Immunomodulators in SLE: Clinical evidence and immunologic actions

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Abstract

Systemic lupus erythematosus (SLE) is a potentially fatal autoimmune disease. Current treatment strategies rely heavily on corticosteroids, which are in turn responsible for a significant burden of morbidity, and immunosuppressives which are limited by suboptimal efficacy, increased infections and malignancies. There are significant deficiencies in our immunosuppressive armamentarium, making immunomodulatory therapies crucial, offering the opportunity to prevent disease flare and the subsequent accrual of damage. Currently available immunomodulators include prasterone (synthetic dehydroeipandrosterone), vitamin D, hydroxychloroquine and belimumab. These therapies, acting via numerous cellular and cytokine pathways, have been shown to modify the aberrant immune responses associated with SLE without overt immunosuppression.

Vitamin D is important in SLE and supplementation appears to have a positive impact on disease activity particularly proteinuria. Belimumab has specific immunomodulatory properties and is an effective therapy in those with specific serological and clinical characteristics predictive of response. Hydroxychloroquine is a crucial background medication in SLE with actions in many molecular pathways. It has disease specific effects in reducing flare, treating cutaneous disease and inflammatory arthralgias in addition to other effects such as reduced thrombosis, increased longevity, improved lipids, better glycemic control and blood pressure. Dehydroeipandrosterone is also an immunomodulator in SLE which can have positive effects on disease activity and has bone protective properties.

This review outlines the immunologic actions of these drugs and the clinical evidence supporting their use.

Keywords

SLE; Immunomodulation; Hydroxychloroquine; Vitamin D; Dehydroeipandrosterone; Belimumab

1. Introduction

Systemic lupus erythematosus (SLE) is a chronic, multisystem, autoimmune condition characterized by the presence of autoantibodies to nuclear material and immune complex

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deposition in involved tissues. Whilst numerous advances have been made in unraveling the pathogenesis of this complex disease, it remains incompletely understood. A multitude of cell types and molecules, participating in many cellular mechanisms have been implicated in SLE. Abberancies in apoptotic pathways and in innate and adaptive immune mechanisms are found in patients with SLE, with genetic, epigenetic, environmental and hormonal factors known to contribute to the disease. There are a number of central events in the development of SLE, these include increased production of autoantibodies during apoptosis, decreased clearance of cellular debris with dysregulated handling and presentation. Subsequent disease activity and tissue damage is mediated by autoantibodies, immune complexes and complement activation with numerous cytokine and interferon pathways implicated. The complexity of these disease mechanisms have meant that there are a multitude of possible targets for immunomodulation in SLE. However, at present, there are few tools in our therapeutic armamentarium which can be considered immunomodulatory. For the most part, we rely on immunosuppressives, in particular for organ specific disease.

Improvements have been made in pharmacotherapy over the past 50 years which have positively impacted upon the prognosis of SLE although, disappointingly, poor renal outcomes [1,2], cardiovascular disease and the accumulation of organ damage often incited by high dose prednisone remain major challenges. Therapeutic advances include anti-malarials, corticosteroids, immunosuppressives, ace inhibitors, antibiotics, B-cell therapies, vitamin D supplementation and dehydroeipandrosterone (DHEA). Despite these therapies SLE continues to associate with premature mortality and morbidity. Current strategies rely heavily on the immunosuppressive properties of corticosteroids to control inflammation. Chronic and high dose corticosteroids associate with significant morbidity and are responsible for much of the long-term damage accrual in SLE. Other immunosuppressives, such as mycophenolate mofetil, methotrexate and azathioprine, are essential in the management of organ specific disease, however they are limited by efficacy, in particular in renal disease.

Immunomodulating therapies that are not immunosuppressive, are a more attractive therapeutic option, offering the opportunity to modify the aberrant immune responses in SLE and thus prevent inflammation and subsequent damage without the risks of infection and malignancy. Current strategies, considered to have immunomodulating properties, include hydroxychloroquine (and other antimalarials), vitamin D, dehydroeipandrosterone and certain B cell therapies. Stem cell transplantation is as of yet un-proven in randomized controlled studies for SLE but offers a fascinating perspective on immunomodulation and may, in the future, be a therapeutic option for those with severe, life threatening disease. Here we review current immunomodulating strategies in SLE, their clinical efficacy and examine their mechanisms of action.

2. Dehydroeipandrosterone

Dehydroeipandrosterone is a weak androgenic steroid and with its metabolite, dehydroepiandrosterone sulphate (DHEAS), is the most abundant adrenal steroid hormone. Dehydroeipandrosterone is a precursor of both androgens and estrogens and is synthesized primarily by the adrenal cortex (zona reticularis) from 17 α-hydroxypregnenolone. It can

then be sulphated, at the 3β'-hydroxyl group, into dehydroepiandrosterone sulphate in the adrenals and in peripheral tissues, dehydroeipandrosterone is also metabolized further into more active steroids including androstenedione, testosterone and estrogen [3]. In its drug form it is called prasterone.

Normal serum levels of dehydroeipandrosterone range from 1 to 50 nM. During fetal development, plasma dehydroepiandrosterone sulphate levels are 100– 200 μg/dL (3–7 μM), falling rapidly after birth and remaining low until adrenarche. Levels then increase rapidly, followed by an age related decline [4]. This decline is possibly mediated by decrease in 17,20-lyase activity [5]. The rate of decline of blood levels is in the region of 2% per year, by the 8th-9th decade residual levels are 10–20% of their peak [6]. There are gender differences to consider with higher levels in males [7]. In addition to these considerations there are genetic variations. Genome-wide association studies (GWAS) indicate that serum levels of dehydroeipandrosterone sulphate are regulated at approximately 60% by genotypes near these genes: BCL2L11, ZKSCAN5, ARPC1A, TRIM4, HHEX, CYP2C9, BMF, and SULT2A1[8].

Dehydroeipandrosterone does not have a specific receptor. It can bind to steroid hormone receptors (reviewed by Triash et al. [9], and by Webb et al. [5]) pregnane X receptor/steroid and xenobiotic receptor (PXR/SXR, NR1I2) [5]; estrogen receptors α and β, androgen receptors [10]; peroxisome proliferator activated receptors [5]; and pregnane X receptor [11]. At most of these sites, dehydroeipandrosterone acts as a partial agonist with weak affinity due to competition for binding. Taking into account the fact that dehydroeipandrosterone is itself a precursor for many of the higher affinity molecules, it is difficult to estimate the degree to which dehydroeipandrosterone itself is effective.

The principal regulator of dehydroeipandrosterone production, is adrenocorticotropic hormone. This in turn depends on corticotropin releasing hormone of hypothalamic origin for regulation [3]. In adults, dehydroeipandrosterone levels peak in the morning, following the circadian pattern of ACTH secretion [12]. The biological effects of dehydroeipandrosterone can be considered both androgenic and estrogenic since it is a precursor of both. Labrie et al. suggest that more than 30% of total androgen in men and over 90% of estrogen in postmenopausal women are derived from peripheral conversion of dehydroeipandrosterone [13]. Elevated dehydroeipandrosterone contributes to disorders associated with hyperandrogenic states such as in polycystic ovarian disease and non-classical 21-hydroxylase deficient congenital adrenal hyper-plasia [14]. Low levels have been associated with many age related disorders and with multiple autoimmune conditions, including SLE.

Women with SLE have been shown in numerous studies, reviewed by McMurray et al., to have significantly depressed concentrations of androgens and elevated levels of estradiol compared with both males with SLE and healthy controls [15]. In female patients with SLE, levels of both dehydroeipandrosterone and dehydroepiandrosterone sulphate are low [15–17]. Lahita et al. demonstrated low levels of all androgens in females with SLE with the lowest amount of both metabolites in those with active disease [16]. The fact that SLE is commonly treated with corticosteroids has been considered to be a confounding factor due

to inhibitory feedback mechanisms. However, steroid naïve SLE patients have also been shown to have low levels of dehydroeipandrosterone [16].

Dehydroeipandrosterone exerts anti-proliferative and anti-inflammatory effects, and modulates immune function. Prasterone (synthetic dehydroeipandrosterone) therapy has been shown in small studies to be beneficial in depression [18] with promise in the management of the negative symptoms of schizophrenia [19,20]. There is little evidence to lend support to the theory that it may have anti-aging effects. As it is known to have some androgenic properties, supplementation has been associated with mild virilization, acne, voice changes and terminal hair growth.

There is evidence that dehydroeipandrosterone has activity on multiple cytokine and immunologic pathways. Numerous studies have reported improvements in immune function with dehydroeipandrosterone supplementation including regulation of the production of pro-inflammatory cytokines such as IL-2, IL-1, IL-6 and TNFα [4,21,22]. There is also some evidence that dehydroeipandrosterone can modulate the proinflammatory cytokine profile associated with SLE, in particular IL-10 levels have been shown to decrease with supplementation [22].

In a murine SLE model (NZBA ~ NZW), decreased severity of lupus-like disease was demonstrated with the administration of dehydroeipandrosterone with data pointing toward decreased antibody production [23]. However, the improved survival in SLE mouse models was demonstrated only in those who were supplemented at 2 months and not in those who received synthetic dehydroeipandrosterone at 6 months [24], leading investigators to question whether there is a crucial point in SLE pathogenesis at which hormonal imbalances trigger a loss of self-tolerance. It is yet to be established in humans whether dehydroeipandrosterone levels are reduced before clinical disease onset.

Dehydroeipandrosterone levels have been found to correlate negatively with IL-6 [25] which is known to play an important role in immune regulation and inflammation in both healthy individuals and in SLE, amongst other autoimmune diseases. IL-6 is a pleiotropic cytokine strongly implicated in particular in lupus nephritis [26,27] and arthritis [28]. It shares several activities with IL-1 and tumor necrosis factor alpha, which have also been implicated in SLE, in the induction of pyrexia and the production of acute phase proteins. Whether IL-6 changes with supplementary DHEA in SLE has not been evaluated.

IL-10 is also implicated in the pathogenesis of SLE and serum from patients with SLE has been shown to stimulate IL-10 production from peripheral blood mononuclear cells [29]. Chang et al. evaluated changes in cytokine profiles in females with active SLE participating in a randomized controlled trial of 200 mg prasterone compared with placebo [22]. The levels of cytokines including interleukin IL-1, IL-2, IL-4, and Il-10 were determined. A significant reduction in IL-10 was demonstrated in those taking dehydroeipandrosterone supplementation. The other cytokines were either undetectable, or in the case of IL-1, there was no difference demonstrated.

These observations led a number of investigators to evaluate the therapeutic utility of supplemental dehydroeipandrosterone in women with SLE. The details of these clinical trials are outlined in Table 1.

Prasterone therapy was first formally evaluated and reported upon in SLE by van Vollenhoven [30] in an uncontrolled, open-label, single center study involving 10 patients in reciept of 200 mg per day. No significant difference in SLE disease activity index (SLEDAI) was demonstrated. However, there was a significant improvement in the physician global assessment of disease activity (PGA). This study was followed by a double-blind placebo controlled study involving 21 patients with 'severe' disease [31]. The primary end-point was clinical response, which was defined prospectively. Patients were considered to be responders if they demonstrated stabilization of their major clinical manifestation at six months (as defined in the protocol).

