The Azrieli
Faculty of Medicine
Bar-Ilan University

The Eli Hurvitz z"l Research Day

The Eli Hurvitz z"l Research Day
The Azrieli Faculty of Medicine
Bar-Ilan University

September 5th, 2022, ט׳ באלול תשפ״ג

The Eli Hurvitz z"l Research Day

דבר הדיקן
ליום המחקר ע"ש אלי הורביץ

שלום רב וברוכים הבאים ליום המחקר לשנה זו לפקולטה לרפואה ע"ש עזריאלי.
אנחנו מאוד שמחים על הזכות (לחזור ולהקדיש את יום המחקר לאיש החזון **אלי הורביץ ז"ל**, ושמחים על השתתפותם של בני המשפחה היקרים.

מדובר בהזדמנות חשובה להזמין מנהיגי מחקר רפואי בישראל כאורחים, ולאפשר במה לסטודנטיות וסטודנטים, חוקרות וחוקרים, צעירות וצעירים של הפקולטה להציג את תגליותיהם במחקר פורץ דרך. אנחנו גאים על האיזון החשוב בין מחקר ביו-רפואי במדעי האדם והבריאות ובהצגת מחקר מהפקולטה ומהמוסדות הרפואיים המסונפים.

ברצוני להודות במיוחד ל**פרופ' ציפי פליק-זכאי, לד"ר זאהר ארמלי, לפרופ' מאיר שמאי, לד"ר קובי ממן, לד"ר רון אורבך, לד"ר לימוד מעודד דנון ולד"ר אבי פרץ** המארגנים המדעיים של הכנס על התמסרותם לרמה המדעית הגבוהה, ששמה זרקור במיוחד על הסטודנטים המציגים גם בפוסטרים וגם בהרצאות, לצד אורחים הנכבדים: **פרופ' בת שבע כרם ופרופ' רן בליצר.**

אני ועמיתיי בהנהלת הפקולטה מודים ומברכים את כל העוסקים במלאכה, במלוא המסירות והאכפתיות.
תודה מיוחדת לסגל המנהלי המסור, האחראי על התמיכה בכל ההיבטים הלוגיסטיים להצלחת יום המחקר, כחלק מהתמיכה בחוקרים במשך כל השנה.

אני מאחל לכולנו יום מהנה ומוצלח,
פרופ' קרל סקורצקי
דיקן הפקולטה לרפואה ע"ש עזריאלי בגליל
אוניברסיטת בר אילן

The Eli Hurvitz z"l Research Day
The Azrieli Faculty of Medicine
Bar-Ilan University

September 5th, 2022, ט' באלול תשפ"ג

Program

08:30-9:00	Registration
09:00-09:20	Welcome remarks: **Prof. Karl Skorecki, Dean** **Prof. Tzipora Falik-Zaccai, Vice Dean for Medical Research**
09:20-10:05	Morning keynote: **Prof. Ran Balicer, Chief Innovation Officer at Clalit: Data-driven innovation in healthcare**
10:05-11:20	**Session 1**, Moderators: **Dr. Hanna Keren, Dr. Ohad Ronen**
10:05	Nature versus Nurture, Environmental involvement in Parkinson's disease **Linoy Mia Frankiensztajn, Ziv Medical Centre, Safed, Azrieli Faculty of Medicine, Bar-Ilan University**
10:20	Upgrade a bioactive peptide to novel customized research tool **Shulamit Fluss Ben-Uliel, Azrieli Faculty of Medicine, Bar-Ilan University**
10:35	Rationally designed attenuated HCV variants for vaccine Development **Roba Dabour, Azrieli Faculty of Medicine, Bar-Ilan University**
10:50	The involvement of iASPP in protecting the male reproductive system from deleterious inflammatory process) **Tal Talya Avitan, Azrieli Faculty of Medicine, Bar-Ilan University**
11:05	Codon optimization of Cyclin Dependent kinase-1 (CDK1) causes changes in cell cycle and apoptosis **Mahua Bhattacharya, Azrieli Faculty of Medicine, Bar-Ilan University**
11:20-11:40	**Coffee Break**
11:40-13:10	**Session 2,** Moderators: **Dr. Liron Rozenkrantz, Dr. Khalaf Kridin**
11:40	Inhibiting cohesin head domain interactions by peptide results in cohesion loss and chromosomal accumulation **Maria Elias, Azrieli Faculty of Medicine, Bar-Ilan University**
11:55	Autophagy controls mucus secretion from intestinal goblet cells by alleviating ER stress **Maria Naama, Azrieli Faculty of Medicine, Bar-Ilan University**

12:10	Combinatorial treatment of GABA, Sitagliptin, and Omeprazole leads to complete recovery in susceptible mice, suffering from Latent Autoimmune Diabetes in Adults **Wisal Sawaed, Azrieli Faculty of Medicine, Bar-Ilan University**
12:25	Changes in State Positive Body Image during Short Visits to Green Wall Environment **Reem Mohsen, Azrieli Faculty of Medicine, Bar-Ilan University**
12:40	Up regulation of the long non-coding RNA PURPL by Kaposi's sarcoma associated herpesvirus to suppress p53 activity **Samia Showgan, Azrieli Faculty of Medicine, Bar-Ilan University**
12:55	Lugol as a chemical enabler of enhanced imaging for bone-muscle tissues in fish Model **Rajashekar Donaka, Azrieli Faculty of Medicine, Bar-Ilan University**
13:10-14:40	**Lunch Break & Poster Session**
14:40-16:15	**Session 3**, Moderators: **Dr. Moran Yadid, Dr. Yuval Cohen**
14:40	A common mechanism for protein aggregation is revealed by a rare neurodegenerative disease **Ashar Masri, Azrieli Faculty of Medicine, Bar-Ilan University**
14:55	Evaluating the Effect of Blood-Derived Concentrated Growth Factors (CGF) on Osseointegration of Titanium Implants **Asaf Zigron, Galilee Medical Center, Nahariya, Azrieli Faculty of Medicine, Bar-Ilan University**
15:10	Characterization of early inflammatory events leading to provoked vulvodynia development in rats **Yaseen Awad-Igbaria, Galilee Medical Center, Nahariya, Azrieli Faculty of Medicine, Bar-Ilan University**
15:25	Examination of multiparous women's opinions regarding the postpartum medical follow-up **Lia Novick, Baruch Padeh Medical Center,Poriya, Emek Medical Center, Afula | Rappaport faculty of medicine, Azrieli Faculty of Medicine, Bar-Ilan University**
15:40	Association between vaccination status and reported incidence of post-acute COVID-19 symptoms in Israel: a cross-sectional study of patients tested between March 2020 and November 2021 **Paul Kuodi Otiku, Ziv Medical Centre, Safed, Baruch Padeh Medical Center, Poriya, Galilee Medical Center, Nahariya, Azrieli Faculty of Medicine, Bar-Ilan University**
15:55-16:40	Afternoon keynote: **Prof. Batsheva Kerem, Department of Genetics, HUJI: The cystic Fibrosis Journey: from gene cloning to drug development**
16:40	**Awards & Concluding Remarks**

Abstracts
Session 1

Nature versus Nurture Environmental involvement in Parkinson's disease

Linoy Mia Frankiensztajn[1,*], Radi Shaheen[2], Sondra Turjeman[1], Orly Avny[1], Omry Koren[1].
[1]Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel
[3] Ziv Medical Center, Safed, Israel.

Aim&Background: Parkinson's disease (PD) is the second most prevalent neurodegenerative disease in the US, affecting an estimated 1 million people or 1% of the US population over the age of 60. It is characterized by the slow and progressive degeneration of dopaminergic neurons in the substantia nigra pars compacta and is often preceded by intestinal inflammation and gastrointestinal abnormalities years before it's diagnosed. Additionally, PD is characterized by the formation of Lewy bodies, the primary structural component of which is α-synuclein (αSyn), in the brain. αSyn aggregates have also been found in the gut of PD patients, suggesting a connection to the gut microbiota in the development of PD. In this study, we investigated the involvement of the functional biomes (the microbiome and the mycobiome) in the manifestation of PD.

Methods: Fecal samples from PD patients and matched controls were collected. These samples were then given by fecal microbiota transplant to germ-free mice at the age of 6-5 weeks in order to transfer parkinsonian symptoms to them. Half of these mice were raised to the age of 18 months and behaviorally tested in order to assess their parkinsonian symptoms, and the other half underwent the same treatment at the age of 10 weeks.

Results & Conclusions: Our preliminary results demonstrate a difference between the groups in the behavioral tests. We saw a significant decline in the PD groups in the beam traversal test and in the inverted grid test, and in the open-field test we found a trend towards reduced movement and an increased stress behavior in the PD mice. Our preliminary results demonstrate a significant link between the functional biome composition and PD in our mice, and strengthen the hypothesis that PD is, at least partly shaped by environmental factors.

Upgrade a bioactive peptide to novel customized research tool

Shulamit Fluss Ben-Uliel[1], Faten Habrat[1], Moriya Slavin[2], Hadas Sibony-Benyamini[1], Nir Kalisman[2], and Nir Qvit[1]

The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Henrietta Szold St. 8, POB 1589, Safed, Israel1,Institute of Life Sciences, The Hebrew University of Jerusalem, Jerusalem, 9190401, Israel2.

Bioactive peptides (BPs) are peptides with hormonal or pharmacological properties. The source of BPs can be from natural peptides of endogenic origin, or it can be synthesized in the lab based on rational design or screening. Herein we demonstrate how a BP of interest can be modified to a highly effective research tool as well as therapeutic lead with minimal modifications that can be done in many labs or ordered in a reasonable price. As a proof of concept, we developed a linear peptide designed to bind to PTEN-induced kinase 1 (Pink1). Pink1 protein is related to mitochondrial dynamics and it was demonstrated to regulate mitochondrial homeostasis. Initially we cyclized the peptide demonstrating that the cyclic peptide has higher binding affinity and improve stability compared to the linear peptide. In addition, the cyclization will gain the peptide drug-like properties. Next we added dye to the peptide and demonstrated its colocalization with the target protein, Pink1 in H9c2 cell line. Finally, using crosslinker we identified the binding domain of the peptide and determined the interaction area, providing novel structural information about the protein. This project presents a general approach using an identified peptide and developing various research tools as well as therapeutic leads, which can be customized to other laboratories easily.

Rationally designed attenuated HCV variants for vaccine Development

Roba Dabour[1], Shaked Bergman[2], Ateret Davidovitz[1], Michal Werbner[1], Tamir Tuler[2] and Meital Gal-Tanamy[1]

1Molecular Virology Lab, Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Safed, Israel.2Department of Biomedical Engineering, Tel-Aviv University, Ramat Aviv, Israel.

Background & Aims: Hepatitis C virus (HCV) is a leading cause of liver disease and no vaccine is currently available for HCV. Live attenuated vaccines are considered very effective since they immitate the natural infection in a non pathogenic manner, but still induce efficient B and T cell anti-viral immunity. In this study we aim to rationally design and generat WT attanated HCV variants by introducing synonymous mutations to disrupt HCV mRNA structure and weaken the virus.

Methods: We utilized novel bioinformatics tools to analyze HCV genomes from databases for identifng 'silent' patterns of HCV mRNA folding, and utilized this information to design HCV variants containing synonymous mutations that affect this structure. By this approach we designed HCV mutants that varies in number and positions of inserted mutations, constructed synthetic HCV genomes containing these mutations and produced the mutant viruses. To evaluate the effect of the synonymous mutations on viral fitness, we measured the ability of the HCV mutants to replicate and spread in Huh7.5 cells. Moreover, we evaluated the mutants' pathogenesis by measuring their effect on expression of host genes related to oncogenic pathways, and examined their invasion properties in Transwell and ECM degradation assays..

Results & Conclusions: In the generated 8 infectious mutants we observed an overall reduction of HCV replication in mutants compared to WT. The spread and replication levels of the mutants varied with correlation to the level and positions of mutations inserted. The pathogenesis of the mutants varied also with correlation to the level viral replication, with minimal effect on oncogenic gene expression for the most attenuated viruses The findings of this study highlight the potential of viral attenuation generated by synonymous mutations affecting viral mRNA folding to reduce viral fitness, as a potential tool for developing rationally designed live attenuated HCV-vaccine.

The involvement of iASPP in protecting the male reproductive system from deleterious inflammatory process

Tal Talya Avitan, Daniel Baumel, Moran Titelbaum, Tzipora C. Falik-Zaccai, and Orly Avni.
Azrieli Faculty of Medicine, Bar-Ilan University

In our previous work, in collaboration with Prof. Tzipora Falik-Zaccai, Arab-Christian-infants, from four families were diagnosed with DCM associated with mild skin, teeth, and hair abnormalities. All passed away before the age of 3 years. Our results demonstrated PPP1R13L, which encoded iASPP, as the gene underlying this novel autosomal recessive cardio cutaneous syndrome in humans, and strongly suggest that the fatal DCM during infancy is a consequence of failure to regulate transcriptional pathways necessary for tuning cardiac threshold response to inflammatory stressors. In this work, using immunostaining, RNA-Seq, molecular and biochemical approaches, we studied the mechanism underlying the functional role of iASPP of another inflammation-sensitive organ, the male reproductive system in a murine model. Our results demonstrate that iASPP functions as a safeguard protecting the testis and epididymis from extended inflammatory response. Our preliminary data strongly suggest that absence of iASPP jeopardizes the immune tolerance to the sperm, and consequently leads to gradually reduced sperm count and motility, also in the heterozygous mice. We hope that understanding the mechanism protecting the male reproductive system from harmful inflammatory processes may lead to the development of new therapeutic approaches for inflammation-associated fertility failure.

Codon optimization of Cyclin Dependent kinase-1 (CDK1) causes changes in cell cycle and apoptosis

Mahua Bhattacharya[1], Gidi Baum[1], Milana Frenkel Morgenstern[1]
[1]Azrieli Faculty of Medicine, Bar Ilan University, Zefat-Israel 1311502

Codon usage plays a significant role in efficient translation of proteins which have an implication on post-transcriptional regulation to changes in translational profiles. Studies have indicated that the codon usage pattern determines the stability of proteins, which in turn affects the post-translational mechanism and functional activity of proteins. Previous studies have shown that cell-cycle regulated genes are biased towards low-affinity codons thereby, adopting non-optimal codon usage, proven in-silico. Our study aimed at understanding the effect of optimized codon on cell cycle regulatory protein. Cyclin dependent kinase 1 (CDK1) is a cell cycle regulator and checkpoint protein G2/M phase transition in mammalian cell cycle. To study the effect of codon optimization of CDK1, various stability and functional assays were performed. We compared the stability of op-CDK1 and found it to be significantly stable than non-op CDK1. In-silico and in-vitro RNA stability experiments showed that mRNA stability of op-CDK1 is higher than non-op CDK1. Using flow cytometric analyses, we observed that optimized CDK1 (op-CDK1) altered the cell cycle pattern compared to endogenous CDK1 (non-op CDK1). We also observed that op-CDK1 showed increased apoptotic activity. Therefore, our findings suggest that op-CDK1 causes perturbation in cell cycle leading to cell death.

Abstracts
Session 2

Inhibiting cohesin head domain interactions by a peptide

Maria Elias[1], Samar Ganai[2], Yana Lerner[2], Katreen Yamin[1], Chen Tor[1], Avi Matityahu[1], Nir Qvit[2*] and Itay Onn[1*]
Chromosome Instability and Dynamics Lab, Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

Abstract
Cohesin, a structural maintenance of chromosome (SMC) complex, mediates the 3-D structure of Chromatin and is involved in maintaining genome stability and function. The core of cohesin is composed of Smc1 and Smc3, elongated-shaped proteins that dimerize through globular domains at their edges. One of these interaction interfaces, called the head, contains two half-ATPase domains. ATP binding to Smc1 and Smc3 induces their dimerization and the formation of two active ATPases, while their hydrolysis results in head dissociation. This ATPase cycle is essential for driving cohesin biochemical activity. We report on the development of the first cohesin-inhibiting peptide derived from Smc1 head domain that binds Smc3. The binding affinity of the peptide to Smc3 decreases when the protein binds ATP. The peptide prevents cohesin's tethering activity in yeast cells and, interestingly, leads to the accumulation of cohesin on Chromatin. Given the sequence conservation of Smc proteins in evolution, we anticipated and provided evidence that the yeast Smc1-derived peptide may inhibit cohesin activity in human cells. This work demonstrates the power of peptides to inhibit cohesin in cells, reveals new aspects of the molecular mechanism, and discusses the potential of cohesion-inhibiting peptides as a future therapeutic approach.

Autophagy controls mucus secretion from intestinal goblet cells by alleviating ER stress

Maria Naama[1], Shahar Telpaz[1], Aya Awad[1], Shira Ben-Simon[1], Sarina Harshuk-Shabso[1], Sonia Modilevsky[1], Elad Rubin[1], Jasmin Sawaed[1], Lilach Zelik[1], Mor Zigdon[1], Nofar Fadida[1], Sondra Turjeman[1], Michal Werbner[1], Supapit Wongkuna[2,3], Bjoern O Schroeder[2,3], Abraham Nyska[4], Meital Nuriel-Ohayon[1] and Shai Bel[1]

Affiliations:
1Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.
2Department of Molecular Biology, Umeå University, Umeå, Sweden.
3Laboratory for Molecular Infection Medicine Sweden (MIMS), Umeå, Sweden
4Sackler School of Medicine, Tel Aviv University, Tel Aviv, Israel.

Colonic goblet cells are specialized epithelial cells that secrete mucus to form a barrier between the host and its microbiota, thus preventing bacterial invasion and inflammation. How goblet cells control the amount of mucus they secrete is unclear. We found that constitutive activation of autophagy in mice via Beclin 1 led to production of a thicker and less penetrable mucus layer by reducing endoplasmic reticulum (ER) stress. Accordingly, inhibiting Beclin 1-induced autophagy via Bcl-2 impaired mucus secretion. Furthermore, alleviating intestinal ER stress with a bile acid, or activating the unfolded protein response (UPR) pharmacologically via eIF2α phosphorylation, led to excessive mucus production in a microbiota-dependent manner. Over-production of mucus altered the gut microbiome, with expansion of mucus-utilizing bacteria, and protected from intestinal inflammation. Thus, ER stress is a cell-intrinsic switch that limits mucus secretion, while autophagy maintains proper mucus secretion and intestinal homeostasis by relieving ER stress.

Combinatorial treatment of GABA, Sitagliptin, and Omeprazole leads to complete recovery in susceptible mice, suffering from Latent Autoimmune Diabetes in Adults.

Wisal Sawaed[1], Sivan Eliyahu[1], Aviad Sivan[1], Nura Aburomi[1], Marina Kurtz[1], Assaf Malka[1], Shira Perez[1], and Ron Piran[1].
1 The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Aim & Background: Latent Autoimmune Diabetes in Adults (LADA) is a relatively new defined disease that has a strong autoimmune component as in T1D, and a late onset (30 years or older) as in T2D. Previously, it has been shown that each of g-Aminobutyric acid (GABA), dipeptidyl peptidase IV inhibitors (DPP-4i), or proton pump inhibitor (PPI) drugs have beneficial effects in various diabetic mouse models. Therefore, we propose that their combined administration can bring forth an additive therapeutic effect and could hold a great promising approach as a novel therapy for LADA.
Methods: We developed a novel mice model for LADA and tested this hypothesis in non-obese diabetic (NOD) mice. Diabetic mice were randomly assigned into different groups: non-treatment diabetic control, GABA plus Sitagliptin plus Omeprazole (GABA+SIT+OMP), GABA+SIT, GABA+OMP or GABA groups. The drugs were administered by oral gavage daily for 7 weeks. Functional tests (Glucose and Insulin tolerance test) and Immunofluorescence analyses were performed.
Results&Conclusion: The combined treatment administration decreased blood glucose levels. Moreover, of the mice that were given GABA+SIT+OMP 31% have completely cured from diabetes. To distinguish responders from non-responders we preformed RNA sequencing from mice's blood prior to the treatment. In the presentation we will illustrate the differences between cured and non-responsive mice.

Changes in State Positive Body Image during Short Visits to Green Wall Environment

Reem Mohsen1, Itamar Lensky2, Keren Agay-Shay1
The Health Environment Research-HER lab, Department of Population Health, Azrieli Faculty of Medicine, Bar-Ilan University1, Department of Geography and Environment, Faculty of Social Sciences, Bar-Ilan University2

Background: Mounting evidence suggests that exposure to natural environments has beneficial effects on health and mental well-being, but less is known about the effects on state positive body image (SPBI) and on the effects of green walls.
Aim of the study: The main aim of the study was to examine the effects of short visits to green walls on SPBI and to examine possible mediation by positive feeling or physiological stress.
Methods: We recruited 200 healthy adult participants and conducted a quasi-experimental study using within-subjects design. We examined the effect of short visits to green wall environment compared to gray wall on: SPBI, positive and negative emotions and physiological stress measured by heart rate variability (HRV -LF/HF). We used mixed-effects linear regression models and mediation analysis.
Results: Short visit to green wall, compared to gray wall, significantly increased the mean SPBI ($\beta = 0.17$, 95% CI: 0.09, 0.25, $p<0.01$) and the mean of positive feelings ($\beta = 2.21$, 95% CI: 1.35, 3.07, $p<0.01$). There was no significant change in negative feelings and physiological stress. Positive emotions were partially (45%) and significantly ($p<0.05$) mediated the association between short visit to green wall and SPBI. We found no mediation effect of Log LF/HF.
Conclusions: Our study suggests a significant association between short visits to outdoor green wall, compared to gray wall, and SPBI, with partial mediation through positive emotions. Further studies should examine more mechanisms influencing this relationship.

Up regulation of the long non-coding RNA PURPL by Kaposi's sarcoma associated herpesvirus to suppress p53 activity

Samia Showgan, Guy Journo, Diana Meary, Vyacheslav Gurevich and Meir Shamay
Daniella Lee Casper Laboratory in Viral Oncology, Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

Kaposi's sarcoma-associated herpesvirus (KSHV, HHV-8) is the causative agent of all forms of Kaposi's sarcoma (KS) and is tightly associated with primary effusion lymphoma (PEL) and multicentric Castleman's disease. Long non-coding RNAs (lncRNAs) are a class of regulatory RNAs with lengths exceeding 200 nucleotides. LncRNAs contribute to the pathogenesis of different malignancies by promoting cell proliferation, migration and invasion. The lncRNA PURPL (p53 up-regulated regulator of p53 levels) is able to repress p53 function, and PURPL over-expression can induce cancer cell proliferation. PURPL is regulated by p53, therefore, it serves as an auto-regulatory feedback loop for p53. Transcriptome analysis and RT-qPCR revealed up-regulation of PURPL in KSHV-infected cells, even in a p53 mutant cell line. We investigated different mechanisms KSHV uses to up-regulate PURPL. We re-analyzed ChIP-seq data of PURPL promoter from available data sources and found that p53 and p73 share a binding region in the PURPL promoter. Reporter assays revealed that expression of p73α and β leads to up-regulation of PURPL in HCT116 p53 knock-out (KO) cells. Furthermore, a mutation at the p53 consensus binding site abrogated all the induction of PURPL promoter by KSHV. Transfection of PEL cells with anti-sense oligos targeting PURPL, reduced cell proliferation. Western Blot of p73 in KSHV infected (BJAB219) and non-infected lymphoma (BJAB) cells revealed that KSHV infection up-regulates p73 protein levels. In summary, we revealed that p73 is a regulator of PURPL expression. The lncRNA PURPL is up-regulated by KSHV and is essential for PEL cell growth.

