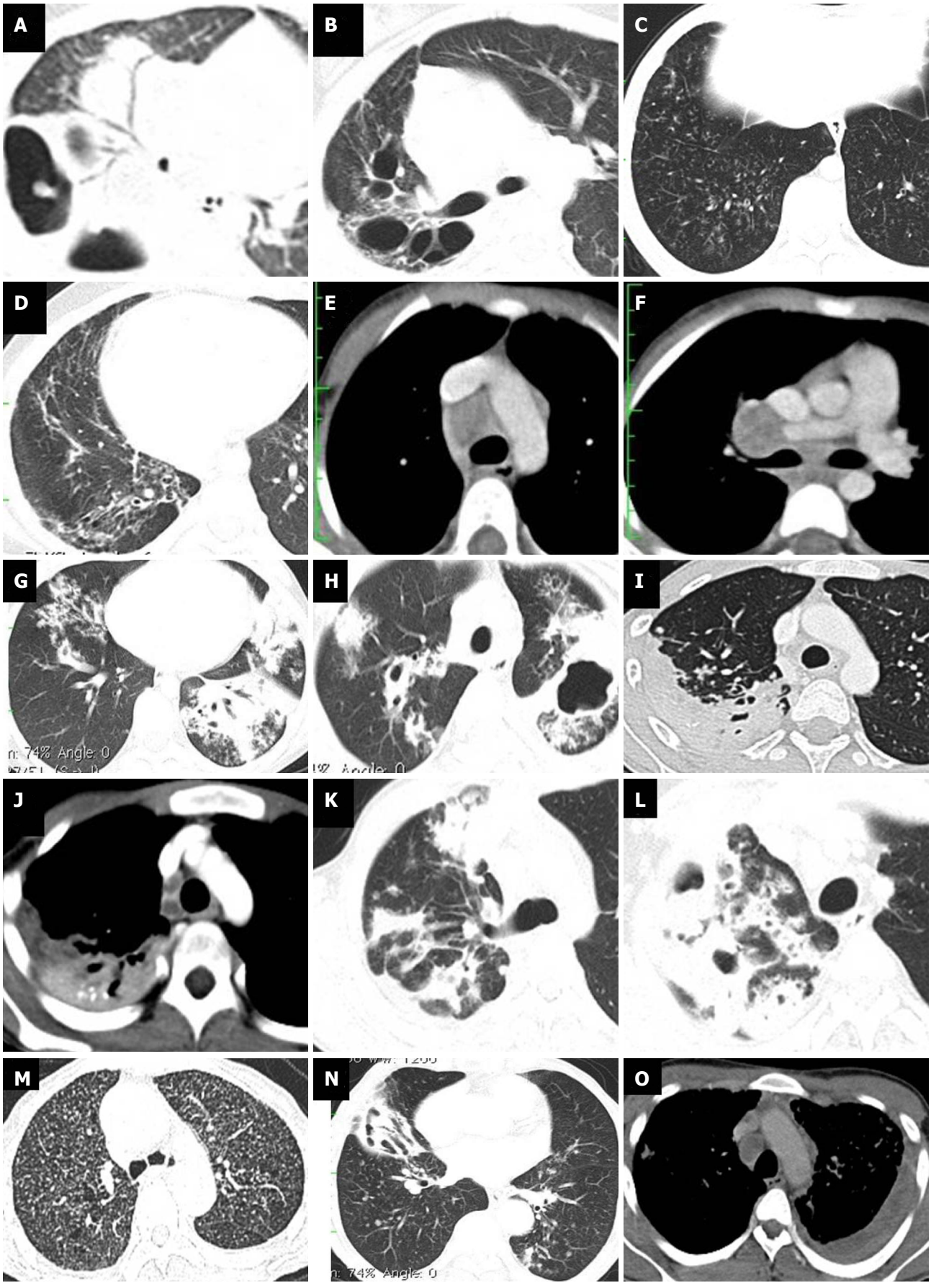
Tuberculosis continues to be a major medical, social and economic problem worldwide with its high morbidity and mortality. Children constitute about 5%-10% of patients suffering from tuberculosis worldwide. World Health Organization (WHO) data shows that in the year 2011 itself about half a million children fell ill with tuberculosis and about 64000 died from the disease[1].

