Association Between Exposure of Ipratropium and Salmeterol and Diagnosis of Multiple Sclerosis: A Matched Case–control Study

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ABSTRACT

Purpose: Ipratropium and salmeterol were found to stimulate oligodendrocyte differentiation in a high-throughput drug screening assay; thus, they may play a role in the risk reduction of multiple sclerosis (MS). So far, they have not been examined in any clinical data. This study aims at investigating the association between ipratropium and salmeterol and reduced diagnosis of MS with the use of real-world clinical data.

Methods: We conducted a 1:10 matched case–control study that compared the exposure of ipratropium and salmeterol between patients with MS and control patients over the past 2 years, using the MS Flowsheet Registry of OSF HealthCare Saint Francis Medical Center. Cases were matched to control patients, based on service year/quarter, age, sex, race, and payer type. The relationship was examined with a Poisson regression model and a generalized structural equation model.

Findings: The sample in our analysis included 217 patients with MS and 2164 matched control patients. The mean (SD) age for both patients with MS and control patients was 41 (11.8) years with a range of 18 to 75 years. The MS group had consistently less prescriptions of ipratropium and salmeterol than the control group in the past 1, 2, and 3 years before the index date. Our multivariable analysis found that the control group had 3.2 more prescriptions (95% CI, 1.4–7.1; $P = 0.006$) of either ipratropium or salmeterol in the past 2 years than the MS group, even if controlling for other confounders. In the generalized structural equation model, we found that use of ipratropium and salmeterol was significantly associated with reduced diagnosis of MS ($P = 0.036$), whereas smokers and people with family history of MS were more likely to have a diagnosis of MS ($P < 0.001$).

Implications: The observed association between ipratropium and salmeterol use and reduced diagnosis of MS indicates that they might potentially serve as agents in the treatment of MS. (*Clin Ther.* 2019;41:1477–1485) © 2019 Published by Elsevier Inc.

Key words: association, case–control study, ipratropium, multiple sclerosis, remyelination, salmeterol.

INTRODUCTION

Multiple sclerosis (MS) is a demyelinating disorder of the central nervous system that affects >400,000 people, with 200 new cases diagnosed each week in the United States.[1] Currently, the cause of MS is not fully understood; however, proposed risk factors include immunologic, environmental, infectious, and genetic factors.[2] Because of the uncertain nature of the disease, current treatments involve managing symptoms, slowing progression of the disease, and

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preventing relapses; there is no known cure. Fortunately, recent research that explored the association between MS and allergies have found promise toward the development of new treatments.

A study performed in 2009 found that patients with a history of atopic allergies were protected against MS.[3] This is further supported by our study done in 2017 which also recognized allergies as a protective factor for MS.[4] The rationale behind this protection is thought to be the T helper type 2 (Th2) cytokine profile among patients with allergies; more specifically, the shift from a Th1 profile (as seen in patients with MS) to a Th2 profile (as seen in patients with allergies).[5] However, there is still much debate on the exact nature of this mechanism. As such, a gap exists in understanding the role of allergies in risk reduction of MS, and there could be much benefit to further studying the Th1 to Th2 transition and its influence on conferring protection against MS, as well as identifying other possible causes for this association. To bridge this gap, it may be useful to explore the relationship between the use of allergy medications and MS.

Such a relationship was explored in a 2017 randomized controlled trial to which it was found that clemastine fumarate, an antihistamine treatment for allergic rhinitis, may potentially induce oligodendrocyte differentiation and myelination.[6] The results of that study found the possibility of exploring other allergy medications that may also have an effect on the central nervous system and subsequent therapeutic values in treating MS. With this in mind, our study focuses on the use of ipratropium and salmeterol, drugs currently used to treat the symptoms of chronic obstructive pulmonary disease and asthma.[7–10] Ipratropium and salmeterol were found to stimulate oligodendrocyte differentiation in a high-throughput drug screening assay[11]; thus, they may play a role in the risk reduction of MS.

We hope to expand the use of ipratropium and salmeterol beyond their current use as medications for chronic obstructive pulmonary disease and asthma. But the evidence for salmeterol and ipratropium being "remyelination drugs" remains lacking, especially in the clinical settings. As such, this study aims to investigate the possible association between the use of ipratropium and salmeterol and reduced diagnosis of MS in an observational study. The significance of this study would be to reveal potential new therapeutic targets in the prevention and treatment of patients with MS.

