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November 9, 2018

Ms. Sarah Pillsbury
New Hampshire Department of Environmental Services
Drinking Water and Groundwater Bureau
29 Hazen Drive
Concord, NH 03301

Via email: Sarah.Pillsbury@des.nh.gov

Re: Request for studies or data that should be considered in deriving the health risk limits PFOA, PFOS, PFNA and PFHxS

Dear Ms. Pillsbury:

The 3M Company (3M) appreciates this opportunity to provide comments to the New Hampshire Department of Environmental Services (NHDES) for its MCL assessments on several perfluoroalkyl substances (PFAS). Specifically, NHDES has asked the following: "Beyond the studies considered in the recently released ATSDR Toxicity Profiles and the existing EPA Health Advisories, are there other studies or data that should be considered in deriving the health risk limits for each contaminant." The following offers 3M's thoughts responsive to this request. 3M believes that it will also be beneficial to share with NHDES specific problems with the ATSDR Draft PFAS Toxicity Profiles and the existing EPA Health Advisories that NHDES should consider during its MCL development activities.

As a science-based company, 3M has substantial experience and expertise with the breadth of topics that NHDES will need to consider in its MCL development. In fact, numerous 3M scientists are authors or contributors to many of the studies referenced in the EPA and ATSDR reports, especially in the areas of toxicology, pharmacokinetics, biomonitoring, and epidemiology. 3M also was first to sponsor the development of several physiologically-based pharmacokinetic models (PBPK) regarding perfluoroalkyls.

The attached documents provide high-level comments primarily for perfluorooctanesulfonate (PFOS) and perfluorooctanoate (PFOA); and to a lesser extent, perfluorohexanesulfonate (PFHxS). These compounds are considered as the end-stage metabolites from precursor molecules, and, for PFOS and PFOA, they have been the subject of the majority of the research studies available to date. We sincerely hope NHDES find these comments to be helpful in your deliberations. Given 3M's in-depth knowledge in this chemistry, we respectfully offer the following general observations and/or suggestions to NHDES.

3M has not manufactured or used these compounds in approximately a decade. Nonetheless, we realize there is ongoing public interest in these chemistries, and we want to share what we have learned. We trust our comments and any others that you may request of us will be viewed as valuable insights to the epidemiology and toxicology of PFAS. We would be pleased to elaborate and discuss these points with you.

Regards,

Oyebode A. Taiwo, MD, MPH

Comments to NHDES by the 3M Company

PRELIMINARY OBSERVATION

The Body of Scientific Evidence Does Not Show Adverse Health Effects in Humans From Perfluoroalkyls

The vast body of scientific evidence does not show that PFOS or PFOA cause adverse health effects in humans at current exposure levels, or even at the historically higher levels found in blood. ATSDR recently acknowledged that there is no cause and effect, when it stated: "The available human studies have identified some potential targets of toxicity; however, cause and effect relationships have not been established for any of the effects, and the effects have not been consistently found in all studies."

A recently released review of studies involving perfluoroalkyls exposed populations commissioned by the Australian government also supports the lack of evidence of harm. That May 2018 report by the Australian Expert Health Panel stated, "The Panel concluded there is mostly limited or no evidence for any link with human disease from these observed differences. Importantly, there is no current evidence that supports a large impact on a person's health as a result of high levels of perfluoroalkyl exposure." The report further stated: "After considering all the evidence, the Panel's advice to the Minister on this public health issue is that the evidence does not support any specific health or disease screening or other health interventions for highly exposed groups in Australia, except for research purposes."

AREAS OF CONSIDERATION REGARDING EPIDEMIOLOGICAL DATA

(1). Declining Concentrations in Human Blood: ATSDR recently noted this decline, stating "[s]erum levels of PFOA and PFOS in the general population of the United States have decreased dramatically in recent years as U.S. production of these substances ceased." (ATSDR 2018). There is a consistent declining trend with the CDC's National Health and Nutrition Examination Survey (NHANES) data in the US, where mean blood levels of PFOS and PFOA in the general population reported in the 2013 – 2014 period have declined by approximately 80% and 60%, respectively, since 1999 – 2000 period (CDC NHANES, 2017). In addition, 3M proactively started a perfluoroalkyl biomonitoring program with American Red Cross adult blood donors looking at the same six regional areas since 2001-2002. The most recent publication, that examined the samples collected in 2015, reported declining trends consistent with NHANES for these six regions (Olsen et al., 2017).

(2). Cross-sectional Epidemiological Studies Do Not Address Temporality or Causality: Many epidemiological studies regarding PFOS or PFOA are cross-sectional by design. Therefore, it is important to acknowledge that this type of study design ***cannot*** address temporality (*i.e.*, time-dependent associations). This issue is important to acknowledge because reverse causation has now been shown to be the explanation for several different health outcomes initially reported in cross-sectional studies as indicating an association between PFOS or PFOA exposure and the outcome. Among the associations explained by reverse causation are <u>chronic kidney disease</u> as explained Watkins et al. (2013), <u>lower birth weight</u> as initially analyzed by a PBPK model by Verner et al. (2015) with

subsequent updated meta-analyses of the epidemiology studies by Negri et al. (2017) and Steenland et al. (2018), early onset menopause as explained by Dhingra et al. (2017) and time to pregnancy as explained in the extensive review paper by Bach et al. (2016). In particular, Verner et al. (2015) showed in their PBPK models that the confounding effect of GFR on the association between lower birthweight and measured PFOA or PFOS concentrations could be minimized if the maternal measurement of PFOA or PFOS occurred in the first trimester. This observation was corroborated in a subsequent meta-analysis of the epidemiological data that was recently published by Steenland et al. (2018) who concluded epidemiologic studies where blood was sampled early in pregnancy "showed little or no association of PFOA with birthweight. These would be the studies in which confounding and reverse causality would be of less concern."

