Clinical Significance of Myositis-Specific and Myositis-Associated Antibody Profiles in Dermatomyositis

Abstract

Background: Myositis-specific autoantibodies (MSA) and myositis-associated autoantibodies (MAA) are clinically useful biomarkers that point to the diagnosis, clinical manifestations, and prognosis of dermatomyositis (DM). Materials and Methods: To estimate the prevalence of MSA as well as MAA and analyze possible clinical correlations of these autoantibodies in patients diagnosed with DM, we conducted a cross-sectional study of 30 patients who were diagnosed with DM. Results: MSA were positive in 19 patients (63%) in which Mi 2 was positive in 8 (27%) patients, and this was the most frequently found MSA. A total of 11 (36.7%) patients showed positive MAA. AntiPM/Scl 75 and anti-Ro 52 were positive in 5 (16.7%) patients each and these were the most commonly found MAA. Anti-La was absent in all our patients. There were 8 (27%) patients in whom both MSA and MAA were positive. Either MSA and/or MAA were positive in 22 (73%) patients. On a bivariate analysis, the patients who were positive for anti-PM/Scl 75 showed a significant difference in manifesting cutaneous ulcers (P value 0.023). It was also found that anti-SAE-positive patients showed a significant difference with malignancy (P value 0.014). Anti-Ro 52-positive patients were less likely to have symmetrical proximal muscle weakness (P value 0.006). Conclusions: All patients who were anti-MDA 5 positive had myositis and none of the anti-MDA 5-positive patients had rapidly progressive interstitial lung disease (RPILD). More than one MSA in the same patient was noted in three patients.

Keywords: Anti-Jo 1, anti-MDA 5, dermatomyositis, myositis-associated antibody, myositis-specific antibody

Introduction

Dermatomyositis (DM) is a progressive autoimmune condition characterized by inflammatory skin changes and muscle weakness. Incidence of DM is about 1 per 100,000 people per year with an estimated prevalence calculated to be about 20 cases per 100,000 people.^[1]

Myositis-specific autoantibodies (MSA) and myositis-associated autoantibodies (MAA) are biomarkers that prove useful in the diagnosis of polymyositis (PM) and dermatomyositis (DM).[2] Many of these are attributed to be associated with unique clinical subsets of DM which helps in predicting and monitoring some clinical manifestations as well as prognosis. MSA described so far are autoantibodies to aminoacyl transfer RNA synthetases (ARS) including histidyl (Jo 1), alanyl (PL 12), threonyl (PL 7), glycyl (EJ), isoleucyl (OJ), (Ha) asparaginyl (KS), tyrosyl and phenylalanyl (Zo), signal recognition

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particle (SRP), melanoma differentiation associated (MDA) 5/CADM 140, transcription intermediary factor 1 (TIF1), nuclear matrix protein (NXP) 2/MJ, Mi 2, 3-hydroxy-3-methylglutaryl-coA reductase (HMGCR) and small ubiquitin-like-modifier activating enzyme (SAE).^[3]

MAA include antibodies to SSA/Ro 52, PM/ Scl 75, PM/Scl 100, U1RNP, Ku and La.^[4]

The objective of our study was to estimate the prevalence of MSA as well as MAA and analyze possible clinical correlations of these autoantibodies in patients diagnosed with DM. The outcome variables included were specific clinical signs, inflammatory markers, interstitial lung disease, malignancy, compatible findings in skin biopsy, imaging studies and electromyogram.

Methods

All DM patients who attended dermatology and rheumatology departments of a

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quaternary care center in South India for five years (April 2016 to March 2021) were recruited for the cross-sectional study. We included 30 patients who were diagnosed with DM who met at least four of the criteria (including the mandatory criterion of typical skin rash of dermatomyositis, including heliotrope rash, Gottron sign and Gottron papules) defined by Bohan and Peter.^[5]

Institutional ethics committee approval was obtained for this study. Informed consent was obtained from the subjects included in the study.

