

Genetic Polymorphisms of ADRB2 and IL10 May Be Associated with the Risk of IgE Sensitization to Digestive Powders in Exposed Medical Personnel

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Key Words

Genetic polymorphism • Specific IgE • Digestive • Pancreatic extract • α -Amylase • ADRB2 • IL10 • IL4

Abstract

Background: It has been reported that pancreatic extracts may induce IgE-mediated respiratory allergy in medical personnel. The aim of the study was to identify genetic factors associated with IgE sensitization to digestive powders containing pancreatic extract. **Methods:** This case-control study was performed on 153 subjects routinely exposed to digestive powder and on 123 nonexposed controls working in Ajou University Hospital. Skin prick testing was performed using 4 commonly used digestive powders and α -amylase. Serum specific IgE levels were measured by ELISA. Three single nucleotide polymorphisms, ADRB2 46A>G, IL10 –1082A>G and IL4 –589T>C, were genotyped using the single base extension method. **Results:** The positive rate of serum specific IgE to digestive powder was significantly higher in the 41 (26.8%) exposed personnel with work-related respiratory symptoms than in controls (24.4 vs. 5.4%, $p = 0.012$). Thirty-nine (25.5%) of the 153 exposed personnel

were found to have an allergy to digestive powder, as determined by a positive skin prick test and/or a high serum specific IgE level to digestive powder. The ADRB2 46A>G and IL10 –1082A>G polymorphisms were found to be significantly associated with the development of an allergy to digestive powder in exposed medical personnel by multiple logistic regression analysis after controlling for age, atopy and job type (pharmacist or nurse; $p = 0.035$ and $p = 0.027$, respectively). **Conclusion:** These results suggest that the genetic polymorphisms ADRB2 46A>G and IL10 –1082A>G are genetic factors that increase IgE sensitization to pancreatic extracts in medical personnel occupationally exposed to digestive powders.

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Introduction

Enzymes in the pharmaceutical, detergent, baking and agriculture industries have been sporadically reported to be occupational allergens [1–5]. Pancreatic extracts, which commonly contain digestive enzymes, have been identified as occupational allergens that result in IgE-me-

diated respiratory allergies such as asthma, rhinitis and conjunctivitis [4–6]. Recently, we reported that digestive powders may have induced IgE-mediated respiratory allergic symptoms in 30% of exposed medical personnel at a university hospital [5], and previous reports have described pancreatic extract-induced allergy and asthma in pharmaceutical workers [6].

The identification of individual risk factors of IgE sensitization is a matter of common interest. Several studies have been undertaken to identify genes that contribute to the development of drug allergies. Vandbriel et al. [7] reviewed candidate genetic polymorphisms, including human leukocyte antigen and transporter associated with antigen processing. In addition, several genetic polymorphisms associated with asthma and atopy share a range of pathogenic mechanisms [7]. Recently, several studies have suggested that genetic polymorphisms in chromosome 5q31–q33 may play roles in atopy and asthma [8, 9]. This region contains many genes, including β_2 -adrenergic receptor gene (ADRB2), IL13 and IL4, which are known to be involved in immune and inflammatory response. IL4 is essentially required for IgE class switch and cooperates with IL13 to increase IgE production [10]. Furthermore, evidence of linkages between the IL4 –589C>T or ADRB2 Arg16Gly polymorphisms and total serum IgE and house dust mite-specific IgE antibody levels has been reported in several different populations [11–13]. However, other investigators found no such association [14, 15].

ADRB2 is another interesting gene and is located in chromosome 5q31. The β_2 -adrenergic receptor is a cell surface receptor that is expressed by a variety of cell types, including mast cells, B and T lymphocytes as well as airway smooth muscle cells. Recent data from mice show that β_2 -adrenergic receptor stimulation on activated B cells increases IgE levels [16].

IL10 is a powerful regulatory cytokine produced by lymphoid cells that exerts its function by inhibiting macrophage/monocyte and T cell lymphocyte replication and the secretions of inflammatory cytokines [17]. Guglielmi et al. [18] reported that a polymorphism in the IL10 promoter region (–1082G/A) was associated with immediate β -lactam allergy in atopic women.

