Tumour necrosis factor and other cytokines in murine lupus

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Investigations into the structure, expression and functional status of cytokines, as well as on the possible utility of their agonists and antagonists as therapeutic agents in lupus, have received prominent attention. In fact, mouse strains predisposed to lupus (NZBxW, BXSB, MRL-*lpr*) have long constituted the primary resource to investigate the role of cytokines in autoimmunity. An additional impetus for such investigations in recent years has been provided by the discovery that T cells may be polarised during an ongoing immune response into the so called T_H1 and T_H2 subsets, which display distinct cytokine profiles and effector functions. Thus, following antigen recognition, cytokines present at the site of priming together with other factors, such as type of antigen presenting cell, amount of antigen, co-stimulatory molecules, affinity and duration of exposure direct the induction of either T_H1 cells, which secrete IL2, IFNγ and TNFβ, or T_H2 cells, which secrete IL4, IL5, IL6, IL10 and IL13.[1] Another cytokine, IL12, produced by activated macrophages and dendritic cells, is a strong inducer of T_H1 cells in which the β-subunit of its receptor is retained but lost on T_H2 cells. The former cells then provide protection from intracellular pathogens, activate phagocytes, induce IgG2a antibodies, and promote DTH responses, whereas the latter cells provide protection from extracellular pathogens, activate eosinophils, induce IgE mediated allergic reactions, and generally promote humoral responses in which IgG1 predominates. The molecular events associated with this polarisation have not been fully elucidated, but certain protooncogenes, kinases and transcription factors seem to play a part.[2–6]

Based on this T cell division, it has been hypothesised that organ specific autoimmune diseases such as IDDM should be mediated by T_H1 cells, whereas humorally mediated autoimmune diseases such as lupus should be mediated by T_H2 cells. As summarised in tables 1, 2 and 3 and reviewed below, such a paradigm does not seem to apply to lupus because both T_H1 and T_H2 cytokines have been found to exert profound effects on spontaneous mouse models of this disease.

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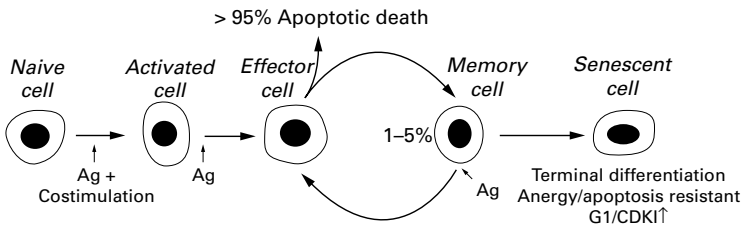

Figure 1 Schematic representation of replicative senescence in repeatedly activated autoreactive memory T cells.

T_H1 cytokines

The earliest cytokine defect identified in all lupus strains was reduced production of, and response to, IL2.[7 8] This defect appears at 4–6 weeks of age in MRL-*lpr* and BXSB mice, and somewhat later in the (NZBxNZW)F_1 mice, wherein it becomes more pronounced with disease advancement. The cause(s) of this defect is unknown, but several possibilities have been considered, including impaired T cell receptor (TCR) signal transduction, IL2R structural defects, abnormalities in certain transcription factors, and exhaustion subsequent to excessive and repetitive activation in vivo.[9–11] Among these possibilities, we favour that of exhaustion and attainment of a replicative senescence state. According to this hypothesis, because of continued autoantigen presence there is a repetitive interconversion of effector T cells into memory cells and vice versa. Upon a defined number of such interconversions, memory cells become refractory to further stimulation and entry into the cell cycle (fig 1). In support of this concept is the finding that the frequency of in vivo cycling T cells in lupus mice declines progressively with age, and this decline is associated with increased expression of cyclin dependent kinase inhibitors, a characteristic of cells that reach replicative senescence.[12 13]

The relation of the IL2 defect to the disease process remains unclear. Several reports have shown correction of the in vitro *lpr* T cell defective proliferation and apoptosis by exogenous IL2.[14–16] Moreover, an early study found reduced serological, cellular and histological abnormalities of the MRL-*lpr* mouse following infections with a vaccinia virus-IL2 construct.[17] Similarly, in a very recent study,[18] MRL-*lpr* mice infected orally by gavage with an attenuated strain of *Salmonella typhimurium* transfected with the IL2 gene (administered at 6 weeks of age and repeated every three weeks to 15 weeks of age) were shown to have reduced double negative T cells, autoantibody levels, glomerulonephritis (GN) and vasculitis. In contrast, intramuscular injections of an IL2 encoding cDNA expression vector in MRL-*lpr* mice were reported to increase autoantibody production and disease.[19] Another study[20] reported no effect on disease progression and severity in (NZBxNZW)F_1 mice treated with low or high doses of human recombinant IL2, while others found suppression of nephritis in (NZBxNZW)F_1 mice treated with anti-IL2R mAb.[21] The divergence of results in these stud-

