THU0014

COMPARATIVE TRANSCRIPTOME ANALYSES ACROSS TISSUES AND SPECIES IDENTIFY TARGETABLE GENES FOR HUMAN SYSTEMIC LUPUS ERYTHEMATOSUS (SLE) AND LUPUS NEPHRITIS (LN)

E. Frangou^{1,2}, P. Garantziotis², M. Grigoriou², A. Banos², N. Panousis³, E. Dermizakis³, G. Bertsias⁴, D. Boumpas^{2,5,6}, A. Filia². ¹Limassol General Hospital, Limassol, Cyprus; ²Biomedical Research Foundation of the Academy of Athens, Athens, Greece; ³University of Geneva, Geneva, Switzerland; ⁴University of Crete, Heraklion, Greece; ⁵University of Athens, Athens, Greece; ⁶University of Cyprus, Nicosia, Cyprus

Background: Systemic Lupus Erythematosus (SLE) is a complex disease associated with the dysfunction of multiple tissues and cells. The causal tissue for each disease phenotype is not known a priori. Despite improvements in diagnosis and treatment, major organ involvement (such as the kidneys) contributes significantly to morbidity and mortality that still remain increased. There is an unmet need for timely targeted therapy.

Objectives: RNA-sequencing was performed to investigate the patterns of transcription variation across tissues between healthy and lupus-prone mice at different stages of lupus, and how these patterns associate with human Systemic Lupus Erythematosus (SLE).

Methods: NZB/W-F1 lupus prone mice were sacrificed at the pre-puberty, pre-autoimmunity and nephritic stage. Age-matched C57BL/6 were used as controls. An "effector" tissue (spleen) and "end-organs" (kidneys, brain) were collected. Total RNA was isolated, and mRNA-sequencing was performed. A time-series analysis was developed and differentially expressed genes (DEGs) were analyzed with DESeq. Hierarchical clustering and functional enrichment analysis were performed with gProfiler. Human orthologs of mouse tissue DEGs were identified in the whole-blood RNA-sequencing dataset comprised of 55 lupus-nephritis (LN), 65 non-LN SLE patients and 58 healthy individuals (HI). Human orthologs were compared to human DEGs. Using machine learning, human orthologs identified in the mouse dataset were used to predict kidney involvement in the human dataset, which was split in training and validation sets.

Results: Lupus susceptibility and progression signatures at different tissues and different stages of the disease were identified. Tissue-specific signatures and a common cross-tissue signature were also described. Previously described and novel biological processes and pathways were revealed. The comparative murine-human transcriptome analysis identified human orthologs from the mouse spleen-signature (including CCL5, IFIT and HLA genes) that are involved in systemic autoimmunity. It also identified human orthologs from the kidney- and brain-signature (including FCGR2A, C1Q, JAK1 and APOA2) that are involved in major "end-organ" damage and response mechanisms. Using a neural network model, 193 human orthologs accurately predicted LN patients vs HI (accuracy=0.86, sensitivity=0.82, specificity=0.91 in the validation set). Using a support vector machine model, 30 human orthologs and age and gender were the best predictors of LN vs non-LN SLE patients (accuracy=0.71, sensitivity=0.73, specificity=0.69 in the validation set).

Conclusion: Murine tissue gene signatures identified by RNA-sequencing analysis revealed biological processes and pathways that could be potentially used as biomarkers or therapeutic targets in human SLE. Comparison of the murine tissue-transcriptome with the whole-blood human-transcriptome revealed common gene signatures, demonstrating similar biological processes and pathways. Machine learning identified a murine kidney lupus signature that can accurately predict kidney involvement in human SLE. Validation in other datasets is ongoing.