PATIENTS AND METHODS

This study was approved by the Peoria Institute Review Board located in the University of Illinois College of Medicine at Peoria.

Research Design

This is a retrospective, matched case–control study of comparing the exposure of ipratropium and salmeterol in the past 2 years between patients with MS and control patients. We hypothesized that patients with MS might have lower exposure rates of ipratropium and salmeterol than control patients. The case group included eligible patients with newly diagnosed MS in the MS Flowsheet Registry of OSF HealthCare Saint Francis Medical Center in Illinois[12] during the 4-year period of January 2014 to December 2017. The date of initial MS diagnosis (newly diagnosed) was defined as the index date of study. Each MS case was matched to 10 controls from outpatients within the OSF HealthCare System, based on service year/quarter, age (difference $\leq$5 years), sex, race, and payer type. The purpose of matching was to control for potential confounders in this study.[13]

Study Setting and Population

The data were collected from the OSF HealthCare, Illinois. From our most recent analysis (quick query) that used the OSF data in 2016 to 2017, the preliminary results found that the proportion of ipratropium prescriptions (at least twice) in the past 2 years was 0% and 1.8% in patients with MS and control patients, respectively. To enhance our statistical power, we set a prescription proportion of 1% (rather than 0%) for patients with MS in the calculation of sample size. We assumed that the count of prescriptions might conform to Poisson distribution. It was estimated of a minimum sample size of 308 (28 patients with MS and 280 outpatients) for a 1:10 matching design, given a statistical power of 90% and a significance level of 0.05. In the 4-year study period, we collected 2481 eligible patients (217 patients with MS and 2164 control patients), which ensures that this study could have a sufficient statistical power to examine the study hypothesis.

Case selection criteria included (1) patients with newly diagnosed MS; (2) adults aged $\geq$18 years, because MS is

most commonly diagnosed in people between the ages of 20 and 50 years; and (3) at least 1-year data available before the index date. Control patients were randomly selected from outpatients without MS in the same HealthCare system (OSF), based on the matching conditions that were mentioned above. The diversity of patients seen on an outpatient basis in the OSF HealthCare System ensures that our controls closely represent the general population. OSF HealthCare patients are from both urban and rural areas, and their race distribution indicates 91% of white American, 7% of black American, and 2% of others.

Study Measurements

Patients in the MS Flowsheet Registry were clinically diagnosed with MS, which was in line with the McDonald criteria.[14] The use of ipratropium and salmeterol was defined as the use of ipratropium bromide (including combinations) and/or salmeterol in the past 2 years before the index date. In sensitive analysis, we also changed the exposure time into 1 year and 3 years. We collected the date, route, dosage, and duration for using those medications, body mass index, smoking, family history of MS, as well as comorbidities besides the aforementioned matching variables (age, sex, encounter year/quarter, race, payer). Age was described in years with four categories: <30, 30 to 39, 40 to 49, and ≥50. Race was categorized into white, black, and others. Payers included Medicaid/Medicare, Blue Cross and Blue Shield of Illinois, and commercial insurers. Body mass index levels were defined as <18.5, 18.5 to 24.9, 25 to 29.9, and ≥30. Smoking was measured based on patient-reported information. We defined current smoker as an adult who had smoked 100 cigarettes in his or her lifetime and who currently smokes cigarettes; former smoker as an adult who had smoked at least 100 cigarettes in his or her lifetime but who had quit smoking at the time of interview; and never smoker as an adult who never smoked or had smoked <100 cigarettes in his or her lifetime. Family history of MS included first- and second-degree relatives. The comorbidities were identified based on diagnosis codes in the past 2 years before the index date with the use of the coding algorithms for Charlson comorbidities.[15]

Data Analysis

A 1:10 matching was performed in SAS 9.4 (SAS Institute Inc, Cary, N C), based on the greedy algorithm, using a macro that was created by Erik Bergstralh and Jon Kosanke in 1995 (updated in 2004).[16] Demographic summaries of cases and controls were presented with mean and SD for continuous variables and with frequency and percentage for categorical variables. The χ^2 test was used to compare overall patient characteristics between patients with MS and control patients.