Table 1 In vitro and in vivo cytokine expression levels in lupus mice

Strains	*IL1β*	*IL2*	*IL4*	*IL6*	*IL10*	*IL12*	*IFNγ*	*TNFα*	*TGFβ*
(NZB X W)F_1	↑	↓	↓	ND	↑	↓	↑	↓	↑
MRL-*lpr*	↑	↓	↓	↑	↑	↑	↑↑	↑	ND
BXSB-*Yaa*	↑	↓	ND	ND	↑	?	↑	↑	↑

↑increased expression, ↓decreased expression, ND no difference from normal controls, ? no data exist.

ies may be attributable to differences in the vectors and lupus strains used as well as the route, timing and dose of IL2. Such differences may affect overall IL2 levels, its expression in the microenvironment of secondary lymphoid organs, and rates of T cell proliferation and apoptosis. Nevertheless, some studies have suggested dissociation of severe lupus disease and the in vitro IL2 deficiency.[22–24] Overall, it seems that the role of IL2 in lupus disease has not yet been conclusively decided, and further investigation is required in view of the presence of autoimmunity in normal background mice with forced deletions of the IL2[25] or the IL2R[26 27] genes.

Among the many cytokine abnormalities found in lupus mice, the most consistent has been high expression of IFNγ.[28–30] The importance of this cytokine in murine lupus pathogenesis was initially suggested by the demonstration that (NZBxNZW)F_1 mice treated with IFNγ showed accelerated disease and conversely, treatment with anti-IFNγ antibody[31] or soluble IFNγR[32] early in life significantly delayed disease progression. A study on a long lived substrain of MRL-*lpr* mice also showed reduced IFNγ levels compared with the early life severe disease developing parental strain concomitant with a shift of Ig isotypes from the C-fixing IgG2a and the cryogenic-nephritogenic IgG3 to the less pathogenic IgG1 isotype.[33] Moreover, MRL-*lpr* mice intercrossed with an IFNγ gene deleted mouse,[34] MRL-*lpr* mice rendered congenic for deletions in either the IFNγ[35] or the IFNγR,[36 37] and (NZBxNZW)F_1 mice congenic for the IFNγR deletion[38] all showed significant reduction in humoral and histological characteristics of the disease. In one of these studies,[35] three notable observations were made. Firstly, hypergammaglobulimaenia was maintained in IFNγ$^{-/-}$ mice with a switch from IgG2a to IgG1 predominance but, importantly, the dramatic decrease in levels of the dominant IgG2a anti-dsDNA autoantibodies was not associated with a compensatory increase in T_H2 associated IgG subclasses. This finding suggested that therapeutic interventions with the goal of reducing IFNγ levels in lupus may selectively affect certain pathogenic autoimmune responses without significantly compromising the person's capacity to respond to exogenous antigens. This is a considerable advantage over other contemplated treatments, such as those with co-stimulation blockers wherein severe and indiscriminate immune compromise may result. Secondly, remarkably, early death and GN were also prevented in IFNγ$^{+/-}$ *lpr* mice (50% reduction in IFNγ levels) despite the fact that autoantibody levels and kidney immune complex deposits were equal to those in the wild type MRL-*lpr* mice. This result suggests that even partial reduction of IFNγ may curtail the deleterious effects it exerts locally on the afflicted organ. A similar uncoupling of the local inflammatory response from autoantibody production and immune complex kidney deposition has also been observed in (NZBxNZW)F_1 mice deficient in the FcγRI and FcγRIII,[39] that is, the FcγR$^{-/-}$ (NZBxNZW)F_1 mice showed similar autoantibody levels and immune complex kidney deposits to the FcγR$^{+/+}$ mice, yet they exhibited prolonged survival, reduced kidney disease and proteinuria.