Lugol as a chemical enabler of enhanced imaging for bone-muscle tissues in fish model

Rajashekar Donaka[1], Iryna Khrystoforova[1], Chen Shochat[1], and David Karasik[1]
1The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Background: Flawless functioning of the musculoskeletal system is essential for healthy life and successful aging. Impairments in genetic and metabolic processes can predispose to aging diseases like osteoporosis and sarcopenia. The knockout (KO) of srebf1 gene in zebrafish demonstrated a decreased BMD and altered lipids in their muscles. We, therefore, utilized srebf1-/- fish as a model to study osteoporosis and sarcopenia.
Purpose: To explore the consequences of a loss of muscle density in srebf1-/- fish, and to test the effect of Lugol staining on the vertebral BMD.
Methods: Adult srebf1-/- and srebf1+/+ (WT) siblings were overfed for 45 days and provided a regular diet (control). Lugol contrasting solution was applied to stain soft tissue. BMD and soft tissue attenuations were measured using micro-CT SKYSCAN 1172 (Bruker) and muscle specific lipids were marked by Oil Red O in the same fish.
Results: Both srebf1+/+ and srebf1-/- overfed fish gained body weight and BMI significantly (n= 13-15 fish per group, $P<0.015$ and $P<0.022$,) compared to WT and KO controls. Lugol staining did not alter vertebral BMD by genotype/diet conditions. Further, fast and slow muscle fibers density significantly ($P<0.0087$, $P<0.000001$) reduced in overfed of srebf1-/- fish compared to srebf1-/- controls, with a concomitant significant increase of slow muscle lipid content.
Conclusion: This study confirmed that Lugol staining allows us to visualize 1) fatty infiltration of muscle tissue; 2) But it does not alter BMD of the adult zebrafish. The study provides a unique framework to analyze simultaneously body composition and reveal manifestations of osteoporosis and sarcopenia using zebrafish.

Abstracts
Session 3

A common mechanism for protein aggregation is revealed by a rare neurodegenerative disease

Ashar Masri, Anwar Dakwar, Gilat Shimon, Mohammad Abo-Raya, Tal Benjamin, Ronit Ilouz
Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.

Dysregulation of cAMP signaling contributes to the etiology of several brain degenerative diseases. A missense mutation in PKA RIb regulatory subunit, the least studied isoform, was found in patients diagnosed with a rare neurodegenerative disease. The gap in knowledge regarding this gene and the devastating outcomes seen in individuals with motor deficits are two critically important problems that we are currently researching. Our recent structural model led us to hypothesize that an amino acid substitution L50R may result in preventing dimer formation. Biochemical studies as well as cell-based high-resolution image analysis suggest that this RIb missense mutation not only prevents RIb homodimerization but also eliminates the binding site that is created by dimer formation for A Kinase Anchoring Proteins (AKAPs) binding. Consequently, PKA holoenzyme localization is affected as evidenced by accumulation of RIb into neuronal inclusions in human brain patients. A quantitative multiplex proteomics revealed that the phospho-signaling cascade is disrupted by the L50R mutation in postmortem brains. This study emphasizes the importance of precisely controlled PKA isoform subcellular localization and demonstrates how a mutation in PKA regulatory subunit drives aberrant cAMP signaling and neurodegeneration. This study provides insights into the molecular and cellular mechanisms of other neurodegenerative diseases where PKA function is dysregulated.

Characterization of early inflammatory events leading to provoked vulvodynia development in rats

Yaseen Awad-Igbaria,[1,2] Eilam Palzur,[2] Jacob Bornstein,[1,2]
Faculty of Medicine in the Galilee, Bar-Ilan University; Safed, Israel[1], The Research Institute of Galilee Medical Center; Nahariya, Israel[2].

Objective: To investigate the development of persistent vulvar mechanical and thermal hypersensitivity in a rat model of vulvodynia developed after three rounds of zymosan challenges.
Methods: Mechanical and thermal vulvar sensitivity and anxiety were measured in rats for 160 days, following three zymosan vestibular challenges that led to inflammatory response. At the early stage of inflammation, Nerve growth factor (NGF) and glutamate were detected. The expression of pain channels (TRPV1, TRPA1), the presence of mast cells (MCs) and hyperinnervation of vulva neurons were assessed by immunohistochemical staining after 20 and 160 days of the 3rd zymosan challenge.
Results: Zymosan-challenged rats developed significant mechanical and thermal sensitivity that persisted for over 160 days after the 3rd zymosan challenge. During inflammation, we depicted localized early increases in NGF and glutamate concentration, as well as a robust increase in MCs degranulation, sensory hyperinnervation and neuromodulation that include an increase in the expression of TRPV1 and TRPA1. 160 days, after the edema and the inflammation subsided, the challenged vulvar vestibule was still characterized by MCs accumulation, sensory hyperinnervation, and neuromodulation. Moreover, we demonstrated a decrease in pain threshold manifesting as allodynia, leading to high anxiety levels.
Conclusion: The present findings suggest that the neuronal alterations, observed during the early inflammatory phase and long after the inflammation subsided, led to allodynia and hyperalgesia. It was mediated by MCs activation and inflammatory agents such as NGF and glutamate. We further propose that discontinuation of the response of MCs with repeated inflammation may prevent the development of PV.

Evaluating the Effect of Blood-Derived Concentrated Growth Factors (CGF) on Osseointegration of Titanium Implants

Asaf Zigron [1,2], Daniel Oren[1], Fares Kablan[1], Samer Srouji[1,2]
[1] Galilee College of Dental Sciences, Oral and Maxillofacial Surgery Department, Galilee Medical Center, Nahariya, Israel
[2] The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

Concentrated growth factors (CGF) are a platelet concentrate obtained by centrifugation of whole blood samples. The CGF produced from this process is composed of a fibrin mesh, platelets, leukocytes, growth factors and stem cells. Bio-coating of dental implants has been proposed to accelerate bone formation at the host-implant interface. Biological coating, such as the coating of titanium implants with different growth factors, showed favorable effects on cells recruited to the implant bio-environment. Here, we examined the effect of CGF bio-coating on implant osseintegration in a big animal model.
Venous blood of 6 male beagle dog was drawn into costume-made implant centrifugation tubes, leading to production of CGF-coated implants. CGF-coated and control uncoated dental implants were implanted in mandibles of animals under general anesthesia. Six-weeks post-op, A micro-computed tomography examination was performed on bone samples removed from the mandibles. The bone implant contact volume (BIC), defined as the direct bone-to-implant interface, was evaluated by both histological evaluation and Micro-CT. Both histological and volumetric analysis indicated denser and more mineralized bone formed in treatment group compared to the control group. These results serve as a clear indication that the use of CGF-coated implants shows superiority in osseintegration non-coated implants.

Examination of multiparous women's opinions regarding the postpartum medical follow-up

Lia Novik[1,2], Tamar shyldkrot[1,2], Zohar Nachum[3,4], Enav Yefet[1,2]
Department of Obstetrics and Gynecology, Baruch Padeh Medical Center, Poriya, affiliated with Azrieli Faculty of Medicine, Bar Ilan University, Israel.1 Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.2 Department of Obstetrics and Gynecology, Emek Medical Center, Afula.3 Rappaport faculty of medicine, Haifa.4

Aim & Background: The postpartum period is the 6 weeks following delivery, in which women may encounter a wide range of health problems. In Israel, it is customary to visit the OB/GYN 4-6 weeks postpartum to assess physical, social, mood and emotional well-being, as well as addressing breastfeeding and family planning. However, the rate of women who perform the postpartum recommendations may be as low as 40%.
In this study, we assessed ways of improving women compliance for postpartum recommendations.
Methods: A cross-sectional study of women who delivered at least once was conducted. We used a 50 items questionnaire, which was designed to evaluate various issues that may have reduced compliance for the previous postpartum checkup. Suggested possible solutions were evaluated. Each item was rated from 1 (not agree at all) to 5 (totally agree). Significant barriers/accelerators were considered when above 50% of participants agreed on (rated 4 or 5).
Results: Three hundred women participated in the study. Women considered the OB/GYN postpartum check-up to be important for physical and mental health, family planning, and reviewing discharge letter recommendations. Major accelerators would be a simultaneous check-up of both mother and newborn and reminders regarding the visit date. Weight measurement, and blood and urine tests during the check-up were also favorable.
Conclusion: Women considered the OB/GYN postpartum check-up to be important. Simultaneous check-up of both mother and newborn, pre-visit reminder notification and laboratory tests may increase compliance for the postpartum OB/GYN check-up.

Association between vaccination status and reported incidence of post-acute COVID-19 symptoms in Israel: a cross-sectional study of patients tested between March 2020 and November 2021

Paul Kuodi[1], Yanay Gorelik[1], Hiba Zayyad[1,3], Ofir Wertheim[3], Karine Beiruti Wiegler[2], Kamal Abu Jabal[1,2], Amiel A. Dror[1,4], Saleh Nazzal[3], Daniel Glikman[1,3], Michael Edelstein[1,2]

Authors' affiliations: Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel
Ziv Medical Centre, Safed, Israel | Baruch Padeh Medical Centre, Poriya, Israel
Galilee Medical Centre, Nahariyah, Israel

Background: Long coronavirus disease 2019 (Long COVID) is a post-severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) infection syndrome characterised by not recovering for several months following the acute episode. The effectiveness of COVID-19 vaccines against long-term COVID-19 symptoms is not well understood. We determined whether vaccination was associated with the incidence of reporting long-term symptoms post-SARS-CoV-2 infection.

Methods: We invited individuals who were PCR-tested for SARS-CoV-2 infection at participating hospitals between March 2020-November 2021 to fill an online questionnaire including demographics, acute episode details and information about symptoms they were currently experiencing. Using binomial regression, we compared vaccinated, unvaccinated and uninfected individuals in terms of self-reported symptoms post-acute infection

Results: 951 infected and 2447 uninfected individuals participated. Of the infected, 637(67%) were vaccinated. The most commonly reported symptoms were: fatigue (22%), headache (20%), weakness (13%), and persistent muscle pain (10%). After adjusting for follow-up time and baseline symptoms, those who received two vaccine doses were less likely than unvaccinated individuals to report any of these symptoms by 64%, 54%, 57%, and 68% respectively, (Risk ratios 0.36, 0.46, 0.43, 0.32, $p<0.04$ in the listed sequence). Those who received two vaccine doses were no more likely than individuals reporting no previous SARS-CoV-2 infection to report any of these symptoms.

Conclusion: Vaccination 2+ doses of COVID-19 vaccine was associated with a substantial decrease in reporting the most common post-acute COVID-19 symptoms. Our results suggest that, in addition to reducing the risk of acute illness, COVID-19 vaccination may have a protective effect against long COVID.

Posters

Session 4

1

Abstract: Before the era of vaccination against the bacterium *Streptococcus pneumoniae*, it was one of the most common contaminants in children, and isolated from many blood cultures. Since the introduction of the vaccine, there has been a change in the distribution of pathogens among children in general, and the incidence rate of *Streptococcus pneumoniae* has decreased.

Goals: Comparing the incidence of latent bacteremia in children, ranging from three months of age to three years, between 2006-2009 (before the vaccination era) compared to 2014-2017 (after the vaccination was introduced), at Padeh Poriya Hospital.
Methods: The study is retrospective, in which all blood cultures taken to children between the ages of three months and three years in 2006-2009 will be reviewed, as well as in 2014-2017, addressing the types and sensitivities of the different bacteria, as well as the clinical and laboratory data.

Results: The total number of cases which included a positive blood culture before the vaccine era was 40 (1%), compared to 29 (0.7%) after the vaccine was introduced. In 14% of children with bacteremia, three criteria were found: Leukocytosis over 15000 cells per microliter, neutrophilia over 70%, and fever ≥ C°39. In addition, *Streptococcus pneumoniae* susceptibility to 6 antibiotics was tested, during the pre- and post-vaccination period, and for the first generation of Cephalosporins – Cefazolin, resistance increased from zero to 27%, before and after vaccination, accordingly.
Conclusion: Detecting and monitoring the bacterial growth of *Streptococcus pneumoniae* in blood cultures of children, in addition to characterizing the various strains of the same bacterium, can provide information concerning the ability of the vaccine to significantly lower infections, and emphasize the importance of taking routine blood cultures for children vaccinated against *Streptococcus pneumoniae* who visit the emergency room with high fever.

Cracking the genetic code of somatic hypermutation

2

Sawsan abo ghali[1], Jukka Alinikula[2], Yaakov Maman[1]
[1]Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel
[2]Institute of Biomedicine, University of Turku, Turku, Finland

Abstract

In mature B cells, the affinity of antibodies to the antigen is improved by introducing point mutations in the Ig genes. This process, termed somatic hypermutation (SHM) is initiated by the activation induced deaminase (AID). AID-induced SHM is linked to transcription, but the preferable targeting to the Ig genes over non-Ig genes in not well understood. Recent studies demonstrated that SHM targeting to Ig loci is dependent on cis-acting elements reside at the Ig enhancers, collectively termed DIVAC (Diversification Activators). These studies highlighted some TFBS (Transcription Factor Binding Sites) that are important for DIVAC activity, but none of them is unique to the Ig loci enhancers. Nevertheless, to date, no non-Ig enhancer has shown SHM activity similar to that of an Ig enhancer. We hypothesize that SHM targeting is enabled by a unique regulatory "language" that, like any other language, contains a highly defined combination of "letters" (individual TFBS), "words" (specific combinations of adjacent binding sites), and "sentences" (DIVAC). To unveil this language, we have developed "DIVAC library" a high-throughput approach that tests millions of synthetic regulatory elements for their capacity to induce SHM. Analyzing the data obtained from this approach, we have identified linguistic determinant of SHM targeting.

3 Maternal and Perinatal Outcome in Vaginal Delivery vs. Cesarean Delivery of Twins with Cephalic/Cephalic Presentation

Amit Hart[1], Jacob Bornstein[1], Lior Lowenstein[1,2], Inshirah Sgayer[*1,2], Maya Wolf[*1,2]
*equal contribution
[1] Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel
[2] Department of Obstetrics and Gynecology, Galilee Medical Center, Nahariya, Israel

Objectives Vaginal delivery (VD) for the breech fetuses has declined in recent years. In twins with cephalic-cephalic presentation, a change in presentation for the second twin might occur after the delivery of first twin. In this scenareo, an emergency cesarean section is performed to deliver the second twin due to a lack in obstetricians who are skilled to manage breech vaginal birth.
Our goal was to compare maternal and perinatal outcomes of cephalic-cephalic twins who undergo a trial of VD versus elective cesarean section (ECS).
Methods Retrospective study between 2015-2019 in which 115 cases were included. The VD group consisted of 57 patients, while the ECS group consisted of 58 patients. Data concerning obstetrical and neonatal complications were compared between the two groups.
Results The rate of succesful VD was 73.68% in the VD group, while 10.5% had VD for first twin and cesarean for the second twin (combined delivery). 15.8% of VD group had emergent cesarean. 8.8% of the second fetuses have changed their presentation from cephalic to breech/transverse after delivery of the first twin. Higher rates of pastpartum hemorrhage was found in the VD compared with ECS group (24.6% versus 6.9%, p=0.01). Maternal hospital stay was longer in the ECS group (5 versus 4 days, p<0.001). After adjustment for gestational age, neonatal adverse outcome of the two groups was similar.
Conclusions Despite the fact that second twin might change his presentation after delivery of first twin, we found a high success rate for cephalic-cephalic twins attemping VD, with no increase in perinatal morbidity.

The rate of incidental carcinoma in thyroid as detected on positron emission tomography-computed tomography (PET-CT) scan among breast cancer patients at Ziv medical center

4

Shlomo Merchavy M.D[a b], Moran Barazani [b], Adi Sharabi-Nov MA, MPH[b c d], Majd Asakly MD[a b].

[a]Otolaryngology, Head & Neck Surgery Unit, Ziv Medical Center, Safed, Israel.
[b]Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel
[c]Research Wing, Ziv Medical Center, Safed, Israel.
[d]Tel-Hai Academic College, Tel-Hai, Israel.

Objective: To study the incidental thyroid malignancy detected by PETCT scans in Ziv-treated breast cancer patients.
Background: Because of the increased use of (18)F-fluorodeoxyglucose positron emission tomography ((18)F-FDG PET) in breast cancer patients, an increasing number of incidentalomas in the thyroid gland have been identified. Up to 35% of thyroid focal incidentalomas are malignant. Early detection of thyroid cancer allows for less invasive surgical treatment and avoids complicated treatment of advanced thyroid cancer.
Methods: Medical records of 166 breast cancer patients who underwent FDG PET-CT imaging in the nuclear medicine department at ZIV medical center from January 2018 to January 2019 and who were treated in the oncology department at ZIV medical center were reviewed for thyroid incindentiloma, maximal standard uptake value (SUVmax), age, and ethnicity were recorded and compared between patients with thyroid malignant incidentaloma and other breast cancer patients.
Results: Eight patients out of 166 had a focal incidental finding in the thyroid gland. Three patients had significantly higher SUVmax values than the other five. One of these patients had a biopsy that revealed papillary thyroid carcinoma; the SUVmax in this case was 17, and the other two patients were referred for FNA of the incidentaloma. The SUVmax of the remaining five patients is less than 10. Malignant thyroid incidental tumors occurred at a rate of 12.5%.
Conclusion: Thyroid gland incidentalomas with a high focal metabolic rate may be malignant. In these patients, further investigation, including ultrasonography-guided fine needle aspiration, is required.

5 The impact of morbid obesity of late GDM diagnosis and related obstetrical and neonatal outcomes.

Raneen Abu Shqara[1, 2], Shany Or[2], Yara Nakhleh[1, 2], Yifat Wiener[3], Lior Lowenstein[1, 2], Maya Frank Wolf[1, 2]

[1]Department of Obstetrics & Gynecology, Galilee Medical Center, Nahariya, Israel [2]Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel

[3]Department of Obstetrics and Gynecology, The Yitzhak Shamir Medical Center, Zerifin, Israel, and Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel.

Objective: To investigate associations of maternal obesity with late GDM diagnosis (>34 weeks) and related obstetrical and neonatal outcomes.

Study Design: This was a retrospective cohort study of 238 women who underwent late repeat oral glucose tolerance tests (OGTT) (>34 weeks). Obstetrical and perinatal outcomes were stratified by GDM and morbid obesity (BMI≥35 kg/m^2) status. Obstetrical complications included pre-eclampsia, induction of labor, cesarean delivery, shoulder dystocia, and third or fourth-degree perineal tear. Neonatal outcomes included Apgar score, arterial cord pH, neonatal hypoglycemia, jaundice, the need for phototherapy, and polycythemia.

Results: Following repeat GDM screening with OGTT (>34 weeks), GDM rate in the total sample was 22.2%. Women with late GDM had a higher risk for macrosomia (p=0.002), large-for-gestational-age fetus (p<0.001), induction of labor (p=0.02); and neonatal hypoglycemia (p=0.001), jaundice (p=0.04), and need for phototherapy (0.026). The higher risk for late GDM diagnosis (33%) was in women with morbid obesity (BMI≥35 kg/m^2). Women with morbid obesity had significantly a higher rate of obstetrical complications including macrosomia (64%) and induction of labor (60%).

Conclusion: Adverse pregnancy outcomes are related to morbid obesity in women with late GDM; this population requires special surveillance. Repeat GDM screening beyond 34 weeks should be considered in this subpopulation.

Revealing the secrets of the Eco1 zinc finger

6

Hanan Zbedat[1], Avi Matityahu[1] and Itay Onn[1]
Chromosome Instability and Dynamics Lab, Azrieli Faculty of Medicine, Bar Ilan University , Safed

The cohesin complex is best known to mediate two functions. It ensures the equal segregation of chromosomes during mitosis, and involved in the repair fedility of double strand breaks in the DNA. The acetyltransferase Eco1 is a key regulator of cohesin in both processes. During the cell cycle, Eco1 acetylates the Smc3 cohesin subunit to induce its chromatin tethering activity. In response to DNA double strand break, Eco1 acetylates another cohesin subunit, Mcd1 which leads to cohesin loading next to the break site. However, the molecuar basis of these different substrate specificity of Eco1 is elusive. Eco1 contains a single C2H2 zinc finge domain. While zinc fingers are best known for their DNA binding activity, we hypothesized that the Eco1 zinc-finger is used to interact with its substrates and determined its specificity. We performed an alanine scan in the zinc-finger domain and examined the effect of each mutation on cells viability. We identified residues that are critical for its cohesin regulation activity. Interestingly we pinpointed residues that are specific for Eco1 activitiy in either sister chromatid cohesion and DNA repair. Based on the scan results we were able to develop a structural model for Eco1-Smc3 interactions. Our work assign a molecular function of Eco1 zinc finger and its non-canonical function in mediating protein-protein interaction. Furthermore, contributes to the understanding how Eco1 and cohesin cooperates in safegurding the stability of the genome.

7 Hypothyroidism following Hemithyroidectomy

Alaa Safia M.Da , Adi Sharabi-Nov MA, MPHb, Shlomo Merchavy MDa .
aOtolaryngology, Head & Neck Surgery Unit, Ziv Medical Center, Safed, Israel.
b Research Wing, Ziv Medical Center, Safed, Israel.

Objective: The aim of this retrospective study was to determine the overall risk of hypothyroidism, both clinical and subclinical, after hemithyroidectomy.

Background: The reported risk of hypothyroidism after hemithyroidectomy shows considerable heterogeneity in literature.

Methods: This retrospective study identified 200 euthyroid patients who underwent hemithyroidectomy between the years 2019 – 2021 in ENT clinics, Ziv Medical Center, Safed and the,- Western Galilee Medical Center, Nahariya in Israel
Laboratory results of TSH , T4 and T3 hormone that was taken before the hemithyroidectomy have been checked and compared with levels of these hormone postoperatively during the first month, three month, six month and last visit after surgery .The incidence of hypothyroidism was analyzed. The independent effect of age, gender, race, diabetes mellitus , obesity and preoperative TSH level on the development of hypothyroidism was evaluated.