Given this background, we aimed to investigate the roles of 3 single nucleotide polymorphisms (SNPs), namely IL4 –589C>T, IL10 –1082G>A and ADRB2 Arg16Gly, which have been previously associated with total and/or specific IgE levels, in subjects with digestive powder-induced occupational respiratory allergies. We studied the prevalence of IgE sensitization to digestive powder, which

has been associated with work-related respiratory symptoms, and the prevalence of possible associations between these 3 SNPs and allergy to digestive powders in medical personnel.

Subjects and Methods

Subjects

We enrolled 153 exposed subjects and 123 nonexposed controls working in Ajou University Hospital (South Korea). The exposed subjects were all nonsmoking women; 135 nurses and 18 pharmacists. The 123 healthy controls were negative by allergy skin prick testing to common inhaled allergens and digestive powder, and had no allergic disease history.

All subjects were provided written questionnaires, modeled on ISAAC questionnaires [19], which addressed histories of allergic disease and work-related symptoms. Work-related respiratory symptoms were defined as rhinitis symptoms with rhinorrhea, sneeze and congestion, and asthma symptoms including a cough, dyspnea and chest tightness during and after handling digestive powders.

Skin Prick Test to Digestive Powders and ELISA for Digestive Powder-Specific IgE Antibodies

We performed skin prick test (SPT) using 4 commonly used digestive powders (Bears®[®], Beszyme®, Festal®, Pancron®) and using α -amylase which is the major active ingredient in these powders. Total serum IgE levels were measured using ImmunoCAP (Phadia, Uppsala, Sweden). ELISA were used to detect serum-specific IgE antibodies to α -amylase and budiastase, which are ingredients common to all 4 powders, and to one powder Bears. All 4 digestive powders contain porcine pancreatic extracts, which include α -amylase and budiastase from *Aspergillus oryzae*. Furthermore, the proportions of ingredients vary only slightly in these powders, and thus, we performed ELISA for specific IgE only on Bears powder, which is the most commonly used digestive powder at our institute. The positive cutoff value used for specific IgE was set at the mean absorbance value of the 123 unexposed healthy controls plus 3 standard deviations. The presence of an allergy to digestive powder was determined by a positive SPT, defined as a wheal size of ≥ 3 mm, to any one of the four digestive powders and α -amylase and/or a positive ELISA result to either α -amylase, budiastase or Bears.

SNP Genotyping

Three genetic polymorphisms were genotyped using SNaP-shot ddNTP primer extension kits (Applied Biosystems, Foster City, Calif., USA), as recommended by the manufacturer.

Statistical Analysis

Differences of clinical characteristics between exposed and nonexposed subjects were examined by independent t test for continuous variables and χ^2 test for categorical variables. Also, we examined differences of clinical characteristics between pharmacists and nurses by independent t test and χ^2 test (SPSS 12.0; SPSS Inc., Chicago, Ill., USA). We analyzed clinical features stratified by an allergy to digestive powders using independent t test for

Table 1. Clinical characteristics of the study subjects

	Exposed subjects			Healthy controls (n = 123)
	total (n = 153)	pharmacists (n = 18)	nurses (n = 135)	
Age, years*,**	27.3 ± 3.2	25.8 ± 3.6	27.5 ± 3.1	37.3 ± 15.2
Working duration, months	41.9 ± 27.8	37.3 ± 35.7	42.5 ± 26.6	NA
Atopy (%)*	51 (33.3)	8 (44.4)	43 (31.9)	12/85 (14.1)
Work-related respiratory symptoms (%)	41 (26.8)	5 (27.8)	36 (26.7)	NA
Total IgE, KU/l	124.4 ± 211.3	170.7 ± 258.8	118.1 ± 204.4	39.0 ± 58.1
SPT on digestives (%)	38 (24.8)	6 (33.3)	32 (23.7)	NA
Specific IgE to digestives (%)*	16 (10.5)	3 (16.7)	13 (9.6)	2 (1.6)
Prevalence of an allergy to digestive powder (%)*	39 (25.5)	6 (33.3)	33 (24.4)	2 (1.6)

Figures in parentheses are percentage. SPT = Skin prick test; NA= not available.

* p value <0.05 for exposed subjects vs. healthy controls; ** p value <0.05 for pharmacists vs. nurses.

continuous variables and χ^2 test for categorical variables, and p values corrected using false discovery rates.

To evaluate the effects of genetic polymorphisms on an allergy to digestive powders, we performed logistic regression controlling for job title, age and atopy. The p values were corrected by false discovery rates (R 2.7.1; R Foundation for Statistical Computing, Vienna, Austria), which is a statistical means to correct for confounding effects of multiple genetic and clinical factors. Associations are expressed as adjusted logistic odds ratio (OR) and 95% confidence intervals (95% CI). p < 0.05 were regarded as significant.

