Use of steroids in the management of low-risk myelodysplastic syndromes with autoimmune features

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Background - The boundaries between myelodysplastic syndromes (MDS) and immune-mediated cytopenias are often difficult to establish and both conditions may benefit from immunosuppressive therapy. The optimal timing and doses of immunosuppressants are largely unknown.

Materials and methods - We systematically evaluated a retrospective cohort of 79 patients with low-risk MDS tested for anti-erythrocyte or anti-platelet autoantibodies to assess their frequency and the efficacy of immunosuppression, particularly with steroids.

Results - We found autoantibody positivity in 43% of cases and overt autoimmune diseases in 18%, including autoimmune hemolytic anemia, immune thromboctyopenia, and Evans syndrome. Steroid treatment improved cytopenia in about half of patients, with 26% achieving a complete recovery lasting for a median of 12 months. Better responses were observed in anemic patients with anti-erythrocyte autoantibodies than in those with anti-platelet autoantibodies, and the combination with recombinant erythropoietin (7/10) had a possible synergistic effect. Steroid doses were heterogeneous depending on the clinical intent (i.e., anti-inflammatory, immunosuppressive, anabolizing). Patients treated with a dose of 1 mg/kg day of prednisone for overt autoimmune cytopenia showed high rates of complete responses (60%).

Discussion - This observation suggests a trial with a short course (2-3 weeks) of standard steroid doses to ascertain efficacy and properly silence the autoimmune pathogenic mechanism. Steroid-related adverse events (16% of cases) should be monitored carefully in this elderly, frail population.

In conclusion, features of autoimmunity are present in more than two-thirds of low-risk MDS patients and a trial with prednisone 0.5-1 mg/kg day for 2-3 weeks, with proper monitoring of adverse events, may be useful to improve cytopenias in selected cases.

Keywords: *low-risk myelodysplastic syndromes, direct antiglobulin test, autoimmune hemolytic anemia, steroids, immune thrombocytopenia.*

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INTRODUCTION

Corticosteroids have several immunomodulating/immunosuppressive activities and are widely used in cytopenic patients, including those with myelodysplastic syndromes (MDS)[1]. In fact, the boundaries between MDS, especially low-risk ones, and

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immune-mediated cytopenias represent a largely uncharted territory with overlapping clinical features and pathogenic mechanisms[2,3]. Both the incidence of overt autoimmune disorders and the detection of autoantibodies without clinically manifest disease have been reported in MDS[4-7]. Their contribution to the cytopenic phenotype is further suggested by the efficacy of steroids and other immunosuppressive treatments in a fraction of MDS patients[8-10]. However, no clear predictors of response have been identified except for a better efficacy of immunosuppression in hypocellular MDS[11].

Here we evaluated a cohort of low-risk MDS patients tested for the presence of autoantibodies to red blood cells and platelets to assess their clinical significance, focusing on the use and efficacy of steroid therapy. Furthermore, we provide a description of peculiar clinical cases, paradigmatic of the impact of autoimmune cytopenias in patients with low-risk MDS.

MATERIALS AND METHODS

Patients and hematological evaluation

This is an observational retrospective study including 79 consecutive patients with low-risk MDS diagnosed from 2001 until the time of writing who underwent an immuno-hematological evaluation during their follow-up at our tertiary hematology center in Milan, Italy. The study was approved by the local Ethical Committee and conducted according to the Helsinki Declaration. Patients gave informed consent to their participation in the study.

Diagnoses were based on the revised 4th edition (2016 update) of the World Health Organization (WHO) Classification of Tumors of Hematopoietic and Lymphoid Tissues[12].

Inclusion criteria encompassed both the presence of a low or intermediate-1 International Prognostic Scoring System (IPSS)[13] score calculated as per published guidelines and the evaluation of at least one between the direct anti-globulin test (DAT) and anti-platelet autoantibodies during the course of the disease.

