ENVIRONMENTAL WORKING GROUP

1436 U Street NW, Suite 100
Washington, DC 20009
T: 202.667.6982
F: 202.232.2592

March 28, 2005

Dr. Suhair Shallal
U.S. Environmental Protection Agency
Science Advisory Board Staff Office
1200 Pennsylvania Avenue, NW
Washington, DC 20460

BY FAX: 202-233-0643

Dear Dr. Shallal:
We are writing to express concern and alarm at what we understand to be DuPont's efforts to inappropriately influence the procedures and alter the conclusions of the Environmental Protection Agency's (EPA) Science Advisory Board (SAB, or the Board) for the Teflon chemical known as C8, PFOA, perfluorooctanoic acid and other names.

From information recently posted on EPA's website, we have learned that DuPont has requested that the Science Advisory Board delay issuing its report until the company releases some new, internal research. DuPont filed this request in new comments submitted for the Board's review, two weeks after the public discussion period, in an attempt to circumvent the open and public process to which the Board is bound.

By requesting that the Board delay issuing its report, DuPont is asking the Board to make a political decision. This is unacceptable and directly contradicts the Board's mandate, which is "to review and comment on the scientific soundness" of your Agency's PFOA human health risk assessment. In addition to this request for a delay, DuPont also provided scientific information that has not been subject to public scrutiny: the information was filed two weeks after the public SAB meeting and your Agency's comment deadline. We are compelled to clarify some of the new information DuPont has provided, and we do so in an addendum to this letter.

DuPont's stated reason for asking the Board to delay its report is that there are studies of interest that may be reported on within the year. More studies will always be forthcoming. At least six PFOA toxicity and exposure studies have been published in the last month alone (included as attachments). If the Board stops to wait for each new study, it will never complete its review, the EPA will never complete its risk assessment, and the American public will never be protected from this indestructible, toxic chemical that pollutes 95% of Americans' blood.

On the second day of the February Board meeting, EPA staff reminded Board members that their charge is to evaluate only the science. The Board's mandate has not changed. The Agency erred in accepting comments after the public discussion period and comment deadline; should the Board acquiesce to DuPont's request for delay contained in those comments, it would compound the EPA's error and discredit the Agency's and the Board's important work. Please do not allow DuPont to politicize this important assessment.

Sincerely,

Timothy J. Kropp, PhD
Senior Scientist

Jane Houlihan
Vice President for Research

Addendum, attachment: 10 pgs.

SAB PFOA Review Panel:
Dr. Deborah Cory-Slechta
Dr. Ernest Abel
Dr. Melvin Andersen
Dr. Germaine Buck Louis
Dr. George Corcoran
Dr. Norman Drinkwater
Dr. William L. Hayton
Dr. Michael A. Kamrin
Dr. James Kehrer
Dr. James E. Klaunig
Dr. Matthew P. Longnecker
Dr. Ronald Melnick
Dr. Franklin L. Mink
Dr. David M. Ozonoff
Dr. Stephen Roberts
Dr. Anne Sweeney
Dr. Thomas T. Zoeller

EPA:
Dr. Charles Auer
Dr. Jennifer Seed

ADDENDUM

SUPPLEMENTAL COMMENTS TO
THE SCIENCE ADVISORY BOARD PFOA REVIEW PANEL

By Timothy Kropp and Jane Houlihan
Environmental Working Group; Washington, D.C.

March 25, 2005

Summary. The purpose of this submission is to clarify issues raised in comments submitted by DuPont to the Science Advisory Board PFOA Review Panel on March 9th, 2005 (Rickard 2005). We are compelled to submit these supplemental comments to the Panel because the Environmental Protection Agency circumvented the normal process that allows for public scrutiny of all comments submitted to the Panel, by accepting DuPont's comments two weeks after the comment deadline and the Panel meeting.

