Setting Allowable Exposures to Ames-positive Candidate Drugs

FDA Genetic Toxicology Workshop

November 4, 2019

Kenny Crump

Kennycrump@email.com

The question posed at the workshop:

How many doses of an Ames positive drug could be administered to healthy subjects without significantly increasing their cancer risk?

An acceptable answer to this question must somehow take into account some measure of the mutagenic or projected carcinogenic potency of the drug.

Even if we had a full suite of typical data on the candidate drug, (animal bioassays, epidemiological studies, etc.) making creditable estimates of cancer risk would be very difficult.

Most of these data typically would be from **chronic exposures**.

Estimates of risk from **short term, low level exposures** would require extrapolating from data on chronic exposures.

An NAS Committee faced a similar problem when asked to assess risk posed by controlled human inhalation exposure (CHIE) studies

In CHIE studies volunteers are exposed to low-level short inhalation exposures (e.g., one hour) to well-studied pollutants (e.g., ozone) under controlled experimental conditions.

Comparing CHIE studies to our situation

SIMILARITIES OF CHIE STUDIES TO OUR PROBLEM

- Exposures to human volunteers
- Controlled conditions
- Short-term exposures

DIFFERENCES IN CHIE STUDIES AND OUR PROBLEM

- Exposures in CHIE studies are to well-studied pollutants
- Lifetime rodent bioassays are often available
- Epidemiological data are often available

Because of the difficulty of making credible estimates of risk from short-term exposures from data on chronic exposues, the NAS committee did not recommend making quantitative estimates of cancer risk.

Instead the committee recommended using "exposure scenario comparators" (ESC) to evaluating the risk in CHIE studies. This involves comparing the exposure in a CHIE experiment to a similar exposure to which people are chronically exposed.

Despite having far less data than is typical of CHIE studies, is there some approach similar to Exposure Scenario Comparator (ESC) approach that might be appropriate for Ames-positive drugs?

In the remainder of my talk I will propose such an approach based on a comparator chemical instead of a comparator exposure scenario.

Nitrite is proposed as the comparator chemical

Nitrite seems to be unique in that it is an Ames-positive chemical that is allowed to be present in food in small amounts.

Nitrite has not been shown to be carcinogenic.

Maximum acceptable daily intakes of nitrite

- **USFDA** allows consumption of some fish containing **200 ppm** nitrite

= **17 mg** of nitrite per serving of 3 oz. fish

- **WHO** maximum recommended daily intake is **0.13 mg/kg** of nitrite

= **8.9 mg** (150 lb person)

- **FAO/WHO** maximum recommended daily intake is **0.05 mg/kg** of nitrite

= **3.4 mg** (150 lb person)

Quantify the mutagenic potential of a candidate drug by applying linear regression to Ames test data

$$Y = \alpha(1 + \beta X)$$

X – Concentration of candidate drug on petri dish

Y – Number of revertants

β – Mutagenic potency of candidate drug

If the entire Ames data are not consistent with a linear response, then sequentially remove highest dose until the remaining data are consistent with linearity.

Estimating potency assuming "low-dose linearity"

Precedent for this approach for low-dose linearity

- "Predicting Rodent Carcinogenicity From Mutagenic Potency Measured in the Ames Salmonella Assay" (1997)
- Bethel A. Fetterman, Byung Soo Kim, Barry H. Margolin, Jonathan S. Schildcrout, Melissa G. Smith, S. Michelle Wagner, Errol Zeiger

Calculation of a maximum daily exposure (MDE) to a candidate drug

$$MDE = 17\ \hat{\beta}_N / \hat{\beta}_D \,\text{mg}$$

17 - the maximum daily exposure to nitrite in mg allowed by USFDA

$\hat{\beta}_D$ - estimated mutagenic potential of candidate drug

$\hat{\beta}_N$ - estimated mutagenic potential of nitrite

Details

- It is suggested that nitrite be tested concurrently with the candidate drug using the same experimental protocol (same salmonella strains, activation protocol, etc.).
- It is suggested that a statistical lower bound be used as the MDE rather than the point estimate.
- Decisions would need to be made regarding how to use data from multiple strains and activation protocols to use in defining the MDE.

