

Independent Inheritance of Serum Immunoglobulin E Concentrations and Airway Responsiveness

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Elevated serum Immunoglobulin E (IgE) levels and increased airway responsiveness (AR) are correlated traits that are characteristic of asthma. It is not known to what extent these traits arise from distinct or shared genetic determinants. We investigated the genetic and environmental components of variance of serum total and specific IgE levels and AR in an Australian population-based sample of 232 Caucasian nuclear families. The inter-relationships of the genetic determinants of these traits were also investigated. Log_e total serum IgE levels had a narrow-sense heritability (h^2_N) of 47.3% (SE = 10.0%). Specific serum IgE levels against house dust mite and timothy grass, measured as a RAST Index, had a h^2_N of 33.8% (SE = 7.3%). AR, quantified by the log_e dose-response slope to methacholine (DRS), had a h^2_N of 30.0% (SE = 12.3%). Extended modeling demonstrated an approximate 70% overlap in the genetic determinants of total and specific serum IgE levels. The genetic determinants of serum IgE levels and AR exhibited less than 30% sharing. These data are consistent with the existence of multiple genetic determinants of the pathophysiologic traits associated with asthma, and suggest that AR is genetically distinct from atopy. These results have implications for gene discovery programs.

Asthma is the most common chronic childhood disease in developed nations (1) and is associated with high economic and social costs (2). Most asthma is closely associated with airway inflammation and narrowing. The familial syndrome of atopy (allergic asthma, seasonal rhinitis, and/or eczema) is accompanied by increased levels of total serum Immunoglobulin E (IgE) and elevated levels of IgE specific to aeroallergens (3). In patients with asthma, specific serum IgE is most commonly detected against inhaled allergens from house dust mites (HDM) and grass pollens (4). Asthma is also typified by non-specific increased airway responsiveness (AR) to inhaled

agents such as histamine or methacholine, which can be quantified by measuring the reduction of expiratory airflow after increasing doses of these agents (5).

In most patients with asthma, elevated serum IgE levels are strongly associated with increased AR (6). However, although symptoms of asthma and increased AR correlate with increased total and specific serum IgE levels, many persons with elevated IgE levels are not asthmatic (6). Similarly, despite its close relationship with atopy, AR is a reasonably specific but not sensitive predictor of asthma (7). It is uncertain to what extent these associations reflect shared genetic or environmental determinants.

Considerable effort is currently being expended in the detection of genetic loci contributing to asthma susceptibility (8). Both binary disease status and asthma-associated quantitative phenotypic traits have been investigated. One measure of the familial aggregation of disease status is the recurrence risk ratio (λ_R) where R refers to a particular class of relatives of an affected individual and λ_R denotes the ratio of the prevalence of disease among these relatives to the prevalence in the general population (9). Generally, the recurrence risk for siblings (λ_s) is calculated; the λ_s of past or current physician-diagnosed asthma has of value < 2 (10). Power calculations based on this value suggest that testing for linkage is significantly enhanced by the use of quantitative traits in preference to a categorical asthma phenotype (10). The use of quantitative "intermediate" phenotypes such as serum IgE levels or AR also permits selection of subjects for the extremes of distribution, with increased power to detect linkage (8, 11). It is therefore important to examine the genetic basis of asthma-associated physiological traits.

The aim of this study was to use Variance Components Analysis to estimate the genetic and environmental components of variance of serum total and specific IgE levels and AR in a population-based sample of nuclear families. A further aim was to investigate the inter-relationships between the genetic determinants of these traits.

METHODS

Study Population

Busselton is a rural town on the coast of South-Western Australia. In 1990 all residents on the Busselton electoral roll (approximately 9,000) were sent a questionnaire regarding current respiratory symptoms and medical history. From approximately 6,500 replies, Caucasian families were ascertained for the present study. The study was undertaken during the winter months of May–July 1992 to avoid seasonal effects on the measured traits. Families with both parents alive and younger than 55 yr of age and having at least two children older than 5 yr of age were recruited serially. All participants older than 5 yr of age were studied, and complete data were obtained on 1,020 subjects, comprising 232 nuclear families. The field methods used have been described in detail elsewhere; the response rate was 72% (12).

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The study was approved by the Human Rights Committee of the University of Western Australia, and informed personal or parental consent was obtained for all subjects.

Questionnaire

Individual and family histories of respiratory symptoms, demographic information, and smoking were assessed at interview using the British Medical Research Council questionnaire (13). Questionnaires relating to children were administered to a parent, generally the mother. Pack-years of smoking were calculated from questionnaire responses. One pack-year was taken to be equivalent to the consumption of 20 cigarettes per day for 1 yr.

IgE Assays

Total serum IgE and specific serum IgE to whole house dust mites (HDM) (*Dermatophagoides pteronyssinus*) and Timothy grass pollen (*Phleum pratense*) were measured using the Immuncap FEIA (Pharmacia AB, Uppsala, Sweden). The levels of specific IgE were converted to RAST units according to Pharmacia recommendations. A "combined RAST index" was calculated for each subject as the sum of the RAST scores to HDM and Timothy grass (8).

