Changes in rate and pattern of caffeine metabolism after cigarette abstinence

Caffeine metabolism is known to be accelerated in cigarette smokers, but the effects of smoking on the kinetics and pattern of metabolism in a daily consumption pattern have not been described. We investigated the effects of tobacco abstinence on the rate and pattern of caffeine metabolism in nine habitual smokers who consumed six cups of coffee per day, each cup containing 2 mg/kg caffeine. Abstinence from smoking for 4 days resulted in a 46% increase in the 24-hour AUC. Thus, significant, although probably not complete, normalization of the enzyme-inducing effects of cigarette smoking can be seen after 4 days abstinence. During abstinence, 24-hour urine ratios of dimethylxanthines to caffeine and monodimethylxanthines to dimethylxanthines were reduced, suggesting that cigarette smoking accelerates both demethylation steps. Other metabolic pathways were unaffected. (CLIN PHARMACOL THER 1988;43: 488-91.)

Christopher R. Brown, MD,* Peyton Jacob III, PhD, Margaret Wilson, MS, and Neal L. Benowitz, MD *San Francisco, Calif.*

Coffee consumption and cigarette smoking have been linked in several studies.[1-4] Accelerated metabolism of caffeine in smokers could be an explanation for the increased coffee consumption seen in cigarette smokers. Although the increased clearance of single doses of caffeine in smokers has been firmly established,[5-8] the effects of cigarette smoking on the kinetics of caffeine in a daily consumption pattern resembling more normal use patterns and on pathways of metabolism of caffeine have not been described.

We investigated the effect of tobacco abstinence on the rate and pattern of caffeine metabolism in habitual smokers receiving caffeine throughout the day.

METHODS

Subjects. Nine healthy men who were regular smokers and coffee drinkers, 25 to 61 years of age (mean 37.7 years), were admitted to the General Clinical Research Center at San Francisco General Hospital Medical Center for 18 days. Written, informed consent was obtained from each subject and the study was approved by the University of California, San Francisco, Committee on Human Research.

Table I. Characteristics of volunteer subjects (n = 9)

	Mean ± SD	*Range*
Age (yr)	37.8 ± 9.7	25-61
Weight (kg)	73.0 ± 8.2	60.9-87.6
No. cigaretes/day	38.9 ± 7.4	30-50
FTC tar (mg/cigarette)	18.1 ± 3.5	14-23
FTC nicotine (mg/cigarette)	1.1 ± 0.2	0.9-1.3
FTC CO (mg/cigarette)	14.4 ± 0.8	13-16
Cotinine (random) (ng/ml)	498.1 ± 186.5	157.7-760.3
Coffee (cups/day)	5.6 ± 2.1	3-8
Cola (cans/day)	1.4 ± 1.3	0-4
Tea (cups/day)	0.3 ± 0.7	0-2

FTC, US Federal Trade Commission.

Experimental protocol. The study was conducted in four treatment blocks, each lasting 3 or 4 days. Each patient went through all four blocks. Blocks consisted of placebo or low- or high-dose caffeine while smoking and high-dose caffeine when abstaining from tobacco. Caffeine, 1 mg/kg (low dose) or 2 mg/kg (high dose) in decaffeinated instant coffee, was given to subjects every 2 hours between 8 AM and 6 PM (total six

From the Division of Clinical Pharmacology and Experimental Therapeutics, Medical Service, San Francisco General Hospital Medical Center School of Medicine, University of California, San Francisco.

Supported by US Public Health Service grants CA32389, DA02277, and DA01696. Carried out in part in the General Clinical Research Center at San Francisco General Hospital Medical Center (RR-00083) with support of the Division of Research Resources, National Institutes of Health.

Received for publication Aug. 14, 1987; accepted Nov. 2, 1987.

Reprint requests: Neal L. Benowitz, MD, San Francisco General Hospital Medical Center, Bldg. 30, Fifth Floor, 1001 Potrero Ave., San Francisco, CA 94110.

