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Title: Association between qPCR bacterial load and airway inflammation in COPD

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Body: Background: In COPD bacterial colonisation is associated with a sputum neutrophilia and increased airway inflammation. Quantitative PCR (qPCR) is more sensitive than culture for bacterial detection. Relationships between sputum inflammatory mediators and pathogens quantified by qPCR in stable state are unclear. Methods: Sputum from 66 stable COPD patients was analysed for bacterial load (semi-quantitative culture [colony forming units/ml, CFU], qPCR for Haemophilus influenzae [HI], Streptococcus pneumoniae [SP] and Moraxella catarrhalis [MC]), differential cell counts (total cell count [TCC], neutrophil %, eosinophil %) and a panel of inflammatory mediators. Associations between bacterial load and both inflammatory mediators and differential cell counts were explored. Correlations were considered significant if $p<0.01$. Results: Positive correlations were found between CFU and TNFα ($r_s=0.42$,$p=0.001$), IL1β ($r_s=0.39$,$p=0.001$) and IL10 ($r_s=0.57$,$p<0.001$). Several positive correlations were found between qPCR HI and inflammatory mediators (table 1). qPCR MC and qPCR SP did not positively correlate with CFU, sputum cell counts or any sputum inflammatory mediator.

Table 1: Correlations between H. influenzae qPCR load and inflammatory mediators

HI	TNFα	IL1β	MMP8	MMP9	CXCL10	TCC	Neutrophil %
r_s	0.46	0.49	0.35	0.34	-0.41	0.39	0.44
p	<0.001	<0.001	0.004	0.005	0.001	0.002	<0.001

Conclusions: Stable state H. influenzae qPCR load is associated with an increased sputum total cell count, neutrophil percent and inflammatory mediators in COPD. The same associations were not seen with qPCR

S. pneumoniae and M. catarrhalis.