CEBP Focus: Tobacco Research

Review

Clinical Trials Methods for Evaluation of Potential Reduced Exposure Products

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Abstract

Potential reduced exposure products (PREPs) to tobacco toxicants may have promise in reducing tobacco-related morbidity or mortality or may promote greater harm to individuals or the population. Critical to determining the risks or benefits from these products are valid human clinical trial PREP assessment methods. Such an assessment involves determining the effects of these products on biomarkers of exposure and effect, which serve as proxies for harm, and assessing the potential for consumer uptake and abuse of the product. This article identifies critical methodologic issues associated with PREP assessments, reviews the methods that have been used to assess PREPs, and describes the strengths and limitations of these methods. Additionally, recommendations are provided for clinical trial PREP assessment methods and future research directions in this area based on this review and on the deliberations from a National Cancer Institute sponsored Clinical Trials PREP Methods Workshop. (Cancer Epidemiol Biomarkers Prev 2009;18(12):3143–95)

Introduction

In recent years, there has been an increasing need to evaluate tobacco products for human toxicity and disease risk because of tobacco company efforts to manufacture and market new products that purportedly decrease exposure to tobacco and tobacco smoke toxicants. Past attempts to manufacture safer cigarettes (e.g., "light" cigarettes) only led to false hopes of reduced health risk. With newer technologies and promotion of another generation of reduced toxicant exposure or reduced risk tobacco products by the tobacco industry, described as potential reduced exposure products (PREP) by the Institute of Medicine, the development of a science base to inform the current debates about whether PREPs present promise or harm is required (1, 2). These newer PREPs include novelly designed combustible products, such as those that have new filter designs or have tobacco processed or cured in a way to reduce some toxicants. They also include cigarette-like devices that heat tobacco, oral tobacco products that may reduce risk by eliminating exposure to toxicants associated with the combustion of tobacco, or electronic cigarettes with tobacco flavors that purportedly deliver only nicotine. Some of these PREPs have been on the market with implicit or explicit health claims that seem to be unsubstantiated. It is recognized that health claims related to PREPs might undermine successful tobacco control by leading to continued use of tobacco products in potential quitters, resumption of smoking among former smokers, or initiation. More so, if these claims were misleading or unsubstantiated, there could be additional harm. The importance of substantiating claims also points to the necessity of scientifically evaluating the effects of these products.

In an effort to avoid the mistakes that were made by the marketing of light, ultralight, and mild cigarettes, a careful study and strategic approach to evaluating tobacco products have been considered (2-6). For the most part, these reports describe three essential components for tobacco product evaluation, which is illustrated by a figure (Fig. 1): (*a*) preclinical evaluation, which involves assessment of type and amount of tobacco constituents, smoke chemistry analysis, and toxicology studies (*in vitro* and *in vivo* analysis); (*b*) clinical evaluation in humans and epidemiology studies involving assessment of pattern of product use, extent of exposure to toxicants and biological effect, abuse potential, and consumer perception of the product; and (*c*) population effect of the product involving post-marketing surveillance, population surveys and monitoring of adverse effects on health. The goal of these evaluations is to ensure that the PREP does not worsen exposure and disease risk compared with conventional products and to assess the amount of risk above complete cessation. Today, given that there are no acceptable biomarkers for cancer risk (6), with the possible exception of 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol and its glucuronides (total NNAL), a biomarker for 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone

