Use of the Rozman Scale in tobacco carcinogenesis

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Outline for presentation

- Definition and use of the Rozman scale
- Background
- Objectives for presentation
- Examples of the use of the Rozman scale in tobacco carcinogenesis
- Conclusions

Definition and use of the Rozman scale

- A dose-response scale where dose is expressed in molecules/kg body weight/day (logarithmic scale) and response is percent of animals with tumors (corrected for zero-dose controls)
- The Rozman scale has been used to estimate thresholds for chemical carcinogenesis
  - Waddell, *Toxicol. Sci.* 2002;68:275-9
  - Waddell, *Toxicol. Pathol.* 2003;31:260-2

Background for presentation –1

- Mainstream cigarette smoke (MSS) contains numerous carcinogens
  - Dose per day to a smoker depends on several factors
    - Brand-style of cigarette
    - Puffing behavior of the smoker (puff volume & frequency)
    - Blocking of filter ventilation (if applicable & occurrence)
    - Number of cigarettes per day
    - Depth of inhalation
    - Toxicants of interest and their retention in respiratory tract
  - Daily intake of a toxicant best measured by human biomonitoring in an in-patient clinical setting
    - This can be difficult and time consuming
    - So estimates of maximum dose made by using toxicant yields determined by machine smoking under intensive conditions and likely maximum number of cigarettes smoked per day
    - For example, Health Canada yields and 60 cigarettes per day

