CHAPTER 3. TOXICOKINETICS, SUSCEPTIBLE POPULATIONS, BIOMARKERS, CHEMICAL INTERACTIONS

3.1 TOXICOKINETICS

Toxicokinetic data on perfluoroalkyls examined in this profile are available from studies in humans and animals. Most studies in animals administered perfluoroalkyls by the oral route. These data are briefly summarized below.

- Absorption
  - Perfluoroalkyls are absorbed following oral, inhalation, and dermal exposure.
  - Quantitative estimates of the fractional absorption of orally administered perfluoroalkyls in animals range from >50% for PFHxS to >95% for PFOA, PFBA, PFNA, PFDA, PFUnA, and PFDoDA.
  - No quantitative estimates of the fractional absorption of perfluoroalkyls following inhalation or dermal exposure were identified.
- Distribution
  - Perfluoroalkyls are widely distributed in the body, with the highest concentrations in the liver, kidneys, and blood.
  - In the blood, perfluoroalkyls bind to albumin and other proteins.
  - Perfluoroalkyls can be transferred to the fetus during pregnancy and to nursing infants.
- Metabolism
  - Results of available oral and *in vitro* studies suggest that perfluoroalkyls are not metabolized and do not undergo chemical reactions in the body.
  - Although no studies examining metabolism of perfluoroalkyls following inhalation or dermal exposure were identified, metabolism by these exposure routes is not expected.
- Excretion
  - Studies of elimination rates (i.e., half-lives) of perfluoroalkyls show that elimination $t_{1/2}$ values are similar following intravenous, intraperitoneal, and oral exposures. Findings suggest that the route of absorption has no substantial effect on rates of elimination of absorbed perfluoroalkyls.
  - Perfluoroalkyls are primarily eliminated in the urine, with smaller amounts eliminated in feces and breast milk.

- Perfluoroalkyls undergo biliary excretion, but substantial reabsorption occurs; therefore, biliary excretion is not a major elimination pathway.
- Rates of elimination of perfluoroalkyls vary substantially across chemical species and animal species, and show sex differences and age-dependencies within certain species.
- In general, perfluoroalkyl sulfonates are eliminated more slowly than perfluoroalkyl carboxylates; elimination rate decreases with increasing chain length, and increases with increased branching.
- In humans, estimates for elimination $t_{1/2}$ range from hours (PFBA: 72–81 hours) to several years (PFOA: 2.1–8.5 years; PFOS: 3.1–7.4 years; PFHxS: 4.7–15.5 years).
- Evidence for sex differences in elimination of perfluoroalkyls in humans is not as strong as in rats. Menstruation may contribute to faster elimination of PFOS in younger women (≤50 years) when compared to men and older women.

3.1.1 Absorption

Inhalation Exposure. Studies of the absorption of perfluoroalkyls in humans following inhalation exposure were not located; elevated serum concentrations of perfluoroalkyls in workers in fluorochemical production industry have been reported (see Table 5-22), indicating that perfluoroalkyls are absorbed following inhalation exposure. Occupational exposures in these workers are likely to have included inhalation of aerosols of perfluoroalkyls complexed with airborne dusts. Higher serum levels in workers compared to the general population (see Table 5-20) probably reflect a predominant contribution from inhaled perfluoroalkyls.

Studies conducted in rodents provide direct evidence for absorption of inhaled perfluoroalkyls. PFOA was detected in plasma of rats within 30 minutes of initiating nose-only exposures to aerosols (mass median aerodynamic diameter [MMAD]=1.9–2.1 µm) of 1–25 mg ammonium PFOA/m^3. Plasma concentrations increased during the 6-hour exposure, with the highest concentrations observed at 9 hours (3 hours after cessation of exposure) in male rats and at 7 hours (1 hour after cessation of exposure) in females (Hinderliter et al. 2006a). Assuming an elimination $t_{1/2}$ of absorbed PFOA of approximately 160 hours in male rats, a peak plasma concentration at 9 hours would correspond to an absorption $t_{1/2}$ of approximately 1.3 hours (see discussion below, Equations 3-1 and 3-2). The earlier time of highest plasma concentration observed in female rats appears to be associated with faster elimination of absorbed PFOA in female rats, compared to male rats (see Section 3.1.4).

Nose-only exposure of male rats to dusts of ammonium perfluorononanoate induced significant increases in absolute and relative liver weight, assessed 5 and 12 days after exposure, providing indirect evidence of absorption of this compound through the respiratory airways (Kinney et al. 1989).

Oral Exposure. Studies of absorption of perfluoroalkyls through the gastrointestinal tract in humans are not available. A study of the general population of Europe and North America estimated that the greatest portion of the chronic exposure to PFOS and PFOA results from the intake of contaminated food, including drinking water (Trudel et al. 2008). Direct evidence of oral absorption of perfluoroalkyls was provided in studies that found associations between environmental levels (e.g., drinking water) and perfluoroalkyl concentrations in human serum (Emmett et al. 2006a; Hoffman et al. 2011; Hölzer et al. 2008; Seals et al. 2011; Wilhelm et al. 2008) and by reductions in serum levels after exposures from water were eliminated or reduced (Bartell et al. 2010; Emmett et al. 2009).

Animal data provide quantitative estimates of the fractional absorption of orally administered PFOA, PFOS, PFBA, PFHxA, PFHxS, PFHpA, PFNA, PFDA, PFUnA, and PFDoDA, with estimates ranging from >50% for PFHxS to >95% for PFOA, PFBA, PFNA, PFDA, PFUnA, and PFDoDA. Greater than 95% of an oral dose of ammonium [^{14}C]PFOA was absorbed in rats that received single gavage doses ranging from 0.1 to 25 mg/kg (Kemper 2003). In male and female mice, comparison of the 24-hour area under the curve (AUC) for oral and intravenous administration showed that 90–100% of the oral dose was absorbed for PFOA (females), PFNA (males and female), PFDA (males and females), PFUnA (males and females), and PFDoDA (males and female); however, absorption of PFOA in males was 80%, compared to 100% in females (Fujii et al. 2015a, 2015b). Gannon et al. (2011) estimated an absorption fraction of 99% based on 168-hour urinary excretion of ^{14}C in male and female rats and mice following single oral doses of 2 or 100 mg/kg ^{14}C-PFHxA. Based on comparison of the AUC for oral and intravenous administration, the estimated oral absorption fractions were 50% in female rats administered a single 10 mg/kg dose of potassium [$^{18}O_{3-}$]PFHxS (Sundström et al. 2012) and 79 and 55% in male and female rats administered a single dose of 4 mg/kg sodium [$^{18}O_{3-}$]PFHxS (Kim et al. 2016b). Sundström et al. (2012) stated that this estimate may not be reliable due to the short (24 hours) observation period. Based on 72-hour urinary excretion of ^{14}C, the estimated fractional absorption of a single dose (50 mg/kg) of ^{14}C-PFHxA was approximately 74% in male rats, 90% in female rats, and 80% in male and female mice (Iwai et al. 2011). A comparison of ^{14}C disposition in rats, mice, hamsters, and rabbits following an oral dose of 10 mg ammonium [^{14}C]PFOA/kg showed that similar fractions of the dose were absorbed (Hundley et al. 2006). The estimated absorbed fractions (i.e., ^{14}C in tissues, urine, and exhaled air measured 120–168 hours after the dose) in males were 89% in rats, 82% in mice, 92% in hamsters, and

88% in rabbits. Corresponding values for females were 76% in rats, 61%, in mice, 75% in hamsters, and 88% in rabbits. These estimates exclude ^{14}C excreted in feces, which may have been absorbed and secreted in bile before excretion (see Section 3.1.4). Fasting appears to increase absorption of PFOA. Plasma PFOA concentrations in rats, 24 hours following a gavage dose of 10 mg ammonium PFOA/kg, were 2–3 times higher when administered to fasted rats, compared to fed rats (Hinderliter et al. 2006b). The estimated absorption fractions of ingested ammonium [^{14}C]PFOA or potassium [^{14}C]PFOS (administered as a 4.2 mg/kg oral dose) were >93 and >95% in rats, respectively (Johnson and Ober 1979, 1999a). Based on combined urinary excretion and retention in the carcass (excluding the gastrointestinal tract and its contents), the estimated oral absorption fraction of [^{14}C]PFOS (administered as a single 4.2 mg/kg dose of potassium [^{14}C]PFOS) in male rats was >95% (Chang et al. 2012). The estimated absorption fraction of PFBA (administered as 30 mg/kg oral dose of PFBA) was >95% in rats (Chang et al. 2008a). Cumulative excretion of PFBA 24 hours after an oral dose (administered as 10, 30, or 100 mg/kg ammonium PFBA) was approximately 35% in urine and 4–11% in feces in male mice; and 65–69% in urine, and 5–7% in feces in female mice (Chang et al. 2008a).

Studies examining the rate of absorption of PFOA, PFHxA, PFBA, and PFBS show rapid absorption from the gastrointestinal tract, with values for absorption $t_{1/2}$ of <2 hours. For PFOA, the highest observed concentrations of ^{14}C in plasma occurred in male rats at approximately 10 hours (range 7.5–15 hours) following single oral doses ranging from 0.1 to 25 mg ammonium PFOA/kg (Kemper 2003). The elimination $t_{1/2}$ of ^{14}C in plasma estimated in these same animals was approximately 170 hours (range 138–202 hours), corresponding to an elimination rate constant (k_e) of 0.0044 hour^{-1} (range 0.004–0.005 hour^{-1}). The corresponding absorption $t_{1/2}$ of approximately 1.5 hours (k_a=0.45 hour^{-1}) can be calculated from these observations (Equations 3-1 and 3-2):

$$t_{\max} = \ln\frac{k_a}{k_e} \cdot \frac{1}{(k_a - k_e)} \qquad \text{Eq. (3-1)}$$

$$t_{1/2} = \frac{\ln(2)}{k_e} \qquad \text{Eq. (3-2)}$$

Where t_{max} = time of maximum concentration of ^{14}C; k_e = elimination rate constant; and k_a = absorption constant. The absorption rate of PFOA appears to be greater in female rats compared to male rats. The time to peak concentrations of ^{14}C in plasma occurred at approximately 1.1 hour (range 0.6–1.5 hours) in female rats and 10 hours (range 7–15 hours) in male rats following single oral doses ranging from 0.1 to 25 mg ammonium PFOA/kg (Kemper 2003). The elimination $t_{1/2}$ of ^{14}C in plasma estimated in these same animals varied with dose and ranged from 3.2 hours at the lowest dose (k_e=0.23 hour^{-1}) to 16.2 hours at the highest dose (k_e=0.059 hour^{-1}). The estimated absorption $t_{1/2}$ from the observations

made at all doses (0.1, 1, 5, and 25 mg/kg), based on Equations 3-1 and 3-2, was approximately 0.25 hours (range 0.12–0.38 hours). The absorption $t_{1/2}$ of PFBA in male and female rats following administration of a single oral dose (30 mg/kg ammonium PFBA) was 0.23 hours (3.04 $hour^{-1}$) in males and 0.17 hours (4.15 $hour^{-1}$) in females (Chang et al. 2008a). In male and female mice administered 10–30 mg/kg ammonium PFBA, the absorption $t_{1/2}$ was <1 hour, although the absorption rate may be dose-dependent in males, with higher absorption $t_{1/2}$ at doses >30 mg/kg (Chang et al. 2008a). Estimated t_{max} values following administration of single doses (2 or 100 mg/kg) of ^{14}C-PFHxA to rats and mice ranged from 0.3 to 0.8 hours (Gannon et al. 2011). Similar results for were reported by Olsen et al. (2009) based on estimated compartmental pharmacokinetic parameters for PFBS in serum of male and female rats following a single intravenous or gavage dose of 30 mg potassium PFBS. Plasma concentration-time profiles were fit to a two-compartment elimination model. The absorption $t_{1/2}$ can be approximated from these data using Equation 3-1, with the elimination rate constant represented by the fast-phase elimination rate constant estimated for either the oral or intravenous dose. Using the oral or intravenous parameters yielded similar values for the absorption $t_{1/2}$ (0.12–0.16 hours). The estimated t_{max} values following the gavage dose were 0.42 hours in males and 0.33 hours in females. The fast-phase elimination rate constants following the gavage dose were 0.892 $hours^{-1}$ ($t_{1/2}$=0.79 hours) in males and 1.308 $hours^{-1}$ ($t_{1/2}$=0.53 hours) in females. The corresponding values for absorption $t_{1/2}$ were 0.14 hours (k_a=5.0 $hours^{-1}$) in males and 0.12 hours (k_a=5.8 $hours^{-1}$) in females. Use of the fast-phase elimination rate constants estimated following intravenous administration (male: 1.143 $hours^{-1}$; female: 1.956 $hours^{-1}$) yielded values for the absorption $t_{1/2}$ of 0.16 hours in males (k_a=4.30 $hours^{-1}$) and females (k_a=4.45 $hours^{-1}$).

Mechanisms of oral absorption of perfluoroalkyls have not been elucidated.

Dermal Exposure. Dermal exposures of rats to ammonium PFOA have been shown to produce systemic (e.g., liver, immunotoxicity) toxicity in animals (see Chapter 2). Estimates of the amount or rates of dermal absorption in humans or animals have not been reported. PFOA was detected in serum of mice following dermal application of PFOA dissolved in acetone (Franko et al. 2012). The investigators noted PFOA ingestion may have occurred during grooming and may have contributed to the body burden. Dermal absorption of PFOS was assessed following application of single doses of potassium PFOS (doses up to 0.30 mg/kg) and the diethanolamine salt of PFOS (doses up to 20 µg/kg) to clipped, intact skin of rabbits (Johnson 1995a, 1995b). Analysis of the liver 28 days after application showed no increase in content of total organic fluoride compared to controls, indicating that dermal absorption was not detectable at low dose levels using this methodology. Dermal penetration of PFOA has been studied in preparations of isolated rat, mouse, and human epidermis (Fasano et al. 2005; Franko et al. 2012). These

studies indicate that the rat and mouse skin may be more permeable to PFOA than human skin. Approximately 24% of a dermal dose of PFOA (0.5 mg in 1% acetone) was absorbed across isolated full thickness human skin in 24 hours and 45% of the dose was retained in skin (Franko et al. 2012); it is noted that the acetone, as well as the glycerol used to pretreat the skin may have enhanced PFOA absorption. Permeability was sensitive to pH and was higher when the skin was buffered at pH 2.5 ($5.5x10^{-2}$ cm/hour) compared to pH 5.5 ($4.4x10^{-5}$ cm/hour), well above the pKa for the terminal carboxylic acid of PFOA (Franko et al. 2012). This suggests that permeability of the unionized acid is greater than that of the dissociated anion. Lower permeability of ionized PFOA is also suggested by relatively low permeability of the ammonium salt of PFOA in isolated preparations of rat and human skin. Following application of the ammonium salt of PFOA to isolated human or rat epidermis (150 µL/cm^2 of a 20% aqueous solution of ammonium PFOA; approximately 30 mg ammonium PFOA/cm^2), approximately 0.048% of the dose was absorbed across human epidermis and 1.44% was absorbed across rat epidermis in 40 hours. The estimated dermal penetration coefficients were $9.49x10^{-7}$ cm/hour in the isolated human epidermis and $3.25x10^{-5}$ cm/hour in the isolated rat epidermis.

The available data suggest that absorption of PFOA and PFOS through the skin is limited and is of minimal concern as an exposure route. No dermal absorption data were located for other perfluoroalkyls.

3.1.2 Distribution

Available information on the distribution of perfluoroalkyls is obtained from oral exposure studies in laboratory animals and occupational exposure studies in which exposure is predominantly by inhalation. Studies specifically examining the distribution of perfluoroalkyls by inhalation or dermal exposure were not identified. As discussed in Section 3.1.3 (Metabolism), perfluoroalkyls do not undergo metabolism. Therefore, distribution is expected to be the same regardless of the route of administration.

Distribution in Blood. In a study of 60 healthy Chinese participants from the general population, whole blood:plasma ratios for PFOS, PFOA, PFHxA, and PFHxS were 0.65, 0.83, 3.0, and 0.57 (Jin et al. 2016). These results indicate that PFHxA, but not PFOA, PFOS, or PFHxS, enters cellular components of blood. In a study of perfluoroalkyl workers, serum:plasma ratios for PFHxS, PFOS, and PFOA were 1:1, and this ratio was independent of the concentrations measured (Ehresman et al. 2007). The ratio of whole blood:plasma (or serum) was approximately one-half, which corresponded to volume displacement by red blood cells, suggesting that these perfluoroalkyls do not enter cellular components of blood. In studies conducted in animals, most of the PFOA in blood is in the plasma fraction. In rats, 24 or 48 hours

following an oral dose of 11.4 mg ammonium [^{14}C]PFOA/kg, the red blood cell:plasma PFOA concentration ratio ranged from 0.2 to 0.3, suggesting that there was no selective retention of PFOA by red blood cells (Johnson and Ober 1999a). Blood:plasma (or serum) ratios of approximately 0.5 have also been observed in rats following intravenous injection of PFOA (Kudo et al. 2007).

Perfluoroalkyls in plasma bind to serum albumin. The dissociation constant for binding of PFOA to serum albumin is approximately 0.4 mM (0.38 mM, ±0.04 standard deviation [SD] for human serum albumin; 0.36 mM, ±0.08 SD for rat serum albumin) and involves 6–9 binding sites (Han et al. 2003). Given a dissociation constant (K_D) of 0.4 mM and an albumin concentration of approximately 0.6 mM, >90% of PFOA in serum would be expected to be bound to albumin when the serum concentration of PFOA is <1 mM (<440 mg/L). This is consistent with observations of the bound fraction of perfluoroalkyls in plasma of rats that received a gavage dose of 25 mg PFOA/kg (Han et al. 2003, 2005; Ylinen and Auriola 1990), and in human, rat, and monkey plasma incubated *in vitro* with perfluoroalkyls (e.g., PFHxA, PFOA, PFOS, PFNA, PFDA) (Kerstner-Wood et al. 2003; Ohmori et al. 2003). Comparison of dissociation constants for binding of PFOA and PFOS to human serum albumin indicates that PFOS (K_D: $8x10^{-8}$) has a higher binding affinity than PFOA (K_D: $1x10^{-4}$) for albumin, consistent with the longer $t_{1/2}$ of PFOS versus PFOA in humans (Beesoon and Martin 2015; see Section 3.1.4 for additional information). PFOS has also been shown to bind to human hemoglobin *in vitro* (Wang et al. 2016). PFBS was found to bind only to albumin, whereas PFOS, PFOA, and PFHxS were found to have the potential to bind to other human serum binding proteins, including plasma gamma-globulin, alpha-globulin, alpha-2-macroglobulin, transferrin, and beta-lipoproteins (Kerstner-Wood et al. 2003).

Distribution to Extravascular Tissues. Absorbed perfluoroalkyls distribute from plasma to soft tissues, with the highest extravascular concentrations achieved in liver. An analysis of samples from human cadavers attempted to quantify PFOA, PFOS, FOSA, and PFHxA concentrations in serum and liver (Olsen et al. 2003c). The route of exposure was unknown. Mean serum PFOS concentration was 17.7 ng/mL (95% CI 13.0–22.5, range of <6.9 [limit of quantification] to 57 ng/mL, n=24) and was not different in males (18.2 ng/mL, n=13) and females (17.2 ng/mL, n=11). The mean liver concentration was 18.8 ng/g (95% CI 14.1–23.5; range <7.3–53.8 ng/g, n=30). The mean liver:serum concentration ratio was 1.3 (95% CI 0.9–1.7, n=23) and was not different in males (1.3, n=13) and females (1.3, n=10). Most liver and serum concentrations for PFOA, FOSA, and PFHxA were below the limit of quantification; these limits were <17.9–<35.9 ng/mL for PFOA, <7.5–<19.6 ng/g for FOSA, and <3.4–<18.5 ng/mL for PFHxA.

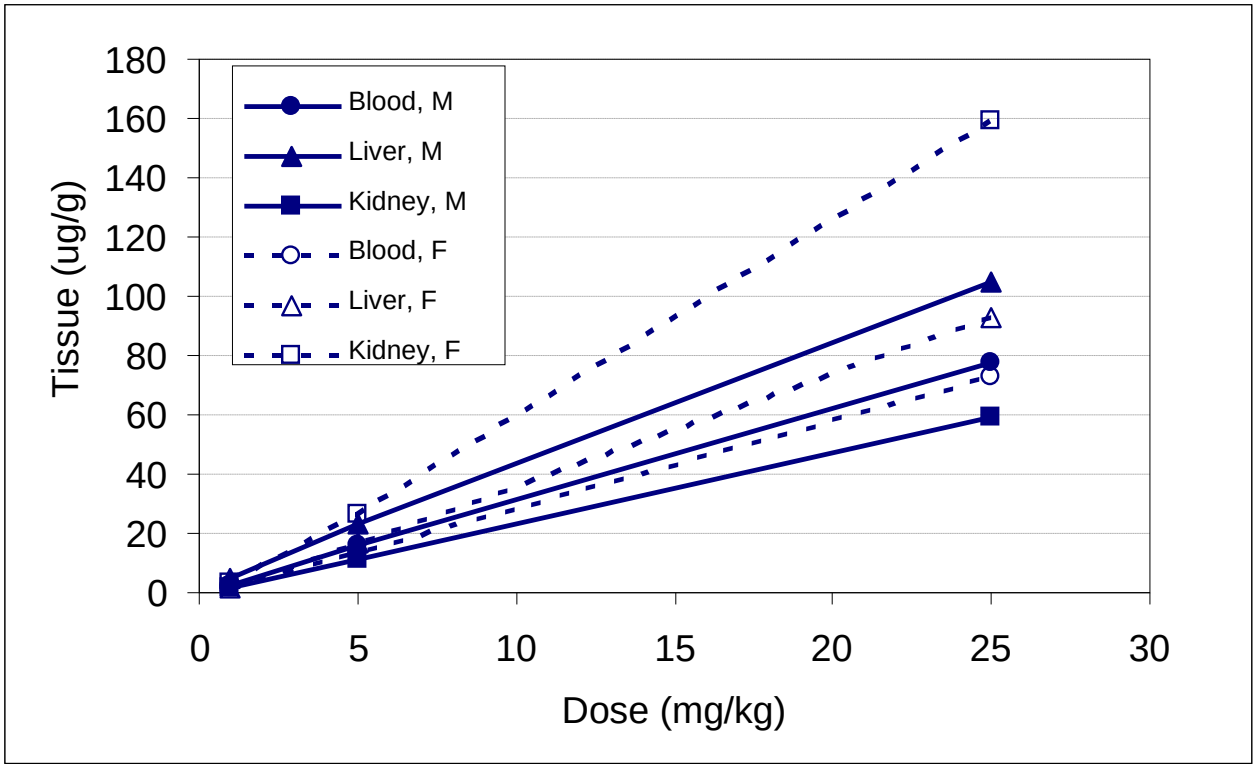
Studies conducted in nonhuman primates and rodents have provided additional information on the distribution of absorbed perfluoroalkyls to extravascular tissues. Distribution, as assessed from tissue perfluoroalkyl concentrations and tissue:serum ratios, exhibits profound species and sex differences as well as dose-dependencies (e.g., tissue levels that change disproportionately with dose). These differences have been attributed, in part, to species and sex differences in elimination kinetics of absorbed perfluoroalkyls and dose-dependence of elimination kinetics (see Section 3.1.4). In general, a consistent finding across species is that the liver receives a relatively high fraction of the absorbed dose and may also experience relatively high tissue concentrations compared with other tissues, with blood (i.e., plasma) and kidney also showing relatively high concentrations. The most extensive investigations of tissue distribution have been conducted in rodents.

Bogdanska et al. (2011) examined distribution of ^{35}S following dietary exposure to adult male C57/BL6 mice to low (environmentally relevant; 0.031 mg/kg/day) and high (experimentally relevant; 23 mg/kg/day) doses of [^{35}S]PFOS for 1–5 days. For both low and high doses after 1, 3, and 5 days of exposure, ^{35}S was distributed to the following tissues: blood, liver, lung, kidney, skin, whole bone, pancreas, spleen, thymus, heart, testes, epididymal fat, fat pads, brain, and muscle; ^{35}S was also detected in tissues throughout the gastrointestinal tract. Similar tissue:blood ratios were observed in both dose groups. In low-dose animals after 5 days of treatment, the highest tissue concentrations (excluding the gastrointestinal tract) were liver (tissue:blood=5.8), followed by lung (tissue:blood=1.4), whole bone, including marrow (tissue:blood=1.1), blood, and kidney (tissue:blood=0.94). In high-dose animals, the highest tissue concentrations were liver (tissue:blood=3.6), followed by lung (tissue:blood=1.6), blood, kidney (tissue:blood=0.81), and whole bone, including marrow (tissue:blood=0.72). A similar pattern of distribution was observed following intravenous administration of [^{14}C]potassium PFOS (4.2 mg/kg) to male rats (Johnson and Ober 1980). For both dose groups, the tissue:blood ratios for all other tissues were <1. In male and female CD-1 mice administered a single oral dose (4.2 mg/kg) of [^{14}C]PFOS, the highest concentrations of ^{14}C was observed in the liver, followed by serum, and then kidney, with similar tissue levels observed in males and females (Chang et al. 2012). In male and female rats fed diets containing 0, 2, 20, 50, or 100 mg/kg [^{13}C]sodium PFOS (equivalent to 0, 0.14, 1.33, 3.21, and 6.34 and 0, 0.15, 1.43, 3.73, and 7.58 mg/kg/day in males and females, respectively) for 28 days, PFOS levels were highest in liver, followed by spleen, heart, and serum. Liver:serum ratios for the 2, 20, 50, and 100 mg/kg/day diets were approximately 52, 42, 41, and 35, respectively, in males and 30, 47, 20, and 23, respectively, in females (Curran et al. 2008). Except for rats fed diets containing 20 mg/kg, the liver:serum ratio in males was higher than in females. No additional data were reported to determine if PFOS distribution differed between male and female rats.

Kemper (2003) determined the distribution of ^{14}C in male and female rats at the approximate time of maximum plasma concentration in both sexes, following single gavage doses of [^{14}C]PFOA (as ammonium PFOA, 0.1–25 mg/kg). This design allows a more direct comparison of patterns of tissue distribution in male and female rats at similar plasma concentrations, even though the elimination kinetics in the female rat are substantially faster than in male rats (see Section 3.1.4). The highest concentrations of ^{14}C were observed in blood, liver, and kidney (Figure 3-1). Liver, blood, and kidney accounted for approximately 22, 22, and 2% of the administered dose of 1 mg/kg in male rats; and 6, 7, and, 3% in female rats (the sex difference reflected more rapid excretory elimination in females). Although blood, liver, and kidney concentrations appeared to increase proportionately with increasing dose in male rats, in female rats, a disproportionately higher concentration in kidney was observed following the 25 mg/kg dose (Figure 3-1). Concentrations in other tissues ranged from 0.1 to 0.25 of that in liver or kidney; concentrations in bone and fat were <0.1 of that in liver or kidney. Profound sex differences and dose-dependencies in tissue concentrations of PFOA were also observed in rats that received oral doses of PFOA for 28 days at doses of 3, 10, or 30 mg PFOA/kg/day (Ylinen et al. 1990; Figure 3-2). Mean serum, kidney, or liver concentrations did not increase proportionally with dose in either sex. Kidney concentrations exhibited a disproportionate increase as the dose increased from 3 to 10 mg/kg/day, with little further increase at the 30 mg/kg/day dose. Sex differences in tissue distribution of PFOA in rats are not explained by sex differences in bioavailability since the differences persist in animals that received parenteral doses of PFOA (Johnson and Ober 1999b; Vanden Heuvel et al. 1991b, 1991c). The differences have been attributed to more rapid elimination of PFOA in female rats, compared to male rats (see Section 3.1.4).

