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Article:
Lawless, D., Geier, C.B., Farmer, J.R. et al. (2018) Prevalence and clinical challenges among adults with primary immunodeficiency and recombination-activating gene deficiency. Journal of Allergy and Clinical Immunology, 141 (6). pp. 2303-2306. ISSN: 0091-6749

https://doi.org/10.1016/j.jaci.2018.02.007

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Prevalence and clinical challenges among adults with primary immunodeficiency and recombination-activating gene deficiency

To the Editor:

Recombination-activating gene (RAG) deficiency has an estimated disease incidence of 1:181,000, including severe combined immunodeficiency (SCID) at a rate of 1:330,000.[1,2] Complete or hypomorphic variants of SCID secondary to low recombinase activity (<5%) present early with severe infections and/or clinical signs of systemic inflammation, such as severe dermatitis, colitis, or both.[3,4] Hypomorphic *RAG1/2* mutations with more preserved residual V(D)J recombination activity (5% to 30%) result in a distinct phenotype of combined immunodeficiency with granuloma, autoimmunity, or both.[1,2,5] Beyond combined immunodeficiency, RAG deficiency has been found in patients with predominantly primary antibody deficiencies[6,7] and naive $CD4^+$ T-cell lymphopenia in most cases. Currently, there is no published systematic evaluation for the presence of an underlying RAG deficiency in patients with primary antibody deficiencies. There is great variability among diagnostic modalities for evaluation and treatment for inflammatory lung disease in case reports of RAG deficiency with no standardized guidelines. Clinical features and lung disease for patients with late presentation of RAG deficiency have not been studied extensively. In addition, no studies have examined the prevalence of RAG deficiency in cohorts of adults with primary immunodeficiency (PID). Here we describe a cohort of 15 patients with late presentation of RAG deficiency. We also estimate the prevalence of RAG deficiency in adults with PID after genetic analysis in 2 separate large cohorts of patients with PID.

We have analyzed the canonical regions of *RAG1* and *RAG2* in a total of 692 patients with PID from 2 separate cohorts, one from the United Kingdom (UK) and one from Austria (Vienna). The UK cohort is part of the National Institute for Health Research BioResource–Rare Diseases PID study, as previously described (Tuijnenburg et al[8]). In the National Institute for Health Research BioResource–Rare Diseases PID cohort of 558 patients (299 adults) and the Vienna cohort of 134 patients (106 adults), we report a total of 5 newly identified cases of RAG deficiency. For details, see the Methods section and Tables E1 to E3 in this article's Online Repository at www.jacionline.org. Based on these findings, we estimate that the prevalence of RAG deficiency in adults with PID ranges from 1% to 1.9%. For all adult patients with PID currently registered with the UK Primary Immunodeficiency Network database (3294 patients older than age 18 years), we expect to find an additional 32.9 to 62.6 cases of RAG deficiency. Gene variants are shown in Fig 1, *A*. Cohort demographics are discussed in the Methods section in this article's Online Repository.

Functional characterization of novel RAG variants is discussed in the Methods section in this article's Online Repository. The activity of mutant RAG1 and RAG2 proteins normally required for catalyzing V(D)J recombination events are shown in Table E2. In addition to the method previously described,[9] we also used a system to measure recombination activity in compound heterozygous cases by means of *in vitro* expression of murine RAG1 and RAG2. Both systems simulate the efficiency of protein expressed in patients in their ability to produce a diverse repertoire of T-cell receptor and B-cell receptor coding for immunoglobulins. More than half of the mutant proteins tested show almost complete loss of activity. All patients tested had an overall low combined RAG activity (6.4% to 28%).

Immune phenotypes and clinical diagnoses are shown in Fig 1, *B*. Persistently low IgG and/or low IgA and IgM levels are seen in approximately 50% of cases (see Table E3). Dominant laboratory features were naive $CD4^+$ T-cell lymphopenia with low absolute numbers and fraction of naive $CD4^+$ cells ($CD4^+CD45RA^+$), and B-cell counts were variably low (see Table E3). ELISA was used to test for anti-cytokine antibodies (targeting IFN-α, IFN-ω, and IL-12) on plasma from 7 patients (data not shown). Four patients had positive results, which is comparable with our previously reported cohort (56%).[5]

Most adult RAG-deficient patients had inflammatory autoimmune complications (87%; see the Methods section and Fig E1, *A*, in this article's Online Repository at www.jacionline.org). Organ-specific manifestations were most common (73%) and similar to previously described reports of 48% to 77%.[5,10] Granulomatous disease was seen in 40% of patients, with 5 of 6 patients showing granuloma localization within interstitial lung tissue. Other complications were also seen (see Fig E1, *E*). Similar to recent reports (21% to 77%),[5,10] cytopenias occurred in 40% of patients: autoimmune hemolytic anemia (27%), immune thrombocytopenic purpura (20%), and autoimmune neutropenia in 1 patient.

