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### Heart Rate Variability in Opioid Use Disordered Participants Undergoing Buprenorphine-Assisted Detoxification

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**OBJECTIVES/GOALS:** This study explored whether gabapentin (GBN) differentially impacted heart rate variability (HRV) and whether HRV was associated with opioid withdrawal ratings among participants with opioid use disorder (OUD) undergoing a randomized, double blind placebo-controlled, trial (RCT) of GBN during a buprenorphine (BUP)-assisted taper. **METHODS/STUDY POPULATION:** Participants (ages 18-64) with OUD, no recent use of benzodiazepines/barbiturates, and no major psychiatric disorders or unstable medical conditions were enrolled in the RCT, induced onto BUP starting week 1 day 1 and randomly assigned to receive adjunct GBN or placebo starting week 1 day 3. All participants began a 10-day BUP-taper beginning week 2 day 3. HRV measures were assessed on week 1 day 2 (before GBN/placebo induction), week 2 day 2, and week 3 day 5 (end of BUP taper). HRV metrics were analyzed using Two Sample T-Test to determine differences between GBN vs. Placebo. Correlations between HRV metrics and opioid withdrawal ratings administered at the above timepoints will be analyzed using Spearman correlation. **RESULTS/ANTICIPATED RESULTS:** 28 participants underwent at least 1 HRV session that resulted in usable data. Preliminary statistical analyses revealed that HRV trended lower for GBN subjects during PB exercises than Placebo subjects, demonstrated by a higher mean heart rate for GBN subjects compared to Placebo subjects ( $p=0.0506$ ) at the end of the BUP-taper (week 3 day 5). We expect future analyses to demonstrate a negative correlation between certain HRV metrics indicative of parasympathetic tone and opioid withdrawal rating assessment scores indicative of withdrawal severity. Such findings would demonstrate an association between opioid withdrawal severity and lower parasympathetic tone and HRV. **DISCUSSION/SIGNIFICANCE:** Individuals with OUD have previously been shown to have a lower parasympathetic tone than individuals without OUD. Additionally, opioid withdrawal has been shown to be associated with reduced parasympathetic tone. Our initial findings suggest that adjunct GBN administration was not associated with lower parasympathetic tone during PB exercises.

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### Hepatomegaly and fatty liver infiltration among unhealthy weight pediatric population in Puerto Rico

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**OBJECTIVES/GOALS:** A quarter of Puerto Rican pediatric population are overweight or obese. Pediatric obesity is established as a

global public health concern that will develop liver complication as a Non-alcoholic fatty liver disease (NAFLD). This study aims to determine the association between body weight, liver size and texture in Puerto Rican pediatric population. **METHODS/STUDY POPULATION:** Twenty-nine ( $n=29$ ) pediatric participants (20 unhealthy weight and 9 healthy weight) between 7 to 19 years underwent panoramic ultrasound imaging of the RLL. Craniocaudal RLL measurement and liver texture were evaluated. Body mass index and waist circumference were also compared. Shapiro-Wilk test and students t-tests were attained with significance at  $p < 0.05$ . **RESULTS/ANTICIPATED RESULTS:** Statistical differences were detected between healthy weight and unhealthy weight pediatric patients: (1) RLL craniocaudal diameter ( $p=0.006$ ); (2) waist circumference ( $p < 0.001$ ); (3) BMI ( $p < 0.001$ ). Unhealthy weight (overweight and obese) pediatric patients presented a greater number of livers with fat infiltration (13) and hepatomegaly (15). **DISCUSSION/SIGNIFICANCE:** This preliminary results showed that liver size was larger and fatty infiltration are relatively frequent among overweight and obese pediatric patients. Prevention of unhealthy weight and the diagnosis of changes in liver texture and size among pediatric patients is important to prevent progressing of liver disease and avoid irreversible damage.

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### High Potency STING Agonists Induce Adaptive Immunity-Dependent Curative Responses in an Immune Checkpoint Blockade-Refractory Glioblastoma Model

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**OBJECTIVES/GOALS:** Glioblastoma (GBM) is the most common and aggressive adult primary brain malignancy. Clinically, GBM is refractory to T cell immune checkpoint blockade (ICB), in part due to its dense immune suppressive myeloid stroma. Here we show that myeloid-targeting STING agonists can repolarize the GBM microenvironment to cure ICB-refractory GBM models. **METHODS/STUDY POPULATION:** Using the synthetic cyclic di-nucleotide STING agonist IACS-8803 (8803) we treated orthotopic ICB-refractory QPP8 orthotopic murine GBM tumors intratumorally. We then analyzed survival and performed high parameter flow cytometry profiling of the tumor immune microenvironment following STING agonist treatment. To assess the contribution of adaptive immunity to STING agonist therapeutic efficacy, we treated orthotopic QPP8 tumors implanted in RAG1 KO mice and monitored survival. **RESULTS/ANTICIPATED RESULTS:** We found that STING agonist therapy cured murine orthotopic QPP8 tumors, in contrast to ICB that showed no survival benefit. In RAG1-/- mice bearing QPP8 tumors STING agonist therapy extended survival, however, the curative effect observed in wild-type mice was lost in the absence of adaptive immunity. STING agonist-treated QPP8 tumors displayed increased counts of CD8 T cells and NK cells, and decreased CD8 T cell PD1 expression. Infiltration of STING-treated gliomas by Ly6C+ F4/80+ Mono-MDSC substantially increased; however, these cells expressed reduced CD206 and CD163, suggestive of reduced immuno-suppression. Finally, in the cervical LN of QPP8-treated mice the frequency and CD80/CD86 expression of cDC1 cells increased. **DISCUSSION/SIGNIFICANCE:** ICB has failed in GBM, and the suppressive myeloid stroma remains a major barrier to generating anti-GBM