Clinical and laboratory data at diagnosis including blood cell counts, markers of hemolysis (lactate dehydrogenase and reticulocyte count), DAT result and presence or absence of anti-platelet autoantibodies, were retrospectively collected. The DAT was performed by the tube technique with monospecific antibodies anti-IgG and anti-complement; in selected cases more sensitive tests included microcolumn Biorad (Hercules, CA, USA) with anti-IgG, anti-C, anti-IgA, and anti-IgM, and solid phase (Capture R Select, Immucor [Norcross, GA, USA]). Anti-platelet autoantibodies were looked for using a solid phase technique (Capture P, Immucor) for the direct test; the indirect test was performed by enzyme-linked immunosorbent assay (Pak Plus, Immucor). This test allows differentiation between autoantibodies against platelet glycoproteins and anti-HLA class I alloantibodies. Patients transfused with platelets were tested for anti-platelets with the direct test at least 7 days after the last transfusion to avoid contamination.

Bone marrow evaluation

For all but six patients bone marrow biopsies from diagnosis were available for evaluation. Histology features were categorized with regard to cellularity, dysplastic changes (either morphological or numerical, especially in the megakaryocytic lineage), reticulin fibrosis, and lymphoid infiltrate.

Moreover, bone marrow multiparameter flow cytometry evaluation was available for 60 patients. The proportions of the various lymphocyte subpopulations (T, B, and NK cells), monocytes, and mastocytes were evaluated using the following monoclonal antibodies directed against CD3, CD10, CD14, CD16, CD19, CD34, CD45, CD56, CD117, and HLA-DR.

Finally, classical chromosome banding for cytogenetic analysis was performed in all patients.

Molecular analysis

In a subgroup of patients (n=41), next-generation sequencing was performed using an Ion Torrents5, with Ion Reporters software 5.2 (Thermo Fisher Scientific, Waltham, MA, USA) which evaluates the mutational status of 69 potentially oncogenic genes present in the Oncomine Myeloid Research Assay diagnostic panel, specifically hotspot genes (*ABL1, BRAF, CBL, CSFR3, DNMT3A, FLT3, GATA2, HRAS, IDH1, IDH2, JAK2, KIT, KRAS, MOL, MYD88, NPM1, NRAS, PTPN11, SETPB1, SF3B1, SRSF2, U2AF1, WT1*), full genes (*ASXL1, BCOR, CALR, CEBPA, ETV6, EZH2, IKZF1, NF1, PHF6, PRPF8, RB1, RUNX1, SH2B3, STAG2, TET2, TP53, ZRSR2*), and fusion transcripts (*ABL1, ALK, BCL2, BRAF, CCDN1, CREBBP,*

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EGFR, ETV6, FGFR1, FGFR2, FUS, HMGA2, JAK2, KMT2A, MECOM, MET, MLLT10, MLLT3, MYBL1, MYH11, NTRK3, NUP214, PDGFRA, PDGFRB, RARA, RBM15, RUNX1, TCF3, TFE3). Only variants with an allelic frequency >5% were reported[14].

Therapy evaluation and outcome measures

Therapy lines and their efficacy were recorded. Hematologic improvement was evaluated according to the International Working Group 2018 criteria for MDS[15]. Regarding steroids, the reason for treatment, the doses administered, the treatment duration, the response rate and its duration, were registered. Adverse events were recorded and graded according to Common Terminology Criteria for Adverse events version 5.0 (CTCAE v5.0). Evolution to acute myeloid leukemia and death were also collected for each patient.

Statistical analysis

For statistical analysis, the Student's *t*-test was used for continuous variables and χ^2/Fisher's exact test for categorical ones. Analysis of variance was performed using mean, median, ranges and standard errors.