Results: Overall Postoperative hypothyroidism developed in 66 patients (33%) out of 200 patients, of them 91% were with subclinical hypothyroidism according to only high level of TSH postoperative and normal level of free T4 and T3 , only 9 % were diagnosed with clinical hypothyroidism according to an increase TSH and low free T4 level after surgery. Those patients main complain were fatigue and increasing weight . and required thyroid hormone replacement, within the first 6 months postoperatively
Conclusions: About one third of euthyroid patients who undergo hemithyroidectomy develop hypothyroidism. It is usually asymptomatic, and can be diagnosed early after surgery. The most significant predictor is the preoperative TSH level, and the patients aged over 46 years .

Molecular, cellular and functional characterization of disease-associated mutations in PKA.

8

Mohammed Aboraya[1], Ronit Ilouz[1].
[1] The Azrieli Faculty of Medicine, Bar-Ilan University.

Abstract:
cAMP dependent protein kinase A (PKA) is a tetrameric enzyme that consists of a regulatory subunit dimer bound to two catalytic subunits. Each of the four isoforms of the regulatory subunits (RIα, RIβ, RIIα and RIIβ) are expressed in different cells and tissues. RIβ is mostly expressed in the brain. The RIβ (L50R) mutation is associated with a late-onset neurodegenerative disorder characterized by frontotemporal dementia and parkinsonism. To expand the molecular analysis of this mutation and to identify a common mechanism by which a disease-associated mutation is linked to the dimerization and docking (D/D) domain of PKA regulatory subunits we search for additional mutation in several databases and chose to work on 4 additional mutations within RIβ regulatory subunit: I27A, I40V, A67V and R68Q in parallel to L50R. Here, we test the effect of each mutation on the ability of the regulatory subunit to interact with its catalytic subunit and to create the holoenzyme complex. Surprisingly, only the L50R variant brake RIβ dimer and a new phenotype was observed with the A67V variant. Moreover, we showed the effect of different mutations on the localization of PKA in two cell lines. Co-transfection of the L50R variant with A Kinase Anchoring Proteins-1C (AKAP-1C) didn't prevent aggregation and the I27A variant can't bind to AKAP-1C. Given the fact that PKA is the prototype of the human kinome, this results may imply on other kinases where a point mutation in their regulatory subunit disrupt kinase space-restricted, localization and action.

9 The development of spesific peptides to inhibit Parkin's interaction with cytosolic P53 and the investigation of the effects of this inhibition in cardiac cells.

Ghunwa Sakran[1], Samar Gani[1], Nir Qvit[1]
The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Henrietta Szold St. 8, POB 1589, Safed, Israel.

E3 ubiquitin-ligase Parkin is essential for the autophagic removal of the mitochondria. Parkin is recruited to defective mitochondria which results in the ubiquitination of the outer mitochondrial proteins. As a result, the mitochondria are removed. P53 protein prevents Parkin translocation from the cytosol to the mitochondria, which reduces the efficiency of the autophagic clearance of damaged mitochondria.
We aim to design specific peptides based on the interaction region between Parkin and P53, and investigate the effect of the peptides using a rat cardiac myoblast cells.
We developed peptide using the L-align tool based on a similar sequence between Parkin and P53. We used solid-phase peptide synthesis (SPPS) to synthesize the peptides, and reverse-phase high-pressure liquid chromatography to determine the purity (RP-HPLC). Further, Matrix-assisted laser desorption ionization (MALDI) Mass spectrometry measurements was performed to establish the the molecular weight. We studied the impact of the designed peptides on a rat cardiac myoblast cell line by XTT assay to quantify the cells' relative metabolic activity, which is inversely linked with cell viability in ischemia-like circumstances by cobalt.
We successfully produce CVP-272a peptide with 100% purity; the desired peptide mass spectra analyzed by MALDI was 2551 m/z, which is equivalent with the molecular weight of the CVP-272a. By using the XTT assay, we were able to demonstrate that CVP-272a had a protective effect. And the treated cells' viability was 77.85 %, compared to 60 % for the untreated cells. These results indicate that CVP-272a can serve as a candidate for reviving mitophagy and treating the elimination of damaged mitochondria.

Computational analysis of differential splicing and transcript alternations in severe COVID-19 infection

10

Sunanda Biswas Mukherjee, Milana Frenkel-Morgenstern
The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Henrietta Szold St. 8, POB 1589, Safed, Israel.

Abstract
Viral infections could modulate the widespread alternations of cellular splicing, which favor the viruses replicating within the host cells by overcoming host immune responses. How the SARS-CoV-2 inducesthe host cell differential splicing, and the landscape of transcript alternation in severe COVID-19 infection remains elusive. Understanding the differential splicing and transcript alternations in severe COVID-19 infection could improve the molecular insights into the SARS-CoV-2 pathogenesis. In this study, we analyzed the publicly available blood transcriptome data of severe COVID-19 patients, recovered COVID-19 patients at 12-, 16-, and 24-weeks post-infection, and healthy controls. We identified 1385 genes that undergo significant transcript isoform switching events. Among these, 414 genes were found to be differentially expressed in gene expression analysis, while 971 genes are not undergoing any changes in expression levels, but they altered at the transcripts level. Altered transcripts show the significant loss of the open reading frame (ORF), functional domains, and changed the coding to the non-coding transcript, impacting normal cellular functions. We identified the expression of several novel recurrent chimeric transcripts in the samples from severe COVID-19 patients. Further, analysis of the isoform switching in recovered COVID-19 patients highlights that there is no significantisoform switching in 16 -, and 24-weeks post-infection, and the expressed chimeric transcripts are also less. This finding emphasizes that SARS-CoV-2 severe infection could induce widespread splicing in the host cells, which could help the viruses alter the host immune responses that might facilitate the viruses to replicate within the host and translate viral proteins efficiently.

11 Paternal microbiota composition has effect on offspring in F1 and F2 generations

Alona Riumin[1], Sondra Turjeman[1] and Omry Koren[1]
[1] Azrieli Faculty of Medicine, Bar-Ilan University

Introduction: Babies are colonized with bacteria at birth hence multiple studies have focused on how maternal exposure to environmental factors influences the establishment of the microbiome. However, paternal exposure to adverse environmental conditions can affect offspring phenotypes in the F1 and F2 generations. Here we examine how paternal microbiota composition, altered by environmental factors, influences metabolic offspring phenotypes.
Methods: Feces from mice fed either high- or low-fat diet (HFD and LFD, environmental stress) were collected and transplanted into adult male mice (FMT). These mice were bred and followed for two generations (F1 and F2). Weight, fat mass, and intraperitoneal glucose tolerance were measured at various time points, and mice were sacrificed for metabolic tests. Feces were collected to characterize gut microbiota and SCFA profiles.
Results: The body weight of the HFD-F1 mice was significantly higher than that of the LFD-F1 mice at 4-6 weeks. At 6 weeks, glucose and fat percent were significantly higher in HFD-F1 mice. Microbiota was altered and SCFA levels were higher in LFD-F1 mice at 5 and 7 weeks. In the F2 generation, there were no differences in metabolic parameters but differences in SCFA levels and microbiota composition were found.
Conclusions: Paternal bacterial composition widely affected offspring in the F1 generation, while in F2, the effect was only on SCFAs and the microbiota. The underlying mechanism remains unknown; future studies should examine the effect of metabolites like SCFAs on paternal epigenetic inheritance.

Immunogenicity after COVID-19 boosting or infection in healthcare workers: Israel, August 2021-May 2022

12

K. Abu Jabal[1, 2], N. Beer[1], H. Ben-Amram[1], K. Wiegler Beiruti[1], Y. Bathish[1], C. Sussan[1], H. Asulin[1], S. Zarka[1, 2], M. Edelstein[1, 2]
Ziv Medical Center, Safed, Israel[1], Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel[2]

Background : Israeli healthcare workers (HCW) are eligible for 4 doses of COVID-19 vaccines. We measured anti SARSCoV-2 spike IgG levels among Ziv Medical Center HCWs who received three or four doses of vaccine (mainly BNT162b2) or were infected, to describe and compare vaccine and virus-induced immunogenicity.

Methods : We regularly measured HCW IgG levels using a quantitative serological assay and reported geometric mean concentrations with 95% confidence intervals (95%CI) according to infection status and number of vaccine doses received. We compared vaccine and virus-induced IgG levels using Kruskal-Wallis tests.

Results : Of 904 HCWs in our cohort, 684 (76%) received three and 112 (12%) received four doses. Uninfected individuals IgG levels increased from 117 (95%CI 107-128) pre-dose 3 to 2746 AU/ml (95%CI 2477-3044) after 1-2 months and reduced to 1174 AU/ml (95%CI 815-1694) after 6-8 months. IgG levels were 710 AU/ml (95%CI 460-1096) pre-dose 4 , 2083 (95%CI 1049-3080) after 1-2 months and 1099 AU/ml (95%CI 389-3106) after 2-4 months. Compared with three-doses recipients, those infected after the second dose had higher IgG levels at the same time point (4678 vs 2746 AU/ml, P<0.003). Infection post dose 3 and a fourth dose led to similar IgG levels (2601 vs 2083 AU/ml, p=0.5).

Conclusions: Receiving a third dose increased 23-fold IgG levels, which remained 10 times higher than pre-dose 3 at 6-8 months. Increase in IgG levels following dose 4 was both smaller (3-fold) and more transient. Infection induced immunity led to similar or higher and longer lasting IgG levels.

13 "Development of a selective peptide modulator of protein-protein interactions in mitochondrial dynamics and its biological implications in cardiovascular diseases "

Faten Habrat-Zoab, Mentor: Dr.Nir Qvit
The Azrieli Faculty of Medicine, Bar-Ilan University

"Mitochondrial dynamics" are the processes that determine mitochondrial morphology and quality through fusion, fission and mitophagy which is important for cells survival and functions. These mechanisms are regulated by mitochondrial proteins, in this project we focus on two of them, Mitofusion2 (Mfn2) and E3 ubiquitin-protein ligase (MARCH5) which involved in mitochondrial fusion. Mfn2 is a dynamin-like GTPase protein that plays a central role in regulating fusion process.

MARCH5 is a ubiquitin ligase of the mitochondrial outer membrane which plays a role in the control of mitochondrial dynamics by regulating Mfn2.

Hypoxic conditions effect the interaction between Mfn2/MARCH5 leading to imbalance of mitochondrial dynamics, these Changes have been associated with the development of various cardiovascular diseases (CVDs) that are one of the leading causes of global mortality worldwide.

Herein, in this project we hypothesis that there is an interaction between Mfn2/MARCH5 proteins, and we can regulate it by developing a specific peptide modulators using rational design. Peptides are vital biological mediators that demonstrate remarkable potency, selectivity, and low toxicity.

We successfully purified component Mfn2 and MARCH5 proteins and proved its interaction by using kinetic method. For the future we are planning to develop peptide modulators that interfere specifically with Mfn2/MARCH5 interaction to regulate the fusion process.

Further, we will investigate the effect of these modulators on chemical hypoxia in cardiac cells and in animal models. We speculate that inhibiting Mfn2/MARCH5 under hypoxic cardiac cells increases cell survival. These studies may be used for development of novel therapeutic agents for treating CVDs.

The association between cell motility, auto-aggregation and biofilm intensity in Clostridiodes difficile

14

Layan Abu Rahmoun[1], Maya Azrad[2], Avi Peretz[1,2*]
[1]Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel
[2]Clinical Microbiology Laboratory, Baruch Padeh Medical Center, Poriya, Israel

Aims & background: *Clostridiodes difficile* is the primary pathogen responsible for nosocomial diarrhea, whose recurrence tightly correlates with biofilm formation. Bacterial motility and auto-aggregation play a role in the initial phase of biofilm development. Auto-aggregation is modulated by genes such as *SpoOA, fliC*, and several surface proteins, including *pilA, Cwp84* and *LuxS*. This study aimed to investigate whether motility and auto-aggregation levels as well as gene expression levels of *SpoOA, fliC, Cwp84 LuxS* and *pilA* affect biofilm production intensity.

Methods: Bacterial motility of 123 strains with different biofilm formation capacities was determined by stabbing a needle with one bacterial colony ~ 0.5 inch into a 0.175% agar tube. After 7 days of incubation at 37°C, the tube was visually examined for signs of bacteria diffusion from the stabbed line into the tube. Auto-aggregation percentage of these strains was evaluated using a formula comparing the optical density of the post- and pre-vortexing of overnight strains' broth culture. The expression levels of the genes were quantitated using real-time PCR.

Results: Strong biofilm producers had a higher percentage of auto-aggregation (57.9%) than non-producers (30.5%), weak producers (40.3%) and moderate producers (48.2%) (p value <0.0001). A significant correlation was observed between motility and biofilm intensity (p value=0.0361). There was no association between expression of the five genes and biofilm intensity.
In conclusion, motility and auto-aggregation levels of *C. difficile* affect biofilm formation.

15 Characterization of the Histological Response to the Butterfly Prostatic Retraction Device in Patients with Benign Prostatic Hyperplasia

Muhamad Abu Ahmed[1], Sze Christina[2], Ali Safadi[1], Wasim Abu Nasra[1], Bilal Chugtai[2], Ran Katz[1]
Ziv Medical Center, Safed, Israel
NewYork-Presbyterian Hospital/Weill Cornell Medical Center NY, USA

Introduction: The Butterfly Prostatic Retraction Device ("Butterfly") is a permanent nitinol implant for BPH, deployed transurethrally using cystoscopy to the prostatic urethra under local anesthesia and intended to retract the lateral lobes of the enlarged prostate. Human histology was found to provide the most relevant data to evaluate the clinical version of this device. This study examines the chronic response of prostate tissue to the Butterfly in histological specimens from patients in the Butterfly pilot clinical study.

Methods: Retrospective qualitative and semi-quantitative review of histological specimens of seven (7) patients who participated in the Butterfly pilot clinical study. Patients had at least one month implantation with the Butterfly prior to implant removal and TURP. Tissue samples were graded by a veterinary pathologist and a uropathologist.

Results: The majority of patients (4/6) had IPSS decreased from baseline. All seven patients' samples had signs of chronic inflammation; one demonstrated acute inflammation and one demonstrated fibrosis. In three cases, intraglandular calcification was identified. There was no ischemic necrosis induced by the implant, and no encrustation, urethral edema, or cellular atypia was noted.

Conclusion:
The Butterfly demonstrated an overall favorable safety profile in terms of tissue response. This study demonstrates that there is no significant tissue reaction in the prostatic urethra due to presence of Butterfly device.

Effects of hemodialysis with cooled dialysate on high-sensitive cardiac troponin I and brain natriuretic peptide

16

Younes Bathish, Karine Beiruti, Hussein Safadi, Adi Sharabi Nov, Elena Bukovetzky, Michael Edelstein, Majdi Halabi, Zeev Israeli
Ziv Medical Centre

Background
Hemodialysis (HD) triggers recurrent and cumulative ischemic insults to the brain and the heart. Cooled dialysate may have a protective effect on major organs and improve hemodynamic tolerability of dialysis. The aim of the study was to compare HD with cooled dialysate with routine dialysis in terms of hemodynamic stability and levels of high-sensitivity Troponin I (hs-TnI) and N-terminal pro b-type natriuretic peptide (NTproBNP) pre and postdialysis.

Methods
The 45 patients were randomized into two groups. The first group received a 35.5°C dialysate first (hypothermic dialysis) and the second group a 36.5°C dialysate first (routine dialysis). Then groups crossed over, so each group received the alternate dialysate (self-controls) For each patient, the first sample was collected at the beginning of dialysis, and a second sample was taken at the end of dialysis. We compared hs-TnI and NTproBNP in the two groups using Wilcoxon signed-rank tests.

Results and conclusion
hs-TnI and NTproBNP increased after routine HD by 10.7 ng\ml ($p < 0.001$) and (12.0 pg/µl) ($p < 0.001$), respectively, and by −3.1 ng\ml ($p = 0.25$) and (4.3 pg/µl) ($p < 0.001$), respectively after hypothermic HD. Our study results showed a tendency towards less rise in hsTnI and NTproBNP during hypothermic HD (35.5°C) as compared to routine HD (36.5°C). Neither arm experienced statistically significant changes in blood pressure. Further studies in larger cohorts and long follow up are warranted in order to confirm that lower rise in (hs-TnI) and NTproBNP actually translate into lower clinical risk for cardiovascular events.

17 Candidate pleotropic genes participating in regulation of the musculoskeletal system's homeostasis

Ben Zvi Eisler Inbar, Shochat-Carvalho Chen, Karasik David.
The Musculoskeletal Genetics Laboratory, The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Osteosarcopenia is a recently described widespread and age-related syndrome, which combines features of both osteoporosis and sarcopenia. Genome-Wide Association study (GWAS) reveals new candidate genes associated with total body lean mass (TB-LM) and bone mineral density (BMD) and complex diseases, such as osteoporosis and sarcopenia. Novel musculoskeletal genes discovered by GWAS require experimental validations to bridge the gap between in-silico discoveries and the mechanisms through which these genes function.
In this study, we aim to validate genes identified by GWAS and find novel genes participating in homeostasis regulation of the musculoskeletal system, using a zebrafish model. We performed preliminary expression screening for genes emerging from GWAS for BMD and TB-LM, first by Real-Time PCR for RNA in larvae at different stages and then for bone, muscle, and fins tissues from adult fish. We then selected relevant candidate genes for mutagenesis by CRISPR-Cas9 technology and generating stable zebrafish mutant lines. Our preliminary analysis showed potential for pleiotropy of the genes *ctnnb1*, *hdac1* and *myoc*. Mutant lines of these genes will allow phenotypic and functional characterization by histology, immunohistochemistry, and micro-CT techniques.
Identifying novel regulators of bone and muscle interactions would promote better understanding of osteosarcopenia pathophysiology and could potentially find new therapeutic targets or re-purpose existing drugs for this syndrome.

Impact of COVID-19 lockdowns on patterns of pediatric emergencies attendances at Ziv Medical Centre: an interrupted time-series analysis, 2018-2021

18

Tomer Bernstine, Michael Edelstein and Danna Krupik
Ziv Medical Center

Background: Israel issued three lockdowns (March-May 2020, September-October 2020, December 2020-February 2021) to mitigate the spread of COVID-19. The impact of lockdowns on childhood injuries in Israel isn't fully described.
Aim: Describe the impact of COVID-19 lockdowns on trends of childhood injuries presenting to Ziv Medical Centre's (ZMC) pediatric-emergency department (PED).
Methods: We extracted 2018-2021 medical records from ZMC PED. We compared the lockdowns to the same time periods in 2018-19 in terms of proportion of attendances for injuries, proportion of injuries hospitalized and age distribution using chi-square tests.
Results: Compared with the 2018-19 average for the same periods, PED attendances decreased during lockdowns, but the proportion of injuries increased: 430 injuries/673 attendances (64%) in lockdown 1 vs. 796/1438 (55%) in 2018-19; 468/613 (76%) vs. 596/1000 (60%) in lockdown 2 and 527/900 (59%) vs. 547/1415 (39%) in the third ($p<0.001$ for all). In the three lockdowns respectively 66(15.3%), 54(11.5%) and 64(12.1%) of injured children were hospitalized vs. 11.9%, 9% and 10.1% in the corresponding 2018-19 periods ($p=0.05$, 0.11 and 0.22 respectively). Compared with the same periods in 2018-19, the proportion of 0-4 year olds attending was higher (44 vs. 29.9%, $p<0.001$, 34.2% vs. 29.1%, $p=0.04$ and 41.5% vs. 32.4%, $p<0.001$ respectively for the three lockdown periods)
Conclusions: Attendance at ZMC PED decreased during lockdowns but the proportion of injury attendances increased. No significant change in hospitalization, and a shift towards younger children were seen. Further studies examining the nature and severity of injuries during lockdowns will help develop injury prevention policies for future pandemics.

19 Unidirectional recruitment is essential for viral latency

Ido Lavi, Supriya Bhattacharya, Ola Orgil, Ankita Awase, Nir Avital, Guy Journo, Vyacheslav Gurevich and Meir Shamay*
Daniella Lee Casper Laboratory in Viral Oncology, Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel 1311502.

Aim & Background:
Kaposi's sarcoma associated herpesvirus (KSHV, HHV-8) is associated with several human malignancies. During latency the viral genomes reside in the nucleus of infected cells as large non-integrated plasmids, known as viral episomes. All KSHV infected cells express LANA, and LANA is essential for viral latency. LANA binding to the viral episomes is critical both for replication of the viral genomes during latency, and for tethering the viral episomes to the cell chromosomes during cell division. Directional recruitment of protein complexes are critical for proper function of many nuclear processes.
Method:
To test for recruitment directionality between LANA and cellular proteins we directed LANA via catalytically inactive Cas9 (dCas9) to a repeat sequence to obtain easily detectable dots. Then, recruitment of nuclear proteins to these dots can be evaluated.
Results & Conclusion:
We found that LANA recruited its known interactors ORC2 and SIN3A. Interestingly, LANA was unable to recruit MeCP2, but MeCP2 recruited LANA. Similarly, histone deacetylase 1 (HDAC1) that interact with the transcriptional-repression domain (TRD) of MeCP2, same as LANA, was unable to recruit MeCP2, but MeCP2 was able to recruit HDAC1. In contrast, HP1a that interacts with MeCP2 through a different domain, was able to recruit MeCP2. We propose that available interacting domains in DNA bound/dimerized form of MeCP2, forces this recruitment directionality. We found that cells derived from Rett syndrome and express a mutant MeCP2 (T158M), impaired in DNA binding, cannot support KSHV genome maintenance. Therefore, this unidirectional recruitment of LANA by MeCP2 identified MeCP2 as a critical factor for viral maintenance.

Vaccine hesitancy and knowledge, attitudes and practices of Obstetrician-Gynecologists in Israel towards COVID-19 and COVID-19 vaccines

20

Michèle Buchinger, Rola Khamisy-Farah
Bar-Ilan University, Faculty of Medicine

Vaccine hesitancy is defined as a delay in acceptance or refusal of vaccination despite the availability of vaccination services [1]. Strategies for addressing COVID-19 vaccine hesitancy include interpersonal-level interventions such as clinician recommendations [2]. Few studies assessed the knowledge, attitudes and practices of health workers, including obstetrician-gynecologists, regarding COVID-19 and COVID-19 vaccines [3,4,5,6,7]. Physicians' perception of the vaccination of pregnant women and those wishing to become pregnant has also been reported [8, 9]. But, to the best of our knowledge, no study has been conducted to investigate the link between clinicians› knowledge and attitudes towards COVID-19 and the vaccine and their recommendations to patients about the COVID-19 vaccine. Our assumption is that the more knowledgeable professionals are about COVID-19 and the COVID-19 vaccine, the more they will recommend it to their patients.

