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Principal Investigator: Jean M. Burris, RN, MPH[1]

Co-investigators: James K. Lundberg PhD[3]
Geary Olsen, DVM, PhD[1]
Cathy Simpson, RN[2]
Jeffrey Mandel, MD, MPH[1]

Study Director: Jeffrey Mandel, MD, MPH[1]

Study Sponsor: Medical Department
3M Company
220-3W-05
Saint Paul, MN 55144

1. Occupational Medicine, Medical Department, 3M Company, 220-3W-05, St. Paul, MN 55114
2. 3M Decatur Specialty Adhesives and Chemicals Plant, P.O. Box 2206, Decatur, AL 35609-2206
3. ET&SS Lab Special Projects, 3M Company, 2-3E-09, St. Paul, MN 55114

ABSTRACT

This is the second interim report for an ongoing study, to determine the serum half-life of several fluorochemicals in humans by obtaining and analyzing multiple serial blood samples from 27 Decatur and Cottage Grove fluorochemical production plant retirees. A first interim report, issued in June 2000, suggested the serum half-life of PFOS in humans was likely in the range of 139-640 days, four-fold lower than the previously estimated range of 1000-1500 days (data derived from a different population of 3 workers). The serum half-life for PFOA in these 27 retirees appeared to be one year. However, the initial interim report was subsequently discounted in a 3M letter to the U.S. EPA, which noted that the first interim report analyzed the serum after each collection period with only one measurement per time period. In an effort to minimize experimental error, including systematic and random error in the analytical method, we initiated a more definitive analytical study on nine of the original 27 subjects. Serum samples collected from each of the nine subjects over four time points (t_0-t_3) spanning 180 days were measured in triplicate, with all time points from each subject being analyzed in the same analytical run.

The results from this second interim analysis suggest that the mean serum half-life for PFOS was 8.7 years (SD = 6.1; range 2.3-21.3). The mean serum half-life for PFOA was 4.4 years (SD = 3.5; range 1.5-13.5). The mean half-life for PFHS, which was deferred in the first interim report due to inconsistent results, was an uninterpretable -2.27 years (SD = 23.1; range -47.63-30.12). Half-life estimates for both PFOS and PFOA remain higher than those reported in laboratory animals.

This self-consistent data-set based on triplicate analysis of nine retirees allowed for an estimation of the serum fluorochemical half-life for PFOS, PFOA and PFHS with several caveats. Chief among these limitations are the following: no effort was made to determine or control for retiree re-exposure to PFOS, PFOA or PFHS during the study time-period, although retirees were not present in the production plant. Second, because PFOS is a metabolic product of compounds known to be present in the subject's blood, PFOS is possibly being produced in the body during the course of the study. Both

exposure to, and metabolic production of target analytes will lead to artificially long half-life estimations.

We will continue to collect retiree serum samples from the 27 subjects on an annual basis for the next two years. The samples will be stored frozen (-70°C) and not analyzed until sample collection is completed. At that time, it is our intention to analyze all study samples, beginning at t_0, during the same analytical run to assure that any experimental biases equally affect every serum sample. We do not foresee any further interim reports until the sample collection and laboratory analysis are completed.

INTRODUCTION

In May, 2000, 3M announced that it would voluntarily cease producing the parent molecule, perfluorooctanesulfonyl fluoride (POSF) and its related products due to concerns about biopersistence and widespread exposure to human populations and wildlife (Hansen et al, 20001; Giesy and Kannan, 2001). These POSF-related products contain chemicals, either as intentional components or as impurities (residuals), which could degrade or metabolize to perfluorooctanesulfonate (PFOS), a compound that apparently does not undergo further metabolic or environmental degradation. PFOS and, in particular, various alkyl-substituted perfluorooctanesulfonamido compounds have been utilized in a wide variety of industrial and consumer products. In addition, 3M has manufactured salts of perfluorooctanoate (PFOA) and perfluorohexanesulfonate (PFHS) for industrial uses. These chemicals may also be found as residual impurities in POSF-related products.

