Effect of Menthol Cigarettes on Biochemical Markers of Smoke Exposure Among Black and White Smokers*

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Study objectives: Black smokers have been reported to have higher serum cotinine levels than do white smokers, and have higher rates of most smoking-related diseases, despite smoking fewer cigarettes per day. Another striking racial difference is the preference for mentholated cigarettes among black smokers. The contribution of menthol to variability in biochemical markers of cigarette smoke exposure (end-expiratory carbon monoxide and serum cotinine) was evaluated in a biracial sample.

Design: Descriptive cross-sectional.

Setting: A university smoking research laboratory.

Participants: Sixty-five black and 96 white adult established smokers who were paid for their participation.

Measurements: Information was obtained through direct observation, self-report (interview and self-administered questionnaires), measurement of butts collected for a week, and laboratory analyses of the biochemical markers of exposure.

Results: Compared with the white smokers, the black smokers had significantly higher cotinine and carbon monoxide levels per cigarette smoked and per millimeter of smoked tobacco rod (both \( p < 0.001 \)). After adjusting for race, cigarettes per day, and mean amount of each cigarette smoked, menthol was associated with higher cotinine levels (\( p = 0.03 \)) and carbon monoxide concentrations (\( p = 0.02 \)).

Conclusions: The use of menthol may be associated with increased health risks of smoking. Menthol use should be considered when biochemical markers of smoke exposure are used as quantitative measures of smoking intensity or as indicators of compliance with smoking reduction programs. In addition, the effect of menthol on total “dose” should be considered in any efforts to regulate the amount of nicotine in cigarettes.

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Key words: carbon monoxide; cotinine; menthol; tobacco

Black smokers report smoking approximately 35% fewer cigarettes per day than do white smokers, yet these “lighter” smokers have higher rates of most smoking-related diseases. It is estimated that the years of potential life lost before age 65 years among black smokers is twice that of white smokers. Black smokers may have significantly higher lung cancer rates for any given level of smoking, an effect particularly marked among black women.

Another striking racial difference is the preference for menthol cigarette brands among black smokers. Up to 91% of young black female smokers and 87% of young black male smokers report smoking menthol cigarettes compared to 34% and 24% of white smokers, respectively. When all age groups are considered, approximately 76% of black smokers choose menthol cigarettes compared to around 23% of white smokers.

Menthol combustion produces carcinogenic compounds such as benzo[α]pyrenes. Kabat and Herbert were not able to demonstrate an association between the use of menthol cigarettes and the risks of lung, esophageal, or oropharyngeal cancers in a series of case-control studies. Those studies were hampered by the inclusion of few blacks and by the subjects’ relatively short-term use of menthol cigarettes. In a later study, Sidney and coworkers found a significant association of menthol use and lung cancer only among men. A finding of higher cotinine concentrations among menthol users could be a marker for higher

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levels of absorption of other components of the particulate phase of tobacco smoke.

Menthol cigarettes have become popular relatively recently. The domestic market share of menthol cigarettes increased from 16 to 28% between 1963 and 1978, and has remained at approximately 29% since that time. If the use of menthol cigarettes is associated with increased risk of smoking-related cancers, the full impact may not be detectable with epidemiologic methods before the next decade. We used laboratory methods to investigate the potential for menthol cigarettes to increase the total effective “dose” of tobacco smoke. Serum cotinine levels and expired air carbon monoxide concentrations were measured in a biracial group of established smokers, comparing the biochemical markers in habitual menthol and nonmenthol smokers.

Materials and Methods

Study Subjects

Subjects were recruited through advertising and personal contacts and were paid $50 for their participation. Eligible subjects were non-Hispanic whites and non-Hispanic blacks between the ages of 18 and 45 years. They smoked at least 5 cigarettes per day for at least 1 year, smoked only 1 brand and type of cigarette, and had not switched brands in at least 3 months, did not use any tobacco or nicotine replacement products, and were willing and able to give informed consent to participate. Black subjects were excluded if they identified themselves as being of other than African or American descent (i.e., Caribbean).

The study was reviewed and approved by the Institutional Review Board for the Protection of Human Subjects of the University of South Florida, Tampa. Written informed consent to participate was obtained from all subjects.

Procedures

The information used in the analyses was gathered through self-report (interview and self-administered questionnaire), observation, collected cigarette butts, and laboratory analyses. A brief structured telephone interview was used to determine eligibility and smoking patterns. Within 2 weeks, each subject came to the laboratory and completed a self-administered questionnaire. Information elicited during the interview and questionnaire included race, gender, age, age at initiation of regular smoking, inhalation frequency and depth, concomitant use of marijuana, time to first morning cigarette, and reported average amount of cigarette left unsmoked. Laboratory personnel examined the subjects’ cigarette packs for a complete description of cigarette brand and type. This was used to obtain machine-smoked tar and nicotine yields from the 1991 Federal Trade Commission (FTC) list and to confirm the use of mentholated cigarettes. An unsmoked cigarette from each subject’s pack was used to measure the cigarette length and length of the filter and overwrap in millimeters.

