

47XXY and 47XXX in Scleroderma and Myositis

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Objective. We undertook this study to examine the X chromosome complement in participants with systemic sclerosis (SSc) as well as idiopathic inflammatory myopathies.

Methods. The participants met classification criteria for the diseases. All participants underwent single-nucleotide polymorphism typing. We examined X and Y single-nucleotide polymorphism heterogeneity to determine the number of X chromosomes. For statistical comparisons, we used χ^2 analyses with calculation of 95% confidence intervals.

Results. Three of seventy men with SSc had 47,XXY ($P = 0.0001$ compared with control men). Among the 435 women with SSc, none had 47,XXX. Among 709 men with polymyositis or dermatomyositis (PM/DM), seven had 47,XXY ($P = 0.0016$), whereas among the 1783 women with PM/DM, two had 47,XXX. Of 147 men with inclusion body myositis (IBM), six had 47,XXY, and 1 of the 114 women with IBM had 47,XXX. For each of these myositis disease groups, the excess 47,XXY and/or 47,XXX was significantly higher compared with in controls as well as the known birth rate of Klinefelter syndrome or 47,XXX.

Conclusion. Klinefelter syndrome (47,XXY) is associated with SSc and idiopathic inflammatory myopathies, similar to other autoimmune diseases with type 1 interferon pathogenesis, namely, systemic lupus erythematosus and Sjögren syndrome.

There was no involvement of patients or the public in this work.

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Data are available on reasonable request.

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Significance & Innovation

- Autoimmune rheumatic diseases generally impact women more than men.
- A strong component to such bias in systemic lupus erythematosus and Sjögren syndrome is mediated by the sex chromosome complement.
- We find that the number of X chromosomes is also important in the sex bias of systemic sclerosis and idiopathic inflammatory myopathies.
- These diseases all share pathophysiology involving type 1 interferon pathways.

INTRODUCTION

Autoimmune diseases are more common among women and girls (1). The sex chromosomes are implicated in autoimmune sex bias (2). Turner syndrome predisposes to some (3) but not all autoimmune diseases (4). Acquired X chromosome monosomy of peripheral blood mononuclear cells is found in primary biliary cirrhosis (PBC) (5), as well as autoimmune thyroid disease and systemic sclerosis (6), but not in systemic lupus erythematosus (SLE) (7). Skewed X inactivation is found commonly among healthy women (8) but may be increased in some but not all autoimmune diseases (9–11).

Klinefelter syndrome (KS) (47,XXY) is found in 1 in 30 men with either SLE or Sjögren syndrome, whereas the known birth rate is about 1 in 500 live-born boys (12–14). Furthermore, women with 47,XXX are also found in excess among those with SLE and Sjögren syndrome (12). In contrast, no increase in X chromosome aneuploidy was found in rheumatoid arthritis (RA) or PBC (12,14). Genes that escape X inactivation (15) are candidates to mediate an X chromosome dose effect and include *CD40* (16), *TLR7* (17), and *CXorf21* (or *TASL*) (18,19).

We undertook the present study to determine whether X chromosome aneuploidies play a role in the sex bias of scleroderma (systemic sclerosis [SSc]) and idiopathic inflammatory myopathies, both of which are female biased with sex ratios of 5:1 and 3:1 (20,21).

PATIENTS AND METHODS

Participants. We studied cohorts with SSc or myositis that had undergone genome-wide single-nucleotide microarray genotyping (22–27). All participants met classification criteria for the disease in question (28–32). Diffuse cutaneous SSc was distinguished from limited cutaneous SSc by previously reported schema (23). The discovery cohorts were assembled from large international collaborative efforts, whereas the Japanese confirmatory myositis cohort was a nationwide effort involving

18 institutions (27). No participant was excluded except by failing to meet classification criteria or failure of genetic data in quality control. The control cohort was assembled at the Oklahoma Medical Research Foundation from healthy volunteers. Each control participant was verified not to have an autoimmune disease by a validated questionnaire and had no serum rheumatic disease autoantibodies (12,33). Participants in the discovery cohorts were of European ancestry, with matched control participants of this same origin. Ethics-committee-approved written informed consent was obtained from all participants at the site of recruitment, and the overall study was approved at the University of Oklahoma Health Sciences Center and Oklahoma Medical Research Foundation.