Airway Responsiveness

Spirometry was performed in the sitting position using a dry wedge spirometer (Vitalograph Model S, Buckinghamshire, UK) calibrated daily with a 3-L syringe. The best FEV₁ was measured according to the guidelines of the American Thoracic Society (14).

Response to methacholine challenge was assessed by the Yan rapid method (15). The FEV₁ was measured after inhalation of saline and increasing (doubling) doses of methacholine chloride delivered from calibrated handheld nebulizers. At each dose step one forced expiratory maneuver was performed unless it was thought to be technically unsatisfactory. The challenge was continued until either the FEV₁ fell by at least 20% from the postsaline value or until the maximal cumulative dose (12 μ mol) of methacholine was delivered. AR was expressed as the two-point dose-response slope (DRS) of methacholine response against percentage fall in FEV₁ (16).

Statistical Analysis

The primary response variables modeled were: (1) total serum IgE, (2) the combined RAST index to HDM and Timothy grass, and (3) the DRS to methacholine challenge. Explanatory variables included sex, age, height, and pack-years of smoking. Total serum IgE, the combined RAST index, and the DRS were also included as explanatory covariates in certain models.

All explanatory variables except sex were analyzed as continuous covariates. Total serum IgE levels and the DRS to methacholine exhibited a skewed distribution with a long right-hand tail; log_e transformation rendered the marginal distributions of these variables approximately Normal. The combined RAST index also exhibited a skewed distribution with a long right-hand tail; there was no obvious transfor-

mation to render the marginal distribution approximately Normal. Therefore, this variable was analyzed in its raw form. A constant of 10 was added to each DRS measurement in order to allow log_e transformation when the DRS was ≤ 0 . All continuous covariates were centered at or close to their mean and were modeled as linear, quadratic, and cubic fixed effects.

The software package FISHER (17) was used to partition observed phenotypic variance into genetic and nongenetic components by maximum likelihood methods. Each model assumed that the distribution of the response phenotype was multivariate normal, with a mean that depended upon a particular set of explanatory covariates. The mean models and specification of variance and covariance structures are given in APPENDIX. Analysis of the combined RAST index relied upon tests of model fit to ensure that the skewed marginal distribution of this variable did not seriously distort assumptions of multivariate Normality. It should be noted that marginal non-Normality need not necessarily imply lack of multivariate Normality once all of the biologic components of a model are included.

Phenotypic variance was partitioned into four components: σ^2_A = additive genetic effects; σ^2_{CS} = common sibling environment; σ^2_C = common family environment; and σ^2_{EC} or σ^2_{EP} = the residual variances (which are assumed to arise from nonfamilial factors) in children and in parents, respectively.

The narrow-sense heritability (h^2_N) was defined as the ratio of variance due to additive genetic effects (σ^2_A) to the total phenotypic variance of each trait (18): $h^2_N = \sigma^2_A / (\sigma^2_A + \sigma^2_C + \sigma^2_{CS} + \sigma^2_{EC})$. Asymptotic standard errors for h^2_N were obtained by reparameterizing the covariance model in terms of h^2_N rather than σ^2_A .

The statistical associations of covariates entered as fixed effects and the current response variable were assessed by removal of terms from the mean model and calculation of a likelihood ratio χ^2 test statistic (18). The same approach was used as an approximate guide to the "significance" of a departure of the value of a variance component from its null value (zero). Statistical significance was taken as the 5% level.

Standard goodness-of-fit tests to check overall validity of models were performed using the FISHER program (17). These included a test of the acceptability of the assumption of multivariate Normality.

Extended Modeling

In order to investigate the extent to which additive genetic effects were shared between phenotypes, the original mean model for each phenotype of interest (e.g., total serum IgE) was extended by adding (one at a time) terms for other outcomes of primary interest (e.g., the RAST index and DRS) (see Tables 2B, 3B, and 4B). Under such circumstances a large reduction in the magnitude of σ^2_A suggested a sharing of additive genetic factors, and therefore a two- or three-way sharing of genetic determinants.

