Chapter 11

Summary and General Discussion

The purpose of this thesis was to expand the knowledge of prognostic factors and outcome in early arthritis. The studies were performed using data from the early arthritis clinic (EAC) of the Jan van Breemen Institute, a large regional rheumatology clinic. The EAC was started in 1995 and has ongoing inclusions. More than 2000 patients have been included and followed since then. Demographic, clinical and laboratory data was used to gain more insight into the distinct nature the two autoantibody systems rheumatoid factor (RF) and antibodies to citrullinated proteins (ACPA), and to assess the prevalence of comorbidity in patients with RA.

Chapter 2 examined whether single nucleotide polymorphisms (SNPs) and haplotypes in immune response genes and human leukocyte antigen (HLA) class II alleles are associated with radiographic progression in patients with early undifferentiated arthritis (UA). The increase in joint damage after two years follow-up was assessed on radiographs in Dutch Caucasians with early UA. The SE and interleukin (IL) 10 GGC haplotype were associated with severe progression, defined as an increase in Sharp/van der Heijde Score ≥5 points. The TNF-308 allele was associated with less radiologic damage, which was an unexpected finding, since this allele is associated with increased TNF-α production.

Chapters three to six concern ACPA. In **chapter 3** the frequency of the SE alleles and ACPA levels were compared in patients with arthralgia, early RA en established RA. When compared to the general population, patients with arthralgia were found to have a higher frequency of SE positivity, but when compared to early or established RA a lower frequency of SE positivity was found. Higher ACPA levels were associated with the presence of the SE alleles but only in arthralgia patients and not in early or established RA patients. Furthermore ACPA levels in arthralgia patients carrying the SE alleles were comparable to the levels found in early or established RA patients carrying the SE alleles. The results suggest that in an early stage the presence of the SE alleles may influence the risk for RA through an effect on ACPA levels.

To confirm earlier findings of the stability of ACPA over time, the increased IgM-RF frequency with age, and the correlation of IgM-RF with the acute phase response, a repository of serum samples of the Jan van Breemen Institute was used in **chapter 4**. In more than 22,000 samples from approximately 18,000 patients, RF and ACPA levels had been determined. The diagnosis was extracted from the diagnosis registration system. It appeared that in patients with RA ACPA status was more stable than RF status. Patients with another diagnosis than rheumatoid arthritis and negative for RF or ACPA seldom became positive. ACPA status was not related to age, but a positive RF status increased with age in patients with a diagnosis other than RA. The correlation between RF and ACPA levels and inflammatory factors was generally low.

In **Chapter 5** it was investigated whether RF or ACPA levels at baseline, and changes in the levels in the following year, are associated with disease activity and functional and radiological outcomes. After one year of follow-up, the median ACPA level had decreased by 31% compared to baseline levels and RF levels had decreased by 56%. But a change from negative to positive status of ACPA or RF was very rare. A positive autoantibody status was associated with the degree of joint damage after two years of follow-up. Changes in levels in the first year were not associated with outcome measures. It was concluded that ACPA and RF levels in the first year do not provide additional information on prediction of outcome at two years compared to the autoantibody status. It is more the presence than the levels of the autoantibodies that counts. Except for a change in RF status from positive to negative, autoantibody status is stable. It therefore does not seem useful to repeat these measurements in patients with early RA.

ACPA comprise a group of antibodies of which antibodies against cyclic citrullinated peptide (anti-CCP) are the most commonly used in clinical practice. Antibodies against mutated citrullinated vimentin (anti-MCV) is another ACPA. **Chapter 6** examined the association between RA disease activity and anti-MCV in patients with early RA or UA during the first two years follow-up. Both the anti-MCV levels and DAS28 decreased clearly in the first and second year. But an association between the anti-MCV and the DAS was very low. The specificity of the anti-MCV test was 92.3% and the sensitivity was 59.3%, and of the anti-CCP test 92.1 and 55.3%, respectively. Patients with a positive anti-MCV status had more joint damage on average at every visit and higher mean levels of inflammatory factors compared to anti-MCV negatives. To monitor the level of disease activity based on anti-MCV levels is not useful.