*Supported by Training Grant GM07546.

Table II. Caffeine pharmacokinetic parameters during smoking and abstinence

	Smoking	*Nonsmoking*	*p Value*
$t_{1/2}$ (hr)	3.09 ± 2.18	3.37 ± 1.66	NS
AUC (ng · hr/ml)	36986.1	53884.2	<0.001
SD	±11095.7	±17693.3	

Table III. Caffeine metabolites as millimolar percentage of the dose in 24-hour urine collection

Metabolite	*Smoking*	*Nonsmoking*	*p Value*
Xanthine	0.60 ± 0.38	0.45 ± 0.34	NS
3-MU	0.12 ± 0.07	0.10 ± 0.04	NS
7-MU	3.06 ± 1.19	2.08 ± 0.51	<0.05
7-MX	4.67 ± 1.54	3.63 ± 1.33	<0.05
1-MU	17.54 ± 5.41	14.55 ± 3.09	NS
3-MX	1.76 ± 0.30	1.50 ± 0.30	NS
3,7-MU	0.09 ± 0.03	0.07 ± 0.02	NS
1-MX	12.76 ± 3.54	10.69 ± 3.35	NS
1,3-MU	1.80 ± 0.34	1.56 ± 0.26	NS
3,7-MX	0.96 ± 0.30	1.06 ± 0.29	NS
1,7-MX	4.23 ± 1.38	4.67 ± 1.23	NS
1,3-MX	0.25 ± 0.13	0.40 ± 0.35	NS
1,7-MU	0.26 ± 0.23	0.18 ± 0.09	NS
1,3,7-MX	0.85 ± 0.21	1.19 ± 0.24	<0.001
Total	48.95%	42.13%	

3-MU, 3-methyl uric acid; 7-MU, 7-methyl uric acid; 7-MX, 7-methylxanthine; 1-MU, 1-methyl uric acid; 3-MX, 3-methylxanthine; 3,7-MU, 3,7-dimethyl uric acid; 1-MX, 1-methylxanthine; 1,3-MU, 1,3-dimethyl uric acid; 3,7-MX, 3,7-dimethylxanthinie (theobromine); 1,7-MX, 1,7-dimethylxanthine (paraxanthine); 1,3-MX, 1,3-dimethylxanthine (theophylline); 1,7-MU, 1,7-dimethyl uric acid; 1,3,7-MX, 1,3,7-trimethylxanthine (caffeine).

cups/day; daily caffeine dose 0, 6, or 12 mg/kg/day). Coffee was prepared by adding anhydrous caffeine or nothing (placebo) to decaffeinated instant coffee mixed with a constant volume of water. Treatment blocks were balanced by Latin squares. During each smoking block the subjects were allowed to smoke their own brand of cigarette freely. Compliance during tobacco abstinence was assured by measuring exhaled carbon monoxide every 4 hours, which should be <8 ppm when abstaining from smoking. For the purposes of this report, data from the two high-dose caffeine treatments, comparing smoking vs tobacco abstinence, are presented.

On the third or fourth day of each block a circadian blood sampling study was performed. Blood was drawn via an indwelling catheter every 2 hours during the day and every 4 hours while asleep and assayed for caffeine concentrations.

Twenty-four-hour urine specimens were collected for

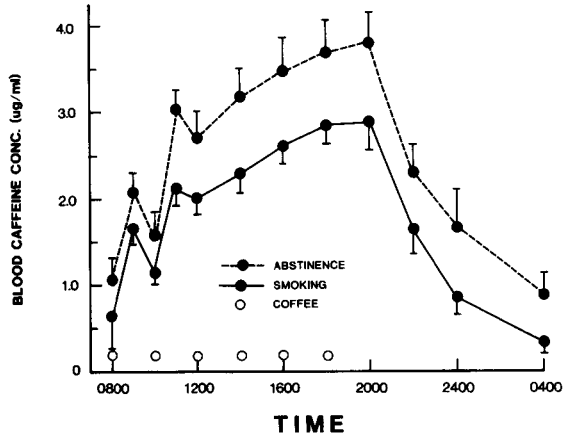

Fig. 1. Circadian blood caffeine concentrations while consuming six cups of coffee per day, comparing conditions of ad libitum cigarette smoking and tobacco abstinence (4 days). Values represent trough concentrations except at 9 and 11 AM. Open circles above the horizontal axis indicate times at which cups of coffee, containing 2 mg/kg body weight caffeine, were consumed. Data represent means of nine subjects ± SE.

