

EDITORIAL

Corticosteroids in sarcoidosis: friend or foe?

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Sarcoidosis is a chronic, multiorgan disorder of unknown aetiology, characterized in affected organs by an accumulation of T lymphocytes and mononuclear phagocytes, noncaseating epithelioid cell ("immune") granulomas and tissue injury [1–3]. The general paradigm of immune granuloma formation suggests a specific, T-cell-mediated response to an antigenic agent that has been processed by macrophages and presented to antigen-specific T-lymphocytes. The T-cell, in turn, directs the accumulation and differentiation of mononuclear phagocytes in the local microenvironment [1, 2, 4]. The disease can present in an acute or subacute form and is often self-limited, but in many other cases it is chronic, with variable disease activity over many years.

Corticosteroids have been the mainstay of treatment for more than 30 yrs, but there is still argument about their place in management. This is coupled to a certain nihilism about a group of drugs of which the side-effect profile is receiving increasing publicity. Whilst it is entirely appropriate that potential benefits of a treatment should be balanced against the risk of side-effect and must be discussed fully with each patient, the enormous benefits gained by many patients with chronic inflammatory diseases must also be part of that discussion.

The aim of this article is to review the use of corticosteroids in sarcoidosis, with particular emphasis on persistent pulmonary disease, by addressing three issues: 1) Is corticosteroid therapy a rational approach?; 2) Do they modulate disease?; and 3) Do they affect long-term outcome?

Is corticosteroid therapy a rational approach?

The rationale on which sarcoid therapy is based centres on the considerable advances which have been made in the understanding of the pathogenesis of the disease over the last decade, and owes much to the advent of cellular and molecular methodologies. An overview of the critical components of pathogenesis is necessary to put into perspective the goals of treatment.

Pathogenesis of sarcoidosis

Aetiology

There is abundant evidence to support the concept that sarcoid granulomas are formed in response to a persis-

tent, poorly degradable, antigenic stimulus. Firstly, immune granulomas are known to form in response to persistent antigenic epitopes present after infection with organisms, such as schistosomiasis or mycobacterial infection. Secondly, pulmonary immune granulomas may be initiated by infectious agents, inorganic agents, and organic particulates: the factor common to all this is their low biodegradability and/or persistence, often within macrophages [5].

The cause of sarcoidosis has been sought for almost 100 yrs. Historically, a relationship with tuberculosis akin to the two forms of leprosy (multibacillary lepromatous disease and paucibacillary tuberculoid disease) has been suspected, but never substantiated. The issue has been given new life by the application of molecular biology techniques. Using mycobacteria-specific primers and the polymerase chain reaction, mycobacterial deoxyribonucleic acid (DNA) molecules were identified in granulomatous tissue (lung skin or lymph node) in one study in 7 out of 16 cases, compared to 1 out of 16 normal controls, but in only 2 out of 4 tuberculosis controls [6]. A second study, by a different group, reported contradictory findings: tissue or cell samples from 16 sarcoidosis patients were compared with 13 control tissue samples, four normal bronchoalveolar lavage samples and 11 normal volunteer lavage samples. Most of the sarcoidosis samples were negative, using *Mycobacterium hominis* specific primers and a technique sensitive enough to detect two mycobacterial genomes - the equivalent of 15 organisms·10⁻⁶ human cells [7]. It is also noteworthy that the coexistence of sarcoidosis and tuberculosis has been recognized for some time [8]. A final judgement on this issue will require more extensive investigations.

T-cell triggering

Whatever the nature of the initiating agent, it is recognized that the antigenic epitopes that drive T-cell responses are "hidden" in the tertiary structure of the native molecules, and are "exposed" by processing within the macrophage and other antigen-presenting cells and co-expressed, as small peptide fragments, on its surface, together with class II major histocompatibility complex (MHC) molecules [9, 10]. This is in contrast to epitopes that drive B cell responses, which are usually on the external regions of the intact protein [10]. Dendritic cells are more potent presenters of antigen than alveolar macrophages. Class II MHC molecules are crucial for

antigen presentation to CD4+ T-cell and, therefore, necessary for the subsequent development of a cell-mediated immune response [11–15].

