EXPERT STATEMENT

The human relevance of the observed lethality of Silanamine, 1,1,1-trimethyl-N-(trimethylsilyl)-, hydrolysis products with silica; pyrogenic, synthetic amorphous, nano, surface treated silicon dioxide (called Silanamine by RAC) (CAS # 68909-20-6) in acute toxicity testing using inhalation exposure in rats

Klaus Weber, PhD, DVM, MSBiol, DJSTP
AnaPath Services GmbH
Hammerstrasse 49
4410 Liestal/Switzerland

Prof. (retired) Dr. Wolfgang Dekant
Rhönstrasse 9
97080 Würzburg/Germany
February 4, 2022

Background. CAS # 68909-20-6 is termed synthetic amorphous silica surface treated with hexamethyldisilazane (HMDZ-SAS). HMDZ-SAS is a low-density particulate material with a hydrophobic surface and is commercially used as non-respirable agglomerates with a mass median aerodynamic diameter (MMAD) of app. 80 µm. Significant shear stress is required to break down the agglomerates into particles in the respirable range (MMAD < 10 µm) for inhalation toxicity testing. The small particles generated by shear stress readily re-agglomerate when shear stress is absent.

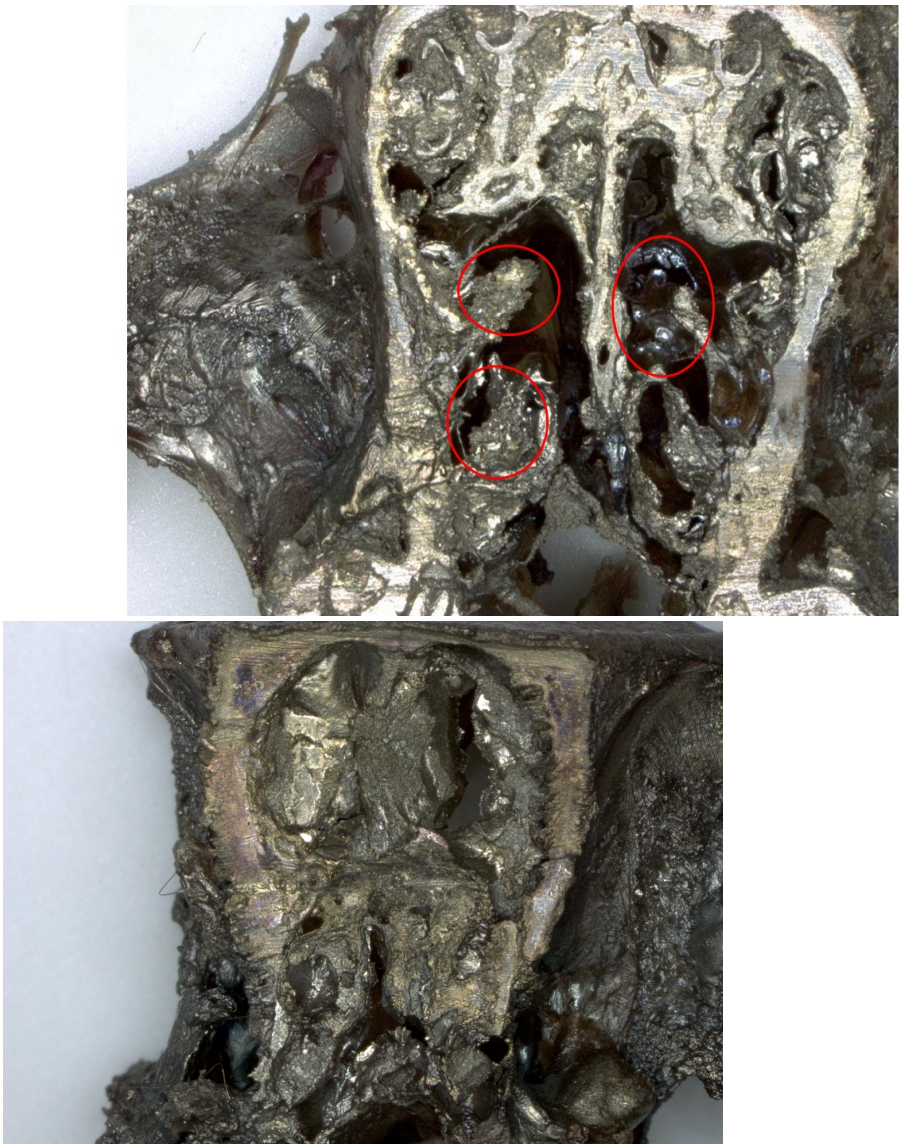
Problem. A HMDZ-SAS (AEROSIL® 812) induced lethality in a 4-hour inhalation exposure in rats at 500 mg/m^3. The derived four-hour LC_{50} may mandate classification of HMDZ-SAS as Acute Tox Cat 2 (fatal if inhaled). However, the particle characteristics of HMDZ-SAS and observations with similar particles suggested suffocation due to airway occlusion as a cause of lethality.