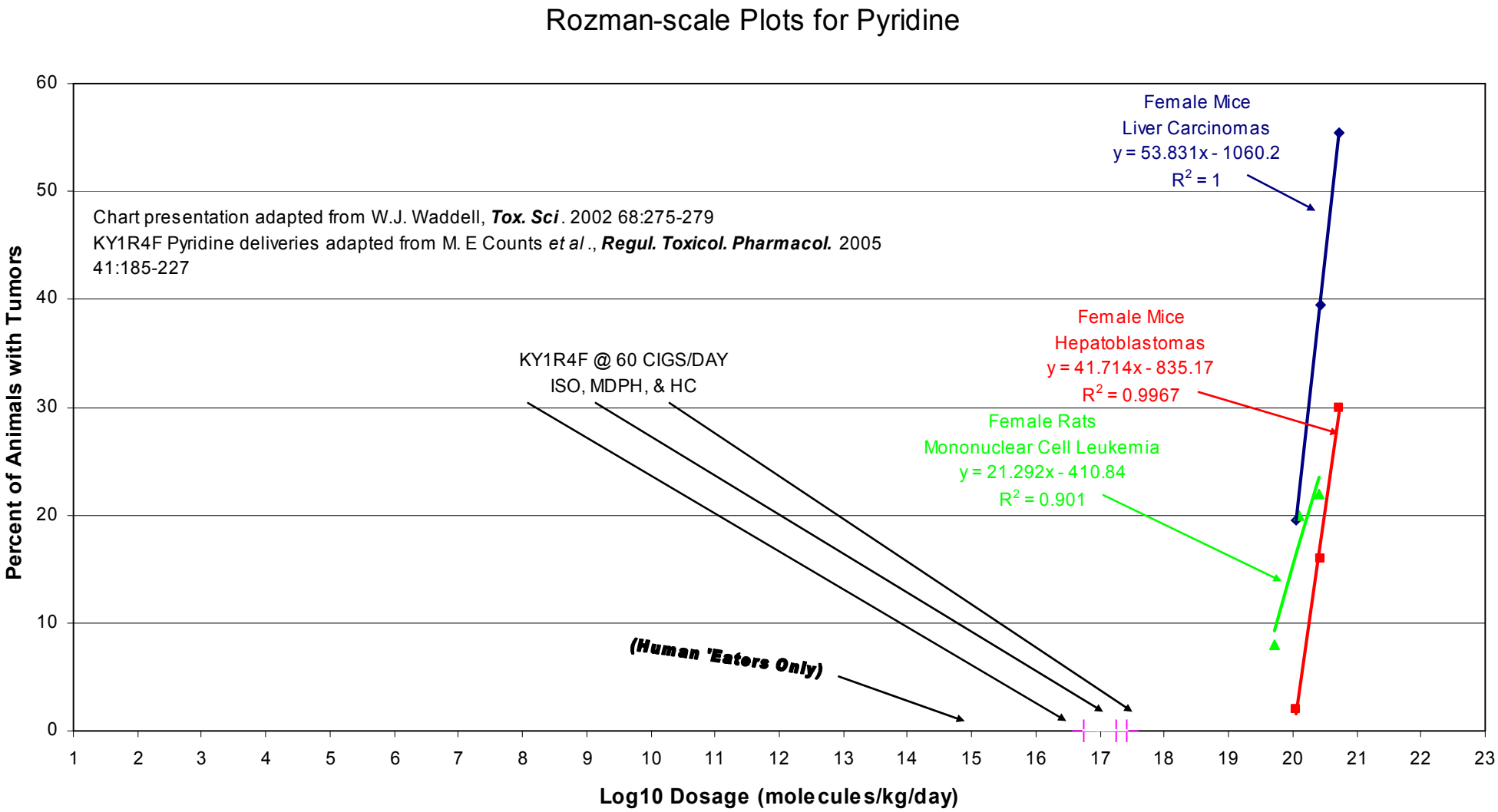
Background for presentation – 2

- It would be desirable to know which toxicants present the greatest danger to the smoker
  - Compounds and levels that should be considered in a regulatory scheme
  - Development of technologies to reduce hazards of smoking
- The Rozman scale provides a way of making such estimates
- Procedure also applicable to other tobacco products such as moist snuff

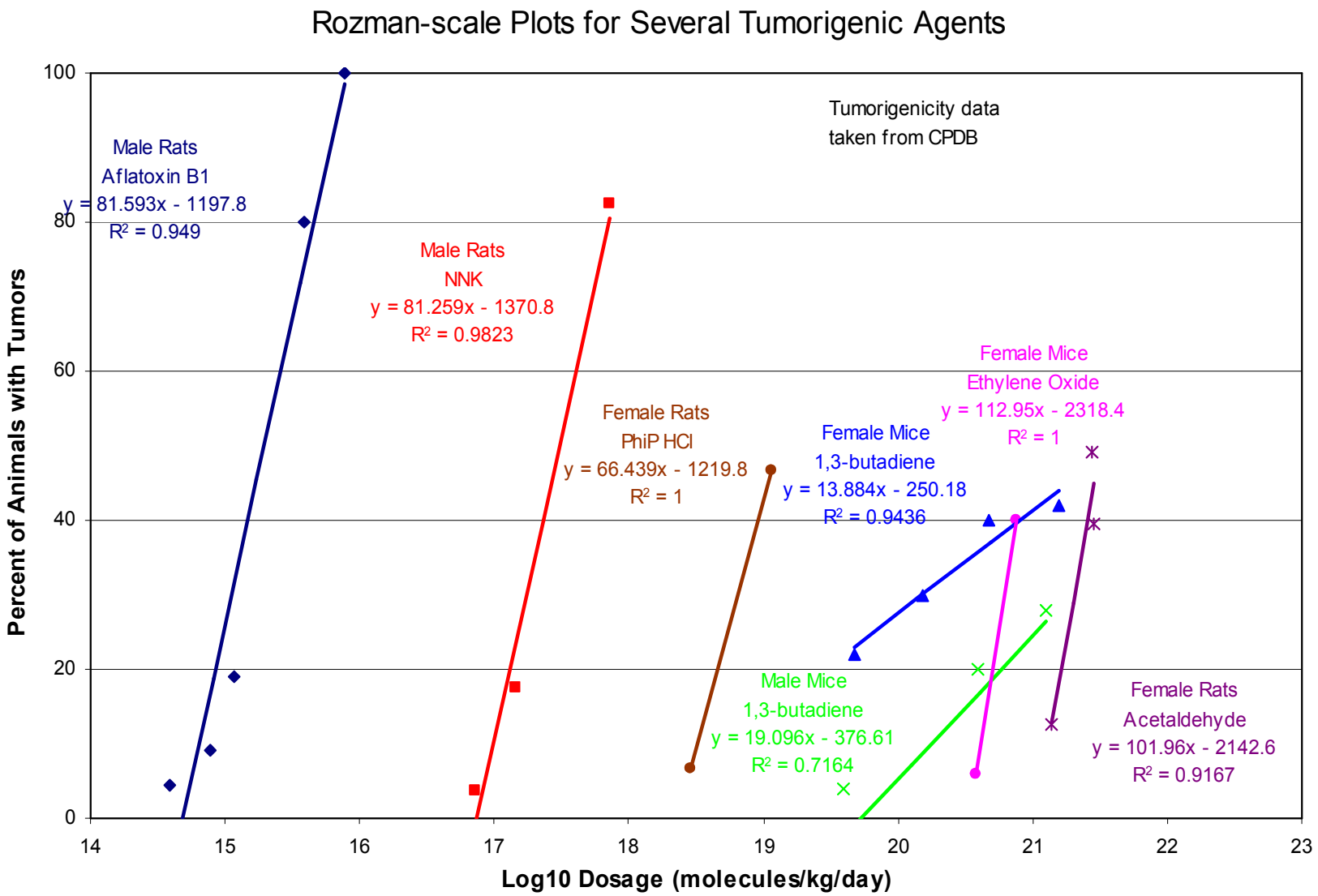
Objectives for presentation

- Demonstrate the use of the Rozman scale with several compounds in MSS
- Demonstrate the use of the Rozman scale for NNK in smokeless tobacco products
- Compare estimates made with Rozman scale with those made with Carcinogenic Potency Project TD50 values

Is there a threshold for tumorigenicity?



