ARTHRITIS ASSOCIATED WITH MUCOSAL INFECTIONS

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SUMMARY

Infectious agents which are well tolerated by the majority of those infected can give rise to persistent pathology in predisposed individuals. Inflammatory arthritis complicates certain infections of the gastrointestinal and genitourinary tract in a minority of those infected; this is reactive arthritis, which is in turn related to other forms of inflammatory arthritis - the spondyloarthropathies (e.g. ankylosing spondylitis) - in which host responses to commensal bacteria rather than specific infectious agents may well be important. Reactive arthritis lends itself to clinical investigation, since it is an acute arthritis triggered by known organisms. Thus persistence and distribution of the organism, together with immune responses to it, can be characterised in detail. Since timing of both the triggering infection and the onset of arthritis are well defined, this facilitates studies of the evolution and outcome of disease.

A current view of the immunopathogenesis of reactive arthritis highlights several findings: Firstly, although the triggering organism cannot be cultured from affected joints, there is good evidence that bacterial antigens and, in at least some cases, transcriptionally active bacteria, reach the synovium of the inflamed joints. Secondly, vigorous T-cell mediated responses to the triggering organism are readily detected in affected joints; analysis of the bacterial antigens targeted by the immune response can be used to identify differences in patients with arthritis and uncomplicated infection. Lastly, various genetic and environmental factors influence the likelihood of infected individuals developing reactive arthritis, and its severity or persistence. The best known of these is HLA-B27, but additional genes including those that play a role in other spondyloarthropathies are involved. There is now some evidence that these genes may determine the nature of the immune response mounted to the triggering bacterium in reactive arthritis, or to commensal bacteria in other spondyloarthropathies.

Lessons gained from the study of reactive arthritis and other spondyloarthropathies may be applied to other diseases associated with infectious agents in which host factors, particularly genes, determine outcome, in contrast to diseases whose course primarily reflects the pathogenic properties of the organism.

INTRODUCTION

For most of the 20th century the field of infectious diseases has been concerned with the diagnosis and treatment of those bacterial and, to a lesser extent,

viral infections which pose an immediate challenge to host survival through multiplication of the organism and, in many instances, the induction of inflammatory responses which themselves damage organs and tissues (e.g. septic shock). Whilst septic shock remains a major cause of mortality, the effects of many of the classical pathogens infections have been dealt with very satisfactorily through the development of antibiotics for bacterial infections, and mainly through vaccine strategies for life-threatening viral infections. In the last 2-3 decades attention has shifted to pathogens which have a more subtle interaction with the host. Such infections are typically very common or even ubiquitous in human populations, with the majority of individuals sustaining either brief self-limiting illnesses (e.g. influenza, food poisoning) or no obvious clinical damage from the infectious agent. Indeed, some of these organisms persist indefinitely in the host without any apparent clinical effects. Obvious examples of this would be the herpes viruses such as Epstein-Barr virus and Cytomegalovirus, and in the case of bacteria, organisms such as *Helicobacter pylori* and *Chlamydia pneumoniae*. Nevertheless, whilst these organisms are not associated with any of the kinds of events which are seen in acute sepsis, it has become clear that there are still pathological consequences of the infection, but that these vary in different individuals. Thus, in the case of *Helicobacter pylori*, many patients maintain chronic infection of the gastric mucosa with minimal clinical effects (other than perhaps more dyspepsia than they would otherwise have suffered), but in others the presence of the organism leads to the development of peptic ulcer, and in another subset, to neoplasms such as gastric lymphoma (*Blaser*, 1990). Epstein-Barr virus infection provides an even more striking example: There is near ubiquitous infection of human populations which is normally asymptomatic, unless the virus is acquired in adolescence, when it often causes infectious mononucleosis. In all subjects the virus persists, because it is exquisitely adapted to an almost silent existence in memory B-cells (*Thorley-Lawson* and *Gross*, 2004). However, immunosurveillance is still required to keep it in check since iatrogenic immunosuppression (usually in the context of transplantation) can result in virus-induced B-cell lymphoproliferative disease (*Loren* et al., 2003). Many of these cases can be brought under immunologic control by removing immunosuppressive drugs, but in others outgrowth of malignant B-cells occurs. In addition, natural infection with the virus contributes substantially to the occurrence of Hodgkin's lymphoma and undifferentiated nasopharyngeal carcinoma, although these neoplasms can also occur without viral infection (*Rickinson* et al., 2000). Thus, at different times and in different individuals the host-virus relationship varies from one which is entirely benign as far as the host is concerned to one which is potentially fatal. Much current interest in infectious diseases concerns those situations in which a stable truce between organism and host is replaced by an outbreak of hostilities and the appearance of disease.