Because this was not a standard nested model problem, an approach based upon profile likelihoods (19) was used to assess the magnitude of any change in the estimate of the genetic component of variance (σ^2_A). $\sigma^2_{A[O]}$ was defined as the estimated value of σ^2_A in the

TABLE 1
POPULATION CHARACTERISTICS

	Mothers (n = 232)	Fathers (n = 232)	Female Offspring (n = 278)	Male Offspring (n = 278)
Age, yr*	38.9 (4.8)	41.5 (4.8)	12.7 (4.9)	12.6 (4.6)
Height, cm*	164.0 (6.2)	176.3 (7.6)	149.9 (17.0)	153 (20.8)
Total serum IgE [†] (IU/ml)	25.7 (20.7 to 32.0)	40.4 (32.8 to 46.6)	62.1 (50.5 to 76.5)	73.7 (60.3 to 88.8)
RAST index*	1.7 (2.4)	1.8 (2.2)	2.3 (3.1)	2.8 (2.9)
DRS to methacholine [†] (% fall in FEV ₁ per μ mol methacholine)	1.8 (1.4 to 2.3)	2.2 (1.7 to 2.7)	2.7 (2.2 to 3.4)	3.7 (2.9 to 4.7)
Pack-years of smoking*	3.1 (7.8)	9.1 (13.5)	0.2 (1.0)	0.2 (1.4)
Asthma (ever), % (n) [‡]	16.4 (38)	15.5 (36)	20.5 (57)	27.0 (75)

* Values are means with SD shown in parentheses.

[†] Geometric mean and 95% confidence intervals (± 1.96 SEM).

[‡] Positive answer to the question: "Has your doctor ever told you that you have asthma or bronchial asthma?"

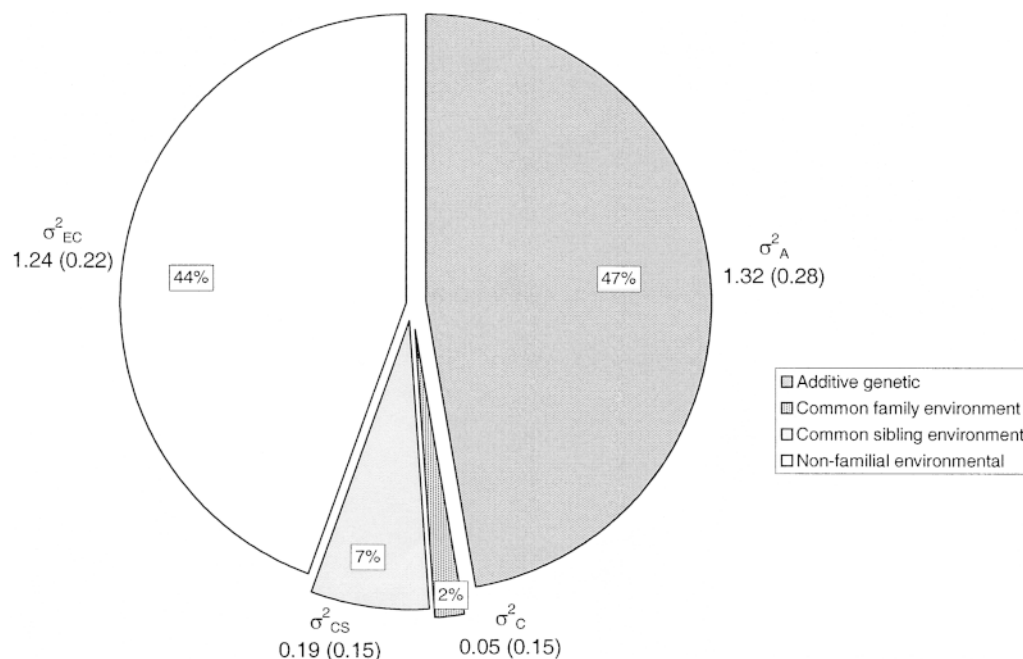


Figure 1. Components of total variance in total serum IgE levels. The estimated value, standard error, and percentage of total variance in total serum IgE concentration are shown for each component of variance.

original model and $\sigma^2_{A(E)}$ as its equivalent in the extended model. The extended model was then refitted and the value of σ^2_A constrained so it was forced to take the value $\sigma^2_{A(O)}$. The ratio of the likelihoods of the free and constrained models measured the plausibility of the hypothesis that the true value of σ^2_A was unchanged by extending the model. In a standard nested model problem, a likelihood ratio of 6.82 approximates to $p = 0.05$. More generally, a likelihood ratio in excess of 10 is moderate evidence that the hypothesis of shared determinants is plausible, and one in excess of 100 is strong evidence.

RESULTS

Characteristics of Families

The mean number of children per family was 2.35. Male and female participants were equally represented in the study population (Table 1). The mean age ($\pm$ SD) of parents was 40.2 ± 5.0 yr and of children 12.6 ± 4.7 yr.

Effects of Age, Sex, Pack-Years of Smoking, and Height

The variance components modeling suggested that the joint age and sex effect (see mean model specification in APPENDIX) was highly significant for total serum IgE ($\chi^2_7 = 68.9$, $p < 0.0001$) and for the combined RAST index ($\chi^2_7 = 44.7$, $p < 0.0001$). Levels were higher in men and in children (Table 1). Neither pack-years of smoking nor height were significantly related to total IgE levels or to the RAST index.

The joint age and sex effect was also significant for DRS levels ($\chi^2_7 = 39.1$, $p < 0.0001$). DRS levels were higher in men and in children (i.e., male children were the most responsive subjects) (Table 1). There was some evidence of an association between increased height and increased AR, although the

relationship was not formally significant ($\chi^2_3 = 7.2$, $p = 0.07$). Pack-years of smoking were not significantly related to the DRS.

