

Effect of IL-13 receptor $\alpha 2$ levels on the biological activity of IL-13 variant R110Q

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Background: IL-13 is a key cytokine associated with the asthmatic phenotype. IL-13 signals via its cognate receptor, a complex of IL-13 receptor (IL-13R) α 1 chain with IL-4 receptor α ; however, a second protein, IL-13R α 2, also binds IL-13. Recently a polymorphic variant of IL-13 (R110Q) has been shown to be associated with atopy.

Objective: To investigate the binding properties of this IL-13 variant to its cognate receptors.

Methods: We used surface plasmon resonance to measure the binding kinetics of R110Q to its receptors. Primary human fibroblasts were grown from endobronchial biopsies obtained from volunteers. Receptor levels were measured by fluorescence-activated cell sorting.

Results: There was no significant difference in the binding of R110Q with soluble human IL-13R α 1 compared with IL-13 (32 ± 5 nmol/L and 36 ± 7 nmol/L, respectively; $P = .625$). However, a small but significant difference was observed in the binding of R110Q to soluble human IL-13R α 2 compared with IL-13 (840 ± 87 pmol/L and $1.1 \pm .05$ nmol/L, respectively; $P = .04$). We observed that primary human lung fibroblasts expressed different levels of IL-13R α 2. Eotaxin release from fibroblasts expressing low IL-13R α 2 levels was significantly higher in response to R110Q compared with IL-13. This was not evident in cells that had high baseline IL-13R α 2 levels.

Conclusion: These results suggest that relatively small changes in functional properties of a ligand combined with variation in receptor levels *in vivo* can result in significant differences in responsiveness.

Clinical implications: Expression of R110Q and low IL-13R α 2 levels can result in important biological differences that may have clinical relevance in an atopic environment. (J Allergy Clin Immunol 2007;120:91-7.)

Key words: Asthma, IL-13R α 2, IL-13, polymorphisms

Abbreviations used

IL-4R: IL-4 receptor
IL-13R: IL-13 receptor
sh: Soluble human
SPR: Surface plasmon resonance
STAT: Signal transducer and activator of transcription
UK: United Kingdom

IL-13 is a pleiotropic cytokine with roles in asthma and allergy.^{1,2} It is produced by activated T cells to promote B-cell proliferation and IgE synthesis.³⁻⁵ It also downregulates the production of TNF- α , increases expression of vascular cell adhesion molecule 1 on endothelial cells, and enhances the induction of MHC class II and CD23 antigens on monocytes.⁶⁻⁸ IL-13 is a key cytokine in asthma not only because of its proallergic role but also because of its wide-ranging effects on epithelial cells and fibroblasts linked to airway wall remodeling. Overexpression of IL-13 in the bronchial epithelium of transgenic mice causes lymphocytic and eosinophilic infiltration, goblet cell metaplasia, subepithelial fibrosis, and smooth muscle proliferation associated with marked bronchial hyperresponsiveness.⁹

IL-13 mediates its effects through interacting with its cognate receptor on hematopoietic and other cell types, although no functional receptors have been identified on human or mouse T cells.⁷ The human IL-13 receptor (IL-13R) is a heterodimer composed of the IL-4 receptor (IL-4R) α chain and an IL-13 binding protein, IL-13R α 1.¹⁰⁻¹⁴ In a cellular context, IL-13 binds to IL-13R α 1 with moderate affinity (disassociation constant [K_D] = 2-10 nmol/L) in the absence of IL-4R α . However, this affinity increases when IL-4R α is present.¹⁵ IL-13 does not bind IL-4R α in the absence of IL-13R α 1. A second IL-13 binding protein, IL-13R α 2, has also been identified.¹⁶ IL-13R α 2 shares a 37% homology with IL-13R α 1 and binds IL-13 with high affinity (50 pmol/L) but not IL-4.^{15,17} Despite this increased binding affinity, IL-13R α 2 is believed to be nonsignaling, and it has been suggested that it acts as a decoy receptor.