(3) Probable Link Reports from C8 Science Panel: Some have pointed to the Probable Link Reports from C8 Science Panel to incorrectly conclude that that the "links" are the same as causation of health effects. This is not the case. It is important to recognize the creation of the 3-member C8 Science Panel (Steenland et al., 2014) was the result of a 2004 settlement of a class action lawsuit in a mid-Ohio river community between residents (plaintiffs) and DuPont (defendant) through the Woods County circuit court (West Virginia). The C8 Science Panel was not a governmental regulatory or authoritative body. As the C8 Science Panel members have acknowledged, its findings were based not on a scientific causation process or standard, but on a standard and process agreed to as part of a legal settlement. (Steenland et al., 2014) Their findings were made solely for the purpose of defining further activities that were to occur in the litigation – they were not regulatory or scientific findings following standard and accepted approaches to determining causation. As a result of this, while considering the underlying studies conducted by members of the C8 Science Panel, ATSDR excluded the probable link findings from its 2018 Draft PFAS Toxicology Profile. (ATSDR 2018).

The C8 Science Panel concluded there was a probable link between PFOA and six diseases: kidney cancer, testicular cancer, ulcerative colitis, thyroid disease, hypercholesterolemia, and pregnancy-induced hypertension (Steenland et al., 2014). However, the C8 Science Panel has also stated that as more scientific evidence accumulates, "some associations may not be confirmed. Others may be identified that we (the C8 Science Panel) had missed".

Subsequent to these probable links reports issued by the C8 Science Panel and their published papers, other researchers have reported inconsistent findings with the C8 Science Panel results. This includes a lack of association with pregnancy-induced hypertension (preeclampsia) and PFOA in the general population (Starling et al., 2014), absence of increased kidney cancer risk and PFOA in production workers (Raleigh et al., 2014), and the occurrence of hypolipidemia (not hypercholesterolemia) in subjects of a phase 1 clinical trial of PFOA (ammonium salt) (Convertino et al., 2018). The findings from this phase 1 clinical trial clearly contradicted with the epidemiological data surrounding higher cholesterol levels noted at much lower PFOA concentrations (as reported in the general population). More data are needed to prove whether such a finding is causal or associative (e.g., physiological), especially given the lack of any increased risk for coronary artery disease incidence (Winquist and Steenland, 2014) or

mortality (Consonni et al., 2013; Raleigh et al., 2014; Woskie et al., 2012) in the more highly exposed (to PFOA) community and occupational populations.

(4). Phase 1 Data Available from Humans with Cancer Treated with Ammonium PFOA: A phase 1 clinical study (Convertino et al., 2018) was conducted at 2 cancer centers in Scotland: Beatson West Cancer Center (Glasgow) and the Aberdeen Royal Infirmary (Aberdeen). CXR Biosciences (Dundee) was the study sponsor who licensed their data to the 3M Company who subsequently requested epidemiologists at the University of Minnesota for statistical analyses of these data. In this phase 1 clinical trial, 49 cancer patients were included to determine the safety, dose-limiting toxicity, and maximum tolerated dose of ammonium PFOA which was evaluted for its chemotherapeutic potential. Ammonium PFOA was administered orally once a week for 6 weeks and comprenhenisve clinical chemistry parameters as well as other medical diagnostic endpoints were monitored.

Increasd plasma PFOA concentrations at approximately 175,000 – 230,000 ng/mL or higher (4 – 5 orders of magnitude higher than the general population) resulted in a decrease in serum total cholesterol and LDL (sometimes called "bad cholesterol"), but not HDL (sometimes called "good cholesterol"). This observation is consistent with animal data and the known mode of action for its lipid-lowering property via the PPARα pathway. This is similar to fibrate drugs that result in hypolipidemia (Roy and Pahan, 2009). These high levels of plasma PFOA concentrations also resulted in an increase in free thyroxine but there were no concomitant changes in TSH. This observation suggested possible transient binding competition between PFOA and thyroxine (which also has been shown to with PFOS). Transient thyroxine displacement without altering TSH status suggests that the re-equilibration of thyroxines (between free and bound) and it does not indicate compromised thyroid function. This is commonly observed with heparin and NSAIDs, too (Koulouri et al., 2013). Liver enzymes including ALT and liver function (e.g., prothrombin time) were unaffected, as well as serum creatinine, with these extremely high PFOA concentrations in this phase 1 clinical trial of PFOA administration. NHDES should consider this important human experimental study regarding ammonium PFOA in evaluating the evidence relating to the association between PFOA exposures and cholesterol levels.

(5). Quantitative Bias Analysis: Most PFASs are highly protein-bound and as such any physiological or pathological processes that could affect blood flow could potentially confound the concentrations of PFASs being measured in the blood. The improvement in the modern analytical capability has evolved greatly in the past decade and it obviously has allowed for the measurement of PFAS at extremely low levels (ng/mL or lower).

In epidemiological studies where the PFASs are being detected near the lower limit of detection, associations between health outcomes in relation to such very low levels of concentrations to PFASs are susceptible to be confounded by study subjects' metabolic status around clearance mechanisms (*e.g.*, glomerular filtration, pregnancy, lactation, and menstruation). To evaluate for this confounding, physiologically-based pharmacokinetic

(PBPK) models have been constructed to evaluate the epidemiological data for this potential confounding (or reverse causation). These include examinations of PFOA/PFOS and lower birth weight (Verner *et al.* 2015), delayed menarche (Wu et al., 2015), early onset of menopause (Ruark et al., 2016), and endometriosis (Ngueta et al., 2017). NHDES should consider these models in evaluating studies relating to PFAS.

(6). Serum elimination half-lives for PFOS, PFOA, and PFHxS should be considered age-, sex-, and concentration-dependent: The ATSDR in their calculation of a draft MRL chose not to use the geometric mean estimate that was discussed in the Olsen et al. (2007) paper. Given the right skewness of their data, Olsen et al. were more favorable to use the geometric mean for a measure of central tendency. The Olsen et al. (2007) study comprised 26 retirees (end of study average age = 66 years) who likely would have had an average glomerular filtration rate (GFR) lower than those calculated from younger ages as reported in Bartell et al. (average age 55) and Li et al. (age range 15 – 55) who had calculated considerable lower serum elimination half-lives for PFOA (Bartell, 2012) and PFOA, PFHxS, and PFOS (Li et al., 2018).