Data regarding demographic features, clinical manifestations, laboratory and radiographic investigations as well as the presence of internal malignancies and interstitial lung disease in these patients diagnosed with dermatomyositis during this period was obtained.

The cutaneous clinical parameters that were included in the study were the presence of heliotrope rash, shawl sign, V sign, Gottron sign, Gottron papules, distorted cuticles with dilated capillary loops in proximal nail folds, mechanic's hands and cutaneous ulcers [Figures 1a-f].

The presence of symmetrical proximal muscle weakness was noted. Medical Research Council classification was used for limb muscle strength grading.^[6]

Lab investigations that were considered in the study were creatine kinase (CK), serum glutamic oxaloacetic

transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT), erythrocyte sedimentation rate (ESR), lactate dehydrogenase (LDH), antinuclear antibody (ANA) by immunofluorescence and anti-double stranded DNA (ant-ds DNA).

Skin biopsy reports of 16 patients who were willing for skin biopsy were noted. Magnetic resonance imaging (MRI) of 23 patients and positron emission tomography (PET) scan of 23 patients were considered in the study. Electromyogram (EMG) findings of 17 patients who were willing for this investigation were recorded.

Interstitial lung disease was diagnosed on the presence of both respiratory symptoms and radiological findings. Associated malignancies which were detected on PET scan and confirmed with histopathology were recorded.

The presence of following autoantibodies in the blood were detected using a commercially available kit (myositis profile-immunoblot, Aster Labs, India): anti-Jo 1, anti-PL 12, anti-PL 7, anti-EJ, anti-OJ, anti-SRP, anti-MDA 5, anti-TIF1, anti-NXP 2, anti-Mi 2, anti-SAE, anti-Ro 52, anti-PM/Scl 75, anti-PM/Scl 100, anti-U1RNP, anti-Ku and anti-La. The results were arbitrarily defined as negative (0/+++), weakly (+/+++), moderately (++/+++), or strongly (+++/+++) reactive. Weak, moderate or strong reactivity results were considered as positive. Continuous variables were presented as median and interquartile



Figure 1: (a) Heliotrope rash (periorbital erythema and edema) with ulceration. (b) Mechanic's hand (hyperpigmented scaly plaques on palms). (c) Gottron papules (erythematous papules on dorsa of hands). (d) Distorted cuticles. (e) V sign (erythema on V area of neck). (f) Shawl sign (erythema on upper back)

range (IQR). Categorical variables were reported as frequency and percentage and compared using the Fisher's exact test. We considered P value <0.05 as statistically significant and the data analysis was done using R version 4.1.1 with RStudio.

Results

The median age of disease onset was 47 (35-61) years with the median duration of 3 (1-8) months between the onset of symptoms and the time of presentation at our center. Nineteen (63.3%) of our patients were females.

Heliotrope rash was present in 26 (86.7%) patients and was the most common cutaneous clinical sign followed by shawl sign in 16 (53.3%), V sign, Gottron sign and Gottron papules in 15 (50%) each, mechanic's hands and distorted cuticles with dilated capillary loops in proximal finger nail folds in 9 (30%) each. Cutaneous ulcer was the least common clinical sign which was seen only in 2 (6.7%) patients.

Symmetrical proximal muscle weakness was detected in 23 (76.7%) patients. In 7 (23.3%) of those patients who had no clinical muscle weakness, only 2 (6.7%) could fit into the definition of amyopathic dermatomyositis (ADM) while the remaining 5 (16.7%) could be defined as clinically amyopathic dermatomyositis (CADM).

Interstitial lung disease was diagnosed in 8 (26.7%) and associated malignancy was detected in 4 (13.3%). Malignancies confirmed by histopathology were non-Hodgkin lymphoma, metastatic squamous cell carcinoma of cervical lymph node with occult primary, adenocarcinoma lung and breast carcinoma respectively in these four patients.

ESR was raised in 18 (60%), ANA (immunofluorescence) was positive in 15 (50%) and anti-ds DNA was positive in 3 (10%).