RESULTS

Clinical and hematological parameters at diagnosis

Table I shows the clinical and hematological parameters at baseline in all patients and according to positivity of autoantibodies. The median follow-up was 54.5 months (range, 3-244). The great majority of the patients were elderly with a slight male predominance. According to the WHO classification, most patients had MDS with multi-lineage dysplasia MDS (36.7%) or MDS with ring sideroblasts (31.6%), and all had low (62%) or intermediate-1 (38%) risk by the IPSS, as per inclusion criteria[8]. According to the Revised IPSS[16] most patients were low risk (53.7%), with the remaining equally divided in the very low and intermediate risk groups.

Bone marrow features, cytogenetics and molecular findings

Most patients displayed a normo- or hypercellular marrow (81%), while 12 patients had a hypocellular trephine (*Online Supplementary* **Table SI**). Megakaryocyte counts were mainly normal or increased, and dysmegkaryopoiesis, dyserythropoiesis, and dysgranulopoiesis were present in 53%, 66%, and 33% of patients, respectively. Reticulin fibrosis was found in 32% of samples (grade 2 in one patient) and a lymphoid infiltrate in 58%, mainly with T-cell phenotype. This was also confirmed by multiparameter flow cytometry showing a predominant T-cell infiltrate (69.5% of total lymphocytes), followed by NK cells (16%), and B-lymphocytes (10%). Cytogenetics was normal in 81% of patients, and chromosomal aberrations included deletions of chromosome Y, 20 and 5, and trisomy of chromosome 8 in two patients. Finally, 41 subjects (52%) were tested for somatic mutations in myeloid genes by next-generation sequencing and 60% had at least one mutation, the most common involving *SF3B1* (n=10), *SRSF2* (n=7), *TET2* (n=7), *ASXL1* (n=7), *IDH1/2* (n=5), *DNMT3A* (n=4), *RUNX1* (n=3) and *ZRSR2* (n=3).

Autoimmune features

As depicted in **Figure 1**, positivity for at least one marker of autoimmunity (DAT and/or anti-platelet autoantibodies) was found in 34 patients (43%). Anti-platelet autoantibodies were significantly more frequent than DAT positivity (22/40 [55%] *vs* 13/50 [26%] tested cases, p=0.004). One patient turned positive to both tests and was later diagnosed with Evans syndrome. Overall, 14 (18%) received a diagnosis of overt autoimmune disease, ten a diagnosis of a hematological disorder (6 autoimmune hemolytic anemia [AIHA], 3 immune thrombocytopenia and 1 Evans syndrome), and four had extra-hematological conditions (rheumatoid arthritis, seronegative arthritis, mixed connective tissue disease and rheumatic polymyalgia). No other extra-haematological autoimmune manifestations were present before the diagnosis of MDS. Additionally, VEXAS syndrome (vacuoles, E1 enzyme, X-linked, autoinflammatory, somatic) was excluded by re-evaluating the history and bone marrow samples of the four subjects with extra-hematological autoimmune diseases.

Finally, hypoplastic MDS subjects more frequently showed a marker of positivity compared to normo/hypercellular ones (67 *vs* 39%, p=ns), and two patients were diagnosed with AIHA and Evans syndrome, respectively.

Comparison between patients with and without autoantibodies

Autoantibody-positive patients more frequently had a diagnosis of multi-lineage MDS (p=0.008), and had a higher rate of thrombocytopenia (50 *vs* 28.9%, p=0.04). Consistently, the median platelet count at presentation was significantly lower (90 *vs* 124×10^9/L, p=0.03), and

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Table I - *Clinical and hematological parameters at baseline in all patients and according to positivity for autoantibodies*