Tumorigenicity thresholds



Threshold versus estimated MSS dosage

Compound	Species	Sex	Strain	Log10 Threshold	KY1R4F HC Avg	Intake	Log10 Dosage
				(molecules/kg/day)	μg/cig	μg/day	molecules/kg/day
NNK	Rat	M	F344	16.9	0.199	12.0	14.7
1,3 Butadiene	Mouse	F	B6C3F1	18.0	99.5	5967	18.0
PhiP HCl	Rat	F	F344/DuCrj	18.4	0.0062	0.370	13.1
1,3 Butadiene	Mouse	M	B6C3F1	19.7	99.5	5967	18.0
Ethylene oxide	Mouse	F	B6C3F1	20.5	50	3000	17.8
Acetaldehyde	Rat	F	Wistar	21.0	1404	84210	19.2

- Assumptions for estimations
  - Cigarette is KY1R4F
  - Smoker weighs 70 kg and smokes 60 cigarettes per day with intake approximating Health Canada protocol
  - MSS delivery of ethylene oxide for KY1R4F under Health Canada intensive conditions similar to that of KY2R4F

NNK – smokeless tobacco products

- Smokeless tobacco products differ by type, NNK levels, and amount consumed each day and these parameters influence NNK dosage (molecules/kg body weight/day
- Product with very low NNK level gives estimated dosage 1000X below Rozman-scale threshold

		Reported*	Est. Daily	Est. Daily	Est. Daily	Formula	Est. Daily	Est. Daily	Log10	Log10
		Concentration	Usage**	Dose	Dosage	Weight	Dosage	Dosage	Est. Daily Dosage	Threshold
Product	Component	Dry wt. basis	(kg/day)	(mg/day)	mg/kg/day	(mg/mmol)	(mmol/kg/day)	(molecules/kg/day)	(molecules/kg/day)	(molecules/kg/day)
Chewing tobacco	NNK (mg/kg)	0.4	8.50E-02	3.40E-02	4.86E-04	207	2.35E-06	1.41E+15	15.15	16.9
Moist snuff, US	NNK (mg/kg)	0.9	3.50E-02	3.15E-02	4.50E-04	207	2.17E-06	1.31E+15	15.12	16.9
Moist snuff, SE	NNK (mg/kg)	0.4	3.50E-02	1.40E-02	2.00E-04	207	9.66E-07	5.82E+14	14.76	16.9
Compressed snuff	NNK (mg/kg)	0.04	1.10E-02	4.40E-04	6.29E-06	207	3.04E-08	1.83E+13	13.26	16.9
Toombak	NNK (mg/kg)	2310	3.50E-02	8.09E+01	1.16E+00	207	5.58E-03	3.36E+18	18.53	16.9

* Concentration data from Rodu & Jansson, *Crit. Rev. Oral Biol. Med*. 2004;15(5):252-63 or Stepanov *et al.*, Nicotine Tob. Res, 2006;8:309-13; **Estimates of daily consumption based on package weights & personal observation

Other potency tools – The CPDB

- Carcinogenic Potency Project TD_{50} values
  - Carcinogenic potency is defined in the CPDB (Carcinogenic Potency Database) in terms of the average daily dose-rate that will halve the probability of remaining tumor-free at end of standard lifespan (TD_{50})
  - CPDB is a unique and widely used international resource of results from 6153 chronic, long-term animal cancer tests on 1485 chemicals
  - TD50 values for the compounds can be used in conjunction with human dosage data to calculate HERP (Human exposure/rodent potency) values