In sarcoidosis, there is evidence that alveolar macrophages, in comparison to those from normal individuals, are capable of increased presentation of nonrelevant antigen, such as tetanus toxoid, to lymphocytes [16, 17].

Further evidence that lung macrophages are presenting antigens to T-cells in sarcoidosis comes from analysis of the numbers of T-cell surface antigen receptors on lung T-cells. Recognition of antigen-class II complexes by specific T-cell antigen receptors (TCRs) is an absolute prerequisite for an antigen to be capable of driving a T-cell response. One consequence of T-cell antigen receptor triggering is the downregulation of surface TCR molecules and the upregulation of TCR messenger ribonucleic acid (mRNA) transcript numbers [18, 19]. If TCR triggering occurs in sarcoidosis, it would be expected that T-cells from active disease sites would exhibit the appropriate surface and mRNA changes. This is the case: compared to blood T-cells, sarcoid lung T-lymphocytes exhibit a decrease in density of surface TCR molecules, and they express increased numbers of TCR β -chain mRNA transcripts [20]. This downregulation of TCR may explain the reduced proliferative response of lung T-cells to recall antigens than lung cells obtained from normal individuals [21].

Consequences of T-cell triggering

There are a number of results of the initial direct macrophage T-cell interaction, which are of importance in the development of the granuloma. These involve the expression of a number of cell surface molecules on macrophages and lymphocytes and their release of key cytokines.

Mononuclear phagocytes

There is evidence from a variety of sources that the mononuclear phagocytes participating in the granulomas in sarcoidosis are recruited both from the blood and from local proliferation, and are activated [22–26]. In this respect, studies of lung macrophages using monoclonal antibodies have demonstrated that the cells bear the phenotype of young, recently recruited monocytes [22, 26]. The pulmonary mononuclear phagocyte population in sarcoidosis is also proliferating at a rate that is two to three times normal [23]. Consistent with the concept that lung mononuclear phagocytes are activated, the surface density of all categories of class II MHC molecules is increased on these cells [24, 25].

Macrophage cytokines are almost certainly involved in the development of sarcoid granulomas, although the evidence for this is not unequivocal. Evaluation of the release of interleukin-1 (IL-1) by alveolar macrophages from individuals with sarcoidosis has led to conflicting reports; some studies show no increase, compared with controls [27–29]. Of two more recent studies, one has

shown that alveolar macrophages from patients with sarcoidosis spontaneously secrete IL-1, tumour necrosis factor- α (TNF- α) and prostaglandin E₂ (PGE₂), but that the amounts secreted from patients with active disease did not differ significantly from patients with inactive disease [30], whereas, the second study demonstrated IL-1 secretion only in "active" disease [31].

Spontaneous alveolar macrophage secretion of TNF- α has been found to be increased in patients with sarcoidosis in three studies [31–33], but no different in another study [34]. However, when normal macrophages are cultured in the presence of IL-2, a cytokine known to be secreted spontaneously by lymphocytes obtained from the lower respiratory tract of patients with sarcoidosis, increased amounts of TNF α mRNA transcripts are found [35]. Because lung CD4+ MHC class II+ T-cells are releasing IL-2 in sarcoidosis [36], it is possible that CD4+ T-cells are activating genes, such as TNF α , in lung macrophages in this disease. Other macrophage products which are released from cells obtained from sarcoidosis patients include the granulocyte colony-stimulating factor (G-CSF) (inducer of granulopoiesis and neutrophil activation) [37].

