

Article type : Original Article-Asthma and Rhinitis

**Longitudinal analysis to better characterize Asthma-COPD Overlap Syndrome
(ACOS): Findings from an adult asthma cohort in Korea (COREA)**

So-Young Park, MD^{1,2*}, Heewon Jung, BS^{2,3}, Jung-Hyun Kim, MD⁴, Bomi Seo, MD⁴, Oh Young Kwon, MD², Sungkyoung Choi, PhD⁵, Bermseok Oh, PhD⁶, Hyouk-Soo Kwon, MD, PhD², You Sook Cho, MD, PhD², Hee-Bom Moon, MD, PhD², Sungho Won, PhD⁷, Taesung Park, PhD⁸, and Tae-Bum Kim MD, PhD².

¹Department of Internal Medicine, Division of Pulmonary, Allergy and Critical care medicine, Konkuk University Medical Center, Seoul, Korea

²Department of Internal Medicine, Division of Allergy and Clinical Immunology, Asan Medical Center, Ulsan University School of Medicine, Seoul, Korea

³Department of Applied Statistics, Chung-Ang University, Seoul, Korea

⁴Department of Internal Medicine, Seoul National University Bundang Hospital, Seongnam, Korea

⁵Department of Pharmacology, Yonsei University College of Medicine, Seoul, Korea

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi: 10.1111/cea.13339

This article is protected by copyright. All rights reserved.

⁶*Department of Biochemistry and Molecular Biology, School of Medicine, Kyung-Hee University, Seoul, Korea*

⁷*Department of Public Health Science, Seoul National University, Seoul, Korea*

⁸*Department of Statistics, Seoul National University, Seoul, Korea*

Corresponding author: Tae-Bum Kim, MD, PhD

Department of Allergy and Clinical Immunology

Asan Medical Center, University of Ulsan College of Medicine

88 Olympic-ro 43-gil

Songpa-gu, Seoul 05505, Korea

Tel: 82-2-3010-3280; Fax: 82-2-3010-6969; e-mail: tbkim@amc.seoul.kr

ORCID: Tae-Bum Kim; <https://orcid.org/0000-0001-5663-0640>

Co-corresponding author: Taesung Park, PhD

Department of Statistics, Seoul National University 56-1 Mountain, Sillim-dong, Gwanak-gu,

Seoul 151-747, Korea

Tel: 82-2-880-8924; e-mail: taesungp@gmail.com

Keywords: Asthma; asthma-COPD overlap syndrome (ACOS); cluster analysis; genome-wide association study (GWAS); longitudinal studies

This article is protected by copyright. All rights reserved.

Short running title

Highly variable disease, ACOS should be defined in a longitudinal way.

Author contributions

SY Park designed this study, acquired the data, analyzed the data, and drafted the manuscript.

H Jung, S Choi, B Oh, and S Won analyzed the data, drafted the manuscript, and revised the manuscript. JH Kim, B Seo, and OY Kwon acquired the data and drafted the manuscript. HS

Kwon, YS Cho, and HB Moon participated in interpretation of the data and revised the manuscript. TB Kim and T Park designed this study, analyzed data, drafted the

manuscript, and revised the manuscript. All authors have read and approved the final

manuscript. All authors have accountability for all aspects of the work ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Conflicts of interest

The authors declare no conflicts of interest.

Abstract

Background: Asthma-chronic obstructive pulmonary disease (COPD) overlap syndrome (ACOS), which has received much attention, has not been unanimously defined.

Objective: In this study, we tried to demonstrate that longitudinally defined ACOS is more useful in the real-world than blending patients with asthma and COPD.

This article is protected by copyright. All rights reserved.

Methods: The study patients had undergone two consecutive pulmonary function tests measured at least 3 months apart (n = 1,889). We selected the patients who had positive bronchodilator response or methacholine provocation tests (n = 959). Next, we defined ACOS as a patient with a persistent airflow obstruction [forced expiratory volume in 1 sec (FEV1)/forced vital capacity < 0.7] that was identified twice consecutively by an interval of at least 3 months (n = 228).

Results: The proportions of patients who were older, male, and smokers were significantly higher, and baseline lung function was lower in patients with ACOS. In the longitudinal analysis, the mean change in lung function was high, and a greater decline in FEV1 was observed in patients with ACOS. In addition, we compared ACOS and severe asthma, and we also performed a cluster analysis and compared the results with our definition of ACOS. According to our definition, ACOS is an independent subtype with distinctive characteristics. Finally, a genome-wide association study (GWAS) was performed to identify genetic variations associated with ACOS, but no significant single nucleotide polymorphisms were identified.

Conclusion: Our findings suggest that ACOS should be defined longitudinally and considered as an independent subgroup distinguished by inherited environmental factors rather than as a genetically distinct independent group.

Introduction

Asthma is a heterogeneous disease characterized by chronic airway inflammation.¹ Asthma-chronic obstructive pulmonary disease (COPD) overlap syndrome (ACOS), which is characterized by a persistent airflow limitation with several features usually associated with asthma and COPD, has received much attention.² However, there is no general agreement on

the definition of ACOS, and the precise prevalence of the disease varies highly depending on physicians' opinions, but has been reported to be between 15 and 55%.³⁻⁶ Many studies on ACOS have been conducted, but most clinical studies have analyzed ACOS according to a particular study definition within patients with asthma or COPD.⁷⁻¹¹ Therefore, when ACOS has been defined in a COPD study, the features associated with typical asthma, such as eosinophilia and history of atopy, have been stressed.¹⁰ In contrast, when a study has been performed in patients with asthma, a persistent airway obstruction and a history of smoking were highlighted in the definition of ACOS.¹¹ In this way, the characteristics of ACOS emphasized in each study appear to vary according to the study group.

The definition of ACOS in most studies is only a cross-sectional definition, so it does not accurately reflect the characteristics of ACOS because asthma and COPD are highly variable chronic diseases. Some longitudinal analyses have been performed.^{9,12,13} The aim of this study was to emphasize the concept that a longitudinal trend in a fixed airway obstruction should be included in the definition of ACOS because the most important feature emphasized in previous ACOS studies was a "persistent airway obstruction".^{11,12,14-16} However, severe asthma may also be confused with ACOS because patients with severe asthma have features of a fixed airway obstruction due to airway remodeling. Therefore, we classified severe asthma and compared it with ACOS. Moreover, we analyzed the clinical characteristics of ACOS and conducted cross-sectional as well as longitudinal analyses over 1 and 3 years. In addition, we performed a cluster analysis and compared patients with ACOS using a longitudinal definition. We then performed a genome-wide association study (GWAS) to identify the factors that explained the genetic background and the pathogenesis of ACOS, defining it as an independent group. In this study, we tried to prove that longitudinally

defined ACOS is a distinct group, and we identified a concrete, real-world definition of ACOS that blended patients with asthma and COPD.

Methods

Study patients and measurements

We studied the clinical data of 3,299 patients with asthma who were registered in the Cohort for Reality and Evolution of Adult Asthma in Korea (COREA) from 23 referral centers.^{17,18} Medical records between May 2005 and September 2015 were retrospectively reviewed. This study was approved by the Institutional Review Board of Asan Medical Center (2015-0435). Additional details are provided in the online supplements.

Definition of ACOS and severe asthma

A total of 1,889 patients with results from two consecutive pulmonary function tests at least 3 months apart were included. Among them, 959 patients satisfied either of the following criteria according to the Global Initiative for Asthma (GINA) guidelines:² (1) positive response to bronchodilator (forced expiratory volume in 1 sec [FEV1] > 200 mL and > 12% increase from baseline), or (2) positive methacholine or mannitol provocation test. To reflect a persistent airway obstruction, we defined the overlapping group with a continuous airflow obstruction as follows: pre-bronchodilator FEV1/forced vital capacity (FVC) ratio < 0.7 at baseline and continuing for at least 3-months of follow-up despite appropriate therapy. Patients who did not meet this condition were classified as the “only” asthma group. A flowchart summarizing the definition of ACOS is shown in Figure 1. Severe asthma was defined as patients treated with the medication corresponding to GINA steps 4 and 5 (n =

198) among those patients previously defined as having “only” asthma (n = 731), and severe asthma was compared with ACOS as defined above.

Longitudinal assessment

A longitudinal lung function analysis was performed for 1 and 3 years as follows: (Method 1) the mean absolute values for the changes in the FEV1, FVC, and the FEV1/FVC (which were measured over 3-month intervals) were analyzed for 1 and 3 years; (Method 2) the personal best of FEV1, FVC, and FEV1/FVC were defined as MAX and the personal worst were defined as MIN, and the difference between MAX and MIN [(MAX) – (MIN)] was analyzed for 1- and 3-year periods; (Method 3) the longitudinal declines of FEV1, FVC, and FEV1/FVC were analyzed using a linear mixed model.

An exacerbation of asthma was defined as the presence of one or more of the following during the previous year: (1) emergency room visit; (2) unexpected outpatient visits; (3) inpatient or intensive care unit treatment; or (4) use of systemic corticosteroids. We also analyzed the exacerbation rate per year, and the changes in Asthma Control Test (ACT) scores were analyzed using a linear mixed model. Mean inhaled corticosteroid (ICS) dose was also analyzed.

Genome-wide association study

Genotype data were produced using the Korean Chip (K-CHIP) available through the K-CHIP consortium (<http://kogo.or.kr>). K-CHIP was designed by the Center for Genome Science, Korea National Institute of Health, Korea (4845-301, 3000-3031). Data from the

Severance Hospital Health Examination Center in Korea was used as the control group (N = 5,915). Among the 1,433 patients with genetic information in the COREA cohort, 77 patients were defined as having ACOS and the remaining patients were classified as having asthma (N = 1,356). We compared normal subjects and those with asthma, patients with ACOS and normal subjects, and finally patients with asthma and those with ACOS. Additional details, including quality control are described in the online supplements.

