XIX Congress of the Catalan Society of Immunology

Immunoevasion:
The art of becoming invisible

Barcelona, November 20th and 21st, 2025

Hybrid Meeting

Acadèmia de Ciències Mèdiques i de la Salut de Catalunya i de Balears.
Carrer Major de Can Caralleu, 1-7, 08017. Barcelona.

Registration available via the SCI website

PROGRAM

SOCIETAT CATALANA D'IMMUNOLOGIA

FUNDACIÓ ACADÈMIA DE CIÈNCIES MÈDIQUES I DE LA SALUT DE CATALUNYA I DE BALEARS

Welcome to the Congress,

On behalf of the organizing committee, we would like to warmly welcome you to the XIX Societat Catalana d'Immunologia Congress (SCI Congress). We believe that our meeting will present high level scientific knowledge with the contribution of immunologists and different specialists in areas related to the Immunoevasion: The art of becoming invisible.

Dr. Francisco Lozano

SCI President

Organization Committe (SCI Board)

President: Francisco Lozano Soto

Vice President: Eva Martínez Cáceres

Secretary: Laura Martínez Martínez

Treasurer: Laura Viñas Giménez

Vocal: Anaís Mariscal Rodríguez

Vocal: Inaki Salvador Corres

Vocal: María José Rodríguez Lagunas

Vocal: Juan Manuel Torres Canizales

Scientific Committe

Eva Martínez Cáceres

Pablo Engel Rocamora

Francisco Lozano Soto

Nuria Izquierdo Useros

Iñaki Álvarez Pérez

Ester Lozano García

José Yélamos López

Conchi Mora Giral

Óscar de la Calle Martín

Contents

Consell Català de Formació Continuada de les Professions Sanitàries

Comisión de Formación Continuada del Sistema Nacional de Salud

Activitat acreditada pel Consell Català de Formació Continuada de les Professions Sanitàries, la Comisión de Formación Continuada del Sistema Nacional de Salud, amb número de registre **09/040566-MD,** i nombre de crèdits 0,5.

Collaborators

Awards to best oral communication and best poster of the XIX Congress SCI 2025, and announcement of the TFG-HIPRA and TFM-HIPRA prize winners

The chairs of the different sessions and the board members of the SCI will select the best oral communication, taking into account its scientific value and the aspects related to the presentation. The congress attendees will elect the awarded poster by voting. The winners will be announced at the end of the congress, along with the awarding of the TFG-HIPRA and TFM-HIPRA prizes recognizing the best undergraduate and master's theses in Immunology.

Sponsors

Platinum

GRIFOLS

werfen

Golden

DIAGNÓSTICA LONGWOOD

Silver

STEMCELL™ TECHNOLOGIES

Palex

Miltenyi Biotec

Pfizer

BD

Bronze

ThermoFisher SCIENTIFIC

Promega

EUROIMMUN

CSL

Condensed program

Day 1

16:00 - 16:15	Welcome to the XIX Congress of the SCI
16:15 - 17:45	OPENING SESSION
17:45 - 18:15	COFFEE BREAK - POSTERS
18:15 - 19:15	ORAL COMMUNICATIONS - SESSION I: Autoimmunity and Inflammation

Day 2

10:00 - 10:45	PLENARY SESSION I
10:45 - 11:15	COFFEE BREAK - POSTERS
11:15 - 12:30	ORAL COMMUNICATIONS - SESSION II: Basic Immunology
12:30 - 14:00	ORDINARY GENERAL MEETING - SOCIETAT CATALANA D'IMMUNOLOGIA
14:00 - 15:00	LUNCH - POSTERS
15:00 - 15:30	SPONSORED TALK
15:30 - 16:15	PLENARY SESSION II
16:15 - 17:30	ORAL COMMUNICATIONS - SESSION III: Immunity and Infection
17:30 - 18:00	COFFEE BREAK - POSTERS
18:00 - 19:15	ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology
19:15 - 20:00	PLENARY SESSION III
20:00 - 20:15	Prize to the best communication and best poster - Awarding of the TFG-HIPRA and TFM-HIPRA prizes - Closing of the Congress

16:00 - 16:15	**Welcome to the XIX Congress of the SCI** **Francisco Lozano** (President of the SCI)
16:15 - 17:45	**OPENING SESSION** • **A novel radiotherapy actionable immunological mechanism of resistance to CDK4/6 inhibitors in HR+ breast cancer** **Lorenzo Galluzzi** (Cancer Signaling and Microenvironment Program, Fox Chase Cancer Center, Philadelphia) • **Myeloid traits of immune evasion** **María Casanova-Acebes** (Cancer Immunity Laboratory, Molecular Oncology Program, Spanish National Cancer Research Center (CNIO), Madrid, Spain) *Chairs: Conchi Mora, Oscar de la Calle Martín*
17:45 - 18:15	**COFFEE BREAK - POSTERS**
18:15 - 19:15	**ORAL COMMUNICATIONS - SESSION I: Autoimmunity and Inflammation** • **Immune modulatory effect of the pro-fibrotic lung microenvironment** **Alba Puebla** (Fundació Clínic per a la Recerca Biomèdica - IDIBAPS (FCRB-IDIBAPS), Barcelona) • **Longitudinal Immunoprofiling Reveals Pharmacodynamic Signatures and Biomarkers Predictive of Dimethyl Fumarate Response in Multiple Sclerosis** **Federico Fondelli** (Institut Germans Trias i Pujol; Universitat Autonoma de Barcelona, Barcelona) • **Immune Monitoring autoreactive T-cell responses in a cohort Multiple Sclerosis patients undergoing disease-modifying therapy** **Alex Agundez** (Institut Germans Trias i Pujol; Universitat Autonoma de Barcelona, Barcelona) • **Complementary Roles of Urine sCD163, sALCAM, and sC5b-9 in the Non-Invasive Assessment of Lupus Nephritis** **Eduardo Gozálvez** (Servei d´Immunologia, Centre de Diagnòstic Biomèdic, Barcelona) *Chairs: Laura Naranjo, Bibiana Quirant*

10:00 - 10:45	**PLENARY SESSION I** • **Immunomodulatory role of exosomes from the infective forms of *Trypanosoma cruzi*, the parasite responsible for Chagas disease** **Antonio Osuna Carrillo de Albornoz** (Department of Parasitology, Biochemical and Molecular Parasitology Group CTS-183, and Institute of Biotechnology, University of Granada, Granada, Spain) *Chair: Joaquim Gascón*
10:45 - 11:15	**COFFEE BREAK - POSTERS**
11:15 - 12:30	**ORAL COMMUNICATIONS - SESSION II: Basic Immunology** • **Dendritic cells exposed to autologous In Vitro–Generated Photopheresates Exert a Suppressive Effect on naive CD4 T cells** **Hendrik Veltman** (Institut Germans Trias i Pujol; Universitat Autonoma de Barcelona, Barcelona) • **CD244 as a Negative Regulator of Human Macrophage Activity** **Aarón Adrián Ochoa** (Universitat de Barcelona) • **CD6 expression impacts B cell biology** **Laura Carrillo-Serradell** (Immunoreceptors del Sistema Innat i Adaptatiu, IDIBAPS, Barcelona) • **Validation of G protein-coupled receptors (GPCRs) antibodies: new CD Assignments and Expression Profiles from the HLDA11 Workshop** **Javier Fernández-Calles** (Faculty of Medicine and Health Sciences,University of Barcelona) • **Towards Large-Scale Immune Profiling: Preliminary Evaluation of a Spectral Panel in Healthy Donors** **Sara Monreal-Peinado** (Institut Germans Trias i Pujol; Universitat Autonoma de Barcelona; Josep Carreras Leukaemia Research Institute (IJC), Barcelona) *Chairs: Jorge Lloberas, Esther Lozano*
12:30 - 14:00	**ORDINARY GENERAL MEETING - SOCIETAT CATALANA D'IMMUNOLOGIA**
14:00 - 15:00	**LUNCH - POSTERS**

15:00 - 15:30	**SPONSORED TALK** • **Respiratory Syncytial Virus. Importance and vaccination in adults.** **Antoni Torres** (Respiratory Intensive Care Unit, Hospital Clínic, Barcelona) *Chair: Sofía Pérez Del Pulgar*
15:30 - 16:15	**PLENARY SESSION II** • **Poxvirus immunoevasion** **Antonio Alcamí** (Centro de Biología Molecular Severo Ochoa, Consejo Superior de Investigaciones Científicas (CSIC), Universidad Autónoma de Madrid (UAM), 28049 Madrid, Spain) *Chair: Ana Angulo*
16:15 - 17:30	**ORAL COMMUNICATIONS - SESSION III: Immunity and Infection** • **The protective potential of vaccination against LCMV-induced Post-Acute Infection Syndromes** **Paula Cebollada Rica** (Universitat Pompeu Fabra, Barcelona) • **Humoral immunity after PHH-1V COVID-19 booster vaccination in adults with immunosuppressive conditions** **Dàlia Raïch-Regue** (IrsiCaixa, Hospital Germans Trias i Pujol, Badalona, Spain) • **Broad-spectrum anti-ACE2 neutralizing antibodies for protection against SARS-CoV-2 variants and emerging coronaviruses** **Pablo Hernández-Luis** (University of Barcelona, Immunology Unit, Department of Biomedical Sciences, Faculty of Medicine, Barcelona) • **Adoptive cell transfer of NK-92 cells expressing a CD5-based Chimeric Antigen Receptor (SRCD5CAR-NK92) to treat a clinically relevant invasive fungal infection** **Maria Velasco-de-Andrés** (Immunoreceptors del sistema innat i adaptatiu, Institut d'Investigacions Biomèdiques August Pi i Sun, Barcelona) • **From Data to Diagnosis: Implementing a multi-omic approach in inborn errors of immunity** **Laura Batlle-Masó** (Translational Immunology research group, VHIR) **Sandra Estiarte** (Coordinator at BCN PID Foundation) *Chairs: Pablo Engel, Meritxell Genesca*
17:30 - 18:00	**COFFEE BREAK - POSTERS**

18:00 - 19:15	**ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology** • **Bone marrow immune and stromal niche remodeling in myelodysplastic neoplasm patients under erythropoietic therapies by 29-color full-spectrum flow cytometry** **Clàudia Pellín Jou** (Vall d'Hebron Institute of Oncology (VHIO), Barcelona) • **Immortalized NK cells expressing a CD6-based CAR effectively target non-hematological tumour cells in vitro** **Lucía Aragón Serrano** (Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), Barcelona) • **Rectifying false-positive results using a competitive antagonist in flow cytometry crossmatch for kidney transplant candidates with rituximab treatment** **Ángel García** (Hospital Clínic de Barcelona, Barcelona) • **Validation of the Luminex Flexmap 3D System for Anti-HLA Antibody Detection Using Single Antigen Beads (SAB)** **David Rodríguez Martín** (Hospital Clínic de Barcelona, Barcelona) • **Comparison and Validation of in vitro extracorporeal photopheresis Protocols for the analysis of its immunomodulatory effects** **Hendrik Veltman** (Institut Germans Trias i Pujol; Universitat Autonoma de Barcelona, Barcelona) *Chairs: Mª José Herrero, Iñaki Salvador*
19:15 - 20:00	**PLENARY SESSION III** • **Harnessing NK cells for Viral Control** **María José Buzón Gómez** (Infectious Diseases Department, Institut de Recerca Hospital Univesritari Vall d'Hebrón (VHIR), Universitat Autònoma de Barcelona, Barcelona, Spain) *Chair: Núria Izquierdo Useros*
20:00 - 20:15	**Prize to the best communication and best poster** **Awarding of the TFG-HIPRA and TFM-HIPRA prizes** HIPRA **Closing of the Congress** **Eva Martinez Cáceres** (Vice-President of the SCI)

ABSTRACTS

ORAL COMMUNICATIONS - SESSION I: Autoimmunity and Inflammation

Immune modulatory effect of the pro-fibrotic lung microenvironment

Alba Puebla 1; Fernanda Hernandez-Gonzalez 1,4; Alvar Agustí 1,2,4; Jacobo Sellares 1,2,4; Rosa Faner 1,2,3; Tamara Cruz 1,2,5

1 Fundació Clínic per a la Recerca Biomèdica - IDIBAPS (FCRB-IDIBAPS), Barcelona, Spain; 2 Centro Investigación Biomédica en Red Enfermedades Respiratorias (CIBERES), Barcelona, Spain.; 3 University of Barcelona, Biomedicine Department, Barcelona, Spain.; 4 Department of Pulmonology, Hospital Clinic, University of Barcelona, Barcelona, Spain.; 5University of Barcelona, Biology, Immunology Department, Barcelona, Spain.

Introduction. Pulmonary fibrosis (PF) is a chronic and debilitating lung disease characterized by irreversible scarring of the lung parenchyma, leading to respiratory failure and high mortality within a few years of diagnosis. Epithelial-fibroblast hyperactivation and the accumulation of senescent cells are recognized as major contributors to the disease. In this scenario, the lung microenvironment profoundly alters immune cell function and the immune system plays a key role modulating the fibrotic progression. Dysregulated immune activation, chronic inflammation, and the presence of exhausted or regulatory immune cells have been described in PF, underscoring that the immune system's impact on fibrosis is highly context- and stage-dependent.

Aim. To explore how the pro-fibrotic lung microenvironment modulates the immune system.

Methods. Precision cut-lung slices (PCLS) were obtained from a donor lung tissue without any lung disease. PCLS were treated with a fibrotic cocktail (TGF-β, TNF-α, PDGF-AB and LPA) during 48h to induce pro-fibrotic features. To assess immune modulation, PBMCs from the same donor were isolated and cultured either directly with the pro-fibrotic PCLS or with their conditioned media (CM) for 48h.

Results. Single-cell RNA sequencing (scRNA-seq) of PBMCs identified 18 immune cell populations, including subsets expressing activation and exhaustion markers (Figure 1a). Exposure to pro-fibrotic PCLS or their CM altered the UMAP distribution of the stimulated immune subpopulations. Notably, distinct clustering patterns were observed within cytotoxic cell compartments (Figure 1b).

Conclusions. Our findings suggests that PCLS can model a pro-fibrotic lung microenvironment in vitro that influence immune cell composition and activation. This model provides a valuable platform to further dissect immune-fibrotic crosstalk and identify potential immune evasion mechanisms in pulmonary fibrosis.

