*Pharmacology Biochemistry & Behavior*, Vol. 40, pp. 229–235. © Pergamon Press plc, 1991. Printed in the U.S.A. 0091-3057/91 $3.00 + .00

# Hormonal and Subjective Effects of Smoking the First Five Cigarettes of the Day: A Comparison in Males and Females

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Received 15 April 1991

MELISKA, C. J. AND D. G. GILBERT. *Hormonal and subjective effects of smoking the first five cigarettes of the day: A comparison in males and females*. PHARMACOL BIOCHEM BEHAV **40**(2) 229–235, 1991.—The effects of smoking normal-nicotine-delivery cigarettes on serum cortisol, plasma beta-endorphin (BE), and mood were measured in 8 male and 8 female smokers; 8 male and 8 female nonsmokers served as sham-smoking controls. Smoking five cigarettes of the smokers' usual type after overnight deprivation, either ad lib or via a quantified smoke delivery system, produced small but reliable elevations in serum cortisol concentrations; BE was elevated somewhat after two, but not after four or five cigarettes. Smoking-induced elevations in serum cortisol were correlated with decreases in self-reported drowsiness after two and five cigarettes. Additionally, female smokers reported more drowsiness at baseline and after smoking nicotine-free cigarettes than did male smokers or female nonsmokers. Results suggest that smoking-induced elevations in serum cortisol, which persist for at least the first five cigarettes of the day, may modulate the arousing effects of smoking under conditions of low arousal. Also, nicotine-deprived female smokers may experience subnormal arousal compared to male smokers or female nonsmokers.

Nicotine Smoking Cortisol Beta-endorphin Affect Mood Arousal Gender Reinforcement

PREVIOUS studies have suggested that cigarette smoking may elevate peripheral levels of beta-endorphin (BE) and cortisol in certain circumstances (20, 27, 33). These findings have prompted the suggestion that elevation of peripheral neuromodulator levels may modulate the reinforcing or rewarding effects of smoking (10, 17, 21). However, reliable elevations in peripheral BE and cortisol have been reported only after rapid smoking of one or two high-nicotine cigarettes (typically in excess of 2.5 mg FTC-estimated nicotine delivery). It has not been adequately established whether normal smoking of cigarettes with more typical nicotine deliveries (0.7–1.0 mg) results in neuromodulator elevations when cigarettes are smoked at a casual pace. Furthermore, since tolerance to many of the effects of nicotine ingestion occurs rapidly (2,18), the extent to which neuromodulator elevations are sustained after the smoking of more than two cigarettes is not known.

Few studies of the neuroendocrine effects of cigarette smoking have carefully assessed the subjective and mood-modulating effects of the smoking procedures employed. Yet, such assessments are important because administering doses of nicotine that are too low to reliably alter mood may not produce physiological effects characteristic of normal smoking. Similarly, administering higher than normal nicotine doses may cause subjective distress and physiological concomitants which are atypical. For example, in one of the few studies in which subjective state associated with smoking-induced changes in neuromodulators was assessed with a psychometric instrument (12), high-nicotine/rapid smoking procedures produced malaise (self-reported nausea, sickness, and unpleasantness) in some subjects, thereby raising questions about whether associated physiological effects are representative of normal smoking. Other work (27) confirms that rapid smoking of high-nicotine cigarettes produces nausea in many, but not all smokers who experience smoking-induced BE and cortisol elevations. Therefore, in the present study, a previously validated psychometric instrument was used to evaluate the mood-modulating effects of the smoking procedures, so that their relationship to the physiological effects produced could be evaluated.

Smokers vary greatly in the amount of nicotine they obtain from ad lib smoking due to variations in the manner in which they smoke (16). Thus different physiological effects may be produced on different occasions, even with the same individual smoking the same cigarette. Therefore, in addition to naturalistic smoking, it is informative to control the dose of nicotine delivered in order to make precise quantitative estimates of the relationship of a particular dose to particular subjective and physiological effects, and to determine whether a controlled dosing procedure produces different effects than ad lib smoking. To achieve this in the present study we used a quantified smoke delivery system (11) which delivers controlled doses of smoke/nicotine in a reliable fashion.

The present study was designed to test whether the smoking

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of typical cigarettes (e.g., those with FTC-estimated nicotine deliveries of 0.7 to 1.0 mg) under relaxed conditions elevates serum cortisol and plasma BE. A second goal was to determine whether hormonal and subjective changes produced by the first two cigarettes of the day are sustained after four or five cigarettes. A third goal was to compare the effects of ad lib smoking and smoking via our Quantified Smoke Delivery System (QSDS). Finally, we wanted to assess the degree to which smoking-induced changes in blood levels of cortisol and BE were related to changes in self-reported mood.

