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REVIEW

Genetics of susceptibility to leprosy

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The ancient disease of leprosy can cause severe disability and disfigurement and is still a major health concern in many parts of the world. Only a subset of those individuals exposed to the pathogen will go on to develop clinical disease and there is a broad clinical spectrum amongst leprosy sufferers. The outcome of infection is in part due to host genes that influence control of the initial infection and the host's immune response to that infection. Identification of the host genes that influence host susceptibility/resistance will enable a greater understanding of disease pathogenesis. In turn, this should facilitate development of more effective therapeutics and vaccines. So far at least a dozen genes have been implicated in leprosy susceptibility and a genome-wide linkage study has lead to the identification of at least one positional candidate. These findings are reviewed here.
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Introduction

Many think of leprosy as a disease of the past and it is true that the worldwide prevalence is declining, in part due to improved case detection and effective multi-drug therapy. Globally, however, there are still 700 000 newly detected cases each year, mainly in Africa, Asia and Latin America[1] and leprosy represents a major health problem in Brazil, India, Madagascar, Myanmar, Nepal and Mozambique. The causative agent of leprosy, *Mycobacterium leprae*, was identified by Armauer Hansen in 1873. It is an obligate intracellular pathogen that mainly infects macrophages and Schwann cells, though it also multiplies in muscles and vascular endothelium, and can infect other tissue such as the brain, eye and testis. Unlike tuberculosis, which also results from infection with a mycobacterium (*M. tuberculosis*), leprosy is not often a direct killer. Instead, due to the infective agent's predilection for skin and peripheral nerves, the common severe consequences of leprosy are deformity and disability. This has significant social and economic impact on both the patient and their community.

Leprosy is like tuberculosis in that in the majority of cases, infection does not lead to clinical disease and when disease does develop, much of the damage is not caused by the infecting organism, but rather by the host's immune responses to that organism. Host factors that influence control of the initial infection and the host's immune response play a significant role in the outcome of infection with either *M. tuberculosis* or *M. leprae*.

In leprosy the significance of the host response to infection is illustrated by the broad clinical spectrum observed amongst those that develop disease. At one pole is tuberculoid leprosy, characterized by strong cell-mediated immunity, a Th1 CD4+ cytokine profile (IL2, IFN-γ), very few bacteria and localized lesions. At the other pole is lepromatous leprosy, characterized by a lack of cell-mediated immunity, Th2 CD4+ responses (IL4 and IL5), a strong humoral response, disseminated progressive disease and large numbers of bacteria. Thus tuberculoid patients can be thought of as those exhibiting the most resistance whereas lepromatous patients are those exhibiting the least. This is not to say that the pathogenesis associated with the more resistant pole is necessarily milder; strong Th1 responses that contain the bacterium can result in rapid and severe nerve damage.

Evidence that host genetic factors contribute to susceptibility to leprosy comes from epidemiological data, segregation and twin studies.[2–6] The goal of identification of host genetic factors that underlie susceptibility to leprosy can be approached in at least two ways: firstly, candidate gene studies can be carried out on genes of known function that have a possible biological role in the control of infection or disease. A second approach utilizes a non-targeted genome-wide linkage analysis, in which increased sharing of chromosomal regions by affected individuals leads to identification of positional candidates. How *M. leprae* targets Schwann cells, exactly how it brings about nerve damage and how it is killed in macrophages have not been fully elucidated and it is possible that host genes which influence these processes may influence the outcome of infection. It is clear, however, that the development of appropriate cell-mediated immunity is important in the control of mycobacterial disease. Several genes that may modulate cell-mediated immunity have been investigated and some appear to have a role in either susceptibility to leprosy *per se*, or to leprosy type (Table 1). These genes and the results of a genome-wide linkage analysis of leprosy susceptibility are discussed below.

Major histocompatibility complex (MHC) region

Linkage and/or association studies have implicated several genes in the MHC region in susceptibility to leprosy. However the presence of strong linkage

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Table 1 Summary of published non-HLA leprosy association studies

Gene(s)	*Population*	*Polymorphisms typed*	*Phenotype*	*Allele/genotype/haplotype*	*Study size*	*Ref.*
COL3A1	India	COL3A1	MB[a]	COL3A1 250 bp susc.[b]	16 PB[d]	132
CTLA4		CTLA4 size allomorphs		CTLA4 104 bp pro.[c]	9 MB 14 con.[e]	
SLC11A1	India	Promoter microsatellite, Exon 2, 469+14G/C, $(TGTG)_n$	Leprosy	No association	124 MB 107 PB 166 con.	128
SLC11A1	Mali	Promoter microsatellite, 469+14G/C, $(TGTG)_n$	Leprosy type	TGTG het.[f]	181 MB 92 PB 201 con.	129
TLR2	Korea	R 677W	MB	677W	45 MB 41 PB 45 con.	160
C4B	Brazil	C4B, C2, BF, C4A	ENL[g]	C4B*Q0	109 cases 46 ENL 172 con.	82
HSPA1A	India	HSPA1A	PB	A	49 PB 38 con.	88
MICA/HLA B haplotype	South China	HLA-DRB1, HLA B, MICA-5A	MB	HLA-B46/MICA-5A5	50 MB 19 PB 112 con.	21
TAP2	North India	TAP1: I333V, N637G, TAP2: V379L, A565T, A665T	PB	TAP2-B	50 PB 40 con.	58
TNFα	Bengali Indians	−308	MB	TNF*2	121 MB 107 PB 160 con.	42
TNFα	Brazil	−308	Mitsuda response	TNF*2	74 PB	75
TNFα	Brazil	−308	Reaction rate	TNF*2 het.	57 TNF*2 positive	79
TNFα	Brazil	−308	MB	TNF*2	90 PB 210 MB 92 con.	74
TNFα	Brazil	−308	Leprosy	TNF*1	T[h] 27 NT[i] 4	32
TNFα/LTA haplotype	Brazil	TNF -308, LTA *Nco*1	Leprosy	TNF*1/LTA*2 susc.	T 26 NT 11	32
				TNF*2/LTA*2 pro.	T 2 NT 16	
VDR	Bengali Indians	*Taq*1	Leprosy type	tt in PB TT in MB	124 MB 107 PB 166 con.	128

[a]multibacillary (includes patients at/near lepromatous pole).
[b]susceptible.
[c]protective.
[d]paucibacillary (includes patients at/near tuberculoid pole).
[e]controls.
[f]heterozygotes.
[g]Erythema Nodosum Leprosum.
[h]Transmitted.
[i]Not Transmitted.

disequilibrium in combination with a relatively large number of closely spaced, polymorphic genes encoding products involved in immune responses has made the task of determining true functional associations a difficult one. The most consistent results indicate that a locus in the class II region is involved in susceptibility to leprosy *per se* and/or modulation of disease.

HLA genes

Located in the MHC region, HLA genes are highly polymorphic and present antigenic peptides to αβ T cells. The HLA genes were one of the first class of genes to be implicated in susceptibility to leprosy, and there have been numerous studies in many populations, probably partly because their gene products were such a likely biological candidate (the main hypothesis being that

presentation of certain antigens in a specific HLA context may influence the type of T-cell response that develops), and partly because serological typings could be performed in the pre-genotyping era.

A series of family studies in various populations have shown non-random segregation of parental HLA haplotypes amongst tuberculoid children[7–11] and lepromatous children.[10,12] As HLA haplotypes segregated randomly among healthy siblings[8,10] it was suggested early on that genes in the MHC region might influence leprosy type rather than susceptibility to leprosy *per se*.[13]

Class I region

Several studies comparing HLA class I gene frequencies in leprosy cases and controls have found associations either with the polar forms of leprosy, or with leprosy itself, however these suggested associations have not been replicated.[14–20]

Recently an HLA-B46, MICA-5A5 haplotype was found less often amongst multibacillary leprosy patients compared to controls in a small south China study, indicating that this haplotype may carry a gene that is protective against multibacillary leprosy in this population.[21] MICA itself is a candidate susceptibility locus: located in the HLA class I region, MHC class 1 chain-related (MIC) genes encode membrane-bound polypeptides that do not bind peptides or associate with β2-microglobulin, but instead act as a co-stimulatory signal by interaction with a receptor NKG2D to augment T-cell activation. MICA cell surface expression can be up-regulated not only by heat shock but also by infection with cytomegalovirus (CMV)[22] or *M. tuberculosis*.[23] The Chinese study found no association with leprosy and B46−/5A5+ or B46+/5A5− haplotypes indicating that neither the HLA B46 nor the MICA-5A5 allele was sufficient to confer protection. The MICA-5A5 polymorphism is located in exon 5 and is a nucleotide triplet repeat that encodes a series of alanine residues in the transmembrane domain. There are numerous other polymorphisms in the MICA gene, including many in exons 2–4, which encode the extracellular domains that bind to NKG2D[24,25] and it is possible that one that influences susceptibility to leprosy occurs on the B46/MICA-5A5 haplotype.

Class II region

Numerous case–control studies have shown associations between the class II HLA genes and leprosy. DQ alleles, especially DQw1 have been shown to be associated with tuberculoid leprosy in India,[26,27] Korea,[19] Thailand[28] and Japan,[29] and with lepromatous leprosy in India[15] and Japan.[18,30] DQw1 was also implicated in a meta-analysis of pooled association studies[31] and in a Venezuelan lepromatous family study[10] whereas in Shaw's recent Brazilian family study significant evidence for linkage and association to tuberculoid leprosy and possibly leprosy *per se* was found with DQB1, DQA1 and DRB1 variants.[32] Unusual DQ restricted antigen-specific CD8 cells that predominantly produce IL4 were proposed to act as suppressor cells in lepromatous patients;[33] however, this does not explain the association with tuberculoid leprosy. Importantly, HLA-DQ1 is in strong linkage disequilibrium with HLA-DR2 in most populations and it has usually been difficult to discern whether the primary association is with the DR or the DQ variant.

