EUROPEAN RESPIRATORY *journal*

FLAGSHIP SCIENTIFIC JOURNAL OF ERS

Early View

Original research article

Allergen immunotherapy effectively reduces the risk of exacerbations and lower respiratory tract infections in both seasonal and perennial allergic asthma: a nationwide epidemiological study

Christian Woehlk, Anna Von Bülow, Muzhda Ghanizada, Marianne Baastrup Søndergaard, Susanne Hansen, Celeste Porsbjerg

Please cite this article as: Woehlk C, Von Bülow A, Ghanizada M, *et al*. Allergen immunotherapy effectively reduces the risk of exacerbations and lower respiratory tract infections in both seasonal and perennial allergic asthma: a nationwide epidemiological study. *Eur Respir J* 2022; in press (https://doi.org/10.1183/13993003.00446-2022).

Allergen immunotherapy effectively reduces the risk of exacerbations and lower respiratory tract infections in both seasonal and perennial allergic asthma: a nationwide epidemiological study

Authors and affiliations:

Christian Woehlk, M.D[1], ORCID: 0000-0003-4605-1308 (corresponding author)

Anna Von Bülow, M.D., Ph.D[1].

Muzhda Ghanizada, M.D[1], ORCID: 0000-0001-5535-0380

Marianne Baastrup Søndergaard, M.D[1], ORCID: 0000-0002-4954-0863

Susanne Hansen, MSc. Public Health, Ph.D[1], ORCID: 0000-0002-9550-6703

Celeste Porsbjerg, Professor, M.D[1], ORCID: 0000-0003-4825-9436

[1]Respiratory Research Unit, Dept. Respiratory Medicine, Ebba Lunds Vej 48, entrance 66, 2400 Copenhagen NV, Denmark.

Corresponding author:

Dr. Woehlk, Respiratory Research Unit, Dept. Respiratory Medicine, Ebba Lunds Vej 48, entrance 66, 2400 Copenhagen NV, Denmark.

Email: cwoe0007@regionh.dk

Tel: +45 53644292 (direct)

Key words: Allergy, asthma, allergen immunotherapy, exacerbation, respiratory infection

Capsule summary: Allergen immunotherapy effectively reduced the risk of lower respiratory tract infections and exacerbations in both seasonal- and perennial allergic asthma and is potentially an early intervention strategy.

Acknowledgement: This study was supported by the Program for Clinical Research Infrastructure (PROCRIN) established by the Lundbeck Foundation and the Novo Nordisk Foundation.

Vancouver criteria: All authors listed contributed according to the Vancouver criteria.

Conflict of interests: An ICMJE Conflict of Interest form has been collected from all authors. Woehlk, C. has received speakers fee from ALK Abelló Nordic A/s. Baastrup, M. report speakers fee from GlaxoSmithKline. Von Bülow, A. has received speakers fee from AstraZeneca, GlaxoSmithKline, and Novartis and has attended advisory board for Novartis and AstraZeneca. Porsbjerg, C. reports; grants or contracts from AstraZeneca, GlaxoSmithKline, Novartis, TEVA, Sanofi, Chesi and ALK Abelló A/s to the institution; consulting fees from AstraZeneca, GlaxoSmithKline, Novartis, TEVA, Sanofi, Chesi and ALK Abelló A/s to the institution and personal honoraria; speakers fee from: AstraZeneca, GlaxoSmithKline, Novartis, TEVA, Sanofi, Chesi and ALK Abelló A/s to the institution and personal honoraria; advisory board honoraria from: AstraZeneca, Novartis, TEVA, Sanofi, and ALK Abelló A/s to the institution and personal honoraria. Ghanizada, M. and Hansen, S. reports no conflicts of interest.

Abbreviations:

AB	Antibiotics
AIT	Allergen Immunotherapy
DST	Statistics Denmark
RR	Rate Ratio
IL	Interleukin
ILC2	Innate lymphoid cell type 2
IRR	Incidence Rate Ratio
LRTI	Lower Respiratory Tract Infection
OCS	Oral Corticosteroids
OR	Online Repository
PAA	Perennial Allergic Asthma
SAA	Seasonal Allergic Asthma
SLIT	Sublingual Immunotherapy
SCIT	Subcutaneous Immunotherapy
TSLP	Thymic Stromal Lymphopoietin

Abstract

Background: Allergic asthma is associated with increased risk of respiratory tract infections and exacerbations. It remains unclear whether this susceptibility is conditioned by seasonal or by perennial allergy.

Aim: To investigate perennial allergy compared with seasonal allergy as a risk factor for lower respiratory tract infections and exacerbations in asthma and whether this risk can be reduced by allergen immunotherapy (AIT).

Methodology: This is a prospective register-based nationwide study of 18–44-year-olds treated with AIT during1995–2014. Based on the type of AIT and use of anti-asthmatic drugs, patients were subdivided into two groups: perennial allergic asthma (PAA) vs. seasonal allergic asthma (SAA). Data on antibiotics against lower respiratory tract infections (LRTI) and oral corticosteroids for exacerbations were analyzed before starting AIT (baseline) and three years after completing AIT (follow-up).

Results: We identified 2688 patients with asthma treated with AIT, among whom, 1249 had PAA and 1439 had SAA. At baseline, patients with SAA had more exacerbations, 23.8%, respectively, 16.5% p=<.001 but there were no differences in LRTI. During the three-year follow-up, we observed a highly significant reduction of exacerbations with an average decrease of 57% in PAA and 74% in SAA. We also observed a significant reduction of LRTI in both PAA and SAA: 17% and 20% decrease, respectively.

Conclusion: AIT effectively reduced the risk of exacerbations and lower respiratory tract infections in both seasonal- and perennial allergic asthma. Perennial allergy is seemingly not a stronger risk factor for respiratory infections and exacerbations than is seasonal allergy.

Introduction

Allergic asthma is associated with more frequent and more severe viral-induced illness[1]. *In vitro* studies have indicated that allergic asthma is linked with impaired immune responses to viral infection[2–6]. This may help understand why the combination of allergic sensitization, allergen exposure and viral infection greatly increases the risk of acute asthma exacerbations and the need for hospitalization[7–10]. We have recently shown that this increased risk also applies for bacterial respiratory infections requiring antibiotics and that, interestingly, allergen immunotherapy (AIT) completely abrogated this risk[11]. These findings suggest a modifiable link between allergy, asthma, and an impaired immune response against respiratory infections.

Mechanisms for impairment of respiratory immunity in allergic asthma are incompletely understood; however, uncontrolled airway inflammation is linked with impaired innate interferon release and seems to be both crucial for the understanding and clinically relevant[12]. Airway exposition to allergen protease activity triggers alarmin release and may lead to development or exacerbation of chronic T2-inflammation[13]. HDM allergen stands out over other aeroallergens in triggering alarmin release[14] and has been shown to directly affect toll-like receptor 3 (TLR3) signaling, thus inhibiting antiviral airway responses[15]. Recently, HDM exposure in HDM sensitized allergic asthma was associated with reduced secretion of anti-viral and anti-bacterial defence molecules[16]. Exposure to pollen has also been shown to suppress innate antiviral immunity independently of allergy and increase viral replication but, in contrast to HDM, is a seasonal burden[17].

AIT is a disease modifying treatment with multiple immunological effects that suppress T2-inflammation and induce immunologic tolerance[18]. The immunological effects of AIT have shown to improve clinical outcome measures, reduce asthma symptoms, reduce use of asthma medication, improve airway hyperresponsiveness, prevent worsening of asthma[19], and also to reduce risk of bacterial infection[11] and exacerbation[11,20]. Accordingly, AIT may be considered to restore respiratory immunity.

We have previously shown that the allergic asthma phenotype is associated with an increased risk of respiratory infections and exacerbations. However, little is known about whether a seasonal or perennial phenotype is associated with increased risk of respiratory deterioration; mechanistic studies indicate that HDM allergen may trigger an increased release of proinflammatory cytokines

and hence a more impaired innate immune defence. Therefore, we hypothesised that perennial allergic asthma is associated with an increased risk of lower respiratory tract infection and exacerbation compared with seasonal allergic asthma. We also hypothesised that AIT has a protective effect on these outcomes. Our primary aim was to assess the risk of infections and exacerbations in patients with seasonal vs. perennial allergic asthma before starting treatment with AIT; our secondary aim was to assess the change in these outcomes after AIT treatment. We undertook a nationwide epidemiologic cohort study of patients with asthma stratified according to seasonal vs. perennial allergy who completed AIT and compared use of antibiotics and oral corticosteroids before and after AIT using data from nationwide population registers in Denmark.

Methodology

Study design

We conducted a prospective register-based nationwide cohort study of patients treated with AIT during 1995–2014 in Denmark. The flowchart and study design are shown in Figures 1 and 2. Patients in the cohort were followed one year before commencing AIT (baseline period), during AIT, and three consecutive years post-AIT (follow-up year 1, follow-up year 2, follow-up year 3).

Data sources

The data sources included the Danish National Patient Register (NPR), the Danish National Prescription Registry (DNPR), the Civil Registration System (CRS), and Statistics Denmark's Education Register. In the NPR, information on all hospital inpatient admissions since 1977 is included through the ICD-8 and ICD-10 codes before and after 1994, respectively, and from 1995 and onwards outpatient and emergency room visits have also been included[21]. The DNPR holds information on all prescription drugs redeemed since 1995[22]. The recorded information is on the individual level and is done using Anatomic Therapeutic Chemical (ATC) codes. Statistics Denmark's Education registry holds information about individuals' highest attained educational level[23]. The Civil Registration System holds vital and demographic records of all people living in Denmark[24]. All Danish residents are assigned a unique identifier that follows the person from birth (or immigration) until death (or emigration) and that identifier can be used to link individuals across the data sources.

Study population

Our study population included all individuals treated with AIT during 1995–2014 living in Denmark. Patients were included if they had redeemed at least five consecutive prescriptions of the same AIT within 4 years of the first prescription during 1995–2014 using ATC codes V01AAxx. Their index date was defined as the date of the last prescription + 244 days. Data were extracted in 2017, and we therefore ended the study period in 2014 to allow for at least three years of follow-up for all those included. This definition was chosen to ensure a full vaccination schedule of a minimum of three years including up-titration.

Patients were excluded from the study if they were <18 years of age or >44 years of age at the index date, if they used specific medicine against chronic obstructive pulmonary disease or cystic fibrosis, if they had died or emigrated, or had missing information in registers according relevance to this study, as illustrated in Figure 1.

