rAsp f3 and f4 are associated with bronchiectasis in allergic fungal airways disease.

Kerry Woolnough (MD)¹, Michelle Craner (DipHE)¹, Catherine H. Pashley (PhD)¹, Andrew J. Wardlaw (MD. PhD)¹.

¹Institute for Lung Health/Department of Infection, Immunity and Inflammation, University of Leicester, Glenfield Hospital, Leicester, UK. ²Department of Health Sciences, University of Leicester, UK.

Corresponding author: Professor Andrew J. Wardlaw, Institute for Lung Health, Respiratory NIHR Biomedical Research Unit, University Hospitals of Leicester, Leicester LE3 9QP, United Kingdom.

Tel: 0116 2583841; Fax: 0116 2702787; Email: Aw24@le.ac.uk

Supported by the National Institute for Health Research (NIHR) Leicester Respiratory Biomedical Research Unit. This report presents independent research funded by the National Institute for Health Research (NIHR). The views expressed are those of the author(s) and not necessarily those of the NHS, the NIHR or the Department of Health.

Manuscript word count: 1039

Figures: 0

Tables: 1

Trial Registration: Not applicable

Kerry Woolnough: No Conflict of Interest

Michelle Craner: No Conflict of Interest

Catherine H. Pashley: No Conflict of Interest

Andrew J. Wardlaw: Has been a member of an advisory board for GSK, Pulmocide, KNOPP and Astra Zeneca

Allergy to thermotolerant filamentous fungi, particularly *Aspergillus fumigatus*, is closely associated with fixed airflow obstruction, bronchiectasis and other radiologically defined abnormalities such as mucus plugging (1). However not all asthmatics who are IgE sensitized to *A. fumigatus* develop these complications. To identify markers of poor outcomes in fungal allergy with asthma (and cystic fibrosis), the term allergic bronchopulmonary aspergillosis (ABPA) was coined. However this syndrome was defined largely using criteria that represent a florid immunological response to *A. fumigatus* rather than clinically relevant outcomes, a problem that remains with the recent modifications recommended by the International Society for Human and Animal Mycology (ISHAM) (2). Moreover asthmatics fulfilling all the criteria for ABPA are unusual, whereas fungal allergy in asthma is common, particularly in more severe disease. Denning and colleagues proposed the term severe asthma with fungal sensitization (SAFS), essentially defined as ABPA with a low IgE, to capture this larger group. However not all fungal sensitized asthmatics have severe disease and not all fungi are associated with lung damage. We have proposed that under the current state of knowledge a more inclusive term such as allergic fungal airways disease should be used to describe all asthmatics IgE sensitized to *A. fumigatus* and related fungi (3). Irrespective of which term is used the need remains to identify biomarkers which distinguish between those asthmatics with fungal IgE sensitization who will develop lung damage as a result of their asthma and those who won't. In this paper we have asked the question whether specific IgE to different allergenic components of *A. fumigatus* could be a more specific marker of lung damage in asthma than total specific IgE. The aim of the study was therefore to determine in a group of fungal sensitized asthmatics, the relationship between sIgE to the allergen components Asp f1-4 and Asp f6 and fixed

airflow obstruction, bronchiectasis and other recognized radiological markers of fungal allergy. A secondary aim was to determine whether the components were useful in distinguishing between asthmatics who meet the criteria for ABPA, SAFS or neither subset.

The majority of fungal extracts used to measure specific IgE use crude extracts which contain non-allergenic and allergenic proteins, polysaccharides and lipids that have been shown to cross-react both between and within species (4). Molecular-based allergy (MA) diagnostics benefit from being standardized, have a low degree of variability and have been developed from either recombinant or purified native molecules (5). Studies that have investigated the ability of specific IgE components to *A. fumigatus* to differentiate between ABPA and sensitization to *A. fumigatus* have shown conflicting results. rAsp f3, rAspf4 and rAsp f6 have been demonstrated to be specific markers for ABPA in both cystic fibrosis and asthma (6, 7); whilst Asp f1 and 2 have also been shown as diagnostic markers of ABPA with AUC values of 0.75 and 0.78 respectively in a Japanese asthma population (8). However as noted above the relevance of these observations to clinical outcomes is not clear.

In this study concentrations of IgE to the molecular allergens rAsp f1-4 and rAsp f6 were measured in asthmatics recruited opportunistically from asthma and fungal allergy clinics by AJW. They were a subset of subjects recruited for a larger study (1). Inclusion criteria included a positive specific IgE to *A. fumigatus* ($>0.35\text{kU/L}$) using the Phadia ImmunoCAP system, with a bias in recruitment towards those with a sIgE of $>1.0\text{ IU/L}$. Demographic details (age, sex, BMI), clinical data (age of asthma diagnosis, asthma duration, GINA status and sputum cell counts) and spirometry were collected during a stable visit using standard methods, recording post-bronchodilator FEV₁ as percentage predicted. High resolution computerised

tomography (HRCT) scans were undertaken of the whole thorax using a Siemens Somaton Definition AS plus spiral scanner (Siemens Healthcare, Knoxville, TN).

