

Transthoracic fine-needle aspiration in the aetiological diagnosis of community-acquired pneumonia

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Abstract

To investigate the safety and practicability of conducting transthoracic fine-needle aspiration (TFNA) in a general hospital setting, we applied the TFNA procedure to 20 patients hospitalized with community-acquired pneumonia (CAP) within 36 h of admission. Also, a preliminary assessment was made of the potential value of adding TFNA to conventional methods of diagnostic microbiology. TFNA was easy to perform and caused little discomfort, and no serious adverse events were observed. In spite of ongoing antimicrobial treatment, a likely aetiological diagnosis was established for 14 of 20 (70%) of the patients. TFNA may provide important additional information on the aetiology of CAP.

Keywords: Aetiological diagnosis, aspiration, CAP, community-acquired pneumonia, fine-needle, pneumococcus

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The main aims of this pilot study were to investigate the safety and practicability of conducting transthoracic fine-needle aspiration (TFNA) to reach the aetiological diagnosis of community-acquired pneumonia (CAP) in a general hospital setting. Also, a preliminary assessment was made of the diagnostic value of adding TFNA to conventional methods of clinical microbiology. We conducted TFNA under fluoroscopic guidance on 20 CAP patients (mean age 57 years, range 21–80 years). TFNA was performed under sterile conditions, using local anaesthesia. A 22-gauge needle, stylet removed, was attached to a 10-mL syringe containing 5 mL of isotonic saline; under fluoroscopic guidance, the needle was introduced into the pulmonary infiltrate. Four millilitres of the saline were injected into the inflamed area, leaving approximately 1 mL as carrier fluid in the syringe. Negative pressure was applied while the needle was slowly retracted. Chest X-ray images were taken during both inspiration and expiration 1–2 h after the TFNA procedure.

Pending PCR examination, 500 μ L of the aspirate was divided into two aliquots and frozen at -70°C . As was the case with TFNA, samples for conventional microbiological examination were collected within 36 h of hospital admission (Table 1).

The pulmonary aspirates were examined using a real-time PCR specific for a 587-bp region (between positions 341 and 927) of the 16S RNA gene of bacteria; the PCR products were sequenced using a 3130xl Genetic Analyzer (Applied Biosystems, Foster City, CA, USA), and the sequences were analysed with Sequencing Analysis 5.2 (Applied Biosystems). The consensus sequences were compared with sequences in the GenBank database for identification using NCBI BLAST. Our diagnostic repertoire included real-time quantitative PCR targeting the fumarate reductase (*frdB*) gene of *Haemophilus influenzae* and the outer membrane protein (*copB*) gene of *Moraxella catarrhalis*, as described by Kais *et al.* [1], and detection of *Streptococcus pneumoniae* using primers targeting *spn9802* [2]. Pneumococcal genomic DNA was amplified in a QantiTect SYBR Green PCR assay (VWR International AB, Stockholm, Sweden), performed with a Rotor-Gene 3000 instrument (Corbett Research, Mortlake, Sydney, Australia). Dilutions of genomic DNA extracted from *S. pneumoniae* CCUG 28588, *M. catarrhalis* CCUG 18284 and *H. influenzae* CCUG 23969 (Culture Collection, University of Gothenburg, Sweden) were used as positive controls. Melting curve analyses were performed to ensure the specificity of the amplified products.

The oropharyngeal swabs were examined using virus-specific real-time PCR assays based on TaqMan technology, all validated for routine diagnostic use.

TABLE 1. Tests performed on conventional samples and pulmonary aspirate

Conventional tests	Blood culture Sputum culture Oropharyngeal swabs	BacT/ALERT PF bottle (BioMérieux) for 5 days <i>Mycoplasma pneumoniae</i> ^{a,b} , <i>Chlamydia pneumoniae</i> ^{a,b} , <i>Bordetella pertussis</i> ^{a,b} , <i>Streptococcus pneumoniae</i> ^b , adenovirus ^b , human bocavirus ^b , coronavirus (OC43, 229E and NL63) ^b , enterovirus ^b , human metapneumovirus ^b , influenza A and B virus ^b , parainfluenza virus types 1–4 ^b , RS virus ^b , and rhinovirus ^b
	Paired serum (sample obtained at hospital admission and after 4 weeks)	<i>B. pertussis</i> ^a (<i>B. pertussis</i> filamentous haemagglutinin IgA and <i>B. pertussis</i> toxin IgG, Pertusscan Trok; Euro-Diagnostica AB, Malmö, Sweden) <i>M. pneumoniae</i> ^a (Novitec <i>M. pneumoniae</i> IgG and IgM antibody test; Hiss Diagnostics, Freiburg, Germany) <i>C. pneumoniae</i> ^a (<i>C. pneumoniae</i> IgG and IgM test; Ani Labsystems Ltd, Vantaa, Finland) Adenovirus ^a (Serion KBR Adenovirus, Würzburg, Germany) Influenza A and B virus ^a (Serion KBR Influenza A virus and Serion KBR Influenza B virus)
Lung aspirate	Urinary antigen test (non-concentrated urine) Culture Gram-stained smear PCR	Binax Now Urinary antigen test for <i>S. pneumoniae</i> and <i>Legionella pneumophila</i> (Binax Inc., Scarborough, ME, USA) BacT/ALERT PF bottle (BioMérieux) for 5 days Microscopy for leukocytes and microbes ^a <i>M. pneumoniae</i> ^{a,b} , <i>C. pneumoniae</i> ^{a,b} , <i>B. pertussis</i> ^{a,b} , adenovirus ^b , human bocavirus ^b , coronavirus (OC43, 229E and NL63) ^b , enterovirus ^b , human metapneumovirus ^b , influenza A and B virus ^b , parainfluenza virus types 1–4 ^b , RS virus ^b , rhinovirus ^b , <i>S. pneumoniae</i> ^{b,c} , <i>Moraxella catarrhalis</i> ^c , <i>Haemophilus influenzae</i> ^c
	Rapid tests	Slidex Méningite kit (BioMérieux, Marcy l'Etoile, France ^a)

