INFECTION WITH CITRULLINATING PORPHYROMONAS GINGIVALIS CAN INDUCE AUTOIMMUNITY TO HUMAN RIBOSOMAL PROTEINS AND TNF ALPHA TREATMENT NONRESPONSE

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Background: Porphyromonas gingivalis (P.g) is involved in triggering self-reactive immune responses when cirtullinating bacterial or human proteins. However, first evidence to link anti-ribosomal T and B cells responses to rheumatoid arthritis (RA) has been published but the mechanism and its influence on therapy is not clear (1). Infection based autoimmunity induced by cirtullination of human proteins with P g. peptidyl arginine deiminase from RA patient (RA-PPAD) and crossreactivity binding induced by P g. was investigated using patient sera, affinity purified RA patient antibodies and monoclonal antibodies to cit-RA-PPAD.

Objectives: Antibodies to RA-PPAD isolated from an RA patients (RA-PPAD) was first time linked to target specific cirtullinated ribosomal proteins and therapy.

Methods: Screening of RA sera was conducted on 37.830 unique human proteins on protein macroarrays (http://www.engine-gmbh.de) with 30 RA sera. The autoantibody response to 840 different proteins was recorded and bioinformatically evaluated. Protein arrays were cirtullinated with human peptidyl arginine deiminase PAD 2, 4, rabbit PAD and RA-PPAD from P.g. Sera and affinity purified antibodies were used to detect reactivity to 840 autoantigens and 15aa CCP peptide form RA-PPAD. Sera from TBA treated sera anti-TNF (adalimumab, etanercept, certolizumab) treatment were tested with the cit-RA-PPAD-peptide of 15aa (CPP).

Results: A human protein macroarray consisting of 840 identified autoantigens from RA patients was modified by human PAD2 and PAD4, rabbit PAD and RA-PPAD form P g. Using cit specific monoclonal antibodies we identified the ribosomal proteins (RP), RPL18a, RPS27a, modified by PAD2, RPL18a and MRPS11 modifies by PAD4, and RPL7L1 modified by rabbit PAD specifically targeted. In addition 6 RA patient sera and 3 collagen arthritis (OA) control sera were used to identify the cirtullinated RA-PPAD specific modified autoantigens not targeted when modified by human PAD2 or PAD4 or rabbit PAD. We identified the RA-PPAD cirtullinated ribosomal Proteins RPL3, RPL21, RPS24, RPL9, RPL15, RPS24, RPS3a, MRPL28 specifically targeted by RA patients. This identifies ribosomal proteins as major specific RA-PPAD cirtullination targets. Moreover, affinity purified antibodies bound to native and cirtullinated RA-PPAD from 6 RA patient sera and 3 OA patient sera were tested for crossreactivity on cirtullinated human proteinarray. Antibodies to cirtullinated ribosomal proteins MRPS11, RPL21, RPS3a, RPL18a, RPS27a, MRPL28 were detected in the RA group but not in the OA control group. High antibody titre against the cit-PPAD-peptide of 15aa (CPP) derived from the auto- cirtullination site (R63) of RA-PPAD correlates with TNF-alpha-inhibitor (TBA) non-response (n=61), DMARD patients refractory to different treatment regimes (n=61), receiving anti-TNF (adalimumab, etanercept, certolizumab), do not respond when maintaining high α-CPP IgG level.

Conclusion: Failure of Porphyromonas gingivalis clearance in RA patients leads to infection induced enzymatic mimicry based autoreactivity targeting evolutionary conserved human ribosomal proteins. Autoimmunity to ubiquitous self-antigens may trigger localized tissue damage in RA.TBA non-response leads to the suggestion to clear Porphyromonas gingivalis infection before α-TNF treatment.

REFERENCE

BARICITINIB IMPROVES JOINT MOBILITY AFTER INJURY IN A RODENT FORCED-AMBULATION MODEL

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Background: Movement-evoked pain and impaired joint mobility are common comorbidities in inflammatory diseases such as Rheumatoid Arthritis (RA) and Osteoarthritis. The Janus kinase (JAK) pathway has been implicated in both inflammation and chronic pain. Clinical data suggests that baricitinib, a selective JAK 1/2 inhibitor, can robustly and rapidly alleviate pain in RA and OA. Therefore, we hypothesized that treatment with baricitinib would improve joint function in both inflammation and chronic pain.

Methods: Unilateral joint injury was induced in female Sprague Dawley rats (Harlan, Indianapolis, IN, USA) by unilateral intra-articular injection of 20 μg Complete Freund’s Adjuvant (CFA). Using the GaitScan (CleverSys Inc., Reston, VA) treadmill system, a composite gait score comprising of range of motion, normalized stance distance, stance/swing ratio, and paw print size was evaluated over 3 days post-injection. 2 rats were treated with vehicle, positive control (40 mg/kg Tramadol), or clinically relevant doses of baricitinib (1, 3, or 10 mg/kg p.o., 2-hrs prior to each test, q.d.). Dorsal root ganglion (DRG) homogenates were harvested post-gait evaluation and Total STAT3 (Cell Signaling, #4904) and phospho-STAT3 (Y705) (Cell Signaling, #9131) protein levels were examined via immunoblotting. The p-values were derived from repeated measures ANOVAs.

Results: In rat DRG homogenates, baricitinib significantly decreased phospho-STAT3 (Y705) protein levels in a dose-dependent manner (p<0.01) by Day 3. These data support a role for JAK-STAT signaling in pain signaling and provide an opportunity to investigate the potential mechanism of action of baricitinib in joint pain.