In a subsequent multi-center, double blind randomized placebocontrolled trial, 191 patients were randomized to receive praster-one, either 100 mg, 200 mg or placebo [32]. This trial was undertaken to evaluate the possibility that dehydroeipandrosterone supplementation could have steroid-sparing properties. The primary end point was reduction in prednisone (or corticosteroid equivalent) dose. There was no difference demonstrated between the three groups. However, differences were demonstrated when patients with quiescent disease were excluded. In the group with disease activity (defined as SLEDAI score >2), comprised of 137 subjects; 45 in the placebo group, 47 in the 100 mg group, and 45 in the 200 mg group, 29%, 38%, and 51%, respectively, were responders (P = 0.031 for 200 mg versus placebo).

Two studies by Chang et al. evaluated dehydroeipandrosterone supplementation, firstly reporting on a multicenter international study evaluating 120 participants with mild to moderate SLE [22,33]. This work demonstrated no significant reduction in disease activity measured by SLE activity measure (SLAM), but improvements were seen in flare, with a 16% reduction, and in the patient global score, which decreased significantly (in keeping with an improved self-reported assessment of disease activity). This was followed by the analysis of cytokines in a subgroup from a single center of Chinese patients. They observed no change in IL-1 and reduced IL-10 with many cytokines proving to be undetectable in this work.

A subsequent large, multicenter, randomized, double-blind, placebo-controlled trial evaluating prasterone supplementation demonstrated no difference in SLEDAI or SLAM in an intention to treat analysis, but when those with disease activity, defined as SLAM >7, SLEDAI >2, were considered there were more responders in those who received dehydroeipandrosterone supplementation compared with placebo (58.5% versus 44.5% (P = 0.017)). There was less flare in the dehydroeipandrosterone group (with SLEDAI >2) and less worsening of patient global (10.9% versus 22.6%, P = 0.007).

Consideration has also been given to whether dehydroeipandrosterone supplementation could influence bone metabolism. Hartkamp and colleagues addressed this question and found no change in overall bone mineral density in women, both pre and post menopausal, with quiescent disease taking less than 10 mg prednisone per day [34]. However, in those

who were postmenopausal there was a mean increase in bone density of 1.80% with prasterone therapy compared with a decrease of 2.32% in the placebo group. This suggested a protective effect on bone mineral density in postmenopausal women with SLE. Mease et al. also evaluated the effect of prasterone on bone mineral density [35]. Significant differences between treatment groups (200 mg prasterone and placebo) for percentage change in bone mineral density for both the lumbar spine and total hip were present. At the lumbar spine, there was a mean gain in bone mineral density of 1.7% in the prasterone group compared to a mean loss of 1.1% in the placebo group (p = 0.003 between groups). At the hip, the mean gain was 2.0% with prasterone compared to a mean loss of 0.3% in the placebo group (p = 0.013). Among those who were postmenopausal, the mean bone density of the lumbar spine increased by 3.1% in the prasterone group compared to a decrease of 1.7% in the placebo group (p = 0.012 between groups). Sánchez-Guerrero et al. [36] also evalauted the effect of supplementary dehydroepiandrosterone on bone mineral density. This was a randomized controlled trial which had three arms; 200 mg prasterone, 100 mg and placebo. There was dose-dependent increase in bone mineral density at the lumbar spine (at 18 months) in patients who received 200 versus 100 mg prasterone (p = 0.021). For patients who received 200 mg, the gain at the lumbar spine was $1.083 \pm 0.512\%$ (p = 0.042). There was no change in bone mineral density at the hip over 18 months with prasterone treatment.

Dehydroeipandrosterone has been shown to improve fatigue in other chronic diseases. Nordmark et al. evaluated the effect of supplementary dehydroeipandrosterone on fatigue and depression in SLE. There was no difference demonstrated between the dehydroeipandrosterone group and placebo, although interestingly, when those who believed they were taking prasterone were considered there was a significant difference compared to those who thought they were in the placebo arm [37].

There are conclusive data that low blood levels of dehydroeipandrosterone associate with disease activity in SLE in numerous populations and various age groups. Clinical trials have, for the most part, demonstrated improvements in disease activity in those with disease activity, although these studies are difficult to pool due to differences in dosing regimens and time-frames. Safety data from these studies were reassuring. Supplementation has not been evaluated in male patients, in whom deficiency is uncommon relative to females with SLE. There is no evidence to suggest that males would benefit from prasterone therapy. Further, in post menopausal women there is concern that the administration of an exogenous source of estrogen could increase the risk of hormone sensitive malignancies such as uterine and breast cancer.

Evidence in human SLE for the immunomodulating properties of dehydroepiandrosterone are at present limited to improvements in some cytokine profiles. It is however likely based on animal data that there are other mechanisms impacted upon by prasterone therapy. There is promising data that disease activity is improved by supplementing dehydroeipandrosterone, this effect is demonstrated in particular in patient reported measures, which are crucially important in this chronic disease. Evidence also exists that dehydroeipandrosterone can protect against bone loss. As fragility fractures are the most commonly reported item on the American College of Rheumatology/Systemic Lupus International Collaborating Clinics damage index [38], bone health is of crucial importance

in this population. The time frame for the bone density studies is relatively short. It is likely that longer durations of therapy and follow-up are necessary to determine if these changes are meaningful and to establish whether they translate into a reduction in fractures. At present, there are insufficient data to determine whether dehydroeipandrosterone has any influence on fatigue in SLE.

3. Vitamin D

Vitamin D, 1,25-dihydroxyvitamin D, is a steroid hormone, principally known for its roles in bone health and calcium homeostasis, now also recognized for its immunomodulatory properties. The chemical structure of vitamin D and its role in the metabolic bone disease, rickets, were first described in the 1930's [39]. Rickets, in children, and its adult equivalent, osteomalacia, are caused by very low levels of vitamin D. This is, for the most part, due to inadequate UV exposure. Malabsorption syndromes, renal failure and poor intake can also play a role. As vitamin D is a fatsoluble vitamin there is evidence that certain GI surgeries, including gastric bypass, decrease absorption [40].

In humans, vitamin D is mainly synthesized in the skin following ultraviolet B (UVB) exposure (wavelength, 290–315 nm) with a minority coming from dietary sources (<10%) [41]. It has two major forms, firstly ergocalciferol (commonly known as vitamin D2), acquired from ultraviolet (UV) irradiation and secondly, cholecalciferol (known as vitamin D3), which is made in the skin and acquired in food sources [41]. Both D2 and D3 forms can be used for food fortification and supplementation. Vitamin D is biologically inert and requires hydroxylation, by D-25- hydroxylase (25-OHase) to 25-hydroxyvitamin D (25(OH)D). This represents the major circulating vitamin D metabolite and is the most reliable parameter in establishing levels [42]. 25(OH)D then requires a further hydroxylation step, by 25(OH)D-1-OHase, to form its biologically active form which is 1,25-dihydroxyvitamin D (1,25(OH) 2D). This process is controlled by parathyroid hormone and the phosphaturic hormone fibroblast growth factor [43].

The highest concentration of 25(OH)D, in humans, is in plasma (usually measured in serum), but the largest pool of 25(OH)D is in adipose tissue and muscle. In the general population, for skeletal health, vitamin D deficiency has been defined by the Institute of Medicine as <20 ng/mL and insufficiency as 21–29 ng/mL [44]. Although there is no consensus in SLE specifically on the optimum vitamin D level, for adult patients at risk of fractures, falls, autoimmune disease or cardiovascular disease, a 25(OH)D level of at least 30–40 ng/ml has been recommended [45]. Given the risks of skin cancer associated with UV exposure, (and in SLE, the risk of disease flare) supplementation is via the oral route. Serum levels higher than 150 ng/mL have been associated with vitamin D intoxication characterized by hypercalcemia, hypercalciuria, and calcifications [45].

Vitamin D insufficiency and deficiency have been implicated in certain malignancies, cardiovascular disease and many autoimmune conditions, including SLE, rheumatoid arthritis and multiple sclerosis [41,46–49]. In general populations, vitamin D insufficiency is common and increases in prevalence with distance from the equator, although cultural practices in which clothing completely shields the skin makes deficiency commonplace,

regardless of latitude [41]. The prevalence of low levels of vitamin D in SLE has been reported, in both adults and children in multiple ethnic populations at different times of the year, at between 36.8% and 75% [49–55]. The reasons for these low levels have not been fully elucidated. Decreased UV exposure in those with SLE compared with controls and the influence of medications, prednisone and antimalarials, have been implicated without conclusion. The presence of photosensitivity, which may imply enhanced sun avoidance behaviors, has been found to associate with critically low levels [52]. Renal disease is also a significant contributing factor. The presence of renal disease was found, by Kamen et al. to be the strongest predictor of deficiency with an odds ratio of 13.3 [52]. There are also general differences in metabolism in individuals with darker skin due to decreased cutaneous conversion of vitamin D to its more active form following UVB exposure [56]. As SLE is more common and more severe in non-caucasians this has also been thought to influence levels. It does not appear that dietary intake is different in SLE compared to control populations [57].

In its active form, vitamin D has numerous immunologic functions mediated through binding to specific nuclear vitamin D receptors. These are present in most cells of the innate and adaptive immune system. Vitamin D receptors are expressed in monocytes, activated macrophages, dendritic cells, natural killer cells, T and B cells. Activation of these receptors has potent anti-proliferative, pro-differentiative, and immunomodulatory functions which can both suppress and enhance the immune response.

In vitro, vitamin D blocks B cell proliferation and differentiation, and suppresses immunoglobulin production [58,59]. It can attenuate the expression of pro inflammatory cytokines, induced by stimulation of toll-like receptor 3,4, 7 and 8 [60].

It can decrease T cell proliferation and shift maturing T cells away from Th1 toward Th2 and regulatory T cell phenotypes. Vitamin D has been shown to suppress dendritic cell differentiation. This is of particular importance in autoimmune conditions, including SLE due to the central role played by dendritic cells in the maintenance of self-tolerance [61]. Aberrant interferon production is also implicated in SLE disease activity and pathogenesis. Low levels of vitamin D have been associated with an increased interferon gene signature in patients with SLE (via the inhibition of dendritic cell maturation) [62]. Contrarily, vitamin D supplementation did not diminish the interferon signature in a placebocontrolled study [63]. In further support of vitamin D as an immunoregulator are the findings that vitamin D supplementation associates with down regulation of the Th1 immune response and the proliferation of activated B cells with up regulation of regulatory T cells.

A suggestion for the immunomodulatory effects of vitamin D in human SLE arises from a large body of data demonstrating an inverse relationship between serum levels and disease activity. Observational studies have, in the majority of cases, demonstrated an inverse relationship between serum vitamin D levels and SLE disease activity, as measured by SLEDAI [64–69], SLAM[70] and British Isles Lupus Assessment Group (BILAG) [71] (in diverse populations at varying latitudes). The interventional studies, which have evaluated the effect of supplementation, are outlined in Table 2.