The average estimated glomerular filtration rate declines with age as shown in the table below.

Age range	Estimated GFR (ml/min/1.73 m^2)	Source:
1-6 months	77	Heilbron et al. 1991 Pediatr Nephrol. Jan; 5(1):5-11.
6-12 months	103	
12-19 months	127	
2-12 years	127	
20–29	116	https://www.kidney.org/sites/default/files/docs/11-10-1813_abe_patbro_gfr_b.pdf
30–39	107	
40–49	99	
50–59	93	
60–69	85	
70+	75	

Renal clearance of perfluorocarboxylates (and perfluorosulfonates) is largely a sum of three processes involving glomerular filtration, renal tubular secretion, and renal tubular reabsorption (Han et al., 2012). Because PFOA and other perfluorocarboxylates vary in their affinities to bind plasma proteins, glomerular filtration of perfluorocarboxylates (and perfluorosulfonates) is a product of the unbound fraction of the perfluorocarboxylate and the glomerular filtration rate (GFR). Thus, the higher estimates of GFR based on the younger ages in the other study populations, especially the younger Li et al. study which had approximately 50% of the follow-up time of Olsen et al., may be due to the age differences of the subjects, and not necessarily the shorter follow-up period considered in these studies. Thus, the serum elimination half-lives of other studies are likely equally

valid for consideration for any calculation that involve estimating the serum elimination half-life of PFOA and PFOS.
As an example, the chart below provides a range of serum elimination half-life calculations for PFOS from a variety of different populations, study designs, and concentrations of PFOS.

Multiple studies that have found serum elimination half-lives for PFOS of less than 5.4 years

Study	Half-life follow-up (years)	Study design
Olsen et al., 2007	4.8 geometric mean	Longitudinal
	5.4 arithmetic mean	
Spliethoff et al., 2008	4.1 (male/female infants)	Cross-sectional
Yeung et al., 2013	4.8 (German males/females)	Cross-sectional
	4.3 (German males/females)	
Wong et al., 2014	4.7 (NHANES males)	Cross-sectional
	4.0 (NHANES females)	
	3.7 (NHANES females)	
Olsen et al., 2017	4.6 (US males/females)	Cross-sectional
Gomis et al., 2017	4.9 (Australia males)	Longitudinal
	5.0 (Australia females)	
	3.8 (NHANES males)	
	3.3 (NHANES females)	
Worley et al., 2017	3.3 (Alabama males/females)	Longitudinal
Li et al., 2017	3.4 (Sweeden, males/females)	Longitudinal
	4.6 (Sweden, males)	
	3.1 (Sweden, females)	

In deriving guidance levels, NHDES should utilize a serum clearance half-life parameter appropriate for the population at issue. For the general population, the above studies suggest that a half-life in the range of 3.4 and 4.8 would be appropriate.

AREAS OF CONSIDERATION REGARDING TOXICOLOGICAL DATA

(7). An Ample Margin of Safety: The hypothesis-driven toxicology research studies for PFOS and PFOA are quite comprehensive and they include studies in mice, rats, rabbits, and monkeys. Many of them are repeat-dose studies that include measurements of PFOS or PFOA in serum and liver, as well as clinical and histological pathology at various times during dosing. The responses to PFOS or PFOA treatment in toxicological studies have shown clear threshold-mediated characteristics that were consistent across species and were related to body burden, as evidenced by serum and liver concentrations of PFOS or PFOA related to cumulative dose which were several orders of magnitude higher than general population. These studies strongly support large margins of acceptable exposure across different species.

(8). Primate Toxicity Data Should not be Minimized: When it comes to human relevance and risk assessment, given the many issues in extrapolating toxicology data from rodents (a lower order species) to humans (the highest order), primate data have

always valued as the most scientifically appropriate species for human risk assessment because it is the second-highest order species next to humans.

In deriving their proposed guidance values, both ATSDR and EPA apply uncertainty factors on the assumption that humans are *more* sensitive than rodents to these effects. This is not the case, however. Published data strongly support that rodents are likely to be much more sensitive to PFAS-induced effects than humans

Given that rodents may not be the most appropriate species for human hazard assessments of PFOS or PFOA due to demonstrated differences in mode of action (Klaunig et al., 2012), the available primate data on PFOS and PFOA should be taken into your consideration (Butenhoff et al., 2002; Chang et al., 2017; Convertino et al., 2017; Seacat et al., 2002).

For example, the 6-month oral capsule study with PFOS in monkeys (Seacat et al., 2002) had been used by several agencies in their risk assessment (Butenhoff and Rodricks, 2015) . Although changes in serum HDL and thyroid hormones were chosen as the basis for point of departure in the risk assessment, the uncertainties in the interpretation of these endpoints prompted a re-evaluation of these clinical parameters in monkeys by Chang et al. (2017). The absence of the effects in thyroid functions, liver functions, renal functions, electrolytes, and coagulations were consistently observed prior to and after PFOS treatment(s) in monkeys when serum PFOS concentration has reached as high as 175,000 ng/mL (ppb). There were some subtle PFOS treatment-related changes in serum HDL, but its biological significance was questionable because the extent of HDL reduction was well within the normal variation. The corresponding lower-bound fifth percentile benchmark concentrations ($BMCL_{1sd}$) was approximately 75,000 ng/mL in terms of serum PFOS concentration in monkeys, which was 4 orders of magnitude higher than that of the currently found in the US general population.

(9). The Toxicity Potency of PFHxS in Animal Models is Lower than PFOS:
Perfluorohexanesulfonate (PFHxS) is the six-carbon homologue of PFOS and even though its toxicological database is not as comprehensive as PFOS, it is imperative to differentiate the fact that the toxicity potency for PFHxS in animal models is not equivalent to PFOS, and, PFOS should not be used as a surrogate for PFHxS for toxicity interpretation. This was erroneous assumption made by Minnesota Department of Health in their 2017 assessment of PFHxS where the agency failed to consider pertinent data from Butenhoff et al. (2009)

While at sufficiently high doses PFOS has been shown to cause maternal systemic toxicity (*i.e.*, body weight loss) in mice and rats and maternal-mediated neonatal effects such as decreased pup birth weight and mortablity, such effects have not been observed after PFHxS exposure in rats or mice (Butenhoff et al., 2009; Chang et al., 2018; Ramhoj et al., 2018). The highest maternal and/or pup serum PFHxS concentrations achieved in rats (Butenhoff et al., 2009; Ramhøj et al., 2018) or mice (Chang et al., 2018) were 4 - 5 orders of magnitude higher than that of the general population and there was

no evidence of PFHxS-induced maternal toxicity nor developmental toxicity in rodents. This data should be considered in evaluating guidance levels for PFHxS.