The pathologic form of pulmonary tuberculosis is classically classified as primary and post primary or secondary tuberculosis and is depended on the sensitivity of infected host. Primary tuberculosis presenting with hilar/paratracheal adenopathy with or without focal parenchymal changes in mid-lower zones is thought to be predominant form of tuberculosis in children. However contrary to the traditional belief incidence of adult form of TB is increasingly seen in pediatric patients in terms of location and severity. Previous authors have already questioned the validity of the terminologies (primary and secondary tuberculosis) in recent literature[10,11].

Our study included 152 patients including 48 children (below 18 years). Previous studies on pediatric tuberculosis used varying age criteria for children (Table 2). We also have separately analyzed data of children below 10 years and adolescents (11-18 years).

Mediastinal lymph nodes

Mediastinal lymphadenopathy is the predominant finding in primary tuberculosis either alone or in association with lung lesions[20]. Our study showed mediastinal adenopathy in 7 (70%), 29 (76.3%) and 74 (71.2%) respectively in children, adolescents and adult patients. Compared to previous studies slightly lower incidence of adenopathy is observed in our study (Table 2). Although there was no significant difference in the incidence of adenopathy in different age groups, there was a definite trend in the extensiveness of nodal involvement. Involvement of multiple nodal groups was seen significantly more common in children. Mediastinal adenopathy was the only finding in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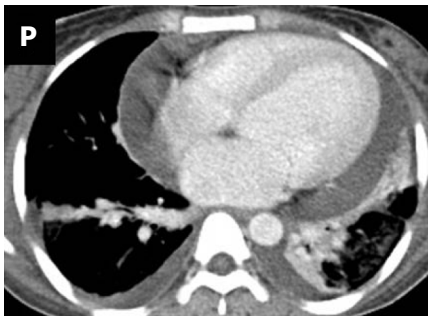

Figure 1 Computed tomography findings in children (less than 10 years), adolescents (10-18 years), and adults (above 18 years) with chest tuberculosis. A-F: Children; A: Parenchymal consolidation in right lung with adjacent thick walled cavities; B: Multiple cavities with adjacent fibrosis in right upper lobe; C: Tiny low density centrilobular nodules seen diffusely in both lungs; D: Fibrobronchiectatic changes in right middle and lower lobes; E-F: Enlarged low attenuating necrotic nodes in right paratracheal, and right hilar location repectively; G-J: Adolescents; G: Parenchymal consolidation with centrilobular nodules in left lung, nodules in right middle lobe; H: Thick walled cavities with surrounding consolidation and air space nodules in both upper lobes; I-J: Pleural based fibrotic changes with calcification in right upper lobe with necrotic right paratracheal node; K-P: Adults; K: Multifocal consolidation in right upper lobe; L: Thick walled cavities in right upper lobe; M: Miliary TB- multiple tiny nodules distributed randomly in both lungs; N: Fibrobronchietasis in right middle lobe with nodules in left lower lobe; O: Necrotic right paratracheal node with empyema in left side; P: Pericardial and bilateral pleural effusion.

Table 1 Incidence of lung and nodal involvement, Parenchymal changes, Mediastinal lymph nodal involvement and Pleural and pericardial effusion in the patients *n* (%)