Several single nucleotide polymorphisms have been identified in the *IL13* locus on chromosome 5q31.^{18,19} Studies in several populations have shown strong association with an *IL13* promoter polymorphism (C-1112T) and asthma, bronchial hyperresponsiveness, total IgE, and food allergies.²⁰⁻²³ A second polymorphic variant of human IL-13, G-2004A, has been found with a minor

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allele frequency of approximately 20% in the Caucasian population. This is the only nonsynonymous coding SNP in IL-13 found to date and results in the positively charged arginine residue at 110 of the mature polypeptide being nonconservatively substituted with neutral glutamine (R110Q). The work of Graves et al¹⁸ and several more recent studies have shown strong associations between this IL-13 polymorphism and atopy and atopic diseases such as atopic dermatitis and rhinitis.²⁴⁻²⁶

In functional assays, Vladich et al²⁷ have shown that R110Q significantly increased signal transducer and activator of transcription (STAT)-6 phosphorylation and CD23 expression in primary human monocytes compared with wild-type IL-13. Interestingly, this group also demonstrated that a soluble form of IL-13R α 2 neutralized wild-type IL-13 more effectively than R110Q. These results support the work performed by Arima et al,²⁸ who examined the functional properties of R110Q and suggested that R110Q had a lower affinity for IL-13R α 2 than wild-type IL-13.

We have previously shown that the responsiveness of primary bronchial epithelial and fibroblasts to IL-13 and IL-4 is inversely correlated to the levels of IL-13R α 2.²⁹ Furthermore, we have observed that the levels of these receptors on the cell surface show considerable variation between subjects. Therefore, we hypothesized that natural variation in the levels of membrane bound IL-13R α 2 has the ability to modulate the functional potential of R110Q-mediated effects on primary bronchial fibroblasts. Because biological potency is a product of both receptor levels and binding affinity, we examined the interactions of R110Q with its 2 cognate receptor chains, IL-13R α 1 and IL-13R α 2. This study describes the first detailed analysis of the interaction of R110Q with its binding chains using surface plasmon resonance and the functional consequences of R110Q against a background of natural variation in IL-13R α 2 expression in primary bronchial fibroblasts.

METHODS

Reagents

CM5 sensor chip, HBS buffer (10 nmol/L HEPES with 0.15 mol/L NaCl, 3.4 mmol/L EDTA, and 0.005% surfactant P20), amine coupling kit, and regeneration agents were supplied by BIAcore (Uppsala, Sweden) unless otherwise stated. The extracellular region of IL-13R α 1 and IL-13R α 2 was fused to the Fc portion of hIgG₁ to generate a soluble form of these IL-13 receptors (soluble human [sh] IL-13R α 1.Fc, shIL-13 α 2.Fc). Eotaxin ELISA kit, recombinant shIL-13R α 1.Fc, shIL-13 α 2.Fc, and neutralizing IL-13R α 2 antibody were obtained from R&D Systems (Abingdon, United Kingdom [UK]). Recombinant IL-13 and R110Q were purchased from Peptrotech (London, UK).

Surface plasmon resonance measurements

For IL-13 receptor analysis, a CM5 sensor chip was activated with a 10- μ L injection of *N*-hydroxysuccinimide/*N*-ethyl-*N'*-(3-diethylamino)propyl]carbodiimide followed by an injection of 10 μ L protein A in 10 mmol/L sodium acetate (pH 4.0). The remaining activated esters were blocked by an injection of 35 μ L 1 mol/L ethanolamine. shIL-13R α 1 or shIL-13R α 2 were then injected and bound noncovalently to the immobilized protein A.

All receptor proteins were diluted from stock to the desired concentration in HBS buffer. To determine kinetic constants, sensograms were collected at 25°C, with a flow rate of 10 μ L/min and data collection rate of 1 Hz. For binding and kinetic analysis, 5 serial dilutions (20-100 nmol/L) of each protein were injected separately over the sensor chip surface. Sensograms were recorded and normalized to a baseline of 0 resonance units. Equivalent concentrations of each protein were injected over an untreated surface to serve as blank sensograms for subtraction of bulk refractive index background. The sensor chip surface was regenerated between runs with a 1-minute injection of 10 mmol/L HCl at 10 μ L/min. The resultant sensograms were then evaluated using the BIA evaluation 2.0 software (BIAcore, Stevenage, UK) to provide kinetic data. Analysis of the rate of binding versus concentration of bound IL-13/R110Q at various time points enabled the derivation of both the association and dissociation rate constants.