A cross-sectional anonymous survey, based on a literature review of previously published relevant questionnaires and the World Health Organization (WHO) recommendations, was conducted between April 2022 and July 2022 among obstetricians-gynecologists working in Israel. The questionnaire was comprised of five sections, sociodemographic data, general knowledge regarding COVID-19, specific knowledge regarding pregnancy, birth, breastfeeding and COVID-19, knowledge regarding COVID-19 vaccination and finally, attitude and practices towards COVID-19 vaccines. Data analysis is currently in process, results will be available shortly.

21 Investigating cellular and molecular aspects of the pathogenic mechanism in severe TOR1AIP1 gene mutations

Volodymyr Chuiko, Amnon Harel
The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

The nuclear envelope is a two-membrane structure that separates the contents of the nucleus from the cytoplasm. Nuclear envelope proteins perform complex functions that affect multiple aspects of cellular physiology. One of these proteins is LAP1, a type 2 transmembrane protein encoded by the *TOR1AIP1* gene. In humans, this gene encodes two isoforms: LAP1B and LAP1C. Each isoform has three domains: a nucleoplasmic domain, a single transmembrane segment (penetrating the inner nuclear membrane) and a luminal domain (located in the lumen between the inner and outer nuclear membranes). Disrupting the function of this protein results in multisystemic diseases, affecting mainly nervous and muscular tissues. Here, we identify a novel *TOR1AIP1* mutation: ΔNK377-8 that is predicted to lead to a partial misfolding of the luminal domain of the protein and a "loss of function" phenotype. Analysis of primary fibroblasts derived from patients carrying the ΔNK377-8 mutation revealed the absence of LAP1 isoforms and a decrease in some of their interaction partners observed by immunofluorescence. Fibroblast migration through constricted spaces in the transwell assay, showed a drastic decrease in the number of cells that managed to pass the barrier. Mutant cells display slower movement and impaired directionality, indicating that multiple aspects of cell motility are affected by severe mutations in the gene encoding LAP1. We next plan to investigate mechanotransduction pathways between the nucleus and the cytoskeleton and to measure the viscoelastic properties of the nucleus in patient-derived cells.

Convalescent Plasma Reduces Mortality and Decreases Hospitalization Stay in Patients with Moderate COVID-19 Pneumonia

22

Maamoun Basheer [1], Elias Saad [1,2], Dorin Shlezinger [1] and Nimer Assy [1,2,*]

[1] Internal Medicine Department, Galilee Medical Center, Nahariya 2210001, Israel; maamon.basheer@mail.huji.ac.il (M.B.); eliass@gmc.gov.il (E.S.); dorinshl307@gmail.com (D.S.)

[2] The Azrieli Faculty of Medicine, Bar-Ilan University, Safad ,2210001, Israel

* Correspondence: nimera@gmc.gov.il

Abstract: Humans infected with SARS-CoV-2 may develop COVID-19, which manifests across a wide spectrum of clinical severity ranging from mild upper respiratory tract illnesses to diffuse viral pneumonia, causing acute respiratory failure. Many therapies have been tested for their efficacy in treating COVID-19. Controversy surrounds convalescent plasma transfusions as an effective treatment for COVID-19. This study discusses the efficacy of this treatment on COVID-19 patients**.** Electronic medical record data were collected from patients diagnosed with COVID-19, from November 2020 to August 2021, in the Galilee Medical Center's COVID-19 departments. Epidemiological, clinical, laboratory and imaging variables were analyzed. Multivariate stepwise regression and discriminant analyses were used to identify and validate the correlation between convalescent treatment and either death or time to negative PCR and hospitalization length. The study population included 270 patients, 100 of them treated with convalescent plasma. The results show that convalescent plasma therapy significantly prevented mortality in moderate patients, reduced hospitalization length and time to negative PCR. Additionally, high BMI, elderly age, high CRP and 4C-scores correlated with the severity and mortality of COVID-19 patients. Convalescent plasma also significantly reduced inflammatory markers, especially in moderate COVID-19 patients. In non-critical hospitalized patients, convalescent plasma therapy reduces morbidity and mortality in moderate COVID-19 patients and hospitalization length. Identifying patients who could benefit from this treatment could reduce the risk of death and shorten their hospitalization stay.

Keywords: convalescent plasma therapy; mortality; moderate COVID-19 patients; time to discharge; hospitalization length; inflammatory markers

23 Human Papillomavirus (HPV)-related Literacy, Attitudes and Practices Towards Anti-HPV Vaccination Among Nurses from a Regional Healthcare Facility from Northern Israel

Rola khamisy -Farah MD, Professor Marwan Odeh, maher endrawis
The Azrieli faculty of medicine

HPV is a highly common infectious agent, which causes one of the most widespread sexually transmitted diseases (STDs), involving approximately 20% of sexually active female adolescents and up to 80% of female adults aged 50 years. There exist two major preventive approaches: namely, anti-HPV immunization and cervical screening. Health-care providers, including nurses, can play a crucial role in HPV immunization campaigns, counteracting vaccine hesitancy and doing advocacy and counseling. To explore the HPV related literacy level and other HPV-related issues, as well as the attitudes and practices of Israeli nurses, a modified and adapted, previously validated knowledge, attitudes, and practices (KAP) questionnaire was administered to a sample of 565 participants, 481 female (85.1%) and 84 male (14.9%). Most of them were married (n = 428, 75.8%), with more than 5 years of experience (n = 405, 71.7%), working in the general/internal medicine ward (n = 432, 76.5%), and Jewish (n = 352, 62.3%). Only 87 nurses (15.4%) got vaccinated against HPV. (9.1%) of the female nurses never underwent a Pap smear. Higher percentages of correct replies were reported only for the item related to available preventative strategies for cervical cancer (82.1% versus 17.9%). Higher percentages of wrong replies were reported for items related to body regions that can be infected by HPV (60.5% versus39.5%), the portion of girls between 15 and18 years old being sexually active (68.0% versus 32.0%).
Concerning the reliability of the third section of the questionnaire, Cronbach's alpha was deemed to be acceptable (α = 0.64). Statistically significant determinants of reporting lower scores in HPV-related knowledge and attitudes scores were religion (OR 1.44 [95%CI 1.02-2.04]), and male offspring (OR 1.22 [1.03-1.44]). The present investigation has major implications for stakeholders, policy- and decision-makers in that they should be aware of the overall poor and unsatisfactory level of HPV-related knowledge among Israeli nurses.

Generation and characterization of SARS-CoV-2 neutralizing antibodies associated with clinical outcome

24

Pratik Das[1], Michal Werbner[1], Modi Safra[2,3], Pazit Polak[2,3], Moshe Matan[4], Avi Peretz[4,5], Moshe Dessau[6], Gur Yaari[2,3], Meital Gal-Tanamy[1]

[1]Molecular Virology Lab, The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

[2]Bio-engineering, Faculty of Engineering, Bar-Ilan University, Ramat Gan, Israel

[3]Bar-Ilan Institute of Nanotechnologies and Advanced Materials, Bar-Ilan University, Ramat Gan, Israel

[4] Clinical Microbiology Laboratory, Baruch Padeh Medical Center, Poriya, Israel

[5] The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

[6] The Laboratory of Structural Biology of Infectious Diseases, The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

Aim and Background: The 'novel coronavirus disease' (2019-nCoV) or severe acute respiratory syndrome (SARS-CoV-2) is the leading cause of the recent global catastrophe known as the COVID-19 disease which originated in December 2019. SARS-CoV-2 enters the host cells via interaction between the angiotensin-converting enzyme (ACE2) receptor and the viral envelop protein 'spike'. Emerging SARS-CoV-2 variants contain mutations that resist antibody neutralization. The use of neutralizing antibodies is currently the most promising approach to prevent the viral infection.
Here, we propose to generate an effective SARS-CoV-2 neutralizing antibodies associated with specific clinical outcome, and investigate their neutralizing abilities, epitopes, resistance and neutralization breath.

Methods: For identifying neutralizing antibody clones against SARS-CoV-2, a dataset containing immunoglobulin heavy and light chain sequences from 13 severe COVID-19 patients and 50 mild/moderate patients were collected. By antibody repertoire analysis, we identified several neutralizing antibody clones prevalent in COVID-19 patients. The full-length antibodies were prepared by cloning the respective heavy and light chain sequences for expression and purification. Neutralization assays were conducted to evaluate the neutralization capacity against VSV and lentiviral based SARS-CoV-2 pseudo-particles. For epitope mapping and selection of resistant mutations, replication competent rVSV expressing SARS-CoV-2-S was prepared.

Results and Conclusion: We identified antibody clones that are prevalent in COVID-19 patients and produced the full-length antibodies successfully. We demonstrated that these antibodies efficiently neutralize SARS-CoV-2. This study is expected to lead towards generation of antibodies with various mechanisms of neutralization that may be efficient as a combination therapy for the cure of COVID-19 patients.

25 B cell mediated immunogenicity following boosting with the BNT162b2 SARS-CoV-2 vaccine in dialysis patients, Israel, 2021-22

Kamal Abu Jabal [1,2], Elizabeth Eshel[1], Neta Tuvia[1], Tali Lange-Tal[1], Netta Beer[1], Michael Edelstein [1,2] ,Younes Bathish [1]
1. Ziv Medical Centre, Safed, Israel
2. Bar Ilan University, The Azrieli Faculty of Medicine, Safed, Israel

Chronic kidney disease patients (CKDP) sometimes require specific vaccination regimens to achieve optimal protection. We determined whether B-cell response following a third BNT162b2 vaccine dose (boosting) differed in CKDP and those with normal renal function, to inform vaccination policy in CKDP.

We recruited double-vaccinated, dialyzed CKDP and compared them to age-matched, double-vaccinated healthcare workers in (i) the proportion of peripheral B cells producing SARS-CoV-2 anti-spike IgG (SARS-CoV-2-B cells), measured using flow cytometry; (ii) geometric mean concentration (GMC) of SARS-CoV-2 anti-spike IgG antibodies in serum (IgG), measured by a quantitative serological assay before and after boosting. We compared the two groups using two-way ANOVA and Kruskal-Wallis tests.

Among the 23 CKDP and 26 controls, the proportion of SARS-CoV-2-B cells changed significantly ($p<0.001$) over time, increasing from pre-boosting to 1-2 months post-boost (0.09% to 0.5% and 0.18% to 0.27% respectively), and decreasing 4-6 months post-boost (to 0.3% and 0.17% respectively). The proportion of SARS-CoV-2-B cells was similar between the two groups at all three time points ($p= 0.110$). Compared with controls, IgG GMC among CKDP was lower pre-boost (29 vs. 178 AU/ml, $p<0.003$) and similar 1-2 months (2429 vs. 1139 AU/ml, $p<0.06$) and 4-6 months later (257 vs 447 AU/ml, $p<0.14$).

BNT162b2 booster immunogenicity was comparable in healthy controls and CKDP for circulating peripheral SARS-CoV-2-B cells and IgG, suggesting that in CKDP, post-vaccination B cell response is preserved and normal vaccine formulation is suitable. Lower pre-boost IgG levels in CKDP require further investigation to determine the need for an earlier booster.

Does Cerebroplacental-ratio in late IUGR fetuses improves outcomes?

26

Netaly Faramand 1,2, Revital Vinitski1,2, Bili Gasman, Zohar Nachum3,4, Enav Yefet1,2

Department of Obstetrics and Gynecology, Baruch Padeh Medical Center, Poriya, affiliated with Azrieli Faculty of Medicine, Bar Ilan University, Israel.1 Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.2 Department of Obstetrics and Gynecology, Emek Medical Center, Afula.3 Rappaport faculty of medicine, Haifa.4

Aim & Background: The management of intrauterine-growth-restriction (IUGR) is controversial. Late IUGR is classified when diagnosed above 32.0 weeks. Placental-insufficiency-associated late IUGR is suspected when the Cerebroplacental-ratio (CPR) (the ratio between the fetal middle-cerebral-artery pulsatile-index (PI) and the umbilical artery PI) increases. CPR is used for management; fetal well-being assessment is recommended once and twice a week in normal and pathological CPR, respectively. In the present study we examined the beneficial role of CPR in surveillance and labor management of IUGR fetuses.

Methods: A retrospective cohort study. We compared neonatal outcomes of 4 groups: suspected IUGR with normal (N=127) and pathological (N=39) CPR and two control groups– IUGR neonates diagnosed only postnatally (N=364) and appropriate for gestational age fetuses (AGA;233). The primary outcome was a composite neonatal outcome (Apgar score <7 at 5 min, cord pH<7.1, neonatal intensive care unit (NICU) admission, cesarean section due to suspected fetal distress, neonatal oxygen support and intrauterine fetal death).

Results: The incidence of the composite outcome was: 26 (67%), 40 (32%), 98 (27%) and 45 (19%) in the pathological CPR, normal CPR, postnatal IUGR and AGA, respectively (P<0.0001 comparing all groups and comparing suspected IUGR with pathological and normal CPR). The surveillance protocol led to preterm delivery in 6 (15%) versus 6 (5%) in the pathological and normal CPR groups, respectively (p=0.04).

Conclusion: IUGR fetuses, particularly with pathological CPR have less favorable outcome. Yet, a strict surveillance did not improve the outcome of fetuses with normal CPR and therefore should be reconsidered.

27 Characterization of Immunodominant Epitopes on Hepatitis C Viral Envelope

Haneen Faris-Gadban[1], Anastasia Brodov[1], Michal Poran[1], Haim Ashkenazy[2], Tal Pupko[2], Jonathan Gershon[2] Meital Gal-Tanamy[1]
[1]Molecular Virology Lab, The Azrieli Faculty of Medicine, Bar-Ilan University.
[2]The George S. Wise Faculty of Life Sciences, Tel-Aviv University.

Background & Aims: Hepatitis C virus (HCV) is a leading cause of liver disease affecting an estimated number of 71 million people worldwide. HCV infection is the major cause of liver transplantation. Although there is an effective HCV treatment by direct acting antivirals, there is still a need for developing effective anti-HCV vaccines since the cancer rates remain high after treatment. Approximately 25% of infected patients clear the virus spontaneously. We therefore aim to investigate antibodies associated with viral clearance and immunodominant epitopes on the HCV-E2 envelope protein contributing to successful antibody-mediated neutralization of the virus.

Methods: For identifying immunodominant epitopes, a random phage display library was used to detect binders to antibodies in sera obtained from HCV spontaneous clearers (SC) and chronically infected (CI) patients. By repetitive rounds of biopanning followed by sequencing of the binding peptides, we identified peptides representing epitopes that were aligned to the HCV E2 protein. These peptides were evaluated for binding to neutralizing antibodies in SC and CI sera using ELISA and neutralization assays.

Results and Conclusion: We discovered a new immunodominant epitope that is SC-unique and demonstrated that it binds neutralizing antibodies abundant in SC compared to CI sera. Mice vaccinated with the new epitope-based peptide showed higher potency of viral neutralization compared to other peptides. Moreover, we aim to discover new antibodies associated with viral clearance using the phage display technique and map and characterize their epitopes. This may lead towards the development of anti-HCV rational vaccine design or neutralizing antibody treatment.

Fetal major anomalies and related maternal and obstetrical outcomes

28

Tal Skliar[2], Lior Lowenstein[1,2], Maya Frank Wolf[1,2], Inshirah Sgayer[1,2]
[1] Department of Obstetrics and Gynecology, Galilee Medical Center, Nahariya, Israel
[2] Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel

ABSTRACT

Objective: We compared maternal and obstetrical outcomes of pregnant women, between those with and without major fetal anomalies.

Methods: A retrospective case-control study between 2010-2019 of 117 women with singleton pregnancies complicated by major fetal anomalies at gestational age ≥23 weeks and a control group of 322 pregnant women.

Results: In the study compared to the control group, the rates were higher of preterm delivery (<37 weeks) (46.2% versus 6.2%, p<0.001) and of cesarean section (CS) (53.8% versus 28.3%, p<0.001). A higher rate of CS due to abnormal fetal presentation in the study group compared with the control group (25.4% versus 7.7%, respectively, p=0.003). In eight cases of the study group CS was performed due to fear of obstructed labor or fetal trauma.

The rate of stillbirth was 17.1% in the study group, compared to 0.3% in the control group (p<0.001). Neonatal death occurred in 12.5% of the study group and none of the control group (p<0.001); and infant death in 2.5% and none, of the respective groups (p=0.019). Major fetal anomalies were found to be associated with adverse maternal outcomes (OR=2.47, 95% CI 1.50-4.09, p<0.001). Polyhydramnios was an independent risk for a composite maternal adverse outcome (OR=4.73, 95% CI 1.65-13.56, p<0.001).

Conclusions: Women with major fetal anomalies should be considered as having high-risk pregnancy, due to the association with adverse maternal outcomes, especially if polyhydramnios exists. There is a need for a structured follow up program for these parents including multidisciplinary team to provide optimal medical and emotional care.

29 Development of a specific inhibitor for the Drp1/Parkin interaction, and investigation of its effect on mitochondrial processes and in myocardial infraction animal models

Samar Gani[1], Nir Qvit[1]
The Azrieli Faculty of Medicine in the Galilee, Bar Ilan university, Henrietta Szold St. 8, POB 1589, Safed, Israel.

Aim & Background: Dynamics mitochondrial processes, such as fission fusion and mitophagy, are the main player in cardiovascular homeostasis. Mitochondria play an important role in myocardial injury. The injury interferes with mitochondrial dynamic processes, in particular, excess mitochondrial fission, playing a major role in dysfunction of the heart.
Dynamin related protein 1 (Drp1) is a guanosine-triphosphate (GTPase) that modulates mitochondrial fission. Parkin is a U3 ubiquitin ligase and ubiquitinates proteins that marks them for degradation by lysosomes. Dysregulated of Drp1/Parkin axis represents one of the early events predisposing susceptibility of neuro cells.
We would like to characterize the Drp1/Parkin protein-protein interactions (PPIs) *in vitro* and in myoblasts cells. We further intend to study the role of Drp1/Parkin in the cardiovascular system and its importance in mitochondrial homeostasis.
Methods: We expressed and purified Drp1 and Parkin by affinity column; tested and validated Drp1/Parkin PPIs by pull down and Field Effect Biosensing (FEB) methods. Next, we designed a specific peptides inhibitor of Drp1/Parkin PPI by L-align software, then we synthesis the peptides by solid-phase peptide synthesis (SPPS) and evaluated their purity by using reverse-phase high-pressure liquid chromatography (RP-HPLC). Finally, using H9c2 cardiomyocyte we tested the peptide and evaluate their effects on cell viability and toxicity using XTT assay. In the future, we plan to study the peptides toxicity in rats.
Results and Conclusion: We successfully get the purified proteins and peptides, and investigate their interaction by pull sown and FEB, and show that all four peptide CVP-001, CVP-002, CVP-032 and CVP-033 protective the stressed cells and the viability around 79% as contrast to only 60% for the untreated cells.
Next, we plan to examine the peptide *in vivo*, in an animal model of myocardial infarction (MI) in rats, and further, study their role in mitochondrial dynamics in cell fate.

So, what are the differences between all these endocrine cells?

30

Lihi Grinberg, Worood Sirhan, Assaf Malka and Ron Piran
The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Abstract

Aim & Background: Previously, we showed that pancreatic injury consisting of acinar cell damage induced by pancreatic duct ligation (PDL) led to islet cell transdifferentiation from α-cells to β-cells. Despite of the robust β-cell regeneration, mice did not survive for long and remained diabetic. Therefore, the complex surgical procedure of PDL was replaced with caerulein. Similarly, to PDL, caerulein plus β-cell ablation induced α- to β-cell transdifferentiation, with many β-cells further transdifferentiating into δ-cells expressing somatostatin, resulting from α- to β- to δ-cell transdifferentiation. That is to say that glucagon secreting α-cells transdifferentiated to β-cells, but they weren't stable and continued to differentiate into δ-cells. Our goal is to stabilize the β-cells and prevent the continued differentiation into δ-cells.

Methods: We intend to study what differentiate α- and δ-cells from each other and from β-cells and to use these findings to explore ways to stabilize the β-cell intermediate during the α- to β- to δ-cell transdifferentiation process. To find the differences between α-, β-, and δ-cells, we used Laser Microdissection technology, which allows for the isolation of the different cell types in their own vile. This innovative technology allows the molecular characterization of each cell type.

Results& Conclusion: We made significant progress establishing the proteomic and transcriptomic characterization of murine α-, β-, and δ-cells, in which we intend to compare the molecular differences between type I, II, and healthy islet cells.

31 P-systems with protein rules: Diabetes Mellitus as a case study.

Yara Hamshawi[a], Florin-Daniel Bîlbîe[b], Andrei Păun[cb], Assaf Malka[a], Ron Piran[a]

[a] The Azrieli Faculty of Medicine, Bar-Ilan University, 8 Henrietta Szold St., Safed, 1311502, Israel

[b] ICUB/Faculty of Mathematics and Computer Science, University of Bucharest, 14 Academiei St. District 1, Bucharest, 010014, Romania.

[c] National Institute of Research and Development for Biological Sciences, 296 Independenței Bd. District 6, Bucharest, 060031, Romania.

In our recently published paper, we succeeded to demonstrate the power of a new methodology by simulating a system with complex conditions, such as glucose homeostasis in health and disease (i.e., diabetes), while bringing the science of membrane-computing closer to the natural world. P-system (aka membrane computing) is a subfield in computer science, which relies on modeling living systems with mathematical tools. In classical P-system, cells are surrounded by a simple membrane and computational events take place in both sides of the membrane. When we tried to model the process of normoglycemia in healthy individuals as well as in diabetes, the main challenge was to prioritize the insulin-producing β-cells over other organs. However, using classical P-system, we could not implement this hierarchy. Therefore, we chose to utilize the membrane actual physiology and add its properties to the current definitions of membrane-computing. To our gratification, we succeeded to develop a new theoretical tool to be complemented into the science of membrane-computing. This development relies on the membrane structure of the cell and on the biochemical reactions which occur on the membrane of different organs in our body, thus simulating the true nature of membranes.

Here, we will present the model we developed, hoping that these new viewing angles on biological conditions will lead to the integration of computational modeling into biological science and to a treatment for diabetes that will also pave the way for additional treatments for many more diseases that currently do not have a cure.

The effect of Metformin on vitamin B12 deficiency and stroke

32

Nizar Horrany, Wadie Abu Dahoud, Yaara Mouallem, Taleb Hajouj, Arnon Blum
Department of Medicine, Padeh Medical Center, Poriya, affiliated with Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel

Background
Type 2 Diabetes Mellitus (T2DM) is a known risk factor to cardiovascular disease (CVD) and stroke. Metformin is an old, relatively safe, first line therapy in T2DM. However, it has been associated with stroke.