Subjects were given containers in which they were to deposit the butts of all cigarettes smoked for 1 week, with separate containers labeled for each day’s butts. The average amount smoked of each cigarette was calculated as the difference between the original length of the cigarette and the measured butt length divided by the total number of returned butts. Average total millimeters smoked per day was obtained by multiplying the reported cigarettes per day by the average millimeters of each cigarette smoked.

At the second laboratory visit, subjects were held in the laboratory for 1 h to assure smoking abstinence. At the end of the hour, blood was sampled by venipuncture for use in serum cotinine analyses. Subjects were shown to a quiet room where they watched an entertainment videotape while smoking one cigarette from their own supply. Only one person smoked in the room at a time. No talking was allowed during the test cigarette. Prior to smoking the test cigarette, and within 120 s, carbon monoxide concentrations were measured from end-expired air after 20 s of breath-holding, using a portable monitor (Vitalograph BreathCo; Vitalograph Inc; Lenexa, Kan). Within 120 s of completing the test cigarette, expired air carbon monoxide was again measured. The postcigarette measurement was used in these analyses, reflecting both the presmoking level (after at least 1-h abstinence) and the boost from the single cigarette.

Serum samples for cotinine analyses were frozen in an ultralow freezer until analyzed by the University of Minnesota Lipid Research Clinic Laboratory. Measurement of serum cotinine was by gas chromatography. Fifteen percent of samples were split for blind reliability testing. The mean difference in split samples was 1.6 ng/mL (95% confidence bounds, 6.15 and 9.35).

Linear regression analyses were used to define the contributions of cigarette type (menthol, cigarette length, machine-smoked tar and nicotine levels), smoking patterns (reported cigarettes per day, mean amount of each cigarette smoked, self-reported inhalation frequency and depth, concomitant use of marijuana), and demographic characteristics (gender, race, age, years of smoking) to serum cotinine levels and expired-air carbon monoxide concentrations. Nonlinear models were explored, but they were not found to increase the amount of variability that could be predicted by linear models.

Results

Of 169 subjects completing the study, 8 were eliminated from the analyses. Six did not have serum cotinine levels because of inadequate blood samples and two had more than one cigarette brand among their collected butts. The remaining 161 subjects consisted of 37 black women, 28 black men, 50 white women, and 46 white men.

Comparisons between black and white smokers are shown in Table 1. Black smokers were more likely to smoke menthol cigarettes and used cigarettes with higher machine-smoked tar and nicotine levels. Blacks reported smoking fewer cigarettes per day, but they smoked significantly more of each cigarette. The total mean millimeters smoked per day was higher for white smokers. Black smokers had significantly higher cotinine levels (mean, 485.1 ng/mL) compared to white smokers (mean, 354.3 ng/mL). The mean difference was significant (p=0.0005). Mean expired-air carbon monoxide after the test cigarette was 38.4 ppm among black smokers and 37.5 ppm among whites. The difference was not significant (p=0.73).

Serum Cotinine Levels

As expected, the most important predictors of serum cotinine levels were cigarettes per day and the mean amount of each cigarette smoked (both measures of “dose” of smoke inhaled per day) and race. Serum cotinine levels were significantly higher among menthol
users (478.2 ng/mL) compared to nonmenthol users (349.1 ng/mL; p value for the difference=0.0005). The mean cotinine level was 84.5 ng/mL higher in menthol cigarette smokers after adjusting for race, cigarettes per day, and the mean amount of each cigarette smoked (p=0.03) (Table 2).

\[ \text{Table 1—Characteristics of the Sample of Established Smokers} \]

<table>
<thead>
<tr>
<th></th>
<th>Black Smokers</th>
<th>White Smokers</th>
<th>Difference (95% CI)*</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>65</td>
<td>96</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female, %</td>
<td>57</td>
<td>52</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean age, yr (SD)</td>
<td>35.1 (6.3)</td>
<td>31.3 (8.4)</td>
<td>3.8</td>
<td>0.001</td>
</tr>
<tr>
<td>Mean years of smoking (SD)</td>
<td>16.3 (7.6)</td>
<td>14.9 (8.4)</td>
<td>1.4</td>
<td>0.27</td>
</tr>
<tr>
<td>Using menthol cigarettes, %</td>
<td>83.1</td>
<td>22.9</td>
<td>60.2</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean cigarettes per day (SD)</td>
<td>16.2 (6.6)</td>
<td>23.1 (10.2)</td>
<td>6.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean mm of tobacco rod smoked (SD)</td>
<td>61.7 (7.2)</td>
<td>55.9 (7.9)</td>
<td>5.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean total mm smoked per day (SD)</td>
<td>1,009.8 (444.8)</td>
<td>1,299.6 (396.7)</td>
<td>298.7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean nicotine yield, mg/cigarette (SD)</td>
<td>1.10 (0.17)</td>
<td>0.97 (0.24)</td>
<td>0.13</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean cigarette length, mm (SD)</td>
<td>95.6 (8.7)</td>
<td>92.7 (8.2)</td>
<td>2.9</td>
<td>0.03</td>
</tr>
<tr>
<td>Mean cotinine, ng/mL (SD)</td>
<td>491.4 (248.9)</td>
<td>354.8 (216.4)</td>
<td>136.6</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean carbon monoxide, ppm (SD)</td>
<td>38.4 (15.7)</td>
<td>37.5 (16.5)</td>
<td>0.9</td>
<td>0.73</td>
</tr>
</tbody>
</table>

