Sex differences in idiopathic inflammatory myopathies: insights from a large multicentre observational study

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Abstract

Objective

Preliminary evidence suggests sex-related variations in clinical and serological features in idiopathic inflammatory myopathies (IIMs), yet comprehensive cohort studies, particularly within the Italian population, remain scarce. The aim of this study is to assess sex differences in clinical phenotype, serological features, and disease activity, and to identify independent associated factors of disease activity in a well-characterised multi-centre Italian IIMs cohort.

Methods

A total of 228 IIMs patients from 5 tertiary Rheumatology Units across Italy were included. Demographic, clinical, serological, and treatment data were collected. Continuous variables are reported as mean ± SD or median (IQR), categorical variables as n (%). Comparisons between the 2 sexes were assessed by Student's t-test, Mann-Whitney U test, χ^2, or Fisher's exact test. Independent associated factors of disease activity (MYOACT) were assessed through multivariable linear regression analysis.

Results

Of 228 patients, 157 (68.9%) were female, and 71 (31.1%) were male. Age at diagnosis was similar between sexes. Males showed significantly higher disease activity measured by the MYOACT index. Multivariable analyses identified male sex, MDA5 autoantibody positivity, and greater disease damage extent as independent associated factors of higher disease activity, whereas dermatomyositis and polymyositis were associated with lower disease activity. Furthermore, MYOACT showed good predictive ability to define patients at risk for intensive care unit admission with an optimal threshold of 0.23.

Conclusions

These findings highlight relevant sex-related differences in clinical expression and may support more personalised management strategies.

Key word

sex, idiopathic inflammatory myopathies, MYOACT, disease activity, disease damage

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Received on September 22, 2025; accepted in revised form on December 19, 2025.

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Funding. PRIN 2022 funded this study [E53D23001200001].
Competing interests: none declared.

Introduction

Idiopathic inflammatory myopathies (IIMs) include a group of rare, heterogeneous autoimmune disorders characterised predominantly by chronic skeletal muscle inflammation and progressive weakness. These conditions frequently involve extra-muscular manifestations affecting organs such as the skin, lungs, and heart. IIMs include dermatomyositis (DM), polymyositis (PM), immune-mediated necrotising myopathy (IMNM), and antisynthetase syndrome (ASyS) (1). Although the etiopathogenesis of IIMs remains largely elusive, a multifactorial model involving a complex interplay of genetic susceptibility, environmental triggers and dysregulated immune responses is considered the key player (2, 3). Lastly, several IIM subtypes are epidemiologically linked to an increased risk of malignancy, particularly during the peridiagnostic period (4, 5).

Gender medicine is a multidisciplinary field investigating the influence of biologic (sex-based) and sociocultural (gender-based) differences on health and disease. It includes studies aimed at understanding how these factors may affect the development, diagnosis, treatment, as well as overall health outcomes (6). However, current evidence on sex-related differences in IIMs remains limited. Notably, data from the international COVID-19 vaccination in autoimmune disease e-survey, based on patient-reported outcomes, indicated that females with IIMs are more likely to report autoimmune multimorbidity and higher fatigue levels than males (7). Preliminary data from a small Italian cohort suggest that women exhibit greater muscle and cutaneous involvement (8). However, extensive data on gender and sex-based differences in disease characteristics, progression, and outcomes in male and female IIMs are still lacking, underscoring the need for further research in this area.

To our knowledge, no published study has comprehensively compared clinical, serological, and extra-muscular features between male and female patients with IIMs worldwide. In this context, the present study aims to elucidate the role of sex in shaping the clinical, serological, and histopathological characteristics of IIMs in a large, multicentre cohort. By providing detailed sex-disaggregated data, our analysis seeks to advance understanding of sex-dependent differences and suggest the development of more personalised diagnostic and therapeutic strategies in IIMs.

Materials and methods

Patients, study design and assessment of disease-related features

The study population included 228 IIM patients from 5 tertiary Rheumatologic Units across Italy with a high experience in the management of this disease. All patients fulfilled the 2017 EULAR/ACR classification criteria for adult IIM (9) and were consecutively enrolled from January 1, 2024, to December 31, 2024. This study was designed as a multicentre, cross-sectional analysis. Clinical, serological, and activity/damage data were obtained from routine follow-up visits conducted in established IIM clinics and were subsequently retrieved retrospectively from medical records for inclusion in the cross-sectional dataset. Because assessments were extracted from a single clinical evaluation per patient, no longitudinal follow-up was performed. The inclusion criteria required the availability of comprehensive clinical, serological, and disease activity and damage data at the time of assessment. No additional exclusion criteria were applied, except for the absence of essential data needed for the analysis. The Ethics committee of the coordinator of the study approved the protocol (Prot. 0006690/i) following the Good Clinical Practice Guidelines and the Declaration of Helsinki. Written informed consent was obtained from all the patients before enrolment.

