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Detection of Pneumococcal Pneumonia in a Phase 3 Trial Comparing Oral Solithromycin versus Oral Moxifloxacin for Treatment of Community-acquired Bacterial Pneumonia in Adults

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Abstract

Background: *Streptococcus pneumoniae* (Spn) is the most common pathogen associated with community-acquired bacterial pneumonia (CABP). However, the detection of Spn in CABP patients can be challenging due to difficulties in quality sputum collection, low recovery of isolates from specimens that must be transported over distances to microbiology laboratories, and the low sensitivity of urinary antigen tests. In this global Phase 3 trial to evaluate the efficacy and safety of solithromycin compared to moxifloxacin in the treatment of CABP in adults, multiple microbiological and diagnostic methods were utilized to identify Spn.

Methods: Patients had acute onset of radiologically confirmed CABP with consistent clinical signs and symptoms. Spn was identified as an etiologic agent on the basis of blood or sputum culture, urine antigen testing (UAT; BINAX), sputum multiplex PCR (Curetis) and quantitative Spn PCR of nasopharyngeal (NP) swabs. The NP swab PCR assay targeted the Spn-specific *lyt*A gene and a threshold of 1000 CFU/mL (determined by receiver operating characteristic curve analysis) was used to differentiate between nasopharyngeal colonization and pneumococcal pneumonia.

Results: 860 subjects were enrolled from 16 countries; not all patients had specimens available for analysis. Spn was the most frequently identified pathogen in the trial (23%). Pneumococcal bacteremia was diagnosed in 15 patients and 28 patients were positive by UAT. Spn was cultured from sputum meeting diagnostic criteria in 55 patients. Pneumococcal pneumonia was identified by NP swab PCR in 133 (15.5%) patients. The overlap of NP swab PCR with more traditional diagnostic modalities is shown in Table 2. NP swab PCR positivity overlapped with blood cultures (67%) and UATs (46%) more frequently than sputum culture did with either (33% and 18%, respectively).

Conclusions: The use of quantitative PCR of NP swabs in this Phase 3 trial significantly increased the rate of identification of Spn as the cause of CABP. Diagnosis by NP swab PCR was better correlated with blood culture, sputum culture and UAT, than any of these more traditional diagnostic methods were with one another.

Introduction

Cempra conducted a randomized, double-blinded Phase 3 trial which evaluated the efficacy and safety of oral solithromycin (5 days) compared to the oral fluoroquinolone, moxifloxacin (7 days), for the treatment of adult patients with CABP. As pneumococcal vaccines are becoming widely used worldwide, including PCV13 (children) and PPSV23 (adults), this phase 3 trial presented a unique opportunity to further evaluate the overall prevalence of pneumococcal types causing CABP, the correlation between the pneumococcal type isolated from the nasopharynx and those strains isolated from blood or sputum and the use of pneumococcal density in nasopharyngeal specimens for detecting pneumococcal pneumonia.

Methods

- Enrollment criteria included the following: ≥ 18 years of age; acute onset or worsening of at least 3 of 4 cardinal symptoms (cough, dyspnea, chest pain, and sputum production); must have fever or hypothermia, and/or physical exam findings consistent with CABP; chest radiograph with lobar or multi-lobar infiltrates; and no long-acting antibiotic use during the prior 7 days.
- Baseline microbiological evaluation included cultures of blood and sputum, detection of *S. pneumoniae* and *Legionella pneumophila* antigen in urine, *L. pneumophila* and *Mycoplasma pneumoniae* serologies (4-fold diagnostic rise in titers), quantitative real-time (*lyt*A) PCR and culture of nasopharyngeal swabs for *S. pneumoniae*, culture and PCR of oropharyngeal swabs for *M. pneumoniae*, and sputum multiplex PCR for lower respiratory pathogens (Unyvero™ by Curetis).

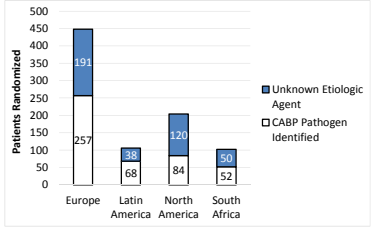
Diagnosis of pneumococcal pneumonia

- Definitive (D-Spn): UAT+, culture positive (blood or high quality sputum)
- Probable (P-Spn): Spn detected using molecular diagnostics (NP swab *lyt*A qPCR, sputum multiplex PCR)

- Spn isolates were serotyped by Quellung reactions using antisera produced by the Staten Serum Institute (SSI). Quellung reactions identify 91 pneumococcal serotypes, including all vaccine types.

