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Autoantibodies in sarcoidosis: Innocent bystander or promising biomarker for organ involvement?

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Abstract. *Background and aim:* Sarcoidosis is a rare inflammatory disease that can affect any organ in the body, but most commonly involves lungs and lymph nodes. Sarcoidosis is often considered an autoimmune disease, attributed to many factors, including autoantigen-specific T cells, antibodies producing B lymphocytes, autoimmune inflammation, although its exact cause and classification are still under debate. The aim of our study was to evaluate the possible role of autoantibodies, such as anti-nuclear (ANA), extractable nuclear antigen (ENA) and antiphospholipids, in sarcoidosis patients. *Methods:* We conduct a retrospective study on our patients with confirmed diagnosis of sarcoidosis involving lungs, lymph nodes and multiple organs, and we collected and analyzed data on blood and urine tests (C-reactive protein, CRP, amount of calcium in blood and urine, CD4/CD8 ratio, lymphocyte count), lung function, radiological patterns, ongoing treatments (steroid therapy, hydroxychloroquine or methotrexate, other immunosuppressive agents). *Results:* We enrolled 328 sarcoidosis patients, and we focused our attention on 32 patients with positive ANA antibodies (11%), observing a high percentage of them with sarcoidosis involving the lungs (77%), but more specifically a significant discrepancy, in percentage terms, in the blood CD4/CD8 ratio. In the ANA-positive group we observed 26% of patients with a high blood CD4/CD8 ratio (average CD4/CD8 ratio of 2.41), whereas in the ANA-negative group, patients with a high CD4/CD8 ratio (average ratio 1.78) represented a much smaller percentage (13%). This finding may be a source of further investigation for other studies on the topic. *Conclusions:* Analysis of autoantibodies expressed in our case series did not identify a specific autoantibodies pattern in sarcoidosis. Few studies have analyzed autoantibody patterns in sarcoidosis patients and involved smaller populations. In conclusion, our study evaluates a sizable population, and underlines the need for further, larger clinical studies to evaluate possible associations between sarcoidosis and autoimmunity.

Key words: sarcoidosis, autoantibodies, lung diseases

Introduction

Sarcoidosis is a rare inflammatory disease that can affect any organ in the body, but most commonly involves the lungs (more than 90% of patients) and lymph nodes. The exact cause of sarcoidosis is unknown, but it is believed to involve an abnormal