All patients were subjected to full history taking, clinical examination, and relevant laboratory and radiological investigations related to their IIMs. At study entry, the following demographic and clinical data were systematically collected (Table I): age, sex, ethnicity, disease duration (in years), and delay of diagnosis (in years), defined as the time between the onset of the first symptom

related to IIM and the diagnosis, borrowing the definition of rheumatoid arthritis (10). Muscular involvement was assessed by the presence of myositis, serum levels of creatinine phosphokinase (CPK), aldolase, and troponin, as well as findings from muscle magnetic resonance imaging (MRI). Clinical features included the presence of ILD and its radiological patterns, intensive care unit (ICU) admission, emphysema, pneumomediastinum (11), and diaphragm involvement. Additional data included the presence of arthritis and joint deformities, and seropositivity for rheumatoid factor (RF) and anti-citrullinated protein antibodies (ACPA). Other systemic and cutaneous manifestations were also recorded, including dysphagia, Raynaud's phenomenon (RP), fever, mechanic's hands, Gottron's papules and sign, shawl sign, V sign, heliotrope rash, cutaneous ulcers, calcinosis, periungual erythema, and the presence of malignancy. IIM clinical subgroups [DM, PM, ASyS, IMNM, and myositis overlap syndrome (OL)] were also recorded for all patients. Due to the small sample sizes in certain phenotypes, no sex-stratified analyses were conducted within each subgroup. However, subtype distribution was included in the descriptive analyses (Table I) and incorporated as a covariate in univariable and multivariable regression models (Table II). Myositis OL syndromes were recorded as a single aggregated category, as specific underlying connective tissue diseases (*e.g.*, Sjögren's syndrome, systemic sclerosis, rheumatoid arthritis, systemic lupus erythematosus) were not individually classified. Autoantibody profiles were systematically recorded, including antinuclear antibodies (ANA), extractable nuclear antigens (ENA), and myositis-specific (MSA) and myositis-associated (MAA) antibodies. The assessment of disease activity of extra-muscular organ systems and muscles was performed with the Myositis Disease Activity Assessment Tool (MDAAT), a combined tool that includes the Myositis Disease Activity Assessment VAS (MYOACT) and the Myositis Intention to Treat Activities Index (MITAX) (12). The manual Muscle test (MMT) was assessed to measure muscle strength as part of the physical examination (13). The Physician Global Assessment (PhGA) was used as an overall rating of the disease activity related to myositis. The damage was assessed using the Myositis Damage Index (MDI) extent and severity indexes (12). Disease activity (MYOACT, MITAX, PhGA, MMT) and disease damage (MDI extent and severity) were assessed cross-sectionally at a single time point, corresponding to the clinical visit at which each patient was included in the study. No longitudinal assessments of disease activity or damage were performed. The current therapy at the time of clinical assessment was also recorded.

All the researchers followed a unified assessment protocol grounded in standardised clinical definitions and vali-

Table I. Main demographic and disease-related characteristics of the study population.