(10). Developmental Toxicity in Rodents and Questionable Human Relevance: Developmental toxicity from two rodent studies were selected in the most recent risk assessments with exposure to PFOS (Luebker et al., 2005) and PFOA (Lau et al., 2006) by several government agencies. It has been demonstrated that in the toxicology studies with PFOS or PFOA, the inactivation of nuclear receptor PPARα in mice can attenuate or completely minimize the developmental toxicity seen in pups, along with several other toxicity endpoints (Abbott et al., 2009; Abbott et al., 2007; Albrecht et al., 2013). Therefore, this endpoint and its relevance to humans becomes questionable given the lower prevalence of PPARα receptors in humans.

(11). Absence of Data That Would Support Direct Reproductive and Developmental Toxicity in Rodents: It is important to recognize that PFOS, PFOA, and PFHxS do not affect male or female reproductive functions in laboratory animals including mice, rats, and/or rabbits. These assessments included estrous cycles, sperm parameters, mating index, fertility index, and reproductive organ morphology. The developmental observations in pups were generally unremarkable in laboratory rodents when maternal-mediated toxicities were taken into consideration (Lau et al., 2007; Lau et al., 2004).

(12). Absence of Data That Would Support Endocrine Disruption: In recent years, there have been toxicological studies reporting on the endocrine disturbance potential with PFOS or PFOA exposures. The endocrine system is a very complicated and evaluation of endocrine functions is a very highly specialized field requiring careful interpretation. In laboratory toxicity studies where endocrine disruption properties with PFAS had been reported, they were typically based on some alterations of hormone levels with thyroid hormones being the most common hormones measured. While many test guidelines have recommendations to measure thyroid hormone such as T4 (thyroxine), it is important to note that many of the test guidelines do <u>not</u> conform to the current science on the assessment of thyroid homeostasis. Therefore, it is important to emphasize and to understand the primary operative roles of H-P-T axis, and that T4 is not a reliable stand-alone index for thyroid hormone status determination even though it is suggested by the test guidelines. Instead, one should recognize that serum thyroid-stimulating hormone (TSH) level should be used for diagnosing the overall thyroid hormone status. Also, when possible, thyroid histology should be evaluated in conjunction with serum TSH data. If and when T4 is being measured, it should be free T4 (FT4), not total T4 (TT4), because FT4 is the biologically active portion of the T4. However, FT4 measurement is subject to various interferences (well-described in human and veterinary medicine) hence extra precaution is necessary to carefully FT4 data (Ekins, 1989; Larsen, 1972; Nelson et al., 1994). As a fact, clinical reference laboratories, such as Mayo Medical Laboratories, cautions that "all free hormone assays should be combined with thyroid-stimulating hormone measurements." (<u>https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/8859</u>). Based on data from the large scale 2-generation reproductive and developmental studies (which are considered as the most comprehensive test by various agencies for evaluating endocrine functions), neither

PFOS or PFOA altered the endocrine functions as the reproductive functions and performances in both males and females were normal across multiple generations. In other words, if they are capable of disrupting endocrine functions, there would be unsuccessful reproduction in the second generation in these studies, which was not the case.

If PFOA and PFOS are indeed true human estrogen or thyroid toxicants, then one would also expect them to act directly with endocrine receptors such as estrogen receptors and thyroid receptors. Ishibashi et al. (2007) reported that neither PFOS or PFOA can directly activate human estrogen receptor α or β. In a collaboration project between the 3M Company and Mayo Clinic, the potential of PFOS, PFOA, and PFHxS to activate human thyroid receptor was examined. While triiodothyronine, tested between 0.000001 – 0.01 uM, elicited a dose-response activation of human thyroid receptor α; under the same study condition, there was no activation of human thyroid receptor α when exposed to PFOS, PFOA, or PFHxS up to 100 uM (Ehresman et al., 2014).

(13). Absence of Data That Would Support Carcinogenicity in Humans: Lifetime treatments of rats receiving PFOS or PFOA developed benign liver adenomas. Detailed mechanistic studies have shown that activation of PPARα in rodents led to these liver tumors via the downregulation of let7c micro RNA which ultimately leads to increased cell cycle progression. This mechanism is not applicable in humans (Shah et al., 2007). Thus, the development of liver tumors in humans with exposure to PFOS or PFOA would not be expected; and no association has been reported.

There was no experimental evidence suggesting the development of kidney lesions in rats with when treated with either PFOS or PFOA for up to two years. The classification of PFOA as a *possible* kidney carcinogen by IARC (Class 2B) meant that while it considered the epidemiological data reviewed to be credible, chance, bias, and confounding could not be ruled out with reasonable confidence (IARC, 2014).

Male rats treated with PFOA for 2 years developed benign testicular Leydig cell adenomas. Given that more than 95% of the testicular cancer seen in humans are of a testicular germ cell tumor type, not Leydig cells, the relevance of this observation to human risk is questionable. The classification of PFOA as a *possible* testes carcinogen by IARC (Class 2B) meant that while it considered the evidence to be credible and unlikely to be explained by bias and confounding, the estimates was based on small numbers (IARC, 2014).