A comparison of PFOA disposition in rats, mice, hamsters, and rabbits showed pronounced species and sex differences (Hundley et al. 2006; Table 3-1). In this study, rats, mice, hamsters, or rabbits received an oral dose of 10 mg ammonium [^{14}C]PFOA/kg and ^{14}C in tissues was measured at 120 or 168 hours (rabbits) hours post-dosing. In male rats, the highest concentrations of ^{14}C occurred in blood, liver and kidney, and all tissues combined accounted for approximately 60% of the dose. However, in female rats, concentrations of ^{14}C in all tissues were below limits of quantification. In mice, liver concentrations were similar in males and females, and liver showed the highest concentrations; ^{14}C levels in all tissues combined were lower in females compared to males. The opposite pattern was evident in hamsters and rabbits, with males having lower tissue levels than females, although, in common with rats and mice,

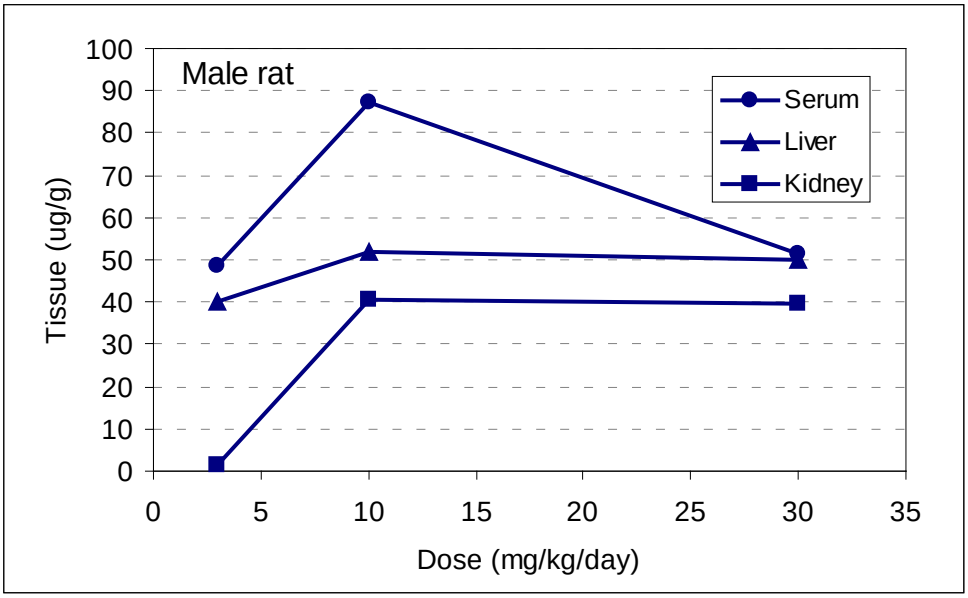
Figure 3-1. Tissue Concentrations of ^{14}C in Male and Female Rats Following a Single Gavage Dose of [^{14}C]PFOA at 1, 5, or 25 mg/kg*

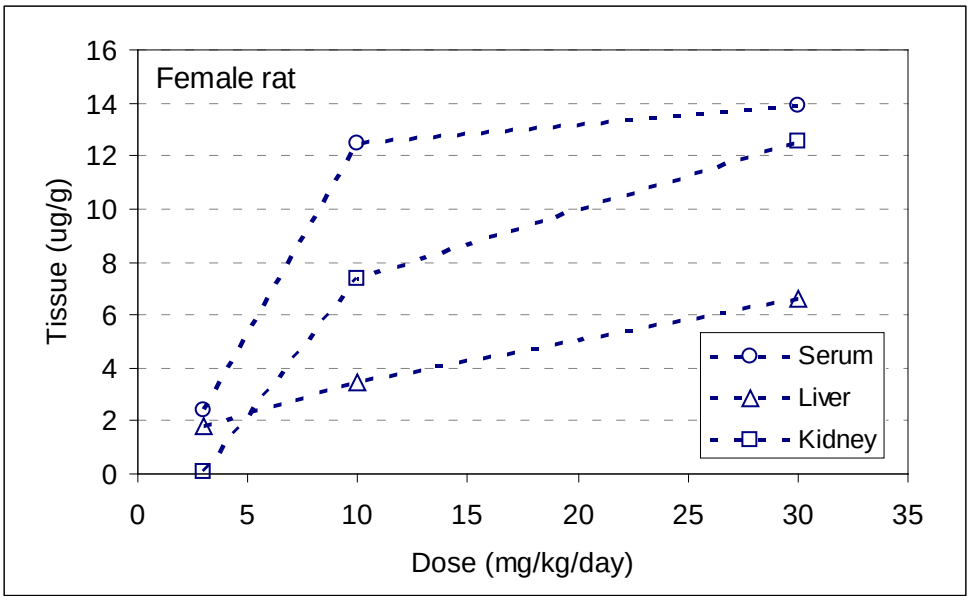


*Tissue levels are measured at time of maximum concentration in each tissue.

Source: Kemper 2003

Figure 3-2. Tissue Concentrations of ^{14}C in Male (Upper Panel) and Female (Lower Panel) Rats Following Oral Doses of PFOA for 28 Days at Doses of 3, 10, or 30 mg/kg/day





Source: Ylinen et al. 1990

Table 3-1. Tissue Distribution and Excretion of ^{14}C-Radioactivity from Both Sexes of Rats, Mice, Hamsters, and Rabbits Dosed with ^{14}C-Labeled APFO[a]

	µg Equivalent per g (mL) wet weight[b]							
	Rat		Mouse		Hamster		Rabbit	
Sample	Male	Female	Male	Female	Male	Female	Male	Female
Blood	23.5	<0.1	13.8	10.1	0.1	8.8	<0.1	0.1
Liver	40.0	<0.1	43.2	45.3	0.3	7.3	0.1	1.5
Kidneys	24.0	<0.1	2.9[c]	2.2[c]	0.2	7.1	0.1	0.4
Lungs	8.7	<0.1	1.4[c]	1.3[c]	<0.1	3.8	<0.1	0.1
Heart	6.4	<0.1	1.2[c]	0.6[c]	<0.1	2.9	<0.1	<0.1
Skin	4.8	<0.01	3.5	0.2	<0.1	3.4	<0.1	<0.1
Testes	3.2	–	0.9[c]	–	<0.1	–	<0.1	–
Muscle	1.9	<0.1	1.1	0.5	<0.1	0.9	<0.1	<0.1
Fat	1.7	<0.1	1.6	1.3	<0.1	1.5	<0.1	<0.1
Brain	0.6	<0.1	0.2[c]	0.8[c]	<0.1	0.3	<0.1	<0.1
	Percent of dose							
Tissues	59.6	0.6	73.6	50.0	0.7	26.5	<0.1	0.3
Urine	25.6	73.9	3.4	6.7	90.3	45.3	76.8	87.9
Feces	9.2	27.8	8.3	5.4	8.2	9.3	4.2	4.6
Expiration	3.6	1.5	5.2	4.4	1.3	2.9	No data	No data
Cage wash	0.6	0.8	4.9	4.9	0.6	2.1	0.5	4.8
Percent recovered	98.5	104.6	95.4	71.4	101.1	86.1	81.6	97.6

[a]The rabbits were sacrificed 168 hours after dosing; all other animals were sacrificed 120 hours after dosing.
[b]The µg equivalent calculations were based on the specific activity of ^{14}C-labeled APFO, which was 1.1×10^6 DPM/mg. The µg equivalent per g wet weight could not accurately be determined below 0.1 µg/g.
[c]Represents the µg equivalents for the entire organ.

APFO = ammonium perfluorooctanoate

Source: Hundley et al. 2006

blood, liver and kidney had the highest concentrations. Male rats that received a single oral dose of 5 mg FOSA/kg had liver FOSA concentrations that were 3–5 times higher than serum concentrations 1 day post-dosing (Seacat and Luebker 2000).

Sex differences in elimination that give rise to sex differences in tissue levels following oral exposure to perfluoroalkyls in rats are not evident in studies conducted with nonhuman primates. Rhesus monkeys that received 3 or 10 mg ammonium PFOA/kg/day for 90 days had liver concentrations of 48 µg/g (one male) or 50 µg/g (one female) at the low dose and 45 µg/g (one male) and 72 µg/g (one female) at the higher dose, with corresponding serum concentrations of 3 and 7 µg/mL, and 9 and 10 µg/mL, respectively (Griffith and Long 1980). Although limited to only one animal per sex, these results suggest

that liver levels did not increase proportionately with increasing dose. A similar observation was made in a study of male Cynomolgus monkeys (Butenhoff et al. 2004c). In male monkeys that received daily oral doses of 3 or 10 mg ammonium PFOA kg/day for 27 weeks, liver PFOA concentrations ranged from 11 to 18 µg/g at the low dose and from 6 to 22 µg/g at the higher dose. Mean serum concentrations measured after 6 weeks of exposure (which may have represented steady-state concentrations) were 77,000 ng/mL in the low-dose group and 86,000 ng/mL in the higher dose group. In this same study, an analysis of serum PFOA kinetics following an intravenous dose of PFOA revealed similar elimination kinetics in males and females (Butenhoff et al. 2004c; see Section 3.1.4). In Cynomolgus monkeys that received daily oral doses of PFOS (0, 0.03, 0.15, or 0.75 mg PFOS/kg/day) for 26 weeks, liver concentrations of PFOS and serum concentration were similar in males and females (liver:serum ratios ranged from 1 to 2) and increased in approximate proportion to the administered dose (Seacat et al. 2002).

Bogdanska et al. (2014) examined distribution of ^{35}S in 20 tissues following dietary exposure of adult male C57/BL6 mice to PFBS (16 mg/kg/day) for 1–5 days. ^{35}S was detected in all tissues and concentrations reached plateau levels after 3 days of exposure. After 5 days, tissue:blood ratios (excluding stomach and small intestine) were >1 for liver (tissue:blood=1.6), kidney (tissue:blood=1.3), whole bone (tissue:blood=1.1), and cartilage (tissue:blood=1.1). At all-time points, approximately 90% of the ingested ^{35}S was recovered in combined blood, liver, bone, skin, and muscle.

Iwabuchi et al. (2017) compared tissue distribution following single doses or 3-month dosing of PFOS (100 µg/kg), PFOA (100 µg/kg), PFHxA (100 µg/kg), and PFNA (50 µg/kg). Following administration of single doses, the tissue:serum (and/or whole blood) ratio was >1 for the liver for PFOS, PFOA, and PFNA, with tissue:serum ratios <1 for kidney, spleen, heart, and brain. For PFNA, the only tissue with a tissue:serum ratio >1 was kidney. After 3 months of exposure, tissue:serum ratios >1 were observed for the liver for PFOA and PFNA, and the liver and kidney for PFOS. For PFHxA, all tissue:serum ratios were <1. Similar to the single dose study, the lowest serum:tissue ratio for all compounds was observed for brain.

Subcellular Distribution. The subcellular distribution of perfluoroalkyls has been examined in rats (Han et al. 2004, 2005; Kudo et al. 2007; Vanden Heuvel et al. 1992b). Two hours following an oral dose of 25 mg ammonium [^{14}C]PFOA/kg, sex differences were noted in the subcellular distribution of ^{14}C in liver; females had approximately 50% of total ^{14}C in the cytosolic fraction compared to 26% in males (Han et al. 2005). The distributions to other cell fractions were: nuclear/cell debris fraction, 30% females, 40% males; lysosomes, 12% females, 14% males; mitochondria, 8% females, 16% males; and

ribosomes, <3% males and females. In kidney, 80 and 70% of the ^{14}C was associated with the cytosolic fraction in males and females, respectively, 16–22% in the nuclear/cell debris fraction, and the remainder in lysosome/mitochondria/ribosome fractions. In liver, approximately 55% of cytosolic ^{14}C was bound to proteins (>6,000 Da) in both males and females, whereas in kidney, 42% of the cytosolic fraction was bound to protein in males and 17% in females. The subcellular distribution of PFOA is dose-dependent. In rats, 2 hours following an intravenous dose of 0.041 mg [^{14}C]PFOA/kg, approximately 5% ^{14}C in the liver was associated with the cytosolic fraction, whereas approximately 45% was in the cytosolic fraction following a dose of 16.6 mg/kg (Kudo et al. 2007). A small component of tissue-associated PFOA and PFDA appeared to be bound covalently to protein. Following an intraperitoneal dose of 9.4 µmol/kg [^{14}C]PFDA or [^{14}C]PFOA (4.2 mg/kg), approximately 0.1–0.5% of liver ^{14}C was bound covalently (i.e., was not removed by repeated extraction with a methanol/ether and ethyl acetate; Vanden Heuvel et al. 1992b). Covalent binding was detected when cytosolic or microsomal fractions of rat liver were incubated *in vitro* with [^{14}C]PFDA (Vanden Heuvel et al. 1992b).

PFOA binds to rat kidney and urine α2u-globulin; dissociation constants were estimated to be approximately 1.5 and >2 mM (for a single binding site) for the proteins isolated from rat kidney and urine, respectively. These values suggest relatively low affinity for the protein, compared to other ligands that are known to induce hyaline droplet nephropathy (10^{-4}–10^{-7} M; Han et al. 2004).

Maternal-fetal Transfer. Perfluoroalkyls can be transferred to the fetus during pregnancy (Cariou et al. 2015; Chen et al. 2017a; Fei et al. 2007; Fisher et al. 2016; Fromme et al. 2010; Glynn et al. 2012; Gützkow et al. 2012; Hanssen et al. 2010, 2013; Inoue et al. 2004; Kato et al. 2014; Kim et al. 2011, Lee et al. 2013; Lien et al. 2013; Liu et al. 2011; Manzano-Salgado et al. 2015; Midasch et al. 2007; Monroy et al. 2008; Needham et al. 2011; Ode et al. 2013; Porpora et al. 2013; Yang et al. 2016a, 2016b). Studies that measured perfluoroalkyls in maternal and fetal cord blood of matched mother-infant pairs found relatively strong correlations (r>0.8) between maternal and fetal serum (or plasma); however, fetal/maternal serum ratios vary depending on the structure of the perfluoroalkyl (Table 3-2). With some exceptions, longer fluoroalkyl chain length and a terminal sulfonate group are associated with lower fetal/maternal ratios (Glynn et al. 2012; Gützkow et al. 2012; Hanssen et al. 2013; Kim et al. 2011; Liu et al. 2011; Needham et al. 2011). PFOS was detected in amniotic fluid obtained from amniocentesis (Jensen et al. 2012). The median concentration in amniotic fluid samples from 300 pregnancies (from the Danish amniotic fluid pregnancy-screening biobank) was 1.1 ng/mL.

Table 3-2. Serum (or Plasma) Concentrations in Matched Human Maternal-Infant Pairs

Study	Perfluoro-alkyl	Perfluoroalkyl chain length	N	Maternal (ng/mL)	Cord (ng/mL)	Ratio[a]	r
Glynn et al. 2012	PFOA	7	413	4	1	NR	0.89
	PFOS	8	413	29	5	NR	0.86
	PFNA	8	413	0.6	0.1	NR	0.53
Cariou et al. 2015	PFHxS	6	59	0.62	0.34	0.56	0.99
	PFOA	7	89	1.05	0.86	0.78	0.83
	PFOS	8	94	3.07	1.11	0.38	0.88
	PFNA	8	22	0.43	0.27	0.51	0.92
Chen et al. 2017a	PFHxS	6	32	0.53	0.33	0.62	ND
	PFOA	7	32	8.67	3.67	0.42	ND
	PFOS	8	32	1.56	1.24	0.79	ND
Fisher et al. 2016	PFHxS	6	315	NR	NR	0.23	NR
	PFOA	7	865	NR	NR	0.28	NR
	PFOS	8	648	NR	NR	0.14	NR
Fromme et al. 2010	PFHxS	6	53	0.60	0.30	0.50	0.89
	PFOA	7	53	2.60	1.70	0.65	0.94
	PFOS	8	53	3.50	1.10	0.31	0.89
	PFNA	8	53	0.60	<0.4	ND	ND
Gützkow et al. 2012	PFHxS	6	123	0.34	0.23	0.68	0.70
	PFOA	7	123	1.25	1.03	0.82	0.82
	PFOS	8	123	5.37	1.78	0.33	0.74
	PFNA	8	123	0.40	0.16	0.40	0.64
	PFDA	9	123	0.10	0.04	ND	ND
	PFUnA	10	123	0.19	0.06	0.32	0.67
Hanssen et al. 2013	PFHxS	6	7	0.26	0.17	0.65	ND
	PFOA	7	7	1.50	1.26	0.84	ND
	PFOS	8	7	10.70	3.93	0.37	ND
	PFNA	8	7	0.89	0.50	0.56	ND
	FOSA	8	7	0.41	0.45	1.10	ND
	PFUnA	10	7	0.33	0.16	0.48	ND
Han et al. 2018	PFBS	4	369	0.19	0.19	1.00	ND
	PFHxS	6	369	0.32	0.31	1.03	ND
	PFHpA	6	369	0.06	0.09	1.50	ND
	PFOA	7	369	42.83	34.67	0.81	ND
	PFOS	8	369	4.55	1.39	0.31	ND
	FOSA	8	369	0.13	0.13	1.00	ND
	PFNA	8	369	0.81	0.44	0.54	ND
	PFDA	9	369	0.55	0.21	0.38	ND
	PFUnA	10	369	0.47	0.17	0.36	ND
	PFDoDA	11	369	0.17	0.14	0.82	ND

Table 3-2. Serum (or Plasma) Concentrations in Matched Human Maternal-Infant Pairs

Study	Perfluoro-alkyl	Perfluoroalkyl chain length	N	Maternal (ng/mL)	Cord (ng/mL)	Ratio[a]	r
Inoue et al. 2004	PFOA	7	15	8.90	2.90	0.32	0.94
Kim et al. 2011	PFHxS	6	20	0.89	0.58	0.65	ND
	PFOA	7	20	1.60	1.10	0.69	ND
	PFOS	8	20	5.60	2.00	0.36	ND
	PFNA	8	20	0.79	0.37	0.47	ND
	PFDA	9	20	0.36	0.01	0.03	ND
	PFUnA	10	20	1.60	0.46	0.29	ND
Kato et al. 2014	PFHxS	6	78	1.20	0.60	0.50	0.89
	PFOA	7	78	3.30	3.10	0.89	0.88
	PFOS	8	78	8.50	3.50	0.31	0.82
	PFNA	8	78	0.66	0.41	0.62	0.79
	PFDA	9	78	0.20	ND	ND	ND
Lee et al. 2013	PFHS	6	70	1.35	0.67	0.57	ND
	PFOA	7	70	2.73	2.09	0.84	ND
	PFOS	8	70	10.77	3.44	0.35	ND
Liu et al. 2011	PFHxS	6	50	0.08	0.06	0.79	0.59
	PFOA	7	50	1.66	1.50	0.91	0.91
	PFOS	8	50	3.18	1.69	0.53	0.75
	PFNA	8	50	0.55	0.33	0.61	0.82
	PFDA	9	50	0.58	0.24	0.41	0.82
	PFUnA	10	50	0.56	0.30	0.53	0.70
	PFDoDA	11	50	0.08	ND	ND	ND
Manzano-Salgado et al. 2015	PFHxS	6	66	0.84	0.40	0.446	NR
	PFOA	7	66	2.97	1.90	0.746	NR
	PFOS	8	66	6.99	1.86	0.299	NR
	PFNA	8	66	0.85	0.32	0.4	NR
Midasch et al. 2007	PFOA	7	11	2.70	3.40	1.30	0.42
	PFOS	8	11	12.10	7.20	0.60	0.72
Monroy et al. 2008	PFHxS	6	101	4.05	5.05	1.25	ND
	PFOA	7	101	2.24	1.94	0.87	0.94
	PFOS	8	101	16.19	7.19	0.44	0.91
	PFNA	8	101	0.80	0.94	1.18	ND
Needham et al. 2011	PFHxS	6	12	12.30	9.10	0.74	0.05
	PFOA	7	12	4.20	3.10	0.72	0.91
	PFOS	8	12	19.70	6.60	0.34	0.82
	PFNA	8	12	0.76	0.37	0.50	0.84
	PFDA	9	12	0.34	0.10	0.29	0.91

Table 3-2. Serum (or Plasma) Concentrations in Matched Human Maternal-Infant Pairs

Study	Perfluoro-alkyl	Perfluoroalkyl chain length	N	Maternal (ng/mL)	Cord (ng/mL)	Ratio[a]	r
Ode et al. 2013	PFOA	7	263	2.30	2.80	1.30	0.74
	PFOS	8	263	17.00	7.40	0.45	0.76
	PFNA	8	263	0.31	0.26	0.93	0.51
Porpora et al. 2013	PFOA	7	38	2.90	1.60	0.55	0.70
	PFOS	8	38	3.20	1.40	0.44	0.72
Yang et al. 2016a	PFHxS	6	50	0.064	0.033	0.52	0.80
	PFOA	7	50	1.24	1.03	0.83	0.93
	PFOS	8	50	2.98	1.23	0.41	0.88
	PFNA	8	50	0.55	0.35	0.64	0.89
	PFDA	9	50	0.56	0.22	0.39	0.92
	PFUnA	10	50	0.55	0.23	0.42	0.88
	PFDoDA	11	50	0.085	0.058	0.68	0.76
Yang et al. 2016b	PFHxS	6	157	0.53	0.26	0.43	0.68
	PFOA	7	157	1.74	1.32	0.71	0.81
	PFOS	8	157	4.23	1.52	0.36	0.63
	PFNA	8	157	0.46	0.23	0.49	0.70
	PFDA	9	157	0.37	0.13	0.35	0.65
	PFUnA	10	157	0.38	0.14	0.36	0.63
	PFDoDA	11	157	0.040	0.026	0.61	0.52

[a]Ratio of cord:maternal perfluoroalkyl level.

FOSA = perfluorooctane sulfonamide; ND = no data (detected but below limit of quantification); NR = not reported; PFBS = perfluorobutane sulfonic acid; PFDA = perfluorodecanoic acid; PFDoDA = perfluorododecanoic acid; PFHxS = perfluorohexane sulfonic acid; PFHpA = Perfluoroheptanoic acid; PFNA = perfluorononanoic acid; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; PFTrDA = perfluorotridecanoic acid; PFUnA = perfluoroundecanoic acid

Studies in rats and mice provide further support for maternal-fetal transfer of perfluoroalkyls. Following gavage administration of 0.1–10 mg/kg/day PFOS to rats during gestation, PFOS was distributed to fetal serum, liver, and brain, with fetal concentrations increasing with maternal dose (Chang et al. 2009; Lau et al. 2003; Luebker et al. 2005a, 2005b; Thibodeaux et al. 2003). Levels in fetal serum and liver generally were similar and higher than in brain. Studies did not report on concentrations of PFOS in other fetal tissues. Paired fetal-maternal levels of PFOS were examined in rats following exposure (gavage) to potassium PFOS at doses of 0.1, 0.4, 1.6, or 3.2 mg/kg/day on GDs 0–20 (Luebker et al. 2005b). On GD 21, fetal:maternal serum ratios were 2.1, 1.7, 1.6, and 1.1 at doses of 0.1, 0.4, 1.6, and 3.2 mg/kg/day, respectively; these results suggest that fetal:maternal serum ratios varied inversely with dose. Fetal:maternal liver ratios (0.37–0.44) were similar across the dose range. In mice administered a single

gavage dose of 12.5 mg/kg [^{35}S]PFOS on GD 16, fetal organ:maternal blood ratios of ^{35}S on GD 18 were 2.8 for kidneys, 2.6 for liver, 2.3 for blood, 2.1 for lungs, and 1.2 for brain (Borg et al. 2010).

Maternal-fetal transfer of PFOA has also been studied in rats and mice (Das et al. 2008; Hinderliter et al. 2005). In rats, PFOA concentrations in amniotic fluid, placenta, and fetus (measured on days 10, 15, or 21 of gestation) increased with increasing maternal oral dose (3, 10, or 30 mg/kg/day, administered daily beginning on GD 4) (Hinderliter et al. 2005). Fetal plasma concentrations of PFOA measured on GD 21 were approximately 40% of maternal plasma concentration. Following gavage administration of 0.01, 1, or 5 mg/kg ammonium PFOA on GD 17 in mice, PFOA was detected in amniotic fluid and pup serum, with dose-dependent increases (Fenton et al. 2009). On PND 1, pup serum PFOA concentrations were approximately 1.7–2.0-fold greater than levels in maternal serum.

Following administration of ammonium PFBA (35, 175, or 350 mg/kg) to pregnant mice on GDs 0–17, fetal serum and liver levels of PFBA were determined on PND 1 (Das et al. 2008). The fetal:maternal serum ratio of PFBA was approximately 0.15 and did not vary with maternal dose. Fetal liver:serum ratios were 0.44, 0.75, and 0.78 at maternal doses of 35, 175, and 350 mg/kg, respectively. PFHxS was detected in fetal blood and in the liver of neonates following exposure of dams to potassium PFHxS (0.3, 1, 3, and 10 mg/kg) throughout gestation (Butenhoff et al. 2009a); concentrations in serum and liver increased with dose.

Maternal-infant Transfer. Perfluoroalkyls can be transferred to nursing infants (Barbarossa et al. 2013; Cariou et al. 2015; Fromme et al. 2010; Kärrman et al. 2007; Kim et al. 2011; Kuklenyik et al. 2004; Liu et al. 2011; Tao et al. 2008a, 2008b). Studies that measured perfluoroalkyls in maternal serum (or plasma) and breast milk in matched mother-infant pairs found highly variable correlations (Table 3-3). Relatively high correlations have been reported for PFOA (Kärrman et al. 2007; Liu et al. 2011). Transfer to breast milk appears to be a significant route of elimination of perfluoroalkyls during breastfeeding. Comparisons of serum concentrations of women who did or did not breastfeed their infants showed that breastfeeding significantly decreases maternal serum concentrations of PFOA, PFOS, PFHxS, and PFNA (Bjermo et al. 2013; Brantsaeter et al. 2013; Mondal et al. 2012, 2014; von Ehrenstein et al. 2009). The decrease was estimated to be 2–3% decrease per month of breastfeeding (Brantsaeter et al. 2013; Mondal et al. 2012, 2014). Concentrations of perfluoroalkyls in breast milk also decrease with breastfeeding duration (Tao et al. 2008b; Thomsen et al. 2010). Numerous perfluoroalkyls (including PFOS, PFOA, PFBS, PFHxS, PFNA, PFDA, PFDoDA, PFUnA, and FOSA) have been detected in breast milk samples in women in China, Korea, Japan, Malaysia, Cambodia, India, Korea, Vietnam, Indonesia,

Norway, Philippines, Sweden, and the United States (Forns et al. 2015; Fujii et al. 2012; Kang et al. 2016; Kärrman et al. 2007; Kim et al. 2011; Liu et al. 2010, 2011; Mondal et al. 2014; So et al. 2006b; Tao et al. 2008a). The mean concentrations for perfluoroalkyls in breast milk collected from 45 women in Massachusetts were 0.131 ng/mL (range of <0.032.0–617 ng/mL) for PFOS, 0.043.8 ng/mL (<0.0301–0.161 ng/mL) for PFOA, and 0.0145 ng/mL (<0.0120–0.0638 ng/mL) for PFHxS (Tao et al. 2008b). PFHpA, PFDA, PFUnA, PFDoDA, and PFBS were also detected in the breast milk; however, ≤4 samples had levels that exceeded the limit of quantitation. Serum concentrations in breastfed infants can be higher than maternal levels. Although cord:maternal serum ratios of PFOA, PFOS, and PFNA at birth are typically lower than 1 (see Table 3-2), infant serum levels increase several-fold during the first 6 months after birth (Fromme et al. 2010; Mondal et al. 2014; Post et al. 2012; Verner 2016a, 2016b). This increase is likely because breast milk concentrations of perfluoroalkyls and fluid intake per infant body weight are highest during this time period. Fromme et al. (2010) also showed increases in serum levels of PFNA in infants fed formula made with contaminated drinking water. Mogensen et al. (2015b) reported that following weaning, significant (<0.0001) decreases were observed in infant serum concentrations of PFOS, PFOA, and PFHxS.