		Children (n = 10)	Adolescents (n = 38)	Adults (n = 104)
Incidence of lung and nodal involvement	Parenchymal Lesions	6 (60)	27 (71)	80 (76.9)
	Mediastinal nodes	7 (70)	29 (76.3)	74 (71.2)
Zonal distribution of parenchymal changes	Right upper	5 (50)	19 (50)	58 (55.77)
	Left upper	2 (20)	15 (39.47)	45 (43.27)
	Right middle	4 (40)	19 (50)	54 (51.92)
	Left middle	1 (10)	16 (42.11)	43 (41.35)
	Right lower	3 (30)	12 (31.58)	35 (33.65)
	Left lower	2 (20)	9 (23.68)	30 (28.85)
Pattern of parenchymal changes	Consolidation	3 (30)	16 (42.11)	35 (33.65)
	Centrilobular nodules[a]	2 (20)	24 (63.16)	67 (64.42)
	Miliary nodules	1 (10)	0 (0.00)	3 (2.88)
	Bronchiectasis	1 (10)	8 (21.05)	17 (16.35)
	Fibrosis	1 (10)	7 (18.42)	14 (13.46)
	Cavitation	3 (30)	10 (26.32)	23 (22.33)
Lymph nodal distribution	Paratracheal	7 (70)	22 (57.59)	57 (54.81)
	Precarinal	6 (60)	9 (23.68)	27 (25.96)
	Subcarinal	7 (70)	17 (44.74)	40 (38.46)
	Hilar	5 (50)	13 (34.21)	27 (25.96)
	AP window	0 (0)	9 (23.68)	12 (11.54)
Characteristics of lymph nodes	Lymphadenopathy	7 (70)	21 (55.26)	50 (48.08)
	Necrosis	6 (60)	17 (44.74)	43 (41.35)
	Matting[a]	5 (50)	11 (28.95)	16 (15.38)
	Calcification	4 (40)	10 (26.32)	25 (24.04)
Pleural and pericardial effusion in the patients	Pleural effusion	2 (20)	5 (13.16)	21 (20.19)
	Pleural loculation	1 (10)	2 (5.3)	10 (9.6)
	Pericardial effusion	0 (0)	1 (2.63)	7 (6.73)

[a]$P < 0.05$.

31.3% of children (group A and B) against 23% of older patients (group C). This is different from data of Kim *et al*[21] and Khatami *et al*[22] who showed isolated nodal involvement in 7% and 10% respectively. Paratracheal followed by subcarinal were the most common locations of nodal involvement in all the groups and is similar to a recent data from Andronikou *et al*[23] and Mukund *et al*[24].

Necrotic nodes characterized by inhomogeneous attenuation or low attenuation centre and enhancing peripheral rim were seen in 48% of children (group A and B) and 41% of adults (group C)[25]. Necrotic nodes were shown in 65% of children in a study by Mukund *et al*[24] which is comparable. Matted nodes were seen in 50% of young children (group A) against 19% of older patients (groups B and C) ($P = 0.02$). Nodal calcification was seen in 40%, 26.3% and 24% of children, adolescents and

Table 2 Previous studies on imaging in pediatric chest tuberculosis

Ref.	Age (yr)	Mod	No	Cons	Nodu	Mil	Cavity	Bects	Node	Plefn
Leung *et al*[4]	< 16	X-ray	191	69%	NS	NS	NS	NS	92%	6%
Kim *et al*[21]	< 14	CT	41	49%	29%	17%	7%	NS	83%	17%
Khatami *et al*[22]	< 15	X ray	30	43.30%	NS	NS	NS	NS	90%	6.70%
Koh *et al*[10]	15-19	X-ray	90	25%	96%	NS	45%	0	2%	0
Mukund *et al*[24]	< 17	CT	91	NS	NS	NS	NS	NS	96.70%	NS
Our study	< 18	CT	48	39.60%	54%	2%	27%	18.80%	75%	14.60%

Mod: Imaging modality used; No: Number of cases; Cons: Consolidation; Nodu: Centrilobular nodules; Mil: Miliary nodules; Bects: bronchiectasis; Node: Mediastinal adenopathy; Plefn: Pleural effusion; NS: Not specified.

adults respectively. Nodal calcification was seen in 12 % and 28.4% of children in studies by Kim *et al*[21] and Mukund *et al*[24] respectively. Hence apart from higher incidence of matting seen in young children no other significant difference in nodal characteristics is seen in different age groups.