In their new comments, DuPont has asked the Panel to delay completing its review of PFOA pending forthcoming DuPont occupational studies, and has also presented new interpretations of existing data. DuPont has asked the Panel to discount statistically significant mammary tumors and pituitary effects, and to discount immune system effects.

In the case of the requested delay, DuPont is asking the Panel to make a political decision that is outside the Panel's mandate. We ask that the Panel not acquiesce to DuPont's inappropriate request. In suggesting that the Panel discount statistically significant effects, DuPont is relying on a range of justifications that fail to meet the standards of sound science, including the use of historic control data contrary to EPA guidelines, and the unjustifiable lumping of data from multi-generational studies that obscures study findings. We ask that the Panel consider the failings in DuPont's new data interpretations that we present below.

Health outcomes observed in occupational settings associated with exposure to PFOA. We understand from DuPont's most recent submission to the Panel (Rickard 2005) that the company is conducting occupational studies and has asked the Panel to delay issuing its report until these studies have been published.

These studies are a much-needed component of worker safety programs at DuPont facilities. But the need for these studies is a separate issue from DuPont's request for delay, which for the Panel would be a political decision, not a scientific one. We ask the Panel to recognize that studies on PFOA will continue to be published for decades to come. At least six significant PFOA toxicity and exposure studies have been published in the last month alone (abstracts are attached). Occupational studies like those currently being conducted by DuPont often inform, but rarely drive, risk evaluations for the general population (the subject at hand for this panel), in cases where the bulk of available data comes from laboratory studies.

Despite their constrained utility in evaluating general population risks, the occupational studies conducted to date do in fact, provide evidence that bolsters findings from the PFOA laboratory studies that form the basis of EPA's risk evaluations (EPA 2005). We mention some of the study findings below because they contradict DuPont's assertion to the Panel that PFOA has not been linked to human health effects (Rickard 2005). To the contrary, at least five studies provide evidence of potential health effects associated with exposure to PFOA in occupational settings:

- **Vascular disease:** Statistically significant, elevated risk of dying from cerebrovascular disease was seen among workers exposed to PFOA versus those not exposed occupationally, at 3M's Cottage Grove, MN plant (Alexander 2001). Findings of vascular disease have been confirmed in a study of DuPont workers exposed to PFOA. SMRs were statistically significant and adjusted for age and gender for rheumatic health disease (SMR = 3.55, 95%CI 1.14-8.3) and atherosclerosis and aneurysm (SMR = 1.98, 95%CI 1.17-3.14) (DuPont, 2003). This evidence directly contradicts DuPont's claim of no human health effects. Furthermore, a statistically significant correlation between cholesterol and PFOA was found in a DuPont study of worker blood (DuPont 2005) and a study of PFOA exposed Italian workers (Shea, 2004). These studies are bolstered by evidence from two animal studies showing changes in blood chemistry associated with PFOA exposures (Goldenthal et. al. 1978; Haughom and Spydevold 1992).

- **Prostate cancer:** Statistically significant association between the length of workplace PFOA exposure and prostate cancer mortality was seen in workers at 3M's Cottage Grove plant. The relative risk per 1 year of employment was 1.13 (95% CI 1.01 to 1.27) whereas the risk was 3.3 (95% CI 1.02-10.6) among 3M workers exposed to PFOA for at least 10 years, compared to the general population (Gilliland and Mandel 1993). When workers were divided into smaller exposure groups, an elevated risk of dying from prostate cancer was found among workers exposed to PFOA (SMR = 1.34), though the 95 percent confidence interval (95%CI) included 1 (Alexander 2001). A non-significant increase in episodes of care for male reproductive cancers (including prostate) was seen at 3M's Decatur, Alabama plant among all workers compared to general population, with greatest increases among those in the long-time, high-PFOA-exposure category (Olsen et al. 2001).