Cluster and statistical analyses

The ACOS and “only” asthma patients were combined (n = 959), and a hierarchical cluster analysis was performed using Ward’s method. The hierarchical cluster analysis using Ward’s method generated a dendrogram to estimate the number of likely clusters within the population. This estimate was pre-specified in a κ -means cluster analysis that was used as the principal clustering technique.^{17,19} The parameters for the cluster analysis were selected based on defined criteria used in previous studies.^{10,20} Seven variables, including age, asthma onset age, smoking status, atopy, FEV1, FEV1/FVC, and serum blood eosinophil count were used to characterize the asthma clusters. The positive response to a bronchodilator parameter was excluded because all patients had to meet bronchodilator response-positive or methacholine provocation test-positive conditions before distinguishing ACOS from “only” asthma.

All statistical analyses were performed with R software (R Project for Statistical Computing, Vienna, Austria; www.r-project.org). The results are presented as mean $\pm$ SD for continuous variables and as counts of events (%) for categorical variables. The baseline characteristics of the ACOS and “only” asthma groups were compared using *t*-tests for

continuous variables and the χ^2 or Freeman–Halton exact tests for categorical variables. We analyzed and compared the exacerbation rate per year by negative binomial regression. Longitudinal analysis of lung function decline was performed by using a linear mixed model. Analysis of variance was used to compare groups in the cluster analysis. When data values for FEV1, FVC, and FEV1/FVC in the pulmonary function test results were missing, the most recent values were imputed. A p-value < 0.05 was considered significant.

Results

Clinical characteristics of ACOS

According to the definition, we diagnosed 23.8% (n = 228) of 959 asthma patients with ACOS. The remaining 76.2% (n = 731) of asthmatics were classified as having “only” asthma (Figure 1). We conducted a cross-sectional analysis by utilizing information at the time of enrollment and compared the clinical characteristics of each group. The mean age of the ACOS group was significantly higher than that of the “only” asthma group (57.56 ± 13.27 vs. 46.19 ± 15.75 years, $p < 0.001$). The percentage of males was higher in the ACOS group (62.7 vs. 37.1%, $p < 0.001$), and the percentage of current or ex-smokers was also higher in the ACOS group (63.5 vs. 42.6%, $p < 0.001$). However, no significant differences were observed between the groups in terms of body mass index (BMI) or family history. The proportion of atopic patients (positive skin prick test) was higher in the “only” asthma group (Table 1), and the results of the skin prick test are summarized in Supplement E1. No significant differences in serum levels of total Ig E, white blood cell (WBC) counts, C-reactive protein (CRP) level, or blood eosinophil count were observed between the groups in this analysis. The results of the induced sputum cell count also did not reveal any significant

Accepted Article

differences between the groups. The average symptom onset age and the asthma diagnosis age in the ACOS group were higher than in the “only” asthma group. Furthermore, the time after the asthma diagnosis was longer for the ACOS group than the “only” asthma group (Table 1). The mean value for the variables in the pulmonary function tests were significantly different between the two groups; the ACOS group had lower pre-bronchodilator FEV1 (63.17 ± 19.42 vs. $84.64 \pm 18.41\%$, $p < 0.001$), pre-bronchodilator FVC (82.37 ± 19.59 vs. $90.47 \pm 17.99\%$, $p < 0.001$), and pre-bronchodilator FEV1/FVC ratio (0.59 ± 0.09 vs. 0.77 ± 0.09 , $p < 0.001$). Patients in the ACOS group also had lower post-bronchodilator FEV1 (71.02 ± 19.02 vs. $87.72 \pm 19.53\%$, $p < 0.001$) and a post-bronchodilator FEV1/FVC ratio (0.61 ± 0.10 vs. 0.79 ± 0.09 , $p < 0.001$). The ACOS patients showed more changes in FEV1 than patients with “only” asthma (22.87 ± 24.58 vs. $9.89 \pm 12.68\%$, $p < 0.001$). Patients with ACOS more frequently presented with positive and very positive bronchodilator test results (69.3 vs. 36.1% , 25.5 vs. 11.7% , $p < 0.001$). The positive rate on the methacholine provocation test was slightly higher in the “only” asthma group, but this difference was not significant (88.8 vs. 68.9% , $p = 0.318$). However, PC_{20} was significantly lower in the ACOS group (3.61 ± 5.10 vs. 6.07 ± 6.03 mg/mL, $p < 0.001$) (Table 1).

Longitudinal analysis

According to Method 1, the longitudinal assessment of the average change in FEV1, FVC, and FEV1/FVC over 1 and 3 years is reported in Table 2. The average change in FEV1 measured at 3-month intervals tended to be greater in the ACOS group than in the “only” asthma group (7.19 ± 5.07 vs. $6.36 \pm 4.26\%$, $p = 0.09$). We also obtained a similar result in terms of the average change in FVC (7.97 ± 6.46 vs. $6.08 \pm 4.90\%$, $p = 0.002$). The average change in FEV1 over 3 years was $4.82 \pm 2.64\%$ in the “only” asthma group and $5.76 \pm 3.46\%$

in the ACOS group ($p = 0.045$). The average change in FVC over 3 years was also higher in the ACOS group (6.13 ± 3.90 vs. $4.69 \pm 3.29\%$, $p = 0.005$). Similar results were obtained when analyzing the change in lung function using Method 2. The mean (MAX – MIN) FEV1 value was $15.92 \pm 11.29\%$ in the “only” asthma group and $18.17 \pm 12.57\%$ in the ACOS group over 1 year. The mean (MAX – MIN) FVC value was also significantly higher in the ACOS group over 1 year (19.19 ± 13.91 vs. $15.98 \pm 11.64\%$, $p = 0.018$). These changes were not significantly different over a 3 year period; however, the differences were still high in the ACOS group. In the results using Method 3, FEV1, FVC, and FEV1/FVC decreased more sharply in the ACOS group, but the differences were not statistically significant. The degree of FEV1 reduction tended to be higher in the ACOS group over 1 year ($0.44[0.18-0.70]$, $p = 0.08$) and 3 years ($0.17[0.07-0.27]$ $p = 0.08$). The degree of FVC reduction was significantly higher in the ACOS group over 1 year ($0.83[0.56-1.10]$, $p < 0.001$) and tended to be higher over 3 years ($0.17[0.07-0.27]$, $p = 0.10$). The medical history of acute exacerbation rate during 1 year was significantly different between the two groups, with the ACOS group showing a higher rate of exacerbation (2.38 ± 1.91 vs. 1.61 ± 2.74 , $p < 0.001$). Long-term follow-up for 3 years showed that the frequency of exacerbation in both groups was also different, albeit not statistically significant. No significant differences were observed in the change in the ACT score after 1 and 3 years (Table 2).

The average dose of ICS used for a year tended to be higher in the ACOS group than the “only” asthma group, but the difference was not significant (Supplement E2).

Comparison of severe asthma and ACOS

We compared the clinical characteristics of severe asthma with those of ACOS. A total of 198 asthmatic patients were defined as having severe asthma, accounting for 27.1% of the “only” patients with asthma. As in the previous analysis, patients with ACOS and severe asthma were compared, and the results are shown in Table 3. Similar to the results of our previous analysis, patients with ACOS had a significantly higher mean age (57.56 ± 13.27 vs. 43.76 ± 15.24 years, $p < 0.001$), and a higher percentage of males (62.7 vs. 39.4%, $p < 0.001$) and smokers (63.5 vs. 47%, $p < 0.001$). The proportions of patients with a family history of allergic disease and a positive skin prick test were higher among those with severe asthma. The blood test and sputum analysis results were not significantly different between the two groups. Patients with ACOS had a longer duration of symptoms, and patients with ACOS were diagnosed with asthma at an older age than those with severe asthma. Additionally, patients with ACOS had lower baseline lung function and more airway obstructions than patients with severe asthma. In contrast, bronchial reversibility and hypersensitivity were significantly higher in the ACOS group (Table 3). The change in pulmonary function was significantly higher in the patients with ACOS than in those with severe asthma. The change in the ACT score was not significantly different at 1 and 3 years. However, acute exacerbation was found to be higher in patients with ACOS than in patients with severe asthma at 3 years (32.7 vs. 10.5%, $p = 0.03$). The average consumption of ICS over 1 and 3 years was significantly higher in the severe asthma group (Supplement E3).

Cluster analysis

We performed a cluster analysis using seven variables. Among the 959 patients, 481 (50.16%) were finally analyzed excluding the missing values. The cluster plot and dendrogram are shown in the supplement (Supplement E4). The patients were divided into three groups for the cluster analysis. In particular, group 3 showed similar clinical features to those of patients with ACOS, as analyzed previously, but differed in character from the other two groups. Additionally, 63% of the patients in group 3 were eligible for ACOS, as defined previously (Supplement E4). The group 3 patients were older, with a higher percentage of males, and more smokers. In addition, a low FEV1 and an obstructive pattern ($FEV1/FVC < 0.7$) were observed in this group. The group 3 patients showed more changes in FEV1 and more frequently presented with positive and very positive bronchodilator tests (Table 4). We presumed that group 1 patients showed characteristics of early onset atopic asthma and that group 2 patients showed characteristics of female obese asthma.

Genome-wide association study

The genetic variations associated with asthma were determined by comparing the genomes of patients with asthma ($N = 1,356$) with those of normal subjects ($N = 5,915$). Although no individual SNPs reached a genome-wide significance level after a Bonferroni correction, the top signals were identified as susceptibility markers for asthma alone (Figure 2A and Supplement E5). No significant SNPs were identified in the analysis of patients with ACOS, nor were the top 10 SNPs similar to those previously analyzed for the "only" asthma group (Figure 2B and Supplement E6). The asthma alone and ACOS groups were then compared

and analyzed. No significant SNPs were found between the two groups (Figure 2C and Supplement E7).