Progressive pulmonary diseases were the leading causes of morbidity and mortality (93%; see Fig E1, *D*), with pneumonia being the most common, followed by bronchiectasis, chronic bronchitis, granuloma, fibrosis, chronic obstructive pulmonary disease, and bronchiolitis (Fig 1, *C*). We observed a transition from acute infectious complications (pneumonia; mean onset, 14 years) to chronic inflammatory complications (mean age, 23 years; Fig 2, *A*).

Progressive pulmonary diseases were also the leading concern for successful hematopoietic stem cell transplantation (HSCT). High-resolution computed tomographic imaging of the lung revealed bronchiectasis and granuloma. Histology of lung biopsy specimens (patients 1 and 3) revealed atypical lymphoid hyperplasia with granulomatous features and giant cell formation (Fig 2, *B*). Germinal center formations in patient 3 were comprised of $CD3^+$ T cells and $CD20^+$ B cells. Patient 1 had peribronchial fibrosis (Fig 2, *B*). Pulmonary lung function data revealed a median forced vital capacity of 79.55%, diffusing capacity of the lungs for carbon monoxide of 75%, and FEV_1/forced vital capacity ratio of 78%. We performed a retrospective analysis of pulmonary function tests over 2 or more years to assess the decrease in respiratory function. Two of 4 patients had a significant decrease, indicating a variable degree of lung function in adult patients with RAG deficiency (see the Methods section and Fig E1, *G*, in this article's Online Repository).

Thirteen (93%) of 14 patients received first-line immunoglobulin replacement therapy (see Fig E1, *B*, and Table E2). Fifty-seven percent received antibiotic prophylaxis, 21% received antiviral drugs, and 14% received disease-modifying

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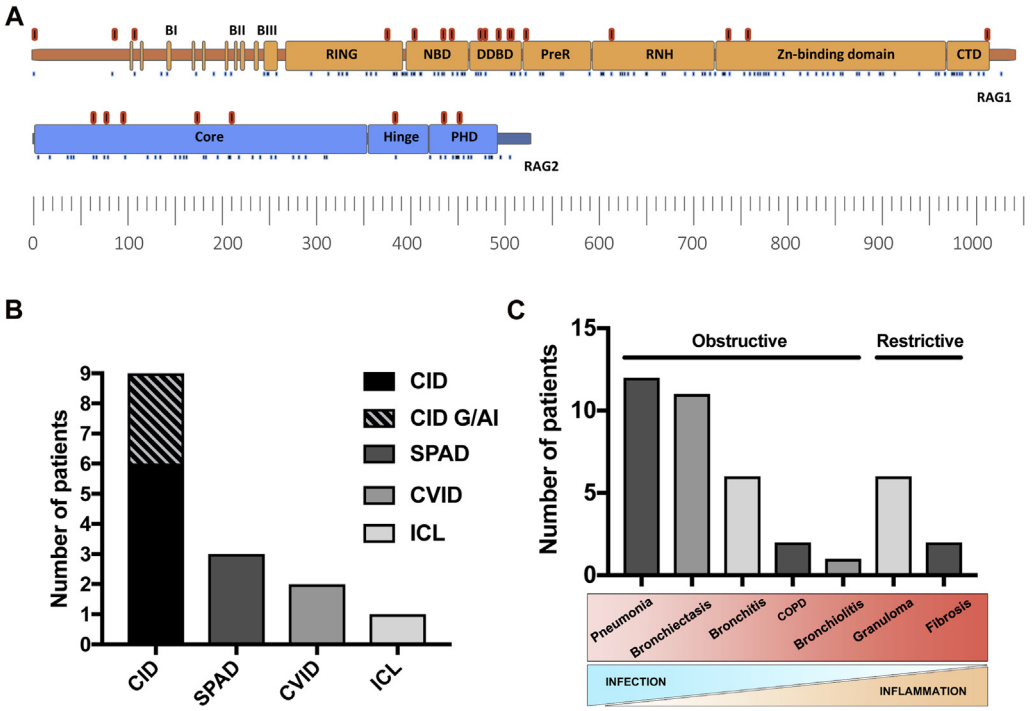