Substantial toxicology and epidemiology research has been conducted on PFOS and PFOA (3M Company, 2000; Alexander et al, 2001a; 2001b; Butenhoff et al, 2001; Gilliland and Mandel, 1996; Kennedy, 1985; Kennedy et al, 1986; Olsen et al, 1998; 1999; 2000; Seacat et al, 2001a; 2001b). As the primary objective of this study was to quantify the serum half-life of PFOS and PFOA in humans, a brief synopsis follows which describes the pharmacokinetics of these two chemicals.

Perfluorooctanesulfonate (PFOS)

In animals, PFOS distributes predominantly to the blood and liver, with liver concentrations being potentially several times higher than serum concentrations, depending on species and dose. After male rats were given single intravenous doses (4.2 mg/kg) of ^{14}C PFOS it was found that the carbon-14 in liver and plasma represented 25

and 3 percent of the dose, respectively, after 89 days (Johnson and Ober, 1979; Johnson et al, 1979). During the 89-day post-dose period, 30.2% of the administered ^{14}C had been excreted in the urine and 12.6% had been excreted in the feces. Whole body elimination in the male rat appeared to be biphasic. Initial redistribution from the plasma yielded a plasma elimination half-life of 7.5 days. However, 89 days post-treatment, measurements of ^{14}C indicated that the half-life of elimination exceeded 89 days in the male rat. Significant enterohepatic circulation was likely as cholestyramine administered in the diet to rats after a single intravenous dose of PFOS increased fecal elimination 9.5 times over control animals (Johnson et al, 1984).

In another study, cynomolgus monkeys were dosed by oral capsule with the potassium salt of PFOS at dosages of 0, 0.03, 0.15 and 0.75 mg/kg/day for 182 days (Seacat et al, 2001a). End-of-treatment PFOS concentrations averaged 0.12, 15, 75 and 172 ppm in serum and 0.12, 20, 64 and 334 ppm in the liver. Liver-to-serum PFOS ratios were comparable in all dose groups, with a range of 1:1 to 2:1. The serum PFOS elimination curves appeared to be multiphasic at the 0.75 mg/kg/day dose whereas at the 0.15 mg/kg/day dose elimination curves appeared more linear (0.03 mg/kg/day dose group was not a recovery group). Toward the end of the one year recovery period, the slope of the two recovery group elimination curves were similar suggesting that the PFOS elimination half-lives were approximately 200 days for both dose groups. Liver PFOS concentrations decreased in the same proportion as the serum concentrations. Liver to serum PFOS ratios showed no dose-related differences. Thus, the whole-body PFOS burden elimination rate was estimated to be proportional to the serum and liver elimination rates, in agreement with kinetic studies in the rat (Johnson et al, 1979).

Ammonium Perfluorooctanoate (PFOA)

Excretion rates of PFOA have been observed in rats, and found to be different by gender and route of excretion. Following single intravenous doses of ^{14}C-ammonium perfluorooctanoate in rats, Johnson et al (1984) reported that females excreted virtually all the administered ^{14}C within 1 day. Urinary excretion for males was about 50% of the dose by day 6 and 83% by day 36. Fecal ^{14}C excretion for females was 1.5% by 3 days and for males was 5.4% by 36 days. Rapid urinary excretion of ^{14}C following oral doses of ^{14}C PFOA was also shown to occur in pregnant rats.

Excretion rates also varied by species studied. Dupont studied excretion of radiolabeled PFOA in four species (DuPont, 1982). Excretion as a percentage of administered dose 120 hours after dosing was in the following order; female rat, male and female rabbit and male hamster (>99%); female hamster (60%); male rat (39%); male and female mice (21%). Of note, the administered dose and routes of exposure and excretion were not specified in this study report.

