Preventive Medicine **31,** 148–152 (2000)
doi:10.1006/pmed.2000.0694, available online at http://www.idealibrary.com on IDEAL®

Effects of the Transdermal Nicotine Patch on Normalization of HDL-C and Its Subfractions[1]

Robert J. Moffatt, Ph.D.,[2] Kyle D. Biggerstaff, M.S.,[3] and Bryant A. Stamford, Ph.D.

Department of Nutrition, Food, and Exercise Sciences, Florida State University, Tallahassee, Florida, 32306-1493; and The Health Promotion Center, University of Louisville, Louisville, Kentucky

Background. **Smoking reduces HDL-C and its subfractions, and smoking cessation leads to normalization of these lipoproteins. Nicotine replacement therapy is an important weapon employed by those attempting to quit smoking. This study examined the effects of the transdermal nicotine patch ("patch") on lipoproteins.**

Methods. **Ten male and 17 female smokers refrained from smoking for 77 days. The patch was utilized during the first 35 days and then removed for the remaining 42 days. Seven male and 9 female nonsmokers were controls.**

Results. **HDL-C, HDL_2-C, and HDL_3-C levels were significantly lower in smokers when compared with controls. These differences were sustained during the initial 35 days when using the patch. Over the following 42 days, however, these lipoproteins normalized to values similar to those of control subjects. Females who quit smoking gained 2.1 kg after the patch was removed.**

Conclusions. **It was concluded that nicotine as administered by the transdermal nicotine patch inhibits normalization of HDL-C, HDL_2-C, and HDL_3-C in those who have quit smoking. Removal of the patch results in normalization of these lipoproteins. The patch appeared to prevent weight gain among female subjects.**

Key Words: **lipoproteins; smoking; nicotine; cessation.**

INTRODUCTION

Cigarette smoking is a major risk factor in coronary heart disease. There may be many mechanisms of effect, including alteration of blood lipoproteins. Smokers have been shown to have lower concentrations of high-density lipoprotein (HDL-C) cholesterol and its antiatherogenic subfraction, HDL_2-C, when compared with nonsmokers [*1–5*]. In addition, environmental tobacco smoke has been shown to have a negative impact on blood lipoproteins [*6*].

Blood lipid and lipoprotein normalization has been demonstrated with smoking cessation [*3,7*]. This raises the question: What factors associated with cigarette smoke specifically impact blood lipids? Nicotine has been studied as a possible cause of atherogenic alterations in lipoprotein profiles. Quensel et al. [*8*] found that chewing nicotine gum for 2 weeks by young healthy volunteers had no effect on blood lipids. The impact on results of using nonsmoking volunteers, rather than smokers seeking to quit, and limiting treatment to 2 weeks is unknown. Allen et al. [*9*] found that HDL-C increased following smoking cessation even though subjects were treated with a transdermal nicotine patch in doses of 21, 14, or 7 mg/day. HDL-C increased significantly in patients exposed to 21 and 14 mg/day, but not in patients exposed to 7 mg/day. It is a surprising finding that the lowest dose of nicotine prevented normalization of HDL-C, while the higher doses and the placebo did not. The present study was undertaken to determine the effects of a transdermal nicotine patch ("patch") on HDL-C and its subfractions in the early stages of smoking cessation among those with a substantial smoking history.

At present, the mechanism responsible for depressing HDL and its subfractions as a result of cigarette smoking is unclear. A continued depression of HDL in association with the patch would indict nicotine as a potential contributor. In addition, a depressed lipoprotein profile

[1] This study was funded in part by Lederle Pharmaceuticals, Pearl River, NY.

[2] To whom reprint requests should be addressed at Department of Nutrition, Food, and Exercise Sciences, 436 Sandels, Florida State University, Tallahassee, Florida 32306-1493. Fax: (850) 645-5000. E-mail: rmoffatt@mailer.fsu.edu.

[3] Now at Department of Kinesiology, Texas Woman's University, Denton, TX.

0091-7435/00 $35.00

contributes to coronary heart disease, and circumstances which sustain the depression represent a clinically significant finding.