Results

Clinical Characteristics of the Study Subjects

All exposed medical personnel were nonsmoking women in their mid-twenties, whereas the mean age of the 123 nonexposed healthy controls (66 males, 57 females) was 37.3 ± 15.2 years, which was higher than the mean age of the exposed personnel by ANOVA with Bonferroni's correction (p < 0.05). The 153 exposed subjects were classified into subgroups based on their likely exposure levels, that is 135 nurses and 18 pharmacists. No significant difference was present between the exposed nurse and pharmacist subgroups in terms of mean age, atopy rate, working duration or history of allergic disease (table 1).

Forty-one (26.8%) of the 153 exposed subjects complained of work-related respiratory symptoms. Working durations at the institute and serum total IgE levels were found to be significantly higher in those with work-related

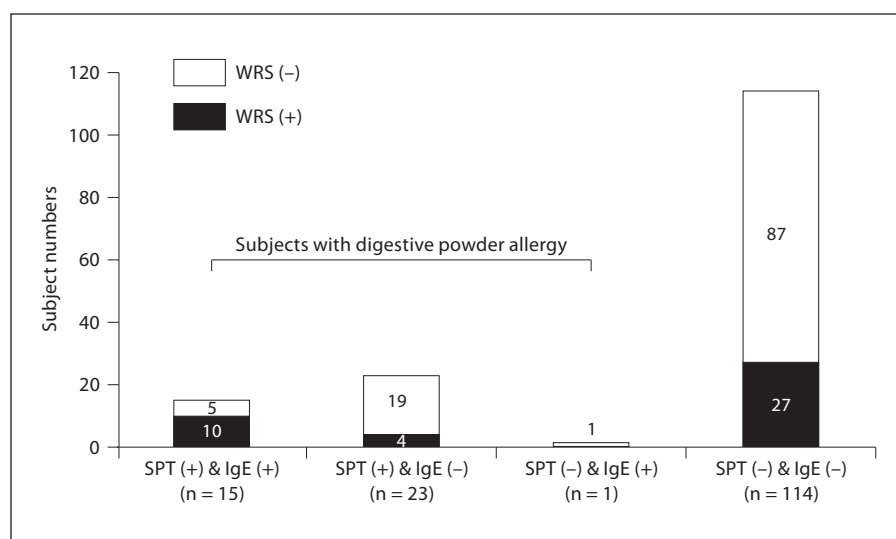
respiratory symptoms (51.93 ± 27.8 vs. 38.27 ± 27.0 months in exposed subjects with vs. without work-related respiratory symptoms and 169.71 ± 260.0 vs. 107.59 ± 188.5 KU/l in exposed subjects with vs. without respiratory symptoms; p = 0.004 and p = 0.047, respectively). On the other hand, no significant difference was found between exposed subjects with and without work-related respiratory symptoms in terms of atopy rate (36.6 vs. 32.1%) or job title (27.8% of pharmacists vs. 26.7% of nurses).

SPT and Specific IgE Antibodies to Digestive Powder

Of the 153 exposed subjects, 38 (24.8%) had a positive SPT response to at least one of powders, and 16 (10.5%) were found to have powder-specific IgE. The positive rates of serum specific IgE were 9.8% (15/153) for α -amylase, 9.2% (14/153) for Barse and 2.0% (3/153) for bio-diastase. SPT positivity among the exposed subjects was 15.7, 16.3, 18.3, 17.0 and 16.3%, for α -amylase, Barse, Beszyme, Festal and Pancron, respectively. Strong SPT-positive rates (defined as the wheal ratio of allergen to histamine ≥ 3) were 5.9% for α -amylase and 3.9% for Barse.

Thirty-nine (25.4%) of the 153 exposed subjects had an allergy to digestive powder: 15 showed a positive SPT finding and specific IgE, 23 a positive SPT finding with no specific IgE and 1 a negative SPT finding associated with specific IgE to digestive powder (fig. 1). The prevalence of serum specific IgE antibodies to digestive powder was significantly higher in subjects with work-related respiratory symptoms than in those without (24.4 vs. 5.4%,

Fig. 1. The prevalences of work-related respiratory symptoms among 153 subjects exposed to digestive powders. Thirty-nine (25.4%) of the 153 had an allergy to digestive powder: 15 showed a positive SPT finding and specific IgE, 23 a positive SPT finding with no specific IgE and 1 a negative SPT finding associated with specific IgE to digestive powder. WRS = Work-related respiratory symptoms.



