Incident asthma and *Mycoplasma pneumoniae*: A nationwide cohort study

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Background: Previous studies investigating the relationship between *Mycoplasma pneumoniae* and incident asthma in the general population have been inconclusive.
Objective: We conducted a nationwide cohort study to clarify this relationship.
Methods: Using the National Health Insurance Research Database of Taiwan, we identified 1591 patients with *M pneumoniae* infection (International Classification of Diseases, Ninth Revision, Clinical Modification code 4830) given diagnoses between 2000 and 2008. We then frequency matched 6364 patients without *M pneumoniae* infection from the general population according to age, sex, and index year. Cox proportional hazards regression analysis was performed to determine the adjusted hazard ratio (aHR) of the occurrence of asthma in the *M pneumoniae* cohort compared with that in the non–*M pneumoniae* cohort.
Results: Regardless of comorbidities and the use of antibiotic or steroid therapies, patients with *M pneumonia* infection had a higher risk of incident asthma than those without it. The aHR of asthma was 3.35 (95% CI, 2.71-4.15) for the *M pneumoniae* cohort, with a significantly higher risk when patients were stratified by age, sex, follow-up time, and comorbidities, including allergic rhinitis, atopic dermatitis, or allergic conjunctivitis. Patients with *M pneumoniae* infection had a higher risk of having early-onset (age, <12 years; aHR, 2.87) and late-onset (age, ≥12 years; aHR, 3.95) asthma. The aHR was also higher within the less than 2-year follow-up in the *M pneumoniae* cohort (aHR, 4.41; 95% CI, 3.40-5.74) than in the cohort without the infection.
Conclusion: This study found that incident cases of early-onset and late-onset asthma are closely related to *M pneumoniae* infection, even in nonatopic patients. (J Allergy Clin Immunol 2016;137:1017-23.)

Key words: Mycoplasma pneumoniae, *asthma, antibiotic, steroid, cohort study*

Asthma, a condition caused by airway inflammation,[1] is characterized by airway hyperresponsiveness.[2-4] *Mycoplasma pneumoniae* infection[1] can lead to or exacerbate airway inflammation[5,6] and contribute to the initial onset of asthma[7] or refractory asthma.[7,8] Additionally, atopic sensitization and a history of asthma might be risk factors for refractory *M pneumoniae* infection.[9] The role of *M pneumoniae* infection in the pathogenesis of asthma has been a subject of continuing debate.[10,11] Case series studies[12] and small-group studies[8,11] have been conducted but with limitations. For example, case series studies generally provide weak evidence for the inference of causality because of bias, confounding factors, and the absence of substantial numbers of patients.

Drug therapy status during *M pneumoniae* infection might affect the airway, lung functionality, and risk of incident asthma.[13] Large-scale studies on the effectiveness of therapies, such as antibiotics and steroids, on severe *M pneumoniae* infection and asthma are limited,[14] and research on outpatient frequency and hospitalization resulting from *M pneumoniae* infection associated with incident asthma is scant.

In comparison with other countries, the protocol for diagnosing *M pneumoniae* infection is relatively well established in Taiwan.[15] The cohort study we describe below investigates whether *M pneumoniae* infection increases the risk of incident asthma by using a longitudinal health insurance database to determine the relationship between the 2 factors.

METHODS

Data source

Claims data were obtained from the Longitudinal Health Insurance Database (LHID 2000), a subset of the National Health Research Institutes National Health Insurance Research Database. The NHI is a universal health insurance program that was established in March 1995. The program provides health care services to more than 99% of the population of Taiwan. The LHID 2000 contains claims data collected from one million randomly selected persons between 1996 and 2011. It contains comprehensive information on the clinical visits of each insured person, such as demographic characteristics, dates of admission and discharge, discharge diagnoses, and discharge status. Data are coded according to the

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Abbreviations used
aHR: Adjusted hazard ratio
AC: Allergic conjunctivitis
AD: Atopic dermatitis
AR: Allergic rhinitis
IRR: Incidence rate ratio
LHID: Longitudinal Health Insurance Database

International Classification of Diseases, Ninth Revision, Clinical Modification (ICD-9-CM). All study patients' identification numbers are encrypted to ensure personal privacy. This study was approved to fulfill the condition for exemption by the Institutional Review Board of China Medical University (CMUH-104-REC2-115). The institutional review board also specifically waived the consent requirement.

Study population

Our study population was recruited by using population-based claims data from the LHID. Fig 1 depicts the cohort selection process. We identified 1591 new cases of *M pneumoniae* infection (ICD-9-CM code 4830)[4,16] diagnosed between the years of 2000 and 2008. The date of diagnosis was set as the index date. We then frequency matched 6364 patients without *M pneumoniae* infection at a ratio of 1:4 according to sex, age, and index year. We corrected for risk factors, including age, sex, index year, and history of comorbidities, in both cohorts. Propensity score matching was performed at a 1:4 ratio. Patients with missing data regarding date of birth or sex and patients with a history of asthma before the index date were excluded from the study.

