DIPARTIMENTO DI SCIENZE CLINICHE APPLICATE E BIOTECNOLOGICHE

UNIVERSITÀ DEGLI STUDI DELL'AQUILA

L'Aquila, 21 febbraio 2024

DISCAB Young Researchers 2024 DAY

Università degli Studi dell'Aquila
EDIFICIO A.C. DE MEIS
II PIANO · AULA C3.5

UNIVERSITÀ DEGLI STUDI DELL'AQUILA

DISCAB
Dipartimento di Scienze Cliniche Applicate e Biotecnologiche

PRESENTAZIONE EVENTO

Il Dipartimento di Scienze Cliniche Applicate e Biotecnologiche (DISCAB) organizza la prima edizione del **DISCAB Young Researchers Day**, un evento scientifico dedicato interamente alla ricerca di base, traslazionale e clinica dei giovani dottorandi di ricerca, specializzandi medici e di area sanitaria, e assegnisti di ricerca afferenti al Dipartimento DISCAB.

La trasmissione delle conoscenze e la formazione di nuove generazioni di ricercatori rappresenta, a tutti gli effetti, la missione principale del "Professore" universitario. In accordo con questa visione, la **Valorizzazione dei giovani** che si avvicinano alla ricerca è uno degli obiettivi strategici del Dipartimento: il **sostegno all'avvio alla ricerca** e la **formazione dei giovani ricercatori** sono aspetti che il Dipartimento ritiene fondamentali, imprescindibili per la propria crescita culturale e scientifica e per la crescita della comunità scientifica in generale. Il Dipartimento è, infatti, il luogo dove i giovani si affacciano per la prima volta al mondo della ricerca scientifica, un mondo creativo, libero, proiettato verso il futuro; è proprio nei nostri laboratori che i giovani possono far crescere le loro radici di conoscenza medico-scientifica, radici forti, che permetteranno loro, negli anni, di nutrire con vigore la pianta che crescerà, di confrontarsi con il mondo e di aprirsi alle sfide della ricerca che verranno.

Il DISCAB Young Researchers Day**,** organizzato in sessioni poster e relazioni orali selezionate, sarà l'opportunità per i più giovani ricercatori DISCAB di presentare al Dipartimento, e a tutti coloro che parteciperanno all'evento, i propri progetti di ricerca e i risultati finora ottenuti.

Il Dipartimento augura a tutti i nostri giovani dottorandi di ricerca, specializzandi medici, specializzandi di area sanitaria, e assegnisti di ricerca di vivere questa giornata come un momento di condivisione e di crescita con tutti i docenti DISCAB, ed anche come un importante momento di incontro e di scambio tra pari, sicuramente su temi scientifici ma anche sui vari aspetti della vita accademica.

La Direttrice
Francesca Zazzeroni

DISCAB Young Researchers 2024 DAY

PROGRAMMA

Mercoledì 21 febbraio 2024

09:30-10:00 **SESSIONE POSTER (I)** – *Atrio secondo piano*
Visita ai poster con numero pari

10:00-10:30 **SESSIONE POSTER (II)** – *Atrio secondo piano*
Visita ai poster con numero dispari

10.30-10.45 **INTRODUZIONE ALL'EVENTO** – *aula C3.5*

Prof.ssa Francesca Zazzeroni
DIRETTRICE DISCAB

Prof.ssa Maria Grazia Perilli
COORDINATRICE SCUOLA DI DOTTORATO IN MEDICINA SPERIMENTALE

Prof. Antonio Barile
PRESIDENTE COMMISSIONE SCUOLE DI SPECIALIZZAZIONE

10:45-12:05 **PRIMA SESSIONE COMUNICAZIONI ORALI SELEZIONATE** – *aula C3.5*
Moderatrici: Maria Grazia Perilli, Daniela Tempesta

10:45-10:55 LORENZA FAGNANI (DOTTORANDA)
Natural compounds as potential inhibitors of viral 3-chymotrypsin-like protease (3CLpro)

10.55-11.05 FANNY PULCINI (DOTTORANDA)
GMP isolation and culture of Human Umbilical Vein Endothelial Cells led to selectively expanded CD34-positive cells

11.05-11.15 LUCIA ROMANO (DOTTORANDA)
Evolution of surgical techniques in proctology: preoperative risk assessment with machine learning models

11.15-11.25 GENNARO SAPORITO (DOTTORANDO)
High Intensity Focused Ultrasound (HIFU) treatment: evaluation of long-term cognitive outcomes

11.25-11.35 ILENIA LE DONNE (DOTTORANDA)
The use of ADOS-2 in early diagnosis for Autism Spectrum Disorder: a Network Analysis and Predictive Risk Assessment study

11.35-11.45 DIANA LUPI (DOTTORANDA)
Kidney transplant from living donor: study of psychological aspects from pre to post intervention

11.45-11.55 FRANCESCO PETRAGNANO (ASSEGNISTA)
In-vitro approaches to investigate the detrimental effect of light on dopaminergic neurons

11.55-12.05 FEDERICO SALFI (ASSEGNISTA)
The Word-Pseudoword Association Learning task (WPAL): development and validation of a novel double-version self-administered tool for sleep-memory studies

DISCAB Young Researchers DAY 2024

12.30-13.30 **SESSIONE POSTER (III)** – *Atrio secondo piano*
Visita plenaria ai poster

14.30-16.10 **SECONDA SESSIONE COMUNICAZIONI ORALI SELEZIONATE** – *aula C3.5*
Moderatori: Alessandra Splendiani, Fabio Vistoli

14.30-14.40 FRANCESCA GABRIELE
SCUOLA DI SPECIALIZZAZIONE IN NEUROLOGIA
Anticoagulant-related intracerebral hemorrhage: no signals of improvements over 10 years in Italy

14.40-14.50 MATTEO RANALLI
SCUOLA DI SPECIALIZZAZIONE IN RADIODIAGNOSTICA
Evaluation of extra-cellular myocardial volume in CT in patients with gastrointestinal cancer disease

14.50-15.00 RITA SEBASTIANI
SCUOLA DI SPECIALIZZAZIONE IN RADIOTERAPIA
Dosimetric comparison of organs at risk using two delineation guidelines for the radiation treatment of cT1 laryngeal cancer

15.00-15.10 GIOVANNI ALLOGGIA
SCUOLA DI SPECIALIZZAZIONE IN PATOLOGIA CLINICA E BIOCHIMICA CLINICA
Flow cytometry in the determination of platelet-associated immunoglobulins

15.10-15.20 ANTONELLA GRASSO
SCUOLA DI SPECIALIZZAZIONE IN CHIRURGIA GENERALE
The role of renal graft preimplantation biopsy. experience of our center

15.20-15.30 EMANUELE VAGNOZZI
SCUOLA DI SPECIALIZZAZIONE IN DERMATOLOGIA E VENEREOLOGIA
Non-invasive techniques for the diagnosis of suspicious skin lesions

15.40-15.50 ALESSIA RUSSO
SCUOLA DI SPECIALIZZAZIONE IN PSICHIATRIA
Mental health between the covid-19 pandemic and the 2009 earthquake in L'Aquila. Changes, socio-demographic and clinical characteristics of the users of the university psychiatric service of diagnosis and treatment of the "San Salvatore" hospital in L'aquila.

15.50-16.00 GIULIA CATALDI
SCUOLA DI SPECIALIZZAZIONE IN REUMATOLOGIA
The evaluation of effectiveness and safety of Guselkumab in patients with psoriatic arthritis in a prospective multicentre "real-life" cohort study

16.00-16.10 GABRIELE DI PASQUALE
SCUOLA DI SPECIALIZZAZIONE IN PEDIATRIA
Dissecting the genetic basis of pediatric developmental epileptic encephalopathies: a multicenter study from central and southern Italy

16.30-17.30 **SESSIONE POSTER (IV)** – *Atrio secondo piano*
Visita finale della commissione per l'assegnazione dei premi

P1

Clinico-pathological and survival characteristics of patients with mutated BRAF and NRAS melanoma: a retrospective monocentric experience

Manfredo BRUNI[1,2], Cristina PELLEGRINI[1], Paolo ANTONETTI[1], Chiara CAPONIO[2], Alessia LODA[1], Mirco MASTRANGELO[1], Ludovica CARDELLI[1], Marco CLEMENTI[1,2], Eleonora CAROSA[1], Annarita LIZZI[1], Maria ESPOSITO[1,2], Maria Concetta FARGNOLI[1,2]

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Dottorato in Medicina Sperimentale - Scienze cliniche, mediche e chirurgiche
Ciclo: XXXIX

Settore Scientifico Disciplinare (SSD): MED/35

Cutaneous melanoma originates through a gradual accumulation of molecular alterations involving various cellular pathways. The MAP kinase is the most important implicated in pathogenesis, being altered in about 80% of cases. Approximately 45-50% of melanomas have a mutation in *BRAF* gene, while *NRAS* is altered in 25-30% of cases. We present the experience of the Skin Cancer Tumor Board (SCTB) of ASL1 Abruzzo, L'Aquila, which managing patients with malignant skin neoplasms since June 2020. Our study aimed to analyze the clinical-pathological and prognostic differences between melanomas with *BRAF* and *NRAS* mutations. Retrospectively, we collected data from 56 patients with primary *BRAF* or *NRAS* mutated melanoma from the SCTB database. Using univariate (Chi-square and/or Mann-Whitney tests) and multivariate analysis (logistic-regression), we compared the two groups based on the following characteristics: gender, age, anatomical site, histotype, ulceration, Breslow thickness, number of mitoses, presence of regression, microsatellite metastasis, inflammatory infiltrate, lymphovascular invasion, perineural invasion, associated melanocytic nevus, solar elastosis, and sentinel lymph node positivity. Differences in disease-free survival (PFS) and overall survival (OS) were evaluated using the log-rank Cox test.
In our cohort (27% women, 73% men), 70% had *BRAF* mutations, and 30% had *NRAS* mutations. *BRAF* mutations were significantly more frequent in younger patients (*BRAF*: median age 63.7 years, IQR = 21.9; *NRAS*: 74.8, IQR 184, p=0.017). *NRAS*-mutated melanomas exhibited a greater thickness (2.3 mm, IQR 2.6 vs. 4.6, IQR 4.65, p=0.040) and were more often nodular (OR 4.3, 95%CI 1.1-16.3, p=0.031), while *BRAF*-mutated melanomas tended to show regression (p= 0.068). No significant differences were found in other clinical-pathological aspects. Patients with *NRAS* mutation had higher survival rates in terms of both disease-free survival (PFS) and overall survival (OS) compared to *BRAF* (average time of 52.0 vs. 41.7 months for PFS and 56.95 months vs. 50.7 for OS respectively), though not statistically significant (PFS for *NRAS*: HR 0.66, 95%CI 0.2-1.9, p=0.43; OS for *NRAS* HR 0.79, 95%CI 0.20-3.2, p=0.74).
In conclusion, *NRAS* mutations seem associated with clinical-pathological factors suggesting a worse prognosis, although survival data indicate better outcomes. Further studies need to elucidate the impact of *BRAF/NRAS* mutational status on melanoma prognosis.

P2

The mismatch repair system in Pit1-derived pituitary neuroendocrine tumors (PitNETs): from bench to bedside

Francesca Carbonara[1], Mariana Moroni[1], Francesco Mancinelli[1], Tiziana Feola[2,3], Francesca Gianno[2,4], Vincenzo Esposito[2,5], Felice Giangaspero[2,4], Marie Lise Jaffrain-Rea[1,2]

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Ciclo: XXXVII

Settore Scientifico Disciplinare (SSD): MED-13

Pit1-derived PitNETs includes lactotroph (PRL), somatotroph (GH), mixed GH/PRL-secreting and rare thyreotroph tumors. Few cases develop in a genetic context, including germline (AIP) mutations and syndromic forms. Although two cases of PRL PitNETs have been associated with Lynch Syndrome, little is known about the potential role of the MMRs system in unselected PitNETs. **Aim:** to analyse the mismatch repair system (MMRs) expression in the Pit1 subgroup and look for individual and/or familial association with malignancies. **Material and Methods:** qRT-PCR for MLH1, MSH2, MSH6 and PMS2 gene expression, normalized for β-actin, was performed in 41 surgical samples. Promoter methylation of MLH1, MSH2/6 was investigated by methylation-specific PCR on tumor DNA. Immunoistochemistry (IHC) was preliminary performed on 7 samples. Sanger sequencing was used to search for GNAS1, SF3B1, BRAF hotpot mutations and AIP LOH where appropriate. The study was approved by the Neuromed IRB, and the data were analysed by non- parametric tests considering potential association with patients/familial malignancies. **Results:** A low MMRs gene expression was observed as follows: 1) all MMRs in invasive tumors (MLH1/PMS2 p=0.02, MSH2/6 p=0.04, vs non-invasive); 2) MLH1 and MSH2 in the youngest patients (p=0.04, vs > 30 years-old), 3) MSH2 in high Ki67 samples (p=0.03, vs low Ki67). MSH2 promoter methylation was observed in 2 samples showing low MSH2 mRNA. MSH6-IHC was negative in 3 samples showing low MSH6 mRNA. No somatic GNAS1, SF3B1 or BRAF mutation was found. LOH was confirmed in 3 PitNETs from patients with a known germline AIP mutation, one showing a low MSH6 mRNA, and another one had low MSH6, MSH2 and MLH1 mRNA. Out of 29 informative patients, 5 had associated malignancies (17.2%), including 3 with some reduced MMRs transcription. Familial data showed a similar prevalence of malignancies in patients (68.4%) and controls (64.7%) (EpidemioPit study). **Conclusion:** These data suggest a role for a reduced MMRs expression in invasive, early- onset and proliferative Pit1-derived PitNETs. Methylation of the MSH2 promoter may occur in a few cases. Further information on MMRs protein expression and association with malignancies are being collected to look for potential indications to germline MMRs sequencing.

P3

Towards the identification of protective mechanisms for retinal pigment epithelium: insights into Age-Related Macular Degeneration

Giulia Carozza, Darin Zerti, Rita Maccarone

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Dottorato in Medicina Sperimentale - Neuroscienze di base e cliniche
Ciclo: XXXVII

Settore Scientifico Disciplinare (SSD): BIO/09

Retinal Pigment Epithelium (RPE) is fundamental in maintaining retinal homeostasis, and its dysfunction is an early event occurring during Age-Related Macular Degeneration (AMD). Oxidative stress is the main cause triggering RPE dysregulation and it has been identified as a main player in AMD pathogenesis. Preserving the functional role of RPE is mandatory to avoid retinal degeneration. Based on this knowledge, the objective of this project was to investigate key factors related to the RPE dysfunction with the final aim to identify molecules able to prevent its degeneration. Recently, it was demonstrated that L-3,4-dihydroxyphenylalanine (L-DOPA) delay the onset of AMD, by activating the signaling of its selective receptor, GPR143, localized on the RPE. Literature on this topic also showed how Curcumin (Cur), a natural compound, could be effective on many retinal diseases, by modulating multiple pathways (e.g. oxidative stress, apoptosis, and autophagy). Interestingly, in an animal model of Parkinson Disease, a combined treatment with Cur and L-DOPA results in a synergic effect of these substances. Therefore, we aimed to explore the outcomes of Cur and L-DOPA treatments on ARPE-19 cells, by focusing on oxidative stress-related mechanisms. First, we evaluated the potential cytotoxicity of Cur and L-DOPA on RPE cells (ARPE-19) through a cell viability assay. We also established a cellular model of oxidative stress using increasing hydrogen peroxide (H2O2) concentrations, and we tested the effectiveness of Cur and L-DOPA in preventing ARPE-19 death. In addition, through western blot technique we evaluated known relevant mechanisms involved in AMD pathogenesis (oxidative stress, autophagy, SIRT1 signaling and GPR143 signaling). We selected safe concentrations of Cur and L-DOPA (up to 20 μM and 100 μM respectively) to treat the ARPE-19 cells exposed to H2O2. ARPE-19 cell viability decreased progressively with increasing concentrations of H2O2, which also modulated the protein expression of: GPR143, catalase and autophagy markers (LC3B and p62), in addition to the NAD+-dependent deacetylase (SIRT1) which is involved in key metabolic functions. So far, we demonstrated that Cur or L-DOPA prevented ARPE- 19 cell death induced by oxidative stress, paving the way to further experiments to identify pathways involved in these protective effects.

P4

Reduction of neuroinflammation through administration of URB597 in an animal model of Alzheimer's disease

Giacomo Cimino[1], Annamaria Tisi[1], Lucia Scipioni[1,2], Giacomo Giacovazzo[2,3], Sergio Oddi[2,3], Mauro Maccarrone[1, 2]

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Dottorato in Medicina Sperimentale - Biotecnologie e scienze biochimiche
Ciclo: XXXIX

Settore Scientifico Disciplinare (SSD): BIO/10

Arachidonoylethanolamide (AEA) is an endocannabinoid with recognized neuroprotective properties. Here, we investigated the potential anti-inflammatory effects of URB597, an inhibitor of fatty acid amide hydrolase (FAAH) responsible for AEA degradation, in an animal model of Alzheimer's disease (AD). 6-month-old AD-like Tg2576 (TG) and wild type (WT) mice were treated intraperitoneally with lipopolysaccharide (LPS) to exacerbate neuroinflammation, and intranasally with URB597 for 3 months. Confocal microscopy was used to identify anti-IBA-1 immunopositive microglial cells. A significant increase in the number of IBA-1(+) cells was found after LPS treatment in the hippocampus of both TG and WT, and was significantly reduced by URB597 in both genotypes compared to LPS alone. Moreover, morphometric analysis was conducted on 360 individual microglial cells in the CA1 and dentate gyrus (DG) of the hippocampus, in order to measure major hallmarks of microglial activation: cell area, area of the soma, number and area of the branches. We found intriguing morphometric changes in LPS- treated groups in the CA1 and DG of both WT and TG, compared to vehicle-treated mice. Indeed, we found a reduction of the cell and branches areas, without alterations in the number of branches and area of the soma, suggesting that the morphometric changes induced by LPS predominantly involved a retraction of the branches. Importantly, URB597 restored the area values of cells and branches in the CA1 of WT and in the DG of TG, indicating a regionalization of the effect. Finally, anti-β-amyloid immunostaining revealed the absence of plaques under all experimental conditions. Overall, our data demonstrate that: (i) URB597 dampens LPS-induced microglia reactivity in both WT and AD-like mice; (ii) neuroinflammation precedes the onset of β-amyloid plaques in Tg2576 mice.

P5

Set-up of an ultrasound-based method to promote the release of putative biomarkers and the uptake of small drugs: study on HCC cell lines

Colaianni Francesco[1], Compagnoni Chiara[1], Zelli Veronica[1], Capece Daria[1], , Barbato Gaetano[2], Alesse Edoardo[1], Rossi Mario[1], Zazzeroni Francesca[1], Tessitore Alessandra[1]

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Dottorato in Medicina Sperimentale - Biotecnologie e scienze biochimiche
Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): MED/46

Background. Hepatocellular carcinoma (HCC) is the most death-causing liver cancer. Ultrasounds (US)-based methods can be considered as innovative approaches to improve HCC diagnosis/therapy. HCC non-invasive early diagnosis is difficult, as serum levels of available biomarkers are mainly suggestive, not precisely diagnostic. Traditional/advanced chemotherapy shows several well-known issues and side-effects, as drugs affect tumor, but also normal cells. US have already shown to promote both the release of putative biomarkers (microRNA/proteins) and the drug uptake in *in vitro* and *in vivo* cancer models. Regarding the latter, the use of this technology (e.g. focused US) would be of great interest to boost the drug uptake especially in the cancer site, with possible dose-reduction.
Aim. A prototype instrument (SonoWell, InnoSol) was used to treat HCC cells with US, with the aim to promote: i) the release of small molecules (microRNAs) and ii) small-molecules uptake.
Methods. HCC cells (HepG2, SNU-387) were cultured. After proper incubation and adhesion, cells were US-treated at different experimental conditions. Untreated cells were incubated in parallel. RNA, including the small fraction, was extracted from supernatants and analysed by qRT-PCR. To assess the release of microRNAs, miR-182 and miR-125a-5p, known to be expressed in HCC, were used. To evaluate the uptake of small molecules, cells were incubated with the FAM-labelled AS1411 aptamer at different timepoints. Aptamer internalization was analysed by fluorescence intensity.
Results. After experimental conditions set-up, US treatment showed HCC cells supernatants' enrichment of miR-182 and miR-125a-5p. Depending on time exposure, US treatment increased also AS1411 uptake.
Conclusions. US treatment increased miRNAs' release and the aptamer's uptake in HCC cells. Further experiments are ongoing to: i) characterize and profile small molecules (e.g. microRNA, proteins), to be putatively considered as biomarkers, released in supernatants, and ii) analyse the dose-dependent effects of chemotherapy drugs (sorafenib) on HCC cells after US treatment.

P6

Carboxylesterase 1 (CES1) as a potential therapeutic target in Acute Myeloid Leukemia

Francesca Dall'Aglio[1], Daniela Verzella[1], Irene Flati[1], Davide Vecchiotti[1], Mauro Di Vito Nolfi[1], Francesca Veglianti[1], Edoardo Alesse[1], Frazoso Guido[2], Daria Capece[1], Francesca Zazzeroni[1]

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Dottorato in Medicina Sperimentale - Biotecnologie e scienze biochimiche
Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): MED/46

Background. Cancer cells undergo metabolic reprogramming in response to low-nutrient microenvironments, allowing tumor cell survival and proliferation, and contributing to drug resistance. Recently, carboxylesterase 1 (CES1) has been identified in colorectal carcinoma (CRC) as an essential NF- κB-regulated lipase promoting metabolic adaptation and tumor progression. Specifically, the lipase CES1 promotes cancer cell survival under energy stress conditions in CRC, by increasing TAG breakdown to fuel fatty acid oxidation and preventing their toxic build-up. As CRC, other cancers such as Acute Myeloid Leukemia (AML), depend on oxidative metabolism.

Aim. This project aims to investigate whether CES1 could play an important role also in the metabolic adaptation of AML cells.

Methods. Public datasets of AML patients were analyzed for CES1 expression. A panel of AML cell lines
was tested for CES1 expression, cultured both in basal and energy stress conditions, by qRT-PCR and Western Blot.

Results. Our analysis of patient datasets showed that CES1 expression is higher in M4 and M5 FAB AML subtype and relates to worse prognosis in AML patients. We showed that CES1 was expressed both in basal and energy stress conditions, suggesting that this lipase could be involved in governing metabolic requirements of AML cells.

Conclusions. Metabolic adaptation is a novel hallmark of leukemogenesis required for tumor initiation, progression, and therapeutic responses. Our preliminary data suggest that CES1 could be a potential druggable target in AML.

P7

Lipid signalling in Space

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Dottorato in Space Science and Technology - Astrobiology, Life Sciences and Space Medicine
Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): BIO/10

Background. Microgravity, a Space-related stressor, is associated with immune dysregulations that impact on the astronauts homeostasis, gastro-intestinal (GI) tract included. Of note, all the inflammation's phases are orchestrated by specific endogenous bioactive lipids, that can be synthesized by specific fatty acid precursors like arachidonic acid (ARA) and docosahexaenoic acid (DHA). Experiments performed aboard the International Space Station (ISS) – like ROALD, ROALD-2/RESLEM and SERiSM – and under simulated microgravity conditions, demonstrated that metabolism and signal transduction of bioactive lipids are dysregulated in microgravity. **Aim**. Hence, we sought to interrogate whether bioactive lipids may influence human intestinal cells' response to authentic or simulated microgravity, thus contributing to maintain GI homeostasis. Accordingly, nutraceutical or pharmacological modulators of the receptors binding such lipids, as well as of their metabolic enzymes, were employed. The final goal was to develop potential countermeasures against Space-related human diseases. **Methods.** Human Caco-2 cells were chosen as a model of intestinal epithelial cells. During ground control studies (1xg) Caco-2 cells were treated with ARA, DHA, and the short chain fatty acids (SCFAs) butyrate (But) and propionate (Pro). Furthermore, Caco-2 cells were subjected to simulated microgravity (10- 5xg) by means of the Rotary Cell Culture System (RCCS) developed by NASA. Western blotting and qPCR were performed to interrogate possible effects on the endocannabinoid (eCB) system (ECS) at a genic and protein level, given its potent pro-homeostatic role. **Results & Conclusions.** In the 1x*g* experiments, Caco-2 cells were treated with the 50% critical micelle concentration (1⁄2CMC) of ARA and DHA (30 μM), as well as of But and Pro (2.5 mM). Briefly, ARA induced a decrease in the expression of the eCB-binding cannabinoid receptors (CB1 and CB2), whereas DHA downregulated the expression of CB2 only. As for the SCFAs, But reduced DAGLα enzyme expression and Pro did not modulate any component of the ECS. Regarding the microgravity studies, Cytodex 3 was found to be necessary for Caco-2 survival [9,10]. Of note, simulated microgravity did not affect protein expression of eCB-binding receptors. Further studies are in progress to elucidate the possible modulation of other components of the ECS.

P8

Preliminary metabolomic investigation and characterization of differentiated and induced stem models of Glioblastoma

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Dottorato in Medicina Sperimentale - Medicina sperimentale, clinica ed endocrinologia
Ciclo: XXXIX

Settore Scientifico Disciplinare (SSD): MED/50

Introduction: Glioblastoma multiforme (GBM) is the most common type of primary malignant brain tumor in adults. Despite the progress made over the years the outcomes remain unsatisfactory due to both the impossibility of complete resection of the tumor mass and, above all, the presence of a stem cell population (GSC) with characteristics of self-renewal and multi-lineage differentiation, refractory to both chemo- and radiotherapy (RT). GSCs are regulated by six main mechanisms that include both intrinsic and extrinsic factors, including metabolism. Indeed, the deregulation of metabolism is one of the hallmarks of cancer and plays a central role in carcinogenesis, drug resistance mechanisms and stemness. Several studies show that GBM cells are characterized by high metabolic plasticity and can adapt their metabolism according to the levels of oxygen and nutrients to which they are exposed, in particular the GSCs. Although glucose is the main energy source, it has been shown that GBM uses unique metabolic pathways and "unconventional" energy sources for its survival.
Aim: In this work we focused on characterizing both from a molecular and metabolic point of view a highly proliferating population and a stem-induced population of a representative GBM line.
Methods: U87-MG cell line and U87-MG stem-induced were used. Flow cytometry, Western Blot, immunofluorescence assay, NMR spectroscopy and Oil Red O staining were perfrmed.
Results: The U87-stem induced cell population is positive for stem cell markers, including CD44, which may be an interesting target as it is involved in processes such as proliferation, maintenance of stemness, regulation of the glycolytic pathway, etc., while the U87 population is positive for its isoform, CD44v6, which also plays a crucial role in the tumor's survival processes. The metabolic profile of the two populations shows a dependency on glutamine metabolism, which could be a promising therapeutic target.
Conclusion: The molecular and metabolic characterization of the two GBM representative cell populations shows how CD44 and its CD44v6 isoform, as well as metabolic pathways could be interesting targets as they play a central role in tumor and GSCs survival.

