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Original Article

Respiratory Outcomes at 5-Year Follow-Up in Children with Mannose-Binding Lectin Deficiency: A Retrospective Cohort Study

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Abstract

OBJECTIVE: Mannose-binding lectin deficiency may predispose children to having increased infection susceptibility. However, there is no conclusive evidence that mannose-binding lectin deficiency is associated with adverse respiratory consequences in children. We aimed to evaluate the effects of mannose-binding lectin deficiency (defined as a level of less than 0.6 mg/L) on clinical, radiological, and microbiological characteristics in children presenting with troublesome respiratory symptoms, as compared to those who are mannose-binding lectin-sufficient.

MATERIAL AND METHODS: We conducted a retrospective cohort study to investigate the association between mannose-binding lectin deficiency and respiratory outcomes in children over a period of 10 years in a large teaching hospital. Children presenting with frequent or persistent respiratory symptoms such as a chronic wet cough lasting more than 4 weeks, recurrent lower respiratory tract infections (≥4 infections in a year), or severe respiratory tract infections requiring admission to intensive care or to high dependency unit were included in the study.

RESULTS: The study showed no significant difference in clinical outcomes with mannose-binding lectin deficiency and sufficiency. Thirty-two percent of children with mannose-binding lectin deficiency and 30% of those with mannose-binding lectin sufficiency had positive respiratory microbiology. Twenty-three percent of children with mannose-binding lectin deficiency and 24% of those with mannose-binding lectin sufficiency had radiological changes on plain radiographs; also the prevalence of bronchiectasis was similar in both groups. The rates of admission to pediatric intensive care unit were comparable in the 2 groups.

CONCLUSIONS: Children with mannose-binding lectin deficiency and sufficiency showed similar clinical, radiological, and microbiological characteristics. Our study suggests that there are no childhood adverse respiratory consequences with mannose-binding lectin deficiency.

KEYWORDS: Child, pediatric, pneumonia, respiratory distress, viruses
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INTRODUCTION

The innate immune system orchestrates immediate defense against infections. It includes mannose-binding lectin (MBL), which is a liver-produced acute-phase protein and an important first-line defense molecule largely involved in antimicrobial recognition and clearing responses.[1,2] It activates the lectin pathway of the complement system, namely cleaving factor C4 through MBL-associated serine proteases.[3]

The levels of MBL vary in the population, with a level of less than 0.6 mg/L defined as MBL deficiency.[4] At birth, the level is two-thirds of the adult level, which increases to the adult level at 1 month of life, after which the level of MBL within each individual is relatively static and there is a small decrease in later life.[5]

The gene encoding MBL, MBL2, is located on chromosome 10q11.2-q21, consisting of 4 exons.[6] Mannose-binding lectin-2 gene polymorphism is very common; variability is greatly influenced by a type of mutation altering complement activation mechanism involved in the opsonization process making bacterial clearance less effective.[7]

Investigation of MBL deficiency can be performed by the quantification of serum MBL or by genotyping for the MBL status.[8] A dilemma with the interpretation of genotyping is that individuals with identical genotypes may differ by 10-fold in MBL levels.[6]

Changes in MBL gene expression associated with low MBL protein levels have been found in healthy individuals commonly and may vary when measured in disease as an acute-phase protein.[9] Emerging studies identified no existing

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association between MBL deficiency and recurrent URTI such as otitis media in children. These data suggest that low MBL concentration does not necessarily indicate a propensity to disease and low levels have to be interpreted alongside clinical features and existing symptoms.[10]

Mannose-binding lectin deficiency is reportedly common, with more than 10% of the general population being MBL-deficient.[8]

The European Society for Immunodeficiencies guideline[11] lists MBL deficiency under complement deficiencies and states that data regarding the clinical impact of MBL deficiency are contradictory and possible effects include susceptibility to bacterial infections and to autoimmunity.