A Poisson regression model was applied to examine if patients with MS had less exposure of ipratropium and salmeterol than control patients in the past few years. In this model, we considered some confounding variables, including age, sex, race, payer type, smoking, and chronic pulmonary disease. In view of ipratropium having a higher market share than salmeterol, we also did a similar analysis for ipratropium only regardless of salmeterol.

To further examine the risk factors for MS, we developed a generalized structural equation model (GSEM). In the GSEM, proposed factors related to MS included smoking, family history, and number of prescriptions of ipratropium and salmeterol. The number of drug prescription could also be affected by smoking, race, payer type, age, sex, and prevalence of chronic pulmonary disease. A statistical significance level of 0.05 was set for all hypothesis tests.

RESULTS

The sample in our analysis included 217 patients with MS and 2164 matched control patients (not every patient with MS had 10 controls). The mean (SD) age for both patients with MS and control patients was 41 (11.8) years with a range of 18 to 75 years. As shown in Table I, both groups had similar distributions in age, sex, encounter year, race, and payer type after matching. In nonmatching variables, we found that the MS group had more current smokers (34.6% versus 20.2%) and family histories of MS (9.2% versus 0.5%) but lower prevalence of chronic pulmonary disease (0.9% versus 5.4%).

As depicted in Figure 1, the MS group had consistently less prescriptions of ipratropium and salmeterol than the control group in the past 1, 2, and 3 years before the index date. To present easily, our main analysis focused on the past 2 years of exposure time. Our multivariable analysis found that the control group had 3.2 more prescriptions (95% CI, 1.4−7.1; $P = 0.006$) of either ipratropium or salmeterol in the past 2 years than the MS group,

Table I. Characteristics of patients with multiple sclerosis and matched control patients.

Variables	Total Patients	Control Patients (n = 2164)	Patients With MS (n = 217)	*P*
Matching				
Age, y, n (%)				0.999
<30	386 (16.2)	350 (16.2)	36 (16.6)	
30–39	764 (32.1)	695 (32.1)	69 (31.8)	
40–49	728 (30.6)	662 (30.6)	66 (30.4)	
≥50	503 (21.1)	457 (21.1)	46 (21.2)	
Female, n (%)	1657 (69.6)	1506 (69.6)	151 (69.6)	0.998
Encounter year, n (%)				1.000
2014	515 (21.6)	468 (21.6)	47 (21.7)	
2015	648 (27.2)	589 (27.2)	59 (27.2)	
2016	823 (34.6)	748 (34.6)	75 (34.6)	
2017	395 (16.6)	359 (16.6)	36 (16.6)	
Race, n (%)				0.998
White	1920 (80.6)	1745 (80.6)	175 (80.6)	
Black and others	461 (19.4)	419 (19.4)	42 (19.4)	
Payer, n (%)				1.000
Medicaid/Medicare	669 (28.1)	608 (28.1)	61 (28.1)	
BCBS	537 (22.6)	488 (22.6)	49 (22.6)	
Commercial and others	1175 (49.3)	1068 (49.4)	107 (49.3)	
Nonmatching				
Body mass index, n (%)				0.111
<18.5	41 (1.8)	33 (1.6)	8 (3.7)	
18.5–24.9	619 (26.5)	568 (26.8)	51 (23.5)	
25–29.9	653 (28.0)	593 (28.0)	60 (27.6)	
≥30	1020 (43.7)	922 (43.6)	98 (45.2)	
Smoking, n (%)				<0.001
Current smoker	512 (21.5)	437 (20.2)	75 (34.6)	
Former smoker	518 (21.8)	473 (21.9)	45 (20.7)	
Never smoker	1351 (56.7)	1254 (57.9)	97 (44.7)	
Family history of MS	32 (1.3)	12 (0.5)	20 (9.2)	<0.001
Top 5 comorbidities, n (%)				
Chronic pulmonary disease	118 (5.0)	116 (5.4)	2 (0.9)	0.004
Diabetes without complications	80 (3.4)	77 (3.6)	3 (1.4)	0.090
Cerebrovascular disease	29 (1.2)	27 (1.2)	2 (0.9)	0.926
Mild liver disease	22 (0.9)	22 (1.0)	0 (0.0)	0.263
Connective tissue disease-rheumatic disease	18 (0.8)	16 (0.7)	2 (0.9)	1.000

BCBS = Blue Cross and Blue Shield of Illinois; MS = multiple sclerosis.

even if controlling for other confounders. The results were also similar when we narrowed to ipratropium only regardless of salmeterol (Table II).