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immune response to certain triggers such as infections, environmental factors, or genetic predisposition. The incidence of the disease is higher among Scandinavians and African Americans and people aged between 40 and 55 years (1). Sarcoidosis is attributed to many factors, including autoantigen-specific T cells, antibodies producing B lymphocytes, autoimmune inflammation (2) and a possible genetic association between sarcoidosis and autoimmune diseases (3) has to be considered; a significant amount of parallels with this kind of diseases and also common variable immunodeficiency has been discovered in recent studies. The pathology is characterized by the presence of non-necrotizing granulomas in the affected organs (4). Pro-inflammatory cytokines produced by T-helper (Th) cells and macrophages trigger the inflammatory cascade and lead to granuloma formation, which is a vital pathological finding of sarcoidosis (5). The clinical presentation is widely heterogeneous: non-specific symptoms are generally cough, dyspnea, fatigue (6,7), low-grade fever, weight reduction, joint pain and night sweats. Sarcoidosis can be often entirely asymptomatic, and the diagnostic result may be completely coincidental; in some cases, the disease determines specific symptoms related to the organs involved. In most cases, pulmonary sarcoidosis presents with mediastinal lymphadenopathy and parenchymal infiltration. Skin involvement occurs in 20–30% of patients with erythema nodosum, profuse sweating, nodules, papules, and plaques. Regarding eye involvement (10–50% of patients), uveitis is the commonest form of ocular manifestation (8). Cardiac, neurological, renal, musculoskeletal, or gastrointestinal involvement is also possible (9). The symptoms of sarcoidosis can also be framed by two syndromic diseases (10): Löfgren's syndrome, characterised by fever, bilateral arthritis of the ankles and/or erythema nodosum and bilateral hilar lymphadenopathy on chest X-ray. This syndrome is highly suggestive of sarcoidosis (11); Heerfordt's syndrome is characterized by uveitis, enlargement of the parotid and submaxillary salivary glands and paresis of the cranial nerves, particularly the seventh cranial nerve (12). The clinical course of sarcoidosis is variable: sometimes it may remain asymptomatic even without treatment; in other cases, it may present acutely or follow a chronic course, which may be stable over time or progressive. In half of the cases, the disease resolves spontaneously within 2 years; in a significant number of patients, remission occurs within 5 years, while after more than 5 years, it is unlikely (13). The diagnosis of sarcoidosis can be difficult because it does not present typical symptoms. Radiological support (CT scan in particular) (14) possibly associated with histological confirmation by biopsy of the affected tissue (search for non-necrotizing granulomas in the affected organs) will certainly be essential (15). The aim of treatment for sarcoidosis, when necessary (e.g. in cases of progressive and/or symptomatic disease), is to reduce inflammation and manage symptoms. Corticosteroids are commonly prescribed to suppress the immune response (16). Other medications, such as immunosuppressants or biologics, may be an option in cases where corticosteroids are ineffective or cause significant side effects. Serious complications may occur if the disease affects vital organs such as the lungs, heart or nervous system. Sarcoidosis is often considered an autoimmune disease, although its exact cause and classification are still under debate (17). It shares some similarities with autoimmune diseases in terms of immune dysregulation, tissue inflammation (18), and involvement of immune cells and proteins implicated in autoimmune diseases, such as T cells and cytokines (19). Then there is the still-discussed role of B cells: polyclonal hyperglobulinaemia has historically been associated with pulmonary sarcoidosis and, more recently, several studies have documented serum positivity of antibodies directed against nuclear antigens (ANA) and other intracellular proteins (20). Autoantibodies are central markers of most autoimmune diseases, produced by inappropriately activated B cells. Data on the presence of autoantibodies in patients with sarcoidosis are limited and the relationship between autoantibodies and clinical laboratory findings remains unclear. Even at the therapeutic level, the use of corticosteroids and other immunosuppressive drugs, commonly used in sarcoidosis to reduce the immune response and inflammation, appears similar to the management of many autoimmune diseases (21). The aim of our study is to understand, within a disease that is not yet fully understood, whether autoantibodies such as ANA, ENA and antiphospholipids are exclusively bystanders of essentially independent immune mechanisms or whether acting on them may represent a different mechanism of approach to disease management.

Materials and Methods

As declared in the introduction, the aim of the study was to evaluate a possible relationship between sarcoidosis and auto-immune disease and at the same time to investigate the clinical immunological abnormalities in sarcoidosis. With this aim, we retrospectively analyzed clinical data from patients with histologically confirmed sarcoidosis.

Study population

This study included 328 patients with histologically confirmed sarcoidosis. Medical records between 2010 and May 2023 obtained from the Cattinara Hospital in Trieste (Italy) were examined. First, other possible causes of granulomatous diseases (tuberculosis, bacterial and fungal infections) and tumours were excluded. Evaluation of antinuclear antibodies (ANA), extractable nuclear antigens (ENA), anti-β2-glycoprotein I (β2GP1) and anti-cardiolipin was performed. The diagnosis of sarcoidosis was made based on clinical manifestations, high-resolution computed tomography (HRCT) of the chest and pathological findings supporting non-caseous granulomas with biopsies collected from lungs and lymph nodes.