For organisms which are able to persist without causing the host's demise, it is important to determine which aspects of the host-pathogen relationship determine whether or not clinical disease occurs. This general paradigm for persistent pathogens can be extended to a consideration of the relationship between host and commensal organisms, particularly bacteria, and the diseases which might result if these relationships are upset. These issues are particularly well illustrated by the association between inflammatory arthritis and infections at various sites, especially the gut.

THE RELATIONSHIP BETWEEN INFECTION OR INFLAMMATION AT 'BARRIER' SITES, AND ARTHRITIS

In addition to the well-recognised ability of particular organisms such as staphylococci or streptococci to disseminate to the joint and cause septic arthritis, there are a number of diseases in which arthritis occurs in relation to either specific infections, or to inflammation at sites where the body encounters commensal bacteria. It is worth noting however that the confidence with which bacteria can be implicated in each of these diseases varies.

Gut inflammation

Examples of arthritis which occurs in the context of gut infection or inflammation include Whipple's Disease, reactive arthritis following enteric infection, spondyloarthropathy complicating ulcerative colitis or Crohn's disease, and arthritis associated with coeliac disease (*Gaston* and *Lillicrap*, 2003). These diseases illustrate a spectrum: Whipple's Disease can almost be regarded as an example of septic arthritis, albeit with a very slow growing organism, *Tropheryma whippelii* (*Relman* et al., 1992), since the organism can be found in joint tissue in an apparently similar state to the gut, i.e. within macrophages (*O'Duffy* et al., 1999). In reactive arthritis, organisms such as *Salmonella* and *Campylobacter* cannot be cultured from the joint but may be readily cultured in stool; nevertheless, there is good evidence that the organism reaches the joint (vide infra). Both ulcerative colitis and Crohn's disease are clearly associated with certain forms of inflammatory arthritis, and both involve inflammation at sites where bacteria are present in large numbers. Furthermore, the normal state of tolerance which is extended towards gut commensals seems to be broken in Crohn's disease (*Duchmann* et al., 1995; *Lodes* et al., 2004). Finally, coeliac disease, due to intolerance for the gliadin fraction of gluten and predominantly involving areas of the gut which are bacteria free, has been associated with arthritis, although this is generally rather mild (*Lubrano* et al., 1996).

Skin

Examples include Lyme disease due to infection with the specific pathogen *Borrelia burgdorferi* or related organisms, which is inoculated into the skin by a tick bite. This produces a characteristic rash which can later be followed by an inflammatory arthritis and other forms of inflammation distant from the skin (*Steere*, 2001). Much more common than Lyme disease is the arthritis which complicates psoriasis, a disease in which specific pathogens are not generally implicated (with the exception of cases of guttate arthritis related to streptococcal infection), but the relationship with the skin flora is disturbed by inflammation.

Urinary Tract

Whilst urinary tract infection is not generally associated with inflammatory arthritis, it is important to note that infection with *Chlamydia trachomatis* produces a clinical syndrome which is indistinguishable from enteric reactive arthritis (*Gaston*, 2000). In addition, an iatrogenic form of reactive arthritis has been repeatedly described when patients have intravesical installation of BCG organisms for the treatment of bladder cancer (*Miossec*, 1996). Thus, particular organisms in the urogenital tract can also predispose to arthritis.

Respiratory tract

The range of associations here also includes specific pathogens such as streptococci, which trigger both the arthralgia of rheumatic fever and a post streptococcal arthritis, and the reactive

arthritis due to the respiratory pathogen, *Chlamydia pneumoniae* (*Deighton*, 1993; *Braun* et al., 1994; *Hannu* et al., 1999), although this infection is a much less common cause of reactive arthritis than *Chlamydia trachomatis* in the urogenital tract. Diseases such as cystic fibrosis, which involve chronic infection and bronchiectasis, have also been asso-ciated with inflammatory arthritis (*Merkel*, 1999; *Bradlow* and *Mowat*, 1983).

Whilst it is useful to point out the association between inflammatory arthritis and infection at each of these different sites, the rest of this review will concentrate mainly on infection/inflammation in the gut, together with *Chlamydia*-induced reactive arthritis.