P9

Clinical usefulness of 18F-FDG PET/CT in dissecting the heterogeneity of patients with FUO and IUO. Results from a "real-life" study

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Dottorato in Medicina Sperimentale - Medicina sperimentale, clinica e endocrinologia Ciclo:XXXIX

Settore Scientifico Disciplinare (SSD): MED/16

Background: Fever of unknown origin (FUO) is defined as a condition characterized by fever (≥ 38.3°C) without explanation despite minimal standard diagnostic tests in an immunocompetent patient.
Aims: To evaluate the usefulness of 18F-FDG PET/CT to guide the diagnosis of patients with FUO.
Methods: We identified all patients with FUO who underwent 18F-FDG PET/CT at the University Hospital of Pitié-Salpêtrière (France) between 2009 and 2022. Inclusion criteria were i. age 16 to 70 years; ii. fever > 38°C in at least two occasions or duration of fever ≥ 3 weeks; elevated inflammatory markers (CRP>5 mg/dL). Exclusion criteria were i. treatment by prednisone or equivalent at a dose > 10 mg/d; ii. treatment by biologic DMARDs at the time of the examination or the 10 days before. Patient medical charts were reviewed and 18F-FDG PET/CT findings were analysed after stratification on 3 disease groups: i. patients with neoplasia, ii. patients with infections, iii. patients with immune-mediated inflammatory disorders (IMID).
Results: Out of 331 assessed patients, 128 (38.7%) showed findings of 18F-FDG PET/CT hypermetabolism in one or more areas. The final diagnoses of these patients were: 16 neoplasia (mainly lymphoma), 42 infections (mainly sepsis and lung infections), and 70 patients with IMID. Patients with neoplasia were characterized by a higher prevalence of hypermetabolism of lymph nodes (p=0.008), and bones (p=0.003), compared to other diagnosis groups. In infection group, 18 patients (42.90%) showed a lymph node hypermetabolism. The IMID group was the most heterogenous. Lymph nodes hypermetabolism was mainly retrieved in Still's disease (83.33%) and sarcoidosis (100%). Furthermore, hypermetabolism of bone marrow was described in 4 patients (6.67%) with Still's disease. Hypermetabolism of blood vessels were described in nearly all patients (85.71%) with vasculitis. Additionally, findings of hypermetabolism of joints were described in patients with polymyalgia rheumatica (100%) and spondylarthritis (100%).
Conclusions: 18F-FDG PET/CT is interesting in patients with FUO to identify hypermetabolism patterns suggestive of neoplasia, infections or IMID. We propose a practical diagnostic algorithm to assist clinicians in making an accurate and timely diagnosis.

P10

Co-existence of blaNDM-5, bla-CTX-M-15, blaOXA-232, blaSHV-182, ompK36 and ompK37 mutations in K. pneumoniae resistant to meropenem-vaborbactam and ceftazidime-avibactam

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Settore Scientifico Disciplinare (SSD): BIO/12

Introduction: Klebsiella pneumoniae is an important pathogen causing severe nosocomial infections. Especially, carbapenem-resistant K. pneumoniae is designed by WHO as one of the ESKAPE microorganisms that poses an urgent threat to public health. K. pneumoniae strains are resistant to various classes of antibiotics and they are considered a reservoir of extended-spectrum β-lactamases and classes A, B and D carbapenemases.

Methods

Whole-Genome Sequencing (WGS) was performed using Illumina MiSeq platform with a 2 x 300 paired-end run (Illumina Inc., San Diego, CA, USA). Raw data from sequencing were quality checked using FastQC tool and assembled with SPAdes Genome Assembler v3.9.0. ResFinder 4.4.2 was used to detect acquired antimicrobial resistance genes (ARGs) and chromosomal mutations. Mobile genetic elements (plasmids and ISs) were identified by MobileElementFinder v1.0.3.

Results

WGS was carried out on two K. pneumoniae isolates collected from urine samples of patients hospitalized in two different wards of L'Aquila Hospital (Central Italy). The K. pneumoniae strains were resistant to most antibiotic tested (imipenem, meropenem, ertapenem, meropenem- varbobactam, cefotaxime, ceftazidime, ampicillin, amoxicillin-clavulanate, ceftazidime- avibactam, cefepime, colistin, ciprofloxacin) but showed susceptibility to cefiderocol. Both strains belonged to ST437. The IncFIB, IncFII and Col-type plasmids and ISs, associated with IS3, IS5 and IS66 families, have been found in these strains. The presence of blaNDM-5, blaCTX-M-15, blaSHV-182, blaOXA-232 genes conferred resistance to several β-lactams, including carbapenems. In addition, we detected ten mutations in ompK36 (N49S, L59V, G189T, F198Y, F207Y, T222L, D223G, E232R, N304E and A217S) and three mutations in ompk37 (I70M, I128M, N230G), which determined resistance to cephalosporins and carbapenems. Other ARGs, including mphA, ARR- 2, aac(6')-Ib-cr, aadA2, rmtB, fosA, ermB, sul1, dfrA12 and qacE were detected. Resistance to fluoroquinolones was also mediated by OqxAb efflux pumps and mutation in acrR (P161R, G164A, F172S, R173G, L195V, F197I, K201M).

Conclusions

K. pneumoniae strains, analysed in the present study, could be considered hypervirulent strains with a complex framework of resistance genes.

P11

Attentional Demands-Task (AD-Task): a new tool to study the modulation of selective and divided attention processes.

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Settore Scientifico Disciplinare (SSD): M-PSI/01

Aim: To develop and validate a new experimental paradigm (Attentional Demand Task, ADT) that measures selective and divided attention, as well as the "cognitive cost" due to switching between them.
Materials: 42 participants (age: 21.0 ± 2.22; 33 F) completed the ADT, a selective attention task (Oddball-Task, OD) and a divided attention task (Dual-Task, DT). To assess learning effects, a sample of 25 subjects (age: 22.3±2.76; 18 F) was divided into an experimental group (13 subjects) and a control group (12 subjects).
Method: To validate the instrument, participants performed the three tasks on three different consecutive days. Regarding the evaluation of learning effects, the experimental group, unlike the control group, performed a training session of the ADT the day before the administration of the original ADT. Performance was evaluated considering both reaction time (RT) and accuracy indices (hit rate, d').
Results: A significant correlation was observed between speed measures and accuracy in the ADT and both the OD (r=.800; p=< .001 / r=.596; p=>.001 / r=.449; p=.003) and DT (r=.678; p=< .001 / r=.418; p=.006 / r=.546; p=>.001). A repeated-measures ANOVA with the within-subject factors "ATTENTIONAL DEMAND TYPE" (selective/divided) and "CONDITION" (single demand/switching demand) showed a main effect of TYPE OF ATTENTIONAL DEMAND on all variables considered, showing better performance, shorter reaction times, and better ability to discriminate the target stimulus in selective attention than in divided attention ($F(1,41) = 90.429$, $\eta 2 = 0.360$, $p = <.001$ / $F(1,41) = 691.77$, $\eta 2 = 0.517$, $p = <.001$ / $F(1,41) = 105.495$, $\eta 2 = 0.291$, $p = <.001$). There was also a significant interaction between the ATTENTIONAL DEMAND TYPE and CONDITION in the RT of selective attention (Mdif = -12.69, t(41.0) = -3.949, p=.0.002). The one-way ANOVA used for comparison between the experimental group and the control group showed no statistically significant differences.
Discussion: From the analyses, the ADT is a candidate instrument for measuring the two attentional processes and the switching between them, devoid of learning effects. In addition, longer RTs and performance decline in divided Vs. selective attention confirm that performance declines when additional demands increase.

P12

Structural proprieties and bisphenol-a release from the aging of orthodontic clear aligners

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Settore Scientifico Disciplinare (SSD): MED/28

Introduction: Orthodontic clear aligners are composed by thermoplastic polymers that during the use undergo to sudden changes of temperature and pH, which can affect the physical properties of the material and cause the release of thermoplastic polymers. Among them there would be the Bisphenol-A (BPA): a compound with weak estrogenic properties. Since in literature there are no clear evidences, the objective of this study is to assess the release of biologically active molecules and BPA from clear aligners and consequently to assess the in- vivo damage caused by the loss of substances. Additionally, a preventive ageing will also be seek to reduce the risk of BPA ingestion.
Materials and methods: Six types of thermoplastic polymers are being tested: PET, PETG, PC, EVA, hybrid TPU and 3D printed resin. The polymer discs were all thermoformed at a pressure of 3 bar and for each polymer, the samples to test were prepared using a mould measuring 4 mm x 4 mm. To isolate and divide the most polar molecules from the less polar ones, two solvents with different polarities have been used: ethanol and saline solution.
Results: The results of the first part of the study showed that the most effective method for the extraction of thermoplastic pomlymers is the solute precipation trought vacuum concentrator. Additionally, there have been identified standard molecules to use for the evaluation of molucles extracted with the gas cromatograph.
Future evaluation: To assess the release of substances with estrogenic-like-action (EA), cellular assays will be used: breast cancer cells MCF-7 and filtered chemicals will be tested measuring cell proliferation. The estrogen-insensitive MDA-MB-231 cells will be used as negative controls. Furthermore, if any of the polymers tested will show EA, the specificity of the effect will be confirmed by the evaluation of MCF-7 proliferation following incubation of polymer-derived solutions together with the strong antiestrogen ICI.182,780. In addition, to confirm EA, a RT- PCR analysis for some estrogen-reactive genes will be perform. Finally, if the the presence of polyhermers with EA will be confirmed, an extra-oral aging protocol will be sought to prevent the dissolution of these molecules in the intra-oral environment.

P13 – SELECTED ORAL COMMUNICATION

Natural compounds as potential inhibitors of viral 3-chymotrypsin-like protease (3CLpro)

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Settore Scientifico Disciplinare (SSD): BIO/12

Background. In response to the urgent requirement to mitigate the progression of SARS-CoV-2 infections, scientific endeavors have concentrated on the formulation of efficacious vaccines, although repurposing approved drugs and natural compounds emerges as a notably viable option. This research targets the principal 3CL protease of the virus. Lichen secondary metabolites, curcuminoids and derivatives, and other natural compounds may be considered good candidates due to their many biological properties. However, their therapeutic potential has not yet been fully explored.
Aim of the study. The present study is the conclusion of a work previously begun. After a preliminary screening of natural compounds, the study focused on the potential inhibitory activity of some of them (protocetraric, salazinic, fumarprotocetraric, and perlatolinic acids, curcuminoids and derivatives, phenylpropanoid compounds) against SARS-CoV-2 3CLpro.
Methods. The kinetic parameters were assessed by measuring the fluorescence intensity with a microtiter plate-reading fluorimeter using the fluorogenic substrate DABCYL-KTSAVLQSGFRKME-EDANS. The cytotoxic effects were evaluated on murine Sertoli TM4 cells. *In silico* analysis was conducted to ascertain the interaction dynamics between natural compounds and the 3CLpro.
Results. Some compounds exhibit no inhibitory effect on 3CLpro, contradicting predictions from in silico studies reported in the literature. Lichen secondary metabolites are found to be slow- binding inactivators of SARS-CoV-2 3CLpro with a Ki value of 3.77 µM (salazinic), 3.95 µM A (protocetranic), 17µM (fumarprotocetraric) and 0.67µM(perlatolinic).Among curcuminoids and derivatives, Ki values range from 3 to 12 µM. The mechanism of inhibition reveals some compounds as competitive inhibitors, while others act as noncompetitive inhibitors. All compounds do not exhibit cytotoxicity, consistent with data in the literature; for example, the viability assay on murine epithelial cells revealed that salazinic and protocetraric acids do not show cytotoxicity up to 80 µM. The molecular modelling confirms the biochemical data.
Conclusion. The findings contribute significantly to the existing knowledge of the biological activities of natural compounds. Based on the outcomes of kinetic analyses, computational studies, and cytotoxicity studies, we can conclude that some examined compounds represent viable frameworks for crafting potent inhibitors targeting the 3CL cysteine protease of SARS- CoV-2.

P14

Assessing Resilience to Sleep Loss Among the Italian Population: A 13-Item Model of the Iowa Resistance to Sleeplessness Test (iREST)

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The present study aimed to validate the Italian version of the Iowa Resistance to Sleeplessness Test (iREST), a 16-item self-report assessing resilience to sleep debt in the affective, cognitive, and somatic domains.
We examined its factor structure, assessed internal consistency and criterion validity, and established test-retest reliability on 768 Italian native speakers (65.8% of women) with a mean age of 25.98. Confirmatory factor analysis (CFA) revealed a new 13-item structure for the Italian iREST (iREST- 13), demonstrating more satisfactory goodness-of-fit values, and exhibiting good internal consistency (Cronbach's α ranging from 0.73 to 0.89), relative to the 16-item original version. Results supported the iREST convergent validity, showing significant independence from established measures of sleep; low correlations with conceptually unrelated measures supported divergent validity, indicating that the iREST effectively measures resistance to sleeplessness without confounding with other constructs. Lastly, test-retest reliability was evaluated by administering the iREST to the same sample with a 2-week interval: the significant correlations supported its temporal stability.
Further studies are needed to evaluate the applicability of the iREST in diverse populations and explore its relationship with objective sleep measures. Nevertheless, the Italian iREST provides a valuable tool for assessing resistance to sleep loss, offering insights into individual differences in resilience. Additionally, the iREST can assist in identifying individuals who require interventions to enhance resilience to sleep debt, as well as help clinicians evaluate the impact of chronic sleep disruption and deliver targeted interventions.

P15

Targeting CES1 to counteract metabolic reprogramming in platinum-resistant ovarian cancer (OvCa) cells

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Ciclo: XXXVI

Settore Scientifico Disciplinare (SSD): MED/46

Background: OvCa is the deadliest gynaecological malignancy worldwide due to the frequent failure of first-line platinum-based chemotherapy. Increasing evidence indicate that drug- resistance is often associated with metabolic reprogramming, a key property of cancer cells, which can adapt to harsh microenvironments. Understandind how metabolism is rewired in tumour cells and which are the molecular mediators underneath could pave the way for developing new target therapies to use in combination with conventional chemotherapy. Recently, carboxylesterase 1 (CES1), has been identified in colorectal carcinoma (CRC) as a lipase promoting cancer-cell survival and metabolic adaptation under energy stress (ES). Specifically, CES1 promoted the survival of CRC cells by increasing TAG breakdown to fuel b-oxidation. Like CRC, OvCa shows addiction to lipid metabolism, preferentially by infiltrating the peritoneal cavity and the omentum, a fat-rich organ.
Aim: We aimed to investigate whether CES1 could be crucial also in OvCa cell metabolic adaptation representing an alternative druggable target, especially in the chemo-refractory subsets.
Methods: CES1 levels, metabolic phenotype, and survival under ES conditions, with or without specific CES1 inhibitor and in combination with carboplatin were evaluated by qRT- PCR, seahorse, WB, and viability assay using a panel of four OvCa cell lines. Public datasets of OvCa patients were analyzed.
Results: We found that OvCa cells become resistant to carboplatin when cultured under ES. Moreover, during ES, OvCa cells metabolically switch to b-oxidation and upregulate CES1 suggesting that lipid metabolism and CES1 could be relevant to cope with nutrient fluctuations. Indeed, pharmacological CES1 blockade by commercially available GR-148672X inhibitor impaired bioenergetic parameters, blocked autophagy flux and caused cell death in carboplatin-resistant OvCa cells. The clinical relevance of CES1 blockade is supported by the inverse correlation between CES1 expression and worse prognosis in OvCa patients.
Conclusions: These data underscore the actionability of CES1 inhibition to impede autophagy, lipid catabolism and survival in platinum-resistant OvCa cells.

P16

Sex differences in the epidemiology of intracerebral hemorrhage over 10 years in a population-based stroke registry

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Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): MED/26

Background: We investigated incidence and outcome of spontaneous ICH in a population-based stroke registry and provided data to inform on the figures of the disease in women and in men. **Methods:** Our prospective population-based registry included patients with first-ever ICH occurring from January 2011 to December 2020. Clinical and neuroimaging records were screened to evaluate the presence and location of ICH. Incidence rates were standardized to the 2011 Italian and European population in men and women. Incidence rate ratios (IRRs) were determined according to Poisson distribution. Poisson regression was used to estimate time trends of ICH incidence. We also estimated 30-days and 1-year case-fatality rates after ICH. Multivariate hazard ratios for 30-day and 1-year fatality were estimated with Cox regression analysis including components of the ICH score and sex. **Results:** We included 748 first-ever ICH (41.3% women). Women were significantly older than men at ICH onset (78.9±12.6 versus 73.2±13.6 years; p<0.001) and showed significantly higher clinical severity on admission (median National Institute of Health Stroke Scale score 11 – IQR 6- 20 versus 9 – IQR 4-15, respectively; p=0.016). The crude annual incidence rate was 20.2 (95% CI 18.0-22.6) in women and 30.2 per 100,000 person-years in men (95% CI 27.4-33.2); incidence was significantly lower in women compared to men (IRR 0.67, 95% CI 0.58-0.78; p<0.001) and did not change significantly over time in both women and men (P for trend=0.073 and 0.904, respectively). Unadjusted comparison of case-fatality rates showed higher 1-year mortality in women compared to men (48.5% versus 40.1%; p=0.026). However, after adjusting for components of the ICH score, female sex lost significance as predictor of mortality in our population. **Conclusions:** We found lower ICH incidence in women than in men. However, in women there was a higher 1-year case-fatality, which was likely related to their older age at ICH onset and higher clinical severity on presentation compared to men. Identification of factors that can explain the reported differences is important to develop targeted interventions to prevent and manage the disease.

P17

Employment of osteoblast-derived extracellular vesicles for the delivery of methotrexate and doxorubicin to MNNG/HOS osteosarcoma cells

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Settore Scientifico Disciplinare (SSD): BIO/17

Osteosarcoma is the primary bone cancer that most commonly occurs and causes bone cancer-related deaths in children. The main approved treatment is based on the administration of high dose chemotherapy, with subsequent whole body side effects related to non-target specificity of the treatment. Currently, synthetics liposomes and nanoparticles are employed as vehicle for drug administration, in order to reduce related side effects and increase specificity. We employed Extracellular Vesicles (EVs) isolated from the human fetal osteoblasts (FOB) cell line as cargo to deliver methotrexate (MTX) and doxorubicin (DXR) to MNNG/HOS human osteosarcoma cells. We firstly evaluated MTX loaded by passive diffusion in FOB-EVs using High- Pressure Liquid Chromatography (HPLC). Next, we treated MNNG/HOS cells with FOB-EVs loaded with MTX, which showed a significant reduction of cells metabolic activity (evaluated by MTT assay), proliferation (evaluated by EdU assay) and number (evaluated by DAPI staining), compared to tumour cells treated with empty FOB-EVs or to untreated cells. Moreover, MNNG/HOS cells treated with FOB-EVs loaded with MTX showed a significant increase of apoptosis (evaluated by flow cytofluorimetry Annexin V assay), compared to tumour cells treated with empty FOB-EVs or to untreated cells. Finally, the effect exerted by FOB-EVs loaded with MTX was comparable to the 0.5 µM dose used of free-MTX selected after IC50 evaluation on MNNG/HOS. Further experiments will be conducted to assess the effect of FOB-EVs loaded with MTX on MNNG/HOS cells organoids. On the other side, preliminary experiments of MNNG/HOS cells treated with DXR passive loaded FOB-EVs did not show any reduction of cells metabolic activity. For that reason, we used electroporation to actively load DXR in FOB-EVs. Next, we treated MNNG/HOS cells with FOB-EVs loaded with DXR, which showed a significant reduction of cells metabolic activity, compared to tumour cells treated with empty FOB-EVs or to untreated cells. Further experiments will be conducted to assess the effect of FOB-EVs loaded with DXR on MNNG/HOS cells proliferation, number, and apoptosis.

P18

Structural Validation of Objective Perspective Taking Test and Santa Barbara Questionaire using Explorative Graph Analysis: Two Measures for Autism

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Visual-spatial skills are fundamental to interaction with the environment, as they are involved in many activities of our daily lives, such as navigating the environment, recognizing, and manipulating objects, and even remembering places. People with autism spectrum disorder (ASD) have peculiar characteristics related to aspects of atypical perceptual processing, particularly complex visual stimuli and visuoperceptual skills. Several tests assessing these abilities exist in the literature, including the Perspective Taking Test (OPT) and the Santa Barbara Sense-of-Direction Scale (SBSOD). The present study aims to validate the OPT and SBSOD tests in the Italian context; to compare spatial orientation skills between ASD and typical developmental (TD) subjects. A total of 254 subjects including 111 males (M=21.86, SD=2.83) and 143 females (M=21.27, SD=2.03), participated in the validation of the measures. An Explorative Graph Analysis (EGA) was performed to assess the structural validity of the instruments. Subsequently, the measures were administered to 13 young adults with ASD (Mean Age=23.46, SD=8.18, Mean age education=12.77, SD=2.65) comparing scores with 13 young adults with TD (Mean Age=24.31, SD=5.96; Mean age of education=14.00, SD=1.95) paired by gender, age, and schooling, to assess the clinical validity of the measures. The results show that the optimal structure for OPT is given by a single dimension investigating Perspective Taking ability (α=0.84), showing good fit indices (CFI=0.95, RMSEA=0.07, GFI=0.96 and Chisq/df= 2.5). Regarding SBSOD, the analyses show that the test consists of two dimensions, Spatial Visualization, and Spatial Orientation, and shows good internal consistency (α=0.82 respectively α=0.70) and good fit indices (CFI=0.88; RMSEA=0.07; GFI=0.97; Chisq/df=2.3). The results show that the ASD group has greater difficulty in the OPT task than the TD group (U=45, p=0.04), while no statistically significant differences emerged in the SBSOD test between ASD and TD. The results indicate that OPT might be a prerequisite for more complex skills such as perspective taking of another person, which is impaired in ASD persons. The fallout of assessing this skill is on the rehabilitation level, it could be useful to train visuospatial skills in activities of daily living, such as driving, and social-type skills such as mentalization, which are impaired in ASD people.

P19

Transoral laser microsurgery (tml) vs radiotherapy (rt) in early glottic carcinomas

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Settore Scientifico Disciplinare (SSD): MED/18

Laryngeal carcinoma is the most common form of cancer of the upper aero-digestive tract worldwide, accounting for 2.5% of all malignancies in men and 0.5% in women. The larynx serves crucial functions including maintaining airway openess, safeguarding the tracheo-bronchial tree from aspiration, and facilitating phonation. These functions can be compromised depending on the location, size, and depth of tumor invasion. Early glottic carcinomas can be treated using: transoral laser surgery (TLM) which offers rapid treatment, repeatable surgery, low morbility, good patient compliance, short hospitalization, and limited costs; radiotherapy (RT) which provides good control rates, but involves prolonged treatment time, mucosal radiation damage, and long-term side effects such as xerostomia, or open partial horizontallaryngectomy (OPHL). The choice of treatment should be based on the cure rate, larynx preservation rate, functional preservation, and the whole impact. From October 2022 to January 2023 we enrolled 8 patients over 18 years-old affected by glottic squamous cell carcinoma pT1 and pT2 who were previously untreated: 6 male patients (aged between 49 and 87) were included, of which 4 were treated with TLM (group 1), and 2 with RT (group 2). Our double-cohort prospective study was based on subjective patient evaluation because the patients' own assessment takes into account their expectations regarding treatment outcomes, with a more favorable outlook on functional outcomes like voice and swallowing.

P20

Angle-Angle Diagrams in the Assessment of Locomotion in Persons with Multiple Sclerosis: A Preliminary Study

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Settore Scientifico Disciplinare (SSD): M-EDF 02

Gait analysis is clinically relevant in persons with multiple sclerosis (PwMS) and consists of several joint angular displacement–time relationships and spatiotemporal parameters. However, it lacks representation by means of diagrams in which knee-angle/hip-angle and knee- angle/ankle-angle variations are plotted against each other at the same points of time. Three- dimensional kinematic analysis was performed on 20 subjects (10 PwMS/10 healthy controls, HCs), and the knee-angle/hip-angle and knee-angle/ankle-angle diagrams of both lower limbs were determined in the sagittal plane while walking on a motorized treadmill. The area (a quantifier of range of motion) and the perimeter (a quantifier of coordination) of angle-angle diagram loops were calculated. PwMS showed reduced knee-angle/ankle-angle loops compared to HCs ($p < 0.05$), whereas the hip-angle/ankle-angle loops between the PwMS and HCs were not significant ($p > 0.05$). Similarly, the activation of leg muscles showed significant differences between PwMS and HCs (p ranged from 0.05 to 0.001). The results indicate that the proposed knee-angle/hip-angle diagram is feasible and could be applied as a reliable tool in future studies aimed at assessing the acute and long-term effects of specific exercise programmes and/or pharmacological treatment in PwMS.

P21

Study of a protocol for the development of organoids derived from dental pulp stem cells

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Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): BIO/13

Bi-dimensional cell cultures are primary research tools widely used. However, lacking of both tissue architecture and complexity, they cannot fully mimic in vivo biological processes. Recent advances in organoid technology have addressed these limitations and are revolutionizing the tools available for in vitro culture. 3D cell culture systems have great potential to fill the gap between cellular and animal models in the studies of infectious diseases, genetic disorders, cancer, drug development, therapies, and tissue and organ repair and replacement. In particular, a novel source of mesenchymal stem cells called dental pulp-derived stem cells (DPSCs) attracted the scientific community. Derived from the neural crest, DPSCs are physiologically involved in dentin homeostasis, but experimental evidence suggests that DPSCs can also differentiate into non-mesodermal cells, including neuronal, epithelial, endothelial, hepatocyte, kidney and lung cells. The aim of this work is to compare the ability of DPSCs to differentiate into odontoblasts when cultured as a monolayer or by developing a 3D cell culture system.

In the preliminary phase of the project, we differentiated DPSCs on a monolayer and subjected them to odontogenic differentiation. We performed colorimetric analysis with alizarin red at different time points, which revealed calcium phosphate deposits typical of odontoblasts. We then carried out a gene analysis using RT-PCR and observed an increase in the expression of differentiation-specific genes and a decrease in the expression of the stemness gene. These results were confirmed by immunofluorescence analysis, which showed a similar trend to the RT-PCR: expression of differentiation markers increase and stemness markers decrease were observed. Also immunofluorescence analysis of the organoids showed an increase in the expression of differentiation markers and a decrease in stemness markers, confirming the results obtained in monolayer cultures. With regard to RT-PCR analysis, we are attempting to standardize a protocol for the extraction of nucleic acids from organoids in order to be able to assess gene expression in this context too. Finally, given the good results obtained with our 3D model, the future aim is to use it for translational studies in the field of dentistry.

P22

Yes we us. The role of ultrasound in the kidney transplant patient in the early recognition of surgical complications

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Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): MED/18

Case report
Stenosis of the iliac segment proximal to the transplant renal artery and transplant renal artery stenosis, are two significant causes of graft dysfunction and hypertension. We present the case of a 30-year-old female living kidney transplant recipient with end-stage renal disease due to urethral stricture. She underwent successful kidney transplantation from her mother. The allograft exhibited primary graft function initially. On the second postoperative day, the patient experienced a sharp decrease in urine output, accompanied by abdominal pain and bowel obstruction. A non-enhanced abdominal CT scan revealed dehiscence of the vesical uretero anastomosis. Surgical intervention was performed to address the uretero-vesical anastomosis, and during the procedure, it became necessary to repackage the arterial anastomosis using PTFE patches. Four months post-transplant, the patient presented with elevated serum creatinine levels, claudication (Leriche Fontaine IIa), and arterial hypertension. Duplex Doppler sonography (ECD) revealed a pathologic flow profile in the iliac artery proximal and distal to the graft, peak systolic velocity (PSV) greater than 400 cm/second in the renal artery, and intrarenal tardus parvus waveforms. CT angiography demonstrated hemodynamically significant stenosis of the external iliac artery proximal to the arterial anastomosis. Subsequent angiography and percutaneous transluminal angioplasty (PTA) of the external iliac artery were performed, resulting in successful vessel caliber reacquisition. Follow-ups indicated a deterioration in renal function and ECD parameters, with a CT scan revealing a recurrence of stenosis in the external iliac artery. Consequently, the patient underwent PTA and hip-axis stenting (open mesh stenting) with successful reacquisition of vessel flow caliber. Serial ECD checks demonstrated excellent flow in the external iliac artery, good vascularization of the renal graft, and substantial recovery of renal function. Duplex Doppler sonography emerges as an excellent method for screening patients suspected of having transplant renal artery stenosis. It aids in identifying individuals who should undergo digital subtraction arteriography. Crucial parameters for evaluation include peak systolic velocity in the external iliac and renal arteries, acceleration time, acceleration in the intrarenal arteries, acceleration time in the renal artery, and resistance index. Early diagnosis facilitated by ECD allows for timely and decisive intervention, contributing to improved patient outcomes.