	All patients (n=79)	Antibody positive (n=34)	Antibody negative (n=45)	p value
Median FU, months (range)	54.5 (3-244)	52.1 (3-243)	55.6 (9-244)	0.92
Median age, years (range)	75 (36-92)	72.5 (42-92)	76 (36-88)	0.43
Sex, N(%)	n=79	n=34	n=45	
Male	47 (59.5)	17 (50)	30 (66.7)	0.1
Female	32 (40.5)	17 (50)	15 (33.3)	0.1
WHO type, N(%)	n=79	n=34	n=45	
MDS SLD	13 (16.5)	4 (11.8)	9 (20)	0.25
MDS MLD	29 (36.7)	18 (52.9)	11 (24.4)	0.01
MDS RS SLD	12 (15.2)	4 (11.8)	8 (17.8)	0.34
MDS RS MLD	9 (11.4)	1 (2.9)	8 (17.8)	0.03
MDS del 5q	4 (5.1)	2 (5.8)	2 (4.4)	0.58
MDS EB1	3 (3.8)	2 (5.8)	1 (2.2)	0.39
MDS/MPN	1 (1.3)	0	1 (2.2)	0.57
CMML-1	1 (1.3)	0	1 (2.2)	0.57
MDS Unclassified	7 (8.9)	3 (8.8)	4 (8.9)	0.65
IPSS, N(%)	n=79	n=34	n=45	
low	49 (62)	20 (58.9)	29 (64.4)	0.39
intermediate-1	30 (38)	14 (41.1)	16 (35.6)	0.39
IPSS-R, N(%)	n=67	n=29	n=38	
very low	16 (23.9)	5 (17.2)	11 (28.9)	0.21
low	36 (53.7)	18 (62.1)	18 (47.4)	0.17
Intermediate	14 (20.9)	5 (17.2)	9 (23.7)	0.37
high	1 (1.5)	1 (3.4)	0	0.43
Laboratory values, median (range)				
Hemoglobin (g/dL)	n=66 10.20 (6.4-15.1)	n=29 10.20 (6.4-14.6)	n=37 10.20 (6.7-15.1)	0.42
ANC (/mm^3)	n=58 2,025 (150-6,660)	n=23 1,580 (370-5,810)	n=35 2,470 (150-6,660)	0.95
Platelet count (x10^9/L)	n=68 114,500 (11- 420)	n=30 90 (20-370)	n=38 124,000 (11-420)	0.03
LDH (mU/mL)	n=56 204.5 (139-393)	n=25 208 (141-393)	n=31 201 (139-383)	0.36
Reticulocytes (x10^9/L)	n=22 44 (6 -181)	n=10 43 (6-121)	n=12 44 (11-181)	0.93
Cytogenetics, N(%)	n=78	n=33	n=45	
Normal	63 (80.8)	25 (75.8)	38 (84.4)	0.25
Abnormal	15 (19.2)	8 (24.2)	7 (15.6)	0.25
NGS, N(%)	n=41	n=16	n=25	
Normal	16 (39)	6 (37.5)	10 (40)	0.57
1 mutation	12 (29.3)	4 (25)	8 (32)	0.45
>1 mutations	13 (31.7)	6 (37.5)	7 (28)	0.38

FU: follow-up; WHO: World Health Organization; MDS: myelodysplastic syndrome; SLD: single lineage dysplasia; MLD: multilineage dysplasia; RS: ringed sideroblasts; EB1: excess blasts-1; MPN: myeloproliferative neoplasm; CMML: chronic myelomonocytic leukemia; IPSS: International Prognostic Scoring System; IPSS-R: Revised International Prognostic Scoring System; ANC: absolute neutrophil count; LDH: lactate dehydrogenase; NGS: next-generation sequencing.

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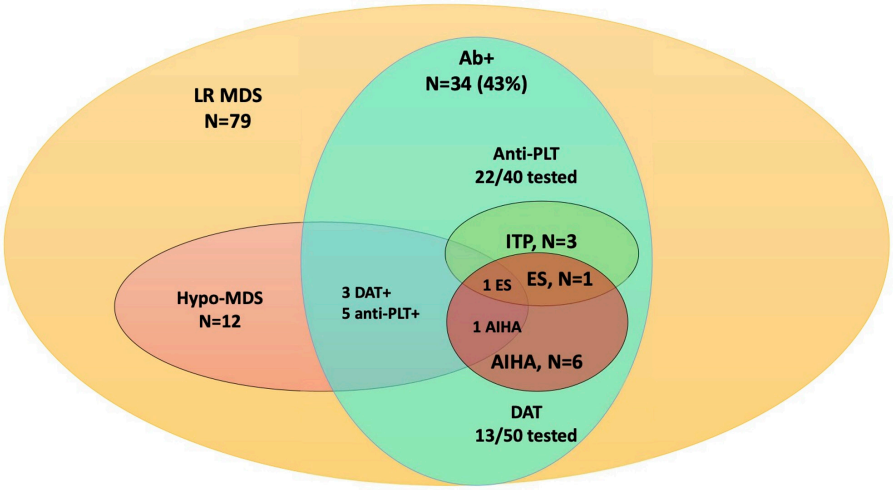


