

Emerging and Established Biomarkers in Pediatric Pneumonia for Diagnostic Prognostic and Therapeutic Implications

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ABSTRACT

Pneumonia is one of the most common causes of morbidity and mortality in children across the globe especially in low and middle income nations. Timely and proper distinction between bacterial and viral etiologies is needed to maximize the treatment, minimize complications, and promote antimicrobial stewardship. Nevertheless, the clinical manifestation and traditional radiographic observations is not specific enough to classify etiologically and predict the severity. Biomarkers have consequently become important supplements in diagnosis and prognosis of pneumonia in children. C-reactive protein (CRP), procalcitonin (PCT) and leukocyte indices are all standard inflammatory markers that have been broadly used in clinical practice. CRP and PCT have moderate diagnostic value, and PCT is more specific to bacterial infection, and can be used to inform antibiotic treatment. The cytokines such as interleukin-6 (IL-6), interleukin-8 (IL-8), and tumor necrosis factor-alpha (TNF-alpha) indicate the severity of the disease and are correlated with the intensive care admission and the risk of complications.

The new biomarkers like presepsin and serum amyloid A (SAA) have potential in enhancing etiological discrimination and prognostic evaluation. Simultaneously, the expression profiling of host genes and molecular pathogen detection with the help of polymerase chain reaction improves the diagnostic sensitivity but should be carefully interpreted because of the colonization and resource restrictions. Even though no individual biomarker can offer unconditional etiological distinction, multi-marker approaches in combination with the clinical assessment and imaging results offer an enhanced predictive efficiency. Further studies in areas of integrated biomarker panels, fast point-of-care diagnostics, and precision-based algorithms are needed to improve the quality of the diagnostic and manage pediatric pneumonia better around the world.

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Introduction

Pneumonia has continued to be a significant cause of morbidity and mortality in young children across the globe especially in low and middle-income nations. The World Health Organization estimates that a high number of children below the age of five years die as a result of lower respiratory tract infections [1]. Even after innovative methods of vaccination, antimicrobial therapy, and community-based health interventions, community-acquired pneumonia (CAP) is still a major clinical and socioeconomic issue in most countries worldwide [2]. The disease is predominant in the vulnerable populations such as the infants, malnourished children and those with underlying comorbid conditions.

Pediatric pneumonia has a heterogeneous etiology. There are still significant causes of severe disease caused by bacterial pathogens (mostly *Streptococcus pneumoniae*) and viral agents (Respiratory syncytial virus and Influenza

virus among others), especially in children younger than five years old [3,4]. Despite the fact that the introduction of pneumococcal conjugate vaccines has dramatically changed the distribution of serotype and lowered the rates of invasive pneumococcal disease, it is still difficult to make a distinction between viral and bacterial pneumonia in the usual clinical practice [5].

It is also essential to diagnose early and perform correct risk stratification to minimize complication, exposure to the inappropriate antimicrobial use, and mortality. In the clinical context, diagnosis mostly depends on clinical manifestations tachypnea, fever, hypoxia, and chest indrawing, in the daily clinical practice. Nevertheless, these signs are delicate signs of lower respiratory tract and lack specificity enough to clearly differentiate the etiology, and they do not accurately indicate disease severity [6]. Radiological data can be used to aid diagnosis though they have low power to differentiate between viral and bacterial etiologies and are prone to interobserver variation [7].

The failure of precise differentiating between etiology of pneumonia has great clinical consequences. False diagnosis of bacterial infection often leads to false prescriptions of antibiotics, which leads to the development of antimicrobial resistance, a perceived global public health crisis [8]. On the other hand, late diagnosis of acute bacterial pneumonia can cause complications that include empyema, bacteremia, systemic inflammation, and sepsis especially in high-risk groups of children [9]. Thus, the increased accuracy of diagnosing is not only important to the outcomes of individual patients, but also to the overall antimicrobial stewardship in general.

In that regard, biomarkers have now become a major point of interest in the study of pediatric infectious diseases. The main aims of biomarker development in childhood pneumonia are the differentiation between bacterial and viral infections, management of disease severity, forecasting of complications (pleural effusion or septic evolution), and treatment response in the long term. The perfect biomarker must be highly sensitive and specific, fast and predictable kinetics, reproducible, cost-effective and applicable in a broad variety of healthcare environments.