- **Leukemia:** Two analyses of leukemia incidence were conducted showing statistically increased odds ratios (2.0 to 8.0, depending on the exposure type) for workers in the Washington Works plant from 1956-1989. The general mortality study (DuPont, 2003) did find an increase in leukemia (Standardized Incidence Ratio = 2.02) though not significant, possibly due to the segregated analysis of different types of leukemias, as suggested by the EPA (EPA, 2005).

- **Cancer of the large intestine:** Significantly increased numbers of episodes of care for intestinal tumors among workers exposed to perfluorochemicals versus those not exposed occupationally were seen at 3M's Decatur, AL plant (Olsen et al., 2001). An elevated yet not significant increase of risk of dying from cancer of the large intestine was seen in those definitely exposed to PFOA compared to the general population in 3M's Cottage Grove, MN plant (Alexander 2001).

- **Pancreatic cancer:** An elevated, yet non-significant risk of dying from pancreatic cancer or pancreatic disease was seen among workers exposed to PFOA versus those not exposed occupationally, at 3M's Cottage Grove, MN plant (Alexander 2001).

- **Testicular cancer**: An elevated, yet non-significant risk of dying from cancer of the testis or other male reproductive cancers was seen among workers exposed to PFOA versus those not exposed occupationally, at 3M's Cottage Grove, MN plant (Alexander 2001).

Taken together, these findings show disease patterns strikingly similar to findings from laboratory studies.

Mammary tumors. As you know, a 3M-sponsored study found a statistically significant increase in fibrademonas with PFOA dose, with tumor incidence increasing from 22 to 42 and then 48 percent of the animals in the 0 (controls), 30, and 300 ppm (in diet) dose groups, respectively (Sibinski, 1987). DuPont argues in its most recent submission to this panel that this statistically significant increase should be discounted because the study tumor rates fall within the maximum tumor rate ever detected in individual study control groups from all available studies for this strain of rat. This data interpretation fails to meet EPA guidance on the appropriate use of historic control data, and is not scientifically defensible.

We want to reiterate that EPA's guidance on the appropriate use of historic control data dictates that comparisons be constrained to only that historical data available from the same laboratory and supplier within two to three years of the study in question (EPA 2003). DuPont's use of historic controls with respect to mammary tumors fails both these requirements. Using the maximum rate ever found in any laboratory has never been the Agency's accepted standard. And even using a related metric such as an *average* incidence in controls from all available historic studies has not won Agency acceptance, because genetic drift over time and variability in lab conditions combine to create significant differences in tumor rates between various control groups. Certainly the *maximum* tumor incidence ever detected in controls in any study in any laboratory is not an appropriate metric for discounting statistically significant results, as DuPont has argued.

DuPont also argues that the mammary tumor findings should be discounted because the incidence of tumors that have progressed to a malignant state does not show an increasing dose-response relationship. Both fibroadenomas and total tumors *do* increase with dose, however, and a final progression to malignancy has never been the accepted criteria for significance. A statistically significant increase in tumors, whether progressed to malignancy or not, is certainly still an adverse finding.

Pituitary Effects. In its March 9th comment submission, DuPont argues that the statistically significant decrease in pituitary weight with PFOA dose in the F1 generation is "spurious and not related to administration of PFOA." As justification, DuPont argues that the effect in the F1 generation is spurious because it was not observed in the F0 generation. DuPont then lumps all pituitary weight data from the F0, F1, and F2 generation together and shows that when taken together the average, absolute pituitary weight does not appear to change with dose. These justifications are baffling, given that the primary purpose of conducting a multi-generation study is to allow for differing effects across generations to be discerned. As we showed in previous comments to the Board, the response shown by the pituitary in the F1 generation mirrors the form of the response for other adverse effects from PFOA exposure; the results are statistically significant, adverse, and should not be discounted for the spurious reasons presented in Rickard (2005).