Chapters 7 to 10 address additional features in early RA. The effect of gender on the incidence and course of rheumatoid arthritis (RA) is complex and there is still debate about whether female sex is a marker of a more severe disease course. Whether there are gender differences in patients with early RA concerning disease activity, functional status, radiographic damage and the use of medication was determined in **chapter 7**. The same analyses were performed in two cohorts, in the EAC of the Jan van Breemen institute and in the two initial monotherapy groups of the BeSt trial. In both cohorts women have more disease activity during follow-up. In the EAC cohort, women had more functional impairment, whereas functionality was similar between genders in the BeSt trial. Furthermore, women receive more medication than men, while the radiographic progression rate is similar in the two groups. Additional other measures than medication may be needed to lower the higher burden of disease that is experienced by women.

Generalized osteoporosis is a well-known complication of RA. In **chapter 8** the prevalence of vertebral deformities in elderly patients with RA was examined, in relation to demographic and clinical variables. In 29% of a group of 98 patients aged ≥60 years vertebral deformities were found, which is higher than in the healthy population. Seven percent had two or more deformities. The prevalence of vertebral deformities was related to high age, RF positivity and low BMD at the hips. In women ESR was also associated with vertebral deformities. No significant difference was found at the lumbar spine.

In RA there often is early involvement of the hand and wrist, causing pain, limited range of motion and/or loss of muscle strength. It is important to gain insight into the sequence of these features in the hand and wrist and how they develop over time in order to provide adequate patient care. The aim of **chapter 9** was to determine the prevalence of hand and wrist symptoms, impairments and activity limitations in relation to disease duration in RA patients. Of all patients 94% suffered from at least one hand or wrist symptom, such as complaints of pain, stiffness, loss of muscle strength, etc. and 80% had at least one hand or wrist impairment. The most common impairment was stenosing tenosynovitis, which was present in 33% of the patients. Hand-related activity limitations were assessed with the Disability of Arm, Shoulder and Hand questionnaire (DASH). The median standardized DASH score of the arthritis patients was significantly higher than the normative DASH score for the general population. In **chapter 10** early predictors of stenosing tenosynovitis in the hand and hand-related activity limitations in patients with RA were identified. Stenosing tenosynovitis was predicted by difficulties with use of the hands indicated by the HAQ category score for grip (HAQ-hand) at the two-year follow-up. Hand-related activity limitations, defined as a DASH score above the normative score plus twice the SD, were present in 30% of the patients. Hand-related activity limitations were predicted by a high DAS28 and the presence of HAQ-hand at the 2-year follow-up.

Discussion

A better understanding of the complex aetiology of RA will eventually lead to improved treatment of the disease. Central and mutually related factors in this respect are the genetic basis of RA and the role of autoimmunity.

Genetic and serological factors

Previous studies have shown that patients with RA can have a genetic predisposition which is also related to the prognosis of the disease.[1-3] The 'shared epitope' (SE) alleles are associated with the development of RA and with more joint damage, which is confirmed in several reports[4,5] as well as in this thesis. Beside the SE alleles there are other genetic factors that

contribute to a different radiographic progression between patients,[6-9] but the prognostic value of genetic factors is not always obvious. This is illustrated by the finding that the Tumour Necrosis Factor (TNF)-308A allele is associated with less radiographic damage in this thesis, while previous research associated it with an increased TNF-α production,[10] leading to more joint damage. Additionally TNF-308A is in linkage disequilibrium with the DQA1*05-DQB1*02 haplotype, which is associated with a mild disease.[11] This might suggest that different genetic factors interact with each other or even overrule each other. Therefore the effect of a single SNP cannot always be interpreted and a (more) complete picture of all involved SNPs is needed. Due to genome-wide association studies (GWAS), more loci are identified, in a recent meta-analysis of GWAS seven new loci were identified.[12] At this time approximately 31 loci have been identified as susceptibility markers for RA in people of European ancestry,[12,13] though these loci are predominantly associated with the subgroup of ACPA positive patients. Specific research is needed to find susceptibility loci for the subgroup of ACPA negative patients.

In earlier reports it was suggested that there is an interaction between the SE alleles and ACPA.[14-16] The contribution of the SE alleles to the development of arthritis is probably mediated by ACPA, this was found both in a cross-sectional and a prospective study.[17] The finding that only in ACPA positive arthralgia patients the SE is associated with a higher ACPA level, and not in RA patients, suggests that the SE only operates on ACPA levels in the early phase up to early arthritis.

