Asthma & Allergy SIG
TP001

EARLY LIFE CHLAMYDIAL LUNG INFECTION ENHANCES ALLERGIC AIRWAYS DISEASE THROUGH AGE-DEPENDENT DIFFERENCES IN IMMUNOPATHOLOGY

MR Starkey, JC Horvat, RY Kim, S Phipps, PG Gibson, KW Beagley, PS Foster, PM Hansbro
Centre for Asthma and Respiratory Disease, University of Newcastle, Newcastle, NSW, Australia

Rationale: Asthma typically originates in early-life and the impact of infection during immunological maturation is a critical factor in disease pathogenesis. Exposure to specific pathogens such as *Chlamydia* may alter immunological programming leading to predisposition.

Methods: We investigated the effect of early life infection on hallmark features of asthma in later-life using an acute mouse model of Ovalbumin-induced allergic airways disease (AAD). Groups were infected with *C. muridarum* as neonates (<24hrs), infants (3 wks) or adults (6 wks) and subjected to AAD 45 days after infection.

Results: Early-life chlamydial infection enhanced the development of hallmark features of AAD in later-life. Notably infection (both neonatal and infant) increased mucus-secreting cell hyperplasia, airways hyper-responsiveness and IL-13 expression in lungs of adult mice after antigen inhalation. Importantly, these effects correlated with differential alterations in T-cell and dendritic cell (DC) responses and lung structure. Infection of neonates suppressed pulmonary inflammatory responses with attenuated eosinophil influx, T-cell and DC responses. However, neonatal infection increased systemic IL-13 release and induced substantial alterations in lung structure. By contrast, infant infection augmented allergic inflammation with increases in eosinophilic inflammation, T-cell and DC responses but without substantially altering lung structure. Adult infection had no effect on AAD in later life.

Conclusion: Early life infection enhances pivotal features of AAD through age dependent, differential and permanent affects on immune responses and lung structure.

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TP002

PREVENAR SUPPRESSES ALLERGIC AIRWAYS DISEASE THROUGH THE EXPANSION OF REGULATORY T CELLS

Alison Thorburn[1], Brendan O'Sullivan[2], Ranjeny Thomas[2], Paul Foster[1], Peter Gibson[1], Philip Hansbro[1]

[1]*Centre for Asthma and Respiratory Disease and Hunter Medical Research Institute, The University of Newcastle, NSW, 2300*

[2]*Immunology and Metabolic Medicine, Diamantina Institute for Cancer, QLD, 4102*

Current therapeutics for asthma treat the symptoms but not the underlying cause of disease. Our recent finding that exposure to *Streptococcus pneumoniae* may suppress pro-asthmatic responses led us to investigate whether the current human *S. pneumoniae* vaccines, Pneumovax and Prevenar, may be used to suppress asthma using mouse models of ovalbumin-(OVA)-induced allergic airways disease (AAD).

Methods: AAD was induced by intraperitoneal sensitisation and intranasal challenge with OVA. At the time of OVA sensitisation, Prevenar or Pneumovax were delivered intranasally either with or without CpG.

Results: Prevenar, but not Pneumovax, suppressed the hallmark features of AAD, including eosinophil influx, OVA-specific T helper (Th) 2 cytokine release in lung draining lymph nodes and spleen, mucus hypersecretion, total serum IgE and airways hyperresponsiveness. We then investigated whether regulatory T cells (Tregs) were important in the suppression of AAD. Prevenar increased the number of CD4+CD25+FoxP3+ expressing Tregs in the lungs, lymph nodes and spleens. The release of immuno-suppressive cytokines IL-10 and TGF-β from lymph nodes was reduced. Proliferation assays demonstrated that Tregs suppressed allergic effector T cell proliferation regardless of Prevenar treatment, suggesting that IL-10 and TGF-β concentrations are not limiting for suppressor function.

Conclusion: A currently available human vaccine increases the infiltration of Tregs and suppresses AAD, and may be useful as a novel therapy for asthma.

Supported by Asthma NSW, Asthma CRC

Conflict of Interest: No

TP003

FEMALE MICE EXPOSED TO INFLUENZA A HAVE GREATER INFLAMMATION AND LUNG RESPONSIVENESS COMPARED TO MALE MICE

Alexander N Larcombe, Elizabeth M Bozanich, Debra J Turner, Graeme R Zosky, Peter D Sly

[1] *Telethon Institute for Child Health Research, Centre for Child Health Research, University of Western Australia, Perth, Western Australia, 6008.*

Background: Murine models have been extensively used to study lung development and physiology, and it is apparent that there is sexual dimorphism in various aspects of these processes. Sex differences in immune function are well established in both animals and humans, with males typically displaying weaker humoral and cell-mediated immune responses.

Methods: We infected male and female weanling (3 wks old) and adult (8 wks old) BALB/c mice with influenza A and measured lung function, hyperresponsiveness to methacholine (Mch) and inflammatory responses during the acute infectious stage (4 days after inoculation) and after recovery (day 21).

Results: During the acute infectious stage, there were significant effects of influenza infection on airway resistance (R_{aw}), tissue damping (G), tissue elastance (H) and number/type of inflammatory cells in the bronchoalveolar lavage (BAL) for both males and females of both ages when compared to media controls. Greater responses were seen in adult females than in adult males (e.g. G: $p = 0.020$; H: $p = 0.016$; macrophages: $p = 0.003$; neutrophils: $p = 0.003$), and in weanling females compared to weanling males, especially in the lung parenchyma. Most parameters had returned to baseline levels 21 days after influenza inoculation in both age groups and sexes.

Conclusions: We've shown increased lung hyperresponsiveness and inflammatory responses in female mice exposed to influenza A compared to male mice. These results are contrary to most previous research, which indicate that males are generally more susceptible to respiratory viral infections.

Supported by the NHMRC.

TP004

HAEMOPHILUS INFLUENZAE INFECTION INDUCES FEATURES OF NEUTROPHILIC ASTHMA

Ama-Tawiah Essilfie, Jodie L Simpson, Margaret Dunkley, Paul S Foster, Peter G Gibson, Phil M Hansbro
Priority Research Centre for Asthma & Respiratory Disease & Hunter Medical Research Institute, University of Newcastle, Newcastle, NSW 2300

Approximately 20% of asthmatics have neutrophilic asthma. These patients have intense neutrophilic inflammation, reduced airways hyperresponsiveness (AHR) and high levels of interleukin (IL)-8. *Haemophilus influenzae* (Hi) colonisation is common in neutrophilic asthmatics however it is not known if colonisation induces neutrophilic asthma or if these patients are predisposed to colonisation. We investigated the nature of this association using mouse models of Hi lung infection and ovalbumin (OVA)-induced allergic airway disease (AAD).
Method: AAD was induced by intraperitoneal OVA sensitisation (day 0) and intranasal challenge (day 12-15), and subsequently characterised (day 16). Live Hi was administered 10 days before, during or 10 days after OVA sensitisation.
Results: At all time points, infection significantly reduced OVA-specific Th2 cytokine production, eosinophil influx and AHR, but induced a potent neutrophil influx into the airways, with increases in IL-17 production. Bacterial clearance was severely hampered in groups that were infected before sensitisation.
Conclusion: This combined model of AAD and Hi infection is consistent with the clinical phenotype of neutrophilic asthma and suggests that Hi infection may promote the development of neutrophilic asthma.

Supported by Asthma NSW and HMRI
Nomination: Ann Woolcock Young investigator Award
Conflict of interest: No

TP005

NEONATAL ANTIGEN AND VIRAL EXPOSURE ALTERS AIRWAY AND PARENCHYMAL RESPONSIVENESS IN ADULT MICE

Elizabeth M Bozanich, Alexander N Larcombe, Debra J Turner, Graeme R Zosky, Peter D Sly
Division of Clinical Sciences, Centre for Child Health Research, Perth WA 6008

Epidemiological data suggests a link between viral infection early in life and long-term symptoms of asthma. We aimed to determine if viral infection in early life exacerbates allergen induced lung hyperresponsiveness (HR) later in life.
Methods: BALB/c mice were exposed to OVA (5µL 2mg/mL or saline i.n.) on d0 then inoculated d7 with Influenza A Mem/1/71(H3N1) (10µL $10^{3.8}$pfu or media). Mice were boosted at 4wks then challenged with 6 aerosols at 8wks (OVA or saline). AHR was assessed to inhaled MCh (0.1-30mg/mL) 24hrs after the final aerosol using a small animal ventilator and a modification of the forced oscillation technique. The Constant Phase Model was fitted to Respiratory Impedance (Zrs) data to produce airway (R_{aw}) and tissue parameters (G, tissue damping; H, tissue elastance).
Results: Both neonatal exposure to Flu (R_{aw}, $p=0.017$; H, $p<0.001$) and antigen (Raw, $p=0.008$; H, $p=0.003$) individually induced HR in adult mice. However, there was neither an additive or synergistic effect of the two exposures (R_{aw}, $p=0.531$, H, $p=0.311$).
Conclusions: These findings demonstrate that exposure to Influenza A virus or OVA during the neonatal period alter lung mechanics when challenged with MCh in adult mice. The addition of viral infection in early life did not exacerbate allergen induced HR.

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TP006

BRONCHODILATOR ACTIONS OF ROSIGLITAZONE IN ISOLATED MOUSE TRACHEA ARE PPARγ-INDEPENDENT

Jane E Ward[1], J Tran[1], SG Royce[2], ML Tang[2], SR Bailey[3]
[1]Pharmacology, University of Melbourne, [2]Immunology, Royal Children's Hospital, [3]Veterinary Science, University of Melbourne, Vic. 3010

Rationale: Peroxisome Proliferator Activated Receptor γ (PPARγ) is a novel target for asthma treatment, with increased expression in bronchial biopsies from asthmatics. The aim of this study was to determine whether PPARγ ligands rosiglitazone (RGZ), ciglitazone (CGZ) or 15-deoxy-PGJ_2 cause direct bronchodilation of mouse trachea through PPARγ activation.
Methods: Tracheal segments mounted in a wire myograph were contracted with methacholine (20%, 75% or 100% maximum response) before addition of PPARγ ligands (0.3-100 μM, 10 min intervals) or salbutamol (0.1 nM-100 μM, 2 min intervals). The effects of RGZ were compared in the absence or presence of PPARγ antagonist, GW9662 (1 μM), indomethacin (10 μM), EP_2 antagonist, AH6809 (3 μM) and/or EP_4 antagonist L-161982 (1 μM).
Results: RGZ caused concentration-dependent and maximal relaxation that was not prevented by GW9662 (threshold response 3 μM, EC_{50} ~ 50 μM, n=5, one way ANOVA, $P<0.001$). CGZ was less effective and 15-deoxy-PGJ_2 did not mediate relaxation. The potency of RGZ was attenuated by indomethacin (n=6, 2 way ANOVA, $P<0.05$), but not by EP_2/EP_4 antagonists. Relaxation to salbutamol was faster than RGZ, but salbutamol and not RGZ lost potency and maximum agonist effect when tissues were maximally contracted. In a mouse model of chronic airways disease, RGZ-induced relaxation was maintained in trachea following ovalbumin-challenge, despite significantly greater contraction to methacholine than in trachea from saline-challenged mice (n=8, $P<0.05$).
Conclusions: Relaxation to RGZ in mouse trachea occurs by a mechanism independent of PPARγ, resistant to functional antagonism and maintained in inflamed airways. Further work is necessary to define indomethacin-sensitive and -insensitive bronchodilator mechanisms for RGZ and similar compounds, to support further exploration of their therapeutic potential in asthma.
Grant support: NHMRC

TP007

BOTH SEX AND TYPE OF VIRUS INFLUENCE LUNG INFLAMMATORY RESPONSES IN BALB/C MICE

Rachel Foong[1], Elizabeth M Bozanich[1], Rosa C Gaulano[2], Anthony Kicic[1], Debra J Turner[1], Graeme R Zosky[1], Alexander N Larcombe[1], Peter D Sly[1]
[1]*Telethon Institute for Child Health Research, Centre for Child Health Research, University of Western Australia, Perth, Western Australia, 6008.*
[2]*Department of Pharmacology, University of Melbourne, Victoria, Australia, 3010.*

Background: The roles of host-specific versus viral-specific factors in determining inflammatory responses in the lung to acute viral infection remain unclear. We examined the role of age, sex and type of virus in determining the inflammatory responses to acute viral infection using a mouse model.
Methods: We infected BALB/c mice of both sexes with $10^{4.5}$pfu flu (A/Memphis/1/71 H3N2) or $5x10^{6}TCID_{50}$ RV (RV1b serotype). Bronchoalveolar lavage (BAL) samples were taken from groups of mice through the acute phase of infection and following recovery. BALs were used to ascertain total cell counts (TCC) and differential cell counts (macrophages: mac, neutrophils: neut) under light microscopy.
Results: Inflammatory responses were more severe and prolonged in mice inoculated with flu compared to those inoculated with RV. Both male and female flu inflammatory responses peaked at day 4, but females had significantly higher responses (TCC, $p = 0.001$; mac, $p = 0.003$; and neut $p= 0.008$). In contrast, the peak of TCC differed between sexes inoculated with RV (female D2; male D4), whereas the pattern of neutrophilic inflammation was the same. The magnitude of the response was the same between sexes (TCC, $p = 0.71$; mac, $p = 0.52$; neut, $p = 0.3$).
Conclusions: These data clearly demonstrate that host-specific and virus-specific effects are important in the inflammatory responses in the mouse lung.

Supported by the NHMRC

TP008

NEUTROPHIL INFLUX DURING CHLAMYDIAL LUNG INFECTION DETERMINES THE PHENOTYPE OF ALLERGIC AIRWAYS DISEASE (AAD)

JC Horvat, MR Starkey, KW Beagley, JA Preston, PG Gibson, PS Foster and PM Hansbro
University of Newcastle, Newcastle, NSW, Australia

C. pneumoniae is linked with asthma, however, it is unknown how a Th1-inducing infection is associated with Th2-mediated asthma. We investigated the association using models of chlamydial lung infection and ovalbumin (Ova)-induced AAD.

Adult mice were infected 45 or 7 days before intraperitoneal (IP) Ova sensitisation. AAD was induced by intranasal Ova challenge 12-15 days after sensitisation. Therefore, mice had a resolved or ongoing infection at sensitisation. The affect of pulmonary neutrophil influx on changes induced by an ongoing infection was also examined by treating mice IP with anti-Keratinocyte Chemokine and anti-Macrophage inflammatory protein 2 antibodies during infection. Seven days after infection, antibody-treated mice were sensitised and challenged with Ova. Features of AAD were compared with un-infected and non-sensitised controls.

Ongoing, but not resolved, infection induced Ova-specific Th1 responses that promote neutrophilic and suppress eosinophilic inflammation. During this neutrophil-dominated AAD, mucus secreting cell (MSC) numbers and AHR were reduced. Depletion of pulmonary neutrophil influx reversed increases in Th1 responses and decreases in MSC numbers and AHR during infection-associated AAD. Significantly, infected, antibody-treated mice no longer mounted robust pulmonary or systemic neutrophil responses upon the induction of AAD, despite cessation of antibody treatment 10 days earlier. These changes correlated with decreased IL-12 and IL-17 expression, increased thymus and activation-regulated chemokine and augmented antigen presenting cell activation compared to infected, isotype-treated controls.