Table 3-3. Matched Serum (or Plasma) and Breast Milk Concentrations in Humans

Study	Perfluoroalkyl	Perfluoroalkyl chain length	N	Serum (ng/mL)	Milk (ng/mL)	Ratio[a]	r
Cariou et al. 2015	PFHxS	6	9	2.28	0.026	0.011	0.36
	PFOA	7	10	1.22	0.041	0.034	0.72
	PFOS	8	19	3.62	0.040	0.011	0.85
Kärrman et al. 2007a	PFHxS	6	12	4.7	0.085	0.020	ND
	PFOA	7	12	3.8	0.49	0.120	0.88
	PFOS	8	12	20.7	0.20	0.010	0.83
	FOSA	8	12	0.24	0.013	0.070	ND
	PFNA	8	12	0.80	0.017	0.010	ND
Kim et al. 2011	PFHxS	6	20	0.89	0.007	0.008	NS
	PFOA	7	20	1.60	0.041	0.026	NS
	PFOS	8	20	5.60	0.061	0.011	0.60
	PFNA	8	20	0.79	<0.0088	ND	ND
	PFDA	9	20	0.36	<0.018	ND	ND
	PFUnA	10	20	1.60	<0.024	ND	ND

Table 3-3. Matched Serum (or Plasma) and Breast Milk Concentrations in Humans

Study	Perfluoroalkyl	Perfluoroalkyl chain length	N	Serum (ng/mL)	Milk (ng/mL)	Ratio[a]	r
Liu et al. 2011	PFHxS	6	50	0.08	ND	ND	ND
	PFOA	7	50	1.66	0.181	0.109	0.77
	PFOS	8	50	3.18	0.056	0.018	0.57
	PFNA	8	50	0.55	0.026	0.048	0.62
	PFDA	9	50	0.58	0.02	0.034	0.54
	PFUnA	10	50	0.56	0.026	0.046	0.44
	PFDoDA	11	50	0.08	ND	ND	ND
	PFTrDA	12	50	0.08	ND	ND	ND

[a]Milk to serum ratio.

FOSA = perfluorooctane sulfonamide; ND = no data (detected but below limit of quantification); NS = not significantly correlated; PFDA = perfluorodecanoic acid; PFDoDA = perfluorododecanoic acid; PFHxS = perfluorohexane sulfonic acid; PFNA = perfluorononanoic acid; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; PFTrDA = perfluorotridecanoic acid; PFUnA = perfluoroundecanoic acid

Studies conducted in rats and mice provide further support for maternal-infant transfer of perfluoroalkyls through breast milk (Fenton et al. 2009; Hinderliter et al. 2005; Lau et al. 2003; Luebker et al. 2005a; Yu et al. 2009b). PFOA concentrations in breast milk of nursing rats increased with increasing maternal oral dose (3, 10, or 30 mg/kg/day, administered daily beginning on GD 4) (Hinderliter et al. 2005). Milk concentrations of PFOA measured on postpartum days 3, 7, 14, or 21 in rats were approximately 0.1 of maternal plasma concentration. In dams exposed to 0.1, 1, or 5 mg/kg PFOA by gavage on GD 17, a dose-dependent increase in PFOA concentrations in breast milk was observed on PND 2, with breast milk:serum ratios of approximately 0.15, 0.38, and 0.25 at 0.1, 1, and 5 mg/kg doses, respectively; milk/serum concentration ratios for PFOA ranged from 0.15 to 0.56 (Fenton et al. 2009). Following lactational exposure of control rat pups to PFOS in breast milk of dams treated with dietary PFOS (3.2 mg/kg diet; approximately equivalent to 0.33 mg/kg/day), pup serum and liver concentrations increased throughout the 35-day lactation period (Yu et al. 2009b). At PND 35, the pup liver:serum PFOS ratios were 2.55 and 2.43 in male and female pups, respectively. Results of a cross-foster study show that pups are exposed to PFOS through breast milk (Luebker et al. 2005a). Postnatal toxicity observed in cross-fostered pups that nursed from exposed dams provides additional evidence of maternal-infant transfer of PFOS in rats and mice (see Section 2.17).

Mechanisms of Distribution. Perfluoroalkyls in plasma bind to serum albumin and various other plasma proteins including gamma-globulin, alpha-globulin, alpha-2-macroglobulin, transferrin, and beta-lipoproteins (Bischel et al. 2011; Butenhoff et al. 2012d; Chen and Guo 2009; Han et al. 2003, 2005; Kerstner-Wood et al. 2003; Luo et al. 2012; Ohmori et al. 2003; Salvalaglio et al. 2010; Vanden Heuvel et al. 1992b; Wu et al. 2009; Ylinen and Auriola 1990; Zhang et al. 2009). The dissociation constant for albumin-bound PFOA in serum is approximately 0.4 mM (0.38 mM, ±0.04 SD for human serum albumin; 0.36 nM, ±0.08 SD for rat serum albumin) and involves 6–9 biding sites (Han et al. 2003). Noncovalent binding appears to be at the same sites as fatty acids (Chen and Guo 2009). Interactions between PFOS and human serum albumin include interaction of PFOS polar sulfonyl groups with albumin hydrophilic sites and interaction of perfluorinated groups with albumin hydrophobic sites (Luo et al. 2012).

Absorbed perfluoroalkyls distribute from plasma to soft tissues, with the highest extravascular concentrations achieved in liver. Mechanisms by which perfluoroalkyls enter the liver have not been elucidated and may involve interactions with organic anion transporters that function in the distribution of fatty acids or other organic anions (Andersen et al. 2008). PFOA appears to be a substrate for organic anion transporters in the luminal and basolateral membranes of renal tubular epithelial cells, which facilitates entry of PFOA into renal tubular cells (Kudo et al. 2002; Nakagawa et al. 2008; Vanden Heuvel et al. 1992b; Weaver et al. 2010). The subcellular distribution of PFOA is sex- and dose-dependent in rats (Han et al. 2005; Kudo et al. 2007) and the association with the membrane fraction of liver cells decreases with increasing dose (Kudo et al. 2007), consistent with limited capacity of membrane proteins that bind PFOA (e.g., membrane transport proteins). Intracellular PFOA binds to proteins; protein complexes formed have not been fully characterized. PFOA exhibits a low affinity for binding to rat kidney and urine alpha-2μ-globulin (dissociation constants 1.5 and >2 mM, respectively) (Han et al. 2004).

3.1.3 Metabolism

Results of available intraperitoneal and *in vitro* studies suggest that the perfluoroalkyls discussed in this profile are not metabolized and do not undergo chemical reactions in the body. The absence of significant metabolism is attributed to the high stability and low reactivity of carbon-fluorine bonds in perfluoroalkyls. Studies conducted in male and female rats did not detect fluorine metabolites in the urine, plasma, or liver following a single injection of 4–150 mg/kg PFOA or 5–50 mg/kg PFDA (Goecke et al. 1992; Vanden Heuvel et al. 1991b, 1991c; Ylinen and Auriola 1990). Following a single intraperitoneal dose of approximately 4 mg/kg of ^{14}C-PFOA, only parent compound was excreted in the

urine and bile (Vanden Heuvel et al. 1991c). PFOA was not metabolized when incubated with microsomal fractions of human or rat intestine, kidney, or liver homogenates (Kemper and Nabb 2005). Although no studies examining metabolism of other perfluoroalkyls, including PFOS, following inhalation, oral, or dermal exposure were identified, metabolism by these exposure routes is not anticipated.

3.1.4 Excretion

As noted in Section 3.1.3 (Metabolism), there is presently no evidence that perfluoroalkyls undergo metabolism. The absence of significant metabolism is attributed to the high stability and low reactivity of carbon-fluorine bonds in perfluoroalkyls. Therefore, route-specific differences in excretion patterns are not expected. Selected studies in which elimination half-lives rates (i.e., $t_{1/2}$) of perfluoroalkyls have been determined (see summaries in Table 3-5) show that, in general, elimination $t_{1/2}$ values are similar following intravenous, intraperitoneal, and oral exposures. Findings suggest that the route of absorption has no substantial effect of rates of elimination of absorbed perfluoroalkyls (Butenhoff et al. 2004c; Chang et al. 2008a; Kemper 2003; Kudo et al. 2002; Ohmori et al. 2003; Vanden Heuvel et al. 1991b; Ylinen et al. 1990). As discussed in this section, perfluoroalkyls are primarily eliminated in the urine, with smaller amounts eliminated in the feces, breast milk (see Section 3.1.2; Distribution, Maternal-fetal Transfer), and menstrual fluid. Perfluoroalkyls undergo biliary excretion, but substantial reabsorption occurs; therefore, biliary excretion does not represent a major elimination pathway. Perfluoroalkyls do not appear to be eliminated in sweat, as induction of perspiration by exercise or sauna does not alter clearance of PFOA, PFOA, PFHxA, or PFNA (Genuis et al. 2013). The elimination of perfluoroalkyls in menstrual fluid appears to contribute to sex differences in serum elimination rates (Wong et al. 2014, 2015; Zhang et al. 2013). Only free (unbound) perfluoroalkyls are available for redistribution, excretion, and renal reabsorption; the interaction of perfluoroalkyls with proteins plays a critical role in bioaccumulation, and the tissue environment highly favors protein bonding.

In humans, absorbed perfluoroalkyls are excreted in urine. Estimates of renal clearance of PFOA and PFOS from serum in humans ranged from 0.8 to 3.3 mL/day for PFOA (serum concentration range: 5–16 ng/mL) and 0.1–1.5 mL/day for PFOS (serum concentration range 9–49 ng/mL). These clearance values were <0.001% of glomerular filtration rate (Harada et al. 2005a). Assuming that 99% of the serum PFOA and PFOS was bound to albumin (see Section 3.1.2), <0.1% of filtered perfluoroalkyls were excreted in urine, suggesting extensive reabsorption of filtered PFOA and PFOS in the renal tubule. Renal clearance was not different in males and females. Mean renal clearances for PFOA were

2.12 mL/day (±0.80 SD, n=5) in males and 1.15 (±0.33 SD, n=5) in five females (mean age 22 and 23 years, respectively). Mean renal clearances for PFOS were 0.66 mL/day (±0.48 SD, n=5) in males and 0.91 (±0.56 SD, n=5) for females. Fujii et al. (2015a) reported renal clearances (mL/day/kg; mean±SD) for several perfluoroalkyls in humans (three males and five females), including PFOA (0.044±0.01), PFNA (0.038±0.01), PFDA (0.015±0.01), PFUnA (0.005±0.00), and PFDoDA (0.005±0.00). Zhang et al. (2013) reported renal clearances for several perfluoroalkyls (mL/day/kg) and found that clearance of PFOS was similar in younger females (≤50 years, 0.050 mL/day/kg, 95% CI 0.037–0.064) and a combined group of males and older females (grouped together since there were no significant differences in serum concentrations) (0.037 mL/day/kg, 95% CI 0.026–0.049). However, there appeared to be differences in renal clearance for PFOA; clearance rates were 0.30 mL/day/kg (95% CI 0.11–0.49) in young females and 0.77 mL/day/kg (95% CI 0.47–1.1) in the combined older women and all males group. Urinary excretion of perfluoroalkyls may show sex and age differences (Zhang et al. 2015b). Urinary excretion of PFOA as a fraction of estimated intake in male adults (n=29) was 31% (p=0.002) higher than in nonpregnant female adults (n=25). In addition, urinary excretion of PFOS was inversely correlated with age (r=0.334; p=0.015).

Absorbed PFOA and PFOS are also secreted into bile in humans, but the biliary pathway is not a major excretory pathway because PFOA and PFOS are reabsorbed after biliary secretion. Estimates of total body clearance, serum-to-urine clearance, and serum-to-bile clearance of PFOA and PFOS in humans are presented in Table 3-4 (Harada et al. 2007). Biliary clearances of PFOA and PFOS were 1.06 and 2.98 mL/kg body weight/day, respectively, and greatly exceeded total body clearance (0.150 and 0.106 mL/kg/day) and urinary clearance (0.030 and 0.015 mL/kg/day). Based on these estimates, approximately 89% of the PFOA secreted into bile and 97% of secreted PFOS was estimated to have been reabsorbed from the gastrointestinal tract. Fujii et al. (2015a) also reported that biliary clearances of several perfluoroalkyls (PFOA, PFNA, PFDA, PFUnA, PFDoDA) were much higher than total body clearance in humans, further supporting that perfluoroalkyls excreted in bile undergo extensive reabsorption.

Table 3-4. Excretory Clearance of PFOA and PFOS in Humans

Parameter	Units	PFOA	PFOS
Serum $t_{1/2}$[a]	day	1,387	1,971
Total clearance[b]	mL/kg/day	0.150	0.106
Urinary clearance[c]	mL/kg/day	0.030	0.150

Table 3-4. Excretory Clearance of PFOA and PFOS in Humans

Parameter	Units	PFOA	PFOS
Biliary clearance[d]	mL/kg/day	1.06	2.98
Reabsorbed from bile[e]	%	89	97

[a]Estimates from Olsen et al. (2005).
[b]$\ln(t_{1/2})$xVd, where Vd is the volume of distribution (300 mL/kg).
[c]Estimates from Harada et al. (2005a).
[d]Estimates from Harada et al. (2007).
[e]1-(Total-Urinary)/Biliary.

PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid

Source: Harada et al. (2007)

Studies conducted in nonhuman primates and rodents provide further evidence that urine is the major route of excretion of perfluoroalkyls, accounting for >93% of absorbed PFOA and PFOS (Benskin et al. 2009; Butenhoff et al. 2004c; Chang et al. 2008a, 2012; Chengelis et al. 2009a; Hanhijarvi et al. 1982, 1987; Hundley et al. 2006; Johnson and Ober 1979, 1980, 1999a, 1999b; Kemper 2003; Kudo et al. 2001; Olsen et al. 2009; Sundström et al. 2012; Vanden Heuvel et al. 1991b, 1991c). Studies conducted in rats have shown that PFDA, PFNA, PFOA, and PFHxA are secreted in bile and undergo extensive reabsorption from the gastrointestinal tract (Kudo et al. 2001; Vanden Heuvel et al. 1991b, 1991c). PFOS, PFHxS, and PFBS are excreted in feces following intravenous dosing of rats, suggesting that these perfluoroalkyls may also be secreted into bile (Chang et al. 2012; Johnson et al. 1984; Olsen et al. 2009; Sundström et al. 2012). The percentage of the dose excreted in the feces appears to vary with compound, 8–13% for PFOS, <0.5% for PFHxS, and 0.13–0.36% for PFBS. Renal clearances of PFOA from plasma in rats were approximately 0.032 mL/minute/kg body weight in male rats and 0.73 mL/minute/kg in female rats; plasma concentrations of PFOA during these measurements ranged from approximately 0.8 to 80 µg/mL (Kudo et al. 2002). In the latter study, approximately >95% of plasma PFOA was bound to high molecular weight protein and the glomerular filtration rate was approximately 10 mL/minute/kg; therefore, urinary excretion of PFOA was approximately 6% of the rate of glomerular filtration of PFOA in males and 146% in females. These estimates indicate that net renal tubular reabsorption of filtered PFOA occurred in male rats, whereas net renal tubular secretion of PFOA occurred in female rats (i.e., clearance of free PFOA in plasma > glomerular filtration rate). The pronounced sex difference in renal clearance of PFOA has been attributed to modulation of renal excretory transport of PFOA by testosterone and estradiol (Kudo et al. 2002; Vanden Heuvel et al. 1992a; see Section 3.1.5).

Rates of elimination of perfluoroalkyls vary substantially across chemical species and animal species, and show sex differences and age-dependencies within certain species. Table 3-5 summarizes estimates of the elimination $t_{1/2}$ for perfluoroalkyls in humans and experimental animals. In compiling the estimates presented in Table 3-5, preference was given to the terminal $t_{1/2}$ when multiple $t_{1/2}$ values were reported. The significance of the terminal $t_{1/2}$ is that it determines the time required for complete elimination of the perfluoroalkyl as well as the exposure duration required to achieve a steady state. Most of the $t_{1/2}$ values in Table 3-5 were estimated from analyses of data on declining serum concentrations of perfluoroalkyls after a single dose or following cessation of a period of repeated dosing. Estimates of the terminal $t_{1/2}$ based on serum concentrations can vary with the length of the observation period following the last dose and with the modeling approach used to estimate the $t_{1/2}$. Longer observation times are required to estimate the slowest phases of elimination. As a result, estimates of $t_{1/2}$ based on observation periods of 1–2 days can be much shorter than estimates for the same perfluoroalkyl based on observation periods of several weeks. Direct comparisons of $t_{1/2}$ values should be made with consideration of whether or not the observation periods were comparable. Differences in estimation methodology can also contribute to differences in $t_{1/2}$ values. Values reported in Table 3-5 are based on fitting data to single or multi-compartment models, or noncompartmental modeling of the data. While the terminal $t_{1/2}$ provides a metric for comparing times required for complete elimination and steady state, it does not always provide a measure of how rapidly the perfluoroalkyl is cleared from the body. A more useful metric for this is the systemic clearance (Cl_S), typically estimated from the absorbed dose (AD) and the area under the serum concentration curve (AUC_S):

$$Cl_S = \frac{AD}{AUC_S} \qquad \text{Eq. (3-3)}$$

Equation 3-3 will provide an accurate estimate of systemic clearance following an oral dose if the oral dose is completely absorbed. Accurate estimation of AUC_S also depends on fitness of the underlying model used to predict serum concentrations. Estimates of systemic clearance based on pharmacokinetics analyses of serum data from animal studies are presented in Table 3-6.

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
PFOA—Human					
Human (n=26), adult, M (n=24) F (n=2)	NA	NA	NA	3.8 years (95% CI 3.1–4.4, GM 3.5)	Olsen et al. 2007a
Human (n=20) 15–50 years, M	NA	NA	NA	2.8 years (95% CI 2.4–3.4)	Li et al. 2018
Human (n=30) 15–50 years, F	NA	NA	NA	2.4 years (95% CI 2.0–3.0)	Li et al. 2018
Human (n=66), >50 years, M, F	NA	NA	NA	2.6 years (SE 0.4, GM 1.2)	Zhang et al. 2013
Human (n=20), ≤50 years, F	NA	NA	NA	2.1 years (SE 0.3, GM 1.5)	Zhang et al. 2013
Human (n=45), M, F	NA	NA	NA	3.9 years	Worley et al. 2017a
Human (n=5), 22±0.9, M	NA	NA	NA	2.3 years	Harada et al. 2005a
Human (n=5), 68±5, M	NA	NA	NA	2.6 years	Harada et al. 2005a
Human (n=5), 23±3, F	NA	NA	NA	3.5 years	Harada et al. 2005a
Human (n=5), 69±5, F	NA	NA	NA	2.9 years	Harada et al. 2005a
Human (n=200) 54±15, M, F	Oral	NA	NA	2.3 years (95% CI 2.1–2.4)	Bartell et al. 2010
Human (n=643), adult, M, F	Oral	NA	NA	2.9 years (<4 years) (95% CI 2.3–3.8) 10.1 years (>4 years)	Seals et al. 2011
Human (n=1,029), adult, M, F	Oral	NA	NA	8.5 years (<9 years) (95% CI 7.1–10.1)	Seals et al. 2011
Humans (n=17), adult, M, F	Oral	NA	NA	5.1 years (SD 1.7, GM 4.8)	Costa et al. 2009
Humans (n=6) adults, F	Inhalation	NA	NA	2.5 (range 1.8–3.1)	Gomis et a. 2016
PFOS—Human					
Human (n=26), adult, M (24) F (2)	NA	NA	NA	5.4 years (95% CI 3.9–6.9, GM 4.8)	Olsen et al. 2007a
Human (n≅1,000), >12–>80 years, M	NA	NA	NA	4.7 years	Wong et al. 2014

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
Human (n≅1,000), >12–>80 years, F	NA	NA	NA	4.3 years (95% CI 4.1–4.5)	Wong et al. 2015
Human (n=66), >50 years, M, F	NA	NA	NA	27 years (SE 3.1, GM 18)	Zhang et al. 2013
Human (n=20), ≤50 years, F	NA	NA	NA	6.2 years (SE 0.5, GM 5.8)	Zhang et al. 2013
Human (n=45), M, F	NA	NA	NA	3.3 years	Worley et al. 2017a
Human (n=20) 15–50 years M	NA	NA	NA	4.6 years (95% CI 3.7–6.1)	Li et al. 2018
Human (n=30) 15–50 years, F	NA	NA	NA	3.1 years (95% CI 2.7–3.7)	Li et al. 2018
Human (n=5), 22±0.9, M	NA	NA	NA	4.9 years	Harada et al. 2005a
Human (n=5), 68±5, M	NA	NA	NA	7.4 years	Harada et al. 2005a
Human (n=5), 23±3, F	NA	NA	NA	4.5 years	Harada et al. 2005a
Human (n=5), 69±5, F	NA	NA	NA	4.6 years	Harada et al. 2005a
PFHxS—Human					
Human (n=26), adult, M (24), F (2)	NA	NA	NA	8.5 years (95% CI 6.4–10.6, GM 7.3)	Olsen et al. 2007a
Human (n=20), ≤50 years, F	NA	NA	NA	7.7 years (SE 0.6, GM 7.1)	Zhang et al. 2013
Human (n=20) 15–50 years, M	NA	NA	NA	7.4 years (95% CI 6.0–9.7)	Li et al. 2018
Human (n=30) 15–50 years F	NA	NA	NA	4.7 years (95% CI 3.9–5.9)	Li et al. 2018
Human (n=45), M, F	NA	NA	NA	15.5 years	Worley et al. 2017a
Human (n=66), >50 years, M, F	NA	NA	NA	35 years (SE 3.9, GM 25)	Zhang et al. 2013
PFBA—Human					
Human (n=3), adult, M	NA	NA	NA	81 hours (SD 41)	Chang et al. 2008b
Human (n=9), adult, M (7), F (2)	NA	NA	NA	72 hours (SD 38)	Chang et al. 2008b
PFBS—Human					
Human (n=6), adult M (5), F(1)				665 hours (SD 266)	Olsen et al. 2009

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
PFNA—Human					
Human (n=66), >50 years, M, F	NA	NA	NA	4.3 years (SE 0.5, GM 3.2)	Zhang et al. 2013
Human (n=20), ≤50 years, F	NA	NA	NA	2.5 years (SE 0.6, GM 1.7)	Zhang et al. 2013
PFDA—Human					
Human (n=66), >50 years, M, F	NA	NA	NA	12 years (SE 1.5, GM 7.1)	Zhang et al. 2013
Human (n=20), ≤50 years, F	NA	NA	NA	4.5 years (SE 0.4, GM 4.0)	Zhang et al. 2013
PFUnA—Human					
Human (n=66), >50 years, M, F	NA	NA	NA	12 years (SE 2.0, GM 7.4)	Zhang et al. 2013
Human (n=20), ≤50 years, F	NA	NA	NA	4.5 years (SE 0.5, GM 4.0)	Zhang et al. 2013
PFHpA—Human					
Human (n=66), >50 years, M, F	NA	NA	NA	1.2 years (SE 0.2, GM 0.82)	Zhang et al. 2013
Human (n=20), ≤50 years, F	NA	NA	NA	1.5 years (SE 0.3, GM 1.0)	Zhang et al. 2013
PFOA—Nonhuman primate					
Cynomolgus monkey, adult, M	Oral	10 mg/kg/day	6 months	20.1 days	Butenhoff et al. 2004c
Cynomolgus monkey, adult, M	IV	10 mg/kg	1 day	20.9 days (SD 12.5)	Butenhoff et al. 2004c
Cynomolgus monkey, adult, F	IV	10 mg/kg	1 day	32.6 days (SD 8.0)	Butenhoff et al. 2004c
PFOS—Nonhuman primate					
Cynomolgus monkey, adult, M	Oral	0.15 mg/kg/day	6 months	170 days	Seacat et al. 2002
Cynomolgus monkey, adult, M	Oral	0.75 mg/kg/day	6 months	170 days	Seacat et al. 2002
Cynomolgus monkey, adult, F	Oral	0.15 mg/kg/day	6 months	170 days	Seacat et al. 2002
Cynomolgus monkey, adult, F	Oral	0.75 mg/kg/day	6 months	170 days	Seacat et al. 2002
Cynomolgus monkey, adult, M	IV	2 mg/kg	1 day	132 days (SE 7)	Chang et al. 2012
Cynomolgus monkey, adult, F	IV	2 mg/kg	1 day	110 days (SE 15)	Chang et al. 2012

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
PFHxA—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10 mg/kg	1 day	5.3 days (SD 2.5)	Chengelis et al. 2009a
Cynomolgus monkey, adult, F	IV	10 mg/kg	1 day	2.4 days (SD 1.7)	Chengelis et al. 2009a
PFHxS—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10 mg/kg	1 day	141 days (SE 30.)	Sundström et al. 2012
Cynomolgus monkey, adult, F	IV	10 mg/kg	1 day	87 days (SE 27)	Sundström et al. 2012
PFBA—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10 mg/kg	1 day	40.3 hours (SD 2.4)	Chang et al. 2008b
Cynomolgus monkey, adult, F	IV	10 mg/kg	1 day	41.0 hours (SD 4.7)	Chang et al. 2008b
PFBS—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10 mg/kg	1 day	15.0 hours (SD 9.8)	Chengelis et al. 2009a
Cynomolgus monkey, adult, F	IV	10 mg/kg	1 day	8.0 hours (SD 2.0)	Chengelis et al. 2009a
Cynomolgus monkey, adult, M	IV	10 mg/kg	1 day	95.2 hours (SE 27.1)	Olsen et al. 2009
Cynomolgus monkey, adult, F	IV	10 mg/kg	1 day	83.2 hours (SE 41.9)	Olsen et al. 2009
PFOA—Rat					
Rat (CR), adult, M	Oral	11.4 mg/kg	1 day	115 hours	Johnson and Ober 1980
Rat (Sprague-Dawley), adult, M	Oral	0.1 mg/kg	1 day	202 hours (SD 38)	Kemper 2003
Rat (Sprague-Dawley), adult, M	Oral	1 mg/kg	1 day	138 hours (SD 32)	Kemper 2003
Rat (Sprague-Dawley), adult, M	Oral	1 mg/kg	1 day	44 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	Oral	5 mg/kg	1 day	174 hours (SD 29)	Kemper 2003
Rat (Sprague-Dawley), adult, M	Oral	25 mg/kg	1 day	157 hours (SD 38)	Kemper 2003
Rat (Sprague-Dawley), adult, M	IV	1 mg/kg	1 day	185 hours (SD 19)	Kemper 2003
Rat (Sprague-Dawley), adult, M	IV	1 mg/kg	1 day	39 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	Oral	0.4 mg/kg	1 day	322 hours (SD 38)	Benskin et al. 2009

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
Rat (Sprague-Dawley), adult, M	Oral	0.022 mg/kg/day	12 weeks	218 hours (95% CL 127–792)	De Silva et al. 2009
Rat (Wistar), adult, M	IV	21.5 mg/kg	1 day	136 hours (SD 24)	Kudo et al. 2002
Rat (Wistar), adult, M	IV	20.1 mg/kg	1 day	135 hours (SD 29)	Ohmori et al. 2003
Rat (Sprague-Dawley), adult, M	IP	3.9 mg/kg	1 day	216 hours (SE 30.9)	Vanden Heuvel et al. 1991c
Rat (Wistar), adult, M	IP	50 mg/kg	1 day	105 hours	Ylinen et al. 1990
Rat (Sprague-Dawley), adult, F	Oral	0.1 mg/kg	1 day	3.2 hours (SD 0.9)	Kemper 2003
Rat (Sprague-Dawley), adult, F	Oral	1 mg/kg	1 day	3.5 hours (SD 1.1)	Kemper 2003
Rat (Sprague-Dawley), adult, F	Oral	1 mg/kg	1 day	3.6 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	5 mg/kg	1 day	4.6 hours (SD 0.6)	Kemper 2003
Rat (Sprague-Dawley), adult, F	Oral	25 mg/kg	1 day	16.2 hours (SD 9.9)	Kemper 2003
Rat (Sprague-Dawley), adult, F	IV	1 mg/kg	1 day	2.8 hours (SD 0.5)	Kemper 2003
Rat (Sprague-Dawley), adult, F	IV	1 mg/kg	1 day	4.6 hours	Kim et al. 2016b
Rat (Wistar), adult, F	IV	21.5 mg/kg	1 day	1.9 hours (SD 0.7)	Kudo et al. 2002
Rat (Wistar), adult, F	IV	20.1 mg/kg	1 day	1.9 hours (SD 0.7)	Ohmori et al. 2003
Rat (Sprague-Dawley), adult, F	IP	3.9 mg/kg	1 day	2.9 hours (SE 0.2)	Vanden Heuvel et al. 1991c
Rat (Wistar), adult, F	IP	50 mg/kg	1 day	24 hours	Ylinen et al. 1990
PFOS—Rat					
Rat (Sprague-Dawley), adult, M	Oral	4.2 mg/kg	1 day	179 hours	Johnson and Ober 1979
Rat (Sprague-Dawley), adult, M	Oral	0.27 mg/kg	1 day	809 hours	Benskin et al. 2009
Rat (Sprague-Dawley), adult, M	Oral	0.023 mg/kg/day	12 weeks	1,968 hours (95% CL 1.584–2.568)	De Silva et al. 2009
Rat (Sprague-Dawley), adult, M	Oral	2 mg/kg	1 day	1,495 hours (SE 50)	Chang et al. 2012
Rat (Sprague-Dawley), adult, M	Oral	2 mg/kg	1 day	635 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	Oral	15 mg/kg	1 day	1,707 hours (SE 270)	Chang et al. 2012