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TABLE 2. Definite and presumptive community-acquired pneumonia pathogens

Patient	Blood culture	Sputum culture	Urinary antigen	Paired sera	Oropharynx PCR	TFNA	
						Culture	PCR
1	–	<i>Staphylococcus aureus</i> , <i>Haemophilus influenzae</i>	–	<i>Mycoplasma pneumoniae</i>	–	<i>S. aureus</i>	<i>S. aureus</i>
2	<i>Streptococcus pneumoniae</i>	Not representative material	<i>Streptococcus pneumoniae</i>	–	–	–	<i>Streptococcus pneumoniae</i>
3	–	No sample obtained	–	<i>M. pneumoniae</i>	–	–	–
4	–	–	–	–	–	–	<i>Streptococcus pneumoniae</i>
5	–	No sample obtained	–	–	–	–	<i>Streptococcus pneumoniae</i>
6	–	–	–	<i>M. pneumoniae</i>	<i>M. pneumoniae</i>	–	<i>M. pneumoniae</i> , PI4
7	–	–	–	–	–	–	–
8	–	–	–	–	–	–	–
9	–	<i>Moraxella catarrhalis</i>	–	–	–	–	–
10	–	–	–	–	–	–	–
11	–	Not representative material	<i>Streptococcus pneumoniae</i>	–	CorOC43	–	–
12	–	–	–	–	Rhinovirus	–	–
13	–	Not representative material	–	–	–	–	<i>Streptococcus pneumoniae</i> , <i>H. influenzae</i>
14	–	Not representative material	–	–	–	–	–
15	–	–	–	Influenza A virus ^a	–	–	<i>H. influenzae</i>
16	–	No sample obtained	<i>Streptococcus pneumoniae</i>	–	–	–	<i>Streptococcus pneumoniae</i>
17	–	–	–	<i>M. pneumoniae</i>	<i>M. pneumoniae</i>	–	<i>M. pneumoniae</i> , <i>H. influenzae</i>
18	–	–	<i>Streptococcus pneumoniae</i>	–	–	–	<i>Streptococcus pneumoniae</i> , <i>H. influenzae</i>
19	–	No sample obtained	–	–	–	–	–
20	–	No sample obtained	–	–	–	–	<i>H. influenzae</i>

–, Negative test; PI4, parainfluenza virus 4; CorOC43, coronavirus OC43; TFNA, transthoracic fine-needle aspiration.
^aThe patient received influenza A vaccination a few days prior to hospital admission.

The TFNA procedure was easy to perform, and there were no serious adverse events. Two patients developed a minor, self-limiting pneumothorax, and seven patients reported pin-point-sized haemoptysis. A definite aetiology of CAP was established in 12 of 20 (60%) of the cases, in eight cases (40%) by TFNA alone (Table 2). When presumptive aetiologies were included, a diagnosis was established for 14 of 20 (70%) of the patients. Except for one patient who was positive both by culture and 16S rRNA gene-targeting PCR, all PCR-positive aspirates were identified as such using real-time PCR methods.

Our study shows that TFNA is technically feasible in a busy general hospital setting, and confirms that the procedure is associated with mild discomfort only, although careful follow-up disclosed a minimal and clinically insignificant pneumothorax in two of 20 (10%) of the cases. Davidson *et al.* [3] reported that their patients preferred TFNA to transtracheal aspiration, and in one study TFNA was considered to be about as painful as venipuncture [4]. Large reviews representing a total of more than 3000 TFNA procedures found the overall risk of pneumothorax to be 3.3%. Chest drainage was required in 0.5% of these cases [5,6].

Whereas aetiological diagnosis based on bacterial culture is likely to suffer from hours of antibacterial treatment before TFNA, PCR-based diagnostics may be relatively unaffected. In a study by Ruiz-Gonzalez *et al.* [7], an aetiological diagnosis of CAP was obtained for 50% of the study group, using conventional methods. This success rate increased to 83% with the use of TFNA.

In patients treated with antibiotics before admission, *S. pneumoniae* was detected by culture of TFNA material in 33% of the patients, whereas PCR revealed the aetiology in the same material in 83.3% of the cases [8]. In our small study, an aetiology was found in 14 of 20 (70%) of the cases, in eight cases (40%) solely by TFNA. This is the first study of the aetiology of CAP in which the diagnostic procedure was routinely performed as late as 36 h after admission. The fact that all patients had received antibiotic treatment before undergoing TFNA may explain why the aetiology was rarely confirmed by culture.

We conclude that, in our hospital setting, TFNA is a safe and practicable method for the aetiological diagnosis of CAP. Studies are ongoing to assess whether extensive use of TFNA will substantially improve the aetiological diagnosis of

CAP and, if so, result in specifically targeted antimicrobial treatment in our hospital.

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Transparency Declaration

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