ACPA status is an early, highly specific and stable marker for RA, indicative of an adverse disease course. Previous research has shown that ACPA are present in the preclinical phase of the disease[18] and precede IgM-RF in the preclinical phase.[19] ACPA has a higher specificity for RA compared to IgM-RF, distinguishing RA from other rheumatic diseases.[20,21] Prospective follow-up of RA patients showed that in time ACPA levels may decrease by a third to a half compared to the baseline measurement, however, this decrease in ACPA levels seldom leads to a change in status. ACPA status is more stable in time and with increasing age than IgM-RF status,[22-24] which was confirmed in a large group of RA patients. Patients with early arthritis also demonstrate differences in prognosis depending on initial ACPA status,[25-27] prospective follow-up added to this knowledge: antibodies against both CCP and MCV are similarly associated with increased radiographic progression. These data contribute to the suggestion that RA can be classified into two subsets by ACPA status.

While autoantibody status is a clear marker of disease outcome, this is not clear for autoantibody levels. Baseline levels of ACPA or IgM-RF are only moderately associated with radiographic progression and are not associated with other outcome measures. In particular,

changes in levels in the first year of disease were not associated with changes in disease outcome.[28] ACPA levels might be predictive in a subset of IgM-RF positive patients,[29] but at the moment there is insufficient information that autoantibody levels are useful as a marker of disease outcome in addition to knowledge of autoantibody status.

ACPA and IgM-RF represent two different autoantibody systems. ACPA appears to have a pathophysiological role in RA,[30,31] whereas IgM-RF is associated with infection and might be a consequence of the rheumatoid inflammation. IgM-RF levels correlate better with markers of inflammation than ACPA levels. ACPA levels respond less to anti-TNF treatment than IgM-RF levels and ACPA changes are not associated with disease activity unlike IgM-RF.[24,32] These results support the suggestion that ACPA is a disease specific marker, while IgM-RF is more a marker of inflammation.[33] Therefore interventions directed to suppress inflammation alone might not be enough to achieve control of the disease.

Additional features

Besides genetic and serological factors the disease course of RA is also influenced by other factors. Three additional topics, namely gender, hand function and osteoporosis were studied in the present thesis. In addition to previous reports it can be concluded that women have more disease activity then men, which leads to more frequent treatment changes in women compared to men. The net result is a similar radiological progression. Additional interventions may be necessary to decrease the higher disease burden that women with RA experience. Hormonal interventions have been tried with varying success.

The finding of frequent hand impairments in early RA, mainly flexor tendon synovitis, opens the way to develop specific interventions for this complication. Similarly, due to the frequent occurrence of osteoporosis in early RA, there is a need for more alertness for this comorbidity.

Conclusion

The results of this thesis support the emerging main division of RA into ACPA positive and negative disease subsets. Future research should search for a genetic basis for ACPA negative disease. For ACPA positive early disease optimal treatment needs to be established to further increase the rate of remission. Since ACPA appear to have pathogenetic significance, future interventions should not only focus on improved anti-inflammatory therapy, but also be directed at the modulation of the ACPA response itself. Future improvement of outcome and decreased effects of comorbidity will, as before, mainly depend on a better management of the underlying disease RA itself.

