Original Research Article

Do Lung surfactant proteins play a role in severity of bronchiolitis

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A B S T R A C T

Background: Bronchiolitis is a common respiratory viral infection in children under 2 years of age. The Surfactant Protein A and D (SP A and SP D) are collagen containing lectin (collectins) help in controlling pulmonary infections, inflammations; and allergies. Thus, these collectins might play a major role in severe bronchiolitis.

Materials and Methods: Thirty (n=30) severe and thirty (n=30) non-severe clinically diagnosed bronchiolitis infants with age and gender matched were recruited. The serum levels of SP A and SP D was quantified using ELISA.

Results: The SP A levels were significantly reduced in severe cases (0.16ng/L ± 0.0028) compared to non-severe cases (0.17ng/L ± 0.0009) (p <0.0001) and SP D levels were significantly higher in severe cases (0.12ng/ml ± 0.001) compared to non-severe cases (0.10ng/ml ± 0.043) (p <0.05).

Conclusion: Reduced level of SP-A and the elevated level of SP-D was associated with severe pulmonary inflammation and infection in bronchiolitis. Thus variation in levels of SP-A and SP D might play a major role in the pathogenesis of severe bronchiolitis.

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1. Introduction

Bronchiolitis is the most frequent cause of lower respiratory tract illness and hospitalization in young infants.¹ It is a viral inflammatory condition of bronchioles mostly in children below two years of age and is characterized by progressive tachypnea, nasal flaring, intercostal, subcostal, & sub sternal retractions leading to severe breathing difficulty.²,³ RSV accounts for 63% of illness; whereas other viruses i.e. human metapneumovirus, influenza virus, Para influenza virus, adenovirus, rhinovirus coronavirus, and bocavirus are responsible in many a cases.⁴

Pulmonary surfactant is a multimolecular complex consisting of phospholipids and cholesterol (total 90%) and surfactant proteins (SP-A, SP-B, SP-C and SP-D) (10%) which forms a lattice called tubular myelin that covers the alveolar surface.⁵ Initially, these proteins were known for their traditional role in reducing surface tension at the air-liquid interface, preventing alveolar collapse during expiration of the lung. The hydrophilic protein SP-A and SP-D belong to collagenous and superfamily of C-type lectin binding domains associated with pulmonary homeostasis and innate immunity.⁶–⁸ The SP-A and SP-D function as pattern recognition molecules; often bind using their carbohydrate recognition domain (CRD) to oligosaccharides on the surface of microorganisms to promote phagocytosis. The immune action of SP-A and SP-D includes induction of pathogens agglutination, preventing pathogens adherence to the cell surface, balancing the activation and inhibition of infectious colonization and invasion. It also activates the lung inflammatory process, regulates the release of oxygen radicals, pro and anti-inflammatory cytokines.⁷ They have been postulated to play a role in bronchiolitis.⁸–¹³ The aim of this study was to evaluate the significance of the levels of SP-A and SP-D in cases of bronchiolitis.

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Table 1: Bronchiolitis Severity Score

<table>
<thead>
<tr>
<th>JLh0</th>
<th>Items</th>
<th>Score 1</th>
<th>Score 2</th>
<th>Score 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Respiratory Rate</td>
<td>&lt;60</td>
<td>61-69</td>
<td>&gt;70</td>
</tr>
<tr>
<td>2</td>
<td>Cyanosis</td>
<td>Normal</td>
<td>Peripheral</td>
<td>central</td>
</tr>
<tr>
<td>3</td>
<td>SPO2</td>
<td>&gt;93-96%</td>
<td>90-93%</td>
<td>&lt;90%</td>
</tr>
<tr>
<td>4</td>
<td>Sensorium</td>
<td>Mild irritable but Easy to console</td>
<td>Difficulty to console</td>
<td>Lethargy/Drowsy</td>
</tr>
<tr>
<td>5</td>
<td>Nasal flare/ Retraction (SC, IC, SCS)</td>
<td>1 of 4</td>
<td>2 of 4</td>
<td>3 or more</td>
</tr>
<tr>
<td>6</td>
<td>Feeding</td>
<td>Normal</td>
<td>&gt;50%</td>
<td>&lt;50%</td>
</tr>
<tr>
<td>7</td>
<td>Air entry/Wheeze</td>
<td>Normal, Scattered Rhonchi, crepitation end expiratory</td>
<td>Fair, Rhonchi, Rales in both inspiration and expiration</td>
<td>Poor, Grunt, Rhonchi and Crepitation</td>
</tr>
</tbody>
</table>

Mild: < 7  
Moderate: 8-14  
Severe: > 14

Table 2: Serum levels of Surfactant Protein A & D.

