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central early phase inflammatory cytokine, we investigated whether inflammasome inhibition affects other cytokines like IL-8 and TNF- α .

Murine macrophages and human lung tissue were stimulated with NTHi 10⁶cfu/ml for 24-48h. To assess the relevance of the inflammasome for the inflammatory response, a caspase-1 inhibitor (CI) was added after *in-vitro* infection. The inflammatory response was measured by cytokine ELISA and Western Blot.

Western Blot analysis showed the activation of caspase-1 after NTHi infection and moreover the expression of the NOD-like receptors NOD1 and NLRP3. In cell culture and human lung tissue experiments IL-1 β production was significantly induced (RAW: control 24h beneath lowest standard vs. NTHi 24h 408 \pm 64pg/ml, n=6, p<0.01). The inhibition of caspase-1 led to a significant reduction of IL-1 β levels and also to a decrease of IL-8 and TNF- α production (IL-1 β : NTHi 24h 408 \pm 64pg/ml vs. NTHi+CI 24h 174 \pm 12pg/ml, n=6, p<0.01).

For the first time we demonstrate the participation of the NLRP3-inflammasome in NTHi-induced inflammation in pulmonary cells and tissues. Our findings concerning caspase-1 mediated IL-1 β -upregulation emphasize the role of the inflammasome in respiratory tract infections. These results may provide new insights into the pathogenesis of persistent airway inflammation in COPD.

P2514**The role of galactomannan in exhaled breath condensate in detecting pulmonary aspergillosis in patients with exacerbated COPD**

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Introduction: Growing evidence suggests that patients with severe COPD are at a higher risk of pulmonary aspergillosis (PA), especially during an exacerbation. The levels of GM in exhaled breath condensate (EBC) might allow earlier diagnosis and extend the diagnostic yield of noninvasive mycological tests.

Objective: Evaluate the role of GM in EBC for early diagnosis of PA in severe COPD patients at exacerbation.

Methods: Serum and EBC were collected from 15 severe or very severe COPD patients at exacerbation and tested for GM using a Platelia[®] *Aspergillus* Ag test. Sera/EBC fluids with an index >0.5 were considered positive. Double diffusion in agarose gel (DD) for antibody response to *Aspergillus* was also determined.

Results: Two patients had probable, 7 possible and 6 had no evidence of PA according to the criteria proposed by Bulpa. Serum positive GM assay was observed in two samples of the patients with probable PA and in one sample of two patients with possible PA. In patients with probable PA also serum precipitins was positive (*A. fumigatus*). EBC analysis yielded GM positive results (range, 0.8–7.5) in one patient with probable PA, in 2 patients with possible PA and in 2 patients without PA. In a patient with probable PA, positivity of the GM in EBC, preceded that of the serum of 4 days. GM in EBC was negative in 5 out of 7 cases with possible PA and in 5 out of 6 without PA. The sensitivity of GM in EBC was lower for the diagnosis of probable and possible PA compared to serum GM. However, considering the discordant results in serum and in EBC of four patients with possible PA we suggest that EBC GM levels can expand the diagnostic yield of PA.

P2515**Clinical features of patients with pneumococcal urinary antigen positivity, in a cohort of hospitalised community acquired pneumonia**

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Streptococcus pneumoniae accounts for up to 50% of hospitalised community acquired pneumonia. Diagnosis of pneumococcal disease has always been a challenge. Urinary antigen testing provides a non-invasive, sensitive and specific diagnostic tool.

We investigated the clinical features of patients with pneumococcal urinary antigen positivity in patients admitted with community acquired pneumonia.

We conducted an observational, prospective cohort study in two large UK teaching hospitals, from September 2008 to September 2010. Consecutive adult patients (aged over 16), admitted with community acquired pneumonia (CAP) were recruited. A standardised proforma was used to collect clinical information. Urine samples were tested using the Binax NOW[®] immunochromatographic test. A total of 920 urine samples were available for analysis. 205(22.3%) had a positive antigen test.

Patients with a positive antigen test were more likely to be hypotensive (16.8% of antigen positive vs. 6.8% of antigen negative patients, OR 2.8, 95% CI 1.7-4.8, p<0.05) and tachypnoeic at presentation. Incidence of parapneumonic effusion and critical care admission rates (OR 2.22, 95%CI 1.49-3.34, p<0.01) were also higher in the antigen positive group. These associations were maintained when adjusted for age and pneumonia severity.

Patients with a positive pneumococcal antigen test were more unwell at presentation with a greater likelihood of complications. This is likely to be due to the higher bacterial load in patients with a positive antigen test. Thus, urinary antigen testing appears to add prognostic value in addition to its diagnostic capabilities, when used in pneumococcal disease.

