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February 10, 2005

U.S. Environmental Protection Agency Science Advisory Board (1400F)
Attention: Dr. Sue Shallal, Designated Federal Officer
1200 Pennsylvania Avenue, N.W.
Washington, D.C. 20460

Re: OPPT Docket 2002-0001

Re: Points of Departure for PFOA Risk Assessment

Dear Dr. Shallal:

The EPA SAB PFOA Review Panel has been charged to comment on the scientific soundness of OPPT's "Draft Risk Assessment of the Potential Human Health Effects Associated with Exposure to Perfluorooctanoic Acid (PFOA) and Its Salts" (January 4, 2005). These comments will focus on one issue the Review Panel is to address: Issue 3: Selection of Endpoints.

I have reviewed the PFOA data in the course of my work as an independent adviser to 3M and as co-author of a published risk assessment, Butenhoff, *et al.*, "Characterization of Risk for General Population Exposure to Perfluorooctanoate," *Regulatory Toxicology and Pharmacology* 39: 363-380 (2004a).¹ This publication reviews pertinent endpoints addressed in the toxicology database and provides benchmark dose calculations for a number of endpoints. A copy is enclosed for the Panel's reference.

¹ My career has been as a toxicologist with experience and expertise in developmental and reproductive toxicology and in risk assessment. I previously served as Assistant Administrator of EPA for Toxics and Pesticides (1983-1989), and prior to that was Deputy Director of the National Toxicology Program and Director of Toxicology Research & Testing, NIEHS, NIH.

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The EPA draft assessment evaluates both prenatal and adult life stages. The draft assessment presents margin of exposure calculations for a number of endpoints, using primate studies, adult rat studies, and developmental studies. My comments are limited to three particular endpoints. I urge reconsideration of the proposed endpoint selected to represent prenatal effects because the endpoint selected appears to reflect toxicity from direct dosing rather than a developmental effect. Second, I submit that the value selected for the assessment of adult female body weight is inappropriate. Finally, adult male body weight and other endpoints would be better addressed using a benchmark dose approach rather than no-effect levels.

In my view, the reduced body-weight gain seen in male pups dosed from weaning to sexual maturation is most plausibly due to a direct effect of the PFOA rather than resulting from gestational exposure. Data from several sub-chronic studies in male rats at approximately the same dose support this interpretation, as effects on body weight and weight gain were observed within the first several weeks of dosing.

The two-generation rat reproduction and developmental study on PFOA (York 2002; Butenhoff *et al.* 2004b) provides a number of endpoints suitable for use in risk assessment. For prenatal exposure, EPA relies on changes in F₁ male rat body weight-gain measured after several weeks of direct dosing that occurred between weaning and sexual maturity.

In the two-generation study, the F₁ generation pups received PFOA exposure via the dams during gestation and lactation; however, at weaning, the F₁ generation male rats were given daily oral doses of PFOA that continued through mating. Although EPA's assessment (p. 10) acknowledges that it is not known whether post-weaning effects are due to prenatal, lactational, and/or post-weaning exposures, for the prenatal life-stage margin of exposure, EPA has used decreased body-weight gain in F₁ male rats that was first observed during the second week of direct dosing at the 10 mg/kg/day dose. This effect occurred prior to sexual maturation and was considered to represent a developmental effect by EPA definition.

The NOAEL for reduced body-weight and body-weight change prior to sexual maturation was 3 mg/kg/day for male pups. EPA's assessment presumes that this effect on male pup body-weight gain could have been the result of prenatal exposure. Therefore, EPA calculates the prenatal life-stage margin of exposure using estimated 24-hour AUC values for pregnant rat dams receiving 3 mg/kg/day.

The assumption that decreased body-weight gain prior to sexual maturation in the post-weaning males being directly dosed with ammonium PFOA is a developmental effect runs counter to evidence from several subchronic studies demonstrating that adult males show body weight effects within the first few weeks of dosing at approximately the same mg/kg/day dose as that given to the post-weaning pups in the two-generation study (Metrick *et al.* 1977; Goldenthal *et al.* 1978; Palazzolo 1993).

- In a 13-week dietary study in male and female rats with dietary doses of 10, 30, 100, 300, and 1000 µg ammonium PFOA/g feed (N=5/group) (Goldenthal *et al.* 1978), weight-gain effects were seen within three weeks at the 100 ppm dietary dose that was equivalent to 7 mg/kg/day.
- In a 13-week dietary study by Hazleton Wisconsin, Inc. conducted at dietary doses of 1, 10, 30, and 100 µg ammonium PFOA/g feed (N = 55/group) (Palazzolo; Perkins *et al.*, 2004), a dietary dose of 100 µg/g feed equivalent to roughly 8 mg/kg/day produced a statistically significant decrease in mean body weight and body-weight gain compared to controls beginning in the second week of dosing.
- A four-week dietary study (Metrick *et al.* 1977) noted a decrease in weight gain over the four-week period at a dietary dose level of 100 µg ammonium PFOA/g feed approximating 10 mg/kg/day.

Benchmark dose calculations for body-weight gain for both F₀ and F₁ males from the two-generation study and for males in the Goldenthal *et al.* (1978) and Palazzolo (1993) studies fall in a range between 1.5 and 5.2 mg/kg/day at the lower 95% confidence limit based on a 10% change in the distribution (see Table 2 below). Considering the data provided by these three studies, the weight of the evidence supports a conclusion that the body weight-gain effects in the F₁ generation male rats is most likely a result of the direct dosing and not an effect resulting from gestational exposure.