Table 2 Effects of cytokine agonists and antagonists in murine lupus

Strain	*Treatment*	*Disease outcome*	*References*
(NZB X W)F_1	rIL1	More severe	63
MRL-*lpr*	rIL1R	Beneficial	93
MRL-*lpr*	rIL1R antagonist	No effect	94
(NZB X W)F_1	human rIL2 high and low doses	No effect	20
(NZB X W)F_1	Anti-IL2R mAb	Beneficial	21
MRL-*lpr*	Vaccinia virus IL2 construct	Beneficial	17
MRL-*lpr*	*S typhimurium* with IL2 gene	Beneficial	18
MRL-*lpr*	IL2 expression vector injected IM	More severe	19
(NZB X W)F_1	Anti-IL4 mAb	Beneficial	44
(NZB X W)F_1	Anti-IL4 mAb + Anti-IL12 mAb	No effect	44
(NZB X W)F_1	rIL6	More severe	49
(NZB X W)F_1	Anti-IL6 mAb	Beneficial	50
MRL-*lpr*	Anti-IL6R mAb	Beneficial	51
(NZB X W)F_1	Anti-IL10 mAb	Beneficial	54
(NZB X W)F_1	Anti-IL10 mAb + Anti-TNFα mAb	No effect	54
(NZB X W)F_1	rIL10	More severe	54
(NZB X W)F_1	AS101 (IL10 inhibiting immunomodulator)	Beneficial	55
MRL-*lpr*	rIL12	More severe	41
(NZB X W)F_1	Anti-IL12 mAb	Reduced anti-DNA, no effect on GN	44
(NZB X W)F_1	rIFNy	More severe	31
(NZB X W)F_1	Anti-IFNγ mAb	Beneficial	31
(NZB X W)F_1	sIFNγR	Beneficial	32
(NZB X W)F_1	rTNFα low dose (2 mo)	No effect	63
(NZB X W)F_1	rTNFα low dose (4 mo)	More severe	63
(NZB X W)F_1	rTNFα intermediate dose (4 mo)	No effect	63
(NZB X W)F_1	rTNFα high dose administered early	Beneficial	58, 73, 74
(NZB X W)F_1	rTNFα high dose administered late	No effect	58, 73
MRL-*lpr*	Anti-TNFα mAb	Beneficial	75
MRL-*lpr*	Transcriptional inhibitor of TNFα	Beneficial	79
MRL-*lpr*	Anti-TGFß mAb	Beneficial	97
MRL-*lpr*	*S typhimurium* with TGFß gene	No effect	18
MRL-*lpr*	TGF-ß expression vector injected IM	Beneficial	19

Table 3 Cytokine gene knockout and transgenic lupus mice

Strain	*Transgene/Knockout*	*Outcome*	*References*
MRL-*lpr*	IFNγ-/-	Beneficial	34, 35
MRL-*lpr*	IFNγ+/-	Beneficial	35
MRL-*lpr*	IFNγR-/-	Beneficial	36, 37
(NZB X W)F_1	IFNγR-/-	Beneficial	38
(NZW X B6-*γaa*)F_1	Transgenic IL4	Beneficial	52
MRL-*lpr*	IL4-/-	Beneficial	34

Thirdly, with regards to mechanisms, the major observation was severe reduction of MHC class I and class II expression by splenic macrophages in IFNγ$^{-/-}$ mice, but not IFNγ$^{+/-}$ mice, whereas reduction in MHC class II expression by tubular epithelial cells was observed in both homozygous and heterozygous IFNγ deleted mice. Thus, insufficient upregulation of MHC on antigen presenting cells, either systemically and/or locally, may be the major mechanism by which the beneficial effects of reduced IFNγ levels are mediated.

The role of IL12 in murine lupus has also been investigated. An intrinsic defect in the in vitro production of IL12 by endotoxin activated macrophages of MRL-+/+ and (NZBxNZW)F_1 mice has been reported.[40] Other studies, however, found that peritoneal macrophages of MRL-*lpr* mice hyperproduce IL12 after stimulation with IFNγ and/or LPS, and exhibit high concentrations of IL12 in the serum[41] as well as kidney.[42] Moreover, daily injections of recombinant IL12 led to increased serum levels of IFNγ and nitric oxide (NO) metabolites, and accelerated GN in this model.[41] These findings, together with previous reports that NO synthase inhibitors can ameliorate autoimmune disease in MRL-*lpr* mice,[43] suggest that the high production and response to IL12 by these mice may be important in disease pathogenesis. Treatment of (NZBxNZW)F_1 mice with anti-IL12, however, was ineffective in preventing the onset or severity of GN.[44]