Associations with Asthma and Inter-relationships

The variance components modeling suggested that total serum IgE levels ($\chi^2_1 = 69.1$, $p < 0.0001$), the combined RAST index ($\chi^2_1 = 88.4$, $p < 0.0001$), and the DRS to methacholine ($\chi^2_1 = 164.0$, $p < 0.0001$) were all closely associated with the presence of physician-diagnosed asthma (ever).

Total serum IgE levels and the combined RAST index were closely associated with each other ($\chi^2_3 = 661.9$, $p < 0.0001$). The DRS was closely associated with both total serum IgE ($\chi^2_3 = 121.4$, $p < 0.0001$) and the combined RAST index ($\chi^2_3 = 217.2$, $p < 0.0001$).

These associations were independent of age, sex, pack-years of smoking, and height, and the covariance model ensured appropriate adjustment for familial correlation.

The fraction (percent) of the total serum IgE explained by the RAST index was calculated with the RAST index expressed in IU/ml. The fraction was log-normally distributed, with a geometric mean of 0.66% (individual range = 0.0% to 83.1%).

Variance Components and Heritability Estimates

After adjustment for all covariates, the h^2_N of serum total IgE levels was 47.3% (SE = 10.0%), i.e., additive genetic effects (σ^2_A) contributed just under half of the total variance (Figure 1 and Table 2A). σ^2_A was significantly greater than zero ($\chi^2_1 = 20.5$, $p < 0.0001$). The remaining variance was largely the result of nonfamilial environmental effects (σ^2_{EC}). Familial environment effects were not significantly different from zero (Table 2A).

TABLE 2A
MAXIMUM LIKELIHOOD (BASELINE) MODEL* SHOWING VARIANCE COMPONENTS ESTIMATES FOR LOG_e SERUM TOTAL IgE LEVELS

Outcome Variable	σ^2_A (SE)	σ^2_C (SE)	σ^2_{CS} (SE)	σ^2_{EC} (SE)	σ^2_{EP} (SE)
Log _e total serum IgE level	1.319 (0.282)	0.047 (0.154)	0.186 (0.148)	1.236 (0.215)	0.736 (0.254)

* Model adjusted for age, sex, pack-years of smoking, and height.

TABLE 2B
MAXIMUM LIKELIHOOD MODELS SHOWING VARIANCE COMPONENTS ESTIMATES FOR
LOG_e SERUM TOTAL IgE LEVELS AFTER SEQUENTIAL ADJUSTMENTS FOR COVARIATES

Model No.	Variables included	σ^2_A (SE) (% of baseline)	σ^2_A profile Likelihood Ratio [†]	σ^2_C (SE) (% of baseline)	σ^2_{CS} (SE) (% of baseline)	σ^2_{EC} (SE) (% of baseline)	σ^2_{EP} (SE) (% of baseline)
1	Baseline + combined RAST index*	0.401 (0.097) 30.4%	$> 1.0 \times 10^{12}$	0.000	0.175 (0.108) 93.8%	0.811 (0.114) 65.6%	0.509 (0.160) 69.2%
2	Baseline + log _e DRS*	1.072 (0.160) 81.3%	3.01	0.000	0.129 (0.135) 69.4%	1.256 (0.176) 101.6%	0.728 (0.208) 98.9%
3	Baseline + combined RAST index* + log _e DRS*	0.555 (0.156) 42.1%	$> 1.0 \times 10^{12}$	0.000	0.172 (0.108) 92.5%	0.747 (0.132) 60.4%	0.494 (0.135) 67.1%

* Linear, quadratic, and cubic terms included in model.

[†] Profile likelihood ratio of free (baseline) model to constrained model.

The h^2_N of the RAST index was estimated to be 33.8% (SE = 7.3%), i.e., σ^2_A contributed approximately one third of the total variance (Figure 2 and Table 3A). The σ^2_A was significantly greater than zero ($\chi^2_1 = 17.5$, $p < 0.0001$). Environmental effects common to siblings (σ^2_{CS}) were also significantly greater than zero ($\chi^2_1 = 11.4$, $p = 0.001$) and contributed approximately 15% of the total phenotypic variance. Environmental effects common to families (σ^2_C) were not significantly different from zero and contributed only minimally, if at all, to total phenotypic variance. The majority of phenotypic variance was attributable to nonfamilial environmental effects (σ^2_{EC}) (Figure 2 and Table 3A).

The h^2_N of DRS levels was estimated to be 30.0% (SE = 12.3%), i.e., σ^2_A contributed approximately one third of total variance (Figure 3 and Table 4A). The σ^2_A was significantly greater than zero ($\chi^2_1 = 5.5$, $p = 0.019$). The remaining variance was largely the result of nonfamilial environmental effects. Familial environmental effects were not significantly different from zero and contributed only minimally, if at all, to total phenotypic variance (Figure 3 and Table 4A).