Variable	Overall (n = 228)	Females (n = 157)	Males (n = 71)	*p*-value
Mean age at diagnosis ± SD (years)	53.92 ± 14.57	53.72 ± 15.54	54.36 ± 12.21	0.217
Diagnostic delay, N (%)	103 (46.6)	73 (47.7)	30 (44.1)	0.621
Disease duration ± SD (years)	8.46 ± 7.42	8.82 ± 8.18	7.66 ± 5.36	0.278
Autoantibodies				
ANA, N (%)	161 (72.2)	115 (74.7)	46 (66.7)	0.217
ENA, N (%)	111 (50.2)	77 (51)	34 (48.6)	0.738
MAA				
SSA, N (%)	**69 (30.8)**	**56 (36.6)**	**13 (18.3)**	**0.006**
Ro52, N (%)	**59 (26.3)**	**47 (30.7)**	**12 (16.9)**	**0.029**
MSA				
MDA5, N (%)	19 (8.5)	11 (7.2)	8 (11.3)	0.308
Jo1, N (%)	89 (39.7)	59 (38.6)	30 (42.3)	0.599
HMGCR, N (%)	1 (0.4)	0 (0)	1 (1.4)	0.317
IIM subgroups				
DM	73 (33)	50 (32.7)	23 (33.8)	0.867
PM	30 (13.6)	23 (15)	7 (10.3)	0.430
ASyS	106 (48)	71 (46.4)	3 (51.5)	0.487
IMNM	8 (3.6)	6 (3.9)	2 (2.9)	1.000
OL	4 (1.8)	3 (1.9)	1 (1.4)	1.000
Muscular involvement				
Elevated CPK, N (%)	**131 (58)**	**83 (53.2)**	**48 (68.6)**	**0.030**
Increased aldolase, N (%)	69 (33.7)	46 (32.9)	23 (35.4)	0.722
Dysphagia, N (%)	49 (21.6)	30 (19.2)	19 (26.8)	0.201
Lung involvement				
ILD (HRCT), N (%)	137 (60.1)	92 (58.6)	45 (63.4)	0.495
ICU admission, N (%)	4 (1.8)	1 (0.6)	3 (4.2)	0.092
Articular involvement				
Arthritis, N (%)	106 (46.9)	75 (48.4)	31 (43.7)	0.509
Cutaneous involvement				
Mechanic's hands, N (%)	56 (24.6)	40 (25.5)	16 (22.5)	0.633
Gottron's papules, N (%)	56 (24.6)	38 (24.2)	18 (25.4)	0.852
Heliotrope rash, N (%)	43 (18.9)	33 (21)	10 (14)	0.053
Other				
Malignancy, (%)	9 (3.9)	6 (3.8)	3 (4.2)	1.000
Fever, N (%)	31 (13.7)	19 (12.2)	12 (16.9)	0.337
Damage assessment				
MDI extent, median (IQR)	0.05 (0.06)	0.05 (0.07)	0.04 (0.06)	0.806
MDI severity, median (IQR)	0.05 (0.05)	0.05 (0.07)	0.06 (0.04)	0.244
Activity assessment				
MITAX, median (IQR)	0.08 (0.16)	0.08 (0.18)	0.085 (0.15)	0.832
MYOACT, median (IQR)	**0.13 (0.14)**	**0.12 (0.14)**	**0.15 (0.12)**	**0.019**
MMT, median (IQR)	146 (16)	145 (18)	148 (12)	0.198
PhGA, median (IQR)	2 (2)	2 (2)	2 (1)	0.426

ANA: antinuclear antibodies; ASyS: antisynthetase syndrome; ATS: anti-topoisomerase I, ENA: extractable nuclear antigen antibodies; Ro/SSA: Sjögren's-syndrome-related antigen A; MDA5: anti-melanoma differentiation-associated gene 5; Jo1: anti-histidyl-tRNA synthetase; Ro52: anti-Ro52/TRIM21; HMGCR: anti-3-hydroxy-3-methylglutaryl-CoA reductase; ILD: interstitial lung disease; ICU: intensive care unit; IMNM: immune mediated necrotising myositis; MITAX: Myositis Intention to Treat Activity Index; MYOAC: Myositis Disease Activity Assessment Tool; MMT: manual muscle testing; PhGA: physician's global assessment. OL: overlap syndrome.

dated evaluation tools. Data collection was centrally reviewed for accuracy and completeness. Any inconsistencies or missing information were promptly addressed through direct coordination with the contributing centres. All assessments were performed by experienced clinicians.

Statistical analysis

Data was collected and analysed using the statistical package SPSS version 20 (SPSS Inc., Chicago, IL, USA). The Shapiro-Wilk test was used to assess the normality distribution of continuous variables. Mean and standard deviation (SD) or median and interquartile range (IQR) as necessary were employed for quantitative variables, while qualitative variables are reported as absolute frequencies and percentages. Cross-tables were analysed by Pearson's Chi-square test, and contingency tables with dimensions greater than 2×2 were also followed by post-hoc analysis with adjusted residuals, while differences in mean/median between groups were tested with the Mann-Whitney U test. Univariable linear regression analyses was conducted to identify potential factors associated with disease activity (MYOACT). The purposeful selection process of covariables started by a univariate analysis of each variable; any variable having a significant univariate test and/or a clinical relevance was considered a possible candidate for the multivariate analysis. At the end of this multistep process of deleting and refitting, the multivariate models were built, and regression coefficients β with 95% CI were reported for significant associations with MYOACT. The final multivariable model included sex as the main exposure and was adjusted for age at diagnosis, disease duration, and IIM subtype distribution. Receiver operating characteristic (ROC) curves were built to evaluate the predictivity of MYOACT on the likelihood of predicting admission to the ICU. The best cut-off for ROC curves was calculated with the Youden's index (14) as previously described. Missing data were handled using a complete-case analysis approach. Variables with substantial missingness were not entered into multivariable models, and no imputation procedures were applied. Statistical significance was defined as a two-tailed p-value <0.05.

Table II. Current treatment of the study population.