$p = 0.002$). Although SPT results tended to be higher in exposed subjects with symptoms, no statistical significance was found (34.1 vs. 21.4%). Furthermore, exposed subjects with a digestive powder allergy had significantly higher levels of serum total IgE (216.9 ± 239.7 vs. 94.6 ± 193.1 KU/l, $p = 0.005$; table 2), although mean working duration at the institute (46.07 ± 29.7 vs. 40.51 ± 27.0 months) and atopy rate (43.6 vs. 29.8%) were higher for exposed subjects with an allergy than in those without, but this was not significant. When we compared prevalence of digestive powder allergy by exposed subject job title, 33% of pharmacists and 24% of nurses were found to have an allergy ($p = 0.52$).

Associations between Genetic Polymorphisms with Allergy to Digestive Powder

In the present study, we investigated 3 genetic polymorphisms, namely ADRB2 46A>G, IL10 -1082A>G and IL4 -589T>C, which have all been previously related to allergy and asthma. These genetic polymorphisms have minor allele frequencies of >5% in healthy control group. All 3 genetic polymorphisms were found to comply with the Hardy-Weinberg equilibrium. It was found that the ADRB2 46A>G and IL10 -1082A>G polymorphisms were significantly associated with allergy to digestive powder ($p = 0.035$ and $p = 0.027$, respectively; table 3). Furthermore, the prevalence of a digestive powder allergy was significantly higher in subjects' AA genotype of ADRB2 46A>G and AG or GG genotype of IL10 -1082A>G than in subjects with other genotypes [40 vs. 60%, OR = 1.694, 95% CI 1.077–2.666 and 35.3 vs. 64.7%,

Table 2. Clinical characteristics of subjects allergic to digestive powder

	Allergy to digestive powder		p_c^1
	positive (n = 39)	negative (n = 114)	
Age, years	27.55 ± 2.6	27.19 ± 3.4	0.548
Job title, P/N	6/33	12/102	0.52
Work duration, months	46.07 ± 29.7	40.51 ± 27.0	0.468
Atopy (%)	17 (43.6)	34 (29.8)	0.288
Total IgE, IU/ml	216.93 ± 239.7	94.56 ± 193.1	0.005

Subjects were dichotomized by job as pharmacists (P) and nurses (N). The presence of an allergy to digestive powder was determined by a positive skin prick test, defined as a wheal size of ≥ 3 mm, to any one of the four digestive powders and α -amylase and/or a positive ELISA result to either α -amylase, biolactase or Barse.

False discovery rate from multiple testing (p_c) was used for adjusting multiple comparison.

¹ Independent t test for continuous variables and χ^2 test for categorical variables were applied and p values were corrected using false discovery rates.

OR = 2.251, 95% CI 1.225–4.138, multiple logistic regression after adjusting for age, job type (nurse or pharmacist) and atopy; table 3]. On the contrary, no significant difference was found in comparison with total IgE levels (96.4 ± 115.6 vs. 116.5 ± 186.3 KU/l for ADRB2 46 AA vs. AG or GG genotype; 104.1 ± 165.1 vs. 214.1 ± 277.4

Table 3. Prevalences of allergies to digestive powder by genetic polymorphism

Genetic polymorphisms		Allergy to digestive powder		p	Adjusted OR (95 % CI)	p _c
		positive (%)	negative (%)			
ADRB2 46A>G	AA	14 (40)	17 (20.7)	0.023	1.694 (1.077–2.666)	0.035
	AG or GG	21 (60)	65 (79.3)			
IL10 –1082A>G	AA	26 (76.5)	74 (90.2)	0.009	2.251 (1.225–4.138)	0.027
	AG or GG	8 (23.5)	8 (9.8)			
IL4 –589T>C	TT	19 (57.6)	50 (58.8)	0.868	0.964 (0.628–1.48)	0.868
	CC or CT	14 (42.4)	35 (41.2)			

Figures in parentheses are 95% CI, unless indicated otherwise. The presence of an allergy to digestive powder was determined by a positive skin prick test, defined as a wheal size of ≥ 3 mm, to any one of the four digestive powders and α -amylase and/or a positive ELISA result to either α -amylase, budiastase or Barse. p values were determined by logistic regression and adjusted for age, job title (nurse/pharmacist) and atopy. False discovery rate from multiple testing (p_c) was used for adjusting multiple comparison.

