tadecadienoic, eicosatetraenoic, eicosatrienoic, docosahexaenoic and docosapentaenoic acids. Pretreatment of MEC with TPP-C18:1 resulted in: i) significant decrease of CL polyunsaturated molecular species and simultaneous elevation of nonoxidizable CL molecular species containing C18:1 and ii) suppression of AcD induced apoptosis. An inhibitor of long chain acyl-CoA synthase, triacsin C, blocked integration of C18:1 into CL molecules and restored MEC's sensitivity to AcD-induced apoptosis. Thus, metabolic remodeling of CL can be a new strategy in regulation of apoptotic cell death pathway and lead to the development of new preventive and therapeutic approaches against pathological conditions where apoptosis is a major contributor, eg, acute radiation syndrome. Supported by NIH U19 AI068021, HL70755, HL094488, ES020693, NIOSH OH008282.

PS 2036 A MITOCHONDRIA-TARGETED IMIDAZOLE-SUBSTITUTED FATTY ACID INHIBITS CYTOCHROME C PEROXIDASE AND MITIGATES RADIATION-INDUCED DEATH.

J. Atkinson[4, 2], A. Kapralov[1, 2], N. Yanamala[1, 2], Y. Tyurina[1, 2], A. Amoscato[1, 2], L. Pearce[1], J. Peterson[1], Z. Huang[1, 2], J. Jiang[1, 2], A. Samhan-Arias[1, 2], A. Maeda[1, 2], W. Feng[1, 2], N. Belikova[1, 2], V. Tyurin[1, 2], H. Wang[1, 2], J. Fletcher[4], Y. Wang[4], I. Vlasova[1, 2], J. Klein-Seetharaman[1, 2], D. Stoyanovsky[1, 2], H. Bayir[1, 2], B. Pitt[1], M. Epperly[3, 2], J. Greenberger[3, 2] and V. Kagan[1, 2]. *[1]EOH, University of Pittsburgh, Pittsburgh, PA, [2]Center for Medical Countermeasures against Radiation, University of Pittsburgh, Pittsburgh, PA, [3]Radiation Oncology, University of Pittsburgh, Pittsburgh, PA and [4]Chemistry, Brock University, St. Catharines, ON, Canada.*

The risk of radionuclide release in terrorist acts or exposure of healthy tissue during radiotherapy demand novel potent radioprotectants. Ionizing radiation induces cell death by initiating the selective peroxidation of cardiolipin (CL) in mitochondria by the peroxidase activity of its complex with cytochrome c leading to release of hemoprotein into the cytosol and commitment to the apoptotic program. We designed and synthesized mitochondria-targeted triphenylphosphonium-conjugated imidazole-substituted oleic (TPP-OA) and stearic (TPP-SA) acids. We found that TPP-IOA and TPP-ISA exerted strong specific liganding of heme-iron in cyt c/CL complex, effectively suppressed its peroxidase activity and CL peroxidation. In addition, TPP-IOA and TPP-ISA prevented cell death induced by irradiation. Both TPP-IOA and TPP-ISA protected/mitigated C57BL6 mice against lethal doses of irradiation. Significant radioprotective/radiomitigative effects of TPP-OA were observed after pretreatment of mice from 1 h before through 24 h after irradiation. This is an important advance in the development of radioprotectors with broad spectrum of applications in biomedicine and biodefense. Supported by NIOSH OH008282; NIH U19 AI068021, HL70755, HL094488, ES020693, ES021068.

PS 2037 MINOCYCLINE AND DOXYCYCLINE CYTOPROTECT BY INHIBITION OF THE MITOCHONDRIAL CALCIUM UNIPORTER AND NOT BY OTHER MECHANISMS.

J. Schwartz[1], X. Zhang[1], G. L. Lovelace[1] and J. J. Lemasters[1, 2]. *[1]Pharmaceutical Sciences, Medical University of South Carolina, Charleston, SC and [2]Biochemistry, Medical University of South Carolina, Charleston, SC.*

BACKGROUND: Minocycline and doxycycline are tetracycline derivatives that protect hepatocytes and other cell types against hypoxia and ischemia/reperfusion (I/R) injury. Previously, we showed the minocycline and doxycycline inhibit Fe2+ and Ca2+ uptake through the mitochondrial Ca2+ uniporter (MCU). The AIM of this study was to evaluate competing hypotheses for the mechanism of cytoprotection by minocycline and doxycycline, namely: 1) mitochondrial depolarization suppressing iron and Ca2+ uptake and formation of reactive oxygen species, 2) MCU inhibition, and 3) inhibition of matrix metalloproteases (MMP). METHODS: Primary rat hepatocytes were incubated in buffer containing fluorescent indicators of Ca2+ (Fluo-5N), membrane potential (Rh123) and cell viability (propidium iodide) with test compounds or vehicle in models of ischemia/reperfusion and chemical hypoxia (KCN and iodoacetic acid). Fluorescence of indicators was assayed by a NovoStar multi-well plate reader. RESULTS: Minocycline and doxycycline (20 and 10 μM respectively) and Ru360, a specific MCU inhibitor, diminished I/R-induced cell death without causing mitochondrial depolarization. By contrast, higher concentrations (100 μM) of minocycline and doxycycline caused depolarization and simultaneously increased cell death. MMP inhibitors MMP2/MMP9 Inhibitor I (5 – 500 μM) and Cis-9-Octadeconyl-N-hydroxylamide (1 – 100 μM) did not protect against any parameter measured. CONCLUSION: Mitochondrial depolarization and MMP inhibition do not explain cytoprotection by minocycline and doxycycline because mitochondria did not depolarize at cytoprotective concentrations and no cytoprotection was observed with MMP inhibitors. Rather, inhibition of Fe2+ and/or Ca2+ uptake through MCU likely underlies cytoprotection by Ru360, minocycline and doxycycline.