P23 – SELECTED ORAL COMMUNICATION

The use of ADOS-2 in early diagnosis for Autism Spectrum Disorder: a Network Analysis and Predictive Risk Assessment study

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Settore Scientifico Disciplinare (SSD): MED/01- MPSI/03

In recent years, awareness of the importance of early diagnosis and the development and revision of screening and diagnostic tools for autism spectrum disorder (ASD) has increased. Currently, the tool considered the gold standard for autism diagnosis is the Autism Diagnostic Observation Schedule, Second Edition (ADOS-2), which allows children at risk to be identified as early as 12 months of age. However, behaviour and social communication patterns may vary during the second year of children's lives.
In this study, we examine two modules of the ADOS-2 that can be administered to children aged 12-30 months and older, namely the Toddler module and Module 1. We performed a network analysis to explore and explain how the ADOS-2 items (Toddler Module and Module 1), which describe typical symptoms of autism, relate to each other and how these relationships may differ between ASD and No ASD children. Exploring how the symptoms investigated by ADOS-2 relate to each other may offer important information on symptom manifestation, early diagnosis and differential diagnosis. A second objective of the study is to explore the predictive agreement between the Toddler module and Module 1, and the marginal diagnostic predictive effect of the two constructs shared by the two modules, namely Social Affect and Restricted and Repetitive Behaviour.

P24

Expression of the endocannabinoid system in a cellular model of atopic dermatitis

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Settore Scientifico Disciplinare (SSD): MED/35

Introduction. The pathogenesis of atopic dermatitis (AD) is multifactorial and includes a combination of immunological alterations and epidermal barrier dysfunction. The endocannabinoid system (ECS) is a pathway including receptors (CB1, CB2, GPR55, TRPV1, PPARalpha, PPARgamma, PPARdelta), their endogenous ligands (anandamide, 2-arachidonoylglycerol) and synthesis and degradation enzymes (NAPE -PLD, FAAH, DAGLalfa, DAGLbeta, MAGL). The ECS has recently been implicated in the regulation of several skin processes, as keratinocyte proliferation and immunotolerance. In this study, we evaluated the expression of ECS pathway members in a cellular model of AD.
Methods. To build the cellular model of AD, HaCaT cells were cultured with DMEM and 10% FBS. HaCaT maturation was achieved by adding 1.8 mM CaCl2 for 7 days. On the last day, cells were treated with IL-4, IL-13, and INFgamma at 30 ng/mL each for 24 h for differentiation into the DA phenotype. We evaluated the expression of all receptors and enzymes of the ECS system both at the protein level (by Western Blot) and at the mRNA level (by RT-Real Time PCR with TaqMan probes).
Results. We observed a significant reduction in PPARα receptor mRNA and protein levels ($p=0.008$, $p=0.002$) in the AD model compared to the control and a 1.7-fold increase in PPARgamma receptor mRNA ($p=0.016$). Regarding ECS enzymes, AD showed significantly lower mRNA levels of FAAH ($p = 0.006$) and NAPE-PLD ($p = 0.008$), compared to HaCaT cells, with a 1-, 4-fold and 2-fold decrease , respectively, At the protein level, the MAGL enzyme was significantly increased compared to the control ($p=0.047$). However, we did not find significant differences in protein and mRNA expression for DAGLα and DAGLβ in AD compared to control ($p=0.264$ and $p=0.150$ for mRNA; $p=0.100$; and $p=0.363$ for protein, respectively). Regarding the CB2 receptor, molecular experiments demostrated and confirmed the presence of two isoforms of CB2, one of exclusively expressed in AD and not in control HaCaT ($p<0.001$).
Conclusions. Our data identified the modulation of CB2, PPARalpha and PPARgamma receptors, and FAAH, NAPE-PLD and MAGL enzymes in AD. We found the presence of a new splicing variant in the AD model, which needs to be sequenced.

P25 – SELECTED ORAL COMMUNICATION

Kidney transplant from living donor: study of psychological aspects from pre to post intervention

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Settore Scientifico Disciplinare (SSD): MED-18, M-PSI-01

Introduction: Kidney transplantation represents the treatment of choice for most patients affected from end-stage renal disease; compared to dyalisis it increases life expectancy and quality of life. Transplantation can be done from a deceased or living donor. The first data relating to the study of some of the most important psychic symptomatological dimensions of the recipients of living donor transplants will be presented here. **Materials and methods:** Use of tests to explore the psychological profile and quality of life both in the pre-transplant and post- transplant phases: SCL-90 R and SF-36. The sample studied to date pre and post surgery is composed of 7 subjects who underwent kidney transplantation exclusively from a living donor, the data is studied through the T test. **Results:** It is noted that post-intervention the symptomatic dimensions of the SCL 90 R and all the scales of the SF36 improve, highlighting how the transplant promotes psychological well-being and the feeling of perceiving a good quality of life. Only one dimension needs attention and that is the Sleep scale where a slight worsening is detected. Further studies will investigate this aspect. From a statistical point of view, a significant p value emerges for the PSY scale. **Conclusions**: living transplantation, given the data, which do not appear to be small since this type of transplant is rare for numerous clinical and psychological reasons, seems to improve the quality of life. At the end of the study foreseen by the doctoral project, it will be verified whether this also happens with cadaveric recipients or whether there are differences in impact.

P26

Management of bilateral temporomandibular joint ankylosis using bilateral custom alloplastic temporomandibular joint prosthesis

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Temporomandibular joint ankylosis (TMJA) is a pathological condition characterized by limitation of the opening of the mouth caused primary by trauma but also by infection, inflammatory and congenital diseases. TMJA can be treated through the utilization of autogenous grafts or alloplastic TMJ prostheses. There are two types of alloplastic TMJ prostheses: stock or custom. In the last years custom alloplastic TMJ prostheses are the emerging treatment modality for TMJ ankylosis. Presentation of the case: A 16-year-old male patient presented with a 1 cm interincisal opening for about 2 years and inability to chew and speak. He referred ankylosis appeared after a trauma. The ankylotic masses were of 17,6 mm on the right side and 19,6 mm on the left side. A bilateral removal of the ankylosis and a custom-made prostheses reconstruction was programmed. Both the ramus-condyle unit and the fossa component of the custom alloplastic temporomandibular joint (TMJ) are fabricated using titanium, while the insert between these two parts is composed of ultra-high molecular weight polyethylene. Treatment options for TMJ ankylosis are various. Stock alloplastic TMJ prostheses may not suit all patients due to anatomical variations. Thus, custom alloplastic TMJ prostheses have emerged as the preferred treatment modality for TMJ ankylosis. We also conducted a review of the literature regarding patients who underwent bilateral custom-made prostheses. Custom alloplastic TMJ prostheses are considered an optimal treatment modality for reconstructing the TMJ in adult and young adult patients with TMJ ankylosis. The implantation of custom-made prostheses is not associated with any peri- and postoperative complications.

P27

Survey of the organisational climate within the Conservatory: an intervention research

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Settore Scientifico Disciplinare (SSD): M PSI/01; M PSI/05

Organisational climate is a multidimensional construct that affects personal motivation, work efficiency, commitment and trust in the organisation, and influences safety because it changes risk perception. When conditions of poor organisational well-being occur, phenomena such as low productivity, low levels of motivation and lack of trust become frequent. The main objective of this research-intervention is to promote organisational well-being by analysing employee satisfaction and motivation. In order to do this, the organisational climate of the L'Aquila Music Conservatory will be analysed. Within this context it would therefore be very useful to start training and survey processes among employees, also taking into consideration the complexity of the environment, characterised by direct interaction between teaching staff and users. The key point is communication, which has a positive correlation with many organisational factors such as commitment, performance and job satisfaction. Conversely, a failure in communication leads to detriments such as stress, job dissatisfaction, decreased commitment but also dismissal and this has a negative impact on corporate efficiency. The methodology to be used will be mixed, by using both quantitative tests and questionnaires to assess communication, leadership, work-related stress and motivation, and qualitative methods such as focus groups. The latter encourages interaction between participants and ensures that everyone has the opportunity to express their opinions. Data is recorded by means of detailed notes or audio/video recordings to enable in-depth analysis at a later stage. Questionnaires will be administered as a follow-up and then a review of the intervention will be carried out. The aim of this research is to assess whether targeted interventions promote an increase in group awareness, communication and well-being in the general working environment.

P28

Neuromuscular characteristics of unilateral and bilateral maximal voluntary isometric contractions following anterior cruciate ligament reconstruction

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Despite the advancement of diagnostic surgical techniques in anterior cruciate ligament (ACL) reconstruction and rehabilitation protocols following ACL injury, only half of the athletes return to sports at a competitive level. A major concern is the neuromechanical dysfunction, which occurs with injuries persisting in operated and non-operated legs following ACL rehabilitation. One of the criteria for a safe return to sports participation is based on the maximal voluntary isometric contraction (MVIC) performed unilaterally, comparing the 'healthy knee' and the 'operated knee'. The present study aimed to investigate MVIC in athletes following ACL rehabilitation during open kinetic chain exercises performed unilaterally and bilateral exercises. Twenty subjects participated in the present investigation: 10 male athletes of regional–national level who were previously operated on one knee and received a complete rehabilitation protocol (for 6–9 months) were included in the ACL group (age: 23.4 ± 2.11 years; stature: 182.0 ± 9.9 cm; body mass: 78.6 ± 9.9 kg; body mass index: 23.7 ± 1.9 kg/m2), and 10 healthy male athletes formed the control group (CG: age: 24.0 ± 3.4 years; stature: 180.3 ± 10.7 cm; body mass: 74.9 ± 13.5 kg; body mass index: 22.8 ± 2.7 kg/m2). MVICs synchronised with electromyographic (EMG) activity (vastus lateralis, vastus medialis, rectus femoris and biceps femoris) were performed during unilateral and bilateral exertions. The rate of force development (RFD) and co-activation index (CI) were also calculated. The differences in the MVIC and RFD between the two legs within each group were not significant ($p > 0.05$). Vastus lateralis EMG activity during MVIC and biceps femoris EMG activity during RFD were significantly higher in the operated leg than in the non-operated leg when exertion was performed bilaterally ($p < 0.05$). The CI was higher in the operated leg than that in the non- operated leg when exertion was performed bilaterally ($p < 0.05$). Vice versa, vastus medialis EMG activity during RFD was significantly higher in the right leg than that in the left leg when exertion was performed bilaterally ($p < 0.05$) in the CG. MVICs performed bilaterally represent a reliability modality for highlighting neuromechanical asymmetries.

P29

Development/Optimization of a Real-Time PCR assay based on TaqMan chemistry for the analysis of the TrkAIII splice variant

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Settore Scientifico Disciplinare (SSD): MED04, MED18

The oncogenic alternative TrkAIII splice variant, originally discovered in human NBs and correlated with advanced stage, metastatic and post therapeutic relapse disease, is characterized by NTRK1/TrkA exons 6, 7 and 9 skipping, and has been subsequently detected in association with advanced stage metastatic cutaneous malignant melanomas, Merkel cell carcinomas and more recently in Pituitary neuroendocrine tumors. In NB models, in contrast to fully spliced TrkA, TrkAIII is not expressed at the cell surface but accumulates in intracellular pre-Golgi membranes, mitochondria and centrosomes, within which it exhibits ligand-independent cell-cycle and stress- regulated activation. TrkAIII oncogenic activity (NIH3T3 transformation and promotion of primary and metastatic tumorigenecity in NB models), associates with pro-survival PI3K/Akt signaling, an angiogenic MMP-9/VEGF/Tsp1 expression equilibrium, centrosome amplification and stress-induced metabolic adaptation. Furthermore, in NB models TrkAIII exhibits similar activity to the TrkA fusion oncogene TrkT3, suggesting that alternative TrkAIII splicing represents a potentially more frequent oncogenic alternative to TrkA gene fusions, cancers driven by which exhibit profound long lived responses to approved Trk inhibitors that also inhibit TrkAIII. Therefore, condsidering therapeutic potential of detecting oncogenic alternative TrkAIII mRNA splicing in tumor tissues, this study was designed to develop, optimize and validate a Real-Time PCR (qRT-PCR) TrkAIII mRNA test, with the aim of identifying TrkAIII expressing tumors, as an alternative to Trk-fusion driven tumors, that may also respond to Trk inhibitory therapy. Several TaqMan-based real time RT-PCR tests were designed to evaluate TrkAIII mRNA expression, one of which exhibited optimal amplification efficiency, sensitivity, dynamic range and reproducibility, associated with sensitivity up to 102 copies of TrkAIII, with test conditions optimized by varying annealing/extension temperatures, and DMSO and BSA concentrations. The next phase of development will focus on performance and accuracy of this particular TaqMan TrkAIII mRNA detection test in complex RNAs mixtures from tumor cell lines, and from fresh and formalin-fixed paraffin embedded tumor tissues.

P30

Molecular profiling of primary melanoma and related nodal and visceral metastases

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Settore Scientifico Disciplinare (SSD): MED/35

Melanoma is a highly aggressive tumour with different molecular and clinical traits. Intra-tumor heterogeneity is high due to different mutations in different clones of neoplastic cells. This makes diagnosis and treatment difficult since metastases may arise from different subclones of the primary lesion, spreading and progressing as an heterogeneous disease. To study the molecular alterations driving melanoma progression, we are working on a project exploring the intra-patient heterogeneity of mutational profiles during progression. For this purpose, we are analyzing the molecular alterations in a panel of 18 genes involved in melanoma pathogenesis and progression, including NRAS, CTNNB1, EPHA3, KIT, TERT promoter, STK19, RAC1, BRAF, TACC1, PREX2, PPP6C, ARID2, WNT1, GRIN2A, TP53, NF1, GNA11, and DDX3X in primary melanoma and related metastases. Finally, we want to integrate diagnostic molecular approaches to evaluate BRAF and NRAS mutations in melanoma. Herein, we reported the first phase of the study, consisting in the evaluation of the molecular heterogeneity in melanoma drivers genes, BRAF, NRAS, NF-1 and c-KIT. DNA was extracted from 80 FFPE tissue samples (34 primary melanomas, and 46 metastasis) from 34 patients. For each patient, normal skin tissue was used to normalize the molecular alterations. BRAF, NRAS, NF1, and c-KIT genes were analyzed trough target sequencing with NGS using the Ion Torrent S5 platform and normalized with corresponding healthy skin. Characteristics of patients, primary tumours, and related metastases were considered during analysis. Overall, 44% of primary tumors had BRAF mutations, 29% NRAS, and 15% NF1; only 15% are triple-wildtype. In metastatic tissues, the mutational profiles included 35% of BRAF mutated samples, 24% NRAS, 17% NF1, and 34,8% triple-wildtype. BRAF subtype correlated with melanoma in younger age ($p<0.01$) while the NRAS in older age ($p<0.01$). NF1 subtype to high Breslow thickness. We identified c-Kit mutations in 11,1% of primary tumours and 4,3% of metastases in triple-wildtype patients. We found 14.7% discordance in BRAF, 11.8% in NRAS and no discordance in NF1 between the primary tumour and related metastases. In conclusion, intrapatient heterogeneity of driver oncogenic mutations BRAF/NRAS/NF1 between primary melanoma and related metastases exists, thus supporting the polyclonal model of melanoma progression.

P31

Characterizing the effects of Pyrroloquinolone Quinone (PQQ) on redox status and mitochondrial bioenergetics of Human Trabecular Meshwork (HTM) cells

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The trabecular meshwork (TM) represents an anatomical structure located at the base of the cornea, near the ciliary body, that play a crucial role in maintaining normal intraocular pressure (IOP) through the correct regulation of intraocular fluid flow, thereby contributing to the preservation of visual function. Accordingly, TM cells exhibit high metabolic activity that require functional mitochondria and the maintenance of redox balance. The ever-changing cellular milieu, however, constantly exposes mitochondria to multiple stressors causing impairment of oxidative phosphorylation and generation of oxidative stress (OS). These conditions alter TM cell viability and function, thereby leading to the obstruction of the aqueous outflow, IOP elevation, and the onset of glaucomatous conditions. Besides OS and altered mitochondrial activities, many studies have also reported TGFß2 as significant contributor to the pathologic changes occurring in HTM cells/tissue in glaucoma. Although the efficacy and clinical application of treatment for OS and mitochondrial injury are still limited, the Pyrroloquinoline Quinone (PQQ) may be a promising compound. PQQ is an aromatic heterocyclic anionic orthoquinone that can potentially play a protective role against TM OS by restoring redox balance, promoting bioenergetic recovery, and supporting mitochondrial health. Unlike coenzyme Q10, PQQ is a recently discovered molecule already present in nature in various foods and different tissues. Moreover, it has also been reported that PQQ, in addition to being a potent antioxidant, can stimulate the biogenesis of new mitochondria. Although there are some studies associated with neurodegeneration/neuroprotection, the effect of PQQ on TM has not yet been explored. The project aims to investigate how PQQ may modulate TM cell functionality under normal and oxidative/bioenergetic stress conditions. In addition, using TGF-B2/dexamethasone treated TM cells, the effects of PQQ will be studied in an in vitro model that replicate the glaucomatous TM phenotype. To this end, by using primary human TM cells, ELISA assays, Western blotting and immunofluorescence analyses, and measures by Seahorse XF technology will be performed to investigate the following endpoints: 1) cell viability and proliferation; 2) redox status and antioxidant capacity; 3) mitochondrial function and dynamics, and energy metabolism; 5) protein levels of myocilin and actin stress fibers organization.

P32

A novel method to differentiate retinal ARPE-19 for the study of the response to NGF treatment: preliminary data of gene expression profiling and setup of a potential in vitro model for the study of pressure increase effects

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Settore Scientifico Disciplinare (SSD): MED/46

Background: RPE is an epithelial polarized monolayer, performing crucial functions in retinal cells homeostasis. NGF is a neurotrophic factor, whose biological mechanism in preventing retinal neurodegeneration is not fully understood. Availability of suitable retinal in vitro models is crucial for NGF response analysis. Age and elevated intraocular pressure (IOP) are the two primary risk factors for glaucoma, which is the leading cause of irreversible blindness. However, the synergic contributions of age, elevated IOP and glaucoma are unknown in the retinal pigment epithelium (RPE).
Aim: Setup and verification of a method to differentiate ARPE-19 cells in RPE for the in vitro study of NGF mechanisms of action: analysis of differentially expressed genes in response to NGF and setup of a potential in vitro model for the study of the pressure increase effects.
Methods: ARPE-19 cells were differentiated for one month in DMEM with high glucose/pyruvate and 1% FBS. The expression of differentiation markers was verified using RNA-seq, western blot and real-time PCR. RNA-seq analysis was used to evaluate the response to NGF (200 ng/ml after 30', 24h and 48h) in RPE. The same RPE model was subjected to an in vitro experimental system (Live Flow) aimed at mimicking pressure increase.
Results: ARPE-19 showed non-epithelial morphology when mitotically active while one month-differentiated cells exhibited the typical phenotype of native RPE. We found increased levels of RPE65, RLBP1, RDH10 genes and protein expression. Accordingly, RNA-seq showed 4,510 significant gene with detectable signals (791 up-regulated, 953 down-regulated) in the RPE. Up-regulated genes were associated with visual cycle, phagocytosis, pigment synthesis and cell differentiation. After NGF treatment, we found increased expression of genes associated with Wnt pathway (e.g WNT7A, WNT5B, CTNNBIP1) in RPE. Preliminary Live Flow results showed an mTOR expression increase after NGF treatment while no changes were detected in NRF2 expression.
Conclusion: A new method for differentiating ARPE-19 was described. RNA-seq preliminary data highlight several differentially expressed genes involved in retinal biological processes. Among the latter, NGF treatment induced Wnt-pathway, playing a critical role in RPE development and maintenance. Further experiments are planned to characterize the RPE responses in relation to pressure increase.

P33

The effects of Slow-wave Sleep in emotional memory consolidation

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Settore Scientifico Disciplinare (SSD): M-PSI/02

It is widely accepted that sleep plays a crucial role in memory consolidation. Recent studies have explored the specific role of different stages of sleep in the relationship between emotional memory and changes in physiological activation, which is linked to memory consolidation. In the context of emotional memories, research focused on the role of eye rapid movement (REM) sleep but the role of slow wave sleep (SWS) in this process remains unclear. Given the limited number of studies on this topic, our purpose is to investigate the impact of SWS on emotional memory and the associated emotional reactivity. To this aim, we recently introduced a new technology for manipulating sleep through vibrotactile stimulation in order to induce experimental sleep fragmentation arousing the subjects during sleep, avoiding their awakening. The present study will employ a within-subjects protocol in which fifteen undergraduate students (age range: 18- 30) will undergo two different experimental conditions: a "Fragmentation" night, during which vibrotactile stimulation will be provided during SWS, and a "Control" night, during which the subjects will sleep undisturbed. Subjects' sleep will be monitored via polysomnography with High Density-EEG. Moreover, we will evaluate emotional reactivity using the physiological indices of skin conductance response (SCR) and heart rate deceleration (HRD) during a rating task of emotional pictures in three sessions: participants will rate the emotional valence and arousal of emotional stimuli in the evening before sleep (T0), the following morning (T1), and 48 hours later (T2). The same sessions will also include the administration of procedural memory (finger tapping) and emotional memory (recognition) tasks involving an initial encoding phase at T0, followed by a re-test phase at T1 and T2. We hypothesize finding a reduction in memory consolidation for both neutral and emotional stimuli after a fragmented night of SWS compared to the control condition, while maintaining a better memory performance for emotional stimuli over the neutral ones. This project might not only yield new information about the role of SWS in memory consolidation, but it might also provide a new paradigm for the experimental manipulation of this sleep stage.

P34

The SOS response in bacteria and the LexA transcriptional repressor: withstanding infections by inhibiting bacterial adhesion and biofilm formation

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Ciclo: XXXVII

Settore Scientifico Disciplinare (SSD): BIO/12

Background. Antimicrobial resistance represents one of the biggest risks to human health, as well as cancer and cardiovascular diseases. To counter this phenomenon, the search for new microbial targets, such as the transcriptional repressor LexA, is useful. This protein regulates the *SOS response*, a network comprising 60 genes involved in bacterial pathogenicity phenomena, like the acquisition of resistance, filamentation, and biofilm formation. **Aim of the project.** The project aims to identify potential inhibitors of LexA self-cleavage within approved or investigational boron-containing drugs, through a drug repurposing approach. Another goal is to validate a co-culture test between bacterial and eukaryotic cells, to assess the phenomena involved in bacterial adhesion and biofilm formation. **Methods and Results.** We selected three boron-containing molecules that belong to the benzoxaboroles class; these molecules are characterized by the same structure, with different substituents and pharmacological applications. Through biochemical assays, we determined the IC50 values for the three boronics under investigation, which were: 8.33 mM for 1-hydroxy-3H-2,1-benzoxaborole, 3.88 mM for Tavaborole, 6.68 mM for Crisaborole; corresponding to ([I]:[LexA]) ratio of 672:1, 313:1, and 539:1 respectively. Subsequently, through microbiological assays, we investigated their ability to inhibit filamentation and biofilm formation in E. coli. We estimated the ability of the boron compounds to reduce the filamentation induced by a sub-inhibitory concentration of levofloxacin, by a fluorescent microscope using fluoroshield with DAPI. All compounds show the ability to interfere with this phenomenon in a concentration-dependent way. By a spectrofluorimetric assay, we evaluated the potentiality of the boron compounds to inhibit biofilm formation and eradicate the biofilm previously formed. Two of the selected compounds interfere with the biofilm formation, with the same MBIC value, while none of these can eradicate the biofilm previously formed. **Conclusions.** From the reported data, it appears that the selected benzoxaboroles show the ability to interfere with both bacterial filamentation and biofilm formation in E. coli, phenomena related to the activation of the SOS response, through their interaction with the LexA protein. The next steps will concern the evaluation of these compounds on other microorganisms, both Gram-negative and Gram-positive, especially in the run-up to their clinical relevance.

P35

Phenotype of the first mouse model of Cole Carpenter Syndrome

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Ciclo: XXXVIII

Settore Scientifico Disciplinare (SSD): BIO/17

The Cole Carpenter Syndrome (CCS) is a rare genetic disease displaying an autosomal dominant inheritance with a prevalence <1:1,000,000. It affects mainly the bone, which becomes brittle and fractures many times. Genetically, the CCS is caused by the heterozygous p.Y393C mutation in the P4HB gene encoding for the Protein Disulfide Isomerase A1 (PDIA1). To date there are no studies showing how the CCS-inducing mutation affects the skeletal. Based on this, we generated a mouse model carrying the aminoacidic substitution tyr393cys (CCS mouse) by constitutive knock-in strategy. Gross evaluation revealed no obvious changes in body length and weight of CCS mice. However, the bone phenotype, assessed by µCT in CCS mice showed marked osteopenia compared to the WT counterpart. In addition, the Indentation Distance and the Total Indentation Distance were dramatically increased in CSS femurs compared to WT, indicating a poor bone quality. Interestingly, the metabolic analysis revealed a significant reduction of postprandial blood glucose levels in CCS male mice compared to WT, along with an unchanged glucose and insulin tolerance. This finding could be linked to renal defects found in CSS mice consisting of an extensive tubular vacuolization, along with intracellular protein deposits, intraluminal protein cylinders and glomerular shrinkage, which can lead to an excessive glucose leakage in the urines. Overall, our data indicate the presence of a severe bone phenotype in CCS mice, consistent with the skeletal features of the human disease, and the pathogenic involvement of organs beyond the bone.

P36 – SELECTED ORAL COMMUNICATION

GMP isolation and culture of Human Umbilical Vein Endothelial Cells led to selectively expanded CD34-positive cells

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Ciclo: XXXVI

Settore Scientifico Disciplinare (SSD): BIO/13

Therapeutic angiogenesis is a critical process in repair and regeneration that can be enhanced by perinatal human umbilical vein endothelial cells (HUVEC). The functional activation of quiescent endothelial cells into migrating tip cells is a key event during the sprouting phase of angiogenesis, and CD34-positive HUVECs have been proposed as a critical substitute for. CD34 is considered an important marker in hematopoietic stem cells, but also a critical cell surface marker to identify angiogenic vascular endothelial cells, both in vivo and in vitro. The isolation and expansion of such progenitor cells are critical steps, as well as ex vivo expansion by Good Manufacturing Practice (GMP). So, guidelines have been developed to replace animal-derived material with human material, such as GMP-grade human platelet lysate (hPL).
We are attempting to standardize reagents and procedures to efficiently expand selectively CD34+ HUVEC collected from full-term placentae. As the first step, clinical translation of cell- based approaches requires the replacement of conventional fetal bovine serum (FBS) with human serum replacement (such as hPL).
We performed HUVEC extraction from human umbilical cords, and we cultured the cells for a few days in a basic endothelial medium supplemented with FBS or hPL. Fluorescent-based immunostainings proved characteristic expression for endothelial markers in the cell products but also highlighted an increase in progenitor markers (CD34) in HUVEC cultured with hPL rather than FBS. Functional assays (angiogenesis assay on Matrigel) confirmed the efficiency in forming tubule-like structures in vitro for donors analyzed, and the analysis of segment length and branching index shows that both parameters efficiently improved in primary HUVEC exposed to hPL than standard xeno-conditions.
In addition to freshly isolated HUVECs, we also performed the same assays on cryopreserved and aged cells (P3) obtaining similar results.
Replacement of xeno-condition with GMP-grade reagents is not only effective but apparently resulted in a superior ability to generate expanded migrating CD34+ HUVECs, promoting the activity of cells with a tip cell phenotype. Data from these experimental conditions and potency assays will help to form the basis for isolation and release criteria of HUVEC for clinical uses.