Example of CPDB and HERP

	Rats	Hamsters	Mice	KY1R4F	KY1R4F	KY1R4F	HERP	HERP	HERP
	TD_{50}	TD_{50}	TD_{50}	HC	HC	HC	Rats	Hamsters	Mice
Compound	**mg/kg/day**	**mg/kg/day**	**mg/kg/day**	**µg/cig**	**µg/day**	**mg/kg/day**	**(%)**	**(%)**	**(%)**
1,3-Butadiene	261	NA	13.9	99	5940	0.0849	0.033	NA	0.610
Acetaldehyde	153	565	NA	1404	84240	1.203	0.787	0.213	NA
Acrylonitrile	16.9	NA	NA	30	1800	0.026	0.152	NA	NA
Benzene	169	NA	77.5	80	4800	0.069	0.041	NA	0.0885
Catechol	84.7	NA	244	88	5280	0.075	0.089	NA	0.0309
Hydroquinone	82.8	NA	225	103	6180	0.088	0.107	NA	0.0392
Formaldehyde	1.35	NA	43.9	61	3660	0.052	3.87	NA	0.1191
B[a]P	0.956	NA	11	0.0136	0.816	0.00001	0.00122	NA	0.00011
NNK	0.0999	NA	NA	0.199	11.94	0.0002	0.171	NA	NA
NNN	NA	10.8	NA	0.244	14.64	0.0002	NA	0.00194	NA

TD50 values taken from http://potency.berkeley.edu ; HERP values estimated from procedure given at http://potency.berkeley.edu/pdfs/herp.pdf

Dosage values based on Health Canada smoke deliveries, 60 cigarettes per day, and 70 kg body weight

Smoke deliveries taken from M. E. Counts *et al*., *Regul. Toxicol. Pharmacol.* 2005 41:185-227

Comments on HERP values

- Values shown are based on three-packs per day, Health Canada yields, and 70 kg body weight
- Volatile components appear to dominate the estimations
  - Acetaldehyde and formaldehyde dominate values for rats as rats reportedly do not give high response with 1,3-butadiene
  - Other than the response in rats to NNK, HERP values for to other particulate-phase compounds are low because of very low levels in smoke
  - Is this approach relevant for particulate-phase compounds in MSS?

Wet snuff – TD50 and HERP values

	Assumed	Rats	Hamsters	Mice	Intake	Intake	Dosage	Dosage	HERP-30	HERP-30	HERP-30	HERP-60
	Concentration	TD_{50}	TD_{50}	TD_{50}	Wet Snuff	Wet Snuff	Wet Snuff-30	Wet Snuff-60	Rats	Hamsters	Mice	Rats
Component	at 50% Moisture	mg/kg/day	mg/kg/day	mg/kg/day	@ 30 g/day	@ 60 g/day	mg/kg/day	mg/kg/day	(%)	(%)	(%)	(%)
NNK (mg/kg)	5*	0.0999	NA	11	0.15 (mg/day)	0.30 (mg/day)	2.143E-03	4.286E-03	2.1	NA	0.02	4.3
NNK (mg/kg)	0.25**	0.0999	NA	11	0.0075 (mg/day)	0.015 (mg/day)	1.071E-04	2.143E-04	0.11	NA	0.001	0.21
NNN (mg/kg)	5*	NA	10.8	NA	0.15 (mg/day)	0.15 (mg/day)	2.143E-03	8.571E-03	NA	0.02	NA	NA
NNN (mg/kg)	0.55**	NA	10.8	NA	0.0165 (mg/day)	0.033 (mg/day)	2.357E-04	4.714E-04	NA	0.002	NA	NA
NDMA (µg/kg)	5	0.0959	NA	0.189	0.00015 (mg/day)	0.0003 (mg/day)	2.143E-06	4.286E-06	0.002	NA	0.001	0.004
BaP (µg/kg)	10	0.956	NA	NA	0.0003 (mg/day)	0.0006 (mg/day)	4.286E-06	8.571E-06	0.0004	NA	NA	0.001

* GothiaTek limit on all TSNAs combined is 5 mg/kg

** Estimated amount in several brands of Swedish snus - adapted from Rodu and Jansson, ***Crit. Rev. Oral Biol. Med***. 2004 15:252-263

- Dosage and HERP values based on 30 g per day or 60 g per day and 70 kg body weight
- NNK appears to dominate the estimations

Conclusions - 1

- The use of the Rozman scale and the CPDB/HERP values provide similar information
  - In this presentation, same sets of experimental tumorigenicity data used as basis for calculations
  - Each approach has its own merits
  - Good estimates of human dosage needed to compare rodent results with potential for human hazard
- Both approaches consider each compound without consideration of other compounds
  - No accounting for possible additivity, synergism, potentiation, antagonism
  - For MSS analytes, there is consideration for reactivity of constituents or partitioning in MSS aerosol

Conclusions - 2

- The Rozman scale is more “chemist friendly” in that it is based on chemical potential and chemical potential is directly proportional to logarithm of concentration
- Results from both approaches need to be used with care as in a given situation rodents may be more susceptible to carcinogenesis from a given chemical than humans