Data collection and analysis

We reviewed information through medical records:

1. demographic features, organ involvement and treatment procedures (steroids, hydroxychloroquine, methotrexate),
2. lung function tests values (FEV1, FVC, DLCO),
3. laboratory parameters, including ACE, lymphocytes count, C-reactive protein (CRP), CD4/CD8 ratio, amount of calcium in urine and blood, an autoantibody profile, including ANA, anti-ENA panel, anti-β2-GP1 and anti-cardiolipin,
4. imaging findings, including chest HRCT, gallium bone scan and PET-CT.

Statistical analysis

Categorical variables were summarized as a number (percentage); continuous variables were summarized using the mean (standard deviation) or median (interquartile range), as appropriate. The normality of the continuous variables was tested using the Shapiro-Wilk test. Between-group differences for categorical and dichotomous variables were tested with the chi-square test, while differences for continuous variables were assessed with the independent-samples t-test or the Mann-Whitney test, when the assumptions for the parametric test were not fulfilled. Statistical significance was tested using a two-tailed p-value <0.005. For confounding adjustment, a multivariable logistic regression was conducted on all variables considered relevant (age, sex, ANA positivity and CD4/CD8 ratio). All analyses were performed with R software.

Results

A total of 328 patients were enrolled, of whom 156 (48%) were women and 172 (52%) were men, with a mean age of 57 years. The mean age at diagnosis of sarcoidosis was 48±8 years. Except for 3 patients (2 black and 1 Pakistani respectively), all were white subjects. 100 patients, or 31% of the total, were smokers. Table 1 summarizes the main characteristics of the patients in our study. In particular: 77% of patients (251) showed lung involvement, with a parallel positive radiological finding for the disease in 72% of subjects undergoing HRCT of the chest and 70% of those undergoing PET-total body CT. Lymph node involvement was detected in a similar number of cases (271), accounting for 83% of enrolled patients. Finally, 42% of patients showed pathological involvement in other organs. The percentage of subjects with pulmonary embolism remained low at 4% of the enrolled patients, which is about half that of patients with positive ANA antibodies (32 individuals) or a high CD4/CD8 ratio (28 patients). Analyzing the average spirometric data of the enrolled patients, the mean percentage value of dynamic flows is within the normal range, as is that of alveolar-capillary diffusion for carbon monoxide.

As for performed treatments, in April 2023 just over half of examined patients were under therapy with oral corticosteroids; about other two therapeutic examined options, from diagnosis time to April 2023 approximately one third of total patients (101) received hydroxychloroquine while just under half (45%) received therapy based on methotrexate.

Table 1. Characteristics of study population.

	N=328
Age, mean (min, max)	57 (50, 67)
Age at diagnosis, mean (min, max)	48 (40, 56)
Male, n (%)	172 (52%)
Race	
Caucasian, n (%) Black, n (%) Asian, n (%)	325 (99%) 2 (0.6%) 1 (0.4%)
Lung involvement, n (%)	251 (77%)
Node's involvement, n (%)	271 (83%)
Other organ's involvement, n (%)	136 (42%)
Serological features	
ANA pos, n (%) Blood CD4+/CD8+ ratio> 2.5, n (%) 24H Calciuria > 250 mg/24 h, n (%)	32 (11%) 28 (14%) 110 (42%)
Lung function	
FEV1, % mean (min, max) FVC, % mean (min, max) Tiffenau index, mean (min, max) DLCO, mean (min, max)	98 (85, 110) 104 (91, 116) 0.78 (0.73, 0.82) 88 (75, 102)
Treatment	
Corticosteroids, n (%)	152 (53%)
Hidroxychloroquine, n (%)	101 (32%)
Methotrexate, n (%)	142 (45%)

Table 2. Characteristics of subgroup divided according to ANA positivity.