P37 – SELECTED ORAL COMMUNICATION

Evolution of surgical techniques in proctology: preoperative risk assessment with machine learning models

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Ciclo:XXXVI

Settore Scientifico Disciplinare (SSD): MED/18

Machine learning is a component of artificial intelligence; it relies on computer algorithms and data analysis to learn patterns that exceeds the capacity of the human mind to comprehend. It uses statistical methods to infer relationships between predictors and outcomes in large datasets, and it has been successfully applied to predict adverse events in health care settings. In our work, the aim was to investigate the potential role of machine learning (ML) versus classical statistical methods (SM) for the preoperative risk assessment in proctological surgery. We used clinical data from a nationwide audit; we collected anthropometric, clinical, and surgical data and we considered ten predictors to evaluate model-predictive performance. The clinical target was the complication rate evaluated at 30-days follow-up. Logistic regression and three ML techniques were compared. ML models included a Decision Tree, a Support Vector Machine, and a classification Extreme Gradient Boosting (XGB).
The database consisted of 1510 patients affected by Goligher's grade III hemorrhoidal disease who underwent elective surgery. The dataset contained ten prognostic factors; it was randomly splitted into a “train” group for the development and validation of the model, and a “test” group used for making predictions.
Our study found no superiority of the ML using structured pre- and intra-operative variables. In detail, the results showed that among the ML techniques, XGB was the most complex and accurate; however, it was overlapping with SM in terms of balanced accuracy, specificity, and negative predictive value. Regarding the relative importance of the input features, all models agreed in identifying the most important factor, which resulted to be anaesthesia.
In our opinion, for non-complex real-life data such as these, ML techniques should only be employed complementary to SM as exploratory tools of model’s performance: they potentially provide a powerful tool to demonstrate correlations that may be missed by traditional methods, but there are some drawbacks to using them uncritically.

P38

Effectiveness of Transcranial Direct Current Stimulation and Monoclonal Antibodies Acting on the CGRP as a Combined Treatment for Migraine (TACTIC)

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Ciclo:XXXVII

Settore Scientifico Disciplinare (SSD): MED/26

Introduction and aims: Migraine is a recurring headache disorder with an unclear pathophysiology involving the central and peripheral nervous systems. Monoclonal antibodies targeting the calcitonin gene-related pathway (CGRP-MAbs) are designed specifically for migraine, acting peripherally on the trigeminal ganglion. In contrast, neuromodulation techniques like transcranial direct current stimulation (tDCS) act centrally by influencing cortical neuronal firing rates. This study aims to assess whether tDCS, when combined with CGRP-MAbs, effectively reduces migraine frequency, intensity, and medication use. Electroencephalographic (EEG) power changes will be analyzed to demonstrate the biological effects of tDCS.

Methods: Our study is a randomized, double-blind, multicenter, sham-controlled trial including patients with migraine in CGRP-Mabs treatment with residual monthly migraine days (MMD) ≥8. After 5-day of tDCS bilateral occipital cathodal and frontal anodal stimulation (sham/active sessions lasting 20 minutes) Before the first and after the last stimulation we recorded a 64-channels high density EEG. We closely monitored patients for 28 days, assessing changes in monthly migraine days and clinical scales such as MIDAS, HIT6, HADS. We analyzed change in MMDs, clinical scales, and EEG spectral changes in the delta (2-4Hz), theta (5-7Hz), alpha (8-12Hz) and beta-bands (13-30Hz) through two-way mixed-design ANOVAs with Session (baseline vs. follow-up), as within-subjects factor, and Treatment (Sham vs. Active) as between-subject factor.

Results: We included 29 patients (median age 48 IQR 30-56, 92.0% female), 15 in the active session and 14 in the sham group. In the active group, tDCS led to a noteworthy reduction in MMDs (mean difference=4.00, standard error (SE)=1.52, p=0.014). Moreover, a noteworthy enhancement was observed in the HIT6 scale for both the active (p = 0.005) and sham (p = 0.003) groups. tDCS exhibited a reduction in occipital alpha band power and delta power specifically over the motor areas, but only in the active stimulation group (both p<0.05); no differences were found in the sham group.

Conclusion: tDCS, as add-on therapy to CGRP-Mabs, exerts a significant effectiveness limited to MMD reduction associated with changed basal cortical activity on stimulated areas.

P39

Molecular characterization and inhibition of a novel stress-induced mitochondrial protecting role for misfolded TrkAIII in human SH-SY5Y neuroblastoma cells

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Ciclo:XXXVI

Settore Scientifico Disciplinare (SSD): MED/04

The inhibition of stress and drug-resistance mechanisms is a major goal in improving therapeutic responses in cancer. In pediatric neuroblastomas (NBs), resistance mechanisms include alternative TrkAIII splicing of the neurotrophin TrkA receptor, which correlates with advanced stage metastatic disease and post therapeutic relapse. In NB models, TrkAIII exerts oncogenic activity by mechanisms including stress-induced mitochondrial TrkAIII importation and activation. Here, using a variety of inhibitors, we characterize novel participants in this mechanism and evaluate their potential targetability in promoting stress-induced death of TrkAIII expressing SH-SY5Y cells. Using DTT, as an activator and a variety of inhibitors in TrkAIII expressing SH-SY5Y NB cells, we report that this mechanism initiates with stress-induced TrkAIII misfolding and complexing with Grp78 and Ca2+-calmodulin, followed by Grp78, Ca2+-calmodulin, adenosine ribosylating factor (Arf1) and Hsp90-regulated mitochondrial importation. Within mitochondria TrkAIII, cleaved by Omi/HtrA2, is activated by a mechanism involving Akt, the mitochondrial uniporter (MCU) and ROS, in association with mitochondrial PTPase oxidation. We show that: i) entrectinib and lestaurtinib Trk, capivasertib Akt, HA-15 Grp78, geldanamycin Hsp90 and W7 calmodulin inhibitors prevent DTT-induced mitochondrial TrkAIII activation and augment sensitivity to DTT-induced death; ii) KN93 calmodulin/CAMK inhibitor prevents DTT-induced mitochondrial TrkAIII activation but does not enhance sensitivity to DTT-induced cell death; iii) LY294002 PI3K inhibitor does not prevent DTT- induced mitochondrial TrkAIII activation but enhances sensitivity to DTT-induced death, and iv) brefeldin A Arf1, DS1657051 MCU and resveratrol ROS inhibitors prevent DTT-induced mitochondrial TrkAIII activation but enhance resistance to DTT-induced death. Overall, the data characterize misfolded N-glycosylated TrkAIII as a novel stress-regulated calmodulin-dependent mitochondrial kinase and pro-survival UPR component involved in mitigating the cytotoxicity of stress disrupted mitochondrial Ca2+ homeostasis, and characterize TrkAIII, Grp78, calmodulin, Hsp90, PI3K and Akt as potentially targetable, and CAMK, Arf1, MCU and ROS as non-targetable participants in this stress and drug resistance mechanism.

P40

Characterization of the endocannabinoid system in organ of Corti UB/OC1 cells and its modulation upon cisplatin-induced ototoxicity

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Ciclo:XXXVIII

Settore Scientifico Disciplinare (SSD): BIO/10

Endocannabinoids (eCBs) are a class of endogeneous bioactive lipids playing a pivotal role in several organs including the cochlea. Cisplatin is a widely used and potent chemotherapeutic to treat several human tumors; however, it has a pleteora of adverse effects including ototoxicity in adults and children. To date, there are no approved therapeutics to counteract these adverse effects, and only a very few studies have interrogated the role of eCB's in cochlear patho-physiology. Here, we investigated for the first time in an organ of Corti cell line (UB/OC1): (i) the expression of the primary eCB system components (i.e., receptor targets and metabolic enzymes) at the protein level; (ii) the possible effects on the eCB system in response to cisplatin. By Western blotting, we found the expression of the following components in the cell line: receptors - cannabinoid receptors 1 and 2 (CB1, CB2), transient receptor potential vanilloid 1 (TRPV1), peroxisome proliferator-activated receptor δ (PPARδ); and enzymes - diacylglycerol lipase α/β (DAGL α/β) and fatty acid amide hydrolase (FAAH). Instead, we failed to demonstrate the expression of the following components: receptors - G-protein coupled receptor 55 (GPR55), PPARα, PPARγ; and enzymes - N-acyl phosphatidylethanolamines-specific phospholipase D (NAPE-PLD), monoacylglycerol lipase (MAGL). We then established a model of cisplatin-induced ototoxicity by treating UB/OC1 cells with a range of concentrations (from 2.5 µM to 50 µM) of cisplatin and determining its IC50 (30 µM) via MTT cell viability assay. Interestingly, we found that the CB2 receptor and the DAGL β enzyme were significantly reduced in cisplatin-treated samples compared to vehicle-treated controls (CB2 by ~22%, p=0.0058; DAGL β by ~40%, p=0.0041), while no differences were observed in the other eCB system elements. Overall, our findings demonstrate for the first time the presence of key eCB system components in UB/OC1 cells and document the selective involvement of CB2 and DAGL β in cisplatin- mediated ototoxicity.

P41

Music-App Based and Music technology: the development of structured play-musical activities for the promotion and acquisition of linguistic, prosodic and grammatical skills

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Ciclo: XXXIX

Settore Scientifico Disciplinare (SSD): M-PSI/04

The "communicative musicality" of early language forms seems to holistically trace the communicative process in its rhythmic-sound, gestural, emotional and linguistic aspects. Moreover, high children's responsiveness to rhythmic components of language turns out to be predictive for the development of expressive components, such as grammatical ability and phonological awareness. Previous research has supported the positive relationship between music and language, showing early language outcomes in children formally or informally exposed to musical activities found in today's leading interactive Apps. The present project aims to promote the creation of interactive Music- Apps for children that could facilitate and support early language development , mainly in terms of phonetic awareness, prosody, grammar, and meta-phonology. Practical implications and future research directions are considered.

P42– SELECTED ORAL COMMUNICATION

High Intensity Focused Ultrasound (HIFU) treatment: evaluation of long-term cognitive outcomes

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Settore Scientifico Disciplinare (SSD): MED/26

Introduction: Magnetic resonance imaging-guided focused ultrasound (MRgFUS) is a recent thermal ablation treatment for Essential Tremor (ET) and Parkinson Disease (PD) related tremor. Data about cognitive changes are controversial.
Objective: Aim of this study was to confirm the long-term cognitive safety following the MRgFUS treatment.
Methods: In this prospective study, patients consecutively undergoing MRgFUS were assessed through a comprehensive neuropsychological and behavioral battery before, six months and 1 year following the treatment. Data were analyzed with paired T-Test or Wilcoxon signed-rank tests and verified with Bonferroni's correction. A p value <0.003 was considered statistically significant.
Results: Fifty patients (male 76%; mean age ± SD 69.0 ± 8.56; mean disease duration ± SD 12.13 ± 12.59) with ET (n=28) and PD (n=22) were included. A significant improvement was detected at six months after the treatment in anxiety feeling (HAM-A 5.66 ± 5.02 Vs 2.70 ± 4.09, p < 0.001), memory (RAVLT: Immediately re-enactment 31.76 ± 7.60 Vs 35.51 ± 8.38; p = < 0.001; RAVLT: Deferred re- enactment 5.57 ± 2.75 Vs 7.03 ± 3.85; p = < 0.001) frontal functions (14.24±3.04 Vs 15.24±2.38; p= 0.003), and in the quality of life [QUEST (35.00±12.08 Vs 8, 93±9.86, p= < 0.001), PDQ-8 (7.86±3.10 vs.3.10±1.52, p=<0.001). A similar improvement in behavioral assessment (HAM-A 5.66±5.02 Vs 2.69±3.76, p=<0.001; BDI-II 3.74±3.80 Vs 1.80±2.78, p= 0.001), memory domains (RAVLT: Immediately re-enactment 31.76±7.60 Vs 35.38±7.72, p= 0.001; RAVLT: Deferred re-enactment 5.57±2 .75 Vs 6.41±2.48), frontal functions (14.24±3.04 Vs 15.16±2.74) and in the quality of life [QUEST 35.00±12.08 Vs 9.03±10 .64, p=<0.001; PDQ-8 7.86±3.10 vs. 3.09±2.29, p= <0.001)] was also detected 1 year following the procedure.
Conclusion: Our study takes a step toward in endorsing the long-term neuropsychological safety of unilateral MRgFUS and encourage the implementation of staged bilateral treatments.

P43

The TrkAIII splice variant oncoprotein enhances PD-L1 expression in human SH-SY5Y neuroblastoma cells.

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Ciclo: XXXVII

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Neurotrophin receptor TrkA alternative splicing, resulting in TrkAIII variant expression, positively correlates with post-therapeutic relapse and advanced stage metastatic disease in pediatric neuroblastomas (NBs), MCPyV positive Merkel cell carcinomas and cutaneous malignant melanomas. TrkAIII is characterized by exons 6 and 7 skipping, which encode regulatory domains, the absence of which result in intracellular accumulation, and ligand- independent cell cycle and stress-regulated activation. TrkAIII activation signals through PI3K- AKT but not Ras/MAPK, exhibits oncogenic activity in NB and melanoma models, and enhances both stress and chemotherapeutic resistance. Immune checkpoint inhibition mediated by programmed death ligand PD-L1 and its receptor PD-1 plays a significant role in NB pathogenesis but the regulation of this important immune-evasion pathway is poorly understood. Here, we investigated whether TrkAIII regulates PD-L1 expression and function in an SH-SY5Y NB cell model. Real Time qPCR and RT-PCR comparisons of PD-L1 mRNA expression, and Western blot and indirect immunofluorescence comparisons of PD-L1 protein expression were made in untreated control SH-SY5Y , untreated and NGF-treated TrkA SH-SY5Y and TrkAIII SH-SY5Y transfectants, and following treatment with entrectinib Trk, LY-294002 PI3K and PD98059 MEK inhibitors. PD-L1 function was assayed by IL-2 ELISA in co-cultures of activated Jurkat and control, TrkA and TrkAIII SH-SY5Y transfectants. Constitutive PD-L1 mRNA and protein expression in untreated control and TrkA SH-SY5Y transfectants was augmented in by NGF in TrkA SH-SY5Y cells, was significantly higher in TrkAIII SH-SY5Y transfectants, and was reduced in both NGF-treated TrkA and TrkAIII SH-SY5Y transfectants by entrectinib and LY-294002. Reduced Jurkat IL-2 production in Jurkat / TrkAIII SH-SY5Y co- cultures was enhanced by PDL1 neutralizing antibody, confirming PD-L1 function. The data confirm that both TrkAIII and NGF-activated TrkA promote PD-L1 expression in SH-SY5Y NB cells via PI3K, supporting a rational for combining clinically approved Trk and PD1/PD-L1 inhibitors in TrkAIII expressing NB.

P44

Endocannabinoid Signaling in Alzheimer's Disease: a Novel Target for Mechanistic Understanding and Potential Therapeutics

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Settore Scientifico Disciplinare (SSD): BIO/10

The pathogenesis of Alzheimer's disease (AD) remains poorly understood, yet the progressive dysfunctional accumulation of neurotoxic amyloid β (Aβ) peptides remains the main explanation for it. In recent years, chronic inflammation has been identified as an additional player in AD onset and progression. Prolonged and continuous activation of microglia - the main immune cells of the brain - can damage neurons by causing immune cells to acquire an exacerbated pro-inflammatory state. This dysfunctional phenotype can aggravate AD by releasing pro-inflammatory cytokines as well as other inflammatory mediators. Among these, endocannabinoids (eCBs) are bioactive lipids that increase or decrease distinct immune functions. Specifically, microglia express an array of receptors and metabolic enzymes (collectively referred to as the "eCB system") that control the immune functions of eCBs. In particular, *N*-arachidonoylethanolamine (AEA) and 2-arachidonoylglycerol (2- AG) - the two most active eCBs - are increasingly recognized for their essential roles in regulating microglial activity, under both normal and AD-associated conditions. We have recently discovered that continuous exposure to Aβ during embryonic development significantly alters the signaling of 2-AG in primary microglia (AD-like microglia). This alteration leads to an increase in the expression of cannabinoid receptor 2, which is typically associated with an activated proinflammatory phenotype. Consistently, when exposed to an inflammatory stimulus, AD-like microglia showed an exacerbated production of nitric oxide. Therefore, we further investigated the immunological and physiological aspects of Aβ-exposed microglia following prolonged and continuous inflammatory stimulation, and found significant changes in oxidative stress, as well as in senescence and dystrophic markers.

P45

Impact of membrane dynamics and molecular sequestration on beta lactam activity and binding upon exposure in MDR Gram negative clinical strains

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Settore Scientifico Disciplinare (SSD): BIO/12

Antimicrobial resistance is now recognized as both an economic and life hurdling burden, with increasing costs on wealth and lifes. Among critical pathogens, Gram Negative bacteria account as the most threatening. While novel molecules are currently available, the interplay between their efficacy and molecular mechanisms is far from being clarified. Indeed, enzymatic expression coupled to membrane permeability and efflux pump account for major determinants in Gram Negative bacteria. However, Outer memebrane vesicles (OMV) may also concur to further complexity, as they may sequester and concentrate eznymes and molecules, in order to extrude and/or degrade antibiotics away from targets (that is, Penicillin Binding Proteins). The aim of this study is to understand OMV dynamics in clinically relevant pathogens (KPC producing *K. pneumoniae* isolates, AmpC overexpressing *P. aeuginosa*, NDM producing Enterobacterales-*E. coli* and *K. pneumoniae*) undergoing carbapenem- or cefalosporin-based therapy and assess: 1) OMV production and content and if beta lactamases degrade the beta lactam content; 2) Verify PBP occupancy in intact cells upon OMV production compared to untreated controls; 3) Assess mutations of beta lactamses and hydrolysis/inhibition of novel beta lactam; 4) beta lactamases combinations and if their efficacy is impacted by OMV sequestration. **Materials and Methods:** Clinically relevant strains will be characterized and parental strains will be treated with antibiotics for 24 hrs. Supernatant will be collected and content will be assessed for OMV. OMV contents will be analyzed for enzymes and functional assays will be performed in order to determine hydrolysis rate of enzymes. Occupancy of PBPs and PBP quantification in intact cells will also be determined and the impact of OMV sequestration will be assessed for each pathogen and molecule combination. Beta lactamase efficiency in intact cells will also be assessed in order to infer the underlying leading mechanism of resistance. **Expected results:** the impact of OMV sequestration is expected to determine increased rate of hydrolysis of beta lactams and/or concomitant sequestration upon beta lactam exposure. The effect on PBP occupancy should determine the activity of the molecules on the pathogen. Other mechanisms such as overexpression of beta lactamases and porin mutations leading to permeability will also be considered and assessed.

P46

A workplace physical activity intervention to increase well-being in the academic community: the 2023/24 project Ateneo in Movimento

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Ciclo: XXXIX

Settore Scientifico Disciplinare (SSD): M-EDF/02

Numerous studies highlighted the importance of engaging in physical activity (PA) in the workplace as a strategy for reducing the sedentary nature of work, carrying on primary and secondary prevention, enhancing psycho-physical well-being, reducing stress, and improving employee productivity, as outlined by the WHO 2020 guidelines. Ateneo In Movimento (AiM) is a project that originated in 2018 as part of the University Strategic Plan, aiming to increase the psycho-physical well-being of the University of L'Aquila employees in the workplace through PA. This project involves the participation of all academic components, including professors, researchers, students, and staff. About 100 subjects voluntarily participate in the project, divided into an intervention group (IG, n=50) and a control group (CG, n=50). Intervention includes a functional circuit training composed of 8 exercise stations, with a work-rest ratio of 1:1 (30 seconds) repeated for 3 rounds, twice a week for 22 weeks. Additionally, one mobility and posture session per week is provided. To monitor if any changes occur during and at the end of the intervention, three evaluations will be conducted, at the beginning (T0), after 11 weeks (T1), and after 22 weeks (T2). The physical fitness assessment will be conducted through the ALPHA Fit Test Battery. Furthermore, IPAQ for daily physical activity level, PSQI for sleep quality assessment, WAI for workplace performance, and STAI Y1 and Y2 for state and trait anxiety will be administered. All these data are collected through a computerized process that automatically creates a database. It is expected that at the end of the intervention IG will report greater levels in terms of physical efficiency, such as muscular strength, cardiorespiratory fitness, body composition, and psychological well-being such as reduced stress and anxiety, which could be related to an increase in work productivity and overall well-being.
The AiM project aims to contribute to the field of adapted physical activity, emphasizing the importance of practicing physical activity in the workplace as a fundamental strategy to reduce and contrast sedentary behaviors, counteract non-communicable chronic diseases, and create a more favourable working environment.

P47

NF-κB axis in the pathology of glaucoma

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Ciclo: XXXIX

Settore Scientifico Disciplinare (SSD): MED/46

Background: Glaucoma is a chronic optic neuropathy considered a major cause of irreversible vision loss. Glaucoma is characterized by retinal ganglion cells (RGCs) loss, axonal degeneration, and lamina cribrosa remodeling. Among the various factors that cause RGCs death (e.g aging, ER stress, neuroinflammation, mitochondrial dysfunction, elevated intraocular pressure (IOP), ocular biomechanics and low ocular perfusion pressure) IOP is the only modifiable ones. In the pathogenesis of glaucoma, neuroinflammation appears to be associated with overstimulation of the NF-κB axis. However, many questions still need to be clarified to demonstrate which transcriptional targets of NF-κB are involved in glaucomatous pathology.
Aims: In order to investigate the role of NF-κB in the glaucoma pathogenesis we organized our project in:
Task 1. Development and validation of an in vitro and potentially in vivo glaucoma models;
Task 2. Molecular characterization of NF-κB pathway in the established glaucoma models; Task 3. NGS analysis.
Results: First data were obtained by culturing a panel of retinal cell lines widely used as model to study retinal pathophysiology: the retinal Müller glial cell line (rMC1), the photoreceptor precursor cell line (R28) and the retinal pigment epithelium cell line (ARPE19). We performed total protein extraction and cell fractionation (nuclei vs cytoplasm), to study various cellular events (eg. protein translocation). Western blotting analysis was performed to analyze the expression of NF-κB pathway (e.g., p65, pp65, IκBα, pIκBα).
Future prospects: The goal of this project is to clarify the role of NF-κB axis in glaucoma pathology by using 2D and 3D models as well as in vivo model. The high IOP will be simulated in vitro by using live Flow technology, a millifluidic bioreactor able to increase idrostratic pressure in vitro . The analysis will be performed under basal conditions and under pressure. Furthermore, next generation sequencing analyses will be conducted in order to elucidate the role of NF-κB in the disease thus contributing to the identification of potential therapeutic targets aimed at counteracting glaucoma-associated damage.

P48

Understanding the role of REM sleep continuity in emotional memory and emotional reactivity through a new methodological approach

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Settore Scientifico Disciplinare (SSD): M-PSI/02

Background: Several evidence supports the role of REM sleep in emotional processing and consolidation of emotional memories. Besides REM duration, it has been recently proposed that REM sleep continuity supports amygdala adaptation to emotional events in subjects with a wide range of insomnia severity. However, sleep features in clinical populations include other macrostructural alterations that limit the generalizability of the findings. Our study aims to understand the role of REM sleep fragmentation in the psychophysiological reactivity to emotional stimuli, experimentally manipulating the continuity of REM sleep in healthy individuals while keeping other sleep characteristics unchanged. **Methods:** Each participant (n=13) underwent two experimental conditions (Fragmentation/Control) consisting of: 1) Stimuli encoding and assessment of emotional Memory/Reactivity (eMR) in the evening; 2) nocturnal polysomnography with/without REM fragmentation; 3) eMR assessment in the morning; 4) follow-up assessment 48hrs later. We manipulated REM sleep continuity in the Fragmentation condition by inducing EEG arousals via vibrotactile stimulations and evaluated the effects on eMR for negative/neutral stimuli by behavioural (Old/New), self-report (SAM), and physiological (SCR, HRD) measures. **Results:** During the Fragmentation night, REM sleep fragmentation index significantly increased, and REM continuity and duration decreased, without changes in total sleep time and awakenings. Preliminary analyses highlighted significant effects of REM sleep fragmentation on physiological measures, but not on behavioural and self-reported ones. SCR amplitude and number decreased overnight in both conditions, but the response's flattening was significantly greater in the Fragmentation one. SCR increased after 48hrs in the Fragmentation condition, while it tended to decrease in the Control condition. HRD showed an overnight reduction and then remained stable at the follow-up in the Control condition, while no variations were detected in the Fragmentation condition. Explorative correlation analyses support a link between REM sleep fragmentation parameters and the overnight changes in autonomic measures. **Conclusions:** Our preliminary results suggest that REM sleep continuity affects physiological reactivity to emotional events, outlining a possible differential effect on the sympathetic and parasympathetic autonomic systems. REM sleep fragmentation seems to impair the habituation of parasympathetic system, showing maladaptive excessive response (HRD) to known emotional events, meanwhile it strongly flattens the sympathetic autonomic response (SCR).

P49

Does ChatGPT have a typical or atypical Theory of Mind?

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Settore Scientifico Disciplinare (SSD): MED/01 – M-PSI/03

In recent years, Large Langauge Models (LLM), such as ChatGPT, have exhibited considerable natural language processing capabilities and a potential ability to mimic human behavioral patterns. It has also been hypothesized that artificial agents will soon become credible interlocutors in social interactions. Although ChatGPT can successfully generate accurate theoretical and inferential information in several fields, its ability to exhibit a Theory of Mind (ToM) is a topic of debate and interest in experimental clinical psychology. ToM is one of the most sophisticated mental abilities of human beings and underlies the ability to converse and interact, as it allows one to interpret the behavior of others based on their mental states (e.g. beliefs, emotions, desires, goals). Impairments in ToM abilities are considered mechanisms underlying social dysfunction in many clinical conditions, including autism spectrum disorder (ASD) and schizophrenia spectrum disorders (SSD). We have recently shown that observing response style in advanced ToM tasks helps to distinguish between clinical and nonclinical populations and supports the differential diagnosis between ASD and SSD. Some studies show that ChatGPT can successfully pass classical ToM tasks, including first-order meta-representations. However, the response style used by LLMs to solve advanced ToM tasks has not been explored in detail. Based on these assumptions, through the exploration of mentalistic reasoning, we investigated whether the reasoning style used for mentalization tasks by ChatGPT overlaps with that used by healthy or pathological populations. In our study, we administered the Advanced Theory of Mind Test and the Emotion Attribution Task to ChatGPT 3.5 and compared its responses with those of ASD (n = 52), SSD patients (n = 59), and typically developing peers (n = 63). Our results demonstrate differences between pathological populations, healthy individuals and ChatGPT. In particular, ChatGPT tend to violate Grice's maxims, respond in an “artificial” manner, and show greater difficulty when faced with more complex mental states including deception and context-dependent emotion inference. The mentalistic reasoning style adopted by ChatGPT seems very similar to that adopted by high-functioning ASD individuals.

P50

Burden of preventive migraine treatment failures on a European population: a cross-sectional multicenter study

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Settore Scientifico Disciplinare (SSD): MED/26

Background and aim: Migraine is a common disabling disorder and despite the advances in its management, patients commonly experience multiple failures to acute and preventive treatments. We performed a cross-sectional analysis of the baseline data from a longitudinal, multicenter, international study.
Methods: The REFINE study aimed at testing the definition of resistant and refractory migraine provided by the European Headache Federation (EHF) consensus published in 2020. Patients with diagnosis of episodic or chronic migraine according to International diagnostic criteria were enrolled from 15 European headache centers. We assessed demographic data, medical history, comorbidities, migraine features, number and class of migraine preventive used in the past and why they were discontinued.
Results: We included 689 patients (82.6% women) with a median age of 48 years (IQR 39-57) and a median age of migraine onset of 17 years (IQR 13-23); 354 patients (51.3%) were non-resistant and non-refractory, 269 (38%) were resistant and 73 (10,6%) refractory. The median number of treatments failed for lack of efficiency or tolerability was 3 (IQR, 0-5), while the median number of contraindicated treatments was 1 (IQR, 1-2). Among patients, 437 (63.0%) had failed treatment with antidepressants, 364 (52.8%) with anti-seizure medications, 255 (37.0%) calcium channel blockers, 128 (18.6%) for monoclonal antibodies targeting the CGRP pathway, 96 (13.9%) ACE- inhibitors or angiotensin II receptor blockers, 149 (21.6%) for BTXA, 115 (16.7%) for other preventive treatments.
Conclusions: Our data highlight that preventive treatment failures are very common in patients with migraine referring to headache centers. The reasons for those failures need pathophysiological investigation and clinical attention.