Ongoing chlamydial respiratory infections modify key allergen-specific immune responses in AAD with the composition of cellular inflammatory responses to infection crucial in determining the outcome of allergic phenotype.

Supported by the NHMRC

TP009 – paper withdrawn

TP010

TUMSTATIN – A NON-COLLAGENOUS DOMAIN OF COLLAGEN IV – EFFECTS ON INFLAMMATION AND ANGIOGENESIS

Markus Weckmann[1], Justine Lau[1], Karryn Grafton[1], Brian Oliver[1], Nicole Hansbro[2], Paul Foster[2], Phil Hansbro[2], Judith Black[1], Janette Burgess[1]
[1] *Respiratory Research Group, University of Sydney, Sydney NSW 2006*
[2] *Discipline of Immunology & Microbiology School of Biomedical Sciences University of Newcastle, NSW 2300*

Remodelling of the airway is a key feature of asthma and involves, among other features, increases in mucus producing cells and angiogenesis. We have previously shown that in lung tissue of patients with asthma, the non-collagenous domain of the collagen IV a 3 molecule (tumstatin) is not detectable. The development of angiogenesis may be suppressed by tumstatin both *in vitro* and *in vivo*. We investigated the potential therapeutic use of tumstatin in a chronic mouse model of ovalbumin (OVA)-induced allergic airway disease (AAD). The effects of tumstatin administration on AAD were determined by assessment of airway hyperresponsiveness (AHR), the levels of the inflammatory cytokine IL-13 and the angiogenic factor VEGF by specific immuno-fluorescence as well as enumeration of eosinophils and mucus-secreting cells in lung sections. Repeated administration of tumstatin during chronic OVA exposure significantly reduced allergen induced AHR (n=6), led to a decrease of 13.5% in VEGF (n=4) levels, and reversed the IL-13 increase of 9.4% in allergic animals back to baseline level. Furthermore, treatment also substantially decreased eosinophilic infiltrates (46.2%, n=6) and mucus over-production (52.9%, n=6). These results suggest that tumstatin suppresses hallmark features of allergen specific immune response in the lung and may have therapeutic applicability.

Supported by the CRC for Asthma and Airways
Conflict of Interest: No

TP011

EXTRACELLULAR MATRIX WITHIN THE AIRWAY SMOOTH MUSCLE LAYER IN ASTHMA

J Elliot[1], R Jones[1], M Carroll[1], T Mauad,[2] M Dolhnikoff,[2] M Abramson,[3] K McKay,[4] T Bai,[5] F Green[6] and A James[1,7]

[1]West Australian Sleep Disorders Research Institute, Nedlands, Western Australia 6009; [2]Sao Paulo University, Sao Paulo, Brazil; [3]Monash University, Melbourne, Australia; [4]Children's Hospital at Westmead, Sydney, Australia; [5]University of British Columbia, Vancouver, BC, Canada; [6]University of Calgary, Calgary, Alta, Canada; [7]Medicine and Pharmacology, University of Western Australia.

The reported increase in the amount of airway smooth muscle (ASM) in asthma may be due to hypertrophy and/or hyperplasia of ASM cells or increased extracellular matrix (ECM) between ASM cells within the ASM layer.

Aim: To estimate the volume fraction of ECM within the ASM layer using ultra-thin (0.5 μm) sections and to calculate and compare the volume of ECM per airway length (mm) in post-mortem tissues from control subjects (C, n=42); nonfatal (NFA, n=39) and fatal (FA, n=29) cases of asthma.

Methods: Point counts of ECM were made within the ASM layer on transverse airway sections stained using the Masson's trichrome technique and the area fraction of ECM (f_{ECM}) was estimated. The volume of ECM per airway length was then calculated ($V_{ECM} = A_{ASMlayer} \times f_{ECM} \times 1mm$). Basement membrane perimeter (Pbm) was used to indicate airway size.

Results: Table shows results (case means) for large airways.

	Sex	Airway	Pbm	ASM layer/Pbm	f_{ECM}	V_{ECM}/1mm
	M/F	n (total)	mm	mm^2		(mm^3)
C	28/14	123	15 ± 6	0.035 ± 0.016	0.21 ± 0.17	0.13 ± 0.10
NFA	19/20	133	14 ± 6	0.043 ± 0.023**	0.18 ± 0.16	0.14 ± 0.12
FA	19/10	92	16 ± 5	0.065 ± 0.026*	0.17 ± 0.12	0.18 ± 0.13

Mean ± SD. (one-way ANOVA) *p <0.05 for C v FA and **NFA v FA.

The volume fraction of ECM and the absolute volume of ECM were not significantly different between case groups, although there was a trend for increased ECM volume in cases of fatal asthma. Results were similar for small airways.

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Conflicts: Nil.

TP012

EXPRESSION OF COX- 1 VARIANT IN EOSINOPHILS AND DENDRITIC CELLS IN ASPIRIN SENSITIVE ASTHMATICS

Jing Shi, Bradley Shelton, Faang Cheah, Joanne Horn, Carolyn Williams, Philip J Thompson. *Lung Institute of Western Australia, Centre for Asthma, Allergy & Respiratory Research and CRC for Asthma and Airways, UWA*

Introduction: Cyclooxygenase (COX)-1 is a key enzyme involved in the synthesis of prostaglandins (PGs) which are important mediators in asthma. Approximately 10% of adult asthma patients have aspirin sensitivity. A splice variant of COX-1 is expressed significantly higher in aspirin sensitive asthmatics (ASA) and aspirin tolerant asthmatics (ATA) compared with healthy subjects (HS). We hypothesise that this variant alters production of PGs in particular PGE_2. Our aim was to determine whether COX-1 variant mRNA expression in eosinophil and monocyte derived dendritic cells (MoDCs) differs between ASA, ATA and HS.

Methods: Six patients with ASA, ATA and HS were recruited. Eosinophils and MoDCs were isolated from peripheral blood using CD16 and CD14 MicroBeads. COX-1 variant and wild-type mRNA expression levels were measured in eosinophils & MoDCs using two-step RT-PCR and real time quantitative PCR.

Results: COX-1 splice variant mRNA transcripts were, for the first time, found to be expressed in circulating eosinophils and MoDCs in all patient groups studied. Preliminary results showed that COX-1 variant mRNA expression levels were approximately 30% of total COX-1 production in HS. Furthermore there was a difference in both the variant and wild-type COX-1 expression levels in asthmatics compared with HS.

Conclusion: Preliminary data confirmed the presence of COX-1 aberrant splice variant expression at varying levels in eosinophils and MoDCs and that this expression is influenced by phenotype. Further studies are underway to confirm these data and the functional significance of COX-1 variant mRNA expression on PG production in particular PGE_2 in ASA, ATA and HS.

Supported by the Lung Institute of WA and the CRC for Asthma and Airways
Conflict of Interest: No

TP013

PRO-FIBROTIC MEDIATORS INCREASE REMODELLING OF COLLAGEN GELS BY HUMAN AIRWAY SMOOTH MUSCLE CELLS

Hayat Dagher, Xin Li, Simon Foster, Nadene Tam, Jane Ward
Department of Pharmacology, University of Melbourne, Vic 3010

Cell-mediated remodelling of three-dimensional collagen gels has been used to assess regulation of pro-fibrotic interactions between cells and their surrounding extracellular matrix. We have previously demonstrated reductions in gel area ("contraction") associated with condensation of collagen fibrils in gels seeded with airway smooth muscle (ASM) cells. In this study, we addressed the regulation of this remodeling by transforming growth factor β (TGFβ) and endothelin-1 (ET-1).

Methods: Serum-deprived human ASM was cast in Type I collagen gels (1.25 x 10^5 cells/0.5 ml). The effects of TGFβ (0.1 – 100 pM) and ET-1 (0.1 – 100 nM) on ASM-mediated reductions in area were determined over 72 hr, in the absence and presence of TGFβ blocking antibody, selective ET_A and ET_B receptor antagonists, and the protein synthesis inhibitor cycloheximide.

Results: Changes in ASM gel area were evident at > 4 hr, with maximum reduction observed by 72 hr (by 33±10%, mean ± SEM, n=6). The rate and extent of contraction was increased by TGFβ, with a threshold response of 0.3 pM. ET-1, acting via ET_A receptors was less potent, with maximal area reduction at 10 nM (by 58±11%, $P<0.05$ *cf* unstimulated). Although CHX did not affect ASM-mediated gel contraction in the absence of TGFβ and ET-1, the augmented response to both mediators required protein synthesis (reduction in area for ET-1+CHX, 33±10%, n=6, $P>0.05$ *cf* unstimulated). However, ET-1-mediated contraction was not associated with increased TGFβ release.

Conclusions: Further studies are required to clarify common or divergent signaling pathways implicated in TGFβ- and ET-1-induced collagen gel remodeling. Under the influence of these pro-fibrotic mediators elevated in asthma, ASM has greater potential to increase collagen density in muscle bundles and in turn, to increase internal resistance in the airways.

Grant support: NHMRC, Asthma Foundation of Victoria

TP014

MAST CELL PRODUCTS REGULATE AIRWAY SMOOTH MUSCLE CELL CXCL10 RELEASE

H Alkhouri[1], K Tong[1], V Cha[1], LM Moir[2], CL Armour[1], JM Hughes[1]
Respiratory Research Group, [1]Faculty of Pharmacy and [2]Discipline of Pharmacology, University of Sydney; [2]Woolcock Institute of Medical Research, Sydney

CXCL10 (IP-10) attracts mast cells (MC) to the airway smooth muscle (ASM) in people with asthma. MC release a variety of granule products, lipid mediators and cytokines that may interact with ASM cells and alter their secretory function contributing to MC localization to the asthmatic airway.

Aim: To determine the effects of MC products on cytokine induced IP-10 release by ASM cells from donors with and without asthma.

Methods: MC were isolated from lung samples from 3 donors and stimulated with IgE/anti-IgE. Supernatants (SN) were collected after 2 and 24h and the MC were lysed. Serum deprived ASM cells were stimulated with IFNγ (10ng/ml) for 30 min before adding either histamine (10μM), tryptase (1nM) or MC SN/lysates (40% in DMEM+10%FBS) with appropriate controls. The protease inhibitor leupeptin (50μM) was added to specific tryptase-treated wells after 3 hours. IP-10 levels in all ASM SN and MC SN/lysates were measured by ELISA.

Results: Histamine did not alter IFNγ induced IP-10 release whereas tryptase significantly reduced it's detection down to 8±4% ($p<0.0001$) and 10±4% ($p<0.0001$) in asthmatic and non-asthmatic ASM SN respectively, and also reduced recombinant human IP-10 by 50%. In non-asthmatic ASM, 0-2h MC SN significantly reduced IFNγ induced IP-10 levels to 54±18% ($p<0.05$) of control, while the 2-24h MC SN significantly increased IFNγ induced IP-10 release to 218± 50% ($p<0.05$). The reduction in IP-10 was completely reversed if tryptase was heat inactivated or leupeptin was added.

Conclusions: Mast cell products differentially modulated IP-10 secretion by ASM cells from donors with and without asthma. Thus mast cells may regulate their own recruitment to the ASM in asthma.

Funded by: NHMRC

TP015

RHINOVIRUS INDUCED EXTRACELLULAR MATRIX DEPOSITION IN PRIMARY BRONCHIAL EPITHELIAL CELLS AND FIBROBLASTS

Brian G Oliver[1,2,6], Sebastian Johnston[3], Nicholas JC King[4], Peter Wark[5], Sam Lim[2], Janette K Burgess[1,6], Judith L Black[1,6], Curtis Kuo[1,2]
[1] Respiratory Research Group and [2] ANZAC Research Institute, Australia, [3] Imperial College London, UK, [4] Department of Pathology, The University of Sydney, Australia, [5] Hunter Medical Research Institute & The University of Newcastle Australia and [6] Woolcock Institute of Medical Research, Australia

Introduction: A hallmark of asthma is remodelling of the airways. Viral infections may promote the development of asthma and are the most common causes of asthma exacerbations. The development of asthma is closely associated with the development of remodelling which is accompanied by increased deposition of extracellular matrix (ECM) proteins. The effect of rhinovirus (RV) infection on remodelling is an area not well understood. In this study we examined whether RV infection induces remodelling assessed by extracellular matrix deposition.
Methods: Primary human bronchial epithelial cells (n= 6-9) and lung parenchyma fibroblasts (n= 4-5) were isolated from patients undergoing resection or transplantation and infected with Rhinovirus-16. Changes in extracellular matrix proteins were measured by ELISA and transwells used to measure cell migration. Proliferation was assessed by MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium] assay.
Results: Rhinovirus increased deposition of fibronectin (54%, $p<0.01$), perlecan (55%, $p<0.01$) and collagen IV (29%, $p<0.05$) in primary bronchial epithelial cells at a multiplicity of infection (MOI) of 0.15 (n=6-9). In addition collagen V increased by 109% ($p<0.005$) at a MOI 0.15 in epithelial cells and 77% ($p<0.01$) at an MOI 1 in fibroblasts. Matrix bound VEGF was increased by 172% at an MOI 0.15 in epithelial cells and 90.7% at an MOI 1 in fibroblasts (n=4). Proliferation of bronchial epithelial cells and fibroblasts on their respective RV modulated matrix was decreased by 11.4% at MOI 0.15 and 8.44% at MOI 1 respectively (n=3). The RV altered fibroblast ECM also inhibited fibroblast migration (n=4). Furthermore,

soluble factors released by RV infected primary bronchial epithelial cells inhibited fibroblast migration (n=5).
Conclusion: The changes in production of ECM proteins, cell migration and proliferation observed in rhinovirus infected epithelial cells and fibroblasts *in vitro* demonstrate that RV has the potential to promote remodelling of the airways.

TP016

THE EFFECT OF VARIABLE TISSUE DEPTH ON ESTIMATION OF AIRWAY SMOOTH MUSCLE CELL VOLUME

R Jones,[1] J Elliot,[1] M Carroll,[1] T Mauad,[2] M Dolhnikoff,[2] M Abramson,[3] K McKay,[4] T Bai,[5] F Green[6], A James[1]

[1]West Australian Sleep Disorders Research Institute, Nedlands, Western Australia 6009; [2]Sao Paulo University, Brazil; [3]Monash University, Melbourne, Victoria; [4]Children's Hospital at Westmead, Sydney; [5]University of British Columbia, Vancouver, Canada; [6]University of Calgary, Alta, Canada.

It is unclear if an increase in the volume of airway smooth muscle (ASM) cells (hypertrophy) contributes to the increased thickness of the ASM layer in asthma. Mean ASM cell volume can be estimated by dividing a measured volume of ASM by the number of cells in that volume in thick tissue sections (30μm). However, there is variation in the depth of ASM tissue within thick sections.

Aim: To examine the effect of tissue depth on the estimation of ASM cell volume.