	ANA-, N=252	ANA+, N=32
Age at diagnosis, mean (min, max)	48 (40, 55)	52 (41, 56)
Lung involvement, n (%)	194 (77%)	27 (84%)
Node's involvement, n (%)	207 (82%)	25 (81%)
Other organs' involvement, n (%)	151 (60%)	18 (56%)
Blood CD4+/CD8+ ratio> 2.5, n (%)	19 (13%)	6 (26%)

Differences between patients with ANA positivity and negativity

Focusing our attention on 32 patients with positive ANA antibodies, we see a high percentage of them with sarcoidosis involving lungs (84%), confirmed by the percentage of compatible pattern at CT scans (81%), like that of subjects with lymph node involvement (25 patients, 81%) (Table 2).

The ANA 1:80 pattern represents the majority of cases of positivity for ANA antibodies (65% of patients). We found the 1:160 pattern in 5 patients, while the 1:320, 1:640, and 1:1280 patterns were found in 3, 2 and 2 patients, respectively. In 5 patients with ANA title 1:160, sarcoidosis showed a pulmonary involvement and in 4 of these 5 subjects the lymph node area was also affected. In all these patients OCS therapy was performed and in 4 of these MTX was associated. In both 2 cases of hypercalciuria, the addition of HCT was decided. We also report that in 2 of these 5 patients ocular involvement was found, in one case of the lacrimal glands, in the other with cases of recurrent uveitis.

In these 7 cases we found multidistrict sarcoidosis, with lung involvement in 100% of cases, lymph node involvement in 6 out of 7 subjects, all treated with OCS; the latter was used in association with MTX in 3 subjects, without distinction based on the titer found. Still remaining in autoimmune field, in our sample we identified 12 patients with a positive history of autoimmune thyroiditis, in two cases with concomitant positive ANA autoantibody pattern, with titers of 1:320 and 1:160 respectively. We also report 5 patients with seronegative spondylarthritis and 3 cases of rheumatoid arthritis. Approximately two thirds of these 32 patients with ANA antibodies were still on oral steroid therapy at the time of data collection, while exactly 50% had received methotrexate since diagnosis and only one third had received hydroxychloroquine. The greatest discrepancy, in percentage terms, between patients with ANA-negative and positive antibodies is evident in the CD4/CD8 ratio, with 26% of ANA-positive patients having a high ratio (mean CD4/CD8 ratio of 2.41) compared to 13% of ANA-negative subjects (mean CD4/CD8 ratio 1.78). The same higher ratio is evident in subjects with clinically confirmed lung involvement compared to those in whom sarcoidosis did not cause respiratory symptoms (Table 3-5).

In subjects undergoing oral steroid (OCS) therapy, no significant differences were highlighted from the point of view of blood exams tests compared to subjects not using OCS.

Pulmonary embolism in patients with sarcoidosis

Regarding the frequency of pulmonary embolism in patients with sarcoidosis, also in this case we

Table 3. Characteristics of subgroup divided according to lung involvement.

	Lung involvement yes, N=251	**Lung involvement no, N=75**
Age at diagnosis, mean (min, max)	48 (40, 57)	47 (39, 55)
Serological features		
24H Calciuria > 250 mg/24 h, n (%) Blood CD4+/CD8+ ratio> 2.5, n (%) ACE, mean (min, max) PCR (mg/L), mean (min, max)	83 (42%) 26 (17%) 27 (14, 48) 2.6 (1.2, 6.1)	27 (47%) 2 (4%) 28 (18, 40) 2.9 (1, 6.1)

Table 4. Serological features of patients treated with steroids at time of data collection.

	Ongoing steroids treatment yes, N=132	**Ongoing steroids treatment no, N=152**
24H Calciuria, mg/24h mean (min, max) Blood CD4+/CD8+, mean (min, max) ACE, U/L mean (min, max) PCR (mg/L), mean (min, max)	226 (135, 320) 1.8 (1.14, 2.75) 24 (13, 46) 2.6 (1, 5.6)	210 (104, 328) 1.8 (1.3, 2.7) 28 (18, 47) 2.2 (1.2, 5.1)

Table 5. Serological features of patients treated with Hydroxychloroquine.