Rats and dogs responded to cholestyramine administration that increased PFOA excretion rates (Johnson et al, 1984). In male rats administered single intravenous doses of ^{14}C PFOA, cholestyramine (4% w/w in feed) increased cumulative 15-day fecal ^{14}C excretion 9.8-fold versus controls. Total ^{14}C excretion (feces plus urine) was also enhanced (84.3% of dose vs. 71.8% for controls). There was no difference between the renal clearances of ^{14}C in male and female dogs either before or after probenecid. Glomerular filtration rates of PFOA were similar in rats and dogs. (Hanhijarvi et al., 1982)

Elimination half-life has been experimentally determined in rats, dogs and primates. In rats, experimental results showed the same differential elimination rate by

gender (i.e. female>male) as well as half-life differences by route of exposure. Following a single oral dose of ^{14}C ammonium perfluorooctanoate in male rats, the plasma half-life was 4.8 days (Johnson et al. 1979). In female rats, over 90% of the intravenous dose was recovered in the urine within the first 12 hours. The whole body elimination half-life of PFOA in male and female rats was 15 days and less than one day, respectively, following a single 4-mg/kg-intraperineal dose (Vanden Heuval et al, 1991). The half-life of PFOA in the liver was 60 hours for female rats and 210 hours for male rats (Ylinen, et al 1990). The decreased excretion rate (i.e., increased elimination half-life) in males was also observed in dogs. The plasma half-life of PFOA was longer in male dogs (473 to 541 hours) than in females (202 to 305 hours) (Hanhijarvi et al., 1982).

The elimination half-life appears to be similar in male rats exposed to either inhalation or dermal exposure. Following repeated inhalation exposures to PFOA over a two-week period, blood organic fluoride levels in male rats showed a half-life of five to seven days (Kennedy et al., 1986). A blood half-life of five to seven days was seen following repeated dermal exposure in male rats (Kennedy, 1985).

Male cynomolgus monkeys received daily oral (capsule) doses of 0, 3 10 and 30 (reduced to 20) mg/kg/day of ammonium perfluorooctanoate for 26 weeks (Butenhoff et al 2001). Dose-dependent increases in liver weight occurred in all treated groups. Body weights were decreased in the 30/20 mg/kg dose group. Serum PFOA concentrations were variable, reached steady state and cleared in weeks and was not directly proportional to dose. Liver PFOA concentrations were also not directly proportional to dose and cleared within three months. At a six-month recovery sacrifice, the two 10 mg/kg monkeys had liver PFOA concentrations that returned to control levels. During the recovery period, serum half-lives of PFOA among the 10 mg/kg/day dose group (only treatment group in the recovery period) was estimated at less than 30 days.

Human Data

Although it was reported that the serum half-life of perfluorooctanesulfonate (PFOS) in humans may range between 1000 and 1500 days (Olsen et al 1999), this estimate was based on just 3 subjects with high variability due to different assays used over time. Published serum half-life of PFOA data is even more limited. Ubel et al (1980) reported an approximate half-life of 18 months for one ammonium perfluorooctanoate production worker whose serum organic fluorine level declined from 70 ppm to 40 ppm. Because of these limited data, a study was designed to quantify serum half-lives for several fluorochemicals, including PFOS and PFOA from retirees of the 3M Decatur and Cottage Grove fluorochemical production plants. A first interim report was issued which suggested the serum half-life of PFOS in humans may be less than originally expected (Burris et al 2000). However, this report was subsequently discounted in a letter (Zobel, 2001) to the U.S. EPA which noted that the first interim report analyzed samples collected from each subject at different time periods with only one measurement per time period. A more definitive analysis was designed to analyze a subset of the serum samples from each time-point in triplicate, with all time points from each subject being analyzed in the same analytical run. This self-consistent data-set would then allow for statistical evaluation of the precision of the measurement and assure that all systematic error inherent in the assay equally affected each sample used for half-life determination. The purpose of this second interim report is to present these findings.

METHODS

This is the second interim report for this ongoing study and summarizes the study activity from November 1998 (t_0) through May, 2000 (t_3), a total of 4 measurements over an 18 month time period.

The overall research design is a prospective experimental study that obtains multiple serial blood samples from retirees throughout the course of a five-year period. Initially, the serum half-lives of seven fluorochemicals were considered: PFOS, PFOA, perfluorohexanesulfonate (PFHS), N-ethyl perfluorooctanesulfonamidoacetate (PFOSAA), N-methyl perfluorooctanesulfonamidoacetate (M570), perfluorooctanesulfonamide (PFOSA) and perfluorooctanesulfonamidoacetate (M556). However, most measurements of PFOSAA, M570, PFOSA and M556 were below the limit of quantitation and thus did not lend themselves to half-life calculations. Therefore, these four fluorochemicals have been discontinued for subsequent assay analyses.

