Focus: Arsenic in Grain-Based Laboratory Animal Diets and Effects on the Rodent Toxicological Phenotype

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In research with animal models, there are many factors to consider when designing an experiment to achieve a particular research goal. Most researchers typically consider the animal model and environmental factors such as housing conditions (i.e. cage density, room temperature and humidity), but one factor that may be overlooked or considered of lesser importance is the diet to be fed. This is usually the case when certain dietary manipulations are not required, and the researcher would then simply default to what diet the animal facility normally uses: 'grain-based chow' or 'chow' for short. While it may not have an obvious influence if all animals are fed chow during a given experiment, this could be a critical factor that may interfere with your expected results now, and in the next study.

Chow vs. Purified Diet

There are 2 basic diet types used in preclinical studies: chow or a purified diet. There are a variety of chows available from manufacturers, which are typically 'closed' or proprietary formulas, and each batch is commonly made with set or variable (depending on the manufacturer) amounts of agricultural by-products such as cereal grains (i.e. soybean meal, ground corn, wheat, and oats) as well as animal by-products (i.e. porcine meat meal, fish meal). Due to their unrefined nature (and as you'd expect), each ingredient provides multiple nutrients, but there are also non-nutrients that usually ride along with these nutrients. In contrast, purified diets are 'open' to the public and each ingredient contains one main nutrient. Due to the refined nature of these ingredients, the presence of non-nutrients in purified diets is limited.

Heavy Metals in Grains

Grains such as ground corn and wheat, which are found in chows, contain a significant amount of carbohydrate, but also contain some protein, fiber, and minerals. What is perhaps less well known is that these grains also contain toxic heavy metals, such as arsenic, which come from the soil; other chow ingredients such as fish meal also can provide arsenic [1–3]. Kozul and colleagues (4) found that the chow they normally used (Purina 5001) contained relatively high concentrations of arsenic (390 ppb total, 56 ppb inorganic), and also several other heavy metals (cadmium, lead, nickel), while they were virtually absent in a purified diet (AIN-76A). Perhaps not surprisingly, when they supplemented chow fed mice with arsenic at 10 to 100 ppb, they didn't find a significant change in gene expression compared to chow fed, non-supplemented mice. Indeed their data showed that chow by itself 'turned on' many genes, including phase I and phase II enzymes involved in metabolism of xenobiotics (i.e. foreign substances) in the liver and also those involved in immune signaling in the lung. These data suggest that arsenic (and perhaps other components in the chow) affected their ability to observe a change with supplemental arsenic already in the chow. When arsenic was given to mice fed the purified diet, a significant change in gene expression profile was observed compared to non-supplemented, purified diet fed mice. Follow-up studies by this group were performed using the same purified diet and found arsenic at 100 ppb could influence immune response, immune protection from disease, and growth of neonatal mice after arsenic exposure in utero, postnatal, or during both periods [5–7]. These subsequent data suggested that changes in gene expression by the presence of arsenic at even lower doses than in chow can profoundly influence an animal's toxicological phenotype.

Clean, Consistent Purified Diets

Because of these data, we were interested in gaining more insight on the levels of toxic heavy metals in different grain-based chows. We found that every grain-based chow analyzed by an independent laboratory had measurable arsenic levels, and differences were observed among chows, within and between manufacturers (Figure 1) (8). We also found that arsenic concentrations of the same chow (NIH-31M) from different manufacturers can vary significantly, suggesting that ingredient sources have highly variable arsenic levels (Figure 1).

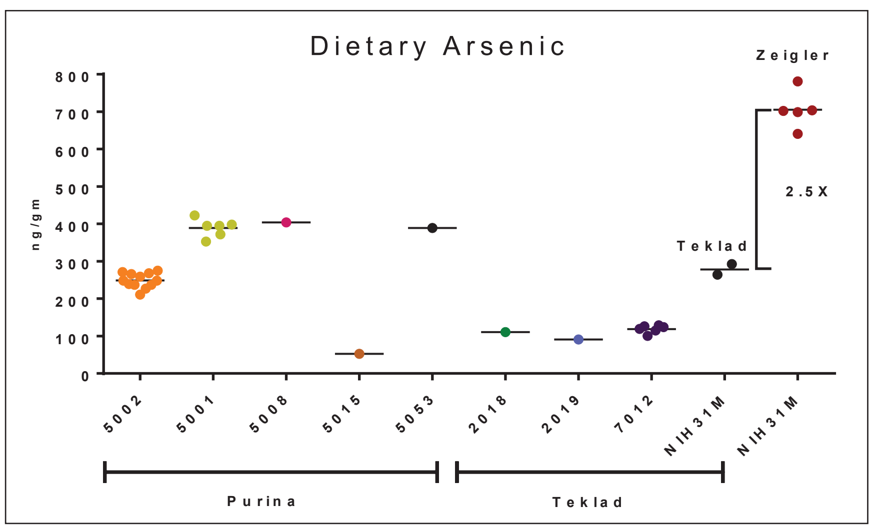

FIGURE 1 - Each dot represents a different chow manufacture date.
ND = Not Detectable. ng/ml = ppb (parts per billion)

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When we fed rats 2 different chows with different arsenic concentrations, it led to concentration-dependent changes of arsenic in their tissues (Figure 2). We also found that individual rats of the chow groups had highly variable tissue arsenic concentrations. In contrast, purified diets contained undetectable levels and significantly reduced tissue arsenic concentrations relative to chow fed rats (Figure 2) (8).