HLA-DR3 was associated with tuberculoid leprosy in Mexico[34] and in the South American populations of Surinam[35] and Venezuela,[10] whereas it was found at lower frequencies in lepromatous patients.[35,36] T-cell proliferation responses to certain epitopes of the mycobacterial hsp65 (p3–13) are DR3 restricted[37] and DR3 was associated with low T-cell responses to *M. leprae* in tuberculoid patients.[38,39] Increased HLA-DR2 frequencies have been reported in both tuberculoid and lepromatous patients compared to controls.[15,18,19,28,29,31,40,41] Association studies have also shown that DR2 is increased in leprosy patients of all subtypes compared to controls.[42,43] Family-based studies have shown a skewed distribution of DR2 alleles in Indian[44] and Egyptian tuberculoid siblings.[11] Molecular HLA typing has allowed DR2 to be refined into HLA-DRB1 subtypes. DRB1*1501 has been associated with lepromatous leprosy[26,30] whereas DRB1*1502 has been associated with tuberculoid leprosy.[45] The *1501 and *1502 alleles differ from each other by a single amino acid at codon 86. Class II molecules have polymorphic pockets that accommodate the side chains of bound peptides. The codon 86 residue lies in binding pocket 1. In another Indian study, both 1501 and 1502 were found to be associated with tuberculoid leprosy,[46] indicating that the residue in pocket 1 may not be involved in determining the outcome of leprosy infection. Instead it appears that certain residues that contribute to the net charge in the putative peptide-specific binding pocket 4 may be more important.[47] It is hypothesized that net negative or neutral charges in binding pocket 4 cause poor binding of the DRB1 molecule to *M. leprae* antigens. As HLA molecules with the highest affinity to peptide produce the greatest T-cell proliferation and IFN-γ response,[48] peptide presentation by low affinity class II molecules may result in muted cell-mediated immunity.[47] Alternatively, peptide presentation by specific class II molecules may result in activation of suppressor/regulatory T-cells.[49]

Transporter 2, ATP-binding cassette, sub-family B (TAP2). TAP2 is critical for peptide translocation from the cytosol into the endoplasmic reticulum and for peptide binding to MHC class I molecules. Various combinations of polymorphisms at codons 379, 565 and 665 result in eight TAP2 alleles: (A–H). In the rat, products of different TAP2 alleles differ in permissiveness to transport peptides into the endoplasmic reticulum, thus modifying the spectrum of peptides presented by class I molecules on the cell surface.[50] Although some immune responses to infectious pathogens and autoimmune diseases are apparently independently associated with human TAP2 alleles,[51–55] it is unclear whether differences in human TAP2 alleles are functionally relevant.[56,57] Nevertheless, TAP2-B has been reported to be associated with tuberculoid leprosy and TAP2-A/F was increased in pulmonary tuberculosis patients in North India,[58] indicating that TAP2 alleles may influence mycobacterial susceptibility in this population.

Class III region

Tumor necrosis factor alpha (TNFα). The TNFα protein has pleiotropic effects which straddle the innate and adaptive immune responses. Produced mainly by macrophages, in mice it is involved in macrophage activation

and, possibly, killing of intracellular *M. leprae* as well as efficient antigen presentation through the class II molecules. It is also an important modulator of the cytokine production required for effective leukocyte localization and thus granuloma formation. One hypothesis for how TNFα may influence infectious disease in humans is that polymorphisms leading to low production could result in insufficient activation of macrophages, such that they are unable to kill mycobacteria. Alternatively, the important role TNFα plays in granuloma formation may mean that low TNFα levels result in a failure to contain infectious foci. On the other hand, TNFα overproduction may influence disease progression by inducing local tissue damage. Finally, a recent study has found that TNFα production supports virulent *M. tuberculosis* growth in human alveolar macrophages, leading the authors to suggest that mycobacteria may deliberately augment TNFα production as a method of immune invasion, for example by inducing apoptosis of infected cells and thus enabling their spread to uninfected macrophages.[59]

There are numerous polymorphisms in the TNFα promoter, several of which may be involved in regulation of TNFα expression. A single nucleotide polymorphism at −863 affects the binding of the NF kappa B p50-p50 dimer to an NF kappa B regulatory site.[60] The base change appears to inhibit p50–p50 binding, thereby reducing p50–p50 repression of reporter gene transcription. Another promoter polymorphism, −376, lies in a region of multiple DNA–protein interactions. The G to A change introduces a binding site for the transcription factor OCT-1. Recruitment of OCT-1 results in an increase in basal TNFα expression in human monocytes and the A allele has been associated with susceptibility to cerebral malaria.[61] The only TNFα polymorphism that has been investigated in relation to leprosy susceptibility is a G to A substitution located in the promoter, at position -308. The alleles have been designated TNF*1 and TNF*2, for G and A, respectively. Initial studies using TNFα promoter reporter constructs suggested the TNF*2 allele is associated with increased TNFα levels[62–64] but this has been disputed by others.[65–68] Recently, it was shown that elevated TNFα levels are produced by TNF*2 only when certain cell types and stimuli are used.[64] Consistent with the hypothesis that the −308 polymorphism is functional, the TNF*2 allele has been associated with a variety of diseases in which excessive TNFα production has been postulated to be involved, including fatal meningococcal disease,[69] mucocutaneous leishmaniasis,[70] scarring trachoma,[71] cerebral malaria[72] and inflammatory bowel disease.[73]

The TNF*2 allele has been associated with lepromatous leprosy in Bengali Indians[42] whereas in southern Brazil this allele appears to be protective against severe leprosy.[74] TNF*2 also appears to be protective against lepromatous leprosy, tuberculoid leprosy and leprosy *per se* in northeastern Brazil, while TNF*1 confers susceptibility.[32] Furthermore, Brazilian leprosy patients that carry the TNF*2 allele have greater skin inflammatory responses to lepromin than those that do not;[75] thus, in Brazil it appears that increased production of TNFα may be important for inducing protective immune responses against leprosy.

TNFα levels have been shown to be increased during reaction responses in leprosy patients.[76,77] Reaction responses are the cause of significant tissue damage and in combination with transforming growth factor β1, TNFα causes significant Schwann cell death *in vitro*.[78] Thus it has been hypothesized that alleles that enhance TNFα production may be associated with the adverse effects of reaction responses. In an analysis of 57 TNF*2-positive leprosy patients, reaction responses were found much more frequently amongst heterozygotes compared to homozygotes; however, TNFα levels were similar for a small series of both TNF*1 and TNF*2 patients during and in the absence of reaction, and both sets of patients showed similar increases in TNFα levels during reaction, indicating that these polymorphisms may not influence TNFα levels *in vivo*.[79] It would be of interest to reassess these associations in a larger data set.

Lymphotoxin α (LTA, formerly TNFβ). The LTA gene is located close to TNFα and encodes lymphotoxin α, a chemokine secreted by lymphocytes and natural killer cells. Soluble lymphotoxin α homotrimers can bind to the same receptors as TNFα whereas heterotrimers formed with membrane-bound lymphotoxin β interact with the lymphotoxin β receptor. Through these receptors, lymphotoxin α exerts pleiotropic immunomodulatory effects. Recently, several loci in the HLA class II region including LTA were linked to susceptibility to leprosy in Brazil.[32] A G to A substitution abolishes an *Nco*I restriction site in intron 1 of LTA. This RFLP was not found to be associated to leprosy susceptibility on its own, however, two locus transmission disequilibrium testing indicated that the haplotype TNF*1/LTA*2 was associated with susceptibility while TNF*2/LTA*2 was associated with protection. As no transmission distortion was observed for the haplotype TNF*1/LTA*1, these data suggest that another polymorphism that occurs on a background of LTA*2 may have a role in leprosy susceptibility. Polymorphisms that occur within the LTA gene and its regulatory regions[80,81] are potential candidates.

Complement component 4B (C4B). Erythema Nodosum Leprosum (ENL) is an immune reaction which can occur in lepromatous leprosy patients and is postulated to be immune complex mediated. C4B is a class III gene which encodes the complement component 4B. The protein is involved in opsonization of pathogens and immune complex clearance. Non-expressed C4B alleles (C4B*Q0) were found to be associated with lepromatous leprosy, and especially ENL in Brazil.[82] The possibility of linkage disequilibrium with other MHC loci was not examined.

Heat shock 70kD protein 1A (HSPA1A). HSPA1A is also located in the class III region of the MHC, between the TNF and C2 genes. One of three 70 kD heat shock proteins encoded in this region, HSPA1A is expressed constitutively at a low level and its expression is increased in response to thermal stress. Antigenic peptides can associate with HSP70s, leading to their uptake and presentation by antigen presenting cells.[83] Furthermore, HSP70 activates macrophages by induction of pro-inflammatory cytokine production via the Myd88/NF kappa B pathway.[84] This HSP70 response is mediated by TLR2 and TLR4, and indeed TLR2 and TLR4 may act synergistically, in a CD14-dependant fashion, to activate NF kappa B.[85] Another role of HSP70s

is to block decay of the AU-rich mRNA of cytokines and proto-oncogenes.[86] Three HSPA1A alleles, A–C, result from nucleotide substitutions at two sites (−110 and +120). The −110 position lies adjacent to a heat shock transcription factor binding site[87] and thus may affect the inducibility of HSPA1A. The HSPA1A-A allele was found to be associated with tuberculoid leprosy in a small North Indian study.[88] In Italians, the HSPA1A promoter polymorphisms were associated with specific HLA class II haplotypes[87,89] including those carrying HLA DR3; however, in North India the HSPA1A association appears to be independent of both class I or II loci, indicating that HSPA1A may play a role in susceptibility to leprosy in this population.

Other candidates

Solute carrier family 11 member 1 (SLC11A1, formerly NRAMP1). Located at 2q35, SLC11A1 is the human homologue of the mouse gene Slc11a1. Mice with a naturally occurring Gly169Asp mutation are susceptible to a range of intracellular pathogens including *Leishmania donovani*, *Samonella typhimurium*, some strains of *M. bovis*,[90] *M. lepraemurium*,[91,92] *M. intracellulare*,[93] *Toxoplasma gondii*,[94] *Candida albicans*[95] and *Leishmania infantum*,[96] but probably not *M. tuberculosis*.[97–99] This substitution of a charged amino acid in one of the putative transmembrane domains may cause misfolding of the Slc11a1 protein, resulting in it being targeted for degradation at the endoplasmic reticulum. Null mutants have the same susceptibility phenotypes as those mice carrying the naturally occurring point mutation[90] indicating that the point mutation results in a completely non-functional gene product.