Identification of patients treated with AIT

The main exposure of interest in our study was a combination of type of allergy (seasonal vs. perennial) and asthma (allergic vs. non-allergic). Patients receiving AIT were classified as having *Perennial Allergic Asthma* (PAA) or *Seasonal Allergic Asthma* (SAA) based on ≥2 prescription fillings of anti-asthmatic drugs with an ATC code starting with R03 (medicine for obstructive airway disease) within 12 months of the index date[25]. Patients not filling prescriptions for anti-asthmatic drugs were classified as *Perennial Allergy* (PA) or *Seasonal Allergy* (SA).

Lastly, patients were categorized as "perennial allergy" if they had filled AIT for *Dermatophagoides Pteronyssinus* (ATC code V01AA03) and *Dermatophagoides Farinae* (AIT code V01AA03). Patients were categorized as "seasonal allergy" if they filled AIT for *Betula verrucosa* (ATC code V01AA05) and *Phleum pratense* (ATC code V01AA02). If patients filled prescriptions for AIT against both a perennial and a seasonal allergen, they were classified as perennial allergy. In Denmark subcutaneous injection therapy (SCIT) is the predominant choice of AIT. Sublingual immunotherapy (SLIT) was introduced in 2006 (Grazax) but received conditional subsidy in 2011 and in 2015 Acarizax was available. Standard of care for SCIT is Alutard® ALK administered by specialists according to national and international guidelines[26]. Standard injection regimens with up-titration are followed by intervals of 6 to 8 weeks but may be up to 10 weeks without dose adjustment. Hence, a prescription filling of AIT lasts approximately 10 months,

assuming an 8-week dosing interval. This definition was chosen to ensure a full vaccination schedule of a minimum of three years including up-titration.

Definition of outcomes

We followed our study cohort for the occurrence of three outcomes identified from the National Prescription Registry or from the National Patient Register.

Lower respiratory tract infection (LRTI):

LRTIs were defined as ≥ 1 prescription filling for antibiotics (AB) targeting LRTI (*appendix 1*) from the National Prescription Registry. This definition was based on national recommendations[27] and international guidelines[28] for the selection of AB to treat LRTI. In Denmark the first line of treatment for LRTI is β-lactam AB (e.g. phenoxymethylpenicillin, ampicillin, amoxicillin + clavulanic acid, etc.) accounting for approximately 50% and macrolides for 36% issued by general practitioners[27].

Exacerbations:

Exacerbations were defined as ≥ 1 prescription filling for oral corticosteroids (OCS) (ATC code: H02AB06) and/or emergency department visit (ED) or hospitalization with an associated ICD-10 code for asthma/or status asthmaticus (*appendix 2)*

Exacerbation related to infection:

Exacerbations related to infections were defined as ≥ 1 prescription filling for AB and OCS within a four-week period. (*appendix 3).*

Statistical analyses

We assessed patient characteristics at time of index (date of the last prescription + 244 days) and compared these between patients with seasonal allergy and perennial allergy using Chi-square tests. Study Aim 1 (LRTIs, exacerbations) was assessed during the baseline period (before treatment) and compared across the exposure groups using Chi-square tests. We also evaluated the within group change in LRTIs and exacerbations from baseline and during the three years of follow-up (post-treatment) by using McNemar's test for repeated nominal data (Figures 4–6). Pre-AIT data shown

in Figures 4–6 were calculated as a mean value as it varies slightly using McNemar's test for three different periods. This analysis was done separately for patients with seasonal allergy and for those with perennial allergy.

For Study Aim 2, we estimated rate ratios (RRs) and 95% confidence intervals from Poisson regression models. We aggregated person-time from time of index and during the three years of follow-up together with counts of LRTIs or exacerbations. In these analyses, we compared the rate of events between patients with seasonal allergic asthma and those with perennial allergic asthma, using seasonal allergic asthma as the reference group. All regression models were adjusted for age, sex, municipality, and education. Statistical significance was set at $p<0.05$, all tests were two-sided, and analyses were performed using SAS software version 9.4 or R.

Results are presented in the main text for patients with allergic asthma; results in patients without asthma are presented in the Online Repository.

Ethical considerations.

Register studies do not require ethics approval in Denmark. Data are handled in accordance with GDPR and the Danish Data Protection Act. All cases are anonymized and handled in accordance with discretionary requirements of personal statistics by the data privacy policy of Statistics Denmark.

Results

We identified 11,198 patients consecutively filling prescriptions for AIT from 1995 to 2014. Baseline demographics in patients receiving AIT are presented in Table 1 according to type of allergy. Patients with PA were less likely to be male (51% vs. 54%), were slightly younger [31 (8) vs. 32 years (7)], more likely to have asthma (39% vs. 18%), and more likely to reside in a urban municipality (62% vs. 55%) compared with patients with seasonal allergy. There was no difference observed regarding attained education.

Of the patients treated with AIT, 2688 (24%) had asthma, of whom 1249 (46%) had PAA and 1439 (54%) had SAA. Prior to commencing AIT, more patients with SAA had ≥1 exacerbation compared with those with PAA (24% vs. 16%, $p<.001$), and there was a tendency toward more patients having exacerbations related to infections in PAA patients compared with those with SAA (2% vs.

1%, p=.054). There was no difference in the frequency of LRTIs at baseline in the two groups (38% vs. 37%, p=0.69) (Figure 3).

In the three-year follow-up post-AIT, we observed a significant decrease in patients with exacerbations and LRTIs (Figures 4 and 5) and (Figures E6 and E7). In patients with PAA, there was a decrease in exacerbations from baseline (n=176) to follow-up year one (n=84), p=<.001; and follow-up year two (n=66), p=<.001; and follow-up year three (n=77), p=<.001, corresponding to an average decrease of 57%. Similarly, LRTIs were reduced from baseline (n=259) to follow-up year one (n=219), p=0.19; and follow-up year two (n=218), p=0.01; and follow-up year three (n=207), p=0.03, with an average decrease of 17%. However, there was no change in the proportion of patients with exacerbations related to infections from baseline to follow-up (Figure 6).

In patients with SAA, there was a decrease in the number of exacerbations from baseline (n=291) to follow-up year one (n=74), p=<.001; and follow-up year two (n=77), p=<.001; and follow-up year three (n=76), p=<.001, with an average decrease of 74%. For LRTIs, a reduction was also seen from baseline (n=316) to follow-up year one (n=246), p=0.01; and follow-up year two (n=256), p=0.02; and follow-up year three (n=255), p=.002, yielding an average decrease of 20%. We also observed a decrease in the number of patients with exacerbations related to infections during the three years of follow-up (Figure 6).

Lastly, we compared the relative risk of exacerbations and LRTIs in patients with PAA compared with patients with SAA and found no overall significant differences between the two groups (Online Repository Figures E1, E2 and E3). However, during the first year of follow-up after completing AIT, patients with PAA had an increased relative risk of having exacerbations of 1.22 (1.01–1.48) compared with patients with SAA, but this difference was not present after the second and third year of follow-up. The same pattern of an increased risk during the first year of follow-up was also seen in patients with PAA regarding LRTI, but the difference had disappeared after two years of follow-up. There was no difference in risk between the two groups regarding exacerbations due to infections.

Discussion

In this epidemiologic study, we associate AIT with a strong protective effect for measures of exacerbations and lower respiratory tract infections in patients with perennial- and seasonal allergic asthma. However, our data did not substantiate the hypothesis of perennial allergy being a stronger risk factor for impaired immune defence. Our findings strongly suggest that sensitization to aeroallergens should be considered a modifiable risk factor for exacerbations and respiratory infections in patients with allergic asthma.

These novel data are consistent with recent robust epidemiologic findings emphasizing the effectiveness of AIT in improving asthma control; reducing prescriptions for asthma; and preventing exacerbations, pneumonias and hospitalizations [29]. Moreover, this study highlights the effect of AIT on exacerbations among patients with both perennial and seasonal allergic asthma, rather than among patients with only HDM-related asthma, as recently shown by Virchow et al.[20].

Our use of nationwide registers ensures data validity and minimizes the risk of selection and recall bias; however, we acknowledge that there are limitations. We recognize the lack of a control group and the potential limitations introduced. We cannot rule out the potential bias of regression towards the mean and a healthy survivor effect, however, in the REACT-study, the effect of AIT on exacerbation was sustained throughout nine years of follow-up for exacerbation and at least five years of follow-up for respiratory infections compared with a control group[29]. Moreover, we cannot rule out that at least some of the effect seen, can be attributed being monitored closely by a Specialist. However, similar findings were made in the recently published REACT-study, where a propensity score-matched control group was included, with a reduction on in exacerbations and respiratory infections, although this study did not examine effects in perineal versus seasonal allergy. Clinical data were not available on the patients and one of the main limitations is the use of surrogate markers. The diagnosis of patients with allergic disease is confirmed by the high adherence to AIT. The asthma diagnosis was based on prescription fillings of medication against obstructive respiratory disease using a widely accepted definition[25]. Due to the study design, we believe there is a very low risk of confounding by indication. A strict upper age limitation was set to avoid interference with COPD. We acknowledge the lack of clinical confirmation of bacterial LRTI and that prescriptions may have been issued for other conditions such as skin infections, tonsilitis, sinusitis or mimicking conditions or conditions mimicking bacterial respiratory infections. This

could potentially overestimate the use of AB; however, we expect these conditions to be evenly distributed in the groups and evenly distributed throughout the observation period and not to be influenced by AIT. However, practitioners in Denmark have been deemed among the most restrictive in Europe regarding prescribing AB for respiratory tract infections due to the extensive use of point of care biomarkers (e.g. C-reactive protein)[30]. We also acknowledge the lack of data on asthma severity defined by daily dose delivery (DDD) of ICS this could potentially confound the effect of AIT. This may explain the difference in pre-AIT exacerbations as PAA requires an increased dose of ICS to maintain asthma control[31]. Conversely, SAA may be well-controlled at a lower DDD of ICS for most of the year but risk seasonal worsening of asthma when pollen exposure exceeds airway thresholds. This has been shown to be effectively preventable with anti-IgE[32]. Patients with polysensitization may have received AIT for just one allergen, thought to be particularly driving asthma, however if the patient had an exacerbation or respiratory infection driven by exposure to another allergen, this would minimize the effect described. Overall, we consider our findings as conservative estimates with low impact from potential biases.