These results strongly suggest that researchers should choose the diet wisely as the presence of arsenic and other toxic heavy metals can have a profound effect on the rodent toxicological phenotype. Purified diets provide a clean, consistent nutrient profile and minimize the presence of any contaminants that can affect the phenotype of your valuable animal models.

For further information regarding any of these data, please contact us at info@researchdiets.com.

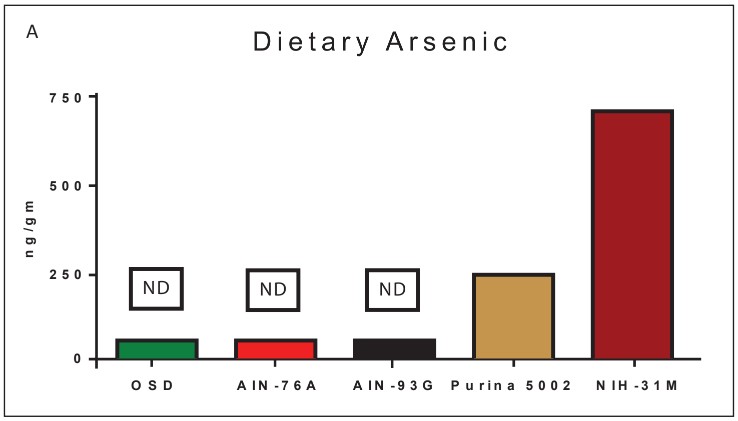

FIGURE 2A - AIN-76A, AIN-93G, and OSD (OpenStandard Diet) are purified diets, Purina 5002 and NIH-31M (Zeigler) are chows. Purified diets had non-detectable levels of arsenic while chows had detectable and variable levels of arsenic. Each dot represents kidney (B) and liver (C) arsenic concentrations for each rat. Groups with different letters are significantly different.

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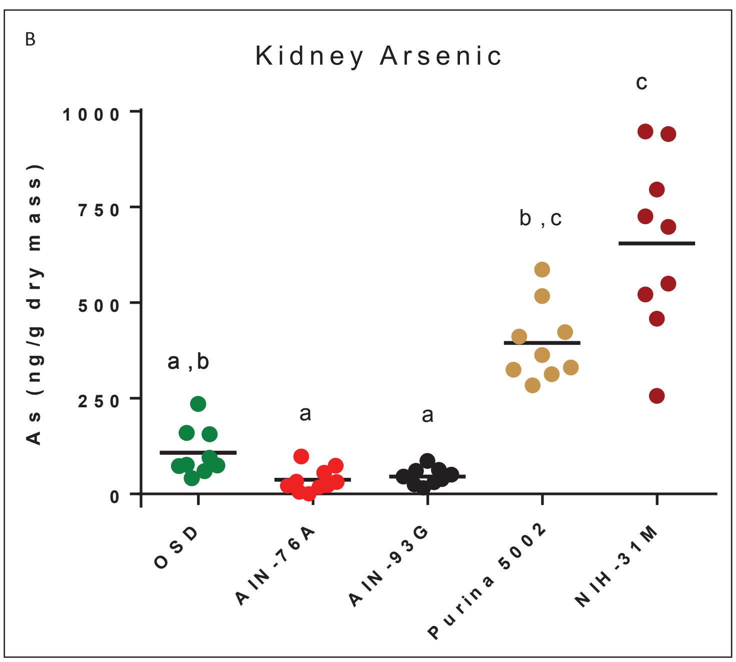

Figure 2B

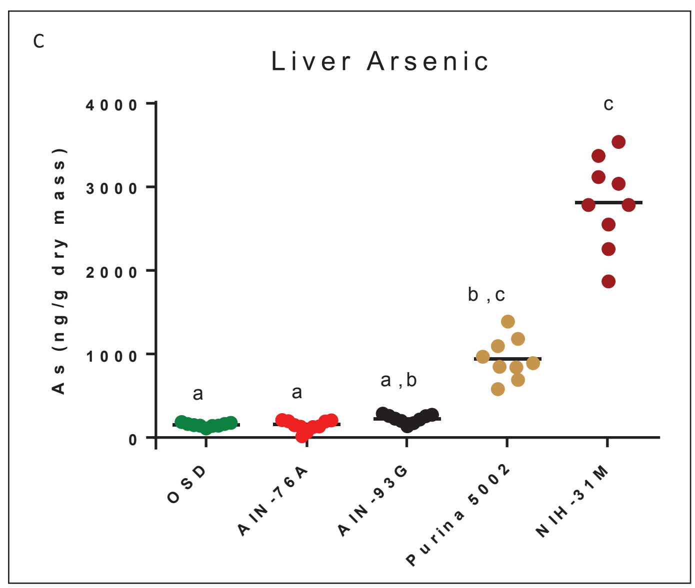

Figure 2C

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