Discussion

In this study, we emphasized that ACOS should be defined longitudinally. We devised this study to determine a real-world definition of ACOS that applies to patients with features of both asthma and COPD. Many of the existing ACOS definitions are complex, and many of the diagnostic criteria are difficult to use in practice. Therefore, a simplified definition involving the longitudinal concept may be appropriate. In this study, ACOS was defined using a step-wise approach with longterm observations. We defined ACOS using the results of continuous pulmonary function tests, namely a longitudinal concept, so that it is easy to apply in practice.

The results of the analysis conducted with our definition were in close agreement with those of many previous studies. The prevalence of ACOS was 23.8% of patients with obstructive airway disease in this study, and this result is consistent with that of previous studies in which the prevalence of ACOS was approximately 20% of patients with obstructive airway disease.^{14,21,22} The clinical features observed in this study were also similar to those of previous studies, including old age, male sex, increased duration of asthma, history of smoking, and low baseline FEV1.^{11,22-24} In this study, ACOS showed significant airway hyper-reactivity, as shown previously.^{10,20,25} Additionally, longitudinal analyses were performed for 1 and 3 years, during which patients with “only” asthma and ACOS showed different characteristics. The results of the longitudinal analyses revealed that pulmonary function decreased more in patients with ACOS after 1 and 3 years, and that

changes in FEV1 fluctuated more widely during the observation period. In addition, patients in the ACOS group tended to experience acute exacerbations more frequently than those in the “only” asthma group. Although not statistically significant, the patients with ACOS tended to have more variation in the symptom score (ACT). In addition, the mean ICS dose was higher among patients with ACOS than those with “only” asthma.

To prove that our longitudinal definition is reasonable, we also performed a cluster analysis in a non-arbitrary and unsupervised manner because we defined ACOS arbitrarily. The parameters for the cluster analysis were selected based on the previous consensus definition.^{10,20} We then compared the ACOS group according to the longitudinal definition of this study with the cluster subgroup considered to be ACOS. The results showed that there was a cluster subtype considered to represent ACOS, and the 63% of cluster 3 patients was fairly consistent with patients with ACOS who were defined according to the definition of this study. In other words, it was confirmed that ACOS could be classified into one stable and independent subgroup, indicating that our definition is reasonable.

According to our definition, ACOS is a subgroup with a strong emphasis on the concept of a fixed airway obstruction, and severe asthma has similar characteristics because it exhibits a fixed airway obstruction due to airway remodeling. However, the characteristics of ACOS were also distinguished from severe asthma in this study; patients with ACOS were more likely to be older, with more severe airway hyper-responsiveness and bronchial variability. In addition, acute exacerbations occurred more frequently in patients with ACOS.

It is clear that the diagnosis of ACOS should be based on objective indicators, such as biomarkers, but the pathophysiological features are poorly understood and few biomarker studies of patients with ACOS have been published.²⁶⁻²⁸ Therefore, it has been difficult to achieve a consensus on the ACOS definition to date.

This article is protected by copyright. All rights reserved.

Too few gene-related studies have been published to explain the pathophysiology of ACOS.^{29,30} In particular, this is the first and only ACOS GWAS study in Korea. In our analysis, significant genes associated with ACOS were not identified as expected, suggesting that ACOS is a disease entity mainly caused by environmental affects and aging.^{31,32} Although there are many reports of asthma-related GWAS results, most of these associations were to specific asthma subtypes. The results of the GWAS analysis were likely not significant because we did not divide the sample sets by subtypes of heterogeneous asthma.

This study had some limitations. First, this was a retrospective study. As the most appropriate design for a longitudinal analysis is a prospective study, further studies should be conducted in these areas to support our results. Second, some values were missing in the data. For this reason, some variables only showed tendencies and yielded statistically insignificant results. In addition, the analysis was only conducted in an asthma cohort, so there was a limitation obtaining clinical information associated with COPD. Third, we only analyzed the ACT score as a disease control assessment of ACOS. The ACT score is an assessment tool for asthma, and may not be suitable as an evaluation system for patients with ACOS combined with COPD characteristics. Appropriate standards to evaluate the quality of life of patients with ACOS must be developed. Fourth, there was no smoking history in the definition of ACOS. Thus, patients with COPD characteristics may not have been included. However, this is why we did not include smoking history in the definition of ACOS. That is, the longitudinal definition in our study emphasized a fixed airway obstruction. If a patient shows a continuous history of $FEV1/FVC < 0.7$ and an additional smoking history of more than 10 PYs, the case is more suitable as typical COPD than ACOS. Fifth, we defined a fixed airway obstruction as a continuous pre-bronchodilator $FEV1/FVC < 0.7$. In general, post-bronchodilator $FEV1/FVC$ is commonly used as a surrogate marker for a fixed airway

obstruction. However, most patients with stable asthma have negative bronchodilator reversibility, and there was no significant difference between post-bronchodilator FEV1 and pre-bronchodilator FEV1. In addition, in the real-world, the majority of patients with asthma do not perform the bronchodilator response in a stable condition; thus, there are insufficient data. Finally, the number of samples used in the GWAS analysis was small and no replication was performed.

Asthma is typically characterized by early onset, a good response to ICS, eosinophilic inflammation involving type 2 helper T (Th2) lymphocytes, and an association with other allergic diseases. In contrast, COPD is characterized by late onset, a poor response to ICS, neutrophilic inflammation involving CD8 lymphocytes, and an association with smoking.^{31,32}

Clinicians can easily distinguish between “only” asthma and “only” COPD patients because of their different pathophysiologies.³¹ However, a considerable number of patients have features of both diseases, which is called ACOS. Many studies conducted in patients with COPD emphasized the characteristics of asthma, and other studies conducted in patients with asthma, highlighted the characteristics of COPD.^{4,8,10,27} As a result, it is difficult to define ACOS.

Nevertheless, this is the first ACOS-related study using a large-scale multicenter cohort in Korea, and it is the first new study to apply a longitudinal definition of ACOS. In addition, this study has strengths, as various analyses were conducted to support this definition.

In conclusion, longitudinal follow-up results are important to determine a real-world definition of ACOS. In this study, we established a definition that distinguished ACOS usefully in practice by a relatively simple method. We define ACOS after observing patients for a long period, and ACOS showed the characteristics of a persistent airway obstruction and fluctuating lung function. Distinguishing ACOS in patients may be important because they

This article is protected by copyright. All rights reserved.

often have more severe, frequent exacerbations and a poorer prognosis than typical patients with asthma; thus, clinicians should be more aware of ACOS.

Acknowledgements

This study was supported by a grant of the Korea Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI) funded by the Ministry of Health & Welfare, Republic of Korea (grant numbers: HC15C1335, HI15C2165 & HI14C1971).

This research was conducted with support from AstraZeneca Korea Ltd.

The COREA Study Group includes the following investigators: Tae-Bum Kim (University of Ulsan, Seoul, Korea); Sae-Hoon Kim (Bundang Seoul National University, Seongnam, Korea); Byoung-Whui Choi and Jae-Woo Jung (Chung-Ang University, Seoul, Korea); Young-Joo Cho and Min-Hye Kim (Ewha Womans University, Seoul, Korea); An-Soo Jang (Soonchunhyang University, Bucheon, Korea); Yoo Seob Shin (Ajou University, Suwon, Korea); Ji-Yong Moon (Hanyang University, Guri, Korea), Jae-Woo Kwon (Kangwon National University, Chuncheon, Korea); Sang-Hoon Kim and So Young Park (Eulji University, Seoul, Korea); Taehoon Lee (University of Ulsan, Ulsan, Korea); Sujeong Kim (Kyungpook National University, Daegu, Korea); Chan Sun Park (Inje University Haeundae Paik Hospital, Busan, Korea); Joo-Hee Kim (Hallym University Sacred Heart Hospital, Anyang, Korea); Jeong-Hee Choi (Hallym University Dongtan Sacred Heart Hospital, Hwaseong, Korea); Young-Hee Nam (Dong-A University, Busan, Korea); Sun-Young Yoon (University of Konkuk, Chungju, Korea); Hyun Jung Jin (Yeungnam University, Daegu, Korea); Min-Suk Yang (Seoul National University Boramae Medical Center, Seoul, Korea); Jaechun Lee (Jeju National University, Jeju, Korea); Hye-Kyung Park (Pusan National University, Busan, Korea); Gyu Young Hur (Korea University, Seoul, Korea); Hee-Kyoo

Kim (Kosin University, Busan, Korea); and Sang Ha Kim (Yonsei University, Wonju, Korea).

References

1. Adkinson F, Bochner B, Burks W, Busse W, Holgate S, Lemanske R, O’Hehir R. Middleton’s Allergy - Principles and Practice. 8th Edn. Elsevier Science, 2013; p.812.
2. Global Initiative for Asthma. Global strategy for asthma management and prevention [Internet]. [place unknown]: Global Initiative for Asthma; 2014 [updated 2015 Aug 12; cited 2017 Nov 12]. Available from: <http://www.ginasthma.org/>.
3. Kauppi P, Kupiainen H, Lindqvist A, Tammilehto L, Kilpelainen M, Kinnula VL, et al. Overlap syndrome of asthma and COPD predicts low quality of life. *J Asthma* 2011;48:279-85.
4. Marsh SE, Travers J, Weatherall M, Williams MV, Aldington S, Shirtcliffe PM, et al. Proportional classifications of COPD phenotypes. *Thorax* 2008;63:761-7.
5. Cosio BG, Soriano JB, Lopez-Campos JL, et al. Defining the Asthma-COPD Overlap Syndrome in a COPD Cohort. *Chest* 2016;149: 45-52
6. Hardin M, Silverman EK, Barr RG, et al. The clinical features of the overlap between COPD and asthma. *Respir Res* 2011; 12: 127.
7. Barrecheguren M, Esquinas C, Miravittles M. The asthma-chronic obstructive pulmonary disease overlap syndrome (ACOS): opportunities and challenges. *Curr Opin Pulm Med* 2015;21:74-9.
8. Rhee CK. Phenotype of asthma-chronic obstructive pulmonary disease overlap syndrome. *Korean J Intern Med* 2015;30:443-9.