Most data indicate a modest beneficial effect on disease activity with vitamin D supplementation (see Table 3). This effect is difficult to quantify as studies in SLE utilize many different supplementation protocols, over various time frames. Supplementation regimens varied from 600 iu to 7500 iu daily (50,000 iu weekly in addition to daily 400 iu). Ruiz- Irastorza et al. found that 600–800 iu over 24 months had no effect on disease activity measured by SLEDAI [72]. Higher doses were utilized in a subsequent study by Terrier and colleagues [73]. The SLEDAI decreased (2.9 ± 2.5 to 2.6 ± 2.5) which was not statistically significant. In the Hopkins Lupus Cohort, there was 0.2 improvement in SLEDAI per each 20 ng/ml increase in vitamin D [74]. The dose of vitamin D was 50,000 iu per week with 400 iu per day. Although statistically significant, the change in disease activity is small and of unclear clinical relevance. The physician global assessment of disease activity also improved with a modest improvement (4%) in proteinuria as measured by the urine protein to creatinine ratio. In an Egyptian population receiving 2000 iu daily, serum levels correlated with disease activity [75] and in juvenile SLE, 50,000 iu per week associated with a decrease in disease activity [76]. As there were no serious adverse events reported in these studies and the safety of vitamin D has been widely reported upon, we consider vitamin D an essential, safe therapy which has, at least, a modest beneficial effect on disease activity.

The mechanisms whereby vitamin D exerts these effects are incompletely understood. An increase in regulatory T cells with supplementation was found by Andreoli et al. [77] and by Terrier et al. [73]. There was no change in interferon signature demonstrated with supplemental vitamin D [63]. It was theorized vitamin D would impact upon was fatigue. Fatigue, measured by visual analogue scale was found to improve with vitamin D supplementation at 400–600 iu daily, but in adolescents, fatigue did not improve with therapy (measured by the kids fatigue severity scale).

Vitamin D deficiency has been associated with increased cardiovascular disease in the general population [78–80]. Patients with SLE are also known to have enhanced cardiovascular risk, which contributes to mortality. The relationship, in SLE, between vitamin D and cardiovascular risk factors has been evaluated, including in a large international inception cohort [69]. It was found that those with replete vitamin D levels were less likely to have hypertension and dyslipidemia. Adverse lipid profiles have also been demonstrated with low vitamin D levels [68]. Both Wu et al. (2009) and Reynolds et al. (2012) have also demonstrated a significant association with insulin resistance and low vitamin D. It is unclear whether there is data to link low vitamin D levels and the development of carotid plaque as the results of Reynolds et al. [67] and Ravenell et al. [81] conflict. It has also been recently reported that vitamin D supplementation improves endothelial repair mechanisms, dysregulation of which may contribute to the enhanced cardiovascular risk associated with SLE [82].

Vitamin D has long been known for its importance in bone health. There is now also ample evidence that vitamin D is important in SLE. The data indicate a modest effect on disease activity with perhaps a greater impact on renal parameters. This is of particular importance as poor renal outcomes are a major challenge with current immunosuppressive therapies. The mechanism whereby vitamin D exerts this effect is incompletely understood, but it may be the result of an increase in T regulatory cells. Vitamin D is a safe therapy in SLE and

therapy should be considered essential in those with deficiency and insufficiency. Whether vitamin D supplementation can impact upon cardiovascular outcomes will require further study.

4. Hydroxychloroquine

Hydroxychloroquine is an essential tool in the medical management of SLE with numerous disease-specific and longitudinal benefits [83–89]. It appears to work through numerous mechanisms in SLE, mediating subtle immunomodulation without causing immunosuppression. Hydroxychloroquine has been shown in multiple, diverse, SLE populations to associate with improved survival [84,90,91] and specifically has been shown to be effective in the treatment of cutaneous disease [92], arthritis [87], with an augmenting effect on the efficacy of mycophenolate mofetil in the management of nephritis [89]. Further, hydroxychloroquine has been shown to decrease thrombosis in those with positive anti phospholipid antibodies [93–95] and to improve pregnancy outcomes for women with SLE, with and without antiphospholipid antibodies [86,96,97]. In addition to disease specific benefits in SLE, hydroxychloroquine has been shown to have lipid lowering properties [98,99], anti-thrombotic effects [100] and hypoglycemic actions [101]. It can decrease progression to SLE in undifferentiated connective tissue disease [102] and in women with the Ro (SSa) antibody it decreases the risk of congenital heart block [103,104]. Hydroxychloroquine is an antimalarial medication. This class includes chloroquine and quinacrine, which can also be used in the treatment of SLE. As hydroxychloroquine is the cornerstone of the medical management of SLE with a multitude of known benefits, we will focus on this drug.

The mechanism of action of antimalarial drugs in SLE includes many molecular pathways [105–109]. Hydroxychloroquine is a weak base is thought to work, in part by increasing lysosomal pH in antigen presenting cells. This interferes with phagocytosis and causes disruption in the presentation of self-antigens [110,111]. T cell responses have been shown to be altered by these medications and numerous cytokines are inhibited (IL-1, Il-2, Il-6, IL-17, IL-22, interferon alpha and tumor necrosis factor alpha) [105–109]. The beneficial effects of hydroxychloroquine, in particular, may be via the inhibition of toll-like receptor activation. The endosomal acidification resulting from hydroxychloroquine therapy results in decreased signaling of toll-like receptors 3,7,8 and 9 [112]. In turn the reduced toll-like receptor signaling results in decreased activation of dendritic cell and the reduction of interferon production [113], amongst other mechanisms [100].

Hydroxychloroquine is effective in the treatment of SLE. Much of the data associating hydroxychloroquine with improved outcomes arises from observational studies. The clinical trials of hydroxychloroquine in SLE are outlined in Table 2. In a randomized, double blind drug withdrawal study, patients were either continued on therapy or switched to a placebo. The risk of flare in those switched to placebo increased by 2.5 with withdrawal of hydroxychloroquine [114]. However, contrarily, it did not reduce flares in a large trial evaluating belimumab [115] and data are conflicting regarding the attainment of therapeutic hydroxychloroquine blood levels and disease control. Costedoat-Chalumeau et al. found lower hydroxychloroquine level in those with active disease and that lower baseline levels

were predictive of flare [116]. In those with cutaneous lupus, hydroxychloroquine levels were significantly higher in patients with complete remission [117]. However, no relationship between blood levels and SLE flare was found in a subsequent clinical trial [118]. In the Hopkins Lupus Cohort, there was a statistically significant trend towards higher disease activity in those who had low hydroxychloroquine blood levels. However, within individual analysis over time did not demonstrate an improvement in disease activity once therapeutic levels were attained [119].

In terms of organ specific effects, increased rates of renal remission have been demonstrated in patients treated with hydroxychloroquine (in addition to immunosuppressives). In fact, nephritis patients treated with hydroxychloroquine with mycophenolate mofetil, had a remission rate which was 5 times higher than those who were treated with mycophenolate alone [89]. Hydroxychloroquine is considered central to the management of cutaneous disease [92] and it is also helpful for inflammatory arthralgias [120]. Numerous other benefits include decreased thrombosis [93], increased survival [84], improved lipid profiles [85,98,99] and lower blood glucose [101].

Pregnancy outcomes are improved in those with SLE taking hydroxychloroquine [86,96] and in particular support of its effect as an immunomodulator, there is some data that it decreases the risk of congenital heart block in children of Ro positive mothers [103,104]. Autoantibody-associated congenital heart block is a serious condition which arises from passively acquired antibodies that target the fetal cardiac conduction system. With a Ro antibody, the risk of having a pregnancy complicated by congenital heart block is in the region of 1–2% and the risk of recurrence in a subsequent pregnancy increases to around 18% [121]. Izmirly et al. demonstrated less congenital heart block in babies exposed to hydroxychloroquine compared with controls [103]. In those with a previous history of having a child with cardiac neonatal lupus, the risk of recurrence has also been shown to be decreased with hydroxychloroquine therapy [104]. The recurrence rate in fetuses exposed to HCQ was 7.5% versus 21.2% in the unexposed group. This effect is thought to be mediated via toll like receptors [122].

In further support of the role of hydroxychloroquine as an immunomodulator, there is evidence that therapy associates with delayed onset of SLE (in patients who have less than 4 American College of Rheumatology classification criteria). Those treated with hydroxychloroquine had a longer time between the first clinical symptom and the diagnosis of SLE and less autoantibodies [102].

Hydroxychloroquine is a well-tolerated medication and side effects are few. However, there are increasing concerns regarding hydroxychloroquine related retinopathy, in particular in light of new screening methods, which are thought to have increased sensitivity [123]. Current American Academy of Ophthalmology guidelines advise monitoring, beyond the dilated retinal examination and automated visual field testing, in an attempt to identify toxicity early [124]. The sensitivity and specificity of these tests are not yet known for hydroxychloroquine related retinal toxicity. Thus the true prevalence of retinal deposition may differ from what was previously reported. Other toxicities, cardiac and neuromyoapthic are rare. These are reviewed in detail by Costedoat-Chalumeau et al. [111].

Hydroxychloroquine is a crucial immunomodulatory medicine in SLE with innumerous disease specific and longitudinal benefits. Clinical data point toward a reduction in flare with therapy, amongst other effects, but at present the mechanism for many of the long-term benefits are incompletely understood. For the prevention of SLE, it is the only medication with any known effect. In women with Ro antibodies, it is the only known strategy to help prevent congenital heart block.

5. B cell therapies

B cells play an important pathologic role in SLE. Abnormal B cell proliferation, maturation, prolonged life-span of auto reactive clones, and autoantibody production are known to be present in SLE, and to associate with immune dysregulation and breakdown of self-tolerance [125]. B cells are involved in several specific pro-inflammatory mechanisms in SLE, including T cell antigen presentation, cytokine release and autoantibody formation. Self-antigens are presented on the cell surface to auto-reactive T cells, triggering B cells into autoantibody production and propagating their function as antigen presenting cells. These cells then release proinflammatory cytokines which are implicated in SLE including interferon α, IL-6 and IL-10, B-cell activating factor (BAFF), TNF α and a proliferation inducing ligand (APRIL) [125]. These pathways contribute to clinical manifestations by inciting inflammation, causing tissue damage and immune complex deposition. As a result of these abnormalities, B cell therapies have always been considered attractive immunosuppressive and immunomodulatory regimens. Although these agents do suppress immune function, they also can have a long lasting effect on B cell populations and disease mechanisms. Belimumab in particular, has only mildly immunosuppressive properties and functions for the most part as an immunomodulatory agent.

B-cell strategies which have been considered in SLE target CD-20 (ocrelizumab [126], rituximab [127]), CD-22 (epratuzumab [128]), BAFF (belimumab [129], blisibimod [130], tabalumab [131], briobacept, atacicept [132]) and a proliferation-inducing ligand (APRIL) (atacicept [132]). Despite robust preclinical and mechanistic data, these agents have been disappointing in clinical trials and have not, with the exception of belimumab, been approved for SLE. Some, such as rituximab, are considered immunosuppressive, as they lead to B-cell depletion.