Study Participants

Twenty-four Decatur, Alabama retirees have voluntarily agreed to participate in this research effort involving multiple serum sample collection cycles. The retirees were invited to participate based on having prior work assignments in the chemical division. These participants were eligible for study selection if they retired from the 3M Decatur Chemical Plant between January 1, 1995 and January 1, 1998. Thirty-four individuals were initially identified and 24 individuals (71%) agreed to participate in the study. In addition, three retirees from Cottage Grove Chemical Division were invited to participate for a total of 27 study participants. Informed consent was obtained from each study participant prior to study initiation. The majority of the twenty-seven participants were

male, only two were female. All participants were long-term 3M employees having worked an average of 28 years at either the Decatur or Cottage Grove Chemical Division. The average age of the participants at the time of the first collection (t_0) was 60 years (range: 55-74). The mean number of months from retirement to the start of the study was 30 months (range: 5-130 months).

Sample Collection and Intervals

To date, there have been five collection periods for the Decatur retirees: November 1998 (t_0), June 1999 (t_1), November 1999 (t_2), May 2000 (t_3) and February 2001 (t_4). Cottage Grove retirees have participated in four collection periods: June 1999 (t_0), November/December 1999 (t_1), May 2000 (t_2) and December 2000 (t_3).

The study participants were notified by letter of the sampling dates. Decatur retirees were invited to the Decatur Medical Department for sample collection. A laboratory technician or the Decatur site occupational health nurse performed the venipuncture. The serum samples were stored frozen until shipped on dry ice to the 3M Medical Clinic in St. Paul, Minnesota. Upon arrival in St. Paul, the samples were stored frozen until transferred to Northwest Bioanalytical Laboratory (NWB, Salt Lake City, UT), the designated contract laboratory. Cottage Grove retirees had their serum collected by the laboratory technician in the 3M Medical Clinic. Each participant completed a brief medical history questionnaire containing information about current medications and disease diagnoses. Participants have received letters informing them of their individual fluorochemical results, as the data became available from the laboratory.

Analytical Method

High-performance liquid chromatography mass spectrometry/mass spectrometry (HPLC/MSMS) has been utilized to analyze all serum samples for PFOS, PFOA and PFHS using methods described by Hansen et al (2001). The analytical method for determining the concentration of specific fluorochemicals in human serum was quite complex and, briefly, involved the following steps. First, serum was diluted with water, buffer, and ion pairing reagent, and then liquid-liquid extracted with methyl tert-butyl ether (MTBE). The organic layer was removed, subsequently dried, and brought up to volume with a methanol/water diluent. The final sample diluent contained the fluorochemicals extracted from the serum along with many other MTBE soluble serum compounds. The extract was then analyzed with HPLC/MSMS and quantified against an extracted standard curve prepared by spiking an aliquot of human serum with varying levels of analyte and extracting the spiked fluorochemical in the same manner as the samples.

Target Analytes

Summarized in Table 1 are the time periods and their corresponding sample identifications, collection dates, number of unique participants, target analytes, analytical laboratory, and matrix for the t_0 through t_3 data collections. At the end of each of the first three collection periods (t_0-t_2) a single serum sample from each study participant was submitted to NWB for fluorochemical analyses. All analytes were quantified by NWB the designated contract laboratory with the exception of M556 and M570 from the t_0 collection date. These analytes were quantified by 3M Environmental Laboratory. Analytes that measured above the limit of quantitation (ULOQ) were reanalyzed after dilution with rabbit sera. Twenty-seven retirees participated in the t_1 collection period.

This was the first collection period that included the three Cottage Grove retirees who agreed to participate in the study. At t_1, a single sample for each retiree was analyzed by NWB for all seven analytes. All samples were quantitated versus a calibration curve prepared using human serum. Samples from the t_3 collection period were analyzed for only three of the seven fluorochemicals (PFOS, PFOA and PFHS) as the decision was made to discontinue analyzing for PFOSAA, M570, PFOSA and M556.

