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CERTIFIED MAIL
Document Processing Center (7407)
Attn: Section 8 (e) Coordinator
Office Toxic Substances
US Environmental Protection Agency
410 M Street, SW
Washington, DC 20460

Re: TSCA 8 (e) SUPPLEMENTAL SUBMISSION: Docket No. 8EHQ-998-374
Perfluorooctane Sulfonate (PFOS): CAS# 2795-39-3

Dear Docket Coordinator:

3M has previously informed the EPA of the results of a multi-generation reproductive/ developmental study in rats conducted with potassium perfluorooctanesulfonate (PFOS) in which perinatal mortality occurred in the F_1 generation pups at the two highest dose levels (1.6 and 3.2 mg/kg/day). A cross-foster study conducted following the multi-generation study demonstrated that the perinatal mortality was primarily due to direct toxicity to the pups, presumably from exposure *in utero*. In order to develop an understanding of the mechanism for the observed toxicity in rats and to confirm and more precisely define the No-Observed-Effect Levels (NOEL), 3M is in the process of completing an additional study. This letter provides preliminary data from the study, supplementing our prior filing.

Findings of the study for various dose groups under different conditions are described in the attachment to this letter. To test the hypothesis that PFOS may cause reductions in cholesterol synthesis and serum lipids which might in turn contribute to the observed perinatal mortality, this study examined whether there is a dose-response relationship between perinatal mortality and serum lipids and whether there is a protective effect of maternal supplementation with mevalonic acid or cholesterol. Serum was collected to measure total cholesterol, high-density lipoprotein, low-density lipoprotein, triglycerides and glucose as well as PFOS concentration. Liver tissue was collected and frozen for determination of PFOS concentrations and other possible endpoints. In addition, due to reports in the scientific literature associating severe hypothyroidism with perinatal toxicity, data on thyroid hormone concentrations were obtained where serum volumes permitted analysis. Thyroid glands and hearts were also collected for possible histopathological evaluation.

As is evident from the attached summary, the preliminary information from this study indicates:

- Perinatal mortality in rats was increased relative to controls at PFOS doses of 0.8 mg/kg/day and higher, corroborating the previous finding of 0.4 mg/kg/day as a NOEL for perinatal effects under the dosing conditions used in these studies (dosing six weeks prior to co-habitation and through gestation and lactation).

- Reductions in serum lipids and cholesterol synthesis do not appear to play a significant role in the causation of the observed perinatal mortality in rats.

- PFOS exposure at doses of 0.4 mg/kg/day and above in rats caused reductions in measured free and total thyroxin (T4) as well as free and total triiodothyronine (T3) and variable responses of thyroid stimulating hormone (TSH). (We have verified that PFOS does not interfere with the measurement of these thyroid hormones.)

The relationship, if any, of the observed perinatal mortality from PFOS exposure to the observed decreases in T3 and T4 is not clear at this time. Further work is in progress to better understand the biological relevance of the observed reductions in T3 and T4 in rats from PFOS exposures.

3M has not observed any statistically significant effect on lipid or thyroid hormone levels in medical monitoring studies of PFOS-exposed workers.

Please contact me at 651-733-5181 for further information.

Sincerely,

LR Zobel MD

Larry R. Zobel, M.D., M.P.H.
Staff Vice President and Medical Director

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Summary of
3M Study of Possible Mechanism of Perinatal Mortality
in Rat Pups Associated with Maternal Exposure to
Potassium Perfluorooctane Sulfonate (PFOS) in Rats
(based on preliminary data)

The study was conducted to better define the dose-response and to explore possible mechanisms of perinatal mortality observed in the prior multi-generation reproduction study in rats.

Experimental Design:

Groups of Female Rats

Control Groups	**Dosed Groups**
Vehicle Control	0.4, 0.8, 1.0, 1.2, 1.6 and 2.0 mg/kg/day PFOS
Cholesterol (500 mg/kg/day) Control	1.6 and 2.0 mg/kg/day PFOS plus Cholesterol (500 mg/kg/day)
Mevalonate (500 mg/kg/day) Control	1.6 and 2.0 mg/kg/day PFOS plus Mevalonate (500 mg/kg/day)

Dosing - oral intubation, 42 days prior to cohabitation with untreated males (mating) and throughout gestation and five days of lactation.

At day 21 of gestation, fetuses from eight litters in the 0, 1.6, and 2 mg/kg dose groups were taken by caesarean section. All remaining rats were allowed to give birth and pups and dams were sacrificed at day 5 of lactation.