Belimumab is unique in showing clinical efficacy in large randomized controlled studies and in its subsequent FDA approval [129,133]. It is a fully humanized monoclonal antibody that specifically binds to soluble BAFF, preventing its interaction with receptors, resulting in a reduction in the numbers of peripheral naive, transitional and activated B cells. Unlike rituximab, it does not deplete B-cell populations. Two large, phase III, multi-center, prospective, randomized, controlled trials have compared belimumab with placebo in SLE [129,133]. Navarra et al. evaluated 867 patients in Latin America, Asia-Pacific and Eastern Europe. All had active disease with a SLEDAI of greater than 6. They were randomized to receive belimumab 1 mg/kg, 10 mg/kg, or placebo. The outcome measure was the Systemic lupus erythematosus responder index (SRI), a composite measure designed to evaluate overall and organ specific disease activity. It is composed of the SLEDAI, BILAG and PGA. Significantly higher rates of response were noted with belimumab 1 mg/kg and 10 mg/kg

than with placebo 125 at week 52. More patients had their SLEDAI score reduced by at least 4 points during 52 weeks with belimumab at both doses than with placebo. Better outcomes were also observed for PGA and BILAG with belimumab treatment. Furie et al. [129] demonstrated similar results and accordingly, belimumab received FDA approval.

Pooled data from these studies indicate that belimumab promotes normalization of serological abnormalities, with reversal of hypocomplementemia and decreased autoantibodies (anti double stranded DNA, anti Sm, anticardiolipin and anti ribosomal P). Both dosing regimens associated with significant reductions in the numbers of CD20 B cells and in multiple B cell and plasma cell subsets, including naive and activated B cells, as well as in CD20 +CD138 + plasmablasts whilst preserving the memory B cell subset and T cell populations, supporting its status as a specific immunomodulatory therapy [134]. Post-hoc analyses demonstrated that this medication is of most utility in those with high disease activity despite standard therapy, low complement levels, antibodies to double stranded DNA and corticosteroid use [135]. It may also be of use in renal disease [136] although specific clinical trials are yet to be reported.

6. Stem cell transplantation

Autologous and allogeneic stem cell transplantation have been reported as having therapeutic benefit in SLE. In severe SLE, refractory to conventional therapy, stem cell transplantation can associate with sustained clinical remission, with rates ranging from 50 to 70%, associated with normalization of many immunologic changes [137,138]. This is a strategy that offers treatment-free remission. However, it has not been evaluated in any randomized controlled studies and associates with considerable short-term morbidity and in some cases mortality [137,139,140]. Transplant mortality has ranged from none to as high as 25% [137]. As such, in the absence of a controlled study, these therapies are unlikely to become available outside of the research setting.

Stem cell transplantation, both autologous and allogeneic, offers a fascinating and instructive insight into SLE disease mechanisms and can be considered the ultimate immunomodulating therapy. Following transplantation, autoantibodies consistently decreases or disappear with normalization of T cell responses [141] with the disappearance of plasmablasts and return of healthy levels of regulatory T cells [142]. There is a CD4+FoxP3+ regulatory T cell in transplanted patients that seems to inhibit abnormal T cell responses [143]. There is evidence that allogeneic mesenchymal stem cell transplant can reverse changes in the T cell-interferon axis which has also been though to contribute to SLE pathogenesis [144].

7. Conclusion

SLE is a potentially fatal autoimmune disease characterized by autoantibodies and immune dysregulation resulting in multiorgan injury. Current treatment strategies for the most part are immunosuppressive and are limited by efficacy and side effects, such as increased infections and long term toxicities. Immunomodulatory therapies offer the opportunity to prevent disease activity and decrease the accrual of damage.

Hydroxychloroquine is a crucial background medication in SLE and has actions on numerous cell types in many molecular pathways. It has disease specific effects in reducing flare, treating cutaneous disease and inflammatory arthralgias in addition to innumerous other effects such as reduced thrombosis, increased longevity, improved lipids, better glycemic control and blood pressure. Dehydroeipandrosterone is also an immunomodulator in SLE which can have positive effects on disease activity and has bone protective properties. Vitamin D is now known to be important in SLE and supplementation appears to have a positive impact on disease activity including proteinuria. Belimumab has specific immunomodulatory properties and is an effective therapy in those with the serological and clinical characteristics predictive of response. Stem cell transplantation is a fascinating therapy in SLE, but is not yet proven in controlled studies and associates with significant morbidity and in some cases, mortality.

Acknowledgments

References

1. Mok CC. Towards new avenues in the management of lupus glomerulonephritis. Nat. Rev. Rheumatol. 2016 Apr; 12(4):221–234. http://dx.doi.org/10.1038/nrrheum.2015. [PubMed: 26729459]

2. Ward MM. Changes in the incidence of end-stage renal disease due to lupus nephritis 1982–1995. Arch. Intern Med. 2000; 160(20):3136–3140. [PubMed: 11074743]

3. Parker LN. Control of adrenal androgen secretion. Endocrinol. Metab. Clin. North Am. 1991; 20(2): 401–421. [PubMed: 1652437]

4. Chen CC, Parker CR. Adrenal androgens and the immune system. Semin. Reprod. Med. 2004; 22(4):369–377. [PubMed: 15635504]

5. Webb SJ, Geoghegan TE, Prough RA, Michael Miller KK. The biological actions of dehydroepiandrosterone involves multiple receptors. Drug Metab. Rev. 2006; 38(1–2):89–116. [PubMed: 16684650]

6. Derksen RH. Dehydroepiandrosterone (DHEA) and systemic lupus erythematosus. Semin. Arthritis Rheum. 1998; 27(6):335–347. [PubMed: 9662752]

7. Rainey WE, Carr BR, Sasano H, Suzuki T, Mason JI. Dissecting human adrenal androgen production. Trends Endocrinol. Metab. 2002; 13(6):234–239. [PubMed: 12128283]

8. Vandenput L, Ohlsson C. Genome-wide association studies on serum sex steroid levels. Mol. Cell Endocrinol. 2014; 382(1):758–766. [PubMed: 23541950]

9. Traish AM, Kang HP, Saad F, Guay AT. Dehydroepiandrosterone (DHEA)–a precursor steroid or an active hormone in human physiology. J. Sex. Med. 2011; 8(11):2960–2982. quiz 83. [PubMed: 22032408]

10. Chen F, Knecht K, Birzin E, Fisher J, Wilkinson H, Mojena M, et al. Direct agonist/antagonist functions of dehydroepiandrosterone. Endocrinology. 2005; 146(11):4568–4576. [PubMed: 15994348]

11. Ripp SL, Fitzpatrick JL, Peters JM, Prough RA. Induction of CYP3A expression by dehydroepiandrosterone: involvement of the pregnane X receptor. Drug Metab. Dispos. 2002; 30(5):570–575. [PubMed: 11950789]

12. Hammer F, Drescher DG, Schneider SB, Quinkler M, Stewart PM, Allolio B, et al. Sex steroid metabolism in human peripheral blood mono-nuclear cells changes with aging. J. Clin. Endocrinol. Metab. 2005; 90(11):6283–6289. [PubMed: 16091484]

13. Labrie F, Belanger A, Simard J, Van Luu-The, Labrie C. DHEA and peripheral androgen and estrogen formation: intracrinology. Ann. N. Y. Acad. Sci. 1995; 774:16–28. [PubMed: 8597456]

14. Goodarzi MO, Carmina E, Azziz R. DHEA, DHEAS and PCOS. J. Steroid Biochem. Mol. Biol. 2015; 145:213–225. [PubMed: 25008465]

15. McMurray RW, May W. Sex hormones and systemic lupus erythematosus: review and meta-analysis. Arthritis Rheum. 2003; 48(8):2100–2110. [PubMed: 12905462]

16. Lahita RG, Bradlow HL, Ginzler E, Pang S, New M. Low plasma androgens in women with systemic lupus erythematosus. Arthritis Rheum. 1987; 30(3):241–248. [PubMed: 3032210]

17. Suzuki T, Suzuki N, Engleman EG, Mizushima Y, Sakane T. Low serum levels of dehydroepiandrosterone may cause deficient IL-2 production by lymphocytes in patients with systemic lupus erythematosus (SLE). Clin. Exp. Immunol. 1995; 99(2):251–255. [PubMed: 7851019]

18. Wolkowitz OM, Reus VI, Roberts E, Manfredi F, Chan T, Raum WJ, et al. Dehydroepiandrosterone (DHEA) treatment of depression. Biol. Psychiatry. 1997; 41(3):311–318. [PubMed: 9024954]

19. Ritsner MS, Strous RD. Neurocognitive deficits in schizophrenia are associated with alterations in blood levels of neurosteroids: a multiple regression analysis of findings from a double-blind, randomized, placebo-controlled, crossover trial with DHEA. J. Psychiatr. Res. 2010; 44(2):75–80. [PubMed: 19665142]

20. Strous RD, Stryjer R, Maayan R, Gal G, Viglin D, Katz E, et al. Analysis of clinical symptomatology, extrapyramidal symptoms and neurocognitive dysfunction following dehydroepiandrosterone (DHEA) administration in olanzapine treated schizophrenia patients: a randomized, double-blind placebo controlled trial. PsychoneuroEndocrinology. 2007; 32(2):96–105. [PubMed: 17208382]

21. Dillon JS. Dehydroepiandrosterone, dehydroepiandrosterone sulfate and related steroids: their role in inflammatory, allergic and immunological disorders. Curr. Drug Targets Inflamm. Allergy. 2005; 4(3):377–385. [PubMed: 16101547]

22. Chang DM, Chu SJ, Chen HC, Kuo SY, Lai JH. Dehydroepiandrosterone suppresses interleukin 10 synthesis in women with systemic lupus erythematosus. Ann. Rheum. Dis. 2004; 63(12):1623–1626. [PubMed: 15547086]

23. Lucas JA, Ahmed SA, Casey ML, MacDonald PC. Prevention of autoantibody formation and prolonged survival in New Zealand black/New Zealand white F1 mice fed dehydroisoandrosterone. J. Clin. Invest. 1985; 75(6):2091–2093. [PubMed: 3159756]

24. Norton SD, Harrison LL, Yowell R, Araneo BA. Administration of dehydroepiandrosterone sulfate retards onset but not progression of autoimmune disease in NZB/W mice. Autoimmunity. 1997; 26(3):161–171. [PubMed: 9550284]

25. Straub RH, Lehle K, Herfarth H, Weber M, Falk W, Preuner J, et al. Dehydroepiandrosterone in relation to other adrenal hormones during an acute inflammatory stressful disease state compared with chronic inflammatory disease: role of interleukin-6 and tumour necrosis factor. Eur. J. Endocrinol. 2002; 146(3):365–374. [PubMed: 11888843]

26. Peterson E, Robertson AD, Emlen W. Serum and urinary interleukin-6 in systemic lupus erythematosus. Lupus. 1996; 5(6):571–575. [PubMed: 9116699]

27. Iwano M, Dohi K, Hirata E, Kurumatani N, Horii Y, Shiiki H, et al. Urinary levels of IL-6 in patients with active lupus nephritis. Clin. Nephrol. 1993; 40(1):16–21. [PubMed: 8358870]