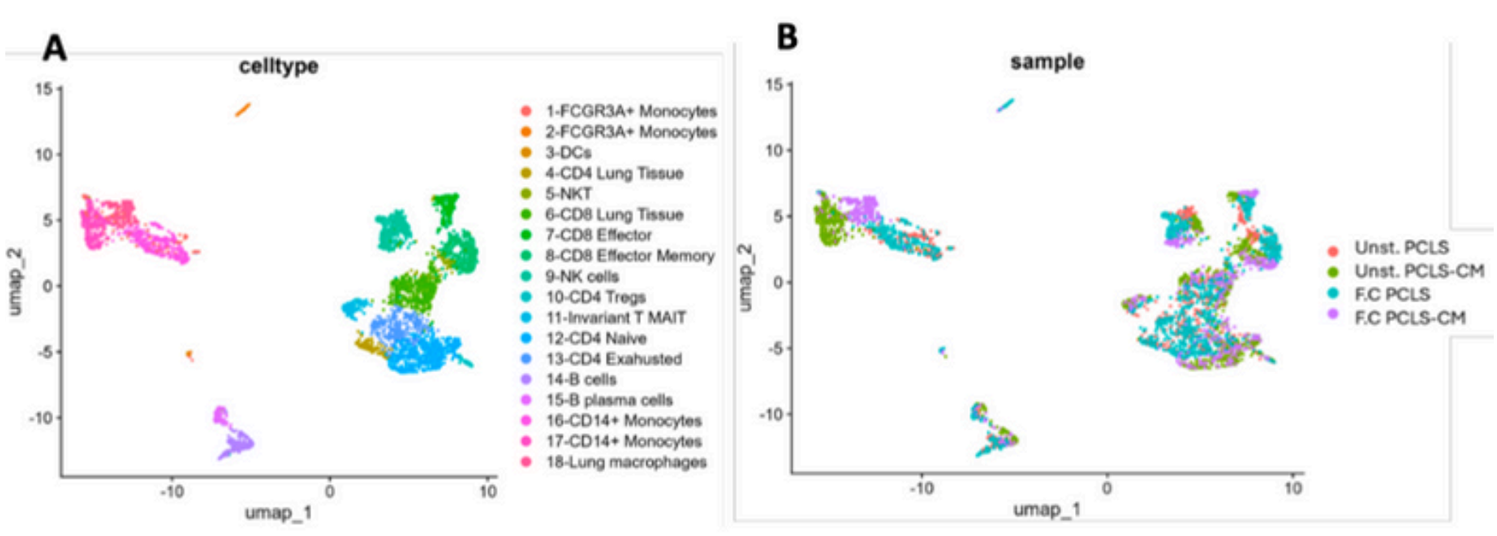

Figure 1. *scRNAseq of PBMC in co-culture or stimulated with CM of PCLS under pro- fibrotic conditions. PBMC were incubated for 48h in co-culture with PCLS unstimulated and stimulated with Fibrotic cocktail (F.C) or stimulated with their CM. 18 populations were identify in the PBMCs including subpopulations with activation/exhaustion markers and co-culture/stimulation induce changes in the proportion of immune subpopulations.*

ORAL COMMUNICATIONS - SESSION I: Autoimmunity and Inflammation

Longitudinal Immunoprofiling Reveals Pharmacodynamic Signatures and Biomarkers Predictive of Dimethyl Fumarate Response in Multiple Sclerosis

Federico Fondelli 1,4; Silvia Presas Rodriguez 3; Josep Calafell 2; Esteban Ballestar 2; Cristina Ramo Tello 3; Eva Martinez Caceres 3,4; Antonio Gonzalez Garcia 3

1 Institut Germans Trias i Pujol; 2 Institut Josep Carreras; 3 Hospital Germans Trias i Pujol; 4 Universitat Autonoma de Barcelona

Introduction: Dimethyl fumarate (DMF) is an immunomodulatory treatment for multiple sclerosis (MS) with heterogeneous clinical responses. Understanding its pharmacodynamic effects and identifying predictive biomarkers of response are crucial for optimising treatment strategies, given the abundance of first-line treatments available for MS patients. This study integrates multiomic approaches to characterise DMF-induced immunological changes and identify baseline biomarkers predictive of treatment response.

Methods & Materials: We conducted a five-year longitudinal study of 89 MS patients undergoing DMF treatment, collecting blood samples every six months for flow cytometry, single-cell RNAseq and ATACseq, and Olink cytokine profiling. Flow cytometry assessed immune cell subpopulations, and patients were classified as good or bad responders based on clinical outcomes. Single-cell transcriptomics and epigenomics were employed to characterise immune signatures at baseline and after 12 months. Cytokine (Olink 96-plex) analysis provided additional insights into cytokine dynamics.

Results: DMF induced a global immunological shift, independent of clinical response, characterised by alterations in multiple immune cell populations. Pharmacodynamic analysis revealed a significant reduction in CD4 Th1Th17 cells and an increase in transitional B cells in good responders at six months. Importantly, baseline levels of these populations predicted treatment response: bad responders exhibited higher pre-treatment CD4 Th1Th17 cell frequencies, whereas good responders showed elevated transitional B cells. Olink identified increased baseline IFNg levels in bad responders. Single-cell RNAseq and ATACseq analyses further delineate T and B cell signatures associated with response dynamics and different clinical outcomes.

Conclusions: This study provides a comprehensive immunological characterisation of DMF treatment in MS. We identify Th1Th17 and transitional B cells as both pharmacodynamic markers and predictive biomarkers of response, with IL-10 as a potential mechanistic link. These findings offer valuable insights for personalised treatment approaches and early stratification of MS patients for DMF therapy, as an opening for DMF use in other diseases.

ORAL COMMUNICATIONS - SESSION I: Autoimmunity and Inflammation

Immune Monitoring autoreactive T-cell responses in a cohort multiple sclerosis patients undergoing disease-modifying therapy

Alex Agundez 1,3; Federico Fondelli 1; Berta Arcos 1; Jana Willemyns 1; Hendrik Veltman 1; Silvia Presas 2; Aina Teniente Serra 1,2,3; Antonio Manuel González García 2; Cristina Ramo 2; Eva Martínez-Cáceres 1,2,3

1 Institut Germans Trias i Pujol; 2 Institut Josep Carreras; 3 Hospital Germans Trias i Pujol; 4 Universitat Autonoma de Barcelona

Introduction: Multiple sclerosis (MS) is a chronic autoimmune disease of the central nervous system (CNS), characterized by the destruction of myelin. Although the exact cause remains unclear, it is believed to involve an immune response where autoreactive T cells target myelin-derived peptides. In this study, we analyzed antigen-specific autoreactive T cell responses against 15 myelin-derived peptides in a cohort of MS patients. We aimed to identify potential specific myelin-derived peptides and assess the differences in T cell reactivity in patients receiving different treatments.

Methodology: T cell reactivity was determined via ELISPOT-IFN-γ assay using 200.000 PBMCs per well. A total of 124individuals were included in the study: 21 untreated MS patients; 83 MS patients undergoing disease-modifying therapies (DMTs), including fumarates, SP1 modulators, nucleoside analogs, anti-CD20, and anti-VLA4; and 20 healthy controls.

Results: Forty-one percent of patients exhibited immune responses to at least one of the 15 myelin-derived peptides,with significant differences in T-cell reactivity depending on treatment. Significant pairwise differencesmainly involved Cladribine and SP1 modulators, which showed lower reactivity compared with other DMTs.Four peptides (10, 12, 13, and 15) were immunodominant, though peptide 15 was excluded due to reactivityin healthy controls (Figure 1).

Conclusions: The response to myelin-derived peptides was heterogeneous among patients, with three peptides demonstrating overall immunodominance. Moreover, lower reactivity was observed in patients treated with SP1 modulators and Cladribine. These findings may provide insights into target peptides for MS immunotherapy development.

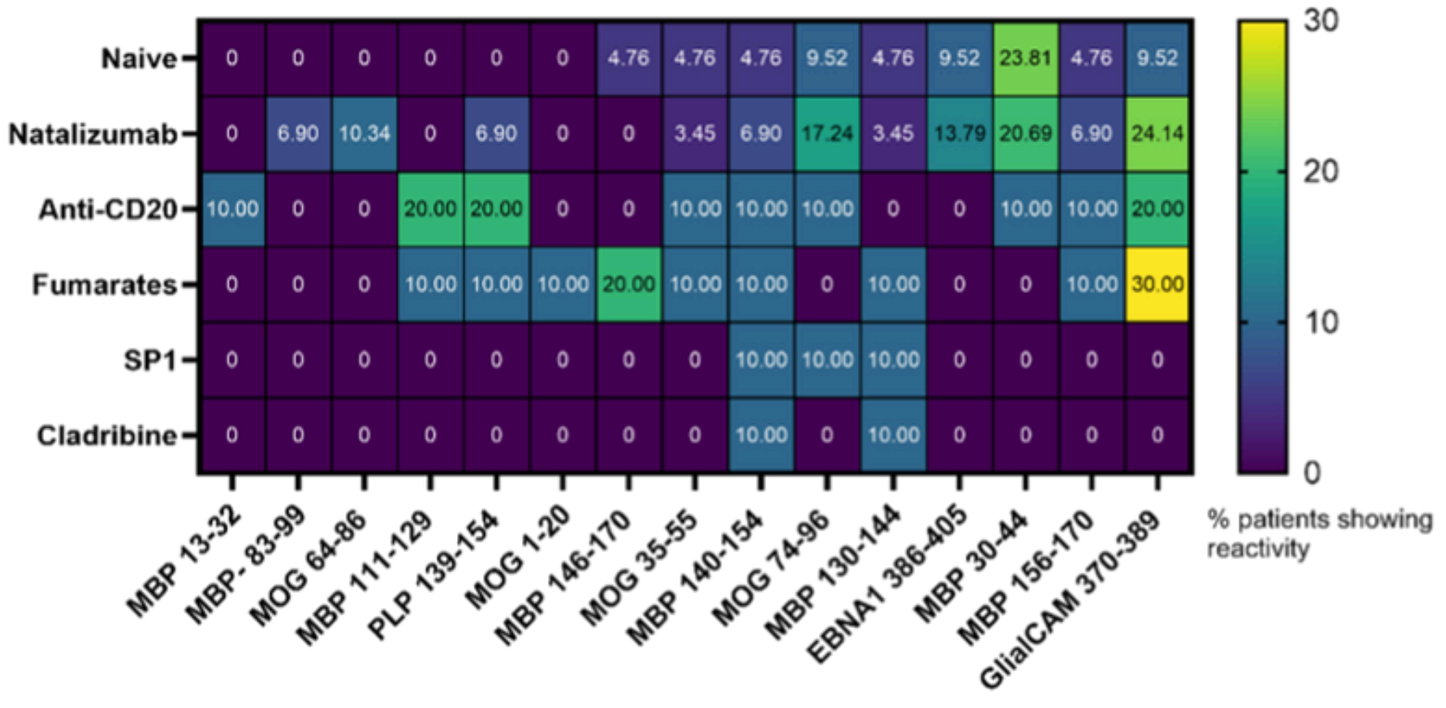

Figure 1. *Immune response against myelin-derived peptides in MS patients and healthy controls. Data were non-normal and analyzed using Dunn's test with a two-stage Benjamini, Krieger, and Yekutieli correction (FDR5%).*

ORAL COMMUNICATIONS - SESSION I: Autoimmunity and Inflammation

Complementary Roles of Urine sCD163, sALCAM, and sC5b-9 in the Non-Invasive Assessment of Lupus Nephritis

Eduardo Gozálvez 1; Jennifer Bolaños 1; Daniel Lorca 1,4; Francisca Santiago 1; Cecilia Garbarino 3; Gerard Espinosa 3,4;Luis F. Quintana 2,4; Marc Xipell 2,4; Francisco Lozano 2,4

1Servei d´Immunologia, Centre de Diagnòstic Biomèdic, Barcelona; 2Servei de Nefrologia i Trasplantament Renal, InstitutClínic de Nefrologia i Urologia, Barcelona; 3Servei de Malalties Autoimmunes, Institut Clínic de Medicina iDermatologia, Barcelona; 4Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), Barcelona

Background & Objective: Lupus nephritis (LN) is a major cause of morbidity in systemic lupus erythematosus (SLE). Although renalbiopsy remains the diagnostic gold standard, its invasive nature limits longitudinal monitoring. Urine offers anon-invasive "renal window," reflecting glomerular and tubular injury through protein biomarkers. Prior studies identified soluble ALCAM/CD166, CD163, and C5b-9 as markers of epithelial, macrophage, and complement activation, respectively. We assessed whether urine sALCAM, sCD163, and sC5b-9 (with serum sALCAM and sCD163) deviate from normal reference values.

Methods: In a cross-sectional cohort of 66 patients with active LN, urine (n=66) and serum (n=64) concentrations of sALCAM, sCD163, and sC5b-9 at the time of renal biopsy were quantified by ELISA. Patient values were compared with normal reference ranges using one-sample t-tests, Wilcoxon signed-rank, and sign tests on both raw and $\log_{10}$-transformed data. Associations between serum and urine biomarkers were examined with Spearman's rank correlation (ρ).

Results: Urine sCD163 was markedly elevated in all patients compared with its undetectable baseline (t = 6.8×10^{-9}; Wilcoxon = 8.2×10^{-13}; sign = 1.4×10^{-20}). Urine sALCAM exceeded the upper limit of normal in 44% of cases (t = 7.98×10^{-4}), though median-based tests were inconclusive; after $\log_{10}$ transformation, differences lost significance, suggesting a high-value subset drove the elevation. Urine sC5b-9 was detectable in 41% (t = 0.0023) yet non-significant by nonparametric tests. Serum biomarkers were not elevated. Correlation analyses showed moderate serum–urine association for sCD163 (ρ = 0.42) and weak for sALCAM (ρ =0.20).

Conclusions: Urine sCD163 emerges as a robust marker of intrarenal inflammation, with potential value for monitoring.sALCAM provides complementary, heterogeneous information, while urinary sC5b-9 identifies a subsetwith complement activation. Together, these biomarkers—particularly sCD163—may enable non-invasive assessment of LN activity pending external validation and correlation with histologic and clinical indices.

ORAL COMMUNICATIONS - SESSION II: Basic Immunology

Dendritic cells exposed to autologous In Vitro–Generated Photopheresates Exert a Suppressive Effect on naive CD4 T cells

Hendrik Veltman 1,3; Jana Willemyns 1,3; Alex Agundez 1,3; Federico Fondelli 1; Jose Cancino Mesa 3; Eva Martinez-Caceres 1,2,3; Dr. Maria Iglesias-Escudero 1,2

1 Immunopathalogy group, Institute Germans Trias I Pujol; 2 Department of Immunology, Institute Germans Trias I Pujol; 3 Department of Cellular Biology, Physiology and Immunology, Universitat Autònoma de Barcelona

The search for alternative therapies specifically controlling allogeneic responses in solid organtransplantation (SOT) is fueling renewed interest in extracorporeal photopheresis (ECP). During ECP, leukocytes are exposed to 8-methoxypsoralen, sensitizing cells to ultraviolet-A light. Irradiation generates a mixture of living and dying leukocytes that is subsequently administered to the patient. The potential of ECP in SOT remains insufficiently explored, partly due to a limited understanding of its mechanism of action. The goal of our research was therefore to elucidate the effects of photopheresates on dendritic cells (DCs) in vitro, as these cells are believed to play a key role in ECP-induced transplant tolerance.