The extended modeling (Tables 2B, 3B, 4B) indicated that adjustment of the total serum IgE for the RAST index resulted in a large fall (to 0.40 [SE = 0.10], 30.4% of baseline model, see Tables 2A and 2B) in the σ^2_A estimate. This was consistent with an approximate 70% overlap in genetic (σ^2_A) determinants, and was associated with a likelihood ratio in ex-

cess of 1.0×10^{12} , providing strong evidence of an important reduction in σ^2_A . Adjustment of the total serum IgE model for DRS resulted in an estimated 18.7% fall (to 1.07 [SE = 0.16], 81.3% of baseline model, see Tables 2A and 2B) in the estimate of σ^2_A . The likelihood of the unconstrained model was only 3.01 times greater than that of the constrained model, consistent with little or no change in σ^2_A . Adjustment of the total serum IgE model for both the RAST index and DRS simultaneously resulted in a large fall (to 0.55 [SE = 0.16], 42.1% of baseline model, see Tables 2A and 2B) in the σ^2_A estimate. This was consistent with an approximate 58% overlap in genetic (σ^2_A) determinants, and was associated with a likelihood ratio in excess of 1.0×10^{12} , providing strong evidence of an important reduction in σ^2_A . However, this reduction was no greater than that observed when the total serum IgE model was adjusted for the RAST index alone (Table 2B).

Extension of the RAST index model to include terms for DRS resulted in a fall of 29.2% (to 2.11 [SE = 0.54], 70.8% of baseline model, see Tables 3A and 3B) in the estimate of σ^2_A . This was associated with a likelihood ratio of 4.9, consistent with little or not change in σ^2_A . Adjustment of the RAST index model for both total serum IgE and DRS resulted in a large fall (to 0.94 [SE = 0.34], 33.8% of baseline model, see Tables 3A and 3B) in the σ^2_A estimate. This was consistent with an approximate 66% overlap in genetic (σ^2_A) determinants, and was associated with a likelihood ratio of 9.7×10^{10} , providing strong evidence of

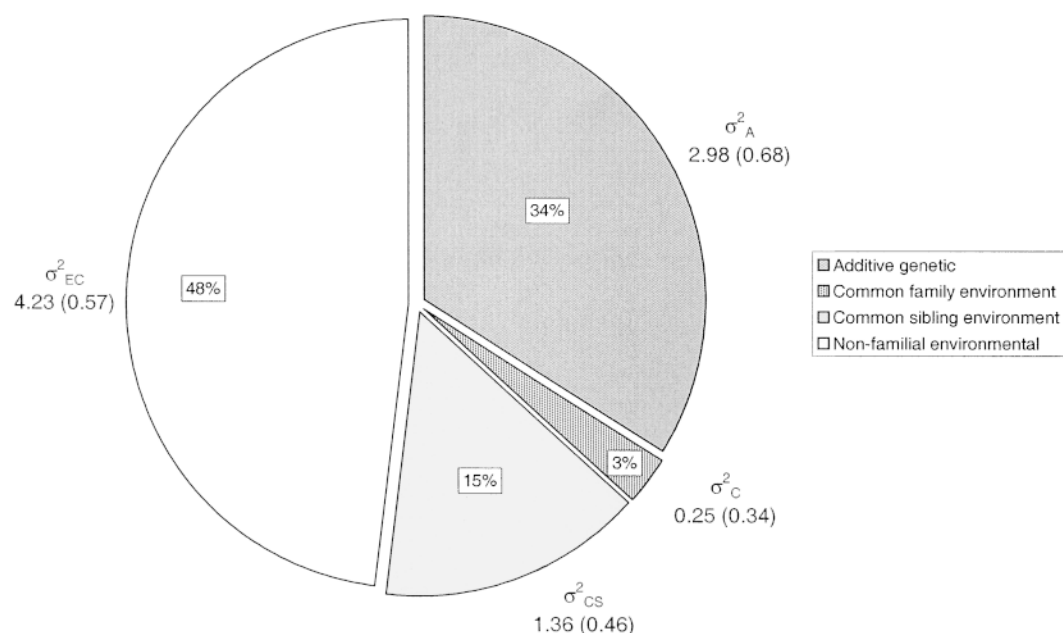


Figure 2. Components of total variance in combined RAST index levels. The estimated value, standard error, and percentage of total variance in RAST index are shown for each component of variance.

TABLE 3A
MAXIMUM LIKELIHOOD (BASELINE) MODEL* SHOWING VARIANCE COMPONENTS ESTIMATES FOR COMBINED RAST INDEX

Outcome Variable	σ^2_A (SE)	σ^2_C (SE)	σ^2_{CS} (SE)	σ^2_{EC} (SE)	σ^2_{EP} (SE)
Combined RAST Index	2.983 (0.677)	0.247 (0.341)	1.365 (0.462)	4.233 (0.570)	0.702 (0.314)

* Model adjusted for age, sex, pack-years of smoking, and height.

an important reduction in σ^2_A . However, this reduction was almost identical to that observed when the RAST index model was adjusted for total serum IgE alone (Table 3B).