The biochemical function of the SLC11A1/Slc11a1 gene product has not been completely elucidated and indeed is the subject of some controversy.[100] The mouse Slc11a1 encodes an integral membrane protein expressed in late endosomal and lysosomal membranes of macrophages. On phagocytosis it is relocated to the phagosomal membrane where it is thought to act as a divalent cation pump. The controversy arises over the direction in which the cations are pumped. One theory is that resistance is mediated by pumping iron out of the phagosome, thus restricting the cations available to the pathogen.[101] The alternative theory is that the iron is pumped into the phagosome, where it is used as a catalyst in the Fenton/Haber Weiss reduction of superoxide anions to generate toxic hydroxyl radicals.[102] In either case, the Slc11a1 gene product seems to have a direct antimicrobial effect, but it also has pleiotropic effects that include macrophage activation and regulation of the Th1:Th2 balance of the adaptive immune response to intracellular pathogens.

Macrophages containing the mutant Slc11a1 have a defect in antigen processing for presentation to T-cells[103] possibly due to a metal ion requirement for metalloprotease activity and/or endosomal fusion events. In addition, Slc11a1 appears to influence MHC class II molecules and cytokines which regulate antigen presentation, such as TNFα and IL1β.[102] Such effects may account for the polarity of Th1 *vs* Th2 responses observed in mice with wild-type Slc11a1 compared to those with the mutant gene after infection with *L. donovani*[104] or exposure to typhoid toxin.[105]

Human susceptibility to infection may in part be determined by the direct influence of SLC11A1 on antimicrobial activity of macrophages; however, the reported pleiotropic effects in regulating Th1:Th2 balance in the immune response may also contribute. The mouse Gly169Asp mutation is unknown in the human homologue, but 11 other sequence variants have been described.[106–108] These include two missence coding changes and a promoter microsatellite reported to affect NRAMP1 expression levels.[109] Variants in SLC11A1, especially the higher expressing promoter microsatellite allele 3, have been reported to be associated with rheumatoid arthritis,[110–112] Crohn's disease,[113,114] type 1 diabetes[115] and sarcoidosis.[116] However, these studies have often been of limited size and some associations have not been replicated in other populations. The promoter microsatellite has also been associated with susceptibility to various infectious diseases including HIV in Colombia (allele 2),[117] visceral leishmaniasis, post kala-azar dermal leishmaniasis in the Sudan and severe meningococcal meningitis (allele 3).[118] Interestingly, although Slc11a1 is not important in the control of *M. tuberculosis* infection in mice,[97] SLC11A1 has been associated with susceptibility to tuberculosis in several human populations including Japanese,[119] Brazilians,[120] Koreans[121] and West Africans.[122,123] In a large Canadian Aboriginal family[124] linkage was found to the chromosomal region containing SLC11A1 but linkage is not found in most studies.[125]

The relevance of SLC11A1 to leprosy susceptibility is less clear-cut. Non-random haplotype segregation in 20 South East Asian leprosy pedigrees implicated SLC11A1 in leprosy susceptibility; however, as haplotype sharing was more pronounced in 16 Vietnamese families compared to four Chinese families, the possibility of ethnic heterogeneity was suggested.[126] Such ethnic heterogeneity may explain why studies in other populations have failed to find SLC11A1 linkage or association with leprosy susceptibility *per se*.[127–130] An association with leprosy type was shown in Mali, where heterozygotes for a TGTG insertion/deletion in the 3′ untranslated region were more common amongst multibacillary cases than in paucibacillary cases.[129] The putative functional microsatellite was also typed in the Mali study but no associations were found with its alleles. It is not known whether the TGTG polymorphism has a functional effect.

Recently, the SLC11A1 region was found to be in linkage with the Mitsuda response among both healthy and affected members of 20 South East Asian leprosy families.[131] The Mitsuda response is a measure of the size of the reaction obtained 28–30 days after intradermal injection of *M. leprae* antigen. It is regarded as a measure of the *M. leprae* induced granuloma-forming capacity. Thus, this finding is in agreement with the hypothesis that, in addition to, or instead of being important in control of mycobacterial infection at the macrophage level, SLC11A1 may play a role in the development of acquired antimycobacterial immune responses in humans.

COL3A1 and CTLA4. In a very small study, polymorphisms in the COL3A1 (procollagen III alpha I) and CTLA4 (cytotoxic T lymphocyte associated antigen) genes located at 2q31–33 were found to be associated with

leprosy.[132] The dinucleotide repeat examined in CTLA4 occurs in the 3′UTR, and at first alleles were postulated to effect mRNA stability but recently strong linkage disequilibrium has been shown between the dinucleotide repeat alleles and known functional polymorphism located in the promoter and exon 1.[133] Interestingly, the allele associated with protection against leprosy in India also displayed increased transmission to IDDM siblings in Russian families.[134] Indeed markers in this region have been linked and associated with several autoimmune diseases.[135] As CTLA4 is an important negative regulator of T-cell activation it is a plausible biological candidate for leprosy susceptibility; however, both COL3A and CTLA4 lie close to the genes encoding CD28 and ICOS, two other T-cell costimulatory receptors which are also likely candidates. To date, no other reports of associations between leprosy and any of these genes have been reported. Furthermore, it is just possible that the associations found could reflect an underlying linkage disequilibrium with alleles of the SLC11A1 gene at 2q35.

Vitamin D receptor (VDR). The vitamin D receptor is a nuclear hormone receptor encoded by the VDR gene (located at 12q12–q14). It is the mediator of the effects of the active form of vitamin D, 1α 25(OH)$_2$D3, which acts as a hormone. VDR contains several domains including a DNA-binding domain (DBD) consisting of two zinc fingers and a ligand-binding domain. On ligand binding, VDR is translocated to the nucleus and a conformational change occurs, allowing interaction with various coactivators. The DBD binds specifically to vitamin D response elements (VDRE) in the regulatory region of vitamin D responsive genes. A stable protein–DNA complex requires a homo- or hetero-dimeric complex with a second partner receptor such as the retinoid X receptor (RXR). This complex of nuclear receptors, vitamin D and coactivators can then act to modulate transcriptional activities. Genes with VDRE in their promoters include oestocalcin, hence the association of VDR polymorphisms with vitamin D resistant rickets, bone density and height in some studies.[136–138]

In addition to its role in calcium metabolism regulation, 1α 25(OH)$_2$ D3, acting through VDR, is a potent immunomodulator involved especially in suppression of inflammation. Some of its immunomodulatory effects may be mediated via VDRE; however, it appears that other complex cross-talk interactions are also involved. Recently, it has been shown that the DBD of VDR interacts with Stat1, the nuclear transcription factor of IFN-γ.[139] In addition to antagonizing the transcriptional activation of a specific VDRE-containing gene by preventing interaction between VDR and RXR, this interaction is proposed to prolong Stat1-mediated induction of IFN-γ-regulated genes in macrophages by protecting Stat1 from inactivation by tyrosine dephosphorylation.

Several cytokine genes including IL2,[140] GM-CSF[141] and IFN-γ[142] are direct targets of 1α 25(OH)$_2$ D3/VDR-mediated repression in activated T-cells. VDR/RXR also represses IL12 p40 expression in activated macrophages and dendritic cells.[143] IL12 is important in the development of a Th1 response and initiation of a cell-mediated response to pathogens. Through VDR, 1α 25(OH)$_2$ D3 inhibits fasL (CD95) mRNA production.[144] FasL mediates activation-induced programmed cell death in activated T lymphocytes, but it also induces maturation of dendritic cells, resulting in upregulation of MHC class II molecule expression and secretion of proinflammatory cytokines.[145] Thus through its receptor, the hormone 1α 25(OH)$_2$ D3 has pleiotropic anti-inflammatory effects that may inhibit the development of a protective Th1 response.

Several polymorphisms have been described in the VDR gene, including three neighboring RFLPs: *Bsm*I and *Apa*I, located in intron 8, and a silent T to C polymorphism in codon 352 that creates a *Taq*1 RFLP in exon 9. Exon 9 encodes part of the ligand-binding domain of VDR. The polymorphisms are designated Aa (*Apa*I), Bb (*Bsm*I) and Tt (*Taq*I), where the uppercase letter signifies absence of the restriction site and lowercase signifies the presence of the restriction site. The AA and BB genotypes have been associated with increased serum osteocalcin levels.[146] The B and t RFLPs were in strong linkage disequilibrium in Australian Caucasians, and the BB genotype correlated with bone mineral density in the same population.[136] In a luciferase reporter gene assay, haplotypes bearing BAt produced more activity than those bearing baT,[(136] although the functional significance of these variants, if any, is unclear.[147,148]

Nevertheless, linkage analysis identified VDR as a positional candidate for inflammatory bowel disease and the tt genotype was subsequently found to be associated with Crohn's disease in Caucasians.[149] In Bengali Indians, the tt genotype was associated with the more resistant form of leprosy, while TT was associated with lepromatous leprosy.[128] Likewise, in The Gambia, tt was found to be associated with resistance to tuberculosis.[150] Consistent with these findings, both 1α 25(OH)$_2$D3 deficiency and the combination of the presence of a T allele and 1α 25(OH)$_2$D3 deficiency were associated with tuberculosis susceptibility in Gujarati Indians living in London.[151] In contrast, susceptibility to pulmonary disease caused by infection with the opportunist *M. malmonese* was associated with increased frequencies of the t allele, the A allele and the At haplotype[152] and in Southern Indian females the tt genotype was associated with susceptibility to pulmonary tuberculosis.[153]

These opposing results are difficult to resolve if the silent *Taq*1 polymorphism does have a direct impact on the immunomodulatory function of VDR; therefore, it seems more likely that other polymorphisms (or combinations thereof) in linkage disequilibrium with the *Taq*1 RFLP may account for the associations with mycobacterial resistance and susceptibility described above. Ethnic differences in the linkage disequilibrium of some VDR polymorphisms have been described.[154] In addition to polymorphisms in the 3′UTR, an RFLP that results in structurally and functionally distinct VDR isoforms occurs in the first ATG start codon of VDR. This T to C substitution abolishes a *Fok*I restriction site, resulting in translation initiation at an in-frame start site three codons downstream. Allele designation follows that for the 3′ RFLPs; the f allele encodes 427 amino acids while the F allele encodes 424 amino acids. The F allele product interacts more efficiently with the human basal transcription factor IIB (TFIIB) and exhibits enhanced transcriptional activity.[155] In addition, this allele is associated with increased bone mineral density

whereas the f allele may be associated with protection against pulmonary disease caused by *M. malonese*.[152] As yet no studies investigating association between the *Fok*I RFLP and leprosy susceptibility have been reported.