MTX, N (%)	51 (22.4)	35 (22.3)	16 (22.5)	0.968
MMF, N (%)	41 (18)	28 (17.8)	13 (18.3)	0.931
AZA, N (%)	25 (11)	21 (13.4)	4 (5.6)	0.083
IVIG, N (%)	7 (3.1)	4 (2.5)	3 (4.2)	0.680
RTX, N (%)	14 (6.1)	11 (7)	3 (4.2)	0.558
CYS A, N (%)	**35 (15.4)**	**17 (10.8)**	**18 (25.4)**	**0.005**
GC daily, median (IQR)	1 (5)	1 (5)	5 (12.5)	0.060
Tacrolimus, N (%)	1 (0.4)	1 (0.6)	0 (0)	1.000
Jaki, N (%)	5 (2.2)	5 (3.2)	0 (0)	0.328
HCQ, N (%)	44 (19.3)	32 (20.4)	12 (16.9)	0.537
CYC, N (%)	1 (0.4)	1 (0.6)	0 (0)	1.000

MTX: methotrexate; MMF: mycophenolate mofetil; AZA: azathioprine; IVIG: intravenous immunoglobulin; RTX: rituximab; CYS A: cyclosporine A; Jaki: Janus kinase inhibitor: HCQ: hydroxychloroquine; CYC: cyclophosphamide; MITAX: Myositis Intention to Treat Activity Index; MYOAC: Myositis Disease Activity Assessment Tool; MMT: manual muscle testing; PhGA: physician's global assessment.

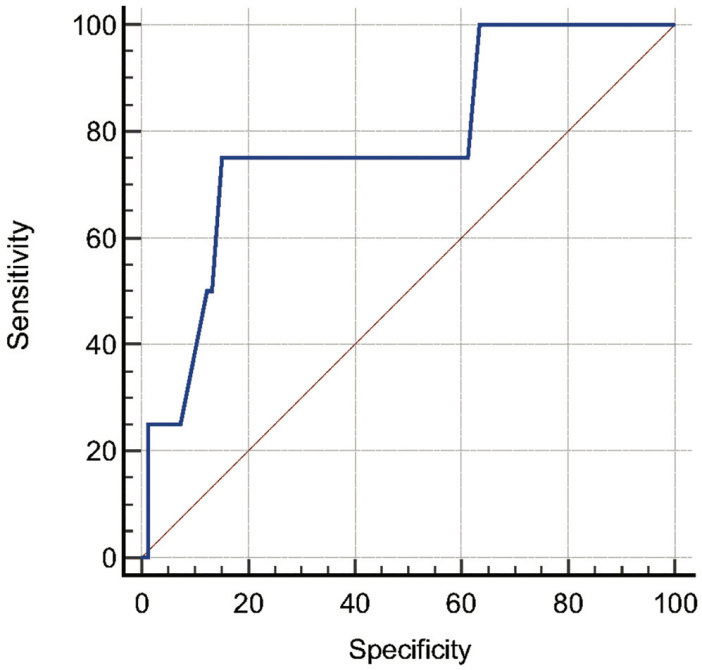

Fig. 1. Receiver operator characteristic (ROC) curve of MYOACT score in predicting the admission to ICU.

Results

Descriptive characteristics of the evaluated IIMs patients

Table I provides a sex-based analysis of the main demographic and disease-related features for all patients. The study cohort included 228 patients with IIMs, of whom 157 were female and 71 were male. The majority of patients were Caucasian (97.8%), with no significant difference in ethnicity distribution between females and males (p=0.207). The mean age at diagnosis was similar between females (53.72±15.54 years) and males (54.36±12.21 years; p=0.217).

The prevalence of ANA and ENA was comparable between sexes (ANA: 74.7% females *vs*. 66.7% males, p=0.217; ENA: 51% females *vs*. 48.6% males, p=0.738). However, female patients showed a significantly higher frequency of SSA antibodies (36.6% *vs*. 18.3%, p=0.006) and Ro52 antibodies (30.7% *vs*. 16.9%, p=0.029) compared to males. Laboratory markers showed that higher CPK levels were more prevalent in males than in females (68.6% *vs*. 53.2%, respectively; p=0.030).

Diagnostic delay did not differ sig-

Table III. Univariable and multivariable regression analyses (MYOACT as dependent variable).