METHODS

The present study of the influence of transdermal nicotine on HDL-C was a secondary project within a larger investigation of the efficacy of participation by a significant partner during a smoking cessation trial. Volunteers to participate in the smoking cessation program were recruited by newspaper advertisements, flyers, and public service announcements. Responses were received from 151 individuals of which 104 individuals were eligible smokers for the cessation program. The remaining 47 respondents were unable to commit to the requirements of the program. From this pool, 96 individuals were randomly assigned to treatment and control groups. Three treatment groups containing 72 individuals received a transdermal nicotine patch as a portion, or all, of their smoking cessation treatment. One group ($n = 24$) was provided the nicotine patch as their only aid in smoking cessation while two groups received group counseling either with ($n = 24$) or without ($n = 24$) spouses participating. The remaining 24 subjects were assigned to a non-counseling, non-patch control group for the larger investigation. From the 72 participants who received the patch, volunteers were recruited to enter the present study. Following a full description of study procedures, 68% of the 72 potential participants (25 male, 24 female) smokers (ex-smokers) volunteered to participate in the present study of HDL-C. All of the 72 potential participants were encouraged to be involved in the present study; however, some were unwilling to have blood drawn over the course of the larger study. For various reasons including skin irritation, scheduling conflicts, inability to stop smoking, and suspected pregnancy, 15 male and 7 female ex-smokers failed to complete the present study. The ex-smokers group, therefore, resulted in 17 females and 10 males at the completion of the study who successfully abstained from smoking for 77 days (see Table 1). Advertisements were placed in the local community requesting nonsmoking volunteers to participate. In response, 9 female and 7 male lifetime nonsmokers volunteered to serve as controls (nonsmokers). Participants were healthy as determined by a medical history questionnaire. No statistically significant differences existed between ex-smokers and nonsmokers with respect to age, height, or body weight (see Table 1). Ex-smokers used cigarettes only and no other tobacco (pipe, cigar, or smokeless tobacco) or nicotine (chewing gum) products. All procedures were approved by the University Human Subjects Board and all subjects signed an informed consent statement before entering the study.

Subjects reported to the laboratory in a 12-h postprandial state on three occasions (days 0, 35, and 77). Ex-smokers abstained from cigarette smoking from day 1 through day 77. From day 1 through day 35, Ex-smokers used a transdermal nicotine patch (22 mg/day). The research of Gorsline et al. [*10*] indicates that the patch dose utilized equates to a steady-state plasma nicotine concentration of approximately 17 mg/ml. From day 36 through day 77, the patch was not used by ex-smokers.

In order to replicate as close to normal conditions associated with smoking cessation as possible, ex-smokers were encouraged to act as naturally as possible, despite their commitment to the study. This means if subjects felt compelled to consume more food upon cessation (a likely occurrence), they should feel free to do so, because weight gain is a typical outcome of smoking cessation [*3*]. Increased caloric intake and the loss of acute metabolic stimulation associated with inhaling cigarette smoke are major contributors to cessation-related weight gain [*3,11*]. The only stipulation regarding diet was to make no qualitative changes. Our previous research has demonstrated that subjects typically will eat more, but will eat more of the kinds of food to which they are accustomed, thus maintaining the same percentage of fat, carbohydrate, protein, and alcohol in the diet [*3*]. With the exception of smoking cessation, subjects were instructed to refrain from making specific

TABLE 1

Age and Anthropometric and Smoking Characteristics of Subjects

	Age (years)	Height (cm)	Body weight (kg)	Cigarettes smoked (cig/day)	Years smoking (years)
Ex-smokers					
Males (n=10)	45.3 ±14.8	175.5 ± 7.2	73.5 ± 8.0	29.2 ± 9.2	25.7 ± 8.7
Females (n=17)	38.2 ±8.6	164.0 ± 3.7	65.3 ± 16.2	28.6 ± 8.5	19.7 ± 7.3
Nonsmokers					
Males (n=7)	41.9 ± 11.1	178.9 ± 4.6	75.9 ± 7.4	N/A	N/A
Females (n=9)	39.9 ±10.8	165.1 ± 4.4	65.7 ± 8.4	N/A	N/A

Note. Mean ± standard deviation.

lifestyle changes, including changes in the volume of alcohol consumed per day and the amount of daily physical activity performed. Oral reports obtained from subjects indicate that subjects complied with all instructions.