Toll-like receptor 2 (TLR2). Toll-like receptors (TLRs) are pattern recognition receptors able to activate direct antimicrobial effector mechanisms in response to various microbial components. Also, activation of mammalian TLRs facilitates transcription of genes that regulate the adaptive response, including cytokines and co-stimulatory molecules. TLR2 is able to respond to many microbial components, including mycobacterial lipoproteins and lipoarabinomannan[156,157] and appears to be very important in generating a pro-inflammatory, protective immune response against mycobacteria in mice.[158] In both mice and humans, TLR2 activation by *M. tuberculosis* lipoprotein leads to killing of intracellular *M. tuberculosis*. Killing is nitric oxide (NO) dependant in mice, but in humans this process is NO independent.[159] Recently, 86 Korean leprosy patients were screened for polymorphisms in a highly conserved part of the TLR2 intracellular domain. A previously undescribed C to T substitution that results in an arginine to tryptophan change at the highly conserved amino acid 677 was found in 22% of the 45 lepromatous leprosy patients.[160] No other polymorphisms were found in the region and it was hypothesized that the substitution may affect the intracellular signaling of TLR2. Another polymorphism of a conserved C-terminal arginine in TLR2 reduces NF kappa B activation in response to Gram-positive bacterial peptides and may be associated with susceptibility to staphylococcal infection,[161] consistent with the hypothesis that TLR2 has an important role in the protection against bacterial infections in humans.

Mannose-binding lectin (protein C) 2 (MBL2) . Located at 10q11.2–q21, MBL2 encodes a calcium-dependant mannose-binding lectin (MBL, also called mannose-binding protein (MBP)) found in serum. MBL binds to arrays of terminal mannose groups on a variety of bacteria. Binding can initiate complement activation, and promote opsonophagocytosis independent of antibodies and Clq. Any one of three exon 1 point mutations (in codons 52, 54 and 57) in MBL2 reduces MBL serum concentrations, probably by interfering with the oligomerization of the protein. Low levels of MBL may be associated with recurrent infections in young children, yet low producing alleles are found with reasonable frequency in all populations, leading to the hypothesis that the disadvantage of increased infection susceptibility in infancy may be counterbalanced by unknown advantages of low MBL concentration.[162] As mycobacteria utilize phagocytosis to gain entry into host cells, it has been hypothesized that low MBL levels may be protective against mycobacterial infection by limiting this entry route.[163] In agreement with this hypothesis, increased MBL levels have been observed in active and fully recovered South African Cape Coloured tuberculosis patients,[164] Tanzanian tuberculosis patients[165] and in a small number of Ethiopian leprosy patients.[163] Exon 1 variants underlying serum MBL deficiency were found less commonly in Cape Coloured meningeal tuberculosis,[164] but not in Gambian pulmonary tuberculosis patients.[166] Intriguingly, it appears that pathogenic mycobacteria may encourage opsonization by production of a molecule that interacts with C2a to form a C3 convertase that results in deposition of C3b on the bacterial wall.[167] However, no associations have been reported between leprosy susceptibility and MBL2 variants, and in contradiction to the above reports, MBL2 variants were found with increased frequency in a limited Indian pulmonary tuberculosis study.[168]

Genome-wide linkage analysis

The development of high throughput genotyping technologies and the identification of thousands of polymorphic microsatellite markers have made genome-wide linkage studies possible. This approach has the advantage that no disease model or prior knowledge of the structure, function or location of the disease gene is required. The chromosomal regions identified by this approach will initially be much larger than that of an association study (several megabases compared to a few hundred kilobases); however; fine mapping and subsequent identification of the genes involved has been aided greatly by the release of the draft human genome sequence. The genome-wide approach also has the advantage that genes of unknown function and those not previously suspected as possible candidates can be identified.

To date, very few genome-wide linkage studies of infection or infectious diseases have been published. The first pathogen to be investigated in this way was *Schistosoma mansoni*. The parasite burden in 11 extended families from Brazil was examined and a region on chromosome 5 (5q31–33) was identified.[169] Further studies of *S. mansoni* and malaria parasite density also showed linkage to this region, which contains a cluster of genes encoding several Th2 cytokines.[170,171] Linkage to this same region of chromosome 5 has also been identified in a study of candidate regions for atopy and asthma.[172–174]

Bellamy *et al* carried out a genome screen for tuberculosis susceptibility loci in 136 African families containing 173 independent affected sib pairs.[125] Two chromosomal regions showed suggestive evidence of linkage (Xq27 and 15q11). Although the evidence of linkage did not reach the criteria for genome-wide statistical significance,[175] further support for the presence of the loci was obtained using the method of common ancestry mapping. Very recently, genome-wide linkage scans have also identified new susceptibility loci for persistent hepatitis B virus infection (Frodsham *et al*, unpublished).

Leprosy is a disease that is well suited to genome-wide linkage studies in that it has a clear phenotype and the disease progression is slow. The disease has a very low mortality rate that not only enables the collection of older family members, but also suggests that if a major susceptibility gene does exist, it might be able to persist in the population. Segregation studies have suggested the presence of a major gene controlling susceptibility to leprosy; however, the gene associations previously detected in candidate gene studies appeared only to

contribute a small proportion to the overall genetic component.

Only one genome screen for leprosy susceptibility has been published to date. A two-stage genome scan was carried out in South India using a total of 224 families (245 independent sib pairs) with mainly the tuberculoid form of leprosy.[176] A region of strongly significant linkage was identified on chromosome 10p13 ($P < 2 \times 10^{-5}$). A gene that lies at the peak of this linkage is the macrophage mannose receptor (MRC1). This receptor mediates phagocytosis and endocytosis of pathogens by recognition of the mannose and fucose structures present on their surface.[177] The terminal mannosyl units of lipoarabinomannan (LAM) from the surface of *M. tuberculosis* were found to be ligands for MRC1 allowing phagocytosis of the organism.[178] The receptor also appears to have a role in the processing of glycolipids derived from mycobacteria.[179]

It is therefore possible that MRC1 could also have a role in the immune response to *M. leprae* infection. To fully define the relevance of this positional candidate gene in leprosy, an extensive analysis of numerous variants in this gene in the South Indian families used for the genome-wide scan is in progress (Tosh *et al*, unpublished).

Although the region of linkage on chromosome 10p13 is thought to contribute substantially to the total genetic component of leprosy susceptibility in this population, a follow-up of markers showing weaker evidence of linkage identified a second region of linkage on chromosome 20p12 ($P = 0.00003$).[180] This linkage is not as strong as that identified on chromosome 10; however, it is interesting that this same region of chromosome 20 (p12) has been found to determine susceptibility to atopic dermatitis and psoriasis.[172,174] These are all diseases caused by inappropriate immune responses to an environmental stimulus. This suggests the presence of a gene involved in the regulation of immune responses, although no strong positional candidates have been identified as yet.

Even though there have been many studies showing linkage and association of the MHC to leprosy, no strong linkage was detected in South India, suggesting heterogeneity in genetic susceptibility. Furthermore, Tosh *et al*, showed that the strength of the chromosome 20 linkage differs in the two geographic regions of India studied. Also, as the sibpairs studied were mainly paucibacillary, it will also be interesting to see if the two regions identified so far contain susceptibility loci for leprosy *per se* or to tuberculoid leprosy.

It is encouraging that regions of linkage, reaching genome-wide statistical significance, have been identified as it seemed possible that infectious diseases were too polygenic to have enough power to detect linkage using the genome-wide approach; however, further genome-wide linkage studies in different populations will be required to determine the extent of genetic heterogeneity that exists for leprosy susceptibility.

Conclusions

Thus far all the genes suggested to have a role in the susceptibility to leprosy either act to directly modulate development of the adaptive response (HLA, MICA, TAP2, CTLA4, VDR), or may bridge the innate and adaptive responses (NRAMP1, TLR2, HSP70, TNFα, MRC1). This is consistent with the idea that an appropriate cell-mediated response is critical in the control of mycobacterial infection. Many of the associations, have only been found in a small series of patients, or in a single population (Table 1), and should be repeated in larger studies. Lack of correlation in results between populations should not necessarily be regarded as a negation of initial associations but may instead reflect heterogeneity in the genetic susceptibility to this enigmatic disease.

