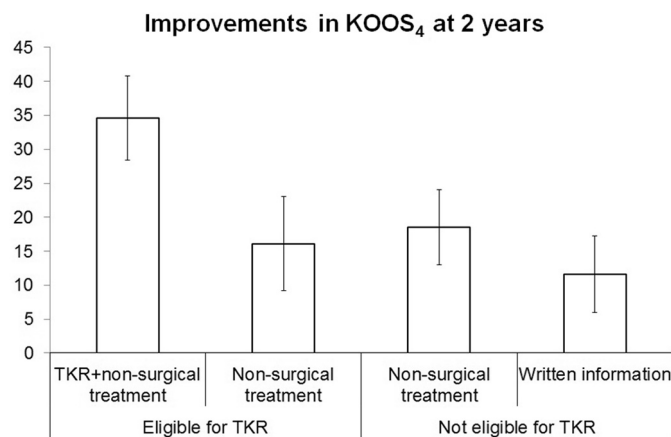


two years in the mean score of four of the five KOOS subscales, covering pain, symptoms, activities of daily living, and quality of life (KOOS₄); scores ranging from 0 (worst) to 100 (best).

Results: In the trial of TKR, 18 (36%) patients randomized to non-surgical treatment alone underwent TKR during the two years (five between one and two years), while eight patients randomized to non-surgical treatment and ten patients randomized to usual care underwent TKR during the two years in the other trial. Patients randomized to TKR followed by non-surgical treatment had greater improvement in KOOS₄ than patients randomized to non-surgical treatment alone in either trial (Fig. 1; 34.6 vs. 16.1 and 18.5 respectively; adjusted mean difference of the trial (95% confidence interval (95% CI)) of 18.0 (–24.1 to –11.9)). Patients randomized to non-surgical treatment alone in either trial improved more than patients randomized to written information and treatment advice (Fig. 1; 16.1 and 18.5 respectively vs. 11.6; adjusted mean difference of the trial (95% CI) of –5.8 (–11.0 to –0.6)).

Conclusions: TKR followed by non-surgical treatment resulted in greater improvements in pain and function than non-surgical treatment alone and non-surgical treatment resulted in greater improvements than written information after two years in patients with knee OA. Nearly two out of three patients with moderate to severe knee OA eligible for TKR postponed surgery for at least two years following non-surgical treatment. These results should encourage clinicians and patients to discuss benefits and harms of both surgical and non-surgical treatment options to decide what treatment best meets the need and expectations of the individual patient.



43 NOVEL TRANSLATIONAL DISCOVERY STUDY OF GENES AND SNPS ASSOCIATED WITH OSTEOARTHRITIS PAIN

R.E. Miller †, D. Syx ‡, S. Ishihara †, R.J. Miller §, A.M. Valdes ||, A.-M. Malfait †, †Rush Univ. Med. Ctr., Chicago, IL, USA; ‡Ghent Univ., Ghent, Belgium; §Northwestern Univ., Chicago, IL, USA; ||Univ. of Nottingham, Nottingham, United Kingdom

Purpose: Genome-wide association studies (GWAS) have been useful in identifying genes that may predispose for osteoarthritis (OA) and for pain, but they demand large patient cohorts. To improve the power of smaller cohorts, we developed a novel approach by combining data generated from a mouse model of OA pain with clinical GWAS data from cohorts with specific OA pain phenotypes. We used the destabilization of the medial meniscus (DMM) mouse model, which results in slowly progressive joint damage accompanied by pain-related behaviors. The development of chronic pain is characterized by molecular changes in the DRG, where the cell bodies of sensory afferent neurons reside. Thus, we performed microarrays on dorsal root ganglia (DRG) cells during the persistent pain period following DMM. Human homologues of the top regulated murine genes were compared to clinical GWAS datasets to see if there were SNPs in these genes that were associated with clinical OA pain phenotypes.