A recent meta-analysis shows that MBL deficiency is associated with death in patients with pneumococcal infection after adjusting other confounders.[12] Neonates with low MBL levels have been deemed to be at higher risk of early-onset sepsis and pneumonia,[13] and children with low MBL levels have often been reported to be at higher risk of severe infections.[14]

Mannose-binding lectin deficiency in children has been reported to lead to decreased opsonizing activity, hence predisposing children to infection.[15] It has been reported to be associated with a severe respiratory infection, due to impaired host response to infective agents.[12,16] Mannose-binding lectin deficiency is especially relevant during the vulnerable period of infancy between 6 and 17 months of age, when maternal antibodies wane and the adaptive immune system is still developing.[17] It has also been associated with more severe lung disease in cystic fibrosis (CF)[18] and also in bronchiectasis not related to CF in adults.[19] In children with bronchiectasis not related to CF, there were no significant differences in growth, annual pulmonary exacerbation rates, pulmonary function tests, radiologic scores, and microbiologic findings between low, intermediate, and high-expressing MBL genotypes over the 1-year follow-up.[20]

However, in another recent study, MBL levels had no association with infection status or with progression from systemic inflammatory response syndrome to sepsis or septic shock in children admitted with a severe or life-threatening illness.[21] It has also been shown that MBL deficiency is not associated with susceptibility to influenza-related critical illness in children.[21] The literature equally reports no link between MBL deficiency and severe lower respiratory tract infection caused by respiratory syncytial virus.[22]

MAIN POINTS

- The current literature is divided about whether mannose-binding lectin (MBL) deficiency predisposes children to having increased infection susceptibility.
- We showed that there are no associations of MBL deficiency with childhood adverse respiratory consequences.
- Children with MBL deficiency and sufficiency showed similar radiological and microbiological characteristics.

So controversy exists as to when MBL deficiency leads to disease. Additionally, there is no conclusive evidence that MBL deficiency is associated with adverse respiratory consequences at follow-up.

The aim of our study was to evaluate any differences in clinical, radiological, and microbiological characteristics in children with MBL deficiency (defined as a level of less than 0.6 mg/L) presenting with troublesome respiratory symptoms (frequent, recurrent, persistent, or very severe), as compared to those who are MBL-sufficient.

MATERIAL AND METHODS

We performed a retrospective cohort study looking at the quantification of serum MBL in children over a period of 10 years from 2004 to 2014 at the Nottingham's Children Hospital.

This study was approved locally as a service evaluation project and ethics approval was not deemed necessary. All the retrospective data collected were anonymized and there was no active intervention for patients; hence, informed consent was not mandated.

We defined a minimum follow-up period of 5 years or more from the time of the MBL measurement.

The inclusion criteria for the study were as follows:

(i) Age less than 18 years and
(ii) Troublesome respiratory symptoms such as

   (a) frequent or persistent respiratory symptoms such as a chronic wet cough lasting more than 4 weeks or
   (b) recurrent lower respiratory tract infections (≥4 infections in a year) or
   (c) severe respiratory tract infections requiring admission to intensive care or to the high dependency unit.

The MBL level was checked as part of baseline immunology testing to exclude a primary immune deficiency.

Mannose-binding lectin deficiency was defined as a serum level <0.6 mg/L. We followed up with a cohort of children with MBL deficiency and compared their respiratory outcomes to those with MBL sufficiency.

The primary outcome was the difference in clinical outcomes in the 2 groups, which included number of admissions to the pediatric intensive care unit (PICU) and responses to childhood immunizations.

We defined a suboptimal response to primary vaccinations based on antibody titers, which were below the laboratory-quoted ranges for *Haemophilus influenzae* B and tetanus. For the pneumococcal conjugate vaccine (Prevenar13), the response was deemed as a suboptimal response when fewer than 8 serotypes were at the desired level.

Secondary outcomes studied were differences in the 2 groups in radiology findings (chest x-ray (CXR) and computed

Table 1. Baseline Characteristics of Study Population

	MBL-Deficient (<0.6 mg/L)	MBL-Sufficient
Number of children	43 (21%)	163 (79%)
Male (%)	22 (51%)	72 (44%)
Median age in years (range)	8 (3.2 to 11)	8 (4 to 11.8)

MBL, mannose-binding lectin.

tomography (CT) of the chest), as well in microbiology findings in respiratory samples for positive viral and bacterial yields.