Interpretation

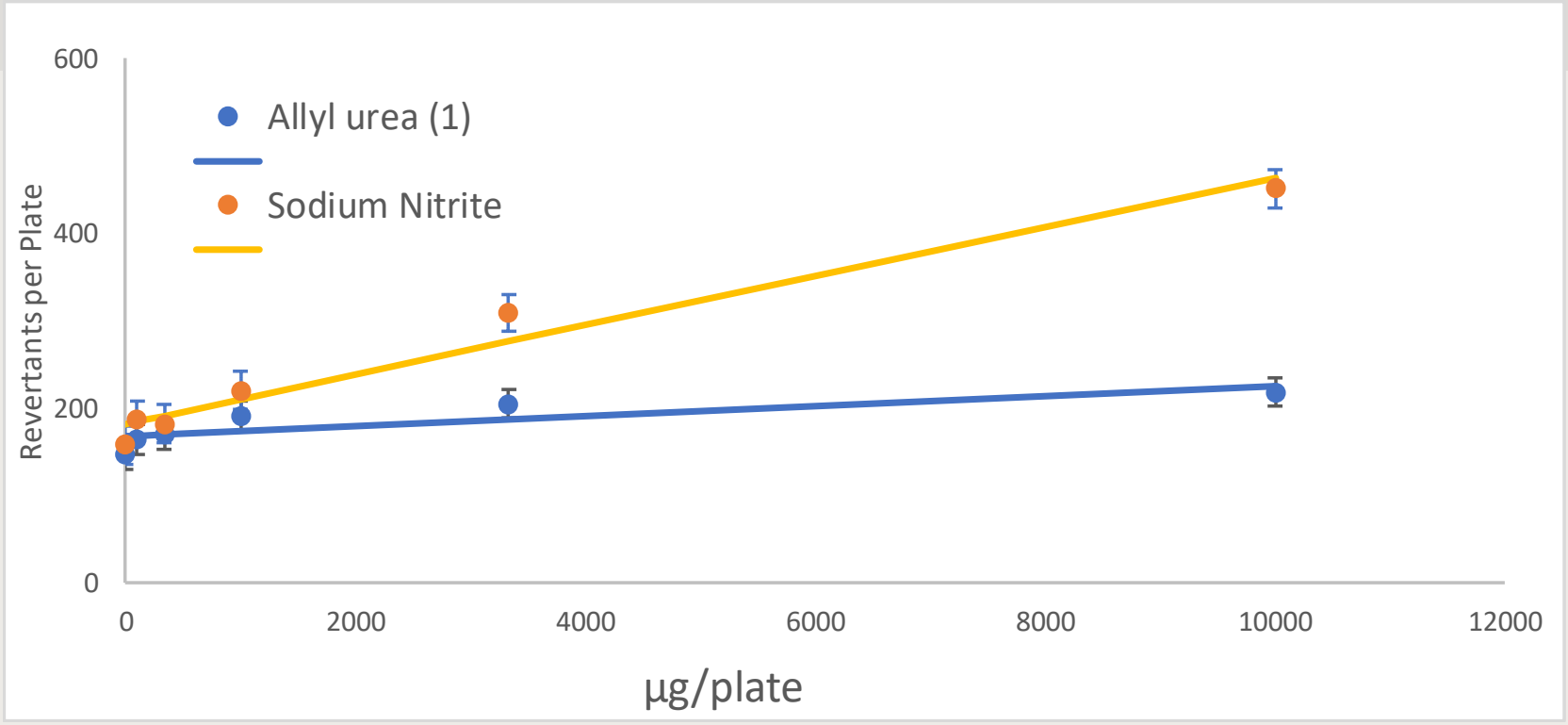
- This approach does not place any restriction on the number of days a volunteer could be exposed. This is in keeping with the fact that it is based on maximum daily exposure limits for nitrite which also do not have any such restrictions.
- Prudence would dictate that exposure should only last (to doses no greater than the MDE) for the minimum number of days necessary to answer the scientific question.
- Exposure to a candidate drug's MDE will entail the same mutagenic potential as exposure to an amount of nitrite allowable by the USFDA, an Ames-positive chemical which has not been found to be capable of causing cancer.