Methods: Post-mortem airways from control subjects (C n=21); nonfatal (NFA n=20) and fatal (FA n=21) cases of asthma were studied. On 30μm sections stained with hematoxylin, the volume density (N_V) of ASM cell nuclei was estimated using the optical dissector method and stereological software. ASM cells were counted within each optical disector volume by scanning through the tissue depth. Partial high power fields (HPF) in which the ASM occupied only part of the tissue depth were recorded. The mean cell volume ($V_C = 1/N_V$) was calculated both <u>including</u> and <u>excluding</u> partial depth HPF.

Results: The mean ASM cell volume was similar in all groups (Table), but for all groups was significantly reduced when partial depth HPFs were excluded.

*p <0.05	**Estimated mean ASM cell volume (μm^3 / $x10^3$)**			
	Large Airways (n=213)		Small Airways (n=130)	
	All HPF	Partial HPF Excluded	All HPF	Partial HPF Excluded
C	6.4 ± 2.6	5.5 ± 2.4 *	5.4 ± 1.3	4.2 ± 1.2 *
NFA	5.8 ± 1.5	5.1 ± 1.3 *	5.4 ± 1.5	4.7 ± 1.4 *
FA	6.4 ± 1.7	5.7 ± 1.6 *	5.5 ± 1.5	4.8 ± 1.6 *

Conclusion: The discontinuous nature of ASM both around and along the length of airways necessitates correction for varying tissue thickness (space between muscle bundles) within the ASM layer when estimating ASM cell volume. **Support:** NHMRC Australia (Grants #343601, #446800) **Nomination:** Nil. **Conflicts:** Nil.

TP017

MOLECULAR MECHANISMS OF AIRWAY NEUTROPHILIA FOLLOWING A LOW ANTIOXIDANT DIET

KJ Baines, LG Wood, PG Gibson
Priority Research Centre for Asthma and Respiratory Disease, School of Medicine & Public Health, University of Newcastle, Newcastle, NSW & Department of Respiratory & Sleep Medicine, HMRI, John Hunter Hospital, Newcastle, NSW

Rationale: Withdrawal of antioxidants through consumption of a low antioxidant diet has been reported to increase airway neutrophilic inflammation and worsen symptoms of asthma; however the mechanisms of this are unknown.
Objective: To investigate differential gene expression of induced sputum samples collected from subjects with asthma before and after 10 days on a low antioxidant diet.
Methods: Induced sputum samples were collected at baseline and after participants consumed a low antioxidant diet for 10 days. Subjects (n=10) were selected for gene expression analysis if they had a >10% increase in sputum neutrophils after the dietary change. Genome wide gene expression profiles were generated from sputum RNA samples using Illumina Sentrix Humanref-8 expression microarrays, and data was analysed using GeneSpring 10. Differentially expressed genes were defined by both significance ($p<0.05$) using paired t test and change of greater than 1.5 fold.
Results: There were 104 genes differentially expressed following the dietary change. 22 genes were upregulated and 82 genes were downregulated post low antioxidant diet. Upregulated genes were involved in the innate immune response and included the innate immune receptors TLR2, IL1R2, CD93, the signaling molecules IRAK2, IRAK3 and neutrophil proteases MMP25 and CPD. Downregulated genes included those involved in endogenous antioxidant defences (GSTA1, GSTA2) and protease inhibition (SLPI, SERPINB3).
Conclusion: Withdrawal of dietary antioxidants in asthma induces significant alterations in airway gene expression characterised by upregulation of genes involved in the innate immune response. These results indicate that diet influences airway inflammation in asthma through potentiation of innate immune gene expression.

TP018

MOLECULAR MECHANISMS OF RHINOVIRUS INFECTION OF PRIMARY BRONCHIAL EPITHELIAL CELLS

KJ Baines[1,2], TV Grissell [1,2], PG Gibson[1,2], PAB Wark[1,2]
[1]Centre for Asthma and Respiratory Disease, Hunter Medical Research Institute, University of Newcastle, Newcastle, NSW, Australia, [2]Department of Respiratory & Sleep Medicine, HMRI, John Hunter Hospital, Newcastle, NSW, Australia

Rationale: The majority of asthma exacerbations are associated with rhinovirus infection, however the molecular mechanisms of this remain unclear.
Aim: To investigate changes in gene expression caused by rhinovirus infection of primary bronchial epithelial cells in vitro.
Methods: Bronchial brushings were obtained from participants with asthma (n=3) and healthy controls (n=3). Primary bronchial epithelial cells were treated with or without RV43 (MOI 10), RV1B (MOI 2), or UV inactivated RV43 and RV1B, and collected at 24 hours post-infection. RNA was extracted, amplified and genome wide gene expression profiles were generated using Illumina Sentrix Humanref-8 expression microarrays. Data was analysed using GeneSpring 10 where differentially expressed genes were defined by tests for both significance (ANOVA) and >1.5 fold change.
Results: Gene expression profiles were generated from epithelial cell cultures for 4 treatment groups (n=6 per group), including media, UV inactivated virus, RV43 and RV1B. There were 37 and 42 genes altered due to infection with RV1B and RV43 respectively. Significant overlap (31 genes) existed between the responses to RV1B and RV43. There was upregulation of genes involved in early virus signalling (e.g. IFIH1 and DDX58), genes induced by type 1 interferon (e.g. CXCL10, IFIT1, IRF7, and OAS1), genes involved in cell signalling (e.g. STAT-1), as well as genes associated with the inflammatory response (e.g. IL-8 and TNF).
Conclusion: Rhinovirus infection significantly alters the expression of a subset of genes involved in immune and antiviral responses in primary bronchial epithelial cells.

TP019

ASSOCIATION WITH A VARIANT ON 17Q21 WITH ASTHMA SEVERITY AND AGE OF ONSET IN AN AUSTRALIAN POPULATION

Carolyn Williams[1,2,3], Amber Bullock[1,2,3], Jing Shi[1,2,3], Joanne Horn[1,2,3], Bernadette Bradley[1,2,3], Bradley Shelton[1,2,3] and Philip Thompson[1,2]. *[1]Lung Institute of Western Australia and Centre for Asthma Allergy and Respiratory Research, School of Medicine and Pharmacology, University of Western Australia, [2]Cooperative Research Centre for Asthma and Airways, Australia, [3]Western Australian Institute for Medical Research, Perth, Australia.*

Introduction: Studying the genetics of complex multifactorial diseases such as asthma poses significant challenges. Identifying genes with a major contributing role in the pathogenesis of disease and their interactions is fundamental to understanding disease susceptibility and progression. A recent association with rs7216389 on chromosome17q21 has been reported with paediatric asthma. We have genotyped a tagged SNP in an extended LD block around rs7216389 in a large highly phenotyped Australian asthma cohort to further elaborate on these findings.

Methods: We assessed the association of rs7216389 with asthma in a cohort of 647 asthma patients and 564 non-asthma controls. Samples were stratified according to age of onset, sex, atopy status and asthma severity.

Results: Genotyping results indicate a significant association between a non-synonymous SNP in this region and early age of onset of asthma but not adult onset asthma. Association with asthma severity was also observed but only amongst the early age of onset subjects. There was no association with atopy.

C**onclusion:** These results demonstrate that the association of rs7216389 is limited to early onset asthma and asthma severity in this group. This finding has the potential to assist in detecting early onset individuals who may be at greater risk of developing severe asthma. Important functional studies related to this variant will help clarify the mechanism by which the 17q21 locus contributes to asthma.

Supported by the Lung Institute of WA and CRC for Asthma and Airways

Conflict of Interest: No

TP020

INCREASED EXPRESSION OF RAGE, AND DECREASED EXPRESSION OF SOLUBLE RAGE, MAY BOTH CONTRIBUTE TO SEVERE ASTHMA

Bradley P Shelton, Michelle C Davies, Carolyn Williams, Philip J Thompson
Lung Institute of Western Australia Inc, Perth, Australia

Activation of the receptor for advanced glycation endproducts (RAGE) leads to prolonged NF-κB signalling and has been associated with chronic inflammation. We have previously identified an association between the RAGE -374T>A promoter polymorphism and both severe and aspirin-sensitive asthma. The effect of this polymorphism on RAGE expression in asthma has not previously been determined. Further, RAGE activation may be offset by an endogenous soluble "decoy" receptor (esRAGE) that binds ligand but lacks signalling capabilities. The role of esRAGE in asthma has not previously been examined.

Objective: Determine RAGE and esRAGE expression levels and correlate with asthma severity and patient genotype at the RAGE -374 loci.

Methods: Serum was collected from asthmatic and non-asthmatic subjects previously genotyped for the -374T>A polymorphism. Levels of circulating RAGE were assessed using an enzyme-linked immunosorbent assay (ELISA).

Results: Preliminary results revealed an almost 1.5-fold increase in RAGE expression levels associated with the -374T>A homozygous mutant genotype. We have previously shown this genotype to be associated with both severe and aspirin-sensitive asthma. Further, decreased levels of esRAGE expression were observed in severe asthmatics relative to healthy control subjects.

Conclusions: Results from this study suggest that the RAGE -374A allele, previously associated with severe and aspirin-sensitive asthma, may be contributing to chronic inflammatory asthma phenotypes by increasing RAGE expression, leading to increased pro-inflammatory signalling. In addition, decreased esRAGE expression in severe asthmatics may indicate a loss of the anti-inflammatory effects of this decoy receptor, further enhancing RAGE signalling in this condition.

TP021

TLR8 UP-REGULATION DURING ACUTE ASTHMA IN CHILDREN, IDENTIFIED BY MICRO-ARRAY AND CONFIRMED BY QRT-PCR, IS ASSOCIATED WITH *TLR8* GENOTYPES

IA Laing[1,2], JA Bizzintino[1], LS Subrata[2], CJ McLean[1], S-K Khoo[1], CM Hayden[1], J Goldblatt[1], PG Holt[2] and P N Le Souef[1]

1. Pop Sci/Genetics & Health, Telethon Inst for Child Health Res, WA, 6008

2. School of Paeds & Child Health, UWA, WA, 6008

TLR8 binds single stranded RNA (ssRNA) and initiates an innate immune response. Most children with acute asthma have a respiratory virus detected and almost all are ssRNA. Micro-array analysis of peripheral blood mononuclear cells (PBMC) from children during both acute asthma (Ac) and convalescence (Cv) detected differential (Diff) expression of TLR8. Therefore, *TLR8* promoter polymorphisms may result in inappropriate TLR8 expression upon viral infection, contributing to acute asthma.

Methods: Fifty children with moderate/severe acute asthma were recruited when they presented to hospital. TLR8 expression was determined using qRT-PCR of mRNA extracted from PBMC collected during Ac and Cv (at least 6 weeks later). *TLR8* polymorphisms -746A/G (rs4830805) and -558C/T (rs1548731) were genotyped using PCR and restriction digestion with *Nla* IV and *Pst* I. Mean Ac, Cv and Diff (Ac-Cv) expression levels were compared between genotypes using SPSS.

Results: Children recruited were mostly male (58%), atopic (88.6%), virus positive (84.2%) and had a mean age of 7.0yrs. TLR8 expression levels (mean±SD) were higher during Ac than Cv (Ac 30.4±33.6, Cv 7.1±9.2, $p<0.001$). Children with *TLR8* -746AA (n=12) or -558CC (n=33) had lower Ac TLR8 expression (0.177fold p=0.004, 0.44fold p=0.006) and lower Diff (0.13fold p=0.008, 0.375fold p=0.018) than children with -746AG/GG or -558CT/TT, respectively. Genotype was not associated with Cv TLR8 expression. *TLR8* -746 and -558 haplotypes were associated with altered Ac and Diff TLR8 expression levels (ANOVA: Ac p=0.003, Diff p=0.003).

Conclusions: *TLR8* genotypes limit TLR8 up-regulation and expression during childhood acute asthma. This failure to upregulate TLR8 may contribute to asthma susceptibility through an inadequate innate immune response to viruses.

Supported by the NHMRC, Nomination for Japanese Respiratory Society Early Career Development Award, Conflict of interest: Nil

TP022

PATIENT AND CLINICIAN SATISFACTION WITH FLUTICASONE/SALMETEROL MDI WITH COUNTER

Q Summers, P Bremner
Suite 323 / Level 3, 25 McCourt Street, Subiaco WA 6008, Australia

Introduction: Until the introduction of the Metered Dose Inhaler with counter (MDIc), there was no accurate and practical way of estimating how much medication remained in the canister of an MDI.

Methods: An open-label study was done to evaluate, in out-patients, the Fluticasone/Salmeterol (Seretide®) MDIc. 132 subjects ≥ 18 years old on ICS/LABA for asthma or COPD were enrolled after written consent. They were given a Seretide® MDI and instructed to use 2 puffs twice daily for 4 weeks. They were then given a Seretide® MDIc and were asked to use it also for 4 weeks. After each treatment period, subjects and clinicians completed a satisfaction questionnaire.

Results: 104 patients (age av. 54 yrs, disease duration av. 20 yrs) were enrolled; 99 completed. With the MDI, >70% could not establish if they were running out of medication, causing anxiety for 28%. Various sub-optimal methods were used by 84% to determine how much medication remained. Use of the MDIc raised confidence in knowing how much medication remained, and led to a higher level of satisfaction in medication use. Patients' satisfaction rose from 62% (MDI) to 85% (MDIc). A majority felt the MDIc allowed them to monitor medication use (86%), gave added assurance about medication use (89%) and informed them when to replace the inhaler (90%). Patients using the MDIc took a high percentage of the prescribed dose (86%). Clinicians' confidence in knowing that patients were able to determine how much medication remained in the inhaler rose from 4.6 to 9.2 on a scale of 1-10 when the MDIc was used. Clinicians' responses indicated that they were more satisfied (84%) with the MDIc than the MDI, and that the counter helped in assessing compliance with medication (76%) and in monitoring medication use (78%).

Conclusions: The Seretide® MDI with counter led to a higher level of satisfaction for both the patients and the clinicians. The counter in the MDI provides an additional tool to help monitor medication use and improve patient management.

Supported by: GlaxoSmithKline

TP023

EXHALED NITRIC OXIDE AND ITS ROLE IN CLINICAL PRACTICE

Hiang Ping Chan[1,2], Juju Zaw[1], Paul Thomas[1,2]
[1]Department of Respiratory Medicine, Prince of Wales Hospital, Randwick, NSW 2031 and [2]Faculty of Medicine, University of New South Wales, Sydney, NSW 2052.

Nitric oxide (NO) is a vital biological mediator which is known to play an essential role within the lungs. Exhaled nitric oxide (FeNO) has been shown to be altered in many respiratory conditions including asthma. The use of FeNO measurements has also been shown to improve asthma control and reduce glucocorticosteroids use. Therefore we aim to establish whether clinicians use FeNO to: 1. diagnose asthma or 2. measure asthma control and response to treatment in clinical practice.

Methods: A retrospective review of FeNO measurements, lung function results and responses by clinicians was made. Archived FeNO and lung function results, and clinical notes were collated and relevant information entered into a database. The concordance of FeNO with clinical diagnosis or clinical asthma was compared.

