Editorial

An important step towards completing the rheumatoid arthritis cycle

Walther J van Venrooij and Ger JM Pruijn

Department of Biomolecular Chemistry, Radboud University Nijmegen, NL-6500 HB Nijmegen, The Netherlands

Corresponding author: Walther J van Venrooij, w.vanvenrooij@ncmls.ru.nl

Published: 19 September 2008
This article is online at http://arthritis-research.com/content/10/5/117

Arthritis Research & Therapy 2008, **10:**117 (doi:10.1186/ar2504)

See related research article by Zhao *et al*., http://arthritis-research.com/content/10/4/R94

Abstract

In the previous issue of *Arthritis Research & Therapy* data are presented showing that circulating immune complexes containing citrullinated fibrin(ogen) are present in anti-citrullinated protein antibody-positive rheumatoid arthritis patients, and that such immune complexes co-localize with complement factor C3 in the rheumatoid synovium. These results corroborate the idea that citrullination is intimately involved in the pathophysiology of rheumatoid arthritis and complete our model (the rheumatoid arthritis cycle) for the development and chronic nature of this disease.

Rheumatoid arthritis (RA) is a chronic, progressive, inflammatory autoimmune disease characterized by the presence of several autoantibodies. Among the autoantibodies detected in RA, only rheumatoid factor and anti-citrullinated protein/peptide antibodies (ACPAs) are considered clinically useful. Both the measurement of antibodies directed against cyclic citrullinated peptides (CCP) and rheumatoid factor determinations are commonly used in daily clinical practice. Because of the extreme specificity of ACPAs, we and others have previously proposed that these antibodies might be involved in the pathophysiology of RA. Since then, experimental evidence supporting several pieces of the RA cycle (Figure 1) has been obtained. In the previous issue of *Arthritis Research & Therapy*, Zhao and colleagues [1] fill in one of the last gaps in our knowledge on the cycle of events leading to RA. The authors utilized complement C1q to capture immune complexes (ICs) from plasma and protein G to capture ICs from pannus tissue derived from human RA and control patients, and demonstrated that ICs containing citrullinated fibrin(ogen) were present in about 50% of anti-CCP-positive patients. They were not detectable in RA patients lacking these antibodies and control patients with other autoimmune diseases. In the rheumatoid synovium, fibrin(ogen)-containing ICs co-localize with complement factor C3, suggesting that they contribute to synovitis in a subset of patients.

There are many citrullinated proteins in the inflamed RA synovium [2] - for example, vimentin, histones, collagen types I and II, and α-enolase - but citrullinated fibrin(ogen) is certainly one of the most abundant and important antigens [3]. The fact that only half of the anti-CCP(+) patients possess ICs with citrullinated fibrin(ogen), however, illustrates the heterogeneity in the ACPA repertoire. It is likely that ICs in the other anti-CCP(+) patients contain one or more of the other citrullinated antigens. The important study of Zhao and collaborators cements into place one of the last pieces of the RA cycle, as depicted in Figure 1 and discussed by us and others previously [4-6]. In this model for the development and chronicity of RA at least five steps can be distinguished.