Blood was sampled on days 0, 35, and 77 from an antecubital vein to determine levels of total cholesterol, HDL-C, HDL_2-C, and HDL_3-C in control and experimental subjects. Total cholesterol concentration was determined by the enzymatic method of Allain et al. [*12*]. HDL was separated from lipoproteins containing apolipoprotein B by a precipitation technique using heparin–manganese [*13*]. The cholesterol concentration of the supernatant was then determined as the HDL-C concentration. The double precipitation method of Gidez et al. [*14*] was used to precipitate HDL_2 using dextran sulfate leaving HDL_3 in the supernatant. The cholesterol concentration of the supernatant was HDL_3-C. HDL_2-C was determined as the difference between total HDL-C and HDL_3-C. Body weight was also determined on each of the three testing days. A three-way mixed analysis of variance (gender × group × day) was employed to determine statistically significant differences. A Scheffé post hoc test was performed to determine single degree of freedom differences. The criterion of statistical significance was set at the $P \leq 0.05$ level.

TABLE 2

Serum HDL-C, HDL_2-C, and HDL_3-C Concentration of Ex-smokers and Nonsmokers

	HDL-C (mg/dl)	HDL_2-C (mg/dl)	HDL_3-C (mg/dl)
Ex-smokers			
Males			
Day 0	36.0 ± 3.3	10.2 ± 3.1	25.6 ± 2.2
Day 35	36.9 ± 3.4	10.9 ± 3.0	26.2 ± 2.4
Day 77	42.6 ± 6.2**	15.5 ± 3.4**	27.0 ± 4.5
Females			
Day 0	43.1 ± 6.0*	14.0 ± 2.9*	29.4 ± 5.8*
Day 35	42.9 ± 5.4	14.2 ± 3.6	28.7 ± 6.5
Day 77	54.3 ± 6.5**	20.7 ± 3.6**	33.3 ± 6.4**
Nonsmokers			
Males			
Day 0	45.3 ± 4.2	15.5 ± 2.6	29.8 ± 2.4
Day 35	45.8 ± 4.8	15.7 ± 3.0	30.1 ± 2.5
Day 77	45.8 ± 3.5	15.1 ± 2.4	30.7 ± 2.4
Females			
Day 0	54.0 ± 2.7*	19.0 ± 3.0*	36.5 ± 5.8*
Day 35	55.7 ± 4.5	18.8 ± 3.0	36.9 ± 5.9
Day 77	55.0 ± 5.0	19.4 ± 3.0	35.6 ± 5.6

Note. Mean ± standard deviation.
* Significantly greater than males of the same group ($P \leq 0.05$).
** Significantly greater than day 0 and day 35 ($P \leq 0.05$).