T_H2 cytokines

The part played by the T_H2 cytokines IL6, IL4 and IL10 in murine lupus has also been the subject of intense study. Unaltered or increased levels of IL6 have been reported.[45–47] Macrophage depletion in in vitro cultures of splenic cells from (NZBxNZW)F_1 mice yielded reduced IL6 levels concomittant to decreased IgG anti-DNA levels.[48] Furthermore, administration of recombinant IL6 to (NZBxNZW)F_1 mice caused accelerated GN that correlated with marked upregulation of mesangial MHC class II antigen and glomerular ICAM-1 expression.[49] Chronic administration of a rat anti-IL6 mAb had no effect on the development of GN in (NZBxNZW)F_1 mice because of the elicitation of an anti-rat response, but tolerance induced against the heterologous Ig by the concurrent administration of anti-CD4 resulted in prevention of autoantibody production, reduced proteinuria and prolonged survival.[50] Blockade of the IL6R by a neutralising mAb to IL6R was also reported to be beneficial in MRL-*lpr* mice.[51]

Reduced levels of IL4 have been found in MRL-*lpr* and (NZBxNZW)F_1 mice, resulting in an increased IFNγ to IL4 ratio.[30] Interestingly, in contrast with the expected paradigm of systemic autoimmunity being causally related to T_H2 response, GN development was completely abrogated in (NZWxC57BL/6.Yaa) F_1 mice rendered transgenic for the IL4 gene under the control of the IgH enhancer,[52] and this protection was associated with retention of overall titres of IgG anti-DNA autoantibodies, but a severe reduction in the nephritogenic IgG3 isotype. Others, in contrast, found that transfer of IL4 (or IL12) stimulated splenocytes from 5 month old (NZBxNZW)F_1 mice into syngeneic recipients increased the production of IgG anti-dsDNA antibodies, while administration of anti-IL4 before disease onset inhibited this production.[44] Moreover, anti-IL4 treatment alone prevented GN, while anti-IL12 alone was ineffectual. It is noteworthy that combined treatment with both antibodies abrogated the beneficial effect of anti-IL4.[44] Finally, IL4 gene deletion[34] as well as recombinant mouse IL4R or anti-IL4 mAb treatments[53] led to significantly reduced lymphadenopathy and end organ disease in MRL-*lpr* mice. BXSB male mice congenic for the IL4 deletion, however, develop disease equally as severe as that seen in the wild type mice (D Kono, A N Theofilopoulos, in preparation), indicating that this cytokine is not an obligatory participant in the autoimmune process.

All lupus strains also show increased IL10 levels,[29] and (NZBxNZW)F_1 mice repeatedly injected (from birth) with anti-IL10 mAb showed substantially delayed onset of autoimmunity,[54] an effect apparently mediated by upregulation of endogenous TNFα, as the anti-IL10 protection was abolished when anti-TNFα was introduced along with the anti-IL10 treatment. Conversely, administration of IL10 accelerated the onset of autoimmunity in these mice, but similar injections into normal mice had no adverse effects. The important role of this cytokine in lupus has also been suggested by the reduced autoantibody levels and kidney disease in (NZBxNZW)F_1 mice treated with an IL10 inhibiting immunomodulator (AS101),[55] as well as by increased Ig and autoantibody production in IL10 treated cultured peripheral blood lymphocytes (PBL) from systemic lupus erythematous (SLE) patients, and inhibition of these manifestations in anti-IL10 treated SCID mice transplanted with PBL from such patients.[56] The inference has been made that IL10 promotes systemic autoimmunity by increasing Fas/FasL mediated apoptosis.[57]

Other cytokines

Additional cytokines have been implicated in lupus pathogenesis, including TNFα, IL1 and TGFβ. TNFα, (a product of macrophages, T_H1 and T_H2 cells as well as B cells) exerts wide ranging effects on immune responses and inflammation and, therefore, its role in autoimmunity has been extensively investigated. It was initially shown by Jacob and McDevitt[58] that LPS activated macrophages of NZW mice produce 5-fold to 10-fold lower levels of TNFα than activated macrophages from non-autoimmune strains, and this low production was associated with a unique BamHI RFLP in

the NZW TNFα gene. Studies by these and other investigators, however, showed that the same RFLP defined allele was present in MRL and BXSB lupus strains as well as several normal strains and wild mice that produced intermediate to high levels of TNFα.[58 59] More recent studies by Jacob and associates[60] identified diverse mutations in the 5' and 3' untranslated region (UTR) of the TNFα gene in several strains of mice. Among them, a three base pair insertion disrupting the AU rich motif of the 3'UTR was detectable in TNFα low producing NZW, B10.KPA44, SM/J strains, and in *Mus spretus*. Transient expression experiments in a macrophage cell line using the luciferase reporter system indicated that presence of the NZW 3'-UTR, indeed, leads to reduced TNFα production.[61]