References

1 WHO. Leprosy. *Weekly Epidemiol Rec* 2002; **77**: 1–8.
2 Abel L, Demenais F. Detection of major genes for susceptibility to leprosy and its subtypes in a Caribbean island: Desirade island. *Am J Hum Genet* 1988; **42**: 256–266.
3 Haile RW, Iselius L, Fine PE, Morton NE. Segregation and linkage analyses of 72 leprosy pedigrees. *Hum Hered* 1985; **35**: 43–52.
4 Feitosa MF, Borecki I, Krieger H, Beiguelman B, Rao DC. The genetic epidemiology of leprosy in a Brazilian population. *Am J Hum Genet* 1995; **56**: 1179–1185.
5 Wagener DK, Schauf V, Nelson KE, Scollard D, Brown A, Smith T. Segregation analysis of leprosy in families of northern Thailand. *Genet Epidemiol* 1988; **5**: 95–105.
6 Chakravartti MR, Vogel F. A twin study on leprosy. *Top Hum Genet* 1973; **1**: 1–123.
7 de Vries RR, Fat RF, Nijenhuis LE, van Rood JJ. HLA-linked genetic control of host response to *Mycobacterium leprae*. *Lancet* 1976; **2**: 1328–1330.
8 Fine PE, Wolf E, Pritchard J *et al*. HLA-linked genes and leprosy: a family study in Karigiri, South India. *J Infect Dis* 1979; **140**: 152–161.
9 de Vries RR, Mehra NK, Vaidya MC, Gupte MD, Meera Khan P, Van Rood JJ. HLA-linked control of susceptibility to tuberculoid leprosy and association with HLA-DR types. *Tissue Antigens* 1980; **16**: 294–304.
10 van Eden W, Gonzalez NM, de Vries RR, Convit J, van Rood JJ. HLA-linked control of predisposition to lepromatous leprosy. *J Infect Dis* 1985; **151**: 9–14.
11 Dessoukey MW, el-Shiemy S, Sallam T. HLA and leprosy: segregation and linkage study. *Int J Dermatol* 1996; **35**: 257–264.
12 Xu KY, de Vries RR, Fei HM, van Leeuwen A, Chen RB, Ye GY. HLA-linked control of predisposition to lepromatous leprosy. *Int J Lepr Other Mycobact Dis* 1985; **53**: 56–63.
13 Casanova JL, Abel L. Genetic dissection of immunity to mycobacteria: the human model. *Annu Rev Immunol* 2002; **20**: 581–620.
14 Serjeantson SW. HLA and susceptibility to leprosy. *Immunol Rev* 1983; **70**: 89–112.
15 Rani R, Zaheer SA, Mukherjee R. Do human leukocyte antigens have a role to play in differential manifestation of multibacillary leprosy: a study on multibacillary leprosy patients from north India. *Tissue Antigens* 1992; **40**: 124–127.
16 Greiner J, Schleiermacher E, Smith T, Lenhard V, Vogel F. The HLA system and leprosy in Thailand. *Hum Genet* 1978; **42**: 201–213.
17 Takata H, Sada M, Ozawa S, Sekiguchi S. HLA and mycobacterial infection: increased frequency of B8 in Japanese leprosy. *Tissue Antigens* 1978; **11**: 61–64.
18 Izumi S, Sugiyama K, Matsumoto Y, Ohkawa S. Analysis of the immunogenetic background of Japanese leprosy patients by the HLA system. *Vox Sang* 1982; **42**: 243–247.

19 Kim SJ, Choi IH, Dahlberg S, Nisperos B, Kim JD, Hansen JA. HLA and leprosy in Koreans. *Tissue Antigens* 1987; **29**: 146–153.
20 Rea TH, Levan NE, Terasaki PI. Histocompatibility antigens in patients with leprosy. *J Infect Dis* 1976; **134**: 615–618.
21 Wang LM, Kimura A, Satoh M, Mineshita S. HLA linked with leprosy in southern China: HLA-linked resistance alleles to leprosy. *Int J Lepr Other Mycobact Dis* 1999; **67**: 403–408.
22 Groh V, Rhinehart R, Randolph-Habecker J, Topp MS, Riddell SR, Spies T. Costimulation of CD8alphabeta T cells by NKG2D via engagement by MIC induced on virus-infected cells. *Nat Immunol* 2001; **2**: 255–260.
23 Das H, Groh V, Kuijl C *et al*. MICA engagement by human Vgamma2Vdelta2 T cells enhances their antigen-dependent effector function. *Immunity* 2001; **15**: 83–93.
24 Robinson J, Perez-Rodriguez M, Waller MJ *et al*. MICA sequences 2000. *Immunogenetics* 2001; **53**: 150–169.
25 Perez-Rodriguez M, Arguello JR, Fischer G *et al*. Further polymorphism of the MICA gene. *Eur J Immunogenet* 2002; **29**: 35–46.
26 Rani R, Fernandez-Vina MA, Zaheer SA, Beena KR, Stastny P. Study of HLA class II alleles by PCR oligotyping in leprosy patients from north India. *Tissue Antigens* 1993; **42**: 133–137.
27 Mehra NK, Rajalingam R, Mitra DK, Taneja V, Giphart MJ. Variants of HLA-DR2/DR51 group haplotypes and susceptibility to tuberculoid leprosy and pulmonary tuberculosis in Asian Indians. *Int J Lepr Other Mycobact Dis* 1995; **63**: 241–248.
28 Schauf V, Ryan S, Scollard D *et al*. Leprosy associated with HLA-DR2 and DQw1 in the population of northern Thailand. *Tissue Antigens* 1985; **26**: 243–247.
29 Miyanaga K, Juji T, Maeda H, Nakajima S, Kobayashi S. Tuberculoid leprosy and HLA in Japanese. *Tissue Antigens* 1981; **18**: 331–334.
30 Joko S, Numaga J, Masuda K, Namisato M, Maeda H. [HLA class II alleles and leprosy (Hansen's disease) classified by WHO- MDT criteria]. *Nippon Rai Gakkai Zasshi* 1996; **65**: 121–127.
31 Todd JR, West BC, McDonald JC. Human leukocyte antigen and leprosy: study in northern Louisiana and review. *Rev Infect Dis* 1990; **12**: 63–74.
32 Shaw MA, Donaldson IJ, Collins A *et al*. Association and linkage of leprosy phenotypes with HLA class II and tumour necrosis factor genes. *Genes Immun* 2001; **2**: 196–204.
33 Salgame P, Convit J, Bloom BR. Immunological suppression by human CD8+ T cells is receptor dependent and HLA-DQ restricted. *Proc Natl Acad Sci USA* 1991; **88**: 2598–2602.
34 Gorodezky C, Flores J, Arevalo N, Castro LE, Silva A, Rodriguez O. Tuberculoid leprosy in Mexicans is associated with HLA-DR3. *Lepr Rev* 1987; **58**: 401–406.
35 van Eden W, de Vries RR, D'Amaro J, Schreuder I, Leiker DL, van Rood JJ. HLA-DR-associated genetic control of the type of leprosy in a population from surinam. *Hum Immunol* 1982; **4**: 343–350.
36 Ottenhoff TH, Converse PJ, Bjune G, de Vries RR. HLA antigens and neural reversal reactions in Ethiopian borderline tuberculoid leprosy patients. *Int J Lepr Other Mycobact Dis* 1987; **55**: 261–266.
37 Van Schooten WC, Elferink DG, Van Embden J, Anderson DC, De Vries RR. DR3-restricted T cells from different HLA-DR3-positive individuals recognize the same peptide (amino acids 2–12) of the mycobacterial 65-kDa heat-shock protein. *Eur J Immunol* 1989; **19**: 2075–2079.
38 Ottenhoff TH, de Vries RR. HLA class II immune response and suppression genes in leprosy. *Int J Lepr Other Mycobact Dis* 1987; **55**: 521–534.
39 Haanen JB, Ottenhoff TH, Lai AFRF, Soebono H, Spits H, de Vries RR. Mycobacterium leprae-specific T cells from a tuberculoid leprosy patient suppress HLA-DR3-restricted T cell responses to an immunodominant epitope on 65-kDa hsp of mycobacteria. *J Immunol* 1990; **145**: 3898–3904.
40 Visentainer JE, Tsuneto LT, Serra MF, Peixoto PR, Petzl-Erler ML. Association of leprosy with HLA-DR2 in a Southern Brazilian population. *Braz J Med Biol Res* 1997; **30**: 51–59.
41 Cem Mat M, Yazici H, Ozbakir F, Tuzun Y. The HLA association of lepromatous leprosy and borderline lepromatous leprosy in Turkey. A preliminary study. *Int J Dermatol* 1988; **27**: 246–247.
42 Roy S, McGuire W, Mascie-Taylor CG *et al*. Tumor necrosis factor promoter polymorphism and susceptibility to lepromatous leprosy. *J Infect Dis* 1997; **176**: 530–532.
43 Joko S, Numaga J, Kawashima H, Namisato M, Maeda H. Human leukocyte antigens in forms of leprosy among Japanese patients. *Int J Lepr Other Mycobact Dis* 2000; **68**: 49–56.
44 van Eden W, de Vries RR, Mehra NK, Vaidya MC, D'Amaro J, van Rood JJ. HLA segregation of tuberculoid leprosy: confirmation of the DR2 marker. *J Infect Dis* 1980; **141**: 693–701.
45 Mehra NK, Verduijn W, Taneja V, Drabbels J, Singh SP, Giphart MJ. Analysis of HLA-DR2-associated polymorphisms by oligonucleotide hybridization in an Asian Indian population. *Hum Immunol* 1991; **32**: 246–253.
46 Zerva L, Cizman B, Mehra NK *et al*. Arginine at positions 13 or 70–71 in pocket 4 of HLA-DRB1 alleles is associated with susceptibility to tuberculoid leprosy. *J Exp Med* 1996; **183**: 829–836.
47 Uko GP, Lu LY, Asuquo MA *et al*. HLA-DRB1 leprogenic motifs in nigerian population groups. *Clin Exp Immunol* 1999; **118**: 56–62.
48 Agrewala JN, Wilkinson RJ. Influence of HLA-DR on the phenotype of CD4+ T lymphocytes specific for an epitope of the 16-kDa alpha-crystallin antigen of *Mycobacterium tuberculosis*. *Eur J Immunol* 1999; **29**: 1753–1761.
49 Mutis T, Cornelisse YE, Datema G, van den Elsen PJ, Ottenhoff TH, de Vries RR. Definition of a human suppressor T-cell epitope. *Proc Natl Acad Sci USA* 1994; **91**: 9456–9460.
50 Joly AL, Le Rolle AL, Gonzalez AL *et al*. Co-evolution of rat TAP transporters and MHC class I RT1-A molecules. *Curr Biol* 1998; **8**: 169–172.
51 Hayney MS, Poland GA, Dimanlig P, Schaid DJ, Jacobson RM, Lipsky JJ. Polymorphisms of the TAP2 gene may influence antibody response to live measles vaccine virus. *Vaccine* 1997; **15**: 3–6.
52 Barron KS, Reveille JD, Carrington M, Mann DL, Robinson MA. Susceptibility to Reiter's syndrome is associated with alleles of TAP genes. *Arthritis Rheum* 1995; **38**: 684–689.
53 Gonzalez-Escribano MF, Morales J, Garcia-Lozano JR *et al*. TAP polymorphism in patients with Behcet's disease. *Ann Rheum Dis* 1995; **54**: 386–388.
54 Hillarby MC, Davies EJ, Donn RP, Grennan DM, Ollier WE. TAP2D is associated with HLA-B44 and DR4 and may contribute to rheumatoid arthritis and Felty's syndrome susceptibility. *Clin Exp Rheumatol* 1996; **14**: 67–70.
55 Kaslow RA, Carrington M, Apple R *et al*. Influence of combinations of human major histocompatibility complex genes on the course of HIV-1 infection. *Nat Med* 1996; **2**: 405–411.
56 Daniel S, Caillat-Zucman S, Hammer J, Bach JF, van Endert PM. Absence of functional relevance of human transporter associated with antigen processing polymorphism for peptide selection. *J Immunol* 1997; **159**: 2350–2357.
57 Faucz FR, Probst CM, Petzl-Erler ML. Polymorphism of LMP2, TAP1, LMP7 and TAP2 in Brazilian Amerindians and Caucasoids: implications for the evolution of allelic and haplotypic diversity. *Eur J Immunogenet* 2000; **27**: 5–16.
58 Rajalingam R, Singal DP, Mehra NK. Transporter associated with antigen-processing (TAP) genes and susceptibility to tuberculoid leprosy and pulmonary tuberculosis. *Tissue Antigens* 1997; **49**: 168–172.
59 Engele M, Stossel E, Castiglione K *et al*. Induction of TNF in human alveolar macrophages as a potential evasion mechanism of virulent *Mycobacterium tuberculosis*. *J Immunol* 2002; **168**: 1328–1337.