measurement of concentrations of caffeine and caffeine metabolites. The diet was a standard hospital diet consisting of 15% to 17% protein, 39% to 45% carbohydrate, and 40% to 46% fat with all caffeine-containing beverages and chocolate prohibited.

Blood was collected in tubes with oxalic acid anticoagulant and frozen at −10° C until analysis. Concentrations of cotinine (the major metabolite of nicotine, used as an indicator of level of nicotine consumption) and caffeine were analyzed by GC with nitrogen-phosphorous detection and capillary column, modified from the assay of Jacob et al.[9] The internal standard for caffeine was 7-ethyltheophylline.

Caffeine metabolites in urine collections from circadian day of each block were analyzed by HPLC according to the method of Muir et al.[10]

Data analysis. Elimination half-life ($t_{1/2}$) and elimination rate constants were computed by linear regression of the log plasma caffeine concentrations vs time after the last dose of the day. The 24-hour AUC for caffeine was computed by the trapezoidal rule. Differences between smoking and nonsmoking conditions were analyzed by a two-tailed paired t test.

RESULTS

Characteristics of the subjects including preadmission cigarette and caffeine consumption are shown in Table I.

Table IV. Ratios of various caffeine metabolites

Metabolite ratio	*Smoking*	*Nonsmoking*	*p Value*
3,7-MX/1,3,7-MX	1.14 ± 0.23	0.89 ± 0.18	<0.05
1,7-MX/1,3,7-MX	4.96 ± 0.80	3.97 ± 0.84	<0.005
1,3-MX/1,3,7-MX	0.30 ± 0.14	0.33 ± 0.24	NS
7-MX/(3,7-MX + 1,7-MX)	1.02 ± 0.64	0.70 ± 0.42	<0.05
3-MX/(3,7-MX + 1,3-MX)	1.54 ± 0.37	1.11 ± 0.30	<0.02
1-MX/(1,7-MX + 1,3-MX)	3.05 ± 0.93	2.24 ± 0.88	<0.05
1-MU/1-MX	1.39 ± 0.23	1.41 ± 0.25	NS
1,7-MU/1,7-MX	0.06 ± 0.05	0.04 ± 0.02	NS

Abbreviations described in Table III.

Plasma caffeine levels throughout the day are depicted in Fig. 1. The peak caffeine concentrations during the smoking and abstinence blocks were 2.9 ± 1.0 and 3.8 ± 0.9 μg/ml, respectively ($p < 0.05$). The time of peak concentration was 8 PM or 2 hours after the last dose in both conditions. The caffeine AUC was significantly smaller and the $t_{1/2}$ tended to be shorter while smoking compared with abstinence (Table II).

The percentage of the administered dose excreted as various metabolites is shown in Table III. Caffeine plus metabolites accounted for approximately 49% of the daily dose in the smoking block and 42% in the nonsmoking block. Table IV shows the ratios of excretion of monomethylxanthines to dimethylxanthines and dimethylxanthines to caffeine in the urine. During abstinence there tended to be less product for a given amount of substrate for each *N*-demethylation step, but the magnitude of effect differed for different metabolic pathways. For example, for caffeine and 1- and 3-demethylation appear to be more affected than 7-demthylation. The ratio of 1-MU to 1-MX, a marker of xanthine oxidase activity,[11] and the ratio of 1,7-MU to 1,7-MX, a marker of microsomal 8-hydroxylation,[12] did not change between smoking and nonsmoking blocks.