THE PATHOGENESIS OF REACTIVE ARTHRITIS

The term "reactive arthritis" is sometimes used loosely to refer to any form of arthritis which follows infection of any kind. However, it is more accurate to confine its use to a specific clinical syndrome, sometimes erroneously called "Reiter's disease", which follows infection with a relatively small number of organisms: In the gastro-intestinal tract, *Salmonella, Campylobacter, Yersinia* and *Shigella*, and in the genito-urinary tract *Chlamydia trachomatis*. A long list of other organisms can occasionally produce the same syndrome, but these five account for the vast majority of cases. The clinical syndrome is classified as a spondyloarthropathy because it share clinical features with other members of this family of arthropathies which comprises: Ankylosing spondylitis, arthritis associated with inflammatory bowel disease and psoriatic arthritis. These include involvement of the spine and entheses (sites of attachment of tendons and ligaments to bone), and extra-articular features such as psoriaform rashes, inflammation of the uveal tract, and gastro-intestinal inflammation. Other forms of arthritis following infection do not show features of spondyloarthropathy and are best termed "post-infectious arthritis".

Progress in our understanding of the pathogenesis of reactive arthritis has occurred in three main areas:
1. Demonstration of the triggering organism or its components in affected joints;
2. Characterisation of the immune response to reactive arthritis triggering organisms, particularly the response within the joints;
3. Exploration of the genes and environmental factors which determine the occurrence of spondyloarthropathies, including reactive arthritis, and their severity.

Bacterial antigens and/or bacteria are present in the reactive arthritis joint

By definition the reactive arthritis joint is sterile, i.e. bacteria cannot be isolated by conventional culture techniques. The first hint that this might not be the end of the story came from electron microscopy studies of synovium in *Chlamydia*-induced arthritis in which *Chlamydia* inclusion bodies, albeit with atypical morphology, were demonstrated (*Ishikawa* et al., 1986). Inevitably, since the organism was not cultivatable the precise status of these electron microscopy findings was much debated. However, it was soon shown that synovial biopsies from reactive arthritis patients stained with *Chlamydia*-specific antibodies (*Keat* et al., 1987). Analogous findings were soon reported by researchers at the University of Turku, Finland, who developed polyclonal and monoclonal antisera specific for enteric organisms known to trigger reactive arthritis. In an important series of studies

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they showed the presence of *Yersinia*, *Salmonella* and *Shigella* in synovial fluid and tissue of reactive arthritis patients (*Granfors* et al., 1989, 1990, 1992; *MerilahtiPalo* et al., 1991). In many cases these bacterial components were demonstrable in the joint many weeks after enteric infection, highly suggestive of organism persistence, although stool cultures were often negative at the same time that the organism was detected in the joint. The antisera identified both bacterial proteins and components of the bacterial cell wall such as LPS, but could not determine whether live or dead organisms were present. Even the presence of dead organisms many weeks after initial infection would be suggestive of persistence of bacterium, perhaps at another site such as the gastrointestinal lymphoid system, from whence dead organisms might traffic to the joint. The cells which contain bacterial antigenic material were both neutrophils and macrophage/monocytes and, in addition to demonstrating these findings in the joint, positive staining for bacterial antigens was evident in the same cells in peripheral blood of some patients. The findings based on immunofluoresence were backed up by immunoblotting studies. Together these findings raise the possibility that the disease might at least in its initial stages (1-2 years) be driven by bacterial antigens and reflect unusual persistence with the organism. Additional evidence for persistence came from serological studies showing that patients with reactive arthritis had longer lasting titres of IgA antibodies to the organisms, a finding pointing to persistence since IgA antibodies have a relatively rapid half-life as compared to IgG (*Granfors* and *Toivanen*, 1986).

The final phase in demonstrating organisms at the site of disease utilised PCR technology. Both PCR to demonstrate organisms-specific DNA and, more recently, RT-PCR to demonstrate organism-derived RNA, have been used.