EPA's draft correctly notes there are a number of endpoints from the two generation study that do bear directly on development which should be used to represent a developmental toxicity endpoint. These include birth weight, weight gain in lactation, mortality, and delays in sexual maturation. Benchmark doses for various developmental endpoints are calculated in the published risk assessment (Butenhoff *et al.* 2004a). A comparison of benchmark values from rat studies indicates that adult body weight effects occur at a lower dose than do adverse developmental effects.

In sum, the available data provide a sound basis for selecting a developmental toxicity endpoint, separate from body weight-gain effects that are best interpreted as a consequence of direct dosing. The two endpoints should not be confused. In addition, I note that gestational and lactational data submitted to EPA after the cut-off date for the risk assessment will allow further refinement of the risk assessment, as will developmental data on mice we understand to be forthcoming from EPA's laboratory.

Use of female rat body-weight and body-weight gain reduction in the second year of dietary dosing with ammonium PFOA in a cancer bioassay does not provide as compelling data for this endpoint as does female body-weight gain and body-weight reduction data from the two-generation study.

EPA's risk assessment appropriately notes (on p. 96) that the significance of body weight for human health is not clear, but that this endpoint is conservative. As discussed above,

male rats clearly experience body weight effects. Female rats exposed to PFOA also experience body weight effects, but these effects occur at a higher doses.

For the adult female life-stage margin of exposure, EPA chose to use an apparent effect of reduced body-weight gain and body weight in a two-year dietary cancer bioassay that included only two dose groups, 30 and 300 µg ammonium PFOA/g feed, which equated to 1.6 and 16 mg/kg/day (Sibinski 1987). The decreased weight in females in the two-year cancer study at the top dose is an uncertain value on which to base an estimate of margin of exposure. Although there is a trend in decreased body weight among females in the 16 mg/kg/day dose group beginning after about week 80 of the study, body weight was reduced at a statistically significant level (0.05) only at weeks 92 and 94 of the 104 week study (see female body-weight data found on pages 847 and 848 of the study report). In addition, obesity is a characteristic of aging in the Sprague-Dawley strain of rat and often clouds effects on body weight. Therefore, the body-weight data used by EPA as a basis for the adult female life-stage margin of exposure are not compelling or relevant for risk assessment.

Two studies with sub-chronic dosing periods (Goldenthal *et al.*, 1978; Butenhoff *et al.* 2004b) provide better insight into adult female rat weight effects.

- In the Goldenthal *et al.* (1978) study, no effect on body weight was observed at doses up to 1000 ug ammonium PFOA/g feed (80 mg/kg/day). It is noted that there were a small number of female rats (5) per dose group.
- In the two-generation rat study (Butenhoff *et al.* 2004b), there was no effect on female body weight in the F₀ generation at the highest dose of 30 mg/kg/day. There was a mild but statistically significant effect on body weight or body-weight gain in females at 30 mg/kg/day in the F₁ generation.² The NOEL for body weight effects in adult female rats was 10 mg/kg/day.

Considering the weight of the evidence, including the nature of the findings in the Sibinski (1987) study and the findings in other studies, use of 1.6 mg/kg/day from the

² The terminal body weights in adult females (Butenhoff et al. 2004b, Table 7) were:

Table 1
Terminal Body Weights in Adult Females in Two-Generation Study

Dose Group mg/kg/day	Terminal Body Wt - Adult Females F ₀ Generation (g)	Terminal Body Wt - Adult Females F ₁ Generation (g)
control (n=28)	345 ± 29	323 ± 23
1 (n=28)	340 ± 24	322 ± 24
3 (n=28)	351 ± 23	329 ± 22
10 (n=28)	343 ± 26	325 ± 24
30 (n=28)	345 ± 18	316 ± 21

Sibinski study as the NOEL for body-weight effects in adult female rats does not fairly represent the available database. I recommend the SAB ask EPA to re-evaluate the adult female body weight effect. The NOEL of 10 mg/kg/day from the F₁ females would provide a more rational basis for the adult female life-stage margin of exposure. Indeed, if a benchmark dose approach were used in accordance with EPA guidance, I expect it would provide an even higher value than the 10 mg/kg/day NOEL from the two-generation study.

A benchmark dose would better represent the data on body weight effects in adult male rats and other endpoints.

EPA uses a LOEL of 1 mg/kg/day for F1 males from the two-generation study as its point of departure for body weight effects in adult males. This is an appropriate endpoint to review, but a benchmark dose approach would better reflect the dose-response relationship than does the LOEL.

Using the EPA software program, BMDL₁₀ values for adult male rat bodyweight are:

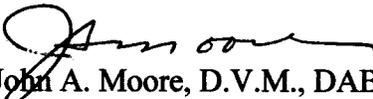
Table 2.
Benchmark dose estimates for a one standard deviation change (~10%) at the lower 95% confidence limit (BMDL₁₀) for decreased body-weight gain in male rats.

Study	Type	Duration	BDML _{10, mg/kg}	Model	p value
2-gen F ₀	Gavage	~15 wks	5.2	Polynomial	0.25
2-gen F ₁	Gavage	~15 wks	1.5	Power	0.20
Palazzolo	Dietary	13 wks	3.0	Power	0.46
Goldenthal	Dietary	13 wks	2.6	Polynomial	0.42

The SAB may wish to suggest EPA provide a benchmark dose calculation for the adult male body weight calculation, and for other endpoints. Various benchmark dose calculations may be found in Butenhoff *et al.* (2004a).

Thank you for the opportunity to provide these written comments. I look forward to an opportunity to respond to any questions the panel members may have.

Sincerely yours,


John A. Moore, D.V.M., DABT

cc: Dr. Seed

References

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