FIG 1. Gene variants and phenotypes of adults with RAG deficiency. **A,** Schematic representation of *RAG1* and *RAG2* adapted from Notarangelo et al.[E19] Variants in this cohort (17 in *RAG1* and 8 in *RAG2*) are shown in red. Known pathogenic variants reviewed previously are shown as *blue dots*.[E19] **B,** Phenotypes of RAG deficiency in 15 patients, including those from the National Institute for Health Research BioResource–Rare Diseases PID and Vienna cohorts. **C,** Presentation of lung disease (n = 15). *CID*, Combined immunodeficiency; *CID G/AI*, combined immunodeficiency with granuloma and/or autoimmunity; *COPD*, chronic obstructive pulmonary disease; *CVID*, common variable immunodeficiency; *ICL*, idiopathic $CD4^+$ T lymphopenia; *SPAD*, specific polysaccharide antibody deficiency.

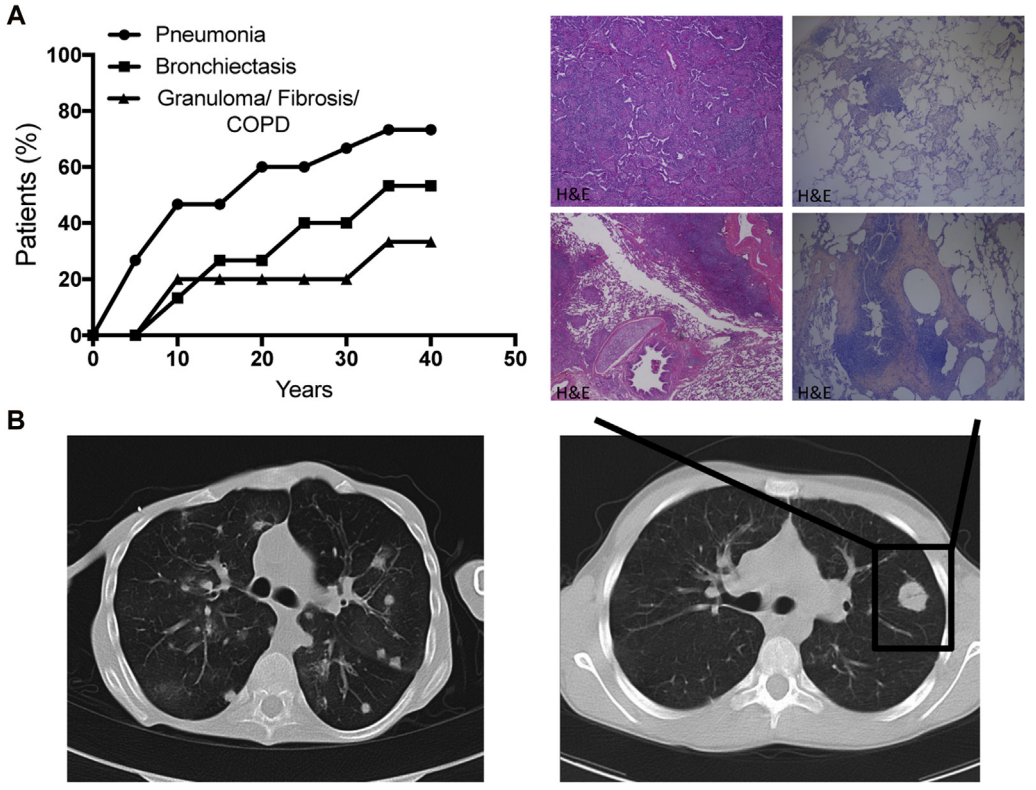

FIG 2. Pulmonary disease. **A,** Onset of pneumonia, bronchiectasis, and granuloma/fibrosis/chronic obstructive pulmonary disease (*COPD*; n = 15). **B,** High-resolution computed tomography of patients 3 and 9. Histologic examination of lung biopsy specimens from patient 1 with atypical lymphoid hyperplasia with granuloma and fibrosis (hematoxylin and eosin).

antirheumatic drugs. Five (36%) patients were considered for HSCT. Comparisons of therapeutic approaches revealed no statistically significant difference in survival. Three of 8 patients who received only immunoglobulin replacement therapy were deceased. Among patients undergoing transplantation, the major mortality cause was infection after HSCT (see Fig E1, *C*).