Methods
A prospective study of patients admitted with ischemic stroke in 12 months (starting March 2020) to our medical center. We studied the clinical impact of Metformin on vitamin B12 deficiency and stroke evolution. Student T-test and ANOVA were used to comapare between the groups of patients and in order to find whether there is any direct or indirect effect of Metformin use on vitamin B12 defiency and on stroke evolution.

Results
80 patients were admitted with ischemic stroke. Clinical status and biochemical data were collected and compared with healthy volunteers. Among the 39 diabetic patients, 16 took Metformin (at least 1 year), 9 of them had vitamin B12 level < 240 pg/ml (56.2%). 23 diabetic patients didn't get Metformin, and only 4 had vitamin B12 level < 240 pg/ml (17.4%) (p=0.014).

Conclusions
T2DM is a significant risk factor to the development of ischemic stroke. We have shown an association between Metformin use and vitamin B12 deficiency. More than that, we have shown an association between vitamin B12 deficiency and stroke in patients with T2DM. Therefore, we suggest to monitor closely levels of vitamin B12 in diabetic patients who are taking Metformin.

33 Designing Biologically Inspired 3-dimensional Constructs for Tissue Reconstruction

Idan Redenski[1,2], Ben Kaplan[3], Shulamit Levenberg[3], Samer Srouji[1,2]
[1]Department of Oral and Maxillofacial Surgery, Galilee Medical Center, Nahariya, Israel
[2]The Azriely Faculty of Medicine, Bar-Ilan University, Safed, Israel
[3]Faculty of Biomedical Engineering, The Technion, Haifa, Israel

When approaching the task of functional regeneration of large-scaled defects, both heavily vascularized tissue grafts and patient-specific anatomy are necessary. Autologous tissue-harvest, considered as the current standard of care, possess considerable drawbacks such as tissue-site morbidity and post-operative pain. Tissue engineering approaches relying on both 3-dimensional design and biological induction of implants can heavily promote grafts survival during the engraftment process.

The development of neo-tissues is initiated by loading co-cultures of stem cells onto soft tissue matrices. Then, bioengineered soft-tissue scaffolds are combined with bone-regeneration constructs to yield composite neo-tissues. These in turn, undergo vascularization and biological induction in a small animal rat model. Both ex vivo and in vivo assessment of maturation and engraftment of the neo-tissues was performed. Creation of 3D-bioprinted maxillofacial bone is followed, with the goal of creating a proof-of-concept up-scaled neo-tissue for clinical purposes. In the current work, pre-vascularization and osteogenic induction of tissue constructs are validated both *in vitro* and *in vivo*. High-resolution micro-computed tomography enabled longitudinal assessment of bone deposition, remodeling and formation of micro-vessels within neo-tissues. Eventually, a maxillofacial bone with complex anatomical features is 3D-printed by employing freeform reversible embedding of suspended hydrogels and bioinks (FRESH). The presented methodology demonstrates neo-tissue fabrication, used to bridge both soft and hard tissue defects. In addition, functional vascular connectivity between hosts and grafts are validated as well, followed by the formation of an exact replica of a maxillofacial bone graft.

PERFORMANCE COMPARISON OF THREE PREDICTIVE MODELS OF OVARIAN MALIGNANCY, IN A SECONDARY HOSPITAL WITH GNECOLOGY ONCOLOGY SERVICE

34

Sapir Zalah[1], Adi Sharabi-Nov[2, 3], Inbar Ben Shachar[1, 2], Yael Sciaky-Tamir[1, 2]

1. Azrieli Faculty of Medicine, Bar-Ilan University, Israel
2. Obstetrics & Gynecology Department, Ziv Medical Center, Safed, Israel
3. Tel-Hai academic college, Tel-Hai, Israel

Abstract

Objective - To examine which one of the three predictive models (Simple Rule – SR, logistic regression model 2 - LR2, Assessment of Different Neoplasia's in the adnexa – ADNEX) of the International Ovarian Tumor Analysis (IOTA) group will give the most accurate prediction of malignancy or benignity of an ovarian mass.

Methods - A retrospective study of women who went through adnexal mass resection at Ziv Hospital between the years 2017-2019. A comparison was made between a diagnosis based on ultrasound examination by the three models to the histopathological diagnosis. The comparison of the prediction of malignancy between the various models was made by examining statistical variables of sensitivity, specificity and negative and positive predictive values.

Results - Out of 120 women who went through the surgery, pathological examination demonstrated: 23 women with normal physiological tissue, 78 women with benign mass, 7 women with borderline tumor and 12 women with malignant tumor. The ADNEX model demonstrated a sensitivity, specificity, PPV and NPV of 72.2%, 86.8%, 56.5% and 93% respectively. The LR2 model demonstrated a sensitivity, specificity, PPV and NPV of 84.2%, 39.6%, 20.8%, and 93%. The SR model demonstrated a sensitivity, specificity, PPV and NPV of 42.9%, 98.9%, 75% and 95.6%.

Conclusion: We demonstrated that the ADNEX model is the most accurate model. This model presents relatively high values of sensitivity and specificity. Unlike the other models, the ADNEX model presented similar values of sensitivity and specificity variables, which makes it the most preferred one.

Keywords: ADNEX , LR2, SR, IOTA group, ovarian carcinoma, CA-125 marker.

35 From genotoxicity to oncogenicity: Uncovering the molecular signature of Helicobacter pylori in gastric cancer

Rose Jbara[1], Hadas Sibony-Benyamini[1], Tamar Leshem[3], Avi Peretz[2,3], Yaakov Maman[1]

[1]Laboratory for Genome Instability and Cancer, Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Zefat, Israel.

[2]Clinical Microbiology Laboratory, Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Zefat, Israel.

[3]Baruch Padeh Medical Center, Poriya, Israel.

Gastric Cancer (GC) is the third most common cause of cancer-related deaths. A major GC risk factors is an early infection with Helicobacter pylori (*H.pylori*). Growing body of evidence portrays a deleterious effect of *H.pylori* on genome integrity of the host. While this may imply a link between *H.pylori* genotoxicity and its oncogenicity, the biological mechanism/s of *H.pylori*-induced DNA damage and their role in GC tumorigenesis remain largely elusive.

By conducting genome-wide mapping of DNA damage in gastric cells upon *H.pylori* infection, we show that *H.pylori* infection results in thousands of recurrent double-stranded breaks (DSBs) across the genome. The structure and localization pattern of these breaks reflected DNA replication stalling. Such harmful event takes place in conditions of replication stress such as nucleotide depletion. Indeed, quantification of the dNTPs concentration in cells revealed dramatic reduction upon H.pylori infection. Consistent with that, *H.pylori* infection results in downregulation of the Ribonucleotide Reductase, the enzyme that responsible for dNTP synthesis. Strikingly, external supplementation of dNTPs strongly attenuated *H.pylori*-mediated damage. Finally, DSBs identified upon *H.pylori* infection show a significant co-localization with the breakpoints of structural variants (SVs) from gastric cancer patients. Together, our data imply that *H.pylori* genotoxicity, as well as its oncogenic potential, are rooted in its capacity to impose dNTP depletion in the host cell, which in turn results in replication fork collapse and DNA damage.

SMURF2 protects nuclear shape and genomic integrity through ubiquitination of Lamin A

36

Venkata Narasimha Kadali, Gal Levy-Cohen, and Michael Blank
Laboratory of Molecular and Cellular Cancer Biology, Azrieli Faculty of Medicine, Bar-Ilan University, 1311502 Safed, Israel

Background

A-lamins encoded by the *LMNA* gene are major structural components of the nuclear envelope (NE), coordinating essential cellular processes. Mutations in the *LMNA* gene and/or alterations in its expression levels linked to human disorders, collectively known as laminopathies, and to cancer. Despite their importance, the molecular mechanisms involved in their regulation remain unknown. We recently discovered that SMURF2, an E3 ubiquitin ligase and suggested tumor suppressor, that directly binds, ubiquitinates and negatively regulates the expression of lamin A and progerin through autophagic-lysosomal pathway. These findings established SMURF2 as a novel regulator of A-lamins: lamin A and progerin. Despite the foregoing, it is yet unknown how lamin A ubiquitination by SMURF2 affects the cellular and molecular processes.

Methods

We used human cancer and transformed cell lines such as U2OS, HeLa and HEK-293T cells. Ubiquitination assays, localization, nuclear structure, and genomic instability were all examined using Western blot and confocal microscopy.

Results and Conclusion

To address the above question, we mapped on lamin A several lysine residues as potential ubiquitination sites of SMURF2, and we discovered Lys32 of lamin A as an authentic ubiquitin acceptor site. Intriguingly, our studies revealed that the lamin A-K32R mutant (Lys mutated to Arg) is unable to properly assemble at the nuclear rim, drastically impairing the cell's nuclear shape. We also found that lamin A mutant cells undergoes genomic instability and excessive DNA damage in unstressed conditions. Overall, our research reveals that SMURF2 ubiquitinating lamin A at lys32 influences lamin A assembly at NE and protects nuclear shape and genomic integrity.

37 The Butterfly device for benign hypertrophy of the prostate – more than 1 year of follow up

Ran Katz, Wasim Abu Nasra, Muhahmd Abu Ahmed, Elly Jarus, Ali Safadi, Tarek Taha, Alexander Visoky
Ziv Medical Center, Safed IL

Introduction & Objectives
The Butterfly Retraction Device is a novel minimally invasive implant that is delivered into the prostatic urethra in order to retract the lateral lobes of the prostate and treat symptoms associated with BPH.

Materials & Methods
64 men, all treated for BPH for at least one year were enrolled. All patients had Qmax≤ 13 ml/sec and IPSS > 12. Patients with previous prostate surgery, prostatitis, cystolithiasis, prostate median lobe, urethral/bladder neck stricture and atonic bladder were excluded.
Insertion of the device was performed under sedation. No catheter was left. Follow up visits were performed at 2 weeks, 1, 3, 6, and 12 months and included uroflowmetry, IPSS, QoL and sexual function questionnaires. Cystoscopy was performed on 3 and 12 months.

Results
Patients' age was 50-83 years. Follow up period ranged from 4 to 35 months. 28 patients completed a minimum of 1 year follow up with an intact device.
In 12 months, mean improvement in Qmax was 2 ml/sec (25%), Mean PVR decrease was 25.9% . Mean IPSS decrease was 40%. The mean improvement of QoL was 1.5 points (38%).
No patient reported deterioration of sexual function and all sexually active patients reported antegrade ejaculation.
On cystoscopy, gradual coverage of the device with urethral mucosa was observed and there were no cases of encrustation.
Conclusions
The butterfly device is an efficient minimally invasive device for the management of LUTS due to BPH with a low complication rate, and good tolerability.

Stabilizing neogenic β-cells by blocking glucagon signaling

38

Worood Sirhan[1], Daljeet Kaur[1], Assaf Malka[1], and Ron Piran[1]
[1]The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

Abstract

Aim & Background: In Diabetes, blood glucose levels increase above physiological concentrations. All diabetes types are characterized by insulin secreting – beta (β) cell loss over time. An approach for replenishing β-cells is to induce endogenous regeneration. Three pathways for endogenous replenishment are proposed for β-cells: replication, neogenesis, and transdifferentiation. We showed that the pharmacological procedure of alloxan plus caerulein (A+C) injections led to islet cell transdifferentiation, but the outcome was large amount of delta (δ) cells resulting from alpha (α) to β- and then to δ-cell transdifferentiation. We hypothesize that the relative excess of α-cells is propagating this process, as δ-cells inhibit glucagon secretion from α-cells. Therefore, we aim to block the β- to δ-cell continued transdifferentiation by desensitizing neogenic β-cells to glucagon signaling.

Methods: To desensitize endocrine cells to glucagon signalling, we constructed α-cell specific Gcg Cre-GcgR floxed knockout (GcgCre-GcgR KO) mice in our laboratory. Two months old littermates of C57BL/6 WT and GcgCre-GcgR KO mice were randomized into two groups that underwent A+C treatment and later were supported with exogenous insulin. Mice blood glucose levels were measured daily until day 170 when the mice were sacrificed.

Results & Conclusion: Mice in the experimental group regenerated their β-cells and became insulin independent whereas the control remained diabetic over the span of 170 days. Thus, proving that α-cell signaling is indeed the driving force, which leads to further transdifferentiation and by inhibiting it neogenic β-cells become stabilized and functional.

39 The mechanism of epigenetic changes imposed by KSHV encoded LANA

Yakeen Kezil, Supriya Bhattacharya, Vyacheslav Gurevich, Meir Shamay
Daniella Lee Casper Laboratory in Viral Oncology , Azrieli Faculty of Medicine Bar-Ilan University, Safed ,Israel 13115025

Kaposi's sarcoma associated herpesvirus (KSHV), belongs to the gamma herpesvirus family and etiologically associated with Kaposi's sarcoma (KS), primary effusion lymphoma (PEL) and Multicentric Castleman's Disease (MCD). KSHV life cycle includes two phases of infection the lytic phase and the latent phase. The Latency Associated Nuclear Antigen (LANA) is one of the few proteins expressed during the latent phase and plays a major role in KSHV pathogenesis. Previous studies showed that epigenetic mechanisms including DNA methylation and Polycomb repressive complexes (PRCs) plays an important role during establishment and maintenance of herpesvirus latency. In normal cells many promoters contain unmethylated CpG islands and already repressed by polycomb repressive complex, but in cancer cells some of these CpG islands become hypermethylated and this is usually correlated with maintained repression. Previous studies found interaction between LANA and the polycomb complex via the EZH2 subunit. LANA also interacts with the de-novo methylatrasnferase DNMT3A. We hypothesized that LANA may bridge between the polycomb and DNA methylatransferases leading to CpG DNA methylation. In order to test our hypothesis, we directed polycomb complex to specific cellular promoters via CRISPR dCas9. Then we tested the ability of polycomb to recruit LANA, and DNA methytransferases, to that specific cellular promoter. In addition, both DNA methylation and expression of these genes will be examined. We expect that these studies will reveal the mechanism by which KSHV impose DNA methylation on the cellular & viral genome, a process linked to maintaince of viral latency and development of cancer.

The effect of the microbiome on lowering aggressive behavior in Drosophila melanogaster

40

Rachel Levin[1], Michal Grinberg[1], Sondra Turjeman[1] and Omry Koren[1]
[1] The Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.

Aim & Background: The social life of animals depends on communication between individuals, with aggression manifesting as one of the common types of behaviors, preserved among most species. An organism will use aggression for self-defense against conspecifics and predators, in acquisition of territory, food and mates, and in defense of progeny.
Recent findings link variations in gut microbiome composition with animal social behavior such and various cognitive and emotional aspects. This study uses *Drosophila melanogaster* as an animal model with the aim of determining the effects of the microbiota on aggression.

Methods: Male flies reared under different treatment [control, mix of antibiotics (abx: tetracycline, rifampicin, streptomycin) and germ-free flies] were subjected to behavioral tests to measure aggression. Microbiome analysis was carried out using QIIME2; in addition, we used real time PCR to test gene expression, western blot to examine protein expression and SEM to study the antenna and maxillary palp.

Results & Conclusions: The absence or presence of microbiome affects aggression in flies. Flies without a microbiome (abx) are significantly more aggressive behavior compared to flies with microbiome. We show that presence of microbiome reduces aggressive behavior in flies; further research will aim to unearth the mechanism as well as the specific effects of the presence or lack of microbiome on aggression.

41 Characterization of PKA holoenzyme complex in a rare neurodegenerative disease derived by PKA point-mutation

Ashar Masri[1], Ronit Ilouz[1]
Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel[1]

Abstract
Protein kinase A (PKA) is an enzyme that recruits various cell signaling by protein phosphorylation. It consists of two regulatory subunits and two catalytic subunits forming an inactive holoenzyme, activated by cAMP. A new neurodegenerative disorder characterized with dementia and/or parkinsonism displays a heterozygous missense mutation (L50R) in the dimerization and docking (D/D) domain of type I-beta regulatory (RIβ) subunit gene of PKA, which creates a docking site for A Kinase Anchoring Proteins (AKAPs), tethering the PKA holoenzyme to distinct cell compartments. Previous work in our lab established that the L50R mutation in the D/D domain of RIβ breaks the dimerization but allow the interaction with its catalytic subunit and form a heterodimer. In this study, by using molecular biology, biochemical and cell-based techniques we aim to understand the molecular mechanism leading to this disease and to distinguish between the two functional defects: PKA mis-localization by impairing it's binding to AKAPs and breaking the two similar R:C heterodimers, we generated a point mutation, I27A, in the D/D domain of RIβ. we revealed that I27A did not disrupt homo-dimerization while still leading to mis-localization of PKA complex. Additionally, we created a double-mutation, L50R+R211K, in the cAMP binding domain A (CNBA) of the regulatory subunit, to increase the binding affinity of the catalytic subunits to the regulatory subunits. Cell dynamic experiments revealed that aggregation is prevented when cAMP rises in the cell in the double-mutant, while is not in L50R, as a result of the dissociation of the catalytic and the regulatory subunit.

A genomic duplication of 83 Kbp is associated with the Mammary-Digital-Nail (MDN) syndrome

42

Ayalla Fedida[1, 2], Golan Nadav[1, 2], Meir Shamay[2], Leonid Kogan[3], Tameema Attallah Moadi[1], Mary Noffi Bahoum[4], Hector I Cohen[5], Alexander Kaplun[6], Rana Nasara[5], Limor Kalfon[1], Malte Spielman[7], and Tzipora C. Falik-Zaccai[1, 2]

[1] Institute of human genetics, The Galilee Medical Center, Nahariya.
[2] The Azrieli Faculty of Medicine, Bar Ilan University, Safed.
[3] Department of plastic surgery, Galilee Medical Center, Naharia, Israel
[4] Pediatric endocrinology service, Clalit, Health Service, Western Galilee, Israel
[5] Department of pathology, Galilee Medical Center, Naharia, Israel
[6] Variantyx Inc., Framingham, USA"
[7] Institute of Human Genetics, University of Lübeck, Lübeck, Germany.

Background: Mammary Digital Nail syndrome (MDN) is linked to a 4.3 Mb interval on chromosome 22q12.3-13.1. We aimed to reveal the causative genetic variant, and molecular mechanism underlying the MDN phenotype. An Irish family with onychodysplasia and a Druze family with MDN were recruited to the study.

Results: Chromosomal microarray analysis revealed a novel heterozygous genomic duplication of 83 Kbp, within the linked interval on chromosome 22 in affected individuals from both families. This duplication contains two ORFs: one encoding the potassium channel KCNJ4 and the other encoding an inositol lipid phosphatase pseudogene (TPTEP2). Relative qPCR confirmed an autosomal dominant segregation pattern within the MDN Druze family. An overlapping duplication including KCNJ4 gene and only the first out of five exons of TPTEP2, was detected in the Irish family presenting ODP. RT-qPCR and WB analysis revealed a significant increase in the transcript as well as protein level of KCNJ4 in skin and breast biopsies derived from affected females. Transcriptome analysis identified dysregulated pathways specific in breast from affected females

Conclusions: A genomic duplication on chromosome 22q12.3-13.1, is linked to the MDN and ODP phenotypes in two unrelated families, strongly suggesting that it is the MDN causative variant. The significant higher abundance of KCNJ4, encoded in the duplication, in the MDN tissues may suggests it's involvement in the molecular mechanism causing the diseases phenotype. Elucidation of the mechanism of pathogenicity may reveal novel insights on the embryonic development of digits and nails, and pubertal breast development.

43 Antimicrobial activity of Black Soldier Fly (Hermetia illucens) larvae hemolymph against Clostridium difficile

Aviel Melchior [1], Maya Azrad [2], Avi Perez [1,2]
[1]Azrieli Faculty of Medicine, Bar Ilan University
[2] Clinical Microbiology Laboratory, Baruch Padeh Medical Center, Poriya

Background: *Clostridioides difficile* (*C. difficile*) is the main pathogen responsible for nosocomial diarrhea. The incidence of *C. difficile* infection (CDI) has increased in recent decades, along with severity of CDI and complication and morbidity rates. High rates of antibiotic resistance pose a major challenge in CDI treatment, thus alternative therapeutics agents are needed.
Antimicrobial peptides (AMPs) hold great promise as novel antibiotics since they can be produced at low costs and are expected to induce low or no resistance compared to antibiotics. Insects, such as the black soldier fly (BSF, *Hermetia illucens*), have gained attention due to the impressive repertoire of AMPs they produce to protect them against pathogens.

Study aims: To investigate the antimicrobial activity of the BSF hemolymph (HIL) against *C. difficile* and to compare this activity between immunized and non-immunized HIL.
Methods: Fourth instar HILs were immunized with *C. difficile* strain 630 (ATCC BAA-1382, which produces toxin A + toxin B) by feeding them brewer's grains medium infected with *C. difficile*. Following a 24-h incubation, antimicrobial activity was tested using the disk diffusion test.

Results: The inhibitory effect of immunized larvae HIL on *C. difficile* growth was significantly greater than that of non-immunized larvae HIL.

Conclusions: The results obtained in current study indicate that HIL hemolymph AMPs are effective tools against multidrug-resistant *C. difficile* strains**.**

The effect of the gut microbiome on aggression

44

Lelyan Moadi[1], Dana Binyamin[1], Nofar Asulin[1], Soha Zgairy[1], Sondra Turjeman[1], Omry Koren[1]
[1]Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel

Background: Numerous studies on animal aggression have been conducted throughout the past century, yet much remains to be deciphered regarding regulation of aggression. The effects of the gut microbiota on metabolism, immunity, general health and behavior have been extensively studied. While there have been indications that the microbiota affects risk taking and mating behavior, its mechanisms and pathways are still a mystery. To date, no study has extensively studied the mechanism by which the microbiota affects aggression in mice.

Methods: To test the effects of microbiota composition on aggression in mice, we eliminated different bacterial strains from the mouse gut using different antibiotic treatments. We compared the mice's tendencies toward aggression and characterized their gut microbiota with 16S rRNA gene sequencing of fecal samples. To exclude the antibiotic effect, we also tested the aggressive tendencies in offspring of treated mice. The offspring received compromised microbiomes from the mothers without direct exposure outside the uterus.
Results: The antibiotic treated groups showed significantly lower bacterial diversity and significant differences in bacterial composition. Mice that received antibiotics were also more aggressive than the control group. The offspring of the dams treated with antibiotics were significantly more aggressive than the control group as well; the number of attacks was higher and the latency to attack was shorter.

Conclusions: Our results indicate that the microbiome affects aggression, and that antibiotics increase aggression via the microbiome, but there is likely a critical window for the effect, as well, as seen in *in utero*-exposed mice.

45 The role of Candida Albicans in the development of Crohn's Disease

Sonia Modilevsky[1], Shahar Tel-Paz[1], Neta Shlezinger[2], Shai Bel[1]
Azrieli Faculty of Medicine, Bar-Ilan University[1], The Robert H. Smith Faculty of Agriculture, Foof and Environment, The Hebrew University of Jerusalem[2].