Statistical Analysis

The data were analyzed using GraphPad Prism 9 software. We tested for normality, and non-normally distributed data were expressed as median and interquartile ranges. The chi-square test was used to compare the follow-up parameters of MBL-deficient and MBL-sufficient children. The odds ratio, the 95% confidence intervals, and the *P*-values were calculated. $P < .05$ was considered statistically significant.

RESULTS

A total of 206 patients undergoing MBL testing were included in the study, out of whom 43 (21%) were MBL-deficient.

The baseline characteristics of the study population are shown below in Table 1.

The clinical, radiological, and microbiological characteristics of the MBL-deficient and MBL-sufficient children are summarized in Table 2. There were no clinical or statistical differences between the 2 groups.

The various follow-up parameters are depicted in Figure 1.

The percentage of children requiring admission to the PICU was small and similar in the 2 groups. Twenty-eight percent of children with MBL deficiency and 33% of those with MBL sufficiency had suboptimal vaccine responses to primary immunizations.

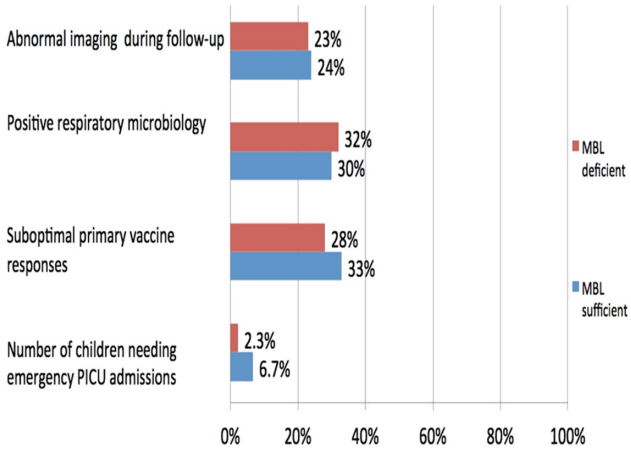

Figure 1. Bar chart showing follow-up features of mannose-binding lectin (MBL)-deficient and MBL-sufficient children.

Thirty-two percent of children with MBL deficiency and 30% of those with MBL sufficiency had positive microbiology (respiratory viruses, bacteria, or both).

Twenty-three percent of children with MBL deficiency and 24% of those with MBL sufficiency had changes on plain radiographs. We performed a subgroup analysis to look at the presence of bronchiectasis in children undergoing a CT chest. Five MBL-deficient children and 10 MBL-sufficient children underwent a CT scan, and the proportion of bronchiectasis was similar in both groups, as shown in Figure 2.

Subgroup Analysis-Booster Immunizations

Our data did not show a difference in the adequacy of response to primary immunizations in the MBL-deficient cohort, compared to the MBL-sufficient cohort. Further subgroup analysis noted that children with MBL deficiency were clinically less likely to respond well to booster immunizations (67% of the MBL-deficient children compared to 40% of MBL-sufficient children), as shown in Figure 3. However,

Table 2. Clinical, Radiological, and Microbiological Parameters over Follow-Up Period

	MBL-Deficient (<0.6 mg/L)	MBL-Sufficient	OR	95% CI	*P*
Number of children needing emergency PICU admissions during follow-up (%)	1 (2.3%)	6 (3.7%)	0.62	0.05 to 4	.66
Suboptimal vaccine responses to primary immunizations, either to Prevenar, *Haemophilus influenzae* B, tetanus, or a combination: N1 (%)	12 (28%)	53 (33%)	0.92	0.43 to 1.97	.83
Suboptimal vaccine responses to booster immunizations: N2/N1 (%)	8/12 (67%)	21/53 (40%)	3.05	0.86 to 9.86	.08
Abnormal imaging at any point during 5-year follow-up (%)	10 (23%)	39 (24%)	0.96	0.45 to 2.16	.96
Bronchiectasis on CT chest	3 out of 5	7 out of 10	0.64	0.08 to 5.21	.64
Any positive respiratory microbiology (respiratory viruses, bacterial pathogens, or both)	14 (32%)	49 (30%)	1.12	0.53 to 2.27	.75

CT, computed tomography; MBL, mannose-binding lectin; OR, odds ratio; PICU, pediatric intensive care unit.