Monocyte-derived immature (iDCs) and mature dendritic cells (mDCs) were generated in vitro and co-cultured for 48 hours with autologous ECP-treated, stimulated CD4 T cells, referred to as photopheresate and stimulated photopheresate. Lipopolysaccharide (LPS)-stimulated DCs and DCs cultured alone served as positive and negative controls, respectively. DC maturation was analyzed by flow cytometry using CD14, HLA-DR, CD83, CD86, and CCR7. Subsequently, DCs were co-cultured with autologous naïve CD4+ T cells to evaluate proliferation and Treg induction over six days.

Co-culture with photopheresate did not increase CD86, HLA-DR, or CCR7 expression and slightly reduced CD83. Naïve CD4 T cell proliferation was reduced by 25% and 60% when co-cultured with iDCs (n=3) and mDCs (n=4), respectively. Interestingly, iDCs - but not mDCs - previously exposed to activated CD4 T cells promoted expansion of Tregs in the secondary co-culture, with the highest increase in iDCs exposed to activated photopheresate (11% vs. ~2.3% in control conditions, n=3).

In conclusion, CD4 T cell death after in vitro ECP has no immunogenic effect on DCs. However, iDCs exposed to activated photopheresate substantially increase Treg frequency, which may underlie reduced naïve CD4 T cell proliferation. Further characterization of these cells is needed to assess their regulatory capacity.

ORAL COMMUNICATIONS - SESSION II: Basic Immunology

CD244 as a Negative Regulator of Human Macrophage Activity

Aarón Adrián Ochoa 1; Javier Fernández Calles 1; Julen Merino Aboitiz 1; Lidia Maestre 2; Giovana Roncador 2; Ana Angulo 1; Pablo Engel 1

1 Universitat de Barcelona; 2 Spanish National Cancer Research Center (CNIO)

CD244 (2B4, SLAMF4) and its ligand CD48 play key roles in immune modulation across viral infections, autoimmunity, cancer, and immunodeficiencies. Their interaction influences immune cell activation, exhaustion, and immune evasion, with context-dependent effects that can either activate or inhibit immune responses. While CD244 engagement enhances the cytotoxicity and cytokine production of NK and cytotoxic T cells, its role in human macrophages remains unclear.

In this study, we characterized the expression of CD244 and CD48 on monocytes and macrophages (M0, M1, and M2) derived from THP-1 cells and primary human cells using flow cytometry. To further explore CD244 function, we used an anti-CD244 blocking antibody (clone 2B4.69) and evaluated its effects on cytokine production and phagocytic activity in LPS-stimulated THP-1 cells.

Our results showed that the CD244 ligand, CD48, was highly upregulated in both THP-1 and primary human macrophages, particularly in M1 cells. While CD244 expression remained unchanged in differentiated THP-1 cells, it was slightly downregulated during the differentiation of primary human macrophages from M0 to M1 and M2. Notably, CD244 blockade significantly increased cytokine production (IL-6, IL-8, and TNF-α) in LPS-activated THP-1 macrophages, with the most pronounced effect observed in M1 cells, where these cytokines are predominantly produced. Furthermore, the blockade also significantly enhanced phagocytic activity in LPS-stimulated THP-1 cells.

Our data reveal an inhibitory role of CD244 in human macrophages, supporting the therapeutic potential of targeting the CD244–CD48 axis to reprogram macrophage function and improve anti-tumor and anti-viral immunity.

ORAL COMMUNICATIONS - SESSION II: Basic Immunology

CD6 expression impacts B cell biology

Laura Carrillo-Serradell 1; Juan Antonio Piñeyroa 2,3; Sílvia Patrícia Fernández Martínez 4; Miguel Ángel de la Fuente García 4; Pablo Bousquets-Muñoz 5,6; Violeta Planells-Romeo 1; Lucía Aragón-Serrano 1; Sergi Casadó-Llombart 1; Dolors Colomer 3,6,7,8; María Velasco-de Andrés 1; María Simarro 4; Xose S. Puente 5,6; Julio Delgado 2,6; Pablo Mozas 2,9; Francisco Lozano 1,11,12

1 Immunoreceptors del Sistema Innat i Adaptatiu, IDIBAPS, Barcelona.; 2 Servei d'Hematologia, Hospital Clínic de Barcelona, Barcelona.; 3 Terapias experimentales en neoplasias linfoides, IDIBAPS, Barcelona.; 4 Instituto de Biomedicina y Genética Molecular de Valladolid, Universidad de Valladolid, Valladolid.; 5 Instituto de Oncología, Departamento de Bioquímica y Biología Molecular, Universidad de Oviedo.; 6 Centro de Investigación Biomédica en Red de Cáncer (CIBERONC), Madrid.; 7 Secció Hematopatologia, Servei Anatomia patològica, CDB, Hospital Clínic Barcelona, Barcelona.; 8 Departament de Fonaments Clínics, Universitat de Barcelona (UB), Barcelona.; 9 Neoplasies Limfoides, IDIBAPS, Barcelona.; 10 Departament de Medicina, Universitat de Barcelona, Barcelona, Spain.; 11 Servei d'Immunologia, CDB, Hospital Clínic Barcelona, Barcelona.; 12 Departament de Biomedicina, Universitat de Barcelona, Barcelona.

CD6 is a signal transducing scavenger lymphocytic receptor mainly expressed by all T cells, but also the small subset of B1 cells and B1-derived cells (e.g., Chronic lymphocytic leukemia, CLL). CD6 is involved 1) in cell-to-cell (T-B, T-DC, T-Mac) adhesive contacts thanks to its interaction with CD166/ALCAM, a member of the Ig-SF of adhesion molecules, as well as 2) in the fine tuning of signals derived from antigen-specific clonotypic receptor from T cells (TCR). Recent signalosome studies reveal that CD6 engagement recruits both stimulatory and inhibitory intracellular effectors upon TCR triggering. However, its involvement in B1 cell physiology remains less defined. To address this, we employed neoplastic B cells (i.e., Daudi and Chronic lymphocytic leukemia (CLL)) as experimental models of CD6-positive and -negative B cells. When CD6 expression was monitored in primary CLL samples - all CD5-positive - we observed higher CD6 mRNA levels correlating with longer time to first treatment (TTFT) and more frequently mutated IGHV status, both indicators of favorable prognosis. Phenotypic and transcriptomic and functional analyses of Daudi cells lentivirally transduced and selected for stable and high-level CD6 expression (Daudi-CD6+) showed relevant changes, including the down/up-regulation of B-cell co-stimulatory surface receptors and down-regulation of cancer-promoting gene programs (e.g., MYC-regulated genes, G2M checkpoint regulators) and dampened energetic profile (respiratory capacity ATP production) with regard to controls (Daudi-ZsGreen). In summary, our findings highlight CD6 as a novel modulator of B cell gene expression, tonic signaling, and tumor progression.

ORAL COMMUNICATIONS - SESSION II: Basic Immunology

Validation of G protein-coupled receptors (GPCRs) antibodies: new CD Assignments and Expression Profiles from the HLDA11 Workshop

Javier Fernández-Calles 1; Daniela Kužílková 2; Fanny Hedin 3; Violeta Bakardjieva- Mihaylova 2; Karolina Škvárová Kramarzová 2; Menno C van Zelm 4; Antonio Cosma 3; Tomas Kalina 2; Pablo Engel 1

1 University of Barcelona Faculty of Medicine and Health Sciences, Biomedical Sciences, Spain; 2 CLIP (Childhood Leukaemia Investigation Prague), Second Faculty of Medicine, Charles University, Dep; 3Luxembourg Institute of Health, Esch-sur-Alzette, Luxembourg; 4Erasmus MC, University Medical Center, Rotterdam, The Netherlands

Monoclonal antibodies (mAbs) directed against cell-surface antigens are indispensable in modern immunology, serving as fundamental tools in basic research, diagnostics, and therapeutic innovation. Their impact has been particularly notable in the development of treatments for cancer and autoimmune disorders. Since its inception by the Human Leukocyte Differentiation Antigens (HLDA) Workshops and subsequent endorsement by the International Union of Immunological Societies (IUIS), the CD nomenclature system has provided a coherent and standardized framework for cataloguing these antibodies and their corresponding molecular targets.

G protein-coupled receptors (GPCRs) constitute the most extensive receptor family on cell membranes and play critical roles in regulating both innate and adaptive immune responses. Despite their biological significance, GPCRs have historically been underrepresented in the catalog of well-characterized monoclonal antibodies compared with other receptor families such as the immunoglobulin superfamily.

During the Eleventh HLDA Workshop (HLDA11), anti-GPCR mAbs were systematically assessed for their reactivity with primary peripheral blood leukocytes, established and transfected cell lines using flow cytometry. These analyses facilitated comprehensive antibody validation and defined expression patterns across immune cell subsets. As a result, fourteen new CD designations were assigned to GPCRs expressed on immune cells: CD198 (CCR8), CD199 (CCR9), CD372 (CCR10), CD373 (CX3CR1), CD374 (XCR1), CD375 (GPR15), CD376 (GPR26), CD377 (SSTR3), CD378 (C3AR1), CDw379 (FPR2), CD380 (LTB4R), CDw381 (GPR138), CDw382 (F2RL1), and CD383 (P2RX7).

Here, we present the updated CD classification for GPCR family members, accompanied by quantitative data on their expression profiles in diverse immune cell populations. We also provide an overview of the antibody validation results and explore the therapeutic potential of targeting these molecules in immune-mediated diseases and cancer. Collectively, these findings highlight the continued evolution of the HLDA workshops and underscore the adaptability and relevance of the CD nomenclature system as immunological research advances.

ORAL COMMUNICATIONS - SESSION II: Basic Immunology

Towards Large-Scale Immune Profiling: Preliminary Evaluation of a Spectral Panel in Healthy Donors

Sara Monreal-Peinado 1,2,3,4; Marco A Fernández 3; María Méndez 1; Annika Nack 1; Agueda Ancochea 1,5; Armando Ruiz 1; Eva M Martínez-Cáceres 1,2,3; Joan Puñet-Ortiz 3,4; Aina Teniente-Serra 1,2

1 Germans Trias i Pujol University Hospital; 2 Univ Autonoma Barcelona; 3 Germans Trias i Pujol Research Institute (IGTP); 4 Josep Carreras Leukaemia Research Institute (IJC); 5 Banc de Sang I Teixits (BST)

Background: Immunophenotypic profiling of healthy donors is essential to establish reference values that support the clinical interpretation of immune alterations. High-dimensional spectral flow cytometry enables simultaneous detection of numerous markers, providing a comprehensive view of the immune system, although validation against conventional methods is required.

Objectives: 1) To compare a 39-color spectral panel with conventional clinical panels defined by the Human Immunology Project Consortium (HIPC); and (2) to evaluate age-related changes in adaptive immune subpopulations, activation markers and immune checkpoints in healthy donors.

Materials and Methods: Fresh peripheral blood samples from immunologically healthy donors were collected through the Blood and Tissue Bank (BST) or hospital outpatient consultations (HUGTIP).Eleven samples were processed in parallel using spectral and conventional cytometry, applying the HIPC analytical strategy. Frequencies of T- and B-cell populations were compared between platforms. In addition, 100 healthy donors (aged 9 months to 78 years) were analyzed using the 39-color spectral panel.In this phase, focusing on adaptative immunity, percentages of T- and B-cell subsets, activation markers, and immune checkpoint molecules were examined for correlations with age.

Results: T- and B-cell frequencies showed strong correlation between both cytometry platforms. With increasing age, $CD4^+$ and $CD8^+$ naïve cells decreased, while $CD4^+$ CM, $CD8^+$ CM, $CD8^+$TEMRA and memory regulatory T cells increased. HLA-DR^+ T-cells increased with age, whereas $CD38^+$ T-cells declined. $TIGIT^+$ cells progressively increased within $CD3^+$, $CD4^+$, $CD8^+$ and $CD8^+$ maturation subsets. No correlation was observed in the other immune checkpointsmarkers analyzed.

Conclusions: The validated spectral panel provides reliable alignment with conventional cytometry approaches and extends analytical depth for immune profiling. Preliminary findings confirm known age-associated patterns in T-cell subsets and offer baseline data on activation and checkpoint markers. These results lay the groundwork for large-scale immune mapping in the upcoming 500-donor cohort.

ORAL COMMUNICATIONS - SESSION III:
Immunity and Infection

The protective potential of vaccination against LCMV-induced Post-Acute Infection Syndromes

Paula Cebollada Rica 1; Sergi López-Serrano 1; Yannick O. Alexandre 6; Beatriz Martin-Mur 2; Anna Esteve-Codina 2; Stefanya Padilla Illescas 1; Raúl Gomez-Riera 3; Nadia Halidi 3; Marta Sisteré-Oró1 ; Valentina Casella 1; Roberto Güerri-Fernandez 4; Montserrat Plana 9; Nicole Ludwig 5; Eckart Meese 5; Scott N. Mueller 6; Gennady Bocharov 7; Jordi Argilaguet8; Andreas Meyerhans 1,10

1 Universitat Pompeu Fabra; 2 Centre Nacional d'Anàlisi Genòmica; 3 Centre de Regulació Genòmica; 4 Hospital del Mar Research Institute; 5 University of Saarland; 6 University of Melbourne; 7 Russian Academy of Sciences; 8 IRTA-CReSA(Centre de Recerca en Sanitat Animal); 9 Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS); 10 ICREA

Post-Acute Infection Syndromes (PAIS) are an insufficiently understood condition that can appear after the recovery from an acute viral infection. While the immune system usually clears acute infections within weeks and establishes virus-specific memory, mounting evidence indicates that persistent or delayed symptoms can occur long after viral control. To investigate the underlying immune mechanisms, we used the Lymphocytic choriomeningitis virus (LCMV) mouse model. Our studies demonstrate that LCMV infection induces a transient but extended state of immune suppression, persisting even after the virus has been controlled. The loss of immune competence is associated with profound disruption of splenic lymphatic tissue, including cellular compartmentalization and extracellular matrix integrity. Notably, the recovery of this tissue damage requires substantially more time than viral control, underscoring a prolonged period of impaired immune function. We found that vaccination against LCMV preserves lymphatic tissue integrity and protects against subsequent immune dysfunction, highlighting the protective role of vaccines beyond viral control. These results establish LCMV infection as a valuable experimental model for immune-related PAIS and reinforce the importance of vaccination in preventing both acute infection-related tissue damage and potential long-term complications. Our study reveals the complex interplay of viral infections, immune responses and tissue integrity, challenging the notion that hosts fully return to their pre-infection health state after acute viral infections. This research sheds light on the long-term effects of acute viral infections and emphasizes the importance of preserving immune tissue structure to ensure sustained health and immunity.

