What do laboratory animal studies tell us about the toxicity of PFAS?

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Summary

- PFAS represent a relatively large number of compounds with many commercial applications due to their unique surfactant properties. However, only a small subset of these compounds have been studied for their toxicity using laboratory animal studies.
- Much of what is known about PFAS toxicity is from laboratory animal studies conducted primarily with two PFAS: Perfluorooctanoic acid (PFOA) and perfluorosulfonic acid (PFOS). Additional data are being developed on some substitutes such as perflurobutanoic acid (PFBS) which seems to have a much shorter half-life in animals after exposure.
- PFAS can exhibit remarkably different rates of elimination from the body across species. For this reason, biomonitoring (measures of PFAS in blood) is the preferred way to measure exposures to PFAS compounds and better helps researchers understand differences in toxicity of PFAS compounds in various animal species.
- Laboratory animal studies conducted via the oral route suggest that developmental toxicity, liver toxicity, and possible immune system effects are the main findings at sufficient dosages. Development toxicity is the most sensitive outcome, followed by liver and immune system toxicity. Impaired mammary gland development in offspring occurred at the lowest maternal exposure levels of PFAS tested.
- Developmental toxicity occurs at blood concentrations of PFAS that are almost 1000 times higher than observed among the general human population.
- The human relevance of these animal toxicity findings is yet to be determined.

Background

Poly- and Perfluorinated alkylated substances (PFAS) are a diverse class of man-made chemicals consisting of a structure in which the Hydrogen (H) atoms on the carbon backbone have been partly (i.e., Poly-) or fully (i.e., Per-) replaced with Fluorine (F) atoms. Figure 1 shows the example of Perfluorooctanoic acid (PFOA), a fully fluorinated PFAS and a partly fluorinated equivalent. PFAS resemble naturally occurring fatty acids which only consist of C-H bonds.

The C-F bond is extremely strong and stable and along with its water-repelling (hydrophobicity) and oil-repelling (lipophobic) qualities render these molecules highly useful for industrial surfactant applications, non-stick cookware (trace), carpets, upholstery, paint, floor polishes, semi-conductors, firefighting foams, food packaging, and many others.[1]

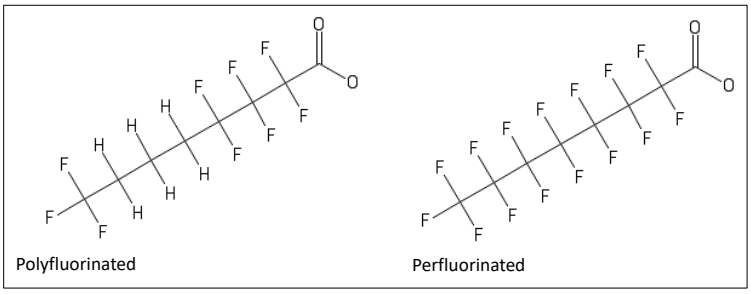

Figure 1: PFOA and a Partly Fluorinated Equivalent Chemical

PFOA and perfluorosulfonic acid (PFOS) are considered “legacy PFAS” (i.e., they are no longer produced) since much of the research has been focused on them and much of the animal toxicity information available for PFAS as a class of compounds are primarily from these two PFAS. Of all the laboratory animal toxicity studies available for PFAS, approximately 50% have been performed with PFOA, 34% with PFOS, and 18% for other PFAS. Based on estimates from the Environmental Protection Agency (EPA), there are more than 600 PFAS currently in commercial use, but only a small subset of these compounds has been studied for their toxicity in laboratory animals.[2]

Toxicity Findings from Laboratory Animal Studies

Animal toxicity studies have been used to study how the body handles PFAS following oral exposures. From the information available for PFOA and PFOS, PFAS are readily absorbed following oral exposure, not metabolized, and their elimination from the body can vary dramatically across animal species and sex, and even life-stage. For example, the elimination half-lives (time for the levels to decrease 50% in plasma) for PFOA range from 3-5 years estimated for humans to 21-30 days in monkeys, 12-20 days in mice, 6 days for the male rat, and as short as 2-4 hours for the female rat. [3,4] Because of these profound differences in clearance, only internal dose metrics such as blood levels are adequate for comparing toxicity effects across different species.

Animal toxicity studies have been carried out at PFAS doses much higher than estimated human exposures in order to identify potential adverse effects and establish dose-response relationships for evaluating such effects at lower exposures. The available studies for PFAS have been mostly carried out by oral exposure (i.e., by adding PFAS to test animal feed or drinking water) at doses which resulted in blood levels of PFAS much higher than those measured in human serum. For example, serum levels of PFOA in mice in the developmental toxicity studies are 2300 ng/mL at the lowest doses tested, as compared to mean serum levels of 4 ng/mL in the general U.S human population.[5] The most relevant toxicity effects to drinking water exposures are those occurring from oral repeated exposures in laboratory animal studies.

