XL®, Trudell Medical International) optimized with its table-top (Ombra®) compressor.
Methods: Each nebulizer (n=5/group) was filled with 2.5-mL, 1.0-mg/mL albuterol (Ventolin®, GSK Canada Inc.), and connected to a breathing simulator (ASL5000, IngMar Medical, Pittsburgh, PA) mimicking adult tidal breathing (V_t= 600-mL; duty cycle = 33%; rate = 10 cycles/min). Emitted aerosol was captured on a filter at the mouthpiece, replaced every minute until onset of sputtering, defining run time. Recovery/assay of salbutamol was undertaken by HPLC-UV spectrophotometry. Fine droplet fraction ($FDF_{<4.7\mu m}$) and mass median droplet diameter (MMD) were determined by laser diffractometry. Total fine droplet mass ($FDM_{<4.7\mu m}$) was the product of total mass and $FDF_{<4.7\mu m}$. Comparative measurements were made with the Sprint® (PARI, Germany) and reusable Sidestream® (Philips-Respironics, Germany) air entrainment nebulizers using PARI BOY®SX® and Inspiration Elite® table-top compressors respectively.
Results: See Table.

Nebulizer/Table-top Compressor Performance Data

Mean ± SD	BAN	Sprint	Sidestream
FDF <4.7μm (%)	70.8±1.0	57.9±3.1	68.6±1.5
MMD (μm)	3.39±0.05	4.13±0.21	3.43±0.11
FDM <4.7μm (μg)	530±22	408±22	233±6
Run time (min)	11	8	10

Conclusions: The BAN/Ombra system provided highly respirable aerosol with $FDM_{<4.7\mu m}$ greater than the benchmark systems. Its run time reflects the fact that aerosol is only delivered during inhalation and not wasted to the environment.

P2182
Formoterol and fluticasone reduce the deposition of pro-inflammatory collagens
Michael Roth, Jun Zhong, Chong Teck S'ng, Michael Tamm. *Pulmonary Cell Research, Dept. Biomedicine, University Hospital, Basel, Switzerland*

Background: Increased deposition pro-inflammatory collagen I and III has been shown in the sub-epithelial airways in asthma. Formoterol has been reported to reduce collagen deposition, while steroid action depends on the presence of inflammation. Under inflammatory condition, steroids increased collagen deposition.
Objective: We investigated the effect of three steroids on the inhibitory effect of formoterol on collagen deposition in TGF-β1 and serum stimulated human airway smooth muscle (ASM) cells.
Methods: ASM cells were incubated for 3 days with serum (5%) or TGF-β1 (1ng/ml) in the presence or absence of formoterol, dexamethasone, budesonide or fluticasone (1nM – 1microM). Collagen deposition was determined by an in house ELISA.
Results: Serum and TGF-β1 significantly increased the deposition of collagen I and III, while they did not affect the collagen IV content. In non-stimulated cells, all 3 steroids reduced the deposition of collagen I and III dose dependently, while they increased collagen IV. In stimulated cells (TGF-β1, serum) collagen I and III deposition were further increased. Formoterol dose dependently reduced the deposition of all three collagens in non-stimulated and stimulated cells. When combined with steroids the inhibitory of formoterol on collagen I and III deposition was dose dependently increased, but had no effect on collagen IV. The combination with fluticasone achieved more often a stronger inhibitory effect than the other 2 steroids.
Conclusion: Our data suggests that formoterol has the potential to reduce airway wall remodelling in asthma and in combination with fluticasone, it is more effective than with other steroids.

P2183
Pharmaceutical development of a liquid formulation for pulmonary application of a peptide
Hendrik Fischer[1], Rudolf Lucas[2], Helmut Pietschmann[1], Susan Tzotzos[1], Bernhard Fischer[1]. *[1]APEPTICO, APEPTICO GmbH, Vienna, Austria; [2]Vascular Biology Center, Georgia Health Sciences University, Augusta, United States*

ALI/ARDS are acute and severe conditions with mortality up to 50%. Currently, no specific drug-related treatment exists. ALI/ARDS are characterized by pulmonary oedema due to increased permeability of the endothelial/epithelial barrier. Oedema clearance is linked to Na uptake at the apical side of Type I/II alveolar cells through the amiloride-sensitive epithelial sodium channel "ENaC". AP301, a synthetic peptide derived from the lectin-like domain of TNFα, enhances Na transport through ENaC and thus oedema clearance. As ENaC is located at the inner surface of alveoli, the water soluble AP301 is administered by inhalation of a nebulised aqueous solution. Hence, pharmaceutical development of AP301 as potent new drug for oedema resolution differs from standard orally or parenterally applied medicine and has to follow specific pharmaceutical and medicinal regulations of the Pharmacopoeia and inhalation medicine guidelines. AP301 is nebulised by a mesh-type not a jet or supersonic type nebulizer due to simplicity and effective, but gentle aerosol generation. In order to reach the alveoli the particle size of the aerosol was optimized to <5μm by testing a range of different concentrations of AP301 in combination with various nebulizers. Analyses of biological activity of AP301 in the generated aerosol showed that it was retained during nebulisation. To

249. Translational respiratory medicine in asthma and COPD

P2181
Comparative *in vitro* performance of a new re-usable breath-actuated nebulizer (BAN) with other high performance systems intended for domiciliary use – 1: Table-top compressors
Jamie Malpass, Mark Nagel, Valentina Avvakoumova, Rubina Ali, Heather Schneider, Jolyon Mitchell. *Medical Aerosol Laboratory, Trudell Medical International, London, ON, Canada*

Rationale: Treatments by portable compressor/nebulizer systems can offer very different delivery characteristics. We evaluated a new, reusable BAN (AeroEclipse-

exactly predict clinically relevant doses of AP301, the amount of aerosol reaching the mouth of subjects was analyzed with a breath simulator. To correlate animal toxicity and safety data to human equivalent dose, scaling between species was done according to the AP301 concentration per lung weight, rather than per body surface area or body weight.