At both time points, total serum cholesterol, HDL, LDL, TG, glucose, free and total T3, free and total T4, and TSH were determined in fetuses/pups and dams.

Results:

Viability Indices

PFOS Dose mg/kg/day	Viability Index (% survival at Day 5)
No Supplementation	
0	97.7
0.4	97.6
0.8	93.5
1.0	89.8
1.2	82.5
1.6	51.1
2.0	17.9
Cholesterol Supplementation (500 mg/kg/day)	
0	98.4
1.6	42.8
2.0	15.8
Mevalonate Supplementation (500 mg/kg/day	
0	98.7
1.6	41.4
2.0	2.6

Serum Lipids and Glucose

	CHOL[a]	HDL[b]	LDL[c]	TG[d]	GLU[e]
			No Supplementation		
Dam GD[f] 21	↓2.0[g]	-	↑2.0	-	-
Fetus GD 21	↑1.6 & 2.0	↑1.6	↑1.6 & 2.0	↓1.6 & 2.0	↑1.6 & 2.0
Dam LD[h] 5	↓0.4 - 2.0	↓0.8-1.6	-	↓1.0-2.0	↑2.0
Pup LD 5	↑1.2 & 1.6	↑0.8 & 1.0	-	-	↓1.0
			Cholesterol Supplementation (500 mg/kg/day)		
Dam GD 21	-	↑1.6 & 2.0	-	↓1.6	-
Fetus GD 21	↑1.6 & 2.0	↑1.6 & 2.0	↑1.6 & 2.0	-	-
Dam LD 5	↓1.6 and 2.0	-	-	↓1.6 and 2.0	↑2.0
Pup LD 5	↑1.6 & 2.0	↑1.6 & 2.0	↑1.6 & 2.0	↓2.0	-
			Mevalonate Supplementation (500 mg/kg/day)		
Dam GD 21	-	-	↑1.6 & 2.0	-	-
Fetus GD 21	↑1.6 & 2.0	↑1.6 & 2.0	↑1.6 & 2.0	↓2.0	-
Dam LD 5	↓1.6 and 2.0	↓1.6 and 2.0	-	↓1.6	-
Pup LD 5[i]	↑1.6	-	↑1.6	↑1.6	-

a) Total serum cholesterol
b) High-density lipoprotein
c) Low-density lipoprotein
d) Triglycerides
e) Glucose
f) Gestation day
g) Reference is to dose groups: 0.4, 0.8, 1.0, 1.2, 1.6 or 2.0 mg/kg/day PFOS
h) Lactation day
i) No sample was obtained from 2.0 mg/kg/day litter pups
↓ or ↑ = statistically significant ($p < 0.05$) change compared to appropriate control
- = no statistically significant (p , 0.05) difference compared to appropriate control

Thyroid Effects

	TT4[a]	FT4[b]	TT3[c]	FT3[d]	TSH[e]
Dam GD[f] 21	↓1.6 & 2.0[g]	↓1.6 & 2.0	↓1.6 & 2.0	↓1.6 & 2.0	-
Fetus GD 21	NC[h]	NC[i]	-	NC[j]	NC[j]
Dam LD[k] 5	↓0.4-2.0	↓0.4-2.0	↓0.8-2.0	↓1.2-2.0	-
Pup LD 5	↓0.4-2.0	↓0.4-2.0	↓1.6-2.0	NC[l]	↓0.4 ↑1.6

a) Total thyroxin
b) Free thyroxin
c) Total triiodothyronine
d) Free triiodothyronine
e) Thyroid stimulating hormone
f) Gestation day
g) Reference is to dose groups: 0.4, 0.8, 1.0, 1.2, 1.6 or 2.0 mg/kg/day PFOS
h) No conclusions are possible as all values (including control) were below detection
i) No conclusions are possible (only one value from control was available)
j) No conclusions are possible (no control value was available)
k) Lactation day
l) No conclusions are possible (most values below detection)
↓ or ↑ = statistically significant ($p < 0.05$) change compared to appropriate control
- = no statistically significant (p , 0.05) difference compared to appropriate control

Results are not yet available for PFOS serum and liver concentrations and liver glycogen content.

Conclusions:

Supplementation with mevalonic acid or cholesterol had no positive effect on perinatal survival through lactation day 5. Thus, reduced serum lipid does not appear to be a factor in neonatal mortality. The data suggest reduced thyroid hormones could play a role in the observed perinatal mortality of this and prior studies, but the evidence is not conclusive.

The no-observed-effect level for perinatal mortality was determined to be 0.4 mg/kg/day