In Taiwan the diagnosis of *M pneumoniae* infection is based on positive serologic test results[13,17] consisting of specific IgM present in a blood sample or a greater than 4-fold increase in specific IgG levels.[18] Suspected cases of infection are then confirmed through clinical examination or thoracic imaging.[19] Consensus by an infection specialist, a chest physician, and a coder with professional training is necessary for all diagnoses of *M pneumoniae* infection because of strict regulations on regular antibiotic use.[20] Therefore this well-established system was used because it minimizes misclassification.[4,16,19,20]

Outcome and potential factor measurement

Asthma is an increasingly common disease in children and adults.[17] The primary outcome this study assessed was the date of asthma diagnosis (ICD-9-CM code 493).[21] All cases were followed from the index date until the date of asthma diagnosis (December 31, 2011), withdrawal from the insurance system, death, or loss to follow-up, whichever occurred first. Comorbidities defined before the index date included hypertension (ICD-9-CM code 401-405), diabetes (ICD-9-CM code 250), anemia (ICD-9-CM code 280-285), allergic rhinitis (AR; ICD-9-CM code 477), atopic dermatitis (AD; ICD-9-CM code 691), and allergic conjunctivitis (AC; ICD-9-CM codes 37205, 37210, and 37214). The presence of AR, AD, or AC was defined as atopy syndrome.[1,22]

We adjusted for the pharmacologic treatment of *M pneumoniae* infection on the basis of antibiotic and steroid use.[12,14] Infected patients who used an antibiotic or steroid within 30 days of the index date were defined and classified into 4 subgroups, as follows: (1) without antibiotic and steroid use; (2) antibiotic use only; (3) steroid use only; and (4) both antibiotic and steroid use.

Statistical analysis

Frequencies and percentages for categorical variables, as well as means and SDs for continuous variables, were used in the χ^2 and Student *t* tests to determine the baseline distribution of the *M pneumoniae* and non–*M pneumoniae* cohorts. Incidence of asthma was stratified by sex, age group (<35, 35-65, and ≥65 years), comorbidity, and follow-up time (<2 and ≥2 years) between the *M pneumoniae* and non–*M pneumoniae* cohorts. We also used the Poisson regression model to determine the incidence rate ratio (IRR) of asthma for these variables between the 2 cohorts. The adjusted hazard ratios (aHRs) and 95% CIs of asthma were determined by using the multivariable Cox proportional hazards model while controlling for sex, age, and comorbidity.

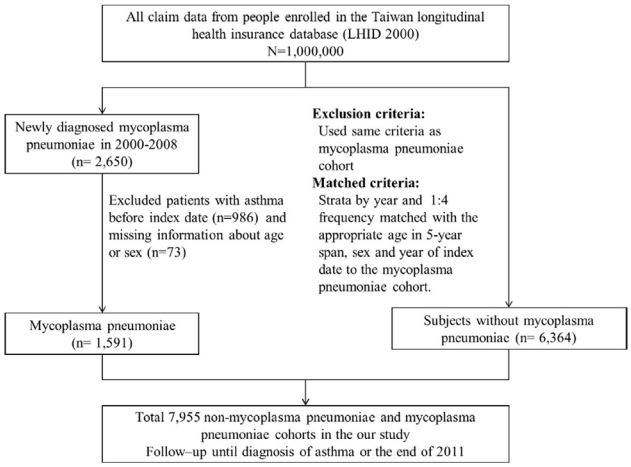

FIG 1. Flow chart presenting the selection of study patients.

To examine the short-term risk, we stratified the follow-up time into 5 periods: less than 0.5 years, 0.5 to 1 years, 1 to 1.5 years, 1.5 to 2 years, and 2 or more years. The phenotype of asthma, intermittent versus persistent, was unavailable through the database.[23] Therefore we further divided those with an incidence of asthma into 2 categories according to the patient's age at which it was diagnosed: early-onset asthma (age at asthma diagnosis, <12 years) and late-onset asthma (age at asthma diagnosis, ≥12 years).[24] We also estimated the aHR of asthma in patients with *M pneumoniae* infection according to the medical service type (ie, only outpatient, only hospitalization, and both outpatient and hospitalization)[14,25] and whether pharmacologic treatment was administered. The risk of asthma-associated *M pneumoniae* interacting with comorbidities was determined after adjusting for age and sex. Before propensity score matching, we performed logistic regression to calculate the propensity score and estimate the probability of *M pneumoniae* infection on the basis of baseline variables, including index year, sex, age, and history of comorbidities. Finally, we performed multivariable Cox proportional hazards model stratification on the matched pairs to estimate the risk of asthma between the *M pneumoniae* and non–*M pneumoniae* cohorts.

All of the statistical analyses were performed with SAS version 9.4 software (SAS Institute, Cary, NC). We also conducted a Kaplan-Meier analysis using R software (R Foundation for Statistical Computing, Vienna, Austria) to measure the cumulative asthma incidence for each study cohort. The log-rank test assessed differences between the 2 cohorts through cumulative incidence curves. A *P* value of less than .05 in a 2-tailed test was considered significant.