P51

Tau localization and phosphorylation status after retinoic acid treatment in neuroblastoma models

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Settore Scientifico Disciplinare (SSD): MED/04

Hyperphosphorylation of the tau protein in neuroblastoma (NB) contributes to cell cycle regulation in proliferating cells. In neuroblastoma, aberrant tau phosphorylation is induced by an increase in kinase activity relative to phosphatases. Treatment with retinoic acid (RA) in NB cells causes an arrest of cell proliferation and the generation of neurites. Our study aims to evaluate whether modulation of phosphorylation and localization of the Tau protein can influence differentiation towards a neuronal phenotype, clarifying its potential role as a therapeutic target in combination with RA treatment. NB SH-SY5Y and CHP126 cell lines were used as reference tumor models. Expression analysis in NB cell lines after synchronization with Nocodazole showed reversible Tau hyperphosphorylation restricted to G2/M phase, in association with a reduction in its tubulin binding affinity. The forced accumulation of p-Tau by okadaic acid, an inhibitor of PP2A phosphatase, was associated with aberrant mitotic spindle and cell death. Starvation by serum reduction in the culture medium, together with RA treatment, induced the differentiation of SH-SY5Y and CHP-126 cells, generating branched extensions similar to those found in neurons, with cell cycle arrest in G1/G0 phase. The spatial and functional reorganization of Tau protein observed in the neuronal phenotype as compared to the tumoral phenotype suggests a role in differentiation by Tau. In fact, when we used okadaic acid in conjunction with RA treatment we detected a greater elongation of the neurites in NB cells compared to RA treatment alone and an overall higher suppression in NB cell proliferation. Besides, upon differentiation we observed tau localization shift towards the forming neurites. Thus, a change of localization can be synonym of a change of function. Our results indicate that RA-induced G1 phase arrest corresponds to increased expression levels of p-Tau and to a different intracellular Tau localization. Therapeutic strategies aimed at increasing the expression of the phosphorylated form of the Tau protein may enhance the differentiation ability of NB cells and counteract potential resistance effects associated with RA treatment in poorly differentiated neuroblastoma.

P52

Exploration of the dimensionality of a self-report measure for the assessment of ADHD in adults

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Settore Scientifico Disciplinare (SSD): M-PSI/03 - MED/01

Attention-deficit/hyperactivity disorder (ADHD) neurodevelopmental disorder characterised by patterns of inattention, hyperactivity and impulsivity. The diagnosis of ADHD in adulthood is often difficult and complex. One of the instruments available in the literature is the Conners' Adult ADHD Rating Scales (CAARS), specifically designed to assess ADHD symptoms in adults (in self and other- report form) and consisting, in its original form, of four factors or scales, namely inattention/cognitive problems, hyperactivity/irritation, impulsivity/emotional lability and problems with self-concept. To date, there is no validation in Italian of this scale. We aim to explore the structure of the CAARS self- report in a sample of Italian subjects, in order to reduce and validate the scale in Italian, for greater applicability in clinical practice. The original self-report scale, composed of 66 items, was administered to a sample of 130 individuals. Exploratory factor analysis of our sample resulted in a six-factor solution that explained 52% of the variance with 50 items. The subsequent confirmatory analysis showed a high model-fit. We support the idea that optimizing the length of the scale reduces the burden on participants and the evaluation time. Furthermore, shorter, high-quality tests, although not sufficient for diagnosis, are important for conducting large studies and as screening tools.

P53

Early Cognitive Dysfunction Hallmarks and Sleep Features in the Pathological Aging: a Pilot Study

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Settore Scientifico Disciplinare (SSD): M-PSI/01

The World Health Organization (WHO) classified dementia among the world priorities in public health. Moreover, the increase in age-related neurodegenerative diseases and life expectancy makes it necessary to differentiate between physiological and disrupted aging. Numerous studies highlight how executive function, emotions, and sleep could be affected early by neurodegeneration. The study aimed to identify specific potential patterns of executive and emotional impairment and sleep alteration in patients with Mild Cognitive Impairment (MCI) compared to elderly not affected by dementia. We enrolled 23 MCI subjects (Group 1) and 16 healthy controls (Group 2). All participants performed in Emotional Task-switching (E- TS), Trail Making Test A and B (TMT), and Free and Cued Selective Reminding Test (FCSRT). The actigraphy sleep-data were acquired for the three consecutive nights. With respect to results, ANOVA one-way showed a significant main effect for the group,indicating a worse performance in Group 1 than Group 2 in the TMT A ($p< 0,001$) and B ($p= 0,002$), FCSRT ($p= 0,002$). Moreover, a Mixed model ANOVA on performance E-TS accuracy showed a significant main effect for the task ($p< 0,001$) and group ($p= 0.005$), revealing a lower accuracy in Group1 with respect to Group in the emotional task. Finally, Group 1 showed an increased total sleep time than group 2 ($p= 0,004$). The attention and episodic memory impairment as well as the partial sleep processes alteration could describe a potential and specific prodromal pattern of early altered cognitive dysfunction, confirming the results of previous studies.

P54

Mitochondrial e3 ubiquitin ligase mul1 reduces proliferation and increases cell death in metastatic osteosarcoma cells

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Settore Scientifico Disciplinare (SSD): MED/06

Osteosarcoma is a highly aggressive and metastatic bone cancer. The identification of new therapeutic targets is important for effective treatments. A possible strategy leverages the inhibition of the hypoxia-inducible factor HIF-1α, upregulated in metastatic osteosarcoma. This study aims to evaluate the effects of HIF-1α downregulation by the mitochondrial E3 ubiquitin ligase MUL1 and the cofactor protein UBXN7 pathway in osteosarcoma cells. Firstly, we observed that MUL1 protein levels inversely correlate with tumor aggressiveness; it was found expressed in the osteoblast-like osteosarcoma Saos-2 cells, while barely detected in the highly aggressive and metastatic 143B cells [Arbitrary Unit (AU), Saos-2: 0.86±0.15, 143B: 0.24±0.07, $p<0.01$); accordingly, the expression of UBXN7 (AU, Saos-2: 0.14±0.04, 143B: 1.06±0.05, $p<0.01$) and HIF-1α (AU, Saos-2:0.12±0.03, 143B:0.90±0.17, $p<0.01$) was higher in 143B cells. MUL1 overexpression in 143B cells caused a 50% reduction of UBXN7 (AU, pEmpty-143B: 1.01±0.08 pMUL1-143B: 0.50±0.03, $p<0.0001$) and HIF-1α protein levels (pEmpty-143B: 0.79±0.10, pMUL1-143B: 0.43±0.07, $p<0.008$). Most importantly, MUL1 induction in 143B cells reduced the proliferative rate of 143B cells (AU, pEmtpy-143B: 1.01±0.04 , pMUL1-143B: 0.79±0.02, $p<0.001$) coupled with induction of apoptosis, as shown by the increase of Annexin V positive cells (AU, pEmtpy-143B: 1.01±0.02, pMUL1-143B: 1.36±0.01, $p<0.01$) and of the cleavage fragments of PARP (AU, pEmpty-143B: 1.01±0.05 pMUL1-143B: 2.65±0.06, $p<0.02$) and Caspase 3 (AU, pEmpty-143B: 1.01±0.05 pMUL1-143B: 3.36±0.09, $p<0.04$). Similar results were obtained through the induction of MUL1 by treatment with Ucf-101, an inhibitor of HTRA2, the upstream mediator of MUL1. Our results show that the MUL1/UBXN7 pathway is dysregulated in metastatic osteosarcoma cells, and that restoring MUL1 levels in the same cells results not only in HIF-1α downregulation, but also in reduced cell proliferation and increased cell death.

P55

From 2D to 3D in vitro models: insights into the molecular mechanisms involved in glaucoma

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Settore Scientifico Disciplinare (SSD): MED/46

Background; Glaucoma is an optic neuropathy characterized by a progressive degeneration of retinal ganglion cells (RGCs). Despite it is known that the major risk of neuronal injury is an elevated intraocular pressure (IOP), molecular mechanisms involved in stress response are poorly understood. Moreover, although many studies are focused on neuronal response, other types of cells in the retina could respond to IOP both promoting or countering RGCs degeneration. To study such a complex system, traditional 2D cultures present many limitations including affected cell polarity and absence of an extracellular matrix (ECM). 3D bioprinting technology can be a useful approach to overcome these challenges allowing the creation of 3D structures composed by living cells encapsulated in specific bioinks which mimic native ECM.
Aim; We propose an in vitro model of glaucoma integrating 2D and 3D models. We also examined the effect of a neurotrophic compound (NTRC) to evaluate its potential role in the treatment of glaucoma. Methods; Glaucomatous condition was reproduced by using millifluidic LiveFlow technology (IVTech). We used 3D bioprinting technology to develop an advanced co-culture system constituted by bioink-embedded Rat Retinal Müller Cells (rMC-1) on top of ARPE19 cell monolayer thus mimicking neuroretinal and retinal pigment epithelium layers respectively. Analysis on 2D and 3D culture were performed by Western Blots and viability of 3D cultures was assessed by PrestoBlue assay.
Results; rMC-1 and ARPE19 respond to hydrostatic pressure activating Akt/mTOR, NF-κB and STAT3 signaling. Interestingly, NTRC modulates these pathways in a context-dependent manner and appears to play an anti-inflammatory role.
Conclusions; Using 2D and 3D models we identified several pathways involved in the response to increased hydrostatic pressure and highlighted a potential role of NTRC in mitigating the inflammatory response. Importantly, the different responses between 2D and 3D cultures underline the importance of using three-dimensional systems.

P56

The key role of cognitive styles and gender in the association between fluid intelligence and divergent thinking in children

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Settore Scientifico Disciplinare (SSD): M-PSI/04

The association between fluid intelligence (Gf) and the ability to think divergently has widely characterized the current research in psychology. Nevertheless, the evidence on the main factors involved in this association during childhood remains a matter of debate. The present study addressed the interplay between Gf and divergent thinking (DT), exploring the involvement of field dependent independent cognitive style (FDI) and gender in a sample of 101 school-age children (Mage = 8.02; SDage = 1.43). Participants performed Raven's Coloured Progressive Matrices, the Children Embedded Figure Test, and the Alternative Uses Task. The results indicated that the FDI mediated the association between Gf and DT. In particular, these findings suggested that field independence, which depicts the disposition to solve problems analytically, represents a function of the efficiency of Gf, which allows children to set up goals and subgoals that are useful to generate divergent ideas. In addition, the findings reported that the interplay between the FDI and DT was moderated by gender, indicating that children's autonomy and analytic thinking involved in field independence are strengthened in boys. Through a multidimensional approach, the current research findings provided further insight into the primary factors involved in the ability of children to find alternative solutions to open- ended problems and think divergently.

P57

Prevention with monoclonal antibodies targeting the CGRP pathway improves the effectiveness of acute migraine treatments: real-world data

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Settore Scientifico Disciplinare (SSD): MED/26

Background: Enhancing the efficacy of acute migraine treatments is a crucial objective of migraine preventive therapies, yet research in this area remains limited. This study aimed to evaluate the potential of monoclonal antibodies targeting the CGRP pathway (CGRP-MoAbs) in improving both the response to acute treatments and their efficacy.
Methods: Patients diagnosed with chronic or episodic migraine from the Headache Centers of Avezzano-L'Aquila and Naples were enrolled between March 2021 and December 2022. We included patients who initiated treatment with any CGRP-mAb (erenumab, fremanezumab, galcanezumab) during their baseline visit and followed them for up to 6 months. At both the initiation of CGRP-mAb treatment and after 6 months, all participants completed the Migraine Treatment Optimization Questionnaire (MTOQ), a validated self-administered tool assessing the response to acute treatments, higher MTOQ scores indicate improved acute treatment optimization. Throughout the study period, participants maintained a headache diary to record migraine days and acute medication usage.
Results: Out of 80 patients initially screened, 65 (81.3%) completed the 6-month follow-up assessment. The majority of participants were female (55, 84.6%), with a median age of 46 years (interquartile range [IQR] 39-56). The median MTOQ tool-score increased from 8 (IQR 4-13) at baseline to 15 (IQR 11-17) at 3 months ($p<0.001$) and 16 (IQR 13-17) at the 6-month follow-up ($p<0.001$). Median migraine days over 90-day periods decreased from 40 (IQR 24-60) to 24 (IQR 15-30) at 3 months ($p<0.001$) and further to 20 (IQR 12-24) at 6 months ($p<0.001$). The median monthly intake of acute medication decreased from 55 doses (IQR 29-80.5) to 24 doses (IQR 15-40) at 3 months and 18 doses (IQR 11-30) at 6 months ($p<0.001$).
Conclusion: Our study demonstrate that 6 months of preventative treatment with CGRP-MoAbs resulted in significantly improved effectiveness of acute treatments, accompanied by reductions in monthly migraine days and use of acute treatments. This effect contributing to better management of migraine-related disability.

P58 – SELECTED ORAL COMMUNICATION

In-vitro approaches to investigate the detrimental effect of light on dopaminergic neurons

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Settore Scientifico Disciplinare (SSD): BIO/14

Parkinson's disease (PD) stands out as one of the most prevalent neurodegenerative conditions, typically manifesting in later stages of life, characterized by the gradual deterioration of motor functions. Recently identified environmental factors either increase the risk or display an inverse relationship with the PD development; exposure to certain pesticides, for example, has been linked to an increased PD risk. Although light pollution hasn't been considered as a potential risk factor for PD, there is an escalating concern for its impact on human health in several scientific fields. Several publications have already shown that prolonged exposure to bright light, emitted by common fluorescent lamps, can significantly reduce dopaminergic neurons in the substantia nigra of rats and mice, *in-vivo*. For instance, we have shown that light wavelengths around and above 600 nm penetrates the scalp and skull of rodents, reaching deep into the substantia nigra. Importantly, computed tomography and magnetic resonance imaging, strongly suggest that the environmental light reaches the mesencephalon of a human brain. Taken together these data suggest that light, especially at 610nm, could directly interact with dopaminergic neurons. In particular, fluorescent white light's electromagnetic spectrum includes wavelength peaks in the blue, green, and red range, collectively producing white light. Monochromatic LED lights were thus used in this work to deepen our knowledge on the detrimental effects of the major wavelength peaks of fluorescent light on mouse and human dopaminergic cells including immortalized dopaminergic MN9D cell lines and dopaminergic neuronal cultures derived from human induced pluripotent stem cells (hiPSCs). Our findings indicate that LEDs with wavelengths shorter than 710 nm are harmful to dopaminergic neurons. Strikingly, The peak at 610 nm that has been shown to reach deep into the brain, induces a clear increase in ROS production, cytotoxicity and neuronal death compared to controls. These observations suggest that the peaks around 610 nm may be responsible for the detrimental effects observed with in vivo exposure to fluorescent white light and may pave the way to the development of in-vitro and drug-free in-vivo models of Parkinson's Disease.

P59

ROS-dependent cytotoxicity and inflammatory effects induced by azoles combinatorial mixtures in Sertoli TM4 cells

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Settore Scientifico Disciplinare (SSD): BIO/13

Sertoli cells (SCs) have a key role in maintaining functional spermatogenesis, providing nutritional and structural support for germ cells, as well as antioxidant factors (*e.g.* superoxide dismutase, SOD; glutathione, GSH). They are also essential for their direct implication in the blood–testis barrier (BTB) homeostasis and in the release of various immunomodulatory factors. SCs are also considered to be one of the main targets of environmental toxicants in male reproductive dysfunctions, though little is known about potential cooperative toxic effects. Triazole and imidazole fungicides, already known as endocrine disruptors, are an emerging class of contaminants with an increasing and ubiquitous presence in the environment. In this regard, a combinatorial activity of tebuconazole (TEB; triazole fungicide) and econazole (ECO; imidazole compound) in inducing mitochondrial impairment, energy depletion, cell cycle arrest, and the sequential activation of autophagy and apoptosis in Sertoli TM4 cells has recently been demonstrated. Given the strict relationship between mitochondrial activity and reactive oxygen species (ROS), and the causative role of oxidative stress (OS) in male reproductive dysfunction, the individual and combined potential of TEB and ECO in inducing redox status alterations and OS was investigated. Furthermore, considering the impact of cyclooxygenase (COX)-2 and tumor necrosis factor-alpha (TNF-α) in modulating male fertility, protein expression levels were assessed. In the present study, we demonstrate that azoles-induced cytotoxicity is associated with a significant increase in ROS production, a drastic reduction in SOD and GSH-S-transferase activity levels, and a marked increase in the levels of oxidized (GSSG) glutathione. Exposure to azoles also induced COX-2 expression and increased TNF-α production. Furthermore, pre-treatment with N-acetylcysteine (NAC) mitigates ROS accumulation, attenuates COX-2 expression and TNF-α production, and rescues SCs from azole-induced apoptosis, suggesting a ROS-dependent molecular mechanism underlying the azole-induced cytotoxicity.

P60

A new insight in PDIA1-induced Cole Carpenter Syndrome

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Settore Scientifico Disciplinare (SSD): BIO/17

The Cole Carpenter Syndrome (CCS) is a neglected autosomal dominant disease affecting skeleton with a prevalence <1:1,000,000. Affected individuals experience non-traumatic fractures, frontal bossing, ocular proptosis, communicating hydrocephalus and craniosynostosis. Genetically, CCS is caused by the p.Y393C heterozygous mutation in the P4HB gene, encoding for the Prolyl 4-hydroxylase β subunit (also known as Protein Disulfide- Isomerase A1, PDIA1), leading to a tyrosine-to-cysteine aminoacidic substitution in the protein chain. The PDIs are the most abundant protein in the Endoplasmic Reticulum (ER), and they are involved in the formation of disulfide bridges in the nascent polypeptide chains playing a pivotal role in protein oxidative folding. Moreover, PDIA1 is involved in the hydroxylation of prolyl residues in type I procollagen and acts as chaperone. To date, there are no studies showing how the CCS-inducing mutation affects the skeletal homeostasis. Based on this, we generated cellular models to investigate the subcellular mechanisms altered by the PDIA1 Y393C mutation. Western Blot and immunofluorescence analyses on HEK-P4HBY393C-GFP showed a higher PDIA1 stabilization compared to HEK-P4HBWT-GFP while protein subcellular localization was unchanged. Of note, Bip1 and p62 expression were higher in mutant compared to WT cells (+3fold and +1.5fold, respectively; p=0.05) along with an unchanged LC3 level, suggesting ER stress activation and a possible involvement of cell autophagy. In addition, analysis conducted in 3T3 fibroblast like cells transfected with the P4HBY393C-GFP revealed a reduction in type I collagen expression (-50%; p=0.009) and secretion (-70%; p=0.01) compared to 3T3 cell transfected with the WT construct. Similar results were obtained in primary mouse osteoblasts isolated from the CCS mouse model carrying the mouse homolog Y395C P4hb mutation. Finally, in vivo analysis in CSS mice sera showed a lower level of PINP1 (N-terminal pro-collagen I propeptide), a marker of type I collagen turnover in the bone matrix, compared to WT littermates (-60%; p<0.01). Overall, our data demonstrated that the CCS-causing mutation induced a defective type 1 collagen biosynthesis and secretion leading ER stress and cell autophagy impairment.

P61

Identification of the molecular mechanisms in preclinical models of high-grade gliomas related to the use of a TORC1/TORC2 inhibitor: RES529

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Settore Scientifico Disciplinare (SSD): MED/50

Introduction: Glioblastoma multiforme (GBM) is the most aggressive primary brain tumor. New therapeutic approaches are needed to improve survival and quality of life. GBM is a highly vascularized tumor, and it is often characterized by treatment-induced resistance. Therefore, it is important to identify and treat pathways involved in resistance and in angiogenic ability. Anti-angiogenetic compounds tested in preclinical and clinical settings demonstrated recurrence associated to secondary activation of the phosphatidylinositol 3- kinase (PI3K)/AKT/mTOR pathway.
Aim: Fo this reason, we would determine the sensitizing effects of the small molecule and oral available dual TORC1/TORC2 dissociative inhibitor, RES529, alone or in combination with the anti-VEGF blocking antibody, bevacizumab, or the tyrosine kinase inhibitor, sunitinib, in human GBM models.
Results: We observed that RES529 inhibited dose-dependently the growth of GBM cells *in vitro*; it was able to reduce the migration and invasion of tumor cells and tubule formation from both brain-derived endothelial cells (angiogenesis) and also it is able to decrease Glioblastoma Stem Cells (GSCs) neurosphere formation.
Conclusions: In summary, these data indicated that RES529 showed to be active in GBM preclinical models and, in association with anti-angiogenic agents; edditionally, the study demonstrated the anticancer efficacy of the compound and support the clinical investigation of RES529 as a potential novel therapy for this aggressive brain tumor type.

P62

Preclinical validation of more predictive 3D cell models in pharmaceutical applications

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Settore Scientifico Disciplinare (SSD): MED/04

Recent advances of in vitro 3D culture technologies, such as bioprinting and spheroid, have opened new avenues for the development of novel tumor models that mimic in vivo tumoral features. These models allow us to obtain more predictive data then in vitro 2D studies. Recent advances in three-dimensional printing techniques and the biomaterials used have provided new tools for tissue cultures development. Our study aims to develop novel 3D tumor models, testing their potential role in predicting the antitumor efficacy of Src inhibitors. In our work two glioblastoma cell lines, U87 and U251, have been cultured in 2D and 3D models. The 3D models are bioprinting and spheroids. These models have been used to evaluate the activity of two newly synthesized compounds which are the [3,4-d] pyrazole pyrimidine inhibitors: si306 and PD1 (pro-drug of si306). U87 and U251 cells were able to proliferate in 3D bioprinted alginate/gelatin structures for two weeks, generating spheroidal aggregates whose diameter tends to grow progressively while maintaining high levels of Src activation. The bioprinted cells were significantly more resistant to si306 and PD1 toxicity, showing higher IC50s than the corresponding 2D cultured cells and the treatment response is dependent on the initial diameter of spheroids formed in the alginate/gelatin hydrogel. We have verified that 3D bioprinting can express new biological features respect to 2D cultures, mimicking resistance features shared by in vivo tumors, and in particular a significant dependence on spheroid dimension. These models can offer effective tools for evaluating tumor phenotypic features in 3D environments and the biological bases of cancer resistance to therapy.

P63– SELECTED ORAL COMMUNICATION

The Word-Pseudoword Association Learning task (WPAL): development and validation of a novel double-version self-administered tool for sleep-memory studies

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Settore Scientifico Disciplinare (SSD): M-PSI/02.

Introduction: This study introduces a novel web-based declarative memory task allowing self- administration in within-subject design protocols. We developed two parallel versions of a Word-Pseudoword Association Learning task (WPAL), comparing their difficulty in a sample of healthy adults. Moreover, we evaluated task's sensitivity to pre-learning sleepiness, and the effect of a post-learning sleep period on long-term memory retention. **Method:** During WPAL, participants try to learn the Italian translations of 40 acoustically presented pseudowords (non-sense phoneme combinations compliant with phonotactic Italian rules). Two 40-item lists of both Italian words and pseudowords were created. Based on normative data, Italian word lists were balanced for familiarity, imaginability, concreteness, pleasantness, arousal, length, and usage frequency. Pseudoword lists were equalized for wordlikeness, length, and consonant-vowel structure. Finally, different task versions (A and B) were generated by forming two lists of randomly coupled word-pseudoword pairs. WPAL consists of five trials: Exposure, three Learning trials, and Test. During Exposure, each pseudoword is presented followed by its Italian translation. During Learning trials, each pseudoword is presented and participants have to type its translation (subsequently shown as feedback). In the Test trial, pseudoword translations are requested without providing correct responses. Task validity was assessed by an experiment involving two 15-subject groups with learning sessions scheduled at distinct times (WAKE group: 9:00AM, mean age ± SD: 27.45 ± 7.31; SLEEP group: 9:00PM, 28.21 ± 9.25) and a re-Test performed after 12 hours. Participants experienced two counterbalanced conditions (task version A and B) a week apart. Pre-learning sleepiness was measured by the Karolinska Sleepiness Scale.
Results: A repeated-measures-ANOVA with "Trial" (Learning 1–3, Test) and "Task version" (A, B) as factors indicated no significant interaction ($p=0.20$), suggesting equivalent learning trajectories over different task versions. An independent-samples t-Test was conducted on the performance changes from Test to re-Test comparing SLEEP and WAKE groups. Analysis highlighted a significant effect ($p<0.001$), suggesting worse re-Test performance in the WAKE group (−8.41%) compared to the SLEEP one (−1.43%). Higher sleepiness predicted lower learning performance ($R=0.48$, $p=0.048$). **Conclusion:** WPAL represents the first double-version, self-administered, sleep-dependent, and sleepiness-sensitive task, holding promise for advancing memory research in within-subject designs and remote settings.

P64

Identification of active molecules for the treatment of Cole-Carpenter syndrome by repositioning FDA-approved drugs

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Settore Scientifico Disciplinare (SSD): BIO/17

The Cole Carpenter Syndrome (CCS) is a rare genetic disease that mainly affects bone, which becomes very brittle. In addition, they present a wide spectrum of bone alterations that cause various and extensive skeletal deformities. All this significantly compromises the life of these people. Unfortunately, there is no cure for this disease, and patients received only palliative treatments. For this reason, the development of a therapy for CCS patients is a very important challenge. Based on this, we identified active molecules that can be reprofiled to be use for treating this disease. Indeed, new drug development is a time-consuming and hight-cost process, that sometimes is not applicable to neglected diseases. Instead, drug repurposing offers various advantage, specifically in the contest of rare and neglected diseases, such as CCS. For these reasons, we tested a drug library composed by 770 active molecules already approved by the *Food and Drug Administration* (FDA) for the use in humans, using a broad-to-narrow approach. To perform the screening, we used primary wild-type (WT) mouse osteoblast cultures and the different compounds of the library have been added for 72 hours. According to the alteration in the type 1 collagen biosynthesis and secretion we observed in in vitro and in vivo models of CCS, we used as primary readout of the screening the measurements of the PINP1 (*N-terminal pro- collagen I pro-peptide*) levels in conditioned medium of the treated cell. DMSO (vehicle)-treated cells were used as control. We selected the top 15 drugs out of the 770 that increased the PINP1 secretion (+50%) in drug-treated osteoblasts compared to the vehicle-treated cells. Next, for the best 15 drugs selected in the initial screening we also analysed their impact on mature type 1 collagen secretion. From this analysis we selected 4 candidate drugs, including Imiquimod, Travoprost, Cidofovir and Cladribine, able to increase the type 1 collagen secretion by 30% (p=0,0024). Overall, in this study we identified possible drugs that can be potentially repurposed for the treatment of CCS, paving the way to their pre-clinical test in the CCS mouse model.