Sensitization and exposure to aeroallergens is a well-established risk factor for exacerbation of asthma[33] and the risk of hospital admission increases dramatically with the contemporary presence of viral infection[9]. Although the mechanistic understanding remains unclear, uncontrolled T2-inflammation and impaired innate interferon release are considered potential main causes[34,35]. Exposure to aeroallergens perpetuates and exacerbates T2-mediated inflammation through increased secretion of epithelial-derived proinflammatory cytokines such as TSLP[36]. Thus exposure increases release of T2 cytokines that exhibit potent negative counter regulation of innate interferon release[6,34]. Hence, the possible rationale that 1) innate interferons may not counter regulate T2-mediated inflammation, thus contributing to retention of inflammation; 2) impaired interferon release may not adequately combat viral infection, leading to prolonged infection and increased inflammation[37]; 3) increased T2 inflammation impairs antibacterial host defence[38] and ultimately increases risk of secondary bacterial infestation of the lower airways[39,40].

The clinical effects of AIT occur through induction of tolerance and modification of a cascade of innate and adaptive immune responses: induction of circulating IgG_4 blocking antibodies prevents binding of allergen-specific IgE to the surface high- and low affinity receptors of the effector- and antigen presenting cells; T2 cell-mediated immunity is suppressed and induction of regulatory T

cells balance immune deviation toward Th1 cell response; homing, activation and degranulation of eosinophils, basophils and mast cells are decreased[41]. Furthermore, a recent study reported that AIT restored IL-10 production in competent ILC2s, hence attenuating Th responses and maintaining epithelial cell integrity[42]. Thus, in epidemiological, clinical and mechanistic studies, AIT has shown to improved quality and causally relevant outcomes in asthma control[43], exacerbation[20] and respiratory infections[11]. Accordingly, AIT could potentially be considered at earlier stages of allergic asthma among patients with recurrent respiratory infections.

Conclusion

AIT effectively reduced the risk of exacerbations and lower respiratory tract infections in both seasonal- and perennial allergic asthma. Perennial allergy does not appear to be a stronger risk factor for respiratory infections and exacerbations compared with seasonal allergy. AIT is a potentially promising early add-on therapy in patients with exacerbation and respiratory infection tendencies. Further research is needed to validate these findings and to establish the underlying immunologic mechanisms.

References

1. Olenec JP, Kim WK, Lee WM, et al. Weekly monitoring of children with asthma for infections and illness during common cold seasons. *J Allergy Clin Immunol*. Published online 2010. doi:10.1016/j.jaci.2010.01.059
2. Bufe A, Gehlhar K, Grage-Griebenow E, Ernst M. Atopic phenotype in children is associated with decreased virus-induced interferon-alpha release. *Int Arch Allergy Immunol*. 2002;127(1):82-88. doi:10.1159/000048173
3. Wark PA, Johnston SL, Bucchieri F, et al. Asthmatic bronchial epithelial cells have a deficient innate immune response to infection with rhinovirus. *J Exp Med*. 2005;201(6):937-947. doi:10.1084/jem.20041901
4. Contoli M, Message SD, Laza-Stanca V, et al. Role of deficient type III interferon-lambda production in asthma exacerbations. *Nat Med*. 2006;12(9):1023-1026. doi:10.1038/nm1462
5. Wark PAB, Grissell T, Davies B, See H, Gibson PG. Diversity in the bronchial epithelial cell response to infection with different rhinovirus strains. *Respirology*. 2009;14(2):180-186. doi:10.1111/j.1440-1843.2009.01480.x
6. Edwards MR, Regamey N, Vareille M, et al. Impaired innate interferon induction in severe therapy resistant atopic asthmatic children. *Mucosal Immunol*. 2013;6(4):797-806. doi:10.1038/mi.2012.118
7. Rakes GP, Arruda E, Ingram JM, et al. Rhinovirus and respiratory syncytial virus in wheezing children requiring emergency care: IgE and eosinophil analyses. *Am J Respir Crit Care Med*. 1999;159(3):785-790. doi:10.1164/ajrccm.159.3.9801052
8. Murray CS. Study of modifiable risk factors for asthma exacerbations: virus infection and allergen exposure increase the risk of asthma hospital admissions in children. *Thorax*. 2006;61(5):376-382. doi:10.1136/thx.2005.042523
9. Soto-Quiros M, Avila L, Platts-Mills TAEE, et al. High titers of IgE antibody to dust mite allergen and risk for wheezing among asthmatic children infected with rhinovirus. *J Allergy Clin Immunol*. 2012;129(6):1499-1505.e5. doi:10.1016/j.jaci.2012.03.040
10. Green RM, Custovic A, Sanderson G, Hunter J, Johnston SL, Woodcock A. Synergism between allergens and viruses and risk of hospital admission with asthma: case-control study. *BMJ*. 2002;324(7340):763. doi:10.1136/bmj.324.7340.763
11. Woehlk C, von Bülow A, Kriegbaum M, Backer V, Porsbjerg C. Allergic asthma is

associated with increased risk of infections requiring antibiotics. *Ann Allergy Asthma Immunol*. 2018;120(2):169-176.e1. doi:10.1016/j.anai.2017.11.015

12. Edwards MR, Strong K, Cameron A, Walton RP, Jackson DJ, Johnston SL. Viral infections in allergy and immunology: How allergic inflammation influences viral infections and illness. *J Allergy Clin Immunol*. 2017;140(4):909-920. doi:10.1016/j.jaci.2017.07.025
13. Kouzaki H, O'Grady SM, Lawrence CB, Kita H. Proteases Induce Production of Thymic Stromal Lymphopoietin by Airway Epithelial Cells through Protease-Activated Receptor-2. *J Immunol*. 2009;183(2):1427-1434. doi:10.4049/jimmunol.0900904
14. Ramu S, Menzel M, Bjermer L, Andersson C, Akbarshahi H, Uller L. Allergens produce serine proteases-dependent distinct release of metabolite DAMPs in human bronchial epithelial cells. *Clin Exp Allergy*. 2018;48(2):156-166. doi:10.1111/cea.13071
15. Akbarshahi H, Menzel M, Ramu S, Mahmutovic Persson I, Bjermer L, Uller L. House dust mite impairs antiviral response in asthma exacerbation models through its effects on TLR3. *Allergy Eur J Allergy Clin Immunol*. 2018;(December 2017):1-11. doi:10.1111/all.13378
16. Cerps S, Sverrild A, Ramu S, et al. House dust mite sensitization and exposure affects bronchial epithelial anti- microbial response to viral stimuli in patients with asthma. *Allergy*. 2022;(November 2021):1-11. doi:10.1111/all.15243
17. Gilles S, Blume C, Wimmer M, et al. Pollen exposure weakens innate defense against respiratory viruses. *Allergy Eur J Allergy Clin Immunol*. Published online 2020. doi:10.1111/all.14047
18. Akdis M, Akdis CA. Mechanisms of allergen-specific immunotherapy: Multiple suppressor factors at work in immune tolerance to allergens. *J Allergy Clin Immunol*. 2014;133(3):621-631. doi:10.1016/j.jaci.2013.12.1088
19. Abramson MJ, Puy RM, Weiner JM. Injection allergen immunotherapy for asthma. *Cochrane Database Syst Rev*. 2010;(8):CD001186. doi:10.1002/14651858.cd001186.pub2
20. Virchow JC, Backer V, Kuna P, et al. Efficacy of a house dust mite sublingual allergen immunotherapy tablet in adults with allergic asthma: A randomized clinical trial. *JAMA - J Am Med Assoc*. 2016;315(16):1715-1725. doi:10.1001/jama.2016.3964
21. Lynge E, Sandegaard JL, Rebolj M. The Danish national patient register. *Scand J Public Health*. Published online 2011. doi:10.1177/1403494811401482
22. Wallach Kildemoes H, Toft Sorensen H, Hallas J. The Danish National Prescription

Registry. *Scand J Public Health*. 2011;39(7 Suppl):38-41. doi:10.1177/1403494810394717

23. Jensen VM, Rasmussen AW. Danish education registers. *Scand J Public Health*. 2011;39(7):91-94. doi:10.1177/1403494810394715
24. Pedersen CB, Gøtzsche H, Møller JØ, Mortensen PB. The Danish Civil Registration System. A cohort of eight million persons. *Dan Med Bull*. Published online 2006.
25. Pont LG, Van Der Werf GT, Denig P, Haaijer-Ruskamp F. Identifying general practice patients diagnosed with asthma and their exacerbation episodes from prescribing data. *Eur J Clin Pharmacol*. 2002;57(11):819-825. doi:10.1007/s00228-001-0395-4
26. Roberts G, Pfaar O, Akdis CA, et al. EAACI Guidelines on Allergen Immunotherapy: Allergic rhinoconjunctivitis. *Allergy Eur J Allergy Clin Immunol*. 2018;73(4):765-798. doi:10.1111/all.13317
27. Bjerrum L, Boada A, Cots JM, et al. Respiratory tract infections in general practice: Considerable differences in prescribing habits between general practitioners in Denmark and Spain. *Eur J Clin Pharmacol*. 2004;60(1):23-28. doi:10.1007/s00228-003-0706-z
28. Woodhead M, Blasi F, Ewig S, et al. Guidelines for the management of adult lower respiratory tract infections - Full version. *Clin Microbiol Infect*. 2011;17(SUPPL. 6):1-24. doi:10.1111/j.1469-0691.2011.03672.x
29. Fritzsching B, Contoli M, Porsbjerg C, et al. Long-term real-world effectiveness of allergy immunotherapy in patients with allergic rhinitis and asthma: Results from the REACT study, a retrospective cohort study. *Lancet Reg Heal - Eur*. 2022;13:100275. doi:10.1016/j.lanepe.2021.100275
30. Cooke J, Butler C, Hopstaken R, et al. Narrative review of primary care pointof- care testing (POCT) and antibacterial use in respiratory tract infection (RTI). *BMJ Open Respir Res*. Published online 2015. doi:10.1136/bmjresp-2015-000086
31. De Vries MP, Van Den Bemt L, Thoonen BPA, Muris JWM, Van Schayck CP. Relationship between house dust mite (HDM) allergen exposure level and inhaled corticosteroid dosage in HDM-sensitive asthma patients on a self management program. *Prim Care Respir J*. 2006;15(2):110-115. doi:10.1016/j.pcrj.2006.01.001
32. Teach SJ, Gill MA, Togias A, et al. Preseasonal treatment with either omalizumab or an inhaled corticosteroid boost to prevent fall asthma exacerbations. *J Allergy Clin Immunol*. 2015;136(6):1476-1485. doi:10.1016/j.jaci.2015.09.008

33. Erbas B, Jazayeri M, Lambert KA, et al. Outdoor pollen is a trigger of child and adolescent asthma emergency department presentations: A systematic review and meta-analysis. *Allergy Eur J Allergy Clin Immunol*. 2018;73(8):1632-1641. doi:10.1111/all.13407