Bronchiectasis (present/absent) was scored based on the radiologist's report.

Descriptive statistics, analyzed using SPSS for windows, were expressed as mean (SD, 95% CI) or median (interquartile range). Non-parametric data were analyzed using Spearman's correlation and Dunn-corrected Kruskal-Wallis test for multiple group comparisons.

85 asthmatics were recruited of which 57.8% (n=44) were male, 91.8% (n=78) had a GINA score ≥ 3 and the majority had either early onset atopic disease (50.6%, n=43) or late onset eosinophilic asthma (40%, n=34). There was no difference in the components when separated by GINA score, endotype or atopic status that reached statistical significance. 37.6% (n=32) met the criteria for ABPA (2), 21.2% (n=18) met the criteria for SAFS, whilst the majority (41.2%, n=35) were sensitized to *A. fumigatus* but did not fit either of these diagnostic classifications. The differences between the levels of IgE to components in these groups were statistically significant (see table 1).

rAsp f6 was the only component to correlate well with total IgE ($r=0.798$, $p<0.001$).

Although significant at the <0.05 level the other components were only weakly correlated with total IgE, *A. fumigatus* IgE, peripheral blood eosinophil count and asthma duration.

No correlations were seen between rAsp f1-4 or rAsp f6 and post-bronchodilator FEV₁. Bronchiectasis was associated with a raised level of rAsp f3 (4.4 (0.81-12.9) vrs 0.34 (0.07-3.91), $p=0.008$) and rAsp f4 (0.47 (0.03-3.16) vrs 0.09 (0.01-0.35), $p=0.033$). No other statistical differences were seen between the components and the frequency of the radiological abnormalities studied (tree-in-bud, bronchial wall

thickening, mucoid impaction, collapse/consolidation, air trapping, fleeting shadows and fibrosis) although receiver-operating characteristic curves did demonstrate an AUC value of 0.71 for rAsp f4 in predicting the development of fleeting shadows. In asthmatics sensitized to crude extracts of *A. fumigatus*, irrespective of their diagnostic label, rAsp f3 and rAsp f4 were found to be associated with the development of bronchiectasis. A potential role for sensitization to the allergens rAsp f3 and f4 in causing bronchiectasis is therefore worthy of further study. rAsp f4 was found to be related to the development of fleeting shadows and total IgE was most closely related to Asp f6 which is commonly increased in atopic dermatitis. Patients who fulfilled the criteria for ABPA had a higher level of components than those with SAFS or neither label, but this is to be expected because ABPA is defined by a florid immunological response to fungal extracts. In summary rAsp f3 and f4 may be markers for the development of bronchiectasis in allergic fungal airway disease

Table 1. Differences in components between those with ABPA, SAFS and those sensitized to *A. fumigatus*. *median (interquartile range); ** AUC value.

	ABPA (n=32)	SAFS (n=18)	Sensitized to <i>A.fumigatus</i> (n=35)	P value
rAsp f1	3.58 (0.27-15.95)* 0.68**	0.14 (0.13-1.05)* 0.25**	0.54 (0.17-5.15)* 0.50**	0.002
rAsp f2	2.32 (0.26-9.72)* 0.73**	0.13 (0.05-0.66)* 0.29**	0.34 (0.07-2.11)* 0.42**	0.001
rAsp f3	7.04 (1.04-14.65)* 0.68**	0.33 (0.01-1.58)* 0.27**	1.06 (0.1-9.79)* 0.49**	0.002
rAsp f4	1.33 (0.15-7.39)* 0.74**	0.02 (0.01-0.07)* 0.24**	0.12 (0.02-0.85)* 0.44**	<0.001
rAsp f6	0.20 (0.02-2.12)* 0.71**	0.01 (0.01-0.01)* 0.23**	0.04 (0.01-0.12)* 0.49**	<0.001
No. of	1-2 21.9%	38.9%	37.1%	
positive	3-4 43.8%	27.8%	37.1%	
components	5 28.1%	0%	5.7%	
Total IgE	2613 (1535-5000)*	244 (219-534)*	1078 (646-2561)*	<0.001
IgE	36.45 (17.2-74.2)*	5.87 (2.88-9.24)*	10.7 (3.36-25.2)*	<0.001
<i>A. fumigatus</i>				

ABPA = allergic bronchopulmonary aspergillosis; SAFS = severe asthma with fungal sensitization

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