P65

Alterations of the endocannabinoid system and microglia reactivity in the retina precede the onset of β-amyloid plaques in the brain of AD-like mice

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Settore Scientifico Disciplinare (SSD): BIO/10

Alzheimer's disease (AD) develops extra-cerebral manifestations in the retina, which is then considered a "window to the brain". Here, we explored for the first time the possible alterations of the endocannabinoid (eCB) system (ECS) and the onset of gliosis in the retina of AD-like mice before the development of β-amyloid plaques in the brain. 12-month-old Tg2576 (TG) mice over- expressing the amyloid precursor protein (APP) were used. Via immunohistochemistry, we confirmed the absence of β-amyloid plaques in TG brains. Then, retinal gliosis was investigated through immunofluorescence on cryosections, showing a significant increase of IBA1 (+) microglia cells in TG *versus* wild type (WT), and an increase in GFAP immunostaining in the inner retina. Analysis of the ECS (receptors/metabolic enzymes) through Western-blotting revealed the up-regulation of cannabinoid receptor 2 (CB2) in TG (1.5 folds over WT), which was consistent with fluorescence intensity analysis of anti-CB2 immuno-stained cryosections. Instead, no statistically significant differences were found for the other enzymes and receptors of the ECS; however, linear regression analysis for individual animals showed a significant correlation between CB2 and fatty acid amide hydrolase (FAAH), diacylglycerol lipase α/β (DAGLα/β), and APP. Finally, liquid chromatography-mass spectrometry (LC–MS) revealed a significant down- regulation of the eCB 2-arachidonoylglycerol (2-AG) in TG retinas (~0,34 ng/mg) compared to WT (~1,7 ng/mg), while a trend towards increase was found for the eCB anandamide (AEA) (WT:~0,15 ng/mg; TG:~0,24 ng/mg). Overall, these findings indicate that the ECS may play a role in AD-associated retinal inflammation, resembling the AD brain, with a central role of CB2 and 2-AG. Importantly, retinal gliosis and ECS dysregulation precede the development of β-amyloid plaques in the brain of AD-like mice.

P66

N-acetyl-L-cysteine (NAC) impairs osteosarcoma aggressiveness: finding new therapeutic opportunities

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Settore Scientifico Disciplinare (SSD): BIO/17

Osteosarcoma (OS) is the most common primary bone tumour, predominantly affecting children and adolescents, and is prone to develop drug resistance and lung metastases, resulting in a drastic reduction in patients' life expectancy. The over-the-counter product N-acetyl-L-cysteine (NAC), a reactive oxygen species scavenger and precursor of glutathione, has been demonstrated to exert controversial effects on cancer growth, however its effect on OS has been poorly elucidated. We aimed to investigate this aspect, to develop alternative therapeutic approaches for counteracting OS growth. The human OS cell line MNNG/HOS was treated with NAC for 48 h (hours), finding a concentration-dependent reduction of cell number and metabolic activity, evaluated by crystal violet and MTT assay, respectively, compared to untreated cells. Due to the mesenchymal/osteoblastic origin of OS, we also treated both primary mouse osteoblasts and the human fetal osteoblastic cell line FOB with NAC, finding a reduction of metabolic activity only at the highest concentration tested (10 mM), thus suggesting that tumour cells are more sensitive to NAC than normal cells. We next evaluated any effect of NAC on MNNG/HOS proliferation by 5- ethynyl-2'-deoxyuridine incorporation assay, finding no effect after 24 h of treatment, while a reduction was observed at 48 h compared to untreated cells. Interestingly, MNNG/HOS pre-treated for 48 h with 5mM NAC also showed a significant lower ability to invade through Matrigel. Consistently, zymography performed on conditioned medium (CM) collected from MNNG/HOS treated with 5mM NAC showed a significant reduction of MMP-2 activity, compared to CM from untreated cells. In conclusion, our data indicates that treatment of OS cells with NAC significantly impairs their aggressiveness. Further experiments will allow to elucidate the mechanisms involved, with the proposal to improve our understanding of how NAC influences OS cells behaviour and elucidate whether it could be a valid antitumoral insult to be used in OS therapy.

P67 – SELECTED ORAL COMMUNICATION

Flow cytometry in the determination of platelet-associated immunoglobulins

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Settore Scientifico Disciplinare (SSD): BIO/12

Background. Thrombocytopenia is the finding of a low blood platelet count. The determination of antiplatelet antibodies or platelet-associated immunoglobulins is one of the possible mechanism to study thrombocytopenia. The majority of tests for determination of antiplatelet antibodies were initially introduced to determine antibodies in patients with idiopathic thrombocytopenic purpura. These methods are now employed for all diseases associated with platelet impairments, when an immunologic pathomechanism of the disease onset is suspected.
Methods. While the majority of clinical test investigate the presence of antibodies against platelet GPIb/IX, GPIIb/IIIa2, flow cytometry can be used to detect antibodies bound on platelet surface using anti-Human IgG F(ab)2 Antibody (FITC conjugated) after isolation of the platelet population. Anti-Human secondary antibodies are affinity-purified antibodies with great specificity for human immunoglobulins and are useful in the detection of its specified target. The measurements were collected in the period June 2023 – January 2024.
Results. On the basis of their platelet count, the patients were organized in three groups: the first group with a count between 150 and 100 (x103/µL) represents 50% of the total exanimated followed by the second group (100-50 x103/µL) representing the 32% and the third group (<50 x103/µL) representing the 18%. Results are expressed in percentage of platelet with antibodies bound on their surface. The first group has shown the best correlation between appropriate clinical request and percentage of platelet with antibodies bound to surface (35%). The third group there is no direct correlation between the severity of thrombocytopenia and percentage of platelet despite a higher mean value (44%).
Conclusion: Further studies are necessary to better investigate the role of this antibodies in vivo and how do they interact with the formation of the blood clot.

P68

The assessment of safety in patients with rheumatoid arthritis treated by JAK inhibitors, results from a single center study

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Settore Scientifico Disciplinare (SSD): MED-16

Background: Janus Kinase (JAK) inhibitors are widely used in treatment of rheumatoid arthritis (RA). Recent safety concerns, which were raised by randomized controlled trials, guided the regulatory agencies to advise some caution in their administration. A higher risk was suggested about the occurrence of major adverse cardiovascular events (MACEs), deep-vein thrombosis (DVT), and malignancies. However, these warnings emerged after the treatment licensing in daily clinical practice, advocating a revaluation of already treated patients with JAKis.
Objectives: To assess the occurrence of severe adverse events (AEs) in patients with RA treated by JAKis in our outpatient clinic.
Methods: A review of clinical charts of patients treated with JAKis, for at least 12 months, was performed to evaluate the occurrence of AEs (coded according to MeDRA-SOC classification) focusing on severe AEs (an episode endangering the patient life and requiring immediate medical intervention), mostly MACEs, VTEs, severe infections, and malignancies.
Results: 51 patients with RA treated by JAKis were assessed (46 women, mean age 69.14 years, mean disease duration 9.2 years). Considering therapies, 31.7% received a previous biologic DMARD, 56.9% were concomitantly treated with a conventional synthetic DMARDs (mainly methotrexate), and 33.3% with glucocorticoids. Furthermore, 43.1% of patients were also affected by high blood pressure and 23.5% by type 2 diabetes, 35.3% were currently or formerly smokers, and 23.5% were obese.
During the follow-up, paralleling with the clinical improvement, a low incidence of severe AEs was recorded. Specifically, during a mean follow-up of 18 months (range 12-36 months), 2 patients developed not-fatal MACE (3.9%) and 2 patients VTEs (3.9%). These events led to the discontinuation of the administered JAKi. No severe infections were observed, and no new malignancies were registered. Comparing these findings with available evidence, a lower cumulative incidence of severe AEs could be descriptively suggested in our cohort. Finally, 13% of patients experienced mild and transient AEs not leading to the discontinuation of the administered JAKi.
Conclusions: Despite our monocentric study design, a lower occurrence of severe AEs was observed than available evidence. Further studies are needed to fully elucidate these issues and the safety of JAKis in patients with RA.

P69

Characterization of oral biofilm

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Settore Scientifico Disciplinare (SSD): MED/28, BIO/16

Oral microbiome is an important component of the oral ecosystem contributing to oral health. Therefore, it is crucial to analyze the characteristics of the biofilm starting from the macroscopic structure to the microscopic architecture of the oral biofilm. The oral biofilm is composed of the microbiome, in which microorganisms proliferate and interact with the different niches of the oral cavity: teeth, tongue, subgingival niche and the restorative materials that could be used in clinical dental practice.Morphological methods can be used to identify species, adhesion mechanisms contributing to biofilm formation and stability and how the action of antibacterial molecules is efficient towards pathological biofilm. Microscopy is the primary technique for the morphological characterization of biofilm. Optical microscopy is the main technique for the identification of bacteria, it includes types such as: stereomicroscopy, confocal laser microscopy combined with fluorescence in situ hybridization (FISH) methods and Live/Dead methods.The other type of microscopic technique is electron microscopy, such as scanning electron microscopy (SEM) and transmission electron microscopy (TEM) which allow us to explore the architecture of the biofilm, therefore the bacterial population, the matrix created by extracellular polymeric substances (EPS) and the mechanisms of physical and chemical forces that contribute to the adhesion of biofilm to substrates, at the nanometric level.Furthermore, microscopic methods lead the way in studying antibacterial treatments, thanks to the capabilities of the latest advanced electron microscopy, such as scanning transmission electron microscopy (STEM) and high-resolution transmission electron microscopy (HR-TEM), and correlative microscopy, to reveal the effectiveness of different molecules in breaking down the biofilm and investigate their shape and size in relation to the mechanism of adhesion and action against oral biofilm.Finally, evidence based on scientific literature shows that the established methods represent the most common tools used to characterize biofilm and its morphology in the field of oral microbiology. Further protocols and studies on the application of advanced microscopic techniques are needed to obtain fine details on the microbiological and pathological aspects of oral biofilm.

P70

Role of secondary lipofilling after breast reconstruction with implants, a case report

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Settore Scientifico Disciplinare (SSD): MED/18

Background: Autologous fat grafting (AFG or lipofilling) is a common technique that involves the transfer of autologous fat tissue from one region of the body to another (in particular abdominal area, trochanteric region and the inside of the thighs and knees). Lipofilling is often used to increase the volume and contour abnormalities in implant-based breast reconstruction. The most practiced fat harvesting technique has been described by Coleman and colleagues, that represents an evolutionary approach to fat grafting. After harvesting, the fat is usually centrifuged to separate the purifed fat (to be reimplanted) from the cellular debris, that contain part of the stem cells (ADSC). AFG has been applied to oncoplastic surgery with satisfactory results since the early 80s.
Matherial and methods: The case examined is that of a 49-year-old woman, who underwent nipple-sparing mastectomy with immediate reconstruction with prosthesis in 2017 for malignant right breast cancer. The patient had scar retraction of the outer quadrants of the right breast with ripple of the skin and emptying into the inner quadrants especially in clinostatism. Two harvesting of adipose tissue were taken from the trochanteric regions, for a total of 60cc of adipose tissue. After centrifugation and purification, the fat was injected into the sites of major cutaneous depression of the right breast.
Results: Comparing with the literature, the result has been satisfactory already in the post- operative seen the immediate filling of the previous cutaneous depressions, with satisfaction from the patient.
Conclusions: Lipofilling is reconfirmed as one of the best breast reconstructive options. In addition to the use of an autologous tissue, avoiding any type of foreign body reaction or rejection, the advantage of this method is the modulation of symmetry. This technique being little invasive, without visible scarring. The disconfort is essentially constituted by the need, in most cases, of different reconstructive times in order to obtain a well-vascularized and durable graft.

P71 – SELECTED ORAL COMMUNICATION

The evaluation of effectiveness and safety of Guselkumab in patients with psoriatic arthritis in a prospective multicentre "real-life" cohort study

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Settore Scientifico Disciplinare (SSD): MED/16

Background: Psoriatic arthritis (PsA) is a chronic disease characterized by widespread musculoskeletal inflammatory manifestations in patients with psoriasis. Indeed, many patients may develop a destructive form of arthritis with a consequent morbidity and disability. Furthermore, PsA is frequently associated with comorbidities which may may worse the outcome of these patients over time. Objectives: To evaluate the 6-month effectiveness of Guselkumab, IL-23 inhibitor, in patients with psoriatic arthritis (PsA) in a "real-life" multicentre patient cohort. To estimate the drug retention rate (DRR) of Guselkumab, also assessing the impact of comorbidities and patient clinical characteristics, in a collective 18-month prospective follow-up.
Methods: Between December 2021 and September 2023, patients with PsA were evaluated if treated at least for 6 months with guselkumab in a prospective multicentre study to evaluate the effectiveness of Guselkumab by means of DAPSA and cumulative DRR.
Results: 111 patients with PsA were evaluated and treated with Guselkumab (age 56.8±9.9, male sex 20.7%). These patients were mainly characterised by active and long-standing PsA with median disease duration of 6.0 (7.0) years (55.9% disease duration ≥ 5 years), 55.0% showed comorbidities, 78.4% of patients were previously treated with bDMARDs, and 60.4% concomitantly with csDMARDs. After 6 months, a significant reduction of DAPSA was observed (β: −15.47, p=0.001, 95%CI: −23.15 to −9.79) with 39.6% of patients who achieved a DAPSA £ 14. At the end of cumulative follow-up, 71.2% of patients were still treated with Guselkumab whereas 24.3% discontinued the drug due to inefficacy. An 18-month DRR of Guselkumab of 66.7% was estimated with a mean time of administration of 9.8 ± 4.1 months. The results of the DRR were stratified according to patient clinical characteristics. The DRR of Guselkumab appeared to be not influenced by long disease duration, comorbidities, obesity, concomitant csDMARDs, and previous bDMARDs.
Conclusions: The "real-life" 6-month effectiveness of Guselkumab was shown in patients with PsA, mainly characterised by active long-standing disease, previously treated with bDMARDs, and with comorbidities. Furthermore, a good DRR of Guselkumab was estimated in the cumulative 18-month of follow-up, which appeared to be not influenced by long disease duration, comorbidities, obesity, and previous bDMARDs.

P72

Downstaging with Yttrium-90 radioembolization prior to liver transplantation in patients with hepatocellular carcinoma: our experience

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Settore Scientifico Disciplinare (SSD): MED/18

Background and Aims: Liver transplantation (LT) is a well-established procedure for hepatocellular carcinoma (HCC) within the Milan criteria. Yttrium-90 microspheres radioembolization (Y90-RE) proved to be an effective and safe loco-regional treatment of primary liver tumors. We retrospectively evaluate the efficacy of the Y90-RE in patients with HCC to downstage the tumor burden within the Milan criteria prior to LT.
Methods: From January 2007 to December 2018, 203 patients referred at the Department of "General Surgery and Transplantation" of the San Camillo Hospital Center with a diagnose of HCC, including patients with portal vein tumoral thrombosis (PVTT). After an assessment of the tumor burden, the baseline biological markers and the liver function, the patients underwent Y90-RE.
Results: One hundred eighty-four patients underwent TARE according to our downstage protocol. At the six months-restaging according to RECIST criteria, downstaging within Milan criteria was achieved in forty patients, with a successful downstaging rate of 22.3%. Alpha-fetoprotein decreased after Y90- RE treatment, with a mean value of 33.41 ± 148.38 ng/ml at the moment of LT. From LT, the overall survival was 49.05 ± 40.62 months (vs 31.03 ± 9.72 months in the group of patients who didn't achieve a successful downstage, p = 0.0001), with a disease-free survival of 20.09 ± 16.45 months and a recurrence rate of 15%. At the pathological examination of the explanted liver, the understaging rate was 32.5%.
Conclusion: Y90-RE becomes a real option to provide successfully downstaging within Milan criteria for patients who were traditionally not considered eligible for surgery, allowing to perform LT.

P73

Embolization of the renal artery before graft nephrectomy: a comparing study to evaluate the possible benefits

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Scuola di specializzazione in Chirurgia Generale

Settore Scientifico Disciplinare (SSD): MED/18

The graft nephrectomy is burdened by immunological and surgical complications. The main surgical complications of graft nephrectomy are hemorrhage, infections, vascular injury and death. The mortality is high, with percentages varying between 1.3 and 38%. Therefore, graft nephrectomy should be recommended only in selected cases. We conducted a retrospective study, comparing the data of 26 patients undergoing an allograft nephrectomy (2009–2013), without embolization of the renal artery (NO EMBO group) with the data of 26 patients undergoing an allograft nephrectomy (2014–2019), with embolization of the renal artery (EMBO group). We included only graft nephrectomies performed at least 6 months after transplantation. The patients included in the study were consecutive because until 2013 we did not perform the embolization of the renal graft artery. Afterwards, from 2014, instead, we routinely carry out embolization to all patients to be subjected to graft nephrectomy. We, therefore, wanted to analyze whether this surgical approach compared to the previous technique can lead to an improvement in morbidity and mortality, reducing the risk of bleeding and operating times. The examination of our data highlights that embolization of renal artery reduces the operating times of the explant, in addition the group subjected to embolization had less changes in hemoglobinemia and less blood loss.

P74

Management of neonatal diabetes: results of an international survey

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Scuola di specializzazione in Pediatria

Settore Scientifico Disciplinare (SSD): MED/38

Introduction: Neonatal Diabetes Mellitus (NDM) is a rare form of diabetes with Mendelian transmission, usually diagnosed within the first 6 months of life. Genetic analysis allows to achieve a molecular diagnosis in over 80% of patients, and it is crucial for clinical course prediction and treatment choice. We investigated clinical practice about diagnosis and clinical management of NDM to identify unmet needs for clinicians.
Methods: A 45 questions multiple-choice survey was conducted among the International Society for Pediatric Diabetes (ISPAD) members through the ISPAD website. Participant were recruited voluntarily via e-mail. Answers about job experience, expertise and clinical practice (investigated through a case scenario) were anonymously recorded.
Results: 108 ISPAD members from 36 different Countries from all Continents agreed to join. 89.8% were pediatric diabetologists, 10% adult diabetologists or other pediatric diabetes team members. 95.4% of them have ever been involved in NDM management. Only 60.2% felt confident to diagnosis NDM in that case scenario, and 82.4% thought that permanent course could be predicted. 88% of participants suggested to start insulin and only 33% would choose an insulin pump. Continuous glucose monitoring (CGM) was preferred by 80.6%. Near all participants would request genetic testing, but 20% of them do not run genetic analysis because of cost/logistic difficulties. 96.3% agreed that genetic testing if crucial to start treatment with sulphonylurea and 73.1% would not try the drug before genetic testing. Interestingly, 63% of participants have never received specific training about NDM, despite 97.2% considered it mandatory for a pediatric diabetologist. 55.5% of the participants felt fairly/completely confident in the management of NDM, 44.5% felt less confident.
Conclusion: NDM is a rare disease requiring specific expertise. International guidelines suggest insulin pump with CGM in these cases. Genetic testing should be run in all the patients to choose the most appropriate treatment, as kids carrying a K-ATP channel gene mutations, which is the most frequent cause of NDM, can be successfully switched to sulphonylurea. Most of the participants have ever been involved in NDM care, nevertheless it seems that medical education and specific training are still required.

P75

Differentially expressed microRNAs as putative biomarkers in a glaucoma mouse model

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Scuola di specializzazione in Patologia Clinica e Biochimica Clinica

Settore Scientifico Disciplinare (SSD): MED/46

Background. MicroRNAs are short, non-coding RNAs, able to fine-tune gene expression involved in maintaining cell homeostasis and fundamental biological processes. Dysregulated miRNAs expression is observed and closely related to many human diseases, including glaucoma, a neurodegenerative disease leading to loss of vision, characterized by progressive death of retinal ganglion cells and optic nerve axons. Among the main factors that play a key role in the onset of glaucoma are aging and increased intraocular pressure (IOP), which can lead to neuronal lesions. Although many glaucoma risk factors have been identified, its pathogenesis is largely unknown.
Aim. In this study, a microRNA profiling was performed in retinas from a glaucoma mouse model to provide novel insights into the potential involvement of such molecules in glaucoma pathogenesis.
Methods. An *in vivo* study was carried out by using DBA/2J mice, described as a model for congenital experimental glaucoma. An optometric test (IOP level) was performed to distinguish 7 months old glaucomatous and non-glaucomatous mice. After sacrifice, retinas were collected, and microRNAs expression levels were analysed by TaqMan-based RT-qPCR. Significantly dysregulated microRNAs were examined *in silico* by DIANA-miRpath and Tarbase to identify target genes and pathways.
Results. MicroRNA profiling showed 8 significantly down-regulated and 16 up-regulated miRNAs in the glaucomatous mice retinas. A total number of 67 miRNAs-related significant pathways (e.g. MAPK, Neurotrophin, Hippo, Wnt, TGF-beta, TNF, mTOR), involved in biological processes already described in retina pathophysiology and also implicated in glaucoma, were identified. Among the target genes, several were highlighted with a proven role in inflammation, regulation of the maintenance of the Trabecular Meshwork structure, IOP, apoptosis, and oxidative stress.
Conclusions. Significantly dysregulated microRNAs were identified in retinas from a glaucoma mouse model. In silico analysis showed several interesting pathways/target genes to be used for further characterizing glaucoma pathogenesis. Given the putative role of miRNAs as biomarkers or therapeutic targets, this study makes available data potentially exploitable in clinical practice.

P76

Prediction of cardiovascular risk in kidney transplant patients using High sensitive troponin I

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Scuola di specializzazione in Patologia Clinica e Biochimica Clinica

Settore Scientifico Disciplinare (SSD): BIO-12

Background. The measurement of cardiac troponin (cTn) by a high sensitivity method now represents the standard method for cTn measurement in the laboratory. Serum high sensitive troponin could be a new tool for the risk assessment of cardiovascular events (CVE) in a proportion of patients who are not suspected of having an acute myocardial infraction but nonetheless low concentration of cardiac troponin is associated with an increase in the risk of major cardiovascular events (1).
Methods. A total of 379 kidney transplant patients composed of 253 males and 126 females were enrolled in the study. The blood samples were collected in the period June 2021 – June 2022. The analysis of hsTnI and BNPs were performed using STAT High sensitive TroponinI kit on Alinity instrument (Abbott, Rome, Italy).
Results. The patients were grouped in accordance of the proposed cardiovascular event risk classification: 204 patients (52,1% males and 57,1% females) had a low CVE risk, 106 patients (25,6% males and 32,5% females) had an intermediate CVE risk and 69 patients (22,1% males and 10,3% females) had a high CVE risk. A correlation between BNPs and hsTnI data was observed especially in patients classified in low CVE risk group. In particular, similar percentage between males and females was observed in low and intermediate CVE risk groups. On the contrary, high CVE risk included almost 22% of males.
Conclusion. The advent of high-sensitivity cardiac troponin assays has allowed the detection of ongoing, clinically silent myocardial injury in stable outpatients with and without established cardiovascular disease.

P77

MigraMeter 2.0: Migraine Monitoring and Prediction via an optimized e-health tool based on machine learning approaches - implementation of biological variables through wearable sensors to predict migraine attacks

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Scuola di specializzazione in Neurologia

Settore Scientifico Disciplinare (SSD): MED/26

Introduction: Migraine is a high prevalent disease that poses significant burden and costs on individuals and on society. E-health tools may offer new opportunities to improve understanding and management of migraine. MigraMeter is made of a highly innovative, secure and user-friendly (e-diary App) application to collect patient-reported information and a back office, accessible by healthcare providers, to customize data collection and download data in a research suitable format.
Methods and objectives: The aim of this project is to advance our already developed e-tool to capture individual data of patients who have migraine with additional technology to obtain a new e-tool, MigraMeter 2.0, that is capable to predict occurrence of migraine attacks, course of the disease, medication overuse and drug resistance. Knowing the commonest clinical patient reported variables (like premonitory symptoms, triggers, and precipitating, predisposing, perpetuating, and protective factors) we want to integrate them with biological parameters obtained using wearable devices/sensors (eg. cardiac frequency, wake-sleep cycle, movement-physical activity), converging into an algorithm (machine learning driven) capable of predicting the onset of migraine attack. We will integrate automatically collected information obtained using smartphone and wearable devices into the current version of MigraMeter. We will investigate the contribution of MigraMeter 2.0 in a sample of patients with migraine, in two separate clinical studies: study 1 will be based on the already available MigraMeter after performing needed edits, study 2 will consider also data collected with wearable devices and smartwatches. Both studies will last 1 year. As final step of this project, we aim to plan a consortium of migraine research centers, at the international level.
Expected Results: We expect that MigraMeter 2.0 will provide the unique opportunity to predict at individual level the occurrence of migraine attacks. We also expect that MigraMeter 2.0 will detect subjects at risk of developing over time high frequency migraine, chronic migraine, medication overuse and resistance or refractoriness to treatments. This new instrument, MigraMeter 2.0, will improve the understanding of the disease and thus offer new opportunities to reduce its burden on individuals and on society.

P78

Excisional Hemorrhoidectomy Versus Dearterialization With Mucopexy for the Treatment of Grade III Hemorrhoidal Disease: The EMODART3 Multicenter Study

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Scuola di specializzazione in Chirurgia Generale

Settore Scientifico Disciplinare (SSD): MED/18

Introduction: Hemorrhoidal disease (HD) is the most common proctological disease. The most widely used classification system for HD is the Goligher classification who ranks the presence and severity of prolapse into 4 grades. For grade III HD, there are several possible surgical approaches. The traditional excisional methods remain the first choice in many centers because of the superior long-term results, but over the past several years, transanal hemorrhoidal artery ligation (HAL) or Doppler-guided HAL (DG-HAL) with mucopexy has become much more popular in this clinical setting, because seems to be associated with decreased postoperative pain, reduced postoperative events, and faster recovery than excisional hemorrhoidectomy (EH).

Materials and methods: We present a multicentre nationwide retrospective study promoted by the Italian Society of Colorectal Surgery (SICCR) to compare transanal HAL with mucopexy and conventional EH for grade III hemorrhoidal disease. The aims were to evaluate the adoption of 2 different surgical techniques and to compare them in terms of symptoms, postoperative adverse events, and recurrences at a 24-month follow-up. Any center belonging to the Italian Society of Colorectal Surgery in which at least 30 surgical procedures per year for HD were performed was able to join the study. Data from 1681 patients were analyzed. The results of both groups were comparable in terms of postoperative clinical score by multiple regression analysis and matched case–control analysis.

Results: Patients who underwent excisional hemorrhoidectomy had a significantly higher risk of postoperative complication (adjusted OR = 1.58; $p = 0.006$). A secondary analysis highlighted that EH performed with new devices and hemorrhoidal artery ligation reported a significantly lower risk for complications than excisional hemorrhoidectomy performed with traditional monopolar diathermy. At the 24-month follow-up assessment, recurrence was significantly higher in the hemorrhoidal artery ligation group (adjusted OR = 0.50; $p = 0.001$).

Discussion and Conclusions: Hemorrhoidal artery ligation is an effective option for grade III hemorrhoidal disease; however, it is burdened by a high risk of recurrences. Excisional hemorrhoidectomy performed with newer devices is competitive in terms of postoperative complications.

P79 – SELECTED ORAL COMMUNICATION

Dissecting the genetic basis of pediatric developmental epileptic encephalopathies: a multicenter study from central and southern Italy

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Scuola di specializzazione in Pediatria

Settore Scientifico Disciplinare (SSD): MED/38

Introduction: Infantile developmental epileptic encephalopathies (DEEs) include a broad variety of (frequently monogenic) conditions characterized by refractory seizures, intellectual disability (ID) and psychomotor delay and/or regression. DEEs are associated with ongoing epileptiform activity on electroencephalography (EEG) and frequent neurological comorbidities. The aetiology underlying monogenic DEEs often remains undetermined, also during this era of next-generation sequencing (NGS).
Materials and methods: In this retrospective (2016-2022) and prospective multicenter study, we used Whole Exome Sequencing (WES) to investigate a cohort of children affected by DEEs. The study was carried out in the Pediatric Departments at the: (i) University of L'Aquila, (ii) University of Catania, (iii) University of Messina, (iv) University of Salerno, (v) University of Palermo, (vi) IRCSS Oasi Troina, and (vii) Federico II University of Naples. In this context, we collected clinical and genetic data from 51 children presenting refractory seizures and neurodevelopmental delay (or regression) and diagnosed with DEEs.
Results: First epileptic manifestation was observed at a mean age of 12 months (± 14 DS), and 68,6 % of children (35/51) experienced their first seizure episode before the age of 1 year. The most frequent epileptic presentation in our cohort was West Syndrome/infantile spasms (13/51; 25,4%), followed by tonic-clonic generalized seizures (5/51, 9.8%). Trio WES led to the identification of a pathogenic variant in 24/51 (47%) children, based on the ACMG criteria. We found: (i) genes known to be implicated in DEEs (e.g., KCNA2, CDKL5, KCNQ2, GRIN1, CASK); (ii) genes involved in neurodevelopmental delay (e.g., ASXL1, SLC25A22, CNTN2) but not implicated in DEEs before; and (iii) a novel disease-causing gene (i.e., MED11) identified as a cause of DEEs and microcephaly.
Discussion and Conclusions: Defining the exact molecular diagnosis underlying DEEs is crucial to evaluating personalized strategies for the follow-up and management of these patients. In a large proportion of children from this cohort, the identification of the exact faulty gene led to individual follow-up and often targeted antiepileptic treatments. Also, we expanded the clinical and genetic spectrum associated with DEEs and identified a novel disease gene; thus, overall improving the knowledge on the molecular mechanisms associated with brain development and epilepsy in children.