Immunological function. DuPont asserts that PFOA does not produce toxicity to the immune system. To date, five studies have investigated immunotoxicity with respect to PFOA. The first (Yang et al., 2000) found that PFOA decreases thymus and spleen weight and the number of thymocytes and splenocytes. The second (Yang et al., 2001) found spleen and thymus weights decreased in response to PFOA exposure for more than 5 days. The third (Yang et al., 2002a), done in PPARα-null mice, found a decrease in the overall weight of the thymus as well as

specific cell types. The fourth (Yang et al., 2002b) found that PFOA potently inhibits humeral immune responses in mice. The latest, a study performed by NIOSH and presented at the Society of Toxicology meeting this month in New Orleans, found that "PFOA was demonstrated to be immunotoxic in a murine model following dermal exposure" (Fairley et al., 2005). Contrary to DuPont's assertion, immunotoxicity is clearly an endpoint of concern and should be addressed by EPA's risk assessment and by additional research as well.

References

Alexander, B. 2001. Mortality study of workers employed at the 3M Cottage Grove Facility. Final Report. Division of Environmental and Occupational Health, School of Public Health, University of Minnesota. AR 226-1136. Washington, DC: U.S. Environmental Protection Agency.

Dupont. 2003. Epidemiology surveillance report: Cancer incidence for Washington Works site 1959-2001. USEPA AR 223-1307.

DuPont 2005. DuPont Reports First-Phase Results of Health Study Examining PFOA Exposure http://www1.dupont.com/NASApp/dupontglobal/corp/index.jsp?page=/content/US/en_US/news/releases/2005/nr01_11_05a.html.

Fairley KJ, Kearns S, Myers LP, Purdy R, Meade BJ. Augmentation of ovalbumin – induced ige and airway hyperreactivity response by perfluorooctanoic acid (PFOA). Abstract No. 1210. 2005 Itinerary Planner. New Orleans, LA: Society of Toxicology.

Gilliland RD, Mandel JS. 1993. Mortality among employees of a perfluorooctanoic acid production plant. Journal of Occupational Medicine 35: 950-4.

Goldenthal EI, Jessup DC, Geil RG, and Mehring JS. 1978. Ninety-day subacute rhesus monkey toxicity study: Fluorad ¨ Fluorochemical FC-143. Report prepared for 3M, St. Paul, MN by Institutional Research and Development Corporation (Mattawan, MN). Study No. 137-090. AR226-1136. Washington, DC: U.S. Environmental Protection Agency.

Haughom B, Spydevold O. 1992. The Mechanism Underlying the Hypolipemic Effect of Perfluorooctanoic Acid (PFOA), Perfluorooctane Sulphonic Acid (PFOSA) and Clofibric Acid. Biochimica et biophysica acta. 1128(1): 65-72.

Olsen GW, Burlew MM., Hocking BB, Skratt JC, Burris, JM and Mandel, JH. 2001. An epidemiologic analysis of episodes of care of 3M Decatur chemical and film plant employees, 1993-1998. Final report. May 18, 2001.

Shea EE. 2004. Report on the meeting held on Friday 20th and Saturday 21st 2004 at the Inn at Montchanin Village (Wilmington, USA) with 3M and DuPont delegations. OPPT TSCA 8(e) docket #8EHQ-0904-15663.

Sibinski, LJ. 1987. Two-Year oral (diet) toxicity/carcinogenicity study of fluorochemical FC-143 (perfluorooctane ammonium carboxylate) in rats. Report prepared for 3M, St. Paul, Minnesota by Riker Laboratories Inc. Study No. 0281CR0012; 8EHQ-1087-0394, October 16, 1987.

U.S. EPA (U.S. Environmental Protection Agency). 2003. Draft final guidelines for carcinogen risk assessment (external review draft, February 2003). Available: http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=55445.

U.S. EPA (U.S. Environmental Protection Agency). 2005. Draft Risk Assessment of the potential human health effects associated with exposure to perfluorooctanoic acid and its salts.

Yang, Q, Xie, Y, and DePierre, JW. 2000. Effects of peroxisome proliferators in the thymus and spleen of mice. Clin. Exp. Immunol. 122: 219-226.