Most of the published data concern detection of *Chlamydia trachomatis*, with occasion reports of detection of *Yersinia* (*Gaston* et al., 1999) and possibly *Salmonella* (*Ekman* et al., 1999; Nikkari et al., 1999. Although PCR for detection of bacteria in reactive arthritis is technically demanding and standardised protocols have not yet been agreed, the weight of evidence suggests that *Chlamydia* nucleic acids are indeed present in the joints of affected cases (*Rahman* et al., 1992; *Taylor-Robinson* et al., 1992; *Bas* et al., 1995; *Schnarr* et al., 2001). There is also evidence to suggest that the quantity of specific nucleic acids present is very small. Thus, careful experimental protocols had to be devised for the handling of synovial fluid and synovial tissue, as compared to, for instance, urine or genitourinary swabs and, although this reflects the different tissue, it almost certainly also reflects the increased sensitivity which is required (*Kuipers* et al., 2002). In addition, in longitudinal studies of particular patients, not all samples of synovial fluid are positive, suggesting that the amount of nucleic acid may hover between undetectable and just detectable levels. It has even been suggested that one explanation for the discrepancies found when several experienced laboratories examine the same sample of synovial fluid could relate to the presence of a small quantity of organisms and nucleic acids so that not all aliquots from one sample will be positive. An additional cause of concern and confusion is that when the same PCR techniques have been applied to patients with other forms of arthropathy, the results are not uniformly negative (*Wilkinson* et al., 1998; *Schumacher* et al., 1999a; *Olmez* et al., 2001). However, although at first sight disconcerting, this is in fact what would be expected, if the joint material which is being examined comes from a population in which the prevalence of *Chlamydia* infection is high. There is no mechanism which

could readily be postulated to prevent the access of *Chlamydia* to the inflamed synovium of a patient with, e.g. rheumatoid arthritis, who acquires *Chlamydia* infection. The main study which showed a significant rate of detection of *Chlamydia* nucleic acids in arthropathies other than reactive arthritis still showed a substantially higher prevalence of positive PCR tests for *C. trachomatis* in the reactive arthritis population (*Schumacher* et al., 1999b). Interestingly, the prevalence of *C. pneumoniae* in the same synovial material did not vary much between reactive arthritis patients and those with other forms of arthropathy. This is consistent with *C. pneumoniae* rarely triggering reactive arthritis, and detecting this organism acts as a kind of internal control in the study indicating that dissemination of organisms to inflamed joints can occur (especially organisms which persist long-term), whether or not the organism is causative of the arthritis. In a recent study this dissemination has been emphasized by the observations that, when RT-PCR is performed using universal primers, almost all inflamed synovia contain significant bacterial ribosomal RNA, mainly derived from species known to be commensal in the gut or skin (*Kempsell* et al., 2000; *van der Heijden* et al., 2000; *Cox* et al., 2003). The results from these studies and others are consistent with the idea that there is a continuing traffic of bacteria through joints, most likely within phagocytes. The more phagocytes recruited to the joint, the higher the likelihood of these organisms and their components reaching the joint. Thus, normal synovium which recruits only small numbers of macrophages to the synovial membrane, was found not to contain bacterial rRNA, but all inflamed synovia, including those from osteoarthritic joints (which is often surprisingly inflamed,) were positive for bacterial rRNA. If this notion is correct, then the traffic of reactive arthritis associated bacteria to the joint does not in itself explain the occurrence of reactive arthritis if this traffic would be expected to happen, at least to some extent, under normal circumstances. It might however provide some insight into why reactive arthritis is commoner in large weight bearing joints. One would postulate the rate of macrophage traffic to such joints, which are subject to micro- trauma in every day activities, to be higher than to non-weight bearing joints, with a greater probability that reactive arthritis bacteria will find their way to those sites. Indeed patients occasionally comment that a joint which was recently injured was one of the first to flare in an episode of reactive arthritis.

Immune responses to reactive arthritis associated bacteria

Studies of T-cell mediated immune responses to organisms associated with reactive arthritis have been studied for more than 20 years, beginning with the landmark studies of Denys Ford (*Ford* et al., 1985). It was apparent early on that very marked T-cell proliferative responses to the organisms responsible for triggering disease were readily detectable in synovial fluid (*Gaston* et al., 1989; *Sieper* et al., 1993; *Hermann* et al., 1990). These studies have been extended, and indeed the synovial fluid has proved a useful source of organism-specific T-cells which have been cloned and used to characterise the immune response to organisms such as *Chlamydia trachomatis* and *Yersinia enterocolitica* (*Hassell* et al., 1993; *Deane* et al., 1997; *Mertz* et al., 1998; *Hermann* et al., 1989). There was an initial hope that measurements of responses to bacterial antigens in synovial fluid might be useful diagnostically, but further consideration of the reasons why such responses should be so prominent in synovial fluid and synovium casts doubt on this possibility. First of all, it is clear that there is preferential recruitment of memory T-