Parenchymal changes

The typical parenchymal change in primary tuberculosis is focal consolidation (Ghon's focus) seen classically in mid and lower zones. Cavitation, fibrosis and bronchiectasis are not commonly seen in primary tuberculosis. In our study, right upper zone was most commonly involved in all age groups and is comparable to the data by Koh *et al*[10] in which upper zone predominance is seen in 49% of patients. 68.75% of children (group A and B) showed lung parenchymal changes of tuberculosis against 76.9% of older patients (group C). Cavitation was seen in 30% of young children and 26% adolescents. Cavitating pneumonia was seen in 7% of children in the study by Kim *et al*[21] and 45% of adolescents by Koh *et al*[10]. The recognition of cavitating disease in children is important because the presence of cavities correlate with organism load, drug resistance, treatment outcome and infectivity[11]. Bronchiectasis was seen in 18.8 % children (group A and B) against 16.4% in adults (Table 1).

There was no significant statistical difference in the incidence of consolidation, miliary disease, bronchiectasis, fibrosis and cavitation among different age groups. Centrilobular nodules were less commonly seen in children. When we compared the incidence of nodules in children (group A) against older patients (group B and C) significantly lower incidence is noted in young children ($P = 0.014$).

Our study showed comparable frequency of pleural effusion, empyema and pericardial effusion in all age groups. It showed 20% and 13% incidence of pleural effusion in children and adolescents (Table 1). Kim *et al*[21] observed pleural effusion in 17% of children with tuberculosis.

To conclude, children with pulmonary tuberculosis are equally prone to develop significant destructive changes in the lung with severe sequelae similar to older patients. The impact is much more severe in cases of children because they have longer life expectancy. Moreover, the cavitating lesions with high bacterial load make these children highly infective and pose an important hazard to community health, than previously thought. The similar location and aggressiveness of parenchymal changes observed in children, blurs the boundary defining primary and reactivation tuberculosis. Hence the need for revision of these terminologies demands urgent attention.

Limitations

First, our study included only those patients who had undergone CT scan at a tertiary institution and hence may not represent the exact patient population at the primary care level. However as all the patients in different age groups had been filtered in the same way the comparisons should remain valid. Secondly, we had only ten patients in below 10 years category and hence further studies including more number of children will help in strengthening the observations.

COMMENTS

Background

Tuberculosis is a contagious lung disease caused by mycobacterium tuberculosis. It is widely prevalent in developing countries and is a major threat to community health. Tuberculosis in children is traditionally believed to be less severe and less contagious as compared to adult disease. However recent evidences suggest that the distinction is far from truth and children are presenting with severe disease in presentation and outcome. This retrospective study is done to compare the disease characteristics based on imaging [computed tomography (CT) of chest] among children and adults who were presented to a tertiary care centre.

Research frontiers

Tuberculosis in children can present in various forms ranging from subclinical infection to destructive parenchymal disease or extensive miliary disease. Early recognition of children having severe disease is essential in the management of disease and in prevention of spread. Although chest radiography is the primary imaging modality in chest tuberculosis, CT is advisable in children presenting with atypical and severe manifestations. Studies on this background are essential in planning and modifying the strategies in the management of pediatric tuberculosis.

Innovations and breakthroughs

Present study has shown that the disease can strike children with similar aggressiveness as in adults and can very much be a source of infection in the community. Hence it is important to take adequate measures to prevent disease spread along with optimal treatment planning.

Applications

Early recognition of severe manifestations of pediatric chest tuberculosis is beneficial in optimal treatment planning. Despite the higher radiation exposure

involved CT of chest is the best investigation to get an accurate estimate of the disease in atypical and complicated cases.

Terminology

CT stands for CT which is a radiological investigation using X-rays to produce tomographic images of the body. In the chest using different post-processing techniques CT provides much greater information than radiography.

Peer review

The study of pediatric *vs* adult pulmonary tuberculosis is very good. The study highlights the incidence of severe manifestations in pediatric chest tuberculosis, the early detection of which is beneficial in optimal treatment planning.