Results: 106 patients attended the respiratory clinic on 143 occasions. FeNO has been demonstrated to be inversely proportionate to FEV1/FVC (r=-0.21, p=0.010, Pearson correlation). FeNO was shown to have a concordance rate of 47.2% when diagnosing asthma and 92.9% when ruling out its diagnosis; 64.8% when demonstrating good control and 60% when demonstrating poor control, with clinical assessment

Conclusion: This study suggests that FeNO could be used as an adjunct to clinical assessment as part of the process for asthma diagnosis and for the assessment of asthma control. It is specific in diagnosing asthma and may thus be of value clinically. The inverse relationship between FeNO and FEV1/FVC provides further evidence that FeNO is useful clinically as the obstructive spirometry pattern (i.e. lower FEV1/FVC) occurs in tandem with a higher FeNO measurement, which would thus suggest higher levels of inflammation occurring in the airway.

Supported by: None
Conflict of Interest: None
Nomination: None

TP024

THE RELATIONSHIP BETWEEN FENO AND ATOPIC STATUS IN PREGNANT WOMEN WITH ASTHMA

H Powell, V Murphy, PG Gibson
University of Newcastle and HMRI, John Hunter Hospital, Newcastle NSW

Forced exhaled nitric oxide(FeNO) is used as a marker of eosinophilic inflammation, but is also modified by atopy and pregnancy. The relationship between FeNO and total and specific immunoglobulin E (IgE) in pregnancy is not known and was evaluated in this study.

Methods: Pregnant women with (n=65) and without asthma (n=60) were recruited prior to 20 weeks gestation. Participants performed FeNO and serum was collected. A fluoroenzymeimmunoassay using the ImmunoCAP250 was conducted on the serum to quantify total IgE and specific allergens (house dust, mould, weed, domestic animal and grass mixes).

Results: Median FeNO, total IgE , and specific IgE to house dust, mould and domestic animal mix were significantly higher for pregnant women with asthma compared to pregnant women without asthma ($p<0.0001$, $p=0.0001$, $p<0.001$, $p=0.008$, $p=0.0001$ respectively). For all subjects FeNO was significantly correlated to total IgE ($r=0.506$), and specific IgE to house dust ($r=0.612$), weed ($r=0.248$), domestic animal ($r=0.443$) and grass ($r=0.282$) mixes. For pregnant women with asthma FeNO was significantly correlated to total IgE ($r=0.449$), and specific IgE to house dust ($r=0.632$), domestic animal ($r=0.357$) and grass ($r=0.367$) mixes. While FeNO was significantly correlated to total IgE ($r=0.348$) and house dust mix IgE ($r=0.283$) in pregnant women without asthma.

Conclusion: In pregnancy, FeNO is related to both asthma and atopic status. The main specific allergen sensitisation driving this relationship is house dust sensitisation, with lesser effects for grass pollen and domestic animal sensitisation in asthma.

Supported by the NHMRC

TP025

A COMPARISON OF TWO REAL-TIME NITRIC OXIDE ANALYSERS

B Jackson[1,2], H Powell[1], PG Gibson[1,2]
[1]*Respiratory & Sleep Medicine, Hunter Medical Research Institute, John Hunter Hospital, NSW 2305*
[2]*School of Medicine and Public Health, University of Newcastle, NSW 2308*

This study aimed to compare exhaled Nitric Oxide (eNO) data collected on devices from two different manufacturers. Airway inflammation is a key characteristic of respiratory diseases such as asthma. Real-time measurement of eNO can be used to non-invasively assess airway inflammation. Various commercial analysers are available, that employ the chemiluminescent reaction between nitric oxide and ozone. The comparability of data collected using devices from different manufacturers is not well known.

Methods: Healthy and asthmatic individuals (n = 55) had their levels of exhaled nitric oxide measured on two eNO analysers; the EcoMedics CLD88 series (ECO MEDICS AG, Bubikonerstr. 45, CH-8635 Duernten, Switzerland) and the NiOx (Aerocrine AB, Smidesvägen 12, S-171 41 Solna, Sweden). For each individual, measurements were made no longer than 30 minutes apart. All measurements were performed according to ATS/ERS guidelines.

Results: A Bland-Altman plot was performed on non-transformed data and showed good agreement between the two analysers, with a small proportional error as magnitude increased. Data was log transformed to allow for normal distribution. A paired t-test of each individual's data showed that eNO measurement using the EcoMedics analyser was significantly lower than with the NiOx device where $p<0.0001$. logEcoMed and logNiOx were highly correlated with $r = 0.981$, $p<0.0001$. Regression equations have been defined to allow for conversion between EcoMed and NiOx measurements.

Conclusion: eNO measurements made on the EcoMedics and NiOx analysers are significantly different, but highly correlated. Consequently, a conversion factor can be used so that data collected on the different machines is comparable.

Supported by the NHMRC
Conflict of Interest: NO

TP026

ASTHMA AND OBESITY ARE LINKED VIA NEUTROPHILIC AIRWAY INFLAMMATION

Hayley Scott[1,2], Peter Gibson[1,2], Manohar Garg[3], Joanne Smart[1,2], Lisa Wood[1,2]
[1]School of Medicine and Public Health, The University of Newcastle, NSW 2308, [2]Respiratory and Sleep Medicine, Hunter Medical Research Institute, John Hunter Hospital, NSW 2305 and [3]Nutraceuticals Research Group, Biomedical Sciences, The University of Newcastle, NSW 2308

The prevalence of both obesity and asthma has increased in recent years. The mechanisms that link asthma and obesity have not been established. We hypothesised that obesity may cause systemic innate immune activation, which potentiates asthmatic airway inflammation. The aim of the study was to assess systemic and airway inflammation in obese and non-obese asthmatic subjects.
Methods: Non-obese (BMI <30kg/m^2) (n=61) and obese (BMI≥30kg/m^2) (n=42) adult subjects with asthma were recruited via ambulatory care clinics at John Hunter Hospital, NSW. Clinical markers, systemic and airway inflammation were assessed. IL-6 and CRP were measured by high-sensitivity ELISA. Airway inflammation was measured using induced sputum total and differential cell counts.
Results: Compared to non-obese asthmatics, obese asthmatics had increased median [IQR] levels of CRP (1.4 [1.0, 3.1] vs 7.4 [2.9, 10.9] mg/L, p<0.0001), IL-6 (1.4 [1.0, 2.2] vs 2.4 [1.8, 3.2] pg/L, p<0.0001) and mean [SD] %sputum neutrophils (37.5 [21.9] vs 49.0 [23.1] %, p=0.008). There was no difference in median [IQR] %sputum eosinophils in non-obese compared to obese asthmatics (1.3 [0.5, 6.6] vs 1.4 [0.3, 3.6] %, p=0.32). Significant positive correlations were found between BMI and %sputum neutrophils (r=0.25, p<0.01), CRP (r=0.53, p<0.0001) and IL-6 (r=0.43, p<0.0001). There was no correlation between BMI and %sputum eosinophils.
Conclusions: This study suggests that obesity is associated with an increase in systemic and neutrophilic airway inflammation in people with asthma. Strategies targeting obesity could be useful in reducing asthma incidence and/or severity.
Supported by a Hunter Medical Research Institute postgraduate support package.

TP027

COMPARISON OF THREE METHODS FOR MEASUREMENT OF FE_{NO}

Melissa A McClean[1,2] and Cheryl M Salome[1, 2,3]
[1]*Woolcock Institute of Medical Research, Glebe NSW 2037,* [2]*CRC for Asthma, Camperdown NSW 2050,* [3]*University of Sydney, NSW 2006*

Assessment of airway inflammation by exhaled nitric oxide (FE_{NO}) can be a useful tool in clinical and epidemiological studies, but simple portable equipment is required. Two new commercially available portable devices, the HypAir FeNO and the Niox Mino®, have been developed that use a chemical sensor to measure FE_{NO}. The Woolcock eNO technique (WeNO) uses a calibrated chemiluminescence analyser (ThermoEnvironmental 42c) to measure FE_{NO} collected offline. It is not known if these techniques are comparable.

Methods: FE_{NO} was collected and measured from 15 adult subjects (11 non-asthmatic, 4 asthmatic) at an expiratory flow rate of 50ml/sec using the HypAir FeNO, the Niox Mino® and the WeNO. FE_{NO} values are expressed as geometric mean (95% confidence intervals) and compared using a paired Student t test. The repeatability of FE_{NO} values was calculated as 95% limits of agreement (95% LoA). Differences between repeat measures were compared between devices by ANOVA.

Results: FE_{NO} measured by the HypAir FeNO (26.9ppb (20.3-35.5)) and the Niox Mino® (25.7ppb (20.0-33.0)) were not significantly different but both devices measure FE_{NO} significantly higher than the WeNO (15.8ppb (10.9-22.8); $p<0.0001$). Repeatability of the HypAir FeNO (95% LoA: -7.1–4.7ppb), Niox Mino® (95% LoA:-3.4–2.7ppb) and WeNO (95% LoA: -3.6–4.2ppb) was not significantly different between devices ($p=0.62$).

Conclusion: There were no differences between the FE_{NO} measured by the HypAir FeNO and the Niox Mino®, but both measure FE_{NO} significantly higher than the WeNO. Repeatability of the HypAir FeNO, the Niox Mino® and the WeNO was similar to values reported previously. The three devices can be used in both clinical and epidemiological studies, but the different methods used to measure FE_{NO} can result in higher or lower FE_{NO} values.

Nominations: None

Conflict of Interest: None

TP028

MEASUREMENT OF FE_{NO} OVER TIME USING THREE DIFFERENT METHODS

Melissa A McClean[1,2] and Cheryl M Salome[1, 2,3]
[1]Woolcock Institute of Medical Research, Glebe NSW 2037, [2]CRC for Asthma, Camperdown NSW 2050, [3]University of Sydney, NSW 2006

Two new commercially available portable devices, the HypAir FeNO and the Niox Mino®, have been developed that use a chemical sensor to measure FE_{NO}. The Woolcock eNO technique (WeNO) uses a calibrated chemiluminescence analyser (ThermoEnvironmental 42c) to measure FE_{NO} collected offline. The stability of the FE_{NO} measurements over time from these devices has not yet been determined.

Methods: FE_{NO} was collected and measured from 7 adult subjects (5 non-asthmatic and 2 asthmatic) at an expiratory flow rate of 50ml/sec using the HypAir FeNO, the Niox Mino® and the WeNO once a week for 6 weeks. FE_{NO} values at week 1 and week 6 are expressed as geometric mean (95% confidence intervals) and compared using a paired Student t test. The stability of the devices over time was determined by ANOVA.

Results: There was no systematic changes in FE_{NO} values over six weeks for any of the methods, HypAir FeNO (p=0.64), Niox Mino® (p=0.22) and WeNO (p=0.58). The table shows FE_{NO} values at Week 1 and Week 6 and the mean±SD difference between repeat FE_{NO} values at week 1 and between week 1 and week 6.

	HypAir FeNO	Niox Mino®	WeNO
Week 1	25.8ppb (10.7-61.7)	15.0ppb (5.8-39.8)	18.2ppb (8.3-39.8)
Week 6	30.4ppb (1.8-57.5)	17.8ppb (7.1-44.7)	28.3ppb (18.6-42.7)
Repeat Wk1	1.14 ± 2.5	0.16 ± 1.6	0.60 ± 2.2
Difference	3.0 ± 5.2	2.72 ± 5.6	3.13 ± 6.8

Conclusions: FE_{NO} is stable over 6 weeks using all three methods. The within day repeatability is better than the repeatability over 6 weeks.

Nominations: None
Conflict of Interest: None

TP029

A HIGH FAT, HIGH ENERGY FOOD CHALLENGE INDUCES AN EXAGGERATED INFLAMMATORY RESPONSE IN ASTHMA

LG Wood[1,2], ML Garg[3], A Wood[1], J Smart[1], PG Gibson[1]
[1]*School of Medicine and Public Health, University of Newcastle ,NSW, 2308,* [2]*Respiratory and Sleep Medicine, Hunter Medical Research Institute, John Hunter Hospital, NSW, 2305 and* [3]*Nutraceuticals Research Group, Biomedical Sciences, University of Newcastle, NSW, 2308.*

Dietary fat has been shown to activate the innate immune response, which has been shown to cause asthma in some individuals. The aim of this study was to examine the effect of dietary fat on inflammation in asthma.

Methods: Non-obese (BMI <30) subjects with asthma were randomized to receive a high fat/ high energy (AHiFHE) (n=8) or low fat/ low energy (ALoFLE) (n=10) food challenge. Non-obese healthy controls (n=10) also underwent a high fat/ high energy (CHiFHE) food challenge. Subjects on the AHiFHE and CHiFHE challenge consumed 200% daily energy requirement in 24 hours, including 50% energy from fat. Subjects on the ALoFLE challenge consumed 75% daily energy requirement in 24 hours, including 20% energy from fat. Clinical assessment and blood samples were collected at 0, 2, 3, 4 and 24 hours. Inflammatory markers, including plasma TNFα, CRP and IL-6, were analysed by high sensitivity ELISAs.

Results: At 4 hours after the commencement of the food challenges, subjects on the AHiFHE challenge, had a significantly higher increase in plasma CRP concentrations, compared to subjects on both the ALoFLE and CHiFHE challenge ($p= 0.021$). In subjects on the AHiFHE challenge, there was also a significantly higher increase in plasma TNFα concentrations at 3 hours, compared to subjects on ALoFLE challenge ($p = 0.034$).

Conclusions: A high fat/ high energy intake causes an exaggerated systemic inflammatory response in subjects with asthma. This suggests that subjects with asthma are more susceptible to fat-induced innate immune activation.

Supported by an NHMRC Project Grant
Conflict of interest: No

TP030

DIETARY FAT AND AN ACTIVATED INNATE IMMUNE RESPONSE ARE ASSOCIATED WITH REDUCED FEV_1

LG Wood[1,2], J Attia[1,3], P McElduff[1], M McEvoy[1,3], V Flood[4], PG Gibson[1,2]
[1]School of Medicine and Public Health, University of Newcastle ,NSW, 2308, [2]Respiratory and Sleep Medicine, Hunter Medical Research Institute, John Hunter Hospital, NSW, 2305, [3]Centre for Clinical Epidemiology and Biostatistics, University of Newcastle, NSW, 2305, [4]NSW Centre for Public Health Nutrition, University of Sydney

Preservation of lung health with aging is an important health issue in the general population, as loss of lung function with aging can lead to the development of obstructive lung disease. Inflammation is increasingly linked to loss of lung function and evidence suggests that consumption of dietary fat exacerbates inflammation. The aim of this study was to examine the contribution of dietary fat to reduced lung function an older population.
Methods: Participants, aged between 55 and 85 years, were recruited from the Hunter Community Study, a population-based cohort, during 2004 and 2005. All participants received a clinical assessment, including baseline spirometry and provided a blood sample. Diets were analysed using food frequency questionnaires. Plasma IL-6 concentrations were measured by ELISA.
Results: Smoking, % energy intake from dietary fat, IL-6 and obesity were all found to be inversely associated with reduced forced expiratory volume in 1 second (FEV_1) in a linear regression model. Using backward stepwise linear regression, former smoking, % energy from dietary fat and plasma IL-6 remained as negative predictors of FEV_1.
Conclusions: An increased proportion of dietary fat and innate immune activation are associated with reduced lung function. Dietary fat restriction may be useful in preserving lung function with aging.