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cells into sites of inflammation (*Thomas* et al., 1992; *Akbar* et al., 2000), such as the joints affected by reactive arthritis. Virtually all of the cells in the inflamed joint express CD45RO, an isoform which is characteristic of memory T-cells, with little expression of CD45RA which is expressed by naive cells (*Matthews* et al., 1993). Therefore, one would anticipate an enrichment in the joint of T-cells responding to any major antigenic challenge which the patient has previously experienced, and indeed T-cell responses to recall antigens such as PPD and tetanus toxin are also readily recorded. Given that reactive arthritis patients have recently been infected with organisms like *Chlamydia* and *Salmonella* it is not surprising to find prominent responses to these organisms in the affected joints. Furthermore, the antigen presenting cells within the joint are enriched for activated dendritic cells, which again enhances the proliferative responses which can be recorded *in vitro* (*Harding* and *Knight*, 1986; *Viner* et al., 1993; *Stagg* et al., 1991). In the light of these considerations, it can be concluded that a vigorous T-cell proliferative response to a reactive arthritis associated organism will normally be present in reactive arthritis patients, but might also be recorded in patients with arthritis due to other causes, if these patients have also experienced infection in the past by one or other of the reactive arthritis associated organisms.

However, whilst for these reasons measurements of synovial T-cell responses to bacteria are of limited use diagnostically, they may still be relevant to the pathogenesis of inflammation. The responses obtained are generally of the 'Th1' kind, i.e. T-cells which make the pro-inflammatory cytokine interferon-γ, in addition to other pro-inflammatory factors such as TNF-α and IL-17 (*Simon* et al., 1993; *Schlaak* et al.,

1992). Indeed, T-cell obtained from synovial fluid make these same cytokines spontaneously *ex vivo*, consistent with their having been activated *in vivo* (*BeacockSharp* et al., 1998). Nevertheless, there are also reports of IL-4 producing T-cells in reactive arthritis synovium and whilst these are in the minority, their presence has led to speculation that the immune response to the organism in the joint is not adequately polarised to Th1, and may therefore allow persistence of the organism (*Simon* et al., 1994). Pathogens such as *Chlamydia* require an interferon-γ producing immune response for clearance of the organism - although paradoxically in certain circumstances interferon-γ may itself drive an organism into a persistent state (*Beatty* et al., 1993). This occurs when interferon-γ induces the enzyme IDO which degrades intracellular tryptophan (*Beatty* et al., 1994). Since Chlamydiae cannot synthesize this amino acid their replication ceases, but there is evidence that they can enter a quiescent state in which they still transcribe a number of genes and can therefore still act as a stimulus to the immune system which could drive persistent joint inflammation (*Gerard* et al., 1996, 2001, 2002).

Much of the research on T-cell responses in reactive arthritis was founded on the hope that bacteria-specific T-cells would be identified which cross-reacted with human proteins expressed at sites of disease (joints or entheses), Thus far this had not been achieved, with the vast majority of T-cells showing specificity for bacterial antigens and no consistent cross-reactivity with self proteins. This general idea- "molecular mimicry" has attracted some criticism recently (*Benoist* and *Mathis*, 2001), though an interesting example involving *Helicobacter pylori* has recently been reported (*Amedei* et al., 2003).

Genetic and environmental influences on the occurrence and severity of reactive arthritis

Three major genetic influences on the occurrence and severity of reactive arthritis can be discussed: (a) HLA-B27; (b) MHC encoded genes other than B27; (c) non-MHC genes; (d) bacterial genes; (e) environmental factors

HLA-B27

The incidence of HLA-B27 in many series of reactive arthritis patients has been reported to be nearly as high as that in ankylosing spondylitis: 60-70% of patients were positive, as compared to approximately 95% of those with ankylosing spondylitis (*Aho* et al., 1973). However, these figures reflect selection of patients with relatively severe reactive arthritis who are referred to secondary care. Other surveys of patients developing reactive arthritis following outbreaks of food poisoning, which include patients with very mild symptoms, have shown much lower incidences of HLA-B27, although the majority have still shown that B27 increases susceptibility to developing the disease. One such survey recently showed a clear effect of HLA-B27 on disease severity, with 75% of the patients who developed definite reactive arthritis being B27+ as compared to around 30% of those who developed any kind of musculo-skeletal symptom following *Salmonella* infection (*Ekman* et al., 2000). It is possible to conclude therefore that B27 positivity is not essential to the development of reactive arthritis, but increases its severity and therefore the likelihood that the disease will present clinically.