RESULTS

We identified 1591 patients with and 6364 patients without *M pneumoniae* infection in the period between the years 2000 and 2008. Table I shows a comparison of the demographic characteristics and comorbidities between the 2 cohorts. The cohorts showed similar distributions of sex and age; 55.1% of patients were women, and 73.1% were less than 35 years old. The *M pneumoniae* cohort had a higher prevalence of comorbidities at baseline than the non–*M pneumoniae* cohort ($P < .002$).

The cumulative incidence of asthma was significantly higher in the *M pneumoniae* cohort ($P < .0001$, log-rank test) than in the non–*M pneumoniae* cohort (see Fig E1 in this article's Online Repository at www.jacionline.org). The cumulative incidences

TABLE I. Characteristics of demographics and comorbidity history between the *M pneumoniae* and non–*M pneumoniae* cohorts

	M pneumoniae				
	No (n = 6364)		Yes (n = 1591)		
Characteristic	No.	%	No.	%	*P* value
Sex					.99
Female	3504	55.1	876	55.1	
Male	2860	44.9	715	44.9	
Age (y)					.99
<35	4652	73.1	1163	73.1	
35-65	1360	21.4	340	21.4	
≥65	352	5.53	88	5.53	
Mean (SD)*	23.7 (20.6)		23.4 (20.8)		.64
Comorbidity					
Hypertension	553	8.69	178	11.2	.0020
Diabetes	272	4.27	97	6.10	.0020
Anemia	256	4.02	100	6.29	<.0001
AR	1464	23.0	666	41.9	<.0001
AD	438	6.88	165	10.4	<.0001
AC	1589	25.0	501	31.5	<.0001

Values were calculated by using the χ^2 test, unless indicated otherwise.
*Student *t* test.

of both early-onset and late-onset asthma showed that the *M pneumoniae* cohort ($P < .0001$, log-rank test) was at greater risk (see Figs E2 and E3 in this article's Online Repository at www.jacionline.org) for asthma.

Table II shows the incidence, IRR, and aHR of asthma between the *M pneumoniae* and non–*M pneumoniae* cohorts. The overall IRR of asthma was 3.91-fold higher in the *M pneumoniae* cohort than in the non–*M pneumoniae* cohort (21.3 vs 5.45 per 1000 person years). After potential risk factors were controlled, aHRs were higher in the *M pneumoniae* cohort for asthma (aHR, 3.35; 95% CI, 2.71-4.15), early-onset asthma (aHR, 2.87; 95% CI, 2.18-3.79), and late-onset asthma (aHR, 3.95; 95% CI, 2.81-5.54). The incidence rate of asthma was higher in male than in female patients. Men in the *M pneumoniae* cohort showed a 4.08-fold increased incidence of asthma compared with men in the non–*M pneumoniae* cohort (men: aHR, 3.66; 95% CI, 2.68-4.99; women: aHR, 3.11; 95% CI, 2.32-4.17). Age-specific incidence was highest in infected patients aged 65 years or greater (43.1 per 1000 person years). However, the relative risk of asthma between the *M pneumoniae* and non–*M pneumoniae* cohorts was highest in patients aged 35 to 65 years (aHR, 3.47; 95% CI, 2.02-5.96). The comorbidity-specific analyses indicated that patients with comorbidity in the *M pneumoniae* cohort exhibited the highest risk (24.4 per 1000 person years; aHR, 3.39; 95% CI, 2.63-4.36). Among the patients without comorbidities, the risk of asthma was 3.77-fold higher in the *M pneumoniae* cohort than in the non–*M pneumoniae* cohort (95% CI, 2.58-5.52). Fig 2 shows the cumulative incidence of asthma in the 4 subgroups stratified by comorbidity, with the highest rates occurring in the patients with *M pneumoniae* infection and comorbidities ($P < .0001$, log-rank test). An association between *M pneumoniae* infection and asthma was observed in both patients without atopy syndrome (aHR, 3.57; 95% CI, 2.52-5.05) and those with atopy syndrome (aHR, 3.46; 95% CI, 2.65-4.53). The aHR for these patients decreased from 4.41 (95% CI, 3.40-5.74) within a 2-year follow-up period to 1.86 (95% CI, 1.25-2.78) after more than 2 years.

We calculated the aHR of asthma within the 2-year follow-up period because of the heightened risk in that period. Table III shows that the incidence of asthma decreased gradually, with increased follow-up time in both cohorts and the highest aHR observed in the less than 0.5-year *M pneumoniae* follow-up group (aHR, 5.99; 95% CI, 4.05-8.87).

The patients with *M pneumoniae* infection were classified into subgroups on the basis of the type of medical service they received (Table IV). The patients with *M pneumoniae* infection had a higher risk of asthma compared with those without *M pneumoniae* infection in all 3 subgroups; specifically, the risk was highest in patients with *M pneumoniae* infection who were outpatients and required hospitalization (aHR, 7.54; 95% CI, 5.21-10.9).