28. Ball EM, Gibson DS, Bell AL, Rooney MR. Plasma IL-6 levels correlate with clinical and ultrasound measures of arthritis in patients with systemic lupus erythematosus. Lupus. 2014; 23(1):46–56. [PubMed: 24243775]

29. Ronnelid J, Tejde A, Mathsson L, Nilsson-Ekdahl K, Nilsson B. Immune complexes from SLE sera induce IL10 production from normal peripheral blood mononuclear cells by an FcgammaRII dependent mechanism: implications for a possible vicious cycle maintaining B cell hyperactivity in SLE. Ann. Rheum. Dis. 2003; 62(1):37–42. [PubMed: 12480667]

30. van Vollenhoven RF, Engleman EG, McGuire JL. An open study of dehydroepiandrosterone in systemic lupus erythematosus. Arthritis Rheum. 1994; 37(9):1305–1310. [PubMed: 7945493]

31. van Vollenhoven RF, Park JL, Genovese MC, West JP, McGuire JL. A double-blind, placebo-controlled, clinical trial of dehydroepiandrosterone in severe systemic lupus erythematosus. Lupus. 1999; 8(3):181–187. [PubMed: 10342710]

32. Petri MA, Lahita RG, Van Vollenhoven RF, Merrill JT, Schiff M, Ginzler EM, et al. Effects of prasterone on corticosteroid requirements of women with systemic lupus erythematosus: a double-blind, randomized, placebo-controlled trial. Arthritis Rheum. 2002; 46(7):1820–1829. [PubMed: 12124866]

33. Chang DM, Lan JL, Lin HY, Luo SF. Dehydroepiandrosterone treatment of women with mild-to-moderate systemic lupus erythematosus: a multi-center randomized, double-blind, placebo-controlled trial. Arthritis Rheum. 2002; 46(11):2924–2927. [PubMed: 12428233]

34. Hartkamp A, Geenen R, Godaert GL, Bijl M, Bijlsma JW, Derksen RH. The effect of dehydroepiandrosterone on lumbar spine bone mineral density in patients with quiescent systemic lupus erythematosus. Arthritis Rheum. 2004; 50(11):3591–3595. [PubMed: 15529389]

35. Mease PJ, Ginzler EM, Gluck OS, Schiff M, Goldman A, Greenwald M, et al. Effects of prasterone on bone mineral density in women with systemic lupus erythematosus receiving chronic glucocorticoid therapy. J. Rheumatol. 2005; 32(4):616–621. [PubMed: 15801015]

36. Sanchez-Guerrero J, Fragoso-Loyo HE, Neuwelt CM, Wallace DJ, Ginzler EM, Sherrer YR, et al. Effects of prasterone on bone mineral density in women with active systemic lupus erythematosus receiving chronic glucocorticoid therapy. J. Rheumatol. 2008; 35(8):1567–1575. [PubMed: 18634158]

37. Nordmark G, Bengtsson C, Larsson A, Karlsson FA, Sturfelt G, Ronnblom L. Effects of dehydroepiandrosterone supplement on health-related quality of life in glucocorticoid treated female patients with systemic lupus erythe-matosus. Autoimmunity. 2005; 38(7):531–540. [PubMed: 16373258]

38. Gladman DD, Urowitz MB, Goldsmith CH, Fortin P, Ginzler E, Gordon C, et al. The reliability of the systemic lupus international collaborating clinics/american College of Rheumatology damage index in patients with systemic lupus erythematosus. Arthritis Rheum. 1997; 40(5):809–813. [PubMed: 9153540]

39. Wolf G. The discovery of vitamin D: the contribution of Adolf Windaus. J. Nutr. 2004; 134(6):1299–1302. [PubMed: 15173387]

40. Peterson LA, Zeng X, Caufield-Noll CP, Schweitzer MA, Magnuson TH, Steele KE. Vitamin D status and supplementation before and after bariatric surgery: a comprehensive literature review. Surg. Obes. Relat. Dis. 2016; 12(3):693–702. [PubMed: 27036669]

41. Holick MF. Vitamin D deficiency. N. Engl. J. Med. 2007; 357(3):266–281. [PubMed: 17634462]

42. DeLuca HF. Overview of general physiologic features and functions of vitamin D. Am. J. Clin. Nutr. 2004; 80(6 Suppl):1689S–1696S. [PubMed: 15585789]

43. Iruretagoyena M, Hirigoyen D, Naves R, Burgos PI. Immune response modulation by vitamin D: role in systemic lupus erythematosus. Front. Immunol. 2015; 6:513. [PubMed: 26528285]

44. Ross AC, Manson JE, Abrams SA, Aloia JF, Brannon PM, Clinton SK, et al. The 2011 report on dietary reference intakes for calcium and vitamin D from the Institute of Medicine: what clinicians need to know. J. Clin. Endocrinol. Metab. 2011; 96(1):53–58. [PubMed: 21118827]

45. Souberbielle JC, Body JJ, Lappe JM, Plebani M, Shoenfeld Y, Wang TJ, et al. Vitamin D and musculoskeletal health, cardiovascular disease, autoimmunity and cancer: recommendations for clinical practice. Autoimmun. Rev. 2010; 9(11):709–715. [PubMed: 20601202]

46. Holick MF. Vitamin D: important for prevention of osteoporosis, cardiovascular heart disease, type 1 diabetes, autoimmune diseases, and some cancers. South Med. J. 2005; 98(10):1024–1027. [PubMed: 16295817]

47. Ascherio A, Munger KL, Simon KC. Vitamin D and multiple sclerosis. Lancet Neurol. 2010; 9(6):599–612. [PubMed: 20494325]

48. Patel S, Farragher T, Berry J, Bunn D, Silman A, Symmons D. Association between serum vitamin D metabolite levels and disease activity in patients with early inflammatory polyarthritis. Arthritis Rheum. 2007; 56(7):2143–2149. [PubMed: 17599737]

49. Müller K, Kriegbaum NJ, Baslund B, Sørensen OH, Thymann M, Bentzen K. Vitamin D3 metabolism in patients with rheumatic diseases: low serum levels of 25-hydroxyvitamin D3 in

patients with systemic lupus erythematosus. Clin. Rheumatol. 1995; 14(4):397–400. [PubMed: 7586974]

50. Ritterhouse LL, Crowe SR, Niewold TB, Kamen DL, Macwana SR, Roberts VC, et al. Vitamin D deficiency is associated with an increased autoimmune response in healthy individuals and in patients with systemic lupus erythematosus. Ann. Rheum. Dis. 2011; 70(9):1569–1574. [PubMed: 21586442]

51. Wright TB, Shults J, Leonard MB, Zemel BS, Burnham JM. Hypovitaminosis D is associated with greater body mass index and disease activity in pediatric systemic lupus erythematosus. J. Pediatr. 2009; 155(2):260–265. [PubMed: 19446841]

52. Kamen DL, Cooper GS, Bouali H, Shaftman SR, Hollis BW, Gilkeson GS. Vitamin D deficiency in systemic lupus erythematosus. Autoimmun. Rev. 2006; 5(2):114–117. [PubMed: 16431339]

53. Huisman AM, White KP, Algra A, Harth M, Vieth R, Jacobs JW, et al. Vitamin D levels in women with systemic lupus erythematosus and fibromyalgia. J. Rheumatol. 2001; 28(11):2535–2539. [PubMed: 11708429]

54. Ruiz-Irastorza G, Egurbide MV, Pijoan JI, Garmendia M, Villar I, Martinez-Berriotxoa A, et al. Effect of antimalarials on thrombosis and survival in patients with systemic lupus erythematosus. Lupus. 2006; 15(9):577–583. [PubMed: 17080912]

55. Mok CC. Vitamin D and systemic lupus erythematosus: an update. Expert Rev. Clin. Immunol. 2013; 9(5):453–463. [PubMed: 23634739]

56. Powe CE, Evans MK, Wenger J, Zonderman AB, Berg AH, Nalls M, et al. Vitamin D-binding protein and vitamin D status of black Americans and white Americans. N. Engl. J. Med. 2013; 369(21):1991–2000. [PubMed: 24256378]

57. Hiraki LT, Munger KL, Costenbader KH, Karlson EW. Dietary intake of vitamin D during adolescence and risk of adult-onset systemic lupus erythematosus and rheumatoid arthritis. Arthritis Care Res. Hob. 2012; 64(12):1829–1836.

58. Chen S, Sims GP, Chen XX, Gu YY, Lipsky PE. Modulatory effects of 1,25-dihydroxyvitamin D3 on human B cell differentiation. J. Immunol. 2007; 179(3):1634–1647. [PubMed: 17641030]

59. Aranow C. Vitamin D and the immune system. J. Investig. Med. 2011; 59(6):881–886.

60. Ojaimi S, Skinner NA, Strauss BJ, Sundararajan V, Woolley I, Visvanathan K. Vitamin D deficiency impacts on expression of toll-like receptor-2 and cytokine profile: a pilot study. J. Transl. Med. 2013; 11:176. [PubMed: 23875738]

61. Monrad SU, Rea K, Thacker S, Kaplan MJ. Myeloid dendritic cells display downregulation of C-type lectin receptors and aberrant lectin uptake in systemic lupus erythematosus. Arthritis Res. Ther. 2008; 10(5):R114. [PubMed: 18811944]

62. Ben-Zvi I, Aranow C, Mackay M, Stanevsky A, Kamen DL, Marinescu LM, et al. The impact of vitamin D on dendritic cell function in patients with systemic lupus erythematosus. PLoS One. 2010; 5(2):e9193. [PubMed: 20169063]

63. Aranow C, Kamen DL, Dall'Era M, Massarotti EM, Mackay MC, Koumpouras F, et al. Randomized, double-blind, placebo-controlled trial of the effect of vitamin D3 on the interferon signature in patients with systemic lupus erythematosus. Arthritis Rheumatol. 2015; 67(7):1848–1857. [PubMed: 25777546]

64. Borba VZ, Vieira JG, Kasamatsu T, Radominski SC, Sato EI, Lazaretti-Castro M. Vitamin D deficiency in patients with active systemic lupus erythematosus. Osteoporos. Int. 2009; 20(3):427–433. [PubMed: 18600287]

65. Amital H, Szekanecz Z, Szücs G, Danko K, Nagy E, Csepany T, et al. Serum concentrations of 25-OH vitamin D in patients with systemic lupus erythematosus (SLE) are inversely related to disease activity: is it time to routinely supplement patients with SLE with vitamin D? Ann. Rheum. Dis. 2010; 69(6):1155–1157. [PubMed: 20439290]

66. Yeap SS, Othman AZ, Zain AA, Chan SP. Vitamin D levels: its relationship to bone mineral density response and disease activity in premenopausal Malaysian systemic lupus erythematosus patients on corticosteroids. Int. J. Rheum. Dis. 2012; 15(1):17–24. [PubMed: 22324943]

67. Reynolds JA, Haque S, Berry JL, Pemberton P, Teh LS, Ho P, et al. 25-Hydroxyvitamin D deficiency is associated with increased aortic stiffness in patients with systemic lupus erythematosus. Rheumatol. Oxf. 2012; 51(3):544–551.