	Univariable analyses			Multivariable analyses		
Variables	**Coeff. β**	**95% CI**	***p*-value**	**Coeff. β**	**95% CI**	***p*-value**
Gender (male)	**0.046**	**0.013 to 0.078**	**0.006**	**0.037**	**0.006 to 0.056**	**0.014**
Age at diagnosis	**0.002**	**0.001 to 0.003**	**0.0015**	0.001	0.000 to 0.002	0.060
Disease duration	0.000	-0.002 to 0.002	0.821	0.001	-0.001 to 0.003	0.225
MAA						
ANA	0.016	-0.018 to 0.049	0.358			
ENA	0.003	-0.028 to 0.034	0.843			
MAA						
SSA/Ro	-0.007	-0.041 to 0.027	0.680			
Ro52	0.007	-0.028 to 0.042	0.699			
MDA5	**0.068**	**0.011 to 0.125**	**0.019**	**0.091**	**0.044 to 0.138**	**0.0001**
Jo1	0.009	-0.022 to 0.041	0.569			
IIM subgroup						
DM	-0.016	-0.049 to 0.017	0.352	**-0.063**	**-0.117 to -0.009**	**0.022**
PM	-0.030	-0.075 to 0.015	0.195	**-0.079**	**-0.138 to -0.020**	**0.009**
ASyS	0.017	-0.014 to 0.048	0.284	-0.032	-0.084 to 0.021	0.233
IMNM	0.023	-0.004 to 0.050	0.096			
OL	0.015	-0.100 to 0.131	0.796			
Muscular involvement						
Elevated CPK, N (%)	**0.041**	**0.010 to 0.071**	**0.010**	0.023	-0.001 to 0.048	0.062
Elevated aldolase, N (%)	0.27	-0.006 to 0.061	0.111			
Dysphagia	0.014	-0.023 to 0.051	0.454			
Lung involvement						
3ILD (HRCT)	0.027	-0.004-0.58	0.089			
ICU admission	**0.125**	**0.010 to 0.239**	**0.033**	0.059	-0.028 to 0.14	0.182
Arthritis	0.003	-0.027 to 0.034	0.219			
Cutaneous involvement						
Mechanic's hands	0.019	-0.017 to 0.054	0.301			
Gottron's papules	0.014	-0.021 to 0.050	0.428			
Heliotrope rash	0.008	-0.032 to 0.047	0.702			
Other						
Malignancy	0.016	-0.072 to 0.104	0.724			
Fever	**0.047**	**0.002 to 0.092**	**0.041**	0.016	-0.019 to 0.051	0.367
Damage assessment						
MDI extent, median (IQR)	**0.870**	**0.717 to 1.022**	**0.0001**	**0.834**	**0.684 to 0.981**	**0.0001**
MDI severity, median (IQR)	0.020	-0.016 to 0.056	0.284			

ANA: antinuclear antibodies; ATS: anti-topoisomerase I, ENA: extractable nuclear antigen antibodies; Ro/SSA: Sjögren's-syndrome-related antigen A; La/SSB: Sjögren's-syndrome-related antigen B; ACA: anti-centromere antibodies; Sm: anti-Smith antibodies; RN: anti-ribonucleoprotein antibodies; Mi2A: anti-Mi-2 alpha; Mi2B; anti-Mi-2 beta; TIF1-γ: anti-transcription intermediary factor 1 gamma; MDA5: anti-melanoma differentiation-associated gene 5; NXP: anti-nuclear matrix protein 2; SAE: anti-small ubiquitin-like modifier activating enzyme (SAE1/2); Ku: anti-Ku antibodies; PL7: anti-threonyl-tRNA synthetase; PL12: anti-alanyl-tRNA synthetase; JO1: anti-histidyl-tRNA synthetase (Jo-1); EJ: anti-glycyl-tRNA synthetase; OJ: anti-isoleucyl-tRNA synthetase; Ro52: anti-Ro52/TRIM21; cN1A: anti-cytosolic 5´-nucleotidase 1; SRP: anti-signal recognition particle; HMGCR: anti-3-hydroxy-3-methyl-glutaryl-CoA reductase; PM/Scl: anti-PM/Scl complex (PM/Scl-75 and/or PM/Scl-100); ANCA: anti-neutrophil cytoplasmic antibodies; Ro60: anti-Ro60; KS: anti-asparaginyl-tRNA synthetase; Zo: anti-phenylalanyl-tRNA synthetase; ILD: interstitial lung disease; NSIP: non-specific interstitial pneumonia; UIP: usual interstitial pneumonia; OP: organising pneumonia; LID: lymphocytic interstitial pneumonia; NSIP/OP: mixed NSIP and organising pneumonia pattern; ICU: intensive care unit; TLC: total lung capacity; RF: rheumatoid factor; ACPA: anti-citrullinated antibodies; MTX: methotrexate; MMF: mycophenolate mofetil; AZA: azathioprine; IVIG: intravenous immunoglobulin; RTX: rituximab; CYS A: cyclosporine A; Jaki: Janus kinase inhibitor: HCQ: hydroxychloroquine; CYC: cyclophosphamide; MITAX: Myositis Intention to Treat Activity Index; MYOAC: Myositis Disease Activity Assessment Tool; MMT: Manual Muscle Testing; PhGA: physician's global assessment.