ORAL COMMUNICATIONS - SESSION III:
Immunity and Infection

Humoral immunity after PHH-1V COVID-19 booster vaccination in adults with immunosuppressive conditions

Dàlia Raïch-Regue 1; Ester Aparicio1; Edwards Pradenas 1; Rytis Boreika 1; Benjamin Trinité 1; Yovaninna Alarcón-Soto 2; Susana Benet 2; Jorge Carrillo 1; Laura Ferrer 3; Julia G. Prado 1,4,5; Beatriz Mothe 2; Julià Blanco 1; Nuria Izquierdo-Useros 1,5

1 IrsiCaixa, Hospital Germans Trias i Pujol, 08916, Badalona, Spain; 2 Fundació Lluita contra les Infeccions, Infectious Diseases Department, Hospital Germans Trias i Pujo; 3 HIPRA, Avinguda de la Selva, 135, 17170, Amer, Girona, Spain; 4 Germans Trias i Pujol Research Institute (IGTP), Badalona, Spain; 5 CIBER Enfermedades Infecciosas (CIBERINFEC), Instituto de Salud Carlos III, Madrid, Spain

PHH-1V is an adjuvanted recombinant protein vaccine based on SARS-CoV-2 receptor-binding domain. It was evaluated as a booster in 238 individuals with pre-existing immunosuppressive conditions (IC) in a Phase IIb/III, open-label, single-arm, multicentre study (HIPRA-HH-4; NCT05303402). The humoral immunogenicity of PHH-1V booster vaccination was analysed in 5 groups of participants with IC at high risk of adverse COVID-19 outcomes: HIV infection (PWH, n=59, <400 CD4+ T cells/mm3), kidney transplant (KTx, n=35), chronic haemodialysis (HD, n=55), primary antibody deficiencies (PAD, n=24), and autoimmune diseases (AID, n=55).

Total binding antibodies measured by electrochemiluminescence immunoassay and neutralising antibodies (nAb) determined by virus/pseudovirus neutralisation assays against 5 SARS-CoV-2 variants were analysed in serum samples at baseline (D0) and day 14 (D14) after PHH-1V booster. Mixed-effects models were used on log-transformed data including the visit, COVID-19 history and age (⩾18 to <65; ⩾65 years) as fixed effects. Results are reported as geometric mean fold rise (GMFR) (95% confidence interval).

At D14, total binding antibodies increased in all IC [GMFR between 7.2 (5.6, 9.3) in PWH and 2.3 (1.6, 3.4 in PAD], except for AID [GMFR 1.2 (1.0, 1.6)]. Boosting of nAb against Omicron BA.2 in PWH [GMFR 4.5 (3.5, 5.8)] was comparable to the HD group [GMFR 5.6 (3.7, 8.5)] and higher than in KTx, PAD and AID (GMFR between 2.6 and 1.2), similarly to nAb against D614G. For Omicron BA.1, Omicron BA.4/5, and Beta, the rise of nAb in HD (GMFR between 7.8 and 5.5) was higher than in KTx (GMFR 4.7 – 3.1) and PAD (GMFR 3.0 – 1.7), and not significant for AID (GMFR 1.4 – 1.2).

Regardless of COVID-19 medical history, PHH-1V booster vaccination elicited significant increases in total binding and neutralising antibodies at D14 in PWH, KTx, HD, and PAD, but not in the AID group, possibly due to the anti-CD20 therapy.

ORAL COMMUNICATIONS - SESSION III:
Immunity and Infection

Broad-spectrum anti-ACE2 neutralizing antibodies for protection against SARS-CoV-2 variants and emerging coronaviruses

Pablo Hernández-Luis 1; Eloi Franco-Trepat 2; Rytis Boreika 2; Itziar Erkizia 2; Nuria Izquierdo-Useros 2,3; Pablo Engel 1; Ana Angulo 1

1 University of Barcelona, Immunology Unit, Department of Biomedical Sciences, Faculty of Medicine; 2 IrsiCaixa, Germans Trias i Pujol Research Institute (IGPT); 3 CIBER Enfermedades Infecciosas (CIBERINFEC), Instituto de Salud Carlos III

The ongoing emergence of new SARS-CoV-2 variants capable to evade both vaccine- and infection-elicited protective immunity, along with the potential threat of novel coronaviruses, underscore the urgent need to develop broad-acting effective therapeutic strategies. Here, we report the development of two chimeric monoclonal anti-human angiotensin-converting enzyme 2 (ACE2) antibodies, mAbB1 and mAbB2, able to efficiently block ACE2-viral receptor-binding domain (RBD) interactions. Importantly, neither of the two monoclonal antibodies (mAbs) affected ACE2 enzymatic activity or triggered significant internalization of he receptor. Surface plasmon resonance (SPR) analysis revealed that these mAbs displayed binding affinities for the ACE2 receptor within the subnanomolar range. ELISA assays confirmed their capacity to recognize different ACE2 polymorphisms. Notably, both mAbs prevented the interaction of RBDs from diverse SARS-CoV-2 variants and other coronaviruses, such as SARS-CoV, HCoV-NL63, or pangolin CoV-GD, to the cellular receptor. In agreement with these results, neutralization studies conducted in Vero E6 cells demonstrated the ability of mAbB1 and mAbB2 to potently inhibit the infection of diverse replication-competent SARS-CoV-2 variants, including D614G, Beta B.1.351, Omicron BA.1.1, KP.2 or XEC. We also demonstrate that the most potent of the two mAbs, mAbB1, when administered therapeutically or prophylactically, protects K18-hACE2 transgenic mice against SARS-CoV-2 Omicron BA1.1 infection, significantly reducing lung viral loads. These mAbs could offer an effective approach to control both circulating and emerging ACE2-dependent coronaviruses, particularly benefiting immunocompromised and other vulnerable populations.

ORAL COMMUNICATIONS - SESSION III:
Immunity and Infection

Adoptive cell transfer of NK-92 cells expressing a CD5-based Chimeric Antigen Receptor (SRCD5CAR-NK92) to treat a clinically relevant invasive fungal infection

Maria Velasco-de-Andrés 1; Pedro Puerta-Alcalde 2,3; Jiri Eitler 4,5,6; Lydia Krutz 4; Mariana Chumbita 2; Marta Español-Rego 7; Cristina Català 1; Laura Carrillo-Serradell 1; Violeta Planells-Romeo 1; Lucía Aragón-Serrano 1; Andrea Vergara 8,9,10,11; Amanda Isabel Pérez-Valencia 12; Patricia Monzó-Gallo 2; Antonio Gallardo-Pizarro 2; María Teresa Cibeira 12; Alexandru Vlagea 7; José Luis Caro 7; Eduard Palou 7; María Suárez-Lledó 12; Alex Soriano 2,3; Gonzalo Calvo 13,14; Álvaro Urbano-Ispizua 12; Torsten Tonn 4,5,6; Carolina Garcia-Vidal 2,3; Francisco Lozano 1,7,15

1 Immunoreceptors del sistema innat i adaptatiu, Institut d'Investigacions Biomèdiques August Pi i Sun; 2 Department de Malalties Infeccioses, Hospital Clínic de Barcelona, Barcelona, Spain; 3 Departament de Medicina, Universitat de Barcelona (UB), Barcelona, Spain; 4 Experimental Transfusion Medicine, Faculty of Medicine Carl Gustav Carus, Dresden University of Tech; 5 Institute for Transfusion Medicine Dresden, German Red Cross Blood Donation Service North-East, Dres; 6 German Cancer Consortium (DKTK), Partner Site Dresden, Dresden, Germany.; 7 Servei d'Immunologia, Centre de Diagnòstic Biomèdic, Hospital Clínic de Barcelona, Barcelona, Spain; 8 Servei de Microbiologia i Parasitologia-CDB, Hospital Clínic de Barcelona, Spain.; 9 Institut de Salut Global (ISGlobal), Barcelona, Spain.; 10 Departament de Fonaments Clínics, University of Barcelona, Barcelona, Spain; 11 CIBER Enfermedades Infecciosas (CIBERINFEC), Madrid, Spain.; 12 Servei de Hematologia, Hospital Clínic de Barcelona, Barcelona, Spain.; 13 Servei de Farmacologia Clínica, Area Medicament, Hospital Clínic de Barcelona, Barcelona, Spain; 14 Clinical Pharmacology, Institut d'Investigacions Biomediques August Pi i Sunyer (IDIBAPS), Barcelona; 15 Departament de Biomedicina, Universitat de Barcelona (UB), Barcelona, Spain.

Invasive fungal infections (IFI) remain a major therapeutic challenge due to limited antifungal options, emergence of multidrug-resistant (MDR) strains and high mortality rates. This has turned up the development of immune-based therapies to restore and/or enhance anti-fungal immune responses into a clinical priority. We have recently demonstrated that the intrinsic antifungal activity of primary NK cells can be potentiated by endowing them with a CD5-based second-generation chimeric antigen receptor (SRCD5CAR) targeting β-glucans, a constitutive and specific component of fungal cell walls. Building on this, we aimed to explore the use of SRCD5CAR-engineered NK-92 cells as ready-to-use adoptive cell transfer therapy for clinically relevant IFI. NK-92 cells were lentivirally transduced and selected for stable and high-level expression of the SRCD5CAR. Antifungal activity of SRCD5CAR-NK92 cells was assessed by in vitro killing assays against different fungal species, and in vivo survival analysis of fungal-infected immunodeficient NSG mice, prior to an authorized compassionate use in an IFI patient.SRCD5CAR-NK92 cells demonstrated enhanced antifungal activity in pre-clinical in vitro and in vivo models of Candida albicans and Cryptococcus neoformans infection. Ex vivo killing assays confirmed activity against a MDR F. petroliphilum isolate obtained from a patient with IFI co-occurring with a hematological malignancy. Following compassionate use approval, the patient received escalating intravenous infusions of irradiated SRCD5CAR-NK92 cells at 2-5 day intervals, without significant local or systemic adverse effects (e.g., cytokine storm or alloreactivity). Despite signs of antifungal activity based on clinical and laboratory monitoring, the patient ultimately succumbed due to progression of the underlying hematological malignancy. In conclusion, the present results highlight the potential of SRCD5CAR-NK92 cell therapy as a safe, off-the-shelf therapeutic strategy for severe IFI, underscoring the need for further investigation on efficacy and long-term outcomes.

ORAL COMMUNICATIONS - SESSION III:
Immunity and Infection

From Data to Diagnosis: Implementing a multi-omic approach in inborn errors of immunity

Laura Batlle-Masó 1,2; Nerea Moreno-Ruiz 3; Laith I Moushib 1,2,4,5; Andrea Martín-Nalda 2,6,7; Jacques Riviere 2,6,7; Aina Aguiló-Cucurull 1,2,8; Janire Perurena-Prieto 1,2,4,8; Pere Soler-Palacín 2,4,6,7; Roger Colobran 1,2,4,8,9

1 Translational Immunology Research Group, Vall d'Hebron Research Institute (VHIR), Vall d'Hebron Barcelona Hospital Campus, Barcelona, Catalonia, Spain; 2 effrey Modell Diagnostic and Research Center for Primary Immunodeficiencies, Barcelona, Catalonia, Spain; 3 Intitute of Evolutionary Biology (UPF-CSIC), Department of Medicine and Life Sciences, Universitat Pompeu Fabra, Barcelona, Spain; 4 Universitat Autònoma de Barcelona (UAB), Bellaterra, Catalonia, Spain; 5 National Center of Hematology, Al-Mustansiriyah University, Baghdad, Iraq; 6 Infection and Immunity in Pediatric Patient, Vall d'Hebron, Research Institute (VHIR), Vall d'Hebron Barcelona Hospital Campus, Barcelona, Catalonia, Spain; 7 Pediatric Infectious Diseases and Immunodeficiencies Unit, Hospital Universitari Vall d'Hebron (HUVH), Vall d'Hebron Barcelona Hospital Campus, Barcelona, Catalonia, Spain; 8 Immunology Division, Hospital Universitari Vall d'Hebron (HUVH), Vall d'Hebron Barcelona Hospital Campus, Barcelona, Catalonia, Spain; 9 Department of Clinical and Molecular Genetics, Hospital Universitari Vall d'Hebron (HUVH), Vall d'Hebron Barcelona Hospital Campus, Barcelona, Catalonia, Spain

Inborn errors of immunity (IEI) are a clinically, immunologically, and genetically heterogeneous group of disorders, with more than 500 monogenic defects identified to date. The advent of next-generation sequencing technologies has accelerated the discovery of novel IEI-causing genes. However, the diagnostic yield achieved across different cohorts using standard NGS-based approaches routinely employed in genetic diagnostics (panels and WES) typically remains below 50%. Therefore, new strategies are needed to close the diagnostic gap in undiagnosed patients.

We performed an integrative analysis combining genome and transcriptome profiling in 25 IEI patients and their families to identify novel genetic causes of IEI. All selected patients had a clinical diagnosis of IEI but no conclusive genetic findings after WES. WGS was performed in patient–parent trios, alongside peripheral blood transcriptome sequencing (RNA-seq) of the patients and age- and sex-matched controls. Interpretation of the coding regions from WGS was complemented by RNA-seq analysis using the DROPpipeline to detect genes with aberrant expression, splicing defects, and monoallelic expression.

In one patient presenting with viral infections and hemophagocytic lymphohistiocytosis episodes, we identified markedly reduced expression and aberrant splicing of STAT2. This led to the discovery of two heterozygous variants in trans: one splice-site and one deep intronic variant, each inherited from a different parent. Pending functional validation, these variants are likely to contribute to the patient's phenotype through the generation of aberrant isoforms from both alleles.

In conclusion, integrating genome and transcriptome profiling can enhance the diagnostic yield in complex IEI cases. This approach has immediate clinical relevance, improving diagnostic precision and informing optimal patient management and treatment selection.

ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology

Bone marrow immune and stromal niche remodeling in myelodysplastic neoplasm patients under erythropoietic therapies by 29-color full-spectrum flow cytometry

Clàudia Pellín Jou 1; Iñaki Salvador Corres 1; Pamela Acha 1; Alba Puyelo 1; Sara Garrido Paniagua 1; Pilar Velarde 1; Maria Landwehr 1; Ana Pérez 1; Laura Gallur Cuenca 1; Silvia Saumell 1; Julia Montoro 1; Andrés Jerez 1; Laura Palomo Sanchís 2; David Valcarcel Ferreiras 1

1 Vall d'Hebron Institute of Oncology (VHIO); 2 Institut Català d'Oncologia

Introduction: The immune and stromal components of the bone marrow (BM) niche are key regulators of myelodysplastic neoplasms (MDS), yet their remodeling under therapy remains poorly characterized. We designed and optimized a 29-color full-spectrum flow cytometry panel to profile hematopoietic and non-hematopoietic populations in low-risk MDS (LR-MDS) patients and applied it to longitudinal samples from treated individuals.