Primary human cell culture

Bronchial fibroblasts were grown from endobronchial biopsies obtained from volunteers over the age of 18 years as previously described.³⁰ Nonatopic control subjects without asthma ($n = 6$) and subjects with asthma ($n = 7$) were characterized according to symptoms, pulmonary function, and medication. All subjects were non-smokers and were free from respiratory tract infections for a minimum of 4 weeks before inclusion in the study. Written informed consent was obtained from all volunteers, and ethical approval was obtained from the Southampton and South West Hampshire Local Ethics Research Committee. Biopsies were placed in chilled Dulbecco modified Eagle medium (Invitrogen, Paisley, UK) with 10% FBS and finely cut using a scalpel. The pieces were then placed into 25-cm³ flasks and incubated for 5 to 7 days until fibroblasts migrated out from the tissue and proliferated. The tissue was then removed and fibroblasts harvested and passaged; all studies were performed on cells between the third and sixth passage. Fibroblasts were characterized for α smooth muscle actin expression with average positive cells <20%.

Enzyme immunoassay for eotaxin 1

For assay, fibroblasts were plated into 24-well trays (2×10^4 cells) and allowed to grow to confluence before being rendered quiescent for 24 hours in Ultraculture serum free medium (Lonza Biologicals, Slough, UK). After 24 hours, the stimuli were added in the indicated concentrations. Supernatants were harvested after 24 hours and stored for analysis of eotaxin concentration by ELISA following the manufacturer's protocol. Cell culture supernatants were assayed without any purification or concentration. Cytokine release was expressed as picograms per milliliter.

Flow-cytometry analysis

Fibroblast cell cultures were detached from culture plates by using trypsin. The cell suspension was then washed in PBS containing 2% FCS. Aliquots of this cell suspension were incubated for 1 hour at 4°C with primary Ab IL-13R α 2 (Dialone, Boldon, UK) before washing. The cells were resuspended in PBS/1% BSA containing fluorescein isothiocyanate-conjugated antimouse secondary antibody. After 30 minutes in dark, the cells were washed in cold PBS for analysis using FACScan flow cytometer (BD Biosciences, Oxford, UK). Fibroblasts expressing with less than 5% fluorescence compared with isotope control were designated as low and above 20% as high.

STAT6 phosphorylation

Primary human fibroblasts were grown to confluence before being serum-starved for 24 hours. Cells were treated with IL-13 or R110Q (10 ng/mL) for 30 minutes. Fibroblasts were also treated in the

TABLE I. Association (K_{on}) and dissociation (K_{off}) rate constants of IL-13 and R110Q†

	K_{on} ($M^{-1} s^{-1}$)	K_{off} (s^{-1})	K_D
IL-13Rα1			
IL-13	4.40×10^5	1.50×10^{-2}	32 ± 5 nmol/L†
R110Q	4.20×10^5	1.50×10^{-2}	36 ± 7 nmol/L†
IL-13Rα2			
IL-13	2.50×10^5	2.0×10^{-4}	840 ± 87 pmol/L*
R110Q	1.95×10^5	1.95×10^{-4}	$1.1 \pm .05$ pmol/L*

*Significant difference in K_D values ($P = .04$).

†No significant difference in K_D values ($P = .625$). Data are means \pm SDs; $n = 10$.

‡Association and dissociation rates for the binding of IL-13 and R110Q to shIL-13Rα1 and shIL-13Rα2. Constants were determined as described under "Methods."

presence or absence of an IL-13Rα2 neutralizing antibody (1 μ g/mL; R&D Systems) for 3 hours before stimulation with either IL-13 or R110Q. The cells were then solubilized in boiling SDS sample buffer before being subjected to SDS-PAGE and Western blotting with an antiphospho-STAT6 antibody (New England Biolabs, Hitchin, UK). Blots were stripped and reprobed with an anti-STAT6 antibody to confirm loading. Levels of immunoreactivity were quantified by densitometry using GeneTools (Syngene Bio-imaging Systems, Cambridge, UK).

Statistical analysis

Because the data were not normally distributed, results were analyzed by using nonparametric tests and summarized by using the median and interquartile range. Statistical comparisons were made using the Wilcoxon rank-sum test and SPSS for Windows software. A P value of $< .05$ was considered significant.

RESULTS

Kinetic analysis of R110Q

Previous studies have shown that a soluble form of IL-13Rα2 cannot neutralize the effects of R110Q as efficiently as wild-type IL-13.^{27,28} Because both receptor levels and ligand affinity are both key factors in the determining the functional outcome of a receptor mediated response, we first evaluated the binding affinity of R110Q for its cognate receptor chains.

