Immuno-Oncology 2022

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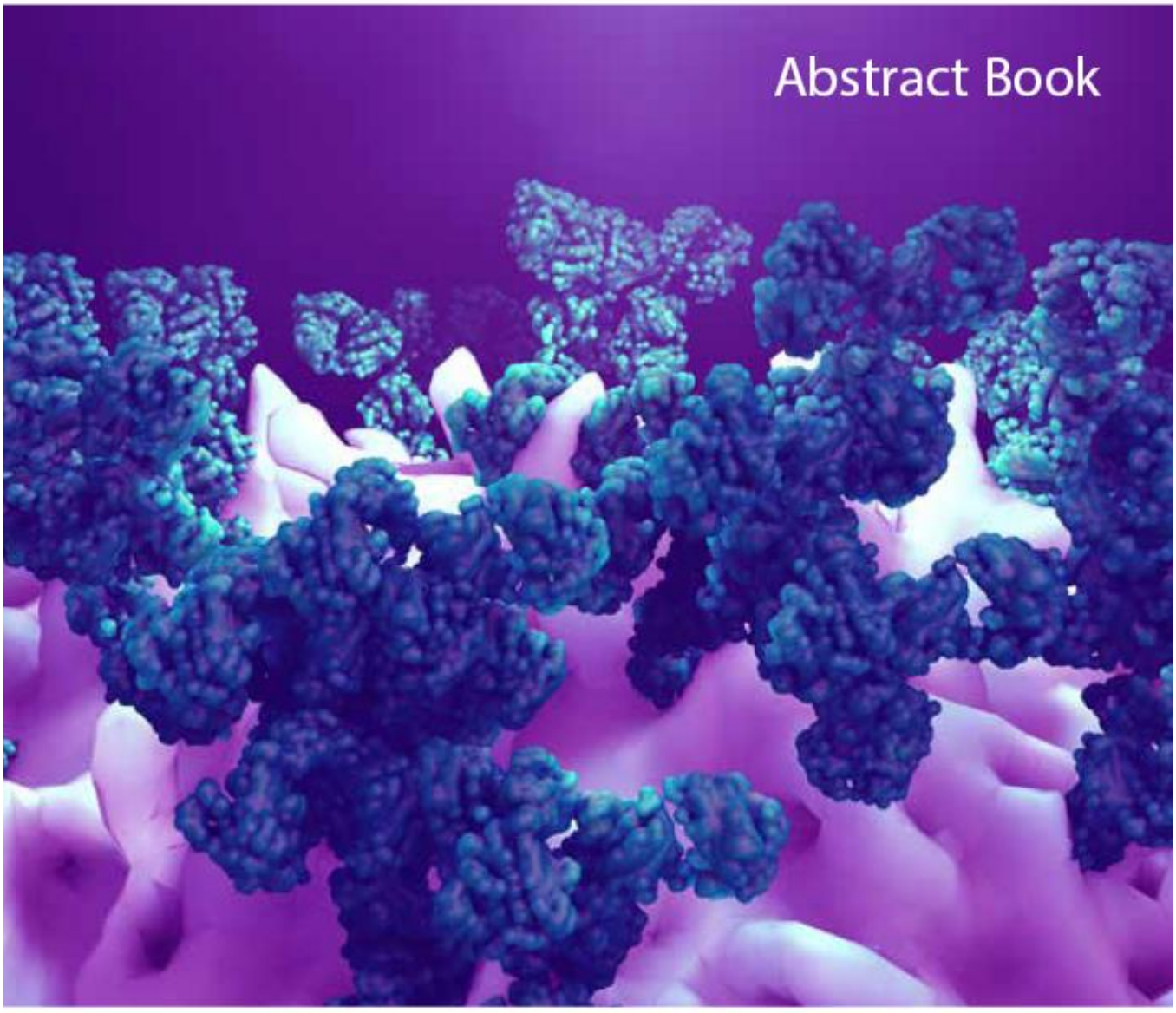

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Poster 1

Biologic markers in Immuno-Oncology

MIF as an Enhancer of Malignancy in Colitis-Associated Colorectal Cancer

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Background. Colorectal cancer (CRC) is the second leading cause of cancer death worldwide. Inflammatory bowel diseases increase the risk of developing CRC by up to 20%. Macrophage migration inhibitory factor (MIF) is a proinflammatory cytokine with chemokine-like functions, overexpressed during the development of CRC, so it has been proposed that MIF plays a fundamental role in attracting immune cells to the CRC microenvironment. Objectives. To determine if MIF enhances the recruitment of immune cells to tumor during the development of CRC. Methods. Male WT (MIF+/+) and Knockout (MIF-/-) BALB/c mice were used to develop colitis-associated colorectal cancer (CAC) induced with AOM/DSS. Clinical signs of mice were recorded weekly until 75 days post-CAC induction, when they were euthanized. Colon was obtained and tumor load was determined. Histology was made and the populations of immune cells infiltrating the tumors were evaluated by flow cytometry. Results. MIF-/- mice exhibited less clinical symptoms, lower numbers of tumors and degree of malignancy, compared to MIF+/+ mice. Importantly, tumor tissue from MIF-/- mice had less infiltration of neutrophils, CD4+ T cells, and myeloid-derived cells than MIF+/+ mice. Conclusion. These results demonstrate that the absence of MIF resulted in less tumor development, probably associated with less infiltration of immune cells in the tumor tissue. This suggests that MIF might enhance tumor malignancy. Funding Support: This work was funded by the National Council of Science and Technology of Mexico (CONACYT), grant number (A1-S-10463) and the Support Program for Research Projects and Technological Innovation (PAPIIT)-UNAM, grant number (IN-217021).

Poster 2

Biologic markers in Immuno-Oncology

Peritoneal Carcinomatosis Induce Down Regulation of Brain-derived Neurotrophic Factor (BDNF) and Interleukin IL 21, a NK Cell Activator

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Background: Cancer diagnosis is life-threatening event and is considerable source of psychological and emotional stress. Depression is common comorbidity observed in cancer patients affecting over 10% of patients.

Objective: To identify the molecular basis of Cerebro-immunitary axis in carcinomatosis animal model

Methods: The behavioral alterations caused by tumor in three different mice models of peritoneal carcinomatosis (using CT-26 and pseudomyxoma from xenograft) were evaluated. After moderate growth of tumor, behavior signs were observed through three widely accepted behavioral tests i.e: .i) marble burying, ii) splash test and iii) open field test. Different gene expressions in several regions (parietal, temporal and occipital) of brain were measured by RT-qPCR. Cytokine array was performed by evaluating total of 144 cytokines in two samples (one control and one cancer) from each model.

Results: The mice injected with cancer cells exhibited significantly less grooming (Splash-test) and less digging behavior (Marble digging test) showing depression-like behavior. By gene array analysis of brain, the preliminary results suggest the strong decrease in gene expression of BDNF and VEGF in cancer mice. However, the gene expression of FGF, NGF and TRKB remained similar in both groups. Cytokines array analysis show that only 3 cytokines showed decreased expression in the model 1 and 2 i.e. IL-20, IL-21 (as an activator of T8, NK cells) and SCF. While, the 4 cytokines that showed increased expression in both the models 1 and 2 were MMP-2, MIP-1gamma, Osteopontin and VEGF-R.

Conclusion: Peritoneal carcinomatosis progression associated with decrease of BDNF and an immunosuppressive state via IL21 downregulation

Poster 3

Biologic markers in Immuno-Oncology

Brain Derived Neurotrophic Factor (BDNF) Down Regulation in Peritoneal Carcinomatosis Patients, Biomarker for Depression

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Aim: Brain-derived neurotrophic factor (BDNF) is described as a factor helping to support the survival of existing neurons by involving the growth and differentiation of new neurons and synapses. Cancer diagnosis impacts the mental health and in consequences, depression arise eventually hinders the recovery, disrupts the quality of life and surviving chances of patients. The focus of this study is to hint upon a prospective biomarker as a promising diagnostic tool for early indicator/predictor of depression prevalence in cancer patients for better care and treatment options.

Materials and Methods: The study aims to analyze peripheral biomarkers from neuro immune axis (BDNF, IL21 as a NK cell activator) using co-relation approach. Samples were obtained from random non cancer candidates and advanced peritoneum carcinomatosis patients with 25% pseudomyxoma, 21% Colon cancer,19% stomach cancer, 10% ovarian cancer, 8% appendices cancer and 10% other area of peritoneum cancer patients. Both groups of study were categorized by gender and age with a range of 18 to 86 years old. Biomarkers were analyzed in collected plasma by performing multiplex sandwich ELISA system. Data were subjected to statistical analysis for the assessment of the correlation.

Results: Our results demonstrate that BNDF and IL 21 down regulated significantly in patient group as compared to non-cancer candidates (ratio of patients/normal is 2.57 for BNDF and 1.32 for IL21).

Conclusion: This preliminary investigation suggested that the neuro immune biomarkers are down regulated in carcinomatosis patients and can be associated with cancer expansion and cancer genesis. Further studies on larger cohort are necessary to validate this hypothesis.

Poster 4

Biologic markers in Immuno-Oncology

Microenvironment Changes during Response to TMZ Therapy in GL261 Preclinical Glioblastoma: what Happens to GAM Population?

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Glioblastomas (GB) are malignant brain tumours with poor prognosis. The participation of immune system is key for sustained response. We have used MRSI metabolomics-based information encoded in nosological images of therapy response which provided the tumour responding index (TRI), the percentage of tumour identified as "responding", supported by a decrease in MRI-based tumour volume. An oscillatory TRI pattern (6-7 days) was shown in longitudinal studies. The purpose of our work was to gain insight into the contribution of immune populations to the MRSI spectral changes recorded from Temozolomide (TMZ)-treated preclinical GL261 GB.

Tumour-bearing mice were treated with TMZ and MRI/MRSI was used to assess response extent. TRI values guided tumour sample obtention. Samples were studied with qPCR for evaluating glioma-associated macrophages (GAM) population and M1-M2 polarisation genes (n=10 control, n=10 TMZ-treated). Histo-cytometry for M1 and M2 markers was also performed (n=5 each group).

Both qPCR and histo-cytometry found a significant increase of GAM in TMZ-treated tumours. However, when it comes to M1 and M2 polarisation, discrepant results were observed, more relevant for the M1 phenotype. While M1 was significantly higher in TMZ-treated samples when studied by qPCR, it was found almost undetectable by histo-cytometry. Differences were less remarkable for the M2 phenotype. These discrepancies raise possible mismatches when using different approaches to assess macrophage populations. We cannot discard that TMZ treatment alters surface features of M1 anti-tumour macrophages, warranting further investigation, since the proposed TMZ treatment produced cure of 50% of treated mice, with immune memory against further tumour generation.

Poster 5

Biologic markers in Immuno-Oncology

Towards Exploiting the Correlation of Metabolomics Data with Molecular Changes in the Tumour Microenvironment for Therapy Monitoring: a Study in a Preclinical Glioblastoma Model

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Glioblastomas (GB) are malignant brain tumours with a dismal prognosis. GAMMs (glioma-associated microglia/macrophages) account for up to 40% of the tumour mass, displaying either a pro-tumour (M2-like) or anti-tumour (M1-like) phenotype. As molecular M1/M2 markers, metalloproteases (MMPs) have been described for early diagnosis of GB as essential for GAMMs reprogramming whereas the PD-1/PDL-1 axis reflects immunosuppressive mechanisms in GB.

Preclinical MRSI metabolomics-based information was encoded in nosological images of therapy response providing the tumour responding index (TRI), the percentage of tumour identified as “responding”, accompanied by tumour reduction. Tumour-bearing mice were treated with TMZ and MRI/MRSI was used to assess therapy response. TRI values guided tumour sample obtention, and qPCR was performed to assess gene expression levels related to GAMM presence and macrophage polarization, as well as for PD-L1 and metalloproteases.

TMZ-treated samples presented increased GAMMs, M1/M2 macrophage ratios and PD-L1 expression. All metalloproteases except MMP9 increased significantly at response, and the correlation between different expression levels was assessed. ADAM17 vs MMP14 and ADAM8 vs PD-L1 presented a noticeable change in responding samples. MMP14 can remodel the extracellular matrix thereby potentiating immune system recruitment while ADAM17 may support antigen presentation steps. Still, mechanisms of cell protection take place through shedding of TNF-R1 by ADAM8 (avoiding TNF-induced cell death) and increased PD-L1, which upon binding to PD-1 might decrease anti-tumour activities of T-cells. Such changes may at least partially explain the metabolomics changes observed in the MRSI pattern, with potential translational interest in noninvasive assessment of therapy response in GB.

Poster 6

Biologic markers in Immuno-Oncology

Tumor-Intrinsic Progranulin Drives Immune Evasion and Early Progression of Pancreatic Ductal Adenocarcinoma

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Objective:

Immune evasion is indispensable for cancer initiation and therapy resistance, albeit the underlying tumor-intrinsic mechanism in pancreatic ductal adenocarcinoma (PDAC) remains elusive. Here we investigated the role of progranulin (PGRN), a fibrotic factor promoting PDAC metastasis, in immunoevasion and tumor initiation of primary PDAC.

Design:

Association of PGRN expression with clinical outcome and immunological parameters was analyzed in multiple PDAC cohorts including CONKO-001 clinical trial. Spatial interaction between PGRN+ tumors and immune cells was assessed by multiplex immunofluorescence imaging and computational analysis in human and mouse PDAC. Functions of PGRN were dissected in cellular model systems by RNA interference, and in spontaneous genetic PDAC mouse model, CKP, by PGRN blockade with antibody during early tumor development.

Results:

High PGRN expression level was associated with immunomodulatory features and predicted poor prognosis in multiple PDAC patient cohorts. Spatial imaging in human PDAC illustrated intratumoral heterogeneity of PGRN tumor expression associated with distinct immune landscapes. In CKP, strong interaction between PGRN+ tumors and tumor-associated macrophages (TAMs) was observed since early PDAC development. In vitro findings demonstrated that PGRN reduced tumor immunogenicity by modulating checkpoint, co-stimulatory and MHCI molecules, TGFß secretion, and TAM polarization. PGRN blockade in CKP at early stage remarkably decelerated PDAC progression. Abated tumor proliferation, TAM reprogramming and revived cytotoxic immunity occurred and were associated with circulating PGRN levels.

Conclusion:

Our findings unveil an imperative role of tumor-intrinsic PGRN in immunoevasion and tumor initiation in PDAC, thereby providing biological basis for targeting PGRN as a potential therapeutic strategy in PDAC.