P2184

Tiotropium enhances the inhibitory effect of the long acting β2-agonist olodaterol on the release of IL-6 and IL8 by primary human lung fibroblasts of asthma patients

Luigi Costa, Michael Roth, Michael Tamm, Pieter Borger. *Pulmonary Cell Research, Dept of Biomedicine, University Hospital, Basel, Switzerland*

Muscarinic and β2-adrenergic receptors of resident lung cells are modulators of airway inflammation and remodeling. Here, we used human primary lung fibroblasts of healthy and asthmatic subjects to investigate the role of β2-adrenergic and muscarinic receptors on the interleukin IL-1β-induced secretion of IL-6 and IL-8. Fibroblasts were isolated from asthmatic (n=5) and non-asthmatic subjects (n=5) and stimulated with IL-1β in the presence or absence of olodaterol (10-6M), tiotropium (10-6M), or with the combination of olodaterol (10-6M) and tiotropium (10-6M). IL-6 and IL-8 levels in the supernatant were measured by ELISA.
Neither olodaterol nor tiotropium alone affected the secretion of IL-6 and IL-8 in unstimulated cells. Tiotropium reduced the IL-1β-induced secretion of IL-6 and IL-8 in both control and asthmatic cells ($p<0.05$). Olodaterol reduced IL-1β-induced cytokines in control (IL-6: 52±1%, $n<0.05$; IL-8: 54±16%, $p<0.05$) and asthmatic (IL-6: 76±5%, $n<0.05$; IL-8: 72±2%, $p<0.05$) fibroblasts. Compared to olodaterol alone the combination of olodaterol (10-6M) with tiotropium (10-6M) further reduced the release of IL-6 (55±7%; $p<0.05$) and IL-8 (50±6%; $p<0.05$) from fibroblasts of asthma patients only.
Both olodaterol and tiotropium exert anti-inflammatory responses in healthy and asthmatic fibroblasts. The combination of olodaterol with tiotropium further improved the anti-inflammatory effect, specifically in asthmatic fibroblasts. These data provide support for combination therapy of long acting β2-agonists plus long acting muscarinic receptor antagonists.
Supported by Boehringer-Ingelheim, Biberach, Germany.

P2185

Low dose inhaled LPS challenge – Reproducibility of the inflammatory response

Olaf Holz, Frank Schaumann, Ole Janssen, Bianca Lavae-Mokhtari, Heike Biller, Norbert Krug, Jens M. Hohlfeld. *Clinical Airway Research, Fraunhofer ITEM, Hannover, Germany*

Introduction: Inhalaled endotoxin (LPS) is used to study pulmonary inflammation. Conventional inhalation methods require substantial amounts of LPS (15–50μg) to induce a detectable inflammatory response. Authorities often permit LPS challenges only with GMP-grade LPS, which is of limited availability. Here we studied the reproducibility of the response to a low dose of LPS (2μg), which was inhaled by a flow/volume controlled procedure to increase lung deposition.
Methods: Twelve healthy non-smoking volunteers were included. Baseline sputum was evaluated 2-4 weeks prior to the first challenge. On two occasions, separated by 4 weeks, subjects inhaled 2μg LPS (Clinical Center Reference Endotoxin CCRE, NIH), which was nebulized using an Aeroneb solo (Inspiration Medical). Sputum was induced 6h after LPS provocation.
Results: The low dose LPS challenge was well tolerated. Both challenges induced a significant ($p<0.001$) increase in sputum neutrophils (median (IQR)% of sputum leukocytes at baseline: 24.4(31.2)%, 1.LPS: 75.3(13.3)%, 2.LPS: 59.0(19.3) %) and increased sputum IL8 and MPO. A significant ($p<0.01$) increase in sputum monocytes was only detected after the 2.LPS challenge (baseline: 4.3(1.5)%, 1.LPS: 7.7(7.0)%, 2.LPS: 11.0(8.1)%). Despite lower increases of neutrophils in the second challenge, the changes compared to baseline were correlated (r=0.79, ICC=0.64).
Conclusion: Low dose LPS caused a reproducible inflammatory response. However, we found evidence for a more pronounced increase in monocytes in the second challenge. This needs to be considered in proof of concept studies for novel inflammatory compounds.
CCRE was kindly provided by Dr. A. Suffredini, NIH, Bethesda.