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
Rat (Sprague-Dawley), adult, F	Oral	0.023 mg/kg/day	12 weeks	1,992 hours (95% CL 1,752–2,280)	De Silva et al. 2009
Rat (Sprague-Dawley), adult, F	Oral	2 mg/kg	1 day	919 hours (SE 56)	Chang et al. 2012
Rat (Sprague-Dawley), adult, F	Oral	2 mg/kg	1 day	564 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	15 mg/kg	1 day	989 hours (SE 48)	Chang et al. 2012
Rat (Sprague-Dawley), adult, M	IV	2 mg/kg	1 day	689 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	IV	2 mg/kg	1 day	595 hours	Kim et al. 2016b
FOSA—Rat					
Rat (Sprague-Dawley), adult, M	Oral	5.0 mg/kg	1 day	125 hours	Seacat and Luebker 2000
PFDA—Rat					
Rat (Sprague-Dawley), adult, M	IP	4.8 mg/kg	1 day	1,008 hours	Vanden Heuvel et al. 1991b
Rat (Wistar), adult, M	IV	25 mg/kg	1 day	958 hours (SD 207)	Ohmori et al. 2003
Rat (Wistar), adult, F	IV	25 mg/kg	1 day	1,406 hours (SD 140)	Ohmori et al. 2003
Rat (Sprague-Dawley), adult, F	IP	4.8 mg/kg	1 day	552 hours	Vanden Heuvel et al. 1991b
PFNA—Rat					
Rat (Sprague-Dawley), adult, M	Oral	0.2 mg/kg	1 day	974 hours	Benskin et al. 2009
Rat (Sprague-Dawley), adult, M	Oral	0.029 mg/kg/day	12 weeks	1,128 hours (95% CL 935–1,416)	De Silva et al. 2009
Rat (Sprague-Dawley), adult M	Oral	1, 3, or 10 mg/kg	1 day	734.4 hours	Tatum-Gibbs et al. 2011
Rat (Wistar), adult, M	IV	22.6 mg/kg	1 day	710 hours (SD 55)	Ohmori et al. 2003
Rat (Sprague-Dawley), adult, F	Oral	1, 3, or 10 mg/kg	1 day	33.6 hours	Tatum-Gibbs et al. 2011
Rat (Wistar), adult, F	IV	22.6 mg/kg	1 day	58.6 hours (SD 9.8)	Ohmori et al. 2003
PFHpA—Rat					
Rat (Wistar), adult, M	IV	17.7 mg/kg	1 day	2.4 hours (SD 1.2)	Ohmori et al. 2003
Rat (Wistar), adult, F	IV	17.7 mg/kg	1 day	1.2 hours (SD 0.2)	Ohmori et al. 2003

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
PFHxA—Rat					
Rat (Sprague-Dawley), adult, M	IV	10 mg/kg	1 day	1.0 hour	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, M	Oral	50 mg/kg	1 day	2.2 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, M	Oral	150 mg/kg	1 day	2.4 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, M	Oral	300 mg/kg	1 day	2.5 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	IV	10 mg/kg	1 day	0.42 hour	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	Oral	50 mg/kg	1 day	2.6 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	Oral	150 mg/kg	1 day	2.2 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	Oral	300 mg/kg	1 day	2.1 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, M	Oral	2 mg/kg	1 day	1.7 hours (SD 0.6)	Gannon et al. 2011
Rat (Sprague-Dawley), adult, M	Oral	10 mg/kg	1 day	0.5 hours (SD 0.1)	Gannon et al. 2011
Rat (Sprague-Dawley), adult, F	Oral	2 mg/kg	1 day	1.5 hours (SD 0.2)	Gannon et al. 2011
Rat (Sprague-Dawley), adult, F	Oral	10 mg/kg	1 day	0.7 hours (SD 0.3)	Gannon et al. 2011
Rat (Sprague-Dawley), adult, M	Oral	50 mg/kg/day	26 days	2.0 hours	Kirkpatrick 2005
Rat (Sprague-Dawley), adult, M	Oral	150 mg/kg/day	26 days	2.1 hours	Kirkpatrick 2005
Rat (Sprague-Dawley), adult, M	Oral	300 mg/kg/day	26 days	2.9 hours	Kirkpatrick 2005
Rat (Sprague-Dawley), adult, F	Oral	50 mg/kg/day	26 days	1.9 hours	Kirkpatrick 2005
Rat (Sprague-Dawley), adult, F	Oral	150 mg/kg/day	26 days	2.2 hours	Kirkpatrick 2005
Rat (Sprague-Dawley), adult, F	Oral	300 mg/kg/day	26 days	3.0 hours	Kirkpatrick 2005
PFHxS—Rat					
Rat (Sprague-Dawley), adult, M	Oral	0.030 mg/kg	1 day	382 hours	Benskin et al. 2009
Rat (Sprague-Dawley), adult, M	Oral	4 mg/kg	1 day	645.6 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	4 mg/kg	1 day	41.28 hours	Kim et al. 2016b

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
Rat (Sprague-Dawley), adult, M	IV	4 mg/kg	1 day	496.8 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	IV	10 mg/kg	1 day	688 hours (SE 14.4)	Sundström et al. 2012
Rat (Sprague-Dawley), adult, F	IV	4 mg/kg	1 day	21.12 hours	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	IV	10 mg/kg	1 day	39 hours (SE 1.9)	Sundström et al. 2012
PFBA—Rat					
Rat (Sprague-Dawley), adult, M	Oral	30 mg/kg	1 day	9.22 hours (SE 0.75)	Chang et al. 2008b
Rat (Sprague-Dawley), adult, M	IV	30 mg/kg	1 day	6.38 hours (SE 0.53)	Chang et al. 2008b
Rat (Sprague-Dawley), adult, F	Oral	30 mg/kg	1 day	1.76 hours (SE 0.26)	Chang et al. 2008b
Rat (Sprague-Dawley), adult, F	IV	30 mg/kg	1 day	1.03 hours (SE 0.03)	Chang et al. 2008b
PFBS—Rat					
Rat (Sprague-Dawley), adult, M	IV	10 mg/kg	1 day	2.1 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), Rat (SD), adult, M	IV	30 mg/kg	1 day	4.51 hours (SE 2,22)	Olsen et al. 2009
Rat (Sprague-Dawley), adult, M	Oral	30 mg/kg	1 day	4.68 hours (SE 0.07)	Olsen et al. 2009
Rat (Sprague-Dawley), adult, F	IV	10 mg/kg	1 day	0.64 hours	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	IV	30 mg/kg	1 day	3.96 hours (SE 0.21)	Olsen et al. 2009
Rat (Sprague-Dawley), adult, F	Oral	30 mg/kg	1 day	7.42 hours (SE 0.79)	Olsen et al. 2009
PFOS—Mouse					
Mouse (CD), adult, M	Oral	1 mg/kg	1 day	1,027 hours	Chang et al. 2012
Mouse (CD), adult, M	Oral	20 mg/kg	1 day	874 hours	Chang et al. 2012
Mouse (CD), adult, F	Oral	1 mg/kg	1 day	907 hours	Chang et al. 2012
Mouse (CD), adult, F	Oral	20 mg/kg	1 day	731 hours	Chang et al. 2012

Table 3-5. Summary Elimination Half-Lives for Perfluoroalkyls Estimated in Humans and Experimental Animals

Species, age, and sex	Route	Dose	Exposure duration[a]	Elimination half-life[b]	Reference
PFHxS—Mouse					
Mouse (CD), adult, M	Oral	1 mg/kg	1 day	732 hours	Sundström et al. 2012
Mouse (CD), adult, M	Oral	20 mg/kg	1 day	671 hours	Sundström et al. 2012
Mouse (CD), adult, F	Oral	1 mg/kg	1 day	597 hours	Sundström et al. 2012
Mouse (CD), adult, F	Oral	20 mg/kg	1 day	643 hours	Sundström et al. 2012
PFNA—Mouse					
Mouse (CD-1), adult, M	Oral	1 or 10 mg/kg	1 day	823.2–1,653.6 hours	Tatum-Gibbs et al. 2011
Mouse (CD-1), adult, F	Oral	1 or 10 mg/kg	1 day	619.2–1,641.6 hours	Tatum-Gibbs et al. 2011
PFBA—Mouse					
Mouse (CD1), adult, M	Oral	10 mg/kg	1 day	13.34 hours (SE 4.55)	Chang et al. 2008b
Mouse (CD1), adult, M	Oral	30 mg/kg	1 day	16.3 hours (SE 7.2)	Chang et al. 2008b
Mouse (CD1), adult, M	Oral	100 mg/kg	1 day	5.22 hours (SE 2.27)	Chang et al. 2008b
Mouse (CD1), adult, F	Oral	10 mg/kg	1 day	2.87 hours (SE 0.30)	Chang et al. 2008b
Mouse (CD1), adult, F	Oral	30 mg/kg	1 day	3.08 hours (SE 0.26)	Chang et al. 2008b
Mouse (CD1), adult, F	Oral	100 mg/kg	1 day	2.79 hours (SE 0.3)	Chang et al. 2008b
PFOS—Rabbit					
Rabbit (New Zealand), adult, F	Oral	0.085 mg/kg/day	102 days	87 days (SD 31)	Tarazona et al. 2016

[a]Exposure durations of 1 day indicate that a single dose was administered.
[b]Reported half-lives are arithmetic means for the terminal elimination phase if multiple elimination phases were observed.

CI = confidence interval; CL = confidence limit; F = female; GM = geometric mean; IP = intraperitoneal; IV = intravenous; M = male; NA = not applicable; PFBA = perfluorobutanoic acid; PFDA = perfluorodecanoic acid; PFHpA = perfluoroheptanoic acid; PFHxS = perfluorohexane sulfonic acid; PFNA = perfluorononanoic acid; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; FOSA = perfluorooctane sulfonamide; SD = standard deviation; SE = standard error

Table 3-6. Summary Systemic Clearance for Perfluoroalkyls Estimated in Experimental Animals

Species, age, and sex	Route	Dose (mg/kg)	Exposure duration	Systemic clearance (mL/day/kg)[a]	Reference
PFOA—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10	1 day	12.4 (SD 7.4)	Butenhoff et al. 2004c
Cynomolgus monkey, adult, F	IV	10	1 day	5.3 (SD 3.3)	Butenhoff et al. 2004c
PFOS—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	2	1 day	1.10 (SE 0.06)	Chang et al. 2012
Cynomolgus monkey, adult, F	IV	2	1 day	1.65 (SE 0.04)	Chang et al. 2012
PFHxA—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10	1 day	569	Chengelis et al. 2009a
Cynomolgus monkey, adult, F	IV	10	1 day	535	Chengelis et al. 2009a
PFHxS—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10	1 day	1.3 (SE 0.1)	Sundström et al. 2012
Cynomolgus monkey, adult, F	IV	10	1 day	1.9 (SE 0.4)	Sundström et al. 2012
PFBA—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10	1 day	2,371 (SE 293)	Chang et al. 2008a
Cynomolgus monkey, adult, F	IV	10	1 day	1,075 (SE 91)	Chang et al. 2008a
PFBS—Nonhuman primate					
Cynomolgus monkey, adult, M	IV	10	1 day	159	Chengelis et al. 2009a
Cynomolgus monkey, adult, F	IV	10	1 day	238	Chengelis et al. 2009a
Cynomolgus monkey, adult, M	IV	10	1 day	12,264 (SE 3384)	Olsen et al. 2009
Cynomolgus monkey, adult, F	IV	10	1 day	8,832 (SE 2880)	Olsen et al. 2009
PFOA—Rat					
Rat (Sprague-Dawley), adult, M	Oral	0.1	1 day	23.1 (SD 5.8)	Kemper 2003
Rat (Sprague-Dawley), adult, M	Oral	1	1 day	20.9 (SD 3.8)	Kemper 2003
Rat (Sprague-Dawley), adult, M	Oral	1	1 day	40.40 (SD 2.29)	Kim et al. 2016b

Table 3-6. Summary Systemic Clearance for Perfluoroalkyls Estimated in Experimental Animals

Species, age, and sex	Route	Dose (mg/kg)	Exposure duration	Systemic clearance (mL/day/kg)[a]	Reference
Rat (Sprague-Dawley), adult, M	Oral	5	1 day	20.4 (SD 5.0)	Kemper 2003
Rat (Sprague-Dawley), adult, M	Oral	25	1 day	27.1 (SD 7.4)	Kemper 2003
Rat (Sprague-Dawley), adult, M	IV	1	1 day	21.5 (SD 2.0)	Kemper 2003
Rat (Sprague-Dawley), adult, M	IV	1	1 day	47.39 (SD 3.40)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	0.1	1 day	778 (SD 144)	Kemper 2003
Rat (Sprague-Dawley), adult, F	Oral	1	1 day	655 (SD 173)	Kemper 2003
Rat (Sprague-Dawley), adult, F	Oral	1	1 day	645.12 (SD 43.44)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	5	1 day	1,164 (SD 118)	Kemper 2003
Rat (Sprague-Dawley), adult, F	Oral	25	1 day	842 (SD 166)	Kemper 2003
Rat (Sprague-Dawley), adult, F	IV	1	1 day	816 (SD 221)	Kemper 2003
Rat (Sprague-Dawley), adult, F	IV	1	1 day	612.84 (SD 32.54)	Kim et al. 2016b
Rat (Wistar), adult, M	IV	21.5	1 day	50.4 (SD 14.4)	Kudo et al. 2002
Rat (Wistar), adult, F	IV	21.5	1 day	2,233 (SD 805)	Kudo et al. 2002
Rat (Wistar), adult, M	IV	20.1	1 day	135 (SD 29)	Ohmori et al. 2003
Rat (Wistar), adult, F	IV	20.1	1 day	2,233 (SD 805)	Ohmori et al. 2003
PFOS—Rat					
Rat (Sprague-Dawley), adult, M	Oral	2	1 day	7.33 (SD 0.55)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	Oral	2	1 day	11.3 (SE 0.56)	Chang et al. 2012
Rat (Sprague-Dawley), adult, M	Oral	15	1 day	4.9 (SE 0.52)	Chang et al. 2012
Rat (Sprague-Dawley), adult, M	IV	2	1 day	9.24 (SD 0.37)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	2	1 day	8.52 (SD 0.37)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	Oral	2	1 day	22.2 (SE 0.28)	Chang et al. 2012
Rat (Sprague-Dawley), adult, F	Oral	15	1 day	5.4 (SE 20)	Chang et al. 2012
Rat (Sprague-Dawley), adult, F	IV	2	1 day	9.82 (SD 0.21)	Kim et al. 2016b

Table 3-6. Summary Systemic Clearance for Perfluoroalkyls Estimated in Experimental Animals

Species, age, and sex	Route	Dose (mg/kg)	Exposure duration	Systemic clearance (mL/day/kg)[a]	Reference
PFDA—Rat					
Rat (Wistar), adult, M	IV	25	1 day	207 (SD 0.054)	Ohmori et al. 2003
Rat (Wistar), adult, F	IV	25	1 day	140 (SD 0.008)	Ohmori et al. 2003
PFNA—Rat					
Rat (Wistar), adult, M	IV	22.6	1 day	6.9 (SD 0.6)	Ohmori et al. 2003
Rat (Wistar), adult, F	IV	22.6	1 day	106 (SD 31)	Ohmori et al. 2003
PFHpA—Rat					
Rat (Wistar), adult, M	IV	17.7	1 day	1,604 (SD 558)	Ohmori et al. 2003
Rat (Wistar), adult, F	IV	17.7	1 day	3,071 (SD 781)	Ohmori et al. 2003
PFHxA—Rat					
Rat (Sprague-Dawley), adult, M	IV	10	1 day	2,784	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	IV	10	1 day	18,600	Chengelis et al. 2009a
PFHxS—Rat					
Rat (Sprague-Dawley), adult, M	Oral	4	1 day	7.15 (SD 0.06)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	IV	4	1 day	9.01 (SD 0.05)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, M	IV	10	1 day	6.7 (SE 0.06)	Sundström et al. 2012
Rat (Sprague-Dawley), adult, F	Oral	4	1 day	124.83 (SD 3.40)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	IV	4	1 day	227.93 (SD 6.73)	Kim et al. 2016b
Rat (Sprague-Dawley), adult, F	IV	10	1 day	53.4 (SE 4.38)	Sundström et al. 2012
PFBA—Rat					
Rat (Sprague-Dawley), adult, M	IV	30	1 day	851 (SE 61)	Chang et al. 2008a
Rat (Sprague-Dawley), adult, F	IV	30	1 day	2,949 (SE 59)	Chang et al. 2008a
Rat (Sprague-Dawley), adult, M	Oral	30	1 day	494 (SE 29)	Chang et al. 2008a
Rat (Sprague-Dawley), adult, F	Oral	30	1 day	1,527 (SE 145)	Chang et al. 2008a
PFBS—Rat					
Rat (Sprague-Dawley), adult, M	IV	10	1 day	946	Chengelis et al. 2009a
Rat (Sprague-Dawley), adult, F	IV	10	1 day	7,464	Chengelis et al. 2009a

Table 3-6. Summary Systemic Clearance for Perfluoroalkyls Estimated in Experimental Animals

Species, age, and sex	Route	Dose (mg/kg)	Exposure duration	Systemic clearance (mL/day/kg)[a]	Reference
Rat (Sprague-Dawley), adult, M	IV	30	1 day	2,856 (SE 816)	Olsen et al. 2009
Rat (Sprague-Dawley), adult, F	IV	30	1 day	11,265 (SE 960)	Olsen et al. 2009
PFOA—Mouse					
Mouse (FVB/NJcl), adult, M	IV	0.13	1 day	14.2 (SD 8.4)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	IV	0.13	1 day	11.8 (SD 6.1)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, M	Oral	1.3	1 day	13.1 (SD 7.4)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	Oral	1.3	1 day	9.0 (SD 1.9)	Fujii et al. 2015a, 2015b
PFOS—Mouse					
Mouse (CD), adult, M	Oral	1	1 day	4.7	Chang et al. 2012
Mouse (CD), adult, M	Oral	20	1 day	4.7	Chang et al. 2012
Mouse (CD), adult, F	Oral	1	1 day	5.0	Chang et al. 2012
Mouse (CD), adult, F	Oral	20	1 day	6.0	Chang et al. 2012
PFHxS—Mouse					
Mouse (CD), adult, M	Oral	1	1 day	2.9	Sundström et al. 2012
Mouse (CD), adult, M	Oral	20	1 day	4.8	Sundström et al. 2012
Mouse (CD), adult, F	Oral	1	1 day	2.7	Sundström et al. 2012
Mouse (CD), adult, F	Oral	20	1 day	3.8	Sundström et al. 2012
PFNA—Mouse					
Mouse (FVB/NJcl), adult, M	IV	0.14	1 day	3.9 (SD 1.9)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	IV	0.14	1 day	5.1 (SD 2.3)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, M	Oral	1.4	1 day	4.0 (SD 1.7)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	Oral	1.4	1 day	2.4 (SD 1.0)	Fujii et al. 2015a, 2015b
PFDA—Mouse					
Mouse (FVB/NJcl), adult, M	IV	0.16	1 day	2.2 (SD 0.9)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	IV	0.16	1 day	2.8 (SD 1.2)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, M	Oral	1.6	1 day	3.9 (SD 1.8)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	Oral	1.6	1 day	2.2 (SD 1.1)	Fujii et al. 2015a, 2015b

Table 3-6. Summary Systemic Clearance for Perfluoroalkyls Estimated in Experimental Animals

Species, age, and sex	Route	Dose (mg/kg)	Exposure duration	Systemic clearance (mL/day/kg)[a]	Reference
PFUnA—Mouse					
Mouse (FVB/NJcl), adult, M	IV	0.17	1 day	2.8 (SD 1.0)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	IV	0.17	1 day	3.4 (SD 1.5)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, M	Oral	1.7	1 day	5.7 (SD 2.6)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	Oral	1.7	1 day	3.1 (SD 1.7)	Fujii et al. 2015a, 2015b
PFDoDA—Mouse					
Mouse (FVB/NJcl), adult, M	IV	0.19	1 day	4.4 (SD 1.6)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	IV	0.19	1 day	4.8 (SD 2.4)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, M	Oral	1.9	1 day	9.4 (SD 4.1)	Fujii et al. 2015a, 2015b
Mouse (FVB/NJcl), adult, F	Oral	1.9	1 day	5.2 (SD 3.2)	Fujii et al. 2015a, 2015b
PFBA—Mouse					
Mouse (CD1), adult, M	Oral	10	1 day	280 (SE 72)	Chang et al. 2008b
Mouse (CD1), adult, M	Oral	30	1 day	296 (SE 640)	Chang et al. 2008b
Mouse (CD1), adult, M	Oral	100	1 day	784 (SE 112)	Chang et al. 2008b
Mouse (CD1), adult, F	Oral	10	1 day	564 (SE 24)	Chang et al. 2008b
Mouse (CD1), adult, F	Oral	30	1 day	696 (SE 32)	Chang et al. 2008b
Mouse (CD1), adult, F	Oral	100	1 day	1,336 (SE 64)	Chang et al. 2008b

[a]As reported in units of mL/day/kg or converted from mL/hour (x24), mL/hour (x24/body weight) or mL/minute (x60x24).

CI = confidence interval; F = female; IV = intravenous; M = male; PFBA = perfluorobutanoic acid; PFBS = perfluorobutane sulfonic acid; PFDA = perfluorodecanoic acid; PFHpA = perfluoroheptanoic acid; PFHxA = perfluorohexanoic acid; PFHxS = perfluorohexane sulfonic acid; PFNA = perfluorononanoic acid; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; SD = standard deviation; SE = standard error

Elimination of Perfluoroalkyls in Humans. Elimination $t_{1/2}$ values for PFOA, PFOS, PFHxS, PFBA, and PFBS have been estimated in humans (Bartell et al. 2010; Costa et al. 2009; Chang et al. 2008a; Glynn et al. 2012; Harada et al. 2005a; Li et al. 2018; Olsen et al. 2007a, 2009; Seals et al. 2011; Spliethoff et al. 2008; Yeung et al. 2013; Wong et al. 2014, 2015; Worley et al. 2017a; Zhang et al. 2013). Estimates in humans are based on measurements of the decline in serum perfluoroalkyl concentrations following cessation or an abrupt decrease in exposure, or on measurements of renal plasma clearance

from serum in a general population sample from Japan (Harada et al. 2005a). The latter clearance estimates were converted to $t_{1/2}$ values, for display in Table 3-5 as follows (Equations 3-4 and 3-5):

$$k_e = \frac{Cl}{V} \qquad \text{Eq. (3-4)}$$

$$t_{1/2} = \frac{\ln(2)}{k_e} \qquad \text{Eq. (3-5)}$$

where k_e is the elimination rate constant (e.g., day^{-1}), Cl is the renal plasma clearance (e.g., mL plasma/day/kg), and V is the plasma volume (L/kg), which is assumed to be 4.3% of body weight (ICRP 1981). In general, these studies show that longer chain length is associated with slower elimination rates. For example, the elimination $t_{1/2}$ for PFBA was estimated to be 70–80 hours (Chang et al. 2008a), whereas the $t_{1/2}$ values for PFHxS, PFOS, and PFOA range from 2 to 35 years (Bartell et al. 2010; Harada et al. 2005a; Li et al. 2018; Olsen et al. 2007a; Seals et al. 2011; Worley et al. 2017a; Zhang et al. 2013). Longer $t_{1/2}$ values for PFOA have been reported with longer monitoring follow-up times, which allow the detection of slower elimination phases of multiphasic elimination kinetics (Seals et al. 2011). Perfluoroalkyl sulfonates are eliminated more slowly in humans than corresponding carboxylates of the same chain length (Zhang et al. 2013). Analytical methods typically used to measure serum perfluoroalkyls do not discriminate between linear and branched isomers and, as a result, these studies estimate elimination rates for the isomer mixture. A study that compared elimination rates of isomers of PFOA found that linear isomers tend to be eliminated more slowly than branched isomers (Zhang et al. 2013), consistent with results of studies conducted in rats (Benskin et al. 2009; De Silva et al. 2009).

An analysis of serum PFOS data from NHANES indicated that $t_{1/2}$ in females may be shorter (4.3 years) compared to males (4.7 years; Wong et al. 2014, 2015). The NHANES data are cross-sectional and, therefore, the estimates of $t_{1/2}$ required fitting the data to age patterns of PFOS intake. An improved fit to the data for females was achieved when estimated losses of PFOS in menstrual fluids were considered, suggesting that menstrual loss of PFOS may account for some, but not all, of the sex difference in the elimination rate (Verner and Longnecker 2015; Wong et al. 2015). Li et al. (2018) also found apparent sex differences in PFOS elimination in male and female residents in Sweden exposed to contaminated drinking water. The estimated $t_{1/2}$ for PFOS were 4.6 years in males and 3.1 years in females. Zhang et al. (2013) estimated serum $t_{1/2}$ for various age and sex strata in a population of 86 individuals. Serum $t_{1/2}$ for PFOS was lower for PFOS in younger females (≤50 years, $t_{1/2}$ = 6.2 years±0.3 SE, n=66) compared to males and older females ($t_{1/2}$ =27 years±3.1 SE, n=20). Zhang et al. (2013) attributed the difference in serum $t_{1/2}$ to clearance in menstrual fluids. However, the estimated serum $t_{1/2}$ of 27 years is much higher than values calculated from other studies; Zhang et al. (2013) noted that the serum $t_{1/2}$ should be

considered as an upper limit estimate. Estimated $t_{1/2}$ for PFOA was not different in younger females (2.1±0.3 SE, n=20) compared to males and older females (2.6±0.4 SE, n=66). Declines in serum PFOA concentrations were observed in populations following initiation of activated carbon filtration of public water supplies that had been contaminated with PFOA (Bartell et al. 2010). The estimated mean serum $t_{1/2}$ for a group of 200 adults followed for 1 year after filtration was initiated was 2.3 years (95% CI 2.1–2.4). Elimination rates were not different in males and females. Serum PFOA concentration ranged from 16 to 1,200 ng/mL. A larger follow-up study measured serum PFOA concentrations in two populations of former residents (n=1,672) of the same water districts (Seals et al. 2011). In one population (n=643), the serum $t_{1/2}$ increased with increasing elapsed time since leaving the water district. The $t_{1/2}$ values were 2.9 years (95% CI 2.3–3.8) for elapsed time of <4 years and 10.1 years for elapsed time of >4 years. In a second population with an elapsed time since residence of <9 years, the $t_{1/2}$ was 8.5 years (95% CI 7.1–10.1). Elimination rates (based on the annual percent decrease in serum concentrations) were faster in males (27%) compared to females (18%) for the first 4 years post-exposure; however, no difference was evident between sexes when elapsed time from exposure was >4 years.