Methods: Approval for these studies was obtained from each institution. DMM or sham surgery was performed in the right knee of 10-week old male C57BL/6 mice. Age-matched naïve mice were also included. Eight and 16 weeks after surgery, ipsilateral L3–L5 DRG (DRG that innervate the knee) were collected and pooled from each mouse, RNA was extracted, and Affymetrix Mouse Transcriptome 1.0 Arrays were performed. A total of 3 mice (3 arrays) were used for each treatment per time point. Two out of three sham 8-week samples did not amplify well and were excluded. To identify genes associated with persistent pain, data pooled from DMM 8- and 16-week samples were compared to data pooled from naïve and sham 8- and 16-week samples by Student's t-test. The top differentially expressed genes were identified by $p < 0.001$. These top genes were investigated for single nucleotide polymorphisms (SNPs) and association with clinical OA pain phenotypes ($p < 0.05$) using three different patient GWAS datasets: (1) Symptomatic vs asymptomatic knee OA; (2) Neuropathic pain symptoms post total joint replacement; (3) Disturbed sleep (potentially related to chronic pain) post total joint replacement. For phenotype (1), genotyping with the Exome BeadChip array was carried out in 458 knee OA cases with a K/L score of 2 or higher in the tibiofemoral compartment. Of these, 212 were asymptomatic, reporting no knee pain. The remaining 246 reported pain in the knee at least 15 days during the past month. For phenotypes (2) and (3), genotyping with the Illumina 610k array was carried out in patients recruited post-total hip or knee replacement for OA ($n = 613$). Individuals were assigned a phenotype by classifying them according to their scores on the painDETECT questionnaire. Scores >12 were classified as “possible neuropathic pain.” Sleep scores from the Medical Outcomes Survey (MOS) were available for these same individuals. We classified the bottom tertile of the MOS sleep subscale as “disturbed sleep.”

Results: A total of 36 genes were differentially regulated in the DRG in the persistent pain phase of the DMM mouse model, according to our cutoff of $p < 0.001$. Sixteen genes were upregulated in DMM samples compared to controls, and 20 genes were downregulated.

Of the 36 differentially regulated murine genes, 27 genes had human homologues, and 34 SNPs within 11 of these genes were associated with clinical OA pain phenotypes ($p < 0.05$). These 11 genes have functions that include roles in circadian rhythm, apoptosis signaling, complement system, autophagy, regulation of cell signaling, and transcriptional regulation. Only 1 of the 11 genes, a cysteine proteinase, has been associated with a pain phenotype in other experimental models. SNPs within 8 genes were associated with disrupted sleep, SNPs within 5 genes were associated with neuropathic pain, and SNPs within 2 genes were associated with symptomatic OA.

The most significant signal ($p = 9.97 \times 10^{-4}$) was a SNP on chromosome 11 within a gene that is implicated in transcriptional regulation and was downregulated after DMM. This SNP had an odds ratio of 1.538 in association with disrupted sleep. Other SNPs in the same gene were found to be nominally associated with symptomatic OA ($p < 0.032$) and with neuropathic pain ($p < 0.005$).

Conclusions: This study suggests that it may be possible to combine microarray data from well-characterized translational models of osteoarthritis pain and GWAS data from well-characterized patient cohorts in order to refine the results of both types of studies. Future work will investigate the roles of these genes in experimental OA, will address the functional implications of these SNPs, and will seek to replicate the findings in other cohorts.

44 NATIONAL CLINICAL AUDIT DATA DECODES THE GENETIC ARCHITECTURE OF DEVELOPMENTAL DYSPLASIA OF THE HIP

K. Hatzikotoulas †, A. Roposch ‡, K.M. Shah §, M.J. Clark §, S. Bratherton §, V. Limbani ||, K. Warsame ‡, M. Ratnayake ¶, M. Tselepi ¶, J. Schwartztruber †, J. Steinberg †, D. DH Case Control Consortium †, J. Loughlin ¶, D.M. Eastwood ||, E. Zeggini †, J.M. Wilkinson §, †Wellcome Trust Sanger Inst., Cambridge, United Kingdom; ‡Univ. Coll. London, London, United Kingdom; §Univ. of Sheffield, Sheffield, United Kingdom; ||Royal Natl. Orthopaedic Hosp., Stanmore, United Kingdom; ¶Newcastle Univ., Newcastle, United Kingdom

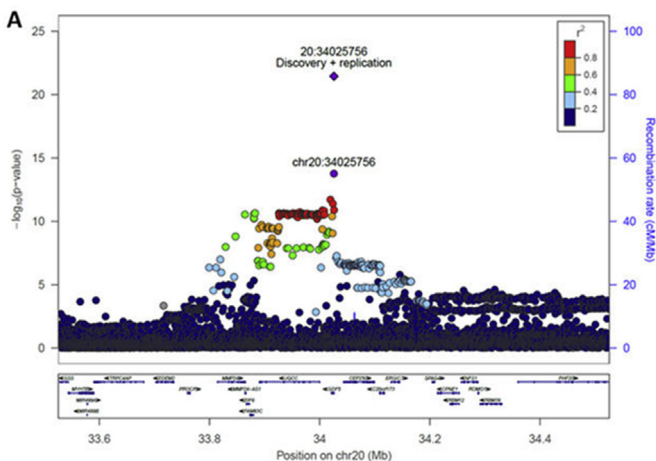
Purpose: Developmental dysplasia of the hip (DDH) is a heritable condition with an incidence of 3–6 per 1000 live births in the United Kingdom. DDH is characterised by abnormal development of the hip joint that results in pain, loss of function, and secondary osteoarthritis. We applied case identification using national clinical audit data and

postal recruitment to conduct the first successful genome-wide study of the genetic architecture of DDH to better understand its biological aetiology.