P2186

Transactivation and transrepression in the repression of inflammatory gene expression by dexamethasone

Elizabeth King[1], Joanna Chivers[1], Mark Giembycz[2], Robert Newton[1]. *[1]Cell Biology and Anatomy, University of Calgary, AB, Canada; [2]Physiology and Pharmacology, University of Calgary, AB, Canada*

Background: The anti-inflammatory activities of glucocorticoids are attributed to the repression of inflammatory gene expression. Dogma states that this occurs via direct repression (transrepression) by the glucocorticoid receptor (GR) of transcription factors, such as NF-κB. However, evidence also suggests that gene induction (transactivation) by GR is important for repression.
Aims: To assess the roles of transrepression and transactivation in the repression of inflammatory gene expression by dexamethasone (Dex).
Methods and results: The effect of Dex on 39 IL-1β-induced genes was examined in human pulmonary A549 cells by real-time PCR. Dex showed a range of activity in terms of the extent (E_{max}) and potency (EC_{50}) of repression on these genes. These parameters correlated, such that the most highly repressed genes were also the most potently repressed. While all 39 genes were NF-κB-dependent, this did not correlate with repression by Dex. Finally, inhibition of protein translation by cycloheximide (CHX) reduced IL-1β-induced expression of 19 genes (secondary response genes). Of the remaining 21 genes, CHX significantly prevented the Dex-dependent repression of 11 (~50%), suggesting a role for transactivation. These 11 genes were significantly more sensitive (E_{max} and EC_{50}) to repression by dexamethasone when compared to genes showing repression that was insensitive to CHX (and which may represent a classical transrepression mechanism).
Conclusions: Repression of inflammatory gene expression by dexamethasone involves multiple mechanisms. Transactivation appears to play a significant role showing both high potency and high level of repression on target genes.

P2187

Tiotropium provides sustained bronchodilation in asthmatics with persistent airflow obstruction uncontrolled despite treatment in accordance with guidelines

Huib Kerstjens[1], Pierluigi Paggiaro[2], Mark Vandewalker[3], Ekkehard Beck[4], Michael Engel[5], Ralf Sigmund[6], Wolfgang Seibold[7], Petra Moroni-Zentgraf[8], Eric Bateman[9]. *[1]Department of Pulmonary Medicine and Tuberculosis, University Medical Center, Groningen, Netherlands; [2]Dipartimento Cardio-Toracico, U.O. Pneumologia e Fisiopatologia Respiratoria, Ospedale di Cisanello, Pisa, Italy; [3]Clinical Trials, Clinical Research of the Ozarks, Columbia, MO, United States; [4]Medical Department, IFG - Institut für Gesundheitsförderung GmbH, Rüdersdorf Brandenburg, Germany; [5]TA Respiratory Diseases, Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach an der Riss, Germany; [6]Biostatistics, Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach an der Riss, Germany; [7]Clinical Research, Germany, Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach an der Riss, Germany; [8]TA Respiratory Diseases, Boehringer Ingelheim Pharma GmbH & Co. KG, Ingelheim am Rhein, Germany; [9]Department of Medicine, University of Cape Town, South Africa*

Introduction: In some asthmatics airflow obstruction persists despite high-dose (HD) inhaled corticosteroid (ICS) and long-acting β_2-agonist (LABA) use. In a recent study, adding a long-acting anticholinergic (tiotropium) showed favourable effects over 8 weeks (wks) (Kerstjens HA, et al. *JACI*. 2011).
Methods: In 2 replicate 48-wk, doubleblind, parallel-group trials a total of 912 asthmatics with postbronchodilator (BD) FEV_1 <80% predicted and asthma control questionnaire score ≥1.5 while on at least HD ICS+LABA were randomised to additional tiotropium Respimat® 5 mcg or placebo. Prespecified co-primary endpoints included peak and trough FEV_1 at 24 wks. Secondary endpoints were FEV_1 at other time-points, FVC, and daily PEFs.
Results: Baseline characteristics were similar between trials and treatment groups (mean post-BD FEV_1 62% [±13]). Mean change from baseline tiotropium vs placebo after 24 wks in peak pre-BD FEV_1 was 86 (±34) mL (P=0.01) or 154 (±32) mL greater ($P<0.001$), and in trough FEV_1 88 (±34) mL (P=0.01) or 111 (±30) mL greater ($P<0.001$) in trials 1 and 2, respectively. Improvements in FVC and daily PEFs were also significantly greater with tiotropium. There were no signs of tachyphylaxis over 48 wks. No deaths occurred; adverse events were balanced across treatments in both trials.
Conclusion: In asthmatics uncontrolled despite at least HD ICS+LABA, adding tiotropium provided significant lung function improvement at 24 wks which was sustained over 48 wks. Tiotropium is likely to improve severe uncontrolled asthma on top of treatment in accordance with guidelines.
Study supported by Boehringer Ingelheim and Pfizer.