KU/l for IL10 –1082 AA vs. AG or GG genotype) and atopy rates (29 vs. 33% for ADRB2 46 AA vs. AG or GG genotype; 35 vs. 13% KU/l for IL10 –1082 AA vs. AG or GG genotype) by genetic polymorphisms of ADRB2 46A>G or IL10 –1082A>G.

Discussion

The purpose of this study was to investigate the associations between 3 genetic polymorphisms, namely ADRB2 46A>G, IL10 –1082A>G and IL4 –589T>C, and specific IgE sensitization to digestive powders containing pancreatic extract in medical personnel. It was found that 26.8% of exposed medical personnel had work-related respiratory symptoms and 25.4% had an allergy to digestive powder. Furthermore, 24.8% of exposed subjects had a positive SPT result to any one of the four digestive powders, and digestive powder-specific IgE was demonstrated by 10.5% of the exposed subjects. Moreover, our genetic association analysis showed that the ADRB2 46A>G and IL10 –1082A>G polymorphisms were significantly associated with allergy to digestive powder in exposed medical personnel by multiple logistic regression analysis after controlling for age, atopy and job title.

Sensitization to specific allergens is a complicated process that is regulated by both environmental and genetic factors. This study indicates that digestive powder allergy (as defined by a single positive SPT and the presence of specific IgE antibodies to digestive powder), working duration and serum total IgE levels are strongly and positively associated with work-related respiratory symptoms

among exposed subjects. We could not evaluate the sensitivity and specificity of SPT and specific IgE measurement to diagnosis of an allergy to digestive powders because a provocative challenge test was not performed in this study. Using standardized extracts, the percentage agreement between in vitro allergen-specific IgE tests and SPTs ranges between 85 and 95%, depending on the allergens being evaluated [20]. However, skin tests appear to be more sensitive but less specific than in vitro allergen-specific IgE tests. In the present study, we found 23 subjects with positive SPT but normal IgE levels and only 1 subject with negative SPT and specific IgE to digestive powder. We cannot exclude the possibility that some patients could respond to other minor digestive components different from amylase, budiastase and Barse, although the proportions of ingredients vary only slightly in the 4 digestive powders used in the SPT.

Furthermore, our findings suggest that total serum IgE level and the ADRB2 46A>G and IL10 –1082A>G genetic polymorphisms are risk factors of specific sensitization to digestive powders. Previous studies suggest that sensitization to enzymes is associated with the intensity and duration of exposure [1, 2, 21]. Thus, we examined relations between the prevalence of allergy to digestive powder and work-related respiratory symptoms in pharmacists and nurses. Pharmacists usually grind and mix these powders in comparatively confined spaces, and thus, were considered to be exposed to higher levels of these powders. Nevertheless, no job title-associated effect was found. Furthermore, although IgE-mediated response is the major pathogenic mechanism of occupational allergies induced by high molecular weight drugs

[22], epidemiological studies on occupational allergies to enzymes among bakers and enzyme-detergent workers have demonstrated exposure-response relationships for IgE sensitization [1, 23, 24].

Some studies have reported that atopy is a risk factor of sensitization to enzymes [1, 25], whereas others have concluded that it is not a significant risk factor [21, 26]. In the present study, serum total IgE levels were found to be significantly associated with the presence of a powder allergy among exposed subjects, but no significant association was found between atopy and working duration in exposed medical personnel with an allergic reaction. However, the present study is limited by uneven numbers of nurses and pharmacists as well as the lack of direct measures of exposure to digestive powder.

Genetic factors have been suggested to be responsible for drug-induced immune responses after repeated contact with a drug [7]. Previous studies on genetic variants and the modulation of IgE responses encouraged us to focus on IL4 -589T>C, ADRB2 46A>G and IL10 -1082A>G. The role of the IL4 -589T>C polymorphism in asthma and its associated phenotypes is controversial. In asthmatic children and adults, the C to T exchange at -589 in the IL4 promoter has been associated with significant increases in total IgE levels and increased reporter gene expression in vitro [17, 27]. However, other studies failed to find any significant association between this SNP and serum total IgE levels [12, 14]. With regard to specific IgE response, Qiao et al. [28] found that serum IL4 levels were significantly elevated in patients positive for specific IgE to penicillin, but they were unable to confirm the association between the IL4 -589C>T polymorphism and penicillin allergy. In the present study, no significant association was found between this SNP and allergy to digestive powder or total serum IgE. This may be why no association has been found between specific IgE to drugs and the IL4 -589C>T polymorphism. It may be that this SNP is only a marker for polymorphisms in the 5q31 chromosome and that its genetic influence is modulated by gene-gene or gene-environmental interactions.