	Ongoing Hidroxychloroquine treatment yes, N=101	**Ongoing Hidroxychloroquine treatment no, N=217**
24H Calciuria, mg/24h mean (min, max) Blood CD4+/CD8+, mean (min, max) ACE, U/L mean (min, max) PCR (mg/L), mean (min, max)	284 (184, 392) 1.8 (1.2, 2.7) 30 (17, 56) 2.5 (1.1, 4.7)	183 (110, 274) 1.8 (1.3, 2.78) 25 (17, 56) 2.6 (1, 6.6)

Table 6. Characteristics of study population divide according to intercurrent pulmonary embolism (PE).

	PE-, N=310	**PE+, N=14**
Age at diagnosis, mean (min, max)	48 (39, 56)	50 (46, 60)
Male, n (%)	166 (54%)	5 (36%)
Smoking history yes	96 (32%)	2 (14%)
Serological features		
ANA pos, n (%) Blood CD4+/CD8+ ratio> 2.5, n (%) 24H Calciuria > 250 mg/24 h, n (%) ACE, U/L mean (min, max)	30 (11%) 25 (13%) 105 (43%) 28 (15, 47)	1 (7%) 3 (30%) 4 (40%) 21 (17, 36)
Treatment		
Corticosteroids, n (%)	143 (53%)	9 (64%)
Hidroxychloroquine, n (%)	94 (31%)	7 (50%)
Methotrexate, n (%)	133 (44%)	8 (57%)

obtain confirmation of already known numbers, with percentage values near 4% (Table 6).

The only patient with a known PE+ and ANA+ autoantibody pattern presented a lymph node sarcoidosis (with joint and skin involvement and hypercalciuria, without lung disease) with multiple comorbidities, and a high CD4/CD8 ratio of 3.3.

Discussion

In this study, we set out to understand whether autoantibodies such as ANA, ENA and antiphospholipids in sarcoidosis are exclusively spectators of essentially independent immune mechanisms or whether acting on them might represent a different