Conventional inflammatory biomarkers like C-reactive protein (CRP) and procalcitonin (PCT) are still actively researched and used in clinical settings, but there has been an increasing body of evidence to the point that single-marker approaches do not yield maximum diagnostic efficacy [10]. Newer methods, such as cytokine signatures, host transcriptomic signature, and multi-biomarker panels, with clinical algorithms, have shown encouraging outcomes in terms of etiological classification and severity predictions [11,12]. Such developments are indicative of a wider change to precision medicine in childhood infectious disease.

This is a review of the existing evidence on the existing and the emerging biomarkers in pediatric pneumonia focusing on their diagnostic expression, their prognostic expression and their clinical implementation in the resource endowed and the resource constrained settings.

Traditional Inflammatory Biomarkers

C-Reactive Protein (CRP)

One of the most widely-researched biomarkers in childhood pneumonia is c-reactive protein (CRP). It is an acute-stage reactant produced by hepatocytes in reaction to pro-inflammatory cytokines, mainly interleukin-6 (IL -6) throughout the execution of system infiltration. CRP serum level normally starts to increase 6-12 hours following infection onset and peaks at 24-48 hours, an indication of the intensity of the inflammatory reaction [13].

CRP has been popularly applied in clinical practice to distinguish between bacterial and viral pneumonia. An increase in CRP levels, which is often more than 40-60 mg/L, is usually linked to bacterial etiologies, especially the

infection by *Streptococcus pneumoniae* and other common bacterial pathogens [14]. Conversely, the decreased level of CRP is also more commonly seen in viral pneumonia infections such as those caused by Respiratory syncytial virus and Influenza virus [15]. A number of cohorts have shown that significantly high levels of CRP (>100 mg/L) have been linked to complicated pneumonia, parapneumonic effusion and empyema [16].

Even though it has been widely used, CRP has significant limitations. CRP values of viral and bacterial pneumonia have a significant overlap, especially at the initial stages of the disease. Due to some viral infections, mixed infections or extreme inflammatory reactions, localized CRP levels may be raised, and hence compromise specificity [17]. Moreover, CRP is an inflammatory marker which is non-specific and can be elevated in non-infectious diseases, including autoimmune disease, trauma, or post-operative.

CRP on its own has moderate etiological differentiation diagnostic accuracy in pediatric populations. Nevertheless, it is better performed in combination with clinical scoring scales, radiographic results, or other biomarkers like procalcitonin (PCT) [18]. Thus, CRP should be utilized in a multimodal approach to diagnosis instead of being a single indicator.

Procalcitonin (PCT)

Compared to CRP, procalcitonin (PCT) has also become a better biomarker of bacterial infection. It is a precursor of calcitonin and is synthesized in insignificant levels by thyroid C-cells. However, in the case of the systemic infection with bacteria, PCT is produced in various tissues, as a result of the action of pro-inflammatory mediators and endotoxins of bacteria [19].

PCT concentrations rise quickly, usually 2-4 hours after the beginning of bacterial attack, and reach their maximum faster than CRP, so it potentially could be used to form an early diagnostic decision-making [20]. Significantly, viral infections are likely to inhibit the production of PCT via interferon-action, which leads to increased specificity against bacterial etiologies [21].

Bacterial infection and severe disease have been linked to high levels of PCT in pediatric pneumonia (above 0.5-2.0 ng/mL) of a bacterial infection. Increased PCT levels are associated with bacteraemia, general inflammatory reaction, and the danger of complications [22]. A number of randomized controlled trials have shown that PCT-guided antibiotic therapy can shorten the length of antibiotic therapy and exposure, as well as showing no adverse effects on clinical outcomes, in support of its application in antimicrobial stewardship [23].

However, PCT has its limitations, too. Its price is still significantly above that of CRP testing, which can restrict its common application to low resource locations. Moreover, high levels of PCT can be witnessed during severe viral

infections, systemic inflammation, major traumas, or post operations, and this can give rise to false-positive interpretations [24]. There are also accessibility limitations and differences in cut-off levels among studies which make it hard to be universal.

All in all, PCT is more specific on bacterial pneumonia than CRP and especially useful in the management of antibiotic starting and stopping.

The count of white blood cells (WBC) and Differential.

White blood cell count (WBC) and leukocyte differential are some of the oldest laboratory tests applied in the examination of pediatric pneumonia. Neutrophilic leukocytosis has traditionally been considered to have bacterial etiology, but lymphocytosis has roundly been regarded as indicating viral etiology [25].