In an effort to minimize experimental error including systematic and random error in the analytical method we initiated a more definitive analytical study on nine of the 27 subjects. These nine individuals represented the range of PFOS and PFOA concentrations that had been measured in prior analyses. All nine subjects were Decatur retirees. Each had their serum from the four time periods (t_0 - t_3) measured in triplicate, with all four time-points analyzed in the same analytical run. This approach allowed for statistical evaluation of the precision of the measurement and assured that all experimental biases equally affected each sample used for half-life determination. This interim report summarizes the findings from these nine individuals. [Note: Single measurements were made on t_3 samples from the remaining 18 individuals but these are not included in this report due to the lack of simultaneous triplicate analyses of all time points (t_0- t_3). Samples collected at the t_4 time period (February 2001) for the 27 subjects are stored frozen at -70° C and will not be analyzed until data collection is completed.]

Reference Material

The reference material purity for PFOS, PFOA and PFHS was not available prior to the conduct of this study. Therefore, the reference material purity was initially assumed to be 100% for samples t_0-t_2. 3M contracted with Centre Analytical Laboratories, Inc. (State College, PA) to determine the absolute concentration of PFOS,

PFOA, and PFHS in the NWB stock solution used to prepare the analytical standards and controls for the reported sample analyses. Based on Centre Analytical's results. the concentrations of the calibration and quality control samples were corrected accordingly: PFOS (correction factor = 0.836); PFOA (correction factor = 0.909); and PFHS (correction factor = 0.855) for all analyses in this interim report.

Statistical Analyses

Serum half-lives were calculated using a one-compartmental model. The mathematical expression of this first-order process is a monoexponential equation, $C = C_0 \cdot e^{-k_{el}t}$, where C is the serum concentration, k_{el} is the first-order elimination rate constant, and t is the time of blood sampling. The logarithmic equation for this exponential function has the general form of an equation describing a straight line: $\log C = \log C0 - (k_{el} \cdot t)/2.303$. The half-life of elimination can be calculated after k_{el} has been determined from the slope of the line where $t_{1/2} = 0.693/k_{el}$.

RESULTS

Presented in Tables 2 and 3 are the individual and mean data, respectively, for the nine Decatur employees whose t_0-t_3 measurements were subjected to triplicate analyses during the same batch analyses. There are seven males and two females. The average age of the nine retirees at the study initiation (t_0) is 61 years (range 55-64; SD = 3.2). The mean number of years employed at Decatur is 27.7 years (range 20-33; SD = 4.8). Their number of months retired from Decatur average 18.9 months (range 5-38; SD = 10.5). Their average body mass index (BMI) is 27.9 (range 22.5-33; SD = 3.6). The mean PFOS, PFOA, and PFHS values at study initiation (t_0) were 0.89 ppm (range 0.11-3.53

ppm; SD = 1.07), 0.72 ppm (range 0.06-1.84 ppm; SD = 0.64), and 0.31 ppm (range 0.02-1.25 ppm; SD = 0.40), respectively. The mean serum half-life for PFOS was 8.67 years (range 2.29-21.3 years; SD = 6.12). The mean serum half-life for PFOA was 4.37 years (range 1.50-13.49 years; SD = 3.53). The mean serum half-life for PFHS was -2.27 years (range -47.63 - 30.12 years; SD = 23.14).

Multivariable regression analyses examined the influence of age, BMI, number of years worked or years since retired on the serum-half life. None of these variables were significant predictors of the serum half-lives.

DISCUSSION

The results from the first interim analysis suggested that the serum half-life of PFOS in humans was likely in the range of 139-640 days with a median half-life of 270 days. The serum half-life of PFOA appeared to be approximately one year. The half-life for PFHS was deferred because the assay measurement of PFHS was inconsistent (e.g., many subsequently collected samples were at higher levels than initial samples). There were several limitations noted in the first interim report, the most important being the limited data available, to date, and the range of the serum levels measured (PFOS range 0.2-2.0 ppm; PFOA 0.1-3.1 ppm). In addition, serum concentrations were based on a single measurement of each collected sample with the analytical measurements being conducted on different days and using slightly different analytical methods. This created an imprecise assessment of the serum fluorochemical concentrations. Finally, reference material purity was not determined until after the t_0-t_2 samples had been analyzed. The lack of adjustment for the reference material likely biased the fluorochemical values from 9-16% depending on the specific analyte. Because of these limitations, a subset of nine retirees had all their serum fluorochemical concentrations remeasured in triplicate.