RAG1/2 are the most common defective genes associated with atypical SCID.[10] Patients can survive into adulthood, and our findings suggest that the prevalence of such cases varies between 1% and 1.9% in adult PID cohorts. Total and naive $CD4^+$ T-cell lymphopenia,[4,10] autoimmunity, and progressive inflammatory lung disease should all prompt further investigations for RAG deficiency in adults with PIDs. The relative absence of RAG deficiency in the pediatric cohort of 216 patients suggests that milder forms of RAG deficiency might not be diagnosed as readily as a PID in childhood. In the era of whole-exome sequencing, the spectrum of RAG deficiency broadens further to include adults with autoimmune and inflammatory manifestations that can result in progressive decrease. Systemic analysis of PID-related genes[9] and functional *in vitro* assays that confirm decreased recombination activity are essential. Laboratory features of naive $CD4^+$ T-cell lymphopenia and the presence of anti-cytokine antibodies can further support the diagnosis of partial RAG deficiency. Where RAG deficiency is confirmed, therapy can be adjusted based on the mechanistic understanding and might ultimately provide a targeted strategy for early intervention.

This study was conducted in accordance with the Declaration of Helsinki. Patients provided written informed consent so that anonymized data could be included in a scientific publication. All results presented in this study were obtained as part of the routine medical care received by the patient.

This study makes use of data generated by the NIHR BioResource–Rare Disease Consortium. A full list of the consortium members who contributed to the generation of the data is available in this article's Online Repository at www.jacionline.org. We thank Professor Christian J. Müller for providing pathology specimens and Dr Karl Waibel for providing pulmonary function testing.

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Supported in part by National Institute of Allergy and Infectious Diseases/National Institutes of Health grant 5K08AI103035 (to J.E.W.), the Jeffrey Modell Foundation (to J.E.W.), the Robert A. Good Endowment, and the University of South Florida.

Disclosure of potential conflict of interest: J. R. Farmer reports a grant from CSL Behring. H. Lango Allen reports employment at the University of Cambridge. S. O. Burns reports a grant from HEFCE and consulting fees or honoraria from CSL Behring; employment by University College London; grants/grants pending from NIHR, UCLH III BRC, and GOSH/ICH BRC; and travel/accommodations/meeting expenses from Immunodeficiency Canada/IAACI, CSL Behring, Baxalta, and Octagam. M. J. Butte reports employment from the University of California–Los Angeles; grants from the National Institutes of Health (NIH) and stock/stock options from Chimera Bio, Alpine Immune Sciences, and Allertope. S. E. Henrickson reports consultancy for Horizon Pharma and grants/grants pending from the and BWF. A. J. Thrasher reports board membership and consultancy for Orchard Therapeutics, Torus Therapeutics, and Rocket Pharmaceuticals and stock/stock options from Orchard Therapeutics. H. M. Wolf reports Consultancy at Baxalta and payment for lectures, including service on speakers' bureaus from Shire and Baxalta and travel/accommodations/meeting expenses from Shire and Kedrion. J. E. Walter reports a grant and pending grant from the National Institute of Allergy and Infectious Diseases (NIAID) and consultancy from CSL Behring and Shire. S. Savic reports payment for development of educational presentations from Grifols, Shire, and Sobi and travel/accommodations/meeting expenses from Grifols, Octapharma, and Shire. The rest of the authors declare that they have no relevant conflicts of interest.

REFERENCES

1. Kumanovics A, Lee YN, Close DW, Coonrod EM, Ujhazi B, Chen K, et al. Estimated disease incidence of RAG1/2 mutations: a case report and querying the Exome Aggregation Consortium. J Allergy Clin Immunol 2017;139:690-2.e3.
2. Kwan A, Abraham RS, Currier R, Brower A, Andruszewski K, Abbott JK, et al. Newborn screening for severe combined immunodeficiency in 11 screening programs in the United States. JAMA 2014;312:729-38.
3. Schwarz K, Gauss GH, Ludwig L, Pannicke U, Li Z, Lindner D, et al. RAG mutations in human B cell-negative SCID. Science 1996;274:97-9.
4. Felgentreff K, Perez-Becker R, Speckmann C, Schwarz K, Kalwak K, Markelj G, et al. Clinical and immunological manifestations of patients with atypical severe combined immunodeficiency. Clin Immunol 2011;141:73-82.
5. Walter JE, Rosen LB, Csomos K, Rosenberg JM, Mathew D, Keszei M, et al. Broad-spectrum antibodies against self-antigens and cytokines in RAG deficiency. J Clin Invest 2015;125:4135-48.
6. Buchbinder D, Baker R, Lee YN, Ravell J, Zhang Y, McElwee J, et al. Identification of patients with RAG mutations previously diagnosed with common variable immunodeficiency disorders. J Clin Immunol 2015;35:119-24.
7. Geier CB, Piller A, Linder A, Sauerwein KM, Eibl MM, Wolf HM. Leaky RAG deficiency in adult patients with impaired antibody production against bacterial polysaccharide antigens. PLoS One 2015;10:e0133220.
8. Tuijnenburg P, Lango Allen H, Burns SO, Greene D, Jansen MH, Staples E, et al. Loss of function NFKB1 variants are the most common monogenic cause of CVID in Europeans. J Allergy Clin Immunol 2018 Feb 22. http://dx.doi.org/10.1016/j.jaci.2018.01.039 [Epub ahead of print].
9. Lee YN, Frugoni F, Dobbs K, Walter JE, Giliani S, Gennery AR, et al. A systematic analysis of recombination activity and genotype-phenotype correlation in human recombination-activating gene 1 deficiency. J Allergy Clin Immunol 2014;133:1099-108.
10. Speckmann C, Doerken S, Aiuti A, Albert MH, Al-Herz W, Allende LM, et al. A prospective study on the natural history of patients with profound combined immunodeficiency: an interim analysis. J Allergy Clin Immunol 2017;139:1302-10.e4.

