

June 3, 2014

Dr. Lori White
Designated Federal Officer for the BSC
Office of Liaison, Policy and Review
Division of NTP, NIEHS,
P.O. Box 12233, K2-03
Research Triangle Park, NC 27709.



PETA

PEOPLE FOR THE ETHICAL
TREATMENT OF ANIMALS

HEADQUARTERS
501 FRONT STREET
NORFOLK, VA 23510
TEL 757-622-PETA
FAX 757-622-0457

Dear Dr. White,

These comments on substance nominations to the National Toxicology Program (NTP) and adverse outcome pathways are submitted on behalf of People for the Ethical Treatment of Animals (PETA) and our more than three million members and supporters. PETA is committed to using the best available science to save animals from suffering in laboratory experiments and promote the acceptance of human-relevant methods for risk assessment.

We welcome the discussion of Tox21 assessments and the proposed development of systems to evaluate volatile chemicals *in vitro*. However, relevant data were overlooked in each of the Research Concepts discussed below. These include data submitted or to be developed under the European Commission Regulation on Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) as well as data evaluated or requested by NTP member agencies in their assessments. We appreciate the potential difficulty in accessing REACH data, but an effort must be made in order to avoid duplicative testing. Likewise, past and present assessments by NTP member agencies must be thoroughly considered in order to ensure that proposed NTP research complements rather than duplicates these efforts.

Bisphenol S

In her Research Concept for bisphenol S (BPS), Vicki Sutherland proposes ADME/TK characterization in rodents by oral and intravenous routes of exposure as well as short-term and subchronic toxicity studies including the perinatal exposure window to assess potential for reproductive toxicity, teratogenicity, and neurotoxicity. Based on NTP study protocols, approximately 500 animals would be killed in these studies.

Sutherland notes that BPS has been assessed in a number of *in vitro* assays as part of NTP's Tox21 High Throughput Screening Program (HTS) and was classified as an estrogen agonist with some affinity for the estrogen receptor. Similar findings have been reported by Grignard et al. (2012) and Kuruto-Niwa et al. (2004); both groups reported that the estrogenic activity of BPS was comparable to that of BPA in transactivation assays. Considering the structural similarity of these chemicals, such findings are not surprising, and the strategy of replacing BPA with BPS is questionable.

BPS is registered under REACH. The dossier includes summaries of screening-level repeated dose and reproduction/developmental toxicity studies. Nephrotoxicity was reported in the 28-day oral gavage study in rats; a 90-day study is planned. In the reproduction/developmental study, no abnormalities were reported with regard to copulation, parturition, delivery, number of corpora lutea, gestation period, parturition status and lactation behavior. However, extended estrus cycle, increased number of animals showing irregular estrus cycle, tendency to decreased number of implantation sites, and significantly decreased implantation were reported. No effects were reported with respect to growth, sex ratio, body weight, viability or anogenital distance of offspring; a full developmental toxicity study is planned.

NTP must consider these data and attempt to coordinate its research program with that of the REACH registrant prior to the initiation of potentially duplicative studies.

References

Grignard E, Lapenna S, Bremer S. Weak estrogenic transcriptional activities of Bisphenol A and Bisphenol S. *Toxicol In Vitro*. 2012 Aug;26(5):727-31.

Kuruto-Niwa R1, Nozawa R, Miyakoshi T, Shiozawa T, Terao Y. Estrogenic activity of alkylphenols, bisphenol S, and their chlorinated derivatives using a GFP expression system. *Environ Toxicol Pharmacol*. 2005 Jan;19(1):121-30

Triclocarban

In her Research Concept for triclocarban (TCC), Sutherland cites an FDA proposed rule on the safety and effectiveness of active ingredients in consumer antiseptic washes, including TCC. This is discussed only briefly in regard to human absorption studies. However, Sutherland's research proposal overlaps several of the data gaps identified by FDA, including ADME and DART studies. Data submitted in response to FDA's proposed rule may, therefore, obviate the need for these studies and must be thoroughly reviewed prior to their commencement.