The negative association between MS and number of prescriptions of ipratropium and salmeterol was validated in our GSEM (Figure 2). In the GSEM, we

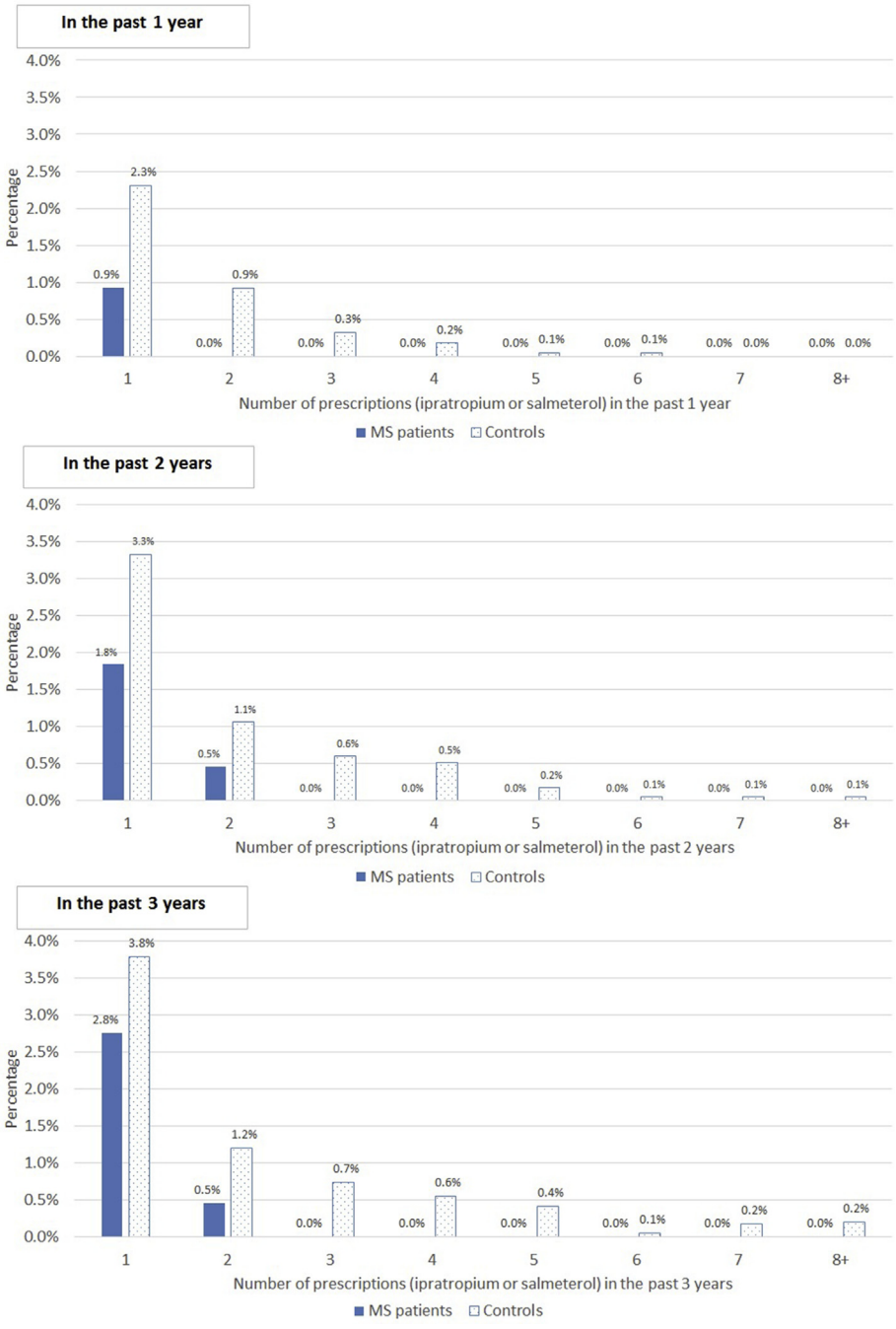

Figure 1. Prescriptions of ipratropium and salmeterol in the past few years between patients with multiple sclerosis (MS) and matched control patients.

found that the use of ipratropium and salmeterol was significantly associated with reduced diagnosis of MS ($P = 0.036$), whereas smokers and people with family history of MS were more likely to have a diagnosis of MS ($P < 0.001$). In addition, the prescription of ipratropium and salmeterol was significantly

Table II. Estimation of the exposure of ipratropium and salmeterol based on multivariable Poisson regression models.