Bartell (2012) and Russell et al. (2015) point out that most studies examining PFOA elimination half-lives fail to account for ongoing background exposure, which could result in an overestimation of elimination half-lives. Bartell (2012) estimated that the bias from background exposure could resulted in 1–26% overestimation of calculated PFOA half-lives and that greater overestimations can occur for half-lives based on longer follow-up times. Russell et al. (2015) estimated that the bias was greatest in populations with serum PFOA levels closest to background levels. In a re-analysis of the Olsen et al. (2007) occupational exposure data, Russell et al. (2015) estimated that overestimation was approximately 1.2% in workers with initial serum concentrations >500 ng/mL (100 times higher than NHANES general population data) and 13% for workers with lower initial serum PFOA levels. Restricting the elimination half-life calculation to workers with initial serum PFOA levels of >500 ng/mL would result in a half-life of 3.0 years (Russell et al. 2015), compared to 3.8 years calculated for the whole cohort (Olsen et al. 2007).

Analysis of kinetics of serum PFOS concentrations in retired U.S. fluorochemical production workers (24 males, 2 females) yielded a mean serum elimination $t_{1/2}$ estimate of 5.4 years (95% CI 3.9–6.9; geometric mean: 4.8 years, 95% CI 4.0–5.8) in subjects whose serum PFOS concentrations ranged from 37 to 3,490 ng/mL (Olsen et al. 2007a). Estimates for the two females in the same study were 4.9 and 6.8 years. Estimates based on renal clearance of PFOS from serum in subjects from the general population of Japan ranged from 2.9 to 7.4 years; these subjects had serum PFOS concentrations that

ranged from 4 to 49 ng/mL (Harada et al. 2005a). Estimates in males (7.4, 2.9 years) were similar to females (4.5, 4.6 years). This same study measured serum PFHxS concentrations in retired U.S. fluorochemical production workers (24 males, 2 females) and yielded a mean estimate of 8.5 years (95% CI 6.4–10.6; geometric mean: 7.3 years, 95% CI 5.8–9.2) for the serum elimination $t_{1/2}$ in subjects whose serum PFHxS concentrations ranged from 10 to 1,295 ng/mL (Olsen et al. 2007a). Estimates for the two females in the same study were 12.2 and 13.3 years.

The elimination rate of PFBA was estimated in fluorochemical workers who may have been exposed to various PFBA precursors (Chang et al. 2008a). In three male workers, the estimated mean $t_{1/2}$ based on serum PFBA kinetics was 81 hours (±41 SD). In a larger study of nine workers (seven males, two females), the mean $t_{1/2}$ was 72 hours (±38 SD). Estimates for the two female subjects were 56 and 118 hours. The combined mean value for the 12 estimates was 75 hours (±38 SD). Olsen et al. (2009) estimated serum $t_{1/2}$ of PFBS in six fluorochemical workers. The mean $t_{1/2}$ was 27.4 days (±11.1 SD). The group included a single female whose $t_{1/2}$ was 45.7 days. Based on these observations, PFBA (chain length 3) and PFBS (chain length 4) are eliminated substantially faster in humans than perfluoroalkyls having longer carbon chain lengths, such as PFHxS (chain length 6), PFOA (chain length 7), and PFOS (chain length 8).

Temporal trends in perfluoroalkyl serum concentrations have also been used to estimate population halving times (Glynn et al. 2012; Olsen et al. 2012; Spleithoff et al. 2008; Yeung et al. 2013). Population halving times are influenced by temporal trends in intakes and may therefore not accurately reflect clearance. Population halving times for PFOS ranged from 4 to 5 years (Olsen et al. 2012; Spleithoff et al. 2008; Yeung et al. 2013). Glynn et al. (2012) monitored serum perfluoroalkyls in a population of pregnant women (n=413) in Sweden over the period 1996–2010. Halving times were 22 years (95% CI 16–38) for PFOA and 8.2 years (95% CI 6.3–12) for PFOS.

Elimination of Perfluoroalkyls in Nonhuman Primates. Elimination $t_{1/2}$ values and systemic clearances for PFOA, PFOS, PFHxA, PFHxS, PFBA, and PFBS have been estimated in Cynomolgus monkeys (Buttenoff et al. 2004c; Chang et al. 2012; Chengelis et al. 2009a; Olsen et al. 2009; Seacat et al. 2002; Sundström et al. 2012). Estimated terminal $t_{1/2}$ values were 20–30 days for PFOA, 100–170 days for PFOS, 90–140 days for PFHxS, 40 hours for PFBA and 8–95 hours for PFBS. Elimination of perfluoroalkyls in monkeys is multiphasic and, as a result, estimates of the terminal $t_{1/2}$ can vary with the duration of the observation period and assumptions made in modeling elimination kinetics (Chang et al. 2012; Chengelis et al. 2009a; Olsen et al. 2009; Sundström et al. 2012). For example, the $t_{1/2}$ values for

PFBS were 8 and 15 hours in female and male monkeys, respectively, when monkeys were monitored for 48 hours following a single intravenous dose (Chengelis et al. 2009a), whereas the $t_{1/2}$ values were 95 and 83 hours in male and female monkeys, respectively, when the monitoring period was extended to 14 days and a three-compartment model was used to estimate the terminal $t_{1/2}$ (Olsen et al. 2009). Studies in monkeys confirm general trends observed in humans that perfluoroalkyl sulfonates are more slowly eliminated than perfluoroalkyl carboxylates and that elimination of longer-chain perfluoroalkyls occurs more slowly than short-chain perfluoroalkyls. Systemic clearances were lower for PFOS, PFHxS, and PFBS compared to the corresponding carboxylates, PFOA, PFHxA, and PFBA (Table 3-6). Systemic clearances were similar in male and female monkeys (Table 3-6).

Elimination of Perfluoroalkyls in Rats. Elimination $t_{1/2}$ values and systemic clearances for PFOA, PFOS, FOSA, PFDA, PFNA, PFHpA, PFHxA, PFHxS, PFBA, and PFBS have been estimated in rats (Benskin et al. 2009; Chang et al. 2008b, 2012; Chengelis et al. 2009a; De Silva et al. 2009; Johnson and Ober 1979; Kemper 2003; Kim et al. 2016b; Kudo et al. 2002; Ohmori et al. 2003; Olsen et al. 2009; Seacat and Luebker 2000; Sundström et al. 2012; Vanden Heuvel et al. 1991b, 1991c; Ylinen et al. 1990). Consistent with observations made in humans and Cynomolgus monkeys, perfluoroalkyl sulfonates are more slowly eliminated than perfluoroalkyl carboxylates and short-chain perfluoroalkyls (e.g., PFBA, PFBS) are eliminated faster in rats than long-chain perfluoroalkyls (e.g., PFOA, PFOS, PFHxA, PFHxS); Tables 3-5 and 3-6. Linear PFOA isomers tend to be eliminated more slowly than branched isomers (Benskin et al. 2009; De Silva et al. 2009).

Elimination of perfluoroalkyls exhibits pronounced sex differences in rats, with faster elimination in females than in males (Benskin et al. 2009; Chang et al. 2008b; Chengelis et al. 2009a; Kemper 2003; Kim et al. 2016b; Kudo et al. 2002; Ohmori et al. 2003; Sundström et al. 2012; Tatim-Gibbs et al. 2011; Vanden Heuvel et al. 1991c; Ylinen et al. 1990). Estimates of systemic clearance for PFOA in male rats ranged from 20 to 50 mL/day/kg, whereas estimates for female rats ranged from 600 to 2,200 mL/day/kg (Kemper 2003; Kudo et al. 2002; Ohmori et al. 2003). Systemic clearances of PFOA, PFOS, PFNA, PFHxA, PFHxS, PFBA, and PFBS are also higher in female rats compared to male rats (Table 3-6). Pronounced sex difference in elimination rates in rats (faster elimination in females) was observed in rats following 30-minute nose-only exposures to aerosols (MMAD=1.9–2.1 μm) of 1–25 mg ammonium PFOA/m^3 (Hinderliter et al. 2006a). Plasma PFOA concentrations were not detectable 12 hours after exposure of female rats, and were approximately 90% of peak plasma concentrations 24 hours after the exposure in male rats. The slower elimination of PFOA in male rats resulted in steady-state plasma concentrations within 3 weeks of repeated exposures (6 hours/day, 5 days/week) in male rats, whereas in

female rats, daily periodic oscillations of plasma concentrations from peak to below detection occurred on each day of exposure. Steady-state plasma concentrations in male rats were approximately 10 times that of daily peak concentrations in female rats.

Pronounced dose dependence appears in the $t_{1/2}$ estimates for PFOA in female rats. With increasing dose, plasma elimination kinetics in female rats converts from monophasic to biphasic. Following an oral dose of PFOA of 0.1, 1, 5, or 25 mg/kg, the terminal $t_{1/2}$ values in female rats were 3.2, 3.5, 4.6, or 16.2 hours, respectively; no apparent dose dependence was observed in male rats over the same dose range (Kemper 2003). Dose-dependent elimination of PFOA has been attributed to a capacity-limited renal tubular secretion of PFOA in female rats (see discussion below on *Mechanisms of Excretion*). The divergence in elimination kinetics between male and female rats appears to be age-dependent, with faster elimination becoming evident in female rats after 30 days of age, consistent with the timing of sexual maturation and involvement of sex hormones in the modulation of the renal excretion of PFOA in rats (Hinderliter et al. 2006b).

Elimination of Perfluoroalkyls in Mice. Elimination $t_{1/2}$ values and systemic clearances for PFOS, PFHxS, and PFBA have been estimated in mice (Chang et al. 2008a, 2012; Sundström et al. 2012). Consistent with studies conducted in rats and monkeys, PFBA is eliminated more rapidly in mice than PFOS and PFHxS. Systemic clearances ranged from 5 to 6 mL/day/kg for PFOS (Chang et al. 2012), from 3 to 5 mL/day/kg for PFHxS (Sundström et al. 2012), and from 300 to 1,300 mL/day/kg for PFBA (Chang et al. 2008a). Sex differences in elimination in mice were observed for PFBA, but not PFOS or PFHxS. Systemic clearances of PFBA in female mice were approximately 2 times that of males (Chang et al. 2008a). Systemic clearance of PFBA in male and female mice appeared to be dependent on dose. Systemic clearance following a single oral dose of 100 mg PFBA/kg was approximately 2 times higher than the systemic clearance following a dose of 10 or 30 mg PFBA/kg. Possible explanations for the apparent dependence of clearance on dose are dose-dependent bioavailability or that the one-compartment model used to estimate elimination rates and serum AUC did not adequately fit the serum kinetics observed at the higher dose (Chang et al. 2008a). The latter could occur if renal tubular reabsorption of PFBA or plasma protein binding of PFBA is saturable in mice. Systemic clearance rates for PFOA were similar in male mice (13.1 mL/kg/day) and in female mice (9.0 mL/kg/day) (Fuji et al. 2015a, 2015b).

Elimination of Perfluoroalkyls in Other Species. Sex differences in elimination of PFOA have also been observed in hamsters; unlike the rat, male hamsters excreted absorbed PFOA more rapidly than female hamsters. Following a single gavage dose of 10 mg/kg as ammonium [^{14}C]PFOA, cumulative excretion

of ^{14}C in urine at 24 hours post-dosing was 96.4% of the dose in female rats and 8.7% in male rats; 24.6% and 84.5% in female and male hamsters, respectively; 4.1% in male and female mice; and 90.5 and 80.2% in female and male rabbits, respectively (Hundley et al. 2006).

Mechanisms of Excretion. Urinary excretion of perfluoroalkyls involves glomerular filtration and renal tubular secretion and reabsorption (for PFOA, see Harada et al. 2005a; Kudo et al. 2002; Ohmori et al. 2003). Glomerular filtration of PFOA is limited by extensive binding of PFOA to albumin and other high molecular weight proteins in plasma (Han et al. 2003, 2005; Ohmori et al. 2003; Kerstner-Wood et al. 2003; Vanden Heuvel et al. 1992a, 1992b; Ylinen and Auriola 1990). Elimination of PFOA and other perfluoroalkyls shows pronounced sex differences in rats, with slower elimination in males for PFOA, PFOS, PFNA, PFHxA, PFHxS, PFBA, and PFBS (Chang et al. 2008a, 2012; Chengelis et al. 2009a; Kemper 2003; Kudo et al. 2002; Ohmori et al. 2003; Sundström et al. 2012). The sex difference in PFOA elimination in rats is dependent on testosterone (Hinderliter et al. 2006b; Kudo et al. 2002; Vanden Heuvel et al. 1992a). The significantly slower elimination of PFOA in adult male rats compared to female rats has been attributed to sex hormone modulation of organic anion transporters in kidney. At similar doses administered to male and female rats, PFOA undergoes net tubular reabsorption in male rats (i.e., urinary excretion rate < rate of glomerular filtration of PFOA) and net tubular secretion in female rats (i.e., urinary excretion rate > rate of glomerular filtration of PFOA) (Harada et al. 2005a; Kudo et al. 2002; Ohmori et al. 2003). In rats, several transporters have been shown to have affinity for C7–C9 perfluoroalkyl carboxylates. The transporters, OAT1 and OAT3, located on the basolateral membrane of the renal proximal tubule, appear to participate in secretion of C7–C9 perfluoroalkyl carboxylates into the tubular fluid (Nakagawa et al. 2008; Weaver et al. 2010). The transporters, OATP1a1 (rat), OAT4 (human), and URAT1 (human), located on the apical membrane, appear to mediate reabsorption of C8–C10 perfluoroalkyl carboxylates from the tubular fluid (Katakura et al. 2007; Nakagawa et al. 2009; Weaver et al. 2010; Yang et al. 2009, 2010). In rats and mice, expression of OAT1, OAT3, and OATP1a1 is controlled by male sex hormones and shows higher activities in males (Buist and Klaassen 2004; Gotoh et al. 2002; Kobayashi et al. 2002; Li et al. 2002; Lu et al. 1996; Lubojevic et al. 2004). The slower elimination of PFOA (and other long-chain perfluoroalkyl carboxylates) in male rats has been attributed to OATP1a1 (Weaver et al. 2010; Yang et al. 2009). Higher activity of OATP1a1 in male rats results in higher reabsorptive transport and lower rates of urinary excretion. However, saturation of this transporter could result in an increase in urinary elimination of perfluoroalkyls due to decreased tubular reabsorption. This is consistent with the apparent plateau in plasma concentration with increasing dose observed in cancer patients treated with PFOA (Convertino et al. 2018). Affinities of OATP1a1 (rat), OAT4 (human), and URAT1 (human) are highest for C7–C10 perfluoroalkyl carboxylates (Weaver et al.

2010; Yang et al. 2009, 2010). Affinity of rat OATP1a1 is strongly correlated with total clearance in rats (r^2=0.98; Yang et al. 2009).

Although sex differences for elimination of perfluoroalkyls have been detected in laboratory animals, human monitoring studies have not consistently detected sex differences in elimination $t_{1/2}$ of perfluoroalkyls; this may reflect limitations in the studies, including numbers and age of subjects (Bartell et al. 2010; Seals et al. 2011; Wong et al. 2014, 2015; Zhang et al. 2013). Menstruation may contribute to faster elimination of PFOS in women (Wong et al. 2014, 2015; Zhang et al. 2013). The effect of menstruation or other variables related to menstruation appear to contribute to faster elimination in younger (≤50 years) women compared to men and older women (Zhang et al. 2013). Two studies have found evidence for elimination of PFOS being affected by menstruation (Wong et al. 2014, 2015; Zhang et al. 2013). The estimated $t_{1/2}$ for PFOA was not different in younger females compared to males and older females. Mechanisms by which menstruation could affect PFOS clearance are not understood. Bulk elimination of blood would be expected to affect serum clearance of both PFOS and PFOA; therefore, other mechanisms must contribute that discriminate between perfluoroalkyl species. A better metric than serum $t_{1/2}$ for evaluating sex differences in elimination for this would be systemic or renal clearance of the perfluoroalkyl. Harada et al. (2005a) measured renal clearance in a small sample of young adults (five males and five females, age 22–23 years) and found that renal clearance was not different in males and females. Zhang et al. (2013) estimated renal clearance of PFOA and PFOS in a population of younger females (≤50 years, n=20), older females (>50 years), younger males (≤50 years), and older males (>50 years) and did not find significant sex or age differences. Studies that measured systemic clearance in monkeys also have not found significant sex differences in systemic clearance of PFOA (Buttenoff et al. 2004c) or PFOS (Chang et al. 2012).

Studies conducted in rats have shown that PFDA, PFNA, PFOA, PFOS, and PFHxA are secreted in bile and undergo extensive reabsorption from the gastrointestinal tract (Johnson et al. 1984; Kudo et al. 2001; Vanden Heuvel et al. 1991b, 1991c). Biliary secretion rates of PFOA are similar in male and female rats when renal excretion is blocked by ligation of the kidneys (Vanden Heuvel et al. 1991a, 1991b). This lack of sex influence on biliary secretion (compared to the sex influence on renal clearance) may reflect a relative sex insensitivity of OAT2 (or other organic anion transporter) expression in liver, compared to kidney; the latter is approximately 7–8 times higher in adult female rats compared to male rats (Kudo et al. 2002).

3.1.5 Physiologically Based Pharmacokinetic (PBPK)/Pharmacodynamic (PD) Models

PBPK models use mathematical descriptions of the uptake and disposition of chemical substances to quantitatively describe the relationships among critical biological processes (Krishnan et al. 1994). PBPK models are also called biologically based tissue dosimetry models. PBPK models are increasingly used in risk assessments, primarily to predict the concentration of potentially toxic moieties of a chemical that will be delivered to any given target tissue following various combinations of route, dose level, and test species (Clewell and Andersen 1985). Physiologically based pharmacodynamic (PBPD) models use mathematical descriptions of the dose-response function to quantitatively describe the relationship between target tissue dose and toxic endpoints.

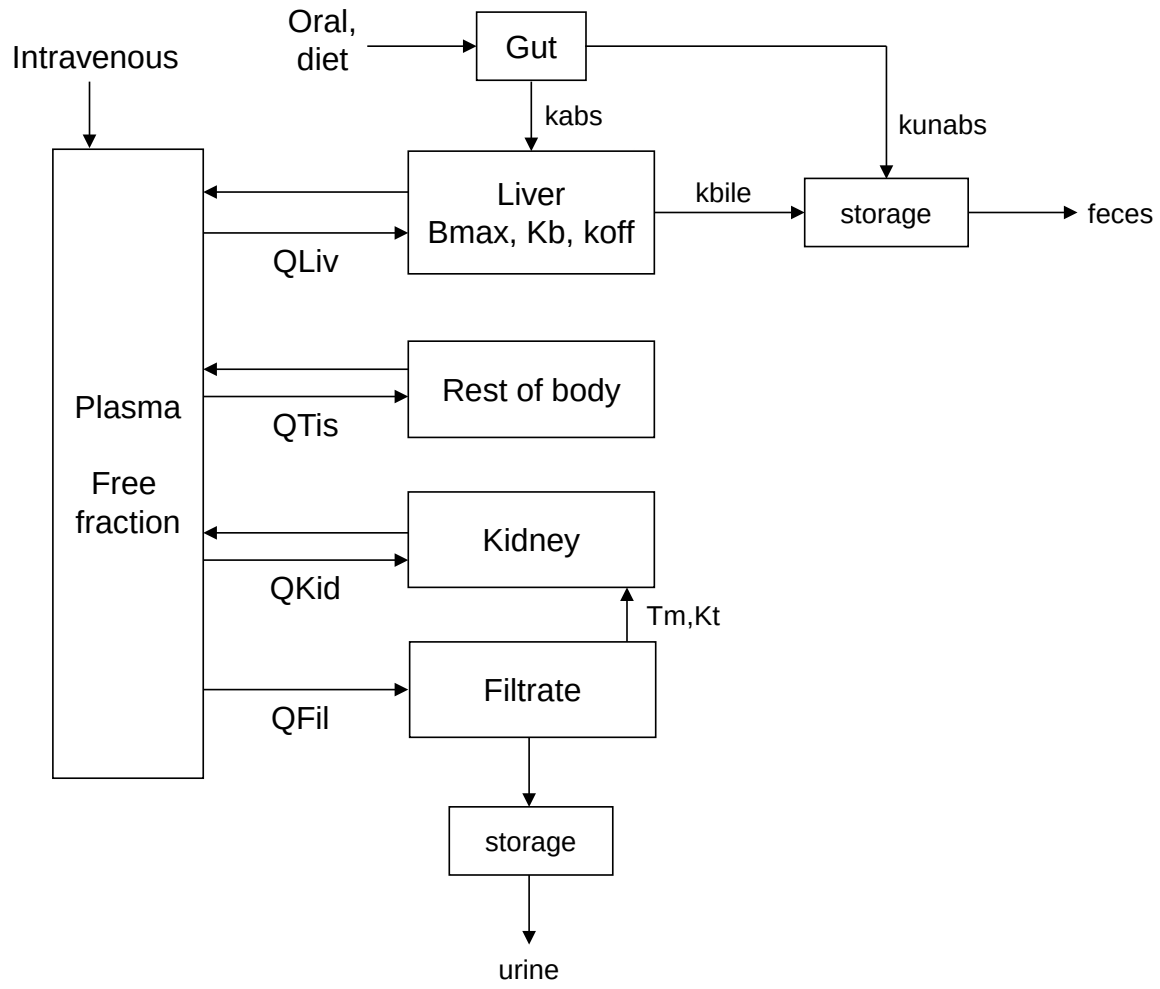
Several PBPK models of PFOA and PFOS have been reported. These include a human model for PFOA and PFOS (Fàbrega et al. 2014, 2016; Loccisano et al. 2011; Worley et al. 2017b), models for PFOA and PFOS in monkeys (Loccisano et al. 2011), models for PFOA and PFOS in rats (Harris and Barton 2008; Loccisano et al. 2012a, 2012b; Tan et al. 2008; Worley and Fisher 2015a. 2015b), and a model for PFOA in mice (Rodriguez et al. 2009). Models of PFOA and PFOS kinetics during gestation and lactation in rats and mice also have been reported (Loccisano et al. 2012a, 2012b; Rodriguez et al. 2009). Various empirical and compartmental models have also been reported (Hoffman et al. 2011; Lorber and Egeghy 2011; Lou et al. 2009; Thompson et al. 2010; Verner et al. 2016; Wambaugh et al. 2013; Wu et al. 2009). Tardiff et al. (2009) utilized a human pharmacokinetic model to estimate an average daily oral dose corresponding to a Reference Dose for PFOA plasma concentration in humans. Cheng and Ng (2017) developed a permeability-limited PBPK model for PFOA in male rats that could be used for *in vitro* to *in vivo* extrapolation. Kim et al. (2018) developed a PBPK model for PFHxS in rats and humans. PBPK models were not identified for other perfluoroalkyls examined in this profile. Given the toxicokinetic differences between compounds, the PFOA, PFOS, and PFHxS PBPK models may not be appropriate for other compounds.

3.1.5.1 Loccisano et al. (2012a, 2012b) Rat Models

Loccisano et al. (2012a) developed a model for simulating the kinetics of PFOA and PFOS in male and female rats. The model was based, in part, on a multi-compartmental model developed by Tan et al. (2008; Andersen et al. 2006). The female rat model (Loccisano et al. 2012a) was subsequently extended to include gestation and lactation (Loccisano et al. 2012b). The general structures of the models are depicted in Figures 3-3, 3-4, and 3-5. Complete lists of parameters and parameter values and the bases

for parameter values and evaluations of model predictions in comparison to observations are described in Loccisano et al. (2012a, 2012b).

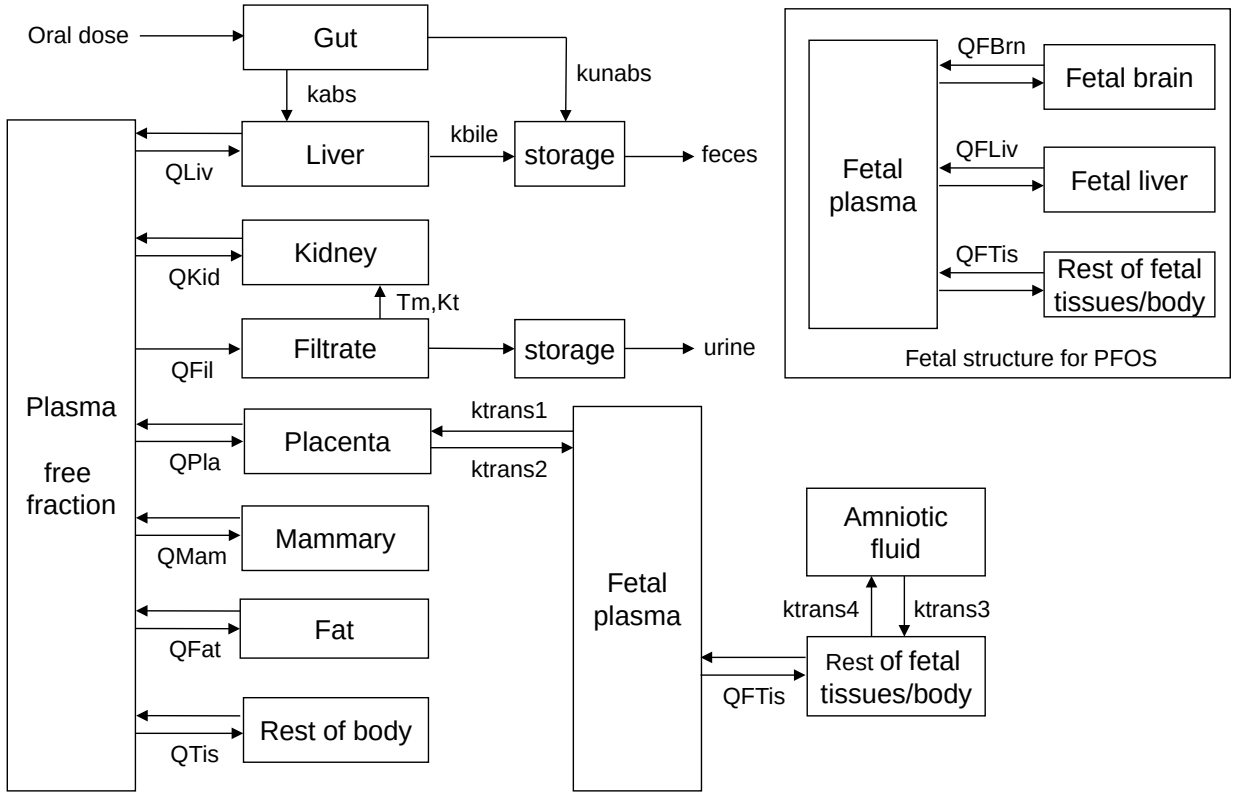
Figure 3-3. Structure of PBPK Model of PFOA and PFOS in the Rat



Bmax = liver binding capacity; kabs = first-order absorption rate constant; Kb = liver binding affinity constant; kbile = biliary excretion rate constant; Koff = liver binding dissociation constant; Kt = affinity constant; kunabs = rate of unabsorbed dose to appear in feces; PBPK = physiologically based pharmacokinetic; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; QFil = clearance from plasma to glomerular filtrate; QKid = blood flow in and out of kidney; QLiv = blood flow in and out of liver; QTis = blood flow in and out of tissues; Tm = transporter maximum

Source: Loccisano et al. 2012a (reproduced with permission of Elsevier Inc. in the format reuse in a government report via Copyright Clearance Center; Reproductive Toxicology by Reproductive Toxicology Center; Washington, DC)