Lymphocytes

Because the aetiology of sarcoidosis is unknown, the major evidence that the granulomas are immune-based comes from analysis of the lymphocyte populations from lavage and tissue samples. Although sarcoidosis is a systemic disorder, the lymphocyte populations are strikingly compartmentalized. At sites of disease activity, such as the lung, there is a marked expansion in lymphocyte numbers, whereas in the blood there is a lymphopenia [38]. Consistent with the concept of compartmentalization, the lung lymphocytes are dominated by CD4+ T-cells, with the ratio of CD4+ to CD8+ populations within the lung being 3–10:1. In comparison, the CD4+ to CD8+ ratio in normal lung is 2:1; approximately 1–1.5:1 in sarcoid blood, and 1.5–3:1 in normal blood [38]. The T-cells within the lung are proliferating spontaneously at an exaggerated rate, and show evidence of activation [39] and express the surface markers of activation, including high affinity IL-2 receptors (IL-2R), human leucocyte antigen-DR (HLA-DR) class II MHC molecules, and the very late activation antigen-1 (VLA-1) marker of late activation [40–42]. Most T-cells are CD45RO+ (in common with normal lung T-cells) which denotes that they are of the "antigen-primed" subset of T-cells and are leu-8-. Furthermore, they can express the epitopes identified by the antibodies Ki67 and PCNA, which indicate that they are proliferating [43, 44]. This is consistent with the observation of spontaneous proliferation *ex vivo* in cells lavaged from patients with active disease. In contrast to lung, blood T-cells in active pulmonary sarcoidosis appear relatively quiescent, with no evidence of lymphokine release [45]. However, although some blood T-cells do express activation markers, such as IL-2R and class II MHC molecules, these cells probably represent T-cells which are activated in tissues, such

as the lung, and which have migrated to blood [45, 46].

Antigen presentation, in association with secondary cytokine signals, stimulates T-cells to release a number of lymphokines that activate macrophages and lymphocytes and provide the building blocks from which granulomas are formed [1, 2, 47]. The defined lymphokines of most relevance to the generation of granulomas are interleukin-2 (IL-2; the T-cell growth factor), and interferon-gamma (IFN- γ ; a potent activator of macrophages). Consistent with this concept, monocytes and alveolar macrophages can be stimulated *in vitro* by IFN- γ to increase the expression of class II surface antigens, Fc receptors, IL-2 receptors, and transferrin receptors. Furthermore, under appropriate circumstances, mononuclear phagocytes will form giant cells under the influence of IFN- γ . In addition, lung lymphocytes express mRNA for the granulocyte-monocyte colony-stimulating factor (GM-CSF; stimulator of granulopoiesis and monopoiesis) [48, 49], and monocyte colony-stimulating factor (M-CSF; stimulator of monocyte proliferation) [50]. The relative contributions of the lymphokines to granuloma formation are not known.

Tissue immunohistochemistry studies have confirmed that lavage cells are representative of those in the lung parenchyma [51, 52] and *in situ* hybridization has reinforced the concept that cytokines have important roles in the generation of immune granulomas. A detailed study of cytokine expression in lymph nodes has demonstrated the presence of IL-1 α and β , TNF α , IL-2, IL-6, and IFN- γ *in situ*; and, importantly, that there is a spatial arrangement of cytokine producing cells: IL-1 β , TNF α and IFN- γ are restricted to the granuloma itself, whereas the other cytokines are distributed in cells more randomly [53]. Strikingly, the most avidly expressed product was IL-1 β , whilst IFN- γ was 32 fold more abundant than IL-2. These data are consistent with the concepts of spatial arrangements of cells in immune granulomas in sarcoidosis, defined by CAMPBELL *et al.* [51] and MUNRO *et al.* [54] using monoclonal antibodies to identify cell phenotypes. It is against this background that we must assess the logic of treating the disease with corticosteroids.

Corticosteroids

This group of drugs has played a crucial role in the management of all forms of chronic inflammatory conditions affecting the lungs and other organs. Corticosteroids are now known to function by entering the cytoplasm of inflammatory cells and binding with steroid receptors located within the cytoplasm. The receptors exist in an inactive form, bound to two 90 kD heat shock protein molecules. Following the association of steroid with steroid receptor, the heat shock protein molecules dissociate, thereby exposing a DNA binding domain. The steroid/receptor complex then enters the nucleus, where binding occurs to glucocorticoid response elements, generally located in the upstream promoter regions of cytokine

and other genes resulting in modulation of gene transcription.

Do corticosteroids modulate disease progression?

Until the cause of sarcoidosis is identified and means found to eradicate the trigger from affected organs, the disease can only be controlled rather than cured. Corticosteroids have been the mainstay of treatment for over 30 yrs and appear to be the most effective drug available, but what is the evidence that they modulate disease? This question can be answered in two ways; firstly, by showing that steroids affect important mechanisms of relevance to pathogenesis; and, secondly, by demonstrating an effect on clinical indices.