## METHOD

### *Subjects*

Subjects were 16 males and 16 females, aged 21–35 years. Half were nonsmokers and half were habitual smokers of at least 15 cigarettes per day, for at least one year. Male smokers were selected from a pool of smokers of cigarettes ranging from 0.9 to 1.1 mg FTC-estimated nicotine delivery (typically Camel Filter®, Winston® or Marlboro® regular brands). Female smokers were selected from a pool of smokers of cigarettes of 0.6 to 0.8 mg FTC-estimated nicotine delivery (typically Winston Lights®, Camel Lights® or Marlboro Lights®). Additionally, females selected for the study were all users of oral contraceptives, and all testing was performed between days 7 and 21 after the end of their last menstrual period.

### *Apparatus*

Exhaled carbon monoxide was monitored with a Catalyst Research CO Monitor, Model 1000. The quantified smoke delivery system [QSDS; (11)] takes 2-second duration, 35 cc, sinusoidal-shaped puffs from a cigarette, drawn at 30-s intervals by a 50-cc glass syringe. The cigarette is connected to the syringe by means of a piece of Penrose® drain tubing and a three-way solenoid valve. The filter end of the cigarette is placed into one end of the tubing, while the other end of the tubing is connected to the valve. Operating the motor draws into the syringe a puff of smoke which is expelled over 0.5 s into the smoker's mouth. The smoker immediately inhales the smoke and holds it in her/his lungs until a signal light goes out, five seconds after inhalation.

### *Procedure*

During a late-afternoon orientation session subsequent to ad lib smoking, subjects were informed about the nature of the study and a sample of their expired breath was monitored for carbon monoxide concentration. Smokers whose afternoon CO levels were below 20 ppm were presumed to smoke too little or to be noninhalers, and were excluded from participation. To familiarize subjects with the experimental procedure, an electrode cap was applied and the EEG recording procedure was simulated. Smoking subjects then practiced smoking both a tobacco cigarette (a Camel Filter® for the males, a Vantage® for the females) and a nicotine-free, herbal cigarette (HoneyRose®) by means of the QSDS. Nonsmokers practiced "sham smoking," going through all the motions of smoking with the QSDS, except no cigarette smoke was administered. Subjects were then instructed to eat breakfast as usual and to consume their usual amount of caffeine before coming to the laboratory for testing. Smokers were also required to abstain from smoking cigarettes or using any tobacco products after 11:59 p.m. before each of the test days. Furthermore, all subjects were informed they would be required to provide a breath sample to demonstrate that they had in fact refrained from smoking on each of the test mornings.

Subjects participated in four test sessions lasting approximately four hours each, beginning at 8:00 a.m. Overnight abstinence was verified by measuring expired CO levels at the start of each test session, and confirmed by subsequent baseline plasma nicotine assays (see the Results section). Subjects whose CO levels exceeded 50% of their afternoon control levels were excused from participation and rescheduled. Prior to testing, a sterile indwelling catheter was inserted into the medial antecubital vein of the subject's arm, and a heparin lock was set. After catheter insertion, recording electrodes for measuring EKG were attached to the subject's chest and an EEG cap was attached. (Data from electrophysiological measures will be reported in a future publication.)

During each test session each smoker smoked five cigarettes under relatively relaxed conditions, while watching moderately interesting videotapes. During one session each smoked ad lib, as much or as little as desired of one cigarette of her/his accustomed brand, during each of five smoking breaks scheduled at 30-min intervals. In the remaining three sessions smokers smoked five cigarettes, again separated by 30-min intervals, via the QSDS. To assess response reliability, during two of these sessions subjects smoked a typical nicotine delivery cigarette (1.0 mg nicotine for males and 0.7 mg nicotine for females; the lower values for females represent an adjustment for lower body weights among female subjects). Puffs of smoke of 35 cc volume were delivered into the smoker's mouth at 30-s intervals, until the cigarette char line reached 3 mm above the filter overwrap. During the other QSDS session subjects smoked placebo (HoneyRose®) cigarettes under the same QSDS puff rate and puff volume conditions. All smokers participated under all smoking conditions, the order of testing being counterbalanced according to a Latin square design.