The mechanism whereby B27 confers increased susceptibility to reactive arthritis is unknown, and indeed the role of HLA-B27 in the pathogenesis of spondyloarthropathies in general has been much debated for 30 years, with no definitive answer at this time (*Gaston*, 1990). An initial attractive hypothesis built on the paradigm of molecular mimicry as an explanation for autoimmunity. This was first demonstrated by showing that antibodies formed in response to infectious agents could bind to self-antigens because the epitopes recognised on antigens from the pathogen 'mimicked' epitopes on normal cell proteins. This idea could be modified by suggesting that pathogen specific HLA-B27 restricted CD8+ T-cells might also recognise peptides derived from self-proteins presented by HLA-B27. Whilst B27 restricted T-cells which recognise reactive arthritis associated pathogens have been described (*Hermann* et al., 1993; *Matyszak* and *Gaston*, 2004), and autoreactive B27 specific CD8 T-cells have also been seen in spondyloarthropathy patients (*Fiorillo* et al., 2000; *Frauendorf* et al., 2003), there is no clear demonstration of molecular mimicry to confirm that this is the main mechanism underlying the association between HLA-B27 and spondyloarthropathy. In recent years, attention has shifted to some extent to the nature of the B27 molecule itself. This has several properties which are not shared with most other Class I MHC alleles including an ability to be expressed on the cell surface in the absence of tapasin (*Peh* et al., 1998), a relatively slow rate of folding in the endoplasmic reticulum and the formation of haemodymeric structures related to a cysteine molecule at position 67 (*Mear* et al., 1999; *Colbert*, 2000). There are several possible consequences from these abnormalities: The slow rate of folding in the endoplasmic reticulum also leads to the accumulation of misfolded B27 heavy chains which is an instigator of a stress response within cells. This could have consequences for how antigen presenting cells interact with T-cells (and this has recently been shown to be defective (*Hacquard-Bouder* et al., 2004), or how the same cells cope with intracellular infection. Alternatively, the expression of surface

B27 molecules which either contain no antigenic peptide or peptides which have not been edited by the TAP/tapasin mechanism may lead to abnormal presentation of self-peptides, with the possibility of breaking self tolerance. In addition, because these B27 molecules containing sub-optimal peptides are relatively unstable they may acquire exogenously self, or even pathogen related, peptides which would not otherwise be presented. Lastly, the formation of B27 homodimers, or expression of free heavy chains, presents an opportunity both for their recognition by autoreactive T-cells (*Boyle* et al., 2001, 2004) or their interaction with other receptors on T-cells such as the KIR family of receptors (*Allen* et al., 1999; *Allen* and *O'Callaghan*, 2004). Which of any of these mechanisms is involved in susceptibility to reactive arthritis and severity of the disease remains unknown.

MHC molecules other than B27

HLA typing studies have shown a remarkable similarity between patients with reactive arthritis due to enteric infection, patients with inflamed joints related to exacerbations in inflammatory bowel disease, and patients with ankylosing spondylitis in the context of inflammatory bowel disease (*Orchard* et al., 2000). This is not confined to HLA-B27 but has been shown to involve other HLA Class I and Class II alleles. The association between spondyloarthropathies and B27 almost certainly relates to the B27 molecule itself - the best evidence for this is the occurrence of disease in B27 transgenic rodents (*Hammer* et al., 1990). However, the association between spondyloarthropathy and other MHC alleles may well be explained by linkage to equilibrium between these alleles and other genes in the MHC which modulate immune and inflammatory responses. The nature of these genes has yet to be elucidated but since the MHC region has now been entirely sequenced and contains a number of candidate genes, this question should be settled in the near future (*Beck* et al., 1999). Obvious candidates would include the genes for TNF-α which is clearly a predominant cytokine in spondyloarthropathy, judged by the striking clinical efficacy of drugs which inhibit TNF (*Braun* et al., 2002).