Step 1

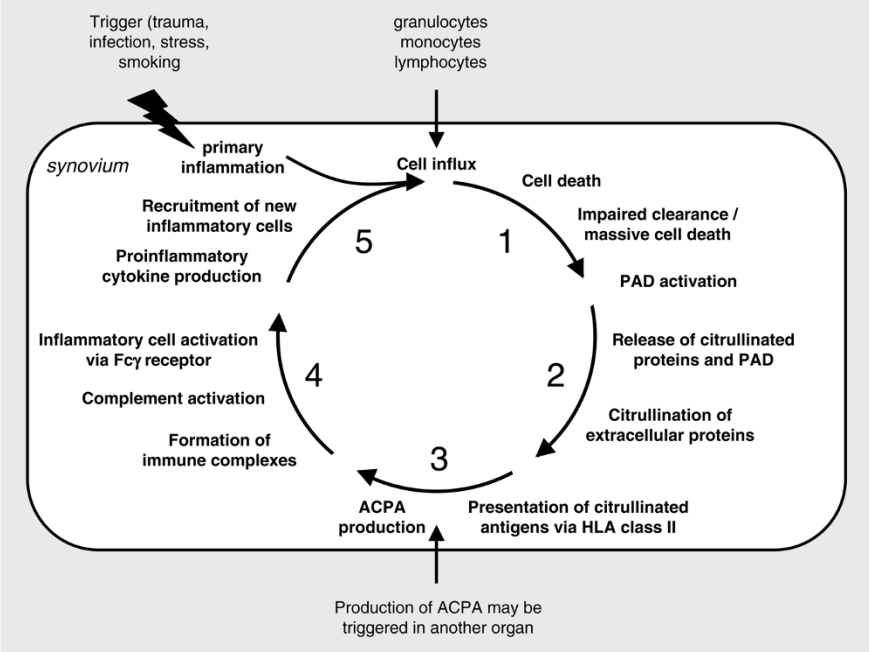
An in itself innocent inflammation of the joint or other tissue in the body leads to infiltration of inflammatory cells (granulocytes, monocytes, lymphocytes). In normal situations many of the infiltrating cells will die via apoptosis and will be cleared (removed) by phagocytes. However, when there is massive apoptosis, for example due to a toxicant or infection, or a (genetic) defect in the clearance system, some apoptotic cells may become necrotic. Granulocytes and monocytes, and macrophages emerging from monocyte differentiation, contain citrullinating peptidylarginine deiminase (PAD) enzymes. These enzymes are activated by the elevation of cytosolic Ca^{2+}-concentrations, for example when cells undergo apoptosis.

Step 2

When the dying inflammatory cells are not cleared properly and become necrotic, they release the intracellular citrullinated proteins (for example, histones, vimentin) and the activated PAD enzymes. These enzymes, PAD2 and/or PAD4, can then citrullinate extracellular synovial proteins like fibrin(ogen).

ACPA = anti-citrullinated protein/peptide antibody; CCP = cyclic citrullinated peptide; IC = immune complex; PAD = peptidylarginine deiminase; RA = rheumatoid arthritis.

Figure 1



The rheumatoid arthritis (RA) cycle. Model for the role of protein citrullination in the pathophysiology of RA. The various aspects of the five major steps are depicted. Step 1, entry and death of inflammatory cells in the synovium; step 2, peptidylarginine deiminase (PAD) activation and protein citrullination; step 3, immune response to citrullinated antigens; step 4, formation of citrullinated immune complexes and their effects; step 5, recruitment of new inflammatory cells. ACPA, anti-citrullinated protein/peptide antibody.

However, the mere presence of citrullinated proteins will not necessarily lead to chronic inflammation because in 99% of individuals these citrullinated proteins are degraded without a (humoral) reaction of the immune system [4]. The presence of a large number of citrullinated proteins in the inflamed synovium has been shown by several groups (for a recent review, see [2]).

Step 3

In those individuals who are able to present citrullinated fragments of proteins to T cells via certain HLA molecules, an immune response to citrullinated antigens might be generated, resulting in the production of high affinity IgG ACPAs [7]. Activation of autoreactive B cells may occur locally in the (inflamed) joint, but may also occur in other inflamed tissues. Via the circulation, these ACPAs or the plasma cells producing them will ultimately enter the joint, for example when a local inflammation occurs or as a result of immune complex-facilitated vascular leakage [6]. Irrespective of the site of B cell activation, there is experimental evidence that ACPAs are produced in RA joints and may mediate tissue injury [2,8].

Recent support for a mechanism as described above has been provided by Hill and collaborators [9]. They showed that citrullinated fibrinogen is able to induce arthritis in DR4-IE transgenic mice. T cell epitope scanning and antibody microarray analysis identified a unique pattern of citrulline-specific reactivity that was not found in DR4-IE transgenic mice immunized with unmodified fibrinogen or in wild-type C57BL/6 mice immunized with citrullinated fibrinogen. These observations directly implicate citrullinated fibrinogen as arthritogenic in the context of RA-associated major histocompatibility (MHC) class II molecules [9]. It is quite likely that this also holds for other citrullinated antigens, such as vimentin and histones.