- Accepted Article
9. Cosio BG, Soriano JB, Lopez-Campos JL, Calle-Rubio M, Soler-Cataluna JJ, de-Torres JP, et al. Defining the Asthma-COPD overlap syndrome in a COPD cohort. *Chest* 2016;149:45-52
 10. Soler-Cataluna JJ, Cosio B, Izquierdo JL, Lopez-Campos JL, Marin JM, Agüero R, et al. Consensus document on the overlap phenotype COPD-asthma in COPD. *Arch Bronconeumol* 2012;48:331-7.
 11. Lee HY, Kang JY, Yoon HK, Lee SY, Kwon SS, Kim YK, et al. Clinical characteristics of asthma combined with COPD feature. *Yonsei Med J* 2014;55:980-6.
 12. Fu JJ, Gibson PG, Simpson JL, McDonald VM. Longitudinal changes in clinical outcomes in older patients with asthma, COPD and asthma-COPD overlap syndrome. *Respiration* 2014;87:63-74.
 13. de Marco R, Marcon A, Rossi A, Anto JM, Cerveri I, Gislason T, et al. Asthma, COPD and overlap syndrome: a longitudinal study in young European adults. *Eur Respir J* 2015;46:671-9.
 14. Gibson PG, Simpson JL. The overlap syndrome of asthma and COPD: what are its features and how important is it? *Thorax* 2009;64:728-35.
 15. Miravittles M, Soriano JB, Ancochea J, Munoz L, Duran-Tauleria E, Sanchez G, et al. Characterisation of the overlap COPD-asthma phenotype. Focus on physical activity and health status. *Respir Med* 2013;107:1053-60.
 16. Menezes AM, Montes de Oca M, Perez-Padilla R, Nadeau G, Wehrmeister FC, Lopez-Varela MV, et al. Increased risk of exacerbation and hospitalization in subjects with an overlap phenotype: COPD-asthma. *Chest* 2014;145:297-304.
 17. Kim TB, Park CS, Bae YJ, Cho YS, Moon HB. Factors associated with severity and exacerbation of asthma: a baseline analysis of the cohort for reality and evolution of adult asthma in Korea (COREA). *Ann Allergy Asthma Immunol* 2009;103:311-7.

- Accepted Article
18. Kim TB, Jang AS, Kwon HS, Park JS, Chang YS, Cho SH, et al. Identification of asthma clusters in two independent Korean adult asthma cohorts. *Eur Respir J* 2013;41:1308-14.
 19. Haldar P, Pavord ID, Shaw DE, Berry MA, Thomas M, Brightling CE, et al. Cluster analysis and clinical asthma phenotypes. *Am J Respir Crit Care Med* 2008;178:218-24.
 20. Sin DD, Miravittles M, Mannino DM, Soriano JB, Price D, Celli BR, et al. What is asthma-COPD overlap syndrome? Towards a consensus definition from a round table discussion. *Eur Respir J* 2016;48:664-73.
 21. Gibson PG, McDonald VM. Asthma-COPD overlap 2015: now we are six. *Thorax* 2015;70:683-91.
 22. Bateman ED, Reddel HK, van Zyl-Smit RN, Agustí A. The asthma-COPD overlap syndrome: towards a revised taxonomy of chronic airways diseases? *The Lancet Respiratory Medicine* 2015;3:719-28.
 23. de Marco R, Pesce G, Marcon A, Accordini S, Antonicelli L, Bugiani M, et al. The coexistence of asthma and chronic obstructive pulmonary disease (COPD): prevalence and risk factors in young, middle-aged and elderly people from the general population. *PLoS One* 2013;8:e62985.
 24. Soriano JB, Davis KJ, Coleman B, Visick G, Mannino D, Pride NB. The Proportional Venn Diagram of Obstructive Lung Disease *Chest* 2003;124:474-81.
 25. Tkacova R, Dai DL, Vonk JM, Leung JM, Hiemstra PS, van den Berge M, et al. Airway hyperresponsiveness in chronic obstructive pulmonary disease: A marker of asthma-chronic obstructive pulmonary disease overlap syndrome? *J Allergy Clin Immunol* 2016;138:1571-9 e10.
 26. Iwamoto H, Gao J, Koskela J, Kinnula V, Kobayashi H, Laitinen T, et al. Differences in plasma and sputum biomarkers between COPD and COPD-asthma overlap. *Eur Respir J* 2014;43:421-9.

- Accepted Article
27. Tamada T, Sugiura H, Takahashi T, Matsunaga K, Kimura K, Katsumata U, et al. Biomarker-based detection of asthma-COPD overlap syndrome in COPD populations. *Int J Chron Obstruct Pulmon Dis* 2015;10:2169-76.
 28. Gelb AF, Yamamoto A, Verbeken EK, Nadel JA. Unraveling the Pathophysiology of the Asthma-COPD Overlap Syndrome: Unsuspected Mild Centrilobular Emphysema Is Responsible for Loss of Lung Elastic Recoil in Never Smokers With Asthma With Persistent Expiratory Airflow Limitation. *Chest* 2015;148:313-20.
 29. Hardin M, Cho M, McDonald ML, Beaty T, Ramsdell J, Bhatt S, et al. The clinical and genetic features of COPD-asthma overlap syndrome. *Eur Respir J* 2014;44:341-50.
 30. Smolonska J, Koppelman GH, Wijmenga C, Vonk JM, Zanen P, Bruinenberg M, et al. Common genes underlying asthma and COPD? Genome-wide analysis on the Dutch hypothesis. *Eur Respir J* 2014;44:860-72.
 31. Barnes PJ. Asthma-COPD Overlap. *Chest* 2016;149:7-8.
 32. Postma DS, Rabe KF. The Asthma-COPD Overlap Syndrome. *N Engl J Med* 2015;373:1241-9.

Table 1. Baseline characteristics of the study patients

	“Only” asthma	ACOS	p-value
Patients, n	731	228	
Age, years	46.19 ± 15.75 (n = 731)	57.56 ± 13.27 (n = 228)	<0.001
Age at diagnosis, years	42.30 ± 16.90 (n = 651)	50.67 ± 16.37 (n = 201)	<0.001
Age at symptom onset, years	38.76 ± 17.22 (n = 723)	46.50 ± 18.17 (n = 226)	<0.001
Males	271/731 (37.1%)	143/228 (62.7%)	<0.001
BMI, kg/m²	23.99 ± 3.55 (n = 725)	24.12 ± 3.11 (n = 227)	0.610
Smoking history			
Never/ Ex/ Current	57.5%/ 28.3%/ 14.3% (n = 722)	36.6%/ 49.8%/ 13.7% (n = 227)	<0.001
Pack-years	4.63 ± 10.3 (n = 653)	19.66 ± 25.45 (n = 212)	<0.001
Family history	339/731 (46.4%)	94/228 (41.2%)	0.173
Atopy test positive	285/491 (58.0%)	46/124 (37.1%)	<0.001
Blood total IgE, IU/mL	389.09 ± 617.78 (n = 381)	367.73 ± 548.46 (n = 127)	0.729
Blood eosinophil, count	383.29 ± 400.66 (n = 627)	343.93 ± 383.27 (n = 200)	0.222
Blood WBC, x10³ /μL	7.16 ± 2.16 (n = 627)	7.47 ± 2.31 (n = 204)	0.075
CRP, mg/dL	0.42 ± 1.51 (n = 353)	0.51 ± 1.82 (n = 137)	0.606
Sputum eosinophil, %	13.52 ± 16.94 (n = 150)	15.15 ± 21.30 (n = 55)	0.611
Sputum neutrophil, %	28.64 ± 28.95 (n = 155)	24.37 ± 24.58 (n = 56)	0.326
Duration of asthma, months	47.81 ± 76.44 (n = 651)	75.08 ± 100.36 (n = 201)	<0.001
Symptom duration, months	89.69 ± 114.89 (n = 723)	133.25 ± 160.66 (n = 226)	<0.001
Pre-bronchodilator FEV1, %	84.64 ± 18.41 (n = 711)	63.17 ± 19.42 (n = 222)	<0.001
Pre-bronchodilator FEV1, L	2.42 ± 0.84 (n = 711)	1.70 ± 0.70 (n = 222)	<0.001
Pre-bronchodilator FVC, %	90.47 ± 17.99 (n = 711)	82.37 ± 19.59 (n = 222)	<0.001
Pre-bronchodilator FVC, L	3.15 ± 1.00 (n = 711)	2.88 ± 1.02 (n = 192)	<0.001
Pre-bronchodilator FEV1/FVC	0.77 ± 0.09 (n = 711)	0.59 ± 0.09 (n = 222)	<0.001
Post-bronchodilator FEV1, %	87.72 ± 19.53 (n = 366)	71.02 ± 19.02 (n = 153)	<0.001
Post-bronchodilator FEV1, L	2.54 ± 0.84 (n = 370)	1.97 ± 0.72 (n = 153)	<0.001
Post-bronchodilator FVC, %	89.38 ± 17.29 (n = 366)	88.80 ± 15.56 (n = 153)	0.721