The association between the IL10 -1082A>G polymorphism and β -lactam allergy was confirmed in a previous study in two different populations [18, 29]. In Chinese patients with penicillin allergy, IL10 levels were markedly reduced in positive responders with specific IgE or IgG, and the -1082 GG genotype was found to be significantly associated with positive specific IgE [30]. Furthermore, this result is consistent with in vivo results that -1082 GG and GA show greater IL10 expres-

sion than the AA genotype [29]. We also found that the prevalence of allergy to digestive powder was significantly higher in exposed subjects with the GG or GA genotypes of the IL10 -1082A>G polymorphism than in those with the AA genotype. However, we were not able to measure IL10 levels and no exposed subject had the GG genotype.

The ADRB2 gene, which is located on chromosome 5q31, is another gene of interest. Two SNPs in the ADRB2 gene, namely, Arg16Gly and Gln27Glu, are known to modify receptor function in vitro and have been associated with asthma, asthma-related phenotypes and variable therapeutic response [31]. β_2 -Adrenergic receptor agonists have anti-inflammatory actions and modulate IgE production through β_2 -adrenergic receptors expressed on B and T lymphocytes in vitro and in vivo [16, 32]. In a previous study, increased adrenoceptor down-regulation was found to be related to the ADRB2 46A>G polymorphism [33]. However, previous studies failed to find significant associations between the Gln27Glu or Arg16Gly ADRB2 polymorphisms and total serum IgE levels in the Korean population [15, 34, 35]. In terms of specific IgE sensitization, Llanes et al. [36] demonstrated no significant association between genetic variations of ADRB2 46A>G polymorphisms and the risk of olive pollen sensitization and asthma. In the present study, we investigated whether the ADRB2 46A>G polymorphism plays a role in specific IgE response to allergens, it was found that the AA genotype of ADRB2 46A>G was significantly associated with IgE sensitization to digestive powder.

Variations in allele frequencies and environments in different populations may be responsible for these inconsistent findings regarding the allergy-related phenotypes of ADRB2 polymorphisms.

We analyzed the combined effects of the ADRB2 46A>G and IL4 -589C>T or IL10 -1082A>G polymorphisms on specific IgE sensitization to digestive powder, but found no evidence of any additive effect. Therefore, we suggest that further studies are required to determine how candidate genetic polymorphisms influence sensitization to specific allergens and whether these genetic factors interact with other genes or environmental factors.

Summarizing, the present study suggests that allergy to digestive powder, as determined by the SPT and the presence of specific IgE to digestive powder or α -amylase, may increase the risk of work-related respiratory symptoms among medical personnel repeatedly exposed to digestive powder. Furthermore, we suggest that the ADRB2 46A>G and IL10 -1082 A>G polymorphisms may be

viewed as potential markers of IgE sensitization to digestive powder, and that duration of exposure and serum total IgE levels are environmental factors that increase the risk of developing respiratory symptoms in subjects with an allergy to digestive powder. However, further studies are needed to investigate how these polymorphisms impact risk and to determine whether these are risk factors of progression to occupational asthma or rhinitis.