266. Prognostic indices in respiratory infections**P2513****Nontypeable haemophilus influenzae leads to activation of the NLRP3 inflammasome – A possible trigger of chronic bronchial inflammation in COPD**

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The inflammasome is a cytosolic protein complex which is involved in a variety of inflammatory diseases. Since it represents a heterogeneous group of proteins, we elucidated which specific set of proteins is recruited after stimulation with nontypeable *Haemophilus influenzae* (NTHi). In view of the fact that IL-1 β is a

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P2516**Effect of *Cryptococcus neoformans* on the immune system of immunocompetent patients**

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On one hand the host immune system regulates the susceptibility and resistance to cryptococcal infection, on the other Cn can also affect T-cell activation and polarization during infection. Cn may potentially interfere with the differentiation of Th1 cells, which may be an escape mechanism of evade host defence and contribute to the cryptococcal infection in immunocompetent patients. However, most of these effects on T-cell biology were only found in cell and animal studies so far.

Objectives: To determine the effect of Cn on the immune system of immunocompetent patients.

Methods: Twenty immunocompetent patients with pulmonary cryptococcal infection were enrolled. Blood plasma concentrations of IFN- γ , IL-4 and IL-12 were measured using Elisa. PBMC were then isolated and incubated with or without IL-12 for 48 hours, followed by the assay of IFN- γ and IL-4 concentration in the supernatant.

Results: Plasma IFN- γ was greatly decreased in the patients when compared to the healthy controls. No significant differences in plasma IL-4 and IL-12 were observed. Although IL-12 treatment can both increase IFN- γ level in PBMC culture supernatant from the two groups, the increment for cryptococcal infection patients was much lower (3.1-fold) compared with that from healthy control (7.4-fold). IL-12 treatment had no observed effect on the IL-4 production of PBMC.

Conclusions: Cryptococcal infection can damage the host immune system, leading to a deficient response to the IL-12 stimulation and an impaired Th1 polarization. This may explain the persistence of Cn in the immunocompetent patients.

P2517**Exhaled breath biomarkers in patients with ventilator associated pneumonia (VAP)**

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Volatile organic compounds (VOC) in breath have been described as biomarkers of metabolism, oxidative stress and cancer. This pilot study was intended to find out whether VAP related breath biomarkers could be recognized by means of a smart and rapid combination of VOC sample preparation and analysis.

20 mechanically ventilated patients (10 with pneumonia, 10 controls) were investigated. 15 mL of alveolar gas were withdrawn from the respiratory circuit. VOCs were pre-concentrated by means of needle trap micro extraction (NTME) at the bedside and identified/quantified by means of gas chromatography-mass spectrometry (GC/MS). Results were analysed using ANOVA on ranks. Expired concentrations of VOC's ranged from (400 pptV to 3000 ppbV (0.02 to 14.2 nmol/L). Exhaled acetone concentrations were higher in control patients (median 2895 ppbV vs. 187 ppbV, $p=0.037$). VAP patients exhaled lower concentrations of C8 aldehydes (median 2.061 ppbV vs. 19.683 ppbV, $p=0.013$) than control patients. Exhaled pentane showed a tendency to higher concentrations in VAP patients (median 9.907 ppbV vs. 6.040 ppbV).

The NTME- GC/MS assay enabled reliable detection of volatile substances from ventilated patients in trace amounts. Elevated pentane concentrations indicate oxidative stress in VAP, reduced aldehyde concentrations may be due to chemical quenching through ROS or ONOO- present in the alveoli of pneumonia patients. Analysis of exhaled oxygenated compounds bears the potential of non invasive monitoring and recognition of pathological pulmonary processes.

P2518**Copeptin predicts early clinical deterioration and persistent instability in community-acquired pneumonia**

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Optimal risk prediction of early clinical deterioration in CAP remains unresolved. We prospectively examined the predictive value of the new biomarkers copeptin and proadrenomedullin (MR-proADM) in comparison to clinical scores and inflammatory markers to predict early high risk prognosis in CAP.

Methods: 51 consecutive hospitalised adult patients were enrolled. We measured CRB-65- and PSI-scores, the ATS/IDSA 2007 minor criteria to predict ICU-admission and the biomarkers CRP, procalcitonin, copeptin and MR-proADM on admission. Predefined outcome parameters were combined mortality or ICU-admission after 7 days and clinical instability after 72 hours.

Results: Copeptin was the only biomarker significantly elevated in patients with either adverse short term outcome ($p=0.003$). In ROC-curve analysis copeptin predicted ICU admission or death within 7 days (AUC 0.81, cut-off 35 pmol/L; sensitivity 78%, specificity 79%) and persistent clinical instability after 72 h (AUC

0.74). In Kaplan-Meier-analysis patients with high copeptin showed lower ICU-free survival within 7 days ($p=0.001$). The diagnostic accuracy of copeptin was superior to the CRB-65 score and comparable to the PSI-score and the ATS/IDSA minor criteria. If copeptin was included as additional minor criterion for combined 7-day mortality/ICU-admission, the diagnostic accuracy of the criteria was significantly improved (AUC 0.85, $p=0.045$).