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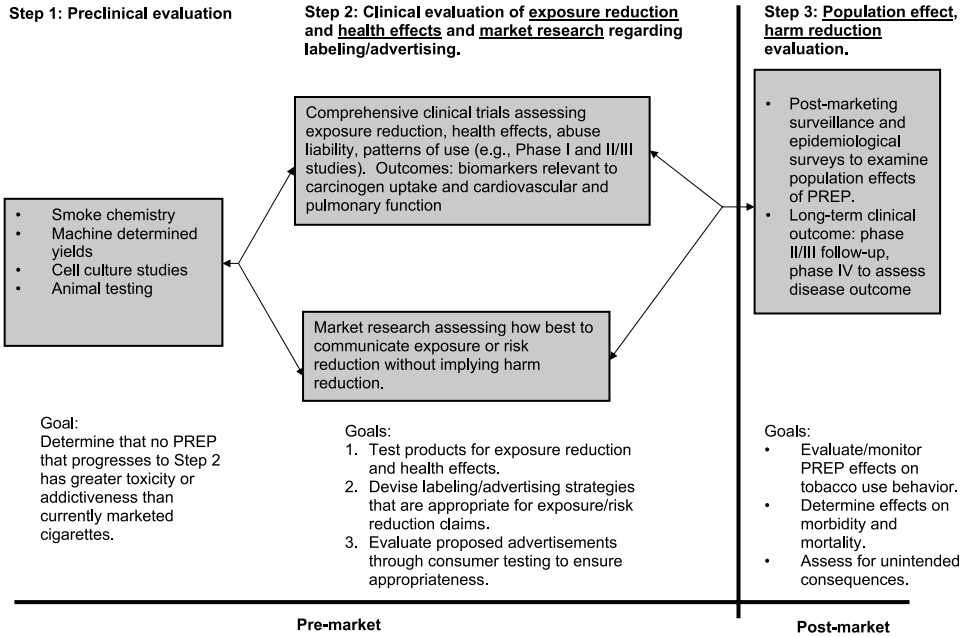

Figure 1. Three-step model for PREP evaluation and marketing with assessment occurring by an independent scientific panel or regulatory agency after each step (6).

(NNK) exposure (7, 8), the best evaluation of a PREP is limited to assessing human exposure reduction. However, exposure reduction is distinct from risk reduction; for example, exposure reduction of one or several tobacco toxicants might not result in reduced disease risk for the individual. Furthermore, although risk reduction in an individual might be feasible, the overall tobacco toxicant exposure in the population might increase because of delayed quitting or resumption of the use of tobacco products due to the availability of the PREPs. Thus, as shown in Fig. 2, there is a spectrum for assessing PREPs in humans that are somewhat distinct, and that exposure and risk reduction refer to individual effects, and harm reduction refers to population effects.

Although frameworks for studying PREPs were proposed by the Institute of Medicine and other reports, the actual methods to be used were left for later evaluation and research by scientists. One critical component of tobacco product assessment that has not undergone extensive review is the methods and measures for conducting clinical trials to assess whether a new PREP results in exposure and risk reduction, and if there can be an inferred effect on harm reduction. The goal of this article is to identify and discuss the challenges and questions associated with clinically assessing PREPs and to review the literature on published human clinical trial studies on methods and measures used in evaluating PREPs.

The choice of trial designs, methods, and measures is complex, and many decisions are necessarily made in the absence of scientific data. To help address questions that are critical to trial designs, and for which there is insufficient data to answer them, a Clinical Trials Workshop of experts (see Supplementary Appendix) was convened. This Workshop was held on June 9-10, 2008, and included presentations and discussions by experts within and outside of the tobacco research area using the review article as a basis for discussion. The deliberations and recommendations from this Workshop are also described along with recommendations for future research directions.

It is important to note that this article is not intended to discuss the relative toxicant levels or risk for disease across the PREP products, which have been described in other reviews (3, 9-11), but to primarily focus on methods used to assess PREPs.

Challenges and Goals for Clinical Trials. Several challenges are associated with assessing PREPs. These challenges and questions are described in Table 1.