Fitting extended models for the other possible combinations of outcomes and covariates (e.g., a model with the outcome DRS adjusted for total IgE levels) gave results consistent with the overlap in genetic determinants described above (Tables 2B, 3B, 4B).

The goodness-of-fit tests (17) did not indicate any significant lack-of-fit problems for any of the models reported. In particular, they always failed to reject the multivariate Normal assumption at the 5% significance level.

DISCUSSION

Our study was designed to recruit a sample of families that were representative of a general white population, and it has shown that the total serum IgE, specific IgE to common allergens, and AR are strongly heritable traits.

The prevalence rates for self-reported asthma for adults and children in the study were similar to those found in other Australian population studies (20, 21). The close associations of total and specific IgE and DRS levels with physician-diagnosed asthma were consistent with previous studies (3–5). As other investigators have reported (22), asthma was more common in boys than in girls. The relationship of serum IgE levels to age and sex was similar to that found in other population-based studies of white individuals (23), as were the relationships of DRS levels to age and sex (24). The smoking rates for parents and offspring were similar to the Australian general population (25, 26).

Theoretical modeling of the relative efficiency and power to estimate heritability of a quantitative trait using the FISHER software (27) indicated that our study had greater than 90% power at $\alpha = 0.05$ to reject the null hypothesis $h^2 = 0$ for a true heritability of 20%. Thus, our study had sufficient power to detect heritabilities greater than 20% for all outcomes investigated.

The importance of familial determinants in the regulation of serum IgE production in humans is well established. Studies of pedigrees and twins have suggested that total serum IgE levels have a heritability of around 50 to 80% (28, 29), consis-

tent with the present results. Our study further shows the correlations to be independent of the effects of age, sex, height, or total pack-years of cigarette consumption. Most of the observed familial correlations are attributable to genetic rather than to environmental factors. The relative contributions of familial and nonfamilial environmental factors to variance in total serum IgE levels are consistent with those reported in other studies (29, 30).

The heritability of specific IgE levels has not previously received much attention and it is of interest that our results show strong genetic effects underlying this trait. The significant contribution (15%) of common sibling environment to this trait is consistent with the effects of domestic allergens on specific IgE responses. Despite the marginal non-Normality of the RAST index, the models we present pass standard tests of multivariate Normality at the 5% significance level. Furthermore, inferences were unaffected if modeling was based on the t-distribution instead of the Normal distribution.

Few studies have directly estimated the heritability or mode of inheritance of AR in humans. One study, of 61 MZ and 46 DZ twin-pairs 6 to 31 yr of age (28), estimated the broad-sense heritability of the area under the log_e methacholine dose-response curve as approximately 66%. This is consistent with our findings of substantial familial correlations in AR that were independent of the effects of age, sex, height, or smoking. The majority of the observed familial correlations in DRS were due to genetic factors transmitted from parents to offspring, with a minor contribution from environmental factors common to families.

Our analysis allowed estimation of the overlap between genetic determinants of the three traits studied. Many studies have shown that total serum IgE levels and specific reactivity to allergens are highly correlated in industrialized populations (31). A similar close association of total serum IgE levels and the combined RAST index in our subjects was reflected in largely shared additive genetic factors (Tables 2B and 3B). There was also some evidence of shared familial (σ^2_C) and nonfamilial (σ^2_{EC}) environmental determinants. In the subjects described in this study, less than 1% of the total serum IgE was attributable to specific IgE against common allergens. This suggests that the evidence of shared determinants was not simply due to repeated modeling of what amounted to the

TABLE 3B
MAXIMUM LIKELIHOOD MODELS SHOWING VARIANCE COMPONENTS ESTIMATES FOR COMBINED RAST INDEX AFTER SEQUENTIAL ADJUSTMENT FOR COVARIATES

Model No.	Variables Included	σ^2_A (SE) (% of baseline)	σ^2_A profile Likelihood Ratio [†]	σ^2_C (SE) (% of baseline)	σ^2_{CS} (SE) (% of baseline)	σ^2_{EC} (SE) (% of baseline)	σ^2_{EP} (SE) (% of baseline)
1	Baseline + Log _e total serum IgE levels*	0.946 (0.238) 31.7%	9.7×10^{10}	0.000	0.641 (0.247) 47.0%	2.628 (0.284) 62.1%	0.931 (0.340) 181.8%
2	Baseline + Log _e DRS*	2.111 (0.536) 70.8%	4.90	0.144 (0.258) 58.3%	1.190 (0.391) 87.2%	3.789 (0.475) 89.5%	0.407 (0.598) 79.6%
3	Baseline + Log _e total serum IgE levels + Log _e DRS*	0.938 (0.338) 31.4%	9.7×10^{10}	0.000	0.749 (0.235) 54.9%	2.242 (0.283) 53.0%	0.670 (0.339) 95.4%

* Linear, quadratic, and cubic terms included in model.