The results from this second interim analysis suggested that the mean serum half-life for PFOS was 8.7 years (SD = 6.1); however, the range of values 2.3 years to 21.3 years suggests variability that remains unaccounted for. This human serum half-life estimate is considerably higher than those reported in laboratory animals (Johnson and Ober 1979; Johnson et al 1979; Seacat et al 2001a; 2001b) although laboratory animal's serum half lives were not short (rats > 89 days; monkeys = 220 days). This may be due, in part, to the fact that serum concentrations in these nine retirees were an order of magnitude lower than the end-of-study PFOS concentrations used to calculate the serum half-lives in the cynomolgus monkey study (Seacat et al 2001a).

The mean serum half-life data for PFOA was 4.4 years (SD = 3.5) for these nine retirees, in contrast to the laboratory animal data that suggested serum PFOA half-lives of 5-7 days (male rat). The inconsistency between these human data and laboratory animal data remains unexplained.

We remain perplexed regarding the serum half-life of PFHS. Three of the nine subjects showed increased serum PFHS concentrations over time, which resulted in an uninterpretable average serum half-life mean of -2.3 years. We are unaware whether these workers may have received occupational or non-occupational exposures that may have distorted their findings. The remaining 6 subjects had mean serum half-lives that ranged between 2.9 and 30.1 years.

The refined approach used in this assessment for nine retirees allowed for an estimation of the serum fluorochemical half-life for PFOS, PFOA and PFHS with several caveats. First, while these retirees rarely entered the plant premises, no effort has been made to determine or control for retiree exposure to PFOS, PFOA or PFHS during the study time period (t_0 - t_3). However, exposure to these compounds could result in an artificially long determination of serum half-life. Because POSF-based fluorochemical

production has been discontinued, the opportunity for future exposures is unlikely. Second, because PFOS is a metabolic product of compounds known to be present in these subjects blood, PFOS is likely being produced in the body during the course of the study, which again will artificially extend the measured half-life. Third, because subjects' blood contained concentrations of fluorochemicals which varied by a factor of 30, the data cannot be pooled or averaged unless the serum concentration decay curve shows first-order kinetics, which do not depend on concentration. Fourth, because subjects have a significant concentration of fluorochemical compounds in their blood in addition to PFOS, it is possible that unknown interactions or processes are at work which affect the actual or measured serum half-lives. Fifth, direct comparison of the PFOS half-life determined from humans and laboratory animals may not be sound due to potentially different protein binding sites and affinities. Finally, the data quality requirements specify that quality control samples and curve points must be within 20% of the actual concentration, therefore, a known systematic error of as much as +/- 20 % may be present and still satisfy the data quality criteria of the analytical method.

Since systematic error is additive, comparing two data-sets analyzed during different analytical runs or on different days may make such discrepancy increase to +/- 40%. This would not include random error. We attempted to remove systematic error by self-consistent data-set analysis of replicate samples. Any comparison of the results from this self-consistent data-set of nine subjects with the results obtained from the 18 subjects over 4 different time periods with systematic error would be difficult to interpret. Therefore, for this interim report, in order to reduce measurement error, we have chosen to report only the self-consistent data-set.

We will continue to collect retiree serum samples from the 27 subjects on an annual basis for the next several years. The samples will be stored frozen (-70°C) and not

analyzed until sample collection is completed. It is our intention to analyze all study samples, beginning at t_0, during the same analytical run to assure that any systematic biases equally affect every serum sample. We do not foresee any further interim reports until the sample collection and laboratory analyses are completed.

REFERENCES

3M Company (2000). SIDS Initial Assessment Report Perfluorooctane Sulfonic Acid and its Salts. St. Paul (MN):3M Company, September 20, 2000.