Subjective, physiological (heart rate and EEG), and neuromodulator (BE and cortisol) measures were assessed in both the ad lib and QSDS smoking conditions. The Fagerstrom Tolerance Questionnaire, Shiffman Withdrawal Questionnaire, Eysenck Impulsivity Inventory, and Jenkins Activity Survey (Form T) were administered during the 60-minute adaptation period at the start of each experimental session after insertion of the indwelling catheter. (Data from these instruments will be reported in a future publication.)

Blood samples (10.5 ml each) were withdrawn on six occasions: 55 min after catheter insertion (baseline); five min before lighting of the first cigarette; seven and eighteen min after completion of the second cigarette; five min before lighting of the fifth cigarette; and seven and eighteen min after completion of the fifth cigarette. Samples were placed on ice and centrifuged under refrigeration at $750 \times g$ for 20 min within 90 min of collection. Plasma and serum were decanted and stored in a freezer at $-90°C$ until shipment on dry ice for assays of cortisol and nicotine. Assays of plasma nicotine and caffeine were conducted in the laboratory of Dr. Neil Benowitz of San Francisco General Hospital, San Francisco, CA; assays of serum cortisol were carried out in the laboratory of Dr. Nancy Haley of the American Health Foundation, Valhalla, NY; assays of plasma beta-endorphin were performed by Dr. Agnieszka Szary in the Central Hybridoma Laboratory of Southern Illinois University. Baseline (presmoking) values for nicotine, caffeine, cortisol, and BE were based on samples drawn five min before cigarette 1. Because smoking-induced elevation of cortisol typically lags behind maximal elevations of proopiomelanocortin-derived hormones by 10–30 min (27,28), cortisol assays were based on samples drawn 18 min after cigarettes 2 and 5. BE, which is maximally elevated within a few min of cigarette completion (28), was assayed 7

min after cigarettes 2 and 5. Assays of blood nicotine and neuromodulator concentrations made from samples drawn five min before cigarette 5 were assigned to cigarette 4.

Each subject's mood was assessed by means of the Feeling State Questionnaire [FSQ; (13)] five min before the lighting of the first and fifth cigarettes, and again 18 min after the end of the second and fifth cigarettes. The FSQ is a self-report instrument that requires the subject to indicate on 11-point Likert scales (0 = none to 10 = extreme) to what degree each of 19 subjective states is experienced. Items include: pleasantness, tension, happiness, nausea, worry, drowsiness, sickness, energy, hand sweating, dizziness, heart pounding, alertness, relaxation, fear, light-headedness, anger, unpleasantness, sadness, and body sweating. EEG, heart rate, blood samples, and subjective measures were obtained before smoking (baseline), after the second cigarette, immediately prior to the fifth cigarette, and after the fifth cigarette.

To provide additional baseline control information, 16 nonsmokers (eight males and eight females) participated in one nonsmoking session. Nonsmokers followed the same QSDS smoking procedure as the smokers but did not actually inhale smoke; during their sole test session they participated in all of the other experimental tasks and provided subjective reports, physiological data, and blood samples at the same intervals as the smokers.

## RESULTS

For preliminary comparisons, data were collapsed across the two QSDS smoking conditions for analyses on variables of interest. Individual plasma nicotine concentrations were low at the presmoking baseline (range: <1.0–2.9, median = 1.6 ng/ml) confirming that all participants complied with instructions not to smoke on the mornings before coming to the lab for testing. Smoking five tobacco cigarettes elevated plasma nicotine levels in a dose-related fashion in both males and females (Fig. 1). Overall, plasma nicotine boosts were marginally lower after QSDS than after ad lib smoking, $F(1,14) = 4.02$, $p = 0.065$. The mean plasma nicotine boost obtained by females was also marginally lower than that obtained by males, $F(1,14) = 2.93$, $p = 0.11$. This difference was associated with a smaller mean plasma nicotine boost obtained by females with QSDS smoking, $F(1,14) = 6.99$, $p = 0.019$, and may reflect the fact that females smoked 0.7 mg, while males smoked 1.0 mg FTC-estimated nicotine delivery cigarettes via the QSDS; after ad lib smoking of 0.7 mg cigarettes, females did not differ in plasma nicotine boost from males smoking 1.0 mg cigarettes, $F(1,14) = 0.5$, $p > 0.05$. Thus plasma nicotine boosts in males and females were quite similar after ad lib smoking, and somewhat lower in females with QSDS smoking. However, the physiological and subjective consequences of these gender and mode-of-smoking differences proved to be relatively small, as reported below.