Methods: Fluorochrome/protein assignments were optimized based on antigen density, co-expression, and spectral spread, following i4MDS consortium guidelines. Dead cells and lymphoid lineages (CD3, CD7, CD19, CD56) were excluded using a dump channel and viability dye. Cryopreserved BM mononuclear cells from three LR-MDS patients with ring sideroblasts were analyzed at diagnosis and after 6 months of erythropoietic therapy (one ESA, two TGF-β ligand traps), alongside an age-matched non-MDS control.

Results: The optimized panel resolved major myeloid and stromal subsets with high sensitivity. Baseline profiling showed increased endothelial progenitor cells (+65%) and reduced mesenchymal stromal cells (−52%) in MDS versus control. After treatment, TGF-β ligand traps induced pronounced remodeling: reduction of endothelial precursors (−79%), expansion of functional MSCs (+390–1161%), decreased classical monocytes, and enrichment of M2/CD169⁺ macrophages, consistent with a niche supportive of erythroid recovery. Non-classical monocytes and MDSC-G/MDSC-M ratios increased, suggesting reduced immunosuppression. Dendritic cell subsets showed heterogeneous modulation, with plasmacytoid DCs markedly increased (+810%) but displaying divergent activation profiles. ESA therapy induced milder and less coordinated immune changes.

Conclusions: This 29-color cytometry panel enables high-dimensional characterization of the BM immune–stromal landscape. LR-MDS patients display an immune microenvironment skewed toward a pro-inflammatory phenotype linked to ineffective hematopoiesis. Under erythropoietic therapy, the niche remodels toward a less inflammatory, more erythropoietic state via distinct cellular routes. Stromal reprogramming appears deeper under TGF-β ligand traps than ESA, supporting therapy-specific modulation of immune–stromal interactions.

ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology

Immortalized NK cells expressing a CD6-based CAR effectively target non-hematological tumour cells in vitro

Lucía Aragón Serrano 1; Laura Carrillo i Serradell 1; Violeta Planells i Romeo 1; Jara Martín Serrano 1; Francisco Lozano 1,2; María Velasco de Andrés 1

1 Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS); 2 Hospital Clinic de Barcelona

Adoptive transfer of Chimeric Antigen Receptor (CAR)-T cells has shown good efficacy in haematological malignancies, while its effectiveness in solid tumours remains limited. CARs often target tumour-associated antigens (TAAs), as truly tumour-specific antigens (TSAs) are difficult to identify. However, TAA targeting can lead to on-target off-tumour toxicity due to spread expression on normal tissues. To mitigate this risk, tuning-down scFv affinity has been explored to enable preferential targeting of TAA-overexpressing cells. Alternatively, physiological receptor–ligand pairs, which naturally exhibit lower binding affinities than scFv, can be used as a strategy to improve safety while maintaining efficacy. On this basis we have generated a CD6-based second-generation CAR (SRCD6CAR) encompassing the whole extracellular region of CD6 and the cytoplasmic activation motifs of CD3zeta and CD137/41BB to target tumour cells overexpressing CD166/ALCAM and/or CD318/CDCP-1, two bona fide CD6 ligands. To avoid some constraints imposed by autologous CAR-T cells (e.g., cytokine-release syndrome and neurotoxicity), we explored the alternative use of allogeneic NK cells, which present milder toxicities. To that end, the NK-derived KHYG-1 cell line was lentivirally transduced and selected for stable and high-level expression of the SRCD6CAR and of a variant lacking the CD6's CD166-binding domain (Δ3SRCD6CAR). Co-culture assays with CD166- and CD318-positive cells of ovary carcinoma (SKOV-3) or triple-negative breast adenocarcinoma (MDA-MB-231) origin demonstrated enhanced SRCD6CAR-KHYG1 cell activation and superior cytotoxicity compared to untransduced (UT) and Δ3SRCD6CAR KHYG-1 cells used as controls. The analysis of co-culture supernatants with SKOV-3 cells also showed higher secretion levels of INF-γ, TNF-α, perforin, granzyme B, CCL-3/MIP-1α, CCL-4/MIP-1β, and CCL-5/RANTES for SRCD6CAR KHYG-1 cells. In conclusion, our results support the notion that SRCD6CAR KHYG-1 cells will specifically target tumour cells expressing high CD166 and/or CD318 surface levels, though further experiments are warranted for efficacy and biosecurity evaluation.

ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology

Rectifying false-positive results using a competitive antagonist in flow cytometry crossmatch for kidney transplant candidates with rituximab treatment

Ángel García 1; Philipp Trummer 2; Nuria Exposito 1; Maria Josef Puges 1; Jose Luis Caro 1; Eduard Palou1; Clemens Schneeweiß 2; Juan Torres Canizales 1

1 Hospital Clínic de Barcelona; 2 Immusyn GmbH Co. KG

Background: Flow cytometry crossmatch (FCXM) plays an important role prior to kidney transplantation in order to prevent acute antibody-mediated rejection, yet unexpected positive results may occur in recipients with autoimmune diseases, pre-existing antibodies undetected by single-antigen bead assays or biological agents, most notably Rituximab, a monoclonal antibody targeting B lymphocytes, which may erroneously deny patients access to life-saving organ transplants.

Aims: This study aims to determine whether a competitive antagonist (RituxiEx®) can correct false positive FCXM results caused by previous or current Rituximab treatment, without compromising the detection of true positive cases in patients.

Patients and Methods: Serum samples were collected from 10 kidney transplant candidates who had received previous or current Rituximab treatment. Isolated donor cells were adjusted to 1×10^7 cells/ml, and crossmatch assays were performed in parallel, both without and with RituxiEx®, by incubating the cells at a 1:1 ratio. After incubation, the standard FCXM protocol was followed. Samples were analyzed using a DxFlex® cytometer. Anti-HLA DSA antibodies were determined by Single Antigen Luminex assay (Immucor).

Results: Before RituxiEx® treatment, all sera without known DSA showed positive results for B cells. After incubation with RituxiEx®, 7 out of 8 DSA-negative sera corrected their false positive B cell results, while one sample remained weakly positive. The two sera with known DSA remained positive for both B and T cells. When comparing results between crossmatches performed with and without RituxiEx®, no significant differences were observed in the fluorescence of negative and positive controls, nor with using different cellular sources.

Conclusions: Preliminary results demonstrate that RituxiEx® effectively eliminates most nonspecific fluorescence in false positive FCXM sera due to Rituximab, without compromising the detection of anti-HLA antibodies. Implementing RituxiEx® in cases with suspected false positives may aid clinical decision-making based on crossmatch results, thereby optimizing organ allocation for transplantation.

ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology

Validation of the Luminex Flexmap 3D System for Anti-HLA Antibody Detection Using Single Antigen Beads (SAB)

David Rodríguez Martín 1; Juan Manuel Torres Canizales 1; Nuria Expósito Capón 1; Ariadna López Priego 1; Eduard Palou Rivera 1; José Luis Caro Oleas 1

1 Hospital Clínic de Barcelona

The Luminex platform is a widely adopted technology for the immunological assessment of transplant recipients, enabling allelic-level detection of anti-HLA antibody through Single Antigen Beads (SAB). The interpretation of fluorescence levels is crucial for positivity assignment and is a semiquantitative indicator reflecting antibody concentrations, making it a useful tool for sensitization monitoring, donor selection, and delisting protocols.

Our laboratory employs reagents from the commercial manufacturer Immucor LIFECODES® and recently transitioned to the Luminex Flexmap 3D system, replacing the previous Luminex IS200. The Flexmap 3D is capable of reading a 96-well plate in approximately half the time compared to the IS200, and offers the ability to analyze up to 500 different beads per sample—five times more than the IS200—thereby expanding the range of detectable antibodies against different HLA alleles. However, it operates with a broader dynamic range, resulting in overall increased median fluorescence intensity (MFI) values, which requires adjustment of data interpretation and positive/negative assignment criteria.

Validation was performed using 98 serum samples (9407 beads for class I and 9216 beads for class II). Each sample was tested on both systems under identical conditions, and data were analyzed using Match IT!® Antibody Analysis Software. A systematic MFI increase was observed across all beads, including class I, class II and control beads, with an average of 1.75-fold for class I and 1.73-fold for class II antibodies. Consequently, the laboratory cut-off for positivity was updated from 750 to 1300. This new threshold maintains high concordance in positive/negative assignment between platforms (99.56% for class I and 99.52% for class II). When reviewing historical results, MFI variations introduced by the system replacement should be considered in the interpretation.

This validation supports the integration of the Flexmap 3D into routine practice, ensuring ongoing reliability, higher analytical capacity, and improved efficiency in anti-HLA antibody testing.

ORAL COMMUNICATIONS - SESSION IV: Clinical Immunology

Comparison and Validation of in vitro extracorporeal photopheresis Protocols for the analysis of its immunomodulatory effects

Hendrik Veltman 1,3; Alex Agundez 1,3; Jana Willemyns 1,3; Federico Fondelli 1,3; Eva Martinez-Caceres 1,2,3; Maria Iglesias-Escudero 1,2

1 Immunopathology group, Institute Germans Triaj I Pujol; 2 Department of Immunology, Institute Germans Trias I Pujol; 3 Department of Cellular Biology, Physiology and Immunology, Universitat Autònoma de Barcelona

The search for alternative therapies specifically controlling allogeneic responses in solid organ transplantation (SOT) is fueling renewed interest in extracorporeal photopheresis (ECP). During ECP, leukocytes are exposed to 8-methoxypsoralen, sensitizing cells to ultraviolet-A light. Irradiation results in a mixture of living and dying leukocytes, which is subsequently administered to the patient. The potential application in SOT has not been adequately investigated, in part due to a limited understanding of the working principal. Thus, the goal of our research wat to design, validate and compare two in vitro ECP protocols.

Three in vitro ECP protocols were compared. Protocol A; leukocytes were isolated and exposed to 200ng/ml 8MOP. Protocol B & C; PBMCs were collected and exposed to 200 & 400ng/ml 8MOP respectively. In all three protocols, irradiation intensity was 2 J/cm2 UV-A. The extend of cell death was validated by analysis of apoptosis and necrosis using Annexin-V and 7AAD every day until 72-hours post-ECP. In addition, CD3 and CD14 were used to analyze T lymphocytes and monocytes respectively.

Virtually no CD14+ monocytes survive 72-hours post-ECP in protocols A&B, whereas 20-30% of the remaining CD3+ T-lymphocytes are viable after this time period. Moreover, in protocol A, CD3+ Tlymphocytes preferentially died through necrosis (70% necrotic and 5% apoptotic at T=72, n=4) whereas in B through apoptosis (70% apoptotic and 0% necrotic at T=72, n=4). Overall viability in protocol A at T72 was 42%, to 18% in B and 7% in C.

Our in vitro ECP models generate pheresates with extensive cell death. Moreover, the three protocols yield significantly different results in terms of type and extend of cell death which may be attributed to different cell products (i.e. leukocytes or PBMCs) and isolation method. These differences are of importance when designing future in vitro experiments aiming to elucidate the mechanisms of action.

POSTERS

Pèptids immunogènics del gluten en femta: avaluació de la seva utilitat en la detecció de transgressions després d'un any de la seva internalització

Clara Esteve Cols 1; Trinidad Alba Cano 1; Carme Ramírez Pineda 1; Ariadna Delgado Gordo 1; Sílvia Miró Cañís 1

1 CLILAB Diagnòstics

Introducció: La Malaltia Celíaca (MC) és una malaltia crònica d'origen autoimmune que afecta la mucosa de l'intestí prim desencadenada pel consum de gluten en persones genèticament predisposades. El tractament és la retirada del gluten de la dieta. Pel seguiment de l'adherència, s'utilitzen qüestionaris nutricionals (però són subjectius) i els anticossos anti-transglutaminasa IgA (aTgA), amb poca sensibilitat (50%). Per això, s'han descrit nous marcadors com els pèptids immunogènics del gluten (GIP), que són els fragments del gluten resistents a la digestió gastrointestinal. La seva detecció en femta o orina indica el consum de gluten (voluntari o involuntari).

Objectius: Avaluar la sensibilitat de la presència de GIP en femta per detectar transgressions de la dieta respecte als aTgA.

Materials i mètodes: Anàlisi retrospectiu dels pacients del nostre hospital (Consorci intercomarcal de l'Alt Penedès i el Garraf, CSAPG) amb determinació de GIP en femta (N=49) des de la implantació d'aquesta determinació al laboratori (juliol del 2024). Per l'avaluació s'ha comparat el resultat del GIP amb els aTgA i s'han revisat les històries clíniques (diagnòstic, grau d'adherència i presència o no de simptomatologia intestinal).

Resultats: Dels 49 pacients, 39 (79.6%) van obtenir un resultat negatiu, i 10 (20.41%) un resultat positiu. D'aquests 10, se'n van excloure tres per no tenir diagnòstic de MC. Dels 7 inclosos, 4 (57.14%) tenien aTgA positius i no referien transgressions ni simptomatologia intestinal, i 3 (42.86%) tenien aTgA negatius. Dos d'aquests (66.67%), referien transgressions i simptomatologia intestinal.

Conclusions: Aquest estudi inicial postula que la determinació de GIP en femta sembla ser més útil que els aTgA per detectar transgressions a la dieta.