We also considered the risk of asthma associated with *M pneumoniae* and antibiotic or steroid use (Table V). We compared the *M pneumoniae* and non–*M pneumoniae* cohorts on the basis of the following pharmacologic treatments: without antibiotic and steroid use, antibiotic use only, steroid use only, and both antibiotic and steroid use. The respective aHRs were 2.67 (95% CI, 2.05-3.48), 2.74 (95% CI, 1.83-4.12), 6.59 (95% CI, 4.80-9.05), and 4.29 (95% CI, 2.33-7.91).

Table E1 in this article's Online Repository at www.jacionline.org shows the interactions of comorbidity and *M pneumonia*. The risk of asthma was significantly higher in the patients with both *M pneumoniae* infection and a comorbidity (aHR, 5.84; 95% CI, 4.37-7.80; interaction $P = .5864$). Table VI displays the joint effect of comorbidities and *M pneumoniae* on asthma. Regarding the joint effect of *M pneumoniae* and each comorbidity, the aHR of asthma was higher in patients with *M pneumoniae* infection with hypertension (aHR, 6.36; 95% CI, 3.49-11.6), diabetes (aHR, 6.96; 95% CI, 3.72-13.1), anemia (aHR, 6.83; 95% CI, 4.08-11.4), AR (aHR, 5.20; 95% CI, 3.92-6.90), AD (aHR, 6.27; 95% CI, 4.15-9.45), and AC (aHR, 5.12; 95% CI, 3.80-6.91).

Because the *M pneumoniae* cohort displayed a greater prevalence of comorbidities and therefore carried a greater risk of asthma than the non–*M pneumoniae* cohort, we performed propensity score matching to balance the distribution of those potential risk factors ($P > .05$, see Table E2 in this article's Online Repository at www.jacionline.org). After propensity score matching to control those risk factors, the risk of asthma was still higher in the *M pneumoniae* cohort than in the non–*M pneumoniae* cohort (overall asthma: aHR, 3.86; 95% CI, 2.94-5.09; early-onset asthma: aHR, 3.78; 95% CI, 2.66-5.38; late-onset asthma: aHR, 4.00; 95% CI, 2.57-6.20; see Table E3 in this article's Online Repository at www.jacionline.org).

DISCUSSION

Asthma is a disease characterized by inflammatory cell infiltration of the airway mucosa and thickening of the basal membrane underlying the airway epithelium.[2,4] Similarly, a respiratory *Mycoplasma* species infection is characterized by subacute hyperplastic suppurative bronchiolitis, which is an inflammatory cell infiltration of the airway.[13] Medina et al[2,26] indicated that toxins in patients with *Mycoplasma* species infections might present a causal factor in the cause and exacerbation of human asthma. These findings imply that *M pneumoniae* can induce airway inflammation[3] and contribute to incident asthma.[7,8,27]

TABLE II. Incidence and aHR of asthma stratified by sex, age, comorbidity, and follow-up time between the *M pneumoniae* and non–*M pneumoniae* cohorts

	M pneumoniae						Compared with non–*M pneumoniae*	
	No			Yes				
Variables	Event	Person years	Rate	Event	Person years	Rate	IRR (95% CI)	aHR (95% CI)
Overall asthma	187	34,332	5.45	168	7,898	21.3	3.91 (3.40-4.48)†	3.35 (2.71-4.15)†
Early-onset asthma	115	34,332	3.35	100	7,898	12.7	3.78 (3.26-4.38)†	2.87 (2.18-3.79)†
Late-onset asthma	72	34,332	2.10	68	7,898	8.61	4.11 (3.55-4.75)†	3.95 (2.81-5.54)†
Sex								
Women	101	18,914	5.34	88	4,386	20.1	3.76 (3.13-4.52)†	3.11 (2.32-4.17)†
Men	86	15,418	5.58	80	3,512	22.8	4.08 (3.32-5.02)†	3.66 (2.68-4.99)†
Age (y)								
<35	139	26,300	5.29	128	6,096	21.0	3.97 (3.38-4.67)†	3.46 (2.70-4.43)†
35-65	28	6,487	4.32	27	1,500	18.0	4.17 (3.11-5.6)†	3.47 (2.02-5.96)†
≥65	20	1,544	13.0	13	302	43.1	3.33 (1.95-5.69)†	3.26 (1.60-6.64)†
Comorbidity								
No	72	18,059	3.99	42	2,723	15.4	3.87 (3.13-4.78)†	3.77 (2.58-5.52)†
Yes	115	16,272	7.07	126	5,175	24.4	3.45 (2.86-4.15)†	3.39 (2.63-4.36)†
Atopy syndrome								
No	87	20,297	4.29	51	3,289	15.5	3.62 (2.98-4.40)†	3.57 (2.52-5.05)†
Yes	100	14035	7.12	117	4610	25.4	3.56 (2.92-4.35)†	3.46 (2.65-4.53)†
Follow-up time, (y)								
<2	106	12,535	8.46	132	2,960	44.6	5.27 (4.59-6.06)†	4.41 (3.40-5.74)†
≥2	81	21796	3.72	36	4938	7.29	1.96 (1.68-2.30)†	1.86 (1.25-2.78)*

aHR represents a multiple analysis, including age, sex, and comorbidities. Atopic syndromes include AR, AD, and AC.