FDA's proposed rule includes a discussion of available human pharmacokinetics, human and animal ADME, carcinogenicity, and hormonal effects data on TCC. In 2009, EPA published a Risk-Based Prioritization of TCC, which was sponsored under the High Production Volume Chemicals Challenge Program. Regarding a three-generation reproductive toxicity study in rats to which Sutherland refers, EPA concluded that TCC showed no systemic, developmental or reproductive toxicity. In addition, both FDA and EPA summarize a carcinogenicity study in rats that is not referenced by Sutherland. From this study, EPA concluded that there was low systemic toxicity and no evidence of increased tumor incidence at any site. FDA noted that while no carcinogenicity findings were seen in this study, male rats exhibited sex organ toxicity.

NTP must review all existing data and coordinate its efforts with those of FDA. In addition, FDA notes that the record does not currently contain sufficient data to show any

additional benefit from the use of consumer antiseptic washes compared to non-antibacterial soap and water. If TCC cannot be shown to be effective, then it may be excluded from these products, which would greatly reduce exposure and concern. Our comments to FDA will recommend that efficacy be demonstrated prior to initiating new safety testing in animals.

C9 Alkylbenzenes

In his Research Concept for C9 alkylbenzenes, Brian Sayers proposes a comprehensive evaluation of toxicity and carcinogenicity for representative trimethylbenzene (TMB) and ethyltoluene (ET) isomers. This evaluation would include reproductive and developmental endpoints. Additional short-term inhalation toxicity studies would be conducted on C9 alkylbenzenes not selected for long-term studies. Based on NTP study protocols, approximately 8,000 animals would be killed in these studies.

Sayers notes that information on mixtures of C9 alkylbenzenes cannot be used to derive safe exposure levels for individual compounds. Both 1,2,4-TMB and 1,3,5-TMB are registered under REACH, and their dossiers include reports of recent prenatal developmental toxicity studies (OECD Guideline 414) by the inhalation route of exposure on test materials of 99% purity. For 1,2,4-TMB, pregnant rats were exposed whole body to concentrations of 0, 100, 300, 600, and 900 ppm. No evidence of embryo-lethal or teratogenic effects was found. Significant decreases in maternal body weight gain and food consumption and reduction in fetal body weight were observed at concentrations of 600 ppm or greater. For 1,3,5-TMB, pregnant rats were exposed whole body to concentrations of 100, 300, 600 and 1200 ppm. Again, no evidence of embryo-lethal or teratogenic effects was found. Significant decreases in maternal body weight gain and food consumption were observed at concentrations of 300 ppm or greater; significant reduction in fetal body weight occurred at 600 and 1200 ppm. These existing data must be considered prior to the initiation of potentially duplicative studies

We appreciate that the capability for *in vitro* assessment of the C9 fraction isomers will be evaluated; however, we question the proposal to do so in parallel to the proposed *in vivo* studies. As Sayers notes, systems designed to generate atmospheres for exposing cells to volatile chemicals *in vitro* already exist, and target tissues can be selected based on the current literature. Therefore, *in vitro* assessment should precede the proposed *in vivo* studies.

Adverse Outcome Pathways

We support a role for the NTP Interagency Center for the Evaluation of Alternative Toxicological Methods (NICEATM) as articulated; that is, forming a bridge between the OECD AOPs program and application to US agencies. For skin sensitizers in particular, PETA supports the development and implementation of an integrated decision strategy that draws upon all sources of information derived from computational and *in vitro* toxicological methods to support hazard classification. We encourage NICEATM to fast-track this effort due to the abundance of information now available on the performance of

these methods and their scrutiny and validation by OECD. The U.S. should coordinate its efforts to develop and accept a skin sensitization AOP model with the work being led by OECD.

Thank you for your attention to these comments. I can be reached at 757-622-7382, ext. 8001 or via email at JosephM@peta.org.

Sincerely,
[Redacted]

Joseph Manuppello, MS
Research Associate
Regulatory Testing Division
People for the Ethical Treatment of Animals