Poster 7

Biologic markers in Immuno-Oncology

Soluble PD-L1 Level is Correlated with its Expression in Tissue and is Associated with Poor Overall Survival in Gastric Cancer

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Background: A strong correlation between the expression of programmed cell death ligand-1 (PD-L1) concentration in cancer tissues and the prognosis of patients has been reported. The possibility of its application as a novel diagnostic tool by testing a soluble biomarker in plasma has recently emerged.

Objectives: We investigated the correlation between the soluble programmed cell death ligand-1 (sPD-L1) concentration in plasma and the PD-L1 expression, in tissue as protein and at mRNA level, in patients with gastric cancer. Also, we evaluated the utility of sPD-L1 in clinical practice.

Methods: The serum and tissue levels of PD-L1 were measured using an enzyme-linked immunosorbent assay. The RNAseq data was used to extract CD274 (encoding PD-L1) gene expression. Correlations between the sPD-L1 concentration, various laboratory parameters, and disease features were assessed on blood samples collected from 85 patients with histologically proven gastric adenocarcinoma.

Results: Correlations between sPD-L1 concentration and the expression of PD-L1 in tumor specimens showed that plasma PD-L1 expression is significantly correlated with tissue protein expression and CD274 mRNA expression. Moreover, plasma PD-L1 was significantly associated with tumor size and lymph node metastasis. The overall survival of the sPD-L1-high group was (95.25 pg/mL) significantly worse than that of the low group ($p = 0.0132$).

Conclusion: The sPD-L1 concentration is correlated with PD-L1 expression in tissue, with tumor progression and the elevated levels are associated with a poor prognosis in gastric cancer patients. Consequently, soluble PD-L1 can be considered an accurate predictor of tumor status and of the treatment effect.

Poster 8

Biologic markers in Immuno-Oncology

Characterization of Colorectal Cancer Cells Using Fluorescent Molecularly Imprinted Polymers and Digital Holographic Cytometry

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Background: Colorectal cancer (CRC) is the second most spread cancer disease with the majority of cases detected in Western countries. Metastases are formed by circulating tumor cells characterized by altered phenotype expressing higher levels of sialic acid (SA) and report changes in morphological properties associated with increased invasiveness and poor prognosis. SA as a terminating part of glycans is expressed in α-2,3 or α-2,6 linkages and plays an important role in cellular responses such as cell-cell communication, cell adhesion, recognition or proliferation. To evaluate a biomarker together with morphological characterization, we have combined the use of SA-targeting molecularly imprinted polymers (SA-MIPs), and morphological measurements using digital holographic cytometry (DHC).

Objectives: To elucidate the glycosylation pattern of both metastatic and primary tumor cell lines derived from CRC using SA-MIPs and the morphological properties of the cell lines using DHC. Further, DHC was used to discriminate between the CRC cell line COLO 205 and peripheral blood mononuclear cells (PBMCs).

Methods: Cells were stained with SA-MIPs, lectins MAL I, SNA, VVL, and PNA, and thereafter analyzed using flow cytometry. DHC analysis of the cell lines with or without SA-MIP staining was performed to measure cellular morphological properties of CRC cell lines. COLO205 and PBMCs were mixed and analyzed for different morphological properties with DHC.

Results: We demonstrate the SA expression patterns of the CRC cells using several lectins and SA-MIPs. The NCI-H508 and COLO 205 cell lines change their morphological properties when treated with SA-MIPs. DHC results show discrimination between CRC cells and PBMCs in vitro by determining differences in the cell area, cell thickness, cell volume, and cell irregularity even when the CRC cells were in minority (5 % out of PBMCs). Here we present DHC as a new powerful tool for discriminating cells of different sizes in suspension together with a combination of biomarkers.

Conclusion: We show that a combination of biomarkers, including SA and DHC, can be a new powerful tool in detecting CRC cells in suspension.

Poster 9

Biologic markers in Immuno-Oncology

Clinicopathological Significance of TIM-3 Expression in TILs and Cancer Cells, and Serum Concentrations in cats with Mammary Carcinoma

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Background: Recently, in order to overcome resistance to current immune checkpoint blockade therapies, T-cell Immunoglobulin and Mucin-domain containing molecule-3 (TIM-3) receptor has emerged. Although feline mammary carcinoma (FMC) is a valuable cancer model, no studies on TIM-3 have been developed.

Objectives and Methods: TIM-3 expression in total (t), stromal (s) and intratumoral (i) tumor-infiltrating lymphocytes (TILs) and cancer cells in 52 cats with mammary carcinoma was evaluated by immunohistochemistry. In parallel, serum TIM-3 levels were quantified using ELISA and TIM-3 gene was sequenced from 19 tumor tissue samples, to identify putative mutations that can be used as biomarkers.
Results: the percentage of tTILs expressing TIM-3 (tTILs-TIM-3+) was negatively associated with tumor metastasis (p=0.01) and tumor recurrence (p=0.013), whereas a decrease of TIM-3+ score in tTILs was correlated with an increased tumor size (p=0.039). Additionally, the solid histological subtype showed a positive association with the percentage of tTILs-TIM-3+ (p=0.047) and sTILs-TIM-3+ (p=0.02). Regarding the expression of TIM-3+ in iTILs, a negative correlation was found with the tumor size and tumor stage (p=0.037 and p=0.035, respectively), whereas a positive correlation was found between the percentage of sTILs-TIM-3+ and cancer cells-TIM-3+, and tumor malignancy grade (p=0.044 and p=0.018, respectively). Moreover, higher densities of cancer cells overexpressing TIM-3+ were associated with positive lymph node status (p=0.025) and with triple negative basal-like (p=0.038). TIM-3 serum levels were lower in the cats with mammary carcinoma than in the healthy group (p0.001) and one mutation was identified in one tumor sample.

Conclusion: our results suggest that TILs-TIM-3+ subpopulations may differently influence the clinical outcome of cats with mammary carcinoma, resembling previous reports in human breast cancer.

Poster 10

Biologic markers in Immuno-Oncology

Mapping the Cellular Architecture of the Tumor Microenvironment by Integrating Hyperplex Immunofluorescence and Automated Image Analysis

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The tumor microenvironment (TME) is composed of malignant cells and the surrounding healthy counterpart. The precise identification of the TME components is crucial to understanding how this microecosystem remodels during tumorigenesis and responds to treatment in order to identify its vulnerabilities and treatment opportunities (1). In the past decade, significant efforts have been made to describe the TME using RNA-based technologies (2,3). These approaches shed light on the tumor heterogeneity and variable response to treatment. However, RNA-based biomarker expression profiling has limited relevance as it might not always accurately reflect the actual protein levels (4). In addition, the increasing number of protein biomarkers available led to the development of new technologies that allow the analysis of dozens of proteins on a single tissue slide (5).

The COMET™ platform is an automated instrument that allows the detection of up to 40 antigens on a single slide using sequential immunofluorescence staining (6). By integrating multiplex immunofluorescence technology, we profiled the expression of 40 protein biomarkers across a tissue microarray composed of primary lung tumors and their corresponding metastatic lymph nodes. The combination of the hyperplex panel with an automated image and data analysis pipeline based on an unsupervised machine learning clustering algorithm allowed for the identification of several classes of immune cells with preferential accumulation sites. We identified distinct myeloid cells that coexist within the TME but infiltrate to a higher extent either the primary tumor or the metastatic loci. Harnessing the same approach, we also observed a higher frequency of T regulatory cells in the primary tumors. Subsequently, newly identified population frequencies determined by unsupervised clustering was confirmed by a complementary approach of supervised single-cell analysis.

Our data highlights the potential that microfluidics-based multiplex technology brings into the fields of both digital pathology and immuno-oncology, thanks to its single-cell resolution and the simultaneous detection of multiple protein biomarkers. We demonstrate here how the combination of hyperplex images obtained using the COMET™ platform, along with machine learning clustering analysis, results in an easy workflow for analyzing the complex TME and obtaining a single-cell atlas of tissue specimens.

1.Binnewies M, et al. Nat Med 2018; 24(5):541–550.

2.Lau D, et al. Trends Cancer 2019; 5(3):149-156.

3.Vries NL, et al. Front Oncol 2020.

4.Vogel C, Marcotte EM. Nat Rev Gen 2012; 13:227–232.

5.Lewis SM, et al. Nat Methods 2021; 18:997–1012.

6.Migliozzi D, et al. Microsyst Nanoeng 2019; 5:59.

Poster 11

Biologic markers in Immuno-Oncology

MIF: a Key Cytokine in Colorectal Cancer Progression

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Background. Colorectal cancer (CRC) is a significant public health problem, as it ranks third in incidence and fourth in mortality among all types of cancer in the world. Macrophages inhibitory factor (MIF) is a pro-inflammatory cytokine with chemokine characteristic, which is expressed significantly in CRC. Objetive. Here, we explored the role of endogenous MIF in the genesis and progression of CRC Methods. We used MIF gene-deficient (mif-/-) and wild-type (mif+/+) C57BL/6 mice to study the impact of MIF on the development of chemically induced CRC with a single intraperitoneal injection of azoxymethane (AOM, 10 mg/Kg), and 1 cycle of 2% dextran sodium sulphate (DSS) diluted in drinking water and 3 cycles of 1.5% DSS. After the induction, the weight of the mice, the clinical signs, and mortality were recorded weekly. Results. At 90 days post-induction, mif-/- CRC mice developed fewer tumors, which presented as characteristic polyps that were up to 3 times smaller than those of mif+/+ CRC mice, which developed larger tumors that featured serrated adenomas and metaplasia. The tumor tissue of mif-/- CRC presented a lower number of infiltrate cells, with low presence of macrophages and NK cells, as well as low expression of arginase and nitric oxide compared to the tumor tissue from mif+/+ mice CRC. In conclusion. The results suggest that MIF has an important role as a regulator of tumor maintenance in the CRC, which suggests that MIF could be a possible therapeutic target in CRC.

Poster 12

Biologic markers in Immuno-Oncology

Fueling the Flames of Colon Cancer – does CRP Play a Direct Pro-Inflammatory Role?

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Background: Systemic inflammation measured by the acute phase C-reactive protein (CRP) has consistently been correlated with poor outcome across cancer types. CRP exists in two isoforms, circulating pentameric CRP (pCRP) and the highly pro-inflammatory monomeric isoform (mCRP). The aim of this pilot study was to map the pattern of mCRP distribution and explore possible functional roles within the tumor microenvironment (TME) of a previously immunologically well-defined colon cancer (CC) cohort.

Methods: FFPE tissue samples from 43 stage II and III CC patients, including 20 patients with CRP of 0-1 mg/L and 23 patients with CRP 30 mg/L were immunohistochemically stained with a conformation-specific mCRP antibody and selected immune and stromal markers. Western blot was performed for mCRP protein verification and antibody specificity.

Results: mCRP was abundantly present in tumors from systemically inflamed patients whereas non-inflamed patients exhibited only modest/no mCRP positivity (median mCRP per area 5.07 (1.32-6.85) vs. 0.02 (0.01-0.04) p0.001). Similarly, tissue-bound mCRP correlated strongly with circulating pCRP (Spearman correlation 0.81, p0.001). Importantly, mCRP was detected exclusively within tumors, whereas adjacent normal colon mucosa showed no mCRP expression. Double IHC staining revealed colocalization of mCRP with endothelial cells and neutrophils. Intriguingly, some tumor cells also colocalized with mCRP, suggesting a direct interaction or mCRP expression by the tumor itself.

Conclusion: Our data strengthen the hypothesis that CRP might not only be an inflammatory marker but also an active mediator within the TME of systemically inflamed CC patients.

Poster 13

Biologic markers in Immuno-Oncology

Cancer Morbidity and Immunological Markers in the Rural Population of Sachkhere District of Georgia

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Background: The study of the spatial and temporal variability of cancer incidence in the population and its potential biomarkers is of great practical and theoretical importance. Objectives: The purpose of the study was to compare the morbidity from cancer (expressed as incidence) to the average levels of blood serum immunological markers in the Sachkhere region (Georgia) population. Methods: healthy residents of the Sachkhere district were examined. In the blood serum samples of patients, the cytokines (IL-1α, IL-10, TGF-β, IL-12, IL-17, TNF-α, IL-6) and NOx content, as well as the total antioxidant activity of the non-enzymatic system (TAA) were determined. Results: Study results show, that cancer incidence in Sareki was statistically significantly higher than in Chorvila and Sairkhe ($p=0.002$; $p=0.004$); in Sareki inhabitant's blood serum levels of the IL-6, NO are increased ($p=0.004$, $p=0.05$), and IL-17, TGFβ, and IL-10 levels are decreased ($p=0.010$, $p=0.001$, $p=0.033$) in comparison to data in Chorvila; in Chorvila inhabitants' indicators of TAA of blood serum were lower ($p=0.001$,$p=0.045$) then in Sairkhe and Sareki. Conclusion: The existence of statistically reliable associations between the levels of cancer incidence in the populations of the surveyed villages and the indicators of immune status in their virtually healthy subpopulations, with a high degree of persuasiveness, allows us to assume a close causal link between them. Clarifying the reasons for the identified patterns and their significance requires more detailed studies.

Poster 14

Biologic markers in Immuno-Oncology

Sezary Cell Phagocytosis in Patients with CTC

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Background

Sézary syndrome (SS) is a rare subtype of cutaneous T-cell lymphoma characterized by the presence of atypical malignant T cells. Monocyte-derived cells are considered to promote tumorigenesis and long-term survival of malignant T cells, but their role is not fully understood in SS.

Objectives

Investigate the role of monocyte-derived cells in SS and discover new biomarkers and potential drug targets.

Methods

Single-cell sequencing data was obtained using the 10xGenomics kit with VDJ enrichment.

Results

Having analyzed scRNAseq data of SS patients, we obtained 8 clusters with clear distinction between a transformed T cell cluster and a monocyte cluster. A subset of monocytes had a mixed signature of T cells (TRAV41, TCF7, CD3E) and monocytes (CD14, FCGR3A). The tumor cluster comprised cells with overexpressed TRAV41 and TRBV7-2 chains. Given the overlap of the TCR genes between tumor cluster and monocyte cluster with mixed signature, we hypothesized that those cells may represent doublets. We then applied the algorithm from the DoubletDetection library, but the cluster of interest remained intact after doublet filtration. Although typically Sezary cells highly express CD47, the tumor cells didn't overexpress it in samples, where we observed a cluster with mixed monocyte/T cell signature. This indicates a potential loss of CD47 expression leading to phagocytosis of Sezary cells.

