Rapid detection of anti-citrullinated protein antibodies in autoimmune patients

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Anti-citrullinated protein antibodies (ACPA) are the most specific serological marker of rheumatoid arthritis (RA). Several ACPA-detection assays are available for clinical use, which are almost all based on ELISA(-like) assays with citrullinated peptides (CCP2). To facilitate ACPA-detection in low-volume laboratories and resource-poor environments, we aimed to develop a rapid and easy to perform test. An agglutination mediator was generated by protein engineering. Addition of this mediator to (diluted) blood samples results in hemagglutination when ACPA are present, which can be detected by the naked eye. A single-chain antibody fragment that binds to glycophorin A, one of the major surface proteins of erythrocytes was conjugated to an ACPA-binding citrullinated peptide. The applicability was assessed by the analysis of fresh blood samples from 200 RA patients and from 100 psoriatic arthritis (PsA) patients as a control group. The addition of the mediator resulted in detectable agglutination in 48-61% of the RA samples. Agglutination correlated well with the results obtained with a commercial anti-CCP2 ELISA (63-67%). Efficient agglutination was observed with only 9% of the PsA samples. We conclude that the agglutination mediator allows the rapid and efficient detection of ACPA by hemagglutination in human blood samples.