A three-factor (Gender × Smoking Status × Time) ANOVA comparing habitual smokers with nonsmokers showed that after smoking nicotine-free cigarettes, smokers (mean = 16.3 μg/dl) did not differ in serum cortisol concentrations from nonsmokers who "sham" smoked (mean = 15.8 μg/dl), $F(1,28) = 0.17$, $p = 0.68$. However, serum cortisol was approximately 2.5 times higher in females (mean = 22.9 μg/dl) than in males (mean = 9.1 μg/dl) across the testing period, $F(1,28) = 120.8$, $p = 0.0001$. As expected, mean cortisol concentrations decreased appreciably in both sham and nicotine-free smokers during the morning test session, $F(3,84) = 34.11$, $p = 0.001$. Separate, within-subjects comparisons on smokers alone showed that smoking nicotine cigarettes, either ad lib or via the QSDS, produced statistically equivalent effects on cortisol levels; after two, four, or five cigarettes, QSDS and ad lib smoking were not significantly different, $F(1,14) = 1.18$, 0.07, and 0.08, respectively; all $ps > 0.05$. Therefore, for ease of presentation, results were averaged across nicotine (QSDS and ad lib) smoking conditions for comparisons with nicotine-free smoking. A Gender × Dose × Time ANOVA performed on changes from baseline showed that cortisol concentrations were higher after two, $F(1,14) = 9.61$, $p = 0.008$, four, $F(1,14) = 8.55$, $p = 0.012$, and five nicotine cigarettes, $F(1,14) = 8.69$, $p = 0.011$, than after the same numbers of nicotine-free cigarettes (Fig. 2A). While overall cortisol levels decreased more in females than in males, $F(1,14) = 16.67$, $p = 0.002$, the Dose × Gender interaction was not significant ($p > 0.05$), suggesting that smoking nicotine cigarettes attenuated the normal diurnal decrease in cortisol levels in both males and females.

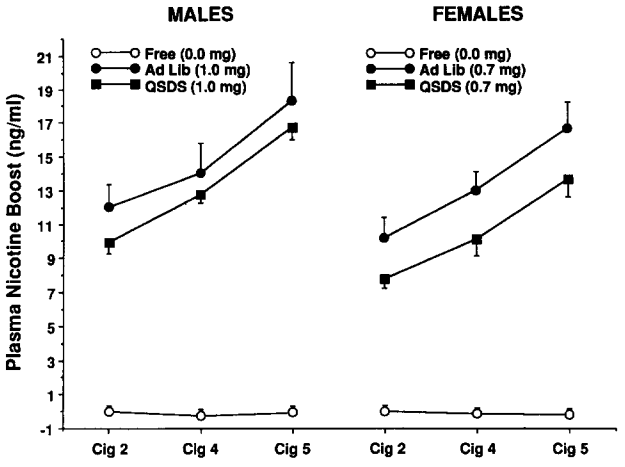


FIG. 1. Effects of different smoking conditions on plasma nicotine boosts after two, four, and five cigarettes in male and female smokers. Data points represent means of N = 8 male and 8 female subjects. Vertical bars represent ± S.E.M.

As with the cortisol analyses, a Gender × Smoking Status × Time ANOVA comparing the plasma BE concentrations of habitual smokers with nonsmokers showed no differences between smokers who smoked nicotine-free cigarettes (mean = 4.8 pmol/l) and nonsmokers who sham smoked (mean = 4.2 pmol/l), $F(1,28) = 0.8$, $p = 0.38$. However, in contrast to the cortisol findings, plasma BE was higher in males (mean = 5.5 pmol/l) than in females (mean = 3.5 pmol/l) across the testing period, $F(1,28) = 11.4$, $p = 0.003$. As with the cortisol data, QSDS and ad lib smoking produced comparable effects on BE concentrations, $F(1,14) = 0.66$, 0.92, and 0.75, respectively, all $ps > 0.05$, so results were averaged across QSDS and ad lib smoking conditions (Fig. 2B). A Gender × Dose × Time ANOVA performed on changes from baseline showed that changes in BE levels were small, and only marginally higher, $F(1,14) = 2.67$, $p = 0.13$, after smoking nicotine cigarettes than after nicotine-free cigarettes (mean = +0.35 vs. −0.28 pmol/l, respectively). Since a greater effect was predicted after the first two cigarettes than after subsequent ones, individual paired comparisons were performed, which showed that BE levels after two nicotine cigarettes were higher than after two nicotine-free cigarettes, $F(1,14) = 6.82$, $p = 0.021$; however, no significant differences were found after four, $F(1,14) = 0.71$, $p = 0.42$, or five nicotine cigarettes, $F(1,14) = 0.18$, $p = 0.68$, relative to the same numbers of nicotine-free cigarettes (Fig. 2B); Gender × Dose interactions were nonsignificant after two, four, and five cigarettes (all $ps > 0.05$). Thus the small increase in BE found after two cigarettes did not