34. Contoli M, Ito K, Padovani A, et al. Th2 cytokines impair innate immune responses to rhinovirus in respiratory epithelial cells. *Allergy*. 2015;70(8):910-920. doi:10.1111/all.12627

35. Gern JE. Virus/allergen interaction in asthma exacerbation. *Ann Am Thorac Soc*. 2015;12(2):S137-S143. doi:10.1513/AnnalsATS.201503-153AW

36. Uller L, Leino M, Bedke N, et al. Double-stranded RNA induces disproportionate expression of thymic stromal lymphopoietin versus interferon-β in bronchial epithelial cells from donors with asthma. *Thorax*. 2010;65(7):626-632. doi:10.1136/thx.2009.125930

37. Gern JE, Busse WW. The Effects of Rhinovirus Infections on Allergic Airway Responses.

38. Beisswenger C, Kandler K, Hess C, et al. Allergic Airway Inflammation Inhibits Pulmonary Antibacterial Host Defense. *J Immunol*. 2006;177(3):1833-1837. doi:10.4049/jimmunol.177.3.1833

39. Antoni T, Blasi F, Dartois N, Akova M. Which individuals are at increased risk of pneumococcal disease and why? Impact of COPD, asthma, smoking, diabetes, and/or chronic heart disease on community-acquired pneumonia and invasive pneumococcal disease. *Thorax*. 2015;70(10):984-989. doi:10.1136/thoraxjnl-2015-206780

40. Hanada S, Pirzadeh M, Carver KY, Deng JC. Respiratory viral infection-induced microbiome alterations and secondary bacterial pneumonia. *Front Immunol*. 2018;9(NOV):1-15. doi:10.3389/fimmu.2018.02640

41. Pipet A, Botturi K, Pinot D, Vervloet D, Magnan A. Allergen-specific immunotherapy in allergic rhinitis and asthma. Mechanisms and proof of efficacy. *Respir Med*. 2009;103(6):800-812. doi:10.1016/j.rmed.2009.01.008

42. Golebski K, Layhadi JA, Sahiner U, et al. Induction of IL-10-producing type 2 innate lymphoid cells by allergen immunotherapy is associated with clinical response. *Immunity*. 2021;54(2):291-307.e7. doi:10.1016/j.immuni.2020.12.013

43. Mosbech H, Deckelmann R, De Blay F, et al. Standardized quality (SQ) house dust mite sublingual immunotherapy tablet (ALK) reduces inhaled corticosteroid use while maintaining asthma control: A randomized, double-blind, placebo-controlled trial. *J Allergy Clin Immunol*. 2014;134(3). doi:10.1016/j.jaci.2014.03.019

Table 1

Baseline demographics					
Variable		Seasonal (n=7957)	Perennial (n=3241)	Total (n=11198)	p-value
Sex, n (%)	Female	3651 (45.9)	1597 (49.3)	5248 (46.9)	
	Male	4306 (54.1)	1644 (50.7)	5950 (53.1)	.001
Age (years)	Mean (±SD)	32.3 (7.4)	31.0 (7.5)	31.9 (7.5)	<.001
Asthma, n (%)	No	6518 (81.9)	1992 (61.5)	8510 (76.0)	
	Yes	1439 (18.1)	1249 (38.5)	2688 (24.0)	<.001
Education, n (%)	Basic School	1357 (17.1)	595 (18.4)	1952 (17.4)	
	High School & vocational	3436 (43.2)	1368 (42.2)	4804 (42.9)	
	Higher (short/medium)	2132 (26.8)	848 (26.2)	2980 (26.6)	
	Higher (long/research)	1032 (13.0)	430 (13.3)	1462 (13.1)	.354
Municipality, n (%)	Urban	4407 (55.4)	1997 (61.6)	6404 (57.2)	
	Rural	2821 (35.5)	978 (30.2)	3799 (33.9)	
	Suburban	729 (9.2)	266 (8.2)	995 (8.9)	<.001

Figure legends

Figure 1: Flowchart: Overall, 20,391 patients filled ≥5 prescriptions of AIT with the same ATC code within 4 years. The study population consisted of 11,198 patients and were divided into "Patients with asthma" and "Patients without asthma" based on ≥2 prescription fillings of ATC R03: Drugs for pulmonary obstructive lung diseases.

Figure 2: Baseline defined as the year preceding first prescription of AIT. Allergen immunotherapy defined as 5 consecutive prescriptions of AIT within 4 years. Index-date defined as 5 consecutive prescriptions of AIT + estimated vaccination period (224 days). Follow-up year one defined as first year after end of AIT. Follow-up year two defined as the second year after end of AIT. Follow-up year three defined as third year after end of AIT.

Figure 3: The percentage of patients with SAA vs. PAA with ≥ 1 event of lower respiratory tract infection, ≥ 1 exacerbation and ≥ 1 exacerbation related to infections before commencing AIT (baseline). Significantly more patients with SAA had exacerbation at baseline compared with PAA and there was also a tendency toward more exacerbation related to infections, albeit statistically not significant. There was no difference in the proportion of patients with lower respiratory tract infection.

Figure 4: The number of patients with ≥ 1 exacerbation is shown between the four observation periods using McNemar's test for repeated measurements. The Pre-AIT value is presented as a mean for graphical reasons because it varies slightly using this statistical method. The figure shows a highly significant decrease ($p<.001$) from Pre-AIT (baseline) to each of the three follow-up years post-AIT for both SAA and PAA. The figure shows a mean decrease from Pre-AIT to follow-up years of 74% for SAA and 57% for PAA.

Figure 5: The number of patients with ≥ 1 lower respiratory tract infection is shown between the four observation periods using McNemar's test for repeated measurements. The Pre-AIT value is presented as a mean for graphical reasons because it varies slightly using this statistical method. Number of events in SAA was reduced from Pre-AIT (n=316) to follow-up year one (n=246), p=0.01; and follow-up year two (n=256), p=0.02; and follow-up year three (n=255), p=.002, yielding a mean decrease of 20%. In PAA, LRTIs were reduced from baseline (n=259) to follow-up

year one (n=219), p=0.19; and follow-up year two (n=218), p=0.01; and follow-up year three (n=207), p=0.03, with an average decrease of 17%.

Figure 6: The number of patients with ≥ 1 exacerbation related to infection is shown between the four observation periods using McNemar's test for repeated measurements. In SAA, we found a significant decrease in the number of patients with events from Pre-AIT (n=34) to follow-up year one (n=18), p=.038; follow up year two (n=9), p<.001; and follow-up year three (n=16), p=.016, yielding an average decrease of 58%. For PAA, there was no difference between Pre-AIT and the three follow-up years.

Appendices

Appendix 1: Lower respiratory tract infection - Defined as a prescription filling one of the following antibiotics targeting lower respiratory tract infections		
Type of antibiotic	Spectrum	ATC-code
Penicillin	Narrow	J01CE02
	Broad	J01CA04, J01CA01
Penicillin combined with beta-lactamase inhibitor	Broad	J01CR02
Cefuroxime	Broad	J01DC02
Macrolide	Narrow	J01FA10, J01FA06, J01FA09, J01FA01
Quinolones	Broad	J01MA14, J01MA02

Appendix 2: Exacerbation - Defined as a minimum of one of the following treatments provided:			
Treatment provided	ATC-code	SKS-codes in NPR	Description
Oral corticosteroids	H02AB06	-	-
Hospitalization	-	DJ45, DJ450, DJ450A, DJ451, DJ451A, DJ458, DJ459	Asthma
	-	DJ46, DJ469	Status asthmaticus (Ivanova et al. 2012).
Emergency department visit	-	DJ45, DJ450, DJ450A, DJ451, DJ451A, DJ458, DJ459	Asthma
	-	DJ46, DJ469	Status asthmaticus (Ivanova et al. 2012).

Appendix 3: Exacerbation related to infection - Defined as a minimum of one prescription filling on antibiotics and oral corticosteroids within a 4-week period.	
Treatment provided	ATC-code
Antibiotics	J01CE02, J01CA02, J01CA04, J01CA01, J01CR02, J01DC02, J01FA10, J01FA06, J01FA09, J01FA01, J01MA14, J01MA02
Oral corticosteroids	H02AB06