Early-onset asthma, Age at diagnosis of asthma less than 12 years; *Late-onset asthma*, age at diagnosis of asthma greater than 12 years; *Rate*, incidence rate (per 1,000 person years).

*$P < .01$.

†$P < .001$.

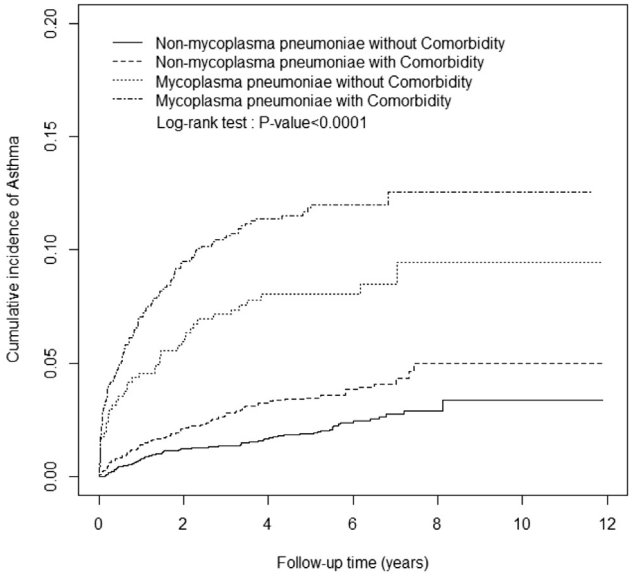

FIG 2. Cumulative incidence of asthma with and without comorbidities in the following 4 subgroups: *M pneumoniae* cohort with comorbidity, *M pneumoniae* cohort without comorbidity, non–*M pneumoniae* cohort with comorbidity, and non–*M pneumoniae* cohort without comorbidity.

The first critical finding of this study is that *M pneumoniae* infection with atopy syndrome increases the risk of incident asthma, regardless of age, sex, and comorbidity. Early-onset (age <12 years) and late-onset (age ≥12 years)[24] asthma constitute different asthma phenotypes. The underlying immune dysfunction and airway inflammation of atopy syndrome might contribute to the *M pneumoniae* infection in patients with early-onset asthma.[22] Therefore atopic sensitization and a history of asthma might be risk factors for refractory *M pneumoniae* infection in affected children.[9] The combined effects of atopy syndrome[28] and *M pneumoniae* infection[1] can contribute to early-onset asthma that continues into adulthood, an age at which a higher frequency of asthma attacks is more likely.[24] Consistent with these findings, Ye et al[1] reported that 402 of 680 children with *M pneumoniae* infection had an allergic condition and that 12 of these 402 allergic children had secondary asthma. The hypothesis of reverse causality between atopy syndrome and *M pneumoniae* infection posited by Juhn[22] fits our conclusions as well.

Our second critical finding is that *M pneumoniae* infection without atopy syndrome increases the incidence of asthma, regardless of age, sex, and comorbidity. The community-acquired distress syndrome toxin[2,26] released at the onset of *M pneumoniae* infection is a critical factor in airway epithelial injuries and activation of innate immunity. Increased airway hyperresponsiveness[29] and exacerbation of airway remodeling[30] in patients with *M pneumoniae* infection increase the risk of incident asthma, even in nonatopic patients. Biscardi et al[31] reported that patients with *M pneumoniae* infection without atopic disease were at an increased risk of asthma. Moreover, atopic disease was not a critical factor for asthmatic patients with *M pneumoniae* infection in a study conducted by Esposito et al.[32] In our study AC alone (aHR, 1.36; 95% CI, 0.99-1.86; $P > .05$) did not increase the risk of asthma. Our conclusion that *M pneumoniae* infection without atopy syndrome contributes to incident asthma is consistent with these reports.[7,8] Collectively,

TABLE III. Incidence and aHR of asthma between the *M pneumoniae* and non–*M pneumoniae* cohorts within the 2-year follow-up

	M pneumoniae						Compared with non–*M pneumoniae*	
	No			Yes				
Variables	Event	Person years	Rate	Event	Person years	Rate	IRR (95% CI)	aHR (95% CI)
Follow-up time (y)								
<0.5	41	3,167	13.0	72	765	94.1	7.27 (6.25-8.44)†	5.99 (4.05-8.87)†
0.5-1	28	3,142	8.91	27	745	36.3	4.07 (3.51-4.72)†	3.35 (1.95-5.76)†
1-1.5	19	3,121	6.09	18	731	24.6	4.04 (3.46-4.72)†	3.91 (2.02-7.56)†
1.5-2	18	3,106	5.80	15	719	20.9	3.95 (2.04-7.65)†	3.00 (1.49-6.06)*
≥2	81	21,796	3.72	36	4,938	7.29	1.96 (1.68-2.30)†	1.86 (1.25-2.78)*

aHR indicates multiple analysis, including age, sex, and comorbidities.
Rate, Incidence rate (per 1000 person years).
*$P < .01$.
†$P < .001$.