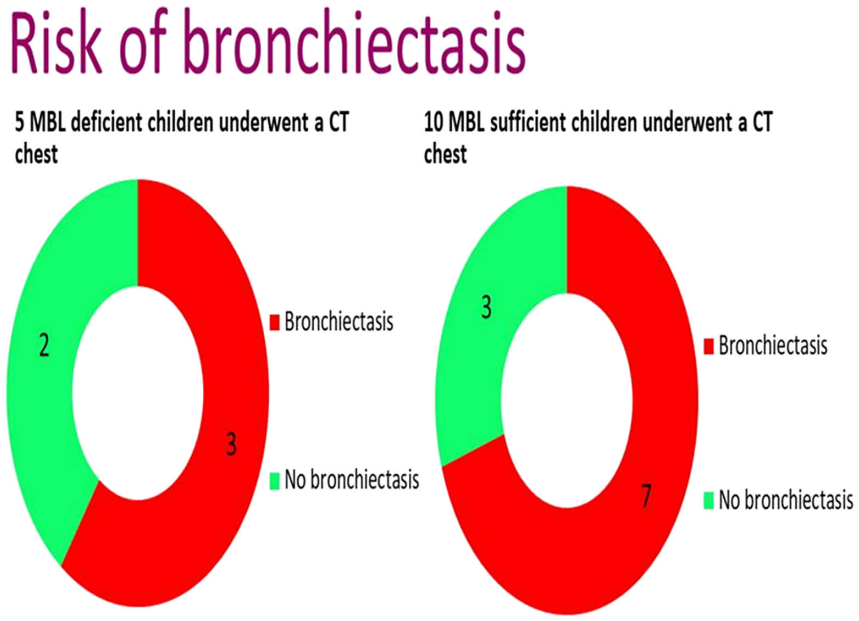

Figure 2. Chart showing risk of bronchiectasis in the 2 groups.

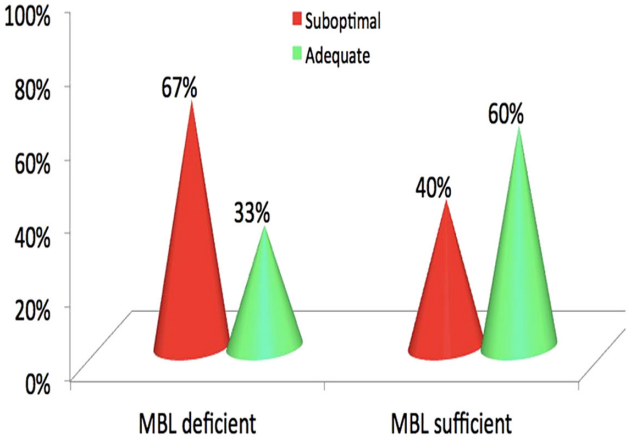

Figure 3. Adequacy of response to booster immunizations in mannose-binding lectin (MBL)-deficient and MBL-sufficient children.

the difference was not statistically significant ($P = 0.08$) due to small numbers.

DISCUSSION

Different studies have explored the relationship between MBL deficiency and increased susceptibility to respiratory infections both in children and adults, with different conclusions.[17,23] Our study did not find adverse clinical, radiological, or microbiological consequences in MBL-deficient children at 5-year follow-up.

Previous studies have shown an association between MBL gene polymorphism and more severe upper respiratory tract infections in children with homozygous mutation of the MBL2 gene over a 2-year follow-up.[24] However, our findings did not reveal a link between MBL deficiency and increased positivity of viral or bacterial respiratory samples.

Even though the literature suggests that children with MBL deficiency are susceptible to infection by specific pathogens such as *Streptococcus pneumoniae*,[25] we did not find an association between MBL deficiency and susceptibility to pneumococcal community-acquired pneumonia and invasive pneumococcal disease, which could have required PICU admissions.