DISCUSSION

We found a 46% increase in caffeine AUC in individuals after short-term tobacco abstinence. In our protocol, plasma caffeine levels were drawn just before the next dose of caffeine was administered (trough levels). This resulted in our underestimating the 24-hour AUC, because caffeine levels peak rapidly (30 to 60 minutes) after each dose.[13]

The time course of the loss of hepatic enzyme induction after short-term abstinence from cigarette smoking has been investigated with theophylline but not with caffeine. Eldon et al.[11] showed that there was no change in clearance of theophylline 36 hours after smoking was stopped, but Lee et al.[14] showed a 37.6% decrease in clearance after 7 days of abstinence. Our study shows that the magnitude of decrease in clearance of caffeine after only 3 to 4 days of cigarette abstinence is similar to that of theophylline after 7 days of abstinence.

We found reduced 24-hour urine ratios of monomethylxanthines to dimethylxanthines and dimethylxanthines to trimethylxanthine (caffeine) after cigarette abstinence. This suggests that smoking accelerates the first and second demethylation steps of caffeine metabolism, probably via induction of hepatic microsomal oxidative P-448 enzymes.[12] The lack of changes in the ratios of 1-methyl uric acid to 1-methylxanthine and 1,7-methyl uric acid to 1,7-methylxanthine indicates that smoking does not substantially affect xanthine oxidase–mediated metabolism or C8-hydroxylation.[8,15] Alternatively, 4 days of abstinence may not have been adequate to see loss of induction for these particular enzymes. That *N*-demethylation is the major pathway induced by cigarette smoking in humans has been observed in studies of the metabolism of theophylline and theobromine.[16,17]

We were able to account for approximately 49% of the administered dose. This figure is close to that of Grant et al.[12] but somewhat less than that of Bonati et al.,[18] Tang-Liu et al.,[19] Callahan et al.,[20] and Blanchard et al.[21] Discrepancies between the results of our study and those of other investigators may be the result of the number of metabolites quantified or the experimental protocol. Several investigators have reported significant amounts (3% to 28% of the administered dose of caffeine) as 5-acetylamino-6-formylamino-3-methyluracil, a metabolite that was not quantified with our assay and that probably accounts for higher recoveries reported by other investigators.[8,15,20] Ours is the only study using a multiple-dosing regimen with 24-hour urine collections. We have assumed that blood concentrations and excretion of metabolites have reached steady state by the end of each study block

(i.e., 3 or 4 days), which is expected according to the relatively short $t_{1/2}$ of caffeine and its major metabolites.[19]

Although not proving a causal link between smoking and coffee drinking directly, our data support a pharmacokinetic explanation. That is, if the level of coffee consumption is determined by a desire to achieve particular levels of caffeine in the body, smokers would need to drink at least one and a half times as much coffee to maintain levels similar to those of nonsmokers.

In summary, we report two new findings concerning cigarette smoking and caffeine metabolism. First, after stopping smoking for only 3 or 4 days the rate of caffeine metabolism is substantially slower. In that cross-sectional studies indicate that clearance of caffeine is 60% greater in smokers compared with nonsmokers,[5-7] our data indicate significant but probably not complete normalization of enzyme-inducing effects of smoking after 4 days abstinence. Insofar as such rapid loss of drug–metabolizing enzyme induction applies to other drugs whose metabolism is accelerated by cigarette smoking, brief changes in smoking habits (such as on admission to or after discharge from a hospital) may result in unexpected changes in drug efficacy or toxicity.

Second, cigarette smoking affects primarily *N*-demethylation pathways of caffeine metabolism. Several *N*-demethylation processes are affected, but to differing degrees. The major metabolites of caffeine, paraxanthine, theobromine, and theophylline are pharmacologically active. Because these metabolites are both generated and eliminated by demethylation, smoking-related differences in magnitudes of induction among different demethylation steps could influence both quantitative and qualitative pharmacologic actions of caffeine.