Post-bronchodilator FVC, L	3.20 ± 0.94 (n = 369)	3.20 ± 0.94 (n = 153)	0.984
Post-bronchodilator FEV1/FVC	0.79 ± 0.09 (n = 366)	0.61 ± 0.10 (n = 153)	<0.001
Change of FEV1, %	9.89 ± 12.68 (n = 368)	22.87 ± 24.58 (n = 153)	<0.001
Change of FEV1 volume, mL	188.89 ± 237.05 (n = 368)	317.97 ± 216.51 (n = 153)	<0.001
Positive bronchodilator test	133/368 (36.1%)	106/153 (69.3%)	<0.001
Very positive bronchodilator test	43/368 (11.7%)	39/153 (25.5%)	<0.001
Positive in MBPT	649/731 (88.8%)	157/228 (68.9%)	0.318
PC₂₀, mg/mL	6.07 ± 6.03 (n = 624)	3.61 ± 5.10 (n = 149)	<0.001

Data are mean ± standard error or n/N (%). BMI: body mass index; family history: asthma, allergic rhinitis, urticaria, atopic dermatitis, drug allergy; WBC: whole blood count; CRP: C-reactive protein. Change of FEV1, %: [(change of volume FEV1)/(baseline volume of FEV1)] × 100; positive bronchodilator test: fulfilled the condition FEV1 > 200 mL and > 12% increase from baseline; very positive bronchodilator test: fulfilled the ACOS diagnostic criteria of the Spanish consensus (FEV1 > 400 mL and > 15% increase from baseline); MBPT: methacholine provocation test.

Table 2. Longitudinal analysis over 1 and 3 years

	“Only” asthma	ACOS	p-value
Method 1			
Mean ± SD ΔFEV1 over 1-year, %	6.36 ± 4.26 (n = 345)	7.19 ± 5.07 (n = 135)	0.09
Mean ± SD ΔFEV1 over 3-years, %	4.82 ± 2.64 (n = 149)	5.76 ± 3.46 (n = 70)	0.045
Mean ± SD ΔFVC over 1-year, %	6.08 ± 4.90 (n = 345)	7.97 ± 6.46 (n = 135)	0.002
Mean ± SD ΔFVC over 3-years, %	4.69 ± 3.29 (n = 149)	6.13 ± 3.90 (n = 70)	0.005
Mean ± SD ΔFEV1 over 1-year, ml	175.82 ± 129.27 (n = 349)	208.55 ± 165.91 (n = 136)	0.040
Mean ± SD ΔFEV1 over 3-years, ml	133.36 ± 91.88 (n = 151)	158.10 ± 105.9 (n = 70)	0.095
Mean ± SD ΔFVC over 1-year, ml	172.89 ± 133.31 (n = 349)	261.53 ± 181.53 (n = 135)	<0.001
Mean ± SD ΔFVC over 3-years, ml	128.65 ± 84.12 (n = 151)	195.33 ± 100.61 (n = 70)	<0.001
Mean ± SD Δ(FEV1/FVC) over 1-year	0.04 ± 0.02 (n = 349)	0.04 ± 0.03 (n = 136)	0.004
Mean ± SD Δ(FEV1/FVC) over 3-years	0.03 ± 0.01 (n = 151)	0.03 ± 0.02 (n = 70)	0.005
Method 2			
Mean ± SD {(MAX)-(MIN)}FEV1 over 1-year, %	15.92 ± 11.29 (n = 345)	18.17 ± 12.57 (n = 135)	0.058
Mean ± SD {(MAX)-(MIN)}FEV1 over 3-years, %	22.25 ± 11.58 (n = 149)	23.85 ± 11.29 (n = 70)	0.336
Mean ± SD {(MAX)-(MIN)}FVC over 1-year, %	15.98 ± 11.64 (n = 345)	19.19 ± 13.91 (n = 135)	0.018
Mean ± SD {(MAX)-(MIN)}FVC over 3-years, %	22.60 ± 13.64 (n = 149)	26.19 ± 15.81 (n = 70)	0.086
Mean ± SD {(MAX)-(MIN)}FEV1 over 1-year, ml	449.48 ± 364.64 (n = 349)	518.90 ± 419.99 (n = 136)	0.092
Mean ± SD {(MAX)-(MIN)}FEV1 over 3-years, ml	616.69 ± 415.65 (n = 151)	649.86 ± 372.18 (n = 70)	0.569
Mean ± SD {(MAX)-(MIN)}FVC over 1-year, ml	441.95 ± 357.38 (n = 349)	636.18 ± 434.69 (n = 136)	<0.001
Mean ± SD {(MAX)-(MIN)}FVC over 3-years, ml	605.36 ± 412.98 (n = 151)	817.29 ± 410.45 (n = 70)	<0.001
Method 3	β	Standard error	p-value
Lung function decline FEV1 over 1-year, %	0.44	0.26	0.080
Lung function decline FEV1 over 3-year, %	0.17	0.10	0.082
Lung function decline FVC over 1-year, %	0.83	0.27	< 0.001

Lung function decline FVC over 3-year, %	0.17	0.10	0.100
Lung function decline FEV1/FVC over 1-year, %	-0.001	0.01	0.840
Lung function decline FEV1/FVC over 3-year, %	0.001	0.0	0.190
ACT score			
Change of ACT score over 1-year	-0.029	0.08	0.730
Change of ACT score over 3-year	0.004	0.03	0.88
Exacerbation rate	“Only” Asthma	ACOS	
Acute exacerbation rate over 1-year	1.61 ± 2.74 (N=62)	2.38 ± 1.91 (N=21)	< 0.001
Acute exacerbation rate over 3-year	0.73 ± 0.59 (N=42)	0.65 ± 0.79 (N=11)	0.070
Data are presented as mean ± SE, n/N (%).			

Table 3. Comparison of baseline demographic and clinical characteristics between patients with ACOS and severe asthma

	Severe asthma	ACOS	p-value
Patients, n	198	228	
Age, years	43.76 ± 15.24 (n = 198)	57.56 ± 13.27 (n = 228)	<0.001
Age at diagnosis, years	39.27 ± 16.24 (n = 174)	50.67 ± 16.37 (n = 201)	<0.001
Age at symptom onset, years	36.28 ± 16.65 (n = 197)	46.50 ± 18.17 (n = 226)	<0.001
Males	78/198 (39.4%)	143/228 (62.7%)	<0.001
BMI, kg/m²	24.20 ± 3.55 (n = 196)	24.12 ± 3.11 (n = 227)	0.81
Smoking history			
Never/ Ex/ Current	53.1%/ 28.6%/ 18.4% (n = 196)	36.6%/ 49.8%/ 13.7% (n = 227)	<0.001
Pack-years	5.55 ± 11.15 (n = 176)	19.66 ± 25.45 (n = 212)	<0.001
Family history	108/198 (54.5%)	94/228 (41.2%)	0.01
Atopy test positive	75/119 (63%)	46/124 (37.1%)	<0.001
Blood total IgE, IU/mL	399.27 ± 585.51 (n = 118)	367.73 ± 548.46 (n = 127)	0.66
Blood eosinophil, count	358.67 ± 337.83 (n = 165)	343.93 ± 383.27 (n = 200)	0.70
Blood WBC, x10³ /μL	7.37 ± 2.17 (n = 165)	7.47 ± 2.31 (n = 204)	0.66
CRP, mg/dL	0.33 ± 0.52 (n = 95)	0.51 ± 1.82 (n = 137)	0.28
Sputum eosinophil, %	8.15 ± 9.80 (n = 24)	15.15 ± 21.30 (n = 55)	0.05
Sputum neutrophil, %	32.98 ± 33.81 (n = 26)	24.37 ± 24.58 (n = 56)	0.25
Duration of asthma, months	58.17 ± 82.13 (n = 174)	75.08 ± 100.36 (n = 201)	<0.001
Symptom duration, months	89.20 ± 107.17 (n = 197)	133.25 ± 160.66 (n = 226)	<0.001
Pre-bronchodilator FEV1, %	84.24 ± 17.42 (n = 192)	63.17 ± 19.42 (n = 222)	<0.001
Pre-bronchodilator FEV1, L	2.51 ± 0.87 (n = 192)	1.70 ± 0.70 (n = 222)	<0.001
Pre-bronchodilator FVC, %	87.38 ± 17.27 (n = 192)	82.37 ± 19.59 (n = 222)	0.01
Pre-bronchodilator FVC, L	3.20 ± 1.07 (n = 192)	2.88 ± 1.02 (n = 192)	0.002
Pre-bronchodilator FEV1/FVC	0.79 ± 0.09 (n = 192)	0.59 ± 0.09 (n = 222)	<0.001

Post-bronchodilator FEV1, %	87.37 ± 16.17 (n = 128)	71.02 ± 19.02 (n = 153)	<0.001
Post-bronchodilator FEV1, L	2.65 ± 0.88 (n = 192)	1.97 ± 0.72 (n = 153)	<0.001
Post-bronchodilator FVC, %	87.34 ± 14.98 (n = 128)	88.80 ± 15.56 (n = 153)	0.42
Post-bronchodilator FVC, L	3.28 ± 1.03 (n = 192)	3.20 ± 0.94 (n = 153)	0.507
Post-bronchodilator FEV1/FVC	0.81 ± 0.08 (n = 130)	0.61 ± 0.10 (n = 153)	<0.001
Change of FEV1, %	9.89 ± 13.15 (n = 131)	22.87 ± 24.58 (n = 153)	<0.001
Change of FEV1 volume, mL	189.62 ± 245.28 (n = 131)	317.97 ± 216.51 (n = 153)	<0.001
Positive bronchodilator test	46/131 (35.1%)	106/153 (69.3%)	<0.001
Very positive bronchodilator test	15/131 (11.5%)	39/153 (25.5%)	<0.001
Positive in MBPT	176/198 (88.9%)	157/228 (68.9%)	0.318
PC₂₀, mg/mL	6.03 ± 6.35 (n = 172)	3.61 ± 5.10 (n = 149)	<0.001