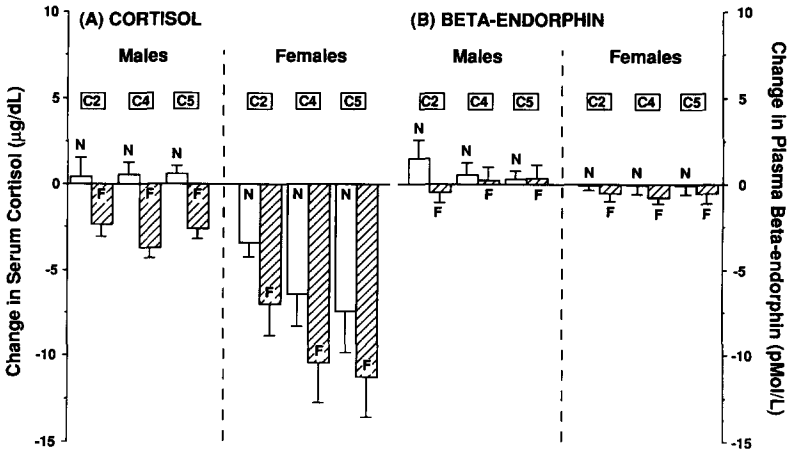


FIG. 2. Effects of smoking nicotine-free (F) and nicotine (N) cigarettes on (A) serum cortisol and (B) plasma beta-endorphin. Data bars represent mean changes from baseline in N = 8 male and 8 female smokers, after two (C2), four (C4), and five (C5) cigarettes. Vertical bars represent ± S.E.M.

persist after four or five cigarettes.

*Effects of Smoking Nicotine Cigarettes on Subjective Measures: Relationship to Blood Cortisol and BE Concentrations*

Subjects' responses on the Relaxation item of the FSQ indicated that they felt relatively relaxed at baseline, prior to smoking (mean = 6.27, range 3–9). ANOVAs performed on subscale items of the FSQ, administered before and after smoking cigarettes 2, 4, and 5, showed that smoking nicotine cigarettes produced no significant effects on self-reported measures of malaise (nausea, sickness, or unpleasantness; all $ps > 0.05$), suggesting that the nicotine doses administered were well tolerated. The effects of nicotine smoking were small for all of the FSQ items except for those relating to arousal (i.e., energy, alertness, drowsiness, and relaxation). Effects were particularly prominent for self-reported drowsiness, which decreased from baseline more after the smoking of nicotine cigarettes than after nicotine-free cigarettes, $F(1,14) = 14.76$, $p = 0.002$. However, comparisons of smokers and nonsmokers (Fig. 3) revealed an unexpected interaction between Gender and Smoker-Nonsmoker Status, $F(1,28) = 11.78$, $p = 0.002$. Prior and subsequent to smoking nicotine-free cigarettes female smokers reported much more drowsiness than male smokers, $F(1,28) = 14.56$, $p = 0.001$. Furthermore, after placebo smoking, female smokers reported more drowsiness than female nonsmokers who "sham" smoked, $F(1,28) = 18.52$, $p = 0.001$; male placebo smokers did not differ from male (sham) nonsmokers, $F(1,28) = .30$, $p = 0.59$. The difference in baseline drowsiness scores of female smokers in the nicotine-free and nicotine conditions was not significant, $F(1,14) = 1.99$, $p = 0.18$. A separate Gender × Dose × Time ANOVA on data obtained from smokers revealed a significant Dose × Time interaction, $F(3,42) = 7.52$, $p = 0.002$; analyses of simple effects showed that smoking nicotine cigarettes reduced drowsiness after two, $F(1,14) = 11.45$, $p = 0.005$, four, $F(1,14) = 17.31$, $p = 0.001$, and five cigarettes, $F(1,14) = 14.41$, $p = 0.002$. Furthermore, while females remained drowsier than their male counterparts after two nicotine cigarettes, $F(1,14) = 6.79$, $p = 0.021$, they were no longer significantly different from male smokers or female nonsmokers who "sham" smoked, after four or five nicotine cigarettes (all $ps > 0.05$). Thus female smokers reported feeling drowsier than female nonsmokers before, but not after smoking tobacco cigarettes.

