

Inhalation Bioassay Chemistry—Walton Horizontal Smoking Machine for Inhalation Exposure of Rodents to Cigarette Smoke^{1,2}

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ABSTRACT—Studies of experimental tobacco smoke carcinogenesis have suffered from the lack of a conveniently available and well-characterized device for exposing animals to tobacco smoke for inhalation. The Walton Horizontal Smoking Machine, a commercially available system designed to expose up to 20 mice to the smoke of a single cigarette, may fulfill this need. This system produced a uniform smoke aerosol of predictable concentration and appropriate composition for cigarettes with high delivery of nicotine (40 mg total particulate matter, 2.6 mg nicotine, and 17 cm³ carbon monoxide per cigarette) and with low delivery of nicotine (30 mg total particulate matter, 0.3 mg nicotine, and 17 cm³ carbon monoxide). In this experiment C57BL and DBA/2Bd strains of mice were used. Limitations of the concept of exposing animals to standing smoke were defined.—JNCI 63: 441-448, 1979.

Biologic studies with the use of cigarette smoke are complicated by the requirement (1) that dosing be extrapolatable to the situation for humans. Unlike many environmental exposures, the route of administration (i.e., inhalation) and the properties^{5,6} of the insult (2, 3) are well defined. Deviations from acceptable conditions of exposure (4) can invalidate the results of biologic studies. Whereas it may be impossible to precisely mimic the human situation, it is generally agreed (1) that the aerosol offered the animals must be physicochemically comparable to that encountered by man and that the interaction between the animal and the smoke during exposure must not produce a "smoke" subsequently inhaled by the animals that deviates excessively from that which is inhaled by man.

Cigarette smoke produced by drawing a puff of 35±2 cm³ over a 2-second period is currently used (4, 5) as the standard. To further standardize experimental conditions, puffs are taken at precisely 1-minute intervals following the lighting puff. The integral of puff volume and duration defines the flow rate of air through the cigarette and therefore the chemical composition of the resulting smoke in a given puff from a given cigarette. Mechanical devices are required to produce smoke of the appropriate composition for inhalation by test animals.

Mechanically assisted smoke devices may be broadly classified (4, 6) as those that provide exposure to "standing" [static (4)] smoke or those that provide exposure to "flowing" [dynamic (4)] smoke. Standing smoke implies that the smoke from a single puff is allowed to remain in a chamber from which the animals breathe and rebreathe the same aerosol. Flowing smoke implies that the animals breathe from a

stream of constantly replenished smoke aerosol so that rebreathing the same smoke is impossible. Devices (4, 6) that expose animals to standing smoke are mechanically simpler and therefore much less costly than those that produce flowing smoke.

The WHSM (7) exposes rodents to standing smoke. A puff of cigarette smoke is introduced into a chamber from which the animals respire. The smoke remains in the chamber for a preselected period, typically 30 seconds, and is then flushed from the chamber with fresh air for the remainder of the 1-minute interval before the subsequent puff is introduced into the breathing chamber. Up to 20 mice may be exposed to the smoke of a single cigarette.

The device exposes the animals to a predictable and determinable quantity of uniformly distributed smoke of appropriate chemical and physical properties. The concentrations of most chemical constituents of the particulate and vapor phases of the smoke in the chamber decrease during the exposure period. The carbon dioxide concentration and the particle size of the smoke increase during exposure. The WHSM may

ABBREVIATIONS USED: OD=outer diameter; RTD=resistance to draw; TPM=total particulate matter; WHSM=Walton Horizontal Smoking Machine.

¹ Received August 29, 1978; accepted January 3, 1979.

² Sponsored jointly by the Council for Tobacco Research-USA, Inc., and the U.S. Department of Energy under contract W-7405-eng-26 with Union Carbide Corporation.

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⁴ We acknowledge the active participation of J. H. Kreisher, Council for Tobacco Research-USA, Inc., in experimental design; the cooperation of L. Florant, Process and Instruments Corporation, in making design modifications on the smoking machine; and the contributions of C. E. Whimire, the National Cancer Institute, and J. E. Caton, Oak Ridge National Laboratory, for the dosimetry information.

⁵ Griest WH, Guerin MR: Influence of tobacco type of smoke composition. In *Recent Advances in Tobacco Science. Tobacco Smoke—Its Formation and Composition* (More GP, Wernsman EA, Kuhn WH, eds), vol 3. Proceedings of 31st Tobacco Chemists' Research Conference, Greensboro, N.C., October 5-7, 1977, pp 121-144.

⁶ Recent Advances in the Chemical Composition of Tobacco and Tobacco Smoke. Proceedings of American Chemical Society Symposium, New Orleans, La., March 20-25, 1977.