Figure 1

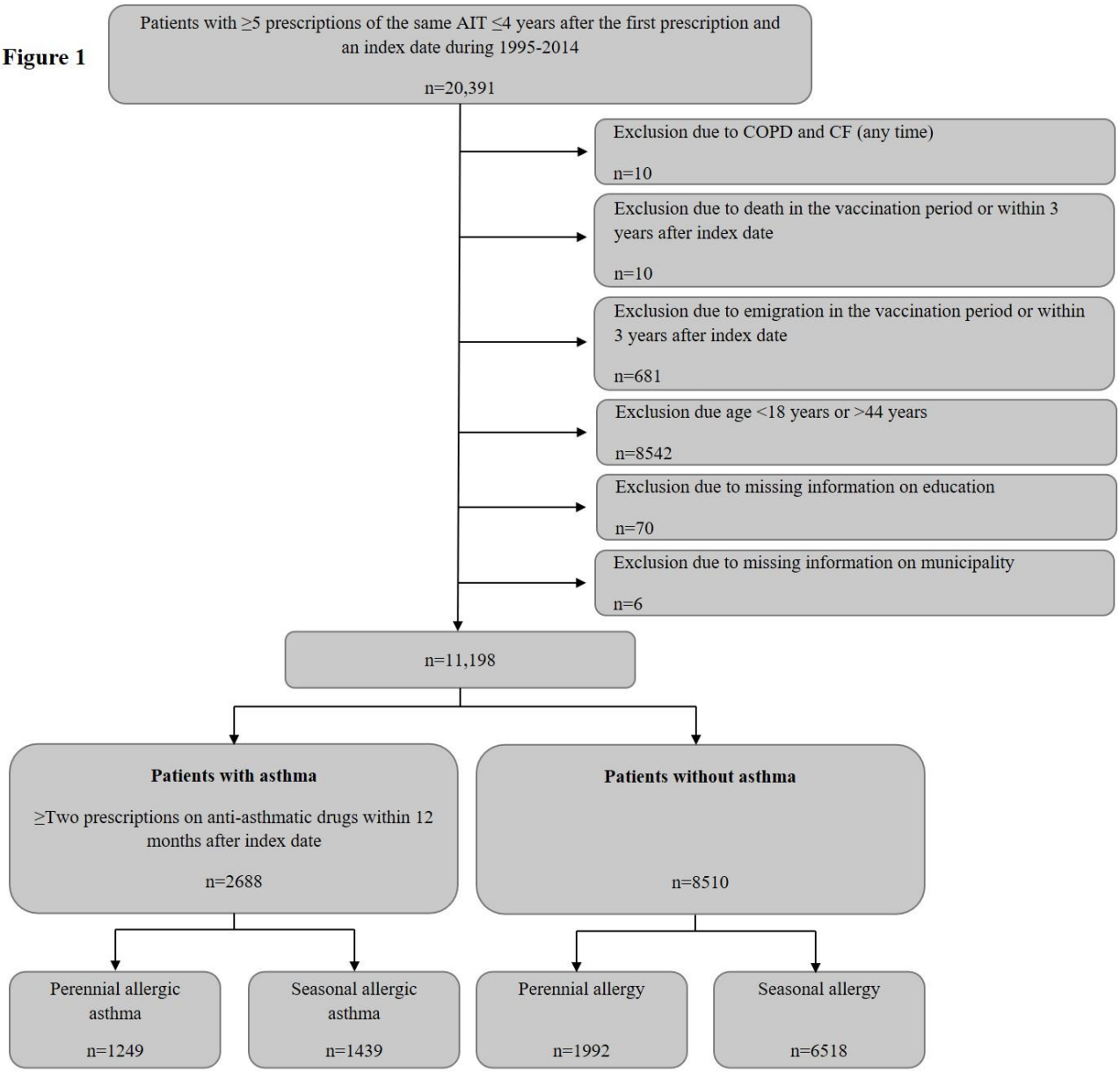

Patients with ≥5 prescriptions of the same AIT ≤4 years after the first prescription and an index date during 1995-2014
n=20,391
Exclusion due to COPD and CF (any time)
n=10
Exclusion due to death in the vaccination period or within 3 years after index date
n=10
Exclusion due to emigration in the vaccination period or within 3 years after index date
n=681
Exclusion due age <18 years or >44 years
n=8542
Exclusion due to missing information on education
n=70
Exclusion due to missing information on municipality
n=6
n=11,198
Patients with asthma
≥Two prescriptions on anti-asthmatic drugs within 12 months after index date
n=2688
Patients without asthma
n=8510
Perennial allergic asthma
n=1249
Seasonal allergic asthma
n=1439
Perennial allergy
n=1992
Seasonal allergy
n=6518

Figure 2

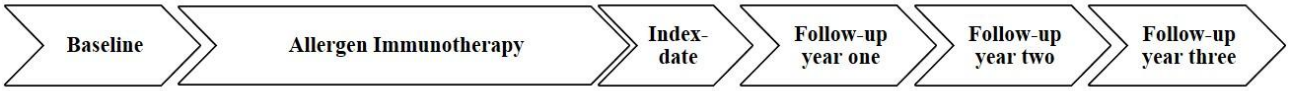

Baseline
Allergen Immunotherapy
Index-date
Follow-up year one
Follow-up year two
Follow-up year three

Figure 3

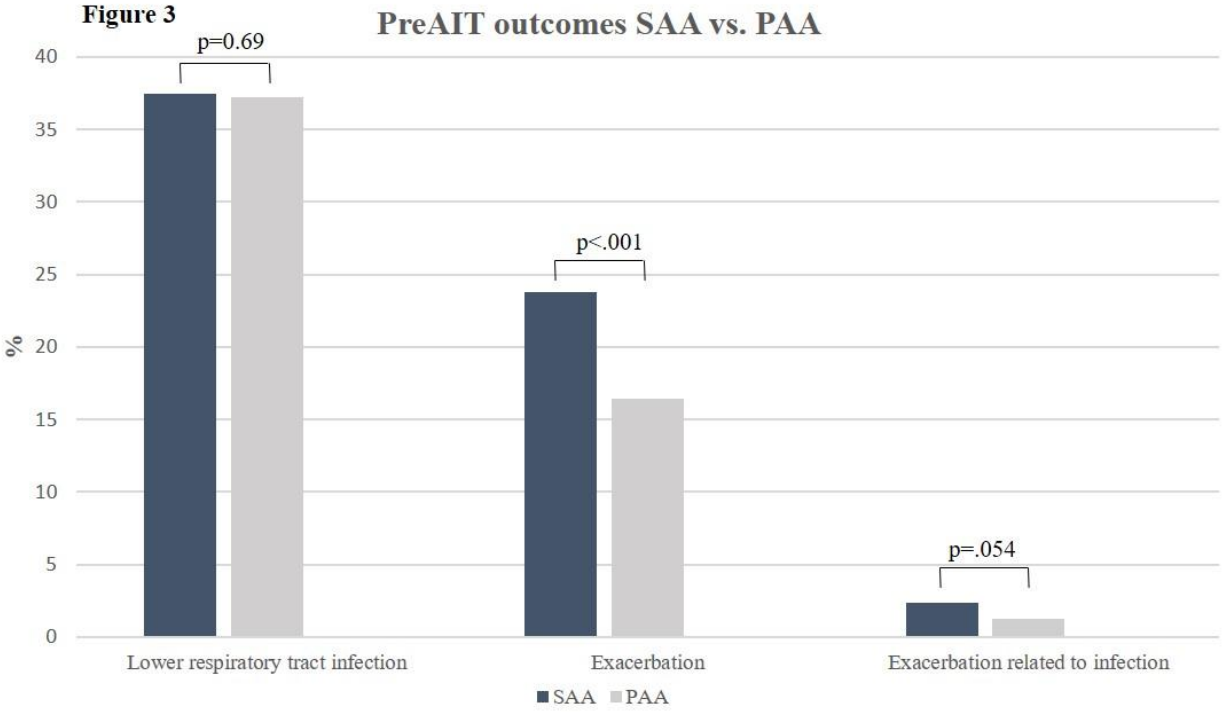

PreAIT outcomes SAA vs. PAA
p=0.69
p<.001
p=.054
40
35
30
25
20
15
10
5
0
%
Lower respiratory tract infection
Exacerbation
Exacerbation related to infection
SAA
PAA

Figure 4

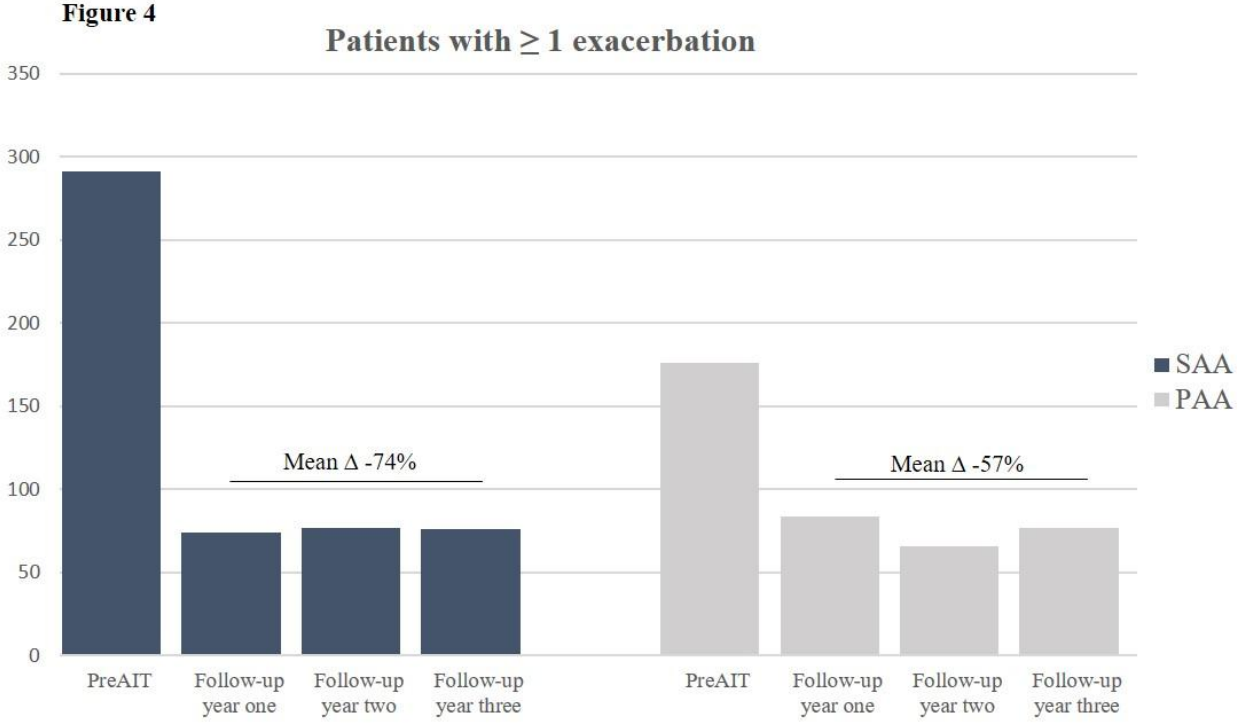

Patients with ≥ 1 exacerbation
350
300
250
200
150
100
50
0
Mean Δ -74%
Mean Δ -57%
SAA
PAA
PreAIT
Follow-up year one
Follow-up year two
Follow-up year three
PreAIT
Follow-up year one
Follow-up year two
Follow-up year three