The binding kinetics of IL-13 and R110Q to shIL-13Rα1 and shIL-13Rα2 were analyzed in real time by surface plasmon resonance (SPR) using a BIAcore 2000 biosensor (BIAcore). The calculated dissociation constants (K_D = dissociation rate constant [k_{off}]/association rate constant [k_{on}]) were found to be 32 ± 5 nmol/L and 36 ± 7 nmol/L, respectively ($P = .625$; Table I).

Similar experiments were also performed for shIL-13Rα2. Fig 1 shows an overlay of a series sensograms obtained after the interaction of various concentrations (20-100 nmol/L) of IL-13 and R110Q with shIL-13Rα2 as described in Methods. In this case, the association rate of R110Q was slower compared with IL-13 (2.50×10^5 and 1.95×10^5 mol/L/s, respectively; $P = .05$). However, the dissociation rate was similar. This gave a lower value in overall affinity of R110Q for IL-13Rα2 compared with IL-13 (Table I). This slower association

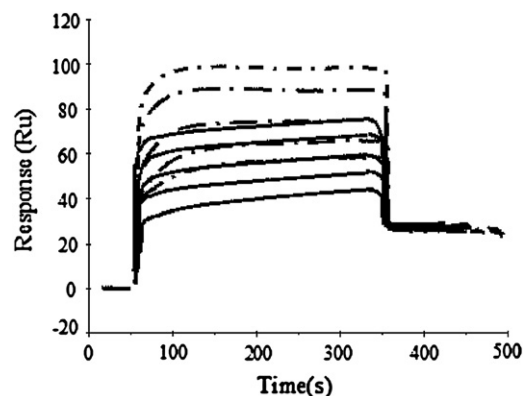


FIG 1. Kinetic analysis of R110Q by SPR. Representative sensograms of R110Q (solid line; lower to upper curve: 20, 40, 60, 80 and 100 nmol/L) and wild type IL-13 (dashed line) binding to the surface of CM5 sensor chip coated with shIL-13Rα2. A slower association rate was observed for the binding of R110Q to shIL-13Rα2 compared with wild-type IL-13. There was no significant difference in the rate of dissociation. Data are means \pm SDs ($n = 5$).

rate may explain why IL-13Rα2 has been found to be unable to regulate R110Q as effectively as wild-type IL-13.

Functional consequences of R110Q expression

In the current study, we found that baseline levels of IL-13Rα2 in bronchial fibroblasts varied considerably between subjects when measured by fluorescence-activated cell sorting. When we examined the release of eotaxin from these fibroblast cultures, we found that those with low IL-13Rα2 levels released significantly more eotaxin compared with fibroblasts with high IL-13Rα2 baseline levels (Fig 2).

The results from the SPR experiments demonstrated that R110Q had a lower affinity for shIL-13Rα2 than wild-type IL-13. Thus, we wanted to examine the effect of this binding affinity difference on the response of primary bronchial fibroblasts to R110Q and wild-type IL-13. Furthermore, we wished to investigate the possibility that differences in IL-13Rα2 levels in these fibroblasts may also lead to variation in response to R110Q compared with wild-type IL-13.

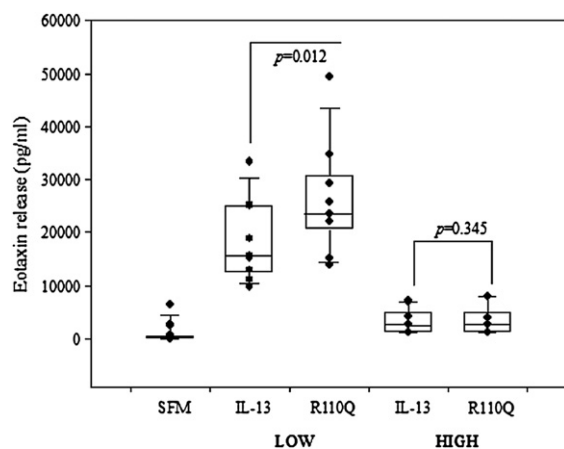


FIG 2. Functional consequences of R110Q expression. Primary human fibroblasts were grown to confluence before being treated with either IL-13 (10 ng/mL) or R110Q (10 ng/mL) for 24 hours. Supernatants were assayed for eotaxin by ELISA. IL-13Rα2 levels were determined by fluorescence-activated cell sorting. Fibroblasts with low levels of IL-13Rα2 showed a significant difference in eotaxin release between cells treated with IL-13 and R110Q. Symbols represent individual volunteers ($n = 13$). Statistical analysis was performed by using the Wilcoxon rank-sum test.