Conclusion

Loss of CD47 expression in Sezary cells leads to their phagocytosis, which may explain better survival rates in these patients. This hypothesis requires further validation.

Poster 15

Biologic markers in Immuno-Oncology

Do Immunotherapy Induced Adverse Events Predict Treatment Efficacy in Patients with Lung Cancer? A Case-Based Study

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Background

Immune checkpoints are built-in mechanisms that regulate the immune response, acting as a “brake”, and not allowing the destruction of self-cells. Immune checkpoint inhibitors (ICIs) are designed to reverse this action allowing T cells to exert their anti-tumour effect. ICIs are currently used in advanced non-small cell lung cancer (NSCLC) with tumours expressing at least 1% PD-L1. Because of their mechanism of action, ICIs can cause Immune Related Adverse Events (irAEs).

Objectives

This paper aims to discuss if irAEs could be used as a biomarker to predict which patients with NSCLC would benefit more from immunotherapy.

Methods

A literature search was performed and identified studies discussed. Interviews with four patients receiving ICIs for lung cancer have been conducted and compared to existing literature.

Results

The development of irAEs is indicative of a strong immune response and has been linked to better overall survival (OS), increased progression-free survival (PFS) and higher objective response rate (ORR) than in patients without irAEs. Certain irAEs (skin, endocrine) are indicative of better treatment efficacy, while more severe irAEs (colitis) indicated lower survival rates. Single organ irAEs occurred more than multiple organ ones. The onset time of irAEs was not significant.

Conclusion

Overall, irAE occurrence was indicative of better OS, PFS and ORR. However, current evidence is contradictory in some respects, thus there is need for more prospective, large-scale studies in order to determine the statistical and clinical significance of irAEs as positive predictive biomarkers for treatment efficacy and patient selection.

Poster 16

Cell therapy

A Non-Genetic Engineering Platform for Rapidly Generating and Expanding Cancer-Specific Armed T Cells

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Background

Cancer-specific adoptive T cell therapy has achieved successful milestones in multiple clinical treatments. However, the commercial production of cancer-specific T cells is often hampered by laborious cell culture procedures, the concern of retrovirus-based gene transfection, or insufficient T cell purity.

Methods

In this study, we developed a non-genetic engineering technology for rapidly manufacturing a large amount of cancer-specific T cells by utilizing a unique anti-cancer/anti-CD3 bispecific antibody (BsAb) to directly culture human peripheral blood mononuclear cells (PBMCs). The anti-CD3 moiety of the BsAb bound to the T cell surface and stimulated the differentiation and proliferation of T cells in PBMCs. The anti-cancer moiety of the BsAb provided these BsAb-armed T cells with the cancer-targeting ability, which transformed the naïve T cells into cancer-specific BsAb-armed T cells.

Results

With this technology, a large amount of cancer-specific BsAb-armed T cells can be rapidly generated with a purity of over 90% in 7 days. These BsAb-armed T cells efficiently accumulated at the tumor site both in vitro and in vivo. Cytotoxins (perforin and granzyme) and cytokines (TNF-α and IFN-γ) were dramatically released from the BsAb-armed T cells after engaging cancer cells, resulting in a remarkable anti-cancer efficacy. Our first BsAb armed T cell product is ready to initiate an investigator-initiated trial in EGFR downstream mutated untreatable colorectal cancer.

Conclusions

The BsAb-armed T cell technology is a simple, time-saving, and highly safe method to generate highly pure cancer-specific effector T cells, thereby providing an effective T cell immunotherapy to patients.

Poster 17

Cell therapy

Cell Avidity a Key Parameter in Understanding Therapeutic Immune Cell Functionality and Predicting in-vivo Response

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T cell killing requires functional engagement between effector-target cell pairs and associated mechanical forces. Whilst legacy techniques often struggle, and can lead to failures in clinic, cell avidity looks more holistically at the immune synapse and is emerging as a key predictor of in vivo performance. Across TCR and CAR-T therapies the affinity of the receptor has not been shown to correlate with a sustained response, in vivo persistence and effective tumour killing. Moreover in vitro killing and cytokine release assays can have similar limitations. However, cell avidity (i.e. overall binding strength between an immune cell and its target) provides a more complete and physiologically relevant live-cell parameter that reflects the true T cell-tumor cell interactions formed.

Here we present Cell Avidity data on TCR-,CAR-, which correlate well with effective anti-tumor responses in vivo. We also present preliminary data on the avidity of therapies using cell engagers This demonstrates the benefits of using cell avidity to improve the prediction of T cell functionality and ultimately for designing better therapies.

Poster 18

Cell therapy

Novel Curcumin Nanoformulation Induces Apoptosis and Reduces Migration and Angiogenesis in Breast Cancer Cells

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Background: Medicinal plants are a promising source for adjuvant, complementary or alternative anti-cancer therapy because they contain natural ingredients with anti-cancer activity. Although the clinical use of curcumin is well known, its low bioavailability and chemical stability limit its therapeutic applications. In this study, we evaluated the efficacy of curcumin nanoparticles (Cur-NPs) coated with PEG/chitosan for the treatment of breast cancer (BC) and elucidated the underlying molecular mechanisms that contributed to its anti-tumor activity.

Methods: Cur-PLGA-PEG/chitosan NPs were synthesized and characterized using dynamic light scattering (DLS). The effect of newly synthesized Cur-NPs was evaluated in two BC cells in addition to MCF10A and WI-38 as normal control cells.
These cells were treated with different non-cytotoxic concentrations of Cu-NPs, and MTT assay and other functional assays were performed.

Results: The average size of Cur-NPs was 139.3 ± 55.16 nm in diameter and the PDI of 0.169. Compared to normal cells, Cur-NPs induced cytotoxicity in MDA-MB-231 and MCF7 cells at 6.25 µg/mL after 48h of treatment. Treatment of MDA-MB-231 cells with 2.5 µg/mL of Cur-NPs inhibited cell migration and this inhibition was augmented at 10 µg/mL (p0.001). Treatment of chicken embryo with 5 µg /mL Cur-NPs reduced angiogenesis (p0.001) which was started on day 2. At the molecular level, Cur-NPs upregulated Bax and p53 and downregulated Bcl-2 in a dose-dependent
manner. Cur-NPS also induced cell apoptosis in MDA-MB-2321 cells.

Conclusion: Treatment of BC cells with Cur-NPs decreased cell proliferation, migration, and angiogenesis, and induced cell death via upregulation of Bax and p53 and downregulation of Bcl-2. Further investigations are warranted to determine the pharmacokinetics of Cur-NPs using a preclinical cancer model.

Keywords: Curcumin nanoparticles, cancer cells, migration, angiogenesis, apoptosis

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Poster 19

Cell therapy

Novel 3'-Diindolylmethane (DIM) Nanoformulation Induces Apoptosis and Reduces Migration and Angiogenesis in Liver Cancer Cells

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Background: Herbaceous plants are valuable sources of complementary, adjuvant, or alternative anti-tumor therapy as they contain natural active ingredients with anti-cancer potential. Here, we evaluated the efficacy of 3, 3′-Diindolylmethane (DIM), which is present mainly in cruciferous plants, and nanoparticles (DIM-NPs) against liver cancer, and elucidated the underlying molecular mechanisms of action.

Methods: The effect of synthesized DIM-NPs was evaluated in HepG-2 and immortal normal liver cells, HUH-7 cells, and WI-38 (normal control cells). These cells were treated with different non-cytotoxic concentrations of DIM-NPs and MTT assay and other functional assays were performed.

Results: Compared to normal cells, DIM-NPs induced cytotoxicity in HepG-2 cells at 6.25 µg/mL after 48h of treatment. Treatment of HepG-2 cells with 2.5 µg/mL of DIM-NPs inhibited cell migration and this inhibition was augmented at 10 µg/mL (p0.001). Treatment of chicken embryo with 5ug/ml DIM-NPs reduced (p0.001) angiogenesis at day 4. Notably, at the molecular level, DIM-NPS upregulated Bax and p53 and downregulated Bcl-2 in a dose-dependent manner. DIM-NPS also induced cell apoptosis in HepG-2 cells.

Conclusion: Treatment of hepatic cells with DIM-NPs decreased cell proliferation, migration, and angiogenesis, and induced cell death via upregulation of Bax and p53 and downregulation of Bcl-2. Further investigations are necessitated to determine the pharmacokinetics of DIM-NPs using a preclinical cancer model.

Keywords: 3, 3′-Diindolylmethane (DIM) nanoparticles, Hepatic cancer cells, migration, angiogenesis, apoptosis.

Funding: This research work was funded by Institutional Fund Projects under grant no (IFPRC-052-248-2020). Therefore, authors gratefully acknowledge technical and financial support from The Ministry of Education and King Abdulaziz University, Jeddah, Saudi Arabia.

Poster 20

Cell therapy

miR-335-laden B Cell-Derived Extracellular Vesicles Promote SOX4-Dependent Apoptosis in Human Multiple Myeloma

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BACKGROUND: Multiple myeloma (MM) is incurable and characterized by the accumulation of malignant plasma cells in the bone marrow, new strategies are needed. Increased expression of the transcription factor Sex-determining region Y-related high-mobility-group box transcription factor 4 (SOX4) has been correlated with tumor development and progression through a variety of distinct processes, including inhibition of apoptosis, increased cell invasion and metastasis, and induction and maintenance of cancer-initiating cells.

OBJECTIVIES: Investigate SOX4 role in MM as therapeutic target.

METHODS: Since SOX4 is a known target of miR-335, we used miR-335 to assess whether SOX4 modulation could promote apoptosis in MM cell lines and patient bone marrow-derived plasma cells. Gene expression levels were analyzed by RT-PCR, protein levels by Western-bloting, functionality by FACS assay.

RESULTS: In model MM cells miR-335 acts on SOX4-related genes such as AKT, PI3K, and on hypoxia-inducible factor 1-alpha (Hif1-α). MiR-335-laden extracellular vesicles induced in B cells (iEVs) were found to be effective at targeting SOX4 and causing apoptosis. SOX4 was found to be expressed in all plasma cells samples obtained from the bone marrow of MM patients. We found that miR-335 was effective in downregulating SOX4, but its effect was more pronounced in plasma cells from patients after second line therapy compared to first line and untreated patients. These data permit to propose mir-335 as an effective therapeutic approach in MM cells characterized by high activity of the SOX4 pathway.

CONCLUSION: SOX4 is a MM gene involved in tumor progression that can be targeted therapeutically by miR335.

Poster 21

Cell therapy

Experimental Comparative Study of Sodium Dichloroacetate, Magnesium Dichloroacetate Efficacy on Pediatric Glioblastoma Tumor and Cells

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Background. Sodium dichloroacetate (NaDCA) is a regulator of mitochondrial metabolism in cancer and immune cells with sex-related effects.

The aim of the study was to evaluate the effect of NaDCA, magnesium dichloroacetate (MgDCA) on the growth of PBT24 (boy's) and SF8628 (girl's) glioblastoma xenografts.

Materials and methods. Pediatric PBT24 and SF8628 xenografts were studied in chicken chorioallantoic membrane (CAM) model groups: control, treated with 10 and 5 mM NaDCA, 5 and 2.5 mM MgDCA. Tumor growth was studied by histological and immunohistochemical methods. The number of blood vessels in CAM, as well as the expression of PCNA, EZH2 in the tumor cells were assessed.

Results. 10 mM NaDCA and 5 mM MgDCA reduced the incidence of PBT24 and SF8628 tumor invasion in CAM (p0.005); whereas 5 mM NaDCA reduced SF8628 tumor invasion (p0.004). The dose of 2.5 mM MgDCA had no effect on the invasion rate of tested tumors (p0.05). In the PBT24-5 mM MgDCA group the reduced neoangiogenesis in CAM under the tumor was observed (p0.02); 10 and 5 mM NaDCA inhibited angiogenesis under SF8628 tumor (p0.05), while MgDCA had no effect (p0.05). DCA preparations decreased PCNA and EZH2 expression in both tumor cells (p0.05), except in the PBT24-2.5 mM MgDCA group (p0.05).

Conclusions. Differences in the response of PBT24 and SF8628 tumors growth to treatment with dichloroacetate or the contrast in efficacy of NaDCA and MgDCA may indicate some differences in the biology of the investigated cells. The study was funded by the Lithuanian Research Council, grant No P-MIP-20-36.

Poster 22

Evaluating response in Immunotherapy

Valproic Acid Effect on NKCC1, KCC2, and SLC5A8 Expression in Rat Thymocytes is Sex-related

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Background. Valproic acid (VPA) has immunomodulatory and anticancer effects depending on gender.

Objectives were to investigate the effect of VPA on NKCC1 (Na-K-2Cl), KCC2 (K-2Cl) co-transporters, and SLC5A8 (Na-dependent short fatty acids transporter) RNA expression in male and female rat thymocytes.

Methods. Wistar rats, aged 4 to 5 weeks, were investigated in VPA-treated gonad-intact and gonadectomized males and females and their controls (n = 6 in a group). The VPA 200 mg/kg/day treatment with drinking water duration was 4 weeks. The NKCC1, KCC2, and SLC5A8 RNA expression in thymocytes were determined by the RT-PCR.

Results. The NKCC1, KCC2, and SLC5A8 expressions were higher in the gonad-intact male control than in the gonad-intact females (p 0.05). VPA treatment decreased the NKCC1 expression in gonad-intact males (p 0.05), increased the KCC2 expression in gonad-intact and gonadectomized males (p 0.05). Treatment with VPA decreased SLC5A8 expression in gonad-intact and castrated males (p 0.05). The SLC5A8 expression in castrated male and female rat thymocytes was higher than in gonad-intact respective sex control groups (p 0.05). The SLC5A8 expression in gonad-intact VPA-treated female thymocytes was higher than control's (p 0.05). Castration and VPA had a significant effect on the correlations among the expression of the studied genes, which are defined by sex-related differences.

Conclusion. The impact of VPA on the expression of NKCC1, KCC2, SLC5A8 and their interrelation in rat thymocytes depends on sex and gonad hormones, may determine the different immunomodulatory and anticancer effects.