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References

- ▶ 1 Houba R, Heederik DJ, Doekes G, van Run PE: Exposure-sensitization relationship for alpha-amylase allergens in the baking industry. *Am J Respir Crit Care Med* 1996;154:130–136.
- ▶ 2 Sarlo K, Kirchner DB: Occupational asthma and allergy in the detergent industry: new developments. *Curr Opin Allergy Clin Immunol* 2002;2:97–101.
- ▶ 3 Park HS, Nahm DH: New occupational allergen in a pharmaceutical industry: serratal peptidase and lysozyme chloride. *Ann Allergy Asthma Immunol* 1997;78:225–229.
- ▶ 4 Brisman J, Belin L: Clinical and immunological responses to occupational exposure to alpha-amylase in the baking industry. *Br J Ind Med* 1991;48:604–608.
- 5 Kim SH, Ye YM, Kim HM, Lee MK, Suh CH, Nahm DH, Park HS: Occupational allergy caused by inhalation of pancreatic extracts in the medical personnel and identification of the major allergens. *Korean J Asthma Allergy Clin Immunol* 2007;27:10–19.
- ▶ 6 Nakamura S: Studies on asthma bronchiale. 6. On the occupational allergy caused by pancreatin among pharmacists. *Alerugi* 1971;20:361–364.
- ▶ 7 Vandebriel RJ: Gene polymorphisms within the immune system that may underlie drug allergy. *Naunyn Schmiedeberg Arch Pharmacol* 2004;369:125–132.
- ▶ 8 Mansur AH, Bishop DT, Markham AF, Britton J, Morrison JF: Association study of asthma and atopy traits and chromosome 5q cytokine cluster markers. *Clin Exp Allergy* 1998;28:141–150.
- ▶ 9 Kauppi P, Lindblad-Toh K, Sevon P, Toivonen HT, Rioux JD, Villapakkam A, Laitinen LA, Hudson TJ, Kere J, Laitinen T: A second-generation association study of the 5q31 cytokine gene cluster and the interleukin-4 receptor in asthma. *Genomics* 2001;77:35–42.
- ▶ 10 Geha RS, Jabara HH, Brodeur SR: The regulation of immunoglobulin E class-switch recombination. *Nat Rev Immunol* 2003;3:721–732.
- ▶ 11 Dewar JC, Wilkinson J, Wheatley A, Thomas NS, Doull I, Morton N, Lio P, Harvey JF, Liggett SB, Holgate ST, Hall IP: The glutamine 27 beta2-adrenoceptor polymorphism is associated with elevated IgE levels in asthmatic families. *J Allergy Clin Immunol* 1997;100:261–265.
- ▶ 12 Dizier MH, Sandford A, Walley A, Philippi A, Cookson W, Demenais F: Indication of linkage of serum IgE levels to the interleukin-4 gene and exclusion of the contribution of the (-590 C to T) interleukin-4 promoter polymorphism to IgE variation. *Genet Epidemiol* 1999;16:84–94.
- ▶ 13 Qiu YY, Zhang XL, Yin KS: Association between beta2-adrenergic receptor genetic polymorphisms and total serum IgE in asthmatic patients of Chinese Han nationality. *Respiration* 2006;73:180–184.
- ▶ 14 Noguchi E, Shibasaki M, Arinami T, Takeda K, Yokouchi Y, Kawashima T, Yanagi H, Matsui A, Hamaguchi H: Association of asthma and the interleukin-4 promoter gene in Japanese. *Clin Exp Allergy* 1998;28:449–453.
- ▶ 15 Kim SH, Oh SY, Oh HB, Kim YK, Cho SH, Kim YY, Min KU: Association of beta2-adrenoceptor polymorphisms with nocturnal cough among atopic subjects but not with atopy and nonspecific bronchial hyperresponsiveness. *J Allergy Clin Immunol* 2002;109:630–635.
- ▶ 16 McAlees JW, Sanders VM: Hematopoietic protein tyrosine phosphatase mediates beta2-adrenergic receptor-induced regulation of p38 mitogen-activated protein kinase in B lymphocytes. *Mol Cell Biol* 2009;29:675–686.
- ▶ 17 Rosenwasser LJ, Borish L: Genetics of atopy and asthma: the rationale behind promoter-based candidate gene studies (IL-4 and IL-10). *Am J Respir Crit Care Med* 1997;156:S152–155.
- ▶ 18 Guglielmi L, Fontaine C, Gougat C, Avinens O, Eliaou JF, Guglielmi P, Demoly P: IL-10 promoter and IL4-Ralpha gene SNPs are associated with immediate beta-lactam allergy in atopic women. *Allergy* 2006;61:921–927.
- ▶ 19 Pearce N, Weiland S, Keil U, Langridge P, Anderson HR, Strachan D, Bauman A, Young L, Gluyas P, Ruffin D, et al: Self-reported prevalence of asthma symptoms in children in Australia, England, Germany and New Zealand: an international comparison using the ISAAC protocol. *Eur Respir J* 1993;6:1455–1461.
- 20 Demoly P, Bousquet J, Romano A: In Vivo Methods for the Study of Allergy; in Adkinson NJ, Bochner BS, Busse WW, Holgate ST, Lemanske RFJ, Simons FE (eds): *Allergy, Principles and Practice*. New York, Mosby, 2009.
- ▶ 21 Cullinan P, Lowson D, Nieuwenhuijsen MJ, Sandiford C, Tee RD, Venables KM, McDonald JC, Newman Taylor AJ: Work related symptoms, sensitisation, and estimated exposure in workers not previously exposed to flour. *Occup Environ Med* 1994;51:579–583.
- ▶ 22 Petrick MM, Slavin RG: Occupational rhinitis. *Immunol Allergy Clin North Am* 2003;23:193–203, vi.
- ▶ 23 Houba R, Heederik D, Doekes G: Wheat sensitization and work-related symptoms in the baking industry are preventable. An epidemiologic study. *Am J Respir Crit Care Med* 1998;158:1499–1503.
- ▶ 24 Juniper CP, How MJ, Goodwin BF, Kinshott AK: *Bacillus subtilis* enzymes: a 7-year clinical, epidemiological and immunological study of an industrial allergen. *J Soc Occup Med* 1977;27:3–12.
- ▶ 25 Hur GY, Koh DH, Kim HA, Park HJ, Ye YM, Kim KS, Park HS: Prevalence of work-related symptoms and serum-specific antibodies to wheat flour in exposed workers in the bakery industry. *Respir Med* 2008;102:548–555.
- ▶ 26 Johnsen CR, Sorensen TB, Ingemann Larsen A, Bertelsen Secher A, Andreassen E, Kofoed GS, Fredslund Nielsen L, Gyntelberg F: Allergy risk in an enzyme producing plant: a retrospective follow up study. *Occup Environ Med* 1997;54:671–675.
- ▶ 27 Meyers DA, Beaty TH, Colyer CR, Marsh DG: Genetics of total serum IgE levels: a regressive model approach to segregation analysis. *Genet Epidemiol* 1991;8:351–359.