Conclusion: Copeptin predicts early deterioration and persistent clinical instability in hospitalised CAP and improves the predictive properties of existing clinical scores. It should be evaluated within a biomarker guided strategy for early identification of high risk CAP patients.

P2519**Correlation of Mycobacterium tuberculosis-specific and non-specific quantitative T cell IFN- γ responses with mycobacillary load in a HIV-prevalent high burden setting**

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Background: Measures of bacillary load in patients with tuberculosis (TB) may be useful for predicting and monitoring response to treatment. The relationship between quantitative T-cell responses and mycobacterial load is poorly studied. We hypothesised that, in a high burden setting, the magnitude of mycobacterial antigen-specific and non-specific T-cell IFN- γ responses would correlate with (a) bacterial load and (b) culture conversion in patients undergoing treatment.

Methods: We compared the magnitude of purified-protein-derivative (PPD) and RD1-specific (TSPOT.TB and QFT-GIT) peripheral blood IFN- γ T-cell responses with associates of sputum bacillary load [liquid culture time-to-positivity, smear-microscopy grade, Xpert-MTB/RIF Ct values, and the presence of cavities on a chest radiograph] in 513 individuals with suspected TB in Cape Town, South Africa. Serial IGRA responses were evaluated at 2 (n=35) and 6 months (n=13) post-treatment initiation.

Results: PPD and RD1-specific IFN- γ responses were not associated with culture TTP (p-values for TSPOT.TB, QFT-GIT and PPD of 0.11, 0.07 and 0.09), smear-grade (0.42, 0.09, and 0.85), Ct values (0.70, 0.91, and 0.49) or the presence of cavities on the chest radiograph (0.12, >0.05, and 0.08). 2-month IGRA conversion rates (positive to negative) were negligible [$<10\%$ for TSPOT.TB (3/28) and QFT-GIT (1/29)] and lower compared to culture [60% (21/35); $p<0.01$].

Conclusions: In a high-burden setting M. tuberculosis-specific and non-specific antigen-driven IFN- γ responses do not correlate with bacillary load and are not useful for prognostication or treatment monitoring.

P2520**LL-37 is produced intrapleurally in infectious pleural effusion**

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LL-37 is an antimicrobial peptide produced by neutrophils, respiratory epithelial and mesothelial cells that has been studied for its broad spectrum activity against microorganisms. It also recruits inflammatory cells and promotes immune responses. It has never been measured in pleural fluid.

Aims and objectives: The objective of our study is to measure the pleural and serum levels of LL-37 in pleural effusion patients, and to compare these levels and the pleural-to-serum LL-37 ratio among pleural fluids of three frequent etiologies: infectious, malignant and congestive heart failure(CHF).

Methods: We obtained 42 pleural effusions and divided them into 3 diagnostic categories. LL-37 was measured in the pleural fluid and serum of 23 infectious effusions, 10 malignant effusions and 9 CHF effusions by ELISA. Statistical analyses were performed using software SPSS 17.0.

Results: Results are presented: mean \pm Std. Deviation (median, minimum-maximum).

Pleural Fluid LL-37 levels: Infectious 3.77 ± 4.81 ng/ml (1.64, 0.38-19.4) malignant 2.58 ± 4.17 (0.87, 0.09-135), CHF 1.59 ± 1.02 (0.99, 0.47-3.3) ($p=0.4$).

Serum LL-37 levels: Infectious 2.09 ± 3.42 ng/ml (0.98, 0.06-16.35) malignant 3.44 ± 4.3 (1.19, 0.17-12.6), CHF 3.44 ± 3.02 (2.6, 0.71-10.3) ($p=0.13$)

Pleural fluid-to-Serum LL-37 ratio levels: Infectious 1.33 ± 1.88 (1.29, 100-0.43) malignant 0.60 ± 0.92 (0.72, 1.11-0.21), CHF 0.46 ± 0.93 (0.44, 1.12-0.24) ($p<0.001$). Infectious vs malignant $p=0.002$, infectious vs CHF $p<0.001$, malignant vs CHF not significant).

Conclusions: Pleural fluid-to-Serum LL-37 ratios are significantly elevated in infectious pleural effusions in comparison with malignant or CHF pleural effusions, suggesting that LL-37 is actively produced intrapleurally in infectious effusions.