Sex chromosome complement determination. We used the Illumina GenomeStudio Software to examine b allele frequency plots of the X and Y chromosomes to determine the number of sex chromosomes, as previously reported (12–14).

Statistics. Descriptive statistics, including frequency and proportion, were calculated for categorical variables. χ^2 Tests were used to examine the association between two categorical variables when no more than 20% of cells had expected frequencies less than five and no one cell had an expected frequency less than one. Otherwise, Fisher's exact tests were used (34). Wilson type 95% binomial confidence interval (CI) was calculated for the proportion in each group because this test has better performance than other types of binomial confidence intervals (eg, Wald, Clopper-Pearson, and Agresti-Coull intervals; see ref. 35). All calculations were performed by using SAS 9.4 (SAS Institute, Inc.).

Patient and public involvement. There was no patient or public involvement in this study.

Ethical approval information. Ethics approval was obtained from local committees at the sites of recruitment of the participants. Thus, there were several dozen human investigation committees that approved this work.

TABLE 1. X chromosome aneuploidies found among participants with SSc

	46,XY	47,XXY	46,XX	47,XXX
Women				
SSc			435	0
Control			1345	0
Men				
SSc	67	3 (4.3%)*		
Control	1253	1 (0.08%)		

Abbreviation: SSc, systemic sclerosis.

* $P = 0.0001$ by Fisher's exact test (see text).

TABLE 2. X chromosome aneuploidies found among participants with PM/DM

	46,XY	47,XXY	46,XX	47,XXX
Women				
PM/DM			1781	2
Control			1345	0
Men				
PM/DM	702	7 (.99%)*		
Control	1253	1 (0.08%)		

Abbreviation: PM/DM, polymyositis or dermatomyositis.

* $P = 0.0016$ by Fisher's exact test (see text).

RESULTS

Of 505 participants with SSc, 70 (13.9%) self-identified as men. Among the men, 3 of 70 had KS (4.3%, 95% CI 0.89%-12.02% or 1 in 112 to 1 in 8; Table 1), substantially higher than the known live-birth rate (0.2% or 1 in 500) (36). We found a statistically significant increase in 47,XXY among the men with scleroderma compared with healthy control men (3 of 70 vs 1 of 1254, $P = 0.0001$ by Fisher's exact test). Among the 435 women, all had 46,XX (Table 1).

We next studied idiopathic inflammatory myopathies with polymyositis and dermatomyositis (PM/DM) grouped together (709 male and 1783 female participants). Among male participants with PM/DM, 7 of 709 had 47,XXY for a ratio of 1 in 101 (0.99%, 95% CI 0.48%-2.03% or 1 in 208 to 1 in 49; Table 2). Compared with the healthy controls, the men with PM/DM had a statistically significant increase in 47,XXY (7 of 709 [0.99%] compared with 1 of 1254 [0.08%], $P = 0.0043$ by Fisher's exact test). We found that two of the women with myositis had 47,XXX (Table 2), which does not differ from the expected birth rate of 1 in 1000 live-born girls or from the prevalence in the control women (36,37). We also studied an independent, confirmatory PM/DM cohort from Japan consisting of 430 women and 146 men. Of the 146 men, 4 had 47,XXY, whereas 2 of 430 women had 47,XXX. Thus, this cohort confirms the finding of X chromosome aneuploidies at an incidence of 6 in 576 samples of both sexes.

The clinical characteristics of the participants with PM/DM with X chromosome aneuploidies are shown in Table 3 and Supplementary Table 1. Five individuals had cancer-associated myositis, and only three of the men with KS had myositis-specific autoantibodies (Table 3).