Data are mean ± standard error, n/N (%). BMI: body mass index; family history: asthma, allergic rhinitis, urticaria, atopic dermatitis, drug allergy; WBC: whole blood count; CRP: C-reactive protein; Change of FEV1, %: [(change of volume FEV1)/(baseline volume of FEV1)] × 100; positive bronchodilator test: fulfilled the condition FEV1 > 200 mL and > 12% increase from baseline; very positive bronchodilator test: fulfilled the ACOS diagnostic criteria of the Spanish consensus (FEV1 > 400 mL and > 15% increase from baseline); MBPT: methacholine provocation test

Table 4. Comparison of the clinical characteristics between the cluster subgroups

	Group 1	Group 2	Group 3	p-value
Patients, n	202	182	97	
Age, years	31.27 ± 9.16 (n = 202)	56.57 ± 8.71 (n = 182)	53.53 ± 12.7 (n = 97)	<0.001
Age at diagnosis, years	26.44 ± 10.61 (n = 202)	53.39 ± 9.71 (n = 182)	48.43 ± 14.51 (n = 97)	<0.001
Age at symptom onset, years	24.33 ± 10.56 (n = 202)	46.67 ± 15.45 (n = 182)	43.31 ± 16.6 (n = 97)	<0.001
Males	101/202 (50%)	55/182 (30.2%)	64/97 (66%)	<0.001
BMI, kg/m²	23.41 ± 3.56 (n = 199)	24.47 ± 3.2 (n = 181)	23.35 ± 2.67 (n = 96)	<0.001
Smoking history				
Never/ Ex/ Current	49%/ 26.7%/ 24.3% (n = 202)	65.9%/ 30.2%/ 3.9% (n = 182)	26.8%/ 59.8%/ 13.4% (n = 97)	<0.001
Pack-years	3.97 ± 8.74 (n = 181)	5.29 ± 22.61 (n = 164)	18.15 ± 21.6 (n = 91)	<0.001
Family history	112/202 (55.4%)	94/182 (39%)	44/97 (45.4%)	0.01
Atopy test positive	164/202 (81.2%)	52/182 (28.6%)	52/97 (30.9%)	<0.001
Blood total IgE, IU/mL	513.15 ± 743.32 (n = 123)	264.9 ± 347.5 (n = 96)	471.12 ± 607.2 (n = 55)	0.01
Blood eosinophil, count	414.21 ± 359.89 (n = 202)	286.51 ± 238.06 (n = 182)	555.1 ± 668.3 (n = 97)	<0.001
Blood WBC, x10³ /μL	7.31 ± 2.08 (n = 202)	7.02 ± 2.24 (n = 182)	7.7 ± 2.63 (n = 97)	0.05
CRP, mg/dL	0.38 ± 1.32 (n = 116)	0.35 ± 0.77 (n = 102)	0.49 ± 0.91 (n = 64)	0.68
Sputum eosinophil, %	14.91 ± 19.9 (n = 31)	17.44 ± 22.96 (n = 33)	14.72 ± 17.4 (n = 25)	0.84
Sputum neutrophil, %	35.41 ± 29.57 (n = 35)	24.35 ± 26.52 (n = 34)	29.53 ± 31.69 (n = 25)	0.29
Duration of asthma, months	57.95 ± 99.97 (n = 202)	38.09 ± 67.76 (n = 182)	61.11 ± 91.7 (n = 97)	0.04
Symptom duration, months	83.36 ± 102.7 (n = 202)	118.7 ± 164.54 (n = 182)	121.3 ± 151.55 (n = 96)	0.02
Pre-bronchodilator FEV1, %	86.81 ± 14.1 (n = 202)	86.21 ± 15.36 (n = 182)	50.96 ± 14.02 (n = 97)	<0.001
Pre-bronchodilator FEV1, L	3.05 ± 0.75 (n = 202)	2.08 ± 0.56 (n = 182)	1.49 ± 0.50 (n = 97)	<0.001
Pre-bronchodilator FVC, %	94.98 ± 15.83 (n = 202)	93.39 ± 16.84 (n = 182)	72.54 ± 16.99 (n = 97)	<0.001
Pre-bronchodilator FVC, L	3.87 ± 0.97 (n = 202)	2.82 ± 0.77 (n = 182)	2.67 ± 0.82 (n = 97)	<0.001
Pre-bronchodilator FEV1/FVC	0.79 ± 0.09 (n = 202)	0.74 ± 0.07 (n = 182)	0.56 ± 0.09 (n = 97)	<0.001

Post-bronchodilator FEV1, %	89.64 ± 14.06 (n = 93)	93.28 ± 15.28 (n = 68)	58.93 ± 16.13 (n = 69)	<0.001
Post-bronchodilator FEV1, L	3.12 ± 0.81 (n = 94)	2.31 ± 0.58 (n = 69)	1.82 ± 0.56 (n = 71)	<0.001
Post-bronchodilator FVC, %	90.86 ± 13.4 (n = 93)	95.46 ± 15.95 (n = 68)	79.92 ± 17.14 (n = 69)	<0.001
Post-bronchodilator FVC, L	3.79 ± 0.93 (n = 93)	2.97 ± 0.73 (n = 69)	3.11 ± 0.82 (n = 71)	<0.001
Post-bronchodilator FEV1/FVC	0.82 ± 0.08 (n = 93)	0.78 ± 0.07 (n = 69)	0.59 ± 0.1 (n = 71)	<0.001
Change of FEV1, %	9.45 ± 10.7 (n = 94)	8.5 ± 9.9 (n = 69)	29.53 ± 22.7 (n = 71)	<0.001
Change of FEV1 volume, mL	238.4 ± 256.9 (n = 94)	161.74 ± 165.32 (n = 69)	386.6 ± 250.24 (n = 71)	<0.001
Positive bronchodilator test	37/94 (39.4%)	19/69 (27.5%)	59/71 (83.1%)	<0.001
Very positive bronchodilator test	18/94 (19.1%)	6/69 (8.7%)	23/71 (32.4%)	<0.001
Positive in MBPT	185/202 (91.6%)	173/182 (95.1%)	49/97 (50.5%)	
PC₂₀, mg/mL	5.46 ± 6.14 (n = 173)	7.03 ± 6.47 (n = 165)	2.9 ± 3.95 (n = 47)	<0.001

Data are mean ± standard error and n/N (%). BMI: body mass index; family history: asthma, allergic rhinitis, urticaria, atopic dermatitis, drug allergy; WBC: whole blood count; CRP: C-reactive protein; Change of FEV1, %: [(change of volume FEV1)/(baseline volume of FEV1)] × 100; positive bronchodilator test: fulfilled the condition FEV1 > 200 mL and > 12% increase from baseline; very positive bronchodilator test: fulfilled the ACOS diagnostic criteria of the Spanish consensus (FEV1 > 400 mL and > 15% increase from baseline); MBPT: methacholine provocation test

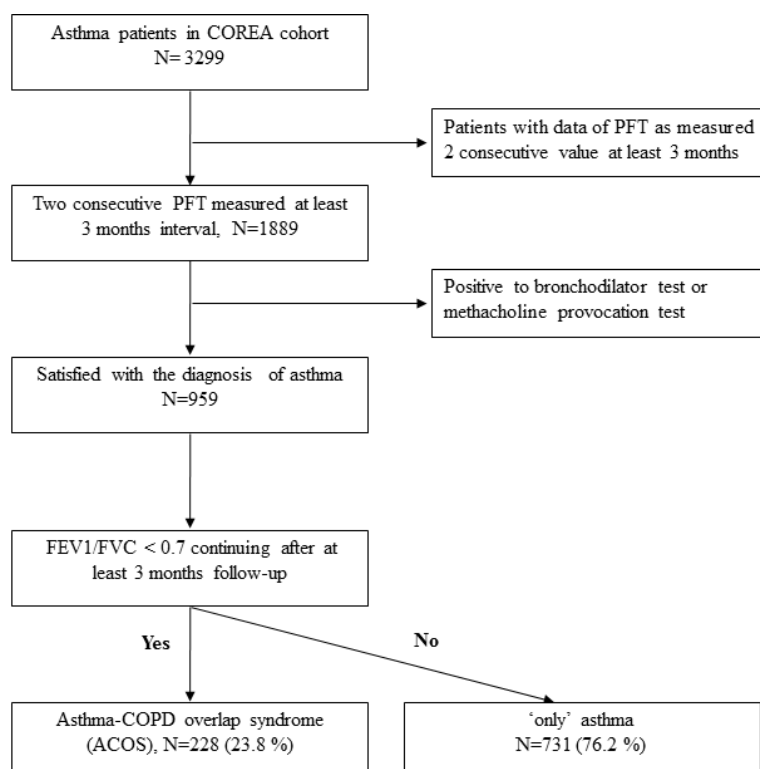
Figure legends

Figure 1. Flowchart summarizing determination of the asthma-chronic obstructive pulmonary disease (COPD) overlap syndrome (ACOS) definition

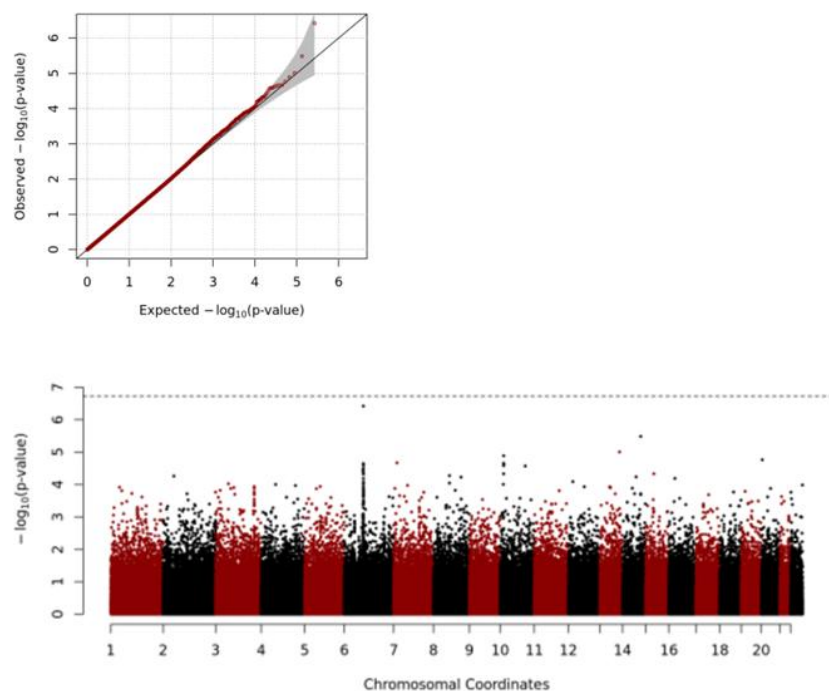
ACOS was defined by sequentially among Korean asthma cohort (COREA) patients with consecutive pulmonary function results.