Figure 1 - Disposition of autoimmune markers and diseases among patients with low-risk myelodysplastic syndromes
LR MDS: low-risk myelodysplastic syndrome; Ab: autoantibodies; anti-PLT: anti-platelet autoantibodies; ITP: immune thrombocytopenia; DAT direct antiglobulin test; AIHA: autoimmune hemolytic anemia; ES: Evans syndrome; hypo-MDS hypocellular myelodysplastic syndrome.

a trend for reduced neutrophils was observed, while values of hemoglobin and markers of hemolysis were comparable (**Table I**).

Patients who were positive for autoantibodies also more frequently had a hypocellular bone marrow than did negative patients (28.6 *vs* 11.1%, p=ns) and showed a higher proportion of mastocytes by multiparameter flow cytometry (1.9 *vs* 1.5%, p=0.04); cytogenetics and next-generation sequencing findings did not differ between autoantibody-positive and -negative patients (*Online Supplementary* **Table SI**).

Treatment and outcome

Therapies administered during the follow-up are summarized in **Table II**. Sixty-six percent received at least one treatment, particularly, recombinant erythropoietin (rEPO, 23/41 responded), followed by luspatercept (2/6 responded), lenalidomide (1/2 responded), and experimental daratumumab in two cases (no response).

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Table II - *Therapy lines administered to all patients and according to positivity for autoantibodies*

Treatment	All patients (n=79)	Antibody positive (n=34)	Antibody negative (n=45)	p value
Treated, N(%)	52 (65.8)	23 (67.6)	29 (42.4)	0.48
Not treated, N(%)	27 (34.2)	11 (32.4)	16 (35.6)	0.48
Type of treatment				
Erythropoietin	41 (51.9)	15 (44.1)	26 (57.8)	0.16
Luspatercept	6 (75.9)	2 (5.9)	4 (8.9)	0.48
Lenalidomide	2 (2.6)	1 (2.9)	1 (2.2)	0.68
Daratumumab	2 (2.6)	1 (2.9)	1 (2.2)	0.68
Eltrombopag	3 (37.9)	2 (5.9)	1 (2.2)	0.39
Immunosoppressive treatment				
Steroids	31 (39.2)	19 (55.9)	12 (26.7)	0.01
Danazol	2 (2.6)	2 (5.9)	0	0.18
Cyclosporine	1 (1.3)	1 (2.9)	0	0.43
Rituximab	1 (1.3)	1 (2.9)	0	0.43

