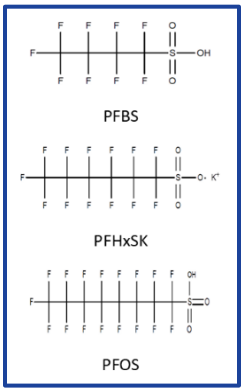
SUMMARY

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 three PFAS: perfluorobutane sulfonic acid (PFBS), perfluorohexane sulfonate potassium salt (PFHxSK), and perfluorooctane sulfonic acid (PFOS). The companion publication (TOX-97) describes the studies of four other PFAS.



Methods: Groups of 10 male and 10 female rats were orally administered PFBS, PFHxSK, or PFOS in deionized water containing a stabilizing agent by gavage for 28 days. Concentrations of PFAS ranged from 0.312 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 known peroxisome proliferator-activated receptor alpha (PPARα) agonist, so its effects could be compared to those of the three 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 PFOS or PFHxSK rats, but reduced survival was observed in PFBS rats. Lower body weights of PFBS rats and, to a lesser extent, PFOS rats were also observed. Plasma and liver concentrations of PFAS were highest in male and female PFOS rats and lowest in male and female PFBS rats. Sex differences in plasma concentrations of PFAS were observed in PFHxSK rats, as male rats showed approximately double PFHxKS concentrations compared to female rats. Common findings that occurred following dosing with PFAS included increased liver weights; increased *Cyp4a1*, *Acox1*, *Cyp2b1*, and *Cyp2b2* expression; and increased acyl-CoA oxidase activity. Several clinical chemistry endpoints were altered in PFBS and PFOS rats, including increased liver enzyme activities; increased bile acid and direct bilirubin concentrations; and decreased globulin, cholesterol, and triglyceride concentrations. Globulin, cholesterol, and triglyceride concentrations were decreased in PFHxSK males, and immature red blood cell counts were decreased in all groups but the PFHxSK females. Histopathological findings included hepatocyte hypertrophy (an increase in the size of liver cells) and/or cytoplasmic alteration, bone marrow hypocellularity (an abnormally low number of cells), and lesions of the nose. Decreases in thyroid hormone levels were present across these chemicals and occurred at almost all doses administered, but thyroid-stimulating hormone levels did not increase in response. Tests evaluating the potential of the three PFAS to damage DNA were mostly negative.

Conclusions: *Under the conditions of these 28-day studies, oral gavage administration of three PFAS to male and female rats showed that the effects of PFHxSK were of lower magnitude (e.g., liver or clinical pathology findings) or not apparent compared to those of PFBS and PFOS. The observed effects corresponded, to some degree, with limited to no increases in liver Acox1 and Cyp gene expression changes. Several of the effects observed in the liver were also observed in rats administered the PPARα agonist, but 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.*