PD1+), Tregs (CD3+ CD4+ CD45RA-Foxp3+), pre-Tfh (CD3+ CD4+ CD45RA-CXCR5^{low} PD1^{int}) and Tfh (CD3+ CD4+ CD45RA-CXCR5^{hi} PD1^{hi}) by multiparametric flow cytometry. Also, the expression of CXCR3 (related to a Th1 profile) and the cytokines IL21, IFNγ and IL17 were analyzed. The frequencies of CD3+, CD4+, CD8+, Tregs and CD19+ cells were not altered in T21 tonsils, but a diminution in Ki67+ CXCR5+ CD19+ cells was observed ($p<0.05$). There is a 40% increase in the fraction of activated non-Tfh cells ($p<0.05$) whereas the ratio of Tfh decreases ($p<0.3$). There seems to be an arrest in the pre-Tfh population which shows an increase in their proportion ($p=0.08$). Interestingly, a higher percentage of CXCR3+ pre-Tfh population ($p<0.01$) is observed. Moreover, the mean fluorescent intensity of CXCR3 is enhanced in all the T cell populations analyzed. When cytokines were studied, an increase in the fraction of Tfh IFNγ+, IL21+ and IFNγ+IL21+ was observed. To explore if dendritic cells (DC) could be involved in promoting a Th1 bias among the non- and pre Tfh cells, we phenotypically analyzed the different DC population using the following markers (CD45, CD3, CD19, CD56, CD11c, HLADR, CD304, CD1c, CD141, CD86, FcεRI, PDL1). No significant changes in the frequencies of the different DCs populations were found. Our results suggest that T21 children have an altered tonsil T cell compartment, with a skewed Tfh differentiation and a Th1 profile among the pre-Tfh population. Further research should be done to understand the mechanisms involved.

285. (524) **HLA EPLET MISMATCH IDENTIFIES PEDIATRIC LIVER TRANSPLANT PATIENTS WITH HIGHER RISK OF DEVELOPING ACUTE REJECTION**

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The quantification of HLA eplet mismatch (eMM) has demonstrated its superiority as a biomarker for the immunological risk stratification in kidney, heart and lung transplantation. The aim of this study is to evaluate the association between HLA eMM and the development of biopsy-proven acute rejection (BPAR).
Patients that received liver transplant between 2018 and 2020 were included and prospectively followed for 9 months. Recipients and donor pairs were HLA typed by NGS and the number of eMM was quantified. Risk loci for BPAR were identified by Cox regression models. ROC analysis was performed to identify specific eMM thresholds for BPAR development and Kaplan-meier curves for the BPAR-free survival according to high or low eMM load were constructed.
Thirteen of our 37 patients included in the study developed BPAR during follow up. The number of antibody verified (ab) eMM in HLA-A (HR: 1.10, CI95% 1.00-1.20; p= 0.045) and HLA-DQ (HR: 1.21, CI95% 0.99-1.47; p= 0.063) were significantly associated with BPAR. Having ≥ 2 ab HLA-DQ eMM and ≥ 9 ab HLA-A eMM were considered as high loads. At 9 months post-transplant, the BPAR-free survival for patients with low and high ab HLA-DQ eMM load was 81.0% (CI95% 65.8-99.6) and 43.8% (CI95% 25.1-76.3), respectively, while for patients with low and high ab HLA-A eMM load was 75.0% (CI95% 60.6-92.9) and 33.3% (CI95% 13.2-84.0), respectively.
The measurement of the ab eMM load for the loci HLA-A and HLA-DQ between donor and recipient prior liver transplantation could identify those at risk of developing BPAR. Individualized immunosuppression protocols and closer surveillance could be performed if the immunological risk is assessed precisely. Further studies in our population are being performed to confirm these results.

286. **(528) DYNAMICS OF SOLUBLE IMMUNE MEDIATORS IN COVID-19 PATIENTS FROM AN ARGENTINEAN COHORT WITH MODERATE AND SEVERE SYMPTOMS**

