

## **MDCPSS Webinar Questions and Answers - Nitrosamines: Evolving Regulatory Landscape and Its Potential Impact on Medical Devices and Combination Products**

### Question 1

*For less than lifetime, it is quite common for carcinogenic responses in non-nitrosamines as well (see Calabrese 1999). Have you compared the chronic dose to short-term doses? To clarify it is quite common for carcinogenic response following a single dose of a non-nitrosamine carcinogen. But what we find is the acute dose is significantly higher than the chronic dose.*

### **Response**

Thank you for your question. The agency reviews acute and chronic toxicity studies but has not conducted a formal comparison of the tumor incidences in acute toxicity studies compared to chronic/lifetime studies.

### Question 2

*Could you please elaborate more on what the non-standard Ames should look like, I understood, plate test, with rat and hamster S9 at several concentrations and without S9. Was it also correct if the Ames is negative then an in vivo test needs to be conducted, i.e. PigA or transgenic mutation assay? If yes, why should an Ames test be conducted at all?*

### **Response:**

Thank you for your question. Please consult the FDA review Division for specific guidance regarding the evaluation of the specific impurity in question.

In general, we currently recommend that the referenced GLP Ames assay include a full complement of bacterial strains, include rat and hamster S9 at various concentrations (10-30%) and utilize the plate incorporation method.

If the GLP Ames test shows a negative result we recommend that you qualify the impurity in an *in vivo* gene mutation assay such as a Transgenic Rodent Somatic and Germ Cell Gene Mutation Assay (OECD Test 488) with a Pig A endpoint to confirm the absence of mutagenic potential, due to the high level of concern for nitrosamine impurities. Options to characterize *in vivo* mutagenicity include Muta™ Mouse, *gpt* delta mouse/rat, or Big Blue (mouse/rat) assays but your choice of assay should be justified in your submission.

The tissue(s) selection for gene mutation assessment must be based on the evidence that the test chemical (nitroso-impurity) reaches the target tissue(s). For example, adequate impurity exposure to bone marrow tissue should be demonstrated for *in vivo* Pig A assay, or adequate impurity exposure should be demonstrated in tissues selected for mutation analysis in the Transgenic Rodent Somatic and Germ Cell assay. These detailed evaluations are necessary due to the high level of concern for nitrosamine impurities.