Phosphatidyglycerol in tracheal aspirates for diagnosis of hyaline membrane disease

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**SUMMARY** Lecithin:sphingomyelin ratio and phosphatidylglycerol were determined by a rapid, simple method in tracheal aspirates obtained from 132 newborn infants with respiratory diseases, sixty-five of whom developed hyaline membrane disease. Phosphatidylglycerol determination was more sensitive (97%) than lecithin:sphingomyelin ratio, but their specificities were similar (76%).

A deficiency in the pulmonary surfactant is believed to cause hyaline membrane disease in newborn infants. Pulmonary maturity has been assessed by the analysis of the phospholipids in the hypopharyngeal and tracheal aspirates of newborn infants. The most widely used test is the lecithin:sphingomyelin ratio described by Gluck et al. but this method is time consuming and affected by contamination of the specimen with blood or meconium.

As a consequence, several investigators have suggested that other phospholipids from surfactant and particularly phosphatidylglycerol (PG) should be studied. PG is synthesised almost exclusively by lung alveolar cells so that there is no interference in assays by blood or meconium. Both one and two dimensional thin layer chromatography are commonly used to analyse PG. These methods are time consuming, however, and require extensive laboratory technician training.

In 1983, Garite et al described a new, rapid, simple, and reliable method to determine PG in amniotic fluid by a semiquantitative immunological slide agglutination test. A complete assay can be performed in about 30 minutes.

We have applied and compared this test to the lecithin:sphingomyelin in the tracheal aspirate.

**Patients and methods**

Tracheal aspirates were obtained from 132 infants who were admitted to the intensive care unit for respiratory disease. Of these, 65 were diagnosed as having hyaline membrane disease after satisfying clinical and x-ray criteria. The other 67 infants had other respiratory diseases, namely, transient tachypnoea of the newborn (30 cases), congenital pneumonia (six), amniotic fluid aspiration syndrome (11), and other respiratory diseases (20). The gestational ages of the infants ranged from 26 to 42 weeks (mean 33 weeks) and their birth weights from 645 to 4700 g (mean 2160 g). Samples were obtained during the first 24 hours after birth.

The lecithin:sphingomyelin ratio was determined using the method of Gluck et al and PG testing by Amniostat-FLM were performed on all samples. A lecithin:sphingomyelin ratio ≥ 2.0 and a PG ≥ + + or 2 μg/ml were interpreted as being 'positive' and as indicating fetal lung maturity and a lecithin:sphingomyelin ratio < 2 and PG < + + were interpreted as being 'negative'.

**Results**

The Table shows the results of the lecithin:sphingomyelin ratio and phosphatidylglycerol in the tracheal aspirates.
gomyelin and PG determination in tracheal aspirates of the 132 newborn infants with respiratory diseases.

For 59 of the 65 newborn infants (91%) who developed hyaline membrane disease, the two tests correlated well with the criteria for hyaline membrane disease, but for six (9%) the results showed no correlation. In four of these six cases the lecithin:sphingomyelin ratio was ≥2 but the PG was absent, and for the other two cases both tests indicated fetal lung maturity.

For the 67 newborn infants who did not have hyaline membrane disease, both biochemical tests predicted pulmonary maturity for 43 of them (64%), but for the 24 others (36%) the results showed no correlation. This lack of correlation was particularly noticeable in these cases with other respiratory diseases (14 of 20), particularly when their gestational ages were less than 30 weeks (80% of the cases). The lack of correlation was not so pronounced, however, in the cases with transient tachypnoea (five of 30), congenital pneumonia (two of six), and amniotic fluid aspiration syndrome (three of 11).

### Discussion

In most cases (102 of 132 (77%)) a good correlation was observed between the results of laboratory tests and the clinical and x-ray criteria for hyaline membrane disease.

The correlation was better, however, for the newborn infants who had hyaline membrane disease (91%). James et al. reported that for babies who did not have hyaline membrane disease, and in particular transient tachypnoea, congenital pneumonia, or amniotic fluid syndrome, the determination of phospholipids in tracheal or gastric aspiration was of doubtful importance. In our study this was observed for other respiratory diseases, in particular when the gestational age of the newborn was very low, but not for transient tachypnoea, congenital pneumonia, or amniotic fluid aspiration syndrome.

For the results that did not correlate (23%), in 11 infants (8%) both biochemical tests differed from the clinical diagnosis. The specificities of lecithin: sphingomyelin ratio and PG determination were similar (76% and 77%, respectively). In 19 cases (14%) there was no correlation between the two tests, but the PG determination was more sensitive (97%) than the lecithin:sphingomyelin ratio (88%) and the results were unaffected by blood or meconium contamination.

In conclusion, the high sensitivity, specificity, rapidity, and simplicity of this assay makes PG determination a useful method for assessing the presence of PG in tracheal aspirates in newborn infants for diagnosis of hyaline membrane disease.

### Table: Determination of lecithin:sphingomyelin (L:S) ratio and phosphatidylglycerol (PG) in tracheal aspirates obtained from 132 newborn infants with respiratory diseases

<table>
<thead>
<tr>
<th>Group</th>
<th>n=</th>
<th>L:S and PG negative</th>
<th>L:S and PG positive</th>
<th>L:S negative and PG positive</th>
<th>L:S positive and PG negative</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infants with hyaline membrane disease</td>
<td>65</td>
<td>59</td>
<td>2</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Infants without hyaline membrane disease</td>
<td>67</td>
<td>9</td>
<td>43</td>
<td>7</td>
<td>8</td>
</tr>
<tr>
<td>Transient tachypnoea</td>
<td>30</td>
<td>2</td>
<td>25</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Congenital pneumonia</td>
<td>6</td>
<td>1</td>
<td>4</td>
<td>—</td>
<td>1</td>
</tr>
<tr>
<td>Amniotic fluid aspiration syndrome</td>
<td>11</td>
<td>1</td>
<td>8</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Other respiratory diseases</td>
<td>20</td>
<td>5</td>
<td>6</td>
<td>4</td>
<td>5</td>
</tr>
</tbody>
</table>

References


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Received 26 August 1986
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Arch Dis Child 1987 62: 193-194
doi: 10.1136/adc.62.2.193