Finally, we studied participants with inclusion body myositis (IBM). Among 147 men, we found six with 47,XXY (4.1%, 95% CI 1.5%-8.7% or 1 in 66 to 1 in 12; Table 4). The findings were statistically different from those for the controls ($P < 0.00001$ by Fisher's exact test). Among 114 women with IBM we found one with 47,XXX, which is similar in magnitude to our findings in SLE and Sjögren syndrome (12); further, the 95% CI for this ratio did not include the known prevalence at birth of ~1 in 1000 (0.0481-0.0016 or 1 in 166 to 1 in 21). The clinical features of these participants are given in Supplementary Table 2.

DISCUSSION

These findings are similar to those in SLE (13,38) and Sjögren syndrome (14) but distinct from RA and PBC. Thus, supernumerary X chromosomes are associated with some but not all sex-biased autoimmune diseases. We conclude that individual autoimmune diseases should be studied for X chromosome abnormalities. The present study adds to the diseases in which an X chromosome dose effect is present. Up to 15% of genes not in the pseudoautosomal regions escape X inactivation (39); therefore, the excess risk in individuals with 47,XXY and 47,XXX is informative concerning the differential risk associated with persons with 46,XX compared with 46,XY. That is, X chromosome biology mediates the sex bias of some autoimmune diseases.

Interferon plays a role in the diseases associated with supernumerary X chromosomes (12,40-43). Two genes lying on Xp, Toll-like receptor 7 (*TLR7*) and *CXorf21*, both contain risk alleles for autoimmune disease and escape X inactivation (17,44). *TLR7* signaling is initiated by binding of RNA, induces interferon- α as well as other cytokine production, and is involved in the pathogenesis of SLE (45). Our recent data show that the TASL

TABLE 3. Participants with PM and DM with X chromosome aneuploidies

Diagnosis	Sex chromosomes	Autoantibody	ILD	Cancer	Other
PM	47,XXY	Anti-HMGCR	No	No	
PM	47,XXY	Anti-Jo1	No	Lymphoma	
PM	47,XXY	IP result negative	No	Liver	
DM	47,XXY	Anti-U1RNP	No	Nasopharyngeal	Possible SSc overlap
DM	47,XXY	IP result negative	No	Metastatic	
DM	47,XXY	Anti-U1RNP/U2RNP	No	Lung cancer	
DM	47,XXY	Anti-MI2, anti-U1RNP	No	No	
PM	47,XXX	Multiple ^a	No	No	Sneddon syndrome, possible lupus overlap
PM	47,XXX	Anti-Pm-Scl	No	No	Asbestosis history

Abbreviations: DM, dermatomyositis; HMGCR, 3-hydroxy-3-methyl-glutaryl-coenzyme A reductase receptor; ILD, interstitial lung disease; IP, immunoprecipitation; PM, polymyositis; SSc, systemic sclerosis.

^a ANA (antinuclear antibodies), anti-RNP (ribonuclear protein), anti-Sm (Smith), anti-SCL70 (anti-topoisomerase), anti-DNA, and anti-FR (folate receptor).

TABLE 4. X chromosome aneuploidies found among participants with IBM

	46,XY	47,XXY	46,XX	47,XXX
Women				
IBM			113	1 (0.88%)
Control			1345	0
Men				
IBM	141	6 (4.1%)*		
Control	1253	1 (0.08%)		

Abbreviation: IBM, inclusion body myositis.

* $P < 0.00001$ by Fisher's exact test (see text).

(or *CXorf21*) protein regulates lysosomal pH as well as interferon and cytokine production in a sexually dimorphic manner (18,46). The SLE-associated haplotype results in a cis expression quantitative trait locus (eQTL) for *CXorf21*, which is an interferon response gene and whose protein product co-localizes with TLR7 (19). These independent studies find that sexually dimorphic expression of *TASL* in immune cells regulates innate immunity (18,19). Thus, X chromosome aneuploidies are found in sex-biased autoimmune diseases in which interferon is known to play a role but are not found in diseases without a known role of interferon.