Following were the elevated muscle enzyme parameters: CK in 16 (53.3%), LDH in 28 (93.3%), SGOT in 22 (73.3%), and SGPT in 21 (70%).

Skin biopsy findings were compatible with dermatomyositis in 12 (75%) out of 16 in whom it was performed. MRI scan was performed in 23 patients, out of which 16 (69.6%) showed myositis. PET scan was done in 23 patients and 12 (52.2%) revealed myositis. In 17 patients who underwent EMG, 12 (70.6%) showed findings compatible with myositis.

Demographic, clinical, laboratory and radiological findings are detailed in Table 1.

MSA were positive in 19 patients (63%) in which Mi 2 was positive in 8 (27%) patients, and this was the most frequently found MSA. A total of 11 (36.7%) patients showed positive MAA. AntiPM/Scl 75 and anti-Ro 52 were positive in 5 patients (16.7%) each and these were the most commonly found MAA. Anti-La was absent in all our patients. There were 8 (27%) patients in whom both MSA

and MAA were positive. Either MSA and/or MAA were positive in 22 (73%) patients. Profile of MSA and MAA are shown in Table 2.

On a bivariate analysis, patients who were positive for anti-PM/Scl 75 showed a significant difference in manifesting cutaneous ulcers (*P* value 0.023). It was also found that anti-SAE positive patients showed a significant difference with malignancy (*P* value 0.014). It was also showed that those patients who had ant-Ro 52 were less likely to have symmetrical proximal muscle weakness (*P* value 0.006, prevalence odd's ratio 0.15 [0.048–0.473]). All patients who were anti-MDA 5 positive had myositis and none of the anti-MDA 5-positive patients had rapidly progressive interstitial lung disease (RPILD). There was only one patient who showed TIF1 γ/α positivity, and this patient had malignancy. There was no significant difference between the other MSA or MAA and any clinical profiles.

Discussion

There are certain specific autoantibodies that are biomarkers associated with certain diseases or clinical features which can serve as diagnostic as well as prognostic pointers in systemic autoimmune rheumatic diseases (SARD). These autoantibodies could be detected much before the onset of signs and symptoms and thus add to the predictive value.^[7]

MSA are almost exclusively found in PM/DM among SARD though some autoantibodies such as ARS are also seen in idiopathic interstitial lung diseases (ILD).^[8] MAA are vague in definition than MSA as autoantibodies are found in PM/DM but not specific for this diagnosis and may be found in other SARD.^[8]

The prevalence of MSA in our study was 63% (19/30), while that in a study by Cruellas *et al.*^[4] from Brazil was only 31.6% (40/127).^[4] In the same study, prevalence of MAA 38.6% (49/127) was almost similar to that in our study of 36.7% (11/30).

Inflammatory myopathy forms a spectrum of diseases that varies from involvement of muscle without skin diseases, muscle as well as skin involvement, skin involvement with minimal muscle disease (CADM) or no muscle disease (ADM). ARS is detected in both PM as well as DM and at times in ADM. Our study showed 23% (7/30) prevalence of ARS. Anti-MDA5 is mostly seen in CADM/ ADM and predominant muscle disease is hardly common in this group.^[8] But, in our study, all four patients who was MDA 5 positive had myositis. Although all MSA are specific for PM or DM, presence of more than one MSA in the same patient is uncommon for unknown reasons.^[9] But, in our study, three patients had more than one MSA positivity out of which two patients showed concomitant positivity of Mi 2 and MDA 5.