Crohn's disease is an inflammatory bowel disease with unclear etiology, yet it is known that environmental factors play a role in disease development. These environmental factors include diet, lifestyle, and certain microorganisms such as the fungi *Candida Albicans* and the bacterium adherent-invasive *Escherichia Coli* (AIEC), which is not found in healthy individuals. While the role of AIEC in Crohn's disease pathogenesis is not clear, it is thought that this bacterium exploits the inflammatory environment to exacerbate disease. Yet how these microbes affect development of intestinal inflammation and whether they interact in driving this inflammation is not clear. We found that, in healthy mice, neither *Candida Albicans* nor AIEC were able to colonize the intestine, when administered individually. However, AIEC was able to colonize the intestine of healthy mice when administered together with *Candida Albicans*, indicating an interaction between the two microbes. Indeed, our *in vitro* co-culture experiments confirmed that AIEC induced expression of *Candida Albicans* virulence factors. Finally, we found that this AIEC-*Candida Albicans* interaction led to development of intestinal inflammation in predisposed mice. Thus, we show that cross-kingdom interactions between the Crohn's disease-associated AIEC and *Candida Albicans* can contribute to disease development. We speculate that AIEC-induced filamentation of *Candida Albicans* leads to activation on an inflammatory response by the latter, which then facilitates colonization and pathogenesis by the AIEC. Our study provides a possible etiology for development of Crohn's disease.

Mechanical Regulation of the Cytotoxic Activity of Natural Killer Cells

46

Lital Mordechay, Guillaume Le Saux, Avishay Edri, Uzi Hadad, Angel Porgador, Mark Schvartzman
Ben-Gurion University of the Negev, Beer Sheva, Israel.

Mechanosensing has been explored for T cells and B cells and is hypothesized to be a part of their activation mechanism.1–4 In this research, we explored the mechanosensing of the third type of lymphocytes – Natural Killer (NK) cells, by showing that they modulate their immune activity in response to changes in the stiffness of a stimulating surface.5 Interestingly, it was found that the immune response of NK cells is biphasic bell-shaped, and peaks for the surface stiffness of a few hundreds of kPa. This bell-shaped behavior was observed for surfaces functionalized with the activating ligand MHC class I polypeptide-related sequence A (MICA), and in the control surfaces with only physical stimulation, but differences were less pronounced. In addition, it was found that stiffness does not affect uniformly all the cells but increases the size of a small group of extra-active cells, which contributes to the overall activation effect of the entire cell population. We further investigated the clustering of costimulatory adapter protein DAP10 on the NK cell membrane and found the same biphasic bell-shaped dependence on surface stiffness. This novel research is the first systematic study on the mechanosensing of NK cells. Our findings open a pathway to reveal the role of mechanical signaling of immune cells in living systems.

47 SMAD4 association in EMT induction in OSCC cancer

Shiraz Mzlbat[1,2], Hagar Tadmor[2], Aysar Nashef[3], Yasmin Ghantous[3], Imad Abu-elnaaj[1,3]

[1]The Azrieli Faculty of Medicine, Bar-Ilan University, zfat, Israel.

[2] Diabetic and Metabolism Lab, Baruch-Padeh poria Medical center.

[3] Oral and cranio-maxillofacial surgery Department, Baruch-Padeh Poria medical center.

Background: Oral squamous cell carcinoma (OSCC) is a prevalent malignant cancer with poor prognosis. The conventional OSCC management includes surgical resection with radical neck dissection. However, many of the patients are negative for neck lymph node (LN) metastasis, therefore, this highly invasive procedure is likely unnecessary.

Epithelial–mesenchymal transition (EMT) is involved in the progression from non-invasive to metastatic disease. EMT is a reversible dynamic process by which epithelial cells acquire characteristics of mesenchymal cells, migrate and format metastasis. Smad4, a tumor suppressor, is an essential mediator of EMT process and is associated with advanced stage cancer, the presence of LN metastasis and low survival. Yet, Smad4 involvement in the progression of OSCC via EMT and metastasis formation in this cancer is unclear.

Aims: we aim to investigate Smad4 association with EMT markers in order to suggest a personalized non-aggressive treatment platform for OSCC patients.

Methods: oral cancer patients will be recruited at the Oral and Maxillofacial Department at the Baruch Padeh Poria medical center. Tumor and healthy tissues will be collected and subjected to biochemical, histological and genetic analyses. Smad4 and other EMT markers level and expression will be compared with pathology results regarding cancer progression and metastasis presence. Cell culture model of OSCC cells with/without mutation in the Smad4 gene will be used to investigate tumor phenotypes (proliferation-XTT, invasion) and their relation with EMT markers.

Results: current immunohistochemistry staining of EMT markers E-cadherin, SMAD4, and Vimentin present high expression compared with N-cadherin, which may indicate early stage of the disease. Preliminary results of cell culture tumor phenotypes show strong association between phenotypes and Smad4 mutation.

A delayed heat shock response is observed in patient-derived cells expressing pathogenic variants of Nup214

48

Vishakh R Nair[1], Boris Fichtman[1], Volodymyr Chuiko[1], Amnon Harel[1]
1 Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel – 1311502

Abstract
Biallelic missense and frameshift mutations in the *NUP214* gene cause acute febrile encephalopathy (AFE), characterized by severe fever-induced brain damage in affected individuals. Previous work from our laboratory revealed a large increase in the presence of central particles (or "plugs") in the nuclear pore channels of patient cells. In this study, we aim to elucidate the effect of heat shock (HS) stress on control and patient-derived skin fibroblasts, both during HS stress and in the recovery from stress. Our working hypothesis is that delayed passage of nuclear transport cargoes through nuclear pore channels disrupts the cellular heat shock response (HSR) and leads to increased apoptosis and decreased patient cell survival. To test this hypothesis, we followed total cell population growth continuously during HS (2 hours, 43°C) and a subsequent recovery phase (7 days, 37°C). We then performed indirect immunofluorescence microscopy on healthy and patient-derived fibroblasts after subjecting them to the aforementioned HS stress. The fluorescence intensity of anti-Nup214 staining at the nuclear envelope was lower in patient compared to healthy controls and was further reduced upon HS stress. Next, we performed qPCR to measure the mRNA expression levels of Nup214 and Hsp70 in nuclear and cytosolic fractions, during and after HS stress. We plan to determine the expression levels and nucleocytoplasmic distribution of key regulators of the cellular HSR in order to test our hypothesis.

49 The efficacy of SGLT2 inhibitors to delay the onset of diabetes mellitus

Narmeen Ghanayiem[1,2], Offir Ertracht[1], Ron Piran[2] , Tali Reuveni[1], Shaul Atar[1,2,3]

[1]The Cardiovascular Research Laboratory, Research institute, Galilee Medical Center, Nahariya, Israel. [2]The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel. [3]The Cardiology Department, Galilee Medical Center, Nahariya, Israel.

Background: Type 2 Diabetes mellitus (T2DM) is a pandemic, results of overweight, sedentary lifestyle, and metabolic syndrome. The NONcNZO10/LtJ is a mice model of human obesity-induced T2DM and metabolic syndrome. In this strain, T2DM develops within 18-24 weeks of age. Empagliflozin a sodium-glucose cotransporter 2 inhibitor (SGLT2i) enables T2DM patients to urinate excessive glucose and reduce blood glucose levels. We hypothesize that early SGLT2i treatment will delay T2DM onset and its complications in NONcNZO10/LtJ mice.

Methods: Three weeks old NONcNZO10/LtJ mice will be feed with high-fat diet (10-11% fat), on week 8 they will be divided into 2 groups, A. Empagliflozin (10 mg/kg/day) P.O. and B. control un-treated mice. Mice body weight, blood glucose level, motoric and sensory parameters will be measured. 24 hours urine collection, will provide electrolytes composition, protein concentration and creatinine level. Measurements will be repeated at 4 and 12 weeks (before sacrifice). At sacrifice, blood will be collected, and specific organs will be harvested. Finally, we will measure survival rate, neuropathy level, renal function, blood glucagon level and specific organs' histological changes.

Expected results: We expect empagliflozin to increase the T2DM latency, improve or preserve physiological function and tissues' morphological, biochemical and histological parameters.

Importance: We are testing an anti-diabetic agent (empagliflozin) for its preventive efficacy. Testing its effects on healthy pre-diabetic animals may shed light on other biological mechanisms of the drug.
Probable implications: The study might add new indications to SGLT2i treatment that will enable T2DM onset delay, or prevented altogether.

Epigenetic and oncogenic signatures induced by Hepatitis C virus infection following virus eradication.

50

Tom Neimarkas, Michal Werbner, Evgeny Tikhonov, Yaakov Maman and Meital Gal-Tanamy.
The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Abstract: Hepatocellular carcinoma (HCC) is the third leading cause of cancer-related deaths worldwide, with approximately 1 million deaths every year. In large parts of the world, hepatitis C virus (HCV) infection is the main factor of HCC development. Until 2011, the standard treatment for HCV infection was a combination of pegylated interferon-alpha (IFN) and ribavirin with low sustained virological response (SVR), around 50%. Direct-acting antiviral (DAA) drugs are the new generation of HCV treatment with over 90% SVR.
Although IFN and DAA treatments can cause virus eradication, the risk of HCC development still persists after cure, and the exact mechanism is not fully understood. We previously showed that HCV induces epigenetic alterations that correlate with HCC progression. Moreover, following DAAs treatment substantial proportion of the epigenetic signature remains persistent after cure.

Here, we focused on the persistence of the HCV-induced epigenetic signature following IFN treatment. We evaluated the alterations in histone modification and gene expression in HCV infected cells and infected liver samples pre and post treatment, which some achieved SVR and some did not. Our results demonstrate that in HCV infected liver samples that were treated with IFN and did not achieve SVR, the epigenetic signature remains persistent while in the SVR samples most of the alterations were reverted, in contrast to SVR following DAAs treatment. Moreover, in HCV infected cells treated with IFN we observed more persistence of the epigenetic signature in high concentration of IFN treated cells, compared to low IFN. These results suggest that the persistence of the epigenetic signature is affected by the efficiency and duration of the anti-HCV treatment.
These results may shade more light on the mechanisms that maintain cancer risk after removal of the cancer-related etiology and its prevention.

51 Clinical implications of the 100-gram oral glucose tolerance test in the third trimester

Raneen Abu Shqara[1, 2], Shany Or[2], Yifat Wiener[3], Lior Lowenstein[1, 2], Maya Frank Wolf[1, 2]

[1]Department of Obstetrics & Gynecology, Galilee Medical Center, Nahariya, Israel.

[2]Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.

[3]Department of Obstetrics and Gynecology, The Yitzhak Shamir Medical Center, Zerifin, Israel, and Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel.

Purpose: The clinical implications of gestational diabetes mellitus (GDM) diagnosed in the third trimester are not well established and controversy continues regarding the performance of diagnostic tests beyond 28 weeks gestation. The study aimed to evaluate the reproducibility of a late 100-gram oral glucose tolerance test (OGTT) in women with risk factors for GDM, and to examine the impact of late GDM diagnosis on obstetric and neonatal outcomes.

Methods: 372 women who completed late (>29 weeks) 100 gram-OGTT due to suspected fetal macrosomia, polyhydramnios, or a personal risk factor for GDM were included. Women with only one abnormal OGTT value were diagnosed with GDM by abnormal glucose follow-up and analyzed separately. Obstetric and neonatal outcomes were compared between the GDM and the non-GDM groups, and in women who presented one abnormal value – with or without GDM that diagnosed later.

Results: GDM was diagnosed in 85/372 (22.8%) women,59.3%) 35) with one abnormal value on OGTT who were later diagnosed.16.5% (33/200) women who had a normal one-hour 50-gram glucose challenge test at 24-28 weeks (GCT,Glucose challenge test) were finally diagnosed with GDM. Among women with versus without GDM, large for gestational age fetuses (LGA), labor induction and elective cesarean section were more prevalent. Women with one abnormal value who were later diagnosed had a higher rate of LGA and a higher mean birth weight compared to women without abnormal values.

Conclusion: Third trimester OGTT showed significant additive value for GDM diagnosis and should be considered in women with risk factors.

The association between reaching glycemic goals and the prevalence of neonatal complications in women with gestational diabetes mellitus

52

Noa Pinchevsky 1,2, Zohar Nachum3,4, Enav Yefet1,2
Department of Obstetrics and Gynecology, Baruch Padeh Medical Center, Poriya, affiliated with Azrieli Faculty of Medicine, Bar Ilan University, Israel.1 Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.2 Department of Obstetrics and Gynecology, Emek Medical Center, Afula.3 Rappaport faculty of medicine, Haifa.4

Aim & Background: Gestational diabetes mellitus (GDM) is the most common complication during pregnancy. Appropriate glycemic control is mandatory to prevent maternal and neonatal complications. Daily self-monitoring blood glucose is the common method for glycemic control monitoring during GDM. It is customary to use the following glucose targets: 95 mg/dL for pre-prandial values and 130 mg/dL for 90-minute postprandial values. Yet those targets were not studied.
In the present study we examined the association between the appropriate glycemic goals and neonatal complications.
Methods: Retrospective observational cohort study of women with GDM who delivered between 2019 and 2021. Women were divided into groups of good and poor glycemic control, defined as a mean daily glucose of up to 100 mg/dL (N =86) and more than 100 mg/dL (N =50), respectively. The composite outcome was defined as at least one of the following: large for gestational age, neonatal hypoglycemia, cesarean delivery due to fetal distress, neonatal jaundice treated with phototherapy, neonatal hypocalcemia and neonatal hypomagnesemia.
Results: Data from 136 women was analyzed. The study showed no statistical significance in the composite outcome between poor and good glycemic control. The rates of admission to the neonatal intensive care unit, respiratory distress and neonatal hospital stay were higher in the poor glycemic control group.
Conclusion: The use of the mean daily glucose values according to the daily glucose charts is limited by its ability to predict and prevent pregnancy and neonatal complications. Additional markers should be explored.

53 Protective effect of BNT162b2 vaccination on aerobic capacity following mild to moderate SARS-CoV-2 infection: a cross sectional study Israel

Yair Blumberg [1,2] Ph.D, Michael Edelstein [1,2] FFPH, Kamal Abu jabal [1,2] M.D, Ron Golan [1] M.D, Ph.D, Neta Tuvia [1], PhD, Yuval Perets [2] Bs.c, Musa Saad [1] M.D, Tatyana Levinas [1,2] M.D, Dabbah Saleem 1 M.D ,Zeev Israeli [1,2] M.D, Abu Raya Alaa 1 M.T, Gabby Elbaz Greener [2,3] M.D, MHA, Anat Amital [1] M.D, Majdi Halabi [1,2] M.D

[1] Rivka Ziv Medical Center, Zefat, Israel; [2] Azrieli Faculty of Medicine, Bar-Ilan University, Zefat, Israel; [3] Department of Cardiology, Hadassah Medical Center, Jerusalem,

Abstract : Patients previously infected with acute respiratory syndrome coronavirus 2 (SARS-CoV-2) may experience post-acute adverse health outcomes, known as Long COVID. Most reported symptoms are fatigue, headache and dyspnea and myalgia. In addition, reduced aerobic capacity has been demonstrated in both mild and moderate COVID19 patients. It is unknown whether COVID-19 vaccination mitigates against reduced aerobic capacity. Our aim was to compare the aerobic capacity of vaccinated and unvaccinated individuals previously infected with SARS-CoV-2

Methods: Individuals aged 18 to 65 years with laboratory confirmed mild to moderate COVID-19 disease were invited to Ziv Medical Centre, Israel, three months after SARS-CoV-2 infection. We Compared individuals unvaccinated at the time of infection to those vaccinated in terms of aerobic capacity, measured using symptom-limited cardiopulmonary exercise test (CPET).

results: We recruited 28 unvaccinated and 22 vaccinated patients. There were no differences in baseline demographic and PFT parameters. Compared with unvaccinated individuals, those vaccinated had higher $V'O_2$/kg at peak exercise and at the anaerobic threshold. The $V'O_2$/kg peak in the unvaccinated group was 83% of predicted vs 100% in the vaccinated ($p<0.002$); In the unvaccinated group, 14/28 subjects (50%) had a $V'O_2$ peak < 80% of predicted vs 2/22 (9%) among those vaccinated.

Conclusion: Vaccinated individuals had significantly better exercise performance. Compared with vaccinated individuals, a higher proportion of those unvaccinated performed substantially worse than expected on CPET. These results suggest vaccination at the time of infection is associated with better aerobic capacity following SARS-CoV-2 infection

Cracking the genomic code underlies the targeting RAG endonuclease activity

54

Rawan Foqara[1], Hadas Sibony-Benyamini[1] Yaakov Maman[1]
[1]Laboratory for Genome Instability and Cancer, Azrieli Faculty of Medicine, Bar-Ilan University.

In early stages of lymphocytes development, the immune repertoire is generated by massive editing of the antigen receptor (AgR) loci. This process - termed **V(D)J recombination** - is initiated by the **RAG endonuclease** that introduces DNA double strand breaks (DSBs) at **recombination signal sequences (RSS)** that are scattered along the variable regions of the loci.

The canonical metric of RSS functionality (i.e the propensity to support RAG-mediated cleavage and RAG binding) is based on the similarity to the RSS consensus sequence. However, RSSs show a considerable level of diversity, and AgR loci from different lymphocyte's lineages demonstrate distinct consensus sequence. Moreover, while millions of potential RAG sites, termed cryptic RSSs (cRSSs) are found throughout the genome outside of the AgR, only small minority of these cRSSs have been demonstrated to support RAG activity. Notably, cleavage of such functional cRSSs is the basis of RAG-mediated tumorigenesis.

Our preliminary analysis showed that functional cRSSs cannot be distinguished from adjacent non-functional cRSSs by the current sequence-based metric.

This ambiguity regarding the genomic language of RSSs and cRSSs impedes our understanding of RAG physiological activity during V(D)J recombination, as well as its role in cancer.

We hypothesize that the stochasticity of the RSS sequence harbors yet uncharacterized linguistic rules for both RAG-mediated cleavage and RAG-binding, that can be revealed by direct measure of these functions.

To test this hypothesis, we designed a sensitive, high-throughput assay based on a library of randomized RSS-like sequences inserted into a recombination construct, where GFP expression is initiated upon successful recombination.

Integrating this library into cells allows us to sort for functional cRSSs in different times following RAG induction to obtain a semi-quantitative measure of cRSS functionality. This measure will be used to build a sequence-based prediction model for functionality.

55 Adipose-Derived Stem Cells Modulate Endometrial Polyp Fibroblasts

Reema Fadoul[1,2], Tharwat Haj Khalil[1], Idan Redenski[1,2], Daniel Oren[1,2], Asaf Zigron,[1,2], Avishalom Sharon[4], Amiel A. Dror[1,3], Mizied Falah[1], Samer Srouji,[1,2]

[1]Department of Oral and Maxillofacial Surgery, Galilee College of Dental Sciences, Galilee Medical Center, Nahariya, Israel

[2]The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

[3]Department of Otolaryngology-Head and Neck Surgery, Galilee Medical Center, Nahariya, Israel

[4]Department of Obstetrics and Gynecology, Galilee Medical Center, Nahariya, Israel

Endometrial polyps (EPs) are benign overgrowths of the endometrium, with the potential to cause severe medical complications such as chronic inflammation or infertility. Dysfunctional endometrial fibroblasts may be a critical component that cause the development of these polyps. Adipose-derived mesenchymal stromal cells (ASCs) are at the focus of modern medicine, as key modulators of tissue homeostasis, inflammation and tissue repair, rendering them prime candidate agents for tissue regeneration and cell-based therapies.

In the current work, endometrial polyps were isolated from patients admitted to the OB/GYN department at the Galilee Medical Center. Endometrial polyps fibroblasts (EPFs) were isolated from these specimens and characterized for their cellular markers and phenotype. Concomitantly, ASCs were isolated from healthy patients. The effect of EPF- and ASC-conditioned media (CM) on polyp-derived fibroblasts was evaluated, by utilizing 2D and 3D assays. Moreover, the expression of matrix-related gene expression was evaluated in cultures exposed to conditioned media. Herein, EPFs exposed to ASC-CM exhibited reduced migration, invasion, contraction of hydrogels, and extracellular matrix deposition, compared to those exposed to EPF-CM. Altogether, the current work suggests a modulating effect ASCs may have on fibroblasts involved in forming endometrial polyps. These results may serve as a basis for the development of conservative treatment strategies aimed at treating severe cases of EPs.

cfDNA as a factor affecting cell-to-cell communication

56

Mina Roth[1], Gidi Baum[1], Milana Frenkel-Morgenstern[1]
[1]Cancer Genomics and BioComputing of Complex Disease lab, Azrieli Faculty of Medicine, Bar-Ilan University

Background: Circulating cell-free DNA (cfDNA) are DNA fragments (166bp, 322bp, 498bp≈) released from cancer as well as normal cells. cfDNA investigation is an important field in cancer research, with an aim to develop an early diagnostic tool that is less invasive. On a different topic, cell-to-cell communication is affected by the intercellular environment that includes different molecules and many different components. Our research hypothesis is that cfDNA is not only a byproduct, but is also involved in intercellular communication and may play an important role in cancer development and progression.

Objectives: Our study aims to investigate the involvement of cfDNA in cell-to-cell communication by means of extracting cfDNA from serum or media of living cancer cells and measuring whether it can bind to or enter other cells.

Methods: Developing a protocol for cfDNA extraction in vitro that yields the sufficient amount of cfDNA that is required for the following steps in the research. Finding the best methods for cfDNA labeling in order to follow its uptake into or binding to cells.

Results: A protocol for cfDNA extraction from the medium of in vitro cultured cells has been developed, this protocol is cheaper and has a better yield than the previous one used in the lab. Different cfDNA labeling methods have been tested and multiple results have been compared.

Conclusions: This study is still in its early stages, but so far it has shown consistent results. The cfDNA was extracted in vitro and it will be followed by the labeling and monitoring in the cells by different visualization techniques.

57 Neuronal SMC3 regulates weight, body composition, and hormonal balance in parallel with sex-dependant effects on anxiety behavior

Natalia Saleev, Evan Elliott.
The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Smc3 is a major component of cohesin complex that regulates higher-order chromatin organization and gene expression. Human genetic studies reveal that de novo mutations in SMC3 gene, found in patients with Cornelia de Lange syndrome (CdLs). This syndrome characterized by intellectual disabilities, and behavioral patterns as self-injury. Nonetheless, little is known about the exact role of SMC3 in neuronal maintenance and gene expression especially in adulthood.
This study aimed to determine the role of SMC3 in adulthood brain, using *in-vivo* models of adulthood excitatory neuron SMC3 knockout in male and female mice. Neuron-specific SMC3 knockout mice displayed dysregulated anxiety-like behavior and self-injury in males and females compared to wild-type littermates. Of interest, female knockouts displayed less anxiety while males displayed more anxiety, although both displayed self-injury, a known phenotype in the human condition. In parallel, significant metabolic changes were displayed in both male and female mice, including overweight phenotype, loss of muscle mass, differences at respiratory exchange, heat production and hormonal changes after knockout of SMC3 gene in excitatory neuron cells of adult brain. RNA-seq in the hypothalamus reveals changes in several peptides that moderate proper hormonal balance. This is interesting, considering reports of adult onset obesity in a subset of individuals with CdLs.