Obviously low TNFα production is not required nor sufficient for induction of lupus. This is the case even in the New Zealand mice, as documented by the presence of delayed but still severe disease in (NZBxNZW.PL)F_1 mice in which the TNFα haplotype is d/d—that is, without the NZW low TNFα defect.[62] Moreover, expression of TNFα is increased in the diseased kidneys of (NZBxNZW)F_1[63 64] as well as MRL-*lpr* mice.[65 66] In the latter strain, a biphasic increase in circulating TNFα was recorded with an initial peak in neonatal mice 703±208 pg/ml followed by normalised levels by 2 months of age 87±13 pg/ml and progressive increases thereafter that were proportional to the severity of renal disease (non-proteinuric, 570±87; proteinuric, 1255±135 pg/ml).[67] While early in life TNFα was detected only in tubular epithelial cells (TEC), in adult MRL-*lpr* mice expression was more ubiquitous and was detected in glomeruli, perivascular infiltrating cells as well as TEC.[67] Although these findings firmly establish the high in vivo TNFα levels in MRL-*lpr* mice, it should be noted that Beller and associates[40 68] observed defective in vitro induction of TNFα in LPS activated macrophages of the congenic non-Fas defective, mild disease manifesting MRL-+/+ mouse. This defect was noted in other lupus strains as well, and was thought to be the basis for defects in in vitro induction of other cytokines, such as IL1 and IL6. Notwithstanding these in vitro findings, the relevance of which is unclear, the fact remains that, in contrast with the NZW and (NZBxNZW)F_1 mice, other lupus strains are characterised by increased TNFα levels. Several studies have also shown that human lupus is characterised by high serum levels of TNFα and soluble TNFR that parallel disease activity.[69–72]

Regardless of variances in TNFα levels, the initial finding of low TNFα production by macrophages of NZW mice prompted Jacob and associates to assess its possible therapeutic effects in the (NZBxNZW)F_1 mouse.[58 73] They found that TNFα replacement therapy at relatively high doses (10 μg/3 × weekly) started up to 4 months of age effectively delayed disease, but was ineffective when started at 6.5 months of age (near the 50% mortality range for these mice). Gordon *et al*,[74] using a similar injection and dose schedule, also reported that TNFα treatment started after the onset of clinical disease led to improved survival relative to controls (92% survival rate in treated versus 42% of control at 10 months of age). They further indicated that while administration of TNFα delayed disease progression, sustained treatment did not prevent the eventual development of severe renal disease. As noted earlier, an additional finding by Ishida *et al*[54] was that sustained anti-IL10 treatment of (NZBxNZW)F_1 mice was beneficial, and that this effect was mediated by the concurrent TNFα upregulation, as simultaneous administration of anti-TNFα antibody abolished the anti-IL10 antibody effect. Overall, the above findings indicated clearly that TNFα replacement therapy is beneficial to the (NZBxW)F_1 mouse.

Additional findings, however, have indicated that the picture is much more complicated than was originally thought. Thus, Kelly and associates,[63] prompted by their finding of increased TNFα expression in the diseased kidneys of the (NZBxW)F_1 mouse, revisited the issue of TNFα therapy for lupus and reported that (NZBxNZW)F_1 female mice treated from 4 months of age with a low dose of recombinant TNFα (0.2 μg/3 × weekly) exhibited accelerated disease, while those receiving an intermediate dose (2 μg/3 × weekly) showed minor or no acceleration. Yet when the low dose treatment was started at 2 months and continued up to 4 months of age, there was no acceleration of renal disease. This latter finding was interpreted to indicate that for TNFα to cause renal injury, it must interact with other pathological features in these animals that appear after 4 months of age. Additional findings have provided further support for the notion that TNFα may, under certain circumstances, exert detrimental effects in lupus. Firstly, treatment of MRL-*lpr* mice with an IgG anti-TNFα antibody was reported to prevent development of pulmonary inflammatory lesions such as lung fibrosis and alveolitis.[75] Secondly, mice rendered defective for the *zfp36* gene, which encodes tristetraprolin, develop systemic autoimmunity apparently mediated by an attendant upregulation of TNFα, as antibodies to TNFα abrogated this syndrome[76]; tristetraprolin was recently shown to be a component of a negative feedback loop that interferes with TNFα production by destabilising its mRNA.[77] Thirdly, treatment with soluble, dimeric TNFR led to reduction of disease in systemic autoimmunity manifesting motheaten mice,[78] and treatment with a novel transcriptional inhibitor of TNFα reduced superantigen induced inflammatory arthritis in MRL-*lpr* mice.[79]