Variables	Prescriptions of Either Ipratropium or Salmeterol			Prescriptions of Ipratropium (regardless of salmeterol)		
	Est.	95% CI	*P*	Est.	95% CI	*P*
Group						
Control versus multiple sclerosis	3.16	1.40–7.13	0.006	3.32	1.22–9.01	0.019
Race						
Black and others versus white	1.40	1.03–1.89	0.032	1.81	1.26–2.60	0.001
Smoking						
Current smoker versus never smoker	1.90	1.38–2.61	<0.001	2.71	1.80–4.09	<0.001
Former smoker versus never smoker	1.37	0.98–1.90	0.062	1.64	1.05–2.55	0.029
Payer						
Medicaid/Medicare versus commercial and others	2.22	1.62–3.05	<0.001	3.00	1.99–4.55	<.0001
BCBS versus commercial and others	1.20	0.81–1.79	0.370	0.70	0.36–1.36	0.291
Chronic pulmonary disease						
Yes versus no	10.50	8.09–13.64	<0.001	9.45	6.81–13.12	<0.001
Age						
One-year change	1.02	1.01–1.03	0.001	1.02	1.01–1.03	0.005
Sex						
Female versus male	1.42	1.05–1.93	0.024	1.53	1.03–2.26	0.034

BCBS = Blue Cross and Blue Shield of Illinois; Est. = estimate of the difference change of expected prescription counts.

influenced by smoking, race, payer type, age, sex, and prevalence of chronic pulmonary disease.

DISCUSSION

This is the first study to investigate the exposure of ipratropium and salmeterol in patients with MS and general outpatients. Our case–control study found that patients with MS had less prescriptions of ipratropium and salmeterol in the past few years compared with control patients, which could imply that use of ipratropium and salmeterol might be associated with reduced diagnosis of MS. We think that ipratropium and salmeterol might coincidently prevent or stop the development of MS in the early stages of disease, such as clinically isolated syndrome or before clinical symptoms of MS develop.

In the cause of MS, environmental factors (eg, smoking and virus infection) might trigger demyelination by a break of immune tolerance in genetically susceptible people.[17–20] Leukocyte infiltration of brain tissue causes inflammation, demyelination, and the subsequent formation of sclerotic plaques, which are a hallmark of MS. Activation of proinflammatory cytokines is essential for regulation of lymphocyte migration across the blood–brain barrier. The role of Th1 lymphocytes (eg, interleukin 2 receptor a and interleukin 17) is thought to be critical in the pathogenesis of brain inflammation in MS.[21] Recently, some population-based case–control studies have concluded that atopic allergies confer protection against MS.[3,4,22] It could be explained that the shift from a Th1 profile to a Th2 profile among patients with allergies might stop the development of MS.[5] However, this phenomenon could also be explained, at least partially, by another surmise that allergy drugs (such as clemastine, ipratropium, and salmeterol) coincidently prevent or stop the development of MS in the early stages of disease. In our study, we found that few patients with MS were prescribed ipratropium or salmeterol twice or more in the past few years. Even if controlling the confounder variable