References

1. Griffin AJ, Wooley P, Panayi GS, Batchelor JR. HLA DR antigens and disease expression in rheumatoid arthritis. Ann Rheum Dis 1984;43[2]:218-21.
2. Furst DE. Predictors of worsening clinical variables and outcomes in rheumatoid arthritis. Rheum Dis Clin North Am 1994;20[2]:309-19.
3. Carrier N, Cossette P, Daniel C, Brum-Fernandes A, Liang P, Menard HA et al. The DERAA HLA-DR alleles in patients with early polyarthritis: protection against severe disease and lack of association with rheumatoid arthritis autoantibodies. Arthritis Rheum 2009;60[3]:698-707.
4. Emery P, Salmon M, Bradley H, Wordsworth P, Tunn E, Bacon PA et al. Genetically determined factors as predictors of radiological change in patients with early symmetrical arthritis. BMJ 1992;305[6866]:1387-9.
5. Mattey DL, Hassell AB, Dawes PT, Cheung NT, Poulton KV, Thomson W et al. Independent association of rheumatoid factor and the HLA-DRB1 shared epitope with radiographic outcome in rheumatoid arthritis. Arthritis Rheum 2001;44[7]:1529-33.
6. Doherty DG, Vaughan RW, Donaldson PT, Mowat AP. HLA DQA, DQB, and DRB genotyping by oligonucleotide analysis: distribution of alleles and haplotypes in British caucasoids. Hum Immunol 1992;34[1]:53-63.
7. Brinkman BM, Huizinga TW, Kurban SS, van der Velde EA, Schreuder GM, Hazes JM et al. Tumour necrosis factor alpha gene polymorphisms in rheumatoid arthritis: association with susceptibility to, or severity of, disease? Br J Rheumatol 1997;36[5]:516-21.
8. Lard LR, van Gaalen FA, Schonkeren JJ, Pieterman EJ, Stoeken G, Vos K et al. Association of the -2849 interleukin-10 promoter polymorphism with autoantibody production and joint destruction in rheumatoid arthritis. Arthritis Rheum 2003;48[7]:1841-8.
9. Khanna D, Wu H, Park G, Gersuk V, Gold RH, Nepom GT et al. Association of tumor necrosis factor alpha polymorphism, but not the shared epitope, with increased radiographic progression in a seropositive rheumatoid arthritis inception cohort. Arthritis Rheum 2006;54[4]:1105-16.
10. Bouma G, Crusius JB, Oudkerk PM, Kolkman JJ, von Blomberg BM, Kostense PJ et al. Secretion of tumour necrosis factor alpha and lymphotoxin alpha in relation to polymorphisms in the TNF genes and HLA-DR alleles. Relevance for inflammatory bowel disease. Scand J Immunol 1996;43[4]:456-63.
11. Verpoort KN, van Gaalen FA, van der Helm-van Mil AH, Schreuder GM, Breedveld FC, Huizinga TW et al. Association of HLA-DR3 with anti-cyclic citrullinated peptide antibody-negative rheumatoid arthritis. Arthritis Rheum 2005;52[10]:3058-62.
12. Stahl EA, Raychaudhuri S, Remmers EF, Xie G, Eyre S, Thomson BP et al. Genome-wide association study meta-analysis identifies seven new rheumatoid arthritis risk loci. Nat Genet 2010;42[6]:508-14.
13. Plant D, Flynn E, Mbarek H, Dieude P, Cornelis F, Arlestig L et al. Investigation of potential non-HLA rheumatoid arthritis susceptibility loci in a European cohort increases the evidence for nine markers. Ann Rheum Dis 2010;69[8]:1548-53.
14. Hill JA, Southwood S, Sette A, Jevnikar AM, Bell DA, Cairns E. Cutting edge: the conversion of arginine to citrulline allows for a high-affinity peptide interaction with the rheumatoid arthritis-associated HLA-DRB1*0401 MHC class II molecule. J Immunol 2003;171[2]:538-41.
15. van der Helm-van Mil AH, Verpoort KN, Breedveld FC, Huizinga TW, Toes RE, de Vries RR. The HLA-DRB1 shared epitope alleles are primarily a risk factor for anti-cyclic citrullinated peptide antibodies and are not an independent risk factor for development of rheumatoid arthritis. Arthritis Rheum 2006;54[4]:1117-21.
16. Kaltenhauser S, Pierer M, Arnold S, Kamprad M, Baerwald C, Hantzschel H et al. Antibodies against cyclic citrullinated peptide are associated with the DRB1 shared epitope and predict joint erosion in rheumatoid arthritis. Rheumatology (Oxford) 2007;46[1]:100-4.

Bos WH, Wolbink GJ, Boers M, Tijhuis GJ, de Vries N, van der Horst-Bruinsma IE et al. Arthritis development in arthralgia patients is strongly associated with anti-citrullinated protein antibody