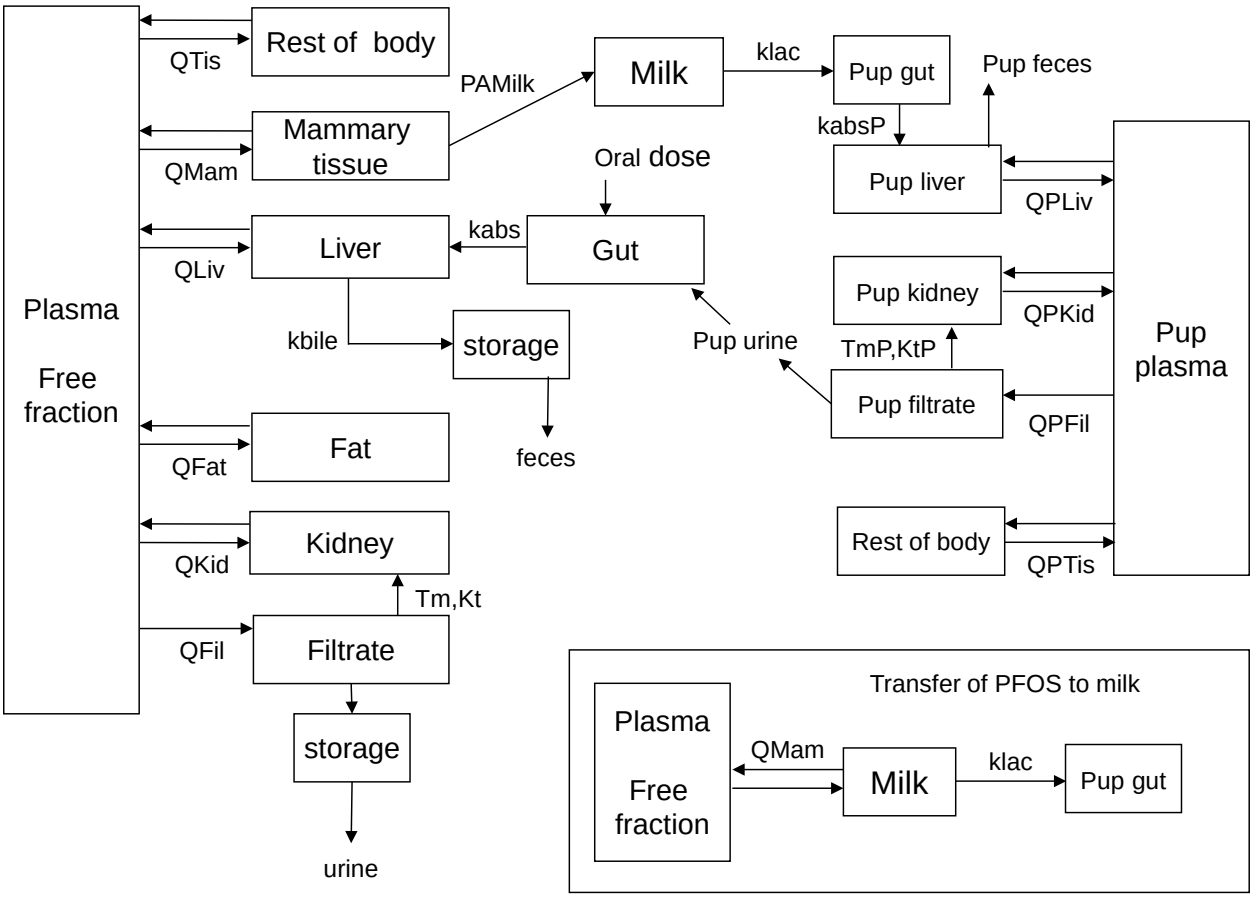
Figure 3-4. PBPK Model Structure for Simulating PFOA and PFOS Exposure During Gestation in the Rat (Dam, Left; Fetus, Right)



kabs = first-order absorption rate constant; kbile = biliary excretion rate constant; Kt = affinity constant; ktrans1/ktrans 2 = transfer between placenta and fetal plasma; ktrans3/ktrans4 = transfer between amniotic fluid and rest of the body; kunabs = rate of unabsorbed dose to appear in feces; PBPK = physiologically based pharmacokinetic; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; QFat = blood flow in and out of fat; QFBrn = blood flow in and out of fetal brain; QFil = clearance from plasma to glomerular filtrate; QFLiv = blood flow in and out of fetal liver; QFTis = blood flow in and out of fetal tissue; QKid = blood flow in and out of kidney; QLiv = blood flow in and out of liver; QMam = blood flow in and out of mammary tissue; QPla = blood flow in and out of placenta; QTis = blood flow in and out of tissues; Tm = transporter maximum

Source: Loccisano et al. 2012b (reproduced with permission of Elsevier Inc. in the format reuse in a government report via Copyright Clearance Center; Reproductive Toxicology by Reproductive Toxicology Center; Washington, DC)

Figure 3-5. PBPK Model Structure for Simulating PFOA/PFOS Exposure During Lactation in the Rat (Dam, Left; Pup, Right)



kabs = first-order absorption rate constant; kabsP = pup first-order absorption rate constant; kbile = biliary excretion rate constant; klac = transfer to pup through milk; Kt = affinity constant; KtP = pup affinity constant; PAMilk = transfer from mammary tissue to liver; PBPK = physiologically based pharmacokinetic; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; QFat = blood flow in and out of fat; QFil = clearance from plasma to glomerular filtrate; QKid = blood flow in and out of kidney; QLiv = blood flow in and out of liver; QMam = blood flow in and out of mammary tissue; QPFil = clearance from pup plasma to glomerular filtrate; QPKid = blood flow in and out of pup kidney; QPLiv = blood flow in and out of pup liver; QPTis = blood flow in and out of pup tissue; QTis = blood flow in and out of tissues; Tm = transporter maximum; TmP = pup transporter maximum

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The basic (i.e., adult nonpregnant rat) model includes compartments representing plasma (including a bound and free fraction), kidney and renal glomerular filtrate, liver, and a lumped compartment representing all other tissues. Two storage compartments are included in the model: one receives perfluoroalkyl from the gastrointestinal tract (unabsorbed) and liver (bile) and the other receives perfluoroalkyl from the glomerular filtrate. The storage compartments were included in the model to simulate time delays between elimination from plasma and appearance of perfluoroalkyl in feces or urine. Absorption from the gastrointestinal tract is simulated as the balance between first-order absorption and fecal excretion of unabsorbed chemical. Absorbed PFOA and PFOS are assumed to be delivered to the liver where saturable binding of PFOS (but not PFOA) to liver proteins occurs. Saturable binding of PFOS in liver was included to simulate the relatively long retention times of PFOS in liver that have been observed in rats. Exchanges between PFOA or PFOS in liver (free fraction), kidney, and other tissues with the free pool in plasma are assumed to be flow-limited (governed by blood flow) with equilibrium determined by the tissue:blood partition coefficient. PFOA and PFOS in plasma are simulated as instantaneous distributions into free and bound fractions. Extensive binding of PFOA and PFOS to plasma proteins has been demonstrated in various animal species including rats (see Section 3.1.2). For PFOA, the free fraction is assigned a constant of 4.5% in females and 0.6% in males. These values were optimized to fit observed kinetics of PFOA in plasma and urine of rats following intravenous and oral exposures (Loccisano et al. 2012a). Adequate fit to observed PFOS plasma kinetics following single doses of PFOS required introducing a time-dependence in binding of PFOS to protein (Loccisano et al. 2012a; Tan et al. 2008). The free fraction for PFOS in plasma decreases from an initial value (after dosing) of 2.2% to a minimum of 0.1% with a $t_{1/2}$ for the change of approximately 14 hours in a 0.25-kg rat ($k=0.035$ hours^{-1}/kg$^{-0.25}$). The relatively short $t_{1/2}$ for the change limits the effects of the time-dependent plasma kinetics over the first 1–2 days of dosing (including peak concentrations) and has no effect on longer-term kinetics or steady state. Although the time-dependence of the free fraction in plasma was needed to simulate short-term plasma PFOS kinetics in rats, the physiological mechanism for a dependence of plasma binding on the time following dosing (i.e., not on concentration of PFOS in plasma or some other dose surrogate) has not been established. Elimination of absorbed chemical occurs by biliary excretion and urinary excretion. Transfer from liver to feces (representing excretion following biliary transfer) is represented as a first-order process acting on the free fraction in liver. Excretion in urine is simulated as the balance between transfer from the free fraction to the glomerular filtrate and renal tubular reabsorption, which removes PFOA and PFOS from the glomerular filtrate and returns it to kidney tissue. Renal tubular reabsorption is simulated as a capacity-limited process with parameters T_m (µg/hour/kg body weight), representing the maximum rate of transport, and K_T (µg/L), representing affinity for the transporter (the concentration in the glomerular filtrate at which reabsorptive transport rate

is half of maximum). This representation of renal tubular reabsorption is used to simulate observed sex differences in elimination of PFOA from plasma, which have been attributed to higher reabsorptive capacity in male rats (see Section 3.1.4). Values for the maximum and affinity parameters for PFOA result in higher reabsorptive clearances from the glomerular filtrate (T_m/K_T=4.1) in male rats compared to female rats (T_m/K_T=0.045), and correspondingly lower urinary clearance of PFOA from plasma in male rats. Reabsorption parameters for PFOS are the same in both sexes and result in reabsorptive clearances that are approximately twice that of PFOA in female rats (T_m/K_T=7.2).

The basic rat model was extended to simulate gestation with inclusion of additional compartments representing adipose and mammary tissue in the dam, placenta, and fetus (Figure 3-4); Loccisano et al. 2012b). Transfer of PFOA and PFOS to the fetus is simulated as a flow-limited transfer to the placenta, with first-order exchange between the placenta and the free fraction in fetal plasma. The free fraction in fetal plasma is simulated as a constant fraction for PFOA and PFOS (i.e., no dependence on time as in the adult). Within the fetus, PFOA in the free fraction of plasma exchanges with a single lumped compartment representing the fetal body, which exchanges with PFOA in amniotic fluid. The fetal PFOS model subdivides fetal tissue into brain, liver, and a lumped compartment for other tissues, all of which undergo flow-limited exchanges with the free fraction of PFOS in fetal plasma. Binding of PFOA and PFOS in fetal liver is assumed to be negligible. Differences in the structure of the fetal models for PFOA and PFOS reflect the differences in the availability of data for estimating parameter values for the various compartments (e.g., perfluoroalkyl concentrations in amniotic fluid, liver).

The lactation model extends the dam portion of the gestational model to include milk and pup (Figure 3-5; Loccisano et al. 2012b). Transfer of PFOA to milk occurs through the mammary gland with flow-limited exchange between plasma and mammary tissue and diffusion into milk from mammary tissue. The model also includes transfer from the pup to the dam, which occurs during maternal stimulation of the neonatal pup to induce elimination and during pup grooming. Data on PFOS in mammary tissue of rodents were not available to establish parameters for a mammary tissue compartment; therefore, the mammary tissue compartment was left out of the PFOS model, and transfer of PFOS to milk is simulated as diffusion directly from plasma. The pup model includes compartments representing the free fraction in plasma, liver, kidney, glomerular filtrate, and a lumped compartment representing all other pup tissues. This structure is essentially identical to the nonpregnant rat model (Loccisano et al. 2012a) with a few differences. Absorption from the gastrointestinal tract is assumed to be complete in pups, and binding in pup liver is assumed to be negligible in pups. There are no storage compartments for biliary or glomerular filtrate perfluoroalkyl in the pup model. Sex differences in renal

tubular reabsorption of PFOA are assumed to develop in response to sexual maturation and, therefore, are not present during lactation (i.e., parameter values are allometrically scaled to pup body weight from the male rat values). Reabsorptive transport parameters for PFOS are allometrically scaled from the lactating dam. The liver/plasma partition coefficient for PFOS in the pups was set lower than that in the dam, based on observations in rats. All other parameters for PFOA and PFOS in the pup were the same or allometrically scaled from values for the dam.

Optimization of parameter values and evaluations of the rat models are described in Loccisano et al. (2012a, 2012b). Data sets utilized in developing and evaluating the nonpregnant rat models included single-dose intravenous and gavage studies and short-term feeding studies (Johnson and Ober 1979; Kemper 2003; Kudo et al. 2007; Perkins et al. 2004). Data used in development and evaluation of the gestation and lactation models included data from gestational and/or lactational exposure studies in rats (Chang et al. 2009; Hinderliter et al. 2005; Kuklenyik et al. 2004; Luebker et al. 2002, 2005a, 2005b; Thibodeaux et al. 2003).

Applications for Dosimetry Extrapolation and Risk Assessment. The wealth of data on pharmacokinetics of PFOA and PFOS in rats allowed an extensive evaluation of the rat models for predicting plasma urinary and liver PFOA and PFOS following single intravenous or single and repeated oral dosing. Inclusion of renal tubular reabsorption parameters in the model provided accurate simulations of sex differences in elimination rates of PFOA from plasma and excretion in urine, and differences in rates of elimination of PFOA and PFOS. The gestation model successfully predicted fetal plasma and liver PFOA and PFOS at the end (or near the end) of pregnancy. Consistent with observations, the model predicts higher fetal plasma concentrations and lower fetal liver concentrations of PFOS compared to maternal, and lower internal exposure (plasma concentrations) to PFOA in the fetus compared to maternal (fetal liver data were not available for PFOA). The lactation model successfully predicted PFOA and PFOS in pup plasma following dosing of the dam. Predicted plasma concentrations of PFOA in nursing pups were approximately 10–50% lower than maternal concentrations, whereas maternal and pup concentrations of PFOS were similar. The model could be used to estimate liver doses and corresponding plasma profiles resulting from single or repeated dosing of adult male or female rats, and maternal-fetal and maternal-pup transfer of PFOA and PFOS. The rat model was evaluated with data from a 14-week oral dosing study and has not been tested for longer exposures. Harris and Barton (2008) developed a PBPK model for PFOS in the rat and found that time adjustments that increased renal clearance and decreased the liver-plasma partition coefficient as a function of time and dose improved predictions of plasma and liver PFOS in adult rats exposed for a period of 105 weeks. Although the

Harris and Barton (2008) model is very different from the Loccisano et al. (2012a) model, these results suggest the possibility that clearance of PFOS may be age- and/or dose-dependent in rats. This may reflect age- or dose-related changes in kidney function, including tubular reabsorption or secretion of PFOS.

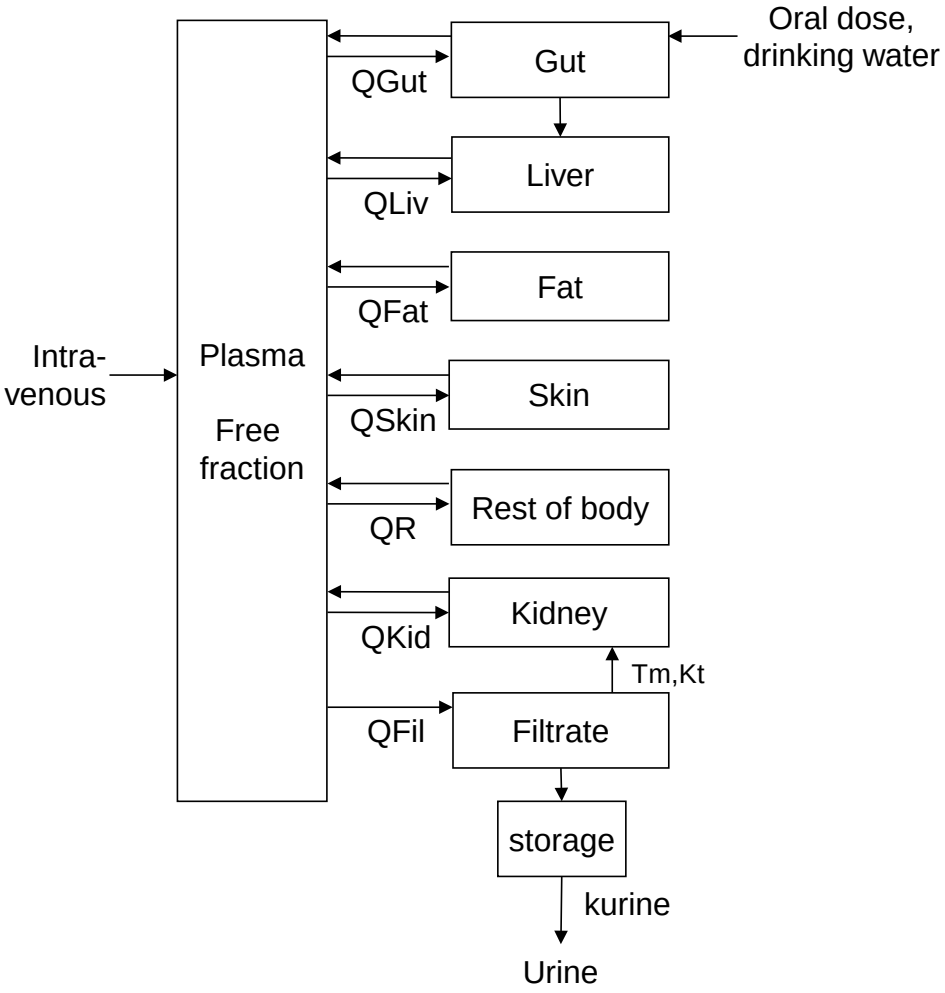
3.1.5.2 Loccisano et al. (2011, 2013) Monkey and Human Models

Loccisano et al. (2011) developed a model for simulating the kinetics of PFOA and PFOS in monkeys and humans. The human model described in Loccisano et al. (2011) was subsequently extended to include simulations of pregnancy and lactation (Loccisano et al. 2013). The monkey model was based, in part, on a multi-compartmental model developed by Tan et al. (2008; Andersen et al. 2006) for simulating the kinetics of plasma and urinary PFOA in monkeys. The structures of the monkey and human models are identical (Figure 3-6) and are very similar to the structure of the rat model (Loccisano et al. 2012a), with inclusion of compartments representing fat and skin, and absence of a storage compartment for biliary transfer. Complete lists of parameters and parameter values and the bases for parameter values and evaluations of model predictions in comparison to observations are reported in Loccisano et al. (2011).

Parameters in the monkey and human models differ in several ways from the rat model. The free fraction in plasma is represented as a constant for both PFOA and PFOS; time-dependency for PFOS in the rat model is absent in the monkey and human models. The parameters for renal tubular reabsorption of PFOA and PFOS are the same for males and females. This is consistent with the absence of evidence for a sex difference in elimination kinetics in monkeys (Butenhoff et al. 2002, 2004a; Seacat et al. 2002).

Values for the affinity constant (K_T) and maximum (T_m) for tubular reabsorption were optimized to plasma concentration kinetics in monkeys. The value for K_T in monkeys was used in the human model. The value for T_m for PFOA in humans was set to yield a plasma elimination $t_{1/2}$ of 2.3 or 3.8 years. The latter two values were derived from estimates of the serum $t_{1/2}$ in populations exposed to PFOA in drinking water (2.3 years; Bartell et al. 2010) or in retired fluorochemical workers (3.8 years; Olsen et al. 2007a). The value for T_m for PFOS in humans was set to yield a plasma elimination $t_{1/2}$ of 5.4 years, based on observations in retired fluorochemical workers (Olsen et al. 2007a). Binding of PFOA and PFOS in the liver was assumed to be negligible in monkeys and humans. Tissue-plasma partition coefficients used in both models were derived from observations in rodents and were the same in the monkey and human models.

Figure 3-6. Structure of PBPK Model for PFOA and PFOS in Monkeys and Humans



Kt = half-saturation constant; kurine = urinary elimination rate; PBPK = physiologically based pharmacokinetic; PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonic acid; QFat = blood flow in and out of fat; Qfil = clearance from plasma to glomerular filtrate; QGut = blood flow in and out of gut; QKid = blood flow in and out of kidney; QLiv = blood flow in and out of liver; QR = blood flow in and out of rest of body; QSkin = blood flow in and out of skin; Tm = transport maximum

Source: Loccisano et al. 2011 (reproduced with permission of Academic Press in the format reuse in a government report via Copyright Clearance Center; Regulatory Toxicology and Pharmacology: RTP by International Society of Regulatory Toxicology and Pharmacology)

Optimization of parameter values and evaluation of the monkey and human models are described in Loccisano et al. (2011). Data sets utilized in developing and evaluating the monkey model included single-dose intravenous and oral studies and repeated-dose oral studies conducted in Cynomolgus monkeys (Butenhoff et al. 2004c; Noker and Gorman 2003; Seacat et al. 2002). Data used in evaluating the human model consisted of serum measurements in people who experienced environmental exposures (Emmett et al. 2006a; Hölzer et al. 2008; Steenland et al. 2009b), adult Red Cross donors (Olsen et al. 2003b, 2008), and retired fluorochemical workers (Olsen et al. 2007a). In general, PFOA and PFOS intakes and exposure durations were not known with certainty in these populations and, as a result, these data do not yield confident evaluations of the ability of the human model to predict intake-plasma level relationships. Follow-up monitoring after a cessation or decrease in exposure can provide data that allow evaluation of the ability of the model to accurately simulate elimination kinetics. Predicted declines in serum PFOA concentrations encompassed observed group mean declines when the T_m for renal tubular reabsorption was set to yield an elimination $t_{1/2}$ of 2.3 or 3.8 years. Group mean declines in serum PFOS were predicted reasonably well for some populations, but not all populations, when the T_m for renal tubular reabsorption was optimized to yield an elimination $t_{1/2}$ of 5.4 years.

The human pregnancy model includes additional compartments representing the free fractions in plasma, amniotic fluid, and a lumped compartment for fetal tissue (Loccisano et al. 2013). The same conceptual approach was used in the rat pregnancy model (Loccisano et al. 2012b, Figure 3-4). Rate constants for placental transfer were initially those from the rat model, adjusted to yield predicated maternal/fetal plasma ratios that agreed with observed maternal/fetal ratios in cord blood (Apelberg et al. 2007b; Fei et al. 2007; Midasch et al. 2007; Washino et al. 2009). Transfers from amniotic fluid to fetus were the same as those used in the rat model, as there were no data on which to base estimates for humans. The lactation model included additional compartments for mammary milk and a lumped compartment representing the infant. Transfer of PFOA to milk is simulated as flow-limited exchange between plasma and milk, governed by mammary tissue blood flow and a milk/plasma partition coefficient. This structure obviated the need to simulate mammary tissue kinetics, for which there were no data in humans. The milk/plasma partition coefficient was calibrated to yield predictions of observed milk/plasma ratios (Fromme et al. 2010; Kärrman et al. 2007). Transfer from maternal milk to infants is the product of the milk concentration and milk production rate (assumed to be equal to sucking rate). The pregnancy model was evaluated by comparing predicted maternal/fetal plasma ratios for PFOA and PFOS with observations from various human monitoring studies (Fei et al. 2007; Fromme et al. 2010; Hanssen et al. 2010; Inoue et al. 2004; Kim et al. 2011; Midasch et al. 2007; Monroy et al. 2008; Tittlemier et al. 2004). The lactation model was evaluated by comparing predicted maternal plasma/milk ratios for PFOA and PFOS

with observations from various human monitoring studies (Fromme et al. 2009; Kärrman et al. 2007; Liu et al. 2011). In general, most model predictions were within plus or minus 2-fold of observations.

Applications for Dosimetry Extrapolation and Risk Assessment. The model predicts plasma concentrations and tissue levels of PFOA and PFOS following intravenous or oral dosing. A skin compartment is included in the model, which may serve for simulating absorption and distribution following deposition onto the skin surface; however, the dermal absorption model was not evaluated in Loccisano et al. (2011). The human model was calibrated to predict $t_{1/2}$ values estimated for human populations (e.g., 2.3 or 3.8 years for PFOA, 5.4 years for PFOS). As a result, comparisons made between observed and predicted serum concentrations evaluate whether or not the populations actually exhibit the $t_{1/2}$ to which the model was calibrated, and not the validity of the model to predict the internal distribution of PFOA or PFOS. It is not currently possible to assess with confidence whether the human model can accurately predict doses to liver or any other tissues. Fábrega et al. (2014) applied the human adult model to estimate plasma concentrations and tissue levels of PFOA and PFOS in human autopsy samples. Exposure inputs to the model were intakes of PFOA and PFOS estimated from public water supply concentrations in the local area where the subjects had resided (Catalonia, Spain) and concentrations in local market basket foods (Domingo et al. 2012a, 2012b). The human model predicted levels of PFOA in plasma and liver that were approximately 10- and 5-fold higher, respectively, than observed. Predicted plasma levels of PFOS were approximately 2-fold higher than observed, and predicted levels of PFOS in kidney were approximately 25% of observed. Fábrega et al. (2014) explored alternative values for tissue/plasma partition coefficients, determined from human autopsy issues (Maestri et al. 2006). The adjusted partition coefficients improved predictions of observed tissue PFOA and PFOS levels. Although the model could be applied to predicting plasma concentrations of PFOA and PFOS or intakes associated with specific plasma concentrations (e.g., oral MRLs), it is not clear what advantages the model offers over simpler empirical or compartmental models similarly calibrated to predict the serum $t_{1/2}$. The monkey model has been more thoroughly evaluated for predicting plasma and urinary kinetics of PFOA and PFOS. This was possible because of the availability of more extensive experimental data on plasma and urine PFOA and PFOS following intravenous and oral (single and repeated) dosing in male and female monkeys. Nevertheless, data on internal distribution were not available to allow evaluation of how well the monkey model predicts doses to the liver or other tissues. Predictions of plasma PFOA and PFOS concentrations from the monkey (and human) model were highly sensitive to values assigned to the maximum rate for tubular reabsorption (T_m) and other parameters that govern urinary elimination of PFOA and PFOS (e.g., free fraction in plasma and glomerular filtration rate; Loccisano et al. 2011). Optimization of the monkey models relied heavily on adjusting these same parameters and, for the human

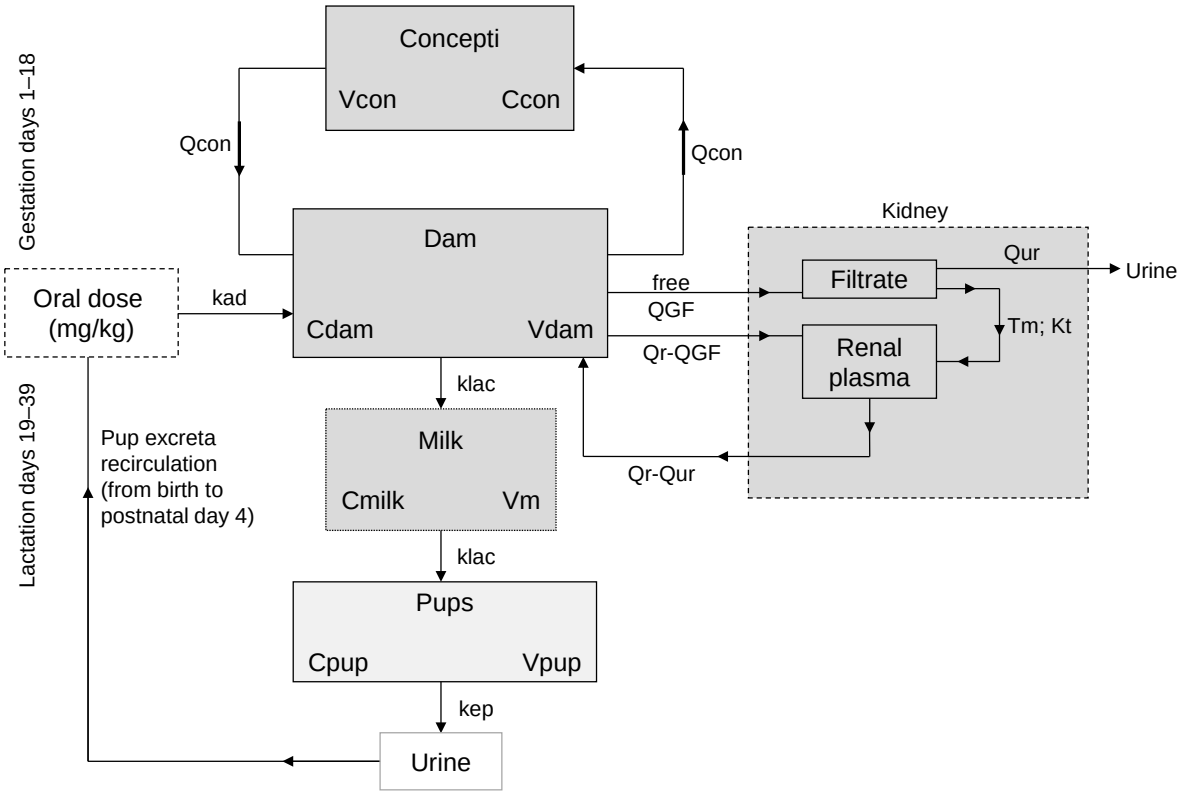
model, the target plasma elimination $t_{1/2}$ was achieved solely by adjusting T_m. Thus, despite the complexity of the models, their potential to accurately predict plasma elimination kinetics and, therefore, steady-state plasma concentrations and associated oral intakes, depends largely on how well they predict plasma clearance. If plasma clearance and the free-fraction in plasma can be reliably predicted empirically for the animal species of interest, then far simpler compartmental models can be used for dosimetry extrapolation of steady-state free plasma concentrations.