This knowledge may provide a novel basis for potential treatment or improvement of quality of life for diagnosed people with CdLs or people who diagnosed with side effects of instable SMC3 function.

Adipose and Blood Derived Substances for Soft and Hard Tissue Bridging

58

Shadi Daoud[1,2], Asaf Zigron[1,2], Daniel Oren[1], Idan Redenski[1], Fares Kablan[1], Samer Srouji[1,2]

[1] Galilee College of Dental Sciences, Oral and Maxillofacial Surgery Department, Galilee Medical Center, Nahariya, Israel

[2] The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel

Tissue engineering, aimed at producing tissue replacements, has provided promising alternatives for regenerating soft and hard tissue defects. Adipose tissues and venous blood, both are easily accessible for harvest, are an excellent source of mesenchymal stromal cells (MSCs) and growth factors. Yet, extensive clinical use of these materials as biological substances for tissue repair has yet to be reported. Both adipose tissues and venous blood were obtained from 350-400 gr Sprague-Dawley rats. Adipose tissue was condensed using a custom-made apparatus to obtain membranes used for a soft tissue implantation model. Venous blood was centrifuged and to obtained concentrated growth factors, used to enhance bone tissue repair. Ex-vivo assessment of samples was employed to evaluate the effect of adipose and blood derived products on tissue repair after implantation. *In vivo* implantation of rat membranes indicated rapid perfusion and integration, and blood-derived growth factors enhanced bone deposition around titanium implants. Preliminary results indicated that adipose and blood derived products may serve as an easily accessible means of producing biologically-induced constructs for tissue repair.

59 Is Zinc Finger domain involved in protein-protein interactions in Eco1?

Shada Shawahni , Avi Matityahu , Itay Onn
The Azrieli Faculty of Medicine, Bar-Ilan University , Henrietta szold street , Safed .

The Eco1 acetyltransferase is a vital factor of Sister chromatid cohesion (SCC), a fundamental process in cells that is essential for maintaining genome integrity. Eco1 harbors a conserved C2H2-type zinc finger domain that has been implicated in Eco1 function. Zinc finger is a common motif proteins in all kingdoms of life, mainly responsible for the interactions with nucleic acids. However, we suspected that Eco1 zinc finger mediates interaction with proteins, a less common non-canonical function of this motif. We aimed to define Eco1 zinc finger interactome. We used two complementary experimental approaches to pull down Eco1 interacting proteins and identified them by mass-spectrometry analysis. We identified 53 proteins that were common to both strategies and their precipitation depends on the zinc finger. Four proteins were selected for validation and further analysis. In order to better understand the molecular basis of Eco1 interactions, we aim to solve the atomic structure of Eco1 zinc finger. We expressed in E. coli and purified to apparent homogeneity the N'-terminal fragment of Eco1 that includes the zinc finger domain. However, under these conditions the protein aggregates and, therefore, is unsuitable for crystallography. In an effort to stabilze it we are co-expressing it with its interacting partner PCNA. If successful, understanding the structural basis protein-protein interaction mediated by zinc fingers would advance our understanding of Eco1 interacting specificity. Altogether, this work will enhance understanding of how Eco1 regulates cohesin and is involved in maintaining genome stability.

Selective peptide modulator of Mfn1-Fis1 protein-protein interaction and its effect on mitochondrial dynamics and cardiovascular disease.

60

Hellena Shumaff[1], Nir Qvit[1]
The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Henrietta Szold St. 8, POB 1589, Safed, Israel.

Mitochondria are dynamic organelles that constantly alter their shape in response to changes in cellular physiological conditions. Mitochondrial dynamics shift between fission and fusion, which are closely associated with mitochondrial function. An irregular balance between fission and fusion has been observed in several human diseases, including cardiovascular diseases, and impacts a broad scale of cellular functions. Mitochondria fusion is regulated by Mitofusins (Mfn1 & Mfn2) and Opa1, while Drp1 is responsible for mitochondrial fission. Another essential protein is Fis1; it is responsible for recruiting Drp1 and initiating mitochondrial fission. Newer reports reported that Fis1 could inhibit the fusion process by binding to the fusion proteins and shifting the balance towards fission.

In this project, we are studying the interactions between Fis1 and Mfn1 through various *in-vitro* and *in-vivo* assays to determine the nature of interaction and its effect on mitochondrial dynamics and CVD. We have successfully expressed and purified both proteins through bacterial origin, and both proteins are stable for at least 16 hours at different temperatures. Our preliminary results also found that in different concentrations, Mfn1 and Fis1 have a strong protein-protein interaction (PPI) *in-vitro* (K_D 31μM) In addition, we designed and synthesized peptide modulators for this PPI.

Finally, we wish to test the effect of the peptides on the PPI in *in-vitro* and *in-vivo* assays, such as myoblast cell assays and animal models to determine if there are any inhibitions or positive interactions and what the impact is on mitochondrial dynamics and heart diseases.

61 Mff interaction with Mid51 as a target for mitochondrial morphology manipulation

Shmuel Silnitsky, Nir Qvit
Bar Ilan University, The Azrieli faculty of medicine

Cardiovascular diseases (CVD's) and myocardial infraction (MI, aka heart attack) are the leading cause of death globally, taking about 18 million lives yearly and are also a major economic burden, with expenditures in the US alone of $216 billion yearly.
Mitochondria provide cellular energy to the cell for survival and functioning. Cardiomyocytes in particular have high energy requirements, so mitochondria's nature to constantly fuse and divide and the ATP supply accompanied by those actions provides a source to control its life and energy production and thus the fate of the cell, especially during MI.
Significant proteins responsible for the mitochondria's division (fission) are Dynamin related protein 1 (Drp1), and two of its main recruiters at the mitochondrial outer membrane (MOM): Mitochondrial fission factor (Mff), and Mitochondrial dynamics protein of 51 kDa (Mid51). Mff/Mid51 interaction (PPI) seems to result in mitochondrial fission. Interestingly it has been proven that in MI excess fission occurs, resulting in energy loss and cell death.
As an effort to restore balance to mitochondria during MI, we wish to interrupt Mff /Mid51 PPI. Herein the proteins were expressed, purified and their PPI was measured in-vitro (K_D = 207 nM). Based on the results and proteins structural knowledge, we developed specific peptides to interrupt with Mff/ Mid51 PPI. The peptides will then be examined in cell culture and in MI animal model (in vivo). We predicted that the novel peptides would regulate Mff/Mid51 PPI and therefore will be protective in animal model of MI.

Mapping the crosstalk between pancreatic endocrine cells: The intra-islet theory debunked

62

Worood Sirhan[1], Daljeet Kaur[1], Assaf Malka[1], Offir Ertracht[2], Shaul Atar[2], and Ron Piran[1].

[1]The Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel. [2]Eliachar Research Laboratory, Galilee Medical Center, Nahariya, Israel.

Abstract

Aim & Background: In Diabetes, blood glucose levels increase above physiological concentrations. To achieve normoglycemia glucagon and insulin are secreted from the pancreatic endocrine α and β-cells, effecting glucose release or absorption by target tissues. Previous studies suggested that intra-islet glucagon and insulin secretion modulate and regulate the function of each other. This intra-islet theory states that glucagon secretion is suppressed when insulin is secreted, and that insulin secretion is modulated when glucagon is secreted. To achieve reciprocal influence on insulin and glucagon secretion, α- and β-cells must have receptors for insulin and glucagon respectively.

Methods: To re-evaluate the intra-islet theory, we used glucagon receptor immunofluorescence staining in mice and looked for positive cells in the pancreatic islets. Surprisingly, we did not find glucagon receptor positive β-cells. To test Glucagon and Insulin reciprocal effects, liver-obstructed and sham-operated rats were tested for their response to exogenous glucagon and insulin on endogenous insulin and glucagon secretion, respectively.

Results& Conclusion: Our preliminary results strengthen our hypothesis that the intra-islet theory is incorrect. Glucagon and insulin influence each other by acting on the liver to absorb or release glucose to and from hepatic glycogen reservoirs. Thus, α and β-cells simply secrete their hormones as a response to blood glucose levels.

63 Field-Effect Biosensing Technology, a promise to unravel Biomolecular Interactions

Surya Sukumaran1*, Yana Lerner[1*], Mei-Sze Chua[2], Samuel K So[2], and Nir Qvit[1]

[1]The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Henrietta Szold St. 8, POB 1589, Safed, Israel.

[2]Asian Liver Center, Department of Surgery, Stanford University School of Medicine, Stanford, CA 94305, USA. Surgery and Asian Liver Center, 300 Pasteur Drive, H3680, Stanford CA 94305-5655, USA.

*These authors contributed equally

Abstract

Aim & Background: Biomolecular interactions play versatile roles in numerous cellular processes by regulating and coordinating functionally relevant biological events. Biomolecules are fundamental building blocks of living beings which assembles into complex networks in biosystems to synchronize a myriad of life events. Proteins typically utilize complex interactome networks to carry out their functions; hence it is mandatory to evaluate such interactions to unravel their importance in cells at both cellular and organism levels. Toward this goal, we introduce a rapidly emerging, field-effect biosensing technology (FEB), to determine specific biomolecular interactions using heat shock protein 90 (Hsp90) and cell division cycle 37 (Cdc37) as experimental model.

Methods: FEB is a benchtop, label-free, and reliable biomolecular detection technique used to determine specific interactions by using high-quality electronic-based biosensors. As a proof of concept, the protein-protein interaction (PPI) between Hsp90 and Cdc37 was elucidated. The FEB technology measures the electric current through the graphene biosensor to which the binding targets are immobilized. Interactions between the immobilized protein (Hsp90) and the analyte (Cdc37) result in alterations in current that are monitored in real-time, enabling accurate kinetic measurements.

Results and Conclusion: The chaperone-kinase pathway connecting Hsp90 and Cdc37 is hyper-activated in multiple malignancies therefore, it is a promising therapeutic target in tumor biology. In the present study FEB detected a strong PPI between Hsp90 and Cdc37 showing consistent KD values of 0.014 µM, 0.053 µM, and 0.072 µM respectively. In summary, we successfully demonstrated the PPI between Hsp90/Cdc37 by using the FEB technology and validated their interaction using ITC, providing promising data to support FEB as an innovative, cost-effective alternative method for PPI detection.

Development of a specific inhibitor for the MFN1/Sirt1 interaction, and investigation of its effect on mitochondrial processes and cardiovascular disease.

64

Nora Syadi[1], Samar Gani[1], Nir Qvit[1]
The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Henrietta Szold St. 8, POB 1589, Safed, Israel.

Abstract
Mitochondrial dynamics requires proteins involved in fission and fusion. Mitofusin1 (Mfn1) control fusion of the mitochondrial outer membrane. Sirtuin1 (Sirt1) hat function in the cellular response to inflammatory, metabolic, and oxidative stressors. Sirt1 has a role in NAD+ dependent deacetylation of histones. Moreover, Sirt1 de-acetylate Mfn1 protein and thus up-regulates Mfn1 protein stability.
Our research goal is to examine the effect of Mfn1/Sirt1 interaction on mitochondrial dynamics especially in cardiovascular disease using specific peptide inhibitors.
We investigated the interaction between the proteins by Field Effect Biosensing (FEB): AGILE R-100. Using the L-align software, we identified the protein regimes that are comparable in both sirt1 and mfn1 since these similar regimes can interact with one another. These overlapping regimes were used to select small peptides. The peptides were created using solid-phase peptide synthesis (SPPS), and their purity was assessed using reverse-phase high-pressure liquid chromatography (RPHPLC). To determine the molecular weight, matrix-assisted laser desorption ionization (MALDI) mass spectrometry was carried out. Then we examined the impact of the special designed peptide on H9C2 cell line derived from embryonic rat heart tissue by XTT-assay to measure cellular metabolic activity as an indicator of cell viability, proliferation and cytotoxicity.

We successfully investigated the interaction between Mfn1 and Sirt1. The molecular weight of the desired peptide, as determined by MALDI analysis was 2404m/z, which is identical to the calculated of the CVP127a peptide, which we successfully produced with 100 percent purity. We were able to show that CVP127a had a protective effect by using the XTT assay. Additionally, the vitality of the treated cells was 95% as opposed to only 60% for the untreated cells.
The results show that the viability of the cells increased in the presence of this peptide. We would also like to examine the peptides affect in a myocardial infarction (MI, aka heart attack) model in rats.

65 The impact of BMI on the outcome of lymphoma treatment

Saleem Jacky[2], Kashlikov Marat[1], Tarabeh Khalil[1], Dally Najib[1,2]
Hematology Institute and Blood Bank[1], Ziv Medical Center, Bar–Ilan University, Azrieli Faculty of Medicine[2]

Background: Lymphomas comprise a diverse group of neoplasms derived from B cells, T cells, or Nk cells, associated with variable clinical presentation.
The main treatment of lymphoma is chemotherapy, though radiation and targeted therapy may be combined according to the type and stage of the disease. Chemotherapy dosage is determined according to body surface area (BSA) thus patients with different body mass indexes (BMI) receive different doses. Patients with high BMI values may not receive the full chemotherapeutic dose due to concerns regarding side effects.

Objective: The aim of the current study was to evaluate the effect of BMI on treatment outcomes of lymphoma patients.

Patients and methods: A retrospective study was conducted to assess the relationship between BMI and treatment outcomes in patients suffering from the most common types of lymphoma: Hodgkin's disease, follicular lymphoma and diffuse large B cell lymphoma. Data was collected from the relevant lymphoma patients' treatment charts at the hematology outpatient clinic at Ziv Medical Center. 117 patients were included in the study; patients were categorized according to BMI. Treatment outcomes were correlated to BMI values.

Results: No difference was found between different BMI groups and treatment outcomes ($P = 0.552$), but patients with higher BMI values received significantly more lines of chemotherapy ($P = 0.011$).
Conclusion: Lymphoma patients with high BMI values do not have worse treatment outcomes compared to patients with normal BMI values, but higher BMI value was strongly correlated to more lines of chemotherapy.

The Significance of Laboratory Inflammatory Indices in Children with Acute Appendicitis

66

Ali Tarabia[1,2], Doua Bakry[2,3], Bian Hino[2,3] & Yulevich Alon[1,3]
1. Department of Pediatric Surgery, Ziv Medical Center, Safed, Israel.
2. Department of Pediatrics, Ziv Medical Center, Safed, Israel.
3. Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel.

Aim of the Study
Acute appendicitis is the most common urgent non-traumatic surgical disease in children. The study aimed to recognize a correlation between white-blood-cell count (WBC), neutrophil cell proportion, and C-reactive protein (CRP) – inflammation indices – and the course of acute appendicitis.
Methods
In a retrospective study, we collected children diagnosed with acute appendicitis between 1.1.2015 and 31.12.2019 at Ziv Medical Center, Israel. Clinical, laboratory, imaging, appendectomy, and histological data were collected and analyzed. Children with a peri-appendicular abscess treated conservatively without early operation were not studied, as were not children with suspected appendicitis.
Main Results
The study included 300 children that underwent appendectomy, 65% boys ($p<0.05$). Their average age was 11-years-old (3-18). Thirty-eight percent of children arrived at the hospital within 24 hours since abdominal pain onset, and 45% after 36 hours. The P-value for WBC, neutrophil percent, and CRP in patients arriving at the hospital after 35 hours or later was 0.03, 0.02, and 0.01, respectively. Abdominal ultrasound was diagnostic in 84% of patients and computerized abdominal tomography in 100%, done in 97% and 10% of patients, respectively. The operative results matched histological examination, with 7% uninflamed appendix. Appendectomies and postoperative courses were uneventful.
Conclusion
All inflammation indices rose significantly 1.5 days since acute appendicitis onset in our study population. Hence, acute appendicitis should be highly suspected in those children.
These tests do not help diagnose or rule out acute appendicitis in children with the shorter unclear disease.

67 Sex-Dependent and cell-type specific effects of autism associated gene CHD8 on behavior during adulthood

Weisberg Orly, Getsleter Dmitriy, Elliott Evan
1Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel

The etiology of autism includes both genetic and an environmental component. CHD8 was identified as one of the top autism-associated genes. CHD8 is a transcriptional regulator expressed in wide cell types and involved in multiple cell pathways. It is not clear if CHD8 is important only during development or has roles in the adult brain. In addition, it is not clear if there are different roles for CHD8 in different sexes or cell types.
The aim of this study was to create conditional deletion of CHD8 in excitatory neurons and study how CHD8 reduction influence behavior in adult male and female mice. using behavioral tests we found females CHD8 cKO mice, moved less in an open arena, while motor function was normal in rotarod. In addition, grooming was increased significantly. Therefore, it is possible that the decrease in movement was due to an increase in time that the mice were doing repetitive movements while stationary. Females cKO mice have increase in contextual fear memory and tendency in cued fear memory. Males cKO mice displayed normal activity in open field however in marble burying their activity was decreased significantly. In contextual fear conditioning test, increased freezing was significant only in last two minutes. Anxiety-like behavior, measured by elevated plus maze and dark light, was normal for both sexes. Sociability and novelty tests were normal in females while male cKO mice showed alteration in novelty but not in sociability. We propose that CHD8 expression in excitatory neurons has novel sex dependent effects.

Beyond endonuclease-understanding the effect of RAG1 on chromatin

68

Wessal Hanout [#], Hadas Sibony-Benyamini [#], Yaakov Maman
Faculty of Medicine in the Galilee, Bar Ilan University, Israel
# Equal contribution

Abstract
The RAG endonuclease initiates V(D)J recombination at the antigen receptor (AgR) loci by introducing DNA breaks adjacent to the V,D and J segments constituting the variable of the loci. RAG ability to induce DNA breakage comprises harmful potential that necessitates a tight regulation to ensure the tunneling of its activity exclusively to the AgR. Our recent studies suggested that RAG is doubly anchored to chromatin. The first anchor is mediated by interaction with promoter's histone mark - H3K4me3. The second is yet unknown, but our data points out that this interaction is mediated by the RAG RING domain - that confers E3 Ubiquitin ligase functionality - and marked H3K27Ac as a candidate for this interaction. Other studies showed that RAG – through its RING domain – can interact and ubiquitylate Histone 3 (H3). This RAG-mediated H3 ubiquitylation on one hand, and the previously demonstrated ability of H3-Ub to facilitate H3 acetylation and promoter activation on the other hand, provides a possible link between RAG localization and activation. In this project we sought to explore this link, and to identify the effect of RAG-mediated ubiquitylation on chromatin. Our preliminary results – obtained by mapping of chromatin accessibility and nucleosome positioning in WT pre-B cells and cells with RAG mutant that lacks the E3-ligase functionality, demonstrate that RAG can shape the structure of the chromatin in its binding regions, suggesting a novel role of RAG as a chromatin modulator, that is dependent on its E3-ligase functionality, but independent of its endonuclease activity. Such function may have a role both in its targeting to the antigen receptor genes, and its off-target activity in lymphoid cancers.

69 Effect of COVID-19 pandemic on maternal obesity and related pregnancy complications

Tomer Yehuda-Fishman[2], Lior Lowenstein[1,2], Maya Frank Wolf[1,2], Inshirah Sgayer[1,2]

[1] Department of Obstetrics and Gynecology, Galilee Medical Center, Nahariya, Israel

[2] Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel

Objectives

The COVID-19 pandemic has had profound effects on different aspects of pregnant women lives. This study aimed to investigate (1) maternal obesity rate and weight gain during pregnancy in laboring women after the intensive first year of COVID-19 pandemic as compared to the era before the pandemic and (2) and to compare obesity-related pregnancy morbidity including gestational diabetes (GDM), hypertensive disorders of pregnancy (HDP) and macrosomia between the two periods.

Methods

The study group consisted of pregnant women who had delivered between 01.12.2020 to 28.02.2021. This group of women have spent their pregnancy course under several lockdowns. The control group consisted of patients who had delivered between 01.12.2019 to 28.02.2019 before COVID-19 outbreak in Israel.

Results

The weight gain during pregnancy was similar in the study versus control group (13.19 versus 12.79 kg, p=0.231). The subgroup of patients with preconceptional obesity (BMI 30-34.99) of the study group have had a higher weight gain during pregnancy when compared with the same sub-group of the control group (11.16 versus 8.69 kg, respectively, p=0.04). We found a lower GDM rate and HDP in the study versus control group (6.6% versus 10.0%, p=0.005) and (2.2% versus 3.9%, respectively, p=0.02).

Conclusions

In accordance with previous studies, some groups of patients have had a higher weight gain during the pandemic. Furthermore, lower rates of GDM and HDP during the pandemic could be related to a reduction of care-seeking due to fear of infection or restrictions on personal mobility. Strategies for antenatal care and screening during pandemic should be revised.

Investigating Mid51 and Fis1 upregulation as overexpression mitochondria fission response in cardiovascular diseases

70

Zerihun Mulate[1] and Qvit Nir[1*]
[1]The Azrieli Faculty of Medicine in the Galilee, Bar-Ilan University, Safed 1311502, Israel;
* Correspondence: nir.qvit@biu.ac.il

Abstract
Mitochondria are dynamic organelles that rely on a delicate balance of opposite processes between fission and fusion. The imbalance in mitochondrial dynamics leads to cell death resulting in the progression of numerous types of cardiovascular diseases (CVDs). Mitochondrial fission 1 (Fis1) and mitochondrial dynamic (Mid51) proteins are regulate mitochondrial fission and dynamics. Furthermore, it has been demonstrated that overexpression of fission leads to cell damage and stress mitochondria functions, as a means of preparing damaged mitochondria for removal via apoptosis. Therefore, our aim was to explore new regulatory mechanisms for the interaction between Mid51/Fis1 that can potentially be therapeutic implications and addressing them by designing novel peptides that improve mitochondrial dynamics, which can be leads for new treatment for CVDs. Thus, we designed peptides that modulate Fis1 and Mid51 protein-protein interaction (PPI) against therapeutic treatment in cardiomyocyte cell function. First, we studied the Mid51/Fis1 PPI and identify high binding affinity (K_D = 0.0539 ± 0.0013 μM), which was not reported before. Then, we developed peptides that modulate Mid51/Fis1 PPI and demonstrated the protein-peptide binding. Next based on the *in vitro* studies we identified CVP-239a, CVP-240a, CVP-241a, CVP-242a, CVP-243a (aka P110) peptides for further cells and animals studies. Meanwhile, we investigated the cardio protective effect in cardiomyocytes cell culture (H9c2 cells) using CVP-240a and CVP-243a peptides. Overall, the results showed highly significant difference in cell viability. In conclusion, we developed peptides that showed promising approach to inhibit the interaction between Mid51/Fis1 proteins as overexpression responses in CVD.