Overall, 31 patients received steroids (**Table III**), for different reasons: constitutional symptoms (n=3), short-term response to red blood cell transfusions (n=7), altered hemolytic markers (n=8), and thrombocytopenia (n=8). Steroids were mainly administered to autoantibody-positive patients rather than to negative patients (55.9 *vs* 26.7%, p=0.008). Overall the median prednisone dose was 25 mg/day (range, 5-65 mg) administered for a median of 12 months (range, 4-60). Autoantibody-positive patients more frequently received higher steroid doses (i.e., prednisone >10 mg/day), whereas those treated for constitutional symptoms and scarce response to transfusions mainly received low doses (≤10 mg/day, p=0.01). Response to steroids was observed in 54% of patients (n=17) with about 50% showing complete cytopenia recovery. The eight patients who received prednisone 1 mg/kg day for overt autoimmune cytopenia, showed better responses (62.5%, 3 complete responses). Additionally, the rate of response was higher in those with a positive DAT compared to those who were positive for anti-platelet autoantibodies (80 *vs* 40%, p=0.01). Erythroid improvement (10/20, 50%) was significantly associated with DAT positivity (70 *vs* 25% in DAT-negative cases, p=0.01), while platelet response (7/15, 47%) was equally distributed among patients with negative and positive anti-platelet autoantibodies (40 *vs* 50%). In fact, 15/20 anemic patients (75%) were already on rEPO treatment at the time of starting steroid therapy, and seven of them responded (47%). The median duration of response was 12 months (range, 2-60). Six patients (5 with positive autoantibodies) received repeated steroid courses during the follow-up, and 15 subjects were still receiving steroids at data cut-off.

Five patients (16%) experienced a steroid-related adverse event, mainly grade ≤2, including insomnia (n=3), diabetes and osteoporosis (n=2). One patient experienced a fatal endocarditis. Forty-two percent of patients required a MDS specific therapy after steroids failure, more often in the group without autoantibodies (50 *vs* 37%).

Other immunomodulating therapies included danazol in three patients (no response), cyclosporine in two thrombocytopenic patients (one response), and rituximab in the patient with Evans syndrome with complete recovery. Finally, three patients received the thrombopoietin receptor analogue eltrombopag and one responded.

The MDS evolved to acute leukemia in only two patients, one with anti-platelet autoantibodies and one with a positive DAT. Eleven patients died, equally distributed between subjects with and without autoantibodies.

Paradigmatic cases

Figure 2 illustrates the treatments and responses in seven patients who were initially diagnosed with low-risk MDS and were later found to have an immune-mediated cytopenia (4 with thrombocytopenia of whom 1 negative and 3 positive for anti-platelet

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Table III - *Steroid treatment in all patients and according to autoantibody positivity*

Steroid treatment (N, %)	All patients	Ab+	Ab−	DAT+	Anti-PLT +
Number treated	31 (39.2)	19 (55.9)*	12 (26.7)	10 (76.9)	10 (45.5)
Low dose	10 (32.3)	5 (26.3)	5 (41.7)	3 (30)	3 (30)
High dose	21 (67.7)	14 (73.7)*	7 (58.3)	7 (70)	7 (70)
Response	17 (54.8)	11 (57.9)	6 (50)	8 (80)	4 (40)
No response	14 (45.2)	8 (42.1)	6 (50)	2 (20)	6 (60)
Complete response	8 (25.8)	4 (21.1)	4 (33.3)	3 (30)	2 (20)
Partial response	9 (29)	7 (36.8)	2 (16.7)	5 (50)	2 (20)
Hb reponse (N=20)	10/20 (50)	7/8 (87.5)*	3/12 (25)	7/10 (70)	0/10
Platelet response (N=15)	7/15 (46.6)	4/9 (44.4)	3/6 (50)	1/10 (10)	4/10 (40)
Median duration of response (months)	12 (2-60)	12 (4-60)	12 (2-60)	12 (4-60)	12 (2-50)
Repeated steroid course	6 (19.4)	5 (26.3)	1 (8.3)	4 (40)	1 (10)
Ongoing steroid therapy	15 (48.4)	9 (47.4)	6 (50)	4 (40)	5 (50)
Therapy after steroids	13 (41.9)	7 (36.8)	6 (50)	2 (20)	5 (50)

Ab: autoantibody; DAT+: positive direct antiglobulin test; Anti-PLT+: positive for anti-platelet autoantibodies; Hb: hemoglobin. *p<0.05.