P2521**Usefulness of procalcitonin as a diagnostic marker of pleural effusion**

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Pleural effusions are common and are associated with many diseases. We investigated the usefulness of procalcitonin (PCT) as a diagnostic marker for the cause of pleural effusion. The study was carried out on 54 patients with pleural effusion divided into groups; transudate (n=6), empyema (n=9), T.B (n=8), parapneumonic effusions (PPE) (n=9) and malignant effusions (n=22). Levels of procalcitonin were measured both in serum & pleural effusions. Pleural fluid procalcitonin was highest in empyema 1.17±0.86 ng/ml, next highest in PPE (0.57±0.56 ng/ml), & lowest in transudative effusions (0.06±0.03 ng/ml). Pleural fluid & serum procalcitonin levels positively correlate in both empyema & PPE. The optimal discrimination of patients with empyema could be performed at a cut-off point of pleural fluid procalcitonin 0.09 ng/ml with area under the curve (AUC) of 0.93 (sensitivity 80%, specificity 95%) and at a serum procalcitonin 0.08 ng/ml with AUC of 0.74 (sensitivity 80%, specificity 60%). However, the optimal discrimination of PPE could be performed at a cut-off point of pleural fluid procalcitonin 0.065 ng/ml (sensitivity 78%, specificity 53%) and at a serum procalcitonin 0.054 ng/ml (sensitivity 89%, specificity 33%). The optimal discrimination of patients with empyema & PPE could be performed at a cut-off point of pleural fluid procalcitonin 0.075 ng/ml (sensitivity 83%, specificity 58%) and at a serum procalcitonin 0.07 ng/ml (sensitivity 83%, specificity 47%). In conclusion: Pleural fluid PCT is a good and early marker of infection in the pleural space and correlates with the serum PCT in patients with PPE or empyema. Pleural PCT had better diagnostic accuracy than the serum PCT in cases of PPE & empyema.

P2522***Pseudomonas aeruginosa* exacerbations in COPD patients**

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Background: *Pseudomonas aeruginosa* is found in COPD patients sputum in 4-15%, mainly in those with advanced disease and/or in those requiring mechanical ventilation. Currently, there are no data to justify an empiric antibiotic therapy against *Pseudomonas aeruginosa* when a new COPD exacerbation occurs in a patient with a previous *Pseudomonas aeruginosa* exacerbation.

Methods: We conducted a retrospective study to analyse microbiological ecology exacerbations in COPD patients with at least one *Pseudomonas aeruginosa* exacerbation

Results: Among the 243 COPD patients hospitalized during the study period (2007-2011), only 23 (9.5%) had at least one *Pseudomonas aeruginosa* exacerbation (between 1 and 11 exacerbations per patient). They presented a new *Pseudomonas aeruginosa* exacerbation in 54% of cases. From one to another exacerbation, the *Pseudomonas aeruginosa* susceptibility changed, with a wild type *Pseudomonas aeruginosa* in 58% of cases during the first exacerbation and 42% during the next one. COPD patients with GOLD stage IV were rarely hospitalized for a wild *Pseudomonas aeruginosa* exacerbation (p = 0.01, 15% vs. 83% in GOLD stage II and III patients)

Conclusion: In this pilot study, the microbiological ecology of COPD exacerbation differed from one exacerbation to another, contrary to that observed in cystic fibrosis patients.

P2523**Prognostic factors for short and long term outcomes of outpatient exacerbations in moderate-to-severe COPD**

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Introduction: Older patients with severe COPD, frequent exacerbations and comorbidities are at higher risk of poor outcomes. Additional risk factors for short and long term outcomes are yet to be fully identified.

Methods: In the MAESTRAL study, COPD patients¹ were treated with 5-day moxifloxacin or 7-day amoxicillin/clavulanic acid for an Anthonisen type I exacerbation², stratified to oral corticosteroid treatment at the physician's discretion. Predictive factors for clinical failure at end-of-therapy (EOT) and 8 weeks post-EOT were compared by post-hoc multivariate stepwise logistic regression analysis.

Results: Patients with frequent exacerbations, purulent sputum, higher respiration rate, low body temperature and bacterial resistance to study drug had a higher risk to fail at EOT. While patients who, in addition to frequent exacerbations and low body temperature, required systemic corticosteroids for their current exacerbation, were on LABA, suffered from sleep disturbances, had longer duration of chronic

bronchitis, increased heart rate, low FEV₁, low BMI and positive sputum culture at EOT were more likely to fail up to 8 weeks post-therapy.