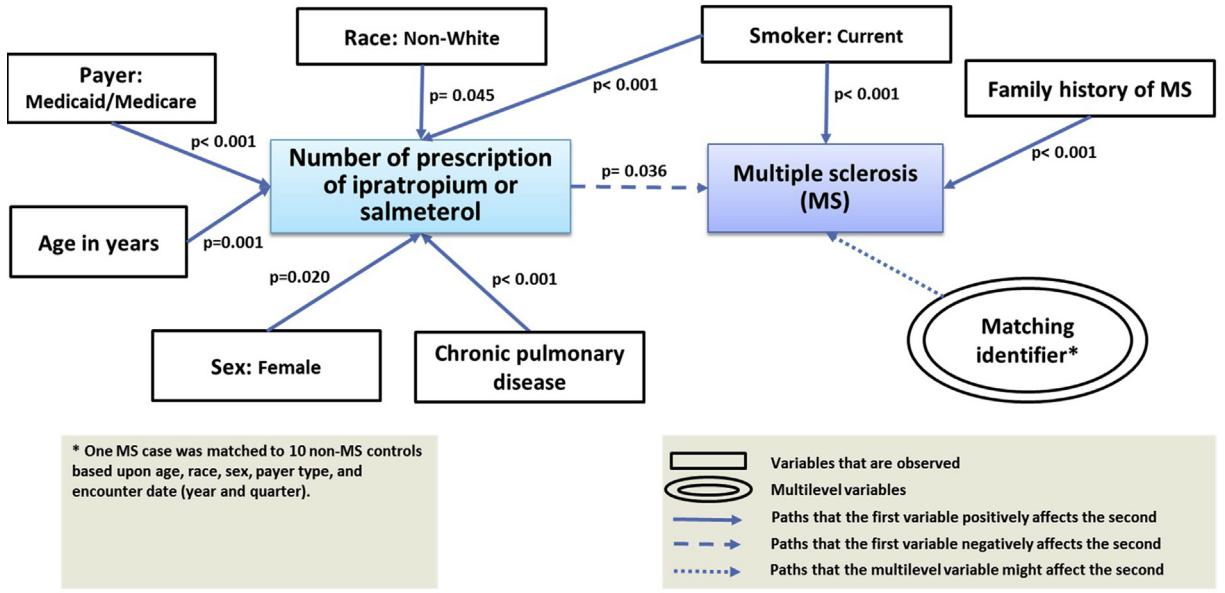

Figure 2. Results of a structural equation model with two outcomes of multiple sclerosis incident and ipratropium and salmeterol.

of chronic lung diseases, patients with MS still have significantly less exposure to ipratropium or salmeterol than control patients.

Repurposing existing drugs might be a shortcut for developing new therapies for some diseases, including MS, because the timeline from taking a drug from the laboratory to the consumer market is long and costly. Some allergy drugs, such as clemastine, ipratropium, and salmeterol, have been identified as potential remyelination drugs by high-throughput drug screening tests.[11,23] Of them, the efficacy of clemastine has been confirmed in multiple animal models.[24,25] After clemastine treatment in a cuprizone mouse model, myelin repair was greatly enhanced in the demyelinated regions with increased mature oligodendrocytes (adenomatous polyposis coli-positive) and myelin basic protein.[25]In addition, clemastine has been used as a remyelinating therapy in a Phase II clinical trial and found a positive impact on reducing the latency delay among patients with MS.[6] However, no clinical data about repurposing ipratropium and salmeterol for MS have been reported yet. The findings in our study suggest to further investigate the feasibility of repurposing ipratropium and salmeterol for MS. The reason that ipratropium has not been selected as a remyelinating therapy might be because it is difficult to pass through the blood–brain barrier, which is associated with common remyelination pathways, including Notch-1, Wnt, and LINGO-1.[26–31] However, ipratropium might change the serum levels of certain chemokines (eg, C-X-C motif chemokine 12, C-X-C motif chemokine receptor [CXCR]4, CXCR7) that modulate remyelination. Treatments that either enhance CXCR4 stimulation or block CXCR7 may be useful in enhancing remyelination in MS.[27,32]

A few limitations in our study should be noted. First, the retrospective study design has its own disadvantages, such as recall bias. We believe the recall bias is limited in our study because the data, including diagnoses and drug prescriptions, were extracted from electronic medical records. Although this retrospective study is not able to certainly confirm that ipratropium and salmeterol reduce the risk of MS, our findings have provided a certain evidence of association between use of ipratropium and salmeterol and reduced diagnosis of MS, which paves the way for future prospective interventional studies. Second, our data were only from one medical system that mainly covers central Illinois. More studies from different sites are needed to validate our findings. Third, the information in the medical records cannot allow us to quantify the dose