Since caffeine consumption may influence subjective arousal, a Gender × Smoker Status ANOVA was performed on baseline plasma caffeine levels. Main effects of Gender, $F(1,28) = 1.15$, Smoker Status, $F(1,28) = 0.54$, and their interaction, $F(1,28) = 1.37$, were all nonsignificant ($ps > 0.05$); thus, subgroups did not differ in plasma caffeine levels prior to testing, suggesting that the observed differences in arousal were not associated with differential consumption of caffeine.

To further explore relationships between changes in blood neuromodulator concentrations and mood, tests of intercorrelations of smoking-induced neuromodulator changes and FSQ affective measures were performed. As with the results reported above, results from QSDS and ad lib smoking were averaged for these analyses. To control for potential confounds associated with baseline differences between males and females in cortisol,

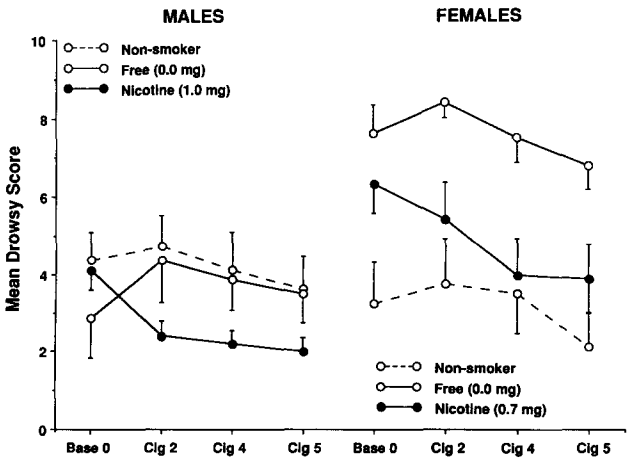


FIG. 3. Effects of sham and nicotine-cigarette smoking on self-reported drowsiness in nonsmokers and habitual smokers. Data points represent means for N = 8 subjects per group. Vertical bars represent ± S.E.M.

TABLE 1

PARTIAL CORRELATIONS RELATING CHANGES (FROM BASELINE) IN FSQ-RELATED MEASURES OF AROUSAL AND MALAISE TO CHANGES IN BLOOD CONCENTRATIONS OF CORTISOL, BETA-ENDORPHIN, AND NICOTINE AFTER TWO AND FIVE CIGARETTES, CONTROLLING FOR GENDER AND HORMONAL CHANGES ASSOCIATED WITH NICOTINE-FREE SMOKING

| | Cortisol | | Beta-Endorphin | | Nicotine | |
|---|---|---|---|---|---|---|
| | Cig2 | Cig5 | Cig2 | Cig5 | Cig2 | Cig5 |
| Drowsy | **−.68** | **−.50** | **−.52** | −.33 | .14 | −.28 |
| | **(0.005)** | **(0.041)** | **(0.036)** | (0.136) | (0.325) | (0.182) |
| Relaxed | −.07 | **−.71** | .03 | −.05 | **.51** | −.30 |
| | (0.414) | **(0.003)** | (0.459) | (0.434) | **(0.039)** | (0.161) |
| Energy | .41 | **.72** | .33 | .27 | −.31 | .32 |
| | (0.081) | **(0.003)** | (0.138) | (0.183) | (0.150) | (0.142) |
| Alert | **.55** | .06 | .33 | .22 | −.35 | −.05 |
| | **(0.026)** | (0.424) | (0.133) | (0.230) | (0.123) | (0.439) |
| Sick | .24 | **.70** | .04 | **.53** | −.16 | .24 |
| | (0.210) | **(0.004)** | (0.442) | **(0.032)** | (0.295) | (0.220) |
| Arousal* | **.53** | **.51** | .27 | .23 | −.39 | .25 |
| | **(0.032)** | **(0.039)** | (0.185) | (0.230) | (0.095) | (0.208) |
| Malaise† | .26 | **.63** | .25 | .38 | −.37 | .22 |
| | (0.196) | **(0.011)** | (0.201) | (0.102) | (0.105) | (0.238) |

*Arousal = Sum z-transformed scores: (Alert + Energy − Drowsy − Relaxed).
†Malaise = Sum z-transformed scores: (Nausea + Sick + Unpleasant). Significant correlations are presented in boldface; *p*-values (one-tailed) in parentheses.

BE, and subjective measures, and for changes produced by smoking nicotine-free cigarettes, correlations were calculated partialling on Gender and changes occurring during nicotine-free smoking conditions. Partial correlations between plasma nicotine boosts and cortisol changes, and between plasma nicotine boosts and BE changes after two, four, and five cigarettes were small and nonsignificant ($ps > 0.05$). While smoking-induced changes in cortisol were positively correlated with changes in BE after two cigarettes ($r = .65$, $p = 0.009$), correlations were nonsignificant after four and five cigarettes ($ps > 0.05$). This independence of cortisol and BE concentrations after five cigarettes is consistent with the finding reported above of persistent cortisol elevations in the absence of BE elevation after cigarettes four and five.