Examples based on data in Zeiger et al. (1992)

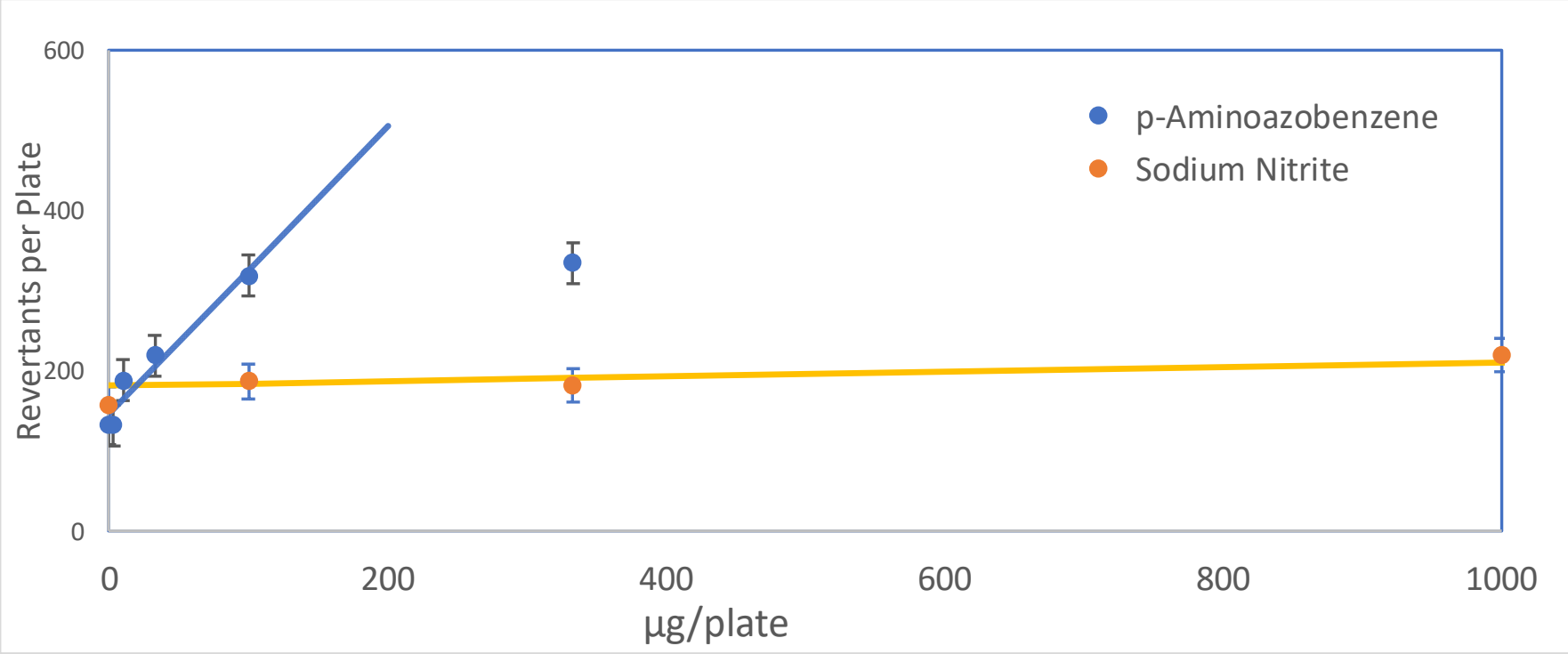
Zeiger (1992) table	Table 282 Sodium Nitrite	Table 11 Allyl urea	Table 11 Allyl urea	Table 11 Allyl urea	Table 13 p-Aminoazo-benzene	Table 14 o-Aminoazo-toluene	Table 18 2-amino-nitrothiazole
Salmonella typhimurium strain	TA(100)	TA100(1)	TA100(2)	TA100(3)	TA100	TA100	TA100
S-9 Fraction	30% HLI	30% HLI	30% HLI	30% HLI	30% HLI	30% HLI	30% HLI
Mutagenic?	+	+W	+W	+W	+	+	+
α (mle)	182.1	168.2	181.4	158.7	176.2	173.0	46.6
β (mle)	1.54E-04	3.40E-05	2.29E-05	4.56E-05	3.19E-03	5.07E-02	2.45E-02
MDE (mg/day)(mle)	17.01*	77.0	114.6	57.5	0.21	0.052	0.1071
MDE (lower 95%)		42.9	59.5	37.7	0.18	0.044	0.0925