As with any clinical trial, the first decision to be made is the goal(s) of the trial. Depending on the research question and the scientific discipline of the researchers, different approaches can be taken. The primary reason to assess PREPs in humans is to gain an understanding for changes in exposure and risk reduction. This would happen via assessment of tobacco use behavior (e.g., cigarettes per day, topography), pattern of use, and biomarkers of exposure and effect. For cigarettes, topographical measures would include number of puffs, puff duration and volume, puff velocity, interpuff interval, and inhalation volume and duration. For smokeless tobacco users, the measures would include number of dips per day, size of dip, dip duration, and interdip interval. Assessment of pattern of product use is also critical, as this could greatly affect individual exposure and risk. This can be assessed in a naturalistic environment by examining (*a*) whether the subject uses the PREP solely or uses the PREP with usual brand tobacco products or other nicotine products; (*b*) the amount of PREP use; (*c*) the time to compensation and stabilization of use; (*d*) the duration of use; and (*e*) the effect of PREP use on the use of conventional products (e.g., eventual cessation, return to usual brand at same rate or reduced rate, or switching to another tobacco product). Equally important for the PREP assessment would be to

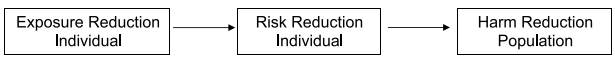

Figure 2. Distinct components for the assessment of PREPs.

Table 1. Challenges and questions associated with clinical trials assessing PREPs

Experimental designs
• What are the primary goals for clinical trials (abuse liability, subjective responses to PREP, tobacco use topography and pattern of use, toxicant exposure, biological effect from the product and/or disease outcome), and what are the experimental designs to address these goals?
• Depending on the goals of the clinical trial, what should be the duration of the study?
• What is the most appropriate control group by which a product should be evaluated?
• What are the limitations and strengths of the different experimental designs to address the goals?
Subject recruitment
• How should subjects be recruited (venues and content of recruitment) for the assessment of PREPs? How should methods of recruitment differ by type and goals of the study?
• What inclusion and exclusion criteria should be used by the studies, and should these criteria differ by type and goals of study?
Subject characteristics
• What subject characteristics should be measured?
• How do volunteers for the study compare with other types of studies (national surveys, people who want to quit, people who do not want to quit)?
• To what extent are the subjects generalizable to the population most interested in trying or using the products? To what extent should the findings from the trials be generalizable?
Subject retention
• Should efforts be made to retain subjects in the study?
• What are the characteristics of dropouts?
• What are the characteristics of those who remain in the study? Do these characteristics reflect those who are interested in using the product?
Compliance with product use
• How should compliance be determined?
• Depending on the goals of the study, how should compliance be maximized?
Predictors of response
• What predictors for PREP response should be used in the study?
Comparative designs
• How should the experimental designs differ from those involved in testing pharmacologic products

determine the extent of toxicant exposure and biological effect from using the product as compared with conventional or usual brand tobacco products, and also compared with either cessation or medicinal nicotine products. To address these main goals, there are different trial designs that can do this, but they generally involve switching a tobacco user from one product to another (e.g., in a randomized trial with crossover or parallel-arm design). Switching studies are particularly challenging because it might be difficult to decipher whether the changes in biomarkers of exposure and effect are due to the product itself, the way the person uses the product, or individual differences in biological response to the product (6).

There is a balance between successful exposure reduction and consumer use. If a product substantially reduces exposure, but consumers do not buy the product, then there would be no harm reduction. Conversely, if a product has a high level of consumer acceptance, but the exposure reduction is minimal, then there is no harm reduction. Therefore, another goal of a clinical trial would be to assess the potential for use of and addiction to the product. This goal would also ensure that products with increased abuse or addiction potential (even with reduced exposure) are not marketed, which might affect future policies such as reducing the addiction potential of all tobacco products to reduce initiation and facilitate cessation (12). These clinical assessments would include determining the pharmacokinetics and pharmacodynamics of a product and subjective responses to the use of the PREP, namely, appeal of the product, satisfaction or liking of the product, withdrawal suppression, and improvement in mood. This assessment would provide initial clues to the extent to which the product would be used or abused.

