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Toxicological evaluation of cigarettes with two banded cigarette paper technologies

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Abstract

A tiered testing strategy has been employed to evaluate the potential of tobacco processes, ingredients, or technological developments to change the biological activity resulting from burning cigarette tobacco. The strategy is based on comparative chemical and biological testing. The introduction of banded cigarette papers in cigarettes to meet New York state "Fire Safety Standards for Cigarettes" constitutes an example of a technological development evaluated utilizing this tiered testing strategy that included a comparison of the chemical and biological effects of cigarettes with and without the banded cigarette paper technologies (BCPT) (representative of current marketed technologies). Specific testing included mainstream cigarette smoke chemistry studies; *in vitro* studies included genotoxicity (Ames and sister chromatid exchange) and cytotoxicity studies (neutral red); *in vivo* studies included a 13-week inhalation study in Sprague–Dawley rats and a 30-week dermal tumor promotion study in SENCAR mice. Collectively, data indicated that cigarettes with and without BCPT had a similar toxicological profile in this test battery.

Keywords: Cigarette; Banded; Cigarette paper; Toxicology; *In vitro*; *In vivo*

Introduction

R.J. Reynolds Tobacco Company (RJRT) has developed a tiered testing strategy to evaluate the potential for new ingredients, tobacco processes, and technological developments to change the biological activity of mainstream smoke (MS) or cigarette smoke condensate (CSC). This tiered testing strategy is based on the US Food and Drug Administration process of evaluating food additives and includes chemical and biological testing (Swauger et al., 2002; US FDA 1982, 1993). The introduction of new banded cigarette paper technologies (BCPT) to meet the New York state "Fire Safety Standards for Cigarettes" regulation constitutes an example of a technological development evaluated in such a tiered testing strategy (Part 429 to Title 19 NYCRR, 2003).

The objective of this paper is to summarize the stewardship test battery evaluating the new BCPTs. These BCPTs are representative of currently marketed

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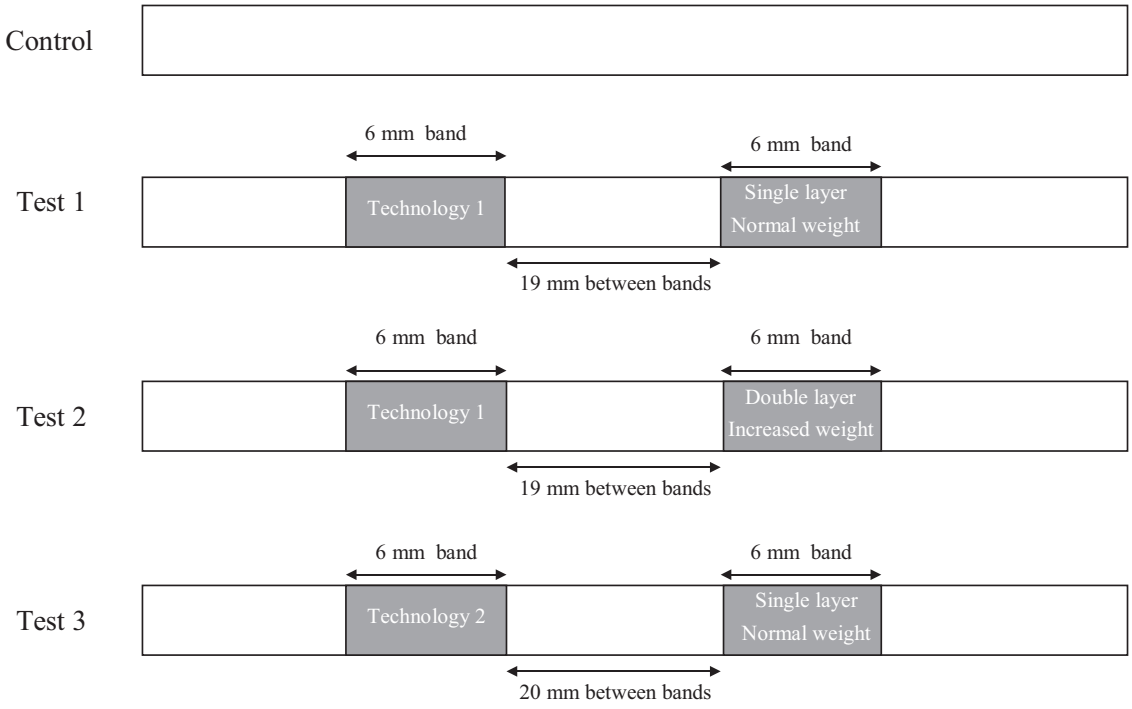

Fig. 1. Experimental design (cigarette papers from control and test cigarettes).

technologies. The test battery compared MS/CSC from cigarettes with (test) and without (control) the bands applied to cigarette papers. The test battery included: MS chemistry studies; *in vitro* studies (Ames, sister chromatid exchange (SCE), and neutral red); and *in vivo* studies (13-week inhalation study in Sprague–Dawley rats and 30-week dermal tumor promotion study in SENCAR mice).

Four cigarettes were tested (control and 3 test). The cigarette designs were similar except for the bands. Cigarettes were designed using a standard American tobacco blend (a blend of Burley, flue-cured and oriental tobacco). The control cigarette had no bands while the test cigarettes had different bands applied to the cigarette paper (Fig. 1). Two BCPTs were tested: (1) test 1 and test 2 cigarettes had one type of BCPT applied to the cigarette base paper as single- or double-layer bands, 6 mm wide and spaced 19 mm apart (with test cigarette 1 having single-layer bands, applied at one weight and test 2 having double-layer bands, applied at increased weight), and (2) test 3 cigarettes had another type of BCPT applied as single-layer bands, 6 mm wide and spaced 20 mm apart, applied at normal weight.

Materials and methods

Smoke chemistry study

Selected MS constituent yields were determined by standard methods (Borgerding et al., 1998; Chepiga et al., 2000). Cigarettes were machine-smoked under Federal Trade Commission (FTC) smoking conditions (35 ml puff volume, 2 s duration, 1 puff/min, butt length: overwrap + 3 mm) (CORESTA, 1969; Pillsbury et al., 1969; Bombick et al., 1997; ISO, 2000). Data were analyzed using a z-test with $p < 0.05$ (two tailed) or ANOVA, with Bonferroni-adjusted p values. The variability of the methods was estimated from analytical monitor data for each of the methods. The analytical monitor is a cigarette that is analyzed periodically to ensure that the test method within specifications. The analytical monitor consists of a large number of cigarettes (a "batch") produced during a single manufacturing run. The cigarettes are as uniform as it is practical to obtain under typical manufacturing conditions. When the monitor is initially introduced for use with a chemical test method, it goes through a qualification process. From the data generated during that period, the mean values for each analyte and the method standard deviation for each analyte are estimated. The standard deviation measured with the monitor cigarette takes into account within day and day-to-day method variability and the number of replicate analyses conducted on the sample.

Ames study

Cigarettes were machine-smoked and mainstream particulate matter was collected on Cambridge filter pads and extracted with dimethylsulfoxide (Bombick et al., 1997). CSCs were tested for mutagenicity in