Nevertheless, WBC alone is not a very useful diagnostic tool. Viral infections can also lead to the increase in leukocyte count, and certain cases of bacterial infections can also be associated with normal or slightly high WBC level. Additionally, age, stress reaction, dehydration, and exposure to corticosteroids could also affect leukocyte parameters and decrease specificity [26].

The neutrophil to lymphocyte ratio (NLR) is a relatively new inflammatory marker that has received interest recently as being a simple and cost-effective patient value. NLR indicates the ratio of innate (neutrophil-mediated) to the adaptive (lymphocytes-mediated) immune responses. In children with pneumonia, high values of NLR have been linked with the severity of the disease, extended hospital stay, and the inflammatory burden [27]. Despite its potential, NLR still needs to be validated in large and multicentric pediatric cohorts to be able to recommend standardized cut-off values.

Overall, conventional inflammatory biomarkers, such as CRP, PCT, and leukocyte indices, are still essential ingredients of the diagnostic process of a child with pneumonia. None of these markers individually, however, has enough sensitivity and specificity to be used in etiological differentiation. Their best use is in combined diagnostic algorithms, which would involve a synthesis of clinical examination with multi-biomarker examination.

Immunological Biomarkers

Presepsin (sCD14-ST)

Presepsin, or soluble CD14 subtype (sCD14-ST), is a soluble fragment of CD14, a Co-receptor on monocytes and macrophages which is involved in bacterial lipopolysaccharide recognition. In the process of bacterial phagocytosis, CD14 is released into the blood which forms presepsin that is a quantifiable biomarker of innate immune action. Due to its liberation being tightly associated with bacterial pathogen recognition, presepsin has been suggested as a more specific predictor of bacterial infection than conventional acute-phase reactants [28].

As shown by a number of studies in the adult sepsis patients, the level of presepsin is associated with the intensity of systemic infection and dysfunction of the organs, implying that it has prognostic value [29]. In the pediatric care, initial studies suggest that high presepsin levels can help distinguish between bacterial and viral pneumonia and can help identify children who are in danger of developing severe disease or septic complications. Despite these limited data, early evidence is giving presepsin greater specificity in bacterial infections as compared to CRP and equal performance as procalcitonin (PCT) in certain cohorts [30]. Nevertheless, further research studies with large sample sizes are necessary to determine standardized cut-off values and the independent predictive ability of it.

Serum Amyloid A (SAA)

Another protein which is produced in the liver as a response to pro-inflammatory cytokines like IL-1, IL-6 and TNF- α is serum amyloid A (SAA). During acute infection, SAA concentrations rise faster than CRP and are sometimes earlier [31].

The emerging evidence that is emerging in pediatric pneumonia indicates that SAA can be more sensitive than CRP in the early phases of the infection especially in bacterial etiologies. Raised SAA levels have been also linked to increased inflammatory reactions and severity of diseases [32]. However, similarly to CRP, SAA does not possess etiological specificity and it can rise in different inflammatory or non-infectious diseases. Its clinical use can thus be maximized as a combination used in other inflammatory markers.

The expression signature of host genes is associated with immune responses in human tissue infections by yeast infection. Host gene expression is a signature of immune responses in human tissue infections caused by yeast infection.

Developments of transcriptomic based technologies have allowed the identification of host gene expression signatures that could be used to differentiate between viral and bacterial infections. This method does not characterize the pathogen per se but rather the pattern of host immune responses, which identifies the biological signals that represent the etiology.

As an illustration, gene expression triggered by interferon (IFN)-stimulation is generally increased during viral infections, and bacterial infections are related to neutrophil activation and inflammatory pathway gene signatures [33]. It has also been shown that multi-gene classifiers are able to achieve high sensitivity and specificity in the distinction of bacterial and viral respiratory infections in children [34].

Even though transcriptomic profiling is a potentially valuable precision-medicine method, its use is mostly restricted to research contexts because of its cost, technological complexity and a lack of fast point-of-care platforms. The simplification into small gene panels or even

simplified and quick molecular tests in the future could help enable more clinical usage.

Lactate

Serum lactate is a proven biomarker of a tissue hypoxia and poor perfusion. An increase in the lactate is indicative of an anaerobic metabolism and is usually associated with serious infections such as septic shock.

Hyperlactatemia has been linked with the severe disease, respiratory failure, systemic inflammatory response, and high mortality risk in pediatric pneumonia [30]. The high levels of lactate can also be an early warning of the occurrence of occult hypoperfusion before an overt hypotension is observed, and thus lactate is a useful risk stratification tool in the emergency and intensive care unit [21].