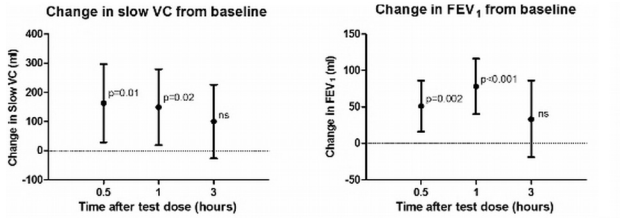
P2188

The effects of sildenafil on lung function in COPD

Andrew Goudie, Pippa Hopkinson, William Anderson, Brian Lipworth, Allan Struthers. *Centre for Cardiovascular and Lung Biology Research, University of Dundee, United Kingdom*

Background: Sildenafil is a pulmonary vasodilator drug used to treat pulmonary hypertension (PH) via PDE5 inhibition. Zaprinast, the precursor to Sildenafil, has moderate bronchodilator effects in exercise induced asthma. A recent study investigating the haemodynamic effects of sildenafil in COPD associated PH (COPD-PH) noted small but significant improvements in FEV1 and FVC.
Aims: To study the effects of sildenafil on bronchodilation and gas trapping in those with COPD-PH.
Methods: COPD patients were invited for echo. RVSP>30mmHg and/or a pulmonary acceleration time of <120ms defined PH. Subjects with COPD-PH were given 50mg of sildenafil(PO). Spirometry was recorded at 0.5, 1 (C_{max}) and 3 hours ($t_{1/2}$).
Results: A total of 33 patients were studied, 61% male. Baseline spirometry: FEV1(l) 1.13 (SD 0.41), FEV1% 45%(14.7), FVC(l) 2.75(0.76), FVC% 86.3%(17.2), slow VC (l) 2.77(0.85), slow VC% 84.9 (18.3). Slow VC significantly increased by 163ml [95% CI 29-297] p=0.01 and 149ml [95% CI 19-280] p=0.018 at 0.5 and 1 hours respectively. FEV1 significantly increased by 51 ml [95% CI 16–86] p=0.002 and 78 ml [95% CI 40-116] p=0.000 at 0.5 and 1

hours respectively. No significant changes were noted for slow VC/FEV1 at 3 hours.



Change in pulmonary function following sildenafil. Data presented as arithmetic means (with 95% confidence interval error bars) as absolute change from baseline. VC = vital capacity. FEV_1 = forced expiratory volume in 1 second. Repeated measures ANOVA analysis with Bonferroni correction, significance set at p<0.05. ns = not significant.

Conclusion: Acute sildenafil use resulted in transient airways dilatation and reduced gas trapping. In the absence of placebo control, spirometric changes due to natural variability cannot be ruled out, although the return to normal at 3 hours suggests a real effect.

P2189

Tolerability and efficacy of budesonide/formoterol via Turbuhaler® vs standard treatment in Japanese patients with moderate to severe COPD: 52-week phase III study results

Masakazu Ichinose[1], Hiroyuki Nakamura[2], Noriharu Shijubo[3], Takefumi Saito[4], Hiroyuki Taniguchi[5], Toru Tsuda[6], Kosho Yoshikawa[7], Lars-Goran Carlsson[8]. *[1]Third Department of Internal Medicine, Wakayama Medical University, Wakayama, Japan; [2]Department of Medicine, Sakaide City Hospital, Kagawa, Japan; [3]Department of Medicine, JR Sapporo Hospital, Hokkaido, Japan; [4]Department of Respiratory Medicine, National Hospital Organization Ibarakihigashi National Hospital, Ibaraki, Japan; [5]Department of Medicine, Tosei General Hospital, Aichi, Japan; [6]Department of Medicine, Kirigaota Tsuda Hospital, Fukuoka, Japan; [7]Department of Medicine, Daido Hospital, Aichi, Japan; [8]Clinical Sciences, AstraZeneca R&D, Molndal, Sweden*

Background: This study evaluated the tolerability and efficacy of budesonide/formoterol (BUD/FORM) vs standard COPD treatment (SCT) in Japanese patients with moderate to severe COPD.
Methods: In this randomised, open-label, parallel-group, phase III study (NCT01070784), patients ≥40 years of age with moderate to severe COPD for ≥2 years received either BUD/FORM 160/4.5 μg 2 inhalations twice daily via Turbuhaler® or SCT (as judged by the investigator) for 52 weeks. Reliever medication: salbutamol via pMDI. Primary outcome: nature, incidence and severity of adverse events (AEs). Secondary outcome variables included: COPD symptoms, lung function and exacerbations.
Results: 260 patients were randomised. BUD/FORM was well tolerated; 404 AEs were reported by 123 patients (94.6%) receiving BUD/FORM vs. 367 AEs by 112 patients (86.2%) on SCT. The majority of AEs were of mild or moderate intensity and the AE profile was similar in the two groups. The most commonly reported AEs (BUD/FORM vs SCT) were nasopharyngitis (42.3% vs 39.2%), COPD (10.8% vs 19.2%) and bronchitis (11.5% vs 11.5%). The frequency of pneumonia-related AEs was similar in both groups (13.1% vs 12.3%) while dysphonia was more frequent with BUD/FORM (5.4% vs 0.8%). Serious AEs were more frequent with SCT (26.2%) vs. BUD/FORM (19.2%). No deaths were reported. Efficacy of BUD/FORM was maintained over 52 weeks.
Conclusions: BUD/FORM 160/4.5 μg 2 inhalations twice daily was well tolerated and efficacy was maintained during 52-week treatment in Japanese patients with moderate to severe COPD.
Funding: AstraZeneca.