60 Udalova IA, Richardson A, Denys A *et al*. Functional consequences of a polymorphism affecting NF-kappaB p50–p50 binding to the TNF promoter region. *Mol Cell Biol* 2000; **20**: 9113–9119.
61 Knight JC, Udalova I, Hill AV *et al*. A polymorphism that affects OCT-1 binding to the TNF promoter region is associated with severe malaria [see comments]. *Nat Genet* 1999; **22**: 145–150.
62 Wilson AG, Symons JA, McDowell TL, McDevitt HO, Duff GW. Effects of a polymorphism in the human tumor necrosis factor alpha promoter on transcriptional activation. *Proc Natl Acad Sci USA* 1997; **94**: 3195–3199.
63 Louis E, Franchimont D, Piron A *et al*. Tumour necrosis factor (TNF) gene polymorphism influences TNF-alpha production in lipopolysaccharide (LPS)-stimulated whole blood cell culture in healthy humans. *Clin Exp Immunol* 1998; **113**: 401–406.
64 Kroeger KM, Carville KS, Abraham LJ. The -308 tumor necrosis factor-alpha promoter polymorphism effects transcription. *Mol Immunol* 1997; **34**: 391–399.
65 Brinkman BM, Zuijdeest D, Kaijzel EL, Breedveld FC, Verweij CL. Relevance of the tumor necrosis factor alpha (TNF alpha) −308 promoter polymorphism in TNF alpha gene regulation [see comments]. *J Inflamm* 1995; **46**: 32–41.
66 Somoskovi A, Zissel G, Seitzer U, Gerdes J, Schlaak M, Muller Quernheim J. Polymorphisms at position −308 in the promoter region of the TNF-alpha and in the first intron of the TNF-beta genes and spontaneous and lipopolysaccharide-induced TNF-alpha release in sarcoidosis. *Cytokine* 1999; **11**: 882–887.
67 Kaijzel EL, Bayley JP, van Krugten MV *et al*. Allele-specific quantification of tumor necrosis factor alpha (TNF) transcription and the role of promoter polymorphisms in rheumatoid arthritis patients and healthy individuals. *Genes Immun* 2001; **2**: 135–144.
68 Stuber F, Udalova IA, Book M *et al*. −308 tumor necrosis factor (TNF) polymorphism is not associated with survival in severe sepsis and is unrelated to lipopolysaccharide inducibility of the human TNF promoter. *J Inflamm* 1995; **46**:42–50.
69 Nadal D, Leppert D, Frei K, Gallo P, Lamche H, Fontana A. Tumour necrosis factor-alpha in infectious meningitis. *Arch Dis Child* 1989; **64**: 1274–1279.
70 Cabrera M, Shaw MA, Sharples C *et al*. Polymorphism in tumor necrosis factor genes associated with mucocutaneous leishmaniasis. *J Exp Med* 1995; **182**: 1259–1264.
71 Conway DJ, Holland MJ, Bailey RL *et al*. Scarring trachoma is associated with polymorphism in the tumor necrosis factor alpha (TNF-alpha) gene promoter and with elevated TNF-alpha levels in tear fluid. *Infect Immun* 1997; **65**: 1003–1006.
72 McGuire W, Hill AV, Allsopp CE, Greenwood BM, Kwiatkowski D. Variation in the TNF-alpha promoter region associated with susceptibility to cerebral malaria. *Nature* 1994; **371**: 508–510.
73 Sashio H, Tamura K, Ito R *et al*. Polymorphisms of the TNF gene and the TNF receptor superfamily member 1B gene are associated with susceptibility to ulcerative colitis and Crohn's disease, respectively. *Immunogenetics* 2002; **53**: 1020–1027.
74 Santos AR, Almeida AS, Suffys PN *et al*. Tumor necrosis factor promoter polymorphism (TNF2) seems to protect against development of severe forms of leprosy in a pilot study in Brazilian patients. *Int J Lepr Other Mycobact Dis* 2000; **68**: 325–327.
75 Moraes MO, Duppre NC, Suffys PN *et al*. Tumor necrosis factor-alpha promoter polymorphism TNF2 is associated with a stronger delayed-type hypersensitivity reaction in the skin of borderline tuberculoid leprosy patients. *Immunogenetics* 2001; **53**: 45–47.
76 Khanolkar-Young S, Rayment N, Brickell PM *et al*. Tumour necrosis factor-alpha (TNF-alpha) synthesis is associated with the skin and peripheral nerve pathology of leprosy reversal reactions. *Clin Exp Immunol* 1995; **99**: 196–202.
77 Kaplan G. Cytokine regulation of disease progression in leprosy and tuberculosis. *Immunobiology* 1994; **191**: 564–568.
78 Skoff AM, Lisak RP, Bealmear B, Benjamins JA. TNF-alpha and TGF-beta act synergistically to kill Schwann cells. *J Neurosci Res* 1998; **53**: 747–756.
79 Sarno EN, Santos AR, Jardim MR *et al*. Pathogenesis of nerve damage in leprosy: genetic polymorphism regulates the production of TNF alpha. *Lepr Rev* 2000; **71** (Suppl): S154–S158 discussion S158–S160.
80 Jongeneel CV, Briant L, Udalova IA, Sevin A, Nedospasov SA, Cambon-Thomsen A. Extensive genetic polymorphism in the human tumor necrosis factor region and relation to extended HLA haplotypes. *Proc Natl Acad Sci USA* 1991; **88**: 9717–9721.
81 Fanning GC, Bunce M, Black CM, Welsh KI. Polymerase chain reaction haplotyping using 3′ mismatches in the forward and reverse primers: application to the biallelic polymorphisms of tumor necrosis factor and lymphotoxin alpha. *Tissue Antigens* 1997; **50**: 23–31.
82 de Messias IJ, Santamaria J, Brenden M, Reis A, Mauff G. Association of C4B deficiency (C4B*Q0) with erythema nodosum in leprosy. *Clin Exp Immunol* 1993; **92**: 284–287.
83 Vanbuskirk A, Crump BL, Margoliash E, Pierce SK. A peptide binding protein having a role in antigen presentation is a member of the HSP70 heat shock family. *J Exp Med* 1989; **170**: 1799–1809.
84 Vabulas RM, Ahmad-Nejad P, Ghose S, Kirschning CJ, Issels RD, Wagner H. HSP70 as endogenous stimulus of the Toll/interleukin-1 receptor signal pathway. *J Biol Chem* 2002; **277**: 15107–15112.
85 Asea A, Rehli M, Kabingu E *et al*. Novel signal transduction pathway utilized by extracellular HSP70: role of toll-like receptor (TLR) 2 and TLR4. *J Biol Chem* 2002; **277**: 15028–15034.
86 Laroia G, Cuesta R, Brewer G, Schneider RJ. Control of mRNA decay by heat shock–ubiquitin–proteasome pathway. *Science* 1999; **284**: 499–502.
87 Cascino I, Sorrentino R, Tosi R. Strong genetic association between HLA-DR3 and a polymorphic variation in the regulatory region of the HSP70-1 gene. *Immunogenetics* 1993; **37**: 177–182.
88 Rajalingam R, Mehra NK, Singal DP. Polymorphism in heat-shock protein 70-1 (HSP70-1) gene promoter region and susceptibility to tuberculoid leprosy and pulmonary tuberculosis in Asian Indians. *Indian J Exp Biol* 2000; **38**: 658–662.
89 Cascino I, D'Alfonso S, Cappello N *et al*. Gametic association of HSP70-1 promoter region alleles and their inclusion in extended HLA haplotypes. *Tissue Antigens* 1993; **42**: 62–66.
90 Vidal S, Gros P, Skamene E. Natural resistance to infection with intracellular parasites: molecular genetics identifies Nramp1 as the Bcg/Ity/Lsh locus. *J Leukoc Biol* 1995; **58**: 382–390.
91 Brown IN, Glynn AA, Plant J. Inbred mouse strain resistance to *Mycobacterium lepraemurium* follows the Ity/Lsh pattern. *Immunology* 1982; **47**: 149–156.
92 Skamene E, Gros P, Forget A, Patel PJ, Nesbitt MN. Regulation of resistance to leprosy by chromosome 1 locus in the mouse. *Immunogenetics* 1984; **19**: 117–124.
93 Goto Y, Buschman E, Skamene E. Regulation of host resistance to *Mycobacterium intracellulare in vivo* and *in vitro* by the Bcg gene. *Immunogenetics* 1989; **30**: 218–221.
94 Blackwell JM, Roberts CW, Roach TI, Alexander J. Influence of macrophage resistance gene Lsh/Ity/Bcg (candidate Nramp) on *Toxoplasma gondii* infection in mice. *Clin Exp Immunol* 1994; **97**: 107–112.
95 Puliti M, Radzioch D, Mazzolla R, Barluzzi R, Bistoni F, Blasi E. Influence of the Bcg locus on macrophage response to the dimorphic fungus Candida albicans. *Infect Immun* 1995; **63**: 4170–4173.
96 Leclercq V, Lebastard M, Belkaid Y, Louis J, Milon G. The outcome of the parasitic process initiated by *Leishmania infantum* in laboratory mice: a tissue-dependent pattern