3.1.5.3 Rodriguez et al. (2009) Mouse Model

Rodriguez et al. (2009) developed a model for simulating the maternal-fetal and maternal-pup kinetics of PFOA in mice. The general structure of the model is depicted in Figure 3-7. Complete lists of parameters and parameter values and the bases for parameter values and evaluations of model predictions in comparison to observations are reported in Rodriguez et al. (2009). The maternal, fetal, and pup systems are simulated as single well-mixed compartments. Absorption from the gastrointestinal tract is simulated as first-order with complete absorption of the ingested dose. Elimination of absorbed PFOA from the maternal system is simulated as the balance between glomerular filtration and renal tubular reabsorption. The latter is represented as a saturable process with parameters T_m and K_T. Transfer to the fetus is flow-limited and governed by a fetus/maternal partition coefficient and placental blood flow. Transfer from the maternal system to the pup by lactation is simulated as first-order governed by a lactation transfer rate constant. Elimination of PFOA from the pup is first-order to urine. Data sets utilized in developing and evaluating the mouse model included oral gestational dosing studies.

Applications for Dosimetry Extrapolation and Risk Assessment. The model predicted observed concentrations of PFOA in maternal, fetal, and pup serum following oral gestational exposures to mice (Abbott et al. 2007; Lau et al. 2006; White et al. 2007). Residuals for predictions are presented, which provide a quantitative measure of how well the model predicted observations (Rodriguez et al. 2009). Similar to the rat, the mouse model predicts higher internal exposure (serum PFOA concentrations) in the maternal system compared to the fetus. It also predicts accelerated loss of PFOA from the maternal system during lactation. The model simulates the maternal, fetal, and pup systems as single compartments. Although this serves for simulating plasma concentrations (the main objective of the modeling effort), it does not allow for simulation of tissue levels of PFOA in the maternal system, fetus, or pup.

Figure 3-7. Renal Resorption Pharmacokinetic Model of Gestation and Lactation used in the Analysis of CD-1 Mice



Ccon = concentration in concepti; Cdam = concentration in dam; Cmilk = concentration in milk; Cpup = concentration in pup; kad = first-order absorption rate; kep = urinary excretion rate; klac = transfer rate via milk; Kt = half-saturation constant; Qcon = blood flow to and from placenta; QGF = glomerular filtrate; Qr = renal plasma flow; Qur = urine flow; Tm = transport maximum; Vcon = volume in concepti; Vdam = volume in dam; Vmilk = volume in milk; Vpup = volume in pup

Source: Rodriguez et al. 2009 (reproduced with permission of Elsevier Inc. in the format reuse in a government report via Copyright Clearance Center; Reproductive Toxicology by Reproductive Toxicology Center; Washington, DC)

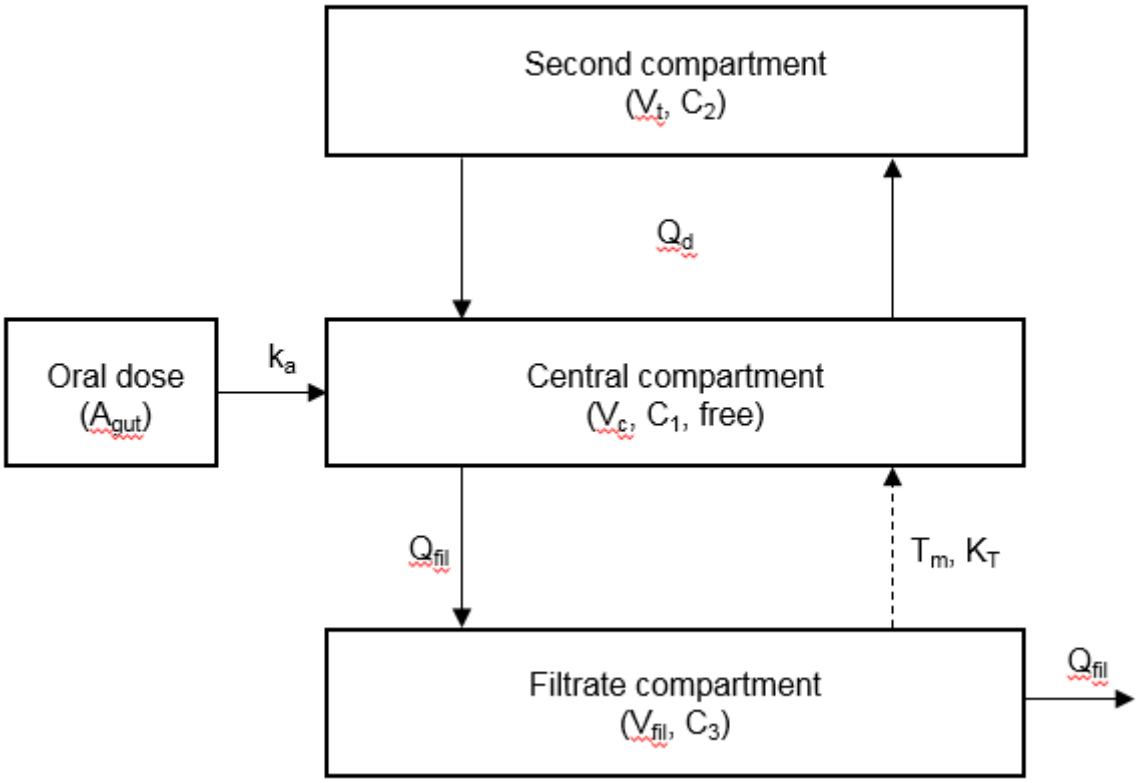
3.1.5.4. Wambaugh et al. 2013 (Andersen et al. 2006) Model

The Wambaugh et al. (2013) model is a three-compartment model based on the three-compartmental monkey model of Andersen et al. (2006). The structure of the two models are identical (Figure 3-8). Parameter values for the Wambaugh et al. (2013) model are presented in Table 3-7. The model includes a central compartment, a secondary distribution compartment, and a renal glomerular filtrate compartment. The central compartment (C1), which includes plasma, receives PFOA or PFOS from oral dosing (first-order k_a, $hour^{-1}$) and exchanges perfluoroalkyl with the secondary compartment (C2, which lumps all other tissues and distribution volumes into a single compartment) and with the glomerular filtrate (C3). A fraction of the perfluoroalkyl in C1 is free (Free) and available for exchange with C2 and C3. Exchanges between C1 and C2 are first order (k_{12}, k_{21}, $hour^{-1}$) with k_{21} assigned a value equal to the $R_{V2/V1}$, where $R_{V2/V1}$ is the ratio of the volumes of the two compartments (V_2/V_1). Transfer of perfluoroalkyl into the glomerular filtrate is first order and governed by the glomerular filtration rate (Q_{filc}, L/hour). Transfer for perfluoroalkyl from the glomerular filtrate to C1 (representing renal tubular reabsorption) is capacity limited (T_{maxc}, µmol/hr; K_T, µM). Perfluoroalkyl that is not reabsorbed is excreted.

Parameter values for the various species and strains were estimated from experimental pharmacokinetic data for each species and strain using Bayesian Markov Chain Monte Carlo (MCMC) analysis. Studies that provided data used to estimate parameter values are listed in Wambaugh et al. (2013). The parameter values shown in Table 3-7 are the mean values and posterior distributions (95% credible interval) from the MCMC analyses.

Applications for Dosimetry Extrapolation and Risk Assessment. Wambaugh et al. (2013) applied the model to predicting internal doses (mean and maximum serum concentrations and plasma AUC) for Benchmark Dose Software (BMDS) modeling and for comparing internal dosimetry from *in vivo* toxicity studies to estimates of potency (AC_{50}, maximum Efficacy) from *in vitro* studies. EPA applied the Wambaugh et al. (2013) model to deriving chronic oral reference doses (RfDs) for PFOA and PFOS (EPA 2016e, 2016f). The model was used to predict internal doses (time-integrated plasma PFOA or PFOS concentrations) achieved in toxicity studies conducted in various laboratory animal models (CD-1 mouse, C57Bl/6 mouse, Sprague-Dawley rat, Cynomolgus monkey). Plasma concentrations were then extrapolated to equivalent steady-state concentrations in humans using a model of first-order elimination of PFOA and PFOS from plasma. The same approach was used to derive MRLs for PFOA and PFOS (see Appendix A).

Figure 3-8. Andersen et al. (2006) Pharmacokinetic Model with Oral Absorption



A_{gut} is the amount of chemical in the gut; k_a is the first-order rate constant for absorption from the gut; Q_{fil} is the flow through the filtrate compartment; C_1, C_2, and C_3 are the chemical concentrations in the central, second, and filtrate compartments, respectively; V_c, V_t, and V_{fil} are the volumes of distribution of the central, second, and filtrate compartments; free is the free fraction of compound in the central compartment; Q_d is the flow between the central and second compartments; the saturable resorption process from the filtrate back into the central compartment is modeled with Michaelis-Menten kinetics, with a maximum rate $T_{maximum}$ and a half-maximum concentration K_T.

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Table 3-7. Estimated and Assumed Pharmacokinetic Parameters for the Modified Andersen et al. (2006) Model for PFOA and PFOS

		Parameter (units)										
Reference	Species	BW (kg)	Cardiac output[a] (L/hour/kg$^{0.74}$)	K_a (hour^{-1})	V_{cc} (L/kg)	k_{12} (hour^{-1})	$R_{V2:V1}$ (unitless)	T_{maxc} (µmole/hour)	K_T (µM)	Free (unitless)	Q_{filc} (L/hour)	V_{filc} (L/kg)
						PFOA[b]						
Lou et al. (2009)	Mouse: CD1 (F)	0.02	8.68	290 (0.6–73,000)	0.18 (0.16–2.0)	0.021 (3.1x10^{-10} to 3.8x10^{4})	1.07 (0.26–5.84)	4.91 (1.75–2.96)	0.037 (0.0057–0.17)	0.011 (0.0026–0.051	0.077 (0.015–0.58)	9.7x10^{-4} (3.34x10^{-9}–7.21)
Dewitt et al. (unpublished)	Mouse: C57Bl/6 (F)	0.02	8.68	340 (0.53–69,000)	0.17 (0.13–2.3)	0.35 (0.058–52)	53 (11–97)	2.7 (0.95–22)	0.12 (0.033–0.24)	0.034 (0.014–0.17)	0.017 (0.010–0.081)	7.6x10^{-5} (2.7x10^{-10}–6.4)
Kemper (2003)	Rat: Sprague-Dawley (F)	0.20 (0.16–0.23)[c]	12.39	1.7 (1.1–3.1)	0.14 (0.11–0.17)	0.098 (0.039–0.27)	9.2 (3.4–28)	1.1 (0.25–9.6)	1.1 (0.27–4.5)	0.086 (0.031–0.23)	0.039 (0.014–0.13)	2.6x10^{-5} (2.9x10^{-10}–28)
Kemper (2003)	Rat: Sprague-Dawley (M)	0.24 (0.21–0.28)[c]	12.39	1.1 (0.83–1.3)	0.15 (0.13–0.16)	0.028 (0.0096–0.08)	8.4 (3.1–23)	190 (5.5–50,000)	0.092 (3.4x10^{-4}–1.6)	0.08 (0.03–0.22)	0.22 (0.011–58)	0.0082 (1.3x10^{-8}–7.6)
Butenhoff et al. (2004b)	Monkey: Cynomolgus (M/F)	7 (m), 4.5 (f)	19.8	230 (0.27–73,000)	0.4 (0.29–0.55)	0.0011 (2.4x10^{-10} to 3.5x104)	0.98 (0.25–3.8)	3.9 (0.65–9,700)	0.043 (4.3x10^{-5}–0.29	0.01 (0.0026–0.038)	0.15 (0.02–24)	0.0021 (3.3x10^{-9}–6.9)
		Parameter (units)										
Reference	Species	BW[d] (kg)	Cardiac output[e] (L/hour/kg$^{0.74}$)	K_a (hour^{-1})	V_{cc} (L/kg)	k_{12} (hour^{-1})	$R_{V2:V1}$ (unitless)	T_{maxc} (µmol/hour)	K_T (µM)	Free (unitless)	Q_{filc} (L/hour)	V_{filc} (L/kg)
						PFOS						
Chang et al. (2012)	Mouse: CD1 (F)	0.02	8.68	1.16 (0.617–42,400)	0.264 (0.24–0.286)	0.0093 (2.63e-10–38,900)	1.01 (0.251–4.06)	57.9 (0.671–32,000)	0.0109 (1.44x10^{-5}–1.45)	0.00963 (0.00238–0.0372)	0.439 (0.0125–307)	0.00142 (4.4x10^{-10}–6.2)
Chang et al. (2012)	Mouse: CD1 (M)	0.02	8.68	433.4 (0.51–803.8	0.292 (0.268–0.317	2,976 (2.8e-10–4.2e4)	1.29 (0.24–4.09)	1.1e4 (2.1–7.9e4)	381 (2.6x10^{-5}–2,900)	0.012 (0.0024–0.038)	27.59 (0.012–283)	0.51 (3.5x10^{-10}–6.09)
Chang et al. (2012)	Rat: Sprague-Dawley (F)	0.203	12.39	4.65 (3.02–1,980)	0.535 (0.49–0.581)	0.0124 (3.1e-10–46 800)	0.957 (0.238–3.62)	1,930 (4.11–83,400)	9.49 (0.00626–11,100)	0.00807 (0.00203–0.0291)	0.0666 (0.0107–8.95)	0.0185 (8.2x10^{-7}–7.34)
Chang et al. (2012)	Rat: Sprague-Dawley (M)	0.222	12.39	0.836 (0.522–1.51)	0.637 (0.593–0.68)	0.00524 (2.86e-10–43,200)	1.04 (0.256–4.01)	1.34e-06 (1.65e-10–44)	2.45 (4.88x10^{-10}–60 300)	0.00193 (0.000954–0.00249)	0.0122 (0.0101–0.025)	0.000194 (1.48x10^{-9}–5.51)
Seacat et al. (2002) and Chang et al. (2012)	Monkey: Cynomolgus (M/F)	3.42	19.8	132 (0.225–72,100)	0.303 (0.289–0.314)	0.00292 (2.59e-10–34,500)	1.03 (0.256–4.05)	15.5 (0.764–4,680)	0.00594 (2.34 x10^{-5}–0.0941)	0.0101 (0.00265–0.04)	0.198 (0.012–50.5)	0.0534 (1.1x10^{-7}–8.52)

[a]Cardiac outputs obtained from Davies and Morris (1993).
[b]Means and posterior distributions from the Bayesian Markov Chain Monte Carlo (MCMC) analysis (95% credible interval in parentheses) are reported.
[c]Estimated average body weight (BW) for species used except with Kemper (2003) study where individual rat weights were available and assumed to be constant.
[d]Average BW for species: individual-specific BWs.
[e]Cardiac outputs obtained from Davies and Morris (1993).

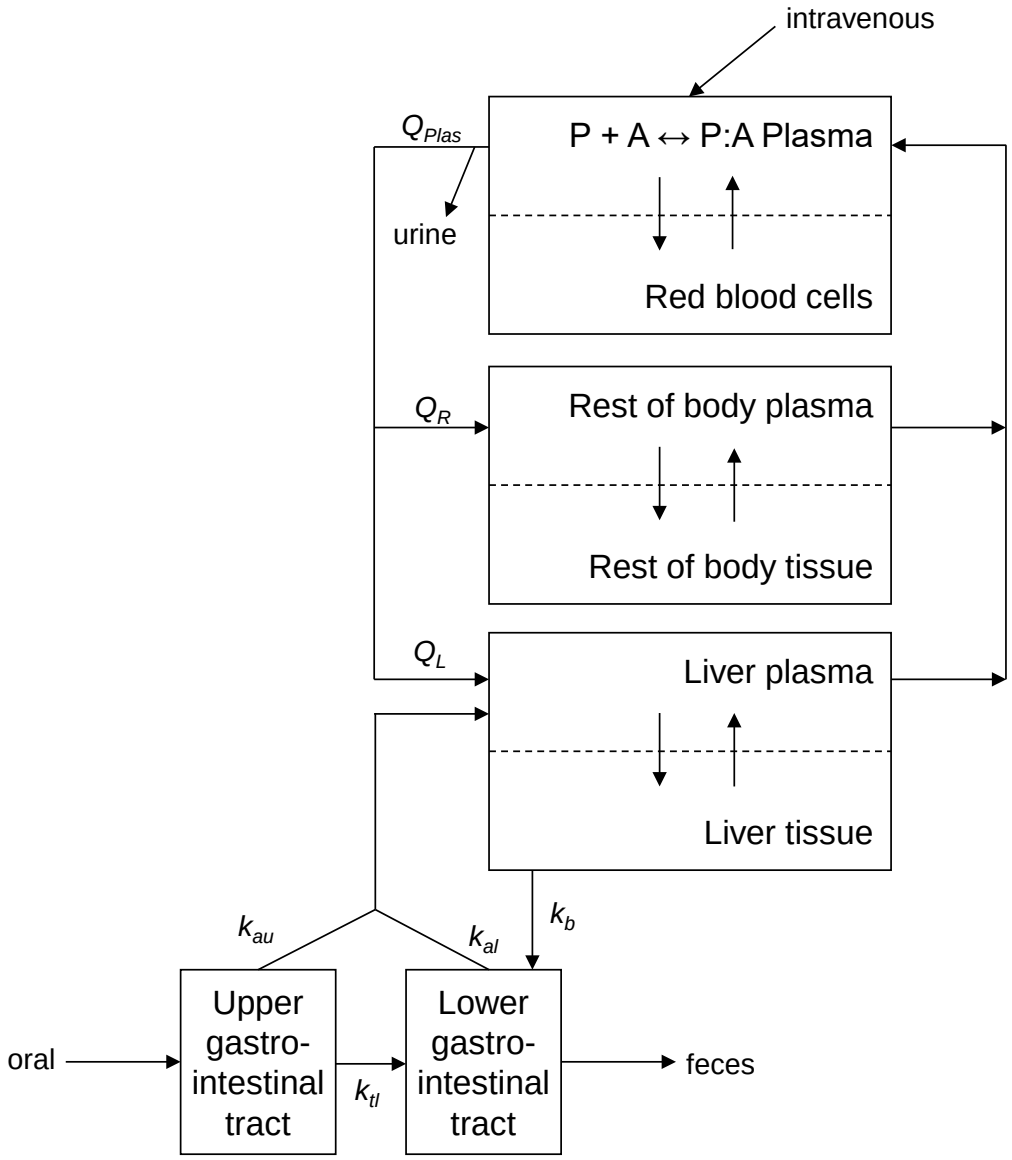
Source: Wambaugh et al. (2013)

3.1.5.5 Harris and Barton (2008) Rat Model

Harris and Barton (2008) developed a model for simulating PFOS kinetics in adult rats. The general structure of the model is depicted in Figure 3-9. Complete lists of parameters and parameter values and the bases for parameter values and evaluations of model predictions in comparison to observations are reported in Harris and Barton (2008). The model includes systemic compartments representing blood (including a bound and free fraction of plasma and red blood cells), liver, and a lumped compartment representing all other tissues. The gastrointestinal tract is simulated as separate compartments representing the upper and lower tracts. Absorption occurs from both the upper and lower tracts, with distinct first-order rate constants assigned to each. Biliary PFOS is transferred from liver to the lower tract. Absorbed PFOS is delivered to the liver where it enters plasma to be distributed to other tissues. Exchanges between PFOS in plasma and all tissues are assumed to be diffusion-limited, with the free pool in plasma participating in the exchange with red blood cells, and the total plasma pool exchanging with liver and all other tissues. Binding of PFOA to plasma albumin is assumed to be saturable, with a dissociation constant 10^{-7} M and a maximum capacity 4.1x 10^{-4} M. This is implemented by assigning bound PFOA to a subcompartment of plasma in which PFOA enters (binds) or exits (unbinds) at rates governed by binding *on* and *off* rates, respectively, that yield a dissociation constant of 10^{-7} M. Elimination of absorbed chemical occurs by biliary excretion and urinary excretion. Transfer from liver to the lower gastrointestinal tract (representing excretion following biliary transfer) is represented as a first-order process acting on the total amount of PFOS in liver. PFOA is transferred to urine from the free fraction of plasma at a rate governed by a urinary clearance parameter, which is assigned a value of 28% of renal plasma flow.

In evaluating performance of the model for simulating PFOS concentrations in a chronic rat feeding study, Harris and Barton (2008) found that the model predicted plasma and liver concentrations measured at 4 and 16 weeks, but over-predicted both at 104 weeks. Performance of the model was improved by having renal clearance increase and the liver/plasma partition coefficient decrease as a function of time (i.e., study duration). These results suggest the possibility that clearance of PFOS may be dependent on age and/or a metric of dose (e.g., cumulative internal dose). This may reflect age- or dose-related changes in kidney function, including tubular reabsorption or secretion of PFOS.

Figure 3-9. Conceptual Representation of a Physiologically Based Pharmacokinetic Model for PFOS Exposure in Rats



k_{al} = rate of absorption from the lower gastrointestinal tract; k_{au} = rate of absorption from the upper gastrointestinal tract; k_b = maximum rate of biliary elimination; k_{tl} = rate of transfer from upper-lower gastrointestinal tract; P:A = PFOS-bound albumin in plasma; PFOS = perfluorooctane sulfonic acid Q_L = plasma flow rate to the liver; Q_{Plas} = plasma flow rate by the heart; Q_R = plasma flow rate to the rest of body

Source: Harris and Barton 2008 (reproduced with permission of Elsevier Ireland Ltd. in the format reuse in a government report via Copyright Clearance Center; Toxicology Letters by European Societies of Toxicology)

Applications for Dosimetry Extrapolation and Risk Assessment. The model simulates kinetics of PFOS following oral or intravenous dosing in adult rats and includes several features that are different from other PBPK models of perfluoroalkyls. The Harris and Barton (2008) model includes a red cell compartment that allows predictions of whole-blood concentrations. The utility of this feature remains to be determined, since PFOS does not appreciably concentrate in red blood cells and PFOS (and other perfluoroalkyls) is typically monitored in the central compartment with measurements of plasma or serum concentrations. The model assumes that the total concentration of PFOS (not just the free concentration) in plasma is available for distribution to liver and other tissues, whereas other models assume that only the free pool in plasma exchanges with tissues. The practical consequence of this difference may not be significant in terms of the toxicokinetics of PFOS if the tissue/plasma partition coefficients in the various models were estimated based on the relevant perfluoroalkyl pool in plasma. However, without basing distribution kinetics on the free concentration, it is not possible for concentration-dependent free fraction to be modeled. The model assumes time-dependence in the liver uptake and urinary excretion of PFOS, which were needed to improve predictions of plasma and liver concentrations of PFOS during chronic exposures. Other rat models (Loccisano et al. 2012a) have not been similarly evaluated. A mechanistic understanding of the time-dependent changes in PFOS kinetics will be important for applications of these models for dosimetry extrapolation across exposure durations.

3.1.5.6 Worley and Fisher (2015a, 2015b) Rat Model

Worley and Fisher (2015a, 2015b) expanded the Loccisano et al. (2012a) adult rat model to include simulation of renal proximal tubule apical (tubule-lumen) and basolateral (tubule-plasma) PFOA transport. This configuration allowed the use of data from *in vitro* studies of kinetics of specific transporters thought to be involved in proximal tubular transport of PFOA in the parametrization of the model. The kidney compartment was expanded to include compartments representing the proximal tubule lumen (glomerular filtrate) and proximal tubule cells. In the model, transfer of PFOA to the tubule lumen is governed by the glomerular filtration rate, represented by a clearance parameter (L/hour/kg kidney). PFOA in the tubule lumen can undergo first-order transfer to urine or saturable transport into the tubule cell (Km, Vmax). PFOA in the tubule exchanges with PFOA in plasma by three mechanisms: saturable transport from plasma into the cell (Km, Vmax), first-order transport from the cell to plasma (kefflux), or bidirectional diffusion between the cell and plasma (kdif). Parameter values (Km, Vmax) for apical and basolateral transport of PFOA were derived from *in vitro* estimates for OATP1a1 (apical) and OAT1 and OAT3 (basolateral) (Nakagawa et al. 2008; Weaver et al. 2010; Yamada et al. 2007). These estimates were scaled to kidney proximal tubule cell mass (Hsu et al. 2014) and the mass-scaled estimates

of Vmax were adjusted with relative activity factors, which were calibrated to *in vivo* observations of plasma PFOA elimination kinetics in rats (Kemper 2003). Values for kefflux (proximal tubule cell to kidney plasma) and kdif (diffusion between kidney plasma and the tubule cell) were also calibrated with *in vivo* data (Kemper 2003; Kudo et al. 2007).

Calibration of the relative activity factor for apical and basolateral membrane transport of PFOA to serum observations made in male and female rats resulted in lower values for activity of both transporters in females compared to males. This resulted in the model predicting lower rates of reabsorptive transfer of filtered PFOA to plasma, and higher renal and systemic (plasma) clearance in females compared to males. Because proximal tubule transporters were assumed to be saturable, the model predicts an increase in clearance with increasing PFOA dose, with larger increases in clearance at lower doses in females compared to males. The model simulated the observed dose-dependent increase in serum clearance (decreasing serum $t_{1/2}$) and higher serum clearance of PFOA (lower $t_{1/2}$) in female rats compared to males (Kemper 2003).

3.1.5.7 Worley et al. (2017b) Human Model

Worley et al. (2017b) scaled and calibrated the Worley and Fisher (2015a, 2015b) rat model to simulate PFOA kinetics in humans exposed to PFOA in drinking water. Physiological parameters were allometrically scaled to the human. Tissue-plasma partition coefficients were derived from human autopsy data (kidney, liver) or studies of distribution of PFOA in rats (Fabrega et al. 2014; Kudo et al. 2007; Perez et al. 2013). Parameter values (Km, Vmax) for apical and basolateral transport of PFOA were derived from *in vitro* estimates for OAT4 (apical) and OAT1 and OAT3 (basolateral) (Nakagawa et al. 2008; Weaver et al. 2010; Yang et al. 2010; Yamada et al. 2007). These estimates were scaled to kidney proximal tubule cell mass (Hsu et al. 2014) and the mass-scaled estimates of Vmax were adjusted with relative activity factors. Parameters that control apical and basolateral transfers of PFOA in the proximal tubule and absorption in the gastrointestinal tract were calibrated against data on serum PFOA concentrations measured in people who drank water from a municipal water supply (Worley et al. 2017b). Model parameter values were adjusted to achieve agreement with geometric mean serum PFOA concentrations measured at two times separated by 6 years. The model was evaluated by comparing predicted and observed serum PFOA concentrations in populations exposed to PFOA in drinking water (Bartell et al. 2010; Emmett et al. 2006b; Steenland et al. 2009a, 2009b). A sensitivity analysis of the model identified that following biokinetic parameters that had standardized sensitivity coefficients >0.1: parameters controlling proximal tubule transport and urinary excretion, plasma-liver partition coefficient,

biliary excretion and protein binding. These parameters, along with drinking water consumption, were assigned probability distributions to conduct a Monte Carlo analysis of predicted serum PFOA predictions associated with exposures to PFOA in drinking water. The probabilistic model simulated interindividual variability in serum PFOA concentrations observed in exposed populations (Bartell et al. 2010; Emmett et al. 2006b; Steenland et al. 2009a, 2009b). These results suggest that that biokinetic variability, as well as exposure variability, may contribute to variability in serum PFOA concentrations observed in populations.