* fixed value based on maximum allowable concentration of nitrite in fish of 200 ppm by USFDA

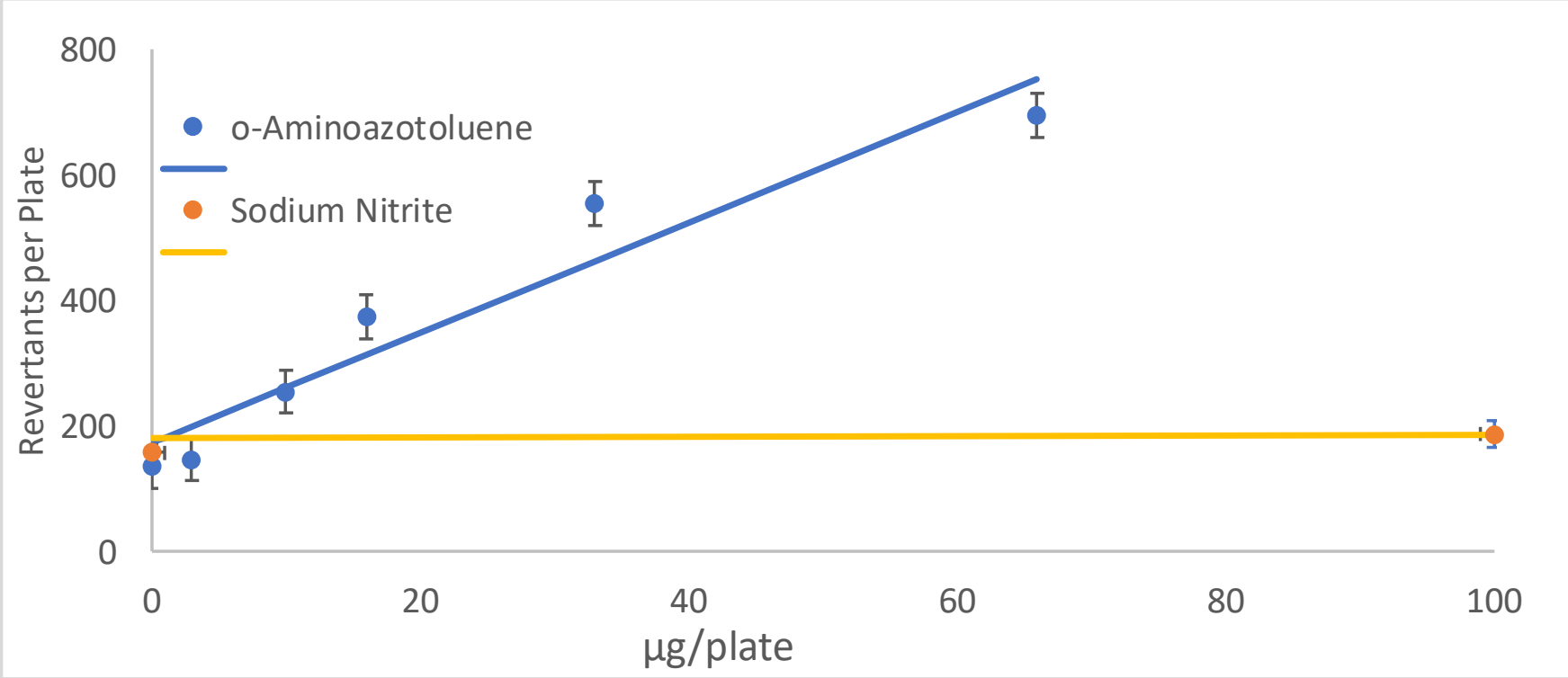
Allyl urea, TA100 30% HLI(1)