Materials and Methods

Using PubMed on April 14, 2008, the following search terms were used and searches were limited to humans and English language: reduced exposure products and tobacco; specific PREP product names (e.g., Eclipse, Accord, Advance, Ariva, Snus, Stonewall, Quest, Next, Omni); denicotinized cigarettes and tobacco; light cigarettes and tobacco (limited to last 20 years); ultralight cigarettes and tobacco (limited to last 20 years); low tar and tobacco; low yield and tobacco, electrically heated cigarette smoking system (EHCSS), and smokeless and tobacco. In addition, references cited in each of the articles were searched through for other relevant articles. Studies were selected based on whether or not they assessed a PREP or low-yield cigarettes and if it was a human clinical trial. The data were compiled to examine (*a*) goals of the study; (*b*) experimental designs that were used; (*c*) measures that were used; (*d*) subject recruitment method, content, and inclusion criteria; and (*e*) methods to determine compliance, particularly in studies that involved using the product outside a laboratory setting.

Results

Human studies of the most recent PREPs or studies of conventional products with applicability to PREPs usually involve examining potential for use and addiction to the product (e.g., nicotine pharmacokinetics and product preferences), subjective and physiologic responses, and biomarkers of exposure. These can be divided into five groups of studies, namely, abuse liability assessment studies, in-laboratory clinical trials (subject uses the product once or a few times, but only in a laboratory setting), short-term clinical trials (<2 weeks of duration on a particular product at home or in a residential facility, and the products are used throughout the day), intermediate-term clinical trials (>2 weeks and ≤12 months), and cross-sectional studies. Studies examining the potential for addiction to the product, which can precede short-term and intermediate-term clinical trials, have generally been short in duration and have typically occurred in the laboratory, although a few studies have occurred outside the laboratory. Other in-laboratory clinical trials are limited to studying only exposure assessments for biomarkers with extremely short time to steady-state levels and half-lives [e.g., exhaled carbon monoxide (CO), carboxyhemoglobin, and nicotine]. They also can measure acute physiologic and subjective responses that may provide insight into potential consumer use or interest in the product. These studies are limited because it is unlikely that subjects adapt to the product, stabilize their pattern of use, and compensate for differences in nicotine delivery.

Table 2. Abuse liability assessment: pharmacokinetic and pharmacodynamic studies

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Benowitz (18)	To characterize pharmacokinetics and pharmacodynamics of ST and NRT	Usual cigarettes, oral snuff, oral chew, and nicotine gum	Controlled smoking of one cigarette, single use ST for 30 min, NRT gum for 30 min	Outpatient smoking laboratory over 4 d balanced by use of Latin squares; recruitment through newspapers; inclusion criteria: healthy men, habitual smokers, and prior use with oral snuff and chewing tobacco	Repeated blood pressure, heart rate, and venous blood samples over 120 min; blood sample measures included average nicotine levels and time course of absorption, maximal blood nicotine concentrations, maximal input rate, estimated absorbed doses of nicotine	*N* = 10; all males; age range, 24-61 y
Benowitz (19)	To characterize intake of nicotine and compensation using reduced-nicotine content cigarettes	Research cigarettes with differing levels of nicotine	*Ad libitum* smoking of 1 cigarette in a smoking laboratory	Outpatient smoking laboratory with semi-double-blinded, within-subject, crossover design, with 6 treatments (usual brand and 5 research cigarettes) balanced by use of Latin squares; recruitment through newspaper and internet; inclusion criteria: healthy (by history), smoke 10 CPD; exclusion criteria: taking cardiovascular or psychiatric medications, pregnant, current alcohol or drug abuse	Smoking behavior (time to smoke cigarette, puffs); plasma nicotine, plasma cotinine, nicotine boost, AUC_{nic}, blood carboxyhemoglobin concentrations, carboxyhemoglobin boost, AUC_{COHb}; ratio of nicotine intake to nicotine content, ratio of nicotine intake to FTC nicotine, compensation based on FTC nicotine, estimated tar exposure; blood pressure, heart rate, exhaled CO, fingertip skin temperature; subjective responses to different contents of nicotine, cigarette acceptability questionnaire (adapted from the Duke Cigarette Evaluation Scale and the Duke Sensory Questionnaire)	*N* = 12; 6 males, 6 females; mean age, 27 y; mean CPD, 17; mean years smoked, 10.4; mean FTND score, 3.9
Fant (17)	To assess nicotine levels produced by ST and physiologic and subjective effects	Four different ST and non-nicotine snuff	Use of each product for 30 min	Five experimental sessions, one for each product, balanced by use of Latin squares; recruitment through newspapers for ST users	Repeated blood samples over 90 min for nicotine analysis; heart rate, blood pressure, electroencephalographam; subjective rating of product strength and sensations	*N* = 10; mean age, 32.2 y; mean dips per day, 6.4; mean years of ST use, 12.5; a test of smokeless tobacco dependence in which scores can range from 4 to 19, mean score, 9.6