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Format

Journals

English journal article (list all authors and include the PMID where applicable)

1 **Jung EM**, Clevert DA, Schreyer AG, Schmitt S, Rennert J, Kubale R, Feuerbach S, Jung F. Evaluation of quantitative contrast harmonic imaging to assess malignancy of liver tumors: A prospective controlled two-center study. *World J Gastroenterol* 2007; **13**: 6356-6364 [PMID: 18081224 DOI: 10.3748/wjg.13.6356]

Chinese journal article (list all authors and include the PMID where applicable)

2 **Lin GZ**, Wang XZ, Wang P, Lin J, Yang FD. Immunologic effect of Jianpi Yishen decoction in treatment of Pixu-diarrhoea. *Shijie Huaren Xiaohua Zazhi* 1999; **7**: 285-287

In press

3 **Tian D**, Araki H, Stahl E, Bergelson J, Kreitman M. Signature of balancing selection in Arabidopsis. *Proc Natl Acad Sci USA* 2006; In press

Organization as author

4 **Diabetes Prevention Program Research Group**. Hypertension, insulin, and proinsulin in participants with impaired glucose tolerance. *Hypertension* 2002; **40**: 679-686 [PMID: 12411462 PMCID:2516377 DOI:10.1161/01.HYP.0000035706.28494.09]

Both personal authors and an organization as author

5 **Vallancien G**, Emberton M, Harving N, van Moorselaar RJ; Alf-One Study Group. Sexual dysfunction in 1, 274 European men suffering from lower urinary tract symptoms. *J Urol* 2003; **169**: 2257-2261 [PMID: 12771764 DOI:10.1097/01.ju.0000067940.76090.73]

No author given

6 21st century heart solution may have a sting in the tail. *BMJ* 2002; **325**: 184 [PMID: 12142303 DOI:10.1136/bmj.325.7357.184]

Volume with supplement

7 **Geraud G**, Spierings EL, Keywood C. Tolerability and safety of frovatriptan with short- and long-term use for treatment of migraine and in comparison with sumatriptan. *Headache* 2002; **42** Suppl 2: S93-99 [PMID: 12028325 DOI:10.1046/j.1526-4610.42.s2.7.x]

Issue with no volume

8 **Banit DM**, Kaufer H, Hartford JM. Intraoperative frozen section analysis in revision total joint arthroplasty. *Clin Orthop Relat Res* 2002; (**401**): 230-238 [PMID: 12151900 DOI:10.1097/00003086-200208000-00026]

No volume or issue

9 Outreach: Bringing HIV-positive individuals into care. *HRSA Careaction* 2002; 1-6 [PMID: 12154804]

Books

Personal author(s)

10 **Sherlock S**, Dooley J. Diseases of the liver and billiary system. 9th ed. Oxford: Blackwell Sci Pub, 1993: 258-296

Chapter in a book (list all authors)

11 **Lam SK**. Academic investigator's perspectives of medical treatment for peptic ulcer. In: Swabb EA, Azabo S. Ulcer disease: investigation and basis for therapy. New York: Marcel Dekker, 1991: 431-450

Author(s) and editor(s)

12 **Breedlove GK**, Schorfheide AM. Adolescent pregnancy. 2nd ed. Wieczorek RR, editor. White Plains (NY): March of Dimes Education Services, 2001: 20-34

Conference proceedings

13 **Harnden P**, Joffe JK, Jones WG, editors. Germ cell tumours V. Proceedings of the 5th Germ cell tumours Conference; 2001 Sep 13-15; Leeds, UK. New York: Springer, 2002: 30-56

Conference paper

14 **Christensen S**, Oppacher F. An analysis of Koza's computational effort statistic for genetic programming. In: Foster JA, Lutton E, Miller J, Ryan C, Tettamanzi AG, editors. Genetic programming. EuroGP 2002: Proceedings of the 5th European Conference on Genetic Programming; 2002 Apr 3-5; Kinsdale, Ireland. Berlin: Springer, 2002: 182-191

Electronic journal (list all authors)

15 Morse SS. Factors in the emergence of infectious diseases. Emerg Infect Dis serial online, 1995-01-03, cited 1996-06-05; 1(1): 24 screens. Available from: URL: http://www.cdc.gov/ncidod/eid/index.htm

Patent (list all authors)

16 **Pagedas AC**, inventor; Ancel Surgical R&D Inc., assignee. Flexible endoscopic grasping and cutting device and positioning tool assembly. United States patent US 20020103498.

2002 Aug 1

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Write as mean ± SD or mean ± SE.

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