P2190

Effectiveness of tiotropium in low-risk patients according to new GOLD severity grading

David Halpin[1], Marc Decramer[2], Bart Celli[3], Antonio Martín[4], Inge Leimer[5], Norbert Metzdorf[5], Donald Tashkin[6]. *[1]Respiratory Medicine, Royal Devon and Exeter Hospital, Exeter, United Kingdom; [2]Respiratory Division, University of Leuven, Belgium; [3]Pulmonary Division, Brigham and Women's Hospital, Boston, MA, United States; [4]Allergy and Respiratory Department, Pfizer Inc, New York, NY, United States; [5]Medical Affairs, Boehringer Ingelheim Pharma GmbH & Co. KG, Ingelheim, Germany; [6]Dept of Medicine, David Geffen School of Medicine, Los Angeles, CA, United States*

Background: GOLD recently updated their COPD severity classification to include risk of exacerbations. Increased risk is typically defined by a FEV_1 of <50% pred and/or ≥2 exacerbations in the previous year (C+D) and low risk by a FEV_1 ≥50% pred and 0-1 exacerbation in the previous year (A+B).
Aims and objectives: To examine the effect of tiotropium 18 μg qd via Handihaler® in GOLD low risk patients (pts) using data from a 4-y, randomized, double-blind, placebo-controlled trial in COPD (UPLIFT®).
Methods: Retrospective analysis of exacerbations, lung function and QoL (SGRQ) in low-risk pts (pts with a baseline postbronchodilator [BD] FEV_1 %pred ≥50% and ≤1 oral steroid/antibiotic course in the previous year). Pts with high risk (FEV_1 %pred <50% or more than 1 course of oral steroids/antibiotics) were also analyzed.
Results: 2012 pts were analyzed (mean age 64.5±8.6 y, male 74%, mean (±SD) baseline postBD FEV_1 1.65 (0.37) L and FEV_1 %pred (±SD) 58.9 (5.8). The HR (tiotropium vs control) for time to first exacerbation was 0.76 (95% CI, 0.68; 0.86; $P<0.0001$); mean annual exacerbation rates were 0.43 (95% CI, 0.40; 0.48) vs 0.61 (0.56; 0.66), rate ratio 0.72 (0.63; 0.81; $P<0.0001$). The SGRQ total score after 4 y was significantly improved by tiotropium vs placebo: −3.63 (95% CI, −5.14; −2.12; $P<0.0001$) and the respective increase for trough FEV_1 was 110 mL (95% CI, 84; 136; $P<0.0001$). SGRQ and trough FEV_1 were significantly improved at all time points. The above-mentioned endpoints were also significantly improved in the high-risk population.
Conclusion: Tiotropium qd was effective throughout 4 y in reducing exacerbations and improving lung function and QoL in low-risk pts with COPD (GOLD A+B).

P2191

Efficacy and safety of fluticasone/formoterol compared to fluticasone alone in patients with asthma

Tetyana Pertseva[1], Sanjeeva Dissanayake[2], Kirsten Kaiser[3]. *[1]Chair of Faculty Therapy and Endocrinology of Dnipropetrovsk Medical Academy, Dnipropetrovsk City Clinical Hospital N° 6, Dnipropetrovsk, Ukraine; [2]European Medical Science, Mundipharma Research Ltd., Cambridge, United Kingdom; [3]Medicinal and Regulatory Development, SkyePharma AG, Muttenz, Switzerland*

Background: To demonstrate efficacy and safety of fluticasone propionate/formoterol fumarate pMDI (FLUT/FORM) compared with fluticasone (FLUT) alone based on the change in FEV_1 from morning pre-dose at baseline to 2 hours post-dose at week 12.
Methods: Patients included in the study were ≥12 years with symptomatic asthma for ≥1 year; steroid-requiring, had FEV_1 40% to 80% [inclusive] of predicted normal values, and documented reversibility within 12 months of the study. Following a 2 week open-label run-in phase with FLUT (100 or 200 μg b.i.d.), 438 patients were randomised to treatment with FLUT/FORM (250/10 μg b.i.d.) or one of two pMDI formulations of FLUT alone (250 μg b.i.d.). The formulations were SkyePharma HFA pMDI (SKP) and GSK fluticasone pMDI (FLO), with n=146 in all groups. Albuterol/salmeterol was given as rescue medication.
Results: There was a clinically important and statistically significant difference in mean change from pre-dose FEV_1 at baseline to 2 h post-dose at week 12 between the FLUT/FORM and both formulations of FLUT (SKP:LS mean difference=0.161 L,P<0.001; FLO:LS mean difference=0.185 L,P<0.001). Results from multiple secondary and tertiary efficacy endpoints assessing lung function, asthma symptoms, exacerbations and rescue medication use supported a superior efficacy of the FLUT/FORM combination over FLUT. Treatment-emergent adverse events were lowest in the FLUT/FORM group (32.9%) compared to SKP (39.7%) or FLO (40.4%). FLUT/FORM was generally well tolerated.
Conclusion: FLUT/FORM was superior to FLUT alone in the management of moderate to severe asthma in adolescents and adults. The overall safety profile of FLUT/FORM was consistent with that of FLUT.

P2192

Does eosinophil cationic protein (ECP) predict asthma outcome and response to treatment in asthmatic patients?