TABLE IV. Hazard ratios of asthma associated with outpatient visits and hospitalizations for the exacerbation of *M pneumoniae* infections

Variables	No.	Event	aHR (95% CI)
Non–*M pneumoniae*	6364	187	Reference
M pneumoniae			
Only outpatient	850	96	3.68 (2.86-4.75)*
Only hospitalization	590	38	1.95 (1.38-2.78)*
With outpatient and hospitalization	151	34	7.54 (5.21-10.9)*

aHR indicates multiple analysis, including age, sex, and comorbidities.
*$P < .001$.

TABLE V. aHR of asthma found in the follow-up period associated with *M pneumoniae* and prescriptions of antibiotics or steroids

Variables	No.	Event	aHR (95% CI)
Non–*M pneumoniae*	6364	187	Reference
M pneumoniae			
Without antibiotic and steroid	921	81	2.67 (2.05-3.48)*
Antibiotic use only	323	27	2.74 (1.83-4.12)*
Steroid use only	267	49	6.59 (4.80-9.05)*
Both antibiotic and steroid use	80	11	4.29 (2.33-7.91)*

aHR indicates multiple analysis, including age, sex, and comorbidities.
*$P < .001$.

the findings imply that *M pneumoniae* infection is an independent factor that can lead to incident asthma.[33]

During the 2-year follow-up period, the aHR of asthma was highest among those patients with *M pneumoniae* within the first 6 months, and it decreased in both cohorts at or after the 2-year duration.[21] Severe inflammation in the initial stage of *M pneumoniae* infection implies that the acute infection is a predisposing factor of an asthma attack episode during the less than 6-month period.[25] Moreover, the persistence of toxins in patients with *M pneumoniae* infection might contribute to refractory asthma[34] and incident asthma with 2 or more years of follow-up.[34,35] A previous report indicated that chronic *M pneumoniae* infections increased the incidence of asthma.[36] In the present study the incidence of asthma in the *M pneumoniae* cohort was higher than that in the non–*M pneumoniae* cohort during a period of 2 or more years and a period of less than 6 years. Grad et al[37] reported that early-onset asthma in young adults results in frequent asthma attacks and presents as refractory asthma, which is also in accordance with our findings. The persistence of *Mycoplasma* species in the human body and their biological properties are major risk factors to consider when studying the development of infection-borne asthma and related relapses.[38] Sabato et al[39] examined 108 children with *M pneumoniae* infection during acute illness and continued to monitor the children for 3 years. Even 2 years after the initial onset of the illness, indications of airway hyperresponsiveness to methacholine were apparent, and ELISA detected IgE antibodies specific to *M pneumoniae*.[29] These findings further evidence a link between chronic *M pneumoniae* infection and refractory asthma.

In outpatients incomplete treatment of the residual *Mycoplasma* species organism in a patient with mild *M pneumoniae* infection can result in chronic canonization and airway inflammation.[5,27,40] Therefore even mild inflammation[26,41] caused by *M pneumoniae* infection poses a risk of an incident asthma attack.[42]

Antibiotics might play a role in the eradication of the *M pneumoniae* organism[14] and reduce the inflammation of the sequelae of the *M pneumoniae* infection.[43] Outpatients with *M pneumoniae* infection usually received antibiotic medication. Patients hospitalized with *M pneumoniae* infection received an early and complete course of antibiotic treatment. We found the risk of an asthma attack among patients with a moderate or moderate-to-severe *M pneumoniae* infection that was treated with antibiotics was similar to the risk of an asthma attack among patients with a mild *M pneumoniae* infection who did not receive antibiotic or steroid treatment (aHR, 2.67 vs 2.74). By contrast, the patients with moderate or moderate-to-severe *M pneumoniae* infection who received only steroids were at the highest risk (aHR, 6.59). The monotherapy of the *M pneumoniae* infection with a steroid was not found to reduce instances of airway inflammation–related asthma attacks, which is in accordance with the findings obtained by Tagliabue et al.[14] Our data suggest *M pneumoniae* infection is a critical factor of incident asthma, especially in patients with a moderate-to-severe *M pneumoniae* infection who do not undergo antibiotic treatment.[44]

Patients with severe *M pneumoniae* infection requiring hospitalization and outpatients who might have received antibiotic and steroid treatment are at the highest risk of asthma attacks. This is due to refractory *M pneumoniae* infection.[12] These findings imply that a severe *M pneumoniae* infection might trigger an asthma attack, even if the patients have received antibiotics and steroids.[25] Either a delayed or brief administration

TABLE VI. aHRs of asthma-associated *M pneumoniae* joint effects with comorbidity