Figure 5

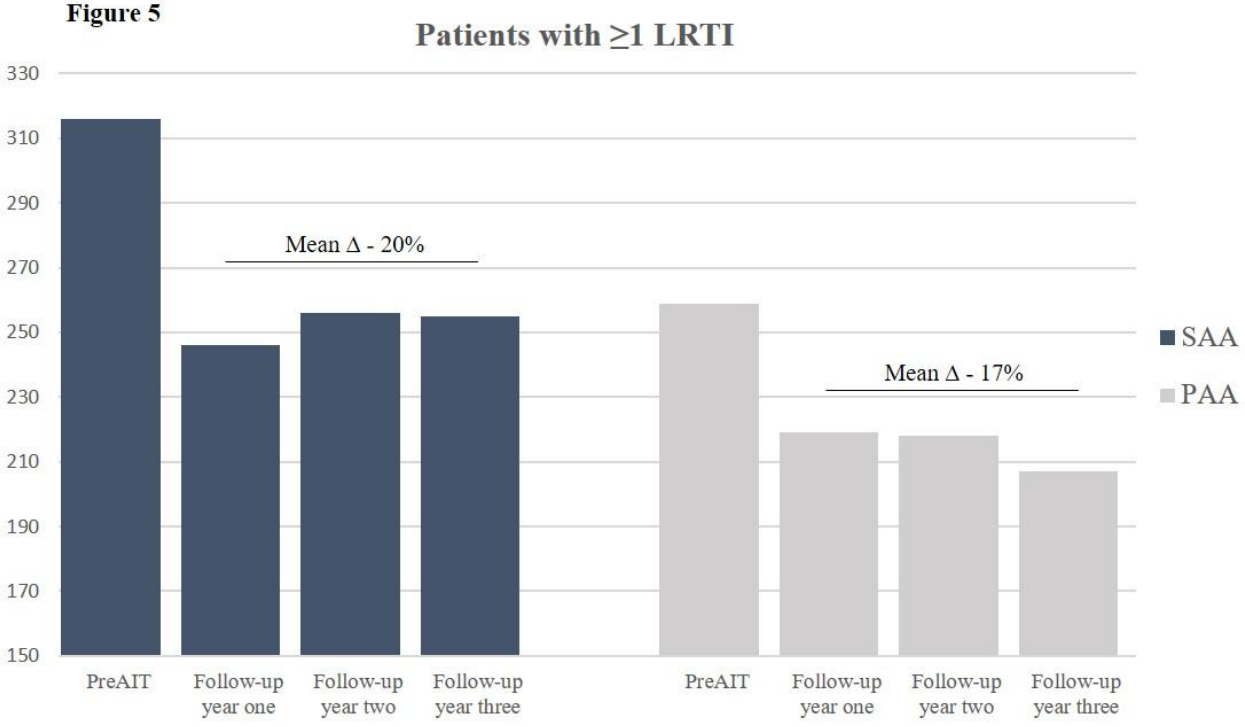
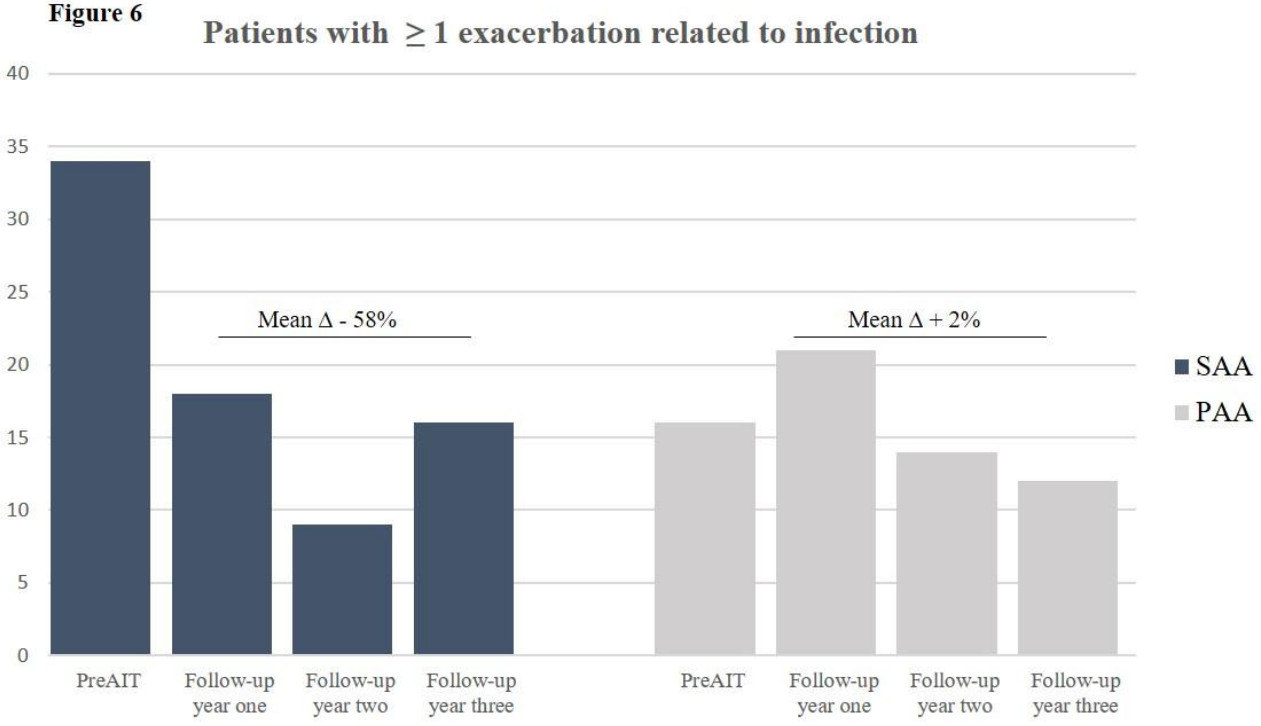

Patients with ≥1 LRTI
330
310
290
270
250
230
210
190
170
150
Mean Δ - 20%
Mean Δ - 17%
SAA
PAA
PreAIT
Follow-up year one
Follow-up year two
Follow-up year three
PreAIT
Follow-up year one
Follow-up year two
Follow-up year three

Figure 6


Patients with ≥1 exacerbation related to infection
40
35
30
25
20
15
10
5
0
Mean Δ - 58%
Mean Δ + 2%
PreAIT
Follow-up year one
Follow-up year two
Follow-up year three
PreAIT
Follow-up year one
Follow-up year two
Follow-up year three
SAA
PAA

Allergen immunotherapy effectively attenuates the risk for exacerbations and lower respiratory tract infections in both seasonal- and perennial allergic asthma

- a nationwide epidemiologic study

Supplementary material for Online Repository.

Authors and affiliations:

Christian Woehlk, M.D[1]. (corresponding author)

Anna Von Bülow, M.D., Ph.D[1].

Muzhda Ghanizada, M.D[1].

Marianne Baastrup Søndergaard, M.D[1].

Susanne Hansen, MSc. Public Health, Ph.D[1].

Celeste Porsbjerg, Professor, M.D[1].

[1]Respiratory Research Unit, Dept. Respiratory Medicine, Ebba Lunds Vej 48, entrance 66, 2400 Copenhagen NV, Denmark.

Corresponding author:

Dr. Woehlk, Respiratory Research Unit, Dept. Respiratory Medicine, Ebba Lunds Vej 48, entrance 66, 2400 Copenhagen NV, Denmark.

Email: cwoe0007@regionh.dk

Tel: +45 53644292 (direct)

Methodology

As described in the original article.

Results for patients without asthma (Online Repository)

In patients with perennial allergy without asthma (PA) we observed increased numbers of ≥1 LRTI compared with seasonal allergy without asthma (SA) Pre-AIT (baseline): 29.7 % vs. 26.7 %, p=.034. Post-AIT, we found similar results: follow-up year one PA vs. SA: 27.5 % vs. 24.5 %, p<.001; follow-up year two: 27.1 % vs. 24.7 %, p=.018. However, in follow-up year three no difference was observed between PA and SA: 26.5 % vs. 25.1 %, p=.074, Table E1.

In patients with SA, we observed a significant decrease in LRTI from baseline (Pre-AIT) to all three follow-up years, Figure E4. From baseline (n=1168) to follow-up year one (n=1017), p=.002; follow-up year two (n=1035), p=.004; and follow-up year three (n=1068), p=.031, yielding an average decrease of 11%,. In patients with PA we observed a decrease in LRTI from baseline (n=383) to follow-up year three (n=350), p=.038, with an average decrease of 12 % from baseline to follow-up, Figure E4.

Lastly, we compared the rate ratio (RR) of LRTIs' in the follow-up period between PA and SA. In follow-up year one PA had increased RR of LRTI compared with SA (ref): 1.23 CI. 95(1.14;1.34), p<.001 and follow-up year two: 1.18 CI. 95(1.09;1.28), p<.001 and follow-up year three: 1.14 CI. 95(1.05;1.23), p=.002, Figure E5.

Discussion (Online Repository)

We have previously shown that allergic asthma is associated with increased risk of lower respiratory tract infections (LRTI) and that AIT completely abrogated this risk. Hence, we commenced this study to evaluate whether a perennial or seasonal allergic phenotype asthma is more susceptible to infections. In the data covering patients with asthma we didn't find any differences in LRTI, however the data shown above suggests patients with perennial allergy without asthma does have persistent (pre and post AIT) increased risk of LRTI despite AIT. The effect of AIT to attenuate infection tendencies was more modest than seen in allergic asthma. These findings suggest that significant allergy symptoms should be addressed appropriately and that more research in driving immunological mechanisms is warranted to fully understand the impact of aeroallergens on the airway epithelium.

Online Repository Tables and Figures.

Table E1				
Period	LRTI	SA (n=6518)	PA (n=1992)	p-value
Baseline	0	4776 (73.3)	1407 (70.6)	
	≥1	1742 (26.7)	585 (29.4)	.034
Follow-up year one	0	4921 (75.5)	1445 (72.5)	
	≥1	1597 (24.5)	547 (27.5)	<0.001
Follow-up year two	0	4911 (75.3)	1452 (72.9)	
	≥1	1607 (24.7)	540 (27.1)	.018
Follow-up year three	0	4879 (74.9)	1465 (73.5)	
	≥1	1639 (25.1)	527 (26.5)	.074

Figure legends:

Table E1: The table shows the number of patients with ≥1 LRTI at Baseline (pre-AIT) and in the post-AIT follow-up period among patients with perennial allergy and seasonal allergy without asthma. The p-value is the result of a chi-squared test.

Figure E1: Shows the results of the adjusted (age, sex, municipality, and education) rate ratio (RR) using a Poisson regression model of a count variable for exacerbations in each of the follow-up years (post-AIT) between PAA and SAA (ref). In follow-up year one, we found a marginal increased rate of exacerbations for PAA after which there is no difference in the rate between the two groups. Overall, we consider our findings conservative with no differences between PAA and SAA.

Figure E2: Shows the results of the adjusted (age, sex, municipality, and education) rate ratio (RR) using a Poisson regression model of a count variable for lower respiratory tract infections in each of the follow-up years (post-AIT) between PAA and SAA (ref). In follow-up year one, we found an increased RR for PAA after which in follow-up years two and three there is no difference. Overall, we consider our findings conservative with no differences between PAA and SAA.

Figure E3: Shows the results of the adjusted (age, sex, municipality, and education) rate ratio (RR) using a Poisson regression model of a count variable for exacerbation related to infection in each of the follow-up years (post-AIT) between PAA and SAA (ref). In this outcome, we found no differences in the RR between PAA and SAA in any of the three observation periods.

Figure E4: The numbers of patients with ≥ 1 lower respiratory tract infection is shown between the four observation periods using McNemar's test for repeated measurements. The pre-AIT value is presented as a mean for graphical reasons because it varies a bit using this statistical method. Overall, we find a mean decrease from baseline to follow-up in SA of 11% and in PA 12 %.