Elena Bacci, Silvana Cianchetti, Manuela Latorre, Lorenza Melosini, Antonella Di Franco, Federico Dente, Pier Luigi Paggiaro. *Cardiothoracic and Vascular Department, University of Pisa, Italy*

In order to test whether sputum Eosinophil Cationic Protein (ECP) predicts asthma outcome and response to treatment, we studied 119 mild/moderate, steroid-naive asthmatic patients. All patients underwent spirometry, methacholine test, induced sputum analysis before and after treatment, and recorded symptom score and rescue beta2-agonist use on a diary card throughout the study. In patients with high (≥75 mcg/L) sputum ECP, baseline FEV1 was lower (high-ECP: 85±17%; low-ECP: 96±14%, p<0.01). Sputum eosinophil percentages were higher in patients with high ECP (high-ECP: 16.3±17.1%; low-ECP: 6.1±8.3%, p<0.01), although the concordance between sputum eosinophils and ECP was poor (r=0.52, p<0.01). Patients were then treated with either inhaled corticosteroids (ICS, n=76) or long-acting beta2-agonist (LABA, n=43) for 3 to 6 months. In LABA-treated patients, symptom score and beta2-agonist rescue use improved regardless of baseline sputum ECP levels; after treatment, however, patients with high baseline ECP levels had greater rescue beta2-agonist use (high-ECP: 0.2±0.2; low-ECP: 0.02±0.03, p<0.05). In ICS-treated patients, PEF, symptom score, beta2-agonist rescue use and PD20FEV1 methacholine improved regardless of ECP levels; however, FEV1 significantly improved in ICS-treated patients with high ECP levels only, possibly because they had lower baseline FEV1 values. Thus, high sputum ECP levels may predict less asthma control, as shown by greater rescue beta2-agonist use, when treatment does not affect airway inflammation; on the other hand, high sputum ECP levels may predict the response to ICS treatment, possibly because they are associated with poorer baseline lung function.

P2193

Methacoline challenge test as an evaluator of response to statins in bronchial hyperresponsiveness

Majid Malekmohammad[1], Fanak Fahimi[2], Atefeh Fakharian[1], Maryam Habibi[2], Parisa Adimi[1]. *[1]Pulmonology and Critical Care, National Research Institute of Tuberculosis and Lung Disease, Tehran, Islamic Republic of Iran; [2]Clinical Pharmacy, National Research Institute of Tuberculosis and Lung Disease, Tehran, Islamic Republic of Iran*

3-hydroxy-3-methylglutaryl-CoA reductase inhibitors (statins), are effective serum cholesterol-lowering agents which they may also have anti-inflammatory properties. The objective of this study was to evaluate the effect of atorvastatin on bronchial hyperresponsiveness.

Adult patients (age 14 to 65 years) with bronchial hyperresponsiveness (BHR) diagnosis based on the spirometry with methacholine challenge test were entered into the study. The study was conducted in the National Research Institute of Tuberculosis and Lung Disease. Patients were randomized to receive either atorvastatin 20 mg/day or placebo for 4 weeks. Spirometric parameters were determined at baseline and at completion of the study. Twenty two patients with the age of 32.95±10.30 years completed the trial.

Changes in airway responsiveness categories (moderate to severe, mild, borderline, normal) after the intervention were not significant in atorvastatin group as in placebo group (p value= 0.131 for atorvastatin group and p value = 0.305 for placebo group). Also, changes in methacholine solution number (different concentrations of methacholine) which caused at least 20% decrease in FEV1 were not significant between groups (p value = 0.089). Although we could not find a significant difference, the patients' fall in FEV1 in atorvastatin group was observed in higher concentrations of methacholine. Median before treatment versus after treatment in atorvastatin group was 1 versus 4 mg/ml, while those were 2 versus 1 mg/ml in placebo group.

This study showed a better but no significant hyperresponsiveness control in the treatment group. The result may be presented more pronounced, if we could increase the sample size.

P2194

The GOLDEN-1 study: Safety and bronchodilatory effects of nebulized glycopyrrolate (EP-101) using high efficiency nebulizer in patients with COPD

Charles Fogarty[1], Edwin Kerwin[2], Karen Dunn[3], David Singh[4], Ahmet Tutuncu[5]. *[1]Clinical Research, Spartanburg Medical Research, Spartanburg, SC, United States; [2]Clinical Research, Clinical Research Institute of Southern Oregon, Medford, OR, United States; [3]Clinical Research, North Carolina Clinical Research, Raleigh, NC, United States; [4]Clinical Research, Medicines Evaluation Unit, Manchester, United Kingdom; [5]Clinical Research, Elevation Pharmaceuticals Inc., San Diego, CA, United States*

Introduction: EP-101 is a long-acting muscarinic antagonist bronchodilator for nebulization using a high efficiency nebulizer for the treatment of COPD. The safety and efficacy of once daily nebulized EP-101 was assessed in this Phase 2b study after 7 days of dosing in patients with COPD.

Methods: This was a multicenter, randomized, double-blind, placebo-controlled, 4-period cross-over, incomplete block design study. A total of 140 patients with moderate-to-severe COPD were randomized to receive 4 of 7 treatments: EP-101 (placebo, 25, 50, 100, and 200 μg) once daily via high efficiency nebulizer, open-label tiotropium 18 μg once daily, and open-label ipratropium 500 μg three times daily via jet nebulizer. There was a 7-day washout period between treatments.