Corticosteroids and pathogenesis

Table 1 illustrates the major cell surface and cytokine targets of corticosteroids. From the table it can be seen that a number of genes which play important roles in the pathogenesis of sarcoidosis are modulated by corticosteroids. Of particular relevance are genes which are expressed soon after antigen triggering of CD4+ T-cells: IL-2, IFN- γ , IL-1 and the IL-2 receptor IL-2R [55]. Other genes which may play important roles in pathogenesis by influencing the expression of cell adhesion molecules (TNF α) and leucocyte/monocyte proliferation and activation (GM-CSF, G-CSF) are also regulated by corticosteroids [55–58].

There are, therefore, good data to suggest that steroids can impact on mechanisms important at an early stage of the pathogenic process.

Corticosteroids and clinical efficacy

Many studies have shown quite conclusively that steroids can improve chest radiographic features and lung function measurements, and these include some of those studies which conclude that there was no long-term benefit on disease outcome (but see below) [59–64]. ODLUM and FITZGERALD [62] studied six patients who had been untreated for a period of 6.8 ± 2.4 yrs and whose disease was demonstrated to be progressing. On treatment with steroids, dyspnoea lessened, lung volumes and gas exchange improved, associated with improvements in chest radiography and gallium scans. In this group of patients defined by their chronicity, sustained improvement (mean of 22 months) was seen despite lengthy periods during which disease had deteriorated and no treatment has been given.

TURNER-WARWICK *et al.* [64] treated 32 patients with a standard treatment which was then titrated to maintain maximum improvement. Results showed that chest radiographs improved in all 32 and were not related to the initial BAL lymphocytes count.

ISRAEL *et al.* [61] studied 83 patients. Treatment consisted of three months of prednisone at 15 mg-day⁻¹.

Table 1. – Cytokines, relevant to sarcoidosis, modulated by corticosteroids

Interleukin 1 and 2	IL-1, IL-2
Tumour necrosis factor- α	TNF α
Granulocyte-macrophage colony-stimulating factor	GM-CSF
Interferon- γ	IFN- γ
Interleukin-2 receptor	IL-2R
IL-1 inhibitor (increased)	

Significant improvement was seen in the treated group with Stage II and III disease by comparison with the untreated group, but no difference was observed in patients with Stage I disease.

SELROOS and SELLERGREN [60] studied 39 patients with Stage II disease of less than 5 yrs' duration. Patients were randomly allocated to a treatment and a nontreatment group. Treatment consisted of a tapering regimen of methylprednisolone, starting at 24–32 mg-day⁻¹ for a total of seven months, with half the treated group receiving alternate day therapy for all but the first two weeks of treatment. Treated patients were found to have improvement in chest radiography, forced vital capacity (FVC) and diffusion capacity for carbon monoxide.

In a study of disease relapse after corticosteroid withdrawal, chest radiography showed increased changes in 7 out of 10 patients who had a symptomatic relapse, by comparison with 0 out of 5 with no relapse; chest radiography improved in all 10 relapsers when steroids were reintroduced and this was accompanied by an improvement in spirometry [63].

Other studies have, however, shown less significant differences between treated and untreated groups. ZAKI *et al.* [65] enrolled 183 patients into a controlled, double-blind therapeutic study and found only trends towards a significant improvement in a treated group. YOUNG *et al.* [66] showed no benefit of treatment at six months or 1–2 yrs.

An inherent weakness of these studies is that mixed groups, including appreciable numbers with "early" disease, were recruited, which meant that a significant percentage of the untreated (and treated) patients would have improved spontaneously, a feature which is well-recognized in sarcoidosis. A second problem is that treatment was not individually tailored, and all patients received the same treatment regimen irrespective of other factors, such as disease extent. These same criticisms apply to the evaluation of the longer term benefits of steroid therapy.

Effect of corticosteroids on long-term outcome

It is the role of steroids in modulating long-term outcome which has drawn the most disagreement among those involved in treating patients with sarcoidosis. Although the prevention of progressive lung injury is one of the most fundamental goals in sarcoidosis, there have been no properly designed studies which have explored this aspect of management, but a number of studies have purported to do so.