References:

[1] Panousis NI, et al. Ann Rheum Dis 2019;78:1079

Acknowledgments: This work was supported by FOREUM, SYSCID and ERC -Advanced Grant

Disclosure of Interests: Eleni Frangou: None declared, Panayiotis Garantziotis: None declared, Maria Grigoriou: None declared, Aggelos Banos: None declared, Nikolaos Panousis: None declared, Emmanouil Dermizakis: None declared,

Table 2. Baseline characteristics and predictors of progression

	PROGRESSORS N= 17	NON-PROGRESSORS N=19	UNIVARIABLE OR (95% CI)	MULTIVARIABLE OR (95% CI)
Mean age y.o.(SD)	50.12 (10.01)	52.53 (9.22)	0.97 (0.91-1.04) p=0.446	-
Female (%)	11 (64.7)	14 (73.7)	1.52 (0.37-6.35) p=0.560	-
RF titre, mean (SD)	77.18 (99.93)	36.58 (53.39)	1.01 (0.99-1.02) p=0.162	1.01 (1.01-1.03) p=0.035
ACPA titre, mean (SD)	123.44 (109.44)	103.77 (116.18)	1.00 (0.99-1.01) p=0.594	0.99 (0.98-1.00) p=0.283
ANA+ (%)	2 (11.8)	5 (26.3)	2.68 (0.45-16.11) p=0.282	-
CRP, mean (SD)	5.41 (5.31)	2.97 (1.96)	1.21 (0.96-1.52) p=0.104	1.31 (0.99-1.73) p=0.055
ESR, mean (SD)	12.24 (11.97)	12.16 (8.44)	1.00 (0.94-1.07) p=0.981	-
Shared epitope (%)	8 (47.1)	10 (52.6)	1.25 (0.34-4.63) p=0.739	-
IFN-Score-A (-ΔCt), mean (SD)	-5.81 (1.07)	-6.23 (1.08)	1.45 (0.76-2.77) p=0.253	1.97 (0.83-4.66) 0.124
IFN-Score-B (-ΔCt), mean (SD)	-4.21 (0.56)	-4.52 (0.45)	3.53 (0.83-14.97) p=0.087	8.82 (1.02-76.38) p=0.048

George Bertsias Grant/research support from: GSK, Consultant of: Novartis, Dimitrios Boumpas: None declared, Anastasia Filia: None declared

DOI: 10.1136/annrheumdis-2020-eular.4102

THU0015

TYPE I INTERFERON SIGNATURE PREDICTS PROGRESSION TO INFLAMMATORY ARTHRITIS IN ACPA+ AT-RISK INDIVIDUALS WITHOUT CLINICAL SYNOVITIS

L. Garcia-Montoya^{1,2}, Z. Wigston¹, A. Burska¹, K. Mankia^{1,2}, E. Vital^{1,2}, P. Emery^{1,2}. ¹LIRMM, University of Leeds, Leeds, United Kingdom; ²Leeds Biomedical Research Centre, NIHR, Leeds, United Kingdom

Background: Interferon (IFN) is known to play a role in the pathogenesis of many autoimmune diseases, among them, rheumatoid arthritis (RA). A study showed that two interferon-stimulated gene expression scores (IFN-Score-A and IFN-Score-B) can be used to predict progression to connective tissue disease (CTD) in at-risk individuals, positive for anti-nuclear antibodies (ANA+) [1]. This validated score could potentially be applied to individuals at-risk of RA.

Objectives: To investigate the role of type I IFN in patients at-risk of RA and assess its potential role as a biomarker to predict progression to inflammatory arthritis (IA).

Methods: PBMC samples were taken from 36 at-risk individuals positive for anti-citrullinated protein antibodies (ACPA+), with a non-specific musculoskeletal complaint but no clinical synovitis and a normal ultrasound scan at baseline (BL). 17 of them developed IA later on, and had a second sample taken at the moment of progression. The other 19 did not progress and had a second sample taken after 1 year. Expression of IFN stimulated genes was assessed using TaqMan. T-test was used to compare the expression of the genes at the two time points of each individual. Binary logistic regression was used to assess BL predictors of progression. Multivariable analysis was adjusted for confounders such as C-reactive protein (CRP) rheumatoid factor (RF) and ACPA titre.