Results: All doses of EP-101 were well tolerated with similar AE rates between placebo and EP-101 (31.2%, 29.7%, 26.9%, 35.5% and 30.7% for placebo, 25, 50, 100 and 200 μg, respectively). There was no apparent dose-response relationship for incidence and severity of AEs. Mean changes in vital signs and ECG parameters from baseline on Day 7 were comparable between the treatment groups. All doses of EP-101 demonstrated dose-related and significant improvements in FEV_1 AUC (0-24hr) on Day 7 compared with placebo, with estimated differences between EP-101 doses and placebo ranging between 110-169 mL.

Conclusion: Once daily dosing with nebulized EP-101 was well tolerated over a 7-day treatment period and provided rapid onset of bronchodilation with clinically meaningful and sustained improvement in lung function over 24 hours in patients with COPD.

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P2195

Spacer cleaning: Nurse and patient survey examines current UK practice

Mark Sanders[1], Mark Levy[2], Keely Thompson[1], Ron Bruin[1]. *[1]HQ, Clement Clarke International Ltd, Harlow, United Kingdom; [2]Senior Clinical Research Fellow, Allergy and Respiratory Research Group, Centre for Population Health Sciences, GP Section, University of Edinburgh, Doorway 3, Medical School, Edinburgh, United Kingdom*

Introduction: Washing spacers improves drug delivery by reduction of electrostatic charges, and is hygienic. Contamination of spacers and masks has been shown (JAMA 2003;290: 2, 195-196). The ERS/ISAM guideline (ERJ 2011;37:1308-1331) identified a lack of standard instructions for good spacer cleaning practice.

Methods: Online structured interview questionnaires by a market research agency, 100 asthma patients (Pts) using pMDI with spacer and 50 respiratory nurses (RN) to establish current practice. Pts: 49 adults, parents of child aged 2-9 yrs (n=30) or 10-15 yrs (n=21).

Results: 74% of RN told Pts to wash spacers weekly, 4% instruct daily and 22% instruct < than weekly. RN reported that Pts describe correct washing; 4% always, 42% mostly, 36% sometimes, 14% rarely and 4% never. RN confidence in patients keeping spacers hygienically was low, with 48% not very or not at all confident, this figure increased to 56% in respect of masks. 71% of Pts said their RN or doctor explained how to wash the spacer, and 63% received drying instructions. 54% were told how to wash and dry, but 24% were not told either; 13% could not recall being told. 21% of adult Pts don't wash + dry their spacer (not been told); 6% of children's spacers were not washed. Of 83 who wash their spacer: 35% do so after each use, 20% every day, 27% once a week and 18% < weekly. 72% report air drying and 27% use a cloth.

Conclusions: RN spacer washing instructions frequently conform to manufacturer's instructions. Many Pts wash spacers more frequently than instructed but some do not wash at all; many dry with a cloth which may dissipate the electrostatic benefits of air-drying and encourage contamination.

P2196

Patterns of bronchodilator reversibility of FEV1 in asthma and COPD patients

Tushar Sahasrabudhe, Madhur Joshi, Vikas Oswal. *Department of Pulmonary Medicine, Padmashree Dr. D.Y. Patil Medical College, Pimpri, Pune, Maharashtra, India*

SAMA (Short Acting Muscarinic Antagonist) has been said to be superior to SABA (Short Acting Beta Agonist) in COPD whereas SABA is preferred bronchodilator in asthma patients. In practice however we observe that the response is variable for individual patient. Therefore we evaluated improvement in FEV1 in 27 persistent asthmatics and 27 COPD patients in response to inhaled SABA (Salbutamol) and SAMA (Ipratropium) on separate days. These patients were in remission with no exacerbation or change of regular treatment in last 4 weeks. The washout period of their regular treatments was adhered to, before performing their spirometries. SABA produced mean increase of 16.66% and 401 ml in FEV1 of asthmatics vs. -1.44% and -15 ml change in FEV1 of COPD patients. SAMA produced 4.07% and 161 ml increase in FEV1 in asthmatics vs. 8.59% and 166 ml in COPD patients. The similar increase in FEV1 in ml in response to SAMA was more significant in FEV1% due to lower baseline FEV1 in COPD patients. An interesting fact observed was that with the increasing duration of Asthma and COPD, the reversibility with SAMA sequentially increased. With a cut off of more than 8 years of duration of disease, the average improvement in FEV1 in response to SAMA was 453ml (vs. 77 ml in less than 8 year duration) in asthma and 222 ml in COPD, whereas there was no significant change in response to SABA in both groups. There was no correlation with patient's age or baseline FEV1 value. We conclude that inhaled muscarinic antagonists can be a beneficial adjunct to beta agonists in persistent asthmatics with longer duration of asthma.

P2197

Bronchodilator response and airway cytology as parameters for asthma control: A randomized clinical trial Protocol ID: APITA, NCT00597064, on August 1st, 2008

Ana Luisa Fernandes, Maria Marta Amorim, Lilian Ballini Caetano, Alexandre Araruna, Ilka Lopes Santoro. *Medicine- Respiratory Division, Federal University of São Paulo, SP, Brazil*

Background: Clinical control is hard to define in asthma and there is little experimental data on the optimal timing, sequence and magnitude of treatment.