Yang, Q, Xie, Y, Eriksson, AM, Nelson, BD and DePierre, JW. 2001. Further evidence for the involvement of inhibition of cell proliferation and development in thymic and splenic atrophy induced by the peroxisome proliferator perfluorooctanoic acid in mice. Biochem Pharmacol 62(8): 1133-40.

Yang, Q, Xie, Y., Alexson, SE, Nelson, BD and DePierre, JW. 2002a. Involvement of the peroxisome proliferator-activated receptor alpha in the immunomodulation caused by peroxisome proliferators in mice. Biochem Pharmacol 63(10): 1893-900.

Yang, Q, Abedi-Valugerdi, M, Xie, Y, Zhao, XY, Moller, G, Nelson, BD and DePierre, JW. 2002b. Potent suppression of the adaptive immune response in mice upon dietary exposure to the potent peroxisome proliferator, perfluorooctanoic acid. Int Immunopharmacol 2(2-3): 389-97.

Attachment

Abstracts from six peer-reviewed papers on PFOA toxicity and exposure published within the last month are provided below.

PFOA INDUCES DYSMORPHOGENESIS IN MOUSE WHOLE EMBRYO CULTURE.

M.R.Blanton1; J.M.Padowski2; S.S.Hunter1; J.M.Rogers1; C.Lau1. 1. RTD, NHEERL, ORD, USEPA, RTP, NC; 2. Curriculum in Toxicology, UNC, Chapel Hill, NC.

Perfluorooctanoate (PFOA) is a perfluoroalkyl acid (PFAA) found in numerous industrial and consumer products. Many PFAAs persist in the environment and are found in humans and animal tissues throughout the world. PFOA is a developmental toxicant in vivo producing embryonic, fetal and postnatal lethality, and altered fetal and neonatal growth. In order to determine if direct exposure of conceptuses to PFOA disrupts development, CD-1 mouse conceptuses (3-6 somite stage, GD8) were exposed to PFOA in whole embryo culture. At the end of a 24H culture period embryonic morphology was assessed and scored using a scoring system developed by our laboratory. Scores ranged from 0, for morphologically normal, to a maximum of 58 for severely affected embryos. In control medium, 82% of embryos (18/22) grew normally and the mean score was 0.40.2. Exposure to 0.1 or 0.2mg/ml PFOA did not alter development (4/4 and 11/17, normal embryos respectively). However, dysmorphogenesis was induced by PFOA at 0.4 (82%), 0.6 (92%), 0.75(100%) and 1.0 (100%) mg/ml. Exposure to 1.25mg/ml produced 100% embryolethality. Prosencephalic and pharyngeal arch hypoplasia and abnormal heart outflow tract development were induced by >0.4mg/ml PFOA. Embryonic scores were increased at PFOA concentrations >0.4mg/ml and were 0.00.0(0.1), 1.70.7(0.2), 11.32.2(0.4), 11.32.0(0.6), 23.51.5(0.75) and 31.13.9(1.0). The benchmark concentration for a 5% increase in dysmorphic embryos by PFOA was 0.040.01mg/ml. These studies demonstrate that a direct exposure to PFOA for 24H disrupts development and induces embryolethality. This abstract does not represent EPA policy.

Citation: M.R.Blanton, J.M.Padowski, S.S.Hunter, J.M.Rogers, C.Lau. PFOA INDUCES DYSMORPHOGENESIS IN MOUSE WHOLE EMBRYO CULTURE.. Abstract No. 2255. *2005 Itinerary Planner.* New Orleans, LA: Society of Toxicology

---

EFFECTS OF PERFLUOROOCTANOIC ACID EXPOSURE DURING PREGNANCY IN THE MOUSE

J.Thibodeaux; R.G.Hanson; J.M.Rogers; C.Lau. RTD, NHEERL, ORD, US EPA, Research Triangle Park, NC.