Supported by Hunter Medical Research Institute Project Grant
Conflict of interest: No

TP031

TOLL-LIKE RECEPTOR AGONISTS STIMULATE CYTOKINE RELEASE FROM BLOOD BUT NOT AIRWAY CELLS

Kevin M Oreo[1,2], Katherine J Baines[1,2], Peter G Gibson[1,2] , Philip M Hansbro[1], Jodie L Simpson[1,2]

(1) Centre for Asthma and Respiratory Disease, The University of Newcastle, NSW, 2308. (2) Department of Respiratory and Sleep Medicine, Hunter Medical Research Institute, NSW, 2305

The innate immune system has a key role in detecting pathogens and priming protective immunity. Toll-Like Receptors (TLRs) are important in sensing pathogens and directing immune responses. Dysfunction of the innate immune system in the airway may be a feature of neutrophilic airways disease, and may be important for resolution of infection and inflammation. Our aim was to investigate innate immune responses of blood and airway cells to TLR2 and 4 activation.

Methods: Blood was collected from healthy volunteers (n=9) and sputum was collected from subjects with airway disease (n=7). Granulocytes and monocytes were isolated from peripheral blood by Percoll separation and cells from sputum were recovered after processing with dithiothreitol. Cells were cultured at $1x10^6$ cells/mL (blood) or $0.5x10^6$cells/mL (sputum) in RPMI1640 and stimulated with either LPS (TLR4 agonist) or Pam3CYSK4 (TLR2 agonist) at a range of concentrations (10 to 10 000ng/mL). Cells were cultured at 37°C and cell free supernatants were collected at 24 hours. Cytokine and protease levels were measured by ELISA.

Results: Both LPS and PAM3CYSK4 stimulation of granulocyte cultures resulted in increased release of IL-8, IL-6 and MMP-9 ($p<0.05$). Neither LPS nor Pam3CYSK4 increased neutrophil elastase release from granulocytes. Similarly IL-6 and IL-8 release was significantly higher in monocyte culture after stimulation with LPS and Pam3CYSK4 compared to unstimulated cells ($p<0.05$). In sputum cell cultures there was no increase in release of IL-8 or IL-6 in response to either LPS or Pam3CYSK4 treatment.

Conclusion: TLR agonists (2,4) cause differential activation of peripheral blood granulocytes and monocytes, whereas airway cells appear refractory to TLR stimulation.

Supported by NHMRC

TP032

STEROID RESPONSIVENESS IN RELATION TO HISTOLOGICAL PHENOTYPE IN ASTHMA

DC Cowan, JO Cowan, R Palmay, CE Landhuis, DR Taylor
Respiratory Research Unit, Dunedin School of Medicine, University of Otago, Dunedin, New Zealand

Airway inflammation in asthma is heterogeneous. Cell types include eosinophilic, neutrophilic, paucigranulocytic and mixed cellularity. Studies have often been confounded by cigarette smoking or inhaled corticosteroid (ICS) treatment. We aimed to ascertain the prevalence of inflammatory subtypes in the absence of confounders and clarify whether steroid responsiveness is exclusive to eosinophilic asthma.

Methods: 78 non-smoking adults with current asthma were recruited. Inclusion criteria: hyper-responsiveness to hypertonic saline OR methacholine OR ≥12% increase in FEV_1 post-bronchodilator. ICS was withdrawn until loss of control (LOC) or one month if no LOC. Patients were classified as eosinophilic (EA: ≥2% sputum eosinophils), or non-eosinophilic (NEA: <2%). Steroid responsiveness was assessed after 4-weeks fluticasone 500mcg bd.

Results: Baseline characteristics for EA and NEA were similar except ICS dose (872 vs. 403 mcg/day, $p<0.05$). After steroid withdrawal, 50 patients (64%) were eosinophilic, 26 (33%) paucigranulocytic, and 2 (3%) mixed cellularity. With steroid, the before/after outcomes for EA/NEA were (mean±SD): ACQ: EA 1.9±0.9/0.7±0.6; NEA 1.0±0.6/0.7±0.5, p-values<0.05; FEV_1: EA 2.20±0.76/ 2.86±0.79, NEA 2.65±0.83/2.83±0.88, p-values<0.05; ΔPC_{20}AMP doubling doses; EA 4.2±2.9; NEA 1.6±2.2, $p<0.001$. Cell type changed: from eosinophilic to mixed cellularity in 2/48 (4%) and neutrophilic in 2/48 (4%); and from paucigranulocytic to neutrophilic in 3/25 (12%). Overall a significant increase in sputum neutrophilia was seen after steroid (19±17% to 29±23%, $p<0.01$).

Conclusions: Steroid responsiveness occurred almost exclusively in EA and was minimal in NEA. In our population, neutrophilic asthma was absent, but inhaled steroid resulted in an increase in sputum neutrophilia. Whether steroid induced sputum neutrophilia is important is unclear.

Supported by Lottery Health New Zealand
Conflict of interest: NO

TP033

TAILORED INTERVENTIONS BASED ON AIRWAY EOSINOPHILIC INFLAMMATION VERSUS CLINICAL SYMPTOMS FOR ASTHMA IN ADULTS AND CHILDREN – A SYSTEMATIC REVIEW

HL Petsky[1], JA Kynaston[2], C Turner[3], CJ Cates[4], TJ Lasserson[4], AM Li[5], AB Chang [1,6]

[1]Qld Children's Resp Centre, [2]General Paediatrics; Royal Children's Hosp, Brisbane,, [3]Sch of Nursing, University of Qld, [4]Community Health Sciences, St George's Uni of London, UK, [5]Dept of Paediatrics, Prince of Wales Hosp, Hong Kong, [6]Menzies SHR Darwin

Markers of airway eosinophilic inflammation (sputum eosinophils and exhaled nitric oxide) have been advocated for asthma monitoring. We combined our Cochrane reviews to evaluate if tailoring of medications based on airway eosinophilic markers improve asthma outcomes.

Methods: Cochrane methodology was used. All randomised controlled comparisons of adjustment of asthma therapy based on sputum eosinophils or exhaled nitric oxide (airway inflammation tailored group) compared to traditional methods (primarily clinical symptoms and spirometry/peak flow) (control group) were included. Results of searches (performed by the Cochrane Airway Group) were reviewed against pre-determined criteria for inclusion.

Results: Eight studies fulfilled the inclusion criteria but had several important differences including the definition of asthma exacerbations and duration of study. The total number of participants randomised was 1148. In the meta-analysis, significantly less adults in the airway inflammation tailored group had >1 asthma exacerbation when compared to the control group; pooled odds ratio (OR) was 0.70 (95%CI 0.54 to 0.91); number needed to treat to benefit was 12 (95%CI 7 to 43). However the airway inflammation tailored group required significantly higher doses of inhaled corticosteroids (ICS), WMD 63.53 (95%CI 11.31 to 115.74). Also there was no significant difference between groups in the asthma exacerbation rate, final FEV_1, FeNO or asthma symptom score.

Conclusion: Tailoring asthma interventions based on eosinophilic inflammatory markers have limited benefits in improving asthma outcomes in adults and significantly increases ICS doses. No conclusion can be drawn for children with asthma.

Supported by: Australian Cochrane Airways Group & Royal Children's Hospital Foundation (Brisbane).

TP034

THE EFFECT OF CIGARETTE SMOKING ON ASTHMA CONTROL AND EXACERBATIONS IN PREGNANT WOMEN

Vanessa E Murphy[1], Vicki L Clifton[2], Peter G Gibson[1]
[1]Centre for Asthma and Respiratory Diseases, University of Newcastle and Hunter Medical Research Institute, NSW, 2308, [2]School of Paediatrics and Reproductive Health, University of Adelaide, SA, 5005.

Smoking is more prevalent among pregnant women with asthma than pregnant women without asthma; however no studies have assessed the clinical implications of smoking on asthma exacerbations in pregnancy.

Methods: Pregnant women with asthma (n=80) were prospectively assessed from recruitment (14.8 weeks [3 (SD)]) to delivery at clinic visits (18, 30, 36 weeks and during exacerbation), and by fortnightly phone calls. There were 27 current smokers (4.0 median pack years), 27 ex-smokers (2.1 median pack years) and 26 never smokers (self-report). The Juniper asthma control questionnaire (ACQ6) was administered at each contact and exacerbations classified as severe (requiring medical intervention) or mild (self-managed).

Results: There were 56 exacerbation events in current smokers (23 severe, 33 mild), 59 in ex smokers (26 severe, 33 mild) and 43 in never smokers (11 severe, 32 mild). Current smokers experienced more severe exacerbations per person (median 1, interquartile range [0, 1]) compared to ex smokers and never smokers (0, [0,1]); however this did not reach statistical significance (P=0.25). ACQ6 during exacerbation (mild or severe) was significantly higher in current smokers (median 2.0, [1.7, 3.0]) compared to never smokers (1.67, [1.2, 2]), while the ACQ6 during exacerbation in ex smokers was intermediate (1.8, [1.3, 2.8]) (P=0.018). The best ACQ6 score recorded when stable by current smokers (median 0.09, [0, 0.46]) and ex smokers (0.09, [0, 0.50]) was not significantly different from never smokers (0, [0, 0.42]) (P=0.49).

Conclusions: During pregnancy, asthma exacerbations are common and are more severe in current smokers than in never smokers. The implications of this may be that the risk of maternal asthma on the baby is greater among smokers.

Supported by the NHMRC, Asthma Foundation of NSW, Hunter Medical Research Institute, Port Waratah Coal Services, University of Newcastle
Conflict of Interest: NO

TP035

COAGULATION FACTORS IN THE AIRWAYS IN MODERATE AND SEVERE ASTHMA AND THE EFFECT OF INHALED STEROIDS

Fraser Brims[1,2], Anoop Chauhan[1,2], Janis Shute[1]

[1]Institute of Biomedical and Biomolecular Sciences, University of Portsmouth, UK and [2]Respiratory Centre, Queen Alexandra Hospital, Portsmouth Hospitals NHS Trust, UK

Rationale: There is evidence for up-regulation and activation of the extrinsic coagulation cascade in the airways in asthma, and that both plasma and locally-derived factors may be involved. Our objective was to test the hypothesis that the normal haemostatic balance of the healthy airway sampled by sputum induction changes in favor of fibrin formation in asthmatic airways, and that inhaled corticosteroids (ICS) and plasma exudation influence this balance.

Methods: 30 stable subjects (10 controls, 10 moderate & 10 severe asthmatics) were recruited and underwent sputum induction using 4.5% hypertonic saline, with analysis of alpha-2 macroglobulin and coagulation factors in sputum using ELISA and activity assays. Additionally, the moderate cohort were weaned off their ICS, followed by further sputum induction 5 days after cessation of steroids.

Results: Weaning of ICS was associated with a significant rise in plasminogen (median (IQR): 13.92 (6.12-16.17) vs. 4.82 (2.14-13.32) ng/ml; $p<0.05$) and tissue-plasminogen activator (tPA) (5.57 (3.57-14.35) vs. 3.88 (1.74-4.05) ng/ml; $p=0.026$) levels in sputum, such that tPA in moderate asthma post steroid withdrawal was significantly ($p<0.0015$) higher than controls (2.14 (0.0-2.53) ng/ml). Severe asthmatics had significantly more alpha-2 macroglobulin ($p<0.001$), tissue factor ($p<0.05$), plasminogen activator inhibitor (PAI-1; $p<0.05$), tPA ($p=0.029$) and thrombin activatable fibrinolysis inhibitor (TAFI; $p<0.01$) in their sputum than control subjects.

Conclusion: Moderate asthma may be associated with increased fibrinolysis that is corrected by ICS. Severe asthma is associated with a pro-fibrinogenic, anti-fibrinolytic environment in the airways. Our study suggests that inhibition of coagulation in severe asthma may be a therapeutic approach.

TP036

WHAT ARE THE PRIORITIES OF OLDER PEOPLE WITH ASTHMA?

Dianne Goeman[1, 3] , Melanie Crane[1, 2], Sinthia Bosnic-Anticevich[4], Jo Douglass[1,3] Christine Jenkins [1,2]
[1]*Co-operative Research Centre f*or *Asthma and Airways, Camperdown, NSW2050,* [2] *The Woolcock Institute of Medical Research, Glebe, NSW 2037,* [3] *AIRMed, The Alfred Hospital & Monash University, Melbourne, Vic 3004,*[4] *Faculty of Pharmacy,University of Sydney, Camperdown, NSW 2006*

Asthma-related mortality and morbidity increase with age and recent Australian Bureau of Statistics data show a continuation of this trend. In 2006, 356/402 (88%) of asthma deaths occurred in those >50 years of age. We designed, validated and then trialled a questionnaire to identify concerns of older people with asthma.
Methods: 152 people over 55 years with asthma were recruited from a random sample of 60 pharmacies in regional, rural and metropolitan Victoria and a cluster sample from 17 metropolitan and regional pharmacies in NSW.
Results: 87% of participants have both preventer and reliever treatments prescribed and self-reported preventer adherence is high. Although most participants reported good asthma control only 10% reported having no asthma symptoms over the last month. Issues identified by patients included: cost of medication (47%), worry about side effects (38%) while 27% report experiencing side effects. Two-thirds (68%) report frustration over asthma stopping them doing all they want to do. Provision of action plans was relatively high at 37% but another 37% stated they would find owning one useful. Less than half of participants reported their GPs had tested their lung function in the past two years, observed their device technique or undertaken a medication review. Findings also suggest that a high proportion of older people with asthma thought more information about asthma would be helpful.
Conclusions: A simplified version of our questionnaire used in general practice could assist GPs to identify and address the needs of older people.

Supported by the Co-operative Research Centre for Asthma and Airways
Nomination: None
Conflict of Interest: No

TP037

SEASONAL PATTERNS (2004-2007) OF RESPIRATORY VIRUSES ISOLATED WITH ACUTE AIRWAYS DISEASE

Peter AB Wark[1,2], Kristy Parsons[1], Jane Ryan[3] Terry Grissell[1], Noreen Bell[1], Jessica Chapman[1], Peter G Gibson[1,2]

1 Centre for Asthma and Respiratory Disease, Hunter Medical Research Institute, University of Newcastle, NSW 2308. 2. Department of Respiratory and Sleep Medicine John Hunter Hospital.3. Biota Australia PtyLtd

Respiratory virus infections are important triggers of acute airways disease. Seasonal variation of these viruses have been seen in the northern hemisphere, but not described in Australia.

Aim: To characterise the viruses associated with acute asthma and COPD.

Methods: From 2004 to 2007 we recruited subjects over 6 years admitted to hospital with acute exacerbations of Asthma, COPD or Cystic fibrosis. Spontaneous sputum, throat and nasal swabs were collected. Samples were assayed by real-time PCR for rhinovirus (RV), enterovirus (EV), non-SARS coronavirus (CoV), human metapneumovirus (hMPV), respiratory syncytial virus types A and B (RSV) and influenza virus types A and B (Flu).