71 The role of IL-1β in protection from Salmonella infection

Mor Zigdon, Shai Bel.
Bar Ilan university

The foodborne pathogen *Salmonella* typhimurium (*S*. Tm) has evolved to manipulate the relationship between the host and its microbiota. By triggering an inflammatory response, *S*. Tm alters the intestinal environment to support its growth and the killing of its competitors, the microbiota. Specifically, *S*. Tm infection leads to neutrophil recruitment to the colon, which kills short-chain fatty acids (SCFA)-producing commensal bacteria, forcing the host to switch energy production from β- oxidation of SCFA, to glycolysis. This metabolic manipulation of the host by *S*. Tm leads to oxidation of the gut, as glycolysis does not consume oxygen, further harming resident microbes which are mostly obligate anaerobes. Yet exactly how *S*. Tm manipulates the host immune system to trigger this cascade is not known. We found that mice lacking the proinflammatory cytokine interleukin 1β (IL-1$\beta^{-/-}$) fail to recruit neutrophils to the gut during *S*. Tm infection. Unexpectedly, this immune deficiency protected IL-1$\beta^{-/-}$ mice from infection, reducing mortality, tissue damage, and impeding *S*. Tm expansion. We also found that infected IL-1$\beta^{-/-}$ mice did not carry out a transcriptional program that indicates metabolic switching to glycolysis. Indeed, hypoxia levels in infected IL-1$\beta^{-/-}$ were significantly higher than infected wild type mice. This implies that IL-1β is a central component of host manipulation by *S*. Tm which allows the pathogen to outcompete the microbiota.

Effect of Probiotics on Different C. difficile strains in a Mouse Model

72

Zohar Hamo[1], Hen Ninio[2], Maya Azrad[2], Halim Roshrosh[2], Dana Binyamin[1], Mor Zigdon[1] Avi peretz[1,2*]
Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel[1]
[2]Clinical Microbiology Laboratory, Baruch Padeh Medical Center, Poriya, Israel

Background: *Clostridioides difficile* (*C. difficile*) represents the leading cause of nosocomial diarrhea. In recent years, there has been an increased interest in the differences between *C. difficile* strain characteristics, and in the possible correlation between these differences correlate with disease severity. Since one risk factor for *C. difficile* infection (CDI) is microbiome disruption, supplemental probiotics are often recommended as microbiota-targeted therapies to improve CDI symptoms. However, evidence for their efficacy is limited. This study aimed to examine the effects of probiotics on different strain type (ST) strains of *C.difficile* in a mouse CDI model.

Methods: Mice were exposed to an antibiotics cocktail for 4 days. Then, they were challenged with different *C. difficile* ST strains and 24 h later, each mouse group was treated with *Bifidobacterium bifidum, Lactobacillus paracasei* or *Lactobacillus acidophilus* for 4 days. Stool samples were then collected for toxin concentration measurements and colonic tissues were assessed for intestinal histopathology by haematoxylin and eosin (H&E) staining.

Results: The different strains of probiotics reduced toxin secretion by *C. difficile,* with *L. paracasei* imparting the greatest effect. Similarly, the colonic morphology of mice treated with the *L. paracasei* showed the greatest improvement. Our results demonstrated that the effect of probiotics, especially *L. paracasei*, against CDI is mediated via a decrease of toxin secretion, which reduce the morphologic damage in the colon epithelium. Therefore, probiotics could be a promising agent in the management of CDI.

73 Thyroid gland dysfunction and covid-19 severity - Is there a correlation?

Nidal El-Khatib[1], Raed Farhat[1], Asakly Majd[1], Alaa Safiy[1], Adi Sharabi-Nov[2], Yaniv Avraham[1] and Shlomo Merchavi[1]

[1]Department of ENT, Ziv Medical Center, Safed, Israel

[2]Statistics Unit, Ziv Medical Center, Safed, Israel and Tel-Hai academic collage, Tel-Hai, Israel

Background: The outbreak of the coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV2) from December 2019 has caused significant a health burden across the world. While this coronavirus disease has the potential to cause multi-organ effects including endocrine disorders, thyroid diseases were not mentioned specifically.

It had been suggested that COVID- 19 can cause thyroid gland dysfunction by direct or indirect effect. Since the relationship between thyroid hormones levels and mortality is well established, this study examined the correlation between thyroid hormone level, disease severity and mortality in hospitalized COVID 19 patients.

Aim - To establish whether Covid-19 could cause thyroid gland dysfunction, and whether there is an association between thyroid hormones dysfunction severity and mortality rate.

Methods – In a retrospective study. 458 Covid-19 patients hospitalized in the Ziv Medical Center were included. Demographic and clinical data were collected from patients' electronic medical records. LR multivariate logistic regression model was used to assess the odds ratios (ORs) and 95% confidence interval (CI) for the correlation between mortality and levels of T3, T4 and TSH.

Results - lower levels of T3, T4 and TSH were more common in patients with severe or critical disease compared to patients defined with mild or moderate disease ($p<0.001$). In a multivariate logistic regression, T4 out of normal range was associated with mortality OR = 5.87 (95% CI: 2.44-14.13, $p<0.001$), TSH out of normal range OR = 3.55 (95% CI: 1.59-7.93, $p<0.01$), hospitalization in the Covid-19 ICU OR = 7.36 (95% CI 2.78-19.44, <0.001), length of hospitalization OR = 1.12 (95% CI: 1.07-1.18, $p<0.001$) and age of patient OR = 1.08 (95% CI: 1.05-1.12, $p<0.001$). No significant correlation was detected with T3 and Gender.

Conclusions – COVID-19 could cause a thyroid gland hormone disorder, and T4 and TSH are strongly associated with severity and mortality of Covid-19 hospitalized patients. T4 and TSH levels may serve as additional tools for early prognosis of Covid-19 severity and mortality.

Pembrolizumab as a first line therapy in a patient with extensive Mucoepidermoid salivary gland carcinoma. A complete clinical, radiological and pathological response. A very specific case

74

Raed Farhat[1], Noam Asna[1] ,Nidal El-Khatib[1], Asakly Majd[1], Alaa Safiy[1], Yaniv Avraham[1] and Shlomo Merchavi[1]
[1]Department of ENT, Ziv Medical Center, Safed, Israel

Abstract
Background: Patients with advanced salivary gland malignancies (SGCs) have few therapy options. Although results from newly published trials suggest that checkpoint inhibition may be useful in a subgroup of patients, there are no clear criteria for PD-L1 score in SGCs. Chemotherapy benets were observed to be limited, with a dismal prognosis in unresectable and high-grade SGC.
Immunotherapies have demonstrated extraordinary ecacy in a variety of cancers, including non-small cell lung cancer and malignant melanoma. Anti-PD-1 antibody pembrolizumab has been shown to have potent anti-tumor action in a number of clinical trials.
Case presentation: We report a unique case of advanced high grade mucoepidermoid carcinoma of the parotid salivary gland after Pembrolizumab treatment as a rst line therapy.
The tumor was downstaged as a result of the pembrolizumab treatment, allowing for a successful surgical excision with no facial nerve sacrice and no major neoadjuvant treatment adverse effects, and the nal specimen pathology was tumor-free. In these types of malignancies, a similar technique resulted in a complete response (CR) radiologically and pathologically has never been discussed before.
Conclusions: In pretreated patients with high-grade salivary gland mucoepidermoid carcinoma, pembrolizumab showed good anticancer activity and provided a clinically, radiologically, and pathological response with a viable treatment choice. More research is needed to bring Pembrolizumab to the front-line of treatment. The time and duration of medication should be compared to the time required for surgery in these investigations.

75 The rate of incidental carcinoma in thyroid as detected on positron emission tomography-computed tomography (PET-CT) scan among breast cancer patients at Ziv medical center

Majd Asakly M.Da b, Moran Barazani b, Adi Sharabi-Nov MA, MPHb c d , Shlomo Merchavy MDa b.
aOtolaryngology, Head & Neck Surgery Unit, Ziv Medical Center, Safed, Israel.
b Azrieli Faculty of Medicine, Bar-Ilan University, Safed, Israel
c Research Wing, Ziv Medical Center, Safed, Israel.
d Tel-Hai Academic College, Tel-Hai, Israel.

Objective: To study the incidental thyroid malignancy detected by PETCT scans in Ziv-treated breast cancer patients.
Background: Because of the increased use of (18)F-fluorodeoxyglucose positron emission tomography ((18)F-FDG PET) in breast cancer patients, an increasing number of incidentalomas in the thyroid gland have been identified. Up to 35% of thyroid focal incidentalomas are malignant. Early detection of thyroid cancer allows for less invasive surgical treatment and avoids complicated treatment of advanced thyroid cancer.
Methods: Medical records of 166 breast cancer patients who underwent FDG PET-CT imaging in the nuclear medicine department at ZIV medical center from January 2018 to January 2019 and who were treated in the oncology department at ZIV medical center were reviewed for thyroid incindentiloma, maximal standard uptake value (SUVmax), age, and ethnicity were recorded and compared between patients with thyroid malignant incidentaloma and other breast cancer patients.
Results: Eight patients out of 166 had a focal incidental finding in the thyroid gland. Three patients had significantly higher SUVmax values than the other five. One of these patients had a biopsy that revealed papillary thyroid carcinoma; the SUVmax in this case was 17, and the other two patients were referred for FNA of the incidentaloma. The SUVmax of the remaining five patients is less than 10. Malignant thyroid incidental tumors occurred at a rate of 12.5%.
Conclusion: Thyroid gland incidentalomas with a high focal metabolic rate may be malignant. In these patients, further investigation, including ultrasonography-guided fine needle aspiration, is required.

The role of the gut microbiota in food allergies

76

Nofar Asulin, Dana Binyamin, Lelyan Moadi1, Alona Riumin1, Soha Zgairy1, Mor Zigdon1, Sondra Turjeman1, Omry Koren1
1 Azrieli Faculty of Medicine, Bar-Ilan university, Safed, Israel

Background: IgE-mediated food allergy (FA) is defined as an unexcepted immune response that occurs within 2 hours of repeated exposure to specific foods. Oral food immunotherapy (OIT) is an active therapeutic strategy for FA and the gut microbiota is now becoming a research focus in the study of FA and particularly in treatment prognosis.

Methods: To characterize the gut microbiota composition, we collected 34 fecal samples from 17 walnut FA patients undergoing OIT, before and after OIT treatment, and an age-matched control group (n=19). DNA was isolated from fecal samples, and 16S rRNA gene sequences were analyzed. To explain the gut microbiota's mechanistic contributions to the altered immune state in FA and to assess whether pre-and post-OIT microbiota differentially impact the immune system, we preformed FMT from allergic walnuts patients before (n=7) and after (n=7) treatment and healthy non-allergic individuals as control (n=4) group to germ-free mice and tested their allergic response.

Results: The gut microbiota composition between the pre-treatment group and the non-allergic control group was significantly different in β-diversity but not α-diversity. This indicates that while richness is similar across groups, the bacterial taxa within the communities differ. The relative abundance of P. copri between the groups, was significantly different when comparing the pre-treatment group to the control group.
In the mice experiment, comparisons of b diversity demonstrated significant differences in the control group in comparison to pre-treatment group.

Conclusions: Our results demonstrate a link between IgE-mediated walnut FA and the composition of the gut microbiota. Because the microbiota is modifiable, this study can set the groundwork for discovery of potential therapeutic interventions that can be used to routinely treat FA.

77 Microbial changes during aging in mice

Dana Binyamin1, Nofar Asulin1, Lelyan Moadi1, Mor Zigdon1, Alona Riumin1, Sondra Turjeman1, Omry Koren1.
1 Azrieli Faculty of Medicine, Bar- ilan University, Safed, Israel

Introduction:
During aging, there is a physiological decline, an increase of morbidity and mortality, and changes in the gut microbiome. Microbial dysbiosis has been associated with diseases or important factors leading to disease onset and progression. In this study, we investigated microbial changes throughout aging in mice.

Methods:
Fecal samples were collected from 81 mice every two months from the age of 2 to 18 months and profiled by 16S rRNA gene sequencing. To determine bacteria with significantly altered abundances during aging, we used MaAsLin2. We also used ANCOM to assess bacterial differences between 4- and 18-month-old mice.

Results:
Bacterial composition (beta-diversity) differed significantly between every pair of time points except 10 vs 12 months, and bacterial richness (alpha-diversity) increased from age 2 to 6 months. We found an increase in Firmicutes/Bacteroidetes ratio over time with significantly different ratios between 2 and 12-months and 2 and 14-months samples which was also an increase in weight until the age of 6 months. MaAsLin2 revealed several taxa that increased over time such as Turicibacter and Oscillospira and decreased over time such as Akkermansia and Prevotella.

Conclusions:
The microbiome changes during aging in mice with increases in alpha diversity and the Firmicutes/Bacteroidetes ratio. We found specific bacterial changes throughout aging such as a decrease in Akkermansia which is associated with beneficial effects on intestinal and extra-intestinal health and displays probiotic potential.

Use of cognitive mapping for developing critical thinking in nursing students during their clinical practice

78

Rachel Kemelman, RN MA, PhD student (Sociology, Iasi University, Romania); Zefat Nursing School, Ziv Medical Center
Alisa Bernotaite, RN MA, Zefat Nursing School, Ziv Medical Center
Adi Sharabi-Nov, MA, MPH, Ziv Medical Center and Tel-Hai academic College
Zipi Regev-Avraham, RN PhD, Zefat Nursing School, Ziv Medical Center, Zefat College

Background: Nursing students struggle to cope with multiple pieces of information during their first clinical practice. Cognitive mapping is a visual technique that allows to present information, complex ideas and processes, helps to develop critical clinical thinking skills and integrate theoretical knowledge with clinical practice through meaningful learning.
Aim: To evaluate the effect of cognitive mapping use during initial clinical practice on nursing students' clinical and theoretical knowledge
Research hypothesis: 1) There is a relationship between cognitive mapping use and theoretical examinations and license examination grades. 2) The license examination grades can predict by the cognitive mapping tasks achievements
Method: A retrospective data survey, collected from the school database during 2020-2021 regarding students who were trained by cognitive map through a workshop. The correlations between cognitive mapping, final medical-surgical and license examination grades were examined.
Study Findings: Data included 169 school graduates. Statistically significant differences were found between Jewish and Arab students' grades in cognitive mappings, final and license exam ($p = 0.055$, $p = 0.020$, $p = 0.003$, respectively).
A multivariate regression for predicting the license exam grade show that, the mapping grades and the med-surg final test had a significant effect on the license exam grades ($p<0.001$).
Conclusions: The study's findings reinforced the need to encourage use of cognitive mapping during initial clinical practice to enable students to actively learn, integrate theoretical knowledge with clinical practice and understand clinical processes. It is important tool for developing critical thinking skills.

79 ACE2 IS A MUTUAL ENTRY FACTOR FOR HCV AND SARS-COV-2

Samer Ayoub[1], Tom Domovitz[1], Michal Werbner[1], Joel Alter[2], Lee Izhaki Tavor[2], Moshe Dessau[2], Meital Gal-Tanamy[1]

[1] Molecular Virology Lab, The Azrieli Faculty of Medicine Bar-Ilan University, Israel

[2] The Laboratory of Structural Biology of Infectious Diseases, The Azrieli Faculty of Medicine Bar-Ilan University, Israel

SARS-CoV-2 entry into its host cell is mediated via its interaction with the cellular receptor ACE2. Hepatocytes, the target cells of hepatitis C virus infection, express low level of ACE2. Recently, we have demonstrated that SARS-CoV-2 and HCV coinfect and coreplicate in hepatocytes. Moreover, we reported the enhanced SARS-CoV-2 entry into HCV-pre-infected hepatocytes via increase in ACE2 expression in HCV-infected cells. Here we aimed to evaluate whether ACE2 plays a role in HCV life cycle as well.

We demonstrate that Huh7.5 cells overexpressing ACE2 were more susceptible to HCV infection, while ACE2 silencing in Huh7.5 cells resulted in decreased susceptibility to HCV infection. We investigated which step in the HCV life cycle is influenced by ACE2, and identified that ACE2 overexpression in Huh7.5 cells increased HCVpp uptake and HCV cell binding as compared to control Huh7.5 cells. In contrast, RNA levels were similar in replicon cells with versus without ACE2 overexpression. These observations show that ACE2 increases viral entry into Huh7.5 cells at the cell binding step, without affecting HCV RNA replication. Furthermore, we demonstrate higher binding of HCV envelop protein E2 to ACE2-expressing Huh7.5 cells compared to control Huh7.5 cells, but not direct ACE2-E2 binding in in vitro assays, providing evidence for the involvement of ACE2 in HCV entry as a cofactor. We also demonstrate that the enhanced ACE2 expression in HCV infected cells is regulated via HCV-induced HIF1-alpha expression. This study reveals that ACE2 is a novel entry cofactor for HCV, which is upregulated in response to HCV infection. The HCV-induced increased ACE2 expression results in enhanced HCV and SARS-CoV-2 infections and efficient coinfection of both viruses in hepatocytes.

Characterization of protein aggregation promoted by PKA-RIβ mutation in a new neurodegenerative disease

80

Gilat Shimon, Anwar Dakwar, Tal Benjamin, Neta Peled, Ronit Ilouz
Azrieli Faculty of Medicine, Bar Ilan University, Safed, Israel.

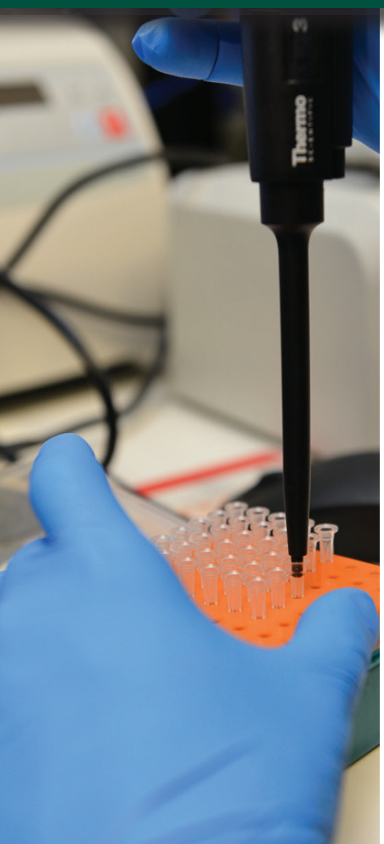
Dysregulation of cAMP signaling contributes to the etiology of several brain degenerative diseases. A missense mutation in PKA RIb regulatory subunit, the least studied isoform, was found in patients diagnosed with a rare neurodegenerative disease. The gap in knowledge regarding this gene and the devastating outcomes seen in individuals with motor deficits are two critically important problems that we are currently researching. Our recent structural model led us to hypothesize that an amino acid substitution L50R may result in preventing dimer formation. Biochemical studies as well as cell-based high-resolution image analysis suggest that this RIb missense mutation not only prevents RIb homodimerization but also eliminates the binding site that is created by dimer formation for A Kinase Anchoring Proteins (AKAPs) binding. Consequently, PKA holoenzyme localization is affected as evidenced by accumulation of RIb into neuronal inclusions in human brain patients. A quantitative multiplex proteomics revealed that the phospho-signaling cascade is disrupted by the L50R mutation in postmortem brains. This study emphasizes the importance of precisely controlled PKA isoform subcellular localization and demonstrates how a mutation in PKA regulatory subunit drives aberrant cAMP signaling and neurodegeneration. This study provides insights into the molecular and cellular mechanisms of other neurodegenerative diseases where PKA function is dysregulated.

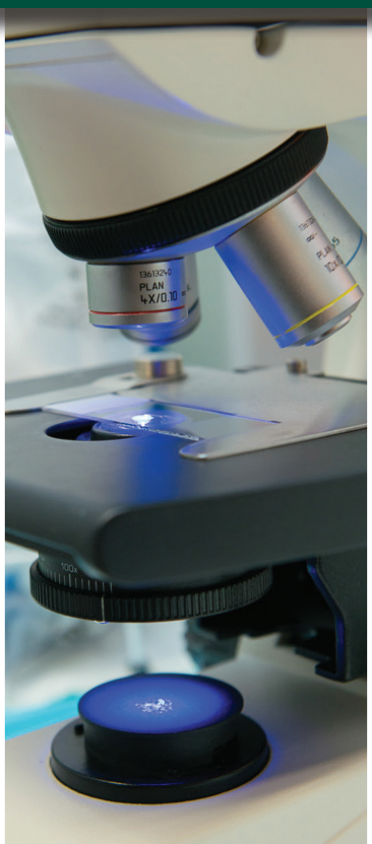
81 The psychedelic psilocybin induces short term anxiety response through a different molecular pathway from the psychedelic response.

Ram Harari, Dmitriy Getselter, Evan Elliott.
Molecular and Behavioral Neurosciences Lab, Bar-Ilan University, Azrieli Faculty of Medicine, Safed, Israel

Psilocybin has emerged as a major research interest due to its potential as a treatment for several neuropsychological disorders, in particular anxiety and depression. Psilocybin is naturally found in some mushroom species commonly known as "magic mushrooms" and has a primary role in the psychedelic and hallucinogenic effects that these mushrooms induce. Psilocybin's hallucinogenic effect is mediated by its metabolite psilocin, a non-selective serotonin 5-HT2A receptor agonist, which through modulation of the serotonergic (5-HT) systems leads to regulation of excitatory neurotransmission and gene transcription in the brain. However, there is little knowledge regarding the brain regions, cell types, genes and mechanisms through which psilocybin may affect depression and anxiety related behaviors. The purpose of our study is to investigate how psilocybin affects anxiety and depressive-related behaviors, and how neuronal activity and molecular pathways change in association with behavioral changes. By using specific anxiety and depressive-related behavioral tests, immunohistochemistry staining for neuronal activation at specific brain sites and pharmacological blocking of 5-HT2A receptor by Ketanserin in mice, we found that Psilocybin produced significant increase in anxiety-related behavior and a specific effect on neurons activation in the amygdala. In addition, we found that pharmacological blocking of 5-HT2A receptor attenuates psilocybin-induced head twitch response, a mouse correlate of psychedelic response, but did not rescue psilocybin effect on anxiety-related behavior. This suggests Psilocybin induces changes in anxiety-related behaviors through a molecular pathway which is distinct from the 5-HT2A psychedelic inducing pathway. These results give important insights into how psilocybin may induce short-term anxiety-causing effects.

The meeting is sponsored by


Thermo


PLAN
4X/0.10