RESULTS

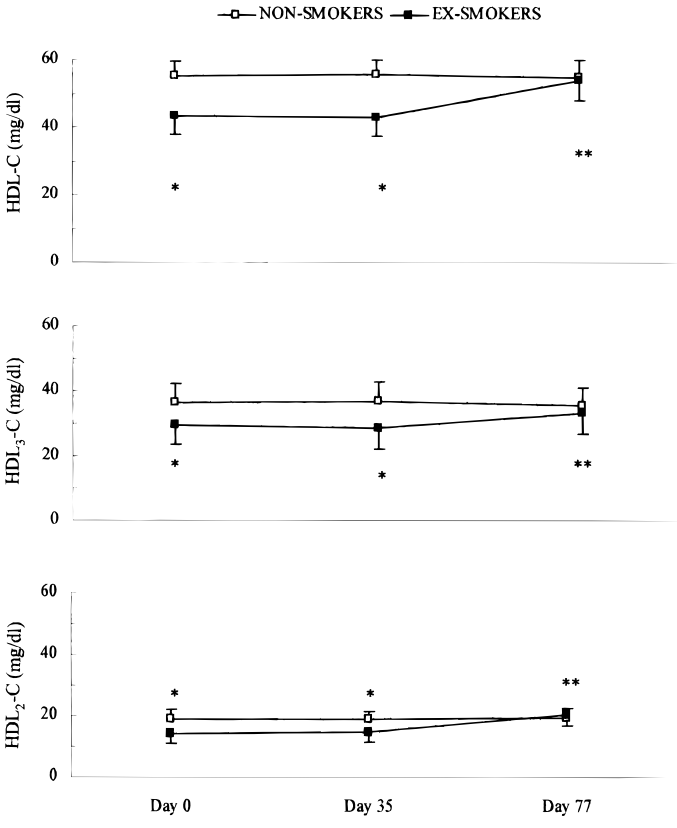
As was expected, precessation levels (day 0) for HDL-C, HDL_2-C, and HDL_3-C for all ex-smokers and nonsmokers were significantly ($P < 0.05$) higher in females when compared with males (see Table 2). Precessation levels (day 0) for HDL-C, HDL_2-C, and HDL_3-C for ex-smokers, males and females combined, were significantly ($P < 0.05$) below those of nonsmokers (see Fig. 1). HDL-C was lower by 24%, HDL_2-C was lower by 47%, and HDL_3-C was lower by 15%. After 35 days of wearing the patch, HDL-C, HDL_2-C, and HDL_3-C blood levels for ex-smokers had not changed. When the patch had been removed for the preceding 42 days, ex-smokers at day 77 demonstrated significantly increased ($P < 0.05$) levels of HDL-C, HDL_2-C, and HDL_3-C. HDL-C increased by 21% (males = 18%; females = 26%), HDL_2-C by 44% (males = 52%; females = 48%), and HDL_3-C by 10% (males = 5%; females = 13%) among ex-smokers (see Fig. 1 and Table 2); however, HDL_3-C did not increase significantly in males.

Male ex-smokers did not gain weight throughout the study (see Table 3.). Female ex-smokers, on the other hand, gained a significant ($P < 0.05$) 2.1 kg at day 77. Male and female nonsmokers did not gain weight. Total cholesterol did not change during the study period in any groups. TC/HDL-C ratios were found to be different ($P < 0.05$) between ex-smokers (day 0 = 4.60 ± 0.86, day 35 = 4.67 ± 0.81) and nonsmokers (day 0 = 3.46 ± 0.41, day 35 = 3.43 ± 0.39) prior to, and during use of, the patch; however, removal of the patch lead to a lower TC/HDL-C ratio at day 77 (ex-smokers = 3.74 ± 0.84 vs nonsmokers = 3.46 ± 0.39).

DISCUSSION

The present data demonstrate that nicotine as administered by a transdermal nicotine patch inhibits normalization of HDL-C, HDL_2-C, and HDL_3-C in those who quit smoking. Following removal of the patch, these lipoproteins normalized from day 35 to day 77. While this investigation is somewhat limited based on the small sample size, these results agree with many other reports which have demonstrated a positive effect on HDL-C with smoking cessation [*3,4,7,15*].

Removal of the patch for 42 days resulted in similar improvements of HDL-C and HDL_2-C that have been documented following smoking cessation for as little as 10 and 30 days [*4,15*]. In the present study, by day 77 of smoking cessation and 42 days after the removal of the patch, HDL-C and HDL_2-C were elevated in males and females alike. In Moffatt et al. [*15*], this response differed from the males, but not the females, where HDL-C and its subfractions were not influenced by 30 days of smoking cessation without the use of the patch. The improvement in HDL-C following removal of the patch in males was primarily due to the increased HDL_2-C; however, in females increased HDL_2-C and HDL_3-C were responsible for the increased HDL-C. The



Mean ± standard deviation.
*EX-SMOKERS significantly different than NON-SMOKERS ($p \leq 0.05$).
**EX-SMOKERS significantly different than Day 0 and Day 35 ($p \leq 0.05$).

FIG. 1. Serum HDL-C, HDL_3-C, and HDL_2-C of ex-smokers and nonsmokers.

combination of the present data and those of Moffatt et al. [*15*] suggest that males and females may have differing lipoprotein-cholesterol responses following smoking cessation and removal of the patch.

