As a member of the CPSC DINP CHAP (2001) and Group Leader of the NIEHS National Toxicology Program’s Peroxisome Proliferation initiative (1995-2006), I became convinced that rodent toxicity data cannot be extrapolated for human risk assessment due to the different responses between rodents and humans resulting from activation of the PPARα nuclear receptor. During the CHAP deliberations, it became clear the mode of action for the hazardous effects of DINP occurred via activation of the PPARα nuclear receptor in rodent models, and that these data would not be relevant for purposes of human risk assessment. Whether PPARα is activated by endogenous fatty acids, environmental xenobiotics such as DINP and metabolites, or by pharmaceutical agents, the biochemical and molecular effects observed in rodents is so different from the effects in humans that the toxic effects of PPARα agonists should be considered to be rodent-specific. The reasons for this conclusion are summarized below and presented in detail in a critical review (Corton et al., 2013) that was, unfortunately, not available prior to the finalization of the Hazard Identification Document (HID) on DINP. Upon exposure to PPARα agonists to rodents, immediate and delayed pleiotropic responses are induced. The immediate responses consist of hepatomegaly, proliferation of peroxisomes in liver parenchymal cells, and the induction of several hepatic enzymes, particularly those responsible for lipid metabolism, and the delayed responses include the development of hepatocellular carcinomas. PPARα agonists consist of a broad spectrum of synthetic and naturally occurring compounds, such as widely used lipid and cholesterol lowering pharmaceutical agents (e.g., clofibrate, nafenopin, ciprofibrate, fenofibrate, gemfibrozil), leukotriene antagonists, phthalate ester plasticizers (e.g. DEHP and DINP), herbicides, solvents, and the naturally occurring steroid dehydroepiandrosterone, among others. The response to PPARα activation in rodents results in increased number and size of liver parenchymal cells, which contain numerous peroxisomes, induction of peroxisomal, mitochondrial and microsomal fatty acid-oxidizing enzymes, including the hydrogen peroxide-generating fatty acyl-CoA oxidase, carnitine acetyl transferase and cytochrome P450 4A isoymes. Peroxisomes in liver parenchymal cells normally occupy <2% of the cytoplasmic volume and in livers with peroxisome proliferation, the peroxisomes may occupy as much as 25% of the cytoplasmic volume. Peroxisome proliferators cause selective induction of H₂O₂—generating enzymes in liver, the organ in which tumors develop following long-term exposure. There are at least three sources of reactive oxygen radical production contributing to oxidative stress. First, peroxisomal fatty acyl-CoA oxidase, the rate limiting enzyme of the classical inducible β-oxidation system increases 20- to 40-fold and has been considered the major source of H₂O₂ in the livers of rats and mice chronically exposed to PPARα agonists.
including DINP. The second source, equally important in magnitude, is the superoxide and \( \text{H}_2\text{O}_2 \)-generating CYP4A subfamily of enzymes that are greatly induced in the livers of rats and mice treated with PPAR\( \alpha \) agonists. The third source is the overall constitutive level of peroxisomal oxidases such as peroxisomal urate oxidase in the livers of rats and mice. In the liver of rodents, there is only about a 2-fold increase in catalase activity, and a disproportionate increase in the levels of \( \text{H}_2\text{O}_2 \)-producing and \( \text{H}_2\text{O}_2 \)-degrading enzymes have been shown to be due to differential regulation of the genes encoding them.

In humans, like rodents, drugs used in the treatment of hyperlipidemia are thought to activate PPAR\( \alpha \) in the liver. However, unlike rodents, activation of PPAR\( \alpha \) in humans does not result in peroxisome proliferation and the concomitant increase in oxidative stress, but instead results in increased apolipoprotein A-II and lipoprotein lipase transcription, and reduced apolipoprotein C-III. These properties are key to the mechanism of action to lower serum triglycerides as well as induction of fatty acid transport protein and acyl-CoA synthetase of cholesterol-lowering pharmaceutical agents. (Apo C-III is a major component of very low density lipoproteins (VLDL) and inhibits lipoprotein lipase and inhibits clearance of lipoproteins by the liver). In humans, the hypolipidemic effects of PPAR\( \alpha \) agonists results from increased transport of lipids from the blood; in rodents, it results in increased catabolism of lipids in peroxisomes and results in oxidative stress.

Rodent-specific toxicity is not uncommon. Indeed, in the HID is a clear description of renal toxicity and carcinogenesis associated with \( \alpha2u \)-globulin nephropathy. It is widely acknowledged that chemicals that meet the IARC criteria for induction of \( \alpha2u \)-globulin nephropathy are likely to be rodent-specific renal carcinogens and should not be considered in human risk assessments. From the data presented above, I believe that DINP activation of PPAR\( \alpha \) produces rodent-specific liver tumors that are irrelevant for human risk assessment.

In summary, activation of PPAR\( \alpha \) in rodents by endogenous substrates, environmental xenobiotics or pharmaceutical agents produces genotypic and phenotypic changes resulting in increased fatty acid catabolism and increased oxidative stress ultimately leading to liver tumors. Activation of PPAR\( \alpha \) in humans induces lipid transport systems that do not result in adverse effects and are not associated with liver tumorigenesis. Therefore, liver tumors in rodents induced by PPAR\( \alpha \) agonists are not relevant for human risk assessment due to the widely different responses induced by these agents in rodents and humans.