Aim: Measuring the effects of short course of oral corticoid in airway inflammation and spirometry values of asymptomatic asthma patients to define control.

Methods: A double blind randomized clinical trial to observe the change in spirometry, nasal and sputum cytology in patient under combination therapy, receiving either short course of oral corticoid (OC) or placebo(P). Controlled asthma was defined by ACQ<1·5. They were submitted to clinical evaluation, nasal and sputum cytology, spirometry before and after 15±5 days of randomization. Intervention efficacy was defined by improvement of 200 mL in FEV_1.

Results: n=70, 73% female, age 46·8±13·1, 35 received OC and 35 P. Who received OC showed significant improvement in all spirometric parameters and abolished their positive bronchodilator response related to P. The eosinophils count in the nasal and induced sputum also reduced in OC arm. The logistic regression model identify for each one year increasing in age there was a 6% decrease in odds of intervention efficacy, associated to 19 times increase in odds related the usage of oral corticoid (related to placebo), and for each increase of 1% in bronchodilator response was associated with 9% increase in odds of efficacy (p= 0·02).

Conclusion: The RCT allowed define uncontrolled asthma at baseline and the degree of bronchodilator response, younger age and usage of OC were significant predictors for functional improvement at the end of the trial.

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P2198

The effect of smoking on severity of asthma and quality of life in patients treated with inhaled corticosteroids and long-acting b2-agonist (ICS/LABA)

Galina Sergeeva, Alexander Emelyanov. *Lung and Allergy Diseases, Mechnokov North West State Medical Univercity, Saint-Petersburg, Russian Federation*

Background: The airm of this study was to assess the effect of smoking on severity of asthma and quality of life in asthmatic patients treated with ICS/LABA in real clinical practice.
Methods: 122 out-patients (aged 20 - 82 yrs, mean age 55 yrs, 28% males) with moderate-to-severe asthma were treated with medium/high doses of ICS (GINA2010) and LABA in one inhaler for ≥2 years. Quality of life was measured by using Russian version of St. George's Respiratory Questionnaire (SGRQ).
Results: Never smoked 36% of patients, former smokers were 31% and current smokers were 33%. Among never smoked asthmatics 80% were treated by medium doses of ICS/LABA and 20% received high doses. Patients with history of smoking (current or previous) have administered medium doses of ICS/LABA in 68% and high doses in 32%. FEV1 differed in nonsmokers and current smokers: 70% vs 56%, $p<0.05$. History of smoking was associated with FEV1 level ($r=-0.25$, $p<0.05$) and severity of disease ($r=0.3$, $p<0.05$). The difference of Symptom score between smokers (current or former) and nonsmokers was significant (57,6 versus 66,7, $p<0.05$). We revealed the impact of smoking to all scores of SGRQ in females: Symptom (57 vs 65, $p<0.05$), Activity (53 vs 51, $p<0.05$), Impact (39 vs 40, $p<0.01$), and Total score (46 vs 48, $p<0.01$) but not in males.
Conclusion: Smoking is common and may decrease effectiveness of ICS and LABA in asthmatic patients in real clinical practice. There were significant correlations between smoking, severity of asthma and SGRQ scores, especially in females.

P2199

The correction of monocyte-derived neohepatocytes from alpha1 antitrypsin deficient patients

Gillian McNab[1], Robert Stockley[2]. *[1]Centre for Translational Inflammation, University of Birmingham, United Kingdom; [2]Lung Investigation Unit, Queen Elizabeth Hospital Birmingham, United Kingdom*

This study explores the culture of monocyte-derived neohepatocytes from PiZ alpha1 antitrypsin deficient (α_1ATD) patients and homologous replacement using small DNA fragments (SDFs) to correct the Z defect.
Monocytes from 6 patients were de-differentiated with MCSF and IL3 and then differentiated into neohepatocytes with FGF-4. Albumin, urea and α_1AT were measured. SDF enclosing the normal sequence at the PiZ mutation site was generated from genomic DNA of a healthy volunteer. SDFs were transfected into neohepatocytes and cDNA checked for the M or Z message.
No albumin was detected from monocytes. Neohepatocytes secreted 250±50 mg/dL albumin/72h. Monocytes secreted both urea (5±2 μg/dL) and α_1AT (272±42 μg/ml) over 72h. Neohepatocytes secreted 103±30 μg/dL urea and 311±34 μg/ml α_1AT.
Neohepatocytes produced PCR products from Z primers. M SDF treated neohepatocytes generated bands using M primers, indicating the generation of a corrected transcript.
Neohepatocytes transfected with a monocyte transfection kit but no DNA control produced 163±42μg/ml α_1AT in 24h, whereas 20μg M SDF significantly increased secretion (173±41μg/ml/24h, p=0.046, n=3). Using a hepatocyte transfection kit caused further increases in the amount of α_1AT released. Control transfected neohepatocytes produced 322μg/ml/24h α_1AT and 20μg M SDF significantly increased secretion (590±104μg/ml/24h, p=0.026, n=3). Moreover, 50 μgM SDF caused more α_1AT production (886±298μg/ml/24h).
Neohepatocytes can be generated from α_1ATD monocytes. The defective gene can be corrected and is associated with an increase in α_1AT secretion. Development of this technique could be beneficial and protect both the liver and lungs.