nificantly between females and males. Abnormal muscle MRI findings, cutaneous manifestations, and ILD prevalence were similar across sexes. Disease damage indexes (MDI extent and severity), as well as activity indexes, including MITAX, MMT, and PhGA, did not show significant differences between females and males. Notably, disease activity measured by the MYOACT was significantly higher in males (median 0.15, IQR 0.12) compared to females (median 0.12, IQR 0.14; p=0.019). Extended baseline clinical and serological characteristics of the study population by sex are reported in Supplementary Table S1.

Current treatments were comparable between sexes, except for cyclosporine A, which was more frequently used in males (25.4% *vs*. 10.8%, p=0.005) (Supplementary Table S2).

Factors associated with higher disease activity as assessed by MYOACT

Among the variables that reached statistical significance, we selected MYOACT as the outcome measure, since it is a validated clinical tool that captures global disease activity across all organ systems involved in IIMs. In contrast, serum CPK levels only reflect muscle injury and fail to provide information on extra-muscular manifestations (13).

Therefore, MYOACT is considered the most appropriate and clinically meaningful parameter for assessing overall disease activity in our cohort.

Univariable linear regression analysis was performed to identify factors associated with disease activity measured by MYOACT in the IIM cohort (Table II). Male sex was significantly associated with higher MYOACT scores (β=0.046, 95% CI: 0.013 to 0.078; p=0.006), indicating greater disease activity in males compared to females. Age at diagnosis was positively associated with disease activity (β=0.002; 95% CI: 0.001 to 0.003; p<0.001). The extent of disease damage as measured by the MDI extent score showed a strong positive correlation (β=0.870; 95% CI: 0.717 to 1.022; p<0.0001), while MDI severity was not significant. Among autoantibodies, positivity for MDA5 (β=0.068, 95% CI: 0.011 to 0.125; p=0.019) showed a significant positive association with MYOACT. Laboratory markers revealed that elevated creatine phosphokinase (CPK) levels were positively correlated with disease activity (β=0.041, 95% CI: 0.010 to 0.071; p=0.010). Clinically, admission to the ICU was significantly associated with increased disease activity (β=0.125, 95% CI: 0.010 to 0.239; p=0.033). Fever presence was also significantly associated with higher MYOACT scores (β=0.047, 95% CI: 0.002 to 0.092; p=0.041). Full univariable regression results are reported in Supplementary Table S3.

A fully adjusted multivariable regression model, incorporating sex, age at diagnosis, disease duration, and IIM subtype distribution, identified several variables that remained independently associated with disease activity as measured by the MYOACT score.

Male sex was significantly associated with higher MYOACT scores (β=0.037, 95% CI 0.006–0.056; p=0.014), confirming a robust sex-related effect on disease activity, after adjustment for disease chronicity and clinical phenotype. MDA5 antibody positivity showed an even stronger association with MYOACT after adjustment (β=0.091, 95% CI 0.044–0.138; p<0.0001), suggesting that positive patients for this antibody have higher disease activity. The MDI extent score remained the strongest associated factor of disease activity (β=0.834, 95% CI 0.684–0.981; p<0.0001), indicating that greater damage extent is linked with increased disease activity.

Conversely, DM (β=-0.063; 95% CI -0.117 to -0.009, p=0.022) and PM (β=-0.079; 95% CI -0.138 to -0.020, p=0.009) were associated with significantly lower MYOACT values, while ASyS did not show a significant independent association (β=-0.032, 95% CI -0.084 to 0.021, p=0.233). Age at diagnosis (β=0.001, 95% CI 0.000 to 0.002, p=0.060), disease duration (β=0.001, 95% CI -0.001 to 0.003, p=0.225), CPK (β=0.023, 95% CI -0.001 to 0.048, p=0.062), ICU admission (β=0.059, 95% CI -0.028 to 0.14, p=0.182), and fever (β=0.016, 95% CI -0.019 to 0.051, p=0.367) were not independently associated with disease activity in the adjusted model. Furthermore, we performed a ROC curve in order to investigate the predictivity of the MYOACT score on the likelihood of predicting admission to the ICU (Fig. 1). The area under the ROC curve was 0.78 (95% CI: 0.72–0.83, p<0.04) for the MYOACT score. The analysis of the ROC curve showed that the optimal threshold for the MYOACT score was 0.23 and provided a sensitivity of 75% and a specificity of 85%.