The main primary effects observed in laboratory animal studies after oral exposures to PFAS are (1) developmental toxicity, (2) liver toxicity, and (3) immune system toxicity. Not all of these effects have been observed or examined for all PFAS; however, these effects can be considered toxicity hallmarks of this class of compounds since they have been seen in different animal species (rats, mice, and monkeys) and with different PFAS including PFOA, PFOS, and others.[2,5] The effects occurring at the administered repeated oral doses of PFAS most relevant to drinking water exposures are those from animal developmental toxicity studies involving maternal oral exposures during pregnancy and lactation and resulting in adverse effects in the offspring, again, at blood levels almost 1000 times higher than observed among the general human population. Such high exposures are used in

toxicology studies to assure that some effects are observed, regardless of the relevance to humans.

Developmental Toxicity: Developmental effects during pregnancy and lactation are one of the most sensitive toxic effects associated with PFAS exposure. The specific effects in rodent studies include pregnancy loss, reduced pup survival, pup body weight deficits, delayed mammary gland differentiation, and delayed eye opening and sexual maturation.[6,7] These effects notably occurred in the absence of serious maternal toxicity, suggesting that the offspring may be differentially susceptible to the toxic effects of PFAS presumably due to critical windows of development.

Both PFOA and PFOS induced developmental effects in the offspring of rat and mice following maternal oral exposures. However, most studies with PFOA have been conducted with mice because of the relatively short elimination half-life in female rats which would prevent the accumulation expected in other species including humans. Studies in mice with PFOA indicate that exposure during pregnancy or lactation leads to decreases in pup growth, although the effect is greater when both types of exposures occur.[2,5] In the case of PFOS, maternal oral exposure during pregnancy (not lactation) was required for decreased pup weight and survival, suggesting that a particular window of development may be involved. Decreased pup survival appears to be linked to a receptor known as Peroxisome Proliferator Activated Receptor (PPARα).[7] However, the mechanism involving this receptor and its implications to toxicity in animals and potential relevance to humans have yet to be delineated.

Impaired mammary gland development is another effect noted in developmental toxicity studies at the lowest doses of PFOA tested of 0.01 mg/kg body weight. This effect noted in lactating females and female pups has been reported following PFOA exposures during pregnancy and lactation. Notably, no effects on the mammary gland have been reported at much higher doses of PFOA in non-pregnant adult mice, suggesting a development-specific effect. [5,8] Researchers are still trying to understand what the impaired mammary gland in mice means to the overall health of the mice. As this becomes more clear, researchers will be better able to assess how this might translate to humans exposed at thousands of times lower levels of specific PFAS chemicals.

Liver Toxicity: One of the most common effects reported in rodents and monkeys following repeated oral exposures to PFAS is liver enlargement. The liver has been described as a target organ for PFAS deposition and toxicity.[2,3] PFAS such as PFOA seem to deposit preferentially in the liver before distributing to the rest of the body via the general blood circulation. The toxicity manifested as liver enlargement involves enlarged liver cells and proliferation of peroxisomes where fatty acids are metabolized by a process known as fatty acid oxidation. Biochemical alterations resulting in decreases in blood cholesterol and triglycerides have also been reported.[2] These effects on the liver are not surprising since the structure of these molecules resemble fatty acids (where H atoms would be present instead of F atoms), except that there is no metabolism due to the strong C-F bond. In longer term studies with PFOA, the liver effects have been shown to be reversible once exposure stops and the recovery parallels decreases in blood levels. It has been proposed that the liver toxicity involves PPARα which is highly expressed in the rodent liver, but not in monkeys where liver enlargement following oral exposure is also a common effect reported. Notably, PPARα is not highly expressed in humans so most researchers believe this effect is not relevant to humans.[2,3]

Immune System Toxicity: The adverse immunological effects reported in laboratory animal studies following oral exposure to PFAS including PFOA and PFOS can be described as related to immune system suppression and include decreased weights of the spleen and thymus (i.e., atrophy), decreased immunoglobulin response, and changes in the specific population of lymphocytes in the spleen and thymus. Mice appeared to be more sensitive than rats to immune toxicity by PFAS. Atrophy of the spleen and lymph nodes have also been reported in other species such as monkeys.[2,5]

Conclusions

The findings from the available laboratory animal studies for PFOA and PFOS inform that PFAS can exhibit remarkably different elimination rates across species once absorbed into the general blood circulation; therefore, internal dose metrics such as blood levels are most relevant for evaluating the potential toxic effects at lower exposures (i.e., dose-response relationships). The findings from animal toxicity studies at higher doses consist primarily

of developmental toxicity, liver toxicity and immunotoxicity. Developmental toxicity effects are reported as the most sensitive with delays in mammary gland development occurring at the lowest administered daily doses of 0.01 mg/kg body weight of PFOA in rodents. The human relevance of these findings have yet to be determined.

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