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THE SIGNIFICANCE OF SPUTUM QUALITY IN THE DIAGNOSIS OF PNEUMOCOCCAL COMMUNITY-ACQUIRED PNEUMONIA (PNCCAP) IN THE ELDERLY BY CULTURE

A. Saukkoriipi[1], A.A. Palmu[2], J. Jokinen[3], T.M. Kilpi[3]

Department of Public Health Solutions, National Institute for Health and Welfare (THL), [1]Oulu, [2]Tampere, and [3]Helsinki, Finland

Contact: annika.saukkoriipi@thl.fi

Background and aims

- The value of sputum culture in the microbiological diagnosis of community-acquired pneumonia (CAP) is controversial. Expectorated sputum is the most readily available specimen, but upper respiratory secretions can contaminate the specimen during the collection process. Potential pathogens of the oropharyngeal flora may give a false positive result in sputum culture or bacteria of the oropharyngeal flora may overgrow pathogens of the lower respiratory tract resulting in a false negative result.
- The degree of contamination of a sputum specimen by oropharyngeal flora is assessed by microscopic screening of a Gram-stained smear of the specimen. If low numbers of squamous epithelial cells and high numbers of polymorphonuclear cells are found, the sample is considered to be representative of the lower respiratory tract, i.e. a high-quality (HQ) sample.
- Generally, sputum culture in the microbiological diagnosis of CAP has been considered valid only if a HQ sample has been obtained. However, evidence of the role of sputum quality regarding evaluation of pneumococcal etiology specifically is lacking.
- We evaluated the relevance of sputum quality assessment for sputum culture in the diagnosis of pneumococcal CAP (PncCAP).

Methods

- We studied patients aged ≥ 65 years with radiologically confirmed CAP and participating in the Finnish CAP Epidemiological study (FinCAP Epi) in 2005–07 (N=323).[1,2] The results of sputum culture for encapsulated pneumococcus (Pnc) were compared with the following pneumococcal tests:
  - Blood culture for encapsulated Pnc
  - Binax NOW® Pnc urine antigen test
  - At least a 2-fold increase in serum antibodies to Pnc surface adhesin A or Choline binding protein A
  - Culture of encapsulated Pnc from nasopharyngeal swab
- Purulent parts of the sputum samples were Gram-stained and quality was assessed microscopically (400-fold magnification). Two sets of quality criteria were applied to delineate HQ from low-quality (LQ) sputa:
  - leukocytes/epithelial cells ratio >5 and ≤2.5 epithelial cells per field (HQ1) or
  - leukocytes/epithelial cells ratio >1 (HQ2).

Results

- A sputum sample was obtained and the quality assessed in 224 (69%) of the CAP cases; 47% were HQ1 (33% of all CAP cases) and 76% HQ2 (53%).

Table. Performance of culture for encapsulated pneumococcus from high-quality and low-quality sputa compared to other pneumococcal diagnostic tests among 224 radiologically confirmed community-acquired pneumonia cases.

Pneumococcal diagnostic test		N	No of cases with encapsulated Pnc cultured from sputum/No of cases analysed (%)			
			HQ1 sputum: leukocytes/epithelial cells >5 AND epithelial cells ≤2.5 per field		HQ2 sputum: leukocytes/epithelial cells >1	
			HQ1, N = 106	LQ1, N = 118	HQ2, N = 170	LQ2, N = 54
Blood culture for encapsulated Pnc	-	214	24/103 (23)	11/111 (10)	32/163 (20)	3/51 (6)
	+	7	1/2 (50)	3/5 (60)	3/5 (60)	1/2 (50)
Pnc urine antigen test	-	174	18/86 (21)	6/88 (7)	23/136 (17)	1/38 (3)
	+	21	3/7 (43)	5/14 (36)	7/14 (50)	1/7 (14)
2-fold increase in PsaA or CbpA anti-bodies in paired sera*	-	161	18/85 (21)	5/76 (7)	22/126 (17)	1/35 (3)
	+	30	6/14 (43)	7/16 (44)	11/24 (46)	2/6 (33)
At least 1 of the above 3	-	184	18/89 **(20)**	5/95 **(5)**	21/139 **(15)**	2/45 **(4)**
	+	40	7/17 **(41)**	9/23 **(39)**	14/31 **(45)**	2/9 **(22)**
NPS culture for encapsulated Pnc	-	190	13/89 (15)	3/101 (3)	16/144 (11)	0/46 (0)
	+	27	12/14 (86)	10/13 (77)	19/22 (86)	3/5 (60)

Pnc, pneumococcus; HQ, high-quality; LQ, low-quality, not high-quality according to the criterion/criteria defined in the column; PsaA, Pnc surface adhesin A; CbpA, Choline binding protein A; NPS, nasopharyngeal swab.
*a serum sample at the acute visit and a convalescent sample 4 – 8 weeks later

- Encapsulated Pnc were cultured from 25 (24%) and 14 (12%) of the HQ1 and LQ1 samples (P=0.02), respectively, and from 35 (21%) and 4 (7%) of the HQ2 and LQ2 samples (P=0.03), respectively.
- If another pneumococcal test was positive, encapsulated Pnc were cultured at similar proportions in LQ1 and HQ1 sputa (for example **39%** vs. **41%**; Table); if the other test was negative, encapsulated Pnc were cultured less often in LQ1 than HQ1 sputa (**5%** vs. **20%**; Table).
- Regardless of the result of the other pneumococcal test, encapsulated Pnc was found less often in LQ2 than in HQ2 sputa (for example **22%** vs. **45%** when other test(s) positive and **4%** vs.**15%** when negative; Table).

Conclusions

- Stringent criteria for HQ sputum reduces the yield of valid sputum samples considerably.
- Positive cultures for encapsulated Pnc from HQ1 and LQ1 sputa showed similar concordance with other pneumococcal tests if the other test was positive. The less stringent HQ2 criterion resulted in equal concordance with other tests suggesting equal sensitivity.
- When other pneumococcal tests were negative, the LQ sputum samples were more commonly negative than the HQ samples suggesting low sensitivity rather than low specificity in the demonstration of pneumococcal etiology of CAP.

References:
[1]Palmu AA et al. (2014) Incidence and etiology of community-acquired pneumonia in the elderly in a prospective population-based study, Scand J Infect Dis (46) 250-259;
[2]Jokinen J et al. Testing pneumonia vaccines in the elderly: Determining a case definition for pneumococcal pneumonia in the absence of a gold standard, Am J Epidemiol, 2017 Dec 15, doi: 10.1093/aje/kwx373. Epub ahead of print.

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