Poster 23

Evaluating response in Immunotherapy

In Vitro 3D Cancer Models for the Screening of Drug Candidates that Modulate the Polarization of Tumor-Associated Macrophages

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Background: In vitro models are important tools in cancer research, enabling screening and evaluation of novel drug candidates. Especially therapies targeting macrophages in tumors have emerged as promising treatments as tumor-associated macrophages (TAMs), often associated with an M2-like phenotype, correlate with tumor progression. Objectives: To select effective therapies targeting TAMs, we established an in vitro 3D multicellular spheroid model including cancer cells, fibroblasts, and monocytes to reliably mimic the microenvironment of solid tumors. Methods: We tested tumor-associated macrophage (TAM)-inhibiting compounds (CCL2 Ab, CSF1R inhibitor, CSF1R Ab) and TAM-reprograming compounds (poly I:C, CD40 Ab, CD40 ligand) for their effects on monocyte infiltration and polarization in tumor spheroids. For characterization of macrophage polarization, we measured the expression of CD206, CD163, CD86, MHC II, CD40, and CD14 and measured 43 soluble factors in the 3D MCTS cultures. Results: Monocytes rapidly infiltrated tumor spheroids and differentiated into mature macrophages with diverse phenotypes in a cancer cell line-dependent manner. Treatment with the CSF1R inhibitor prevented infiltration of monocytes into pancreatic cancer spheroids, and macrophages treated with the inhibitor showed decreased expression of M2 markers. Treatment with a CD40 ligand and poly I:C induced M1 macrophage polarization in our models. Conclusion: We show that these models can be used to evaluate compounds that modulate monocyte infiltration and macrophage phenotype in in vivo-like tumors and propose that these models can be used to improve the drug screening process of anti-cancer immunotherapies targeting TAMs.

Poster 25

Evaluating response in Immunotherapy

A Personalized Multi-Antigenic Tumor Vaccine Induces Significantly Stronger CD4 and CD8 T Cell Immune Responses against B16 Melanoma than the Adenovirus Vector-Based Vaccine Encoding B16 Neo-Antigens.

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Background: Immunotherapy using personalized tumor vaccines are developed on different platforms such as synthetic peptide neoantigens, recombinant mutated proteins, RNA- or DNA-vectors encoding tumor antigens. It is not known which of the vaccine platforms is more efficient.
Objectives: Here we compare two vaccine platforms particularly the neo-antigen encoding recombinant adenoviral vector-based vaccine and the tumor tissue-derived multi-antigenic vaccine according to their immunogenicity.
Methods: Recombinant adenovirus vector, encoding B16F10 melanoma mutant antigen, was used as a neo-antigenic tumor vaccine (Ad-B16). A multi-antigenic tumor vaccine (MTV-B16) was made out of the B16F10 tumor tissue. For this, a B16F10 tumor was surgically removed, and tumor tissue was minced and blended with molecular immunoadjuvants that activate dendritic cells and reprogram myeloid suppressors. The number of antigen-reactive IFNγ-secretory T effector and T effector memory cells was analyzed by ELISPOT. Serum antibodies specific to intracellular antigens of B16F10 melanoma cells were analyzed using ELISA, while FACS was applied to detect B16F10 surface antigens.
Results: Both vaccines induced tumor-specific immune responses although MTV-B16 was much more immunogenic according to the number of tumor-specific IFNγ-secretory T cells in the spleen of mice. Thus, immunization with MTV-B16 generated up to 6500 IFNγ CD4 and 3500 IFNγ CD8 T-effector cells versus 600 IFNγ CD4 and 650 IFNγ CD8 T-effector cells generated by Ad-B16. Both vaccines induced serum antibodies specifically recognizing antigens of B16F10 melanoma cells.
Conclusion: MTV-B16, a personalized multi-antigenic tumor vaccine easily produced out of tumor tissue and empowered with molecular adjuvants, induces tumor-specific Th1-type CD4 and CD8 T cell responses, one order of magnitude higher intensity than those induced by the Ad-B16, an adenovirus-based vaccine.
This study was supported by the Russian Science Foundation (project №20-15-00391).

Poster 26

Future directions in Immuno-Oncology

Knowledge, Attitudes, Practice and Other Factors Associated with Human Papillomavirus Vaccine of Young Children in the View Point of Professors and Health Staffs of Shahid Beheshti University of Medical Sciences in 2018

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Background and Aim: A study aimed to evaluate knowledge, attitudes, practice and other factors, which are associated with Human Papillomavirus Vaccine of Young Children, in the view point of Professors and health staffs of Shahid Beheshti University of Medical Sciences in 2018.

Materials and Methods: It was a cross-sectional study. Participants were 220 adults with five different specialties, which were randomly selected from SBMU of Tehran. Data were collected using researcher-made questionnaire then, it was filled by relevant field specialists. Knowledge, attitude and practice questionnaire included 45 items, which was developed through literature review. Knowledge, attitude and practice had (12 items) (18 items) and (15 items), respectively. The demographic questionnaire included information on age, gender, level of education, occupation, and marital status. Content validity was calculated by content validity ratio (CVR) and content validity index (CVI). Reliability was evaluated using test-retest and by Cronbach's Alpha coefficient, internal consistency was calculated values %0.81 which considered as satisfactory.

Results: The mean age of the studied population was 37.70± 8.07 (23-67) years. Of 220 subjects 80 (36.4%) were male and 140 (63.6%) were female. In evaluating knowledge, attitude and practice in the men and women, mean and standard deviation of knowledge were estimated at good level and one-way ANOVA analysis showed significant differences between women and men (p 0.019). There was no significant difference in men and women related to attitude (p 0.92) and practice (p 0.38).

Conclusion: Knowledge of subjects was estimated at good levels. Women`s knowledge was significantly higher than men. Attitude and practice in men and women were not significantly different. Attitude and practice could be increased because there was consensus to vaccine among the specialists to prevent human papillomavirus.

Poster 27

Future directions in Immuno-Oncology

A MelARV Vaccine with Mutations in the Immunosuppressive Domain Elicits Increased Immunity and Elimination of Established Tumors in Mice

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Endogenous retroviruses (ERVs) are mainly silenced in healthy tissues, but they become reactivated in cancer. Their envelope (Env) protein contains an immunosuppressive domain (ISD) that promotes immune evasion and tumor progression. To investigate ERV-targeting in mice, the murine melanoma-associated retrovirus (MelARV) Env protein was chosen as a target for our virus-like vaccine technology. To potentially increase its efficiency, we mutated the vaccine's Env ISD abrogating its immunoinhibitory properties.

Vaccine-induced cellular immune responses were tested in vaccinated BALB/c mice by restimulating mouse splenocytes and staining CD8+ T cells intracellularly for IFNγ and TNFα, detected by flow cytometry. The vaccine`s therapeutic efficacy was tested on established CT26 tumors in BALB/c mice 10 days after the challenge. Anti-PD1 treatment was given four times every 3-4 days. Survivors were rechallenged with a distinct cancer cell line (4T1) to assess cross-protection.

Immunization with the modified vaccine generated strong CD8+ T cell responses against MelARV. ISDmut in combination with anti-PD1 antibodies, showed high curative efficacy (80% survival) against established CT26 tumors, compared to the wild type (30%) and control (22%). Furthermore, ISDmut prevented growth of 4T1 cancer cells in almost 40% of the mice that cleared CT26 tumors.

Mutation of the vaccine ISD significantly enhanced MelARV-specific T cell responses. ISDmut in combination with a-PD1 treatment prevented the growth of established CT26 tumors and protected against rechallenge with a distinct cancer cell line (4T1). Therefore, our ISDmut vaccine can be used therapeutically and prophylactically against MelARV-expressing tumors, with the prospect of translation into a human ERV-targeting vaccine.

Poster 28

Future directions in Immuno-Oncology

LAG-3 Expression in Tumor Environment of Feline Mammary Carcinoma Subtypes

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Background: Several clinical trials are investigating alternative immune checkpoint targets, such as lymphocyte activation gene-3 (LAG-3, CD223), as well as, potential synergistic effects of combined immune checkpoint inhibitors, including anti-LAG-3 with anti-PD-1. Similar to women, feline mammary carcinoma (FMC) is one of the most common causes of cancer-related death in female cats, wherein more than 80% are malignant and clinically very aggressive.

Objectives: The identification of novel diagnostic biomarkers and therapeutic targets is needed, not only to improve the clinical outcome of cats with mammary carcinoma but also because FMC shares clinicopathological, histopathological and epidemiological features with human breast cancer.

Methods: In this study, a set of 62 animals were included, 48 female cats with mammary carcinoma and 14 healthy controls that underwent for ovariohysterectomy. The cases were recruited from Small Animal Hospital of the Faculty of Veterinary Medicine/ULisbon and private clinics around Lisbon. Immunohistochemistry was carried out to optimize the detection of the LAG-3 and PD-1, in human tonsil and feline lymph node samples, and after this step, immunofluorescence was performed to evaluate the LAG-3 and PD-1 expression in total (t), stromal (s) and intratumoral (i) tumor-infiltrating lymphocytes (TILs).

Results: Preliminary results showed that triple negative mammary carcinomas had higher densities of TILs overexpressing tLAG-3 and sLAG-3, comparing with luminal and HER2-positive subtypes. Moreover, all tumors that expressed LAG-3 also contained PD-1. However, no statistically differences were observed regarding the co-expression of PD-1 and LAG-3 in the different subtypes.

Conclusion: These results suggest that the use of combinatorial immunotherapies targeting multiple tumor microenvironment immune checkpoints could increase the therapeutic efficacy, particularly in cats with triple negative mammary carcinoma subtype.

Poster 29

Future directions in Immuno-Oncology

H_1-antihistamines Desloratadine and Loratadine Show Therapeutic Potential in Tumors Responsive to Anti-Immune Checkpoint Inhibition

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Background: Evidence that H_1-antihistamines may be effective against tumors is mounting[1-12], and while several potential mechanisms have been proposed, most are either wholly or partly histamine receptor H_1 independent[2-5,8,9], possibly involving immunological pathways[15]. We have previously found that use of desloratadine and loratadine is associated with improved survival in both breast cancer[16] and melanoma (submitted).

Objectives: We set out to find whether a similar association can be seen in tumors with and without a known response to treatment with anti-immune checkpoint inhibition, such as anti-CTLA-4 or anti-PD-1.

Methods: We investigated survival and use of desloratadine, loratadine, and four other common H_1-antihistamines among 424,181 Swedish patients with ten types of immunogenic and six non-immunogenic tumors.

Results: We found that desloratadine use was associated with an improved survival for all the immunogenic tumors, but none of the non-immunogenic ones. Loratadine use was also associated with improved survival for some of the immunogenic tumors. Use of other antihistamines was not consistently associated with improved survival.

Conclusion: Our hypothesis is that our findings result from immune checkpoint inhibition. We believe that there is already a compelling case for initiation of clinical trials of desloratadine and loratadine as adjuvant treatment of both breast cancer and melanoma. Based on what we present here, that pool should be extended to include other immunogenic tumors, with priority given to trials of desloratadine as treatment of pancreatic cancer and other tumors with dismal prognoses and limited treatment options.

1. Faustino-Rocha AI, Ferreira R, Gama A, Oliveira PA, Ginja M. Antihistamines as promising drugs in cancer therapy. *Life sciences.* 2017;172:27-41.

2. Blaya B, Nicolau-Galmes F, Jangi SM, et al. Histamine and histamine receptor antagonists in cancer biology. *Inflammation & allergy drug targets.* 2010;9(3):146-157.

3. Jangi SM, Diaz-Perez JL, Ochoa-Lizarralde B, et al. H1 histamine receptor antagonists induce genotoxic and caspase-2-dependent apoptosis in human melanoma cells. *Carcinogenesis.* 2006;27(9):1787-1796.

4. Jangi SM, Ruiz-Larrea MB, Nicolau-Galmes F, et al. Terfenadine-induced apoptosis in human melanoma cells is mediated through Ca2+ homeostasis modulation and tyrosine kinase activity, independently of H1 histamine receptors. *Carcinogenesis.* 2008;29(3):500-509.

5. Nicolau-Galmes F, Asumendi A, Alonso-Tejerina E, et al. Terfenadine induces apoptosis and autophagy in melanoma cells through ROS-dependent and -independent mechanisms. *Apoptosis: an international journal on programmed cell death.* 2011;16(12):1253-1267.

6. Garcia-Quiroz J, Garcia-Becerra R, Barrera D, et al. Astemizole synergizes calcitriol antiproliferative activity by inhibiting CYP24A1 and upregulating VDR: a novel approach for breast cancer therapy. *PloS one.* 2012;7(9):e45063.

7. Garcia-Quiroz J, Garcia-Becerra R, Santos-Martinez N, et al. In vivo dual targeting of the oncogenic Ether-a-go-go-1 potassium channel by calcitriol and astemizole results in enhanced antineoplastic effects in breast tumors. *BMC cancer.* 2014;14:745.

8. Ellegaard AM, Dehlendorff C, Vind AC, et al. Repurposing Cationic Amphiphilic Antihistamines for Cancer Treatment. *EBioMedicine.* 2016;9:130-139.

9. Laverdiere I, Boileau M, Neumann AL, et al. Leukemic stem cell signatures identify novel therapeutics targeting acute myeloid leukemia. 2018;8(6):52.

10. Varbanov HP, Kuttler F, Banfi D, Turcatti G, Dyson PJ. Screening-based approach to discover effective platinum-based chemotherapies for cancers with poor prognosis. *PloS one.* 2019;14(1):e0211268.

11. Fernandez-Nogueira P, Noguera-Castells A, Fuster G, et al. Histamine receptor 1 inhibition enhances antitumor therapeutic responses through extracellular signal-regulated kinase (ERK) activation in breast cancer. *Cancer letters.* 2018;424:70-83.

12. Soule BP, Simone NL, DeGraff WG, Choudhuri R, Cook JA, Mitchell JB. Loratadine dysregulates cell cycle progression and enhances the effect of radiation in human tumor cell lines. *Radiation oncology (London, England).* 2010;5:8.

13. Kuzu OF, Toprak M, Noory MA, Robertson GP. Effect of lysosomotropic molecules on cellular homeostasis. *Pharmacological research.* 2017;117:177-184.

14. Aits S, Jaattela M, Nylandsted J. Methods for the quantification of lysosomal membrane permeabilization: a hallmark of lysosomal cell death. *Methods in cell biology.* 2015;126:261-285.

15. Crawford G, Hayes MD, Seoane RC, Ward S, Dalessandri T, Lai C. Epithelial damage and tissue gammadelta T cells promote a unique tumor-protective IgE response. 2018;19(8):859-870.

16. Olsson HL, Einefors R, Broberg P. Second generation antihistamines after breast cancer diagnosis to improve prognosis both in patients with ER+ and ER- breast cancer. *Journal of Clinical Oncology.* 2015;33(15_suppl):3062-3062.