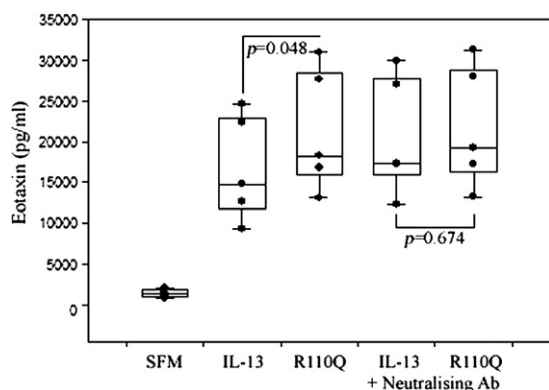


FIG 3. Addition of neutralizing IL-13Rα2 antibody on eotaxin release. Primary human fibroblasts were grown to confluence before being treated with either IL-13 (10 ng/mL) or R110Q (10 ng/mL) for 24 hours in the presence or absence of a neutralizing IL-13Rα2 antibody. Significant difference in eotaxin release between those cells treated with IL-13 and R110Q was abolished by the addition of the neutralizing antibody. Symbols represent individual volunteers ($n = 13$). Statistical analysis was performed by using the Wilcoxon rank-sum test.

Primary human fibroblasts were treated with R110Q or wild-type IL-13 for 24 hours. The supernatants were collected and analyzed for eotaxin release by ELISA. Initial analysis suggested that there was little difference in the amount of eotaxin released from cells treated with either R110Q or wild-type IL-13. However, when the baseline expression levels of IL-13Rα2 were taken into account, a significant difference was observed in the response to R110Q of those fibroblasts with low IL-13Rα2 levels compared with those with high levels compared with wild-type IL-13 (Fig 2). The addition of an

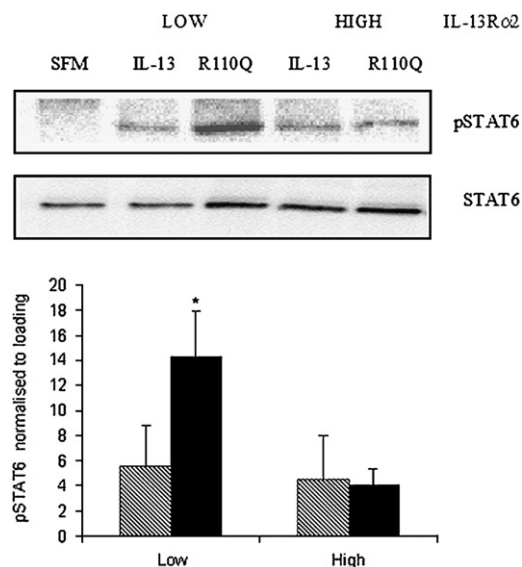


FIG 4. IL-13 and R110Q induced STAT6 phosphorylation. **A**, Primary human fibroblasts were treated with serum free medium (SFM), IL-13, or R110Q as indicated. Cells were then solubilized in sample buffer before Western blotting and probing with a phospho-STAT6 antibody. **B**, STAT6 phosphorylation was quantified by densitometry and normalized to STAT6 loading. Data are presented as means \pm SDs ($n = 3$). ▨ IL-13. ■ R110Q. * $P < .05$.

IL-13Rα2 neutralizing antibody completely abolished this observed difference in R110Q-mediated eotaxin release in fibroblasts with low IL-13Rα2 levels (Fig 3) compared with wild-type IL-13.³¹ The addition of this blocking antibody to cultures treated with TNF-α had no effect on TNF-α-mediated eotaxin release (data not shown).