Results: There were 201 acute episodes were specimens were collected, viruses were detected in 102 (51%) of these. RV was the most frequent virus isolated in 52% of positive samples, followed by EV (18%), CoV (14%), RSV (9%), Flu (7%), and hMPV (7%). The lowest virus detection occurred in summer. Peak RV detection occurred in autumn, but was the most prevalent virus in all seasons. Both Flu and RSV detection were confined to winter.

Conclusion: RV is the most prevalent virus associated with acute asthma and COPD all year round. This occurs even in winter though the proportion of Flu and RSV increase during this time.

Conflict of Interest: None

Funding: NHMRC Australia, Biota Australia

TP038

THE PROFILE OF SPUTUM EOSINOPHILIA, EXHALED NITRIC OXIDE AND PERCEPTION OF DYSPNOEA IN OLDER ASTHMATICS

Kheng Sit Lim[1], Tracy Greer[2], Kerrie Wade[2], Matthew Peters[2], Christine Jenkins[2]
[1]*Graduate Medical Program, Faculty of Medicine, University of Sydney, Australia.*
[2]*Department of Thoracic Medicine, Concord Hospital, Sydney, Australia*

Introduction: Poor perception of dyspnoea together with under-reporting of the severity of breathlessness contributes to suboptimal management of asthma in adults over 65. The Fraction of exhaled Nitric Oxide (FeNO) is well established as a research tool to assess airway inflammation but its role in clinical practice is unclear. Despite this, it has recently been recommended by the ATS/ERS for use in clinical settings to assess the nature and severity of airway inflammation.
Aim: In this observational study we assessed the relationships between FeNO, sputum eosinophilia and other biomarkers of asthma controls in older asthmatics.
Methods: 18 participants over the age of 55 were recruited. At visit 1, FeNO samples, Juniper Asthma Control Questionnaire (ACQ) and Marks Asthma Quality of Life Questionnaire (AQLQ) were conducted. Perception of dyspnoea was assessed using the difference between Visual Analog Scale (VAS) both before and after pre and post salbutamol spirometry to measure subjective breathless against quantitative changes in spirometry. Sputum induction was performed and sent for analysis of sputum eosinophils. At visit 2, FeNO samples, perception of dyspnoea, ACQ and AQLQ were reassessed. Airway hyperresponsiveness was tested using methacholine. Both visits were conducted within 2 months of each other.
Results: FeNO at visit 1 correlated with AHR (r=0.573, p=0.013) and ACQ (r=0.540, p=0.021). There were no correlations between FeNO and other biomakers including sputum eosinophils. There were no correlations between perception of dyspnoea and bronchodilator reversibility. FeNO was non reproducible within the two visits (Mean FeNO of 21.3 ppb and 15.9 ppb at visit 1 and 2 respectively with an error range of 16.9 ppb).
Conclusions: FeNO does provide useful clinical information as it correlates with AHR and ACQ. The observed high variability of measured FeNo in this population warrants further investigation.

Conflict of Interest: No

TP039

TROPICAL PLANT POLLENS AND ALLERGIC RHINITIS

Fay Johnston [1,2], Ivan Hanigan [2,3,4]
1. Menzies Research Institute, University of Tasmania 2. School for Environmental Research, Charles Darwin University 3. National Centre for Epidemiology and Population Health, Australian National University 4. School of Plant Science, University of Tasmania

Background: Little has been reported about the allergenicity of pollens from tropical plant families or their relative contributions to allergic disorders in these regions. We investigated associations between daily average fungal spore and pollen counts of several tropical plant families and sales of four medications commonly used to treat allergic rhinitis in Darwin, Australia, from April 2004 to November 2005.
Methods: Five pharmacies provided daily sales data for loratadine/pseudoephedrine and fexofenidine tablets and beclomethasone and budenoside nasal sprays. We used Poisson generalized linear and categorical modelling to examine outcomes. All analyses accounted for the potential confounding effects of time trends, season, holidays, respiratory viral illnesses, meteorological conditions and air pollution.
Results: The peak total pollen count was 94 grains/m^3 while the peak Poaceae (grass) pollen count was just 24 grains/m3. Daily sales of all products increased 5% with each interquartile range rise (3 grain/m^3) in Poaceae pollen, greatest at a lag of 1 day (5.07 95%CI 1.04, 9.25). No associations were observed with fungal spores, the total pollen count or pollen from other plant families.
Conclusions: We found an association between tropical grass pollen and allergic rhinitis measurable at low pollen concentrations. High allergenicity and increasing population exposure to pollen from some species of tropical grasses could be contributing an increasing prevalence of allergic rhinitis in tropical regions.

Supported by the Australian Research Council
No conflict of interest

TP040

EDUCATION INTERVENTION FOR CHILDHOOD ASTHMA BY INDIGENOUS HEALTH WORKERS IN THE TORRES STRAIT, AUSTRALIA

PC Valery[1,2], IB Masters[3], B Taylor[4], PO'Rourke[1,2], Y Laifoo[5], AB Chang[3,6]
[1]Qld Institute of Medical Research, [2]Australian Centre for International and Tropical Health and Nutrition, [3]Royal Children's Hosp., [4]Asthma Foundation Qld, [5]Thursday Is. Primary Health Care Centre, [6]Menzies School of Health Research

The prevalence of childhood asthma in the Torres Strait is high with 30% having persistent asthma and parental asthma knowledge is poor. We conducted a randomised controlled trial of additional education intervention by Indigenous Health Care Workers (HCW) on asthma outcomes.

Methods: Children with paediatric respiratory physician diagnosed asthma were enrolled and randomly allocated to: (1) three additional asthma education sessions with a trained HCW or (2) no additional education, and re-assessed at 12 months. Primary endpoint was the difference in the number of unscheduled hospital/doctor visits due to asthma exacerbation between the groups. Secondary outcomes were improvement of quality of life and functional severity scores, asthma knowledge, interpretability of asthma action plans and school days missed due to wheezing.

Results: We enrolled and followed up 88 children (81%) aged 1-17 years, 97% Aboriginal and/or Torres Strait Islanders (35 intervention; 53 controls). The groups were mostly comparable at baseline (except for asthma severity which was adjusted for in the analysis). There were no significant differences ($p=0.25$) in the number of unscheduled hospital/doctor visits due to asthma exacerbation (intervention group median=1.0, control group median=0.0). Compared to the control group, carers in the intervention group were significantly better in knowledge of asthma medication ($p<0.05$), possession ($p=0.01$) and ability to interpret asthma action plans ($p=0.02$). Children in the intervention group missed fewer school days due to wheezing ($p=0.04$) compared to the control group. Both groups improved in quality of life and functional severity scores (baseline vs follow up) but there were no significant differences between the intervention and control groups.

Conclusions: Additional asthma education sessions conducted by trained local HCW improved some but not all asthma outcomes.

Funding: NHMRC, RHSET, Telstra Foundation, Royal Children's Hosp. Foundation.

TP041

COMPLEMENTARY AND ALTERNATIVE MEDICINE USE IN ASTHMA

Sarah Appleton[1], Catherine Hill[2], Tiffany Gill[3], David Wilson[1], Richard Ruffin[1], Anne Taylor [3], Robert Adams[1]

[1]*Health Observatory (University of Adelaide Discipline of Medicine) and* [2]*Dept of Rheumatology at The Queen Elizabeth Hospital.* [3] *Population Research and Outcome Studies Unit, South Australian Department of Health; Adelaide, South Australia.*

Complementary and alternative medicines (CAMs) including fish oils and zinc may benefit people with asthma by reducing airway inflammation and hyperresponsiveness. Our aim was to determine the prevalence of CAM use in asthma and its relationship with morbidity.

Methods: We assessed CAM use in the North West Adelaide Health Study, (n=3206 adults). Respondents completed surveys and underwent biomedical assessment including spirometry. Asthma was identified by self-reported physician diagnosed asthma or bronchodilator responsiveness. Current CAM use was identified at the clinic visit and included fish oil; other oils (flaxseed, emu); glucosamine; vitamins (multi-, B/C/E/CoQ10); herbal (celery, garlic, ginseng); minerals (magnesium, zinc, "minerals").

Results: Asthma prevalence was 16.8%. Use of any CAM (29.5%, n=157), all herbal and all vitamin supplement was not significantly different between people with and without asthma. Compared to those without asthma, males with asthma were more likely to use fish oils [7.1 vs 4.2% OR=1.71 (0.93-3.87), p=0.08], and mineral supplements, [4.4 vs 1.6%, OR=2.40 (1.05-5.47)] specifically zinc, and females with asthma were less likely to use fish oil [5.5 vs 9.2%, OR=0.51, (0.29-0.91)]. In the asthma population, effects of CAMs on lung function were seen. Males using zinc (n=7) demonstrated significantly higher mean FEV_1 (105.5 vs 90.9, p=0.03), and FVC (105.3 vs 95.0, p=0.057) than those not using zinc. Although the differences were not statistically significant, females using fish oils demonstrated increased lung function.

Conclusions: Sex-specific differences in CAM use occurred in our asthma population. Further investigation of the effects of fish oil and zinc supplements on lung function and respiratory symptoms is warranted.

Conflict of interest: None

TP042

THE RELATIONSHIP BETWEEN PATIENT PERCEIVED RISK OF INHALED CORTICOSTEROIDS IN PREGNANCY AND MEDICATION ADHERENCE

K McLaughlin[1], V Murphy[1], K McCaffery[2], H Powell[1], VL Clifton[3], W Giles[4], MJ Hensley[1], PG Gibson[1]

[1]University of Newcastle and HMRI, John Hunter Hospital, Newcastle NSW. [2]University of Sydney, Sydney, NSW. [3]University of Adelaide, SA. [4]Royal North Shore Hospital, Sydney, NSW

Asthma affects 12% of pregnancies in Australia. Variable changes in asthma during pregnancy have been well documented, and it is important to continue using preventer medications (inhaled corticosteroids, ICS) in pregnancy to maintain adequate asthma control. We investigated the relationship between women's perceived risks of medication and their use of asthma medication.

Methods: Subjects with current asthma (n = 40) were recruited prior to 20 weeks gestation and had monthly visits. Women completed a 10 cm visual analogue scale indicating their perceived risk of salbutamol and ICS on the baby, with a score of 0% indicating no side effects (healthy baby) and a score of 100% indicating severe side effects (eg: deformity). Asthma self-management education was provided at each visit and self-reported ICS adherence assessed.

Results: The median perceived risk of ICS medication was 24% (range 0 – 70%) at visit 1, 15% (0 – 61%) at visit 2, 13% (0 – 61%) at visit 3 and 12% (0 – 46%) at visit 5 (non-parametric repeated measures ANOVA, P=0.005). By comparison, the median perceived risk of salbutamol was 10% (range 0 – 80%) at visit 1 and 5% (range 0 – 45%) at visit 5. At visit 1, 30% of women perceived low risk ($\leq$10%) of ICS on the baby, while at visit 5, 43% of women perceived low risk. There was a significant relationship between ICS non-adherence and perceived risk of ICS at visit 2 (Spearman r = 0.592, P = 0.012), however, this relationship was no longer significant at visit 5 (r = 0.180, P=0.411).

Conclusion: Pregnant women with asthma perceive that there is some risk to their baby of the use of ICS. Higher levels of perceived risk were associated with ICS non-adherence. Their perception of risk was improved following asthma education, and provision of such information may improve adherence rates.

Supported by the NHMRC

TP043

HIGH FLOW OXYGEN CAUSES CARBON DIOXIDE RETENTION IN SEVERE ASTHMA: A RANDOMISED CONTROLLED TRIAL

Kyle Perrin[1], Meme Wijesinghe[1], Mark Weatherall[2], Richard Beasley[1]
[1]*Medical Research Institute of New Zealand, Wellington, New Zealand*
[2]*University of Otago, Wellington, New Zealand*

The use of high flow oxygen in acute exacerbations of COPD can result in CO_2 retention. High flow oxygen is often used in acute severe asthma, but it is uncertain whether this causes an increase in $PaCO_2$. In this randomised controlled study we investigated the effects of high flow versus titrated oxygen therapy on $PaCO_2$ in acute asthma.

Methods: 80 patients with severe exacerbations of asthma (FEV1 $\leq$ 50% predicted) presenting to the Wellington Hospital Emergency Department were randomised to high flow oxygen (8l/min via a medium concentration mask) or titrated oxygen (to a saturation of 93 to 95%) for 60 minutes, along with routine treatment. Transcutaneous carbon dioxide measurements (tCO_2) were made at 0 and 60 minutes. The primary outcome variable was the proportion of patients with a rise in $tCO_2 \geq 4$ mmHg at 60 minutes. The secondary outcome variable was the proportion of patients with a rise in $tCO_2 \geq 8$ mmHg.

Results: Three subjects withdrew from the high flow group leaving 36 for analysis and 41 in the titrated group. A rise in $tCO_2 \geq 4$ mmHg was seen in 15/36 (41.7%) of the high flow group and 6/41 (14.6%) of the titrated group, a relative risk of 2.8 (CI 1.2 to 6.6, p=0.008). A rise in $tCO_2 \geq 8$ mmHg was seen in 5/36 (13.9%) of the high flow group and 3/41 (7.3%) of the titrated group, a relative risk of 1.9 (CI 0.5 to 7.4, p=0.35). The mean (SD) FEV1 percent predicted was 33.4% (10.5) in the high flow group and 35.4% (9.7) in the titrated group (P = 0.35).

Conclusion: High flow oxygen therapy results in an increase in tCO_2 when delivered to patients with severe exacerbations of asthma, and excessive oxygen delivery should be avoided.

Supported by: The Health Research Council
Conflict of Interest: No

TP044 Paper withdrawn

TP045

THE CHILDHOOD ASTHMA PREVENTION STUDY (CAPS): DEVELOPMENT OF ALLERGEN SENSITISATION IN THE FIRST 8 YEARS OF LIFE

E Belousova[1], C Weber-Chrysochoou[1], BG Toelle[1], GB Marks[1], AS Kemp[2]
[1]*Woolcock Institute of Medical Research, NSW 2037.*
[2]*Children's Hospital at Westmead, NSW 2145*

There have been few detailed studies of the course of atopy during childhood.

Aim: To examine the development of sensitivity to ingested and inhaled allergens in the first 8 years of life in a high risk birth cohort.

Methods: Children with a family history of asthma were recruited antenatally into a randomized trial of house dust mite (HDM) avoidance and of dietary modification. Neither of these interventions reduced the prevalence of atopy or asthma by age five years [1]. Skin prick tests to common ingested and inhaled allergens were performed at 18 months, 3, 5, and 8 years. A positive result was a wheal ≥3mm . The p-values for the trends were estimated from the generalised estimating equations controlling for the interventions and gender**.**

Results: Sensitization to any allergen increased from 14% at 18 months to 45% at 8 years. Sensitization to ingested allergens either decreased (egg) or plateaued (eg. salmon, tuna, peanut) and sensitization to inhalant allergens increased with age.