Alexander BH (2001a). Mortality study of workers employed at the 3M Decatur facility. Final Report. Minneapolis (MN):University of Minnesota, April 26, 2001.

Alexander BH (2001b). Mortality study of workers employed at the 3M Cottage Grove facility. Final Report. Minneapolis (MN):University of Minnesota, April 26, 2001.

Burris J, Olsen G, Simpson C, Mandel J (2000) Determination of Serum Half-Lives of Several Fluorochemicals, Interim Report #1. St. Paul (MN):3M Company, June 8, 2000.

Butenhoff J, Costa G, Elcombe C, Farrar D, Hansen K, Iwai H, Jung R, Kennedy G, Lieder P, Olsen G, Thomford P (2001). Toxicity of ammonium perfluorooctanoate (APFO) in male cynomolgus monkeys after chronic oral dosing. Toxicol Sci (submitted).

DuPont Company, Haskell Laboratory (1982). Excretion and disposition of ^{14}C-ammonium perfluorooctanoate in male and female rats, mice, hamsters, and rabbits. Unpublished Report.

Gilliland FD, Mandel JS (1996). Serum perfluorooctanoic acid and hepatic enzymes, lipoproteins and cholesterol: a study of occupationally exposed men. Am J Ind Med 129:560-568.

Giesy JP, Kannan K (2001). Global distribution of perfluorooctane sulfonate and related perfluorinated compounds in wildlife. Environ Sci Tech 35:1339-1342.

Hanhijarvi H, Phaug R, Singer L (1982). The sex-related difference in perfluorooctanoate excretion in the rat. Proc Soc Exp Biol Med 171:51-55.

Hansen KJ, Clemen LA, Ellefson ME, Johnson HO (2001). Compound-specific, quantitative characterization of organic fluorochemicals in biological matrices. Environ Sci Technol 35:766-770.

Johnson JD, Ober RF (1979). Absorption of FC-95-14C in rats after a single oral dose. St Paul (MN):Riker Laboratories, Inc. Project No., 890310200.

Johnson JD, Gibson SJ, Ober RE (1979). Extent and route of excretion and tissue distribution of total carbon-14 in rats after a single intravenous dose of FC-95-14C. St Paul (MN):Riker Laboratories, Inc. Project No. 8900310200.

Johnson JD, Gibson SJ, Ober RE (1984). Cholestyramine-enhanced fecal elimination of carbon-14 in rats after administration of ammonium [^{14}C]perfluorooctanoate or potassium [^{14}C]perfluorooctanesulfonate. Fund Appl Toxicol 4:972-976.

Kennedy G, Hall G, Brittelli J, Chen H (1986). Inhalation toxicity of ammonium perfluorooctanoate. Fd Chem Toxicol 24:1325-1329.

Kennedy G (1985). Dermal toxicity of ammonium perfluorooctanoate. Toxicol Appl Pharmacol 81:348-355.

Olsen GW, Gilliland FD, Burlew MM, Burris JM, Mandel JS, Mandel JH (1998). An epidemiologic investigation of reproductive hormones in men with occupational exposure to perfluorooctanoic acid. JOEM 40:614-622.

Olsen GW, Burris JM, Mandel JH, Zobel LR (1999). Serum perfluorooctane sulfonate and hepatic and lipid clinical chemistry tests in fluorochemical production employees. JOEM 41:799-806.

Olsen GW, Burris JM, Burlew MM, Mandel JH (2000). Plasma cholecystokinin and hepatic enzymes, cholesterol and lipoproteins in ammonium perfluorooctanoate production workers. Drug Chem Toxicol 23:603-620.

Seacat AM, Thomford PJ, Hansen KJ, Olsen GW, Case MT, Butenhoff JL (2001a). Subchronic toxicity studies on perfluorooctanesulfonate potassium salt in cynomolgus monkeys. Toxicol Sci (in press).

Seacat AM, Thomford PJ, Hansen KJ, Clemen LA, Butenhoff JL (2001b). Sub-chronic dietary toxicity of potassium perfluorooctanesulfonate in rats. Toxicology (submitted).