POSTERS

X-Linked Neutropenia Caused by Novel WASp G299R Variant: Functional and X-Inactivation Insights From a Family Study

Anna Calvet Lacruz 1; Eder Mateus Medina 2; Teresa Franco Leyva 1; Montserrat Torrent Espanyol 3; Josep Maria Valle T Figueres 3; Oscar de la Calle Martín 1; Laura Martínez Martínez 1

1 Hospital de la Santa Creu i Sant Pau, Servei d'Immunologia; 2 Sant Pau, Institut de Recerca; 3 Hospital de la Santa Creu i Sant Pau, Servei de Pediatria

X-linked Neutropenia (XLN) is a rare immune disorder caused by gain-of-function (GOF) mutations in the GTPase domain of the WAS gene, leading to constitutive activation of the WAS protein (WASp). This condition is associated with congenital neutropenia, recurrent infections, increased myeloid cell apoptosis, and lymphocyte anomalies. We present the case of an 11-year-old Caucasian boy with neutropenia since birth, recurrent mild infections, thrombocytopenia, atopic dermatitis, and small lymphadenopathies. Immunological studies revealed an inverted CD4/CD8 ratio, low NK cell percentage, reduced IgA levels, and a B-cell profile skewed toward immature transitional cells. Notably, IgA+IgD− switched B cells were diminished, compared to aged-matched healthy controls. Bone marrow disorders were excluded, prompting suspicion of XLN. Sanger sequencing of exon 9 of WAS identified a novel missense mutation (c.895G>A, p.G299R), located near a key residue for WASp activation (Tyr291). The patient's mother and grandmother, both asymptomatic carriers of the mutation, displayed neutropenia, thrombocytopenia with large platelets, and low memory B-cell counts. Functional studies revealed hyperactive neutrophils in our patient and his mother with increased viability, oxidative capacity, enhanced migration, and higher phagocytosis of Gram-negative bacteria. The mother also showed heightened phagocytosis of Gram-positive bacteria, whereas the patient demonstrated significantly reduced capacity compared to controls, correlating with his recurrent Streptococcus infections. Neutrophil hyperactivity persisted up to five days post-extraction. In agreement with murine models, neutrophils were scarce in peripheral blood but present in saliva. X-chromosome inactivation (XCI) analysis revealed skewed inactivation in the mother and random inactivation in the grandmother. This case identifies c.895G>A as a novel WAS GOF mutation and demonstrates its pathogenic role in XLN. It also highlights how skewed XCI in female carriers contributes to immune dysregulation. Our findings expand the understanding of WASp-associated disorders and underscore the importance of functional and genetic studies in diagnosing rare immunological conditions.

POSTERS

Antiphospholipid Autoantibodies: Agreement between ELISA, CIA, and PMAT in a Large Cohort of Patients with Suspected APS

Miriam Cárdenas Gómez 1; Andrea García López 1; Carmen Domènech 1; Lucía Marcelo 1; María Torradeflot 1; Ignasi Rodríguez-Pinto 2; Estíbaliz Ruiz-Ortiz 1; Sandra Forner Civera 1

1 Immunology Department, Centre Diagnòstic Biomèdic, Hospital Clínic de Barcelona, Spain; 2 Department of Autoimmune Diseases, Reference Centre for Systemic Autoimmune Diseases (UEC/CSUR) – Me

Introduction: Antiphospholipid syndrome (APS) is an autoimmune disorder characterized by thrombosis and/or pregnancy morbidity associated with antiphospholipid antibodies (aPL), including anticardiolipin (aCL) and anti-β2-glycoprotein I (aβ2GPI) IgG/IgM isotypes. Multiple platforms exist for aPL detection, potentially influencing interpretation.

Objective: To evaluate concordance of aPL antibody results using ELISA, chemiluminescence immunoassay (CIA), and particle-based multi-analyte technology (PMAT).

Patients & Methods: Serum from 791 patients with suspected APS was tested for aCL and aβ2GPI (IgG, IgM) using ELISA (QUANTA Lite®), CIA (QUANTA Flash®), and PMAT (Inova Diagnostics, USA). Cutoffs were applied (20 U/mL, 20 CU, 5 FLU). Results were classified as negative, low-positive, or moderate/high-positive. Positivity was graded: ELISA—negative <20, weak 20–40, strong ⩾40; CIA—negative <20, weak 20–90, strong ⩾90; PMAT—negative <20, weak 20–85, strong ⩾85. Concordance was assessed by Cohen's kappa (κ).

Results: Agreement varied by antibody and isotype. Discrepancies occurred mainly in low-positive samples; moderate/high positives showed stronger concordance.

Conclusion: Inter-assay agreement depends on antibody and isotype. Among the four aPL markers, aβ2GPI IgM showed the highest consistency. PMAT and CIA exhibited the strongest concordance, suggesting PMAT could substitute CIA in clinical practice. Most discrepancies occurred in weakly positive samples, while moderate/high positives showed strong agreement. Stratifying results by antibody level improves interpretation, and a thorough review of patients' clinical data is needed to better understand inter-assay discrepancies.

Antibody and Isotype	ELISA vs CIA (κ and level of agreement)	ELISA vs PMAT (κ and level of agreement)	PMAT vs CIA (κ and level of agreement)	3×3 Discrepancies
aCL IgG	0.37 (low)	0.63 (moderate)	0.52 (moderate)	ELISA–/CIA+: 126/147 weak ELISA+/CIA–: 5/7 weak
aCL IgM	0.59 (moderate)	0.50 (moderate)	0.79 (good)	ELISA–/CIA+: 32/37 weak ELISA+/CIA–: 23/31 weak
aβ2GPI IgG	0.51 (moderate)	0.66 (good)	0.77 (good)	ELISA–/PMAT+: 39/44 weak ELISA–/PMAT+: 39/44 weak
aβ2GPI IgM	0.64 (good)	0.67 (good)	0.63 (good)	ELISA–/PMAT+: 32/37 weak; PMAT+/CIA–: 46/51 weak

POSTERS

Thymic and Splenic Lymphocyte Profiles of Suckling rats depend on the Composition of their Maternal Diet

Ceballos-Sánchez Daniela 1,2; Casanova-Crespo Sergi 1,2; Rodríguez-Lagunas, M.J 1,2; Castell, M 1,2,3; Pérez-Cano, F.J 1,2; Massot-Cladera M 1,21

1 Department of Biochemistry and Physiology, Faculty of Pharmacy and Food Science, University of Barc; 2 Institut of Research in Nutrition and Food Safety (INSA-UB), Barcelona, Spain; 3 CIBER Physiopathology of Obesity and Nutrition (CIBEROBN), Institute of Health Carlos III

Introduction: Maternal Mediterranean diet, may shape lymphocyte development and support early-life immune health. This study evaluated the impact of a maternal high-fiber and polyphenol-enriched (HFP) diet on the immune profile of Wistar rat offspring by analyzing lymphocyte populations in the thymus and spleen at two postnatal time points.

Methods: Offspring from five maternal dietary groups differing in fiber and polyphenol exposure were analyzed at postnatal days 1 and 21. The HFP animals derived from mothers fed the enriched diet throughout pregestation, gestation, and suckling, whereas REF offspring came from control-fed dams. P, G, and S offspring corresponded to maternal enrichment limited to pregestation, gestation, or suckling, respectively. Thymic and splenic lymphocyte subsets were characterized by specific surface markers. Thymic populations included SP ($CD4^+$ or $CD8^+$), DP ($CD4^+CD8^+$), and DN ($CD4^-CD8^-$) cells, with DN1–DN4 subtypes defined by CD25/CD44 expression. Splenic subsets comprised B, NK, NKT, Th, Tc, and TCRγδ⁺ cells, which were further classified as CD8αα⁺/CD8αβ⁺ according to their marker profiles.

Results: The maternal diet influenced offspring immune profile both at birth and at the end of suckling. At d1, the P group showed increased DN and DN4 thymocyte proportions, while the HFP diet elevated DN4 cells in the HFP group. By d21, gestation and lactation groups (G, S) increased $CD4^+$ SP thymocyte proportion. In the spleen, the G group showed a reduced T/B ratio at d1, while at d21, the P, G, and S groups exhibited lower TCRαβ⁺ and T/B ratios accompanied by higher NK and B cell frequencies. The P group also showed a higher Th cell proportion.

Conclusion: Maternal diet composition critically shapes early immune development, with gestation and lactation identified as key programming windows. The timing of dietary exposure appears to be more decisive

POSTERS

Anti-GAD65 antibody-associated stiff-limb syndrome: A case report

Manuel Guillén Navarro 1; Eduardo Villegas Martín 2; Jorge Eguía Núñez 2; Ainhoa Iglesias López 1; Carmen Pérez Gomariz 1

1 Hospital del Mar de Barcelona; 2 Laboratori de Refèrencia de Catalunya

Background: Stiff-limb syndrome (SLS) is a focal variant of stiff-person spectrum disorders characterized by progressive rigidity and spasms, frequently associated with anti–glutamic acid decarboxylase 65 (GAD65) antibodies. Unlike classic stiff-person syndrome, SLS predominantly affects a single limb and may be misdiagnosed as orthopedic or spinal pathology. Early recognition is essential, as immunotherapy can improve function.

Case Report: A 49-year-old man presented with chronic rigidity and spasms of the left lower limb, predominantly affecting the leg, foot, and toe flexors. Although partial improvement had occurred, episodes of increased stiffness were triggered by exertion or external stimuli, limiting mobility. The patient relied on a cane for short distances but reported no sensory deficits or weight loss.

Neurological examination revealed increased tone in the left lower extremity with difficulty achieving full extension, flexed toe posture, preserved reflexes (ROTS 2/4; left patellar 3/4), and intact tactile and vibratory sensation. No upper motor neuron signs were present. Neurophysiological testing demonstrated continuous motor unit activation in agonist and antagonist muscles of the affected limb, consistent with SLS.

ELISA anti-GAD65 test was positive (81,5 U/mL; reference range: 0-4.9] and the presence of anti-GAD65 antibodies was confirmed by immunoblot and indirect immunofluorescence assays performed on cerebellar and pancreatic tissue substrate. The rest of the panel of paraneoplastic and neuronal antibodies was negative. Lumbar MRI showed multisegmental degenerative disc disease but no findings explaining the hypertonia.

Conclusions: This case supports the diagnosis of anti-GAD65–mediated SLS based on clinical presentation, EMG findings, and seropositivity. As up to 20% of cases may be seronegative, electrophysiology remains essential for diagnosis.

Monthly intravenous immunoglobulin for three months was proposed, with continuation every 4–6 weeks based on response. PET-CT was requested to exclude paraneoplastic etiology.

POSTERS

Anti-GAD65 Antibody-Associated Limbic Encephalitis in a 12-Year-Old: A Case Report

Manuel Guillén Navarro 1; Eduardo Villegas Martín 2; Jorge Eguía Núñez 2; Ainhoa Iglesias López 1; Carmen Pérez Gomariz 1

1 Hospital del Mar de Barcelona; 2 Laboratori de Refèrencia de Catalunya

Background: Autoimmune encephalitis associated with anti–glutamic acid decarboxylase 65 (GAD65) antibodies is an uncommon but important cause of limbic encephalitis, particularly in children. Anti-GAD65 antibodies target the enzyme responsible for GABA synthesis, leading to neuronal hyperexcitability and seizures. Diagnosis relies on clinical features, neuroimaging, and demonstration of antibodies in serum and cerebrospinal fluid (CSF). Early immunotherapy improves neurological outcomes.

Case Report: A 12-year-old boy with a history of mild autism spectrum disorder was admitted for seizures and suspected encephalitis. Initial episodes involved unresponsiveness, ocular and facial spasms, and limb jerks. EEG revealed right rolandic epileptiform discharges and later mild diffuse encephalopathy. MRI demonstrated a Chiari I malformation and, on second review, enlargement and hyperintensity of the left amygdalar complex, compatible with limbic involvement. CSF analysis showed normal glucose and protein levels and negative infectious PCRs. Anti-GAD65 antibody testing revealed:

- CSF immunoblot: strong positive.
- Serum immunoblot: weak positive.
- Indirect immunofluorescence performed on cerebellar and pancreatic tissue substrates showed high-titer positivity (>1:100) in serum and positivity in cerebrospinal fluid.
- ELISA: positive for anti-GAD65 (>100 U/mL; reference range: 0-4.9).

These findings confirm intrathecal antibody synthesis, supporting autoimmune limbic encephalitis rather than passive serum diffusion. Other onconeural and paraneoplastic antibodies were negative. The patient was clinically stable, afebrile, and cognitively normal at readmission. He was treated with methylprednisolone 1 g/day IV for 3 days and levetiracetam 500 mg every 12 h, with good tolerance and no further seizures. Endocrine evaluation was initiated due to the association of anti-GAD65 antibodies with diabetes and thyroid autoimmunity.

Conclusions: This case highlights pediatric anti-GAD65 limbic encephalitis confirmed by strong CSF antibody positivity and compatible imaging. Early immunotherapy led to clinical stabilization. Early identification of anti-GAD65 antibodies and prompt initiation of immunotherapy can lead to favorable outcomes, emphasizing the need for awareness of autoimmune causes in pediatric encephalitis.

POSTERS

Different strategies to prevent Celiac Disease (CD) antibody impairment: effect of cocoa and the gluten-degrading enzyme Celiacase in a mouse CD model

Marina Girbal González 1,2; Arturo Rodríguez-Banqueri 3; Ulrich Eckhard 3; F. Xavier Gomis-Rüth 3; Maria José Rodríguez-Lagunas 1,2; Angels Franch-Masferrer 1,2; Francisco J. Pérez-Cano 1,2

1 Section of Physiology, Department of Biochemistry and Physiology, Faculty of Pharmacy and Food Scien; 2 Research Institute of Nutrition and Food Safety (INSA-UB), University of Barcelona, Av. Prat de la R; 3 Proteolysis Laboratory, Department of Structural Biology, Molecular Biology Institute of Barcelona

Celiac disease (CD) is characterized by an abnormal immune response to dietary gluten, leading to the production of specific autoantibodies (Abs) and intestinal inflammation. Nutritional and enzymatic approaches have been proposed to reduce gluten-induced immune activation. This study aimed to evaluate the effects of cocoa or a gluten-degrading enzyme (Celiacase) on antibody responses in a mouse model of CD. DQ8-Dd-villin-IL-15tg mice (n = 14/group) were assigned to five groups: gluten-free diet (REF), gluten diet with oral gliadin (GLI), or the same diet supplemented with either defatted cocoa (GLI+COCOA) or Celiacase at two enzyme:gliadin ratios (1:500 and 1:100). Treatments lasted 25 days, after which plasma and intestinal levels of immunoglobulin (Ig) isotypes (IgG1, IgG2a, IgG2b, IgG3, IgA, IgE, IgM, total IgG) and CD-specific autoantibodies were quantified by ELISA or Luminex.

A gluten-containing diet significantly increased intestinal total IgG, IgG1, IgG2b, IgE, IgM, and IgA compared with REF, confirming mucosal immune activation. Cocoa supplementation partially mitigated this effect, particularly in males, indicating an immunoregulatory role. Celiacase treatment prevented the rise in intestinal IgA, IgE, IgM, and total IgG levels, also predominantly in males. Regarding autoantibodies, cocoa administration effectively reduced both intestinal and plasma anti-gliadin Abs but did not affect antitransglutaminase Abs. In contrast, Celiacase prevented the generation of both anti-gliadin and antitransglutaminase Abs.

Overall, both cocoa and Celiacase showed immunomodulatory potential against gluten-induced immune activation in this CD model. These findings support further investigation into their mechanisms and potential application as complementary strategies to prevent or attenuate immune responses associated with CD.