Despite the fact that lactate fails to distinguish viral and bacterial etiology, it is a valuable prognostic factor which can be used to determine whether intensive monitoring and aggressive supportive treatment should be administered.

Severity and Prognosis Biomarkers.

In addition to etiological distinction, there are a number of biomarkers that have been strongly correlated with disease severity and negative clinical outcomes in childhood pneumonia. High levels of PCT have been continually linked to severe bacterial pneumonia and systemic involvement. The elevated concentrations of interleukin-6 (IL-6) are associated with the necessity to be admitted to the intensive care unit (ICU) and the abundance of inflammatory load. Excessive levels of lactate are indicative of the risk of mortality and the development of septic shock. On the same note, significantly high levels of CRP (>100 mg/L) have also been associated with complicated pneumonia such as empyema and pleural effusion [16,22,25].

Notably, the predictive power of combining biomarkers is better than when using single-markers. Multi-biomarker panels Multivariate models combining CRP, PCT, cytokines and clinical variables form better discrimination of severe outcomes, and the approach of multi-biomarker panels is supported in the management of pediatric pneumonia [31].

Correlation of Biomarkers based on Imaging.

The biomarker profile is often related to radiological findings. Lobar consolidation in the radiograph of the chest-usually typical of bacterial pneumonia- is likely to relate to increased CRP and PCT levels. Interstitial infiltrates, which occur more often in viral infections, are in contrast linked to reduced concentrations of inflammatory biomarkers [27].

Even though the use of imaging cannot be considered a reliable etiology determinant, radiographic pattern integration with data on biomarkers can contribute to an improved diagnostic confidence and inform the choice of treatment.

Biomarkers and antibiotic Stewardship.

Biomarkers are important in antimicrobial stewardship measures. It has been demonstrated that PCT-informed antibiotic algorithms shorten the length of antibiotic treatment and overall exposure to antibiotics in children with lower respiratory tract infection and do not raise the rate of treatment failure and adverse events [23]. On the same note, serial CRP testing can help to measure treatment progress and establish when the antibiotics should be stopped.

Randomised clinical trials and meta-analyses also suggest that the biomarker-based strategies can minimise unneeded antibiotic prescriptions, thus helping diminish antimicrobial resistance [38]. The strategies have been specifically applicable in pediatric population, where the frequency of antibiotic overuse is still high.

Weaknesses in Existing studies.

Regardless of promising evidence, there are a number of obstacles to the broad clinical use of emerging biomarkers. Research usually includes heterogeneous groups of people with differences in age scale, disease severity, and etiological distributions, making it difficult to compare them directly. There are no standardized cut-off values of most of the biomarkers, and the variability of the assays also adds to the inconsistency [22].

Also, the majority of developed biomarker research is based in high-income nations and there is little information available on low- and middle-income areas (where the burden of pneumonia is the highest). A large amount of intercession between viral and bacterial biomarker patterns remains, particularly when there is co-infection [12].

The future studies need to focus on large, multicenter trials in children and the creation of multi-marker panels coupled with machine learning-based predictive models that can enhance the accuracy of the diagnosis and generalizability.

Future Directions

Recent studies have focused on the need to combine multi-omics methods such as proteomics and metabolomics to discover new diagnostic and prognostic biomarkers. Innovations in rapid point-of-care molecular diagnostics with the ability to identify both host-response markers and pathogens simultaneously are also an important direction of innovation.

These potentially include artificial intelligence (AI)-powered biomarker panels, which are a combination of laboratory data, clinical parameters, and imaging functions, which are capable of real-time risk stratification and tailored treatment algorithms [34]. Combination of biomarkers and established clinical severity measures can lead to increased diagnostic accuracy, better antibiotic stewardship and better therapeutic outcomes.

Conclusions

The biomarkers are important in enhancing the accuracy of diagnosis, assessment of the severity, and optimization

of treatments in pediatric pneumonia. Conventional markers like CRP and PCT are the most clinically defined to be used in normal practice. Cytokine profiling and host gene expression signature are attractive areas of research that have the potential to offer specific diagnostics.

Nevertheless, a single biomarker is not yet sensitive and specific enough to be used as a definite etiological classifier. Multi-marker approaches, coupled with clinical examination, and imaging outcomes have better predictive capability than single-marker roles.

The further exploration of the host-response signatures, economical point-of-care devices, and integrative diagnostic algorithms is the key to improve the management of pneumonia in children worldwide and decrease the morbidity and mortality.

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