Effect of IL-13Rα2 levels on STAT6 phosphorylation

To explore this difference in more detail, we examined phosphorylation of the transcription factor, STAT6, which lies immediately downstream of the IL-13 receptor signaling pathway. This showed that in fibroblasts with high levels of IL-13Rα2, there was no difference in the amount of STAT6 phosphorylation induced by equimolar concentrations of IL-13 or R110Q. However, in cells with low IL-13Rα2 levels, there was a significant increase in R110Q-induced STAT6 phosphorylation compared with wild-type IL-13 (Fig 4). We interpret these findings to suggest that when IL-13Rα2 levels are low, the difference in binding affinity of IL-13 and R110Q is the major contributor to the overall response, and thus the amount of phosphorylated STAT6 is greater for R110Q than IL-13. In contrast, when levels of IL-13Rα2 are high, mass action drives its binding to R110Q, and the effect of the difference in binding affinity is minimized with consequently similar levels of STAT6 phosphorylation.

To demonstrate that IL-13Rα2 levels are important for STAT6 regulation, a neutralizing antibody for IL-13Rα2 was added to the fibroblast cultures before stimulation with either IL-13 or R110Q. This resulted in a significant

increase in the amount of STAT6 phosphorylation in cells stimulated with either IL-13 or R110Q (Fig 5). The addition of the neutralizing antibody to fibroblasts with low baseline levels of IL-13R α 2 completely abolished the differences observed in STAT6 phosphorylation for IL-13 and R110Q. Thus, the levels of IL-13R α 2 appear to have a significant effect on the ability of the cells to regulate IL-13 and R110Q-mediated responses.

DISCUSSION

Several studies have established a link between genetic factors such as polymorphic variation in components of the IL-4 and IL-13 pathways and the development of allergic inflammation.³²⁻³⁴ However, it is an understanding of the functional consequences of these variants that will eventually reveal how these genetic susceptibilities translate into the development of asthma or other allergic diseases. Furthermore, when polymorphic variation occurs in more than 1 component of a pathway, as occurs in the case of IL-4/IL-13 signaling, the contribution of individual components has the potential to be modified (augmented or suppressed), leading to additional complexity in interpreting their functional effects. For example, Chen et al³⁵ have previously shown that coexpression of R110Q and an IL-4R α polymorphism (V50R551) increased CD23 expression.

Studies by Vladich et al²⁷ and Arima et al²⁸ have suggested that a variant of IL-13 (R110Q) may be an important genetic determinant in allergic disease. In the current study, we have extended the concept of interacting elements by showing that the potential of R110Q to contribute to an allergic response is dependent not only on its reduced affinity for IL-13R α 2 but also on naturally occurring levels of IL-13R α 2. These studies surpass previous reports in which a recombinant form of IL-13R α 2 (which lacks the transmembrane domain and cytoplasmic tail of the receptor) was assessed for its ability to act as a decoy receptor for R110Q compared with wild-type IL-13.

We used SPR to characterize the binding of R110Q to its receptor subunits and compared it with wild-type IL-13. There was no significant difference between the binding of R110Q and wild-type IL-13 to IL-13R α 1. Both IL-13 and R110Q had a slow dissociation rate from IL-13R α 2, which is characteristic of a negative regulator as previously reported. However, a small but distinct difference in association rate of R110Q to IL-13R α 2 was observed that resulted in an overall lower affinity of R110Q for IL-13R α 2. These data suggest that IL-13R α 2 is unable to bind R110Q as quickly as wild-type IL-13. These results may provide an explanation for the results obtained by Vladich et al,²⁷ who showed that a soluble IL-13R α 2.Fc chimera was unable to neutralize R110Q as effectively as wild-type IL-13.

We have observed that the expression of IL-13R α 2 in primary human fibroblast cells varied considerably between subjects. The variability in IL-13R α 2 expression

