coding RNAs (sncRNAs) of EVs derived from nasopharyngeal secretions (NPS) of children with episodes of viral infections and exposed to SHTS; ii) test the biological activity of EVs released from upper airway mucosa on target/recipient lung cells METHODS/STUDY POPULATION: EVs were isolated from ten NPS samples of children with episodes of acute respiratory infections. EVs were characterized by particle sizing (size and concentration), EV markers, and protein arrays for interferons, cytokines, and other immune mediators content. RNA was extracted from ten samples of NPS-derived EVs by column for next generation high throughput sequencing (NGS) to identify sncRNAs in EVs. In studies currently in progress, EVs will be isolated from RSV-infected human nose organoids (HNOs) cells in air liquid interface (ALI) culture, with or without pre-exposure to tobacco smoke. EVs will be then tested for antiviral activity on recipient RSV-infected lower airway cells. Viral titers will be measured in recipient infected lung cells. RESULTS/ANTICIPATED RESULTS: We isolated EVs from NPS samples and confirmed by immunoblot EV markers CD63 and Alix. The average size of NPS-derived EVs of virus positive and negative patients was 170 nm and 145 nm, respectively. We determined the particles number of EVs, concentrations of IFN-βand IFN-Î»in NPS and NPS-derived EVs of these children. While IFN-βlevels were below the limit of detection in both NPS and NPS-derived EVs of all children, IFN-Î»was detected in NPS and NPS-derived EVs from infected patients, except for the two patients with no viral infections. We extracted RNA from control-, virus infected- and/or SHTS- EVs and found that piR-36511, piR-40926, piR-49645, piR-32679 and piR- 53263 were all significantly enriched in EVs derived from NPS of children exposed to TS compared to those not exposed. DISCUSSION/SIGNIFICANCE: RSV leads to approximately 22,000 hospitalizations of children due to second-hand smoke. A vaccine is not currently available for RSV infection. EVs represent a novel translational approach to target undruggable. Airway mucosa EVs in viral respiratory infection function as antiviral messengers and tobacco smoke impairs the EV antiviral activity.

490

Nursing Professionals'Experiences during the COVID-19 Pandemic in Puerto Rico: A Phenomenological Study

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OBJECTIVES/GOALS: The COVID-19 pandemic has impacted nursing frontline professionals. The aims of this study were to explore experiences of nursing professionals in Puerto Rico during the pandemic, examine the impact on their health and provide research development opportunities enhance research capacity. METHODS/STUDY POPULATION: This interpretative phenomenological study recruited graduate nurses who participated in one in-depth semi-structured virtual interviews. Interviews were audio recorded and transcribed. The data analysis process was guided using the following steps: 1. Reading and re-reading, 2. Initial noting, 3. Developing emergent themes, 4. Searching for connections across emergent themes, 5. Moving to the next case, 6. Looking for patterns across cases, and 7. Writing up. In addition, Van Manen's thematic structure of four foundations was used as a complement to guide reflection and interpretation. Faculty and students participated throughout the process. RESULTS/ANTICIPATED RESULTS: Seven nursing professionals'lived experiences caring for Covid-19 patients were gathered. Their ages ranged from 31 to 45 and had worked between 2 and 14 years providing direct care. Themes that emerged from narrations include compassion fatigue, teamwork, working beyond clinical role, and gratification. Nurses expressed dealing with a very difficult situation, fear of being infected , or infecting my family , and working together to get through it and better help patients . Nurses also expressed feelings of anxiety and lack of institutional support. Additionally, the impact of working with patients, feeling good for being there, good or bad and support from families. DISCUSSION/SIGNIFICANCE: Nurses'narrations point to the complexities of their experiences working during the pandemic. They had to transcend usual demands even though they often lacked needed support. We must recognize the value of nursing and reflect upon changes in healthcare that are essential to move nursing forward.

491

Outcomes of an Integrated Research Ethics Consultation Service

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OBJECTIVES/GOALS: The need for mechanisms of ethical discourse and guidance has increased as translational research collaborations become more complex. The goal of this project is to analyze the stakeholder engagement and ethical issues our research ethics consultation service (RECS) conducted over a two year period. METHODS/STUDY POPULATION: We conducted a retrospective review of our RECS database from 2020 to 2022. We examined the nature of the research and ethical issues of concern from consult requestors, including whether or not consults were preventative. In addition, we assessed the educational outreach conducted during that timeframe as a measure of service awareness. RESULTS/ANTICIPATED RESULTS: There was a total of 42 consults conducted over the previous year. There were a wide variety of issues related to informed IRB-related processes (31%), consent (24%), QA/QI determination (12%), authorship (10%), confidentiality (7%), diversity/inclusion (7%), grant preparation (7%). Many of the consults (n=28, 67%) included secondary issues. A few consults (n=4, 10%) were preventative, meaning that the consult was requested in anticipation or consideration of a potential ethical issue. Outreach efforts extended to a diverse array of institutional stakeholders and trainees. DISCUSSION/SIGNIFICANCE: The RECS serves numerous constituencies throughout our institution on ethical issues spanning nearly all aspects of research design, conduct, and analysis. These data highlight initiatives to increase study efficiency (in collaboration with institutional research oversight) and helps to direct educational efforts and outreach.