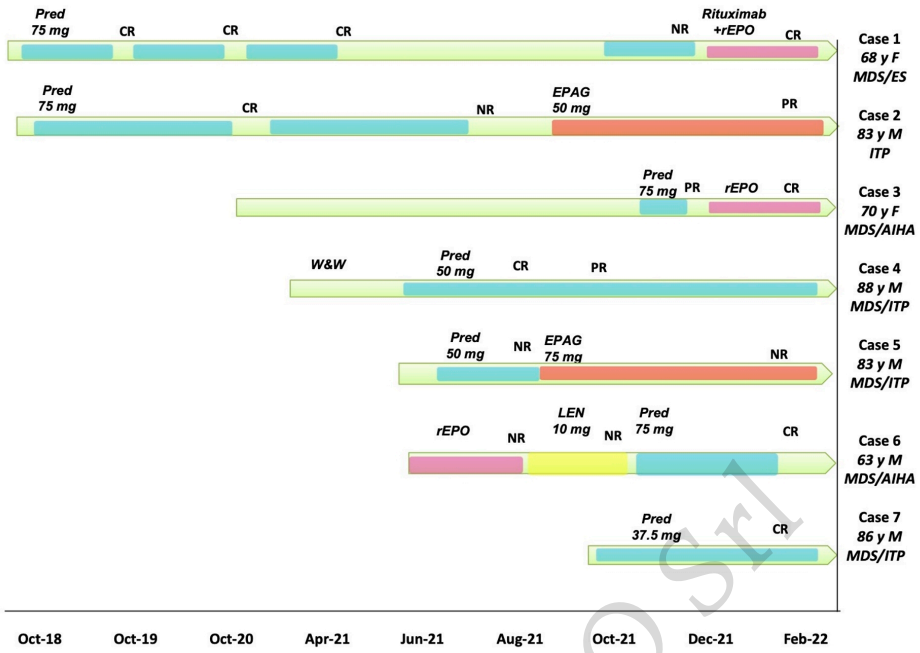


Figure 2 - Paradigmatic cases of low-risk myelodysplastic syndrome receiving therapy for immune mediated cytopenia
AIHA: autoimmune hemolytic anemia; ITP: immune thrombocytopenia; ES: Evans syndrome; MDS: myelodysplastic syndrome; Pred: prednisone; LEN: lenalidomide; rEPO: recombinant erythropoietin; EPAG: eltrombopag; PR: partial remission; CR: complete remission.

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autoantibodies, 2 with DAT-positive hemolytic anemia, and 1 with DAT-positive Evans syndrome) requiring treatment with steroids, eltrombopag, and rituximab. Cases 4 and 7 were two elderly patients, one positive and one negative for anti-platelet autoantibodies, referred to our center for severe thrombocytopenia and low-risk MDS. Both were treated with 1 mg/kg steroid therapy obtaining a complete response in 1 month. Unfortunately, one of the two died from severe recurrent endocarditis. Two further patients positive for anti-platelet autoantibodies (cases 2 and 5) were refractory to steroids and received eltrombopag with only one responding but experiencing thrombophlebitis.

Concerning AIHA, a 70-year-old woman (case 3) with MDS and IgG DAT positivity (detected 2 years after MDS diagnosis) received steroids 1 mg/kg obtaining a partial response that ameliorated with the addition of rEPO. A second patient with 5q- MDS (case 6) was treated with lenalidomide, with no response and worsening transfusion need. He was later found to be DAT-positive for IgG and complement and was treated with steroids 1 mg/kg achieving complete response. Steroids were thereafter tapered, and the patient continued only with the lenalidomide treatment.

Finally, an elderly thrombocytopenic patient (case 1) with positive anti-platelet autoantibodies responded to repeated steroid cycles and was later diagnosed with low-risk MDS (*DNMT3*-mutated at next-generation sequencing evaluation). Subsequently, IgG and complement DAT-positive AIHA (namely Evans syndrome) was diagnosed and rituximab was administered which produced a complete response.