68. Mok CC, Birmingham DJ, Leung HW, Hebert LA, Song H, Rovin BH. Vitamin D levels in Chinese patients with systemic lupus erythematosus: relationship with disease activity, vascular risk factors and atherosclerosis. Rheumatol. Oxf. 2012; 51(4):644–652.

69. Lertratanakul A, Wu P, Dyer A, Urowitz M, Gladman D, Fortin P, et al. 25-hydroxyvitamin D and cardiovascular disease in patients with systemic lupus erythematosus: data from a large international inception cohort. Arthritis Care Res. Hob. 2014; 66(8):1167–1176.

70. Becker A, Fischer R, Schneider M. Bone density and 25-OH vitamin D serum level in patients with systemic lupus erythematosus. Z Rheumatol. 2001; 60(5):352–358. [PubMed: 11759235]

71. Bonakdar ZS, Jahanshahifar L, Jahanshahifar F, Gholamrezaei A. Vitamin D deficiency and its association with disease activity in new cases of systemic lupus erythematosus. Lupus. 2011; 20(11):1155–1160. [PubMed: 21680639]

72. Ruiz-Irastorza G, Gordo S, Olivares N, Egurbide MV, Aguirre C. Changes in vitamin D levels in patients with systemic lupus erythematosus: effects on fatigue, disease activity, and damage. Arthritis Care Res. Hob. 2010; 62(8):1160–1165.

73. Terrier B, Derian N, Schoindre Y, Chaara W, Geri G, Zahr N, et al. Restoration of regulatory and effector T cell balance and B cell homeostasis in systemic lupus erythematosus patients through vitamin D supplementation. Arthritis Res. Ther. 2012; 14(5):R221. [PubMed: 23075451]

74. Petri M, Bello KJ, Fang H, Magder LS. Vitamin D in systemic lupus erythematosus: modest association with disease activity and the urine protein-to-creatinine ratio. Arthritis Rheum. 2013; 65(7):1865–1871. [PubMed: 23553077]

75. Abou-Raya A, Abou-Raya S, Helmii M. The effect of vitamin D supplementation on inflammatory and hemostatic markers and disease activity in patients with systemic lupus erythematosus: a randomized placebocontrolled trial. J. Rheumatol. 2013; 40(3):265–272. [PubMed: 23204220]

76. Lima GL, Paupitz J, Aikawa NE, Takayama L, Bonfa E, Pereira RM. Vitamin D supplementation in adolescents and young adults with juvenile systemic lupus erythematosus for improvement in disease activity and fatigue scores: a randomized, double-blind, placebo-controlled trial. Arthritis Care Res. Hob. 2016; 68(1):91–98.

77. Andreoli L, Dall'Ara F, Piantoni S, Zanola A, Piva N, Cutolo M, et al. A 24-month prospective study on the efficacy and safety of two different monthly regimens of vitamin D supplementation in pre-menopausal women with systemic lupus erythematosus. Lupus. 2015; 24(4–5):499–506. [PubMed: 25801893]

78. Kim DH, Sabour S, Sagar UN, Adams S, Whellan DJ. Prevalence of hypovitaminosis D in cardiovascular diseases (from the national health and nutrition examination survey 2001 to 2004). Am. J. Cardiol. 2008; 102(11):1540–1544. [PubMed: 19026311]

79. Poole KE, Loveridge N, Barker PJ, Halsall DJ, Rose C, Reeve J, et al. Reduced vitamin D in acute stroke. Stroke. 2006; 37(1):243–245. [PubMed: 16322500]

80. Kendrick J, Targher G, Smits G, Chonchol M. 25-Hydroxyvitamin D deficiency is independently associated with cardiovascular disease in the Third National Health and Nutrition Examination Survey. Atherosclerosis. 2009; 205(1):255–260. [PubMed: 19091317]

81. Ravenell RL, Kamen DL, Spence JD, Hollis BW, Fleury TJ, Janech MG, et al. Premature atherosclerosis is associated with hypovitaminosis D and angiotensin-converting enzyme inhibitor non-use in lupus patients. Am. J. Med. Sci. 2012; 344(4):268–273. [PubMed: 22222338]

82. Furie RA, Petri MA, Wallace DJ, Ginzler EM, Merrill JT, Stohl W, et al. Novel evidence-based systemic lupus erythematosus responder index. Arthritis Rheum. 2009; 61(9):1143–1151. [PubMed: 19714615]

83. A randomized study of the effect of withdrawing hydroxychloroquine sulfate in systemic lupus erythematosus. N. Engl. J. Med. 1991; 324(3):150–154. [PubMed: 1984192]

84. Alarcon GS, McGwin G, Bertoli AM, Fessler BJ, Calvo-Alen J, Bastian HM, et al. Effect of hydroxychloroquine on the survival of patients with systemic lupus erythematosus: data from LUMINA, a multiethnic US cohort (LUMINA L). Ann. Rheum. Dis. 2007; 66(9):1168–1172. [PubMed: 17389655]

85. Cairoli E, Rebella M, Danese N, Garra V, Borba EF. Hydroxychloroquine reduces low-density lipoprotein cholesterol levels in systemic lupus erythematosus: a longitudinal evaluation of the lipid-lowering effect. Lupus. 2012; 21(11):1178–1182. [PubMed: 22641182]

86. Clowse ME, Magder L, Witter F, Petri M. Hydroxychloroquine in lupus pregnancy. Arthritis Rheum. 2006; 54(11):3640–3647. [PubMed: 17075810]

87. Williams HJ, Egger MJ, Singer JZ, Willkens RF, Kalunian KC, Clegg DO, et al. Comparison of hydroxychloroquine and placebo in the treatment of the arthropathy of mild systemic lupus erythematosus. J. Rheumatol. 1994; 21(8):1457–1462. [PubMed: 7983646]

88. Espinola RG, Pierangeli SS, Gharavi AE, Harris EN, Ghara AE. Hydroxychloroquine reverses platelet activation induced by human IgG anti-phospholipid antibodies. Thromb. Haemost. 2002; 87(3):518–522. [PubMed: 11916085]

89. Kasitanon N, Fine DM, Haas M, Magder LS, Petri M. Hydroxychloroquine use predicts complete renal remission within 12 months among patients treated with mycophenolate mofetil therapy for membranous lupus nephritis. Lupus. 2006; 15(6):366–370. [PubMed: 16830883]

90. Fessler BJ, Alarcon GS, McGwin G, Roseman J, Bastian HM, Friedman AW, et al. Systemic lupus erythematosus in three ethnic groups: XVI. Association of hydroxychloroquine use with reduced risk of damage accrual. Arthritis Rheum. 2005; 52(5):1473–1480. [PubMed: 15880829]

91. Sutton EJ, Davidson JE, Bruce IN. The systemic lupus international collaborating clinics (SLICC) damage index: a systematic literature review. Semin. Arthritis Rheum. 2013; 43(3):352–361. [PubMed: 23786872]

92. Kuhn A, Ruland V, Bonsmann G. Cutaneous lupus erythematosus: update of therapeutic options part I. J. Am. Acad. Dermatol. 2011; 65(6):e179–e193. [PubMed: 20739095]

93. Petri M. Use of hydroxychloroquine to prevent thrombosis in systemic lupus erythematosus and in antiphospholipid antibody-positive patients. Curr. Rheumatol. Rep. 2011; 13(1):77–80. [PubMed: 20978875]

94. Kaiser R, Cleveland CM, Criswell LA. Risk protective factors for thrombosis in systemic lupus erythematosus: results from a large multiethnic cohort. Ann. Rheum. Dis. 2009; 68(2):238–241. [PubMed: 18782792]

95. Tektonidou MG, Laskari K, Panagiotakos DB, Moutsopoulos HM. Risk factors for thrombosis and primary thrombosis prevention in patients with systemic lupus erythematosus with or without antiphospholipid antibodies. Arthritis Rheum. 2009; 61(1):29–36. [PubMed: 19116963]

96. Petri M, Howard D, Repke J. Frequency of lupus flare in pregnancy. The Hopkins lupus pregnancy center experience. Arthritis Rheum. 1991; 34(12):1538–1545. [PubMed: 1670196]

97. Mekinian A, Lazzaroni MG, Kuzenko A, Alijotas-Reig J, Ruffatti A, Levy P, et al. The efficacy of hydroxychloroquine for obstetrical outcome in antiphospholipid syndrome: data from a European multicenter retrospective study. Autoimmun. Rev. 2015; 14(6):498–502. [PubMed: 25617818]

98. Kerr G, Aujero M, Richards J, Sayles H, Davis L, Cannon G, et al. Associations of hydroxychloroquine use with lipid profiles in rheumatoid arthritis: pharmacologic implications. Arthritis Care Res. (Hoboken). 2014 Nov; 66(11):1619–1626. http://dx.doi.org/10.1002/acr.22341. [PubMed: 24692402]

99. Durcan L, Winegar DA, Connelly MA, Otvos JD, Magder LS, Petri M. Longitudinal evaluation of lipoprotein variables in systemic lupus erythematosus reveals adverse changes with disease activity and prednisone and more favorable profiles with hydroxychloroquine therapy. J. Rheumatol. 2016 Apr; 43(4):745–750. http://dx.doi.org/10.3899/jrheum.150437. [PubMed: 26834214]

100. Wallace DJ, Gudsoorkar VS, Weisman MH, Venuturupalli SR. New insights into mechanisms of therapeutic effects of antimalarial agents in SLE. Nat. Rev. Rheumatol. 2012; 8(9):522–533. [PubMed: 22801982]

101. Gerstein HC, Thorpe KE, Taylor DW, Haynes RB. The effectiveness of hydroxychloroquine in patients with type 2 diabetes mellitus who are refractory to sulfonylureas-a randomized trial. Diabetes Res. Clin. Pract. 2002; 55(3):209–219. [PubMed: 11850097]

102. James JA, Kim-Howard XR, Bruner BF, Jonsson MK, McClain MT, Arbuckle MR, et al. Hydroxychloroquine sulfate treatment is associated with later onset of systemic lupus erythematosus. Lupus. 2007; 16(6):401–409. [PubMed: 17664230]

103. Izmirly PM, Kim MY, Llanos C, Le PU, Guerra MM, Askanase AD, et al. Evaluation of the risk of anti-SSA/Ro-SSB/La antibody-associated cardiac manifestations of neonatal lupus in fetuses of mothers with systemic lupus erythematosus exposed to hydroxychloroquine. Ann. Rheum. Dis. 2010; 69(10):1827–1830. [PubMed: 20447951]

104. Izmirly PM, Costedoat-Chalumeau N, Pisoni CN, Khamashta MA, Kim MY, Saxena A, et al. Maternal use of hydroxychloroquine is associated with a reduced risk of recurrent anti-SSA/Ro-antibody-associated cardiac manifestations of neonatal lupus. Circulation. 2012; 126(1):76–82. [PubMed: 22626746]

105. van den Borne BE, Dijkmans BA, de Rooij HH, le Cessie S, Verweij CL. Chloroquine and hydroxychloroquine equally affect tumor necrosis factor-alpha, interleukin 6, and interferon-gamma production by peripheral blood mononuclear cells. J. Rheumatol. 1997; 24(1):55–60. [PubMed: 9002011]

106. Bondeson J, Sundler R. Antimalarial drugs inhibit phospholipase A2 activation and induction of interleukin 1beta and tumor necrosis factor alpha in macrophages: implications for their mode of action in rheumatoid arthritis. Gen. Pharmacol. 1998; 30(3):357–366. [PubMed: 9510087]

107. Silva JC, Mariz HA, Rocha LF, Oliveira PS, Dantas AT, Duarte AL, et al. Hydroxychloroquine decreases Th17-related cytokines in systemic lupus erythematosus and rheumatoid arthritis patients. Clin. (Sao Paulo). 2013; 68(6):766–771.