Discussion

This multicentre, real-world study represents one of the most comprehensive evaluations, to date, of sex-related differences in IIMs, a group of rare autoimmune diseases, within the well-characterised Italian cohort, and consistent with established epidemiological results in other autoimmune diseases (15-19), we showed how some clinical or serological biomarkers have a different expression between the 2 sexes. The mean age at disease onset (53.9±14.6 years) aligns with previous reports (20), reinforcing the generalisability of our population. Despite higher disease frequency in females, males showed significantly higher disease activity as measured by the MYOACT index, suggesting potentially more aggressive disease phenotype and/or distinct immunopathogenic mechanisms. Males may experience higher disease activity due to immune, hormonal, or behavioural differences, thus tailoring the management of the disease within higher-risk groups. This may reflect a sex-related impact on inflammatory pathways rather than on cumulative damage or muscle strength. In various autoimmune diseases, such as systemic sclerosis, systemic lupus erythematosus, and primary Sjögren's syndrome, male patients consistently present with a more aggressive disease phenotype, including higher disease activity, greater organ involvement, and worse clinical outcomes compared to females (21-28).

In our study, by descriptive statistical analysis, we observed that females had a significantly higher prevalence of SSA and Ro52 autoantibodies, suggesting their potential role in the pathogenesis of the disease (29). On the contrary, we observed that male patients displayed higher disease activity as measured by the MYOACT index, which was not reflected in other activity indices (MITAX, MMT, PhGA) or damage scores (MDI severity). This discrepancy may be explained by the broader construct of the MYOACT, which reflects global multisystem disease activity, while MITAX is mainly driven by treatment decisions, and MMT focuses solely on muscle strength. Our findings mirror what already observed in other autoimmune diseases. In systemic lupus erythematosus, systemic measures such as SLEDAI and treatment-oriented tools like BILAG have shown divergent validity and sensitivity to change (30). Similarly, in spondyloarthritis, BASDAI and ASDAS often provide discordant assessments (31). These parallels support that multidimensional global indices may better capture systemic disease burden, while organ-specific or treatment-driven measures can underestimate subgroup differences.

In this study, univariable linear regression analysis showed key clinical and immunological factors associated with higher disease activity in IIMs, as measured by the MYOACT score, including male sex, age at diagnosis,

positive MDA5 antibody status, ICU admission, elevated CPK, and fever.

Notably, male sex emerged as a significant predictor of higher disease activity, highlighting potential sex-related differences in disease severity. This finding aligns with the general observation in autoimmune diseases that despite the female predominance is higher, males may experience more aggressive disease phenotypes. In contrast, DM and PM were independently associated with lower MYOACT scores, indicating that these subtypes exhibited comparatively lower global disease activity in our cohort.

Although IIMs comprise heterogeneous clinical subgroups, subgroup-specific sex analyses were not performed due to limited sample size in some categories. Instead, subtype distribution was included in the regression models to control for potential confounding. Notably, male sex remained independently associated with higher MYOACT scores even after adjustment for age, disease duration, and subtype, supporting a true sex-related difference in disease activity. Overlap connective tissue diseases were also documented, as they may influence serological profiles, and this is acknowledged as a study limitation.

Aging is known to drive immunosenescence, characterised by impaired immune cell function, alterations in self-antigens, and a narrowed receptor repertoire, all of which increasing vulnerability to autoimmune diseases. Furthermore, senescent immune cells and their secretory profiles exacerbate chronic inflammation and tissue injury, while mitochondrial dysfunction accelerates the accumulation of these harmful cells, thereby promoting disease progression (32).

MDA5 autoantibodies were significantly associated with increased MYOACT scores, consistent with their known link to aggressive clinical phenotypes. Anti-MDA5 dermatomyositis is a rare but severe subtype characterised by a high prevalence of ILD, particularly rapidly progressive ILD (RP-ILD), which significantly increases mortality risk, with male sex identified as a strong predictor of poor prognosis (33). Notably, a prospective study of 251 patients with IIM positive for anti-MDA5 antibodies showed that the 6-month mortality risk increases with age, rising sharply in male patients over 54 years old, whereas in female patients the risk increases more gradually and linearly (33). Our multivariable model confirmed that male sex and anti-MDA5 positivity were independent factors significantly associated with a more active disease phenotype. Importantly, after adjusting for age, disease duration, and IIM subtype distribution, male sex remained independently associated with higher MYOACT scores, supporting a true sex-related difference in disease activity. These findings strengthen the hypothesis that males may exhibit a more inflammatory or aggressive disease phenotype despite the overall female predominance of IIM. Such sex-related differences may serve as an important biomarker panel for identifying patients at risk of more severe or treatment-resistant disease who may benefit from closer surveillance or early aggressive therapy.