Male rats treated with PFOA for 2 years also developed benign pancreatic acinar cell adenomas without excessive malignant carcinomas. It has been well-documented that there is a distinct species difference in pancreatic cell proliferation regulation. In rats, it is via direct binding with CCK receptors while in humans, it is via secretory neuronal cholinergic pathway. Therefore, in light of the difference in the biological behavior, the increased pancreatic acinar cell adenoma associated with PFOA exposure in rats and its relevance to humans is questionable. By way of example, Myer et al. (2014) published a paper that summarized several long-term studies in mice, rats, monkeys, and humans on a

novel CCK receptor agonist (code name GI181771X, developed by GSK) for the treatment of obesity. While the CCK receptor agonist, given for up to 26 weeks, induced dose-dependent increases in pancreatic hyperplasia in rodents, no changes were seen in monkeys at even higher doses and a longer treatment period (up to 52 weeks) or in humans (up to 24 weeks in a clinical trial).

(14). Insufficient Evidence to Support Immunotoxicity with PFOS or PFOA: Albeit the US National Toxicology Program (NTP) conducted a systemic review in 2016 and concluded that PFOS and PFOA are both presumed to be immune hazards to humans, there were several areas of the NTP systematic review on PFOA and PFOS in which insufficient animal data were used as supporting evidence for human findings and its final hazard conclusion. Among those deficiencies, NTP used suppression of the TDAR in mice, which evaluates suppression of the "primary" IgM response, to support its claim of possible suppression of antibody titers to vaccinations in humans. However, because vaccine antibody titers actually represent the secondary IgG response, the observation in some human epidemiological data of decreased antibody titers was in great discrepancy with animal data in that no suppression of the secondary IgG response was observed in mice. Similarly, there were incongruences between humans and animal data to support the final hazard conclusions reached by the NTP in the areas of hypersensitivity for PFOA, infection disease resistance for PFOS, and NK cell activity for PFOS.

(15). Validity of Anecdotal Mechanistic Studies: The heightened attention to PFAS research is reflected by the enormous amount of publications that have become available. Many of these studies that were done under *in vitro* conditions to which high concentrations of PFOS or PFOA were employed. Given that PFOS and PFOA are strong surfactants, the toxicity effects reported from the typical mono-layered *in vitro* tissue culture system offered very little insight and scientific value because the data were often comprised by the surfactant-induced toxicity. In addition, *in vitro* tissue culture cells lack vascular system an intact animal can offer. When *in vivo* designs were involved, they tend to be small in scale with few number of animals used, and, often times a limited set of endpoints were reported (e.g., selected gene expressions at a certain target organ) which do not represent full functionality. Given these issues, in vivo and in vitro mechanistic studies must be interpreted with care.

OBSERVATIONS ON ATSDR DRAFT TOXICOLOGICAL PROFILE FOR PERFLUOROALKYLS

3M submitted detailed comments in response to ATSDR's request for comments on ATSDR's draft Toxicological Profile for Perfluoroalkyls. The following summarizes some of the key concerns with ATSDR's document.

(16). The Proposed MRLs Fail to Reflect the Best Available Science. Overall, the provisional MRLs proposed by ATSDR for PFOA, PFOS, and PFHxS were not derived using best available science. There were many deficiencies and unnecessarily conservative and scientifically flawed assumptions associated with these MRLs.

Greater consideration should have been given by ATSDR to the non-human primate studies that exist in the literature for PFOA and PFOS, as was done by ATSDR in 2015. In addition, ATSDR selection for PFOA and PFOS did not consider the more recently available human and non-human primate studies. ATSDR should have considered these studies in their approach to MRL as they either do or more closely represent human physiology; and, have relevance to questions regarding thyroid, cholesterol, and liver evaluations. These include a Phase 1 clinical trial in humans for PFOA (Convertino et al. 2018) and a one-year evaluation of clinical chemistries in non-human primates for PFOS (Chang et al. 2017).

ATSDR selected inappropriate studies to serve as basis for the proposed MRL for PFOA which ***lacked fundamental scientific rigor***, including such shortcomings as: (1) use of only single dose level, making it impossible to confirm a dose-response effect, or to determine the point of departure level; (2) involved too few animals to generate reliable results; (3) provided no details on the reproductive nor the developmental hallmarks; (4) litter bias; (5) used non-standard testing methods; and (5) provided no internal serum PFOA dosimetry data. The corresponding study results should not be used in any meaningful risk assessment for humans and are wholly inadequate to form the basis for a PFOA MRL.

PFOA, PFOS, and PFHxS MRLs are biased (downward) because ATSDR used serum half-lives that do not accurately reflect the most reliable and current evidence on human serum half-lives applicable to the general population. Had it done so MRL values would have ranged between 9 - 40% higher for PFOA, 12 – 38% higher for PFOS, and 14-38% higher for PFHxS;

ATSDR applied scientifically flawed uncertainty factors that lowered the MRLs by as much as an order of magnitude or more, including: (1) use of an uncertainty factor of three for interspecies extrapolation (animal to human) for PFOA, PFOS and PFHxS, even though that rodents are known to be more sensitive than humans to the effects at issue; (2) use of an uncertainty factor of 10 in its PFOS and PFHxS MRL derivations to account for potential immunological effects that was arbitrary, not justified by toxicology and epidemiologic studies, and contrary to ATSDR's acknowledgement that the human evidence for immune effects is insufficient to support causation; and (3) use of an inappropriate uncertainty factor of 10 for PFOA for a LOAEL-to-NOAEL extrapolation because the study design was so deficient so as to preclude even establishing any LOAEL or NOAEL values.

(17). Epidemiological Associations Claimed by ATSDR are Not Supported by the Science. In addition, the draft Toxicological Profile for Perfluoroalkyls identified eight potential epidemiological associations between perfluoroalkyl exposure and health outcomes. The relevant body of science for these chemicals does not support ATSDR's position. As our detailed comments to ATSDT show, the scientific evidence clearly refutes the claimed associations and shows that ATSDR must revisit its analysis. In addition, ATSDR acknowledges that none of these associations indicate causality.

(18). Additional Many Other Concerns and Deficiencies Require Revisions to the Draft Our detailed comments outlined many other concerns with the draft Toxicological Profile for Perfluoroalkyls, including, but not limited to: (1) significant new studies were not

considered by ATSDR; (2) a lack of transparency in ATSDR's synthesis of its weight-of-the-evidence review for the eight epidemiological associations or key toxicological endpoints; and (3) a failure to address declining levels of PFOS and PFOAs in the general population.