Poster 30

Future directions in Immuno-Oncology

Interest of Mannose 6-Phosphate Analogues for Therapeutic Antibodies Engineering

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Currently, therapeutic antibodies arise as a strategic choice for cancer therapies. Their action mainly lies in the stimulation, the neutralization, or the inhibition of a specific target. However, the target is rarely degraded. Here, the objective is to internalize the target in cells via the cation-independent mannose 6-phosphate receptor (M6PR) to degrade it in the lysosomes. For this purpose, analogues of mannose 6-phosphate functionalized on aglycone position (called AMFA) are grafted to therapeutic antibodies. This engineering of the oligosaccharidic chains on the Fc fragment does alter neither the structural conformation nor the target specificity of the antibody. These AMFA analogues allow the antibodies to be recognized by the M6PR with a high affinity. Thanks to this specific binding, a higher cell uptake of AMFA-grafted antibodies and their antigens is demonstrated and associated with a decreased antigen activity. In conclusion, this innovative glycoengineering technology can generate a new class of therapeutic antibodies.

Poster 31

Future directions in Immuno-Oncology

Hyperoxic Treatment Modulates Inflammatory Responses in the Tumor Microenvironment.

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[2]*CIBERONC, Biomedical Research Centre in Cancer, Spain*
[3]*Program in Solid Tumors, CIMA-University of Navarra, Spain*
[4]*Hematology-oncology, Navarra Health Research Institute (IDISNA), Spain*

Background

The conditions of tumor cell growth are characterized by low oxygen supply (hypoxia) caused by insufficient blood delivery. Hypoxic cancers have a strong invasive potential, metastasis, and resistance to therapy. The key regulator of adaptation to tumor hypoxia is hypoxia inducible factor 1-α (HIF-1α). Hyperoxia could be a clinically relevant treatment to fight tumors that present with hypoxia through the modification of tumor microenvironment.

Objectives

We aimed to study how supplemental oxygen can modulate tumor adaptation to hypoxia through re-wiring metabolic adaptation and the inflammatory response.

Methods

Western Blot to evaluate how hyperoxia affected hypoxic mimetic-induced HIF-1α accumulation in uveal melanoma and normal epithelial lung cells. Glycolytic activity using Abcam Assay kit. Crystal Violet Assay for determining proliferation. Inflammatory responses by qPCR, Abcam Human Inflammation Antibody Array and NF-kB pathway using Promega Dual-Luciferase® Reporter Assay.

Results

Our data suggested that hyperoxia prevented HIF-1α accumulation induced by hypoxia mimetic. Hyperoxia decreased glycolytic activity, which is upregulated in cancer, and cell proliferation. There is an up-regulation in the expression of genes involved in inflammatory pathways and, interestingly, non-tumor cells were much less affected. The inflammation array revealed an immunomodulatory effect of hyperoxia on the tumor microenvironment. An in vivo study of lung metastasis in mice showed a strong reduction in the incidence of metastatic foci in the hyperoxia-treated group.

Conclusion

Counteracting hypoxia with supplemental oxygen modifies tumor progression and slows down metastatic spread. This could be due to its action as an immunomodulatory therapy in which tumor cells are more vulnerable.

Poster 32

Future directions in Immuno-Oncology

Active Immunization against Human Endogenous Retrovirus Type K (HERV-K) as an Immunotherapeutic Strategy against Solid Tumors

Peter Johannes Holst[1], Emeline Ragonnaud[1], Lasse Neukirch[1], Joana Daradoumis[1], Lara Duvnjak[1], Amaia Bermejo[1], Bang Nguyen[1], Silke Schroedel[2], Christian Thirion[2], Anne-Marie Carola Andersson[1]

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[2]*Vector Development, Sirion Biotech, Germany*

Background: Endogenous retrovirus are well establish natural cancer immune targets in mice and men. In humans, the HERV-K virus family is the most recently active. HERV-K Gag and Env genes are reported as structurally intact, and detected in human cancers, including on cell surfaces and exosomes, with minimal detection in healthy tissues.

Objectives: To establish if a self-surface exosomal antigen benefit from a collaborative B cell, CD4+ T cell and CD8+ T cell immune response

Materials and methods: HERV-K Gag and Env consensus sequences were encoded in human adenovirus type 5 and 19a/64 adenoviral vectors with or without point mutations in a putative immune suppressive domain (ISD). Cellular effects was characterized in tumor and antigen presenting cells. Immune responses were analyzed in vivo and tumor control experiments performed with HERV-K expressing RENCA and with CT26 cells using adaptive transfer of immune cells.

Results: HERV-K transgene expression let to the formation of virus like particles with increased expression of ISD mutated antigen. Adenoviral vectors expressing HERV-K Gag and Env were highly immunogenic with potent T cell and antibody responses rapidly inducible.

Adoptive transfer experiments established that CD8+ T cells alone lost tumor control whereas combinations of CD8+ and CD4+ T cells and B cells exhibited tumor control in most animals.

Conclusions: The HERV-K-ISDmut antigen holds promise for directing a broad and effective immune response to a major proportion of human cancers.

Poster 33

Future directions in Immuno-Oncology

The Turnaround of a Nightmare
How a Life-Threatening Interventional Complication Triggered a Sustainable Immunotherapeutic Response in End-Stage Pseudomyxoma Peritonei

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[3]*Gastroenterology, St.Josefskrankenhaus Heidelberg, Germany*
[4]*Oncology, Onkologische Schwerpunktpraxis Heidelberg, Germany*
[5]*Institute of Pathology, University Hospital Heidelberg, Germany*

Background:

Characterized by dissemination of atypical mucinous cells within the abdominal cavity, Pseudomyxoma peritonei (PMP) is an extremely rare and usually slowly progressive malignancy. Despite aggressive treatment (CRS/HIPEC) PMP frequently recurs with significant morbidity & mortality.

Case history & presentation:

In May 2015 debulking laparotomy was performed on a 34-year-old female teacher diagnosed with a widespread peritoneal carcinomatosis. Histology revealed a low-grade mucinous neoplasm of the appendix (LAMN: pT4a; pN0; pM1a, G1; R1). Following a diagnosis of pseudomyxoma peritonei stage IV, the patient received an aggressive treatment regimen (8 x FOLFOX & 2x CRS & HIPEC) for 3 years, while having repeated relapses. In October 2018 the patient was hospitalized with obstructive ileus. A subsequent salvage laparotomy failed due to hostile abdomen, resulting in PEG drainage. Thereafter, tumor-board advice from 2 comprehensive cancer centers independently recommended best supportive care. The patient was transferred for implantation of a permanent ascites–drainage, presenting in devastating condition despite PEG-drain and parenteral nutrition.

Complication:

Two days after a CT-guided ascites-drain implant the patient returned complaining of left abdominal pain and rectal bleeding. Fluoroscopy revealed a complete piercing of the descending colon by the drain. The drain was removed immediately.

Outcome:

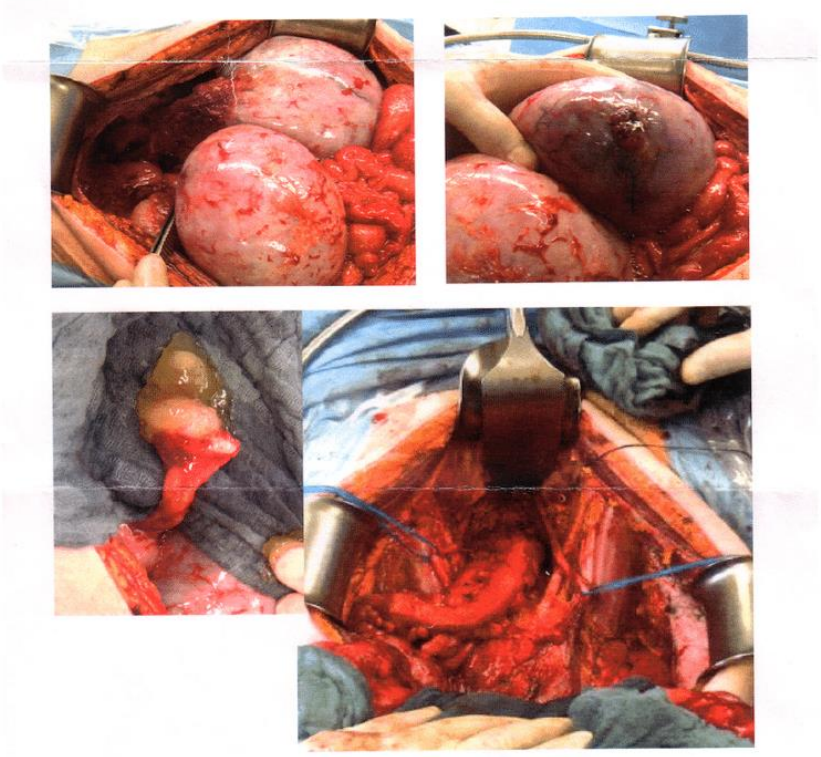
The patient was hospitalized with severe peritonitis. Therapy with Piperacillin/Tazobactam was started and microbiological sampling of ascites revealed infection with Escherichia coli and Enterococcus faecalis. After initial deterioration, clinical and laboratory findings normalized. MRI showed severe inflammatory response of the peritoneal surface on DWI and distinct reduction of ascites. The patient was discharged in surprisingly good condition. One year after the failed ascites drainage the patient is in a clinically highly ameliorated condition, with no need for any further ascites puncture. Abdominal MRI confirms nearly complete vanishing of mucus collection in the main peritoneal cavity.

Conclusions:

The unintended gut-microbiome exposure to the peritoneal cavity has revealed a highly significant antitumoral effect in a case of progressive end-stage PMP. The observed immunological mechanism which led to this sustainable tumor regression may become a game changer in the therapeutic approach for advanced PMP and opens up a promising path for future research.

Surgical situs during tumor debulking

Ovarectomy; Hysterectomy; Adnexectomy; Appendectomy; Omentectomy; Peritonectomy; Lymph node resection;



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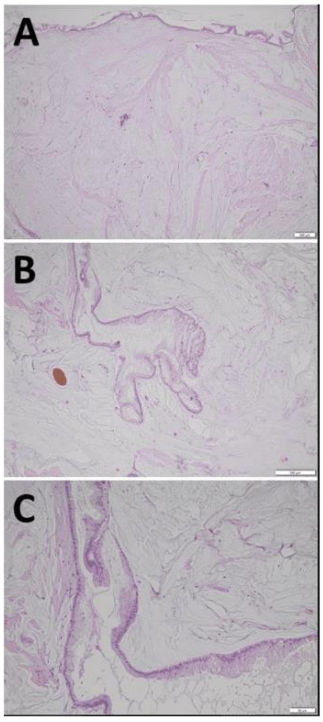
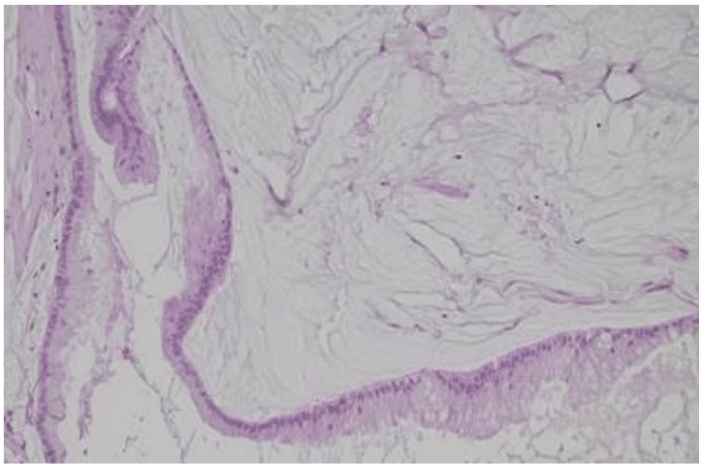
Histology of abdominal biopsies (A-C) showed a low-grade mucinous neoplasia with abundant mucinous material and low-grade mucinous epithelia, consistent with pseudomyxoma peritonei.





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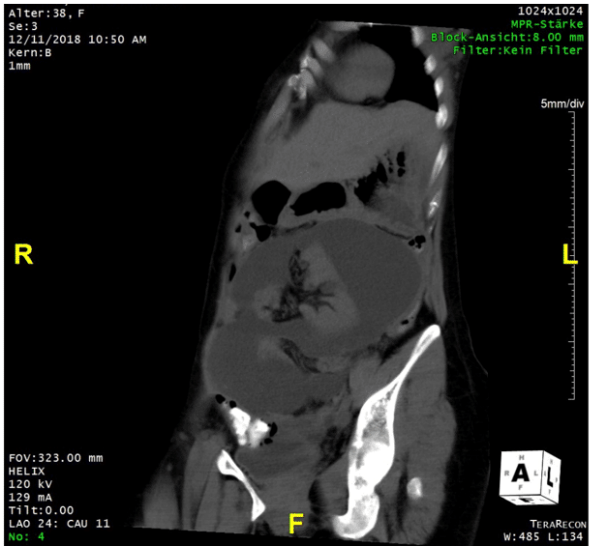
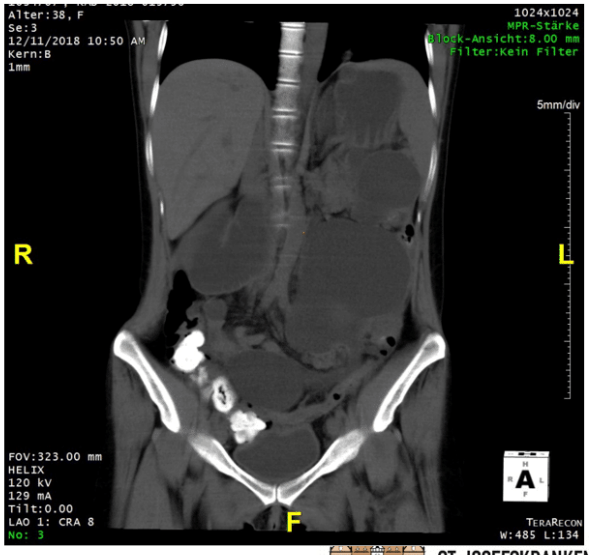
CT – view before drainage