controlled by the Lsh and MHC loci. *J Immunol* 1996; **157**: 4537–4545.
97 North RJ, LaCourse R, Ryan L, Gros P. Consequence of Nramp1 deletion to *Mycobacterium tuberculosis* infection in mice. *Infect Immun* 1999; **67**: 5811–5814.
98 Medina E, Rogerson BJ, North RJ. The Nramp1 antimicrobial resistance gene segregates independently of resistance to virulent *Mycobacterium tuberculosis*. *Immunology* 1996; **88**: 479–481.
99 Medina E, North RJ. Evidence inconsistent with a role for the Bcg gene (Nramp1) in resistance of mice to infection with virulent *Mycobacterium tuberculosis*. *J Exp Med* 1996; **183**: 1045–1051.
100 Wyllie S, Seu P, Goss JA. The natural resistance-associated macrophage protein 1 Slc11a1 (formerly Nramp1) and iron metabolism in macrophages. *Microbes Infect* 2002; **4**: 351–359.
101 Jabado N, Jankowski A, Dougaparsad S, Picard V, Grinstein S, Gros P. Natural resistance to intracellular infections: natural resistance-associated macrophage protein 1 (Nramp1) functions as a pH-dependent manganese transporter at the phagosomal membrane. *J Exp Med* 2000; **192**: 1237–1248.
102 Blackwell JM, Searle S, Goswami T, Miller EN. Understanding the multiple functions of Nramp1. *Microbes Infect* 2000; **2**: 317–321.
103 Lang T, Prina E, Sibthorpe D, Blackwell JM. Nramp1 transfection transfers Ity/Lsh/Bcg-related pleiotropic effects on macrophage activation: influence on antigen processing and presentation. *Infect Immun* 1997; **65**: 380–386.
104 Kaye PM, Blackwell JM. Lsh antigen presentation and the development of CMI. *Res Immunol* 1989; **140**: 810–815; discussion 815–822.
105 Soo SS, Villarreal-Ramos B, Anjam Khan CM, Hormaeche CE, Blackwell JM. Genetic control of immune response to recombinant antigens carried by an attenuated *Salmonella typhimurium* vaccine strain: Nramp1 influences T-helper subset responses and protection against leishmanial challenge. *Infect Immun* 1998; **66**: 1910–1917.
106 Liu J, Fujiwara TM, Buu NT *et al*. Identification of polymorphisms and sequence variants in the human homologue of the mouse natural resistance-associated macrophage protein gene. *Am J Hum Genet* 1995; **56**: 845–853.
107 White JK, Shaw MA, Barton CH *et al*. Genetic and physical mapping of 2q35 in the region of the NRAMP and IL8R genes: identification of a polymorphic repeat in exon 2 of NRAMP. *Genomics* 1994; **24**: 295–302.
108 Buu NT, Cellier M, Gros P, Schurr E. Identification of a highly polymorphic length variant in the 3′UTR of NRAMP1. *Immunogenetics* 1995; **42**: 428–429.
109 Searle S, Blackwell JM. Evidence for a functional repeat polymorphism in the promoter of the human NRAMP1 gene that correlates with autoimmune versus infectious disease susceptibility. *J Med Genet* 1999; **36**: 295–299.
110 Shaw MA, Clayton D, Atkinson SE *et al*. Linkage of rheumatoid arthritis to the candidate gene NRAMP1 on 2q35. *J Med Genet* 1996; **33**: 672–677.
111 Singal DP, Li J, Zhu Y, Zhang G. NRAMP1 gene polymorphisms in patients with rheumatoid arthritis. *Tissue Antigens* 2000; **55**: 44–47.
112 Sanjeevi CB, Miller EN, Dabadghao P *et al*. Polymorphism at NRAMP1 and D2S1471 loci associated with juvenile rheumatoid arthritis. *Arthritis Rheum* 2000; **43**: 1397–1404.
113 Hofmeister A, Neibergs HL, Pokorny RM, Galandiuk S. The natural resistance-associated macrophage protein gene is associated with Crohn's disease. *Surgery* 1997; **122**: 173–178; discussion 178–179.
114 Kojima Y, Kinouchi Y, Takahashi S, Negoro K, Hiwatashi N, Shimosegawa T. Inflammatory bowel disease is associated with a novel promoter polymorphism of natural resistance-associated macrophage protein 1 (NRAMP1) gene. *Tissue Antigens* 2001; **58**: 379–384.
115 Esposito L, Hill NJ, Pritchard LE *et al*. Genetic analysis of chromosome 2 in type 1 diabetes: analysis of putative loci IDDM7, IDDM12, and IDDM13 and candidate genes NRAMP1 and IA-2 and the interleukin-1 gene cluster. IMDIAB Group. *Diabetes* 1998; **47**: 1797–1799.
116 Maliarik MJ, Chen KM, Sheffer RG *et al*. The natural resistance-associated macrophage protein gene in African Americans with sarcoidosis. *Am J Respir Cell Mol Biol* 2000; **22**: 672–675.
117 Marquet S, Sanchez FO, Arias M *et al*. Variants of the human NRAMP1 gene and altered human immunodeficiency virus infection susceptibility. *J Infect Dis* 1999; **180**: 1521–1525.
118 Blackwell JM. Modern genetics and leprosy susceptibility. *Lepr Rev* 2001; **72**: 352–356.
119 Gao PS, Fujishima S, Mao XQ *et al*. Genetic variants of NRAMP1 and active tuberculosis in Japanese populations. International Tuberculosis Genetics Team. *Clin Genet* 2000; **58**: 74–76.
120 Shaw MA, Collins A, Peacock CS *et al*. Evidence that genetic susceptibility to *Mycobacterium tuberculosis* in a Brazilian population is under oligogenic control: linkage study of the candidate genes NRAMP1 and TNFA. *Tuber Lung Dis* 1997; **78**: 35–45.
121 Ryu S, Park YK, Bai GH, Kim SJ, Park SN, Kang S. 3′UTR polymorphisms in the NRAMP1 gene are associated with susceptibility to tuberculosis in Koreans. *Int J Tuberc Lung Dis* 2000; **4**: 577–580.
122 Cervino AC, Lakiss S, Sow O, Hill AV. Allelic association between the NRAMP1 gene and susceptibility to tuberculosis in Guinea-Conakry. *Ann Hum Genet* 2000; **64**: 507–512.
123 Bellamy R, Ruwende C, Corrah T, McAdam KP, Whittle HC, Hill AV. Variations in the NRAMP1 gene and susceptibility to tuberculosis in West Africans. *N Engl J Med* 1998; **338**: 640–644.
124 Greenwood CM, Fujiwara TM, Boothroyd LJ *et al*. Linkage of tuberculosis to chromosome 2q35 loci, including NRAMP1, in a large aboriginal Canadian family. *Am J Hum Genet* 2000; **67**: 405–416.
125 Bellamy R, Beyers N, McAdam KP *et al*. Genetic susceptibility to tuberculosis in Africans: a genome-wide scan. *Proc Natl Acad Sci USA* 2000; **97**: 8005–8009.
126 Abel L, Sanchez FO, Oberti J *et al*. Susceptibility to leprosy is linked to the human NRAMP1 gene. *J Infect Dis* 1998; **177**: 133–145.
127 Shaw MA, Atkinson S, Dockrell H *et al*. An RFLP map for 2q33–q37 from multicase mycobacterial and leishmanial disease families: no evidence for an Lsh/Ity/Bcg gene homologue influencing susceptibility to leprosy. *Ann Hum Genet* 1993; **57** (Pt 4): 251–271.
128 Roy S, Frodsham A, Saha B, Hazra SK, Mascie-Taylor CG, Hill AV. Association of vitamin D receptor genotype with leprosy type. *J Infect Dis* 1999; **179**: 187–191.
129 Meisner SJ, Mucklow S, Warner G, Sow SO, Lienhardt C, Hill AV. Association of NRAMP1 polymorphism with leprosy type but not susceptibility to leprosy per se in west Africans. *Am J Trop Med Hyg* 2001; **65**: 733–735.
130 Roger M, Levee G, Chanteau S, Gicquel B, Schurr E. No evidence for linkage between leprosy susceptibility and the human natural resistance-associated macrophage protein 1 (NRAMP1) gene in French Polynesia. *Int J Lepr Other Mycobact Dis* 1997; **65**: 197–202.
131 Alcais A, Sanchez FO, Thuc NV *et al*. Granulomatous reaction to intradermal injection of lepromin (Mitsuda reaction) is linked to the human NRAMP1 gene in Vietnamese leprosy sibships. *J Infect Dis* 2000; **181**: 302–308.
132 Kaur G, Sachdeva G, Bhutani LK, Bamezai R. Association of polymorphism at COL3A and CTLA4 loci on chromosome 2q31–33 with the clinical phenotype and in-vitro CMI status