[†] Profile likelihood ratio of free (baseline) model to constrained model.

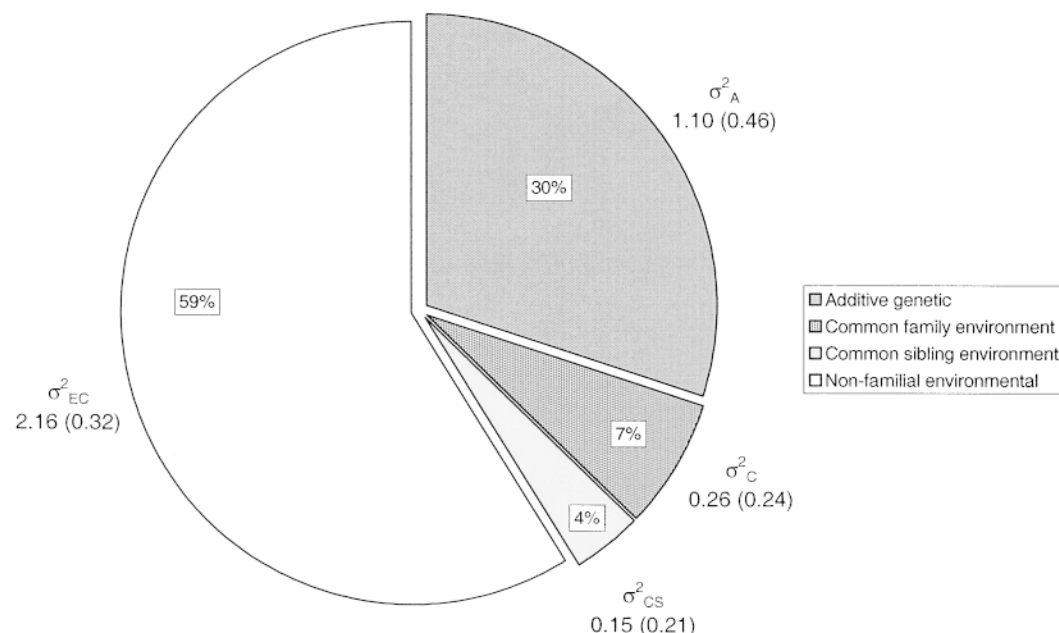


Figure 3. Components of total variance in airway responsiveness (measured as DRS to methacholine). The estimated value, standard error, and percentage of total variance in airway responsiveness are shown for each component of variance.

same trait (as may have been the case if, e.g., most of the total serum IgE was attributable to specific IgE against common allergens). There was also evidence of unshared genetic and environmental determinants influencing the two traits. The unshared genetic determinants suggest the existence of at least two distinct genetic pathways modulating atopic IgE responses independently of age- or sex-dependent expression or genetic susceptibility to tobacco smoke exposure: one genetic pathway modulating general potentiation of IgE immune response and the other modulating specific reactions to individual allergens.

In asthmatic and nonasthmatic subjects, total serum IgE levels and specific IgE levels to domestic allergens are strongly associated with AR (6, 32). Previous studies have suggested that around 30% of individual variance in AR can be attributed to total serum IgE levels (3, 6). In the present study, the observed close relationships of total and specific serum IgE levels to the DRS were consistent with the previous reports. However, we found only borderline evidence that the association of the DRS to methacholine with both total serum IgE levels and the RAST index might be partially due to the sharing of additive genetic determinants.

The extended modeling to investigate three-way sharing of genetic determinants (Model 3 in Tables 2B, 3B, 4B) showed that addition of two outcomes as covariates did not cause any further reduction in the σ^2_A component of phenotypic variance for any of the outcomes. This suggests that any pairwise overlap between the additive genetic determinants of total serum IgE levels and airways responsiveness to methacholine is likely due to sharing of genetic determinants common to all 3 traits, i.e., methacholine responsiveness is unlikely to share genetic determinants with total serum IgE levels that are not

also shared with the RAST index. The same argument also applies to pairwise overlap between the genetic determinants of specific serum IgE and AR. However, the profile likelihood ratios observed for these reductions in σ^2_A were sufficiently small to be consistent with the hypothesis that the genetic determinants of methacholine responsiveness are entirely distinct from those of total and specific serum IgE levels.

The current study thus suggests that a large proportion (though not all) of the genetic determinants of total serum IgE levels are shared with those of specific IgE levels to house dust mite and Timothy grass. In contrast, the genetic determinants of serum IgE immune responses were largely independent of those for AR. Some independence of the familial determinants of total and specific IgE responses have been previously suggested (3, 33), but the degree of overlap has not been previously quantified. Independence of the genetic determinants of IgE levels and DRS has not been previously demonstrated.