108. Sacre K, Criswell LA, McCune JM. Hydroxychloroquine is associated with impaired interferon-alpha and tumor necrosis factor-alpha production by plasmacytoid dendritic cells in systemic lupus erythematosus. Arthritis Res. Ther. 2012; 14(3):R155. [PubMed: 22734582]

109. Willis R, Seif AM, McGwin G, Martinez-Martinez LA, Gonzalez EB, Dang N, et al. Effect of hydroxychloroquine treatment on pro-inflammatory cytokines and disease activity in SLE patients: data from LUMINA (LXXV), a multiethnic US cohort. Lupus. 2012; 21(8):830–835. [PubMed: 22343096]

110. Ziegler HK, Unanue ER. Decrease in macrophage antigen catabolism caused by ammonia and chloroquine is associated with inhibition of antigen presentation to T cells. Proc. Natl. Acad. Sci. U. S. A. 1982; 79(1):175–178. [PubMed: 6798568]

111. Costedoat-Chalumeau N, Dunogue B, Morel N, Le Guern V, Guettrot-Imbert G. Hydroxychloroquine: a multifaceted treatment in lupus. Presse Med. 2014; 43(6 Pt 2):e167–e180. [PubMed: 24855048]

112. Lafyatis R, York M, Marshak-Rothstein A. Antimalarial agents: closing the gate on Toll-like receptors? Arthritis Rheum. 2006; 54(10):3068–3070. [PubMed: 17009223]

113. An J, Woodward JJ, Sasaki T, Minie M, Elkon KB. Cutting edge: antimalarial drugs inhibit IFN-β production through blockade of cyclic GMP-AMP synthase-DNA interaction. J. Immunol. 2015; 194(9):4089–4093. [PubMed: 25821216]

114. A randomized study of the effect of withdrawing hydroxychloroquine sulfate in systemic lupus erythematosus. The Canadian Hydroxychloroquine Study Group. N. Engl. J. Med. 1991; 324(3): 150–154. [PubMed: 1984192]

115. Petri MA, van Vollenhoven RF, Buyon J, Levy RA, Navarra SV, Cervera R, et al. Baseline predictors of systemic lupus erythematosus flares: data from the combined placebo groups in the phase III belimumab trials. Arthritis Rheum. 2013; 65(8):2143–2153. [PubMed: 23754628]

116. Costedoat-Chalumeau N, Amoura Z, Hulot JS, Hammoud HA, Aymard G, Cacoub P, et al. Low blood concentration of hydroxychloroquine is a marker for and predictor of disease exacerbations in patients with systemic lupus erythematosus. Arthritis Rheum. 2006; 54(10):3284–3290. [PubMed: 17009263]

117. Frances C, Cosnes A, Duhaut P, Zahr N, Soutou B, Ingen-Housz-Oro S, et al. Low blood concentration of hydroxychloroquine in patients with refractory cutaneous lupus erythematosus: a French multicenter prospective study. Arch. Dermatol. 2012; 148(4):479–484. [PubMed: 22508872]

118. Costedoat-Chalumeau N, Galicier L, Aumaitre O, Frances C, Le Guern V, Liote F, et al. Hydroxychloroquine in systemic lupus erythematosus: results of a French multicentre controlled trial (PLUS Study). Ann. Rheum. Dis. 2013; 72(11):1786–1792. [PubMed: 23144449]

119. Durcan L, Clarke WA, Magder LS, Petri M. Hydroxychloroquine blood levels in SLE: clarifying dosing controversies and improving adherence. J. Rheumatol. 2015 Nov; 42(11):2092–2097. http://dx.doi.org/10.3899/jrheum.150379. [PubMed: 26428205]

120. Williams HJ, Egger MJ, Singer JZ, Willkens RF, Kalunian KC, Clegg DO, et al. Comparison of hydroxychloroquine and placebo in the treatment of the arthropathy of mild systemic lupus erythematosus. J. Rheumatol. 1994; 21(8):1457–1462. [PubMed: 7983646]

121. Gleicher N, Elkayam U. Preventing congenital neonatal heart block in offspring of mothers with anti-SSA/Ro and SSB/La antibodies: a review of published literature and registered clinical trials. Autoimmun. Rev. 2013; 12(11):1039–1045. [PubMed: 23684701]

122. Clancy RM, Alvarez D, Komissarova E, Barrat FJ, Swartz J, Buyon JP. Ro60-associated single-stranded RNA links inflammation with fetal cardiac fibrosis via ligation of TLRs: a novel pathway to autoimmune-associated heart block. J. Immunol. 2010; 184(4):2148–2155. [PubMed: 20089705]

123. Melles RB, Marmor MF. The risk of toxic retinopathy in patients on long-term hydroxychloroquine therapy. JAMA Ophthalmol. 2014; 132(12):1453–1460. [PubMed: 25275721]

124. Marmor MF, Kellner U, Lai TY, Lyons JS, Mieler WF. Revised recommendations on screening for chloroquine and hydroxychloroquine retinopathy. Ophthalmology. 2011; 118(2):415–422. [PubMed: 21292109]

125. Dorner T, Giesecke C, Lipsky PE. Mechanisms of B cell autoimmunity in SLE. Arthritis Res. Ther. 2011; 13(5):243. [PubMed: 22078750]

126. Mysler EF, Spindler AJ, Guzman R, Bijl M, Jayne D, Furie RA, et al. Efficacy and safety of ocrelizumab in active proliferative lupus nephritis: results from a randomized, double-blind. phase III study, Arthritis Rheum. 2013; 65(9):2368–2379. [PubMed: 23740801]

127. Merrill JT, Neuwelt CM, Wallace DJ, Shanahan JC, Latinis KM, Oates JC, et al. Efficacy and safety of rituximab in moderately-to-severely active systemic lupus erythematosus: the randomized, double-blind, phase II/III systemic lupus erythematosus evaluation of rituximab trial. Arthritis Rheum. 2010; 62(1):222–233. [PubMed: 20039413]

128. Wallace DJ, Gordon C, Strand V, Hobbs K, Petri M, Kalunian K, et al. Efficacy and safety of epratuzumab in patients with moderate/severe flaring systemic lupus erythematosus: results from two randomized, double-blind, placebo-controlled, multicentre studies (ALLEVIATE) and follow-up. Rheumatol. Oxf. 2013; 52(7):1313–1322.

129. Furie R, Petri M, Zamani O, Cervera R, Wallace DJ, Tegzova D, et al. A phase III, randomized, placebo-controlled study of belimumab, a monoclonal antibody that inhibits B lymphocyte stimulator, in patients with systemic lupus erythematosus. Arthritis Rheum. 2011; 63(12):3918–3930. [PubMed: 22127708]

130. Furie RA, Leon G, Thomas M, Petri MA, Chu AD, Hislop C, et al. A phase 2, randomised, placebo-controlled clinical trial of blisibimod, an inhibitor of B cell activating factor, in patients with moderate-to-severe systemic lupus erythematosus, the PEARL-SC study. Ann. Rheum. Dis. 2015; 74(9):1667–1675. [PubMed: 24748629]

131. Isenberg DA, Petri M, Kalunian K, Tanaka Y, Urowitz MB, Hoffman RW, et al. Efficacy and safety of subcutaneous tabalumab in patients with systemic lupus erythematosus: results from ILLUMINATE-1, a 52-week, phase III, multicentre, randomised, double-blind, placebo-controlled study. Ann. Rheum. Dis. 2016; 75(2):323–331. [PubMed: 26338095]

132. Isenberg D, Gordon C, Licu D, Copt S, Rossi CP, Wofsy D. Efficacy and safety of atacicept for prevention of flares in patients with moderate-to-severe systemic lupus erythematosus (SLE): 52-week data (APRIL-SLE randomised trial). Ann. Rheum. Dis. 2015; 74(11):2006–2015. [PubMed: 24951103]

133. Navarra SV, Guzman RM, Gallacher AE, Hall S, Levy RA, Jimenez RE, et al. Efficacy and safety of belimumab in patients with active systemic lupus erythematosus: a randomised, placebo-controlled, phase 3 trial. Lancet. 2011; 377(9767):721–731. [PubMed: 21296403]

134. Stohl W, Hiepe F, Latinis KM, Thomas M, Scheinberg MA, Clarke A, et al. Belimumab reduces autoantibodies, normalizes low complement levels, and reduces select B cell populations in patients with systemic lupus erythe-matosus. Arthritis Rheum. 2012; 64(7):2328–2337. [PubMed: 22275291]

135. van Vollenhoven RF, Petri MA, Cervera R, Roth DA, Ji BN, Kleoudis CS, et al. Belimumab in the treatment of systemic lupus erythematosus: high disease activity predictors of response. Ann. Rheum. Dis. 2012; 71(8):1343–1349. [PubMed: 22337213]

136. Dooley MA, Houssiau F, Aranow C, D'Cruz DP, Askanase A, Roth DA, et al. Effect of belimumab treatment on renal outcomes: results from the phase 3 belimumab clinical trials in patients with SLE. Lupus. 2013; 22(1):63–72. [PubMed: 23263865]

137. Illei GG, Cervera R, Burt RK, Doria A, Hiepe F, Jayne D, et al. Current state and future directions of autologous hematopoietic stem cell transplantation in systemic lupus erythematosus. Ann. Rheum. Dis. 2011; 70(12):2071–2074. [PubMed: 21873334]

138. Sun L. Stem cell transplantation: progress in Asia. Lupus. 2010; 19(12):1468–1473. [PubMed: 20947559]

139. Marmont AM. Immune ablation with stem-cell rescue: a possible cure for systemic lupus erythematosus? Lupus. 1993; 2(3):151–156. [PubMed: 8103697]

140. Jayne D, Passweg J, Marmont A, Farge D, Zhao X, Arnold R, et al. Autologous stem cell transplantation for systemic lupus erythematosus. Lupus. 2004; 13(3):168–176. [PubMed: 15119545]

141. Traynor AE, Schroeder J, Rosa RM, Cheng D, Stefka J, Mujais S, et al. Treatment of severe systemic lupus erythematosus with high-dose chemotherapy and haemopoietic stem-cell transplantation: a phase I study. Lancet. 2000; 356(9231):701–707. [PubMed: 11085688]

142. Alexander T, Thiel A, Rosen O, Massenkeil G, Sattler A, Kohler S, et al. Depletion of autoreactive immunologic memory followed by autologous hematopoietic stem cell transplantation in patients with refractory SLE induces long-term remission through de novo generation of a juvenile and tolerant immune system. Blood. 2009; 113(1):214–223. [PubMed: 18824594]