Illustrates diffuse spread of mucin within peritoneum &



Mechanical obstruction of the intestines



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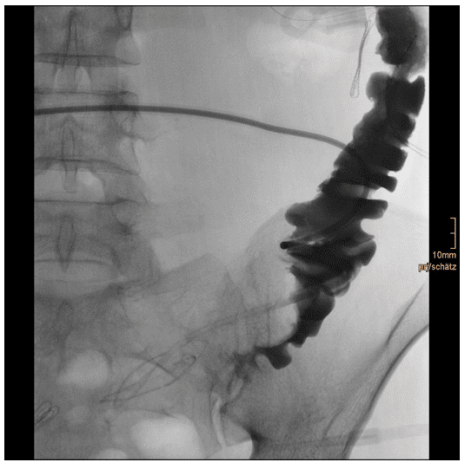
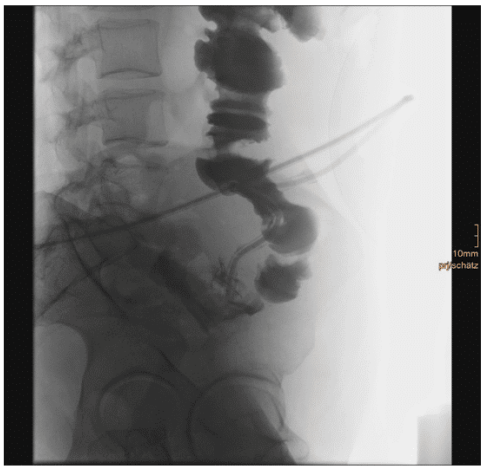
Flouroscopy: prior drain removal

Colonography of descending colon via PleurX drain



lateral view reveals in & out perforation of colon by drain



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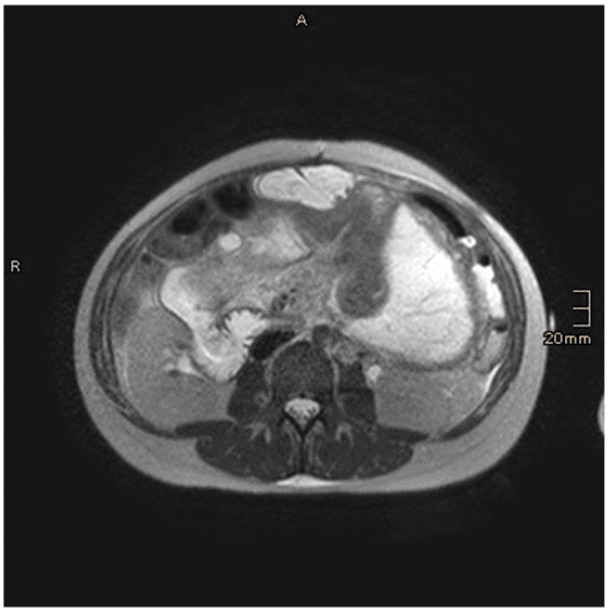
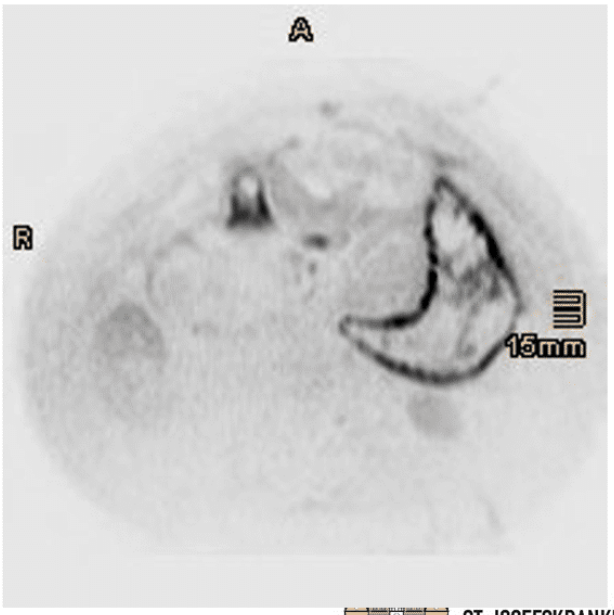
MRI one week after drain removal reveals massive immun-response of peritoneum to gut-microbiome exposure

T2



DWI (B-value 1000)



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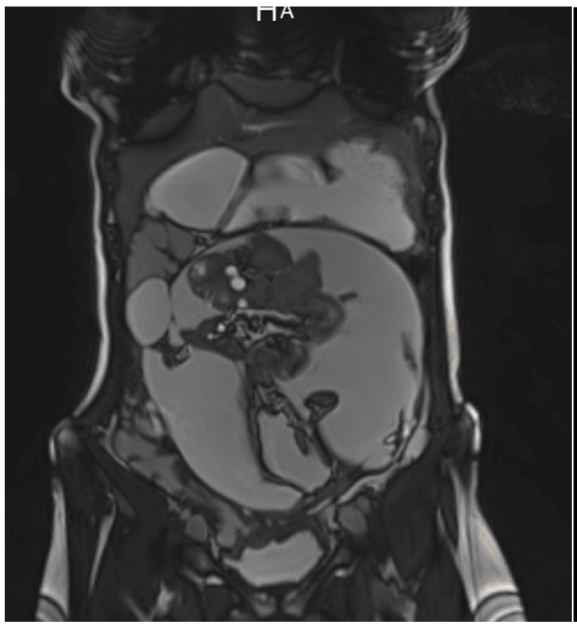
Z

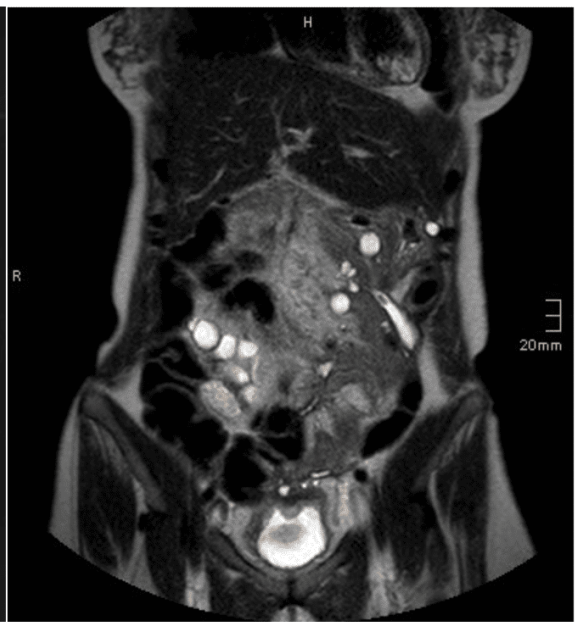
6 weeks before 13 Month after

Gut Microbiome Exposure

MRI Oktober 2018



MRI Januar 2020



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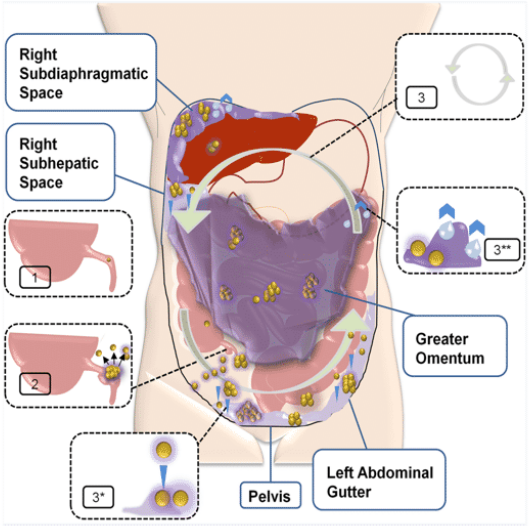
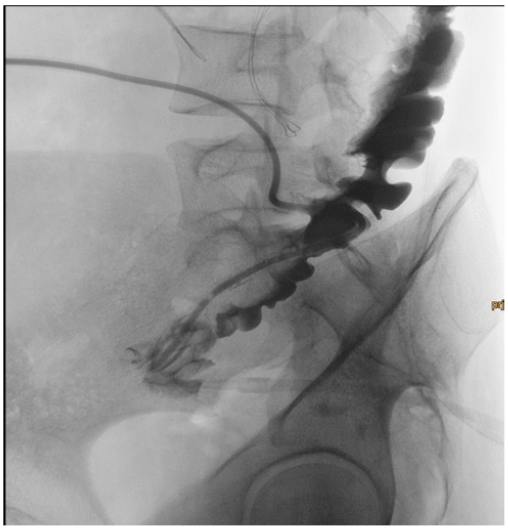
Conclusions:

What almost killed the patient, was a perforation of the appendix with invasion of the peritoneal cavity by globlet tumor cells



Amini et al. Orphanet Journal of Rare Diseases 2014, 9:71

What almost saved the patient, was a perforation of the desc. colon with invasion of the peritoneal cavity by gut microbiota



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Poster 34

Future directions in Immuno-Oncology

Geriatric and Pharmacological Consultation in the Hospital`s Hemato-Oncology Unit

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Background

New cancer medications based on biological mechanism are safer for use. Their efficiency, together with relatively high safety, allow a vast of geriatric patients to use those medications. The geriatric patient usually has chronic diseases and consumes chronic medications, and so a specific compatibility is needed between those, and the medication used for oncological protocols.

Objectives

The aim of our project is to supply a personal package, including geriatric evaluation and pharmacological consultation, for geriatric patient that are being treated in the hemato-oncology unit in Carmel Medical Center. All for improving the patient treatment in all aspects and providing him with a "tailored suit" treatment.

Method

1. A multidisciplinary team was formed: a geriatrician, hematologist, a pharmacist, and a nurse.

2. Patient aged 75+ who are scheduled to begin chemotherapy are invited for a 1-hour consultation with the team.

3. The team perform a full geriatric and pharmacologic assessment.

4. After the meeting a detailed summery is provided including recommendations for the hematologic doctor, for the family physician and for the patient himself

Results

In 2021, 20 patients were examined.

All were given detailed recommendation letters. Drug protocols were changed according to patient`s specific clinical data.

Conclusions

There is an advantage for multidisciplinary team in treating the geriatric patient in the hemato-oncology unit:

· Performing a detailed assessment of the patient`s ability both clinical and mental to undergo chemotherapy.

· Performing medication revaluation of total drug consumption.

· Providing the medical team with an advanced and accessible pharmacological knowledge.

Poster 35

Future directions in Immuno-Oncology

Targeting Sphingolipids May Be an Effective Approach to Rebalancing the Tumor Microenvironment's Immune Response

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[2]*Oncology, CTI, USA*

[3]*Comprehensive Cancer Center, University of Cincinnati, USA*

[4]*Cancer Center, University of New Mexico, USA*

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[6]*Neuro-Oncology, MD Andersen Cancer Center, USA*

[7]*Cancer Center, University of New Mexico, USA*

[8]*Neuro-Oncology, University of Kentucky Cancer Center, USA*

[9]*The Taussig Cancer Institute, The Cleveland Clinic Foundation, USA*

Background:

Sphingolipids are bioactive signaling molecules implicated in multiple cellular processes and molecular pathways. Sphingosine-1-phosphate (S1P) is a key sphingolipid that induces cancer cell proliferation, activates multiple oncogenic pathways, and stimulates immuno-suppressor cells; ceramides, a different class of sphingolipids, induce apoptosis, down regulate oncogenic pathways, and stimulate immuno-effector cells.

BXQ-350 is a novel biologic that modulates sphingolipid metabolism, lowering S1P and increasing ceramides. BXQ-350 was investigated in a Phase 1 dose-escalation safety study in an all-comer cancer patients with advanced solid malignancies to determine its safety profile and its potential clinical activity as a single agent.

Objectives:

To determine which immune-suppressor / effector cells BXQ-350 impacts preclinically and clinically.

Methodology:

Preclinical experiments (in vitro, in vivo and ex vivo) and analysis of biomarker samples, collected during the Phase 1 clinical study, were performed to determine the impact of BXQ-350 on S1P and ceramide concentrations, cytokines, and immuno-effector / -suppressor cells.

Results:

Preclinical and clinical results revealed that BXQ-350 decreases S1P and increases ceramide concentrations in most of patients experiencing a clinical benefit. Results also showed that it repolarizes macrophages towards the M1 phenotype, inhibits MDSCs' proliferation, and expands CD4/8+ Tcells and enhance their cytotoxicity. Also, it seems that there is a concurrent change in circulating levels of pro/antitumoral cytokines.

Conclusion:

While these results are exploratory and preliminary and need to be further confirmed, they suggest that lowering the immuno-suppressor S1P and simultaneously increasing immuno-effector ceramides may be an effective strategy to rebalance the immune system towards tumor suppression.

Poster 36

Future directions in Immuno-Oncology

High Immunogenicity of a Personalized Antitumor Vaccine Made from 4T1 Breast Carcinoma Tissue and Molecular Adjuvants that Reprogram Dendritic Cells and Myeloid Suppressors.

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Department of Immune Biotechnology, FRC Institute of Immunology, Moscow, Russia

Background: Neoantigenic vaccines induce specific adaptive immune responses that can detect and eliminate the cancer cells. Despite all advantages, the neoantigenic vaccine technology is not sufficiently developed to be replicated for millions of patients.
Objectives: To estimate the immunogenicity of a personalized multi-antigen cancer vaccine (PMV-4T1) made from 4T1 carcinoma tissue with the addition of molecular immunoadjuvants.
Methods: The solid tumor was surgically removed eleven days after the subcutaneous inoculation of 15,000 4T1 cells in BALB/c mice. Tumor tissue was used to prepare a multi-antigenic vaccine and molecular adjuvants were added that activate dendritic cells and reprogram myeloid suppressors. Antigen-reactive IFNγ-secretory T cells were counted using ELISPOT with FACS-sorted CD4 and CD8 T cells. Serum antibodies specific to 4T1 intracellular antigens were measured using ELISA while flow cytometry was applied to detect antibodies specifically recognizing 4T1 surface antigens.
Results: An immunization with PMV-4T1 induced intensive Th1-type antitumor immune responses. The IFNγ-secreting CD4 and CD8 T effector cell counts in the spleen reached 6,000 and 1,000 (per 1 million), respectively. The numbers of CD4 and CD8 T effector memory cells recognizing 4T1 carcinoma antigens reached 4000 and 3500 (per 1 million), respectively, which was 10-times higher than the level of T-cell antitumor immune responses in mice with a progressively growing 4T1 tumor. The intensity of T effector and T effector-memory cell production and the CD4-to-CD8 T cell responses ratio strongly depended on the composition of molecular adjuvants. Immunization with PMV-4T1 induced high levels of antibodies specific for both intracellular and surface antigens of 4T1 cancer cells.
Conclusion: The results obtained evidence for the high efficacy of our method of personalized antitumor vaccination.
This study was supported by the Russian Science Foundation (project №20-15-00391).