Solution. As the guidelines for acute toxicity assessment (OECD 403 and 436) only require counting dead animals and a macroscopic examination of the surfaces of the organs in the abdominal and thoracal cavity, causes for lethality are not assessed. Experiments including a detailed histopathological examination of the respiratory tract were conducted to characterize mechanisms of lethality of HMDZ-SAS after inhalation exposure.

Methods. Three male and three female rats were exposed to 500 mg/m^3 of AEROSIL® 812 (MMAD of < 4 µm) using nose-only exposure and a standard inhalation toxicity testing system for four hours. A detailed assessment of particle deposition in the entire respiratory tract was included. To avoid removing of deposited particles, nasal cavities and segments of the respiratory tract were shock-frozen and trimmed into specific sections.

Observations. Within three hours after start of the inhalation exposure, three rats of the six rats died. Two more rats died within one hour after termination, the poor condition of the remaining rat required sacrifice. The nasal cavities of all rats contained deposits of re-agglomerated HMDZ-SAS (red circles in example in Fig. 1, top) that almost completely blocked the nasal cavity at level 4 (Fig. 1, bottom). In addition, the lungs of the animals showed clear signs of respiratory failure including focal to multifocal hemorrhage, alveolar fibrin, focal to multifocal acute emphysema. Deposition of HMDZ-SAS (as Si) was not detected in lung tissues.

Figure 1: Nose sections from rats exposed to 500 mg/m^3 of AEROSIL® 812 (MMAD of < 4 µm). Top, male rat, decedent 2 ¾ hours after start of inhalation. Nasal cavity level 4 (frozen, dried, sputtered sample): Partial blockage by deposited test item. Bottom, female rat, decedent 1 hour after end of inhalation. Nasal cavity level 4 (frozen, dried, sputtered sample): complete blockage by deposited test item. Digital microscopy, lens x30.



Conclusions. The results confirm a physical blockade of the upper respiratory tract by agglomerated HMDZ-SAS as cause of death by suffocation. The blocking of the upper respiratory tract in rats is initiated by the rapid re-agglomeration of HMDZ-SAS and an interaction of the formed agglomerates with liquids on the surface of the airways. These forms large and viscous droplets due to the inhalation of a very large number of low-density particles.

Implications. Lethality is not due to an intrinsic toxic property of HMDZ-SAS. The observations confirm the caveats regarding lethal effects by physical obstruction of airways after inhalation of high particle loads described in OECD guidance 39. In addition, lethality due to physical obstruction of the nasal cavity in rats after exposures to determine a LC_{50} has no human relevance. This is due to differences in breathing patterns and airway structures.
Rats, due to a close apposition of the rodent epiglottis to the soft palate, are obligate nose-breathers. Blockade of the nasal passage will result in asphyxia. In contrast, humans are both nose and mouth breathers, specifically under stress. Therefore, blockade of the nasal passages will not result in asphyxia since mouth breathing remains possible in humans.
The upper airways in rats and humans also show major differences regarding airway geometry (shape of nasoturbinate and maxilloturbinate) and diameter (see Fig. 2). These influence air flow characteristics, location and amount of particle deposition, and tendency for blocking of airways. Basic laws of fluid dynamics of particulate containing air in low diameter tubing predict a very low airflow in rats exactly at the site of airway blockade seen with HMDZ-SAS (transition from nasal cavity section 4 to larynx, see Fig. 1). The low airflow promotes re-aggregation and deposition of HMDZ-SAS on the moist surfaces of the airways result in blockade of the airways in rats. In humans, such low airflows are not predicted, and deposition will not occur at these sites. In addition, the much wider airways of humans require formation of larger droplets. These cannot form due to absence of moisture on the surface of an already deposited particle film.

If particles inhaled at high concentrations reach the lower airways in humans, the major structural differences will also result in different outcomes and blocking of the lower airways in humans is considered as highly unlikely.

Therefore, lethality due to physical obstruction of airways under conditions of assessing an LC_{50} should not be used in classification and labeling.

Figure 2: Predicted air flow rates in human and rat nose (from Shang et al., 2016; DOI: 10.3109/08958378.2015.1088600). Areas with highest airflow are in red, lowest airflow in green. In rats, the lowest airflows are predicted in the transition from the nasal cavity to the larynx, which has also a low diameter. These two factors contribute to sedimentation of low-density particles such as HMDZ-SAS and blockade of airways in rats. In humans, both diameter and airflows are larger and thus do not result in airway blockade by low-density particles.