Figure 2. Graphical summary of the association results between asthma and asthma-chronic obstructive pulmonary disease (COPD) overlap syndrome (ACOS) (Manhattan plot)

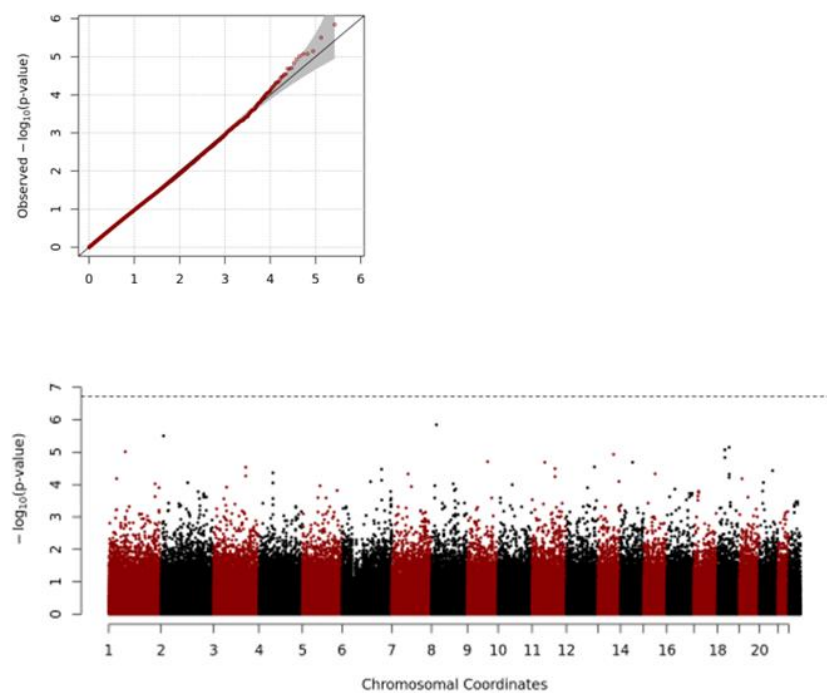
Plot of P values by chromosome for asthma and ACOS. (A) Comparison of asthma and normal subjects (B) Comparison of ACOS and normal subjects (C) Comparison of asthma and ACOS.



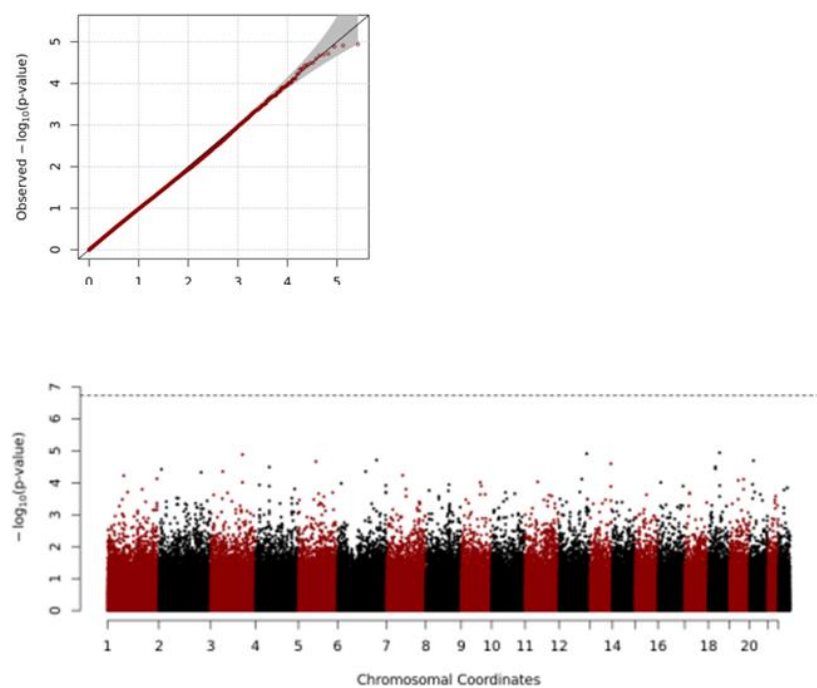
A.



B.



C.



Online Supplements

Methods

Study patients and measurements

Demographic parameters, body mass index (BMI), complete blood cell count, total serum IgE levels, and serum eosinophil counts were recorded. An induced sputum analysis was performed for these patients to determine the airway inflammatory cell counts. The atopic response was measured by skin prick tests to 12 common allergens (Allegopharma, Reinbek, Germany). The history of hospital visits due to exacerbation was also recorded. The pulmonary function test was implemented in as many 3-month intervals as possible and included the forced expiratory volume in 1 sec (FEV1), forced vital capacity (FVC) and the FEV1/FVC ratio. Patients were administered a bronchodilator response test or methacholine provocation test. Blood samples were obtained to determine the number and percentage of blood eosinophils and to quantify total immunoglobulin E.

Genome-wide association study (GWAS)

Genome-wide single nucleotide polymorphism (SNP) genotyping was performed using approximately 200 ng of genomic DNA from the patient's peripheral blood lymphocytes. Call rate (>98%) and minor allele frequency (MAF > 0.05) were measured as quality controls. A total of 267,429 SNPs were passed through strict quality control measures. The principal component score was used to compare and analyze the control group (N = 5,915) with three times the total number of patients (N = 1,433).

Supplement E1. Skin prick test results

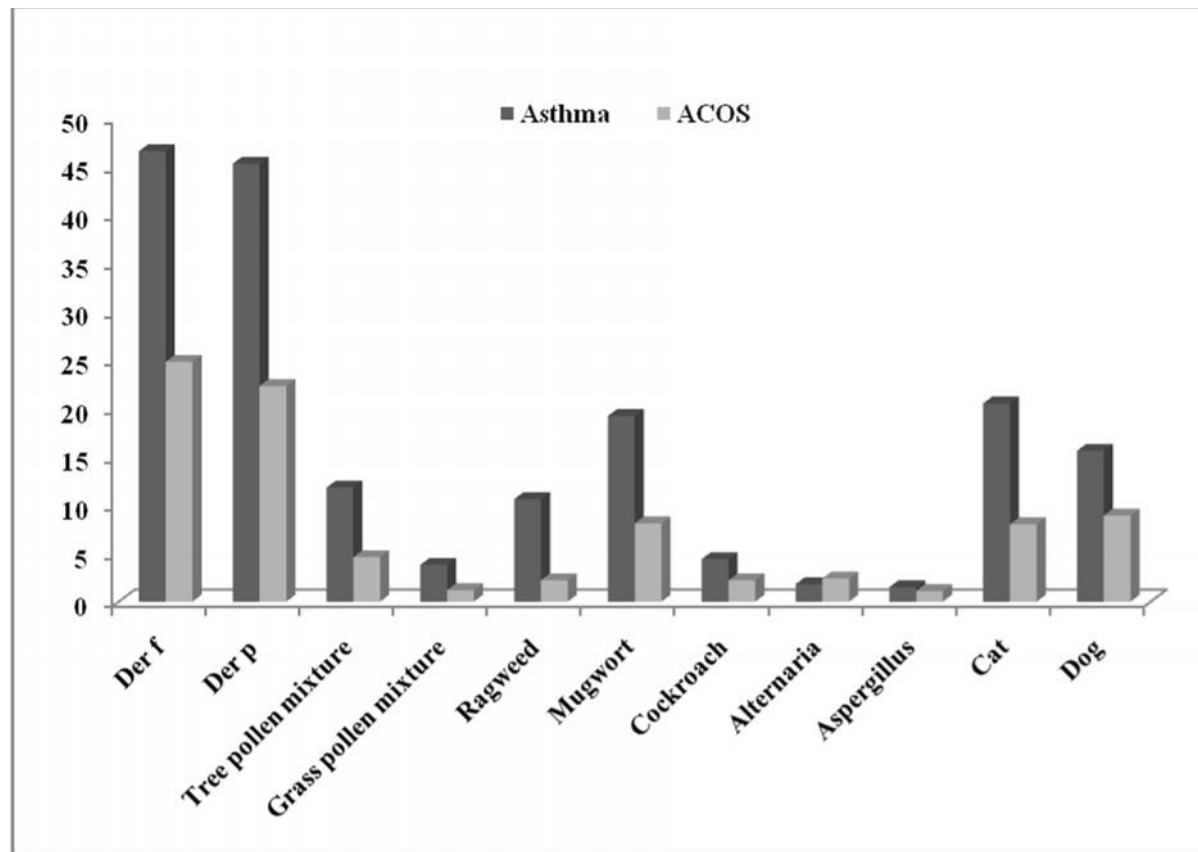
	“Only” asthma	ACOS	p-value
Subjects, n	731	228	
Atopy test positive	285/491 (58.0%)	46/124 (37.1%)	<0.001
<i>Der f</i>	204/450 (45.3%)	25/112 (22.3%)	
<i>Der p</i>	208/446 (46.6%)	29/117 (24.8%)	
<i>Tree pollen mixture</i>	44/374 (11.8%)	4/88 (4.6%)	
<i>Grass pollen mixture</i>	13/344 (3.8%)	1/82 (1.2%)	
<i>Ragweed</i>	38/360 (10.6%)	1/46 (2.2%)	
<i>Mugwort</i>	71/369 (19.2%)	7/87 (8.0%)	
<i>Cockroach</i>	16/367 (4.4%)	2/91 (2.2%)	
<i>Alternaria</i>	6/338 (1.8%)	2/82 (2.4%)	
<i>Aspergillus</i>	5/344 (1.5%)	1/88 (1.1%)	
<i>Cat</i>	79/386 (20.5%)	7/88 (8.0%)	