The finding of increased KS among men with IBM is unexpected, in that this disease does not preferentially affect women (47). Although a neurodegenerative pathogenesis has been proposed (48), other studies support an autoimmune mechanism with autoantibodies (49). Highly differentiated CD8⁺ T cells infiltrating muscle tissue behave similarly to natural killer cells (50). Antigen-driven transformation of CD20⁺ B cells into clonal CD138⁺ plasma cells and CD19⁺ plasmablasts (48) led to the detection of circulating autoantibodies, along with identification of the target antigen as cytosolic 5'-nucleotidase 1A (NT5C1A) (51). Presence of this autoantibody may identify a subset of patients with IBM who are more likely to be female (51,52). Purified anti-NT5C1A antibodies cause modest myodegenerative changes with protein aggregation (53). Thus, these findings implicate an autoimmune etiology for a subset of patients. However, type 2, not type 1, interferon may be a part of IBM pathogenesis (43). Perhaps X chromosome aneuploidies are found among the patients with IBM with autoimmunity, but this has not been borne out in our results thus far.

There are limitations to the present study. Selection bias is a possibility, but patients were recruited without exclusion or inclusion criteria related to X chromosome aneuploidies. There is no clinical phenotype of either 47,XXY or 47,XXX that would lead to misclassification of participants with SSc or inflammatory myopathy. We have studied the heterogeneity of single-nucleotide polymorphisms on the X chromosome to determine 47,XXY or 47,XXX. This approach will miss patients who have a duplicated X chromosome from a nondisjunction in meiosis II, which occurs in about 15% of patients with KS. Thus, we might have missed some patients with X chromosome abnormalities, and our

numbers can be considered a lower estimate of X chromosome aneuploidies in these diseases.

We posit that our data demonstrate remarkable complexity in the female sex bias of inflammatory disease as well as sex-based differences in immune function. Thus far, diseases with evidence of pathological involvement of type 1 interferon, or type 2 interferon in the case of IBM (43), show an X chromosome gene dose effect. The present data extend these findings to SSc, PM/DM, and IBM. Further investigation is needed to fully define the mechanisms related to these findings.

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AUTHOR CONTRIBUTIONS

All authors were involved in drafting the article or revising it critically for important intellectual content, and all authors approved the final version to be published. Dr. Scofield had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study conception and design. Scofield, Miller.

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Analysis and interpretation of data. Scofield, Lewis, Cavitt, Kuri, Lamb.

REFERENCES

- Billi AC, Kahlenberg JM, Gudjonsson JE. Sex bias in autoimmunity. *Curr Opin Rheumatol* 2019;31:53–61.
- Libert C, Dejager L, Pinheiro I. The X chromosome in immune functions: when a chromosome makes the difference. *Nat Rev Immunol* 2010;10:594–604.
- Jorgensen KT, Rostgaard K, Bache I, Biggar RJ, Nielsen NM, Tommerup N, et al. Autoimmune diseases in women with Turner's syndrome. *Arthritis Rheum* 2010;62:658–66.
- Cooney CM, Bruner GR, Aberle T, Namjou-Khales B, Myers LK, Feo L, et al. 46,X,del(X)(q13) Turner's syndrome women with systemic lupus erythematosus in a pedigree multiplex for SLE. *Genes Immun* 2009;10:478–81.
- Invernizzi P, Miozzo M, Battezzati PM, Bianchi I, Grati FR, Simoni G, et al. Frequency of monosomy X in women with primary biliary cirrhosis. *Lancet* 2004;363:533–5.
- Invernizzi P, Miozzo M, Selmi C, Persani L, Battezzati PM, Zuin M, et al. X chromosome monosomy: a common mechanism for autoimmune diseases. *J Immunol* 2005;175:575–8.
- Invernizzi P, Miozzo M, Oertelt-Prigione S, Meroni PL, Persani L, Selmi C, et al. X monosomy in female systemic lupus erythematosus. *Ann N Y Acad Sci* 2007;1110:84–91.
- Shvetsova E, Sofronova A, Monajemi R, Gagalova K, Draisma HHM, White SJ, et al. Skewed X-inactivation is common in the general female population. *Eur J Hum Genet* 2019;27:455–65.
- Chitnis S, Monteiro J, Glass D, Apatoff B, Salmon J, Concannon P, et al. The role of X-chromosome inactivation in female predisposition to autoimmunity. *Arthritis Res* 2000;2:399–406.