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The cytokine storm, a form of systemic inflammatory response syndrome, is one of the most dreadful complications that can occur during COVID-19. In this work, we aimed at studying the occurrence of a cytokine storm in a cohort of COVID-19 patients (Cpts) from Córdoba (Argentine). During first wave, we collected sera from individuals with RT-PCR+ for SARS-CoV2 hospitalized in Hospital Privado with moderate (MOD) and severe (SEV) disease (n=62, aged 21-80 years) as well as healthy controls (HC n=24, age matched), to determine the concentrations of IL-1β, IL-6, IL-8, IL-10, IL-12, IL-28A/B, IL-29, TNF, IP-10, IFNα2, IFNβ, IFNγ and GM-CSF by LEGENDplexTM. Males represented 74% of MOD and 67% of SEV Cpts. Hypertension (HT, 48%), obesity (31%), dyslipidemia (DL, 24%), and diabetes (24%) were the most frequent comorbidities. All cytokines, except IL-28A/B, were significantly increased in total Cpts in comparison with HC ($p<0.01$). Elevated levels of IL-6 and CRP ($p<0.01$) between 4-7 day after hospitalization were found in all Cpts who died, but the cytokine profiles were different in deceased SEV than in MOD Cpts. Mortality in SEV group was associated with high levels of IL-6 ($p<0.001$), GM-CSF ($p<0.01$), IL-8, CRP, leukocytosis and decreased platelets ($p<0.0001$ for all). Comorbidities were linked to particular patterns of immune mediators on admission but not afterwards during infection, with HT Cpts exhibiting increased IP-10 levels but DL Cpts showing lower concentration of IL-1β, GM-CSF and IL-10 compared to no-HT and no-DL Cpts, respectively ($p<0.05$). The frequency of Cpts who required O2 support was higher in HT (84%) vs DL (67%). Although our data have similarities with those in international reports, the complete profiling of different parameters (cytokine/chemokines, risk factors, epidemiological and clinical characteristics) in the local cases add value by identifying particularities that may be relevant for the management and prognosis during SARS-CoV2 infection.

287. (533) **ALTERATIONS OF BLOOD IMMUNE CELLS IN COVID-19 ELDERLY PATIENTS WITH OR WITHOUT TYPE 2 DIABETES**

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Elderly individuals, especially those with pre-existing conditions like diabetes mellitus (DM), have a high risk for developing severe cases of COVID-19. The aim of this work was to characterize the alterations of blood immune cells (BIC) in patients with symptomatic COVID-19 and confirmed SARS-CoV-2 infection, ≥ 60 years and who needed hospitalization in the Centro de Salud Hospital of Tucuman during the second peak of the pandemic. Blood samples

were taken at the time of admission (d0) and five days after (d5) for routine laboratory tests and the characterization of BIC by flow cytometry. Most of the patients were men (70%) aged between 60 and 78 years. The 70% of patients had DM while 50% had arterial hypertension. At d0, all the patients had increased neutrophils and inflammatory markers (C reactive protein and D-dimers) and reduced numbers of lymphocytes, HLA-DR^{hi} monocytes, CD16^{+}CD56^{+} NK cells, CD3^{+}HLA-DR^{+}CD25^{+} cells, CD4^{+} and CD8^{+} T cells in blood. Patients received a standard treatment for COVID-19 care (O_2, corticosteroids and antibiotics). The treatment normalized the levels of BIC (d5) in 30% of patients who were those with no comorbidities. In patients with DM, BIC recovery was variable. In DM patients who required administration of plasma (30%), prolonged O_2 therapy (40%) or referral to the intensive care unit (10%) significant reductions of CD16^{+}CD56^{+}, CD3^{+}HLA-DR^{+}CD25^{+}, CD4^{+} and CD8^{+} cells were observed between d0 and d5. In line with previous studies, our results shows that absolute counts of major lymphocyte subsets in blood are significantly and substantially decreased during the course of severe COVID-19 disease in elderly patients. These BIC alterations may persist despite clinical care in elderly patients with DM. Further studies are needed to investigate the utility of early lymphocyte subset measurements as prognostic biomarkers of disease severity, mortality, and response to treatment in COVID-19 elderly patients with DM.

288. (543) **MOLECULAR DETECTION OF *CLOSTRIDIOIDES DIFFICILE* BY DIRECT PCR: NEW TOOLS FOR THE DIAGNOSIS OF *C. DIFFICILE* INFECTION**

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Clostridioides difficile infection (CDI) is the major cause of hospital-acquired diarrhea associated to antibiotics treatment in developed countries. CDI has become a health security threat and a considerable challenge to public health worldwide. The increased incidence and the severity of disease have been linked to the emergence and fast spread of hypervirulent epidemic strains. Moreover, a further increase in community-acquired infections and the zoonotic potential of *C. difficile* lead to a highly dynamic epidemiology of CDI. Enzyme immunoassay (EIA), a technique with high specificity but low sensitivity, is widely used as a diagnostic tool for *C. difficile* nowadays. To optimize the diagnosis and provide information for epidemiological surveillance strategies, the expression of glutamate dehydrogenase and toxin B (TcdB) of *C. difficile* was determined by EIA, direct PCR of stool samples and colony PCR of anaerobic culture. We also conducted comparative analysis to determine the performance of the direct PCR for *C. difficile*.