- ▶ 28 Qiao HL, Yang J, Zhang YW: Relationships between specific serum IgE, cytokines and polymorphisms in the IL-4, IL-4R α in patients with penicillins allergy. *Allergy* 2005;60:1053–1059.
- ▶ 29 Turner DM, Williams DM, Sankaran D, Lazarus M, Sinnott PJ, Hutchinson IV: An investigation of polymorphism in the interleukin-10 gene promoter. *Eur J Immunogenet* 1997;24:1–8.
- ▶ 30 Qiao HL, Wen Q, Gao N, Tian X, Jia LJ: Association of IL-10 level and IL-10 promoter SNPs with specific antibodies in penicillin-allergic patients. *Eur J Clin Pharmacol* 2007; 63:263–269.
- ▶ 31 Hawkins GA, Weiss ST, Bleecker ER: Clinical consequences of ADR β 2 polymorphisms. *Pharmacogenomics* 2008;9:349–358.
- ▶ 32 Fedyk ER, Adawi A, Looney RJ, Phipps RP: Regulation of IgE and cytokine production by cAMP: implications for extrinsic asthma. *Clin Immunol Immunopathol* 1996;81:101–113.
- ▶ 33 Green SA, Turki J, Hall IP, Liggett SB: Implications of genetic variability of human β 2-adrenergic receptor structure. *Pulm Pharmacol* 1995;8:1–10.
- ▶ 34 Cho SH, Oh SY, Bahn JW, Choi JY, Chang YS, Kim YK, Min KU, Kim YY: Association between bronchodilating response to short-acting β -agonist and non-synonymous single-nucleotide polymorphisms of β -adrenoceptor gene. *Clin Exp Allergy* 2005; 35:1162–1167.
- ▶ 35 Kim WJ, Oh YM, Sung J, Kim TH, Huh JW, Jung H, Lee JH, Kim EK, Lee SM, Lee S, Lim SY, Shin TR, Yoon HI, Kwon SY, Lee SD: Lung function response to 12-week treatment with combined inhalation of long-acting β 2 agonist and glucocorticoid according to ADR β 2 polymorphism in patients with chronic obstructive pulmonary disease. *Lung* 2008;186:381–386.
- ▶ 36 Llanes E, Quiralte J, Lopez E, Sastre B, Chacartegui M, del Pozo V, Palomino P, Lahoz C, Cardaba B: Analysis of polymorphisms in olive pollen allergy: IL13, IL4RA, IL5 and ADR β 2 genes. *Int Arch Allergy Immunol* 2009;148:228–238.