



May 2, 2007

Delivered Via Email

Barbara Shane, Ph.D.
Executive Secretary
National Toxicology Program Board
NTP Liason and Scientific Review Office
NIEHS
P. O. Box 12233
MD A3-01
Research Triangle Park, NC 27709

Re: Public Comment Addressing NTP's Technical Report on the Toxicology and Carcinogenesis Studies of Cresols in Male F344/N Rats and Female B6C3F1 Mice (NTP TR 550).

Dear Dr. Shane:

The Cresols Panel of the American Chemistry Council wishes to comment on the draft NTP Technical Report on the Toxicology and Carcinogenesis Studies of Cresols in Male F344/N Rats and Female B6C3F1 Mice (NTP TR 550) (Draft Technical Report). The Cresols Panel consists of representatives from the following five companies: Dakota Gasification Company, Degussa Corporation, LANXESS Corporation, Merisol USA LLC, and Sumitomo Chemical America Inc.

The Panel's comments focus on two areas of NTP's Draft Technical Report on cresols: (1) reproduction and developmental toxicity; and (2) rodent forestomach tumors.

First, the Panel would like to point out that in addition to the studies referenced in the Draft Technical Report for reproductive and developmental toxicity, rat and rabbit developmental toxicity and rat two-generation reproductive function studies are available for each cresol isomer. These studies, which are not referenced in the Draft Technical Report, were completed in 1988 and 1989 as part of an industry effort to comply with a TSCA Section 4 test rule issued on cresols. There were nine bioassays, each used oral gavage as the route of exposure, and each was GLP-compliant and followed U.S. Environmental Protection Agency (EPA) Office of Prevention, Pesticides and Toxic

Substances (OPPTS) guideline protocols. A full report for each study was submitted to EPA. The citations for those studies are included in the references provided with this letter.

Second, the Panel urges that NTP consider the relevance of rodent forestomach tumors for human cancer in the presentation of results. Mouse forestomach squamous papillomas are reported in the NTP Draft Technical Report. The Cresols Panel suggests that to assist readers of the Draft Technical Report with understanding the importance of this lesion to human cancer, NTP should consider adding additional explanation of the relevance to human health of these specific rodent tumors.

As an example, context for these mouse lesions to human cancer has been provided by the International Agency for Research on Cancer (IARC) in its technical publication titled Predictive Value of Rodent Forestomach and Gastric Neuroendocrine Tumours in Evaluating Carcinogenic Risks to Humans (IARC, 2003). In that technical document, IARC advises that in evaluating the relevance of rodent forestomach tumors (for human cancer) test exposure conditions be considered, specifically conditions that lead to high local concentrations of the test substance in the rodent forestomach. Such conditions favor prolonged exposure of forestomach epithelial tissue and “may contribute to responses that may be unique to the forestomach” (page 13). With this exposure, according to IARC, non-DNA reactive agents appear to cause squamous epithelial papillomas of the forestomach through initial cytotoxicity and subsequent sustained cell proliferation and hyperplasia (page 12). More precisely, Erik Dybing and Tore Sanner writing in the IARC technical publication (page 23), state that *Salmonella*-negative rodent forestomach carcinogens can have promotional (tumorigenic) effects by inducing a local proliferative response of the forestomach epithelium, which is accompanied by inflammation, ulceration and erosion of the epithelium. In the Summary Report for the document, IARC states that “the relevance (of rodent forestomach papillomas) is probably limited for agents that have no demonstrable genotoxicity and that are solely carcinogenic for the forestomach squamous epithelium in rodents after oral administration, since the exposure conditions are quite different between the experimental animal and humans. Consequently, for these agents, the mode of carcinogenic action could be specific to the experimental animal” (page 12). The Cresols Panel would appreciate NTP including in the Discussion and Conclusions section of the Technical Report language similar to that used by IARC with respect to the relevance of mouse forestomach epithelial papillomas to human cancer.

To support this position, the Cresols Panel points out that important elements of a well-established mode-of-action for these papillomas are common to m-/p-cresols---they are a test material with considerable contact irritation potential and they do not appear to act as genotoxic agents or interact with DNA. Earlier work established that cresols can act as promoters of dermal carcinogens (Boutwell and Bosch, 1959). The mouse forestomach tumors seen in the m-/p-cresols chronic study occurred in the high-dose group only and tumors at other sites did not form in the study.

The cancer endpoint observed by NTP in mice treated with cresols (m-/p-cresol mix) was squamous cell papilloma of the forestomach. This lesion did not occur in the untreated control animals and was reported in 1/50 low-dose mice, 1/49 mid-dose mice and 10/50 high-dose mice. Only female mice were used in cresols testing. The lesion, a benign tumor, is thought not to invade the gastric wall and not metastasize (IARC, 2003).

As pointed out in the NTP Draft Technical Report, the genetic toxicology of cresols has been extensively investigated. Single cresol isomers and mixtures of isomers have been uniformly negative for mutation when tested with *Salmonella typhimurium* with and without exogenous metabolic activation. Higher tier *in vitro* testing was predominantly negative for mutation or DNA interaction and the weight of evidence from *in vivo* genetic toxicity testing is strongly in favor of nongenotoxicity. The NTP Draft Technical Report describes the m-/p-cresol test material as a nongenotoxic irritant (page 63) and the absence of tumors at any other site support a nongenotoxic mechanism of tumorigenicity.