Allergen	18 months n=535	3 years n=521	5 years n=488	8 years n=402	p
Egg, %	6.2	2.7	2.7	2.5	0.001
Peanut, %	4.1	3.8	3.9	5.0	0.504
Salmon / tuna, %	1.3	0.4	0.8	2.5	0.115
HDM/grasses/mould/cat	7.3	21.3	36.3	43.0	0.0001

Conclusion: The pattern of sensitization differed between ingested and inhaled allergens suggesting different immunological mechanisms. In addition, by 8 years of age peanut was the most common food allergen sensitization .

Support: NHMRC Australia and AsthmaCRC.

[1] Marks GB et al. J Allergy Clin Immunol. 2006;118:53-61.

TP046

EARLY LIFE RISK FACTORS AND INCIDENCE OF ALLERGIC RHINITIS

MC Matheson[1], SC Dharmage[1], MJ Abramson[2], J Sunyer[3], J Heinrich[4], D Jarvis[5], D Norbäck[6], C Raherison[7], M Wjst[4], C Svanes[8]

[1]MEGA Epidemiology, Uni of Melbourne, Australia; [2]Monash Uni, Australia; [3]Centre for Recerca Epidemiología Ambiental, Spain; [4]GSF-Institute of Epidemiology, Germany; [5]King's College, UK; [6]Uppsala Uni, Sweden; [7]Faculté de Médecine Xavier Bichat, France; [8]Uni of Bergen, Norway

Background: Allergic rhinitis (AR) is an increasingly common condition. AR may predispose to asthma and early aggressive treatment might prevent asthma. Understanding risk factors associated with AR is important. We examined the association between early life factors and development of AR using European Community Respiratory Health Study (ECRHS) data.

Methods: In 1992-94, community based samples of 20-44 year old people were recruited from 48 centres in 22 countries. On average, 8.9 years later, 28 centres re-investigated their samples using similar methods. Onset of AR was reported in interviewer-led questionnaires. Cox regression was used to assess independent predictors of AR risk in childhood (0-10 yrs), adolescence (11-20 yrs) and adult life (21+ yrs).

Results: Out of the 10839 participants in ECRHS II n=3533 (Prev=32.6%; 95%CI 31.7-33.5%) had AR but there was significant heterogeneity in the estimates across countries (p=0.0001). Data on age of onset of AR was available for 10,373 participants. Males more often got AR in childhood (HR 0.80, 95%CI 0.69-0.93), while females more often got AR in adulthood (1.48, 1.33-1.65). Siblings were associated with a reduced risk of AR in childhood (0.86, 0.82-0.91), adolescence (0.89, 0.86-0.93) and adulthood (0.98, 0.95-1.01). Attendance at pre-school/ nursery/day care before 5 years (0.84, 0.78-0.90) and sharing bedrooms with older children (0.86, 0.80-0.92) were associated with a reduced risk of AR; this did not vary with age. Cats and dogs in the first year of life were associated with a significantly reduced risk of AR only in adolescence. A serious respiratory infection before 5 years and parental allergies were associated with increased risk of AR.

Conclusion: Early life factors (siblings, day care and bedroom sharing) have the strongest effect in childhood consistent with the 'hygiene hypothesis'. The apparent protective effects of pets appeared restricted to adolescent onset AR.

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TP047

REFINING NATIONAL ASTHMA INDICATORS USING A DELPHI SURVEY

P Espinel, E Belousova, LM Poulos, P Correll, HK Reddel, GB Marks
Australian Centre for Asthma Monitoring, Woolcock Institute of Medical Research Sydney, NSW 2037

Background: Currently there are 24 indicators recommended for monitoring to guide policy about the prevention and management of asthma in Australia. There is a consensus that this is number is too great for an efficient monitoring program.
Aim: A Delphi survey was used to identify a smaller set of core indicators as the focus of future asthma monitoring activities in Australia and elsewhere.
Methods: Practising respiratory physicians, paediatricians, general practitioners, asthma researchers, epidemiologists and representatives of other relevant stake-holders were identified at a national level by investigators and were invited via email to participate. A web-based survey is currently being conducted in three rounds. For the 2nd and 3rd rounds, panellists are given feedback including their own previous responses, pooled results and anonymized comments of other participants and asked to consider refining their answers based on this feedback.
Results: Sixty two asthma experts from different disciplines were invited to participate. Thirty two panellists (52%) completed the 1st survey and 72% of these (preliminary results) have completed the 2nd survey. C*urrent asthma* (defined as doctor diagnosis plus symptoms or treatment in the last 12 months) and *hospital separations for asthma* were consistently ranked by the panellists as indicators recommended for retention. On the other hand, *Asthma Cycle of Care uptake* and *airway hyperresponsiveness* were identified as potential indicators for exclusion.
Conclusions: The Delphi survey has helped to obtain consensus about the most important asthma indicators for monitoring asthma at a national level. This core set of standardized indicators should be used to gain population-based information on asthma in Australia and other countries.
Support: ACAM is a collaborating unit of the AIHW and is funded by the Australian Government Department of Health and Ageing.

TP048

RISK FACTORS FOR THE DEVELOPMENT OF ASTHMA BETWEEN AGE 4 AND 7 YEARS IN A NATIONAL COHORT STUDY

A-M Waters, R Ampon, LM Poulos, W Xuan, A Zinoviev, HK Reddel, GB Marks

Australian Centre for Asthma Monitoring (ACAM), Woolcock Institute of Medical Research, NSW 2050

Background: The risk of developing asthma is associated with genetic, environmental and lifestyle factors. The aim of this study was to estimate the incidence of, and examine risk factors for developing, asthma using data from the child cohort of the Longitudinal Study of Australian Children.

Methods: The child cohort (aged 4–5 years at baseline) was recruited in 2004 and re-assessed two years later via face-to-face interviews with the primary carer. Asthma diagnosis was ascertained from the question "*Has a doctor ever told you that your child has asthma?*". Multivariate logistic regression was used to examine associations between risk factors reported at baseline and new asthma diagnosis two years later among children with no diagnosis of asthma at baseline.

Results: At baseline, 20% of children aged 4–5 years had ever-diagnosed asthma and the estimated incidence of newly diagnosed asthma over the next two years was 8.6%. Independent risk factors significantly ($p \leq 0.013$) associated with new asthma diagnosis among 6–7 year olds were wheeze (OR=3.0); food/digestive allergies (OR=2.3); and neonatal intensive care after birth (OR=1.6). No association was observed for eczema, passive smoke exposure, ever breastfed, no siblings, 1+ pets in household, English-speaking primary carer, socioeconomic disadvantage, sex or overweight/obesity.

Conclusions: While several of the observed associations are similar to those reported in comparable populations elsewhere, the lack of association with sex, passive smoke exposure, and breastfeeding status suggests that these factors do not have an impact on the incidence of asthma after early childhood.

Support: ACAM is a collaborating unit of the AIHW and is funded by the Department of Health and Ageing.

TP049

INCIDENCE OF ASTHMA AND WHEEZE IN INDIGENOUS CHILDREN LIVING IN URBAN AND REGIONAL AREAS

LM Poulos, R Ampon, A Zinoviev, A-M Waters, W Xuan, HK Reddel, GB Marks
Australian Centre for Asthma Monitoring (ACAM), Woolcock Institute of Medical Research, NSW 2050

Background: There is a large disparity in asthma and asthma-related outcomes in Aboriginal and Torres Strait Islander Australians compared with other Australians.
Aim: The purpose of this study was to compare the incidence of asthma and wheeze over a two year interval among indigenous and non-indigenous children.
Methods: In 2004, the Longitudinal Study of Australian Children recruited two cohorts aged 0–1 years (infant cohort, n=5,107) and 4–5 years (child cohort, n= 4,983). Asthma and wheeze were diagnosed by questionnaire and indigenous status was assessed by self-report. Prevalence rates at baseline and incidence rates over a two year follow-up period were compared between indigenous and non-indigenous children by calculating rate ratios.
Results: In the infant cohort, of whom 4.9% were indigenous, the prevalence of wheeze at baseline was 1.86 times (95% CI 1.52–2.27) higher in indigenous than non-indigenous children but no significant difference was found in the incidence of wheeze over the following two years (IRR 1.21; 95% CI 0.93–1.58). In the child cohort, of whom 3.9% were indigenous, there was no difference in the prevalence of wheeze at baseline among indigenous (19.5%) and non-indigenous children (15.0%) (RR 1.30; 95% CI 0.96–1.75). In this cohort, the prevalence of asthma at baseline was 1.62 times (95% CI 1.18–2.21) higher in the indigenous children but the incidence of newly-diagnosed asthma over the next two years did not differ between the indigenous and non-indigenous children (IRR 0.7; 95% CI 0.33–1.44).
Conclusions: The findings confirm a higher prevalence of reported asthma and wheeze in indigenous compared with non-indigenous children and show that the disparity diminishes with age during childhood. This suggests that the prevalence of wheezing illness in indigenous children is affected by events in early childhood.
Support: ACAM is a collaborating unit of the AIHW and is funded by the Australian Government Department of Health and Ageing.

TP050

GEOGRAPHIC VARIATIONS IN HOSPITAL RE-ADMISSION FOR ASTHMA

Wei Xuan, Leanne M Poulos, Anne-Marie Waters, Guy B Marks

Australian Centre for Asthma Monitoring (ACAM), Woolcock Institute of Medical Research, NSW 2050

Background: Re-admission to hospital within 28 days has been used as an indicator of health system performance in the care of patients with asthma. However, socio-demographic factors may confound its interpretation at a local level.

Aim: The aim of this study was to use national hospital admission data in estimating expected rates of hospital re-admission for asthma at a statistical local area (SLA) level, adjusted for socio-demographic factors.

Methods: Nationwide hospitalisation data (excluding Queensland) between 1996 and 2005 were used to identify hospital re-admissions for asthma within 28 days for the same individual using a linkage key. Expected re-admission rate was calculated for each SLA by logistic regression using data on age and sex distribution, state/territory and socio-economic index for areas (SEIFA) as predictors. Observed-to-expected ratio was then calculated for each SLA.

Results: The overall rate of re-admission within 28 days for asthma was 4.7%. Age group, sex, state/territory and SEIFA were significant predictors of re-admission rates. The median of the observed-to-expected ratio was 0.93 and the 10^{th}, 25^{th}, 75^{th} and 90^{th} percentiles were 0.0, 0.64, 1.22 and 1.59 respectively.

Conclusions: This analysis has identified important local variation in re-admission rates for asthma that are not attributable to measured socio-demographic factors. Examination of the causes of this variation may improve health system performance for asthma care.

Support: ACAM is a collaborating unit of the AIHW and is funded by the Australian Government Department of Health and Ageing.

TP051

THE ATOPIC MARCH: GENETIC, SHARED ENVIRONMENT OR DIRECT EFFECT?

Bircan Erbas[1], Melanie Matheson[2], Mingh Bui[2], John Burgess[2], Hayden Walters[3], Michael Abramson[4], John Hopper[2], Shyamali Dharmage[2]

1. School of Public Health, La Trobe University, Bundoora, VIC 3086

2. School of Population Health, University of Melbourne, Vic 3010

3. Menzies Research Institute, University of Tasmania, Hobart, Australia.

4. School of Public Health & Preventive Medicine, Monash University, Melbourne.

Background: The "atopic march" hypothesis - eczema precedes the development of allergic rhinitis and asthma - is controversial. Little is known about whether the influence of eczema on hay fever and asthma is direct or mediated by other factors such as genes and/or shared environment. We sought to examine the contributions of genes and/or shared environment, to the atopic march hypothesis.

Methods: We used data from the baseline survey of the Tasmanian Longitudinal Health Study. In 1968, 8,583 7-year old school children and their siblings (21,000) were investigated for asthma and other allergies. A novel twin-sibling regression model was used to examine the association between infantile eczema and hay fever and asthma separately.

Results: 182 dizygotic (DZ) twin pairs and 3,696 sib pairs were included in the study. The association between infantile eczema and hay fever was mediated by parental phenotype ($p<0.001$) and infantile eczema in the sibling ($p=0.002$). Hay fever was strongly associated with asthma ($p<0.001$). In the sib model examining the association between hay fever and asthma the effect of hayfever in a sib was no longer significant ($p=0.9$) after adjusting for parental phenotype ($p<0.001$). Infantile eczema was significantly associated with asthma. There was no effect of infantile eczema in a sib ($p=0.86$) on the association between infantile eczema and asthma.

Conclusion: Our findings suggest that different mechanisms are triggered at different stages of the atopic march. There seem to be strong genetic and shared environment components for the infantile eczema - hay fever associations and a stronger genetic component for the hay fever – asthma associations. Conversely, there seems to be a direct effect of infantile eczema on asthma.

Grant Support: NHMRC.

TP052

SKIN PRICK TESTS USING A TWO MILLIMETRE CUT POINT ARE BEST AT IDENTIFYING ELEVATED SPECIFIC IgE CONCENTRATIONS

Daniele Crisafulli[1], Christina Weber-Chrysochoou[1], Warwick Britton[2], Euan Tovey[1], Andrew Kemp[3], Brett Tolle[1], Rita Amin[1], Guy Marks[1]
[1]The Woolcock Institute of Medical Research, NSW 2037 and [2]The Centenary Institute of Cancer Medicine and Cell Biology, NSW 2050 and [3]Department of Allergy and Immunology, Children's Hospital at Westmead, NSW 2145

In epidemiological studies of children, using a skin prick test (SPT) cut point of 2 mm or 3 mm for most allergens, attracts controversy due to a lack in evidence based guidelines. The Childhood Asthma Prevention Study (CAPS) provided an opportunity to assess the wheal size cut point offering the best trade-off between sensitivity and specificity in identifying elevated specific IgE concentrations.
Methods: Subjects were eight year old children who were born in Sydney and had at least one parent or sibling with asthma. SPTs were performed using extracts of *D. pteronyssinus* (HDM), cat hair and epidermis (cat), *A. alternata* (Alternaria) and *L. perenne* (Rye grass) pollen. Serum specific IgE against the same allergens was determined using the Pharmacia ImmunoCAP 250 system. Levels ≥ 0.35 kUA/L were classified as positive. ROC curves were used to examine the relation between weal size and positive ImmunoCAP for each allergen. The agreement of SPTs using cut-points of ≥ 2mm and ≥ 3mm with ImmunoCAP was assessed by kappa (K).
Results:

	HDM n=331	Cat n=331	Alternaria n=328	Rye grass n=330
ROC AUC[1]	0.96	0.90	0.82	0.86
≥ 2mm K	0.87	0.83	0.72	0.75
≥ 3mm K	0.82	0.81	0.70	0.73

[1] Area under ROC curve

Conclusions: Amongst 8-year olds, there is good agreement between SPT and serum-specific IgE for the four inhaled allergens examined. A SPT threshold of ≥ 2mm yields results that are in close agreement with specific IgE in this population.

Supported by the NHMRC.