Non-MHC genes

The importance of these has been clear for some time in ankylosing spondylitis where the frequency of disease is much higher in B27+ individuals who have a first degree relative with ankylosing spondylitis as compared to B27+ individuals with no such relatives. The nature of the genes involved is under intensive investigation by whole genome surveys in large groups of patients from multicase families. Whilst some of these genes will relate to ankylosing spondylitis specifically, there may be others which play a part in other forms of spondyloarthropathy. It has also been noted that B27 associated diseases 'breed true', i.e. those with B27 and a family history of reactive arthritis have a higher risk of reactive arthritis rather than ankylosing spondylitis, and conversely for those with B27 and relatives with ankylosing spondylitis. It is, however, striking that inflammation in the gut or in the skin can in some ways 'substitute' for HLA-B27 in the development of ankylosing spondylitis, since, in the absence of these factors 95% of patients are B27+, whereas if these diseases co-exist only 50% of the patients are B27+. Therefore, the genes which underlie psoriasis and inflammatory bowel disease are worth attention, and particular interest has been aroused by the recent description of the CARD 15 molecule which is associated with Crohn's Disease and possibly also with psoriatic arthritis (*McGovern* et al., 2001; *Rahman* et al., 2003). Given that this molecule has properties suggesting

that it modulates the response to bacteria products such as LPS, and other molecules detected at the-cell surface by Toll-like receptors (*Pauleau* and *Murray*, 2003), it may be worthwhile looking for similar molecules which would be associated with reactive arthritis and/or ankylosing spondylitis. CARD 15 itself does not appear to be associated with ankylosing spondylitis (*van der Paardt* et al., 2003). Likewise IL-11 has recently been linked to ulcerative colitis (*Klein* et al., 2002); IL-11 can down-regulate the response to LPS which requires signalling through NFκB.

Bacterial genes

Little is known about the specific bacterial genes which influence the occurrence of reactive arthritis, but these much exist since markedly different rates of arthritis are seen after infection with similar organisms. It has already been noted that *Chlamydia pneumoniae* is a much less common trigger than *Chlamydia trachomatis*, and the same applies to *Chlamydia psittaci*. More striking still is the difference between *Shigella sonnei* and *Shigella flexneri*, with the former only rarely reported as a trigger of arthritis (*Lauhio* et al., 1988). Genetically all Shigellae resemble *E. coli* which also does not trigger reactive arthritis.

Environmental factors

These clearly play a role in spondyloarthropathies including reactive arthritis. Thus, in ankylosing spondylitis concordance for disease is not 100% in monozygotic twins (*Brown* et al., 1997; *Jarvinen*, 1995). The role of environmental factors in reactive arthritis is not well defined but two pieces of evidence are worth considering:

The influence of HIV on the incidence of reactive arthritis and other forms of spondyloarthropathy

Prior to the epidemic of HIV in sub-Saharan Africa, reactive arthritis was very rare despite a high incidence of infection with triggering organisms such as *Shigella* and *C. trachomatis*. HLA-B27 is virtually absent form many of these populations, and this might have been taken to be a protective factor. However, since the appearance of HIV patients with reactive arthritis have been commonly reported (*Njobvu* et al., 1998; *Njobvu* and *McGill*, 1999, 2000). This suggests that there is a genetic background in these populations which allows reactive arthritis to occur (or is not protective), but this has only been revealed by the alterations in the immune system brought about by HIV infection.

The "Chlamydia paradox"

The incidence of *Chlamydia* infection in Western countries continues to rise and this is probably not just due to better diagnostic methods. However, no rise in the incidence of *Chlamydia*-triggered reactive arthritis has been seen in these countries, and indeed these cases are somewhat uncommon. This paradox is unexplained. However, investigations of the T-cell-mediated response to *C. trachomatis* have mapped epitopes which are identical in *C. pneumoniae* (*Deane* et al., 1997; *Goodall* et al., 2001). This raises the question of whether prior infection with *C. pneumoniae* might make reactive arthritis more likely by "priming" the immune system to make excessive responses to the infection. This concept has not been proven, but the prevalence of *C. pneumoniae* infection is relatively low in the cohort most likely to acquire *C. trachomatis* (teens and

twenties). The general concept that previous antigenic experience, due to infection with related organisms, might affect the likelihood of post-infectious inflammatory diseases is worthy of further investigation.

CONCLUSIONS

Reactive arthritis illustrates how bacterial infection of the gut or genito-urinary tract can result in severe and occasionally chronic inflammation at distant sites. Although many questions remain about its pathogenesis, host genes which affect the immune response to the organism are implicated, together perhaps with the host's previous antigenic experience.

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