mechanism of approach to disease management. 77% of patients (251) showed lung involvement, with a parallel positive radiological finding for the disease in 72% of patients undergoing thoracic HRCT and 70% of those undergoing total body PET-CT (6,22,23). However, 30-60% of patients with pulmonary sarcoidosis are asymptomatic, an aspect of this disease that makes it more difficult to understand. Lymph node involvement was evidenced in a similar number of cases (271), i.e. 83% of enrolled patients (8). Finally, 42% of patients showed pathological involvement in other organs. Both the numbers of pulmonary and lymph node involvement reflect what is known in the literature (1). The percentage of individuals with pulmonary embolism (PE) remained low at 4% of the enrolled patients, however, this is a significantly higher rate (24) than that found in the general population, where PE cases are approximately 60-70 per 100,000 individuals. More specifically, patients with a reduced blood ratio of CD4 to CD8 lymphocytes and antiphospholipid antibody positivity are at increased risk of PE. A 35-year record linkage study by Crawshaw et al. reported a two-fold higher rate of PE in patients with sarcoidosis. Several studies have reported that sarcoidosis, like other chronic inflammatory conditions, is associated with an increased risk of venous thromboembolism (VTE) (25–28). One of the hypotheses to be considered is that chronic inflammation due to sarcoidosis induces damage at the level of endothelial cells, with release of inflammatory cytokines and activation of the coagulation cascade; in addition, a reduced ratio of CD4 to CD8 lymphocytes may be involved (27–36). However, the underlying cause of the association between sarcoidosis and PE remains speculative (27,28,31–44). Regarding the relationship between sarcoidosis and autoantibodies, the main focus of this study, as reported in the results, there is a significant proportion (26%) of patients with ANA-positive and high CD4/CD8 ratio (mean CD4/CD8 ratio of 2.41) compared to 13% of patients with ANA-negative (mean CD4/CD8 ratio 1.78). This finding is especially significant considering that it occurs in subjects with confirmed lung involvement (84%) compared to those in whom sarcoidosis did not cause respiratory symptoms. The involvement is confirmed by the percentage of compatible pattern on CT scan (81%), which is also similar to that of subjects with lymph node involvement (25 patients, 81%). This autoimmune pattern requires further confirmation before it can be recognized as a sarcoidosis phenotype, considering a fair but not so large number of patients, i.e. there are no clinical/therapeutic elements to be able to recognize it as a phenotype. In the presence of an ANA title greater than or equal to 1:160 in 83% of cases, multi-organ forms of sarcoidosis are found, with an indication for combination with MTX, with a view to containing the side effects of OCS therapy, given the probable need for therapy over several months. Remaining in the field of autoantibodies, the presence of antiphospholipid antibodies in patients with sarcoidosis, on the other hand, is already known to be correlated with a poor prognosis (24,31–38,45,46) with more frequent extrathoracic involvement, the persistence of radiographic changes (38–42) and an increased risk of venous thromboembolism (47). On the other hand, the association between sarcoidosis and antiphospholipid antibodies has a reduced frequency (41,42). The percentage of patients with ANA antibodies in our study is lower than in other works dealing with this topic, where 11 of the 26 patients with sarcoidosis (328 subjects enrolled in our study) had ANA antibodies, i.e. 42.3% (vs. 11% in our work) (48); in general, positivity is usually detected in up to 28-30% of patients with sarcoidosis (49,50). It is always advisable to investigate the autoantibody pattern in patients with sarcoidosis because it may highlight an increased risk of developing connective tissue diseases, both in the short and long term, as well as being an indirect indicator of B-cell hyperactivity potentially attributable to sarcoidosis activity. All the previously mentioned studies considered smaller populations than our case series, so our results corroborate literature on a larger scale. Starshinova et al. (51) give different examples of how pulmonary sarcoidosis can coexist with other autoimmune diseases, suggesting the possibility of a common pathogenesis and genetic predisposition between sarcoidosis and Sjogren's syndrome (both present an association with the HLA-DR3 genotype and high levels of CD4+ lymphocytes) (52,53), between sarcoidosis and SLE (the onset of SLE with the detection of non-caseating granulomas in the lungs and skin has been highlighted; in both pathologies, we see an increase in immunoglobulin values and changes in the ratios of B and T lymphocytes) (54), between sarcoidosis and ankylosing spondylitis (again, cases of non-caseating granulomas in the lungs have been highlighted in conjunction with an