DISCUSSION

Here we confirm that patients with low-risk MDS frequently show features of autoimmunity, namely autoantibody positivity in 43% of cases and overt autoimmune disease in 18%, particularly cytopenias. Among cases of autoantibody positivity, anti-platelet autoantibodies were more frequent than DAT, notwithstanding the low sensitivity and specificity of tests for the former and the higher prevalence of anemia in this population of patients. Moreover, autoantibody-positive patients more frequently had hypocellular bone marrow,

consistently with the well-known overlapping features of hypoplastic MDS and classic immune-mediated bone marrow failure[17-20]. Notably, positivity for autoantibodies was not associated with typical clinical or laboratory features of autoimmune cytopenias (i.e., alterations of hemolytic markers for anemic patients or presence of hyperplastic megakaryocytes for thrombocytopenic ones), thus supporting the systematic evaluation of such inexpensive and widely available tests. This is particularly true for DAT which may also be useful during the follow-up as some of our patients (i.e., case 3) developed positivity years after MDS diagnosis and benefited from steroids and rituximab. Finally, "reactive" dysplastic features have been reported even in the bone marrow of patients with autoimmune cytopenias[2] so that strict adherence to WHO criteria for MDS diagnosis, including morphology, cytogenetics, and, more recently, molecular analysis, should be pursued even in the case of antibody positivity[12,21]. In our series, all patients met WHO criteria for MDS, including cytogenetic and molecular markers of clonality.

A novel finding of our study regards the efficacy of steroid treatment in about half of patients with cytopenia, with 26% achieving a complete recovery lasting for a median of 12 months. Interestingly, 19% of patients benefited from repeated steroid courses during the follow-up, while 42% required a further line after steroids (eltrombopag, cyclosporine, danazol, rituximab).

Erythroid and platelet responses to steroids were generally equally distributed; however, significantly better responses were observed in anemic patients with DAT positivity than in those with anti-platelet autoantibodies. Most anemic patients received steroids in combination with rEPO (7/10), due to non-response to the latter. Hence, we cannot exclude a possible synergistic effect, as observed in case 3 in whom the addition of rEPO further improved the erythroid response obtained with steroids. *Vice versa*, thrombocytopenic low-risk MDS patients have very limited treatment options, and steroids are mainly given *ex-adjuvantibus*[22-25]. Bone marrow stimulation by eltrombopag is under investigation with preliminary encouraging results deserving further confirmation[26-28]. In our series, three subjects received eltrombopag and one patient positive for anti-platelet autoantibodies responded.

Steroid doses were heterogeneous and largely dependent on the clinical intent (anti-inflammatory, immunosuppressive, or anabolizing). In particular, patients treated with a dose of 1 mg/kg day for overt autoimmune cytopenia reached a better response (60% complete responses), significantly more frequently than in the overall population (26%). This may advise the use of standard immunosuppressive doses, at least for a short course, to ascertain efficacy and properly silence the autoimmune pathogenic mechanism[29-31]. Short-term side effects should be managed, rather than maintaining the patient on a long-term, possibly harmful low dose. In fact, 16% of subjects experienced a steroid-related adverse event, including one case of fatal endocarditis, pinpointing the importance of assessing and preventing such events in this elderly frail population.

CONCLUSIONS

To our knowledge, this is the first systematic analysis of steroid use in low-risk MDS patients. Our study has several limitations, particularly regarding the limited number of patients and the retrospective nature. However, it provides a snapshot of real-life management of cytopenic low-risk MDS and suggests systematical testing for anti-erythrocytes and anti-platelets autoantibodies which may prompt steroid therapy.

AUTHORSHIP CONTRIBUTIONS

BF, FS, FV, GC, MDP and WB followed the patients, collected data, wrote the article and revised it for important intellectual content. LMC and MB performed flow cytometry tests and revised the article for important intellectual content. GAC revised bone marrow trephines and evaluated the manuscript for important intellectual content. NR performed immunohematological tests and revised the article for important intellectual content.

All data are available within the manuscript and are available upon reasonable request to the corresponding Author.

The Authors declare no conflicts of interest.

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