Poster 37

Selecting patients for treatment with immunotherapy

The Effect of Prognostic Nutritional Index on Survival in Patients Receiving Immune Checkpoint Inhibitor Therapy

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[2]*Public Health Department, ÇUkurova University, Turkey*

Background: It is important to know the prognostic factors in cancer patients receiving immunotherapy.

Objective: In this study, we aimed to show the effect of prognostic nutritional index (PNI) on survival in patients receiving immune checkpoint inhibitors.

Methods: 117 patients who received immune checkpoint inhibitor (ICI) therapy in the Oncology Department of Çukurova University between 2010 and 2022 were included in the study.

Results: The patients' 75.2% are male and 24.8% female. The median age of the patients was 59 years. Diagnostic distribution of the patients included in our study, 34 malignant melanoma, 32 lung cancer, 11 renal cell carcinoma, 9 hepatocellular carcinoma, 9 hodgkin lymphoma, 6 mesothelioma, 5 nonhodgkin lymphoma, 3 bladder cancer, 2 head and neck tumor and 1 gastric cancer, 1 testicular cancer, 1 prostate cancer, 1 osteosarcoma, 1 colon cancer, and 1 Kaposi`s sarcoma. The distribution of immune checkpoint inhibitor used in the patients included in our study was nivolumab in 83 patients, pembrolizumab in 14 patients, atezolizumab in 9 patients, ipilimumab in 7 patients, and nivolumab-ipilimumab in 4 patients. Of the patients followed, 50 were alive and 67 were dead. 117 patients were studied. It was found that the area under the curve of the PNI was important in predicting the exitus status at the end of 1 year and the optimum cut-off value was 42.75(sensitivity 55%, specificity 75%). Patients with a PNI at admission 42.75 had a 3.53-fold increased risk of death.

Conclusion: PNI is an important prognostic and predictive marker for mortality in patients using ICI.

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Selecting patients for treatment with immunotherapy

Rectal non-Hodgkin's Lymphoma Presented as Ulcerative Proctitis in Practice

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Non-Hodgkin's lymphomas are malignant tumors of lymphatic system, characterized by extranodal involvement especially in gastrointestinal tract. Primary rectal lymphoma is a very rare condition, accounting for only %0.1-0.6 of colorectal malignancies. Here we report a case of non-Hodgkin's extranodal marginal zone lymphoma (MALT) in rectum and discuss literature.

A 25-year-old female admitted with abdominal pain, rectal bleeding, alternating diarrhea. Colonoscopy revealed ulcerative proctitis, Mayo 2. Rectal biopsy showed that she had stage 2A B cell non-Hodgkin extranodal marginal zone lymphoma (MALT). The results of immunohistochemical staining were; CD20(+), CD23(+), CD43(+), IgM(+), Bcl-2(+), CD10(-), Cyclin D1(-) and Ki67=%20. The patient received CHOP (cyclophosphamide, doxorubicin, vincristine, and prednisone) chemotherapy. After second session of CHOP, a repeated PET scan assessed partial metabolic progression in the rectum. Bone marrow biopsy showed no malignant infiltration, hence bone marrow transplantation was not needed. CHOP treatment was finished after 6th session. During the follow up, at the 7th year of her disease, the patient achieved a full remission.

Primary rectal lymphoma is a rare condition. It is generally seen in men older than 50 years old. The main clinical manifestations are abdominal pain, rectal bleeding and changing in bowel habits. Colonoscopy with biopsy and computerized tomography (CT) are valuable tests for diagnosis. CHOP chemotherapy is the first line therapy in disease management.

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The Road from Genetic Testing to Immunotherapy in Pediatric CNS Tumors

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Background
Pediatric cancers present a distinct group from their adult counterparts, in their rapid growth, their developing tissue origin, somatic mutation burden and driver gene numbers. Over 50% of all tumors harbor potentially druggable driver genes. Survival rate of pediatric cancer has increased to 84% as of 2020. Current treatment protocols are highly toxic, yet prognosis remains poor for high-risk and refractory cancers. In these patients personalized treatment approaches can hold key to recovery, including immunotherapies.
In this study we present how next generation sequencing (NGS) directed choice of therapy in pediatric central nervous system (CNS) tumors leading to a patient receiving monoclonal antibody (trastuzumab) therapy.

Methods
23 high-risk and relapsed/refractory CNS tumor samples were included in our study, from which 15 were deemed eligible for sequencing.
We identified NGS-based alterations and implement targeted treatment where feasible.

Results
The percentage of actionable alterations was 26%. 80% (12/15) of patients received targeted therapy (11 kinase inhibitors, 1 monoclonal antibody). Clinical benefit was only seen in one patient treated with immunotherapy.
A 11 year old female patient presented with dizziness, nausea, blurred vision and clumsiness in 2016. The MRI examination found a tumor in the posterior fossa with multiple metastases in the uprasellar region and the ventral horn of the left ventricle. After surgical resection, the histology examination confirmed a non-WNT/non-SHH (WHO GRADE IV) tumor witouth C-myc or N-myc gene amplifications.
Her oncology treatment consisted of the MBL 2008 protocol HR arm (chemotherapy) in November 2017, total CNS irradiation in December, and autologous bone marrow transplantation (Bu-Thio-Ve) in August 2018.
She relapsed in July 2020, when the MRI scan showed multiplex infa-and supratentorial malformations. In August 2020 she received multi-agent oral antiangiogenic (metronomic) therapy: thalidomide+celecoxib+etoposide/cyclophosphamide).
Progression occured in July 2021 in the posterior temporo-medial region as a new finding. Her NGS sequenging showed a ERBB-P489L driver gene mutation, which was shown to be sensitive to irreversible HER2 inhibitors in leukemia cell lines in a preclinical trial. She received Topotecan+ Temozolomide treatment alternating with Trastuzumab. ON her last MRI examination all of the lesions regressed and new lesions could not be detected. After 6 months of stable disease and 2 control MRI examinations the therapy was suspended due to the lack of further regression and strong side effects.
Conclusions
NGS sequencing is feasible in patient selection for monoclonal antibody therapy.
Due to global collaborative initiatives, the integration of genomic and (pre) clinical data can be used to direct the treatment choice and further clinical trials/drug development.

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IL-8, S100 and IL-10 as Independent Predictors of Response and Survival in Melanoma (MEL) Patients (pts) Treated with Immune Checkpoint Inhibitors (ICI)

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Background. Prognostic scores are needed to optimize ICI treatment. Methods: ICI treated advanced MEL and NSCLC pts were prospectively enrolled to test association between IL-1b, Il-2, IL-4, IL-5, IL-6, IL-8, IL-10, TNFα, GM-CSF, S100, LDH and DCR, PFS and OS. Blood collection: before the first 6 ICI cycles (T1-T6) and at each tumor assessment until PD or maximum for 2 years. Results: the first 43 MEL pts' blood samples analyses at T1- and T2-time points, showed: IL-6, IL-8, LDH, S100 and T2-IL-10 significantly higher for pts with PD (K-W test) as IL-8, IL-10, LDH and S100 median relative increase (RI) T1T2 (32vs -11%, 104 vs 41%, 4 vs 1% and 39 vs 0%). Multiple logistic analysis confirmed higher T2-IL-8 (34.22pg/ml) and IL-8-RI (1%) as independent factors (accuracy 88.6%) associated with a lower probability of DC (odd ratio-OR-=0.02, 95%CI: 0.00─0.18 and OR= 0.06, 95%CI: 0.00─0.65 respectively). Moreover, increased T2-S100 serum levels (0.15 μg/L) showed a trend (OR=0.16, p-value 0.0726) for PD. In multiple Cox regression: elevated T2-IL-8 (36.98pg/ml, HR=7.89, 95% CI: 2.66─25.78), T2-IL-10 (2.66pg/ml, HR=2.99, 95%CI: 1.09─8.49) and IL-8-RI (11%, HR=3.61, 95%CI: 1.27─13.81) were significantly associated with a worse PFS. Higher T2-IL-8 (31.55pg/ml, HR=15.67, 95%CI: 3.54─107.19), IL8-RI (30%, HR=5.12, 95%CI: 1.57─19.50), and T2-S100 (0.15μg/L, HR=14.35, 95%CI: 2.61─170.9) significantly associated with shorter OS. Conclusions: T2-IL8 and IL8-RI were independently associated to PD, PFS and OS, S100 with OS, and T2-IL-10 with PFS. These preliminary data will be validated in the whole cohort of 92 MEL pts already accrued. Ongoing further analyses. Funding: Italian Ministry of Health RF-2018-12367604 and IOV-IRCCS BIGID219SILE.

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Using Immunotherapy in Paediatrics - Targeted Therapy Usage Experience in a Medium Sized Paediatric and Adolescent Unit in Australia

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Background and aims: Increased use of targeted therapies is being seen in paediatric oncology with limited open studies, resulting in these medications mostly being available individually, at significant expense and resources. We review aspects of targeted therapy usage in a medium sized paediatric cancer centre on Melbourne, Australia treating over 75 new oncology patients from 0-19 years annually.

Methods: Patients were identified through the unit records; some were enrolled in a nationwide Precision Medicine Program (PRISM). Data was difficult to obtain, and tried to gain information on access to clinical trials, how the medications were obtained, costs if available and any issues, side effects or benefits.

Results: Over twenty patients were identified from October 2018 to December 2021. Of twenty-five patients enrolled on PRISM, twenty had recommendations of a single target, and three had multiple targets. There were over eighteen targets identified. Fifteen different medications were recommended. Fifteen of the twenty-three patients had accessible medications and eleven commenced on the recommended agents. Eight patients remain on the medication with stable disease or better. Non- PRISM patients commenced on either Government funded targeted therapies with established indications or novel agents suggested by pathology, biology and /or clinical course. Most patients were discussed at multidisciplinary meetings. Most needed individual access to medications, through hospital drug usage committee or special compassionate access from drug companies. Parent did not pay for medications.

Conclusion: Targeted therapies are being increasingly used in paediatric oncology, with potentially improved outcomes, but at significant resource and financial costs. Recommendations include improved records of patients on novel agents. Better systems will track usage, cost access, outcomes, and allow more cost/benefit analysis.

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The current drugs available in immunotherapy (and combinations)

The β-carboline Harmine Increases the MHC-I-Dependent Antigen Presentation Machinery and Improves the Therapeutic Benefit of Anti-PD-1

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Dual-specificity tyrosine-regulated kinase 1A (DYRK1A) inhibitor harmine displays a number of biological and pharmacological properties. We have previously reported that harmine, also referred to as ACB1801 molecule, increases major histocompatibility complex (MHC)-I-dependent antigen presentation on melanoma cells. Here we showed that ACB1801 upregulates the expression of Transporter associated with antigen processing proteins 1 (TAP1) in several murine and human cancer cells. Treatment of mice bearing melanoma B16-F10 with ACB1801 inhibits tumor growth and weight and induces a profound modification in the tumor immune landscape. Combining ACB1801 improves the therapeutic benefit of anti-PD-1 in B16-F10 melanoma-bearing mice. These results suggest that, through upregulating the expression of TAP1, ACB1801 can be combined with anti-PD-1/PD-L1 therapy to improve the survival benefit in cancer patients expressing low levels of TAP1. This statement is supported by our data showing that survival is significantly improved in melanoma and colorectal (CRC) patients expressing high TAP1 relative to those expressing low TAP1. Interestingly, we reported that high expression of TAP1 in melanoma and CRC patients was associated with increased expression of CD8 and NK cell markers and upregulation of proinflammatory chemokines.

The current drugs available in immunotherapy (and combinations)

Effectiveness of Conventional and Targeted Therapies in Pediatric Oncology

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Pediatric oncological treatment places a heavy burden on healthcare, economy and families worldwide. Conventional therapies (including surgical resection, chemotherapy and irradiation) cause both short and long-term detrimental side effects.
Personalized (targeted) therapies improve outcomes in high risk patients according to the MAPPYACTS trial in 2022.
Targeted therapies could improve survival with an acceptable side effect profile.
Our primary aim was to estimate the therapeutic effect of additional targeted therapies (including small molecule drugs, monoclonal antibodies, interleukins) compared to conventional treatments in pediatric oncology (comparative studies). Our secondary aim was to analyze all pediatric oncology studies focusing on targeted therapies (non-comparative studies).
Methods:
We conducted a systematic search on the 13th of October 2021, in five databases (MEDLINE, Embase, CENTRAL, Scopus, Web of Science). Out of 23343 hits and 17786 asticles remaining after duplicate removal, we found 236 eligible studies.
Upon analyzing the survival outcomes for pediatric patients treated with conventional therapies alone (control group) versus conventional therapies with additional personalized treatments (interventional group), we estimated the survival benefit of additional targeted treatments. We also compared the number of side effects between patients receiving targeted therapies only versus targeted therapies in combination with targeted treatments.
Results:
The event-free survival probability favored the interventional group, with a significant difference at 5 years follow-up time (survival probability 0.55 [0.50;0.60] vs. 0.44 [0.41;0.47]).Upon comparing the hazard ratios for events, we found that the targeted therapy group showed significantly improved likelyhood for event-free survival (HR=0.82, p=0.08). The overall survival also favored the interventional group (5 year survival probability 0.72 [0.64;0.81] vs. 0.64 [0.51;0.79]).

Upon examining the hazard ratio for mortality in the two groups, we could see a significant benefit of targeted therapy for survival. (HR=0.80, p=0,02).
The number of adverse events in the non-comparative studies were not statistically significantly different between the targeted therapy only group versus the targeted therapy and conventional therapy group. (Proportion: 0.869 [0.517;0.976] vs. 0.919 [0.805;0.969], p=0.558)

Strength: Comprehensive and complex meta-analysis in pediatric oncology implementing targeted therapies in hematological malignancies AND solid tumors.

Limitations: The comparative studies focis on hematological disease, solid tumors are underrepresented.

Conclusion:
Additional targeted therapy improves event-free survival and overall survival in pediatric oncology patients. Targeted therapies can be administered alone or in combination with conventional therapies with similar number of adverse events.
Since the population is heavily pretreated in both groups, further studies should be conducted implementing targeted treatments as first-line therapies to assess non-cumulative toxicity profiles.