Results: Table 1 shows the list of tested genes. There were no differences in the gene expression of progressors at BL and at the moment of IA diagnosis. Similarly, no differences were found between the non-progressors at BL and after 1 year. Comparing BL samples from progressors and non-progressors, there was a trend to higher expression of IFN-score-B in the progressors (Mann Whitney p=0.055), whereas IFN-score-A was similarly expressed in both groups. This is similar to the results found in a cohort of ANA+ individuals at risk of progressing to CTD [1]. Multivariable analysis showed that IFN-score-B and RF titre were predictive for IA progression with an odds ratio (OR) of 2.18 (p=0.048) and OR 1.01 (p=0.035).

Table 1. Interferon stimulated genes and scores A and B

GENE	IFN score	GENE	IFN score
CCL8	A	BST2	B
CEACAM1		IFI16	
CXCL10		IFIH1	
GBP1		LAMP3	
IFI27		NT5C3B	
IFI44		PHF11	
IFI44L		SERPING1	
IFIT1		SOC31	
IRF7		SP100	
ISG15		STAT1	
RSAD2		TAP1	
XAF1		TRIM38	
		UBE2L6	
		UNC93B1	

Conclusion: These exploratory results suggest that IFN changes in IA progressors precede subclinical inflammation on ultrasound. A preliminary risk model shows that IFN-score-B could be useful to predict progression to IA in at-risk of RA individuals; however, these results require validation in a larger at-risk cohort.

References:

[1] Md Yusof MY. Ann. Rheum. Dis. 2018;77(10):1432-9.

Disclosure of Interests: Leticia Garcia-Montoya: None declared, Zoe Wigston: None declared, Agata Burska: None declared, Kulveer Mankia: None declared, Edward Vital Grant/research support from: AstraZeneca, Roche/Genentech, and Sandoz, Consultant of: AstraZeneca, GSK, Roche/Genentech, and Sandoz, Speakers bureau: Becton Dickinson and GSK, Paul Emery Grant/research support from: AbbVie, Bristol-Myers Squibb, Merck Sharp & Dohme, Pfizer, Roche (all paid to employer), Consultant of: AbbVie (consultant, clinical trials, advisor), Bristol-Myers Squibb (consultant, clinical trials, advisor), Lilly (clinical trials, advisor), Merck Sharp & Dohme (consultant, clinical trials, advisor), Novartis (consultant, clinical trials, advisor), Pfizer (consultant, clinical trials, advisor), Roche (consultant, clinical trials, advisor), Samsung (clinical trials, advisor), Sandoz (clinical trials, advisor), UCB (consultant, clinical trials, advisor)

DOI: 10.1136/annrheumdis-2020-eular.4981

THU0016

EPIGENOMIC ANALYSIS OF RA PATIENTS SHOWS DISTINCT BIOLOGICAL PROCESSES ASSOCIATED WITH ANTI-TNF RESPONSE

A. Julià¹, A. Gómez¹, A. Fernández Nebro², F. J. Blanco³, A. Erra⁴, S. Sánchez Fernandez⁵, J. Monfort⁶, M. Alperi-López⁷, I. González-Álvarez⁸, R. García de Vicuña⁹, R. Sanmartí⁹, C. Díaz Torne¹⁰, C. Marras Fernandez Cid¹¹, J. Tornero Molina¹², N. Palau¹, R. M. Lastra¹, J. Lladós¹, S. Marsal¹. ¹Rheumatology Research Group, Vall d'Hebron Research Institute, Barcelona, Spain; ²Rheumatology Department, Hospital Regional Universitario de Málaga, Universidad de Málaga, Málaga, Spain; ³Rheumatology Department, INIBIC-Hospital Universitario A Coruña, A Coruña, Spain; ⁴Rheumatology Department, Hospital San Rafael, Barcelona, Spain; ⁵Rheumatology Department, Hospital General La Mancha Centro, Ciudad Real, Spain; ⁶Rheumatology Department, Hospital del Mar, Barcelona, Spain; ⁷Rheumatology Department, Hospital Universitario Central de Asturias, Asturias, Spain; ⁸Rheumatology Department, Hospital Universitario La Princesa, Madrid, Spain; ⁹Rheumatology Department, Fundació Clínica per a la Recerca Biomèdica, Barcelona, Spain; ¹⁰Rheumatology Department, Hospital de la Santa Creu i Sant Pau, Barcelona, Spain; ¹¹Hospital Universitario Virgen de la Arrixaca, Murcia, Spain; ¹²Servicio de Reumatología, Hospital Universitario Guadalajara, Guadalajara, Spain