*CI=confidence interval.

The mean serum cotinine level for black smokers in this sample was 32.9 ng/mL per cigarette smoked (median, 29), compared to a mean of 16.1 (median, 14.1) among white smokers. The use of menthol contributed to this discrepancy, accounting for 23% of the black-white difference in cotinine levels.

We offer mechanisms by which menthol use may increase serum cotinine levels or expired-air carbon monoxide concentration. The menthol smokers in our laboratory reported, anecdotally, an increased feeling of "wetness" in the mouth with menthol cigarettes. Duner-Engström and coworkers\(^25\) reported that chewing menthol gum gave a significantly higher amount of stimulated saliva compared to nicotine chewing gum or a placebo for nicotine gum. Most of the body burden of nicotine is delivered by way of inhalation into the lungs, but a part of each puff is held in the mouth. If menthol delivered by way of a burning cigarette also increases salivary flow (relative to nonmenthol cigarettes), the result may be an increase in dissolution in the mouth of the particulate phase of tobacco smoke. Because saliva raises pH, this would not be a particularly efficient delivery mechanism, but it may contribute somewhat to total nicotine absorption. It cannot explain the increased carbon monoxide levels.

It is known that 1-menthol increases drug solubility in some vehicles and increases skin permeability for some drugs.\(^26,27\) It is not known whether menthol could increase transport of the products of smoking across oral membranes.

Differences in puffing and inhaling patterns could
account for higher cotinine and carbon monoxide levels. Menthol stimulates cold receptors, providing a sensation of coolness, possibly resulting in larger puff volumes and deeper, longer inhalation of cigarette smoke. McCarthy and coworkers\textsuperscript{25} tested intrindividual differences in smoking behavior with menthol and nonmenthol cigarettes and found a significant increase in puff volume and puff frequency with a mentholated brand. In other studies, the same authors reported no changes in number of puffs taken from menthol cigarettes compared to regular,\textsuperscript{20} and decreased puff volume, but increased carbon monoxide absorption with menthol cigarettes.\textsuperscript{30} Inhalation and breath-holding may be modified by menthol. However, there are no good self-report measures for these aspects of smoking topography.\textsuperscript{24}

The higher cotinine and carbon monoxide levels among users of menthol cigarettes has several implications. The preference for menthol cigarettes is unequally distributed among ethnic and gender groups; thus the use of biochemical markers of exposure, without adjusting for menthol use, may result in inaccurate estimates of smoking levels or inappropriate assumptions about the lack of accuracy of self-reports of daily smoking rates in some groups. These results may also have regulatory implications. There have been recent federal level suggestions that the amount of nicotine allowed in manufactured cigarettes should be controlled.\textsuperscript{31} We found that menthol increases cotinine, the major metabolite of nicotine. To achieve equal availability of nicotine, it may be necessary to require lower concentrations of nicotine in menthol cigarettes. Finally, in addition to the health risk of functionally “stronger” cigarettes, with greater availability of nicotine and carbon monoxide, the higher levels of these biochemical markers may be indicators of higher levels of absorption of other components of the gas and particulate phases of tobacco smoke.

\textbf{REFERENCES}

12 US Department of Health and Human Services. Tobacco use in 1986, methods and basic tabulations from adult use of tobacco survey. Public Health Service, Centers for Disease Control, Center for Chronic Disease Prevention and Health Promotion, Office on Smoking and Health, Atlanta, 1986
16 Kabat GC, Hebert JR. Use of mentholated cigarettes and oropharyngeal cancer. Epidemiology 1994; 5:183-88
22 English PB, Eskenazi B, Christiansen RE. Black-white differences in serum cotinine levels among pregnant women and subsequent effects on infant birthweight. Am J Public Health 1994; 84:1439-43
31 H. R. 2147, Fairness in Tobacco and Nicotine Regulation Act of 1993 (May 18, 1993)

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Clinical Investigations