One important element of the mode-of-action for rodent forestomach squamous epithelial cell papilloma formation described in the IARC 2003 technical publication, sustained cell proliferation and hyperplasia, does not appear to be a feature of cresols papilloma production. Sustained proliferation was not reported in the 90-day or two-year rodent bioassays with cresols. Contrary to language in the Discussion And Conclusion section of the Draft Technical Report,¹ m-/p-cresol did produce irritation (minimal) and hyperplasia of the squamous epithelium of the forestomach in 1/5 high-dose animals (30,000 ppm) in the 28-day study and hyperplasia in a small percentage of high-dose mice in the two-year study. The data in the table below are taken from Tables B4 and 11 of the Draft Technical Report. IARC observed, for that matter, that small papillomas may be difficult to distinguish from focal regenerative hyperplasia (page 7).

Lesion	Control	Low dose 1000 ppm	Mid dose 3000 ppm	High dose 10000ppm
Forestomach squamous epithelial hypertrophy	0/49	0/49	0/49	2/49
Forestomach epithelial papilloma	0/50	1/50	1/49	10/50

This evidence of forestomach irritation, albeit weak irritation, is augmented in each NTP bioassay of cresol isomers. The 28-day dietary study, the 90-day dietary study and the chronic study produced ample evidence of profound irritation at the portal-of-entry. Even though the studies employed dietary admixture as the route of cresol administration, atrophy and regenerative changes of nasal epithelium and even lower respiratory tissue were described and a hyperplastic response was noted.

¹ The NTP Draft Technical Report states on page 63: "...there was no evidence of injury to the gastric mucosa; neither hyperplasia, other nonneoplastic lesions, nor inflammation were observed in the forestomach of cresol-exposed animals. Further, no evidence of forestomach irritation was reported in the 13-week studies of m-/p-cresol in male or female mice or in the 28-day study of female mice."

Cresol isomers are strong contact irritants, therefore, portal-of-entry tissues are targets for irritation from cresols. Orally-administered cresol comes into direct contact with mouse forestomach epithelium. The food storage function of the forestomach affords opportunity for prolonged contact of the forestomach epithelium with the cresol/dietary admixture. The m-/p-cresol test mix, which produced papillomas at a dietary concentration of 10,000 ppm in a two-year study, produced hyperplasia of forestomach at dietary concentrations of 30,000 ppm administered for 28 days.

Evidence exists for each of the key events, described above, critical to mode-of-action for the induction of tumors by m-/p-cresols. The possible exception to this is the apparent absence of sustained irritation at the target site and subsequent hyperplasia.

For these reasons, the Cresols Panel respectfully requests that NTP consider including language in the Technical Report that would provide a greater context for understanding these observations in terms of what is known about them and human cancer.

The Cresols Panel of the American Chemistry Council thanks NTP for the opportunity to comment. The scientific comments presented here were prepared on behalf of the Cresols Panel by John H. Butala, Toxicology Consultants Inc., 7 Glasgow Road, Gibsonsia PA, 15044. email: butala@jhbutala.com.

Please contact me if you have any questions.

Sincerely,



Jonathon T. Busch
Manager, Cresols Panel
Director, Chemical Products and Technology Division
American Chemistry Council (ACC)
1300 Wilson Boulevard
Arlington, Virginia 22209
Phone: (703) 741-5633
Email: jon_busch@americanchemistry.com

References

Boutwell, R. K. and Bosch, D.K., The tumor-promoting action of phenol and related compounds for mouse skin. *Cancer Res.*, 19, 413-424, 1959

IARC, Predictive Value of Rodent Forestomach and Gastric Neuroendocrine Tumours in Evaluating Carcinogenic Risks to Humans. International Agency for Research on Cancer Technical Publication Number 39, World Health Organization, Lyon, 2003.

Developmental and Reproductive Toxicity References

R. W. Tyl, Unpublished Report Number 51-508: "Developmental Toxicity Evaluation of o-, m-, or p-cresol Administered by Gavage to New Zealand White Rabbits," Bushy Run Research Center, Export, Pa., June 27, 1988.

R. W. Tyl, Unpublished Report Number 51-509: "Developmental Toxicity Evaluation of o-, m-, or p-cresol Administered by Gavage to Sprague-Dawley Rats," Bushy Run Research Center, Export, Pa., June 29, 1988.

T. L. Neeper-Bradley and R. W. Tyl, Unpublished Report Number 51-634: "Two Generation Reproduction Study of m-Cresol, Administered by Gavage to Sprague-Dawley Rats," Bushy Run Research Center, Export, Pa., February 28, 1989.

T. L. Neeper-Bradley and R. W. Tyl, Unpublished Report Number 51-614: "Two Generation Reproduction Study of o-Cresol, Administered by Gavage to Sprague-Dawley Rats," Bushy Run Research Center, Export, Pa., December 19, 1989.

T. L. Neeper-Bradley and R. W. Tyl, Unpublished Report Number 51-512: "Two Generation Reproduction Study of p-Cresol, Administered by Gavage to Sprague-Dawley Rats," Bushy Run Research Center, Export, Pa., March 28, 1989.