PS 2038 ETHYLMERCURY INDUCES ER STRESS AND MITOCHONDRIA-MEDIATED APOPTOSIS.

J. Choi, Y. Park and M. Dong. *School Lifesciences and Biotechnology, Korea University, Seoul, Republic of Korea.* Sponsor: B. Lee.

Mercury is one of the most important environmental and industrial pollutants throughout the world. Exposure to mercury causes strong damage to organs including the brain, blood, liver, bone and kidneys. Renal proximal tubular cells represent the primary target site where mercury is taken up, accumulated, and expresses toxicity. We previously investigated the cytotoxicity of seven kinds of mercury compounds in human renal proximal tubule (HK-2) cells. Our experimental data demonstrated that ethylmercury was shown the strongest cytotoxicity among them at 24h and 48h exposure in HK-2 cells. In this study, we explored the mechanism of ethylmercury induced cytotoxicity in HK-2 cells. Ethylmercury chloride (EMC) induced cytotoxicity with 2.4 and 0.76 uM of IC50 values exposed for 24h and 48h, respectively. For 24h exposure to 1 and 2 μM of EMC, The cells were dose-dependently undergone the apoptosis in FACS analysis. After 4h treatment of cells to 2 μM EMC, cleaved of caspase-9 and caspase-12 which are activated by ER stress, were markedly activated. EMC was up-regulated the mRNA of CHOP (transcription factor, C/EBP homologous protein), XBP1 (X-box binding protein-1), ERdj4 (endoplasmic reticulum-localized DnaJ homologues) and GRP78 (78 kDa glucose-regulated protein) expression. In HK-2 cells exposed to 2 μM EMC, phosphorylated eIF2 and CHOP protein were increased at 3h and GRP78 protein at 12h. To determine the EMC induced oxidative stress, we observed the changes in mitochondrial membrane potential with JC-1 and gene expression using RT-PCR analysis in HK-2 cells. ECM was increased the accumulation of JC-1 monomers in mitochondria from 0.5 to 2 μM concentrations in a dose-dependent manner. The expression of Hic (hydrogen peroxide-inducible clone)-5, which were related to oxidative stress, was also induced by EMC treatment. Collectively, these results suggested that EMC exposure induced apoptosis via ER stress and mitochondrial dysfunction in human renal proximal tubule (HK-2) cells. * This research was support by a grant (10182KFDA992-2101) from Korea Food & Drug Administration in 2011

PS 2039 CADMIUM-INDUCED OXIDATIVE STRESS DAMAGE CAUSES NEURON CELLS APOPTOSIS THROUGH JNK/MITOCHONDRIA-DEPENDENT/ENDOPLASMIC RETICULUM STRESS PATHWAYS.

L. Tsai[1], T. Tseng[2], T. Lu[1], D. Hung[3], C. Su[4], C. Yen[5], C. Huang[6], S. Liu[7] and Y. Chen[8]. *[1]Graduate Institute of Basic Medical Science, China Medical University, Taichung, R.O.C., Taiwan, [2]Department of Anatomy, China Medical University, Taichung, R.O.C., Taiwan, [3]Toxicology Center, China Medical University Hospital, Taichung, R.O.C., Taiwan, [4]Department of Otorhinolaryngology, Head and Neck Surgery, Changhua Christian Hospital, Changhua, R.O.C., Taiwan, [5]Department of Occupational Safety and Health, Chung Shan Medical University, Taichung, R.O.C., Taiwan, [6]School of Chinese Medicine, China Medical University, Taichung, R.O.C., Taiwan, [7]Institute of Toxicology, National Taiwan University, Taipei, R.O.C., Taiwan and [8]Department of Physiology, China Medical University, Taichung, R.O.C., Taiwan.*

Cadmium (Cd), a well-known toxic metal, is an important pollutant throughout the world. In mammalian, exposure to Cd causing injuries of kidney, liver and osseous system has been demonstrated. Although some studies have shown the possible connections between neurodegenerative disorders and Cd exposure, the toxic effects of Cd on neuron cell are still unclear. In this study, we designed to investigate the effects and possible mechanisms of Cd-induced neuron cell death. Our results found that after exposed to Cd in cultured Neuro-2a cells for 24 h obviously decreasing the viable cells,mitochondrial membrane potential, and led to glutathione depletion in a dose-dependent manner with a range from 1 to 20μM, which accompanied by a marked Cd accumulation in cytosol. Cd also induced the protein phosphorylation of JNK, the disruption of mitochondrial function, Bcl-2 down-regulation, Bax up-regulation, the activation of PARP and caspase cascades, displaying features of mitochondria-dependent apoptotic signaling pathways. Furthermore, exposed Neuro-2a cells to Cd could trigger endoplasmic reticulum (ER) stress as indicated by the alterations of glucose-regulated protein (GRP) 78,

The Toxicologist

Supplement to *Toxicological Sciences*

51st Annual Meeting and ToxExpo™

March 11–15, 2012 • San Francisco, California

SOT 2012 San Francisco California

An Official Journal of the Society of Toxicology

SOT | Society of Toxicology

Creating a Safer and Healthier World by Advancing the Science of Toxicology

www.toxicology.org

OXFORD UNIVERSITY PRESS

ISSN 1096-6080
Volume 126, Issue 1
March 2012

www.toxsci.oxfordjournals.org