Perfluorooctanoic acid (PFOA), a member of the perfluoroalkyl acids that have wide commercial applications, has recently been detected in humans and wildlife. The current study characterizes the developmental toxicity of PFOA in the mouse. CD-1 mice were mated overnight and the day on which copulatory plugs were found was designated as GD 1. Animals were given 1, 3, 5, 10, 20, or 40 mg/kg PFOA by oral gavage daily from GD2 to GD 18; controls received an equivalent volume (10 ml/kg) of water. PFOA treatment produced early, full-litter

resorptions in a dose-dependent manner, leading to 100% loss in the 40 mg/kg group. Significant deficits in maternal weight gains were detected in dams of the 20 mg/kg group that carried pregnancy to term. On GD 18, some dams were sacrificed for maternal and fetal examinations while the rest were allowed to give birth for monitoring of postnatal survival, growth, and development of the pups. PFOA induced liver enlargement in the dams at all doses, but did not alter the number of implantations. In the 20 mg/kg group, the percent of live fetuses was reduced to 74% (compared to 97% in controls) the day before birth, and fetal weight (0.86 0.11 g) was significantly lower than that of controls (1.05 0.02 g). However, no discernable anatomical malformations were noted in any of the treatment groups. The incidence of live births was significantly lowered by PFOA in the 10 and 20 mg/kg groups (70% compared to 93% in the control group), and postnatal survival of the neonates was severely compromised in these treatment groups. Dose-dependent deficits of weight gains were detected in all PFOA-treated pups. Significant delays in eye-opening (by about 2 days) were noted in the 10 and 20 mg/kg groups, and pubertal indices were significantly delayed (by about 3 days in males and females) in the 20 mg/kg group. These data indicate maternal and developmental toxicities of PFOA in the mouse measured as early pregnancy loss, compromised postnatal survival, and delays in postnatal growth and maturation. This abstract does not necessarily reflect U.S. EPA policy.

Citation: J.Thibodeaux, R.G.Hanson, J.M.Rogers, C.Lau. EFFECTS OF PERFLUOROOCTANOIC ACID EXPOSURE DURING PREGNANCY IN THE MOUSE. Abstract No. 280. *2005 Itinerary Planner*. New Orleans, LA: Society of Toxicology

---

AUGMENTATION OF OVALBUMIN - INDUCED IGE AND AIRWAY HYPERREACTIVITY RESPONSE BY PERFLUOROOCTANOIC ACID (PFOA)

K.J.Fairley1; S.Kearns1; L.P.Myers1; R.Purdy2; B.J.Meade1. 1. Agriculture and Immunotoxicology Group, NIOSH, Morgantown, WV; 2. Independent Toxicologist, River Falls, WI.

Studies were conducted to investigate the role of dermal exposure to Perfluorooctanoic acid (PFOA), an immunosuppressant with widespread use as a carpet and fabric protectant, on the hypersensitivity response to ovalbumin in a murine model. BALB/c mice were exposed dermally to concentrations of PFOA ranging from 0.01-2.0% (0.25-50mg/kg) for 4 days. In hypersensitivity studies, mice were also intraperitoneally injected with 7.5g ovalbumin and 2mg alum on days 1 and 10 and in some studies, intratracheally challenged with 250g ovalbumin on days 17 and 26. Endpoints for studies included body and organ weights and cellularities, IgE, airway hyperreactivity, and lung histopathology. Following exposure to PFOA, an increase in liver weights and a decrease in thymus and spleen weights and cellularities were observed. Similar immunomodulatory trends were demonstrated in mice co-administered PFOA and ovalbumin. Greater than a 2-fold increase in total IgE was demonstrated when mice were co-exposed with concentrations of PFOA ranging from 0.75-1.5%, while the ovalbumin-specific IgE response peaked after a 3-fold increase ($p<0.01$) in 0.75% PFOA co-exposed animals as compared to the ovalbumin alone exposed animals. Antigen-specific airway hyperreactivity was increased ($p<0.05$) in the in the 1.0% PFOA co-exposed group, with a dose-responsive pleiotropic cell response characterized by eosinophilia and mucin production, in animals co-exposed to concentrations of PFOA up to 1.0%, as compared to the ovalbumin alone exposed

animals. PFOA was demonstrated to be immunotoxic in a murine model following dermal exposure, with an enhancement of the hypersensitivity response to ovalbumin, suggesting that PFOA exposure may augment the IgE response to environmental allergens.