How can one reconcile the apparent dichotomous results reviewed above in which some studies reported beneficial and others detrimental effects of TNFα in murine lupus? On one hand, it could be argued that low doses of TNFα or short duration of treatment may promote autorecognition by, for example, upregulating MHC[80 81] or by increasing membrane expression of autoantigens.[82] In contrast, high doses or chronic exposure to TNFα may

inhibit autoimmunity by a variety of specific and non-specific immunosuppressive effects. With regard to non-specific immunosuppressive effects, studies by Gordon and Wofsy[83] have shown that mice treated with high doses of TNFα exhibit severe lymphopenia, while, with regard to specific effects studies by Cope *et al*[84 85] in TCR transgenic animals showed that prolonged exposure to TNFα caused severe suppression in a broad range of T cell responses, including proliferation and cytokine production. The possible relevance of this immunosuppression has been suggested by the finding that a small, but significant, percentage (6–7%) of RA patients treated with anti-TNFα mAb developed anti-dsDNA autoantibodies.[86] Diverse effects of TNFα are obviously not confined to lupus, but seem to apply to other autoimmune disorders. Thus, TNFα treatment has been shown to inhibit or promote diabetes in NOD mice depending on the time of treatment initiation,[81] while anti-TNFα mAb treatment[81] as well as high transgenic expression of soluble TNFRp55-FcIgG3 fusion molecules[87 88] have been shown to avert disease. Apparently, because of reported beneficial and detrimental effects of TNFα in autoimmunity, terms such as "Trojan horse"[89] and "pretty girl or old witch"[90] have been coined to illustrate the unpredictable effects of this molecule.

Increased IL1 levels have been described for all lupus strains.[29 91 92] Recombinant IL1 given to (NZBxNZW)F_1 mice increased nephritis,[63] while recombinant IL1R given to MRL-*lpr* mice inhibited GN, splenomegaly/lymphadenopathy and autoantibody levels in one study,[93] while in another study, an IL1R antagonist had no effect.[94] Surprisingly, in vitro experiments showed that IL1 and IL1R antagonists both induced significant suppression in IgG production by B cells derived from diseased MRL-*lpr* mice, but they had no effect on B cells from young animals.[92 95 96]

Male BXSB and MRL-*lpr* mice show increased levels of TGFβ,[29] which was shown in MRL-*lpr* mice to adversely affect host defence against both Gram negative and positive bacterial infections because of the failure of initial polymorphonuclear leucocyte migration to the infection site.[97 98] The role of increased TGFβ levels in this defect was directly demonstrated by the fact that this abnormality was duplicated in MRL-+/+ mice injected with TGFβ at the time of bacterial infection, and lethality of infected MRL-*lpr* mice was ameliorated by administration of anti-TGFβ mAb.[97] Such findings suggest that increased TGFβ production in lupus may suppress host defence mechanisms against bacterial infection, thereby providing an explanation for the increased risk of such infections in SLE patients. Nevertheless, direct injections into the skeletal muscle of a TGFβ cDNA expression vector was reported to reduce autoantibody levels in MRL-*lpr* mice,[19] while infection with a non-pathogenic strain of *Salmonella typhimurium* carrying the TGFβ gene was without effect.[18]

Conclusions

The summarised findings on the role of cytokines in murine lupus cast doubt on the widely held view that T_H2 cytokines play the primary part in this humorally mediated autoimmune disease. It would appear that both T_H1 and T_H2 cytokines are involved, and that agonists and antagonists can disturb these cross talking classes of cytokines to exert disease inhibiting or promoting effects without any dogmatic predictability and in a far more complex manner than the simple T_H1 versus T_H2 dualism dictates. It is also evident that the effects of a given cytokine agonist or antagonist in the lupus models frequently differ from one study to another. These differences can be accounted for on the basis of variables in study design, including administration, dose, and timing of treatment initiation. Therefore, caution should be exercised in generalisations derived from one or another experiment wherein beneficial or detrimental effects are observed. Obviously, further work is needed before we fully comprehend the complex interplay between these molecules and confidently design cytokine treatments for human lupus.

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