P80

Blood donors with hemochromatosis: when can they be accepted? A clinical case of a donor with a heterozygous mutation

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Scuola di specializzazione in Patologia Clinica e Biochimica Clinica

Settore Scientifico Disciplinare (SSD): MED/05

Hemochromatosis is a hereditary disease determined by defects in the mechanisms regulating iron metabolism, which lead to the progressive accumulation of iron in the body and, in advanced stage, to the development of serious organ damage. Patients with hemochromatosis were previously excluded from donations. Today, in Italy, they are allowed to donate whole blood thanks to the decree of the Ministry of Health of 2 November 2015, entitled "Provisions relating to the quality and safety requirements of blood and blood components". Indeed, in the current legislation, subjects found to be carriers of hemochromatosis, with clinical documentation of absence of organ damage, can be accepted for whole blood donation. The number of donations in the year must not exceed 4 for men and menopausal women, and 2 for women of childbearing age.
The aim of this work was therefore to follow a donor with heterozygous hemochromatosis to evaluate how whole blood donations can determine the reduction of ferritin values over time, until optimal levels are reached and maintained. At the time of the aspirant course, the donor, through preliminary analyses, had high levels of ferritin. Subsequently, the mutation of the hemochromatosis gene was searched for, which was found to be mutated in heterozygosity. He was therefore eligible for donations. In conclusion, it was observed that by removing a maximum of 450 ml of whole blood at each donation, there was a progressive reduction in ferritin. In these donors, periodic donation is therefore desirable.

P81

3T RM hypervascular foci and lesions: the importance of second look ultrasound

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Scuola di specializzazione in Radiodiagnostica

Settore Scientifico Disciplinare (SSD): MED/36

Type of study: Retrospective.
Purpose: Evaluation of the reliability and sensitivity of breast MRI performed on 3T equipment in identifying hypervascular lesions and/or suspicious hypervascular foci (BIRADS 4 and 5) and the role of the second look ultrasound in their possible confirmation or downstaging.
Materials and methods: From May 2023 to December 2023, 40 hypervascular lesions >6mm and 25 hypervascular foci detected in breast MRI performed on 3T equipment, subsequently subjected to second look ultrasound, were retrospectively re-evaluated. The MRI evaluations were performed in high-risk patients as screening and/or in case of equivocal correlation of mammographic and ultrasound suspicions, in the presence or absence of a personal history of breast cancer.
Results: Of the 40 hypervascular lesions, 13 did not have a clear ultrasound counterpart and were therefore subjected to biopsy under MRI guidance at another center with the finding of 3 B2, 8 B3, 2 B5. The other hypervascular lesions detected at the second ultrasound look were biopsied under ultrasound guidance and were found to be 12 B3, 7 B5, 8 B2. At the second ultrasound look, only 8 foci were detected which however had topographic correlation with suspicious mammographic areas and therefore subjected to biopsy under stereotactic/ultrasound guidance (5 B3, 3 B5). The foci without ultrasound and mammographic evidence were followed over time with no evidence of malignancy arising during follow-up.
Conclusions: 3T MRI highlights malignant or benign hypervascular lesions with limited specificity, so the role of the second look ultrasound is frequently crucial although there is a small spectrum of hypervascular lesions without certain ultrasound correspondence for which histological verification is still necessary.

P82 – SELECTED ORAL COMMUNICATION

Anticoagulant-related intracerebral hemorrhage: no signals of improvements over 10 years in Italy

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Scuola di specializzazione in Neurologia

Settore Scientifico Disciplinare (SSD): MED/26

Background and aims: direct oral anticoagulants (DOACs) are increasingly adopted as alternatives to vitamin K antagonists (VKAs). We aimed to provide an updated account on the epidemiology of oral anticoagulant-related intracerebral hemorrhage (OAC-ICH) over a decade of increasing use of DOACs to assess whether the introduction of DOACs was associated with a change in time trends in incidence and case-fatality rates (CFRs) of ICH.
Methods: all patients with a first-ever spontaneous ICH residing in the district of L'Aquila (298,343 inhabitants) from 2011 to 2020 were prospectively included. We defined OAC-ICHs as an ICH occurring within 48 hours from intake of VKAs or DOACs. To assess whether OAC intake was independently associated with 30-day and 1-year CFR, Cox regression analyses were performed with ICH score components (age, NIHSS and Glasgow Coma Scale (GCS) scores, systolic blood pressure at ICH onset, ICH volume at onset, hemorrhage location, intraventricular extension) plus intake of OACs.
Results: we recorded 748 ICHs of whom 108 (14.4%) were OAC-related, 75 (69.4%) with VKA and 33 (30.6%) with DOAC intake, respectively. There was a non-significant trend toward an increase in OAC-ICHs from 2.35 (95% confidence interval [CI], 0.94-4.83) in 2011 to 4.02 cases per 100,000 person-years (95% CI, 2.08-7.03) in 2020 (p=0.482). Among OAC-ICHs, we observed a relative increase of DOAC-ICHs (0% in 2011; 83.3 % in 2020). 30-day and 1-year CFRs for OAC-ICHs were 48.1% and 51.9%, with a non-significant increase over time (p=0.847 and p=0.941). Univariate Cox regression revealed that OAC use was not a predictor of 30-day CFRs (HR 1.33, 95% CI, 0.98-1.80; p=0.064). GCS (HR 0.82, 95% CI, 0.68-0.97; p=0.025), non-lobar location (HR 0.45, 95% CI, 0.20-1.00; p=0.049), uncertain location (HR 3.29, 95% CI, 1.08-10.02; p=0.037) and intraventricular extension (HR 4.60, 95% CI, 1.94-10.86; p<0.001) were the only factors independently associated with 30-day CFRs for OAC-ICHs.
Discussion: during the study period, VKA-ICHs were gradually overcome by DOAC-ICHs. However, the incidence and case-fatality of OAC-ICH did not significantly change. Conclusions: despite the wider adoption of DOACs representing a safer alternative to VKAs, OAC-ICH remains a devastating complication of the use of oral anticoagulants.

P83

Characterization of Volatile Organic Compounds (VOCs) in Breath via Thermal Desorption, Gas Chromatography, and Mass Spectrometry (TD-GC-MS) for Early Diagnosis of Prostate Cancer in Patients Undergoing Multiparametric Prostate MRI and Subsequent Fusion Biopsy

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Scuola di specializzazione in Radiodiagnostica

Settore Scientifico Disciplinare (SSD): MED/36

Objective: The primary objective of this study is to identify and characterize a pattern of volatile compounds capable of discriminating, through appropriate statistical treatment, patients with prostate cancer (PCa) from healthy reference individuals.
Materials and Methods: This prospective pilot trial was conducted in the Radiodiagnostics Department of L'Aquila in collaboration with the research and development company Predict srl (Bari, Italy), between May 2022 and June 2023. Alveolar breath was collected from 147 volunteers aged between 45 and 75 years on the day of multiparametric MRI (MRmp) execution. The sample was collected before MRmp execution using Mistral technology (Predict). Molecules were analyzed using gas chromatography and mass spectrometry (TD-GC-MS). Artificial intelligence algorithms were utilized to evaluate the selection of significant variables and the precision of the breath test in identifying the group of each sample.
Results: After a qualitative selection of 257 molecules, the breath test's ability to accurately distinguish the group of each sample was tested using AI algorithms, demonstrating a predictive accuracy exceeding 74%.
Conclusions: The results obtained highlight that information about metabolic alterations induced by the presence of prostate tumor pathology is contained in the breath. The goal is to develop a new non-invasive diagnostic monitoring approach that supports and complements the gold standard reference techniques for this pathology.

P84

Unusual features in Systemic Lupus Erythematosus, a single center experience

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Scuola di specializzazione in Reumatologia

Settore Scientifico Disciplinare (SSD): MED/16

Background: Systemic lupus erythematosus (SLE) is a chronic autoimmune disease with heterogeneous clinical manifestations, also uncommon clinical features may be observed complicating the management of these patients.
Objectives: This study was designed to describe and characterize the unusual features in SLE patients referring to our rheumatology clinic. Moreover, we assessed the possible disease clinical characteristics which could be predictive of unusual manifestation development.
Methods: A 10-year retrospective evaluation of SLE patients referred to our center was performed. SLE was classified according to ACR 1997, SLICC-12, and/or EULAR/ACR 2019 criteria. Unusual manifestations were codified as a rare and/or peculiar clinical features, due to no other cause than SLE. Patients with and without these features were also compared, and by logistic regression analysis, possible predictive clinical factors were assessed.
Results: Our cohort comprised 55 SLE patients, 44 women (80%) and 11 men (20%). Average age at the time of diagnosis was 40.2 years old. Disease duration was 15.9 years (1-44), beginning from first symptom to the time of evaluation. All these patients showed positivity for ANA, 15 (28.8%) for anti- dsDNA and 17 (41.5%) hypocomplementemia. Out of 55, 23 (41,8%) of these patients exhibited at least one unusual manifestation. Specifically, 8 patients showed an unconventional involvement of nervous system (Devic's disease, retrobulbar optic neuritis, psychosis), 7 of respiratory system(lymphocytic interstitial pneumonia), 4 cardiovascular (heart attack, intracardiac thrombosis, pancarditis), 2 osteoarticular (Jaccoud's arthropathy, bone necrosis), 1 gastrointestinal (alithiasic cholecystitis), 1 mucocutaneous (ulcer), and 1 hematologic (macrophage activation syndrome). All patients with unusual features were treated with high dosages of steroids, mycophenolate mofetil, belimumab, or rituximab. Among these patients with unusual manifestations, a significantly higher percentage showed neurological diseases (16, 30.2%, p=0.027), anaemia (9, 17.7%, p=0.018), and hypocomplementemia (12, 29.3%, p=0.03). Regression multivariate analysis demonstrated that neurological features (OR: 5.35, CI95%: 1.06- 27.04, p=0.042) and hypocomplementemia (OR:7.56, CI95%:1.44-39.72, p=0.017) may be predictive factors of uncommon clinical signs.
Conclusions: We described the unusual manifestations of SLE patients referred to our rheumatology clinic. In this retrospective study, neurological diseases and hypocomplementemia resulted could be predictive factors for those uncommon features.

P85 – SELECTED ORAL COMMUNICATION

The role of renal graft preimplantation biopsy: experience of our center

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Scuola di specializzazione in Chirurgia generale

Settore Scientifico Disciplinare (SSD): MED/18

Introduction: Kidney transplantation is currently the most effective therapy for end stage renal disease. Organ shortage is nowadays the main constraint to kidney transplantation, In order to increase the donor pool and the chance of transplantation to patients on wait list, most transplant programs are increasingly accepting suboptimal, so called "extended criteria," donors (ECD). Preimplantation biopsy (BRPI) is used to assess the state of the renal graft, identifying any abnormalities or pathologies that could affect the outcome of the transplant. Indications to BRPI are: older donors (>60y), persons aged 50–59 years with hypertension, SCr>1.5 mg/dL or death from cerebrovascular accidentor, donor with kidney disease or with high immunological risk. Technical procedures are: wedge, needle core or punch biopsy. BRPI is defined appropriate when the diameter is 1 cm, it includes at least 25 glomeruli and 2 arteries, adequate number of tubules and arterioles. The scoring system mostly used is the Karpinski Score.

Materials and methods: We present the retrospective study performed at our center on the role of BRPI on the long-term outcome of kidney transplantation. 152 kidneys from 76 cadaver donors who died between February 2015 and September 2020 were considered. BRPI was performed with the wedge resection technique. Indications to biopsy were: Age> at 60 years or <at 60 years in the presence of comorbidities. The number of discarded kidneys and of transplanted kidneys were evaluated.

Results: The results were analyzed by evaluating serum creatinine at 7 days, 30 days, 60 days, 12 months, 19 months, 24 months, 36 months and the last follow up at 48 months, comparing with Karpinski score preimplantation. The score 0-1 is associated with a greater temporal stability of creatinine and therefore a better outcome. Only 6% of the 90 recipients returned to dialysis and only 4% of patients died.

Discussions: The use of marginal donors have been considered but it still controversial for the lower longevity of the transplant and the need to expand the donor pool.

Conclusions: Our study confirms the usefulness of pre-implantation biopsy for the assessment of chronic damage in ECDs, suggesting it could be performed as routine.

P86

An unusual case of gastroduodenal occlusion: a rare tale of a hairy tail

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Scuola di specializzazione in Chirurgia Generale

Settore Scientifico Disciplinare (SSD): MED/18

Introduction: Gastric trichobezoars are a rare form of bezoar formed from swallowing humain or othersources of hair , due to a psychiatric disorder named trichophagia and trichotillomania.
Case Report: A case of a 26 year orld girl with a backgroung of serious loss of hairs and associated anorexia,admitted into the emergency room with acute abdominal pain, vomiting and fever.
Discussion: Clinical examination revealed the presence of a mobile and sensitive abdominal mass witch filled the upper audrant. Blood test were negative. An abdominal CT scan showed a heterogeneous mass occupying the whole stomach cavity with extension into the third portion of the duodenum. The patient's mother reports many dermatological visits for several hair loss and no other problems in her medical story.
The removel of the trichobezoar endoscopically it was not possible. She was subjected to an exploratory laparotomy. An antral gastrostomy were performed and a 20 x10 cm trichobezoar was extracted.
Conclusion: Rapunzle syndrome is an uncommon trichobezoar, it's commonly found in young females usually with an underlying prichiatric disorder. Management requires gastrostomy. The patient was discharged after a week of hospitalization in excellent clinical conditions. A psychiatric assessment and long term follow up are advocated as a regular part or treatment to prevent recurrence.

P87

Multidisciplinar approach to complex pelvic floor prolapse

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Settore Scientifico Disciplinare (SSD): MED/18

Introduction – Disorders of the pelvic floor are frequently characterized by rectal and genital organs'prolapse and/or disturbed function (bowel incontinence/ obstruction symptomps, bulge, bladder incontinence or urinary retention), and lastly significant patient distress and reduced Quality of Life. In such instances, a complex surgical strategy is mandatory with the aim of combining different repair of pelvic prolapse.
Materials and Methods – In our Specialized University unit of Surgical Coloproctology we established a protocol of treatment in case of multiple involment of pelvic organs involving urogynecologists in the surgical treatment. In the period 2012 – 2022, 145 patients underwent surgical treatment of Pelvic Complex Prolapse. 23 males, 122 females of those, 74 women presented symptoms of obstructive defecation and vaginal prolapse/vescical prolapse.
68 reported some degree of fecal incontinence. After physical evaluation with the uro-gynecologist and after evaluation with Wexner's Score for obstructed defecation and defecating proctogram, surgical indication was posed as follows: Delorme proctopexy, 31 patients; Resection rectopexy, 14 patients; Stapled Transanal rectal resection (STARR) , 43 patients; STARR+ Colpopexy, 11 patients.
Results – Results of all surgical procedures were characterized by a negligible amount of post - operative complication: 1 bleeding after Delorme, 2 post-operative intestinal obstruction after Resection-Rectopexy. None required re-intervention. STARR + Colpopexy was conducted by two surgical teams (urogynecologists and colorectal) according to standard techniques with nihl post-operative morbidity. **Conclusion** – Multidisciplinary approach and combined surgery in case of complex disorders of the pelvic floor is feasible with low morbidity. In a single combined operating session, the problem of multicompartment prolapse is solved by offering the advantages of a reduced risk of anesthesia, a single hospital stay and fast recovery period, reduced post operative pain. Improvement of urinary, vaginal and bowel function has been reported immediately in the early post-operative period. Long term follow up is necessary to evaluate functional results and improvement in Quality of Life.

P88

Assessment of the glymphatic system function in patients with essential tremor and Parkinson's Disease who are eligible for thalamotomy via MR-guided Focus Ultrasound (MRgFUS)

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Settore Scientifico Disciplinare (SSD): MED/36

Background: Tremor is the most disabling symptom in essential tremor and Parkinson's disease. Evidence suggests the involvement of the glymphatic system in the pathophysiology of neurodegenerative diseases such as Parkinson's.
Objectives: This study aims to assess the presence of glymphatic system alterations in patients with ET and PD through the analysis of diffusion tensors along perivascular spaces (DTI-ALPS index).
Materials and Methods: we retrospectively evaluated 35 patients (19 PD, 16 TE, 28 males, mean age 67 years) who were eligible for Vim thalamotomy using MRgFUS (2018-2022) and 17 healthy controls (13 female, mean age 39 years). DTI images were analyzed using open-source software. The ALPS indices were compared between the ET and PD patient populations, respectively. Both these populations were compared to healthy controls.
Results: PD and ET groups didn't show statistically significant differences in clinical parameters (disease duration, age, gender, cognitive score). The ALPS index in PD patients showed a mean of 1.33, with no statistically significant differences compared to ET patients (1.31, p-value = 0.31), both reduced compared to healthy controls (1.62; p-value < 0.001). We observed a more intense tremor in ET patients rather than in PD patients (FTM 34,14 VS 28,64; p-value = 0.041). There was a significant correlation between the ALPS index and tremor intensity in ET patients (R = -0.76; p-value < 0.001).
Conclusions: There were no differences observed in the MRI indices of the glymphatic system between ET and PD patients, and both are reduced in confront of healthy subjects. Our results suggest the need for further studies to better define the role of the ALPS index as a marker of disease progression and to evaluate the possibility of neuro-degenerative physiopathology in the Essential Tremor.

P89

Long-term iloprost-treated patients with SSc showed a low occurrence of vascular manifestations: findings from a single center study

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Settore Scientifico Disciplinare (SSD): MED/16

Background: Systemic sclerosis (SSc) is a connective tissue disease characterized by inflammation, fibrosis, and vascular dysfunction. The latter is associated with severe complications such as digital ulceration (DU), necrotizing raynaud phenomenon (RP), scleroderma renal crisis, and pulmonary arterial hypertension (PAH). All these manifestations significantly contribute to morbidity and mortality in SSc. Vasodilating agents, such as prostanoids and mostly iloprost, represent a therapeutic option in the treatment of SSc vasculopathy. However, a standardized therapeutic administration strategy of these drugs and their efficacy in long-term are to be fully determined.
Objectives: To evaluate the cumulative incidence of SSc-related vascular complications in patients treated with a monthly infusion of iloprost [Wigley FM. J Rheumatol. 1992;19:1407-14] and to compare the results with available evidence (EUSTAR data). Methods: We retrospectively evaluated 102 patients fulfilling the 2013 EULAR criteria for SSc prospectively followed-up in our center from January 1st 2004 to January 31st 2024 and treated with a monthly administration of iloprost with a focus on the incidence of vascular manifestations.
Results: Out of 102 patients with SSc 96 were female; 83 (81.3%) showed typical SSc autoantibodies, specifically 27 (26.5%) anti-topoisomerase I, 55 (54.0%), anti-centromere and 1 (0,9%) anti-RNA polymerase III. Among assessed patients, 59 presented vascular complication before the administration of iloprost, specifically 56 DUs (54.9%), 11 necrotizing RP (10.8%), 9 PAH (8.8%), 1 renal crisis (0.9%). The mean duration of iloprost treatment was 6.4 ± 5.0 years. Concerning the incidence of new vascular complications, we registered the occurrence of new DUs in 30 patients (29.4%), all characterised by a clinical low severity, the development of PAH in 6 patients (5.8%), and no new event of necrotizing RP, or renal crisis. Finally, these findings may descriptively suggest a lower incidence of vascular complications during a long-term iloprost administration in our SSc cohort than available literature. In EUSTAR dataset, the occurrence of DUs is reported in almost 50% of SSc patients, PAH in 13% and scleroderma renal crisis in 10% of SSc patients.
Conclusions: SSc patients treated with infusional long-term iloprost regimen presented a decreased cumulative incidence of vascular complications, lower than available evidence.

P90

A nail psoriasis study integrating clinical and high frequency ultrasonographic evaluation

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Settore Scientifico Disciplinare (SSD): MED/35

Introduction: Plaque type psoriasis (PSO) is a chronic immune-mediated inflammatory disease, characterized by consistent morbidity and quality of life impact. Disease progression may lead to nail and joint disease. Nail involvement is esteemed to be present in up to 70% of the patients affected by PSO, with a broad spectrum of clinical characteristics. Recent studies have revealed the importance of nail involvement in the transitioning from PSO to psoriatic arthritis (PsA).
Aim: To evaluate morphostructural aspects and nail vascularity throughout high frequency ultrasonography (HFUS) in the nail unit of patients with PSO in the context of a multidimensional assessment, and to evaluate whether there are differences among psoriatic patients with/without PsA and with/without nail involvement.
Material and Methods: Patients with PSO and PsA were evaluated and compared as well as patients with/without diagnosis of nail psoriasis in a cross-sectional single centre study. Nail abnormalities were evaluated by HFUS using high frequency probes (27 MHz). After a descriptive assessment, the nail-psoriasis-and-severity-index NAPSI and the Brown-University-Nail-Enthesis- Scale (BUNES) were used to clinically and ultrasonographically assess nails.
Results: Sixty patients were enrolled (23/60 PSO-37/60 PsA). HFUS evaluation identified nail alterations characterized by thickened matrix, inhomogeneous echogenicity of the nail bed, and increased blood flow by PD. The sample was characterized by a majority of female patients (63.3%), mean BMI 30.5 ± 27.2, mean disease duration of 12.8 ± 12.3, mean PASI 6.8 ± 7.6, mean NAPSI 19.3 ± 16.4, mean PEST 2.4 ±1.6, mean BUNES morphometry 1.6 ± 0.5, mean BUNES PD 2.4 ± 2.1. Among the studied sample, 43/60 (71.7%) presented nail psoriasis, with a major proportion (69.7%) presenting a coexisting PsA as compared to PSO patient (30.3%) ($p = 0.04$) confirming the strong link between nail psoriasis and PsA. Besides, the statistical analysis showed significant correlation between NAPSI and BMI, BSA, PGA, PEST, BUNES. **Conclusion:** Nail abnormalities were demonstrated by HFUS, suggesting a practical additional tool to be used in clinical setting. Although further studies are needed to demonstrate the role of nail involvement in the transition process from PSO to PsA, our results confirm the association between nail psoriasis/NAPSI and PsA.

P91

Sentinel lymph node biopsy in malignant melanoma of the skin, comparative analysis between standard technique with technetium99m and indiocyanin green. Experience of a high-flow regional reference center

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Settore Scientifico Disciplinare (SSD): MED/18

Introduction: In cutaneous melanoma as well as other solid organ tumors lymphatic spread is a fundamental prognostic factor. Sentinel lymph node biopsy (SLNB) with preoperative lymphoscintigraphy and technetium99m (Tc99m) is currently the gold standard, however the use of indocyanine green (ICG), as reported in several recent studies, is a promising option for mapping the Sentinel lymph node (SLN) in melanoma. The main aim of our study is to compare this method with radio tracer and the technique with the fluorescent dye ICG considering if it as a valid alternative nonradioactive for sentinel lymph node labeling.
Materials & Methods: We conducted a prospective observational study in a single center by a single operator, at the San Salvatore Hospital in L'Aquila in a period between March 2017 and May 2023. 113 patients diagnosed with malignant melanoma 0.8 mm thick without clinical- radiological evidence of metastasis were consecutively included in the study. The comparative statistical analysis between the Tc99m Vs ICG procedure was obtained through Concordance Rates Analysis. The impact of some covariates related to ICG use was assessed with multivariable statistical models. All patients in the study period underwent SLNB with the standard technique with Tc99m on with an additional application of a near infrared fluorescent dye. 127 lymph nodes were removed, 77 located in the groin and 50 in the axillary cavity.
Results: Preoperative (before skin incision) visualization of the SLN by ICG was successful in only 36,8 of 127 lymph node basins (29%). The number of SLNs identified using the near infrared fluorescence technique in the intraoperative (after skin incision and initial tissue preparation) stage was 116 of 127 (92%). In the multivariate analysis the inguinal location and low BMI in the preoperative phase and BMI alone in the intraoperative phase were statistically related a correct response with ICG.
Conclusions: The SLNB in melanoma with Tc99m remains the gold standard at present, however the undoubted advantages of the fluorescence technique alone, the speed of execution, the lower costs and the absence of radiation, are not to be overlooked even in light the development of improved dyes and new fluorescence detection technologies.

P92

Disturbi dello spettro autistico e comorbidità psichiatriche: una revisione narrativa

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Settore Scientifico Disciplinare (SSD): MED/25

Background. Autism spectrum disorders (ASD) are characterized by persistent difficulties in sociality, restricted interests, repetitive behaviors. Diagnosis is difficult because of the heterogeneity of symptoms among individuals. ASD people are also more likely to experience psychiatric comorbidities, worsening the clinical course of ASD. Distinguishing comorbidities symptoms from ASD's one can be difficult. Aim of this narrative review is to make an overview of psychiatric comorbidities of ASD, updating prevalence, highlighting symptoms and available diagnostic tools and defining possible treatment strategies.
Methods. This narrative review was conducted by defining the objective, researching the scientific literature and data evaluation and then presenting results. Search terms were entered into ERIC, MEDLINE, PsycARTICLES, PsycINFO, Scopus, and PubMed. Only studies on children, adolescents, and adults, published in English, which analyzed autistic traits in severe mental disorders and studies of patients with ASD and severe mental disorders in comorbidity were included. Given the breadth of the topic, studies on psychosis, schizophrenia, gender dysphoria, trauma and post-traumatic stress disorder, substance use, and suicidal behavior were not included. Terms and databases were combined using the Boolean search technique to make the search more restrictive and detailed. Tables were then constructed, and results sorted by prevalence, symptoms, diagnostic tools and treatment and discussed in a narrative way.
Results. Studies were divided into eight categories: ADHD; anxiety disorders; personality disorder; repetitive behaviors and OCD; sleep disorders; mood disorders; Tourette and tic disorder studies; feeding and eating disorder studies. ADHD has the highest prevalence among psychiatric comorbidities in ASD, followed by anxiety disorders. The clinical presentation is often overlapping with symptomatology of ASD, although some peculiarities help clinicians to recognize the comorbidity. Assessment tools available for the diagnosis of psychiatric disorders in ASD are often not adjusted. The co-occurrence of these disorders is usually associated to a worsening of the clinical presentation and to a resistance to conventional treatments. Among non-pharmacological treatments, cognitive behavioral therapy seems to be effective in almost all the psychiatric disorders evaluated in this study.
Conclusions. Prospective studies with homogeneous samples are needed to develop specific diagnostic tools and dedicated treatments for psychiatric comorbidities in ASD

P93

Temperature, neutrophils and multiple organ failure (tnm) score in the early prediction of mortality in acute pancreatitis

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Settore Scientifico Disciplinare (SSD): MED/18

Introduction: The grading systems for acute pancreatitis (AP) are not employed commonly in clinical practice because they are too complicated or inability to obtain a final score until 48 hours after admission (Ranson criteria). We suggest to grade the severity of AP with a simple system, TNM: Temperature, Neutrophils and Multiple organ failure (MOF). This study evaluates the predictive value of TNM on mortality of patients with AP.
Methods: TNM was developed in a training cohort of 176 patients with AP from April 2006 to December 2013 (retrospective data). To verify the prognostic value of TNM, we prospectively recruited 181 patients treated from January 2014 to November 2020 as the validation cohort. After defining the categories T, N and M, patients were grouped in stages (0-IV). We analyzed specific variables for their relation to death: age, gender, ASA, blood transfusion, temperature, neutrophils, organ failure, immune-compromised status, stage 0-IV.
Results: In the training cohort 32 patients were in stage 0; 52 in stage I; 49 in stage II; 26 in stage III; 17 in stage IV. Death occurred in 14 (7.9%) patients. ASA score (III-IV), neutrophils, organ failure, stage III-IV emerged as statistically significant different prognostic factors. ASA score (III-IV) and stage (III-IV) were significant independent predictors of postoperative death in multivariate analysis. Comparable results were observed in the validation cohort.
Conclusions: AP is a dynamic process and TNM is a dynamic and simple score. TNM helps to define the mortality risk, and is useful to objectively compare patients with AP.