Genetic linkage studies are consistent with these observations. Linkage to the 5q31-33 chromosomal region has been shown with total serum IgE levels, but not specific serum IgE levels (34). Linkage of bronchial hyperresponsiveness to the same region may be due to separate gene(s) (35). A recent whole genome screen for quantitative trait loci underlying asthma (8) was based on a subset of the population described in this report. In that study, the $\log_e$ total serum IgE levels and $\log_e$ DRS showed significant linkage to different chromosomes.

The present study suggests the presence of multiple genetic determinants of the pathophysiologic traits associated with asthma, and shows that two of the important phenotypes associated with asthma (IgE levels and AR) are distinct traits with largely unshared additive genetic determinants. Programs of gene identification will be facilitated by the recognition that

TABLE 4A
MAXIMUM LIKELIHOOD (BASELINE) MODEL* SHOWING VARIANCE COMPONENTS
ESTIMATES FOR $\log_e$ DOSE-RESPONSE SLOPE (DRS) TO METHACHOLINE

Response Variable	σ^2_A (SE)	σ^2_C (SE)	σ^2_{CS} (SE)	σ^2_{EC} (SE)	σ^2_{EP} (SE)
$\log_e$ DRS level	1.100 (0.458)	0.262 (0.244)	0.150 (0.207)	2.156 (0.318)	2.121 (0.400)

* Model adjusted for age, sex, pack-years of smoking, and height.

TABLE 4B
MAXIMUM LIKELIHOOD MODELS SHOWING VARIANCE COMPONENTS ESTIMATES
FOR LOG_e DOSE-RESPONSE SLOPE (DRS) TO METHACHOLINE AFTER
SEQUENTIAL ADJUSTMENT FOR COVARIATES

Model No.	Variables Included	σ^2_A (SE) (% of baseline)	$\sigma^2_{A\text{profile}}$ Likelihood Ratio [†]	σ^2_C (SE) (% of baseline)	σ^2_{CS} (SE) (% of baseline)	σ^2_{EC} (SE) (% of baseline)	σ^2_{EP} (SE) (% of baseline)
1	Baseline + Log _e total serum IgE*	0.830 (0.409) 75.5%	1.24	0.199 (0.214) 75.9%	0.026 (0.188) 17.3%	2.160 (0.296) 100.2%	2.119 (0.354) 99.9%
2	Baseline + RAST index*	0.726 (0.374) 66.0%	1.64	0.211 (0.194) 80.5%	0.126 (0.179) 84.0%	1.941 (0.267) 90.0%	1.823 (0.330) 85.9%
3	Baseline + Log _e total serum IgE + RAST index*	0.705 (0.372) 64.1%	1.26	0.208 (0.191) 79.4%	0.116 (0.178) 77.3%	1.957 (0.266) 90.8%	1.814 (0.329) 85.5%

* Linear, quadratic and cubic terms included in model.

[†] Profile likelihood ratio of free (baseline) model to constrained model.

different genetic mechanisms are likely to regulate serum IgE levels and AR: these traits are not proxies for one another and should be examined separately and together in molecular and cellular studies.

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APPENDIX

1. The mean models were specified as follows:

$$\text{Mean} = \beta_1 + \delta \cdot (\beta_2 + \beta_3 \cdot \text{age} + \beta_4 \cdot \text{age}^2 + \beta_5 \cdot \text{age}^3) + (1 - \delta) \cdot (\beta_6 \cdot \text{age} + \beta_7 \cdot \text{age}^2 + \beta_8 \cdot \text{age}^3) + \beta_9 \cdot (\text{cigarette pack years}) + \beta_{10} \cdot (\text{cigarette pack years})^2 + \beta_{11} \cdot (\text{cigarette pack years})^3 + \beta_{12} \cdot \text{height} + \beta_{13} \cdot \text{height}^2 + \beta_{14} \cdot \text{height}^3$$

where β_n was the n^{th} fixed regression coefficient and δ is a binary indicator variable taking the value 1 in male and 0 in female subjects. This model permitted independent age-specific profiles for the phenotype of interest in male and female subjects.

2. The total phenotypic variance (conditional on the mean model) was based on a conventional covariance structure (18) and was specified as: $(\sigma^2) = \sigma_A^2 + \sigma_{CS}^2 + \sigma_C^2 + \gamma \cdot \sigma_{EC}^2 + (1 - \gamma) \cdot \sigma_{EP}^2$

The residual variance was allowed to differ between children and parents; the binary indicator variable γ took the value 1 in children and 0 in parents.

3. The conditional covariances within a family were specified as:

- (1) $1/2\sigma_A^2 + \sigma_{CS}^2 + \sigma_C^2$ between two siblings;
- (2) $1/2\sigma_A^2 + \sigma_C^2$ between a parent and a child; and
- (3) σ_C^2 between two parents.