Variable		No.	Event	aHR (95% CI)
M pneumoniae	Hypertension			
No	No	5811	158	Reference
No	Yes	553	29	3.82 (2.31-6.34)†
Yes	No	1413	153	4.16 (3.33-5.20)†
Yes	Yes	178	15	6.36 (3.49-11.6)†
M pneumoniae	Diabetes			
No	No	6092	177	Reference
No	Yes	272	10	1.85 (0.94-3.66)†
Yes	No	1494	156	3.78 (3.05-4.69)†
Yes	Yes	97	12	6.96 (3.72-13.1)†
M pneumoniae	Anemia			
No	No	6108	180	Reference
No	Yes	256	7	1.09 (0.51-2.33)
Yes	No	1491	152	3.66 (2.95-4.54)†
Yes	Yes	100	16	6.83 (4.08-11.4)†
M pneumoniae	AR			
No	No	4900	121	Reference
No	Yes	1464	66	1.84 (1.36-2.48)†
Yes	No	925	88	4.09 (3.11-5.39)†
Yes	Yes	666	80	5.20 (3.92-6.90)†
M pneumoniae	AD			
No	No	5926	165	Reference
No	Yes	438	22	1.75 (1.12-2.74)*
Yes	No	1426	141	3.77 (3.01-4.72)†
Yes	Yes	165	27	6.27 (4.15-9.45)†
M pneumoniae	AC			
No	No	4775	130	Reference
No	Yes	1589	57	1.36 (0.99-1.86)
Yes	No	1090	104	3.72 (2.88-4.82)†
Yes	Yes	501	64	5.12 (3.80-6.91)†

The model was adjusted for age and sex.
*$P < .05$.
†$P < .001$.

of steroids or macrolide-resistant *M pneumoniae* infection[34] might contribute to residuals of the *Mycoplasma* species organism in the airway.[34,45] The combined effect of steroids and antibiotics among patients with *M pneumoniae* infection has varied in different study groups.[9,12,14,25] In the current study the combined effect of antibiotics and steroids was lower than the effect of antibiotics only. Tagliabue et al[14] found an obstructed airway among patients with *M pneumoniae* infection, regardless of combined therapy with antibiotics and steroids. Collectively, these findings imply that *M pneumoniae*[1] increases the relationship between airway infections and incident asthma,[33,41] especially in patients with a severe *M pneumoniae* infection.

M pneumoniae infection seems to be a precipitating factor for asthma development in predisposed persons.[31] *M pneumoniae* causes an acute inflammation of the airway[46] and can lead to an initial asthma attack.[7,21] Chronic airway inflammation and infection[26,34] can contribute to refractory asthma.[6,7] In a previous study the impairment of lung functionality caused by infection at the acute[13,14] or chronic[39] stage was more likely to result from *M pneumoniae* than from other pulmonary infections.[35] We have found *M pneumoniae* infection[41,47] contributes to the incident development or progression of early-onset and late-onset asthma, regardless of comorbidities, antibiotic and steroid use, and outpatient and hospitalization status.

Strengths

This study cohort is representative of the general population, and our findings can be generalized to the general population of Taiwan. Physicians follow asthma treatment guidelines,[48] and education programs on asthma care[49] for public health nurses have been conducted in Taiwan. These policies might avoid bias in the diagnosis of asthma.

Limitations

There are several limitations in this study. First, the association between *M pneumoniae* infection and asthma seem to be stronger in the less than 2-year follow-up period in this study. These patients might have had undiagnosed asthma or a disease with few symptoms[50] at the time of the diagnosis of *M pneumoniae* pulmonary infection.

Second, we did not analyze reverse causality or the effect of asthma on the *M pneumoniae* infection. Pathology data regarding *M pneumoniae* infection and incident asthma were unavailable for the patients included in this study.

Finally, the number of patients hospitalized versus the number treated as outpatients was also unavailable in the National Health Insurance Research Database. These factors warrant further investigation.

Conclusion

This study found that incident cases of early-onset and late-onset asthma are closely related to *M pneumoniae* infection, even in patients without atopy syndrome.

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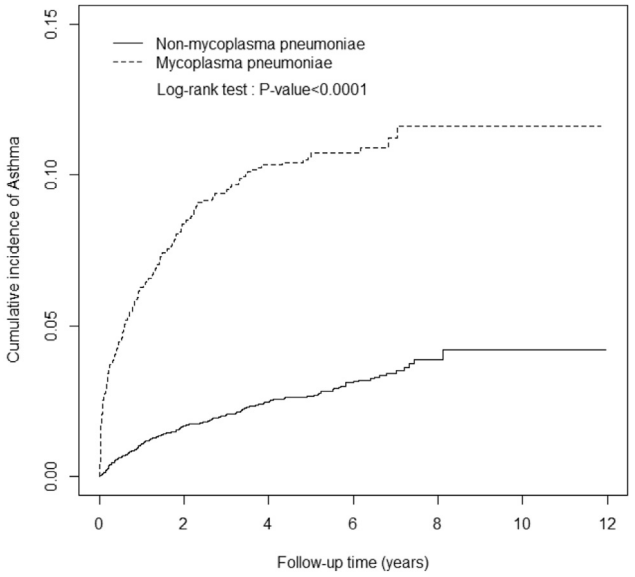

FIG E1. Cumulative incidence of asthma in the *M pneumoniae (dashed line)* and non–*M pneumoniae (solid line)* cohorts.