ISRAEL *et al.* [61] used a three month treatment period of 15 mg-day⁻¹, in a placebo-controlled, double-blind study and evaluated outcome at a mean of 5.4 yrs. YOUNG and co-workers [66, 67] compared treatment for six months (six patients) and up to 2 yrs (six patients) against no treatment and assessed outcome at 1–2 yrs and, in a subsequent report, at 10–15 yrs: 11 of the 12 treated patients received no treatment beyond the 2 yrs period. Both of these studies showed no significant differences between treated and untreated groups at the end of the follow-up period. However, neither these nor other studies have attempted to produce maximum improvement and to then titrate the dosage of therapy to maintain that improvement (this also entails an estimate of prevention of disease progression). It is, perhaps, not surprising that a standard regimen administered for a limited period has no effect on outcome 5–10 yrs later; and, indeed, even ISRAEL [68], in a critical editorial in 1987, entitled "Corticosteroid treatment of sarcoidosis - who needs it?" concluded ".....patients frequently relapse after termination of treatment, and close surveillance is necessary for several years afterward". At present, the body of evidence indicates that steroids can modulate disease but that it has not been proven if longer term treatment can prevent disease progression, lung injury and fibrosis.

Inhaled corticosteroids

Few studies have explored the role of inhaled steroids in the management of sarcoidosis, even though the disease is widely recognized to be "bronchocentric", with the major site of the inflammatory infiltrate involving the peribronchiolar regions and airflow obstruction being a feature of more longstanding disease and, in one study, being present in a significant number of patients at presentation [69]. If pulmonary disease could be modulated by inhaled therapy, there would be less reluctance to intervene with steroids, as the cost/benefit ratio of commencing treatment early would be low. Definitive studies have yet to be performed, although SELROOS [70] reported some success in a small number of patients treated with budesonide. In an open study, 20 patients were treated with up to 1,600 μ g of budesonide daily for 3–6 months followed by a taper to 400 μ g, if possible, and continued for 18 months. Outcome was assessed at 3–6 monthly intervals by chest radiography, lung function tests and serum angiotensin converting enzyme levels. Fifteen of 16 patients improved symptomatically; 15 of 20 chest radiographs had improved; and 16 patients had increased FVC (but not to statistical significance). Although these results are encouraging and side-effects were minimal (two individuals with hoarse voice), the interpretation of these findings is limited by the lack of a control group and the recruitment strategy, which involved a "run-in" period of only 6 months. Even though "progressive" disease was noted during this period, it is also possible that spontaneous resolution could have followed.

Another study compared 10 patients with symptomatic pulmonary sarcoidosis with a placebo group of 10

normal individuals and five patients with sarcoidosis. In this series, all 10 patients on active treatment improved symptomatically after budesonide 1,600 $\mu\text{g}\cdot\text{day}^{-1}$, but the chest radiographic pattern improved in only 3 out of 10, and no lung function improvement was observed over the 16 week study period [71]. However, BAL lymphocyte numbers fell in the treated group and the percentage of macrophages with the phenotype associated with immune response suppression (RFD1/D7) was also reduced in the treated group only [72, 73]. The observation on lymphocyte numbers is matched by a reduction of CD4:CD8 T-cell ratio of bronchoalveolar lymphocytes after inhaler treatment reported in another study [74].

Conclusion

Corticosteroids are effective in the management of sarcoidosis, by downregulating genes known to play a key role in pathogenesis and by improving indices of disease. Their effect on disease outcome needs further study. The ideal requirements of such a study are: 1) a homogeneous population of patients; 2) disease present for at least one year (more likely to be persistent); 3) predefined outcome measures; 4) individualized treatment to maximize and maintain response; 5) comparison with a nontreatment or placebo-treated group; and 6) it will almost certainly need to be multicentre (internationally) and will, therefore, need tightly applied recruitment criteria.

Further longer term studies of inhaled therapy should be undertaken, and should also be double-blind, placebo-controlled and should fulfil the above recommendations.

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