status: a prospective cohort study. Ann Rheum Dis 2009.
18. Rantapää-Dahlqvist S, de Jong BA, Berglin E, Hallmans G, Wadell G, Stenlund H et al. Antibodies against cyclic citrullinated peptide and IgA rheumatoid factor predict the development of rheumatoid arthritis. Arthritis Rheum 2003;48[10]:2741-9.
19. Nielen MM, van Schaardenburg D, Reesink HW, van de Stadt RJ, van der Horst-Bruinsma IE, de Koning MH et al. Specific autoantibodies precede the symptoms of rheumatoid arthritis: a study of serial measurements in blood donors. Arthritis Rheum 2004;50[2]:380-6.
20. Avouac J, Gossec L, Dougados M. Diagnostic and predictive value of anti-cyclic citrullinated protein antibodies in rheumatoid arthritis: a systematic literature review. Ann Rheum Dis 2006;65[7]:845-51.
21. Nishimura K, Sugiyama D, Kogata Y, Tsuji G, Nakazawa T, Kawano S et al. Meta-analysis: diagnostic accuracy of anti-cyclic citrullinated peptide antibody and rheumatoid factor for rheumatoid arthritis. Ann Intern Med 2007;146[11]:797-808.
22. Bobbio-Pallavicini F, Caporali R, Alpini C, Moratti R, Montecucco C. Predictive value of antibodies to citrullinated peptides and rheumatoid factors in anti-TNF-alpha treated patients. Ann N Y Acad Sci 2007;1109:287-95.
23. Vis M, Bos WH, Wolbink G, Voskuyl AE, Twisk JW, van de Stadt RJ et al. IgM-rheumatoid factor, anti-cyclic citrullinated peptide, and anti-citrullinated human fibrinogen antibodies decrease during treatment with the tumor necrosis factor blocker infliximab in patients with rheumatoid arthritis. J Rheumatol 2008;35[3]:425-8.
24. Bos WH, Bartelds GM, Wolbink GJ, de Koning MH, van de Stadt RJ, van Schaardenburg D et al. Differential response of the rheumatoid factor and anticitrullinated protein antibodies during adalimumab treatment in patients with rheumatoid arthritis. J Rheumatol 2008;35[10]:1972-7.
25. Jansen LM, van Schaardenburg D, van dH-B, I, van der Stadt RJ, de Koning MH, Dijkmans BA. The predictive value of anti-cyclic citrullinated peptide antibodies in early arthritis. J Rheumatol 2003;30[8]:1691-5.
26. van Gaalen FA, Linn-Rasker SP, van Venrooij WJ, de Jong BA, Breedveld FC, Verweij CL et al. Autoantibodies to cyclic citrullinated peptides predict progression to rheumatoid arthritis in patients with undifferentiated arthritis: a prospective cohort study. Arthritis Rheum 2004;50[3]:709-15.
27. Nielen MM, van der Horst AR, van Schaardenburg D, van der Horst-Bruinsma IE, van de Stadt RJ, Aarden L et al. Antibodies to citrullinated human fibrinogen (ACF) have diagnostic and prognostic value in early arthritis. Ann Rheum Dis 2005;64[8]:1199-204.
28. Rönnelid J, Wick MC, Lampa J, Lindblad S, Nordmark B, Klareskog L et al. Longitudinal analysis of citrullinated protein/peptide antibodies (anti-CP) during 5 year follow up in early rheumatoid arthritis: anti-CP status predicts worse disease activity and greater radiological progression. Ann Rheum Dis 2005;64[12]:1744-9.
29. Miriovsky BJ, Michaud K, Thiele GM, O'Dell JR, Cannon GW, Kerr G et al. Anti-CCP antibody and rheumatoid factor concentrations predict greater disease activity in men with rheumatoid arthritis. Ann Rheum Dis 2010;69[7]:1292-7.
30. Klareskog L, Stolt P, Lundberg K, Kallberg H, Bengtsson C, Grunewald J et al. A new model for an etiology of rheumatoid arthritis: smoking may trigger HLA-DR (shared epitope)-restricted immune reactions to autoantigens modified by citrullination. Arthritis Rheum 2006;54[1]:38-46.
31. Sebbag M, Chapuy-Regaud S, Auger I, Petit-Texeira E, Clavel C, Nogueira L et al. Clinical and pathophysiological significance of the autoimmune response to citrullinated proteins in rheumatoid arthritis. Joint Bone Spine 2004;71[6]:493-502.
32. De Rycke L, Verhelst X, Kruithof E, Van den BF, Hoffman IE, Veys EM et al. Rheumatoid factor, but not anti-cyclic citrullinated peptide antibodies, is modulated by infliximab treatment in rheumatoid arthritis. Ann Rheum Dis 2005;64[2]:299-302.
33. Klareskog L, Rönnelid J, Lundberg K, Padyukov L, Alfredsson L. Immunity to citrullinated proteins in rheumatoid arthritis. Annu Rev Immunol 2008;26:651-75.