Figure E5: Shows the results of the adjusted (age, gender, municipality, and education) rate ratio (RR) using a Poisson regression model of a count variable for lower respiratory tract infections in each of the follow-up years (post-AIT) between PA and SA (ref). In each of the follow-up years, we find an increased RR of LRTI among PA compared with SA yielding a mean increased RR of 1.23 95CI% (1.14;1.34).

Figure E6: Shows the percentage reduction in exacerbation in the follow-up years.

Figure E7: Shows the percentage reduction in LRTI in the follow-up years.

Figure E1

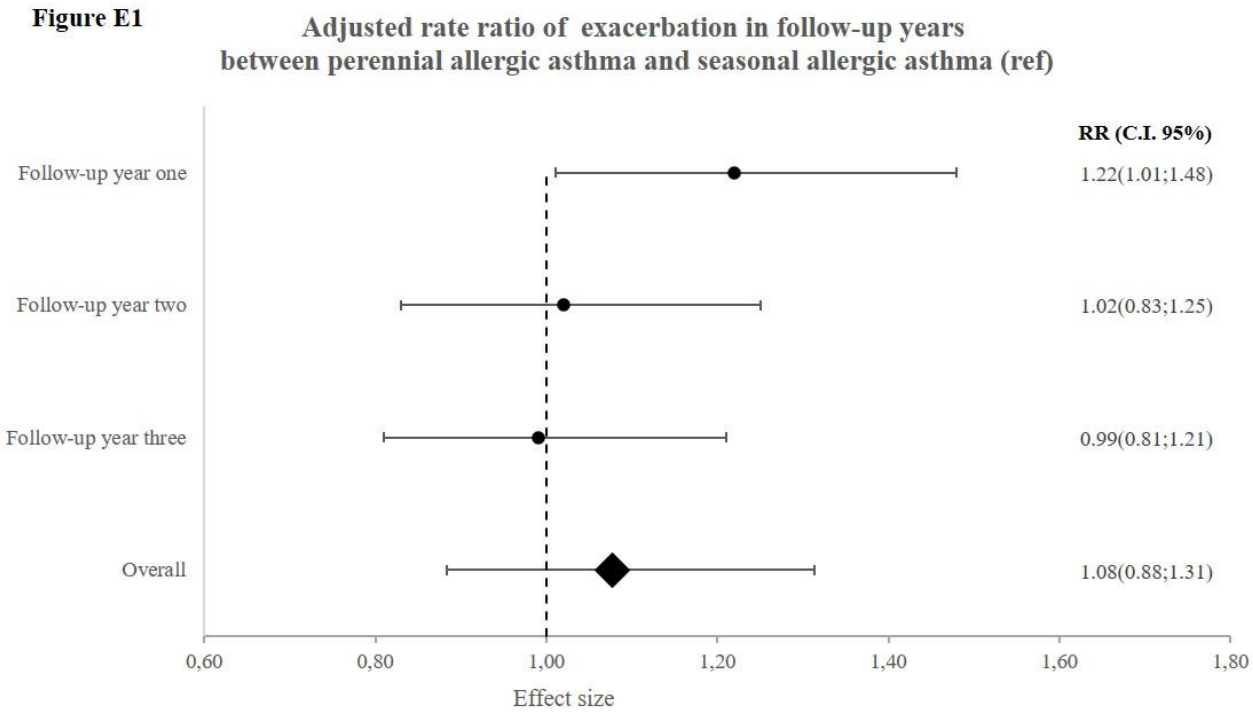

Adjusted rate ratio of exacerbation in follow-up years between perennial allergic asthma and seasonal allergic asthma (ref)
RR (C.I. 95%)
Follow-up year one
1.22(1.01;1.48)
Follow-up year two
1.02(0.83;1.25)
Follow-up year three
0.99(0.81;1.21)
Overall
1.08(0.88;1.31)
0,60
0,80
1,00
1,20
1,40
1,60
1,80
Effect size

Figure E2 **Adjusted rate ratio of lower respiratory tract infection in follow-up years between perennial allergic asthma and seasonal allergic asthma (ref)**

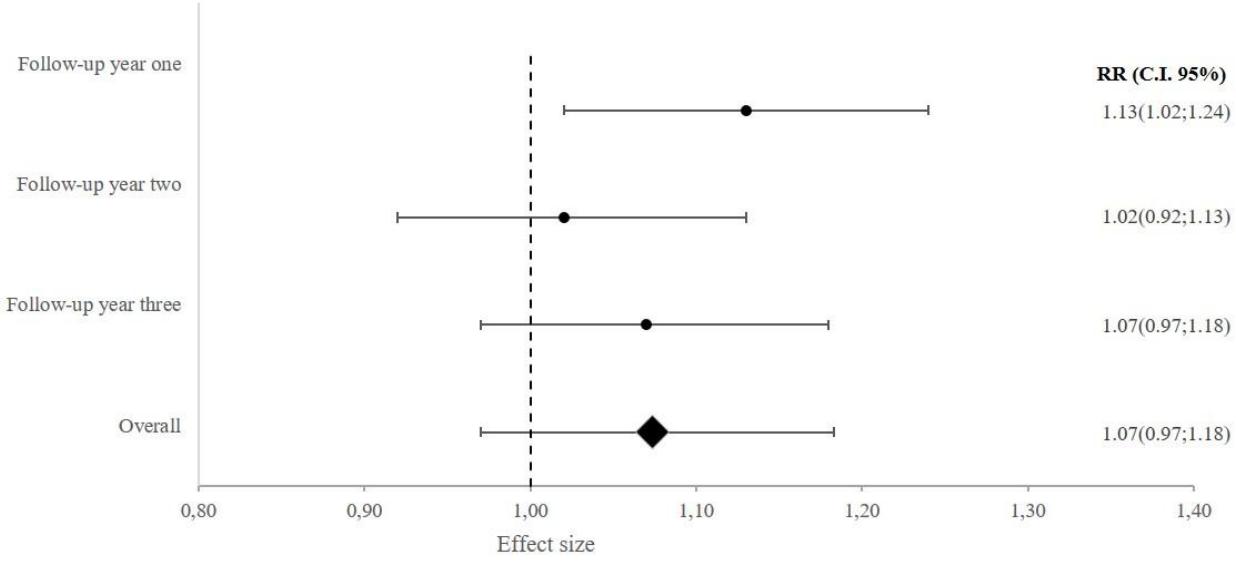


Figure E3 **Adjusted rate ratio of exacerbation related to infection in follow-up years between perennial allergic asthma and seasonal allergic asthma (ref)**

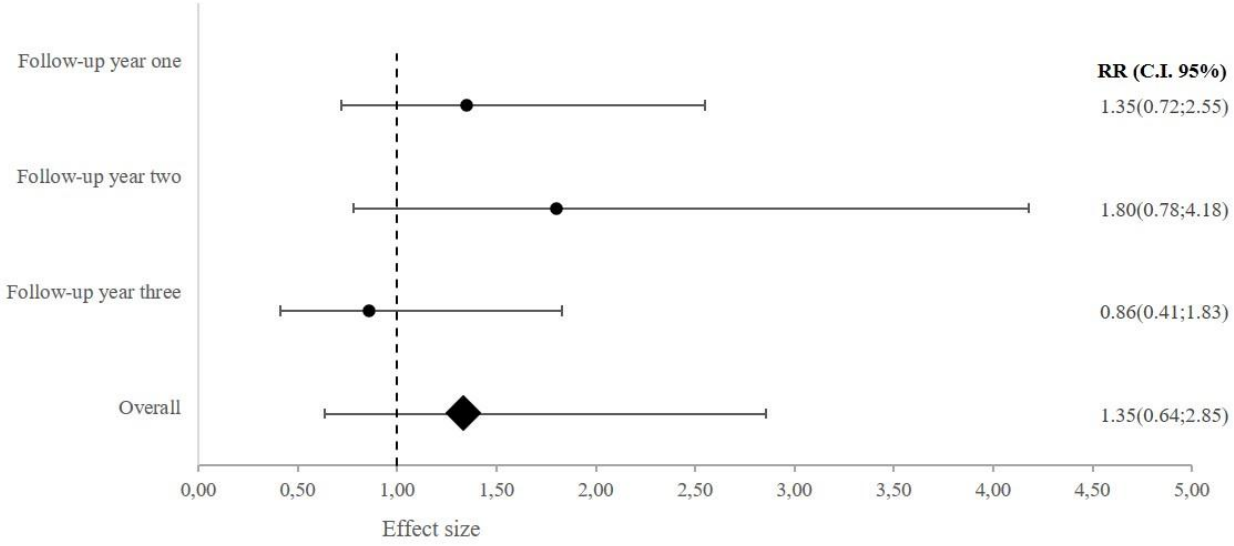


Figure E4

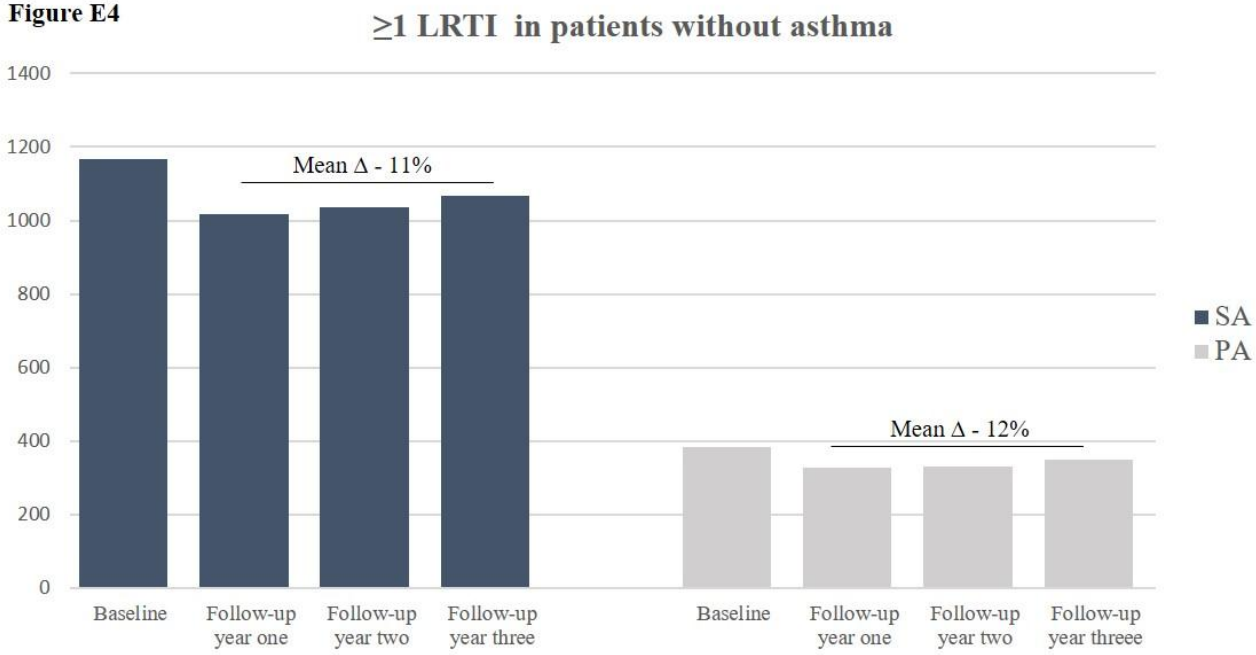

≥1 LRTI in patients without asthma
1400
1200
1000
800
600
400
200
0
Mean Δ - 11%
Mean Δ - 12%
Baseline
Follow-up year one
Follow-up year two
Follow-up year three
Baseline
Follow-up year one
Follow-up year two
Follow-up year threee
SA
PA