3.1.5.8 Fàbrega et al. (2014, 2016) Human Model

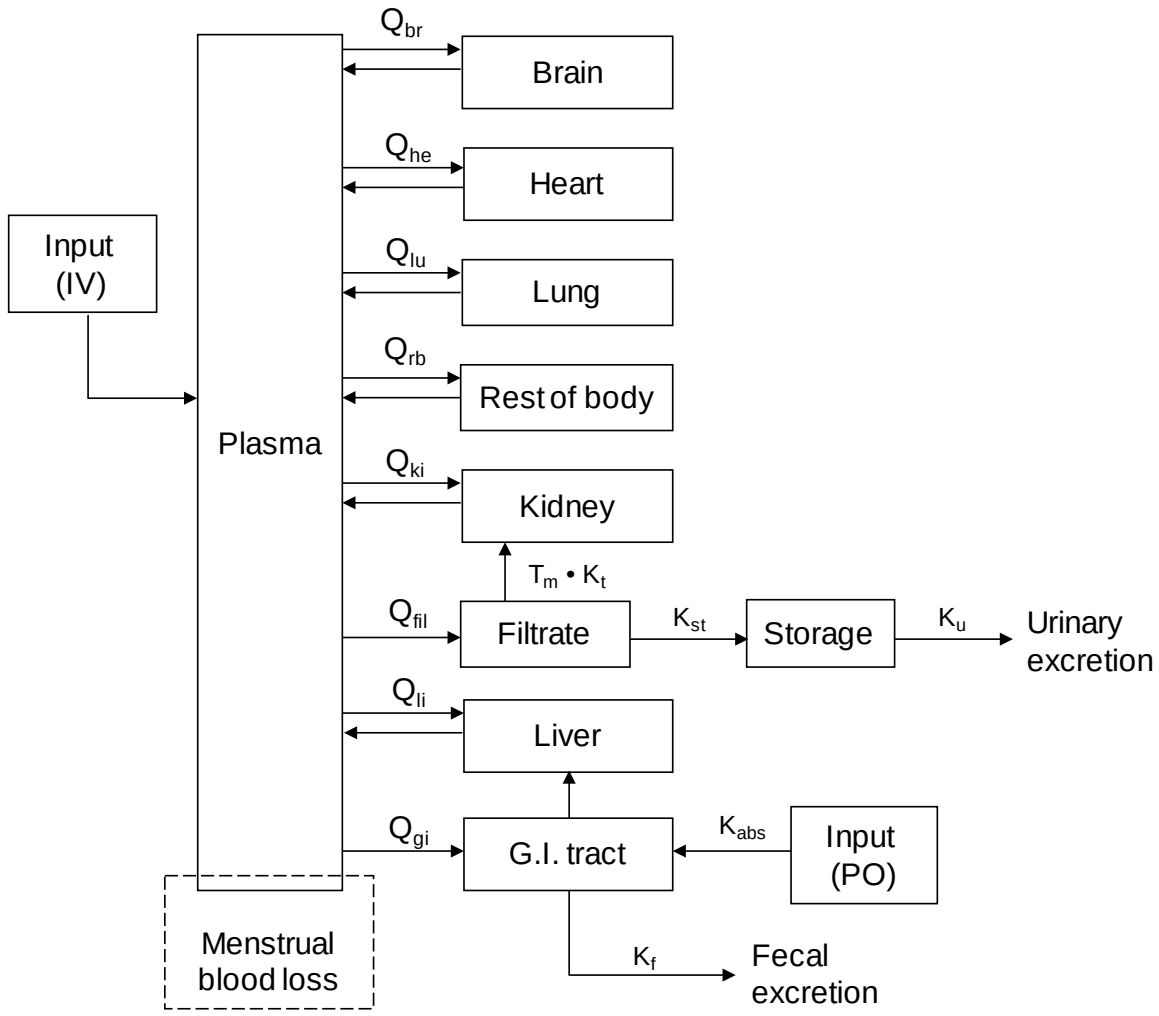
Fàbrega et al. (2014, 2016) modified the Loccisano et al. (2011, 2013) human models for PFOA and PFOS with inclusion of brain and lung compartments and removal of the skin compartment. Tissue-plasma partition coefficients were re-estimated using data from human cadavers (Maestri et al. 2006) in place of estimates based on rat data (Loccisano et al. 2011). The major differences in the partition coefficients for PFOA were lower values for liver in humans (1.03) compared to rats (2.20), higher values for fat in humans (0.47) compared to rats (0.04), and inclusion of partition coefficients for brain (0.17) and lung (1.27). For PFOS, the major differences in the partition coefficients were lower values for liver in humans (2.67) compared to rats (3.72) and higher values for fat in humans (0.33) compared to rats (0.14). Values for parameters that control urinary excretion (Tm and Km for reabsorptive transport from glomerular filtrate to kidney tissue) were recalibrated based on plasma concentration data (Ericson et al. 2007). Fàbrega et al. (2014) compared predictions to observed concentrations of PFOA and PFOS in cadaver samples (from Tarragona County, Spain) for constant intakes of 0.11 µg/day for PFOA or 0.13µg/day for PFOS. Better agreement with observations was achieved with partition coefficients based on cadaver data. Fàbrega et al. (2016) performed a quantitative uncertainty analysis of predictions of tissue PFOA and PFOS concentrations by assigning lognormal probability distributions to renal transport parameters, the unbound fraction in plasma, and intake. Probability distributions for PFOA and PFOS intakes were based on data from Domingo et al. (2012a, 2012b). Distributions for biokinetic parameters were established to achieve a coefficient of variation of 0.3 (Allen et al. 1996; Brochot et al. 2007; Sweeney et al. 2001). Observations of tissue PFOA and PFOS were within uncertainty bounds on predictions.

3.1.5.9 Kim et al. (2018) Rat and Human Model

Kim et al. (2018) developed a model for simulating the kinetics of PFHxS in rats and humans. The structures of the rat and human models are identical (Figure 3-10). Complete lists of parameters and parameter values and the bases for parameter values and evaluations of model predictions in comparison to observations are reported in Kim et al. (2018). The model includes compartments representing plasma (including a bound and free fraction), brain, gastrointestinal tract, heart, lung, kidney and renal glomerular filtrate, liver, and a lumped compartment representing all other tissues. A storage compartment receives PFHxS from the glomerular filtrate and is included in the model to simulate the time delay between elimination from plasma and appearance of PFHxS urine. Absorption from the gastrointestinal tract is simulated as the balance between first-order absorption and fecal excretion of unabsorbed PFHxS. Absorbed PFHxS is assumed to be delivered to the liver. Exchanges between PFHxS in tissues with the free pool in plasma are assumed to be flow-limited (governed by blood flow) with equilibrium determined by the tissue:plasma partition coefficient. Partition coefficients were estimated from the tissue:plasma concentration ratios measured in female and male rats 14 days after a single intravenous dose of PFHxS (0.5–10 mg/kg). Values for each sex were significantly different for brain, lung, liver, spleen, gastrointestinal tract, adipose, and skeletal muscle; in each case, male>female. The highest partition coefficient was in male liver (approximately 0.13), with the value for female being approximately half of the male value. PFHxS in plasma is simulated as instantaneous distributions into free and bound fractions. The free fraction was estimated from ultrafiltration studies of rat and human plasma. The free fraction was assigned a constant of 0.069% in female and 0.076% in male rats.

Elimination of absorbed PFHxS in the rat model occurs by fecal and urinary excretion. Fecal excretion of absorbed PFHxS is represented as flow-limited transfer from plasma to the gastrointestinal tract and first order transfer from the gastrointestinal tract to feces. Excretion in urine is simulated as the balance between transfer from the free fraction of plasma to the glomerular filtrate and renal tubular reabsorption, which removes PFHxS from the glomerular filtrate and returns it to kidney tissue. Renal tubular reabsorption is simulated as a capacity-limited process with parameters T_m (µg/hour), representing the maximum rate of transport, and K_T (µg/L), representing affinity for the transporter (the concentration in the glomerular filtrate at which reabsorptive transport rate is half of maximum). This representation of renal tubular reabsorption is used to simulate observed sex differences in elimination of PFHxS from

Figure 3-10. Structure of the PBPK Model for PFHxS in Rats and Humans*



*PFHxS can be resorbed into the kidney with transporter maximum (T_m) and transporter affinity constant (K_t). Ks indicates a rate constant; K_{st}, the rate constant to the storage compartment; K_u, the urinary elimination rate constant; K_f, the transfer rate constant from the G.I. tract to fecal elimination; and K_{abs}, the oral absorption rate constant. Q_s refers to the blood flows between plasma and tissues, except for Q_{fil}, which is a clearance from the plasma to the filtrate compartment. Menstrual blood loss (dotted square) is only applicable to female humans.

G.I. = gastrointestinal; IV = intravenous; PBPK = physiologically based pharmacokinetic; PFHxS = perfluorohexane sulfonic acid; PO = *per os*

Source: Reprinted by permission from Springer Nature, Kim et al. 2018

plasma, which have been attributed to higher reabsorptive capacity in male rats (see Section 3.1.4). Values for the maximum and affinity parameters for PFHxS result in higher reabsorptive clearances from the glomerular filtrate (T_m/K_T=5.2) in male rats compared to female rats (T_m/K_T=0.057), and correspondingly lower urinary clearance of PFHxS from plasma in male rats. Values for T_m and K_t in humans were assumed to be the same as those in rats. Tissue volumes and blood flows were assigned values based on various sources (Davies and Morris 1993; Igari et al. 1983). Glomerular filtrate volume and flow were assigned values from Loccisano et al. (2012a).

The rat model was calibrated and evaluated against data on plasma and tissue levels of PFHxS measured following a single intravenous (0.5–10 mg/kg) or gavage dose (1 or 4 mg/kg) of PFHxS (Kim et al. 2018). Temporal profiles of plasma PFHxS and cumulative urinary excretion following intravenous or oral dosing were within ±1 SD of observations. Predicted cumulative urinary excretion of PFHxS reproduced the observed sex differences in urinary excretion with slower excretion and higher plasma levels in males compared to females. Terminal levels of PFHxS in heart, kidney, liver, and lung predicted for 14 days following oral dosing were within the range of observed values.

The human model was developed from the rat model with the following attributes:

- Human tissue volumes and blood flows were assigned values based on various sources (Davies and Morris 1993; Igari et al. 1983).
- Glomerular filtrate volume and flow were assigned values from Loccisano et al. (2011).
- Values for the free fraction in human plasma were 0.023% in females and 0.025% in males, based on results from ultrafiltration studies.
- Sex-specific values for renal tubular reabsorption parameters, T_m and K_t, were assumed to be the same in rats and humans.
- First order rate constants were scaled by 0.25 power of body weight ($BW^{0.25}$).
- Loss of PFHxS in menstrual blood was included in the human female model. This is represented as a direct loss of loss of 42.5 mL blood (25 mL plasma) per month (Verner and Longnecker 2015).

Kim et al. (2018) does not report an evaluation of the human model.

Applications for Dosimetry Extrapolation and Risk Assessment. The rat and human models were applied interspecies dosimetry extrapolation of a rat NOAEL for PFHxS (1 mg/kg/day). The rationale for the rat NOAEL is described in Kim et al. (2018). The dosimetry extrapolation was applied to the rat

model to predict a steady-state plasma concentration of PFHxS corresponding to a chronic oral dose of 1 mg/kg/day (value not reported). The equivalent human dose was predicted from the human PBPK model as the daily dose required to achieve the same steady state concentration in the human.

3.1.6 Animal-to-Human Extrapolations

Interspecies differences in the toxicokinetics of perfluoroalkyls and possible differences in the mechanisms of toxicity have been found. The elimination rate for PFOA in female rats is approximately 45 times faster than in male rat, 150 times faster than in Cynomolgus monkeys, and approximately 5,000–9,000 times faster than in humans (Bartell et al. 2010; Butenhoff et al. 2004c; Kemper 2003; Olsen et al. 2007a). Elimination of PFOS in male rats is approximately 3 times faster than in Cynomolgus monkeys and approximately 40 times faster than in humans (Chang et al. 2012; De Silva et al. 2009; Olsen et al. 2007a; Seacat et al. 2002). These large differences in elimination rates imply that similar external PFOA or PFOS dosages (i.e., mg/kg/day) in rats, monkeys, or humans would be expected to result in substantially different steady-state internal doses (i.e., body burdens, serum concentrations) of these compounds in each species. In addition, exposure durations required to achieve steady state would be expected to be much longer in humans than in monkeys or rats. Assuming a terminal elimination $t_{1/2}$ of 1,400 days for PFOA in humans (Olsen et al. 2007a), a constant rate of intake for 17 years would be required to achieve 95% of steady state. Steady state (i.e., 95%) would be achieved in approximately 110 days in monkeys ($t_{1/2}$=25 days, Butenhoff et al. 2004c), 30 days in male rats ($t_{1/2}$=7 days; Kemper 2003), and 1 day in female rats ($t_{1/2}$=0.2 days; Kemper 2003). Using an internal dose metric such as serum perfluoroalkyl concentration and PBPK models that can account for these differences in elimination rates can decrease the uncertainty in extrapolating from animals to humans.

The mode of action for most health outcomes associated with perfluoroalkyl exposure has not been fully characterized in humans or laboratory animals. Some perfluoroalkyl-induced effects observed in rats and mice appear to be mediated through the PPARα-dependent and -independent mechanisms (see Section 2.20 for additional information). Interpretation of the relevance of the effects observed in laboratory animals is complicated since it is generally agreed that humans and nonhuman primates are refractory, or at least less responsive than rodents, to PPARα-mediated effects (Corton et al. 2014; Klaunig et al. 2003; Maloney and Waxman 1999). While studies in mice have identified specific effects that require PPARα activation, for example, postnatal viability (Abbott et al. 2007) and some immunological effects (Yang et al. 2002b), other effects such as hepatomegaly and antigen-specific antibody response (DeWitt et al. 2016) were reported to be PPARα-independent (Yang et al.

2002b). Therefore, further studies are needed to expand the knowledge regarding PPARα-dependent and -independent effects that would allow selection of an appropriate animal model for perfluoroalkyls toxicity. In the absence of data to the contrary, ATSDR assumes that the health effects observed in laboratory animals are relevant to humans. The exception is some of the hepatic effects observed in rodents; increases in liver weight and hepatocellular hypertrophy observed in rats and mice were considered adaptive and not relevant to humans (see Section 2.9 for details).

3.2 CHILDREN AND OTHER POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

This section discusses potential health effects from exposures during the period from conception to maturity at 18 years of age in humans. Potential effects on offspring resulting from exposures of parental germ cells are considered, as well as any indirect effects on the fetus and neonate resulting from maternal exposure during gestation and lactation. Children may be more or less susceptible than adults to health effects from exposure to hazardous substances and the relationship may change with developmental age.

This section also discusses unusually susceptible populations. A susceptible population may exhibit different or enhanced responses to certain chemicals than most persons exposed to the same level of these chemicals in the environment. Factors involved with increased susceptibility may include genetic makeup, age, health and nutritional status, and exposure to other toxic substances (e.g., cigarette smoke). These parameters can reduce detoxification or excretion or compromise organ function.

Populations at greater exposure risk to unusually high exposure levels to perfluoroalkyls are discussed in Section 5.7, Populations with Potentially High Exposures.

The possible association between serum perfluoroalkyl levels in children and health effects has been examined in participants of the C8 Health Project and in the general population. The studies examined a number of health effects including alterations in serum lipid levels, adverse renal outcomes, neurodevelopmental alterations, and reproductive development. Immunotoxicity has been examined in children in several general population studies. Additionally, a large number of studies have examined the possible association of elevated serum perfluoroalkyl levels and adverse birth outcomes.

Similar to adults, associations between serum PFOA and PFOS and serum cholesterol levels were observed in a study of over 12,000 children (Frisbee et al. 2010); an increased risk of high cholesterol was also observed in children with higher serum PFOA and PFOS levels. A smaller study of children (n=43)

living in the Mid-Ohio Valley did not find associations between serum PFOA levels and hematology parameters, total cholesterol and liver enzymes, indices of kidney function, or serum TSH levels (Emmett et al. 2006b). Another study of highly-exposed residents did not find any associations between serum PFOA levels in children aged 6–12 years and IQ, reading and math skills, language, memory, learning, or attention (Stein et al. 2013). Similarly, no association between serum PFOA, PFOS, or PFNA levels in children 5–18 years old and the likelihood of ADHD diagnosis was observed in a study of highly-exposed residents, although the study did find an increased risk associated with higher PFHxS levels (Stein and Savitz 2011). A general population study that utilized the NHANES data found an association between serum PFOA, PFOS, and PFHxS levels and the risk of ADHD diagnosis (as reported by the parent) (Hoffman et al. 2010). Another smaller-scale study found associations between serum PFOS, PFNA, PFDA, PFHxS, and FOSA and impulsivity; no association with PFOA was found (Gump et al. 2011). A study of children 8–18 years of age participating in the C8 studies found reduced odds of reaching puberty at higher serum PFOA levels (Lopez-Espinosa et al. 2011); however, the biological significance of the short delay (4–5 months) is not known.

Several studies have evaluated immunotoxicity in children and adolescents. These studies have found impaired antibody responses associated with serum PFOA, PFOS, PFHxS, and PFDA (Grandjean et al. 2012, 2017; Granum et al. 2013; Mogensen et al. 2015a; Stein et al. 2016a). An increased asthma diagnosis was also associated with serum PFOA levels (Dong et al. 2013; Humblet et al. 2014; Zhu et al. 2016). Marginal evidence of an association with asthma diagnosis was also found for PFOS, PFHxS, PFNA, PFDA, PFBS, and PFDoDA (Dong et al. 2013; Zhu et al. 2016), although some studies found no associations for these compounds (Humblet et al. 2014; Smit et al. 2015; Stein et al. 2016a).

Hines et al. (2009) showed that *in utero* exposure (GDs 1–17) to low levels of PFOA (0.01–0.3 mg/kg/day) resulted in increases in body weight gain in 10–40-week-old mice; by 18 months of age, the body weights in these mice were similar to controls. Increases in serum insulin and leptin levels were also observed in the mice exposed to 0.01 and 0.1 mg/kg/day. The study also compared body weight and body composition of *in utero* exposed mice (exposed on GDs 1–17) and adult exposed mice (exposed for 17 days starting at 8 weeks of age) and found that *in utero* exposure to 1 mg/kg/day resulted in significantly higher body weight, brown fat weight, and white fat weight; this was not observed in mice exposed to 5 mg/kg/day. The results of the study suggest that gestational exposure to low doses of PFOA may result in increased susceptibility to PFOA toxicity.

A number of studies of highly exposed residents and the general population have examined the potential associations between serum perfluoroalkyl levels and alterations in birth weight. Decreases in birth weight have been found to be associated with higher PFOA (Fei et al. 2007; Lee et al. 2013; Maisonet et al. 2012; Savitz et al. 2012b) or PFOS levels (Maisonet et al. 2012), but not with lower levels of perfluoroalkyls (Fei et al. 2007; Hamm et al. 2010; Inoue et al. 2004; Kim et al. 2011; Monroy et al. 2008; Washino et al. 2009; Whitworth et al. 2012b). The decreases in birth weight were small (<20 g or 0.7 ounces per 1 ng/mL). Additionally, no increases in the risk of low birth weight infants were found in highly exposed populations (Darrow et al. 2013; Nolan et al. 2009; Savitz et al. 2012b; Stein et al. 2009). No apparent alterations in the risk of birth defects were found in C8 Health Studies (Darrow et al. 2013; Savitz et al. 2012b; Stein et al. 2009) or in another study of these communities (Nolan et al. 2009).

The developmental toxicity of PFOA and PFOS has been investigated in a number of rat and mouse studies. The observed effects include PFOA- and PFOS-induced increases in prenatal losses and decreases in pup survival, decreases in pup body weight, and neurodevelopmental toxicity (Abbott et al. 2007; Albrecht et al. 2013; Case et al. 2001; Chen et al. 2012b; Era et al. 2009; Fuentes et al. 2006, 2007a, 2007b; Grasty et al. 2003; Hu et al. 2010; Johansson et al. 2008; Lau et al. 2003, 2006; Luebker et al. 2005a, 2005b; Onishchenko et al. 2011; Thibodeaux et al. 2003; White et al. 2007, 2009, 2011; Wolf et al. 2007; Xia et al. 2011; Yahia et al. 2008, 2010). Additionally, delays in mammary gland development were observed in mice exposed to PFOA (Macon et al. 2011; White et al. 2007, 2009, 2011). A limited number of developmental endpoints have been examined in rats and mice exposed to PFDA, PFHxS, or PFBA (Butenhoff et al. 2009a; Das et al. 2008; Harris and Birnbaum 1989; Johansson et al. 2008; Viberg et al. 2013). A more in-depth discussion of the developmental toxicity of perfluoroalkyls in animals is included in Section 2.17.

PFOA and PFOS, as well as other perfluoroalkyls, are valid biomarkers of exposure to these compounds in children, as they are in adults. No relevant studies were located regarding interactions of perfluoroalkyls with other chemicals in children or adults.

No studies examining increased susceptibility to the toxicity of perfluoroalkyls were identified. The available epidemiological data identify several potential targets of toxicity of perfluoroalkyls, and individuals with pre-existing conditions may be unusually susceptible. For example, it appears that exposure to PFOA or PFOS can result in increases in serum lipid levels, particularly cholesterol levels. Thus, an increase in serum cholesterol may result in a greater health impact in individuals with high levels of cholesterol or with other existing cardiovascular risk factors. Associations have been found between

PFOA and PFOS levels and an increased risk of hypertension/pre-eclampsia in pregnant women. The liver has been shown to be a sensitive target in a number of animal species and there is some indication that it is also a target in humans. Therefore, individuals with compromised liver function may represent a susceptible population. Likewise, individuals with a compromised immune system may have an increased risk of perfluoroalkyl-induced immunotoxicity.

3.3 BIOMARKERS OF EXPOSURE AND EFFECT

Biomarkers are broadly defined as indicators signaling events in biologic systems or samples. They have been classified as biomarkers of exposure, biomarkers of effect, and biomarkers of susceptibility (NAS/NRC 1989).

A biomarker of exposure is a xenobiotic substance or its metabolite(s) or the product of an interaction between a xenobiotic agent and some target molecule(s) or cell(s) that is measured within a compartment of an organism (NAS/NRC 1989). The preferred biomarkers of exposure are generally the substance itself, substance-specific metabolites in readily obtainable body fluid(s), or excreta. Biomarkers of exposure to perfluoroalkyls are discussed in Section 3.3.1. The National Report on Human Exposure to Environmental Chemicals provides an ongoing assessment of the exposure of a generalizable sample of the U.S. population to environmental chemicals using biomonitoring (see http://www.cdc.gov/exposurereport/). If available, biomonitoring data for perfluoroalkyls from this report are discussed in Section 5.6, General Population Exposure.

Biomarkers of effect are defined as any measurable biochemical, physiologic, or other alteration within an organism that (depending on magnitude) can be recognized as an established or potential health impairment or disease (NAS/NRC 1989). This definition encompasses biochemical or cellular signals of tissue dysfunction (e.g., increased liver enzyme activity or pathologic changes in female genital epithelial cells), as well as physiologic signs of dysfunction such as increased blood pressure or decreased lung capacity. Note that these markers are not often substance specific. They also may not be directly adverse, but can indicate potential health impairment (e.g., DNA adducts). Biomarkers of effect caused by perfluoroalkyls are discussed in Section 3.3.2.

A biomarker of susceptibility is an indicator of an inherent or acquired limitation of an organism's ability to respond to the challenge of exposure to a specific xenobiotic substance. It can be an intrinsic genetic or other characteristic or a preexisting disease that results in an increase in absorbed dose, a decrease in the

biologically effective dose, or a target tissue response. If biomarkers of susceptibility exist, they are discussed in Section 3.2, Children and Other Populations that are Unusually Susceptible.

3.3.1 Biomarkers of Exposure

Measurement of serum or whole-blood perfluoroalkyl concentrations is the standard accepted biomarker of perfluoroalkyl exposure in humans. Perfluoroalkyls have been detected in the serum of workers, residents living near perfluoroalkyl facilities, and the general population. As part of NHANES, CDC has been measuring serum levels of perfluoroalkyls in the U.S. general population since 1999. Of the 12 perfluoroalkyls examined in this toxicological profile, blood concentrations of 7 compounds (PFOA, PFOS, PFDA, PFHxS, PFNA, and PFUnA) were detected in enough subjects to allow for estimation of the geometric mean. As compared to the general population, serum PFOA and PFOS levels are much higher in individuals with occupational exposure to these compounds (Olsen et al. 2003a; Sakr et al. 2007a) and PFOA levels are much higher in individuals living near a PFOA manufacturing facility (Emmett et al. 2006a; Steenland et al. 2009a), suggesting that serum levels are a good biomarker of exposure. Due to the long half-life of some perfluoroalkyls, particularly PFOA and PFOS, elevated serum levels may not be indicative of recent exposure. Although elevated serum levels are likely to be indicative of exposure to the parent compound, their presence in blood can also indicate exposure to other perfluoroalkyls. For example, PFOS can be derived from metabolism of FOSA (Olsen et al. 2005; Seacat and Luebker 2000). PFOA can be derived from metabolism of 8-2 fluorotelomer alcohol (Fasano et al. 2006; Henderson and Smith 2007; Kudo et al. 2005; Nabb et al. 2007). Exposure of mice to 8–2 telomer alcohol also generated PFNA as a metabolite (Kudo et al. 2005). Most epidemiological studies measured serum perfluoroalkyl levels as a biomarker of exposure. In general, these studies provided a one-time serum perfluoroalkyl level, but lacked information on actual environmental exposure concentrations or doses, route of exposure, and exposure duration. The differences in elimination half-lives between perfluoroalkyls also confounds the interpretation of one-time measurements; the relative concentration of the perfluoroalkyls measured in serum may not be reflective of the actual mixture to which the individual was exposed.

Two studies have also evaluated the use of perfluoroalkyl levels in hair as a biomarker of exposure. In rats administered PFOA, PFOS, or PFNA in the drinking water for 90 days, significant correlations between hair perfluoroalkyl levels and serum and tissue (liver, heart, lung, kidney) levels were found, suggesting that hair perfluoroalkyl levels may be a reliable biomarker of exposure (Gao et al. 2015). A study in humans (Alves et al. 2015) has also found detectable levels of PFBA, PFHxA, PFOA, PFBS, and

PFHxS in hair samples, but PFHpA, PFNA, and PFOS were not detected in hair samples. The study did not evaluate the potential relationship between serum perfluoroalkyl levels and hair levels, which does not allow for an assessment of whether hair is a viable biomarker of exposure.

Urinary perfluoroalkyl levels have also been evaluated as a biomarker of exposure (Worley et al. 2017a). A study of highly exposed residents measured urinary PFOA, PFOS, PFNA, and PFHxS levels. With the exception of PFOA, the proportion of values below the detection limit was too high to calculate mean or median values. The study found a strong linear correlation between serum PFOA levels and urinary PFOA levels in men and a nonsignificant weak correlation between serum and urinary PFOA levels in women.

3.3.2 Biomarkers of Effect

There are no specific biomarkers of effect caused by perfluoroalkyls.

3.4 INTERACTIONS WITH OTHER CHEMICALS

There are limited data on the interactions of perfluoroalkyls with other chemicals. Particularly absent are studies examining toxicological and toxicokinetic interactions of a perfluoroalkyl with other perfluoroalkyls. Olestra decreased the absorption of PFOA from the gastrointestinal tract of mice (Jandacek et al. 2010). No additional information was located regarding interactions among chemicals of this class or between perfluoroalkyls and other chemicals. Both PFOA and PFOS (and many other diverse chemicals) can activate the PPARα, as well as other PPARs to a lesser extent (Takacs and Abbott 2007; Vanden Heuvel et al. 2006). Therefore, it is not unreasonable to speculate that interactions at the receptor level might occur; however, there are no experimental data to support or rule out this presumption. PPARα-independent mechanism are also involved in the toxicity of perfluoroalkyls and interactions between compounds are also likely to influence these mechanisms.