Available online March 2, 2018.
http://dx.doi.org/10.1016/j.jaci.2018.02.007

Proximity to traffic and asthma among Mexican American children: Independent and interactive effects

To the Editor:

Studies linking proximity to traffic, a surrogate measure of exposure to traffic-related air pollution (TRAP), with an increased risk of respiratory conditions in children have produced inconsistent findings. Some studies showed positive associations of TRAP with asthma and wheezing in children with a family history of asthma or allergies,[1,2] whereas other investigations[3,4] found positive associations only in children without parental history of atopic conditions. In the United States, there is evidence demonstrating that racial/ethnic minority populations including Mexican Americans (MAs) are more likely to live near major roads or reside in neighborhoods with increased traffic and therefore experience a disproportionate share of air pollution exposure. However, MAs remain underrepresented in research on the role of TRAP in the etiology of respiratory disease. In this study, we tested associations of proximity to traffic with childhood respiratory outcomes (lifetime asthma, lifetime wheezing, and current wheezing) and whether these associations differed by parental history of asthma or allergies, individual-level sociodemographic characteristics, immigration-related factors, and environmental stressors.

Details of the research methodology are provided in this article's Methods section in the Online Repository at www.jacionline.org. Briefly, individual-level data were collected as part of the Study of Asthma in Children of Mexican Descent, a population-based, cross-sectional study.[5] Between August 2004 and April 2005, the parents of 2023 children (grades K-8) completed a mailed questionnaire or telephone interview about respiratory conditions and factors related to asthma in their children. We geocoded the addresses of all children using geographic information system software. The locations of all primary arterials were obtained from the Illinois Department of Transportation for the year 2004. Proximity to traffic was examined as a 4-level (0-150 m; 151-250 m; 251-500 m; >500 m) and a binary (0-250 m; >250 m) variable. The study outcomes include parent report of lifetime asthma, lifetime wheezing (wheezing or whistling), and current wheezing (in the last 12 months). The final analysis was based on data for 1925 children with complete data. The current analysis was approved by the Institutional Review Board of the University of Illinois at Chicago.

We performed multilevel logistic regression analyses using generalized estimating equations to estimate the odds ratio (OR) for the association of proximity to traffic with respiratory outcomes. In separate analyses, we examined interactions between the binary traffic-related exposure variable and potential moderators. All models accounted for clustering of children within neighborhoods.

Of the 1925 participants, 6.7%, 17.0%, and 6.0% had lifetime asthma, lifetime wheezing, and current wheezing, respectively, and 7.8% had parental history of asthma or allergies. In fully adjusted multilevel logistic regression analyses treating proximity to traffic as a 4-level variable, the odds of lifetime asthma were greater among children who lived between 0 and 150 m (OR, 2.39; 95% CI, 1.39-4.10) and 151 to 250 m (OR, 2.37; 95% CI, 1.34, 4.19) relative to those who lived more than 500 m from a primary traffic arterial (Table I). When we examined proximity to traffic as a binary exposure, the odds of lifetime asthma (OR, 2.21; 95% CI, 1.50-3.25) and lifetime wheezing (OR, 1.52; 95% CI, 1.17-1.97) were significantly higher for children who lived 0 to 250 m compared with those who lived more than 250 m from primary traffic arterials (Table I).

The associations of proximity to primary traffic arterials with lifetime asthma and lifetime wheezing, but not current wheezing, were modified by parental history of asthma or allergies (*P* for