OBSERVATIONS ON US EPA's DRINKING WATER HEALTH ADVISORIES

(19). EPA's Drinking Water Health Advisories are Not Peer Reviewed

Contrary to statements made by EPA and others, the drinking water health advisories were not peer reviewed. EPA also issued these values with no opportunity for public comment.

On May 26, 2016, EPA announced, the release of "final" health effects support documents for PFOA and PFOS and the release of lifetime drinking water health advisories for PFOS and PFOA. EPA stated that "EPA's HAs for PFOA and PFOS are supported by peer-reviewed health effects support documents that summarize and analyze available peer reviewed studies on toxicokinetics, human epidemiology, animal toxicity, and provide a cancer classification and a dose response assessment for noncancer effects." This announcement, as well as subsequent statements by the agency conveys the impressions that the health effects documents and the health advisories were peer reviewed. This is misleading.

While early drafts of PFOA and PFOS health assessment documents were subject to peer review and public comment in 2014, the health assessment documents released in 2016 differed significantly from the 2014 documents, yet they were issued without public comment or peer review. EPA relied on these documents to develop the drinking water health advisories.

Both 2016 PFOA and PFOS health effects documents were significantly longer and materially different than their 2014 counterparts. EPA changed, without explanation, the point of departure study and critical endpoints for both PFOA and PFOS. For PFOS, EPA changed the point of departure study from a neonatal neurodevelopmental rat study by Butenhoff et al. to a two-generation rat study by Luebker et al (2005a). The critical endpoint changed from neurodevelopmental delay (increased motor activity and decreased habituation on postnatal day 17) in pups to decreased pup weight. For PFOA, EPA changed the point of departure studies from rodent studies by Palazzolo et al. (rats), Lau et al. (mice) and York et al. (rats) to a mouse study by Lau et al. (2006). The critical endpoint changed from liver weight gain to developmental effects in pups.

The drinking water health advisories themselves were never issued in draft, nor underwent any public comment or peer review. The lifetime drinking water health advisories issued in 2016 replaced provisional short-term exposure drinking water health advisories from 2009. EPA issued the 2016 advisories without review or comment, even though EPA deviated from its standard exposure assumptions (including exposed individual, water consumption rate, body weight) for developing drinking water health advisories.

(20). Concerns with EPA PFOA Reference Dose

EPA (2016) derived an RfD of 0.00002 mg/kg-day for PFOA based on a reproductive and developmental study in mice by Lau *et al.* (2006). In this study, pregnant mice received oral doses of PFOA at 0-20 mg/kg-day from gestation day (GD) 1 to GD 17 or 18. Fetuses from some dams were checked for malformations on GD 18, and pups from the dams that were allowed to give birth were monitored for growth and development. The authors reported delayed skeletal ossification (bone development) and accelerated male puberty in mice, starting at the lowest dose group (1 mg/kg-day). However, neither the delayed skeletal ossification nor the accelerated male puberty in mice showed increased effects in a dose-related manner. EPA concluded that the lowest observed adverse effect level (LOAEL) from this study was 1 mg/kg-day, corresponding to a predicted serum concentration in the mouse of 38 μg/mL. EPA calculated a human equivalent dose (HED) of 0.0053 mg/kg-day based on differences between mouse and human PFOA pharmacokinetics. A total uncertainty factor (UF) of 300 (10 for intraspecies differences, 3 for interspecies differences, and 10 for use of a LOAEL rather than a no observed adverse effect level [NOAEL]) was applied to the HED to derive an RfD of 0.00002 mg/kg-day for PFOA.

There are a number of concerns with EPA's development of the PFOA RfD, including the following: (1) use of a rodent study instead of primate studies as point of departure study; (2) study interpretation; (3) use of the interspecies uncertainty factor of 10; (4) use of the intraspecies uncertainty factor of 3; and (5) use of an uncertainty factor of 10 to account for the use of a LOAEL.

Use of a rodent study as point of departure study: As discussed above, rodents are likely to be much more sensitive to PFAS-induced effects than humans. Given that rodents may not be the most appropriate species for human hazard assessments of PFOA due to demonstrated differences in mode of action (Klaunig et al., 2012), EPA should have looked to the available primate data on PFOA, instead of the rodent study it selected.

The basis of the RfD for PFOA is not consistent with the underlying science. EPA's choice of these endpoints as the basis of the RfD for PFOA is not consistent with the underlying science. Specifically, none of the study endpoints used for RfD derivation exhibits a regular dose-response. For example, reduced ossification of the calvaria (part of the skull) has a U-shaped response, with the largest effects in the two lowest dose groups (1 and 3 mg/kg-day). Each of the other two reduced ossification endpoints, for forelimb and hindlimb proximal phalanges (toe bones), also have U-shaped responses. The dose-response for sexual maturation in males is reversed, with the largest effect at the lowest dose and the smallest effect at the highest dose. Irregular dose-response patterns such as these call into question whether the effects are actually related to the agent being tested.

There are also irregularities in the way the endpoints are reported. Ossification effects are typically reported as the number of animals exhibiting the effect, as opposed to the number of affected sites (US EPA, 1986). This is important because if one animal has multiple affected sites, the effect reported as number of sites appears larger compared to

the effect reported as number of animals. Thus, it is not possible to tell in the Lau *et al.* (2006) study whether the results would be statistically significant if they were reported in the more standard "number of animals exhibiting the effect" format.

In addition, reduced or delayed ossification is not necessarily an adverse effect. Reduced or delayed ossification is usually considered a variation, not a malformation, because ossification reduction or delay is mild and reversible. Variations are alterations in the body that are considered to have no adverse effect on health or body structure, may have a high incidence, and represent slight deviations from normal morphology (Stump *et al.*, 2012). It is also notable that other agencies, including ATSDR (2015), the European Food Safety Authority (EFSA) (2008), and the United Kingdom Food Standards Authority (UK FSA) (2006), did not include the ossification or puberty effects of PFOA reported by Lau *et al.* (2006) in their determinations of relevant endpoints. ATSDR (2015) considered the LOAEL from Lau *et al.* (2006) to be 3 mg/kg-day based on the "less serious" effect of a 20% body weight reduction in the pups on postnatal day (PND) 23, and a LOAEL of 5 mg/kg-day based on the "more serious" effect of full litter resorptions. EFSA (2008) considered the developmental LOAEL to be 3 mg/kg-day based on growth deficits in the pups.