10. Santiwatana S, Mahachoklertwattana P, Limwongse C, Khlairit P, Pongratanakul S, Roothumngong E, et al. Skewed X chromosome inactivation in girls and female adolescents with autoimmune thyroid disease. *Clin Endocrinol (Oxf)* 2018;89:863–9.
11. Kanaan SB, Onat OE, Balandraud N, Martin GV, Nelson JL, Azzouz DF, et al. Evaluation of X chromosome inactivation with respect to HLA genetic susceptibility in rheumatoid arthritis and systemic sclerosis. *PLoS One* 2016;11:e0158550.
12. Liu K, Kurien BT, Zimmerman SL, Kaufman KM, Taft DH, Kottyan LC, et al. X chromosome dose and sex bias in autoimmune diseases: increased prevalence of 47,XXX in systemic lupus erythematosus and Sjögren's syndrome. *Arthritis Rheumatol* 2016;68:1290–300.
13. Scofield RH, Bruner GR, Namjou B, Kimberly RP, Ramsey-Goldman R, Petri M, et al. Klinefelter's syndrome (47,XXY) in male systemic lupus erythematosus patients: support for the notion of a gene-dose effect from the X chromosome. *Arthritis Rheum* 2008;58:2511–7.
14. Harris VM, Sharma R, Cavett J, Kurien BT, Liu K, Koelsch KA, et al. Klinefelter's syndrome (47,XXY) is in excess among men with Sjögren's syndrome. *Clin Immunol* 2016;168:25–9.
15. Tukiainen T, Villani AC, Yen A, Rivas MA, Marshall JL, Satija R, et al. Landscape of X chromosome inactivation across human tissues. *Nature* 2017;550:244–8.
16. Lu Q, Wu A, Tesmer L, Ray D, Yousif N, Richardson B. Demethylation of CD40LG on the inactive X in T cells from women with lupus. *J Immunol* 2007;179:6352–8.
17. Souyris M, Cenac C, Azar P, Daviaud D, Canivet A, Grunenwald S, et al. TLR7 escapes X chromosome inactivation in immune cells. *Sci Immunol* 2018;3:eaap8855.
18. Harris VM, Harley IT, Kurien BT, Koelsch KA, Scofield RH. Lysosomal pH is regulated in a sex dependent manner in immune cells expressing CXorf21. *Front Immunol* 2019;10:578.
19. Odhams CA, Roberts AL, Vester SK, Duarte CS, Beales CT, Clarke AJ, et al. Interferon inducible X-linked gene CXorf21 may contribute to sexual dimorphism in systemic lupus erythematosus. *Nat Commun* 2019;10:2164.
20. D'Amico F, Skarmoutsou E, Mazzarino MC. The sex bias in systemic sclerosis: on the possible mechanisms underlying the female disease preponderance. *Clin Rev Allergy Immunol* 2014;47:334–43.
21. Meyer A, Meyer N, Schaeffer M, Gottenberg JE, Geny B, Sibilia J. Incidence and prevalence of inflammatory myopathies: a systematic review. *Rheumatology (Oxford)* 2015;54:50–63.
22. Marquez A, Kerick M, Zernakova A, Gutierrez-Achury J, Chen WM, Onengut-Gumuscu S, et al. Meta-analysis of immunochip data of four autoimmune diseases reveals novel single-disease and cross-phenotype associations. *Genome Med* 2018;10:97.
23. Mayes MD, Bossini-Castillo L, Gorlova O, Martin JE, Zhou X, Chen WV, et al. Immunochip analysis identifies multiple susceptibility loci for systemic sclerosis. *Am J Hum Genet* 2014;94:47–61.
24. Miller FW, Cooper RG, Vencovsky J, Rider LG, Danko K, Wedderburn LR, et al. Genome-wide association study of dermatomyositis reveals genetic overlap with other autoimmune disorders. *Arthritis Rheum* 2013;65:3239–47.
25. Miller FW, Chen W, O'Hanlon TP, Cooper RG, Vencovsky J, Rider LG, et al. Genome-wide association study identifies HLA 8.1 ancestral haplotype alleles as major genetic risk factors for myositis phenotypes. *Genes Immun* 2015;16:470–80.
26. Rothwell S, Cooper RG, Lundberg IE, Miller FW, Gregersen PK, Bowes J, et al. Dense genotyping of immune-related loci in idiopathic inflammatory myopathies confirms HLA alleles as the strongest genetic risk factor and suggests different genetic background for major clinical subgroups. *Ann Rheum Dis* 2016;75:1558–66.
