volved with FSH-mediated signaling in Sertoli cells, 14-3-3 Theta and 14-3-3 Epsilon modulate protein kinase C activity and are also found in Sertoli cells. 1-Cys Peroxiredoxin and 14-3-3 Zeta have phopholipase A2 activity. These data reveal disruption in Sertoli cells and imply that germ cell compromise may be secondary to Sertoli cell insult.

1326 DI(*n*-BUTYL) PHTHALATE INTERFERES WITH FETAL TESTICULAR STEROIDOGENESIS AT THE LEVEL OF CHOLESTEROL TRANSPORT AND CLEAVAGE.

C. Thompson, S. M. Ross and K. W. Gaido. *CIIT Centers for Health Research, Research Triangle Park, NC.*

The phthalate ester di(*n*-butyl) phthalate (DBP) produces antiandrogenic effects on male reproductive development in rats. In the fetus, these effects are mediated, not by interaction with the androgen receptor, but rather through diminution of testosterone (T) production by the testes. Previous studies have shown that several genes involved in cholesterol transport and steroidogenesis are downregulated at the mRNA level following *in utero* exposure to DBP. The purpose of this study was to make a functional determination of the points in the cholesterol transport and steroidogenesis pathways affected by DBP. We cultured fetal testis explants with T precursors and assessed cholesterol uptake and T production. Pregnant Sprague-Dawley rats were treated with 500 mg/kg DBP or corn oil control *via* oral gavage from gestational days 12 to 19. Following the final treatment, testes were removed from the fetuses and cultured for 3 h with ^{3}H-cholesterol, leuteinizing hormone (LH), Bt_2-cAMP, 22(R)-hydroxycholesterol, pregnenolone, progesterone, or 17-hydroxyprogesterone. T production in unsupplemented cultures of DBP-exposed testis was roughly 10% of that seen in corn oil controls (164.7 ± 32 pg/h vs. 1684.1 ± 347 pg/h). Both control and treated explants could be stimulated by LH or Bt_2-cAMP, but T production by DBP-treated testes remained less than 50% of control levels. Incorporation of ^{3}H-cholesterol by mitochondria of DBP-treated explants was 67% of that observed in controls, although this difference was not statistically significant ($p = 0.08$). Pregnenolone, progesterone, and 17-hydroxyprogesterone all significantly increased T production compared to unsupplemented DBP-treated explants. However, there was no significant difference between the unsupplemented explants and those treated with the membrane-permeable 22(R)-hydroxycholesterol. These data indicate that the toxic effects of DBP on the fetal testis are mediated at the level of cholesterol cleavage by $P450_{scc}$ and possibly at the level of cholesterol transport into the mitochondria.

1327 EFFECTS OF METHOXYCHLOR (M) OR ITS ACTIVE METABOLITE, 2, 2-BIS(*p*-HYDROXYPHENYL)-1, 1, 1-TRICHLOROETHANE (HPTE), ON TESTOSTERONE (T) FORMATION BY CULTURED NEONATAL (FETAL) LEYDIG CELLS (LC).

E. P. Murono and R. C. Derk. *Pathology and Physiology Research Branch, CDC/NIOSH, Morgantown, WV.* Sponsor: V. CASTRANOVA.

M is a pesticide developed as a replacement for dichlorodiphenyltrichloroethane (DDT). Its active metabolite is thought to be HPTE. Both M and HPTE have been reported to exhibit weak estrogenic or antiandrogenic activities, their proposed mechanism(s) of action. In the present studies, we examined the effects of M or HPTE on T biosynthesis by cultured LC from neonatal rats, which represent fetal LC. Increasing concentrations of M or HPTE (100-1000 nM) caused a progressive decline in both basal and 10 mIU/ml human chorionic gonadotropin (hCG)- or 1 mM 8 Br-cAMP-stimulated T following exposure for 4 or 24 h, although the declines with HPTE were greater. To localize the site(s) of action of HPTE, LC were exposed to HPTE (100-1000 nM) for 24 h (both alone or with hCG), then fresh media containing steroid precursors of T were added to assess their conversion to T over 4 h. The conversion of 0.01 mM pregnenolone, progesterone or androstenedione to T was unaffected by prior exposure to HPTE; however, the conversion of 22(R)hydroxycholesterol to T progressively decreased, suggesting that among the enzymes involved in converting cholesterol to T, P450 cholesterol side-chain cleavage activity is inhibited by HPTE. The concomitant inclusion of the "pure" estrogen antagonist, ICI 182, 780, did not alter the inhibitive effects of HPTE, suggesting that the effects of HPTE are not mediated through the estrogen receptor (ER) pathway. Furthermore, the antiandrogen 4-hydroxyflutamide (100-1000 nM) had no effect on hCG-stimulated T following 24 h exposure, suggesting that HPTE is not acting as an antiandrogen through the androgen receptor (AR). These results suggest that the two prevailing proposed modes of action of MC/HPTE in altering male reproductive function (weakly estrogenic through the ER or antiandrogenic through the AR) do not apply with respect to their inhibitive effect on T formation by fetal LC.