Interspecies Uncertainty Factor. EPA applied an interspecies uncertainty factor of 3 to account for the possibility that humans are more sensitive than rodents to the effects at issue. As already discussed, the known data suggests that humans are less sensitive to PFOA-mediated effects than rodents. Accordingly, this uncertainty factor should be eliminated.

Intraspecies Uncertainty factor. EPA applied an interspecies uncertainty factor of 10. This factor could have been lower or eliminated because human variability is much less than the factor of 10. This is already accounted for in the conservative intake/exposure assumptions used by EPA

LOAEL-to-NOAEL Uncertainty Factor. EPA should not have applied an uncertainly factor of 10 for LOAEL-to-NOAEL. EPA implicitly determined the study by Lau et al. (2006) did not have a no observable adverse effect level (NOAEL) and based its assessment on a lowest observable adverse effect level (LOAEL). This is incorrect. Lau et al. (2006) clearly showed in Table 6 of the paper that NOAEL-equivalent benchmark doses (BMDs) were indeed available. BMD values are preferable to NOAELs. Therefore, EPA should have based its PFOA evaluation on the BMD values and LOAEL-to-NOAEL uncertainty factor of 10 should be eliminated. In fact, EPA chose not to follow its own guidance, which outlines the use benchmark dose instead of LOAELs or NOAELs where data are available. U.S. EPA, Benchmark Dose Technical Guidance, EPA Doc. No. EPA/100/R-12/001 (2012).

(21). Concerns with EPA PFOS Reference Dose

US EPA (2016) derived an RfD of 0.00002 mg/kg-day for PFOS based on two reproductive and developmental studies in rats by Luebker *et al.* (2005a,b). In the first study (Luebker *et al.*, 2005a), male and female rats were dosed with PFOS for 6 weeks prior to mating, during mating, and through gestation and lactation across two

generations. US EPA chose reduced weight gain in the F2 pups as the critical effect and cited a NOAEL for developmental effects of 0.1 mg/kg-day and a LOAEL of 0.4 mg/kg-day. The study authors noted that they did not consider the reduced weight gain in the F2 pups a toxicologically significant effect because it was transient (seen at days 7 and 14 but not at day 21 post-partum), could have been related to litter size (*i.e.*, larger litters tend to have smaller pups), or could have been a random effect of culling. The F1 pups did not exhibit this effect; the NOAEL for the F1 pups was 0.4 mg/kg-day (the highest dose tested in the F1 pups). Luebker *et al.* (2005a) considered the NOAEL for offspring effects to be 0.4 mg/kg-day.

The second study by Luebker *et al.* (2005b) was a one-generation reproduction and development study in rats of the same strain as those used by Luebker *et al.* (2005a). In this study, females only were dosed for 6 weeks prior to mating through day 4 of lactation, and the lowest dose tested was 0.4 mg/kg-day. At this dose, the F1 pups exhibited lower birth weight and reduced weight gain. This is contrary to the first study, in which the F1 pups did not exhibit effects at this dose. The authors did not address the differences in results between the two studies. US EPA cited a LOAEL for this study of 0.4 mg/kg-day and considered it to be support for their choice of a NOAEL of 0.1 mg/kg-day for the critical effect of reduced weight gain in the F2 pups in the first study (Luebker *et al.*, 2005a).

US EPA used a pharmacokinetic model to calculate an HED of 0.00051 mg/kg-day based on differences between rat and human PFOS pharmacokinetics. A total UF of 30 (10 for intraspecies differences and 3 for interspecies differences) was applied to the HED to derive an RfD of 0.00002 mg/kg-day.

There are also a number of concerns with EPA's development of the PFOS RfD, including the following: (1) use of a rodent study instead of primate studies as point of departure study; (2) serum elimination half-life; (3) interspecies uncertainty factor of 10; and (4) intraspecies uncertainty factor of 3.

Use of a rodent study as point of departure study: As discussed above, rodents are likely to be much more sensitive to PFAS-induced effects than humans. Given that rodents may not be the most appropriate species for human hazard assessments of PFOS due to demonstrated differences in mode of action (Klaunig et al., 2012), EPA should have looked to the available primate data on PFOS, instead of the rodent study it selected. For example, the 6-month oral capsule study with PFOS in monkeys (Seacat et al., 2002) had been used by several agencies in their risk assessment (Butenhoff and Rodricks, 2015) . Other primate studies are also available. See Chang et al. (2017).

Serum elimination half-life: EPA selected of 5.4 years as the serum elimination half-life for PFOS based on the paper by Olsen et al. (2007) to calculate the HED. This value is not representative of values applicable to the general population as documented by the recent literature. The literature documents serum elimination half-lives between 3.4 and 4.8 years. Please refer to Point 6 above, which further discusses the elimination half-life for PFOS.

In addition, a study has been published since EPA's drinking water advisories that would suggest an average serum elimination half-life for PFOS of 3.4 years would be appropriate. Li et al. (2018), Li et al. (2018) analyzed individual data from Ronneby, Sweden. A serum elimination half-life for PFOS of 3.4 years was reported. Relying on values from this study would be consistent with EPA's practice of having chosen a serum elimination half-life of 2.3 years for PFOA based on another population that had drinking water contaminated with PFOA mitigated by GAC filter installation (Bartell *et al.*, 2010).

Interspecies uncertainty factor of 3. EPA applied an interspecies uncertainty factor of 3 to account for the possibility that humans are more sensitive than rodents to the effects at issue. As already discussed, the known data suggests that humans are less sensitive to PFOA-mediated effects than rodents. Accordingly, this uncertainty factor should be eliminated.