Citation: K.J.Fairley, S.Kearns, L.P.Myers, R.Purdy, B.J.Meade. AUGMENTATION OF OVALBUMIN - INDUCED IGE AND AIRWAY HYPERREACTIVITY RESPONSE BY PERFLUOROOCTANOIC ACID (PFOA). Abstract No. 1210. *2005 Itinerary Planner.* New Orleans, LA: Society of Toxicology

---

EFFECT OF PERFLUOROOCTANE SULFONATE (PFOS) AND PERFLUOROOCTANOATE (PFOA) ON L - TYPE CALCIUM CURRENT IN GUINEA PIG VENTRICULAR MYOCYTES.

K.Harada1; F.Xu2; K.Ono2; T.Iijima2; A.Koizumi1. 1. Health Environmental Sciences, Kyoto University Graduate School of Medicine, Kyoto, Japan; 2. Pharmacology, Akita University School of Medicine, Akita, Japan.

Perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) are chemicals ubiquitously found in the environment, wildlife and humans. Their toxicity is still unknown. There are several reports that PFOS and PFOA may change physiological properties of cell membranes. These effects on membrane properties, however, have not been investigated from viewpoint of cellular functions. We herein show the effect of PFOS and PFOA on L-type calcium current in isolated guinea pig ventricular myocytes using the whole cell patch-clamp recording technique. PFOS (1, 3, 10, 30, 100 M) increased the voltage-activated peak amplitude of I(Ca, L) in a concentration-dependent manner. PFOS decreased the maximal activation voltage of I(Ca,L) (EC50 = 6.7 M). PFOS shifted the steady-state inactivation curve of I(Ca,L) to the left (EC50 = 12.6 M). PFOA also had the same effects of PFOS but its dose-response curves shifted to the higher doses by about 10-fold larger than those of PFOS. These effects were also observed in sodium dodecyl sulfate, which is known to change surface charge, suggesting the importance of amphiphilic nature for the effects on Ca-channel. In this study, perfluoroalkyl acids were considered to alter the L-type calcium channel kinetics in ventricular myocytes through the change of surface charge. This surface charge alteration may modify physiological properties of other channels, leading perturbation of signal transduction through various ion channels by PFOS and PFOA. Further studies are needed to bridge over a gap between in vitro finding and in vivo toxicity.

MAMMALIAN METABOLISM AND DISTRIBUTION OF PERFLUOROOCTYL ETHANOL AND ITS OXIDATION METABOLITES

W.M.Henderson1,2; E.J.Weber2; S.E.Duirk2; M.Smith1. 1. Interdisciplinary Toxicology Program, University of Georgia, Athens, GA; 2. NERL, USEPA, Athens, GA.

Perfluorinated compounds (PFCs) have been shown to be globally distributed, bioaccumulative, persistent and potentially toxic. It has been hypothesized that many precursor fluorinated compounds, including the telomer alcohols, degrade or metabolize to the common metabolite PFOA. Currently, little information is available on the mammalian metabolism of the telomer