Clinically, ICU admission and the presence of fever were significant markers of heightened disease activity. Importantly, the ROC curve analysis showed that MYOACT has a good discriminatory ability in predicting ICU admission, with an area under the curve of 0.78. The identified MYOACT threshold of 0.23 offers a practical cut-off with balanced sensitivity (75%) and specificity (85%), suggesting that MYOACT may serve as an effective early warning indicator for critical disease progression requiring intensive care to interfere with the clinical deterioration. A retrospective study aimed to delineate the clinical features, the mortality risk factors, as well as to evaluate outcomes of IIMs patients admitted to a medical ICU, over an 8-year period, confirming the mortality risk associated with conditions such as dermatomyositis, acute respiratory failure, and opportunistic infections. In this study, non-survivors after ICU admission were more often male, diagnosed with DM or clinically amyopathic DM, complicated by ILD, and treated with methylprednisolone pulse (34).

The extent of disease damage, quantified by the MDI extent score, showed a significant association with MYOACT score, reinforcing the link between cumulative tissue injury and ongoing disease activity. This relationship supports a model in which chronic persistent inflammation increases the damage accrual and loss of function in IIMs. Notably, the MDI severity score did not reach statistical significance, suggesting that the overall breadth of damage may have greater relevance to disease activity assessments than the severity of localized lesions. This distinction underscores the importance of evaluating the full extent of damage to better assess patient risk and guide therapeutic priorities. Finally, DM and PM showed significantly lower MYOACT values in the adjusted model, indicating comparatively lower global disease activity relative to other IIM subtypes. For DM, this apparent "protective" effect is largely explained by the fact that the highly aggressive MDA5-positive phenotype, characterised by marked systemic inflammation, rapidly progressive ILD, and increased short-term mortality, was modelled separately (35). PM, on the other hand, typically exhibits less multisystem inflammatory involvement than other IIM subtypes, which likely may account for its lower adjusted MYOACT scores (36).

In cross-sectional observational studies, the lack of preassigned therapy can confound outcome-exposure associations, risking incorrect conclusions. Thus, to avoid the well-known "confounding by indication bias" in our study, we did not assess the impact of different treatments on IIM patients (37, 38).

We are aware of some limitations of this study. The cohort consisted predominantly of Caucasian patients from Italy, which may limit the generalisability of the findings to populations of different ethnic backgrounds or geographic regions. Some IIM subgroups (IMNM, OL) were represented by very small numbers. Therefore, the study is underpowered to detect sex-related differences within these rare phenotypes, and no firm conclusions can be drawn for these subgroups. Missing data were

handled with a complete-case approach without imputation, with minimal impact given the low proportion of missing values for the main clinical and serological variables. Patient-reported outcomes and quality of life measures were not included, limiting insight into symptom burden and functional impact from the patient perspective. Another limitation is that disease activity and damage were assessed only cross-sectionally, at the time of study inclusion; therefore, longitudinal changes over time could not be evaluated. Moreover, due to the inherent "confounding by indication bias" of cross-sectional designs, treatment effects and sex-specific treatment responses could not be reliably assessed. Similarly, sex-related differences in remission could not be assessed, as time to remission and remission status were not systematically or uniformly collected across centres, and given the cross-sectional design, analysing remission outcomes would have introduced substantial bias. Future prospective studies with standardised and universally validated remission criteria are needed to address this question. Furthermore, myositis OL syndromes were recorded only as a single aggregated category, and specific underlying diagnoses (such as Sjögren's syndrome, systemic sclerosis, rheumatoid arthritis, or systemic lupus erythematosus) were not individually classified, limiting a more granular assessment of overlap conditions. Lastly, although overlap connective tissue diseases were recorded, their possible influence on SSA/Ro52 patterns remains a residual confounder, as specific connective tissue diseases were not individually classified.

On the other hand, this study constitutes one of the most comprehensive evaluations to date of sex-related differences in IIMs within a well-defined Italian cohort. The multicentre design facilitated the inclusion of a wide spectrum of clinical phenotypes, thereby enhancing the external validity and generalisability of our findings. Employing an integrative analytical approach that combined detailed clinical, serological, functional, and biomarker data, we achieved a robust characterisation of determinants influencing disease activity in different sexes. Our results uncover novel sex-related insights into disease activity as well as serological profiles. These findings underscore the critical importance of incorporating sex as a biological variable in both IIM research and clinical management. Furthermore, the utilisation of validated composite indices, such as MYOACT strengthened the reliability and clinical relevance of our conclusions.

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