Ubel FA, Sorenson SD, Roach DE (1980). Health status of plant workers exposed to fluorochemicals, a preliminary report. Am J Ind Assoc J 41:584-589.

Vanden Heuvel J, Kuslikis B, Van Refelghem M, Peterson R (1991). Tissue distribution, metabolism and elimination of perfluorooctanoic acid in Male and Female Rats. J Biochem Toxicol 6(2):83-92.

Ylinen M, Koho A, Hanhijarvi H, Peura P (1992). Disposition of perfluorooctanoic excretion in the rat. Proc Soc Exp Biol Med 171:51-55.

Zobel L (2001). Serum Half-Life of Perfluorooctane Sulfonate. Letter to EPA. St. Paul (MN):3M Company, April 25, 2001.

Table 1 Chronology of Serum Sample Collection

Time Period	Collection Date	Unique Participants	Samples per Participant	Target Analytes	Laboratory	Matrix
t0 (samples: 9276-9299)	11/1998	24 (All Decatur Retirees)	1	PFOS, PFOA, PFHS, PFOSA, PFOSAA M556, M570	NWB, 3M Environ-mental	Human, select samples diluted with rabbit sera
t1 (samples: 9406-9432)	06/1999	27 (3 Cottage Grove Retirees agree to participate)	1	PFOS, PFOA, PFHS, PFOSA, PFOSAA M556, M570	NWB	Human
t2 (samples: 9569-9595)	11/1999	27	1	PFOS, PFOA, PFHS, PFOSA, PFOSAA M556, M570	NWB	Human
t3 (samples: 9871-9897)	05/2000 and Repeat analysis for: 11/1998, 06/1999, 11/1999	18 9	4* 12**	PFOS, PFOA, PFHS	NWB	Human
t4	02/2001	26	1	***	***	***

* one serum sample from each of the following screening periods: t0, t1, t2, and t3

** three serum samples from each of the following screening periods: t0, t1, t2, t3

*** samples stored frozen -20°

Table 2 Individual Data for Employees with Triplicate Analyses

Employee/ Usual Job	Age	Years at Plant	Months Retired	BMI	Gender	PFOS @ t0 (ppm)	½ Life PFOS (years)	PFOA @ t0 (ppm)	½ Life PFOA (years)	PFHS @ t0 (ppm)	½ Life PFHS (years)
A (chemical operator)	60	31	11	28.1	Male	3.5	13.1	1.8	13.5	1.3	- 17.6
B (supervisor)	57	33	18	27.4	Male	1.1	3.7	0.7	4.3	0.4	30.1
C (process engineer/ supervisor)	64	27	38	28.1	Male	0.2	21.3	0.4	3.6	0.1	- 47.6
D (chemical operator)	64	20	27	31.0	Female	0.4	4.4	1.7	3.1	0.1	7.1
E (process operator)	55	20	5	33.0	Female	1.3	12.0	0.9	3.9	0.2	11.0
F (elect/control mech)	63	27	11	24.4	Male	0.5	10.3	0.2	3.9	0.1	12.4
G (cell operator)	62	29	23	32.2	Male	0.3	6.6	0.4	3.5	0.6	- 22.4
H (QC technician)	58	32	11	22.5	Male	0.6	2.3	0.2	1.5	0.2	2.9
I (process engineer)	62	30	26	24.8	Male	0.1	4.4	0.1	2.1	0.02	3.6

Table 3 Summary Half-Life Data for Employees with Triplicate Analyses

	Age @ t0	# Years at Plant	Months Retired	BMI	PFOS @ t0 (ppm)	½ Life PFOS (years)	PFOA @ t0 (ppm)	½ Life PFOA (years)	PFHS @ t0 (ppm)	½ Life PFHS (years)
Average	61	27.7	18.9	27.9	0.9	8.7	0.7	4.4	0.3	- 2.3
Minimum	55	20	5	22.5	0.1	2.3	0.1	1.5	0.02	- 47.7
Maximum	64	33	38	33.0	3.5	21.3	1.8	13.5	1.3	30.1
Std Dev	3.2	4.8	10.5	3.6	1.1	6.1	0.6	3.5	0.4	23.1