It is difficult to determine why the present results contrast with those of Allen et al. [*9*] who found that lipoproteins normalized despite nicotine dosages from 21 and 14 mg/day patches, while a nicotine dosage from a 7 mg/day patch prevented normalization. The same study also demonstrated normalization in the placebo group. These inconsistent results are confusing. The authors cite the fact that blood sampling was nonfasting. The serum concentration of lipids and lipoproteins is closely impacted during postprandial periods [*16,17*].

A different approach was taken by Quensel et al.[*8*] in which nonsmoking healthy young volunteers consumed nicotine chewing gum for 2 weeks. There were no changes in HDL-C or apolipoproteins A-1 and B. Whether lack of reduction in HDL-C among nonsmokers during 2 weeks on nicotine chewing gum is the same as normalization of HDL-C among smokers who quit smoking and who use the patch is open to question. If it is, then the preponderance of evidence prior to the present study was that nicotine does not affect lipoprotein concentrations. The present data challenge these previous findings. Additional research is required to resolve inconsistencies in the cumulative available data in order to determine whether nicotine is responsible for reductions in healthful lipoproteins among smokers.

The nicotine transdermal patch is in wide use as a nicotine replacement mode which helps cigarette smokers quit smoking. If the patch, and therefore nicotine, prevents normalization of lipoproteins as the present data suggest, the effect is acute and lasts only as long as the patch is in place. The health tradeoffs involved (quitting smoking versus a delay in normalization of lipoproteins) would in the long run seem worthwhile. This is especially true when considering that female ex-smokers gained weight when the patch was removed. Thus, although the patch may have prevented normalization of lipoproteins, it also prevented weight gain in females. The latter is a positive outcome, particularly when considering that unwanted weight gain is often cited as the reason for returning to smoking. The present data indicating that weight gain does not occur while the patch is in use are in conflict with other published data that suggest weight gain ranging from 1.6 to 4.0 kg occurs while ex-smokers are using the patch [*18–20*]. Long-term follow-ups (12–62 months) of those who have successfully abstained from smoking with the aid of nicotine patches indicate that weight gain does occur [*21,22*].

In conclusion, the present data indicate that the nicotine patch inhibits the normalization of HDL-C and its subfractions, however; it also limits weight gain. After the removal of the nicotine patch with continued smoking cessation, the typical normalization of HDL-C following smoking cessation is noted; however, with the

TABLE 3

Body Weight and Total Cholesterol of Ex-smokers and Nonsmokers

	Ex-smokers		Nonsmokers	
	Males	Females	Males	Females
	Body Weight (kg)			
Day 0	73.5 ± 8.0	65.3 ± 16.2	75.9 ± 7.4	65.7 ± 8.4
Day 35	74.0 ± 7.7	65.7 ± 17.0	75.2 ± 7.9	65.8 ± 8.7
Day 77	73.1 ± 9.0	67.4 ± 18.1*	75.5 ± 6.3	66.2 ± 8.4
	Total Cholesterol (mg/dl)			
Day 0	197.1 ± 20.0	197.5 ± 35.0	190.3 ± 23.8	189.4 ± 17.0
Day 35	197.6 ± 18.9	198.9 ± 36.6	191.1 ± 27.0	190.2 ± 17.2
Day 77	199.7 ± 22.5	199.9 ± 39.4	189.4 ± 25.7	187.9 ± 20.5

Note. Mean ± standard deviation.
* Significantly different ($P \leq 0.05$) than day 0 and day 35.

removal of the nicotine patch weight gain may also result.