Intraspecies uncertainty factor of 10. EPA applied an interspecies uncertainty factor of 10. This factor could have been lower or eliminated because human variability is much less than the factor of 10. This is already accounted for in the conservative intake/exposure assumptions used by EPA.

(22). Drinking Water Health Advisory Exposure Assumptions

Using the RfD of 0.00002 mg/kg-day for both PFOA and PFOS, EPA (2016, 2016) derived Lifetime HAs of 0.07 μg/L for each of these chemicals, or for both chemicals combined when they are present together, in drinking water according to these equations:

$$DWEL = RfD \times \frac{BW}{DWI}$$

and

$$Lifetime\ HA = DWEL \times RSC$$

where:

RfD	=	Reference Dose (mg/kg-day) = 0.00002 mg/kg-day
DWI/BW	=	Drinking Water Intake/Body Weight (L/kg-day) = 0.054 L/kg-day
DWEL	=	Drinking-water-equivalent Level (mg/L) = 0.00037 mg/L
RSC	=	Relative Source Contribution (unitless) = 0.2
Lifetime HA	=	0.000074 mg/L (rounded to 0.00007 mg/L) = 0.07 μg/L

In the derivation of the HAs, EPA made assumptions about relative source contributions, drinking water intake, and the average body weight of lactating women. The discussion that follows describes the bases of these assumptions and why the chosen values for these parameters are not reliable. Added together, these assumptions yield an overly restrictive concentration of PFOA and PFOS in drinking water corresponding to the selected toxicity criteria.

Drinking Water Intake and Body Weight. The DWI/BW used to derive the Lifetime HAs for both PFOA and PFOS is 0.054 L/kg-day, the 90th percentile value for lactating

women according to US EPA (2011). EPA chose this parameter because of the potential for increased susceptibility during pregnancy and lactation, based on the studies of developmental toxicity that were used for the RfD determinations (Lau *et al.*, 2006; Luebker *et al.*, 2005b).

The 0.054 L/kg-day value was derived from a study by Kahn and Stralka (2008). This study has a number of limitations that render the DWI value unreliable. EPA (2011) notes in its Exposure Factors Handbook that regarding the Kahn and Stralka (2008) study, the overall confidence rating for the values it presents is low. EPA (2011) states, "[e]stimates are less statistically reliable based on guidance published in the *Joint Policy on Variance Estimation and Statistical Reporting Standards on NHANES III and CSFII Reports: NHIS/NCHS Analytical Working Group Recommendations* (NCHS, 1993)...The sample size does not meet minimum reporting requirements to make statistically reliable estimates as described in the *Third Report on Nutrition Monitoring in the United States*, 1994–1996 (FASEB/LSRO, 1995)." The sample size was 34.

The intake value for lactating women reported by Kahn and Stralka (2008) is based on self-reported water intake and an average self-reported body weight of 54.8 kg (121 lbs.). Considering the recommended value for an adult body weight in the US was historically 70 kg (154 lbs.) and is currently 80 kg (176 lbs.) (US EPA, 2011), the average body weight for lactating women in the Kahn and Stralka (2008) study is likely underestimated. This would have the effect of overestimating the 90th percentile water intake for lactating women, normalized to body weight, and would cause the HAs to be unnecessarily low.

EPA overall rating of this consumption estimate was "low." (US EPA, 2011). It rated study "soundness" as low, in part because of the small sample size, lack of physical measurements, and reliance on recent recall of standardized volumes of drinking water containers. EPA rated its applicability and utility as low to medium. Id.

EPA guidance for health advisories suggests using 2 liters of water per day and a 70-kg individual for a consumption rate of 29 mL/kg-day. The 54 mL/kg-day consumption rate used by EPA for the PFOA and PFOS health advisories is nearly twice as much. If EPA had used a consumption rate of 29 mL/kg-day, the resulting health advisories would have been 0.138 ug/L instead of 0.070 ug/L. EPA also calculated a lifetime health advisory for alternative general population exposure scenario using an assumed drinking water rate of 2.5 L/day (90th percentile for those 21 or older) and a mean body weight for all adults. (EPA 201, 2016). This resulted in a water consumption rate of 31 mL/kg-day and health advisories of 0.10 ug/L.

In sum, the exposure assumptions used by EPA are based on a very limited and statistically invalid data set, exaggerate potential water consumption through very conservative assumptions, deviate from standard practices and produce artificially low health advisory levels.

Relative Source Contribution (RSC). EPA used 20% for the relative source contribution (RSC) for PFOA and PFOS through the consumption of water. A 20% RSC is the default assumption used by EPA for exposure through drinking water under this method when information is not adequate to attribute sources of exposure. EPA said it used the Decision Tree methodology to derive the relative source contribution, but it provided no specific

reasoning for the selection of this value. There was no discussion of sources of PFOA or PFOS other than water.

The use of a 20% RSC for deriving drinking water values is not scientifically supported and is more stringent than is necessary to protect public health. There have been several studies of dietary, dust, and inhalation exposure to PFOA (Fromme *et al.*, 2009; Lorber and Egeghy, 2011; Tittlemier *et al.*, 2007; Schecter *et al.*, 2010; Haug *et al.*, 2010; Noorlander *et al.*, 2011; EFSA, 2012; Vestergren *et al.*, 2012) and PFOS (Fromme *et al.*, 2009; Egeghy and Lorber, 2011; Tittlemier *et al.*, 2007; UK FSA, 2009; Clarke *et al.*, 2010; Noorlander *et al.*, 2011; Haug *et al.*, 2011; Domingo *et al.*, 2012; EFSA, 2012; Vestergren *et al.*, 2012; Klenow *et al.*, 2013; Gebbink *et al.*, 2015). None of these studies indicate that exposures other than drinking water are likely to add up to 80% of the allowable daily intakes of PFCs at the US EPA RfDs. In addition, the Minnesota Department of Health (MDH) recently chose a 50% RSC that was based on serum concentrations in both the local Minnesota and general US populations when deriving Health Based Value (HBV) derivations for PFOA and PFOS (MDH, 2017b, c).

An RSC value of 50% would be better supported by the intake studies and would still ensure a margin of safety. Moreover, it is still conservative and health protective.

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