As Table 1 indicates, smoking-induced changes in cortisol and BE were moderately correlated with changes in some affective measures after two nicotine cigarettes. Consistent with results reported above, increased cortisol concentration correlated significantly with reductions in drowsiness and relaxation, with increases in energy and alertness, and with a composite measure of arousal (Energy + Alertness − Drowsiness − Relaxation) after two and five cigarettes. However, self-reported sickness and a composite measure of Malaise (Nausea + Sickness + Unpleasantness) were also correlated significantly with changes in cortisol, and to a lesser extent with changes in BE and nicotine. Therefore, correlations between changes in cortisol and BE and the various arousal measures were calculated as before with the addition of malaise and nicotine boost as controlling variables. The various measures of arousal remained correlated with neuromodulator elevations (Table 2) even after controlling for the effects of malaise, supporting the conclusion that neuromodulator-associated arousal changes occurred independently of changes in malaise; furthermore, while change in drowsiness was associated with change in BE after 2 cigarettes, the various arousal measures were more closely related to changes in cortisol after both two and five cigarettes. Thus, smoking-induced elevations in cortisol, and to a lesser degree, BE, were associated with increases in self-reported arousal.

## DISCUSSION

Serum cortisol normally reaches its maximal elevation at night, during sleep, and declines throughout the day in smokers and nonsmokers (3). Our earlier work (12) suggested that smoking two 1.0 mg nicotine cigarettes under relaxing conditions might elevate afternoon serum cortisol levels modestly. Results of the present study indicate that smoking five tobacco cigarettes

TABLE 2

PARTIAL CORRELATIONS RELATING CHANGES (FROM BASELINE) IN FSQ-RELATED MEASURES OF AROUSAL TO CHANGES IN BLOOD CONCENTRATIONS OF CORTISOL AND BETA-ENDORPHIN AFTER TWO AND FIVE CIGARETTES, CONTROLLING FOR GENDER, HORMONAL CHANGES ASSOCIATED WITH NICOTINE-FREE SMOKING, PLASMA NICOTINE BOOST AND MALAISE

| | Cortisol | | Beta-Endorphin | |
|---|---|---|---|---|
| | Cig2 | Cig5 | Cig2 | Cig5 |
| Drowsy | **−.90** | **−.52** | **−.65** | −.36 |
| | **(0.001)** | **(0.050)** | **(0.016)** | (0.138) |
| Relaxed | .01 | **−.52** | .25 | .07 |
| | (0.493) | **(0.050)** | (0.229) | (0.424) |
| Energy | .40 | **.63** | .24 | .14 |
| | (0.113) | **(0.020)** | (0.235) | (0.344) |
| Alert | **.63** | .26 | .31 | .27 |
| | **(0.019)** | (0.222) | (0.275) | (0.210) |
| Arousal* | **.63** | .46 | .25 | .18 |
| | **(0.019)** | (0.079) | (0.227) | (0.294) |

*Arousal = Sum z-transformed scores: (Alert + Energy − Drowsy − Relaxed). Significant correlations are presented in boldface; *p*-values (one-tailed) are in parentheses.

under relaxing conditions, either with a controlled-dosing (QSDS) or ad lib procedure, reduces the morning decline in serum cortisol that normally occurs between 9:00 a.m. and noon. While smoking-induced elevation of serum cortisol has been reported previously [(33,34); see (9) for review], not all studies have confirmed this finding [e.g., (3,29)]. Methodological differences relating to levels of quiescence of the subjects tested, small sample sizes, or tolerance to nicotine-induced cortisol elevations after several hours of smoking (3) may account for discrepancies among studies. However, if serum cortisol elevation is a reliable correlate of normal cigarette smoking under relaxed, quiescent conditions as the present results suggest, it may be an important biological mediator of nicotine's behavioral, and possibly reinforcing, actions (10). For example, the observed increases in subjective arousal associated with smoking-induced cortisol changes may be a cortisol-mediated reinforcing effect of smoking under low-arousal conditions. A report that exogenous cortisol administration enhances electrocortical and subjective arousal (4) supports the interpretation that cortisol plays a role in arousal processes.

