



(PCR)

600 µl 100% ethanol 3 M sodium acetate 30 µl  
 . 4 12,000 rpm 20  
 DNA 80% cold ethanol  
 20 µl dH<sub>2</sub>O DNA .

*H. influ - enzae*

.<sup>7)</sup>

.<sup>7)8)</sup>

(PCR) PCR premix( ( )  
 primer . *H. in -*  
*fluenzae* P6  
 , *S. pneumoniae* PBP 2P , *M. catarrhalis*  
 M46 clone (Table 1).<sup>6)</sup>  
 DNA thermal cycler(MJ  
 Research, INC, U.S.A) 94 (dena -  
 turation), 50 (annealing), 72 (poly -  
 merization) 3 40 .  
 (PCR) nest primer  
 ( *H. influenzae*, *S. pneumoniae*,  
*M. catarrhalis*)

nest PCR

5 µl 2% agarose gel  
 120 volt, 20 0.5% Ethi -  
 dium bromide  
 Chi - square

1998 5 1999 2

52

8

3 8 5.6 ,  
 1 : 1.6 (Table 2).

bottle

70

collector 30 /52 (57.7%), 24  
 /52 (46.2%) . *H. influenzae*

DNA

DNA

lysis coc -

tail(10 mM Tris - HCl ; 5 mM EDTA ; 0.5% SDS ; 0.2  
 mg/ml proteinase K)

37

300 µl

phenol : chloroform : isoamyl alcohol(25 : 24 : 1)

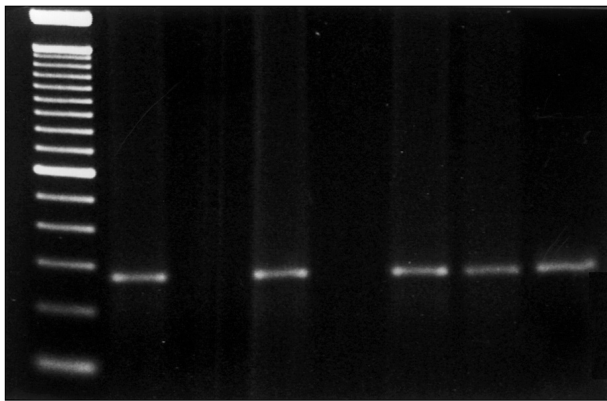
12,000 rpm 15

**Table 1.** Primers used for the specific detection of bacterial pathogens

<i>Haemophilus influenzae</i> from the P6 gene	
5` primer :	ACT TTT GGC GGT TAC TCT AT
3` primer :	TGT GCC TAA TTT ACC AGC AT
<i>Streptococcus pneumoniae</i> from the PBP2P gene	
5` primer :	GAT CCT CTA AAT GAT TCT CAG GTG G
3` primer :	ACT ATA GAA GAA AGG GAA GTT TCC A
<i>Moraxella catarrhalis</i> from the M46 clone	
5` primer :	TTG GCT TGT GCT AAA ATA TC
3` primer :	GTC ATC GCT ATC ATT CAC CT

**Table 2.** Age and sex distribution of patients

Age	Sex		Total
	Male	Female	
3	4	4	8
4	4	8	12
5	11	10	21
6	3	2	5
7	2	2	4
8	0	2	2
Total	24	28	52



**Fig. 1.** Electrophoresis of *H. influenzae* by PCR. Left lane ; 100 bp marker. Lane 1 - 3 ; PCR product from effusion. Lane 4 - 6 ; PCR product from adenoid tissue. Lane 7 ; positive control. Positive bands are shown in lane 1, 3, 5, and 6.

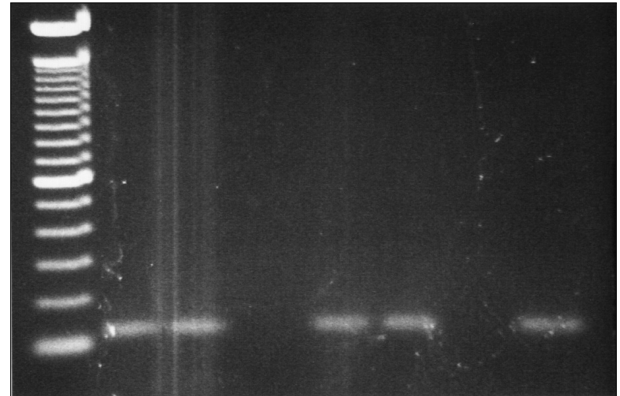
**Table 3.** Detection of *H. influenzae* in OME and adenoids