Faecal samples from 81 hospitalized individuals with diarrhea were collected. Clinical and demographic data were analyzed. We found a frequency of 18.5% for toxigenic strains. Treatment with antibiotics or proton-pump inhibitors were the main risk factors for CDI present in our cohort. No differences were observed between CDI^{+} and CDI^{-} individuals for the aforementioned risk factors, nor comorbidities or age distribution. However, we did detect an increase in leukocytes, lymphocytes and monocytes counts in CDI patients ($p<0.05$).

To validate our direct PCR method we used the EIA as the reference test. Our results showed a sensitivity of 1.0 and a Negative Predictive Value of 0.85 compared to EIA. Although a larger number of samples is needed to validate the method and determine specificity, this technique could be a useful method for *C. difficile* infection screening.

289. (546) **EFFECT OF TOFACITINIB ON THE ACTIVATION OF T LYMPHOCYTES IN PATIENTS WITH RHEUMATOID ARTHRITIS**

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Tofacitinib (Tofa) is a Jak1/3 inhibitor that blocks the intracellular signaling of inflammatory cytokines and is used as 3rd line of treatment in Rheumatoid Arthritis (RA). Tofa is very effective to achieve disease remission but it is associated to higher incidence of herpes zoster reactivation likely due to alterations in cellular immunity. While several studies have evaluated on the effects of Tofa on the immune system in the context of RA, knowledge about its impact on the activation and differentiation of T lymphocytes (TL) is scarce. We aimed to study this aspect in vivo and in vitro by determining the functional status of TL in different groups of treated RA patients (Tx RA) and the effect of Tofa in the activation of T cells from healthy donors (HD), respectively. Thirty-one HD and 106 RA patients were recruited in the Rheumatology Service (HNC) to evaluate numerous biochemical and immunological parameters. Principal component analysis showed that 82 of these variables explain around 70% of the variance, with variables related to the activation and differentiation of TL as the main difference between HD and different groups of Tx RA. Compared to HD, Tofa Tx RA patients presented a significant increase in the % of populations with terminal differentiation characteristics including CD27-CD28- of CD4+ TL ($p<0.01$) and KLRG1+CD57+ CD4+ and CD8+ TL ($p<0,05$). In addition, in vitro studies showed that Tofa reduced the activation of purified CD4+ and CD8+ TL as evidenced by a decrease in the upregulation of CD25, T-bet and the frequency of Ki-67+ cells. These effect were a dose-dependent and observed in total, naïve and, mainly, memory TL. Interestingly, Tofa increased the expression of senescent marker p21 in memory CD8+ TL. Altogether, our findings suggest that Tofa-induced replicative immunosenescence could underlie the biological effects of this drug in RA and be also involved in side effects, restraining the activity of memory TL involved in viral control.

290. (558) **CHEDIAK HIGASHI SYNDROME: CASE REPORT**

Introduction: Chediak Higashi Syndrome (CHS) is a rare autosomal recessive disorder, characterized by partial oculocutaneous albinism, prolonged bleeding, immune and neurologic dysfunction, and risk for the development of hemophagocytic lympohistiocytosis. The presence of giant secretory granules in leukocytes is the classical diagnostic feature, which distinguishes CHS from closely related Griscelli and Hermansky Pudlak syndromes.

The accelerated phase or HLH, is the primary cause of mortality in CHS and can occur at any age

Objective: Present patient with late diagnosis without development of accelerated phase

Clinical case: 5 year old male referred by Hematology due to the presence of intracytoplasmic granulations in neutrophils and gray hair

First child of healthy parents, not consanguineous. Recurrent obstructive bronchitis treated with budesonide with good response. No relevant infections. Difficult management of epistaxis and mild neurocognitive delay. Physical examination only shows gray hair and nystagnmus.

In laboratory, moderate neutropenia and mild anemia. Negative EBV and CMV serologies

Hair´s microscopic evaluation detects dispersal of pigment clumps throughout the hair shaft.

Normal abdominal ultrasound.