Results

Figure 1: Patients Randomized by Region and Percent of Patients in which Etiologic Agent was Identified



- The most frequently identified CABP pathogens in order of prevalence:
  - *S. pneumoniae* (23%)
    - Most common serotypes
      - 3 (10.6%)
      - 19F (7.3%)
      - 19A (5.7%)
  - *H. influenzae* (16%)
  - *L. pneumophila* (14%)
  - *M. pneumoniae* (9%)
  - *M. catarrhalis* (6%)
  - *S. aureus* (4%)

* Some patients had >1 pathogen

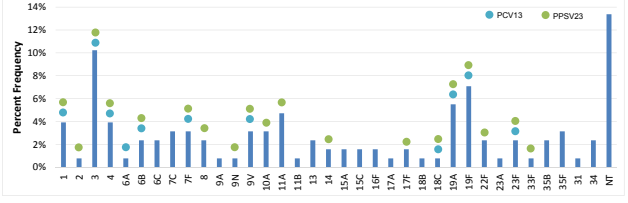
Table 1: Correlation of serotype between isolates from the NP versus those from blood or sputum

Definitive diagnosis	NP culture (%)	Same strain in NP and blood or sputum (%)
Blood culture (N=15)	9 (60)	8 (89)
Sputum (N=55)	28 (50.1)	26 (93)
UAT (N=28)	10 (35.7)	NA
Blood and sputum (N=5)	4 (80)	4 (100)
Blood and UAT (N=6)	4 (66.7)	4 (100)
Sputum and UAT (N=5)	2 (40)	2 (100)

Table 2: Overlap of NP swab PCR with more traditional diagnostic modalities (definitive diagnosis) for pneumococcal pneumonia

	Blood Culture N (%)	Sputum Culture N (%)	UAT N (%)	NP Swab lytA PCR N
Blood Culture	15			
Sputum Culture	5 (33)	55		
UAT	6 (40)	5 (18)	28	
NP Swab PCR	10 (67)	30 (55)	13 (46)	133

Figure 2: Overall Distribution of *S. pneumoniae* Serotypes



Results

Figure 3: ROC curve analysis for *lyt*A qPCR diagnostic accuracy of pneumococcal pneumonia using a threshold of 1x10³ copies/ml.

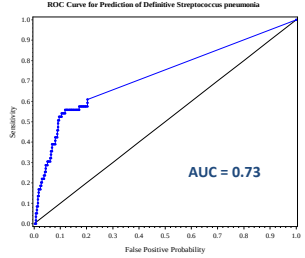


Figure 4: Pneumococcal nasopharyngeal density in Spn CABP patients is significantly different than that in CABP patients due to other etiology.

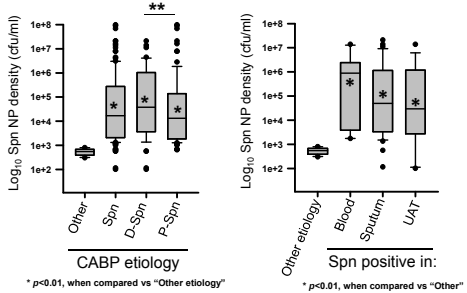


Table 3: Correlation of NP density ≥1x10³ copies/ml with definitive diagnosis of pneumococcal CABP.

	D-Spn CABP		
NP *lytA*	Positive	Negative	Total
Positive	33	40	73
Negative	26	256	282
Total	59	296	355
Sensitivity: 55.9%			
Specificity: 86.5%			
Positive Predictive value: 45.2%			
Negative predictive value: 90.8%			
Accuracy: 81.4%			

* $p<0.01$, when compared vs "Other etiology"

** $p=0.063$. D=Definitive; P=Probable.

CABP	NP lytA positive (%)*	NP Spn density median	NP Spn culture Positive (%)#
Spn CABP (N=198)	126 (63)	$2.3x10^4$	75 (59.5)
Other etiology (N=263)	16 (6.1)	$5.6 x10^2$	3 (18.7)
D-Spn CABP (N=84)	45 (54)	$5.1x10^4$	37 (82.2)
P-Spn CABP (N=114)	81 (71)	$1.5x10^4$	37 (46)

*cutoff >$1x10^3$ cfu/ml; #Percentage of NP *lyt*A positive samples

Conclusions

- *S. pneumoniae* was the most prevalent bacterial pathogen isolated in the Phase 3 CABP trial. Whereas 35 different Spn serotypes were identified, vaccine escape types were found in >38% of patients with Spn CABP.
- We observed a very strong correlation (>89%) between the pneumococcal serotype isolated from blood or sputum, utilized as definitive diagnoses of pneumococcal pneumonia, and those isolated from the nasopharynx of Spn pneumonia patients.
- The use of qPCR of NP swabs significantly increased the rate of identification of Spn as the cause of CABP.
- Diagnosis by NP swab PCR was better correlated with blood culture (67%), sputum culture (55%) and UAT (46%), than any of these more traditional diagnostic methods were with one another.

References

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