Methods: We recruited 770 patients (639 female) with a history of DDH from the English National Joint Registry (NJR) and 3364 controls (3048 female) from UK Household Longitudinal Study (UKHLS) for the discovery cohort. All participants were of UK European ancestry. Genomic DNA was genotyped using the Illumina HumanCoreExome beadchip. Following quality control checks at the sample and genotype level, association analyses were conducted under an additive model. Identified independent signals were followed up in an independent replication cohort of 1129 (1004 female) children with DDH, recruited prospectively, and 4652 independent controls from the UKHLS. Finally, a meta-analysis of both cohorts was conducted. We defined genome-wide significance as $p < 5 \times 10^{-8}$. We estimated the heritability of DDH using genetic complex trait analysis (GCTA). To test for shared genetics of DDH with OA, we employed high-resolution polygenic risk scoring (with data from a previous GWAS of OA) by using evenly spaced p-value thresholds between 0.001 and 0.50.

Results: Using genome-wide single nucleotide polymorphism (SNP) data and GCTA analysis we find that common-frequency autosomal SNPs explain 55% ($\pm se = 6\%$, $p < 0.0001$) of the liability-scale heritability of DDH. In the discovery case-control analysis we find 53 SNPs, comprising 25 independent signals, showed suggestive association with DDH at $p < 9 \times 10^{-5}$. Eleven correlated variants reached genome wide significance, with rs 143384 in the *GDF5* promoter as the lead variant (OR 1.57, 95% CI 1.3–1.77, $p = 1.72 \times 10^{-14}$). At replication, the rs143384 variant was also associated with DDH at genome-wide significance (OR = 1.37, 95% CI 1.24 to 1.51, $p = 1.33 \times 10^{-10}$). Finally, at meta-analysis the rs143384 variant was associated with DDH with OR = 1.44 (95% CI 1.34 to 1.56, $p = 3.55 \times 10^{-22}$, Fig. A; regional association plot showing variants within *GDF5* locus and strength of association). We also identify two further replicating loci with suggestive association to DDH near the *NFIB* (rs4740554, OR 1.30 [95% CI 1.16–1.45], $p = 4.44 \times 10^{-6}$) and *LOXL4* (rs4919218, 1.19 [1.10–1.28] $p = 4.38 \times 10^{-6}$) genes. We identify *RETSAT* and *PDRG1* association with DDH through gene-based analysis ($p = 1.19 \times 10^{-6}$ and $p = 3.77 \times 10^{-6}$, respectively). To identify potential shared genetic aetiology and hence common biological pathways underpinning DDH and idiopathic hip OA we constructed polygenic risk scores using the arcOGEN dataset and tested their predictive potential in the DDH GWAS. We find no significant association between polygenic risk scores for hip OA and DDH.

Conclusions: Using the NJR as a proof-of-principle, we describe the genetic architecture of DDH and establish the first robust DDH genetic locus. Fine mapping of the 5'UTR of *GDF5* indicates rs143384 (rather than 143383) as the causal variant. We also demonstrate a robust and scalable national clinical audit-based recruitment strategy for genetic studies that is transferrable to other complex diseases.