in healthy and leprosy subjects: a preliminary study. *Hum Genet* 1997; **100**: 43–50.
133 Holopainen PM, Partanen JA. Technical note: linkage disequilibrium and disease-associated CTLA4 gene polymorphisms. *J Immunol* 2001; **167**: 2457–2458.
134 Chistiakov DA, Savost'anov KV, Nosikov VV. CTLA4 gene polymorphisms are associated with, and linked to, insulin-dependent diabetes mellitus in a Russian population. *BMC Genet* 2001; **2**: 6.
135 Kristiansen OP, Larsen ZM, Pociot F. CTLA-4 in autoimmune diseases–a general susceptibility gene to autoimmunity? *Genes Immun* 2000; **1**: 170–184.
136 Morrison NA, Qi JC, Tokita A *et al*. Prediction of bone density from vitamin D receptor alleles. *Nature* 1994; **367**: 284–287.
137 Hughes MR, Malloy PJ, Kieback DG *et al*. Point mutations in the human vitamin D receptor gene associated with hypocalcemic rickets. *Science* 1988; **242**: 1702–1705.
138 Lorentzon M, Lorentzon R, Nordstrom P. Vitamin D receptor gene polymorphism is associated with birth height, growth to adolescence, and adult stature in healthy caucasian men: a cross-sectional and longitudinal study. *J Clin Endocrinol Metab* 2000; **85**: 1666–1670.
139 Vidal M, Ramana CV, Dusso AS. Stat1-vitamin D receptor interactions antagonize 1,25-dihydroxyvitamin D transcriptional activity and enhance stat1-mediated transcription. *Mol Cell Biol* 2002; **22**: 2777–2787.
140 Alroy I, Towers TL, Freedman LP. Transcriptional repression of the interleukin-2 gene by vitamin D3: direct inhibition of NFATp/AP-1 complex formation by a nuclear hormone receptor. *Mol Cell Biol* 1995; **15**: 5789–5799.
141 Towers TL, Freedman LP. Granulocyte-macrophage colony-stimulating factor gene transcription is directly repressed by the vitamin D3 receptor. Implications for allosteric influences on nuclear receptor structure and function by a DNA element. *J Biol Chem* 1998; **273**: 10338–10348.
142 Cippitelli M, Santoni A. Vitamin D3: a transcriptional modulator of the interferon-gamma gene. *Eur J Immunol* 1998; **28**: 3017–3030.
143 D'Ambrosio D, Cippitelli M, Cocciolo MG *et al*. Inhibition of IL-12 production by 1,25-dihydroxyvitamin D3. Involvement of NF-kappaB downregulation in transcriptional repression of the p40 gene. *J Clin Invest* 1998; **101**: 252–262.
144 Cippitelli M, Fionda C, Di Bona D *et al*. Negative regulation of CD95 ligand gene expression by vitamin D3 in T lymphocytes. *J Immunol* 2002; **168**: 1154–1166.
145 Rescigno M, Piguet V, Valzasina B *et al*. Fas engagement induces the maturation of dendritic cells (DCs), the release of interleukin (IL)-1beta, and the production of interferon gamma in the absence of IL-12 during DC-T cell cognate interaction: a new role for Fas ligand in inflammatory responses. *J Exp Med* 2000; **192**: 1661–1668.
146 Morrison NA, Yeoman R, Kelly PJ, Eisman JA. Contribution of trans-acting factor alleles to normal physiological variability: vitamin D receptor gene polymorphism and circulating osteocalcin. *Proc Natl Acad Sci USA* 1992; **89**: 6665–6669.
147 Verbeek W, Gombart AF, Shiohara M, Campbell M, Koeffler HP. Vitamin D receptor: no evidence for allele-specific mRNA stability in cells which are heterozygous for the Taq I restriction enzyme polymorphism. *Biochem Biophys Res Commun* 1997; **238**: 77–80.
148 Mocharla H, Butch AW, Pappas AA *et al*. Quantification of vitamin D receptor mRNA by competitive polymerase chain reaction in PBMC: lack of correspondence with common allelic variants. *J Bone Miner Res* 1997; **12**: 726–733.
149 Simmons JD, Mullighan C, Welsh KI, Jewell DP. Vitamin D receptor gene polymorphism: association with Crohn's disease susceptibility. *Gut* 2000; **47**: 211–214.
150 Bellamy R, Ruwende C, Corrah T *et al*. Tuberculosis and chronic hepatitis B virus infection in Africans and variation in the vitamin D receptor gene. *J Infect Dis* 1999; **179**: 721–724.
151 Wilkinson RJ, Llewelyn M, Toossi Z *et al*. Influence of vitamin D deficiency and vitamin D receptor polymorphisms on tuberculosis among Gujarati Asians in west London: a case-control study. *Lancet* 2000; **355**: 618–621.
152 Gelder CM, Hart KW, Williams OM *et al*. Vitamin D receptor gene polymorphisms and susceptibility to *Mycobacterium malmoense* pulmonary disease. *J Infect Dis* 2000; **181**: 2099–2102.
153 Selvaraj P, Narayanan PR, Reetha AM. Association of vitamin D receptor genotypes with the susceptibility to pulmonary tuberculosis in female patients & resistance in female contacts. *Indian J Med Res* 2000; **111**: 172–179.
154 Ingles SA, Haile RW, Henderson BE *et al*. Strength of linkage disequilibrium between two vitamin D receptor markers in five ethnic groups: implications for association studies. *Cancer Epidemiol Biomarkers Prev* 1997; **6**: 93–98.
155 Jurutka PW, Remus LS, Whitfield GK *et al*. The polymorphic N terminus in human vitamin D receptor isoforms influences transcriptional activity by modulating interaction with transcription factor IIB. *Mol Endocrinol* 2000; **14**: 401–420.
156 Means TK, Wang S, Lien E, Yoshimura A, Golenbock DT, Fenton MJ. Human toll-like receptors mediate cellular activation by *Mycobacterium tuberculosis*. *J Immunol* 1999; **163**: 3920–3927.
157 Lien E, Sellati TJ, Yoshimura A *et al*. Toll-like receptor 2 functions as a pattern recognition receptor for diverse bacterial products. *J Biol Chem* 1999; **274**: 33419–33425.
158 Underhill DM, Ozinsky A, Smith KD, Aderem A. Toll-like receptor-2 mediates mycobacteria-induced proinflammatory signaling in macrophages. *Proc Natl Acad Sci USA* 1999; **96**: 14459–14463.
159 Thoma-Uszynski S, Stenger S, Takeuchi O *et al*. Induction of direct antimicrobial activity through mammalian toll-like receptors. *Science* 2001; **291**: 1544–15447.
160 Kang TJ, Chae GT. Detection of Toll-like receptor 2 (TLR2) mutation in the lepromatous leprosy patients. *FEMS Immunol Med Microbiol* 2001; **31**: 53–58.
161 Lorenz E, Mira JP, Cornish KL, Arbour NC, Schwartz DA. A novel polymorphism in the toll-like receptor 2 gene and its potential association with staphylococcal infection. *Infect Immun* 2000; **68**: 6398–6401.
162 Garred P, Madsen HO, Kurtzhals JA *et al*. Diallelic polymorphism may explain variations of the blood concentration of mannan-binding protein in Eskimos, but not in black Africans. *Eur J Immunogenet* 1992; **19**: 403–412.
163 Garred P, Harboe M, Oettinger T, Koch C, Svejgaard A. Dual role of mannan-binding protein in infections: another case of heterosis? *Eur J Immunogenet* 1994; **21**: 125–131.
164 Hoal-Van Helden EG, Epstein J, Victor TC *et al*. Mannose-binding protein B allele confers protection against tuberculous meningitis. *Pediatr Res* 1999; **45** (Pt 1): 459–464.
165 Garred P, Richter C, Andersen AB *et al*. Mannan-binding lectin in the sub-Saharan HIV and tuberculosis epidemics. *Scand J Immunol* 1997; **46**: 204–208.
166 Bellamy R, Ruwende C, McAdam KP *et al*. Mannose binding protein deficiency is not associated with malaria hepatitis B carriage nor tuberculosis in Africans. *Qjm* 1998; **91**: 13–18.
167 Schorey JS, Carroll MC, Brown EJ. A macrophage invasion mechanism of pathogenic mycobacteria. *Science* 1997; **277**: 1091–1093.
168 Selvaraj P, Narayanan PR, Reetha AM. Association of functional mutant homozygotes of the mannose binding protein gene with susceptibility to pulmonary tuberculosis in India. *Tuber Lung Dis* 1999; **79**: 221–227.
169 Marquet S, Abel L, Hillaire D *et al*. Genetic localization of a locus controlling the intensity of infection by Schistosoma mansoni on chromosome 5q31–q33. *Nat Genet* 1996; **14**: 181–184.
170 Muller-Myhsok B, Stelma FF, Guisse-Sow F *et al*. Further evidence suggesting the presence of a locus, on human chromosome 5q31–q33, influencing the intensity of

infection with *Schistosoma mansoni*. *Am J Hum Genet* 1997; **61**: 452–454.
171 Garcia A, Marquet S, Bucheton B *et al*. Linkage analysis of blood *Plasmodium falciparum* levels: interest of the 5q31–q33 chromosome region. *Am J Trop Med Hyg* 1998; **58**: 705–709.
172 Cookson WO, Ubhi B, Lawrence R *et al*. Genetic linkage of childhood atopic dermatitis to psoriasis susceptibility loci. *Nat Genet* 2001; **27**: 372–373.
173 Nair RP, Henseler T, Jenisch S *et al*. Evidence for two psoriasis susceptibility loci (HLA and 17q) and two novel candidate regions (16q and 20p) by genome-wide scan. *Hum Mol Genet* 1997; **6**: 1349–1356.
174 Trembath RC, Clough RL, Rosbotham JL *et al*. Identification of a major susceptibility locus on chromosome 6p and evidence for further disease loci revealed by a two stage genome-wide search in psoriasis. *Hum Mol Genet* 1997; **6**: 813–820.
175 Lander E, Kruglyak L. Genetic dissection of complex traits: guidelines for interpreting and reporting linkage results. *Nat Genet* 1995; **11**: 241–247.
176 Siddiqui MR, Meisner S, Tosh K *et al*. A major susceptibility locus for leprosy in India maps to chromosome 10p13. *Nat Genet* 2001; **27**: 439–441.
177 Taylor ME, Drickamer K. Structural requirements for high affinity binding of complex ligands by the macrophage mannose receptor. *J Biol Chem* 1993; **268**: 399–404.
178 Schlesinger LS, Hull SR, Kaufman TM. Binding of the terminal mannosyl units of lipoarabinomannan from a virulent strain of *Mycobacterium tuberculosis* to human macrophages. *J Immunol* 1994; **152**: 4070–4079.
179 Prigozy TI, Sieling PA, Clemens D *et al*. The mannose receptor delivers lipoglycan antigens to endosomes for presentation to T cells by CD1b molecules. *Immunity* 1997; **6**: 187–197.
180 Tosh K, Meisner S, Siddiqui MR, Balakrishnan K, Ghei S, Golding M *et al*. A region of chromosome 20 is linked to leprosy susceptibility in a South Indian population. *J Infect Dis* 2002; **186** (8).