Variable	EOT			8 weeks post-therapy		
	OR	P value	95% CI	OR	P value	95% CI
Increasing number of AE episodes in the last 12 months ^b	1.448	0.0004	(1.178, 1.779)	1.285	0.0033	(1.087, 1.520)
Sputum purulence ^a		0.0081		n/a	>0.05	n/a
Clear vs purulent	6.674		(0.571, 78.038)			
Mucoid vs purulent	10.218		(0.568, 183.619)			
Mucopurulent vs purulent	1.850		(1.211, 2.827)			
Increasing respiration rate ^{a,b}	1.038	0.0450	(1.001, 1.077)	n/a	>0.05	n/a
Low body temperature ^{a,b}	0.557	0.0035	(0.376, 0.825)	0.58	0.0001	(0.438, 0.787)
Presence of pre-therapy resistance to study drug (yes vs no)	0.492	0.0122	(0.282, 0.857)	n/a	>0.05	n/a
Co-administration of systemic steroid ^a	n/a	>0.05	n/a	2.132	0.0001	(1.557, 2.881)
(no vs yes)						
Presence of sleep disturbance ^a	n/a	>0.05	n/a		0.0024	
Not at all vs A lot/Extreme				1.932		(1.324, 2.820)
Moderate vs A lot/Extreme				1.726		(1.143, 2.606)
Longer duration of chronic bronchitis ^b	n/a	>0.05	n/a	1.024	0.015	(1.005, 1.043)
Increasing heart rate ^{a,b}	n/a	>0.05	n/a	1.017	0.0091	(1.004, 1.029)
Lower % predicted FEV ₁ ^{a,b}	n/a	>0.05	n/a	0.979	0.0019	(0.966, 0.992)
Lower BMI ^{a,b}	n/a	>0.05	n/a	0.963	0.0143	(0.934, 0.992)
Treatment with LABA ^a	n/a	>0.05	n/a	0.625	0.0062	(0.446, 0.875)
Presence of positive sputum culture at EOT (yes vs no)	n/a	>0.05	n/a	0.561	0.0011	(0.395, 0.795)

¹ Outpatients (ITT population, N=1352): ≥60 yrs, ≥20 pack/yr smoking history, FEV₁ ≤ 60%, ≥2 previous exacerbations in the last year.

² Wilson R et al., *Eur Respir J* 2012, epub ahead of print.

^a at current exacerbation, ^b continuous variables, n/a, not applicable.

Conclusion: Several new risk factors have been identified that may help identify exacerbation patients who are at risk of failure despite adequate antibiotics. These patients should be closely monitored during and after treatment of their exacerbation.

P2524**Colonization in advanced chronic obstructive pulmonary disease**

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Background: Isolation of potentially pathogenic organism from the sputum is associated with at least one hospitalization for COPD exacerbation (Martinez-Garcia et al. *Chest* 2011; 140: 1130-1137). But there is still a lack of examinations in larger populations of patients with COPD and pathogenic colonization.

Aims: This examination was performed to evaluate the colonization in patients with advanced COPD.

Methods: In this single-center evaluation, 379 patients with advanced COPD (GOLD III and IV) in our pre-transplant outpatient clinic were screened between October 2008 and June 2011 by lung function, exacerbation rate within the last 12 months and sputum analysis.

Results: The median exacerbation rate within the last 12 months was 2 (IQR 1-3). 51.7% of the patients had expectoration and 40.9% had none (7.4% remains unknown). We analyzed the sputum of 196 patients and had a positive sputum culture in 31.6% of the patients, which is 16.4% of the whole examination group. Patients with a positive sputum culture were significant more often hospitalized due to exacerbation (p=0.02). 94 patients (24.8%) underwent lung transplantation in the observation period. 19% of the explanted lungs had a proof of pathogenic organism. In 71 patients (75.5%) analysis of the sputum before transplantation was concordant with the results of the explanted lung. 11 patients (11.7%) had a proof in the explanted lung and no positive sputum or expectoration before.

Conclusion: Even in patients with end stage COPD chronic bacterial colonisation does play a role only in a minority of the patients (16%). The proof of pathogenic organism correlates with significant more hospitalization due to exacerbations.

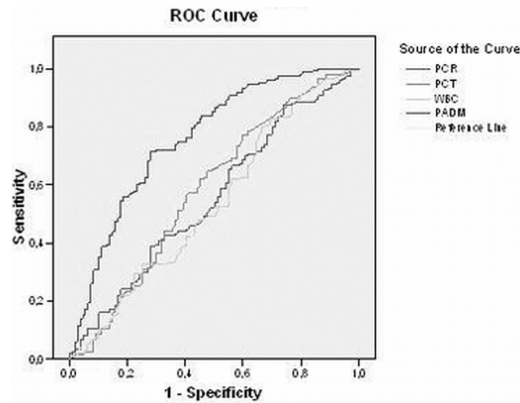
P2525**Biomarkers and severity in community-acquired pneumonia (CAP)**

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Introduction: To evaluate the prognostic value of biomarkers in community-acquired pneumonia (CAP), we studied the correlations between C-reactive protein (CRP), procalcitonin (PCT), leukocyte count (WBC) and proadrenomedullin (proADM) with the widely used PSI severity score.