<i>H. influenzae</i>	Effusions		Total
	+	-	
Adenoids	7	12	19
	9	24	33
Total	16	36	52

( $p > 0.05$ ) + : Number of detected samples  
- : Number of non-detected samples

16 / 52 (30.8%),  
7 / 52 (13.5%)  
10 / 52 (19.2%)  
8 / 52 (15.4%)  
6 / 52 (11.5%)  
7 / 52 (13.5%)

*H. influenzae*  
7 ,  
21 ,



**Fig. 2.** Electrophoresis of *S. pneumoniae* by PCR. Left lane ; 100 bp marker. Lane 1 - 3 ; PCR product from effusion. Lane 4 - 6 ; PCR product from adenoid tissue. Lane 7 ; positive control. Positive bands are shown in lane 1, 2, 4, and 5.

**Table 4.** Detection of *S. pneumoniae* in OME and adenoids.

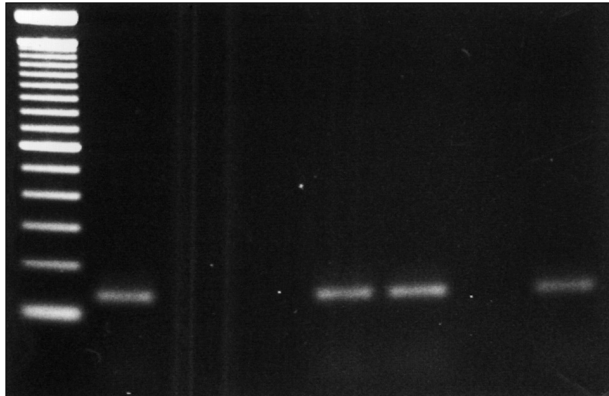
<i>S. pneumoniae</i>	Effusions		Total
	+	-	
Adenoids	4	4	8
	6	38	44
Total	10	42	52

( $p > 0.05$ ) + : Number of detected samples  
- : Number of non-detected samples

24  
13.5%  
*H. influenzae*  
( $p > 0.05$ )(Fig. 1, Table 3).  
*S. pneumoniae*  
4 ,  
10 ,  
38  
7.7%  
*S. pneumoniae*  
( $p > 0.05$ )  
(Fig. 2, Table 4).  
*M. catarrhalis*  
2 ,  
9 ,  
41  
3.8%  
*M. catarrhalis*  
( $p > 0.05$ )(Fig. 3, Table 5).

(otitis media with effusion)

(PCR)



**Fig. 3.** Electrophoresis of *M. catarrhalis* by PCR. Left lane ; 100 bp marker. Lane 1 - 3 ; PCR product from effusion. Lane 4 - 6 ; PCR product from adenoid tissue. Lane 7 ; positive control. Positive bands are shown in lane 1, 4, and 5.

**Table 5.** Detection of *M. catarrhalis* in OME and adenoids

<i>M. catarrhalis</i>	Effusions		
	+	-	Total
Adenoids	2	5	7
	4	41	45
Total	6	46	52

( $p > 0.05$ ) + : Number of detected samples  
 - : Number of non-detected samples

가  
 9-11) 1) 12)13)  
 3가  
 가  
 3가 ,  
 (reservoir) 14)15)  
*H. influenzae*, *S. pneumoniae*, *M. catarrhalis*  
 8  
 20%, 4 10% 2 8% 4)  
*H. in-*  
*fluenzae*가 30 64% 9)

6) 가  
 (lysozyme) 18)  
 가 19)  
 2/3 4)  
 가 (me -  
 tabolically activity)  
 가 GA - PDH  
 mRNA - (reverse  
 transcriptase - polymerase chain reaction)  
 가  
 20)  
*H. influenzae*  
 50%, 80%  
 가 5)21)  
*H. influenzae*가 61%, *S. pneumoniae*가  
 13% 22)  
*H. influenzae* 30.8%, *S. pneumoniae*  
 19.2%, *M. catarrhalis* 11.5%  
*H. influenzae*가 36.5%, *S. pneumoniae* 15.4%,  
*M. catarrhalis* 13.5% ,  
 57.7% 46.2%  
 Bernstein  
*H. influenzae*가 25.4%  
 19.7%  
*H. influenzae* 8)  
 가 가

가 가  
가 가

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