POSTERS

Characterization of Cyclin D3 as a novel regulator of beta cell fitness targeted by autoimmune diabetes

Joan Florensa 1,3; Marta Torres 1,3; Celia Vived 3; Júlia Luna 3; Francisco Ciruela 4,5; Joan Verdaguer 1,3; Núria Pujol-Carrion 1,3; Maria Angeles de la Torre-Ruiz 1,3; Anaïs Panosa 1,3; Baldo Oliva 2; José CE. Serrano 1,3; Conchi Mora 1,3

1 Universitat de Lleida; 2 Universitat Pompeu i Fabra; 3 IRBLleida; 4 Universitat de Barcelona; 5 IDIBELL Barcelona

In Type 1 diabetes (T1D), the inflammatory niche around the islets has been reported to impair pancreatic beta cell (pBC) function and induce pBC apoptosis. We have previously reported that Cyclin D3 is downregulated in pBCs during the progression of autoimmune attack in the NOD (Non-Obese Diabetic) murine model of T1D. Furthermore, we have demonstrated that the abrogation of Cyclin D3 expression in pBCs impairs pBC viability and function without affecting proliferation. These observations revealed a novel cell-cycle independent role for Cyclin D3 in beta cell homeostasis. Our goal is to unveil these novel, cell-cycle independent signaling pathways triggered by Cyclin D3 responsible for pBC viability and fitness, since they become new potential targets for therapeutic intervention in T1D. To that end we performed the yeast two-hybrid (Y2H) analysis to identify proteins that physically interact with Cyclin D3. The first Cyclin D3 partner identified (Candidate 1 or C1) to bind cyclin D3 has been reported to be involved in protein processing in the Endoplasmic Reticulum (ER). We further aimed to confirm this interaction in a NOD insulinoma (NIT-1) cell line, by either co-immunoprecipitation and/or the Lumit technology assays. We confirmed the interaction Cyclin D3-C1 by both approaches. Furthermore, by the biocomputational platforms Ifrags and iLoops, we identified point amino acid residue positions in Cyclin D3 that are potentially essential for its interaction with C1 and tested their relevance in vitro by Lumit technology. At present we are validating the functional implications of the Cyclin D3-C1 interaction on pBC survival and fitness upon exposure to several ER stress stimuli. Altogether, these data suggest that Cyclin D3 could be acting as a promoter of pBC proteostasis to alleviate the ER-protein synthesis overload, since secretory cells such as pBCs are submitted physiologically to ER-Stress, for instance, by high demand of insulin production.

POSTERS

Differential modulation of CD38⁺ immune subsets after daratumumab in refractory antibody-mediated renal allograft rejection

Eduardo Gozálvez 1; Carolt Arana 3,4; Nuria Expósito 1; Ana Belén Larque 4,5; Jose Luis Caro 1,2,4; Fritz Diekmann 3,4; Eduard Palou 1,2,4; Juan Manuel Torres 1,2,4

1 Servei d'Immunologia, Centre de Diagnòstic Biomèdic, Barcelona; 2 Secció d'Immunologia del trasplantament, Hospital Clínic de Barcelona; 3 Servei de Nefrologia, Institut Clínic de Nefrologia i Urologia, Barcelona; 4 Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS); 5 Servei d'Anatomia Patològica, Centre de Diagnòstic Biomèdic, Barcelona

Background: Antibody-mediated rejection (ABMR) with persistent microvascular inflammation (MVI) remains a major cause of late renal allograft failure and lacks standardized therapy. Anti-CD38 monoclonal antibodies such as daratumumab, primarily targeting plasma cells, may also modulate other CD38⁺ immune populations implicated in ABMR pathogenesis. This study aimed to characterize the temporal dynamics of lymphocyte subsets after daratumumab in refractory ABMR.

Methods: Renal transplant recipients with biopsy-proven refractory ABMR and persistent MVI treated with two doses of daratumumab were included. Immunophenotyping by flow cytometry was performed before treatment, and at 15 days, 1 month, and 3 months post-treatment, compared with healthy controls. Donor-derived cellfree DNA (dd-cfDNA) and anti-HLA antibodies were monitored, and follow-up biopsies were assessed for MVI activity.

Results: Seven patients were analyzed. A progressive depletion of CD38⁺ subsets was observed following daratumumab. CD38⁺CD19⁺ B cells declined markedly from baseline to 3 months (−46.3%), with values significantly lower than controls at 1 and 3 months ($p = 0.041$ and $p = 0.028$, respectively). CD38⁺ NK cells also decreased steadily (−56.7% at 3 months; $p = 0.037$ vs controls; $p = 0.083$ vs pre). Trends toward reduction were seen in CD38⁺CD3⁺ T cells (−40%, $p \approx 0.09$) and total NK percentage (−25%, $p \approx 0.11$). In contrast, activated CD4⁺ effector-memory and antibody-secreting populations increased transiently after depletion, suggesting compensatory immune activation. Notably, dd-cfDNA levels declined significantly at 1 month post-treatment, and MVI scores in follow-up biopsies showed improvement, indicating reduced allograft inflammation and injury.

Conclusions: Daratumumab therapy in refractory ABMR induces significant and sustained depletion of CD38⁺ B and NK cell compartments, along with dynamic shifts in T- and B-cell activation states. The concomitant decline in dd-cfDNA and MVI scores supports an overall reduction in immunologic activity. These findings highlight potential cellular and molecular biomarkers for monitoring anti-CD38 therapy and optimizing patient management in ABMR.

POSTERS

Combined Utility of GDF-15 and KL-6 as Biomarkers for Interstitial Lung Disease in Systemic Autoimmune Diseases

Melisa Toro Ayuso 1; David Roselló Martínez 1; Diego Castillo 1; Iván Castellví 1; Esther Ortiz 1; Elisabeth Moltó 1; Yolanda Álvaro 1; Laura Martínez Martínez 1; Anaís Mariscal Rodríguez 1

1 Hospital de la Santa Creu i Sant Pau

Some systemic autoimmune diseases (SAD) can present with interstitial lung disease (ILD), a disorder involving pulmonary fibrosis and inflammation. ILD can increase morbidity/mortality in SAD, so early detection and close monitoring is important. Krebs von den Lungen-6 (KL-6) has previously been described as a biomarker of detection and evolution of pulmonary fibrosis, however it is not a biomarker for the presence of SAD. To increase KL-6 usefulness as ILD biomarker we aimed to investigate the potential combined utility of KL-6 and GDF-15 as well as their individual diagnostic value. We analysed KL-6 and GDF-15 levels in a cohort of 170 patients categorized as: SAD (affected or not with ILD), or ILD (without SAD). We observed that KL-6 differentiates between patients with and without ILD, independently from SAD. Nevertheless, there were differences in GDF-15 levels between patients with ILD and SAD and those with ILD without SAD. Additionally, both biomarkers, combined or independently, correlate with a higher erythrocyte sedimentation rate (ESR), so indicating a major systemic inflammation. In terms of respiratory damage, KL-6 and GDF-15 are indicators of ILD, since the levels of diffusing capacity of the lung for Carbon Monoxide % (DLCO%) showed a gain of function only in the group that had low levels of both biomarkers. These findings support that both KL-6 and GDF-15 are complementary biomarkers to evaluate the pulmonary involvement in patients suffering from SAD.

POSTERS

Development of a Composite Immunological Score to Improve the Laboratory Diagnosis of Antisynthetase Syndrome

Juan Francisco Luchoro-Cerdà 1; María Torradeflot 1; Carmen Domènech 1; Lucía Marcelo 1; Mario Framil-Seoane 1; Andrea García-López 1; Sergio Prieto-González 2; Estíbaliz Ruiz-Ortiz 1

1 Immunology Department, Centre Diagnostic Biomèdic CDB, Hospital Clínic de Barcelona, Barcelona, Spai; 2 Department of Autoimmune Diseases, Vasculitis Research unit, Hospital Clínic

Introduction: Antisynthetase syndrome (ASS) is a rare autoimmune disease within the spectrum of idiopathic inflammatory myopathies (IIM), characterized by the presence of autoantibodies directed against aminoacyl-tRNA synthetases. Laboratory diagnosis is challenging due to false positives results in standard techniques (e.g., immunoblot), motivating an integrated approach that improves interpretability and diagnostic accuracy.

Patients & Methods: We developed a composite immunological score integrating IIM line immunoblot (Euroimmun®), indirect immunofluorescence (IIF) on HEp-2 cells (Werfen®), and chemiluminescent immunoassays (CIA) for Jo1 and Ro52 (Werfen®). A training cohort of 151 patients (19 with confirmed ASS) was analyzed. Associations between each assay and diagnosis were assessed using Fisher's exact test. Based on discriminative performance, weighted values were assigned to each test to compute an "immunological probability" score.

Results: Significant contributors were: anti-synthetase antibody by immunoblot ($p<0.001$), anti-Jo1 by CIA ($p<0.001$), compatible HEp-2 IIF pattern (AC-19 or AC-20, $p<0.001$), absence of polyreactivity ($p<0.05$) and anti-Ro52 antibodies copositivity ($p<0.05$). The parameter set that maximized diagnostic performance produced an AUC of 0.988 (95% CI 0.975–1) in a ROC curve analysis. Assigned values were: antisynthetase antibody by immunoblot (+++: 5, ++: 3, +: 1, Negative: 0); Jo1 by CIA conditional on immunoblot positivity (Positive: 3, Negative: −3 when Jo1 is positive by blot); compatible IIF pattern (Positive: 3, Negative: 0); Anti-Ro52 copositivity by immunoblot (+++: 0.75, ++: 0.5, +: 0.25, Negative: 0); and by CIA (Positive: 0.75, Negative: 0). The optimal cutoff yielding maximal sensitivity and specificity (1.00 and 0.909, respectively) was 3.5.

Conclusions: The score shows excellent discrimination in the training cohort and will be validated in a larger independent cohort. If confirmed, it could reduce diagnostic uncertainty, improve reporting results to the clinician in order to enable optimal interpretation of immunological data, and optimize management of patients with suspected ASS.

POSTERS

Autoinflammatory Disorders: A Case Report Highlighting IRF3 Transactivation and Autoinhibition

Àngels Santaolalla 1; Laura Martínez-Martínez 1; Anna Calvet Lacruz 1; Andrea Bravo Gómez 1; Anaís Mariscal 1; Melisa Toro 1; Luis Querol 1; Teresa Franco-Leyva 1

1 Hospital de la Santa Creu i Sant Pau

Autoinflammatory diseases arise from dysregulated innate immune pathways and often present with heterogeneous and overlapping clinical features, complicating diagnosis and treatment. Recognition of many such disorders as inborn errors of immunity has facilitated understanding of their molecular bases.

This study reports an atypical early-onset autoinflammatory syndrome resembling Schnitzler Syndrome, but with unusual clinical and laboratory features, including early-onset, IgG-kappa monoclonal gammopathy, and progression to multiple myeloma. Genetic analysis identified a variant of uncertain significance in IRF3, a key innate immune regulator, which was also found in an unaffected relative, suggesting complex inheritance or additional genetic factors.

Protein expression studies revealed abnormal IRF3 expression patterns in the patient, indicating functional consequences of genetic variation. These findings highlight the complexity of diagnosing autoinflammatory disorders and underscore the potential role of IRF3 dysregulation in disease pathogenesis. The results underline the need for integrated immunogenetic and proteomic approaches to clarify disease mechanisms, ultimately improving diagnostic precision and targeted therapies in autoinflammatory disorders.

POSTERS

G-protein coupled receptor (GPR)55 deficiency protects against lung damage by regulating neutrophil activation in an LPS-induced acute lung injury mouse model

P. Goncalves Romeu 1; G. Li 2; G. Shakir 2; M. Matute 3; T. Marín 3; M. Galdeano 3; I. Aldàs 3; A. Herraiz 3; J. M. Hernández 1; L. Fluvià 1; S. Steffens 2; R. Guillamat-Prats 1

1 Germans Trias i Pujol Research Institute (IGTP); 2 Institute for Cardiovascular Prevention (IPEK), Ludwig-Maximilians-Universität (LMU); 3 Respiratory Department, Hospital Universitari Germans Trias i Pujol (HUGTiP)

Severe respiratory infections can progress to acute respiratory distress syndrome (ARDS), which accounts for ~10% of ICU admissions. Alveolar neutrophil infiltration is a hallmark of ARDS, their excessive activation and release of proinflammatory cytokines impair their reparative function, worsening the lung damage.

G-protein coupled receptor 55 (GPR55) and its endogenous ligand, lysophosphatidylinositol (LPI), have been implicated in the regulation of immune cell function across different pathological processes. ARDS patients exhibit reduced GPR55 expression in circulating neutrophils and lower plasma LPI concentrations, suggesting a role for GPR55/LPI axis in ARDS pathogenesis. Ex vivo pharmacological blockade of GPR55 in blood neutrophils from ARDS patients enhanced their phagocytic activity and reactive oxygen species (ROS) production, highlighting its regulatory role.

To further investigate the impact of GPR55 deficiency on neutrophil activity and lung injury, we used an in vivo mouse model of acute lung injury induced by intratracheal instillation of lipopolysaccharide (LPS; 2 mg/mL in 75 µL) in wild-type (WT) and GPR55 knockout (KO) mice. GPR55 deficiency increased neutrophil mobilisation and infiltration into the bronchoalveolar space. GPR55-KO mice exhibited reduced lung inflammation (IL1β, IL6, TNFα), although GPR55-deficient neutrophils showed enhanced ROS production, activation (Ly6C expression) and degranulation (MPO, NE) after LPS stimulation.

Transcriptomic profiling of WT and GPR55-KO circulating neutrophils revealed the role of GPR55 in cytokine production, innate immune response, and antiviral response. In vitro, we confirm that GPR55-deficient neutrophils showed attenuated proinflammatory cytokine expression following LPS stimulation.

In conclusion, GPR55 deficiency dampens proinflammatory cytokine expression and lung inflammation while preserving neutrophil defence mechanisms in mice. Targeting GPR55 signaling may represent a promising therapeutic strategy to modulate neutrophil activation and mobilisation, providing a potential therapeutic target for ARDS.

POSTERS

Probiotic Supplementation Mitigates Antibiotic-Induced Changes in Systemic and Mucosal Immunoglobulin Profiles in a Rat Model of Antibiotic-Associated Diarrhea

Rodríguez-Martínez, Nuria 1,2; Grases-Pintó, Blanca 1,2; Kapravelou, Garyfallia 1,2; Massot-Cladera, Malén 1,2; Castell, Margarida 1,2; González-Hedström, Daniel 3; Martínez-Ríos, Verónica 3; Martorell, Patricia 3; Pérez-Cano, Francisco J. 1,2; Rodríguez-Lagunas, Maria José 1,2

1 Departament de Bioquímica i Fisiologia, Facultat de Farmàcia i Ciències de l'Alimentació, University; 2Institut de Recerca en Nutrició i Seguretat Alimentària (INSA), C/ Prat de la Riba 171, Santa Coloma; 3ADM Biopolis, Parc Científic Universitat de València, Catedrático Agustín Escardino Benlloch, 9.