Material and methods: We prospectively studied 282 immunocompetent, adults patients hospitalized with CAP, calculated their PSI score and measured on admission the mentioned four blood biomarkers. Subsequently, we established the ROC curves to determine which of the biomarkers had a better discriminating power from mild CAP (PSI 1-3) to severe ones (PSI 4-5).

Results: PCT and proADM significantly discriminated severe from mild CAP, although the area under curve was significantly higher for proADM (0.757 vs. 0.581). The other two biomarkers did not reach statistical significance.



Area Under the Curve

Test Result Variable(s)	Area	Std. Error(s)	Asymptotic Sig. (b)	Asymptotic 95% Confidence Interval	
				Lower Bound	Upper Bound
PCR	.562	.037	.152	.481	.654
PCT	.581	.037	.027	.508	.654
WBC	.526	.037	.229	.463	.609
PADM	.757	.031	.000	.696	.818

The test result variable(s) PCR, PCT, WBC, PADM has at least one tie between the positive actual state group and the negative actual state group. Statistics may be biased.
 a. Under the nonparametric assumption
 b. Multihypothesis: true area = 0.5

Conclusions: ProADM is a good predictor of CAP severity at the time of admission, and can be useful, with the clinical scores to identify severe CAP. This may help us to make decisions of patients site of care and management in the early hours.

P2526

Role of basophils in immunological memory responses to pneumococcal protein antigens and *S. pneumoniae* infections in mice

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Basophils have been shown to play an important role in memory immune responses to vaccination with pneumococcal protein antigens. We here examined whether increased basophil counts would provide increased humoral immune responses and thus protection against *S. pneumoniae*. Mice underwent primary and secondary immunization with pneumococcal surface protein A (PspA). Prior to secondary immunization, mice were treated with IL-3 or IL-3 complexed with α -IL-3 antibody (IL-3 complex) to increase basophil pool sizes. Subsequently, mice were challenged with invasive *S. pneumoniae* and developing bacteremia and survival were monitored over time. Treatment of mice with IL-3 and even more so IL-3 complex resulted in strongly expanded basophil pool sizes and significantly increased PspA-specific antibody titers that protected mice from pneumococcal sepsis but unexpectedly did not improve their survival. However, passive immunization of mice with antiserum of IL-3 complex-treated, PspA-immunized mice significantly improved their survival after challenge with invasive *S. pneumoniae*. These data show that although IL-3 complex treatment of mice boosts their basophil counts and protects mice from pneumococcal sepsis, it still exerts severe side-effects in mice after intratracheal challenge with *S. pneumoniae*, and as such does not offer as adjuvant-independent approach to improve lung protective immunity against lung-tropic pathogens.

P2527

Bacterial biofilms in bronchiectasis of primary ciliary dyskinesia (PCD) in comparison with cystic fibrosis (CF)

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Background: Bronchiectasis (B.) is induced by different mechanisms, one of these is primary ciliary dyskinesia (PCD). Genetic aberrations lead to a lack of mucociliary clearance. The bacterial biofilm in B. of patients with PCD in comparison to CF was studied by fluorescence *in situ* hybridisation (FISH).

Material and methods: An explant and 2 middle lobe resections of 3 patients (age between 5 and 50 years) were investigated using conventional histology. Diagnosis of PCD was confirmed by transmission electron microscopy. For comparison 10 explants of CF patients were available. Of all cases, at least 2 locations were studied by FISH using a pan-bacterial and a *Pseudomonas* (Ps.) specific probe.

Results: Histology revealed typical B. In all 3 PCD cases no bacterial biofilms were detected by FISH, although in at least one case Ps. was detected by culture previously. In comparison all CF cases showed colonization with Ps.

Conclusions: Significant differences exist concerning bacterial biofilms in PCD versus CF. This might be of relevance for the clinical practise.

P2528

Biomarkers and community-acquired pneumonia (CAP) etiology

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Introduction: It is a controversial matter if virus alone, without a copathogen is able to cause pneumonia in immunocompetent adults. We studied leukocyte count (WBC), C-reactive protein (CRP), and procalcitonin (PCT) serum levels to test their behaviour in each etiologic group.

Materials and methods: In a prospective study, we obtained serum CRP, PCT, WBC and PADM from 282 patients with CAP at their admittance. A complete test battery was performed, including two multiple polymerase chain reaction (PCR) for respiratory viruses.