27. Kochi Y, Kamatani Y, Kondo Y, Suzuki A, Kawakami E, Hiwa R, et al. Splicing variant of WDFY4 augments MDA5 signalling and the risk of clinically amyopathic dermatomyositis. *Ann Rheum Dis* 2018;77:602–11.
28. Subcommittee for Scleroderma Criteria of the American Rheumatism Association Diagnostic and Therapeutic Criteria Committee. Preliminary criteria for the classification of systemic sclerosis (scleroderma). *Arthritis Rheum* 1980;23:581–90.
29. Bohan A, Peter JB, Bowman RL, Pearson CM. Computer-assisted analysis of 153 patients with polymyositis and dermatomyositis. *Medicine (Baltimore)* 1977;56:255–86.
30. Rose MR, ENMC IBM Working Group. 188th ENMC International Workshop: inclusion body myositis, 2–4 December 2011, Naarden, the Netherlands. *Neuromuscul Disord* 2013;23:1044–55.
31. Riggs JE, Schochet SS Jr, Gutmann L, McComas CF, Rogers JS II. Inclusion body myositis and chronic immune thrombocytopenia. *Arch Neurol* 1984;41:93–5.
32. Hilton-Jones D, Miller A, Parton M, Holton J, Sewry C, Hanna MG. Inclusion body myositis: MRC Centre for Neuromuscular Diseases, IBM workshop, London, 13 June 2008. *Neuromuscul Disord* 2010;20:142–7.
33. Karlson EW, Sanchez-Guerrero J, Wright EA, Lew RA, Daltroy LH, Katz JN, et al. A connective tissue disease screening questionnaire for population studies. *Ann Epidemiol* 1995;5:297–302.
34. Bewick V, Cheek L, Ball J. Statistics review 8: qualitative data: tests of association. *Crit Care* 2004;8:46–53.
35. McGrath O BK. Binomial confidence intervals for rare events: importance of defining margin of error relative to magnitude of proportion. *Cornell University Library*. September 7, 2021. URL: <https://arxiv.org/abs/2109.02516>.
36. Nielsen J, Wohler M. Chromosome abnormalities found among 34,910 newborn children: results from a 13-year incidence study in Aarhus, Denmark. *Human Genet* 1991;87:81–3.
37. Tartaglia NR, Howell S, Sutherland A, Wilson R, Wilson L. A review of trisomy X (47,XXX). *Orphanet J Rare Dis* 2010;5:8.
38. Bentham J, Morris DL, Graham DS, Pinder CL, Tomblinson P, Behrens TW, et al. Genetic association analyses implicate aberrant regulation of innate and adaptive immunity genes in the pathogenesis of systemic lupus erythematosus. *Nat Genet* 2015;47:1457–64.
39. Carrel L, Willard HF. X-inactivation profile reveals extensive variability in X-linked gene expression in females. *Nature* 2005;434:400–4.
40. Chen S, Pu W, Guo S, Jin L, He D, Wang J. Genome-wide DNA methylation profiles reveal common epigenetic patterns of interferon-related genes in multiple autoimmune diseases. *Front Genet* 2019;10:223.
41. Muskardin TL, Niewold TB. Type I interferon in rheumatic diseases. *Nat Rev Rheumatol* 2018;14:214–28.
42. van den Hoogen LL, van Roon JAG, Mertens JS, Wienke J, Lopes AP, de Jager W, et al. Galectin-9 is an easy to measure biomarker for the interferon signature in systemic lupus erythematosus and antiphospholipid syndrome. *Ann Rheum Dis* 2018;77:1810–4.
43. Pinal-Fernandez I, Casal-Dominguez M, Derfoul A, Pak K, Plotz P, Miller FW, et al. Identification of distinctive interferon gene signatures in different types of myositis. *Neurology* 2019;93:e1193–204.
44. Morris DL, Sheng Y, Zhang Y, Wang YF, Zhu Z, Tomblinson P, et al. Genome-wide association meta-analysis in Chinese and European individuals identifies ten new loci associated with systemic lupus erythematosus. *Nat Genet* 2016;48:940–6.
45. Clancy RM, Markham AJ, Buyon JP. Endosomal Toll-like receptors in clinically overt and silent autoimmunity. *Immunol Rev* 2016;269:76–84.