Background: Blocking Tumor Necrosis Factor (TNF) activity is a successful therapeutic approach for approximately 60% of patients with rheumatoid arthritis (RA). To date, however, the biological basis of the lack of efficacy of anti-TNF agents is unknown.

Objectives: The objective of present study was to characterize the biological basis of anti-TNF lack of efficacy in RA using an epigenomic data approach in two steps: first, to assess the differential methylation changes between responders and non-responders and second, to use this differential methylation profile in a systems biology approach to infer differential methylated biological modules according to anti-TNF response.

Methods: A total of n=68 patients diagnosed with RA according to the ACR-EULAR criteria belonging to 16 Hospitals across Spain were recruited. All patients were >18 years old, with more than 6 months of disease evolution and a baseline disease activity of DAS28 > 3.2. Treatment response was defined according to the EULAR criteria at week 12. Good and moderate responders were aggregated into a single responder group. Genomic DNA was collected at baseline and the methylation profile was assessed using the Illumina Infinium EPIC array, which interrogates 850,000 methylation CpG sites across the genome. Differential Methylation analysis, biological pathway association and the systems Biology approach using Protein-Protein Interaction Networks, were conducted using the R statistical language and the Bioconductor libraries.

Results: From 68 anti-TNF treated patients, n=27 (39.7%) were good responders, n=26 (38.2%) moderate responders and n=15 (22.05%) non-responders at week 12 of treatment. Differential methylation analysis identified two distinctive biological profiles associated with the clinical response: responders were associated to interleukin and cytokine production, and non-responders were associated with biological pathways associated to TGF-Beta production and T cell regulation. Using these differentially methylated profiles, epigenetic modules with differentially methylated hotspots between responders and non-responders were also found. Two epigenetic modules with significant enrichment in inflammatory and interleukin production and immune regulatory processes were validated in an independent patient cohort.

Conclusion: The epigenetic analysis of whole blood from RA patients using a module-based approach shows reproducible biological mechanisms associated with the response to anti-TNF therapy.

Acknowledgments: We would like to thank the clinical researchers and patients participating in the IMID Consortium for their collaboration

Disclosure of Interests: Antonio Julià: None declared, Antonio Gómez: None declared, Antonio Fernández Nebro: None declared, Francisco J. Blanco Grant/research support from: Sanofi-Aventis, Lilly, Bristol MS, Amgen, Pfizer, Abbvie, TRB Chemedica International, Glaxo SmithKline, Archigen Biotech Limited, Novartis, Nichi-iko pharmaceutical Co, Genentech, Janssen Research & Development, UCB Biopharma, Centrexion Therapeutics, Celgene, Roche, Regeneron Pharmaceuticals Inc, Biohope, Corbus Pharmaceutical, Tedec Meiji Pharma, Kiniksa Pharmaceuticals, Ltd, Gilead Sciences Inc, Consultant of: Lilly, Bristol MS, Pfizer, Alba Erra: None declared, Simon Sánchez Fernandez: None declared, Jordi Monfort: None declared, Mercedes Alperi-López: None declared, Isidoro González-Álvarez Grant/research support from: Roche Laboratories, Consultant of: Lilly, Sanofi, Paid instructor for: Lilly, Speakers bureau: Abbvie, MSD, Roche, Lilly, Rosario García de Vicuña Grant/research support from: BMS, Lilly, MSD, Novartis, Roche, Consultant of: Abbvie, Biogen, BMS, Celltrion, Gebro, Lilly, Mylan, Pfizer, Sandoz, Sanofi, Paid instructor for: Lilly, Speakers bureau: BMS, Lilly, Pfizer, Sandoz, Sanofi, Raimón Sanmartí Speakers bureau: Abbvie, Eli Lilly, BMS, Roche and Pfizer, Cesar Díaz Torne: None declared, Carlos Marras Fernandez Cid: None declared, Jesús Tornero Molina: None declared, Núria Palau: None declared, Raquel M Lastra: None declared, Jordi Lladós: None declared, Sara Marsal: None declared