Results: PCT had significant higher levels in bacteria-involved CAP (bacterial an mixed) than viral/atypical ones. Mixed bacterial/viral CAP had a similar value for PCT, whereas viral pneumonias showed lower PCT levels. CRP values were also different between viral and mixed CAP. When Influenza was removed from viral group, differences were maintained. ROC curve to try to discriminate bacteria-involved from viral CAP, had an AUC for PCT of 0.658 (p<0.001).

	Bacterial	Mixed	Viral	Bacterial vs. Mixed	Bacterial vs. viral	Mixed vs. Viral		
	n	Median	n	Median	n	Median		
PCT 64	2,106	38	1,903	58	0,312	0,963	0,001	0,013
CRP 64	18,635	39	28,710	58	16,385	0,051	0,422	0,018
WBC 64	14,000	39	11,600	58	11,900	0,027	0,043	0,750

Conclusion: Biomarkers CRP and specially, PCT have a different behavior in viral than mixed bacterial/virus CAP, even without including Influenza. That suggests that viruses can be, without associated copathogen, cause of CAP.

Levels of PCT are similar in bacterial CAP than mixed virus/bacterial CAP, and higher than the viral pneumonia ones, and it can help us to differentiate bacteria-involved pneumonia from viral CAP.

P2529

Microorganisms isolated in COPD patients hospitalised for acute exacerbations and their clinical correlations

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Microorganisms isolated in hospitalised patients with COPD increase morbidity and mortality. We analysed the microorganisms isolated in blood, sputum, tracheal aspirate and/or bronchial lavage cultures of COPD patients hospitalised for acute exacerbations. 135 COPD patients were included in the study. Mean age of patients was 69.4±11.6 year and 74.1% of the patients was male. The length of hospital stay was 13.9±8.3 day and 57.0%(n=77) of the patients were followed up in intensive care unit (ICU). Mortality rate was 28.1%. In 51.1%(n=69) at least one

Rates of microorganisms isolated in COPD patients

Microorganism	n (%)
Acinetobacter	28 (20.7)
Pseudomonas aeruginosa	16 (11.9)
MRSA	9 (6.7)
Coagulase-negative staphylococci	9 (6.7)
Stenotrophomonas maltophilia	8 (5.9)
Klebsiella	8 (5.9)
E. coli	5 (3.7)
Streptococcus pneumoniae	4 (3)
Enterococcus	4 (3)
H. influenzae	2 (1.5)
Enterobacter spp.	1 (0.7)

culture was positive. Cultures of blood, sputum, tracheal aspirate and bronchial lavage were positive in 34.3%, 48.8%, 71.2% and 6.7% respectively. Acinetobacter was the most isolated microorganism.

The most common microorganism in the ward, and ICU patients were Pseudomonas (8.6%) and Acinetobacter (32.5%) respectively. Mean age was 72.2 ± 9.4 vs 66.4 ± 12.9 ($p=0.004$), mortality rate was 47.8% vs 7.6% ($p<0.001$), length of hospital stay was 15.7 ± 9.7 vs 12 ± 6 days ($p=0.009$) and hospital costs were 6949 ± 6606 vs 2913 ± 1743 Turkish Liras ($p<0.001$) in culture positive patients and culture negative's. In hospitalised COPD patients in our clinic, Acinetobacter was the most common isolated microorganism. In culture positive group, mean age, mortality and hospital costs were higher compared to culture negative group.

P2530

Cellular composition of bronchial brush-biopsies at COPD exacerbation

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46 COPD exacerbation patients were examined. Cytological research of brush-biopsies were taken at bronchoscopy was made; for verification of infectious nature of COPD exacerbation the quantitative bacteriological sputum research, definition of diagnostic main IgG, IgM levels to Ch.pneumoniae, M.pneumoniae in serum by means immuno-assay method, definition of their genomes fragments in sputum by means of PCR method were made. Kruskal – Wallis criterion was used.