TP053

USE OF MEDICINES IN CHILDREN WITH ASTHMA: THE AUSTRALIAN ASTHMA EDUCATOR PERSPECTIVE

Bandana Saini [1], Julianne Chong [1], Kirsti Vainio [2], Anders Davidsson [3], Rebekah Moles [1]

1.Faculty of Pharmacy, University of Sydney, Australia 2. Department of Social Pharmacy, University of Kuopio, Finland 3. Department of Pharmaceutical Biosciences, Uppsala University, Sweden

The global burden of childhood asthma is significant. Health care systems are faced with increasing financial costs due to childhood asthma, while children and their carers are affected through reduced quality of life and reduced emotional and physical health. Despite the availability of effective treatment, the quality use of asthma medicines in children remains suboptimal. An investigation was undertaken to explore issues related to children's asthma medicine usage from the perspective of the health care professional. Literature evidences problems from the patient's perspective, but an informed reality is expected from health care professional's views about 'issues' in medicines use and this has been relatively unexplored in the past. Semi-structured qualitative interviews were conducted with a convenience sample of 21 Australian asthma and respiratory educators. Interviews were audiotaped, transcribed verbatim, and transcripts thematically analysed with the assistance of NVivo 7. Emergent themes associated with health care professionals, parents, medicines and children were found. Major issues included a lack of information provided to parents, poor parental understanding of medicines, the high cost of medicines and devices, child self-image, the need for more child responsibility over asthma management and the lack of standardisation, access to and funding for educational resources on childhood asthma. There are therefore a multitude of key issues that may affect asthma medicines usage in children. This research will help inform the development of educational tools on the use of medicines in childhood asthma that can be evaluated for their effectiveness in getting key messages to their target audience (children, carers, and teachers).

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Conflict of Interest: NONE

TP054

RESISTIN AND AIRWAY INFLAMMATION IN CHILDREN WITH ALLERGIC ASTHMA

Mandana Arshi [1], John Cardinal [2], Rebecca J Hill [1], Peter SW Davies[1], Claire Wainwright [3,4]

[1]The University of Queensland, Children's Nutrition Research Centre, Royal Children's Hospital, QLD, 4029 Australia. [2]Department of Chemical Pathology, Queensland Health, Royal Brisbane Hospital, QLD, 4029, Australia. [3]Queensland Children's Respiratory Centre, Royal Children's Hospital, QLD, 4029, Australia. [4]The University of Queensland, Department of Paediatrics and Child Health, Royal Children's Hospital, Australia.

Background: A relationship between obesity and asthma has been evidenced by numerous studies on large populations. However, little is known about the linking mechanism. Products of adipose tissue, known as adipokines, including leptin, resistin, TNF-α, PAI-1 and IL-6, have been found to be associated with inflammatory states. This study aimed to determine the relationship between adipokines and respiratory inflammation in a cohort of children with persistent allergic asthma.

Methods: Thirty-one children (20 with allergic asthma (AA) and 11 non-allergic healthy control (HC)) aged 6.0-17.9 years of age were recruited. Fasting blood samples were obtained to test for serum levels of leptin, resistin, TNF-α, PAI-1 and IL-6. Inflammation in the airways was tested by measuring levels of exhaled nitric oxide (Fe_{NO}), adiposity was determined by calculating the percentage of body weight made up of fat mass (FM %) using air displacement plethysmography, and allergic state was assessed by skin prick test.

Results: No significant differences were found between AA and HC groups with respect to %FM, leptin, resistin, TNF-α and IL-6. However, in AA group Fe_{NO} was significantly higher (mean=31.14±28.01) compared with HC (mean=7.38±3.91), and a significant negative correlation was found between Fe_{NO} and resistin ($r = -0.46$, $p=0.04$), but not with other adipokines, which was not dependent on %FM.

Conclusions: Patients with persistent allergic asthma had significantly higher Fe_{NO} levels, which were negatively related to the levels of resistin. This suggests that resistin may have protective effects on airway-inflammation in children with persistent allergic asthma.

Supported by: The University of Queensland

Conflict of interest: NO

TP055

DOES VENTILATION HETEROGENEITY PREDICT AIRWAY HYPERRESPONSIVENESS IN COPD OR OLDER ASTHMATIC SUBJECTS?

Kate Hardaker[1], Sriram Mahadev[2], Sue Downie[1,3], Norbert Berend[1,3], Gregory King[1,2,3], Cheryl Salome[1,3]

[1] Woolcock Institute of Medical Research, University of Sydney, NSW 2006
[2] Department of Respiratory Medicine, Royal North Shore Hospital, NSW 2065 [3] Cooperative Research Centre for Asthma and Airways, NSW, 2050

Ventilation heterogeneity in the conducting airways (S_{cond}) measured by the multiple breath nitrogen washout (MBNW) predicts airway hyperresponsiveness (AHR) in asthmatic subjects. We hypothesise that this is an underlying mechanism for AHR, independent of disease. To test this we compared the relationship of AHR to ventilation heterogeneity in COPD and age matched asthmatics.

Methods: 12 COPD and 15 asthmatic subjects (60-86yrs) underwent baseline spirometry, MBNW, and methacholine (MCh) challenge. AHR was expressed as dose response ratio (DRR = %fall FEV_1/µmol MCh). Ventilation heterogeneity of the conducting (S_{cond}) and acinar (S_{acin}) airways were calculated from the MBNW.

Results: Values are mean ± SD or geometric mean (95% CI).

	FEV_1 (% pr)	**S_{acin}**	**S_{cond}**	**DRR**
COPD	69.9 ± 7.1	0.60 ± 0.13	0.06 ± 0.02	2.3 (1.3–3.5)
Asthma	71.7 ± 9.6	0.21 ± 0.04	0.07 ± 0.02	9.5 (3.1–22.6)
p-value	0.32	<0.01	0.79	0.12

In COPD, DRR correlated with S_{cond} (r= 0.63, p=0.03), but not with S_{acin} (r= -0.25, p=0.44). In asthmatic subjects, DRR correlated with S_{acin} (r= 0.66, p=0.008) but not with S_{cond} (r= -0.08, p=0.77).

Conclusions: S_{cond} is related to airway responsiveness in COPD, but most subjects in this study did not have AHR. In contrast to younger asthmatics, AHR is predicted by S_{acin}, not S_{cond} in older asthmatics, suggesting more peripheral disease processes. Thus, increased baseline S_{cond} does not predict AHR in either COPD or older asthma.

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Conflict of interest: No

TP056

REFERENCE EQUATIONS FOR IMPEDANCE PARAMETERS MEASURED BY THE FORCED OSCILLATION TECHNIQUE

NJ Brown[1], CM Salome[1,2], GG King[1,2,3], M Hunter[4], AW Musk[4,5], AL James[4,5]
[1]Woolcock Institute of Medical Research, Sydney, Australia, [2]The University of Sydney, Australia, [3]Royal North Shore Hospital, Sydney, Australia, [4]Sir Charles Gairdner Hospital, Perth, Australia [5]University of Western Australia, Perth, Australia

Previous studies to determine reference equations for forced oscillatory parameters are limited either by small sample sizes, inclusion of only a single sex, or selection of "normal" subjects from people presenting at clinics or hospitals.

Aim: To determine normal predictive equations for respiratory system resistance (Rrs) and reactance (Xrs) in a large randomly selected sample from a general population.

Methods: Prospective respiratory health survey of the general population in Busselton, WA, between 2005 and 2007. Subjects had measures of spirometry, atopy by allergen skin prick tests, and Rrs and Xrs (6, 11, 19 Hz) by forced oscillation technique. Eligible subjects were never smokers, with no history of respiratory disease, no symptoms of cough, shortness of breath or chest tightness in the previous 12 months, and no respiratory tract infections in the previous 4 weeks.

Results: 459 eligible subjects (167 male) aged 18 to 93 yrs had technically satisfactory FOT measurements. Rrs6 (95%CI) was 2.94 (2.8 to 3.1) in males and 3.49 (3.4 to 3.6) in females. Xrs6 was -0.34(-0.43 to -0.30) in males and -0.54(-0.58 to -0.49) in females. The predictive normal equations were: lnRrs6 = 4.19 - 0.0017(Age) + 0.0088(Wt.kg) - 0.0211(Ht.cm), R^2=0.24, and expXrs6 = -1.30 - 0.0014(Age) - 0.0033(Wt.kg) + 0.0134(Ht.cm), R^2=0.28.

Conclusion: In this study, Rrs6 and Xrs6 were predicted by age, height and weight but not sex. These data provide predictive equations for forced oscillatory parameters, in well-characterised normal subjects from a large general population.

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Nomination: None
Conflict of Interest: None

TP057

THE EFFECT OF INHALED CORTICOSTEROIDS ON AIRWAY DISTENSIBILITY IN ASTHMA

Jessica Kermode[1,2], Nathan J Brown[1,2], Cheryl Salome[1,2], Lucy Keatley[3], Robyn Schoeffel[3], Norbert Berend[1,2], Greg King[1,2,3]

[1]*Woolcock Institute of Medical Research, Glebe NSW 2037,* [2]*The University of Sydney, Camperdown NSW 2006,* [3]*Department of Respiratory Medicine, Royal North Shore Hospital, St Leonards NSW 2065*

Airway distensibility has been proposed as a potential marker of airway remodelling and is reduced in asthma. The contribution of current inflammation to distensibility is unknown. The aim of this study was to determine the effect of inhaled corticosteroids (ICS) treatment on airway distensibility in asthma.

Methods: Twelve asthmatic patients (7 males, 20-60yrs) underwent 18 weeks of treatment with fluticasone propionate 500µg bd. Exhaled nitric oxide (FeNO), spirometry, methacholine challenge, and distensibility between 75% TLC and TLC by FOT were measured at baseline, 4, 8, 14 and 18 weeks.

Results: At baseline, FEV_1 was 81.7±17.8 % of predicted and distensibility was 0.20±0.19 $L.s^{-1}.cmH_20^{-1}.L$ lung volume, indicating reduced distensibility in this group. Baseline distensibility correlated with disease duration (p=0.005), but not with age (p=0.78), DRS (p=0.22), FeNO (p=0.64) or spirometry (p=0.85). After 18 weeks of treatment, FeNO mean(95%ci) decreased from 8.5(6.8-10.5)ppb to 6.3(95%ci 5.0-7.4)ppb (p=0.04). DRS decreased from 8.38(6.4-10.4) to 5.25(3.6-6.9) %/µmol (p=0.0003). There was no change in distensibility (p=0.46) or spirometry (p=0.54).

Conclusion: 18 weeks of ICS treatment did not change distensibility in this group of well-controlled asthmatics, despite improvements in FeNO and DRS. This suggests that current inflammation does not contribute to distensibility. The relationship between distensibility and disease duration implies that reduced airway distensibility in asthma is related to long-term structural changes, such as airway remodelling.

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Nomination: None
Conflict of Interest: None

TP058

SIMILAR BETWEEN-DAY REPEATABILITY OF FORCED OSCILLATION MEASUREMENTS IN ASTHMATICS COMPARED WITH NORMALS

Nicholas Coatsworth[1], Robin Schoeffel[1], Cheryl Salome[2,3], Norbert Berend[1,2,3], Gregory King[1,2,3]

[1]Department of Respiratory Medicine, Royal North Shore Hospital, St Leonards 2065, the [2]Woolcock Institute of Medical Research, University of Sydney 2006 and the [3]CRC for Asthma and Airways, Camperdown 2050

Introduction: FOT measurements of resistance (Rrs), conductance (Grs) and reactance (Xrs) are measures of airway function that are effort independent and therefore may be used for asthma monitoring by patients.

Aims: Compare the day-to-day variability in FEV1, Rrs and Xrs in asthmatics and non-asthmatics in the laboratory.

Methods: Ten asthmatics (6 males) and 9 normals (4 males) performed repeated spirometry and FOT over 10 consecutive days in the laboratory. Subjects were tested at the same time each day and were asked to continue all usual medications, including bronchodilators. The within subject day-to-day variability was measured from the mean variances.

Results: Asthmatic subjects mean±SD age of 38±12 years was similar to 31±3 years for non-asthmatics. FEV1 was lower in asthmatics (3.22±0.67L vs 3.64±0.88L, $p<0.001$), while Rrs was higher (3.30±1.00 vs 2.44±0.76 cmH20/L/s, $p<0.001$) and Xrs was lower (-0.95±0.42 vs -0.75±0.28). For Rrs, the within subject SD correlated with mean ($r=0.86$, $p<0.0001$) but not for FEV1, Grs or Xrs. The within subject day-to-day variability (mean within subject SD) was similar between asthmatics non-asthmatic subjects for FEV1 (0.26L vs 0.15L), Grs (0.04 vs 0.03L/s/cmH20) and Xrs (0.21 vs 0.17cmH2O/L/s).

Conclusion: Day to day variability in Grs and Xrs measurements in asthmatics are comparable to those in non-asthmatics. Variability of Rrs between days is not a useful measure of variability in lung function since it is strongly related to mean Rrs. This has implications for the use of FOT in monitoring lung function in asthmatics.

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TP059

VENTILATION-PERFUSION ABNORMALITIES IN SEVERE ASTHMA

Camelia Borta, Peter McLaughlin, Dee Nandurkar, Teresa Young, Matt Salamonsen, Phil Bardin

Department of Respiratory and Sleep Medicine, Monash Medical Centre

Background: In severe asthma, ventilation-perfusion relationships (V/Q) in the lung may be abnormal and little is known regarding differences compared to a normal population. Studies are also limited examining associations between V/Q abnormalities, severity of airflow limitation and zonal distributions.

Aim: We examined regional differences of V/Q in patients with asthma and non-asthmatic normal subjects and related measurements to the degree of airflow limitation.

Methods: Ventilation-perfusion (V/Q) radionuclide scans were obtained in 10 patients with stable severe asthma and in 10 age-matched control subjects. Individual V/Q scans examined V/Q mismatch, were graded for heterogeneity (scored on a scale of 1-3), and the geometric mean of maximal extent of radiotracer present in lung fields was used to assess zonal distribution (upper vs. lower zones). We correlated the degree of heterogeneity with measurements of FEV_1. To ascertain if there is any significant difference in the zonal distribution in the 2 groups a two sample independent t-test was used.

Results: The asthma cohort had reduced lung function as reflected by FEV_1 measurements (pre-BD 55±5.1, post-BD 66+/- 6%predicted, mean±SD). V/Q abnormalities were matched in all but one patient. Clumping of radiotracer was noted in one patient. Heterogeneity was mild in 4 patients, moderate in 3 patients and severe in one patient and the degree of heterogeneity correlated significantly with severity of airflow obstruction (n=9; r=0.75, p=0.03). The mean percentage difference of ventilation in upper versus lower zones in the asthmatic group was 4.6± 18.60(mean±SD) and of perfusion 15.75±15.33 respectively. The mean percentage difference in ventilation in the normal cohort was -7.29±7.30 (mean ± SD) and in the perfusion scan -1.29 ±7.29 (mean±SD). In the 2 groups there was a statistically significant difference between upper and lower zone distributions of ventilation (p=0.01).

Conclusion: Ventilation-perfusion is abnormal in stable severe asthma and reflects the degree of airway obstruction. We identify maldistribution of ventilation as a novel abnormality suggesting that predominant upper zone ventilation may accompany airflow limitation in severe asthma.