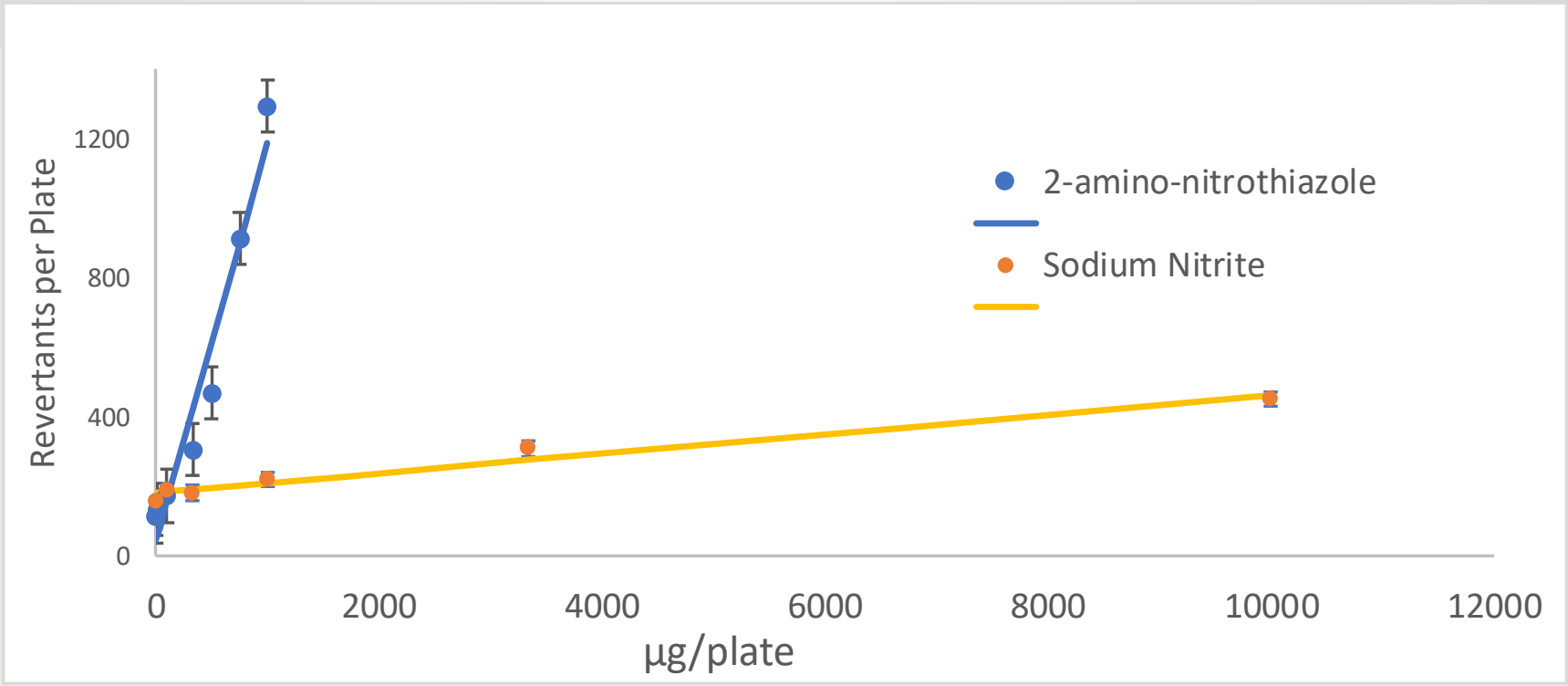
p-Aminoazobenzene, TA100 30% HLI



o-Aminoazobenzene, TA100 30% HLI



2-amino-nitrothiazole, TA100 30% HLI



Characteristics of this approach

- Straight-forward and easily implemented
- Takes into account the mutagenic potency of a candidate drug
- Does not restrict the number of daily exposures, but rather the amount of exposure per day
- Based on the precedent set by USFDA for an Ames-positive chemical. (This approach ensures that an MDE for a candidate drug has equal mutagenic potential as FDA's MDE for nitrite.)