involvement of the HLA-B27 genotype typical for ankylosing spondylitis) (55). Furthermore, observing the granuloma formation pathway typical of sarcoidosis, a mechanism opposite to what occurs in a pathology with a different origin such as tuberculosis is observed, with a prevalence of M2 type macrophages (typically anti-inflammatory) and the interest of the mTOR pathway in granuloma formation (56). A recent Chinese study provides a randomized study which suggests significant genetic evidence supporting a causal effect of predictors of autoimmune diseases, such as celiac disease, T1DM, and IBD, on sarcoidosis (3). On the other side, a study from D'Alessandro et al. provided substantial elements confirming the immunological peculiarity of sarcoidosis. It shows a reliable dysregulation of expression of immune checkpoint molecules (PD1, CTLA4, TIGIT) in different immune cells which may significantly contribute to the formation and chronicization of granuloma (57). All these experiences continue to deepen the fundamental theme of the pathogenesis of sarcoidosis in all its facets, both from a purely immunological and genetic point of view. The definition of this aspect of the disease represents an essential element to progress also from a therapeutic point of view, with a view to identifying effective target-specific therapies as soon as possible (2). A recent experience (58), following this last option, chose Tofacitinib, a type of Janus kinase (JAK) blocker approved for several refractory immune-mediated inflammatory diseases (rheumatoid arthritis, psoriatic arthritis, ankylosing spondylitis and ulcerative colitis) in order to act as an inhibitor of inflammatory immune responses. Favorable clinical responses have been described in patients with refractory cutaneous and/or pulmonary disease and could represent a fourth-line therapy in severe refractory sarcoidosis with multiorgan involvement. Another example is efzofitimod, a first-in-class biologic which selectively binds neuropilin-2 (NRP2); it seems to be associated with improved quality of life with a trend towards reduced glucocorticoid use and stable to improved pulmonary function (59,60), but further evaluation of this approach is needed. Returning to the results obtained, as mentioned above, the average spirometric data of the enrolled patients did not show any significant alterations with respect to normal values. From a functional point of view, alterations such as ventilatory limitation, pulmonary involvement and deconditioning represent the main causes of activity limitation in patients with sarcoidosis (61). However, as pulmonary function tests (PFTs) are not sufficiently reliable to predict functional limitation during exercise in patients with sarcoidosis, the implementation of functional analysis by means of cardiopulmonary exercise testing (CPET) could be considered a useful method to detect exercise tolerance in these patients (62). Another useful, low-cost, renewable, repeatable, and acceptable tool to investigate exercise capacity could be the Six-Minute Walk Test (6MWT), also for prognostic purposes in patients with heart failure and lung disease (61). Focusing on the therapies used in sarcoidosis, in April 2023, slightly less than half of the patients examined were receiving oral corticosteroids; since diagnosis, approximately one third of the sample had received hydroxychloroquine, while slightly less than half (45%) had received methotrexate therapy. Prednisone is one of the most widely used corticosteroids (13). The minimum effective corticosteroid dose differs from patient to patient and for different organs. Corticosteroid therapy is associated with significant long-term toxicity, and a worse quality of life for those who received higher cumulative dose of OCS. Looking at the treatment of the 32 patients with ANA positivity, the percentages do not differ from what has already been seen, with about one third of the patients receiving OCS at the time of data collection. Other studies also recommend close monitoring of haematochemical and radiological status in patients with sarcoidosis and ANA antibody positivity (48). Several agents have been shown to be steroid sparing for sarcoidosis, including methotrexate (63). As noted in the literature, in a double-blind, placebo-controlled study, methotrexate did not demonstrate a worsening of lung function, but did lead to lower prednisone use and weight gain. In conclusion, hydroxychloroquine is usually chosen to treat cutaneous, bone and joint sarcoidosis, as well as hypercalcemia and certain types of uveitis (64,65). Its use is therefore also related to blood and urine calcium levels, unlike the other two treatment options, where, according to our data, haematochemical results do not significantly influence the choice of therapy. Comparing the therapeutic management of our patients with that of the work of Cattelan et al. (48), a lower percentage of patients (53%) received OCS after a diagnosis of sarcoidosis, compared with 19 of the 26 subjects in their work. On the other hand, a higher share received MTX

(45% vs 30.8%), probably due to a more long-term data collection in our case.

Conclusions

The results obtained in our study are in line with others treating sarcoidosis, with regard to the prevalence of pulmonary and extrapulmonary involvement, the number of cases of pulmonary thromboembolism, the low relevance of pulmonary function tests and the treatment options. The results concerning the autoimmune pattern are consistent with the data already known in the literature and confirm them on a larger scale. Statistical analysis of our data, however, did not reveal an autoantibody pattern that could be associated with sarcoidosis. The finding of immune dysregulation patterns in sarcoidosis and different autoimmune diseases may contribute to give us an opportunity for the development of biological therapies to treat sarcoidosis. Although our study has limitations, e.g. the single-centre design of the study and the need for further confirmation to define an autoimmune pattern as a phenotype of sarcoidosis, it allows us to further confirm the available data and to identify a different area of investigation of a disease that is still often difficult to manage, indicating the autoimmune pattern (and in particular the ANA-CD4/CD8 ratio correlation) as deserving at least classification and monitoring from diagnosis to the subsequent therapeutic phases.

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