Anti-Jo1 antibodies are associated with a particular clinical subset manifesting myositis, ILD, arthritis, mechanic's hands, and Raynaud's phenomenon, which has been

Table 1: Demographic, clinical, laboratory and radiologica Characteristics	Number of patients n (%) ($n=30$)	
Age in years at the time of presentation, median (IQR)	47	(35-61)
Male	11	(36.7%)
Female	19	(63.3%)
Duration in months between the onset of symptoms and the time of presentation, median (IQR)	3	(1-8)
Symmetrical proximal muscle weakness	23	(76.7%)
Heliotrope rash	26	(86.7%)
Shawl sign	16	(53.3%)
V-Sign	15	(50.0%)
Gottron sign	15	(50.0%)
Gottron papules	15	(50.0%)
Mechanic's hands	9	(30.0%)
Distorted cuticles with dilated capillary loops on the proximal nail folds	9	(30.0%)
Cutaneous ulcer	2	(6.7%)
Interstitial lung disease	8	(26.7%)
Associated malignancy	4	(13.3%)
CK elevated	16	(53.3%)
LDH elevated	28	(93.3%)
SGOT elevated	22	(73.3%)
SGPT elevated	21	(70.0%)
ESR elevated	18	(60.0%)
Skin biopsy compatible with DM	*12	(75.0%)
MRI Scan compatible with DM	**16	(69.6%)
PET Scan compatible with DM	***12	(52.2%)
EMG compatible with DM	****12	(70.6%)
ANA	15	(50.0%)
Anti-ds DNA	3	(10.0%)

*n=16, **n=23, ****n=23, ****n=17. IQR - interquartile range, CK - creatine kinase, LDH - lactate dehydrogenase, SGOT - serum glutamic oxaloacetic transaminase, SGPT - serum glutamic pyruvic transaminase, ESR - erythrocyte sedimentation rate, DM - dermatomyositis, MRI - magnetic resonance imaging, PET - positron emission tomography, EMG - electromyogram, ANA - antinuclear antibody , and ant-ds DNA - anti-double stranded DNA

designated as anti-synthetase syndrome.^[10] In our study, out of the four patients who had anti-Jo 1, all four had myositis, and two of them had both ILD and mechanic's hands; thus, all satisfied the criteria for anti-synthetase syndrome. Anti-Jo1, usually found in 15-25% of PM/DM patients, are by far the most common among anti-ARS antibodies while all other ARS are usually found only in 0.5-6% of patients.^[10] When it comes to the prevalence of anti-Jo 1 in patients with DM, it varies from 4.5 -13%.[8] This was agreeable in our study as well wherein the prevalence was 13% (4/30). Though patients with any anti-ARS show features of anti-synthetase syndrome, many studies point that those with autoantibodies to non-Jo1-ARS are associated with earlier and more severe ILD as well as poor prognosis than those with anti-Jo 1 patients. Also, non-Jo1-ARS patients are more prone to develop ILD without typical myositis.^[8] But this pattern was not followed in this study. Literature reveals that anti-SRP is specifically seen in PM and present with severe myopathy resistant to treatment, which is histologically characterized by necrotizing myopathy.^[11] In our study, we had only two patients with DM who were anti-SRP positive, and both had severe myopathy.

Reported prevalence of anti-Mi 2 varies widely from 2% to 60% in different studies and is found to be different even in the same country.^[8] In the subset of MSA, anti-Mi 2 was the most common with a prevalence of 27% (8/30) in our study. Studies point that anti-Mi 2 is associated with classical clinical features of DM including Gottron papules, shawl sign, heliotrope rash and V-sign and a risk to develop ILD and cancer is uncommon.^[12] The same was seen in our study as well. These patients also respond well to steroids and have a fairly good prognosis.

Studies on anti-MDA 5/CADM 140 in DM have shown a prevalence that ranges from 3% to 58% which increased even up to 100% when patients with only CADM were considered.^[8] The prevalence of anti-MDA 5 positive patients was 13% (4/30) in our study. But only one patient out of seven patients with CADM/ ADM had positive anti-MDA 5. Many reports describe that anti-MDA 5 antibodies are specific for DM and a majority of patients have CADM and high prevalence of rapidly progressive ILD (RPILD) leading to poor prognosis.^[13-15] However, in our study, only two out of four patients who were anti-MDA 5 positive manifested