alcohols. Both and oxidation pathways are proposed, resulting in the formation of perfluorononanoic acid (PFNA) and perfluorooctanoic acid (PFOA), respectively. An animal model is needed to fully elucidate the mechanisms of metabolism and translocation as it relates to humans. The objective of this study was to investigate the distribution and metabolism of the telomer alcohol in mice. Male CD-1 mice received a gavage dose of 20 mg 8-2 telomer alcohol/kg BW in a propylene glycol/water (1:1) vehicle. Animals were serially sacrificed at 1, 3, 6, 12 and 24 hours post-treatment and organs excised for PFC analysis. Telomer alcohol and metabolite concentrations were analyzed with EI-GC/MS. Based on preliminary studies, the telomer alcohol temporally decreased becoming non-detectable by 12 hours in both liver and serum. In liver, the telomer alcohol was seen at peak concentrations at 1 hour and in serum was quantifiable only at 1 and 3 hours. In serum the most abundant metabolite was PFOA with a concentration of 186-212 ng/mL at 6 hours post-treatment and decreased by 22% at 12 and 24 hours. In liver, PFOA is also the major metabolite reaching peak concentrations (38-44 ng/g) at 12 hours post-treatment. The highest concentration of PFNA coincides with peak PFOA concentrations and both metabolites decreased at 24 hours. In conclusion, the appearance of PFNA in the liver suggests that oxidation occurs although it does not appear to be the predominant metabolism pathway. However, these results suggest the telomer alcohol is metabolized by both and oxidation pathways. Analysis of the brain, kidney, spleen, gonads and fat is ongoing.

Citation: W.M.Henderson, E.J.Weber, S.E.Duirk, M.Smith. MAMMALIAN METABOLISM AND DISTRIBUTION OF PERFLUOROOCTYL ETHANOL AND ITS OXIDATION METABOLITES. Abstract No. 2065. *2005 Itinerary Planner*. New Orleans, LA: Society of Toxicology

---

ROLONGATION OF K+ - INDUCED BACKWARD SWIMMING OF *PARAMECIUM CAUDATUM* BY PFOS AND PFOA

Y.Yamazaki; K.Harada; A.Morikawa; T.Yoshinaga; A.Koizumi. Health Environmental Sciences, Kyoto University Graduate School of Medicine, Kyoto, Japan.

Paramecium caudatum is a protozoan and has been used to investigate the effects of chemicals on physiological processes. When cells are transferred to a K+-rich solution, increase in Ca2+ conductance by stimulation of Ca2+-channel, results in initiation of backward swimming. Cells begin whirling in consequence, when the increase in Ca2+ conductance is attenuated. Thus if a given chemical has some effects on the processes of Ca2+-channel kinetics, a series of swimming behavior will be modified. In the present study, we investigated the effects of perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) on K+-induced swimming behavior. After cultivating for four days, cells were transferred in K+-rich solution [20mM KCl, 1mM CaCl2 and 1mM Tris-HCl buffer (pH 7.2)], PFOS solutions (1, 10 and 100 ppm in K+-rich solution) and PFOA solution (1, 10 and 100 ppm in K+-rich solution) and were observed with a stereoscopy. In comparison with a period in K+-rich solution (MSE, sec) [25.51.6 (n=10)], backward swimming was significantly prolonged by PFOS: 60.78.8 at 1ppm (n=10), 162.420.3 at 10 ppm (n=10). PFOA prolonged significantly only at 100 ppm [65.853.0 (n=10)]. At 100 ppm PFOS was lethal and all cells died within 5 min. We further investigated the effect of a typical anionic amphiphile, sodium dodecyl sulfate: it also prolonged backward swimming at 10 ppm [317.231.0 (n=5)]. The present study clearly demonstrated that PFOS has effects on Ca2+ conductance at pharmacological dose levels likely through an action on the cellular membrane. Since the Ca2+ signaling plays important roles in homeostasis, PFOS may perturb cellular

functions through Ca2+ signaling.

Citation: Y.Yamazaki, K.Harada, A.Morikawa, T.Yoshinaga, A.Koizumi. PROLONGATION OF K+ -INDUCED BACKWARD SWIMMING OF *PARAMECIUM CAUDATUM* BY PFOS AND PFOA. Abstract No. 2067. *2005 Itinerary Planner*. New Orleans, LA: Society of Toxicology