P94

Sarcopenia and Visceral Adipose Tissue as Risk Factors for Incisional Hernia

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Settore Scientifico Disciplinare (SSD): MED/18

Purpose: Incisional hernia, defined as a failure of the abdominal wall at a site of previous surgical scar, is a notable complication post-laparotomy, particularly in emergency median incisions. The aim of this study was to assess the impact of radiologically determined visceral fat and skeletal muscle mass deficit in the development of incisional hernias.
Materials and Methods: A retrospective analysis was conducted on patients who underwent emergency median xiphopubic laparotomy and continuous suture repair, from 2017 to 2020, in our center. Various variables were considered and measured based on CT scans from the initial surgery, comparing groups of patients with and without incisional hernias.
Results: Out of 222 emergency median laparotomy patients over 18 years old, 85 met the study's inclusion criteria. Statistically significant differences were observed between the two groups regarding obesity and visceral fat area (VFA, cutoff > 130 cm2), but not for sarcopenia (cutoffs: <41 cm2/m2 for women; <43 cm2/m2 for men with BMI < 25; <53 cm2/m2 for men with BMI > 25).
Conclusions: Visceral fat area emerged as the sole factor linked to the development of incisional hernia. It also proves to be a more reliable criterion than body mass index due to its easy measurability through preoperative CT scans. Future studies with larger sample sizes, categorized by surgical diagnosis, are recommended to minimize selection bias and further validate these findings.

P95

Cancer mortality after kidney transplantation: a multicenter cohort study in Italy

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Settore Scientifico Disciplinare (SSD): MED/18

Introduction: The Transplant center of L'Aquila join a multicenter cohort study with the aim to assess whether cancer mortality among Italian kidney transplant (KT) recipients was higher than in the corresponding general population. Amoung Kidney transplant (KT) recipients, cancer was the most common cause of death constituting 32.4% of all deaths. KT recipients are known to be at risk of developing several cancer types; however, cancer mortality in this population is underinvestigated.
Materials and Methods: A cohort study was conducted among 7373 individuals who underwent KT between 2003 and 2020 in 17 Italian centers. Cancer was the most common cause of death among the 7373 KT recipients, constituting 32.4% of all deaths, followed by cardiovascular disease (23.0%) and genitourinary disease (14.3%).
Results: A 1.8-fold excess mortality (95% CI: 1.59-2.09) was observed for all cancers combined. Lymphomas (SMR = 6.17, 95% CI: 3.81-9.25), kidney cancer (SMR = 5.44, 95% CI: 2.97-8.88) and skin melanoma (SMR = 3.19, 95% CI: 1.03-6.98) showed the highest excess death risks. In addition, SMRs were increased about 1.6 to 3.0 times for cancers of lung, breast, bladder and other hematopoietic and lymphoid tissues. As compared to the general population, relative cancer mortality risk remained significantly elevated in all age groups though it decreased with increasing age. A linear temporal increase in SMR over time was documented for all cancers combined ($P < .01$). Our study documented significantly higher risks of cancer death in KT recipients than in the corresponding general population.
Discussion and Conclusion: Prevention and screening play an important role in reducing the cancer burden in this at-risk population. Although routine cancer screening is recommended for all individuals undergoing KT, current surveillance strategies are largely based on data from the general population while they should also be individualized based on the risk factors specific for the transplant population. Our results support further investigation into the prevention and early detection of cancer in KT recipients.

P96 – SELECTED ORAL COMMUNICATION

Evaluation of extra-cellular myocardial volume in CT in patients with gastrointestinal cancer disease

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Settore Scientifico Disciplinare (SSD): MED/37

The aim of our study is to evaluate the change in myocardial extracellular volume (ECV) over time, using thoracic computed tomography without and with contrast, performed for staging and follow-up, in patients with gastric cancer, pancreatic and colorectal (GI) treated with chemotherapy.
Materials and methods: 83 patients with GI tumors were selected from July 2021 (including 68 males and 49 females, mean age 64.9 ±13 years), subjected to a first staging CT examination at the beginning of treatment and, re-evaluation CT oncological, after therapy with anti-HER, antiVEGF/VEGFR and fluoropyrimidine drugs. The study was performed with 320 detector row CT.
The results available to us, although still limited, nevertheless demonstrate a statistically significant increase in the ECV value which represents the myocardial damage induced by chemotherapy treatment.

P97

Myasthenia Gravis and Bipolar Disorder clinical presentation, diagnosis, and pharmacological management in a 26-year-old female: a case report and mini review

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Settore Scientifico Disciplinare (SSD): MED/25

A 26-year-old female was admitted to the psychiatric ward after manifesting a symptomatology characterized by psychomotor alterations, delusions, hallucinations, disorganized thinking, and grossly disorganized behavior. Such symptomatology began while she was hospitalized in a neurology ward because of symptoms suggestive of myasthenia gravis (MG), a diagnosis later confirmed by clinical and laboratory investigations, completed while she was already in the psychiatric ward. This peculiar comorbidity challenged the clinicians in the choice of a pharmacological treatment, as it was necessary to consider that both diagnoses excluded the possibility to employ specific and appropriate drugs. The patient was discharged after 69 days of hospitalization with a diagnosis of "bipolar disorder with mood-congruent psychotic features and catatonia", and pharmacological treatment was set up upon both the neurological and psychiatric diagnosis. Subsequent radiological investigations confirmed the diagnostic suspicion of a thymic hyperplasia, which was surgically treated with a thymectomy. After the surgical procedure, the patient manifested complete remission of symptoms related to both diagnoses and return to a premorbid level of social, study, and daily functioning. Several hypotheses were examined during the hospitalization to perform an accurate differential diagnosis and avoid pitfalls in pharmacological management. The first and most probable hypothesis contemplated a CNS involvement in MG as a paraneoplastic syndrome that could explain the psychiatric symptomatology. On the other hand, another hypothesis was grounded on the possibility that the employment of specific psychotropic drugs exacerbated the MG symptoms. It also seemed necessary to include the possibility of a spurious correlation between the onset of the neurologic and psychiatric pathology. In this article, we aim to delineate challenges encountered by clinicians in the diagnostic and treatment process while conducting a rapid review of the literature available on this topic. Common and shared guidelines regarding the clinical management of such complex comorbidities are still lacking, and further studies seem necessary to fill this gap to improve our possibility as clinicians to manage similar clinical cases.

P98

Benign painful bone lesions of the elbow: treatment evaluation of a single center by interventional radiology with RFA and MRgFUS

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Settore Scientifico Disciplinare (SSD): MED/36

Purpose: Interventional radiology is the gold standard for the treatment of osteoblastoma and osteoid osteoma. The main interventional radiology techniques include radiofrequency ablation (RFA) and focused magnetic resonance guided ultrasound (MRgFUS). The main difference between these two types of procedure concerns the minimal invasiveness of the MRgFUS which guarantees the ablation of the lesion without using needles.
Materials and Methods: We included 8 patients, 6 of were treated with MRgFUS and 2 with RFA. All patients were studied with both MRI and CT scan and the pain evaluation was considered in terms of the VAS score (Visual Analog Scale). The improvement in pain relief and functional recovery were evaluated with a clinical follow up to 6 years.
Results: All patients showed a significant reduction of pain (VAS 8 pretreatment - VAS 0 post treatment) and improvement of the functional limitation during short- and long-term follow-up. As the only complication we observed the fracture of the humerus but, even in this case, no treatment was required.
Conclusions: Both interventional radiology treatments have proven to be safe and effective with a complete success in terms of pain relief and functional recovery.

P99 – SELECTED ORAL COMMUNICATION

Mental health between the Covid-19 pandemic and the 2009 earthquake in L'Aquila. Changes, socio-demographic and clinical characteristics of the users of the University Psychiatric Service of Diagnosis and Treatment of the "San Salvatore" Hospital in L'Aquila

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Settore Scientifico Disciplinare (SSD): MED/25

Objective: Traumatic events can affect recovery patterns in the psychiatric ward. The objective of this cross-sectional study is to analyze the trend of admissions at the SPUDC of the Hospital of L'Aquila in the five years 2007-2011 and 2018-2022, with focus on the second quarter of 2009 (L'Aquila earthquake) and 2020 (lockdown from COVID-19).
Materials and Methods: Data from hospital discharge records of adult patients with psychotic disorders, depression, bipolar disorder and substance use in the five-year periods 2007- 2011 (n=1627) and 2018-2022 (n=1404). The average number of weekly admissions for the 20 quarters of each five-year period was analysed by means of ANOVAs. To assess possible differences in each five-year period according to gender, diagnosis and age, the average weekly hospitalisations in the year before the traumatic event were compared with those of the year including the earthquake/lockdown through a two-factor ANOVA (Pre-post-earthquake/lockdown + Gender/Diagnosis/Age).
Results: The ANOVA for both quinquennials showed significant differences in the number of weekly accesses between quarters (both $p<0.001$). Weekly admissions in the earthquake quarter were significantly lower than in previous quarters (all $p<0.001$). Three quarters after the earthquake, the number of admissions remained stable (all $p>0.550$) and increased again from quarter 14 (all $p<0.048$). For the five-year period 2018-2022, no significant differences appeared between the lockdown quarter and the others (all $p>0.075$), except for an increase in the quarters 17 ($p<0.001$) and 18 ($p=0.049$). Significant interactions came to light between the Earthquake Pre-post factor and the Diagnosis factors ($p<0.001$) and Age ($p=0.027$). Post hoc comparisons showed a greater reduction in admissions for psychotic disorders ($p<0.001$) compared to other diagnoses (all $p<0.039$), while the age group 18 -25 was the only one not to show a decrease ($p=0.592$) compared to the others (all $p<0.028$).
Conclusion: The post-earthquake year was associated with a transient reduction in admissions, probably attributable to the population's developed post-traumatic resilience. This decrease is more evident for psychotic disorders. In the five-year period 2018-2022, the number of admissions remained generally stable, except for a substantial increase in the first half of 2022, suggesting possible long-term effects of the pandemic period.

P100

Synchronous breast cancer with different tumour histology. A case report

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Settore Scientifico Disciplinare (SSD): MED/18

Introduction: Advances in imaging and screening standardization have led to an increase in diagnoses of unilateral, contralateral breast cancer both synchronous and metachronous. Diagnosis is based on clinical examination supported by mammography, ultrasound and second-level examinations such as Magnetic Resonance Imaging (MRI), which has been shown to be more accurate in detecting multifocality and bilaterality.
Case report: We present the case of a 48-year-old woman with a palpable swelling of the right breast's Upper Outer Quadrant (UOQ). Bilateral ultrasound and mammography were negative. The diagnosis of an invasive lobular carcinoma was made by a biopsy. Given the typical bilaterality of this type of carcinoma the patient underwent a MRI with contrast, which showed an area that also affects the controlateral Lower Outer Quadrant (LOQ). It was decided to perform a biopsy on the contralateral area, which resulted a ductal carcinoma in situ. The dilemma with this case of two histological different synchronous neoplasm was what treatment the patient should undergo. The volume of the right neoplasm and the risk of multifocality required a mastectomy while the localization in situ of the left neoplasm required a conservative treatment. However the patient's wish of a contextual bilateral breast reduction finally led us to perform a bilateral skin reducing mastectomy. Hormone therapy followed. Follow-up 6 months.
Discussion: Biologically different synchronous breast cancer (SBBC) is a difficult clinical challenge. Several studies have shown that the prognosis of SBBCs depends strictly on the aggressiveness of the worst neoplasm, how it is worse than that of metachronous neoplasms and how the choice of treatment changes. There is evidence that both quadrantectomy and mastectomy are effective in terms of prognosis and survival.
Conclusion: It was decided to perform a bilateral demolition with mastectomy and contestual reconstruction with prosthesis, followed by hormonal therapy and with the advantage of not performing RT. After consulting the data of literature, the therapeutic choice was discussed to a multidisciplinary oncology board.

P101 – SELECTED ORAL COMMUNICATION

Dosimetric comparison of organs at risk using two delineation guidelines for the radiation treatment of cT1 laryngeal cancer

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Purpose: A number of contouring methods have been used for the delineation of the primary tumor clinical target volume (CTV-P) in Head and Neck Cancers. They include the "anatomic method" proposed by Lapeyre and colleagues the so called French contouring methods. More recently an international guidelines proposed by Gregoire and colleagues tried to integrate the anatomical method with the "geometric methods" proposed by the DAHANCA group in a single contouring method. This new contouring guideline may have the interesting characteristic to obtain reduced contouring volumes with respect to the French method with the potential to spare the surrounding organs at risk (OARs) especially in cT1 GC. This specific aspect is important since factor influencing the choice between radiotherapy and for the treatment of glottic cancer (GC) is focused not just on oncological outcome but also on reduction of treatment related toxicity. Our objective is to compare the OARs sparing properties of plans generated by VMAT of the French and the International consensus contouring methods in laryngeal SCC.
Methods: Ten CT of patients with T1 GC were contoured using the two contouring guidelines. For all cases, the GTV was delineated by a single radiation oncologist. The OARs were outlined and VMAT plans were generated. Dosimetric parameters of OARs, carotid arteries (CAs) and carotid bulbs (CBs) were compared.
Results: The target volumes contoured by the two delineation methods were VMAT plans translated into dose distributions favoring the International guidelines. Better dosimetric sparing was observed for spinal, supraglottic larynx, cricopharyngeal inlet, thyroid gland and medium and inferior PCM. Mean ipsilateral CBs and CAs D0,1cc were lower in VMAT plans generated from the International guidelines. Similarly, contralateral CBs and CAs D0,1cc, Dmean and V35 were spared after the use of the International guidelines.
Conclusions: The use of the International guidelines of Gregoire and colleagues translated into dosimetric advantages in OARs sparing showing large differences in CTV delineation between the 2 delineation methods.

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Clinical and functional outcomes of subtotal colectomy for slow transit constipation

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Background: Slow transit constipation is a rare condition that is more commonly observed in middle-aged women. The pathophysiology and aetiology are poorly understood, but a multifactorial pathogenesis seems likely and in its most severe form, total colectomy with ileorectal anastomosis is the final treatment option. In this study we present a long-term clinical and functional outcomes in patients who had surgery for laxative-resistant slow transit constipation.

Methods: A retrospective study on prospectively collected data of 14 patients treated with subtotal colectomy with ileorectal anastomosis between January 2013 and April 2023 was performed to assess bowel frequency, PAC-QOL score, satisfaction with procedure, likelihood to choose the procedure again.

Results: 12 patients (male =2) with mean age of 55,85 years (45-76 yrs.) were available for follow-up out of an initial cohort of 16 with mean follow-up time was 50,0 months (range 12-120). Mean time since surgery was 9,3 years (range 4-15). There was a sharp improvement in bowel movements ($p<0.05$ from mean 1 in 9 days to 2,8 per day), Bristol stool type (from 83,3% with type 1 to 8,33% type 1) and in PaCQoL score ($p<0,05$ from mean 91 to mean 49). Satisfaction and likelihood to choose surgery were 91,6% where one female patient has more than 5 daily evacuation and one male patient still needs laxatives with 4 bowel movements per week. Two patients had early postoperative complication; thus, it was performed loop ileostomy. Two patients had a late complications like incisional hernia and one female patient has a blind loop syndrome.

Conclusions: In conclusion subtotal colectomy results in satisfactory functional results in patients with severe slow-transit constipation. Surgery, is the only viable alternative in these patients, where neither laxatives nor enemas can provide an acceptable quality of life. It is clear that meticulous selection of patients for subtotal colectomy is mandatory. However, our results bear limitations due to small sample of patients, to retrospective nature of the investigation and being single-centre, single-operator.

P103

Late assessment of genitourinary, gastrointestinal and vaginal toxicity in women treated with vaginal brachytherapy or pelvic radiotherapy plus vaginal brachytherapy in Stage I endometrial cancer

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Aim: To study acute the late toxicity of women receiving vaginal brachytherapy (VB) versus pelvic radiotherapy plus vaginal brachytherapy (RT-VB) in Stage I endometrial cancer (EC).
Methods: The records of 191 women with postoperative Stage I EC and subsequently treated with vaginal brachytherapy (7Gy/3 fractions or 5.5Gy/4 fractions) (71 ptz.) or pelvic radiotherapy (45 to 50.4 Gy over 5 weeks) plus vaginal brachytherapy (5Gy/2 fractions) (120 ptz.) were analyzed. Tolerance to treatment was graded according to RTOG scale, for late GU and GI toxicity. Differences in toxicities were compared using Fisher exact test. Significance was defined as two tailed p-value <0.05.
Results: Nine patients (7,5%) in the group treated with RT-VB [mean follow-up: 45, 9 months (95%CI 44,1-54.7)] and 1 patient (1,4%) in the group treated with VB [mean follow-up: 36,9 months (95%CI 31,9-41,9)] suffered from G2 or higher late GI toxicity (p=0,42). Grade 3 (0,8%) and grade 4 (3,3%) late GI toxicity was only observed in the group treated with RT-VB. Four patients (3,3%) in the group treated with RT-VB and 1 patient (1,4%) in the group treated with VB suffered from G2 or higher late GU toxicity (p=0,65). Similarly to the late GI toxicity, grade 3 (0,8%) but not grade 4 late GU toxicity was only observed in the group treated with RT-VB. Thirteen patients (10,8%) in the group treated with RT-VB and 5 patient (7,2%) in the group treated with VB suffered from G2 or higher vaginal toxicity (p=0,45). Grade 3 late vaginal toxicity was observed in 7 out of 120 (5,8%) patients treated with RT-VB and in 1 patient treated with VB, respectively (p=0,26). Vaginal recurrence and distant metastasis rate was 2,8% for both indicators [mean follow-up of 36,9 months (95%CI 31,9-41,9)] in the group treated with VB and 2,5% and 3,3% in the group treated with RT-VB [mean follow-up: 45,9 months (95%CI 44,1-54.7)].
Conclusion: The low rate of relevant late GI, GU and vaginal toxicity seems to indicate that the combined use of pelvic radiotherapy with VB is well tolerated. Superiority of RT-VB compared with VB was not demonstrated.

P104

Anal Fissure in Immunocompromised and solid organ Recipients

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Chronic anal fissure is a common clinical finding. Anal fissures may manifest in individuals with compromised immune systems, such as those affected by conditions like HIV/AIDS, organ transplantation, specific medical treatments like chemotherapy or radiotherapy for neoplasm and patients with lymphoproliferative disorders (LPD). The occurrence of anal fissures in immunocompromised individuals presents unique challenges due to underlying health conditions and potential complications.These patients are more susceptible to infections, including perianal infections, which predispose them to the development of abscesses and fissures. Pathogens involved encompass bacteria, fungi, and viruses. Gram-negative bacilli, followed by gram-positive cocci, are the most commonly isolated bacteria. Candida represents the predominant fungus, while herpes simplex type 2 is the most prevalent virus. Cutaneous manifestations, albeit rare, may arise from Cytomegalovirus, inducing ischemic changes in vessels. Immunocompromised patients may also encounter challenges with aciclovir-resistant HSV strains, drugs such as foscarnet,vidarabine and ciclofovir are possible therapeutic options.Obtaining an etiological diagnosis is crucial, involving swab analysis for bacteria and PCR analysis for viruses. Multi-antibiotic resistance of microorganisms complicates therapeutic decisions. In immunocompromised/immunosuppressed patients with anal fissures, antibiotic therapy may be preferable to surgical treatment, considering the heightened risks and complications associated with surgery in this population. Medical treatment should precede any surgical intervention, recognizing the challenges of surgical procedures, including the risks of sepsis and delayed healing.Immunocompromised patients necessitate personalized and multidisciplinary care. Collaborative efforts among healthcare providers are essential to tailor treatment approaches to the specific needs and medical history of each patient. Timely reporting of new or worsening symptoms is crucial for healthcare professionals to conduct appropriate evaluations and interventions.

P105

Use of quantitative fluorescence in systematic research of first non-sentinel lymph node in melanoma sentinel lymph node biopsy (FLASH-NODE Project): preliminary data of a feasibility study

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Backgrounds: Indocyanine Green (ICG), already proved efficient in melanoma sentinel lymph node biopsy (SLNB), exhibits potential for diffusion to the following LNs, particularly to the first nonsentinel lymph node (NSLN). This study investigates quantitative fluorescence to identify NSLN in patients with melanoma undergoing SLNB.
Methods: SLNB patients underwent Technetium-99m-MIBI scintigraphy 3-6h before and periscar intradermal injection of 1,5-3ml of ICG 15min before. SLN research was performed using the scintiprobe and the SPY-cam probe, that defines the presence of fluorescence in the drainage LNs, both qualitatively and quantitatively. SLN was defined as LNs with fluorescence > 400% relative to the injection site (figure 1). After SLN removal, other lymph nodes – defined as NSLN - showing fluorescence rates beyond 250 % with respect to the injection site were sistematically researched and excised.
Results: 60 SLNB were done in 50 patients. SLN was positive in 28% (17/60). At least one NSLN was identified in 55 cases (91,7%) resulting positive in 8% (5/55) of cases. More than one NSLN was identified in 23 procedures. In two cases (3,6%) the NSLN was positive with SLN negative. Neither demographic nor histopathologic factors proved relevant to detect NSLN.
Conclusions: NSLN search via quantitative fluorescence analysis proves feasible in 92% of cases. In two cases NSLN positivity was highlighted in the absence of disease in the SLN, hypothesizing that the systematic search for the NSLN can reduce the number of false negatives in SLNB. The study provides insights in possible upgrades of current SLNB standards, worthy of further investigation.

P106 – SELECTED ORAL COMMUNICATION

Non-invasive techniques for the diagnosis of suspicious skin lesion

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Since melanoma and non-melanoma skin cancers in advanced stages are difficult to treat, early detection is indispensable to reduce morbidity and mortality. Therefore, several in vivo non-invasive diagnostic techniques have been developed, helping the clinician in real-time diagnostics of suspicious lesions at an earlier, curable stage. The dermatological scenario during last decades has been enriched with several advancements of diagnostic techniques, experiencing a succession of multiple eras, historical periods characterized by the development and widespread use of a specific technique.
In the context of melanoma diagnosis, the first era was centered on signs and symptoms of disease, such as hitching, blooding and ulceration that allowed, albeit late, the detection of a suspected melanoma. Subsequently, the period of the "ABCDE" was characterized by the attention to clinical aspects that for their simplicity could also be used by patients in their periodic self-examination. The diagnostic accuracy of clinical signs in the detection of melanoma is estimated at 60%, a value that can be certainly improved. In 1990 started the era of dermoscopy that involves the use of a device with specific contact lens which uses light emitting diodes for illumination, generating a beam of light that falls on the cutaneous surface at an angle of 20. Modern digital dermatoscopes provide magnifications of up to 70- fold, with maintenance of image definition. This technique increases diagnostic accuracy by 15-35%, achieving good diagnostic results that can be further improved with the aid of innovative techniques such as the reflectance confocal microscopy (RCM). RCM allows images of the skin at cellular resolution by providing horizontal sections, using diode laser technology with a wavelength of 830 nm that can reach a depth of 300 micrometers. When compared with standard therapeutic care only (clinic+dermoscopy), adjunctive use of RCM further increases diagnostic accuracy. Finally, we entered in the period of line-field confocal optical coherence tomography (LC-OCT), a technique based on a combination of the optical principles of optical coherence tomography and reflectance confocal microscopy with line-field illumination, which can generate cell-resolved images of the skin, in vivo, in vertical section, horizontal section and in three dimensions.

P107

Dose intensified (70 Gy) versus standard dose (66 Gy) in salvage post-prostatectomy image-guided VMAT: preliminary assessment of genitourinary and gastrointestinal toxicity

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Aim: To study acute and late tolerance of men receiving dose-escalated radiotherapy (39 ptz) (70 Gy) versus standard dose (57 ptz) (66 Gy) in a salvage post-prostatectomy (SRT) setting.
Methods: The records of 96 men treated with prostatectomy and subsequent SRT to the prostate bed were analyzed. Men were treated with image-guided VMAT. Tolerance to treatment was determined from the chart and graded according to RTOG scale.
Results: Acute toxicity: Fifteen days after RT, 7 men (18%) in the group treated with dose-intensified SRT and 10 patient (17,5%) in the group treated with standard dose suffered from G1-2 acute GI toxicity (p=1.0). Of these, 3 (7,7%) in the group treated with dose-intensified SRT and 3 (5,2%) in the group treated with standard dose suffered from G2 acute GI toxicity. Men treated with dose-intensified and standard SRT experienced higher incidence of acute GU than GI toxicity. Ten men (25,6%) in the group treated with dose-intensified SRT and 19 patient (33,3%) in the group treated with standard dose suffered from G1-2 GU toxicity (p=0.4). Two men (3,5%) in the group treated with standard dose and 5 patient (12,8%) in the group treated with dose-intensified SRT suffered from G2 acute GU toxicity. Late toxicity: The late toxicity was assessed six months after the end of postoperative RT in 76 men. Only 5 out 37 patients (13.5%) in the group treated with dose-intensified SRT and 4 out of 39 patients (10.2%) in the group treated with standard dose suffered from G1-2 late GU toxicity (p=0.7). Three men (8.1%) in the group treated with dose-intensified SRT and 2 men (5.4%) in the group treated with standard dose suffered from G2 late GU toxicity. No late G2 or higher GI toxicity in the group treated with standard dose was observed while 2 men in (5.4%) in the group treated with dose-intensified SRT suffered from G2 late GI toxicity.
Conclusion: With a low rate of relevant acute and late GI and GU toxicity no significant difference of symptoms burden in the SRT with a dose-intensified protocol was found with respect to standard SRT dose in short-term period.

P108

Pancreas Preserving Duodenal Resection in elective and emergency setting: our experience

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Introduction: Pancreas Preserving Duodenal Resection (PPDR) can be considered the surgical treatment of first choice for benign and malignant neoplasm of distal duodenum.
Materials and Methods: From January 2019 to May 2022 twelve PPDR were performed at our institution. Ten patients underwent elective surgery and were affected by primitive or secondary duodenal neoplasm: 5 duodenal adenocarcinoma, 3 duodenal gastrointestinal stromal tumor, 1 duodenal lymphoma and 1 duodenal involvement from right colon adenocarcinoma. Two patients underwent urgent PPDR for digestive hemorrhage caused by aorta-duodenal fistula.
Results: Patient population included 9 males and 3 females. Mean age was 57,8 years (42-70 years); mean operative time was 275 minutes (210-360 minutes) and mean postoperative stay was 11 days (7-32 days). One patient died within 30 days of surgery (8,3%) from massive hemorrhage due to acute pancreatitis. One of the two patients undergoing emergency surgery for aorta-duodenal fistula developed an aortic graft infection and was reoperated.
Discussion: Survival rates are not statistically different as compared with duodenopancreatectomy but PPDR is associated with a significant reduction in postoperative morbidity and mortality. Moreover, PPDR can be performed as an emergency operation in case of aorta-duodenal fistula.
Conclusion: PPDR is a feasible and safe operation that can be proposed as a definitive surgical treatment for primary and secondary diseases of the duodenum, in elective and emergency setting.

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