of exposure. For example, ipratropium bromide is usually prescribed to use as needed, and it is difficult to know the actual dose that patients receive. Thus, our study used the frequency of prescriptions to reflect the dose of exposure. Fourth, the interactions between ipratropium and salmeterol and other drugs are unclear. Side effects should be prudently monitored when ipratropium and salmeterol are used to treat MS. Fifth, our study might still miss a few probable confounding factors. Because of the limit of sample size, we were not able to conduct further stratified analyses to examine the differences in MS between patients with asthma treated with ipratropium and salmeterol and patients treated with other bronchodilators. Finally, the low exposure rate of ipratropium and salmeterol increases our challenges in the analysis. As such, we applied Poisson regression model instead of logistic regression model to investigate the exposure of ipratropium and salmeterol in both groups. In addition, overmatching could introduce new confounding or biases in a matched case–control study. Therefore, we should cautiously select an appropriate analysis when matching is used in a study. Recently, it has been suggested that a "matched" (conditional) analysis may not be required or appropriate for a matched study design.[33] In our study, we chose the unconditional regression model to compare the exposure of drugs between two groups and controlled for those potential confounders.

CONCLUSION

This study will provide a platform for personalized medicine for the high-risk populations of MS by tailoring treatments based on lifestyle and environmental factors. It would allow for future research opportunities focused on developing intervention plans for high-risk populations. It would also allow us to proceed forward and to develop a randomized clinical trial that used ipratropium and salmeterol for patients with MS to explore new treatment strategies.

CONFLICTS OF INTEREST

The authors declare that they have no competing interests. The authors have indicated that they have no conflicts of interest regarding the content of this article.

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REFERENCES

1. Multiple sclerosis by the numbers: facts, statistics and you. April 26, 2019, [http://www.healthline.com/health/multiple-sclerosis/facts-statistics-infographic].
2. What Causes MS. April 26, 2019, [https://www.nationalmssociety.org/What-is-MS/What-Causes-MS].
3. Pedotti R, Farinotti M, Falcone C, et al. Allergy and multiple sclerosis: a population-based case-control study. *Mult Scler*. 2009;15:899–906.
4. Ren J, Ni H, Kim M, Cooley KL, Valenzuela RM, Asche CV. Allergies, antibiotics use, and multiple sclerosis. *Curr Med Res Opin*. 2017;33:1451–1456.
5. Oreja-Guevara C, Ramos-Cejudo J, Aroeira LS, Chamorro B, Diez-Tejedor E. TH1/TH2 Cytokine profile in relapsing-remitting multiple sclerosis patients treated with Glatiramer acetate or Natalizumab. *BMC Neurol*. 2012;12:95.
6. Green AJ, Gelfand JM, Cree BA, et al. Clemastine fumarate as a remyelinating therapy for multiple sclerosis (ReBUILD): a randomised, controlled, double-blind, crossover trial. *Lancet*. 2017;390:2481–2489.
7. Atsmon J. Ipratropium bromide in COPD and asthma. *Am J Med Sci*. 1988;296:140–142.
8. Donohue JF, Wise R, Busse WW, et al. Efficacy and safety of ipratropium bromide/albuterol compared with albuterol in patients with moderate-to-severe asthma: a randomized controlled trial. *BMC Pulm Med*. 2016;16:65.
9. Paik J, Scott LJ, Pleasants RA. Fluticasone propionate/salmeterol MDPI (AirDuo RespiClick(R)): a review in asthma. *Clin Drug Investig*. 2018;38:463–473.
10. Frith PA, Ashmawi S, Krishnamurthy S, et al. Efficacy and safety of the direct switch to indacaterol/glycopyrronium from salmeterol/fluticasone in non-frequently exacerbating COPD patients: the FLASH randomized controlled trial. *Respirology*. 2018;23:1152–1159.