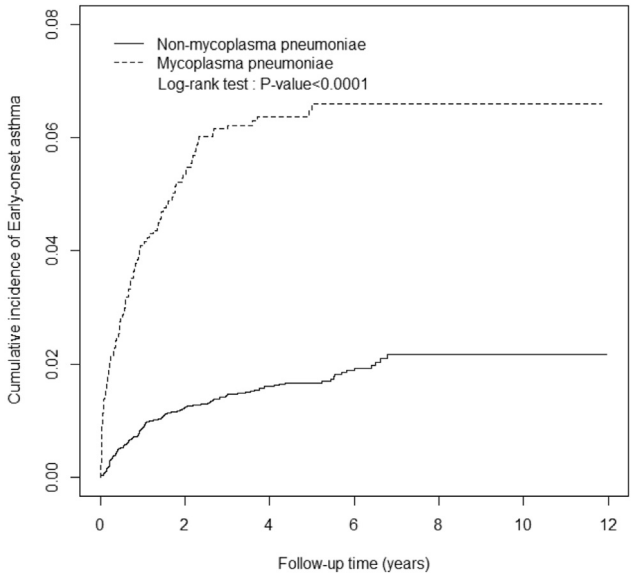

FIG E2. Cumulative incidence of early-onset asthma in the *M pneumoniae (dashed line)* and non–*M pneumoniae (solid line)* cohorts.

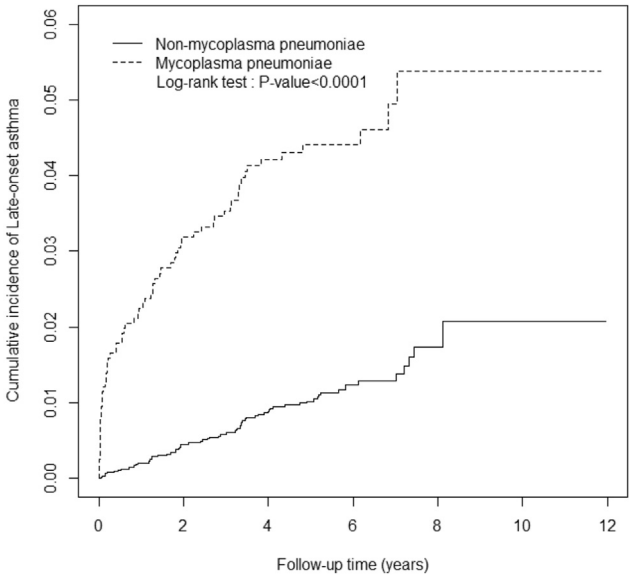

FIG E3. Cumulative incidence of late-onset asthma in *M pneumoniae (dashed line)* and non–*M pneumoniae (solid line)* cohorts.

TABLE E1. aHRs of asthma-associated *M pneumoniae* interactions with comorbidity

Variable		No.	Event	aHR (95% CI)	*P* value
M pneumoniae	Comorbidity				.5864
No	No	3204	72	Reference	
No	Yes	3160	115	1.73 (1.29-2.33)*	
Yes	No	506	42	3.83 (2.62-5.60)*	
Yes	Yes	1085	126	5.84 (4.37-7.80)*	

The model was adjusted for age and sex.
P value for interaction test: *$P < .001$.

TABLE E2. Comparison of demographics and comorbidity between the *M pneumoniae* and non–*M pneumoniae* cohorts by means of propensity score matching

	M pneumoniae				
	No (n = 4200)		Yes (n = 1050)		
	No.	%	No.	%	*P* value
Sex					.656
Women	2360	56.2	582	55.4	
Men	1840	43.8	468	44.6	
Age (y)					.555
<35	3088	73.5	759	72.3	
35-65	865	20.6	232	22.1	
≥65	247	5.88	59	5.62	
Mean (SD)*	23.6 (20.9)		23.7 (20.9)		.796
Comorbidity					
Hypertension	414	9.86	113	10.8	.383
Diabetes	200	4.76	60	5.71	.203
Anemia	168	4.00	45	4.29	.675
AR	570	13.6	148	14.1	.659
AD	256	6.10	72	6.86	.362
AC	990	23.6	255	24.3	.627

Values were determined by using the χ^2 test, unless indicated otherwise.
*Student *t* test.

TABLE E3. Incidence and aHR of asthma between the *M pneumoniae* and non–*M pneumoniae* cohorts after propensity score matching

	M pneumoniae							
	No			Yes			Compared with non–*M pneumoniae*	
Variables	Event	Person years	Rate	Event	Person years	Rate	IRE (95% CI)	HR (95% CI)
Overall asthma	126	23,286	5.41	105	5,346	19.6	3.63 (3.07-4.29)*	3.86 (2.94-5.09)*
Early-onset asthma	75	23,286	3.22	61	5,346	11.4	3.54 (2.96-4.24)*	3.78 (2.66-5.38)*
Late-onset asthma	51	23,286	2.19	44	5,346	8.23	3.76 (3.15-4.49)*	4.00 (2.57-6.20)*

Early-onset asthma, Age at diagnosis of asthma less than 12 years; *Late-onset asthma*, age at diagnosis of asthma greater than 12 years; *Rate*, incidence rate (per 1000 person years).
*$P < .001$.