This is a repository copy of Innate Lymphoid Cells are Present at Normal Human Enthesis Providing a Potential Mechanism for Spondyloarthritis Pathogenesis.

White Rose Research Online URL for this paper:
http://eprints.whiterose.ac.uk/103206/

Version: Accepted Version

**Proceedings Paper:**

https://doi.org/10.1136/annrheumdis-2016-eular.2270

(c) 2016, EULAR/BMJ Publishing Group. This is an author produced version of a paper published in Annals of the Rheumatic Diseases. Uploaded in accordance with the publisher's self-archiving policy.

**Reuse**
Unless indicated otherwise, fulltext items are protected by copyright with all rights reserved. The copyright exception in section 29 of the Copyright, Designs and Patents Act 1988 allows the making of a single copy solely for the purpose of non-commercial research or private study within the limits of fair dealing. The publisher or other rights-holder may allow further reproduction and re-use of this version - refer to the White Rose Research Online record for this item. Where records identify the publisher as the copyright holder, users can verify any specific terms of use on the publisher’s website.

**Takedown**
If you consider content in White Rose Research Online to be in breach of UK law, please notify us by emailing eprints@whiterose.ac.uk including the URL of the record and the reason for the withdrawal request.
Innate Lymphoid Cells are Present at Normal Human Enthesis Providing a Potential Mechanism for Spondyloarthritis Pathogenesis

1. RJ Cuthbert, EM Fragkakis, P Millner, R Dunsmuir, Y El-Sherbiny, D McGonagle

1. 1Leeds Institute of Rheumatic and Musculoskeletal Medicine, University of Leeds, Leeds, UK
2. 2Department of Spinal Surgery, National Health Service, Leeds, UK

Background and objectives

The pathogenesis of murine spondyloarthritis (SpA) has been intimately linked to the presence of IL-23 responsive, innate like lymphocytes at peripheral and spinal enthesis. Human SpAs are associated with SNPs in genes related to the IL-23 pathway and drugs that block IL-12/23 have shown efficacy. We hypothesised that the normal human enthesis has a population of resident innate lymphoid cells (ILCs) that could be key in governing entheseal immune homeostasis partly via interaction with resident mesenchymal stromal cells (MSCs).

Materials and methods

Normal spinal enthesis were harvested from patients undergoing spinal decompression surgery and enzymatically digested prior to sorting or flow cytometry. Immunophenotyping and cell sorting was performed on enthesis samples harvested from 6 patients and unmatched peripheral blood. The expression of RORγt and key immunomodulatory transcripts was tested in sorted populations by RTqPCR. Anterior cruciate ligament and Achilles enthesis were obtained from patients with knee OA and Achilles tendon rupture and analysed by immunohistochemistry (IHC). Adherent cells from entheseal digest were cultured under standard MSC culture conditions and expression of known MSC markers was assessed by flow cytometry.

Results

All sorted samples contained ILC3s, median proportion 0.09% (range 0.015-0.63). Transcript analysis confirmed the expression of RORγt transcript in sorted ILC3 populations. ILC3s expressed 51-fold greater relative expression of RORγt in comparison to unsorted mononuclear cells. 5 of 6 sorted samples contained ILC2s, median proportion 0.20% (range 0-0.49). RORγt expression was detected in knee OA and there was widespread expression of RORγt in inflammatory infiltrates in injured enthesis as shown by IHC. Culture expanded adherent cells grew in characteristic fibroblastoid colonies and expressed phenotypic markers consistent with bone marrow derived MSCs.

Conclusions
Our findings show that both ILCs and MSCs are present in the normal human spinal enthesis. ILCs may also be greatly increased in frequency following injury. The co-localisation of ILC and MSC populations at the enthesis suggests a potential link between cellular dysregulation of the IL-23/17 axis and human SpA pathology.