492

Phagocyte heterogeneity and T cell dependence of cellular host defense mechanisms in tuberculosis

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OBJECTIVES/GOALS: Phagocytes, diverse cells that ingest material, are the primary cell type infected by Mycobacterium tuberculosis (Mtb) and the executors of protective mechanisms. T cells play a critical role by helping phagocytes control the infection. Understanding the precise T cell-dependent mechanisms by which

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phagocytic cell types contain Mtb is critical. METHODS/STUDY POPULATION: To determine the impact T cells have on different phagocyte cell populations' host defense mechanisms, groups of wild–type and T cell deficient TCRa-/- mice were infected with an Mtb strain expressing fluorescent mScarlet protein. At four weeks post-infection, a time when T cell help contributes to control of Mtb, lungs were homogenized and cells sorted based on detection of mScarlet, indicating Mtb-infected cells. Cell suspensions from each mouse background were underwent single-cell RNA sequencing analysis to reveal the heterogenous cellular transcriptional response of different phagocyte populations. RESULTS/ANTICIPATED RESULTS: We found that Mtb-infected phagocytes from wild-type and TCRa-/- mouse lungs contain the same dominant cell phenotypic clusters, but these have different patterns of gene expression. Without T cells, phagocytes are prone to a more inflammatory phenotype. DISCUSSION/SIGNIFICANCE: This will translate fundamental biological data to test the hypothesis that Mtb encounters different environmental stresses exerted by different phagocytic cell types. This work could reveal host intracellular niches that enable bacterial persistence and elucidate new pathways that could be targeted for traditional antibiotic therapies for TB.

493

Prevalence and Clinical Presentation of Chronic Neck Pain in Individuals with Generalized Joint Hypermobility

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OBJECTIVES/GOALS: Evidence suggests that individuals with generalized joint hypermobility (GJH), or excessive joint range of motion, are at higher risk of developing chronic neck pain. The objective of this study is to determine the prevalence and clinical presentation of chronic neck pain in GJH and investigate its associations with other measures of spine health. METHODS/STUDY POPULATION: Data was collected at the Driven to Discover Research Facility at the 2022 Minnesota State Fair. Individuals 18 years and older were invited to participate. All enrolled participants completed Phase 1, which included: the Beighton Score (measure of GJH), the 5-Point-Questionnaire (self-report survey for current or historical GJH), and a custom self-report survey for demographics and musculoskeletal pain. A subset of participants was also asked to complete Phase 2 of the study. Phase 2 consisted of additional self-report surveys (Neck Disability Index (NDI) and PROMIS-10 Global) and the following physical measures: neck range of motion in all planes, neck strength in flexion-extension and lateral bending, and grip strength. RESULTS/ANTICIPATED RESULTS: A total of 559 participants were enrolled in the study. All participants completed Phase 1, and 285 of those individuals completed Phase 2. Those with a Beighton Score≥4 were categorized as having GJH. The overall prevalence of GJH was 23.8% for females and 9.1% for males. Consistent with previous studies, multiple linear regression analysis (R2=0.20, F(2,552) = 69.37, p DISCUSSION/SIGNIFICANCE: This is one of the largest studies investigating GJH, pain, and physical measures of neck function in the general population. The results highlight the higher prevalence of chronic neck pain in those with GJH and will form the basis for a subsequent study to identify mechanisms and potential therapeutic targets for individuals with GJH and chronic pain.

495

Radon and Fracking Exposures and Lymphoma Risk in a Canine Model of non-Hodgkin Lymphoma*

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OBJECTIVES/GOALS: The objective of this study was to determine whether residential radon and proximity to horizontal oil and drilling (fracking) are risk factors for the development of multicentric lymphoma in pet dogs, a spontaneous, immunocompetent model for non-occupational risk for NHL in humans. METHODS/STUDY POPULATION: Two case-control populations of dogs with multicentric lymphoma were utilized, with a focus on two dog breeds at high risk for lymphoma. Control dogs were matched for age, breed, and sex. Home addresses were collected for 54 Golden retrievers with lymphoma and 108 Golden retriever controls, and for 56 boxer dogs with lymphoma and 84 unaffected boxer controls. Counties of residence were matched to radon zones and percentage of home radon tests that exceeded the actionable level of 4 pCi/L, available by county through the EPA and the CDC National Environmental Public Health Tracking Network from 2008 to 2017. Locations of horizontal oil and gas wells were obtained from the Enverus Database, and distances from dog homes to the closest well, and well density by county, were calculated for each case and control. RESULTS/ANTICIPATED RESULTS: We found no significant differences in radon zones, county level radon measurements, or residential proximity to active fracking wells between dogs with lymphoma and unaffected controls in either the Golden Retriever or boxer populations. DISCUSSION/SIGNIFICANCE: Canine multicentric lymphoma resembles human NHL and is a valuable model of non-occupational environmental risk for NHL in people. Although we did not find geographic associations between radon and fracking wells, follow-up studies will measure household radon, as well as household air, water, and dog urine for potentially genotoxic chemicals.

498

Structural Determination of the CqsR CACHE Domain and its Autoinducer*,†

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OBJECTIVES/GOALS: Our goal is to determine the structure of the CACHE domain of the Vibrio cholerae quorum sensing receptor CqsR as well as its autoinducer (AI). We are performing X-ray crystallography on the protein in its apo form, with the fractions containing the AI, and with known ligand ethanolamine (ETA). METHODS/STUDY POPULATION: We have transformed BL21(DE3) E. coli cells with a pTB146 vector to contain the gene for the CqsR CACHE domain. We grow these cells to high optical density and induce protein expression, at which point we harvest them and purify the protein. This entails lysing the cells, separating the protein with Ni-NTA resin, cleaving our protein tag, and column chromatography. With purified protein, high-throughput screens are set up to find crystallization conditions of apo CqsR, CqsR-ETA, and CqsR-AI. We then determine conditions that best lead

†Wai-Leung Ng's name has been corrected. Additionally, middle initials for two authors have been added and the affiliations have been corrected. A corrigendum detailing these changes has been published (doi:10.1017/cts.2023.551).

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