Figure E5

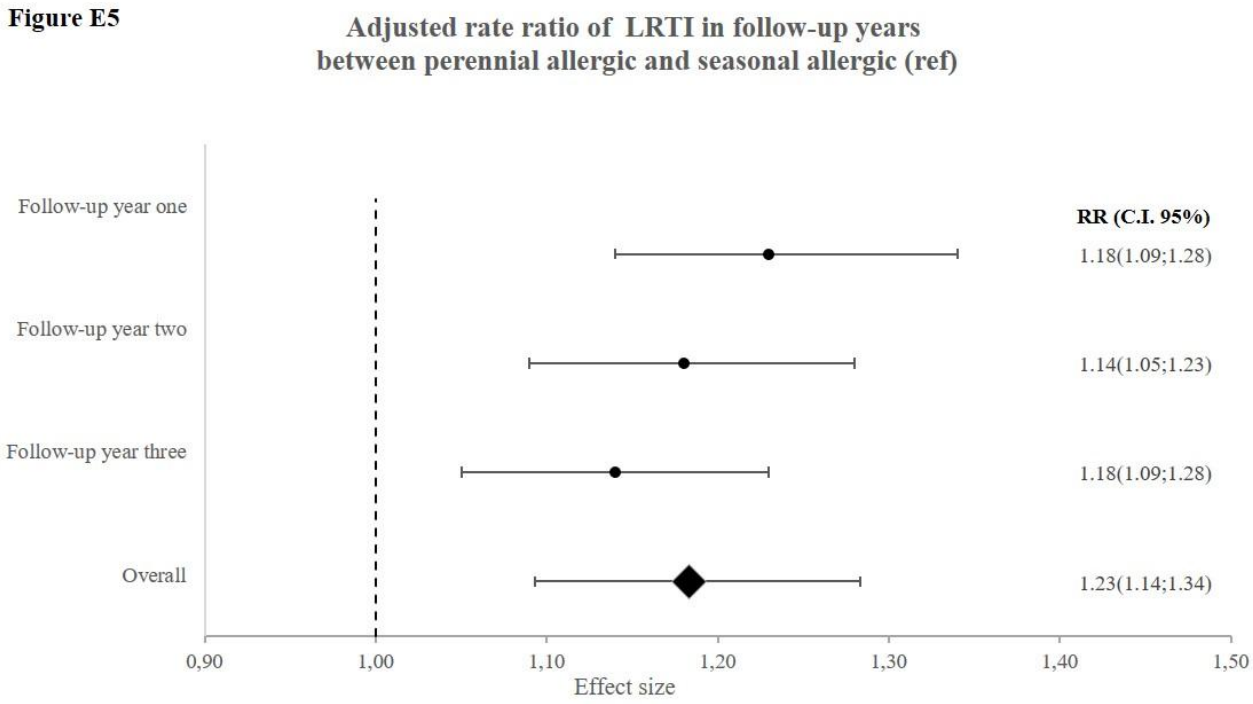

Adjusted rate ratio of LRTI in follow-up years between perennial allergic and seasonal allergic (ref)
Follow-up year one
Follow-up year two
Follow-up year three
Overall
RR (C.I. 95%)
1.18(1.09;1.28)
1.14(1.05;1.23)
1.18(1.09;1.28)
1.23(1.14;1.34)
0,90
1,00
1,10
1,20
1,30
1,40
1,50
Effect size

Figure E6

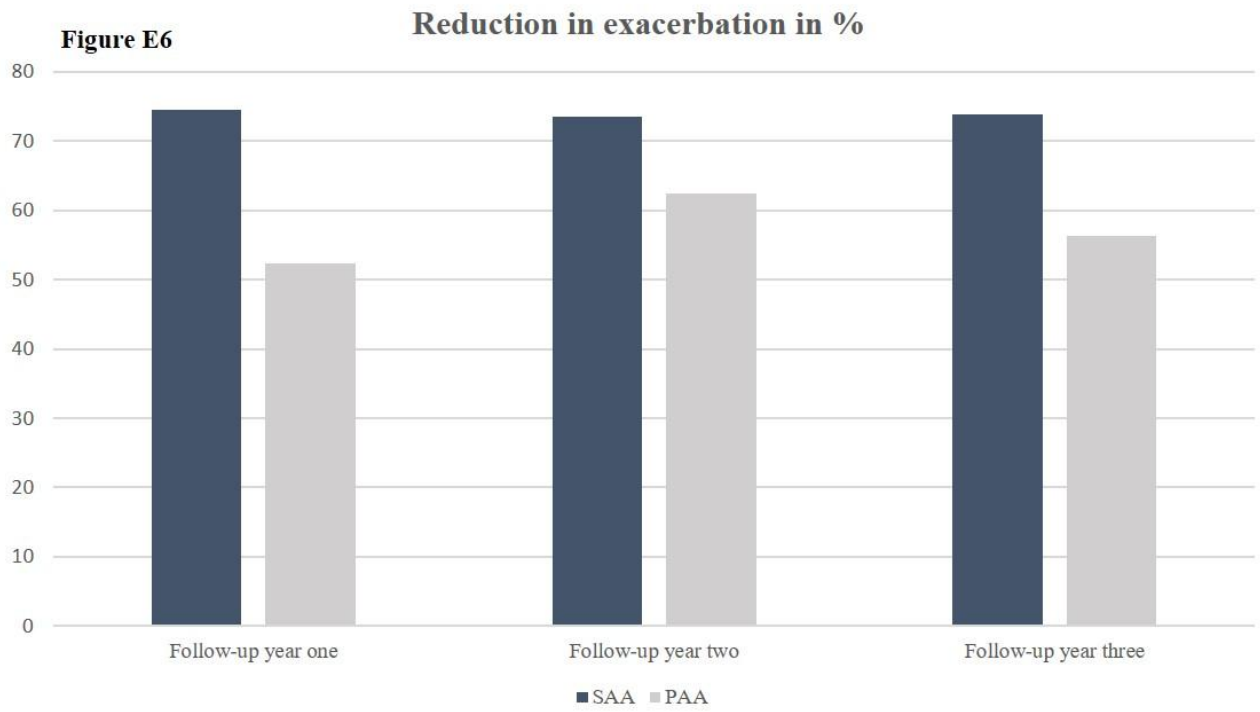
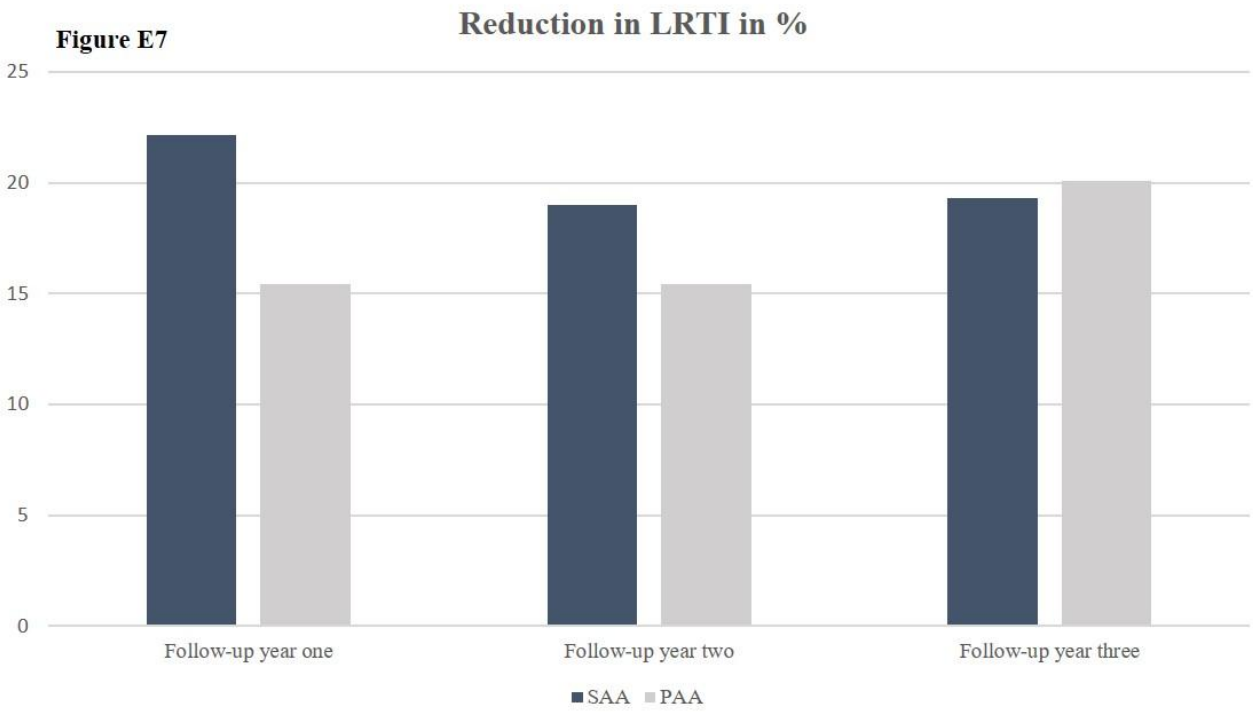

Reduction in exacerbation in %
80
70
60
50
40
30
20
10
0
Follow-up year one
Follow-up year two
Follow-up year three
SAA
PAA

Figure E7


Reduction in LRTI in %
25
20
15
10
5
0
Follow-up year one
Follow-up year two
Follow-up year three
SAA
PAA