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Inhibition of MIF by CPSI-1306 in Colitis-Associated Colorectal Cancer

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Background. Macrophage Migration Inhibitory Factor (MIF) is a key factor in chronic inflammatory processes, such as colitis. In line with this, MIF is overexpressed in several carcinogenic processes, such as colitis-associated colorectal cancer (CAC). Previous studies suggest that MIF enhances the development and malignancy of CAC, however, the effect of chemically inhibiting MIF in vivo in CAC still not completely understood. Objective. To determine the effect of the MIF inhibitor CPSI-1306 in vitro and in vivo. Methods. In vitro assays were performed on CRC cell lines treated with several CPSI-1306 doses per 48h; later, production of TNF-α, IL-6 and IL-10 were quantified. CAC was induced in 6-to 8-weeks-old female BALB/c mice, with azoxymethane via intraperitoneal, followed by 3 cycles of dextran sulfate sodium (DSS) (2%) in drinking water for 7 days with intervals of DSS-free water for 14 days. On day 40 post-induction (p.i.), CPSI-1306 (1mg/kg) was administered by gavage. Clinical evolution was recorded weekly. Mice were euthanized on day 68 p.i. and tumoral burden was measured. Results. Inhibition of MIF in cell cultures reduced the production of TNF-α, IL-6 and IL-10. In addition, CAC mice treated with CPSI-1306 had fewer clinical symptoms and smaller tumors, and a lower degree of malignancy. These preliminary results suggest that inhibition of MIF improves the prognosis of this pathology. Funding Support: This work was funded by the National Council of Science and Technology of Mexico (CONACYT), grant number (A1-S-10463) and the Support Program for Research Projects and Technological Innovation (PAPIIT)-UNAM, grant number (IN-217021).

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The current drugs available in immunotherapy (and combinations)

Pertuzumab, Trastuzumab, and Hyaluronidase-zzxf in A Patient of Locally Advanced Breast Cancer: Experience from the Delhi State Cancer Institute

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Background

Delhi State Cancer Institutes (DSCI) is an autonomous institution established by the Government of Delhi.Pertuzumab, Trastuzumab and Hyaluronidase-zzxf, a drug used for the treatment of HER2Neu positive breast cancer, is the first formulation to combine pertuzumab and trastuzumab, in one vial for subcutaneous injection.

Objective

A 39 year old patient who had HER2 Neu amplified invasive ductal carcinoma(grade III) breast cancer, presented to DSCI.The administration of an expensive new molecule that needs to be given free of cost in a government funded institute was a pivotal challenge. Patient's apprehension about receiving a new molecule needed to be allayed while working on the training of the nursing staff with regards to the oil based formulation administration by subcutaneous route.

Method

Patient was prescribed 6 courses of docetaxel, carboplatin, trastuzumab, pertuzumab (DCHP) based neoadjuvant chemotherapy (NACT), followed by response assessment.Patient and the nursing staff were counseled about the risks and benefits of the formulation and its administration technique.

Results

Patient has received 2 cycles of NACT till date and has experienced no discomfort while receiving the drug. Both the patient and the nursing staff were satisfied with the short drug administration time.

Conclusion

Response to the regimen is awaited. pCR will be assessed by the end of the planned treatment and will serve as a guide for identification of the ideal cohort of patients who will draw the maximum benefit after the drug administration.

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Use of Helminth-Derived Molecules to Improve Conventional Chemotherapy during Experimental Colorectal Cancer Development

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Background: Colorectal cancer (CRC) is among the deadliest neoplasias worldwide. 5-Fluorouracil (5-FU) is a chemotherapeutic agent utilized to treat CRC, notwithstanding its low efficiency. The factors that give rise to CRC include chronic inflammation; however, helminth infections have been reported to inhibit inflammatory responses. Despite this, the role of helminths in cancer development is still not thoroughly explored.

Objectives To determine the effects of excreted/secreted products of the helminth Taenia crassiceps (TcES) as an adjuvant with 5-FU in a murine model of colitis-associated colon cancer (CAC).

Methods Eight-to ten-week-old female BALB/c mice were induced to CAC with azoxymethane (12.5mg/kg) and dextran sulfate sodium (DSS) (2%) in drinking water. On day 54 after CAC-induction, mice were inoculated with 200µg of TcES 3-times per week, and on day 60, mice received 5-FU (30mg/kg) together with TcES or single 5-FU or saline and were euthanized on day 80.

Results. Treatment with TcES plus 5-FU reduced IL-1β, TNF-α, and IL-17 and inhibited colon tumorigenesis by downregulating genes related to drug resistance and KI-67 and Cyclin D1. Also, TcES plus 5-FU increased the recruitment of NK cells, which secreted more granzyme.

Conclusion Our study demonstrates a remarkable effect of TcES on suppressing ongoing colorectal cancer by downregulating proinflammatory and pro-tumorigenic signaling pathways, improving the 5-FU effect.

Funding Support: This work was funded by the National Council of Science and Technology of Mexico (CONACYT), grant number (37879), and the Program for Research Projects and Technological Innovation (PAPIIT)-UNAM, grant number (IN-212722).

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Toxicity management of Immunotherapy drugs

Fatal Immune Hemophagocytic Lymphohistocytosis Secondary to Sequential Treatment of Pembrolizumab Followed by osimertinib

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Background

The combination immune checkpoint inhibitor (ICI) and chemotherapy is the standard of care in first-line metastatic non-small-cell lung carcinoma (NSCLC) without molecular target. In case of metastatic EGFR-mutated NSCLC, osimertinib, a third generation EFGR tyrosine kinase inhibitor (TKI), is the gold standard. Side effects of immunotherapy, secondary to excessive inflammatory response, lead to a large spectrum of immune-related adverse events. Hematologic immune-related adverse events (Hem-irAEs) are rare and mostly fatal. There are currently no consensus on the diagnosis and the management of Hem-irAEs.

Case description

A 59-year old woman diagnosed with a metastatic EGFR (L747_T751N exon 19 deletion)-mutated NSCLC was treated by the combination pembrolizumab, an ICI anti-PDL1, plus chemotherapy followed by osimertinib at second-line. Four months after EGFR TKI beginning, she developed a grade IV pancytopenia (anemia 9.1 g/dL; thrombopenia 14 G/L; neutropenia 0.01 G/L) associated with hyperferritinea and hypertriglyceridemia. Osteomedullar biopsy concluded to hemophagocytosis lymphohistiocytosis. Despite high dose corticosteroid and immunosuppressive therapies including etoposide and cyclosporine, myelosuppression lasted. As no specific treatment could not be begin, the patient dead due to tumoral progression.

Conclusion

This case reported a rare cortico-resistant immune HLH syndrome induced by the sequential treatment of ICI followed by EGFR TKI. Physiopathological mechanism remain unclear. The lethality of Hem-irAEs implies close monitoring during and after ICI stopping and an early management consisting in steroid therapy.

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Toxicity management of Immunotherapy drugs

Financial Toxicity in Immunotherapy for Gastrointestinal Cancers: A Case Series from India

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Background:

Immunotherapy has changed the paradigm of modern cancer treatment. In India, immunotherapy is unaffordable to 99% of cases because of its cost.

Objectives:

To present our patient data on response to Immune checkpoint inhibitors (ICIs) in metastatic gastrointestinal (GI) cancers and the associated toxicities.

Methods:

We prospectively collected data of 6 patients with advanced GI cancers who received ICIs with or without chemotherapy at our center. The datasets had information on diagnostic tests, therapy response and the toxicity.

Results:

Our series included Hepatocellular carcinoma [HCC] (2), Rectal adenocarcinoma (2), Gastro-esophageal junction adenocarcinoma (1) and Esophageal squamous cell carcinoma [ESCC] (1). Five (83%) patients were dMMR/MSI-H. One patient (Rectum) had complete response after 2 years of therapy and one (HCC) had progression after 28 months. Commonest adverse event observed was financial toxicity in 4 (66%) patients that resulted in, increased interval during therapy in 2 patients, patient discontinued chemotherapy in one and one patient had to stop ICI and switched to mono-chemotherapy. Among all 6 patients, 2 (33%) had Grade II (CTCAE, V.5) transaminitis and dermatitis; 1 (16%) had Grade I diarrhoea, pain and mucositis with Grade III Anaemia and Non-neutropenic Fever; 1 (16%) had Grade III polyradiculopathy. All six patients are alive (median Progression Free Survival is 24 months) with good quality of life.

Conclusions:

Less than one percent of Indian patients can afford immunotherapy for managing advanced GI cancers. There is an urgent need to reduce the cost of immunotherapy in India.

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Tumor PD-L1 Expression and Molecular Profiling are not Associated with Immune Checkpoint Inhibitor-Induced Thyroid Dysfunction in Advanced NSCLC Patients

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Background: Immune checkpoint inhibitors (ICIs) have revolutionized the treatment of advanced non-small cell lung cancer (NSCLC). ICIs are frequently associated with thyroid-related adverse events.

Objectives: To investigate the association between patient characteristics, tumor PD-L1 expression and molecular profile with the development of thyroid dysfunction in NSCLC patients treated with PD-1/L1 inhibitors.

Methods: This single center, retrospective study included 107 NSCLC patients treated with PD-1 or PD-L1 inhibitors from April 2016 to July 2020. All patients were euthyroid at baseline and had at least two TSH measurements post-treatment initiation. The primary outcome was the difference in tumor PD-L1 expression in patients who developed any thyroid dysfunction versus those who remained euthyroid with treatment. Additional outcomes included the development of overt thyroid dysfunction, the association of specific molecular alterations with the development of thyroid dysfunction, and time to develop thyroid dysfunction as a function of tumor PD-L1 expression.

Results: Overall, 37 (34.6%) patients developed any thyroid dysfunction and 18 (16.8%) developed overt thyroid dysfunction. Tumor PD-L1 staining intensity was not associated with the development of thyroid dysfunction. TP53 mutation was less likely to be associated with any thyroid dysfunction (p 0.05) and no association was found between EGFR, ROS, ALK or KRAS mutations. There was no association between PD-L1 expression and time to develop thyroid dysfunction.

Conclusion: PD-L1 expression is not associated with the development of thyroid dysfunction in advanced NSCLC patients treated with PD-L1/PD-1 inhibitors, suggesting that the thyroid-related adverse events are unrelated to tumor PD-L1 expression.

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Identification and Management of Important Risks Due to Checkpoint Inhibitors in Mexican Cancer Patients: Real-World Evidence

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Background: The checkpoint inhibitors have changed the cancer treatment; have been used for more than 10 years. From clinical trials, checkpoint inhibitors are well tolerated when compared with traditional chemotherapy. However, is the study population representative of the real life patient to be treated? How is their safety profile in the real life population? How to anticipate, reduce, and prevent the toxicities?

Objectives: To explore the important identify risks of pembrolizumab, durvalumab and nivolumab in the Instituto Nacional de Cancerología (INCan), Mexico.

Methods: A retrospective study of adverse drug reactions reported by INCan health professionals between 2017-2019 was carried out. Identified important risks and risk minimization measures for these drugs were analyzed.

Results: From 9223 cases of adverse reactions received spontaneously between January 2017 and December 2019, only 1.6% (147 cases) were associated with checkpoint inhibitors. 59.9% cases associated to pembrolizumab (peripheral neuropathy 11.3%, arthralgia 9.1%, nausea 7.9% and fatigue 6.8%); 21.8% to durvalumab; (fatigue 43.8%, mucosal inflammation 9.4%, dysgeusia, nausea and peripheral neuropathy 6.3% each) and 18.4% to nivolumab (fatigue 18.5%, xerosis 11.1%, photosensitivity reaction, peripheral neuropathy and skin hypopigmentation 7.4% each). The most frequent reactions did not become significant risks because they do not affect the benefit-risk balance.

Conclusion: knowledge of adverse reactions and important risks of real-world patients allows us to propose institutional strategies for their management. We did not identify new important risks for pembrolizumab, durvalumab, and nivolumab. Some common reactions can be managed without medication, which has a favorable impact on the patient`s life.

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Efficiency of "Consilium" Smartphone App for monitoring Patient Reported Symptoms (PROs) in Cancer Patients depending on Medication and Outpatient Characteristics

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Background: ePROs are gaining imminent importance in cancer immunotherapeutic approaches.

Objectives: Using the "Consilium"smart phone app we examine the number, characteristics and intensity of electronically captured and patient reported outcome (ePROs) depending on treatment and outpatient characteristics.

Methods: 600 patients with cancer of breast, colon, prostate, lung, hematological malignancies are included at different stages of disease. During a three-month period from the initiation or change of therapy, patients electronically capture standardized and structured symptoms according to CTCAE, regarding treatment related side effects. Since a subset of these patients is treated with checkpoint inhibitors and combinations with monoclonal antibodies, here we provide insight in the real world patient reported outcome measures (ePROMs). In this clinical trial, we have in particular focused on the assessment and capture of high quality outcome data. For this, a statistical coefficient is implemented in the app`s system in predetermined intervals. This mechanism empowers patients and doctors to review and confirm the data entries in a specific manner.

Results: Upon all patient questionnaires the „trustworthiness" was highly associated with "Consilium" use together with high usability comfort as more than 91% of the participants surveyed confirmed the high reliability of the app, confidentiality of data and positive perception during doctor visits. Furthermore, we aim to demonstrate that patient empowerment and early intervention can avert patient damage in immunotherapeutic settings.

Conclusion: Besides current study conduct, statistical analysis and results we will also present outlook into novel trial conduct.

References: Egbring M, et al. J Med Internet Res. A Mobile App to Stabilize Daily Functional Activity of Breast Cancer Patients in Collaboration with the Physician. 2016 Sep 6;18(9):e238.

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FocuSCOPE: A Single Cell, Multi-Omics Solution to Simultaneously Analyze Tumor Variants and Microenvironment

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Recent advances of high-throughput single cell sequencing technologies have greatly improved our understanding of the complex biological systems. Heterogeneous samples such as tumor tissues commonly harbor cancer cell-specific genetic variants and gene expression profiles, both of which have been shown to be related to the mechanisms of disease development, progression, and responses to treatment. Furthermore, stromal and immune cells within tumor microenvironment interact with cancer cells to play important roles in tumor responses to systematic therapy such as immunotherapy or cell therapy. However, most current high-throughput single cell sequencing methods detect only gene expression levels or epigenetics events such as chromatin conformation. The information on important genetic variants including mutation or fusion is not captured. To better understand the mechanisms of tumor responses to systematic therapy, it is essential to decipher the connection between genotype and gene expression patterns of both tumor cells and cells in the tumor microenvironment. We developed FocuSCOPE, a high-throughput multi-omics sequencing solution that can detect both genetic variants and transcriptome from same single cells. FocuSCOPE has been used to successfully perform single cell analysis of both gene expression profiles and point mutations, fusion genes, or intracellular viral sequences from thousands of cells simultaneously, delivering comprehensive insights of tumor and immune cells in tumor microenvironment at single cell resolution.