46. Harris VM, Koelsch KA, Kurien BT, Harley IT, Wren JD, Harley JB, et al. Characterization of cxorf21 provides molecular insight into female-bias immune response in SLE pathogenesis. *Front Immunol* 2019;10:2160.
47. Lilleker JB, Vencovsky J, Wang G, Wedderburn LR, Diederichsen LP, Schmidt J, et al. The EuroMyositis registry: an international collaborative tool to facilitate myositis research. *Ann Rheum Dis* 2018;77:30–9.
48. Greenberg SA. Inclusion body myositis: clinical features and pathogenesis. *Nat Rev Rheumatol* 2019;15:257–72.
49. Herbert MK, Stammen-Vogelzangs J, Verbeek MM, Rietveld A, Lundberg IE, Chinoy H, et al. Disease specificity of autoantibodies to cytosolic 5'-nucleotidase 1A in sporadic inclusion body myositis versus known autoimmune diseases. *Ann Rheum Dis* 2016;75:696–701.
50. Allenbach Y, Chhara W, Rosenzweig M, Six A, Prevel N, Mingozzi F, et al. Th1 response and systemic treg deficiency in inclusion body myositis. *PLoS One* 2014;9:e88788.
51. Amlani A, Choi MY, Tarnopolsky M, Brady L, Clarke AE, Garcia-de la Torre I, et al. Anti-NT5c1A autoantibodies as biomarkers in inclusion body myositis. *Front Immunol* 2019;10:745.
52. Rietveld A, van den Hoogen LL, Bizzaro N, Blokland SL, Dahnrich C, Gottenberg JE, et al. Autoantibodies to cytosolic 5'-nucleotidase 1A in primary Sjögren's syndrome and systemic lupus erythematosus. *Front Immunol* 2018;9:1200.
53. Tawara N, Yamashita S, Zhang X, Korogi M, Zhang Z, Doki T, et al. Pathomechanisms of anti-cytosolic 5'-nucleotidase 1A autoantibodies in sporadic inclusion body myositis. *Ann Neurol* 2017;81:512–25.

APPENDIX A: MEMBERS OF THE MYOSITIS GENETICS CONSORTIUM

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