Antibiotic treatment, as a side effect, can disrupt mucosal immunity and gut microbiota, leading to Antibiotic-Associated Diarrhea (AAD). This study aimed to evaluate whether probiotic administration could prevent or mitigate immune alterations in an AAD model. For that purpose, eight-week-old Wistar rats were divided into five groups: reference (R), Amoxicillin + clavulanic acid (A), A plus probiotic (AP), antibiotic cocktail consisting of ampicillin + streptomycin + clindamycin (D), and D plus probiotic (DP). During the first 7 days, groups A, AP, D and DP received the suitable antibiotic once a day by oral gavage to induce diarrhea, while R received water. During the whole period (14 days), AP and DP received the probiotic while R, A and D groups received maltodextrin by oral gavage once a day. At the end of the study, immunoglobulins (Ig) concentration was determined in the plasma or in the intestinal content of the ileum by ProcartaPlex immunoassay and ELISA, respectively. Plasma analysis showed decreased IgA levels in A and D groups, which were not restored by probiotic treatment. IgM levels showed a tendency to decrease in AP group compared to R group, while total IgG levels remained unchanged. Regarding IgG subclasses, antibiotic treatment was able to reduce IgG1 levels in A group, while D group showed a decrease in IgG2a levels. The probiotic supplementation could not revert the decrease observed in IgG1 levels, but restored IgG2a levels in the DP group. At mucosal level, antibiotic treatment did not affect IgA concentration. In counterpart, male rats from A and D groups showed lower IgM levels, a trend partially reversed in DP animals. Overall, antibiotic administration altered systemic immunoglobulin while maintaining mucosal Ig concentration, whereas probiotic treatment showed limited capacity to reverse these effects. The pattern of immunoglobulin modulation suggests a general dampening of humoral immune responses that could be attributable to the dysbiosis caused by the antibiotics.

POSTERS

Innate Lymphoid Cells Bridge Innate and Adaptive Immunity via Antigen Presentation in Type 1 Diabetes

Ariadna Roig 1; Sebastián Pérez 1; David Perna-Barrull 2; Eva Aguilera 3; Marta Vives-Pi 2; Raquel Planas 1

1 Departament de Biologia Cel.lular, Fisiologia i Immunologia, Universitat de Barcelona; 2 Servei d'Immunologia, Institut de Recerca Germans Trias i Pujol, Badalona; 3 Servei d'Endocrinologia, Hospital Germans Trias i Pujol, Badalona

Innate lymphoid cells (ILCs) are emerging as key modulators of immune responses, but their role in human autoimmune diseases remains unclear. Recent evidence suggests that ILCs may acquire antigen-presenting capabilities under inflammatory conditions. Type 1 diabetes (T1D) is characterized by autoimmune destruction of pancreatic β-cells, involving both innate and adaptive immunity. We investigated whether circulating ILCs from T1D patients exhibit features of antigen-presenting cells (APCs) and contribute to autoreactive T cell activation.

We used spectral cytometry to phenotype ILCs and performed cell sorting for in vitro stimulation with proinflammatory cytokines and glycolysis inhibitors. Cell-sorted, cytokine-stimulated ILCs were loaded with Cytomegalovirus/Epstein Barr/Influenza (CEF) or insulin peptides and co-cultured with autologous memory $CD4^+$ T cells from healthy donors or patients with T1D. T cell activation was assessed by expression of activation-induced markers.

Circulating total ILCs, ILC1, and ILC3 from recent-onset T1D patients (n=11) overexpress HLA-DR compared to controls (n=12). Patient-derived ILCs (n=4) also show increased CD80 expression upon mitogen stimulation. Healthy donor-derived purified ILCs stimulated with IL-1β (n=3) upregulate HLA-DR and CD80, an effect partially reduced by the glycolysis inhibitor 2-deoxyglucose. Co-cultures of cytokine-stimulated ILCs and autologous memory $CD4^+$ T cells, with CEF or insulin B9-23 peptides, demonstrate that ILCs can present antigens and induce peptide-specific $CD4^+$ T cell responses—to pathogens in healthy donors (n=4) and to insulin in T1D patients (n=3).

In T1D, circulating ILCs show HLA class II overexpression and higher predisposition to upregulate costimulatory molecules by mitogens. Proinflammatory cytokines enhance class II and costimulatory molecule expression, partly via glycolysis in ILC3. Under inflammatory conditions, human ILCs may contribute to antigen presentation and activation of autologous memory $CD4^+$ T cells. Their potential role as APCs in contributing to autoimmunity warrants further investigation.

POSTERS

Exploring the impact of the fibrotic microenvironment on lung tumors using a 3D multicellular spheroid model

Marina Tuxans Serrano 1; Isabel Alfaya Fiaño 1; Paloma Millan Billi 2; Maria Saigi 3; Karina Portillo Carroz 2; Llorenç Coll Mulet 4; Miguel Angel Peinado 4; Raquel Guillamat Prats 1

1 Germans Trias i Pujol Research Institute (IGTP); 2 Germans Trias i Pujol Hospital; 3 Institut catala d'oncologia (ICO); 4 Aniling

Lung cancer is the leading cause of cancer-related deaths globally. The risk of developing LC increases more than 7-fold in patients with chronic respiratory diseases, such as pulmonary fibrosis, leading to a worse prognosis and making treatment more complicated.

Our hypothesis is that the fibrotic tumor microenvironment and cell-cell interaction under this environment modulate tumor cell growth and modify their transcriptome.

We recreated tumor growth in vitro by culturing spheroids with non-small cell lung cancer cell lines (H23-adenocarcinoma, EBC1-squamous) alone or together with human lung fibroblasts (HLF) and macrophages (HM) under fibrotic (TGF-β, PDGF, FGF and IL-4) and non-fibrotic conditions. We measured cell proliferation by flow cytometry, gene expression by bulk-RNA-sequencing and qPCR, and measured cell-cell interactions using microscopy.

We observed reduced proliferation of tumor cells from mono- and multicellular spheroids under fibrotic conditions in both LC lines. HLF in contact with EBC1 and HM in contact with H23 reversed these changes in cell proliferation. Multivariable RNA sequencing analysis of the two lung cancer spheroid models under fibrotic conditions revealed upregulated immune modulatory pathways, including complement activation and humoral responses, alongside downregulation of hypoxia, proliferation and mitochondrial activity. Fibrotic conditions also increased the expression of key immune checkpoints, CD73 and PD-L1, promoting immunosuppression. In H23, additional immunoregulatory genes—Galectin-9, PD-L2, and CD276—were differentially regulated under fibrotic conditions when tumoral cells where in contact with HLF and HM vs alone, whereas EBC1 showed a synergistic downregulation of CD40 and CD47, CD112, CD155, suggesting enhanced immune evasion under fibrotic conditions.

HM from multicellular spheroids under fibrotic conditions displayed higher expression of genes linked to anti-inflammatory and reparative phenotype and immunosuppressive response; as well as increased phagocytic activity.

Our data highlight the impact of the fibrotic microenvironment on tumoral cell functionality and its response to potential immunotherapies.

POSTERS

Diagnostic Performance of the Kappa Index Versus Oligoclonal Bands in Multiple Sclerosis and Clinically Isolated Syndrome

Diego Ruiz Nieto 1; Joan Climent Martí 1; Àlex Soriano Martínez 1; Eva María Martínez Cáceres 1

1 Hospital Universitari Germans Trias i Pujol

The diagnosis of multiple sclerosis (MS) relies on clinical, radiological, and laboratory criteria. Oligoclonal bands (OCBs) in cerebrospinal fluid remain a key diagnostic tool despite the inherent subjectivity in their interpretation. In the 2024 revision of the McDonald criteria, the kappa index was introduced as an alternative biomarker.

In this study, we compared the diagnostic performance of OCBs and the kappa index in 40 patients: 21 with clinically isolated syndrome (CIS) and 19 without demyelinating events. Sensitivity, specificity, positive and negative predictive values (NPV), and concordance between both methods were calculated.

The kappa index showed a sensitivity of 90% and a specificity of 63.6%, compared to OCBs, which exhibited 70% sensitivity and 90% specificity. All OCB-positive samples were also kappa-positive, whereas 68.4% of kappa-positive cases were OCB-positive. Among patients with CIS (n=21), ten were ultimately diagnosed with MS.

N=21	BOC +	BOC -	Kappa +	Kappa -
MS (N=10)	7	3	9	1
No MS (N=11)	1	10	4	7

Two sequential diagnostic algorithms were assessed:
a) Initial screening with OCBs, followed by the kappa index in negative cases.
b) Initial screening with the kappa index, followed by OCBs in negative cases.

Both algorithms correctly identified 9 of the 10 MS cases. Given its high sensitivity, negative predictive value (87.5%), and lower interpretative bias, the kappa index appears to be the most suitable screening tool, eliminating the need for OCB testing in kappa-negative cases. However, due to its lower specificity and higher false-positive rate, laboratories capable of performing both tests are advised to complement the kappa index with OCB analysis in cases where differentiation from other demyelinating disorders may aid diagnostic accuracy.

POSTERS

Peripheral Immune Signatures Define c-Kit⁺ T and CD11c⁺ B Cell Alterations in Multiple Sclerosis

Joan Puñet-Ortiz 1; Federico Fondelli 2,3; Marco A Fernández 1; Hammad Alzayat 1; Silvia Presas 4; Cristina Ramo-Tello 4; Eva M Martínez-Cáceres 2,3; Aina Teniente-Serra 2,3

1 Mixed Cytometry Unit IGTP-IJC; 2 Immunology Department, Germans Trias Hospital and Research Institut; 3 Department of Cell Biology, Physiology, Immunology, Universitat Autònoma Barcelona; 4 Neurology Department, Germans Trias Hospital and Research Institute

Background: Multiple Sclerosis is an autoimmune disease in which immune responses target central nervous system, leading to neuroinflammation, demyelination, and progressive neurological disability. The disease typically manifests as relapsing–remitting MS (RRMS) or as primary progressive MS (PPMS). Although several therapies are available for RRMS, effective treatments for PPMS remain scarce. A precise characterization of the immune subsets driving RRMS and PPMS is crucial for the development of more selective, mechanism-based therapies. To address this, we used high-dimensional spectral flow cytometry to generate a comprehensive peripheral immune atlas of RRMS, PPMS, and healthy donors (HC).

Objective: To identify peripheral immune fingerprints associated with disease.

Methods: We analyzed peripheral blood PPMS (n=15), RRMS (n=17) and healthy controls (n=21) using a 37-color spectral cytometry panel. Unsupervised algorithms were applied to analyse T and B-cell subpopulations, and cluster profiles were compared.

Results: Regarding the T cell compartment, both RRMS and PPMS showed similar alterations, characterized by a decreased frequency of clusters identified as memory and effector cytotoxic T cells, as well as proinflammatory NKT cells and $CD4^{-}CD8^{-}$ T cells, compared with HC. In contrast, both RRMS and PPMS groups showed an increased frequency of CD4 T cells with a Th17-like phenotype and c-Kit expression relative to HC. Within the B cell compartment, we observed an increase of transitional B cells in RRMS patients compared with HC, whereas PPMS patients displayed a higher proportion of switched memory B cells compared with HC. Moreover, only PPMS patients, but not those with RRMS, exhibited an expansion of clusters of CD11c-expressing B cells.

Conclusions: c-Kit-expressing CD4 T cells and CD11c-expressing B cells are rare immune subpopulations that have been poorly characterized in multiple sclerosis. Here, we report for the first time that both populations display differential abundance in the peripheral blood of RRMS, PPMS and HC.

POSTERS

Maternal nutrition with fiber and polyphenols influences rat lymphoid tissues in adult offspring

Sergi Casanova Crespo 1,2; Daniela Ceballos Sánchez 1,2; Malen Massot Cladera 1,2; Maria José Rodríguez Lagunas 1,2 ; Margarida Castell 1,2,3; Francisco José Pérez Cano 1,2

1 Department of Biochemistry and Physiology, Faculty of Pharmacy and Food Science, University of Barce; 2 Institut of Research in Nutrition and Food Safety (INSA-UB), Barcelona, Spain; 3 CIBER Physiopathology of Obesity and Nutrition (CIBEROBN), Institute of Health Carlos III, 28029

Maternal diet is an element of the exposome that can shape immune development in offspring. This preclinical study aimed to determine whether maternal consumption of a fiber- and polyphenol-enriched (FP) diet in a period beginning 3 weeks before gestation and lasting until the end of lactation influences the lymphoid composition of spleen, mesenteric lymph nodes (MLNs), and thymus in the adult offspring upon antigenic challenge. For this purpose, two groups were constituted by the Wistar rat offspring whose mothers received either a reference (REF) diet or a FP diet. After weaning, offspring from both groups were maintained on a reference diet. At 7 weeks of age, animals were immunized with ovalbumin (OVA) and after a 4-week period the proportion of the main lymphocyte subsets from thymus, spleen and MLNs was established by flow cytometry.

In both spleen and MLNs, the proportions of T lymphocyte populations such as TCRαβ+, TCRγδ+, NKT, and also that of NK cells were not significantly altered. However, FP OVA offspring showed a higher proportion of CD8+ and lower proportion of CD4+ cells within TCRαβ+ and NKT subsets, resulting in a reduced CD4/CD8 ratio compared to REF OVA group. In MLNs, OVA immunization increased the proportion of CD103+ cells in B, CD4+, and CD8+ cells of OVA animals from REF mothers, whereas this effect was prevented in OVA animals from FP mothers. In the thymus, no major differences in maturation stages were detected, although FP OVA offspring displayed reduced proportion of DN1 cells and an increased that of DN4 cells within double-negative (CD4-CD8-) thymocytes.

In conclusion, maternal FP diet durably modulated offspring immunity by reprogramming lymphoid composition in both central and peripheral compartments. These results highlight the immunomodulatory role of maternal nutrition, capable of shaping adaptive immune responses in offspring during adulthood.

To all Sponsors, Speakers, Chairs and Curious Listeners, THANK YOU

for making this Congress possible!

Catalan Society of Immunology

2025

Best regards from the Organizing Committee of the Congress

Programme Brochure designed by Rebeca Gutiérrez Cozar

SCi
SOCIETAT CATALANA D'IMMUNOLOGIA

L'Acadèmia
FUNDACIÓ ACADÈMIA DE CIÈNCIES MÈDIQUES I DE LA SALUT DE CATALUNYA I DE BALEARS