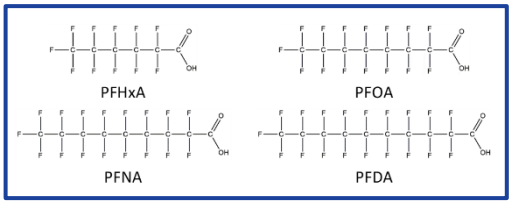
SUMMARY

Background: Per/polyfluorinated alkyl substances (PFAS) are a group of chemicals used in the manufacturing of a variety of consumer products. Widespread exposure to several PFAS is associated with a variety of toxicities, including liver, immune, and endocrine toxicity. In two companion NTP studies, the effects of 28-day exposure to PFAS were evaluated in male and female rats to identify potential toxicity in humans. The current report evaluated four PFAS: perfluorohexanoic acid (PFHxA), perfluorooctanoic acid (PFOA), perfluorononanoic acid (PFNA), and perfluorodecanoic acid (PFDA). The companion publication (TOX-96) describes the studies of three other PFAS.



Methods: Groups of 10 male and 10 female rats were orally administered PFHxA, PFOA, PFNA, or PFDA in deionized water containing a stabilizing agent by gavage for 28 days. Concentrations of PFAS ranged from 0.156 to 1,000 milligrams (mg)/kilogram (kg) of body weight/day. An additional group of rats was administered 0.625 to 25 mg/kg/day of a peroxisome proliferator-activated receptor alpha (PPARα) agonist, so its effects could be compared to those of the four PFAS. Control animals received deionized water with the stabilizing agent but no PFAS added (0 mg/kg/day). Rats were assessed at the end of the studies for plasma and liver concentrations of each PFAS, body weight, clinical observations, mortality, blood parameters, thyroid hormone levels, and liver expression of PPARα-related genes (*Cyp4a1*, *Acox1*) and constitutive androstane receptor-related genes (*Cyp2b1*, *Cyp2b2*). Liver enzyme activities were evaluated in both sexes, but acyl-CoA oxidase enzyme activity in the liver was evaluated only in males. At the end of the studies, tissues from more than 20 sites were examined for signs of disease.

Results: There was no effect on survival in PFHxA, PFOA, or PFDA rats, but reduced survival of PFNA rats was observed. Lower body weights of male PFHxA, PFOA, PFNA, and PFDA rats and female PFNA and PFDA rats were also observed. Plasma and liver concentrations of PFAS were generally higher in animals administered the longer-chain PFAS, and PFHxA and PFOA females had lower plasma concentrations than did males. Common findings following dosing with PFAS included increased liver weights; increased *Acox1*, *Cyp4a1*, *Cyp2b1*, and *Cyp2b2* expression; and increased acyl-CoA oxidase activity. Clinical chemistry endpoints were altered following dosing with several PFAS, including increased liver enzyme activities, decreased globulin and cholesterol concentrations, and increased bile acids and direct bilirubin concentrations. Male and female PFHxA rats had a decrease in red blood cell mass, with an increase in immature red blood cell counts. In most PFAS, total thyroxine (T4) hormone and free T4 levels were decreased with no compensatory increase in thyroid-stimulating hormone levels. Histopathological findings included hepatocyte hypertrophy (an increase in the size of liver cells), cytoplasmic alteration, and necrosis (cell death); bone marrow hypocellularity (an abnormally low number of cells); atrophy of the thymus (a gland that produces and trains immune cells); and lesions in the nose, testes, and epididymis (a tube where sperm mature). Tests evaluating the potential of the four PFAS to damage DNA were negative, except in male rats administered PFOA.

Conclusions: *Under the conditions of these 28-day studies, oral gavage administration of four PFAS to male and female rats showed that the effects of PFHxA were of lower magnitude (e.g., liver or clinical pathology findings) or not apparent compared to those of PFNA and PFDA. Several of the effects observed in the liver were also observed in animals administered the PPARα agonist, but some PFAS-induced effects observed outside the liver were not observed with the PPARα agonist. These data provide a basis for comparisons across the PFAS class.*