

The proportion of FcεRI⁺ blood monocytes increases with the degree of IgE sensitization in asthma

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Atopic sensitization is an important and frequent driver of asthma development, and the interaction between allergen-specific IgE and its receptor FcεRI is central to the disease. Monocytes have been shown to accumulate in the airways of children with fatal asthma, possibly as a result of recruitment from the blood.[1] A fraction of peripheral blood monocytes expresses FcεRI as a trimeric complex of one alpha and two gamma chains (without the signal-amplifying beta chain). The trimeric FcεRI expression in monocytes is higher in atopic individuals and children with elevated IgE. [2-4] As IgE-crosslinking induces NFκB activation and production of TNF-α in monocytes *in vitro*, [5] FcεRI⁺ monocytes may play a pro-inflammatory role. However, FcεRI⁺ monocytes might also internalize and degrade IgE, thereby contributing to IgE clearance and immune homeostasis.[6] In our study, the proportion of FcεRI⁺ monocytes was evaluated as a potential asthma biomarker in a follow-up study of an Uppsala-based cohort of young adults with asthma, and control subjects without an asthma diagnosis.

The 39 asthma patients were 16-41 years old (mean 26±1), had total IgE levels between 14-1604 kU/L (geometric mean 245±1), and well-controlled asthma with an average asthma control test (ACT) score of 21±0.5. The 28 controls were 15-37 years old (mean 24±1), had total IgE levels between 0.05-315 kU/L (geometric mean 13±1), and no asthma symptoms (see supplemental methods and patient characteristics in Table S1). Isolated peripheral blood

mononuclear cells were analyzed for FcεRI expressing CD14⁺ blood monocytes by flow cytometry (Figure S1A).

A relationship between FcεRI expression in CD14⁺ monocytes (henceforth monocytes) and total serum IgE levels was previously shown in children, with the highest proportion of FcεRI expression in those with both atopy and asthma.[3] In our study, total IgE levels correlated positively with the proportion of FcεRI⁺ monocytes in adult asthma patients (Figure S1B). As expected, asthma patients had higher total IgE levels than the controls (Figure 1A). The asthma patients with >50% FcεRI⁺ monocytes tended to have higher levels of total IgE (Figure 1B), and higher levels of IgE antibodies against aeroallergens (ImmunoCAP Phadiatop; not shown; p=0.08). Most asthma patients were sensitized to aeroallergens (Phadiatop >0.35 kU/L), and had a higher proportion of FcεRI⁺ monocytes than the controls (Figure 1C). There was a trend towards a correlation between the degree of sensitization to aeroallergens and the proportion of FcεRI⁺ monocytes in asthma patients (Figure S1C). A fraction (40%) of the patients with asthma were also sensitized to food allergens (ImmunoCAP fx5 >0.35 kU/L). The fx5 test measures IgE levels to hen's egg white, cow's milk, peanut, wheat, soy bean, and cod allergens, but does not discriminate which allergen an individual is sensitized to. Food allergen sensitization (fx5) correlated positively with the proportion of FcεRI⁺ monocytes in asthma patients (Figure S1D). Further, individuals sensitized to food allergens had a higher proportion of FcεRI⁺ monocytes (Figure 1D). Phadiatop⁺/fx5⁺ individuals had higher total IgE levels than asthma patients that were Phadiatop⁺/fx5⁻, or Phadiatop⁻/fx5⁻ (Figure 1E). Furthermore, asthma patients who were Phadiatop⁺/fx5⁺ had a higher frequency of FcεRI⁺ monocytes than Phadiatop⁻/fx5⁻ (Figure 1F). We speculate that IgE levels are central for the induction of FcεRI expression on human

blood monocytes. A limitation is that the induction of FcεRI expression on monocytes by specific IgE was not assessed. Thus, the increase in FcεRI⁺ monocytes in asthma patients might be due to a greater atopic status.

Subsequently, we investigated whether having a high fraction of FcεRI⁺ monocytes was associated with other parameters. Having >50% FcεRI⁺ monocytes was not linked to a worse or better forced expiratory volume in one second, peak expiratory flow, asthma control test, fraction of exhaled nitric oxide, age, or body mass index (BMI) (Supplemental Table 2). However, as all subjects had well-controlled asthma, a possible link between the induction of FcεRI on monocytes and disease severity cannot be excluded. While the basophil count was similar in asthma patients and controls ($p=0.13$), asthma patients with >50% FcεRI⁺ monocytes had slightly more basophils (Figure 1G). There was a positive correlation between the proportion of FcεRI⁺ monocytes and the blood basophil count in asthma patients (Figure S1E). This relationship was expected since a strong association between FcεRI expression in basophils and total serum IgE levels has previously been demonstrated.[3] No relationships were found with other white blood cell counts (not shown).

Correlation analyses between the proportion of FcεRI⁺ monocytes and the concentration of 180 plasma proteins (using Olink Inflammation and Immune response panels) were performed among all subjects, and in asthma patients and controls separately. Only the aryl hydrocarbon nuclear transporter (ARNT) correlated positively with the proportion of FcεRI⁺ monocytes in asthma patients (Figure S1F, not adjusted for false discovery rate). There were no correlations between ARNT levels and age or BMI (not shown). ARNT levels were higher in asthma patients than in controls (Figure 1H) and tended to be higher in asthma patients

with >50% FcεRI⁺ monocytes (p=0.06). Asthma patients sensitized to both aeroallergens and food allergens had the highest ARNT levels (Figure 1I). The nuclear protein ARNT forms a heterodimer with the aryl hydrocarbon receptor (AhR) after the activation of AhR in epithelial cells by, for example, allergens, particulate matter or environmental toxins, and allow for AhR nuclear translocation. The formation of the AhR/ARNT heterodimer induces expression of pro- and anti-inflammatory cytokines.[7] Interestingly, tryptophan-derived metabolites generated by microbiota can activate AhR.[8] Patients with atopic dermatitis have reduced levels of an AhR-activating tryptophan-metabolite, which mediates tolerance via AhR in a mouse model of atopic dermatitis. [9] Further, AhR signaling in mice is associated with suppression of allergic sensitization to egg and peanut. [10] Whether AhR-mediated tolerance induction is related to the high levels of plasma ARNT in patients sensitized to both aero- and food allergens remains unknown.

In conclusion, a high proportion of FcεRI⁺ monocytes is associated with a high degree of IgE sensitization in young adults with asthma, suggesting that the proportion of FcεRI⁺ monocytes is a biomarker of sensitization in these patients. Moreover, ARNT is a possible biomarker of atopic asthma with a food allergy component.

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Conflict of interest

The authors declare no conflict of interest

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Figure legend

Figure 1. Multi-sensitized individuals have a high proportion of FcεRI⁺ CD14⁺ monocytes and ARNT levels. (A-D, G-H) Groupwise comparisons of subjects with asthma (A) and controls (C), based on their % FcεRI⁺ monocytes, or a positive or negative Phadiatop or fx5 test. (E-F, I) Multiple comparisons of subjects with positive or negative Phadiatop or fx5 test, and the specified parameters. Groupwise comparisons were tested by unpaired Student's t-test, whereas multiple comparisons were assessed by one-way ANOVA (Tukey's post-hoc test).