<table>
<thead>
<tr>
<th>Case (Mean ±SD)</th>
<th>Control (Mean ±SD)</th>
<th>P value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>SP - A 0.16 ± 0.0028</td>
<td>0.17 ± 0.0009</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>SP - D 0.12 ± 0.001</td>
<td>0.10 ± 0.043</td>
<td>0.046</td>
</tr>
</tbody>
</table>

*p<0.05 is consider as significant

2. Materials and Methods

Infants under 2 years of age with bronchiolitis were recruited in pediatrics casualty and outpatient department of JIPMER, Puducherry. Consecutive infants diagnosed as bronchiolitis were categorized into non-severe (n=30) and severe (n=30) bronchiolitis using Bronchiolitis Severity score\textsuperscript{14,15} (Supplementary Table 1) as controls and cases respectively after obtaining informed consent with age and gender matched. All the samples were collected on the 2nd or 3rd day of illness.

2.1. Quantifying Serum SP-A and SP-D levels

One milliliter (1ml) blood was collected, centrifuged at 3000rpm for 10mins and separated serum was stored at -40°C. The sandwich ELISA kit (SP-A - E3802Hu and SP-D – E3597Hu) from Bioassay Technology Laboratory was used for accurate quantification of serum levels of SP-A and SP-D as per the manual instruction.

2.2. Ethics

This study was approved by Institute Ethics Committee (Human studies) Ref No: JIP/IEC/2015/13/544. Written informed consent was obtained from all the parents before participation.

2.3. Statistical analysis

Analyses were performed using Graphpad prism version 6. Variables like age and weight were expressed in mean and other variables were described in frequency and percentage. Un-paired t-test was used to compare the levels of SP-A and SP D in case and control. The P value <0.05 was consider as significant.

3. Results

The mean age in both groups was 5 months (1 month – 10 months) and each group has 20 male and 10 female infants with the mean weight of 5.6 ± 1.3 and 5.75 ± 1.9Kg in control and case groups respectively. In Case group 70% belong to joint family, 50% lies in upper lower class status, 84% live in rural areas whereas 57% belong to joint family, 50% lies in lower middle class status, 74% live in rural areas in control group.

The SP A levels were significantly reduced in severe cases (0.16ng/L ± 0.0028) compared to non-severe cases (0.17ng/L ± 0.0009) (p <0.0001) and SP D levels were significantly higher in severe cases (0.12ng/ml ± 0.001) compared to non-severe cases (0.10ng/ml ± 0.043) (p <0.05) (Table 2). The levels of SP-A and SP-D in different viruses.
4. Discussion

In this study the mean serum SP-A level was significantly reduced whereas the mean serum SP-D level was significantly higher in severe cases compared to the controls irrespective of the infecting virus. Synthesis of surfactant protein by type II pneumocytes is reduced due to the viral invasion. At the same time the SP- A binds to viruses and opsonin by alveolar macrophages for phagocytosis and viral clearance. It is also associated with reduced immunity and increased severity. This is similar to the conclusion drawn from other studies across the world.

The elevated levels of SP-D in severe cases in our study have also been observed in studies conducted in Japan and Korea. However the result also disagrees with some Scottish and Brazilian studies. Pulmonary SP- D concentrations may vary during the progress of viral infection. It may rise in the early viral infection to enhance D concentrations may vary during the progress of viral infection in bronchiolitis. Thus variation in levels of SP-A and SP-D might play a major role in the pathogenesis of severe bronchiolitis.

5. Conclusion

Reduced level of SP-A and the elevated level of SP-D was associated with severe pulmonary inflammation and infection in bronchiolitis. Thus variation in levels of SP-A and SP-D might play a major role in the pathogenesis of severe bronchiolitis.

6. Conflicts of Interest

All contributing authors declare no conflicts of interest.

7. Source of Funding

None.

References


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