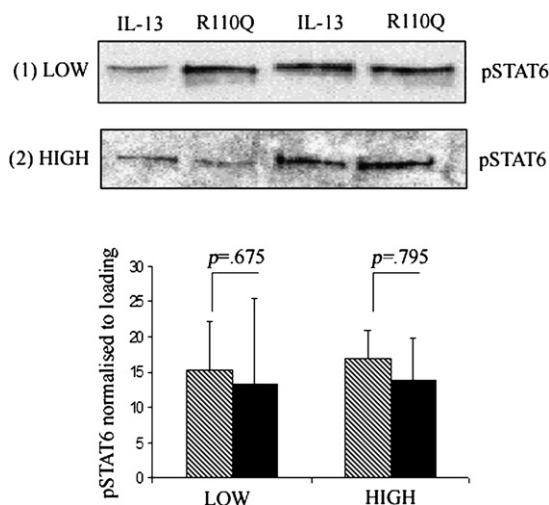


FIG 5. Surface levels of IL-13R α 2 are an important regulatory factor. **A**, Fibroblasts with either low (1) or high (2) surface levels of IL-13R α 2 were treated with IL-13 or R110Q as indicated in the presence or absence of a neutralizing IL-13R α 2 antibody. **B**, The presence of the neutralizing antibody completely abolished the difference in STAT6 phosphorylation observed in fibroblasts with low surface levels of IL-13R α 2. Data are representative of 3 individual experiments. pSTAT6, PhosphoSTAT6.

remains unclear because there are no known polymorphisms in the promoter region of the IL-13R α 2 gene. Receptor expression was not linked to the age or sex of patients, and there was only a slight association with atopy, so we speculate that some form of posttranscriptional regulation may be involved. The levels of IL-13R α 2 inversely correlated to their responsiveness to IL-13 despite responding normally to TNF- α .²⁹ Chen et al³⁵ had previously shown that coexpression of R110Q and an IL-4R α polymorphism (V50R551) increased CD23 expression.³⁵ Because receptor expression and polymorphisms are key factors in signal transduction, we were interested to see whether differences in naturally occurring receptor levels would affect R110Q-mediated effects.

We compared eotaxin release from fibroblasts treated with R110Q or wild-type IL-13. The difference in binding affinities is not substantial, and our initial observations suggested that R110Q was as equipotent as wild-type IL-13. This was consistent with our evidence that it is only the association rate that is slightly slower. In other words, R110Q may not bind as fast to IL-13R α 2 as wild-type, but once it has bound, it is not released. However, when the fibroblasts were analyzed for receptor expression, a significant difference was found between the amount of eotaxin released from fibroblasts with low levels of IL-13R α 2 when they were treated with R110Q compared with wild-type IL-13. This observed difference was abolished by the presence of an IL-13R α 2 neutralizing antibody. In fibroblasts that maintained high levels of IL-13R α 2 on their surface, no difference in the amount of eotaxin released compared with unstimulated controls was observed.

The difference in binding affinity of R110Q may decrease the ability of IL-13R α 2 to regulate its responses compared with wild-type IL-13. If IL-13R α 2 is unable to bind R110Q as quickly as wild-type IL-13, then R110Q will be free to bind to the IL-13 receptor complex consisting of IL-13R α 1 and IL-4R α , thus leading to a more sustained response than that observed for wild-type IL-13. In fibroblasts with high levels of receptor expression, an increase in the number of binding sites probably negates any difference in binding affinity.

Computer modeling of the structure of IL-13 may give us an insight into this decrease in R110Q affinity. Expression of R110Q leads to the substitution of arginine with glutamine in IL-13 molecule. This substitution occurs in the D helix of the IL-13 molecule that has previously been identified as important region for the binding of IL-13 to IL-13R α 1. Oshima et al³⁶ demonstrated that substitution of an arginine residue with the negatively charged amino acid, aspartic acid, produced agonist with a 5-fold increase in affinity. Arima et al²⁸ also demonstrated a lower affinity for IL-13R α 2, but not IL-13R α 1. Results from recent site-directed mutagenesis experiments performed by this group suggest that the substitution of arginine for glutamine at position 110 may change the conformation of the IL-13 molecule itself rather than disrupt an important binding interaction with IL-13R α 2.³⁷

Asthma is a complex disease in which there is often an underlying genetic predisposition. R110Q has been consistently associated with atopy and atopic diseases such as atopic dermatitis and rhinitis. We have demonstrated that IL-13R α 2 has a lower affinity for R110Q and thus is unable to regulate this cytokine as effectively as wild-type IL-13. This leads to a more sustained response than that observed for wild-type IL-13. If this is then linked with a natural variation in IL-13R α 2, the ability R110Q to contribute to an allergic response is dependent not only on its reduced affinity for IL-13R α 2 but also on naturally occurring levels of IL-13R α 2.

In this study, we have shown a relatively small difference in binding affinity of R110Q may explain its association with atopy and the asthmatic phenotype. However, if the expression of R110Q is combined with low IL-13R α 2 expression, this may result in important and significant biological differences that may have clinical relevance for the progression of allergic asthma.

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