Step 4

After their production in/entry into the inflamed synovium, ACPAs can react with the abundantly available citrullinated antigens. The paper of Zhao and collaborators [1] is the first to show the presence of such ICs in circulating plasma as well as in the inflamed synovium. These ICs stimulate the inflammatory process by activation of the complement system and further recruitment and activation of granulocytes, monocytes, and macrophages via both complement receptor- and Fcγ receptor-dependent pathways. In this way, ACPAs contribute to the perpetuation of joint inflammation and to the chronicity and severity of RA.

Step 5

New monocytes and granulocytes will enter the synovium, where they will be activated, subsequently die and release another load of activated PAD enzymes. A new round of

citrullinated antigens and ACPA production will take place, leading to a new flare of inflammation. It has been noted that novel IgM-producing B cells are continuously recruited to the inflamed RA joint, demonstrating that the ACPA response is continuously reactivated during the course of arthritis [10].

The continuation of this vicious circle for years, eventually accompanied by fresh traumas or environmental events that stimulate inflammation, will ultimately lead to a chronic inflammation that manifests as the disease we know as RA.

The paper of Zhao and collaborators proves one of the last suppositions of the RA cycle. This model will allow us to design new strategies to interfere with the chronic cycle of events in the inflamed joints, which may ultimately lead to the development of new and more specific therapies for RA.

Competing interests

The authors declare that they have no competing interests.

References

1. Zhao X, Okeke NL, Sharpe O, Batliwalla FM, Lee AT, Ho PP, Tomooka BH, Gregersen PK, Robinson WH: **Circulating immune complexes contain citrullinated fibrinogen in rheumatoid arthritis.** *Arthritis Res Ther* 2008, **10:**R94.
2. Van Venrooij WJ, Van Beers J, Pruijn GJM: **Anti-CCP antibody, a marker for the early detection of rheumatoid arthritis.** *Ann NY Acad Sci* 2008, in press.
3. Masson-Bessière C, Sebbag M, Girbal-Neuhauser E, Nogueira L, Vincent C, Senshu T, Serre G: **The major synovial targets of the rheumatoid arthritis-specific antifilaggrin autoantibodies are deiminated forms of the alpha- and beta-chains of fibrin.** *J Immunol* 2001, **166:**4177-4184.
4. Vossenaar ER, Van Venrooij WJ: **Citrullinated proteins: sparks that may ignite the fire in rheumatoid arthritis.** *Arthritis Res Ther* 2004, **6:**107-111.
5. Vossenaar ER, Zendman AJ, Van Venrooij WJ: **Citrullination, a possible functional link between susceptibility genes and rheumatoid arthritis.** *Arthritis Res Ther* 2004, **6:**1-5.
6. van Gaalen F, Ioan-Facsinay A, Huizinga TW, Toes RE: **The devil in the details: the emerging role of anticitrulline autoimmunity in rheumatoid arthritis.** *J Immunol* 2005, **175:**5575-5580.
7. Hill JA, Wang D, Jevnikar AM, Cairns E, Bell DA: **The relationship between predicted peptide-MHC class II affinity and T-cell activation in a HLA-DRbeta1*0401 transgenic mouse model.** *Arthritis Res Ther* 2003, **5:**R40-R48.
8. Kuhn KA, Kulik L, Tomooka B, Braschler KJ, Arend WP, Robinson WH, Holers VM: **Antibodies against citrullinated proteins enhance tissue injury in experimental autoimmune arthritis.** *J Clin Invest* 2006, **116:**961-973.
9. Hill JA, Bell DA, Brintnell W, Yue D, Wehrli B, Jevnikar AM, Lee DM, Hueber W, Robinson WH, Cairns E: **Arthritis induced by posttranslationally modified (citrullinated) fibrinogen in DR4-IE transgenic mice.** *J Exp Med* 2008, **205:**967-979.
10. Verpoort KN, Jol-van der Zijde CM, Papendrecht-van der Voort EA, Ioan-Facsinay A, Drijfhout JW, van Tol MJ, Breedveld FC, Huizinga TW, Toes RE: **Isotype distribution of anti-cyclic citrullinated peptide antibodies in undifferentiated arthritis and rheumatoid arthritis reflects an ongoing immune response.** *Arthritis Rheum* 2006, **54:**3799-3808.