Dog

60/385 (15.6%)

8/90 (8.9%)

Der f: *Dermatophagoides farinae*; Der p: *Dermatophagoides pteronyssinus*



Supplement E2. The longitudinal analysis of inhaled corticosteroid (ICS) dose

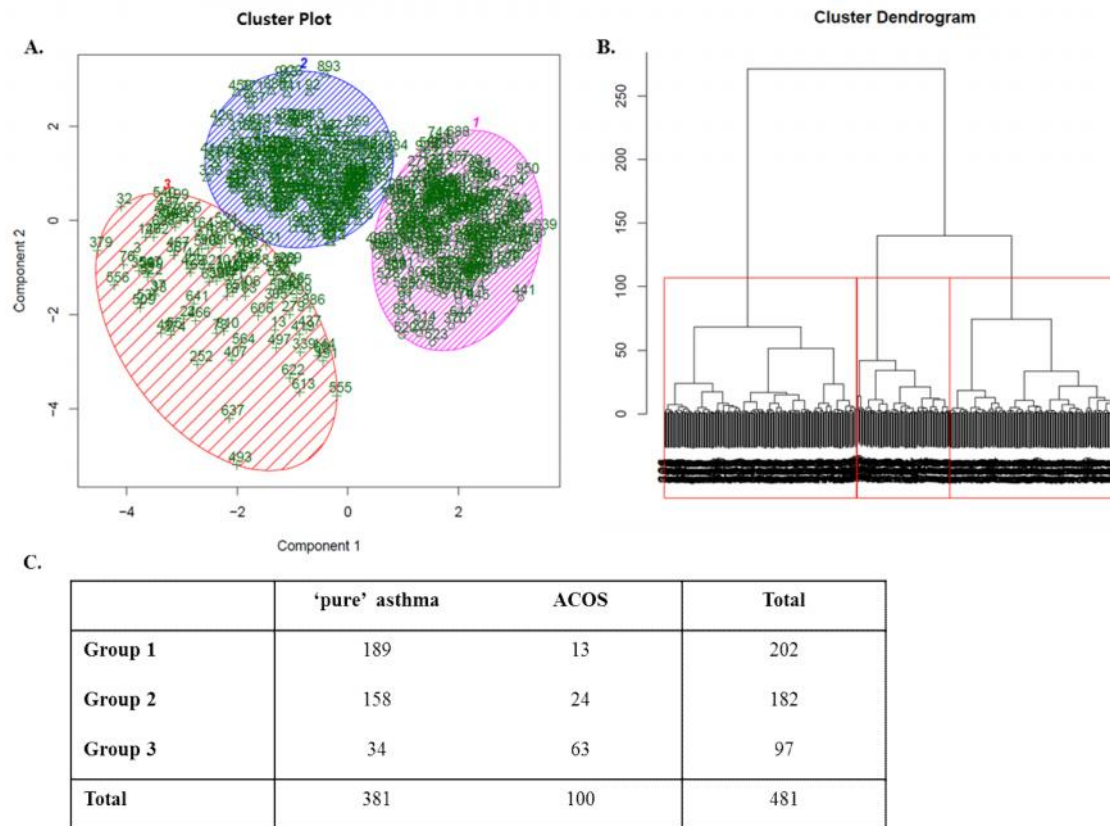
	“Only” asthma	ACOS	p-value
Average ICS dose over 1 year, µg/day	583.11 ± 332.92 (n = 230)	657.67 ± 352.34 (n = 105)	0.063
Average ICS dose over 3 years, µg/day	601.87 ± 402.71 (n = 110)	670.12 ± 397.89 (n = 52)	0.313
Data are mean ± standard error or n/N (%);ICS dose: converted to equivalent dose of budesonide.			

Supplement E3. The longitudinal analysis of ICS dose and exacerbation

	Severe asthma	ACOS	p-value
Experience of acute exacerbation over a 1-year	12/81 (14.8%)	17/109 (15.6%)	0.99
Experience of acute exacerbation over a 3-years	4/38 (10.5%)	17/52 (32.7%)	0.03
Average ICS dose over a 1-year, µg/day	792.23 ± 275.73 (n = 94)	657.67 ± 352.34 (n = 105)	<0.001
Average ICS dose over a 3-years, µg/day	958.4 ± 338.91 (n = 36)	670.12 ± 397.89 (n = 52)	<0.001

Supplement E4. Cluster analysis results and patient composition table

The results of a cluster analysis using seven variables [age, asthma onset age, smoking status, atopy, forced expiratory volume in 1 sec (FEV1), FEV1/forced vital capacity (FVC) and serum blood eosinophil count] are shown. Cluster plot, dendrogram (A and B), and the patient composition table (C) are shown.



Supplement E5. Top 10 single nucleotide polymorphisms (SNPs) associated with asthma alone

SNP ID	Chromosome	Gene	Location	Variation	MAF	HWE	OR	P value
rs2858892	6	---	downstream	G>A	0.347	0.0016	0.7466	3.80E-07
rs2402306	14	UNC79	intron	T>G	0.2697	0.7415	1.255	3.27E-06
rs7338473	13	---	upstream	A>G	0.3706	0.3025	1.22	9.85E-06
rs1775551	10	---	downstream	A>C	0.2286	0.79	1.249	1.29E-05
rs6137337	20	---	intron	A>G	0.4855	0.4219	0.8238	1.72E-05
rs4720756	7	---	intron	A>G	0.3272	0.01883	1.223	2.14E-05
rs1149911	10	---	downstream	T>G	0.2394	0.8718	1.239	2.20E-05
rs2213568	6	HLA-DQA2	intron	C>A	0.3158	0.2322	0.7801	2.25E-05
rs1912584	10	---	downstream	T>C	0.4068	0.4959	1.209	2.38E-05
rs2213571	6	HLA-DQB2	downstream	G>C	0.2482	0.2561	0.7468	2.56E-05

Supplement E6. Top 10 single nucleotide polymorphisms (SNPs) associated with asthma-chronic obstructive pulmonary disease (COPD) overlap syndrome (ACOS)

SNP ID	Chromosome	Gene	Location	Variation	MAF	HWE	OR	<i>P</i> value
rs117733692	8	MSRA	intron	C>A	0.055	0.012	3.332	1.43E-06
rs3772010	2	RNF144A	intron	T>C	0.386	0.567	0.385	3.14E-06
rs11665213	18	---	intron	C>T	0.183	0.100	2.232	7.12E-06
rs1677005	18	TAF4B	downstream	T>C	0.073	0.068	2.618	8.40E-06
rs35614679	18	TAF4B	intron	G>A	0.050	0.276	2.996	8.46E-06
rs4601609	1	LINC01135	intron	T>C	0.160	0.177	2.258	9.69E-06
rs2149063	13	GPC5	intron	T>C	0.058	0.473	2.877	1.17E-05
rs1786253	18	TAF4B	downstream	C>G	0.052	0.793	2.932	1.46E-05
rs12336157	9	---	upstream	T>G	0.114	0.337	2.332	1.96E-05
rs34186721	14	RAD51B	intron	G>A	0.069	0.012	2.585	2.06E-05

Supplement E7. Top 10 single nucleotide polymorphisms (SNPs) associated with asthma-chronic obstructive pulmonary disease (COPD) overlap syndrome (ACOS) compared to asthma alone

SNP ID	Chromosome	Gene	Location	Variation	MAF	HWE	OR	<i>P</i> value
rs11665213	18	---	intron	C>T	0.180	0.655	2.257	1.15E-05
rs7967352	12	---	upstream	T>G	0.065	0.663	2.904	1.23E-05
rs1046706	3	SLC9A9	UTR-3	G>T	0.095	0.646	2.591	1.31E-05
rs78276030	6	---	intron	A>G	0.067	0.087	2.728	1.93E-05
rs8115801	20	PAK7	intron	C>T	0.158	0.488	2.304	2.02E-05
rs7732758	5	LHFPL2	intron	T>C	0.434	0.106	2.055	2.15E-05
rs4773144	13	COL4A2	intron	A>G	0.376	0.572	2.063	2.53E-05
rs35614679	18	TAF4B	intron	G>A	0.053	1.000	3.008	3.17E-05
rs6837266	4	ATP10D	upstream	G>C	0.101	0.771	2.497	3.22E-05
rs1786253	18	TAF4B	downstream	C>G	0.053	0.794	3.026	3.52E-05