Based on the results of studies of rapid smoking of high-nicotine cigarettes it was proposed (20,21) that smoking-induced elevation of plasma BE modulates the reinforcing effects of nicotine ingestion. In a recent replication of procedures used in earlier work (12), we confirmed that rapid smoking of two high-nicotine cigarettes increases plasma BE, but also increases feelings of nausea, sickness and unpleasantness in some smokers, feelings which were correlated with BE release. Hence, it was important to establish whether elevation of plasma BE occurs consistently under normal smoking conditions without associated subjective malaise. Using more normal smoking conditions and nicotine deliveries in the present study, BE was elevated relative to nicotine-free cigarette smoking without significant increases in malaise after the first two but not subsequent cigarettes. The absence of nicotine-induced BE elevation after four or five cigarettes suggests that plasma BE elevation is not a consistent feature of normal cigarette smoking under low-stress conditions. Since smoking is apparently reinforcing under such conditions, it appears that elevation of plasma BE is not a necessary condition for the reinforcement of smoking.

ACTH and beta-endorphin are thought to be coreleased into the blood stream via the cleaving of a common parent molecule (15). The fact that serum cortisol levels were elevated after four and five nicotine cigarettes in the present study without concomitant elevations in BE suggests that cortisol was increased by nicotine administration independently of ACTH secretion. Increases in peripheral cortisol without concomitant increases in ACTH concentrations have been reported in humans in conjunction with diurnal cortisol variations (8); after cigarette smoking (27); and in cats after nicotine administration (24). It has been proposed that nicotine may stimulate cortisol release via a direct action on the adrenal cortex (24). However, the fact that nicotine-induced corticosteroid elevation is suppressed by dexamethasone pretreatment in smokers (22), and abolished in hypophysectomized rats (5), argues against a non-ACTH-mediated mechanism of nicotine-induced cortisol release. Nevertheless, it is conceivable that while nicotine-induced cortisol release is suppressed or abolished when ACTH is totally absent, it is preserved when even low levels of circulating ACTH are present. Thus nicotine may enhance ACTH-mediated cortisol release.

The finding that smoking tobacco but not nicotine-free cigarettes decreased drowsiness and increased alertness is consistent with reports that smoking enhances concentration (25,31) and vigilance performance (23,32). However, while drowsiness decreased after nicotine cigarettes in both males and females, female smokers reported feeling drowsier prior to smoking than their male counterparts and female nonsmokers. In contrast, nicotine-deprived male smokers did not differ in drowsiness from male nonsmokers. To our knowledge, differences between female smokers, male smokers, and female nonsmokers in self-reported drowsiness have not been reported previously.

While the unexpected difference between female smokers and nonsmokers in baseline drowsiness could have arisen from chance factors (e.g., sampling error), the magnitude of the smoker-nonsmoker and gender differences ($ps<0.001$), and the fact that these differences were eliminated by smoking, suggests that female smokers may experience greater reductions in arousal after overnight smoking deprivation than their male or nonsmoking female counterparts. Alternately, it is possible that females who take up smoking represent a subgroup that is typically less aroused than those who do not acquire the habit; smoking may be reinforcing in these individuals, in part, because it provides a ready means of enhancing subnormal arousal/alertness. Conversely, if nonsmoking females are inherently more aroused/alert than smoking females, they might not experience smoking-induced arousal as reinforcing, and therefore would be less likely to take up the smoking habit. Large prospective longitudinal studies have consistently demonstrated that individuals who later take up smoking differ in genetically influenced personality/temperament traits from those who do not (6, 19, 26), supporting the view that constitutional factors influence motivation to smoke. On the other hand, the gender and smoker/nonsmoker differences in drowsiness we observed may be related to any of several differences in personality-related behavioral patterns. For example, the female smokers may have been more depressed and/or slept less than the males and female nonsmokers. As a group, smokers tend to be more depressed and experience more negative affect than nonsmokers (1, 7, 14).

A limitation of the present study is that only females who were oral contraceptive users were tested. It is possible that some of the behavioral and physiological differences between males and females we observed may in fact be related to contraceptive pill use, rather than gender differences. For example, it has been reported that while men and noncontraceptive pill-using women did not differ in morning cortisol levels, oral contraceptive users were approximately 2.2 times higher in resting plasma cortisol levels than men (30), confirming a finding of the present study. A comparison of the effects of smoking in nonoral contraceptive pill users is needed to test the generality of the present findings.

ACKNOWLEDGEMENTS

Richard Welser and Stephanie Scott provided invaluable technical assistance in this project. Portions of this research were supported by grants from the R. J. Reynolds Tobacco Company.

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