References

1. Mathias S, Garland C, Barrett-Conner E, Wingard DL. Coffee, plasma cholesterol, and lipoproteins: a population study in an adult community. Am J Epidemiol 1985;121:896-905.
2. Murray SS, Bjelke E, Gibson RW, Schuman LM. Coffee consumption and mortality from ischemic heart disease and other causes: results from the Lutheran Brotherhood Study, 1966-1978. Am J Epidemiol 1981;113:661-7.
3. Dawber TR, Kannel WB, Gordon T. Coffee and cardiovascular disease: observations from the Framingham Study. N Engl J Med 1974;291:871-4.
4. Friedman GD, Siegelaub AB, Seltzer CC. Cigarettes, alcohol, coffee and peptic ulcer. N Engl J Med 1974; 290:469-73.
5. Parsons WD, Neims AH. Effect of smoking on caffeine clearance. Clin Pharmacol Ther 1978;24:40-5.
6. Kotake AN, Schoeller DA, Lamber GH, Baker AL, Schaffer DD, Josephs H. The caffeine CO_2 breath test: dose-response and route of *N*-demethylation in smokers and nonsmokers. Clin Pharmacol Ther 1982;32: 261-9.
7. May DC, Jarboe CH, VanBakel AB, Williams WM. Effects of cimetidine on caffeine disposition in smokers and nonsmokers. Clin Pharmacol Ther 1982;31: 656-61.
8. Kalow W. Variability of caffeine metabolism in humans. Arzneimittelforschung Drug Res 1985;35:319-24.
9. Jacob P III, Wilson M, Benowitz NL. Improved gas chromatographic method for the determination of nicotine and cotinine in biologic fluids. J Chromatogr 1981; 222:61-70.
10. Muir KT, Jonkman HG, Tang D, Kunitani M, Riegelman S. Simultaneous determination of theophylline and its major metabolites in urine by reversed-phase ion-pair high-performance liquid chromatography. J Chromatogr 1980;221:85-95.
11. Eldon MA, Luecker PW, MacGee J, Ritschel WA. Lack of effect of withdrawal from cigarette smoking on theophylline pharmacokinetics. J Clin Pharmacol 1987; 27:221-5.
12. Grant DM, Tang BK, Kalow W. Variability in caffeine metabolism. Clin Pharmacol Ther 1982;33:591-602.
13. Blanchard J, Sawers SJA. The absolute bioavailability of caffeine in man. Eur J Clin Pharmacol 1983;24:93-8.
14. Lee BL, Benowitz NL, Jacob P III. Cigarette abstinence, nicotine gum and theophylline disposition. Ann Intern Med 1987;106:553-5.
15. Grant DM, Tang BK, Campbell ME, Kalow W. Effect of allopurinol on caffeine disposition in man. Br J Clin Pharmacol 1986;21:454-8.
16. Grygiel JJ, Birkett DJ. Cigarette smoking and theophylline clearance and metabolism. Clin Pharmacol Ther 1981;30:491-6.
17. Miners JO, Attwood J, Wing LMH, Birkett DJ. Influence of cimetidine, sulfinpyrazone, and cigarette smoking on theobromine metabolism in man. Drug Metab Dispos 1985;13:598-601.
18. Bonati M, Latini R, Galletti F, Young JF, Tognoni G, Garattini S. Caffeine disposition after oral doses. Clin Pharmacol Ther 1982;32:98-106.
19. Tang-Liu DD, Williams RL, Riegelman S. Disposition of caffeine and its metabolites in man. J Pharmacol Exp Ther 1983;224:180-5.
20. Callahan MM, Robertson RS, Branfman AR, McComish MF, Yesair DW. Comparison of caffeine metabolism in three nonsmoking populations after oral administration of radiolabeled caffeine. Drug Metab Dispos 1983; 11:211-7.
21. Blanchard J, Sawers SJA, Jonkman JHG, Tang-Liu DD. Comparison of the urinary metabolite profile of caffeine in young and elderly males. Br J Clin Pharmacol 1985;19:225-32.