The association between MBL protein levels and recurrent respiratory infections in children remains debatable. Confounders such as atopy, parental smoking, and environmental factors, as well as small sample sizes of the studies, make it difficult to attribute the cause solely to MBL deficiency.

It is increasingly recognized that MBL genetic polymorphisms may have an impact on the serum level and function of the molecule. It has been reported that 3 single nucleotide exonal substitutions in the MBL2 gene lead to a decrease in functional circulating MBL; the structural variant alleles interfere with the assembly of MBL trimers and lead the degradation of the protein. Promoter variants may also influence the expression of MBL, hence impacting MBL serum concentrations.[26]

Low MBL levels, as well as low-producing MBL2 variants, are described to lead to an increased susceptibility to infections and also to worse severity of illness.[27] This is especially relevant when in cases of an existing weak immune system, for instance, for infants and young children and patients with CF, or on immunosuppressive treatment.

We advocate that MBL testing performed as part of baseline immunology testing in children is unnecessary, as the results seem to guide neither the management nor the prognosis. This recommendation would be in line with the recommendations of the recent evidence-based European Respiratory Society (ERS) guidelines[28] and result in more cost-effective practice, given that each MBL level assay costs £24.[29] The International Union of Immunological Societies Expert Committee has in fact not included MBL deficiency in their recently updated classification of inborn errors of immunity.[30]

While our study showed that children with MBL deficiency were more likely to respond inadequately to appropriate booster immunizations, a previous study did not find an association between MBL deficiency and response to pneumococcal vaccination.[31] Equally, MBL polymorphisms have been shown not to affect the production and persistence of antibodies for acellular pertussis.[32] Longer-term prospective studies may shed more light on this area.

The most important strength of the study is the size of the cohort, with the inclusion of a large number of children, and a substantial period of follow-up of 5 years or longer. Our study is hence a robust addition to the existing evidence regarding the specifics of respiratory infections in children with MBL deficiency.

Our study had a number of limitations. It was a retrospective study, which may introduce selection and recall bias. While undertaking the subgroup analyses, our subgroup numbers were relatively low. Also, we did not include a symptom score and hence could not quantify the degree of symptoms that children with MBL deficiency and sufficiency subjectively experienced.

Finally, we defined MBL deficiency on biochemical values, rather than by genetic polymorphisms. Most MBL-disease association studies to date have assessed MBL2 genotypes with or without MBL levels, with MBL insufficient genotypes being XA/O or O/O.[11] This is different to our study, where we looked at serum MBL protein levels, rather than the genotype and ideally both measures could have been included. We note that the literature reports that variant alleles can lead to increased MBL serum levels, but the variant MBL is of a lower molecular weight and is dysfunctional when compared to normal MBL.[26]

CONCLUSION

To conclude, we showed no difference at 5-year follow-up in clinical, radiological, and microbiological characteristics between children who are MBL-deficient, compared to those who have sufficient levels. The rates of admission to PICU were comparable in the 2 groups. About a quarter of the children in both groups had long-term pulmonary changes on the CXR and the prevalence of bronchiectasis was similar. Children with MBL deficiency in our cohort are more likely to respond poorly to appropriate booster immunization, but the significance of this is unclear.

The study found no adverse respiratory consequences with MBL deficiency in childhood. This adds to the existing body of literature that shows no statistically significant association between MBL deficiency and susceptibility to recurrent respiratory tract infection in children.

Ethics Committee Approval: This study was approved locally as a service evaluation project and ethics approval was not deemed necessary.

Informed Consent: All the retrospective data collected were anonymized and there was no active intervention for patients; hence, informed consent was not mandated.

Peer-review: Externally peer-reviewed.

Author Contributions: Design - J.M.B., M.R.; Data Collection - M.R., A.P., J.A.; Data Analysis - M.R.; Writing the Manuscript - M.R., A.P., J.M.B.; Critical Review - M.R., A.P., E.M., L.C., J.M.B.

Declaration of Interests: The authors have no conflict of interest to declare.

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