DOI: 10.1136/annrheumdis-2020-eular.4572

THU0017

ATTEMPTS TO LINK EXONIC GENE POLYMORPHISMS TO SYSTEMIC LUPUS ERYTHEMATOSUS (SLE)-ASSOCIATED PROTEIN MODIFIED FUNCTIONALITY: A STRUCTURAL BIOLOGY APPROACH

E. Eliopoulos¹, G. Goulielmos², M. Matalliotakis^{2,3}, D. Vlachakis¹, T. Niewold⁴, M. Zervou². ¹Agricultural University of Athens, Department of Biotechnology, Athens, Greece; ²University of Crete, School of Medicine, Heraklion, Greece; ³Aristotle University of Thessaloniki, Third Department of Obstetrics and Gynecology, Thessaloniki, Greece; ⁴NYU School of Medicine, Colton Center for Autoimmunity, New York, United States of America

Background: Gene association studies and genome wide association studies (GWAS) have played a primary role in depicting genetic contributions to systemic lupus erythematosus (SLE) development, while accommodating the exonic polymorphisms on the protein structure level, when available, enhances our understanding of protein function modification or depletion. Linking human genetics with therapeutic targets requires the biological function of the causal gene and variant to be known.

Objectives: To investigate recently identified SLE-associated functional gene polymorphisms, such as *PARP1*, *ITGAM*, *TNFAIP3*, *NCF1*, *PON1*, *IFIH1*, *SH2B3* and *TYK2* [1-4] by correlation to protein structure and function.

Methods: Three-dimensional (3D) homology modeling and molecular mechanics/dynamics studies were applied for the localization of the polymorphisms under study on the respective proteins. The mutants were constructed using molecular modeling with the program Maestro (Schrodinger, LLC), which was also used to analyze the conformational changes caused by the mutation. All figures depicting 3D models were created using the molecular graphics program PyMOL V.2.2 [5].

Results: Modeling revealed that rs1136410 SNP encodes the less common polymorphism Val762Ala on *PARP1* that reduces enzymatic activity of Poly(ADP-ribose) polymerase 1 (Figure 1), *ITGAM* polymorphism rs1143679 (Arg77His) on Integrin alpha M, component of the macrophage-1 antigen complex affects protein surface recognition, *TNFAIP3* rs2230926 polymorphism encodes Cys instead of Phe at residue 127 of the ubiquitin editing A20 protein, while rs201802880 polymorphism of the neutrophil cytosolic factor 1 (*NCF1*) gene modifies the function of the cytosolic subunit of neutrophil NADPH oxidase with the mutation Arg90His. *PON1* is involved in the oxidative stress process that cause tissue damage observed in SLE and anti-phospholipid syndrome (APS). The *PON1* Gln192Arg mutation (rs662 SNP) affects shape and recognition of the ligand recognition site as part of the evolutionary process, while *IFIH1* (rs35667974) helicase C domain1 mutant I923V is located on an essential RNA beta loop interacting directly with the nucleic acid (Figure 2). Finally, the rs3184504 SNP of *SH2B3* gene generates mutant Arg262Trp on SH2 adapter protein 3, acting as a signaling pathway involved in autoimmune disorders, while in *TYK2* gene, one of the Janus kinases, the rs35018800 producing mutant Ala928Val modifies the ADP binding site.