1328 EVIDENCE FOR THE PRESENCE AND ACTIVITY OF SOLUBLE EPOXIDE HYDROLASE IN THE RAT EPIDIDYMIS AND SPERM.

S. B. DuTeaux[1], J. W. Newman[2], C. Morisseau[2], E. A. Fairbairn[1], B. D. Hammock[2] and M. G. Miller[1]. *[1]Environmental Toxicology, University of California, Davis, CA and [2]Entomology, University of California, Davis, CA.*

Soluble epoxide hydrolase (sEH) is proposed to play a role in mediating ion balance in the kidney through the catalytic transformation of bioactive arachidonic acid metabolites. The epididymis and kidney are related embryonically. In addition, the epididymis regulates ion concentrations through diffusion, active transport, and leaky epithelium much like the kidney. Based on these similarities, the present study used immunohistochemistry, Western blotting, and enzyme assays to address the possibility that sEH is present in the epididymis. Tissue sections probed with an antibody to sEH showed immunoreactive proteins in the clear cells of the caput, corpus, and cauda epididymis. The sEH antibody also reacted with proteins on the ventral acrosome and the principal piece of sperm. In Western blots, immunoreactive proteins in kidney cytosol were consistent with a 65 kD recombinant sEH standard. The same antibody recognized a single 75 kD protein in epididymal segments and a single 71 kD protein in sperm. Epoxide hydrolase activity was measured in kidney, epididymal, and sperm samples using [^{3}H]-*trans*-diphenylpropene oxide (tDPPO) as a substrate. Results indicate that all portions of the epididymis as well as sperm are capable of generating diols from the tDPPO epoxide. The specific activity of the corpus epididymis (110 pmol/min/mg) was similar to the activity in the kidney cortex (113 pmol/min/mg). Evaluation of *in vitro* arachidonic acid metabolism showed that the epididymis can generate epoxides and diols. In addition, epididymal metabolism was chemically inhibited by cyclohexyldodecyl urea, a potent sEH inhibitor. The present study indicates the presence of epoxide hydrolase activity in the epididymis at levels similar to the kidney. Because the protein detected in the epididymis is larger than the known mass of sEH, immunoinhibition assays and protein sequencing studies are planned to further elucidate the nature and identity of epoxide hydrolase in the epididymis.

1329 IDENTIFICATION OF TRICHLOROETHYLENE AND ITS METABOLITES IN HUMAN SEMINAL FLUID OF WORKERS EXPOSED TO TRICHLOROETHYLENE.

P. Forkert[1], L. Lash[2], R. Tardif[3], N. Tanphaichitr[4], C. VanderVoort[5] and M. Moussa[6]. *[1]Anatomy & Cell Biology, Queen's University, Kingston, ON, Canada, [2]Department of Pharmacology, Wayne State University School of Medicine, Detroit, MI, [3]Department of Environmental and Occupational Health, University of Montreal, Montreal, QC, Canada, [4]Hormones/Growth/Development Research Group, Ottawa Health Research Institute, Ottawa, ON, Canada, [5]California National Primate Research Center, University of California, Davis, CA and [6]Department of Pathology, London Health Sciences Centre, London, ON, Canada.*

We have investigated the potential of the male reproductive tract to accumulate TCE and its metabolites including chloral, trichloroethanol (TCOH), trichloroacetic acid (TCA)and dichloroacetic acid (DCA). Human seminal fluid and urine samples from eight mechanics diagnosed with clinical infertility and exposed to TCE occupationally were analyzed. In *in vivo* studies, TCE and its metabolites were determined in epididymis of mice exposed to TCE (1000 ppm) by inhalation for 1 to 4 weeks. In other studies, incubations of monkey epididymal microsomes were performed in the presence of TCE and NADPH. Our results showed that seminal fluid from all 8 subjects contained TCE, chloral and TCOH. DCA was present in samples from 2 subjects, and only 1 contained TCA. TCA and TCOH were also identified in urine samples from 2 subjects. TCA, chloral and TCOH were detected in murine epididymis after exposure to TCE for 1 to 4 weeks. Levels of TCE and chloral were similar throughout the exposure period. TCOH levels were similar at 1 and 2 weeks, but increased significantly after 4 weeks. Chloral was identified in microsomal incubations with TCE in monkey epididymis. CYP2E1, a P450 enzyme with a major role in TCE metabolism, was localized in human and monkey epididymal epithelium and testicular Leydig cells. These results indicated that TCE is metabolized in the reproductive tract of the mouse and monkey. Furthermore, TCE and its metabolites were identified in seminal fluid, and suggested associations between TCE metabolites, reproductive toxicity and impaired fertility. Supported by Health Canada/Environment Canada.

1330 2, 3, 7, 8-TETRACHLORODIBENZO-*P*-DIOXIN (TCDD) INHIBITS PROSTATIC EPITHELIAL BUD FORMATION IN THE UROGENITAL SINUS (UGS) OF C57BL/6J MICE VIA MESENCHYMAL BUT NOT EPITHELIAL ARYL HYDROCARBON RECEPTOR (AHR).

R. W. Moore, K. Ko, N. T. Rasmussen and R. E. Peterson. *School of Pharmacy, University of Wisconsin at Madison, Madison, WI.*

We previously reported that *in utero* TCDD exposure prevents prostatic epithelial buds from forming in the ventral region of the UGS and reduces the number of buds formed in the dorsal and lateral regions of the UGS. These effects of TCDD