REFERENCES

1. Criqui, MH, Wallace RB, Heiss G, et al. Cigarette smoking and plasma high-density lipoprotein cholesterol: The Lipid Research Clinics Program Prevalence Study. Circulation 1980;62:70–6.
2. Stamford EA, Matter S, Fell R, et al. Cigarette smoking, physical activity, and alcohol consumption: relationship to blood lipids and lipoproteins in premenopausal females. Metabolism 1984; 33:585–90.
3. Stamford, BA, Matter S, Fell R, et al. Effects of smoking cessation on weight gain, metabolic rate, caloric consumption, and blood lipids. Am J Clin Nutr 1986;43:486–94.
4. Moffatt, RJ. Effects of cessation of smoking on serum lipids and high-density lipoprotein-cholesterol. Atherosclerosis 1988;74: 85–9.
5. Craig WY, Palonaki GE, Haddow JE, et al. Cigarette smoking and serum lipid and lipoprotein concentrations: an analysis of published data. Br Med J 1989; 298:784–8.
6. Moffatt RJ, Stamford BA, Biggerstaff KD. Influence of worksite environmental tobacco smoke on serum lipoprotein profiles of female nonsmokers. Metabolism 1995;44:1536–9.
7. Stubb I, Eskilsson J, Nilsson-Ehle P. High-density lipoprotein concentrations increase after stopping smoking. Br Med J 1982;284:1511–3.
8. Quensel M, Agardh CD, P Nilsson-Ehle. Nicotine does not affect plasma lipoprotein concentrations in healthy men. Scand J Clin Invest 1989;49:149–53.
9. Allen SS, Hatsukami D, Gorsline J, et al. Cholesterol changes in smoking cessation using the transdermal nicotine system. Transdermal Nicotine Study Group. Prev Med 1994;23:190–6.
10. Gorsline J, Gupta SK, Dye D, et al. Steady-state pharmacokinetics and dose relationship of nicotine delivered from Nicoderm (Nicotine Transdermal System). J Clin Pharmacol 1993;33: 161–8.
11. Moffatt RJ, Owens SG. Cessation from cigarette smoking: changes in body weight, body composition, resting metabolism and energy consumption. Metabolism 1991;40:465–70.
12. Allain CC, Poon LS, Chan CSG, et al. Enzymatic determination of total serum cholesterol. Clin Chem 1974;20:470–5.
13. Warnick GR, Albers JJ. A comprehensive evaluation of the heparin–manganese precipitation procedure for estimating high density lipoprotein-cholesterol. J Lipid Res 1978;19:65–76.
14. Gidez LI, Miller GJ, Burnstein M, et al. Separation and quantitation of subclasses of human plasma high density lipoproteins by a simple precipitation procedure. J Lipid Res 1982;23:1206–23.
15. Moffatt RJ, Stamford BA, Owens SG, Chitwood LF. Cessation from cigarette smoking: lipoprotein changes in men and women. J Smoking-Related Dis 1992;3:11–9.
16. O'Meara NM, Lewis GF, Cabana VG, et al. Role of basal triglyceride and high density lipoprotein in determination of postprandial lipid and lipoprotein responses. J Clin Endocrin Metab 1992; 75:465–71.
17. Zhang JQ, Thomas TR, Ball SD. Effect of exercise timing on postprandial lipemia and HDL cholesterol subfractions. J Appl Physiol 1998;85:1516–22.
18. Jorenby DE, Leishcow SJ, Nides MA, et al. A controlled trial of sustained-release buproion, a nicotine patch, or both for smoking cessation. N Engl J Med 1999;340:685–91.
19. Dale LC, Schroeder DR, Wolter TD, Croghan IT, Hurt RD, Offord KP. Weight change after smoking cessation using variable doses of transdermal nicotine replacement. J Gen Intern Med 1998; 13:9–15.
20. Jorenby DE, Hatsukami DK, Smith SS, Fiore MC, Allen S, Jensen J, Baker TB. Characterization of tobacco withdrawal symptoms: transdermal nicotine reduces hunger and weight gain. Psychopharmacology 1996;128:130–8.
21. Richmond RL, Kehoe L, de Almeida Neto AC. Effectiveness of a 24-hour transdermal nicotine patch in conjunction with a cognitive behavioural programme: one year outcome. Addiction 1997; 92:27–31.
22. Daughton DM, Fortmann SP, Glover ED, et al. The smoking cessation efficacy of varying doses of nicotine patch delivery systems 4 to 5 years post-quit day. Prev Med 1999;28:113–8.