Table 2: Profile of MSA and MAA		
Autoantibodies	Number of positive patients n (%) (n=30)	
Anti-PM/Scl 100	2 (6.7)	
Anti-PM/Scl 75	5 (16.7)	
Anti-Ro 52	5 (16.7)	
Anti-U1RNP	1 (3.3)	
Anti-Ku	1 (3.3)	
Anti-La	0 (0.0)	
MAA	11 (36.7)	
Anti-Jo 1	4 (13)	
Anti-PL 12	3 (10)	
Anti-PL 7	2 (7)	
Anti-EJ	1 (3)	
Anti-OJ	1 (3)	
SRP	2 (7)	
Anti MDA 5	4 (13)	
Anti-TIF1-γ	1 (3)	
Anti-NXP-2	2 (7)	
Mi -2	8 (27)	
SAE	2 (7)	
MSA	19 (63)	
MAA and MSA	8 (27)	
MAA and/or MSA	22 (73)	

MSA - myositis-specific autoantibodies,

MAA - myositis-associated autoantibodies

with ILD and none had RPILD. Anti-MDA 5 was also associated with ILD in juvenile DM (JDM).^[16] Sato *et al.*^[17] reported that anti-MDA 5 antibody levels in those who showed good response to therapy and survived were significantly lower than those who did not respond and expired.

The association of anti-TIF1 γ/α positive DM particularly with cancer has been confirmed in many reports.^[18] Most of anti-TIF1 γ/α positive patients have DM with classical skin rashes but the prevalence of ILD was low. Prevalence of malignancy in anti-TIF1 γ/α positive patients is 42–100%.^[8] Our study was consistent with this as it showed a 100% prevalence of malignancy in anti-TIF1 γ/α positive DM.

The prevalence of anti-NXP 2 in our study was 7% (2/30). A higher prevalence of calcinosis was noted in a few studies and some studies showed an association of malignancy with this antibody. Out of the two patients with anti-NXP 2 positivity in our study, one had associated malignancy.

Literature reports a prevalence of anti-SAE in 8-10% of patients with DM and majority present with skin disease prior to the onset of myositis.^[8] The prevalence of anti-SAE in our study was 7% (2/30).

The association of MAA anti-Ro 52 with anti-Jo 1 has been described in 10% of the cases of idiopathic inflammatory myositis and this co-existence causes an increased risk of malignancy and more severe myositis in anti-synthetase syndromes. But anti-Ro-52 was

associated with pulmonary disorders in DM, independent of anti-Jo-1 reactivity.^[4] But none of the these reflected in our study.

Clinical implication of this study is that by deriving the prevalence of various MSA and MAA in DM and further understanding the correlation of multiple variables, the presence of these antibodies may be considered in the diagnostic criteria of DM.

Longitudinal studies on the profile of MSA and MAA are recommended which can be helpful in confirming significant associations and thereby conceiving various subsets of DM.

Strengths and limitations

In this cross-sectional study, the prevalence of various MSA and MAA could be determined. Also, the correlations of multiple variables could be studied. These can be considered as the strengths of this study.

Autoantibody positivity may precede clinical disease by many years. The median duration between the onset of symptoms in our patients and their time of presentation at our center was 3 months. Taking this into account, we assume that our patients might have presented much early in disease timeline and hence did not manifest certain associated clinical features. This was a limitation in our study. Moreover, the small sample size was a limitation which made it difficult to arrive at significant differences.

Conclusion

Anti-MDA 5 is predominantly seen in CADM/ADM as per most of the previous studies. In contrast, this study revealed that all patients who were MDA 5 positive had myositis. In general, more than one MSA in the same patient is uncommon. However, in our study this pattern was not seen to be followed. More than one MSA in the same patient was noted in three patients. Anti-MDA 5 antibodies are known to be specific for DM and a majority of patients have CADM and high prevalence of RPILD leading to poor prognosis. But in this study, anti-MDA 5 positive patients who had CADM was minimal and none of the anti-MDA 5 positive patients had RPILD.

Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form the patient(s) has/have given his/her/their consent for his/her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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Conflicts of interest

There are no conflicts of interest.

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