143. Zhang L, Bertucci AM, Ramsey-Goldman R, Burt RK, Datta SK. Regulatory T cell (Treg) subsets return in patients with refractory lupus following stem cell transplantation, and TGF-beta-producing CD8+ Treg cells are associated with immunological remission of lupus. J. Immunol. 2009; 183(10):6346–6358. [PubMed: 19841178]

144. Wang D, Feng X, Lu L, Konkel JE, Zhang H, Chen Z, et al. A CD8 T cell/indoleamine 2,3-dioxygenase axis is required for mesenchymal stem cell suppression of human systemic lupus erythematosus. Arthritis Rheumatol. 2014; 66(8):2234–2245. [PubMed: 24756936]

145. Petri MA, Mease PJ, Merrill JT, Lahita RG, Iannini MJ, Yocum DE, et al. Effects of prasterone on disease activity and symptoms in women with active systemic lupus erythematosus. Arthritis Rheum. 2004; 50(9):2858–2868. [PubMed: 15452837]

146. Hartkamp A, Geenen R, Godaert GL, Bijl M, Bijlsma JW, Derksen RH. Effects of dehydroepiandrosterone on fatigue and well-being in women with quiescent systemic lupus erythematosus: a randomised controlled trial. Ann. Rheum. Dis. 2010; 69(6):1144–1147. [PubMed: 19854713]

147. Piantoni S, Andreoli L, Scarsi M, Zanola A, Dall'Ara F, Pizzorni C, et al. Phenotype modifications of T-cells and their shift toward a Th2 response in patients with systemic lupus erythematosus supplemented with different monthly regimens of vitamin D. Lupus. 2015; 24(4–5):490–498. [PubMed: 25801892]

148. Levy RA, Vilela VS, Cataldo MJ, Ramos RC, Duarte JL, Tura BR, et al. Hydroxychloroquine (HCQ) in lupus pregnancy: double-blind and placebo-controlled study. Lupus. 2001; 10(6):401–404. [PubMed: 11434574]

Table 1

Clinical Trials in SLE involving Dehydroeipandrosterone.

Author & year	Design	N	Patients	Dose + time	Outcome	Result
van Vollenhoven et al., 1994 [30]	Single center Open label Uncontrolled	10	Female	200 mg	SLEDAI Patient assessment (0–100) Physician assessment (0 –100)	SLEDAI: decreased (non significant) Physician assessment: improved (P = 0.04) Patient assessment: unchanged
van Vollenhoven et al., 1999 [31]	Number of centers not stated, in San Fransisco area. Double-blind Randomized Placebo-controlled	21	Female 'Severe'	200mg/placebo 6 months, 6 months open label extension	'Responder analysis' SLEDAI SLAM BMD	Responder: DHEA 7/9 achieved response, placebo 4/10 (non-significant). SLEDAI: No significant change SLAM: No significant change BMD: Non significant increase in lumbar spine.
Petri et al., 2002 [32]	Multi-center Double-blind Randomized Placebo-controlled	191	Female Prednisone 10–30 mg	100mg/200mg/ placebo 7–9 months	Sustained reduction <7.5 mg prednisone SLEDAI SF-36 KFSS Damage index	No reduction in prednisone dose. 200 mg: 55% responder 100 mg: 44% Placebo:41% SF-36: Non significant KFSS: Non significant change
Chang et al., 2002 [33]	Multicenter Randomized Double blind Placebo controlled	120	Female	200 mg/placebo 24 weeks	SLAM Flare SLEDAI	SLAM: Non significant Flare: 16% decrease Patient global: Improved −5.5 versus 5.4; P = 0.005
Chang et al., 2004 [22]	Single center Randomized Double blind Placebo controlled (subgroup of Chang et al., 2002(22))	32	Female Active SLE Chinese	200mg/placebo	Il 10 Il 1β TNFα	IL 10: Reduced (9.21–1.89 pg/ ml with DHEA) IL1β: UnchangedTNFα: Undetectable
Petri et al., 2004 [145]	Multicenter Randomized Double-blind Placebo-controlled	381	Female SLAM >7 SLEDAI >2 (n = 293)	200mg/placebo	SLEDAI SLAM KFSS Patient global Time to flare	ITT analysis: No difference SLEDAI: >2 Response: 58.5% versus 44.5% (P = 0.017) Flare: Less with SLEDAI>2 Patient global: Less worsening SLEDAI: Less worsening
Harkamp et al., 2004 [34]	2 centers Randomized Double-blind Placebo-controlled	58: Initial DEXA 56: Both baseline and follow up DEXA	Female 10 mg prednisone	200 mg/placebo 12 months	BMD SLEDAI	BMD: No significant difference (Postmenopausal mean change was 1.80% with DHEA, −2.32% with placebo. Premenopausal: No change) SLEDAI: No change

Author & year	Design	N	Patients	Dose + time	Outcome	Result
Mease et al., 2005 [35]	Multicenter Double-blind Randomized Placebo-controlled	66: Initial DEXA 55: Both baseline and follow up DEXA	Female SLE 10 mg prednisone: (6 months)	200mg/placebo 12 months	BMD (% change) SLEDAI SLAM KFSS SLICC damage index Patient VAS Physician VAS	BMD: Lumbar spine, gain in BMD of 1.7% in versus loss of −1.1% (P = 0.003). Total hip, 2.0% gain versus a loss of −0.3% with placebo (p = 0.013) No significant change in other outcomes
Nordmark et al., 2005 [37]	2 centers Double-blind Randomized Placebo-controlled	41	Female >5 mg prednisone Age 20–65	200mg/placebo 6 months with 6 month open extension	SF-36 BMD Body composition SLEDAI/mSLEDAI MCS HSCL-56 PGWB	SF-36: Improved emotional components BMD: No increase Body composition: Increased waist hip ratio SLEDAI: No change mSLEDAI: No change MCS: Improved (unsustained) HSCL-56: Improved PGWB: Nonsignificant
Hartkamp et al., 2010 [146]	2 centers Double-blind Randomized Placebo-controlled	60	Female No prednisone	200mg/placebo 12 months	Fatigue: MFI Depression: Zung self-rating scale SF-36 Pain VAS SLEDAI	Fatigue: No change Depression: No change Well-being: No change
Sanchez-Guerrero et al., 2008 [36]	Multicenter Double-blind Randomized Placebo-controlled	155	Female >5 mg prednisone No osteoporosis/ bisphosphonate	200mg/placebo in phase 1, then 200mg/100 mg in extension phase Calcium Vitamin D	BMD	BMD: Increased at lumbar spine, 200 mg dose, (non-significant at 6 months, significant at 18 months). Maintenance of BMD at hip. (100 mg no effect)

SLEDAI: Systemic lupus erythematosus disease activity index, DHEA: Dehydroepiandrosterone SLAM: Systemic lupus activity measure, BMD: Bone mineral density, SF-36: Short form health survey, KFSS: Krupps fatigue severity scale, DEXA: Dual-energy X-ray absorptiometry, mSLEDAI: Modified systemic lupus erythematosus disease activity index, MCS: Mental component summary, HSCL-56: Hopkins symptom check list, PGWB: Psychological general wellbeing index, MFI: Multidimensional fatigue inventory, VAS: Visual analogue scale.

Table 2

Vitamin D supplementation studies in SLE.

Author & year	Design	N	Patients + location	Dose	Outcome	Result
Ruiz-Irastorza et al., 2010 [72]	Longitudinal Observational (prospective-cohort) Single center	80	SLE Spain	600–800iu/day 24 months	SLEDAI SDI Fatigue (VAS)	Fatigue: VAS, 4.1 versus 3.3 P = 0.015. SLEDAI: No effect SDI: No effect
Terrier et al., 2012 [73]	Single center Open-label	20	SLE France	100,000Iu weekly for 4 weeks, then 100,000 monthly for 6 months	Safety SLEDAI B cells T cells Cytokines	SLEDAI: 2.9 ± 2.5 to 2.6 ± 2.5 at 2 months, non significant. Anti dsDNA: Decreased at 2 and 6 months. CD4: Non significant increase CD*: Decreased in frequency but not in number. T regs: Increased
Petri et al., 2013 [74]	Longitudinal Cohort	1006	SLE USA (37% African American)	50,000 iu weekly + 400 iu calcium/vitamin D daily	SLEDAI Physician global (0 –3) UPCR	SLEDAI: Significant decrease. Physician global: Improved significantly UPCR: 20-ng/ml increase in the 25(OH)D value was associated with a 4% decrease in UPCR
Abou-Raya et al., 2013 [75]	Randomized (2:1)	267	SLE Egypt	2000iu daily/placebo	SLEDAI	SLEDAI: Correlated negatively with vitamin D. SLEDAI improved with supplementation.
Andreoli et al., 2015 [77]	Randomized Unblinded	34	SLE Italy	300,000 bolus, 50,000iu monthly compared with	T cell and B cell populations	Promotion of regulatory T cells Production of Th2 cytokines
Piantoni et al., 2015 [147] (reported in two parts)			SLE Italy	25,000iu monthly	SLE serology	Serology: Unchanged
Arno w et al., 2015 [63]	Randomized Double blind Placebo controlled	54	SLE USA (54% African American)	2000iu, 4000iu/placebo	Interferon signature	No effect on interferon signature
Lima et al., 2015 [76]	Randomized Double blind Placebo controlled	50	Juvenile SLE Brazil (30% non-caucasian)	50,000 iu weekly versus placebo	K-FSS SLEDAI ECLAM	SLEDAI: Improved (P = 0.01) ECLAM: Improved (P = 0.006)

SLEDAI: SLE disease activity index, SDI: ACR/SLICC damage index, UPCR: Urine protein to Creatinine ratio, VAS: visual analogue scale, ECLAM: European Consensus Lupus Activity Measurement, K-FSS: Kids fatigue severity scale.

Table 3

Hydroxychloroquine clinical trials in SLE.

Author & year	Design	N	Patients	Dose + Time	Outcome	Result
Williams et al., 1996 [87].	Randomized, Double blind Placebo-controlled	73	SLE Arthritis/Arthralgia	48 weeks	Joint count Disease activity Pain (patient reported)	Improved patient reported pain
Levy et al., 2001 [148].	Randomized Placebo-controlled	26	SLE & DLE	Pregnancy duration	SLEPDAI Birth outcomes	Decreased prednisone in HCQ group Improved SLEPDAI Higher birth weight
The Canadian Hydroxychloroquine Study Group [114].	Randomized Double blind Placebo-controlled withdrawal study	47	SLE	6 months	Flare	Relative risk of flare 6.1 times higher with withdrawal of HCQ.
Costedoat-Chalumeau et al. 2013 [118]	Randomized Double-blind Placebo controlled	171	SLE (with HCQ level <1000 ng/ml)	7 months	SLEDAI Flare	No difference in SLEDAI with attaining therapeutic blood level (p = 0.70)

SLE: Systemic lupus erythematosus, DLE: Discoid lupus erythematosus, SLEPDAI: Systemic lupus erythematosus pregnancy disease activity index, HCQ: Hydroxychloroquine, SLEDAI: Systemic lupus erythematosus disease activity index.