Infectious character of COPD exacerbation was confirmed at 36 patients. Typical cells count in H.influenzae, Ch.pneumoniae, M.pneumoniae infection was $14.6 \pm 2.2\%$, $15.1 \pm 2.5\%$, $15.5 \pm 3.1\%$, that was reliable low ($p<0.05$), than in S.pneumoniae, M.catarrhalis infection ($32.6 \pm 3.6\%$, $37.1 \pm 5.1\%$). Reserved cells count was reliable high ($p<0.05$) in M.pneumoniae ($12.0 \pm 2.6\%$), than S.pneumoniae H.influenzae, Ch.pneumoniae, M.catarrhalis infection ($5.3 \pm 1.0\%$, $8.4 \pm 1.5\%$, $8.9 \pm 1.9\%$, $5.0 \pm 2.3\%$). Squamous metaplasied cells was reliable high ($p<0.05$) in H.influenzae, Ch.pneumoniae, M.pneumoniae infection ($6.3 \pm 1.2\%$, $7.5 \pm 1.2\%$, $7.4 \pm 1.6\%$), than in S.pneumoniae, M.catarrhalis infection ($2.5 \pm 0.9\%$, $2.2 \pm 0.7\%$). Dystrophical cells count was reliable high ($p<0.05$) in H.influenzae, Ch.pneumoniae infection ($56.6 \pm 2.1\%$, $54.9 \pm 2.8\%$), than S.pneumoniae, M.pneumoniae M.catarrhalis infection ($46.9 \pm 3.0\%$, $44.7 \pm 3.5\%$, $38.9 \pm 4.3\%$). Macrophages quantity was reliable high ($p<0.05$) in M.pneumoniae infection ($55.1 \pm 0.9\%$) as compared with S.pneumoniae H.influenzae, Ch.pneumoniae, M.catarrhalis infection ($33.7 \pm 3.7\%$, $27.4 \pm 3.0\%$, $25.2 \pm 3.5\%$).

Infectious agent species influence on degree of bronchial mucosa damage. H.influenzae, Ch.pneumoniae have more injured effects and M.catarrhalis have least injured effects.

P2531

Inflammatory cells composition of bronchial brush-biopsies in dependence on infectious agent species at COPD exacerbation

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46 COPD exacerbation patients were examined. Cytological research of brush-biopsies were taken at bronchoscopy was made; for verification of infectious nature of COPD exacerbation the quantitative bacteriological sputum research, definition of diagnostic main IgG, IgM levels to Ch.pneumoniae, M.pneumoniae in serum by means immuno-assay method, definition of their genomes fragments in sputum by means of PCR method were made. Kruskal – Wallis criterion was used. Infectious character of COPD exacerbation was confirmed at 36 patients. Inflammatory cells composition in bronchial brush-biopsy were researched. Macrophages quantity was reliable high ($p<0.05$) in M.pneumoniae infection ($55.1 \pm 0.9\%$) as compared with S.pneumoniae H.influenzae, Ch.pneumoniae, M.catarrhalis ($33.7 \pm 3.7\%$, $27.4 \pm 3.0\%$, $25.2 \pm 3.5\%$; $36.2 \pm 4.9\%$ accordingly). Neutrophiles count was reliable high ($p<0.05$) in H.influenzae, Ch.pneumoniae ($50.3 \pm 4.5\%$, $56.5 \pm 4.2\%$ accordingly) as compared with S.pneumoniae, M.pneumoniae M.catarrhalis ($36.1 \pm 4.0\%$, $30.8 \pm 4.7\%$, $34.8 \pm 4.6\%$ accordingly). Eosinophiles count was reliable high in S.pneumoniae infection ($3.7 \pm 1.4\%$) as compared with H.influenzae, Ch.pneumoniae, M.pneumoniae, M.catarrhalis ($3.0 \pm 1.5\%$; $2.0 \pm 1.1\%$; $1.2 \pm 0.1\%$; $1.4 \pm 0.6\%$ accordingly) ($p<0.05$).

Infectious agent species influence on intensification and character of inflammation in bronchial mucosa in COPD exacerbation. M.pneumoniae induces mononuclear response, H.influenzae, Ch.pneumoniae induces polymorphonuclear response.

P2532

Nosocomial pneumonia control programs: The most difficult questions

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Objective: To develop and to study nosocomial pneumonia control programs (NPCP) in general surgical departments (GSD).

Significance: We've developed and carried out specific NPCP for GSD and also have studied level of surgeons knowledge and revealed most problem questions.

Study design: Prospective study.

Setting: 6 surgical departments in a city hospital #6.

Study population: Surgeons.

Methods: During 2001-2003 NPCP were developed and carried out among 65 surgeons. There have been considered questions covered control and prophylaxis of nosocomial pneumonia (NP) and other nosocomial infections (NI). Special questionnaire included 27 questions has been created to estimate an initial and final level of medical staff knowledge (LMSK).

Results: After surgeons completed questionnaire at the first time, level of correct answers (LCA) has totaled 48.8%. Taking into account the results of received data there have been developed and carried out special NPCP aimed to increase the LMSK (lectures, discussions, printed issues). Comparative exercise was repeated 1.5 yrs later. Test showed the increase of LCA up to 60% ($p<0.05$). We determined that NP-rate was statistically decreasing during NPCP ($p<0.05$). It was fixed that the most difficult questions were the following: sources of NI agents, risk factors of NI, rationale antibiotic use, hand-hygiene and other NI-preventive measures, mortality rates under NI.

Conclusions: There is a necessity to carry out NPCP among surgeons in GSD to increase LMSK and to decrease NP level. Therefore, the most difficult questions require to be studied more deeply.