45

WHOLE GENOME SEQUENCING FINDS RARE HIGH-RISK GENOTYPES FOR HIP OSTEOARTHRITIS IN THE *COMP* AND *CHADL* GENES

T. Ingvarsson †, U. Styrkarsdottir †, H. Helgason †, A. Sigurdsson †, G. Norrdahl †, A. Agustsdottir †, L. Reynard §, A. Garcia ||, G. Halldorsson †, A. Jonasdottir †, A. Magnusdottir †, A. Oddson †, G. Sulem †, F. Zinc †, G. Sveinbjornsson †, A. Helgason †¶, H. Johannsdottir †, A. Helgadóttir †, T. Rafnar †, I. Almdahl #, A. Brækhus †††, T. Fladby §§|||, G. Selbæk ¶¶##, F. Hosseinpanah †††, F. Azizi †††, J. Koh §§§, N.L.S. Tang |||||, M. Daneshpour ¶¶¶, J. Mayordomo ###, C. Welt ††††, N. Samani ††††, P. Braund §§§§, L. Kiemeny |||||, S. Lohmander ¶¶¶, C. Christiansen #####, O.A. Andreassen †††††, O. Magnusson †, arcOGEN consortium G. Masson †, G. Masson †, A. Kong †, A. Kong †, I. Jonsdottir †††††, D. Gudbjartsson †, D. Gudbjartsson †, P. Sulem †, H. Jonsson §§§§§, J. Loughlin ||, U. Thorsteinsdottir †, K. Stefansson †. †Fsa Hosp. /Univ. of Iceland, Reykjavík, Iceland; ‡Decode Genetics, Reykjavík, Iceland; §Musculoskeletal Res. Group, Inst. of Cellular Med., Newcastle Univ., Newcastle-upon-Tyne, United Kingdom; ||Musculoskeletal Res. Group, Inst. of Cellular Med., Newcastle Univ., Newcastle-upon-Tyne, United Kingdom; ¶Dept. of Anthropology, University of Iceland, Iceland; #Dept. of Neurology, Akershus University Hospital, Lørenskog, Norway; ††Norwegian Natl. Advisory Unit on Ageing and Hlth., Vestfold Hospital Trust, Tønsberg, Norway; †††Dept. of Geriatric Med., Oslo University Hospital, Oslo, Norway; §§Dept. of Neurology, Akershus University Hospital, Lørenskog, Norway; |||||Inst. of Clinical Med., Univ. of Oslo, Oslo, Norway; ¶¶Norwegian Natl. Advisory Unit on Ageing and Hlth., Vestfold Hosp. Trust, Tønsberg, Norway; ##Faculty of Med., Univ. of Oslo, Oslo, Norway; †††††Endocrine Res. Ctr., Res. Inst. for Endocrine Sci., Shahid Beheshti Univ. of Med. Sci., Tehran, Islamic Republic of Iran; †††††Endocrine Res. Ctr., Res. Inst. for Endocrine Sci., Shahid Beheshti Univ. of Med. Sci., Tehran, Islamic Republic of Iran; §§§Div. of Endocrinology and Metabolism, Asan Med. Ctr., Univ. of Ulsan Coll. of Med., Seoul, Democratic People's Republic of Korea; |||||Dept. of Chemical and Lab. for Genetics of Disease Susceptibility, Li Ka Shing Inst. of Hlth.Sci., Faculty of Med., The Chinese Univ. of Hong Kong, Hong Kong, China; ¶¶¶Cellular and Molecular Endocrine Res. Ctr., Research Institute for Endocrine Sciences, Shahid Beheshti University, Tehran, Iran; ###University of Zaragoza, Zaragoza, Spain; †††††Reproductive Endocrine Unit, Massachusetts General Hospital, Boston, MA, USA; †††††Department of Cardiovascular Sciences, University of Leicester, Leicester, United Kingdom; §§§§Department of Cardiovascular Sciences, University of Leicester, Leicester, United Kingdom; |||||Department of Epidemiology, Biostatistics & HTA (133 EBH), Department of Urology (659 URO), Radboud University, Nijmegen Medical Centre, Nijmegen, The Netherlands; ¶¶¶¶Department of Orthopaedics (Lund), Lund University, Lund, Sweden; ¶¶¶¶Center for Clinical and Basic Research A/S, Ballerup, Denmark; †††††KG Jebsen Centre for Psychosis, Oslo University Hospital, Oslo, Norway; †††††Department of Immunology, The University Hospital of Iceland, Reykjavík, Iceland; §§§§§Landspítalinn University Hospital, Reykjavík, Iceland

Purpose: Genome-wide association studies (GWAS) of hip OA have reported several common sequence variants, all conferring small to moderate effects. Here, we report on a GWAS of total hip replacement (THR) for end-stage hip OA including both common and rare sequence variants

Methods: The study group included 4,657 Icelanders who have undergone THR because of OA and 207,514 individuals as controls. The sequence variants were identified through whole-genome sequencing of 8,453 Icelanders, and subsequently imputed into the study population. We tested for association between THR and 31.6 million sequence variants under the multiplicative model, and 19.2 million variants under the recessive model. We directly genotyped populations of European, Middle-Eastern and East-Asian descent for replication purposes and estimation of population frequencies. We used RNA-sequencing to explore gene expression in cartilage and tested for nonsense-mediated decay in an *in-vitro* reporter system.

Results: We discovered two rare signals that strongly associate with THR; a missense mutation, p.Asp369His, in the *COMP* gene (allelic frequency = 0.026%, $P = 4.0 \times 10^{-12}$, OR = 16.7 under a multiplicative model), and a frameshift mutation that introduces a premature stop codon, p.Val330GlyfsTer106 (rs532464664[ins8]), in the *CHADL* gene that associates through a recessive mode of inheritance (homozygote frequency = 0.15%, $P = 4.5 \times 10^{-18}$ and OR = 7.71). None of the carriers of the p.Asp369His mutation in *COMP* ($n = 77$) have been diagnosed with MED or other dysplasia. The *COMP* p.Asp369His carriers form a