11. Lariosa-Willingham KD, Rosler ES, Tung JS, Dugas JC, Collins TL, Leonoudakis D. A high throughput drug screening assay to identify compounds that promote oligodendrocyte differentiation using acutely dissociated and purified oligodendrocyte precursor cells. *BMC Res Notes*. 2016;9:419.
12. Cooley KL, Paris BL, Asche CV, Valenzuela R. Development of an integrated multiple sclerosis flowsheet registry to bridge the gap between electronic health record functionality and clinical information needs. *Austin J Mult Scler Neuroimmunol*. 2015;2:1023.
13. Pourhoseingholi MA, Baghestani AR, Vahedi M. How to control confounding effects by statistical analysis. *Gastroenterol Hepatol Bed Bench*. 2012;5:79–83.
14. Polman CH, Reingold SC, Banwell B, et al. Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. *Ann Neurol*. 2011;69:292–302.
15. Quan H, Sundararajan V, Halfon P, et al. Coding algorithms for defining comorbidities in ICD-9-CM and ICD-10 administrative data. *Med Care*. 2005;43:1130–1139.
16. Match. April 26, 2019, [http://www.mayo.edu/research/documents/matchsas/doc-10027556].
17. O'Gorman CM, Broadley SA. Smoking increases the risk of progression in multiple sclerosis: a cohort study in Queensland, Australia. *J Neurol Sci*. 2016;370:219–223.
18. Ockinger J, Hagemann-Jensen M, Kullberg S, et al. T-cell activation and HLA-regulated response to smoking in the deep airways of patients with multiple sclerosis. *Clin Immunol*. 2016;169:114–120.
19. Hedstrom AK, Sundqvist E, Baarnhielm M, et al. Smoking and two human leukocyte antigen genes interact to increase the risk for multiple sclerosis. *Brain*. 2011;134(Pt 3):653–664.
20. Mori M. [Association between multiple sclerosis or neuromyelitis optica and Epstein-Barr virus]. *Brain Nerve*. 2015;67:881–890.
21. Khaibullin T, Ivanova V, Martynova E, et al. Elevated levels of proinflammatory cytokines in cerebrospinal fluid of multiple sclerosis patients. *Front Immunol*. 2017;8:531.
22. Sahraian MA, Jafarian S, Sheikhbahaei S, Safavi F. Respiratory tract rather than cutaneous atopic allergy inversely associate with multiple sclerosis: a case-control study. *Clin Neurol Neurosurg*. 2013;115:2099–2102.
23. Mei F, Fancy SPJ, Shen YA, et al. Micropillar arrays as a high-throughput screening platform for therapeutics in multiple sclerosis. *Nat Med*. 2014;20:954–960.
24. Liu J, Dupree JL, Gacias M, et al. Clemastine enhances myelination in the prefrontal cortex and rescues behavioral changes in socially isolated mice. *J Neurosci*. 2016;36:957–962.
25. Li Z, He Y, Fan S, Sun B. Clemastine rescues behavioral changes and enhances remyelination in the cuprizone mouse model of demyelination. *Neurosci Bull*. 2015;31:617–625.
26. Zhang Y, Zhang YP, Pepinsky B, et al. Inhibition of LINGO-1 promotes functional recovery after experimental spinal cord demyelination. *Exp Neurol*. 2015;266:68–73.
27. Nadeem M, Sklover L, Sloane JA. Targeting remyelination treatment for multiple sclerosis. *World J Neurol*. 2015;5:5–15.
28. Hammond TR, Gadea A, Dupree J, et al. Astrocyte-derived Endothelin-1 inhibits remyelination through Notch activation. *Neuron*. 2014;81:1442.
29. Ye F, Chen Y, Hoang T, et al. HDAC1 and HDAC2 regulate oligodendrocyte differentiation by disrupting the beta-catenin-TCF interaction. *Nat Neurosci*. 2009;12:829–838.
30. Mi S, Hu B, Hahm K, et al. LINGO-1 antagonist promotes spinal cord remyelination and axonal integrity in MOG-induced experimental autoimmune encephalomyelitis. *Nat Med*. 2007;13:1228–1233.
31. Mi S, Miller RH, Lee X, et al. LINGO-1 negatively regulates myelination by oligodendrocytes. *Nat Neurosci*. 2005;8:745–751.
32. Brunn A, Utermohlen O, Mihelcic M, et al. Differential effects of CXCR4-CXCL12- and CXCR7-CXCL12-mediated immune reactions on murine P0106-125 -induced experimental autoimmune neuritis. *Neuropathol Appl Neurobiol*. 2013;39:772–787.
33. Pearce N. Analysis of matched case-control studies. *BMJ*. 2016;352:i969.

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