(*Continued on the following page*)

Table 2. Abuse liability assessment: pharmacokinetic and pharmacodynamic studies (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Kotlyar (21)	To evaluate nicotine pharmacokinetics and subjective effects of ST PREPs, moist snuff, and nicotine lozenge	Ariva, Stonewall, Revel, Commit, and Copenhagen	Use of each product for 30 min	Randomized, within-subject, crossover study; one product at each of the five laboratory sessions; recruitment through flyers and advertisements in the local media; inclusion criteria: 18-65 y old and Copenhagen ST use daily for ≥1 y; exclusion criteria: subjects with unstable medical or psychiatric conditions, taking drugs likely to interact with the products being tested, using any other tobacco or nicotine products, severe periodontal or other oral lesions, history of substance misuse within past year	Repeated blood samples to assess maximal nicotine concentration and AUC; nicotine craving, withdrawal symptoms; ratings of product effects and liking the product	*N* = 10; all males; mean age, 30.9 y; mean tins per week, 2.4; mean dips per day, 8.1
Lunell and Lunell (22)	To evaluate plasma nicotine levels for ST and NRT gum and estimate the amount of sodium chloride–extracted ST that might affect heart failure and hypertension	Swedish ST and NRT gum	Use of ST and NRT gum for 30 min	Crossover design, open-label, partly randomized; subjects were given 12-hourly repeated doses of 4 different types of ST administered in randomized order; NRT administered on separate occasion; inclusion criteria: healthy, nonsmoking for at least 1 y, regular snus users	Repeated blood samples to measure maximum nicotine plasma concentration and time to peak plasma concentration; residual nicotine and sodium chloride levels in used snus, extracted dose of nicotine from the tobacco product	*N* = 12; all males; age range, 18-23 y

Abbreviations: AUC, area under the concentration time curve; COHb, carboxyhemoglobin; CPD, cigarettes per day; ST, smokeless tobacco.

Short-term clinical trials less than 2 weeks long have similar limitations, although the available biomarkers that can be used increase where the time to steady-state and half-lives can be longer (but less than 2 weeks). Intermediate-term studies provide more information about toxicant exposures where biomarkers with longer half-lives and physiologic responses after the subject has adapted to the product can be assessed. Cross-sectional studies of tobacco-users in the population can provide information about longer periods of product use, larger numbers of subjects, outcome measures on persons who self-selected their use of a product, and data analysis by subgroups. However, cross-sectional studies are limited because they do not describe changes in use over time or information about delayed quitting and cessation and are subject to cohort effects.

The study designs, outcome measures, subject recruitment methods, subject characteristics, and product compliance procedures are described for each of the five categories or types of studies. A summary of the results, Clinical Trials Workshop recommendations, and future research questions with regard to study design are addressed under each of the study type subsections. The summary and Workshop recommendations are combined for the short-term and intermediate-term trials. Clinical Trials Workshop recommendations for issues that are cross-cutting are reserved for later discussion.

Abuse Liability (Pharmacokinetics, Drug Choice) and Other In-Laboratory Studies

Study Design. The assessment of harm reduction potential of a PREP should include the assessment of its abuse liability, a term used interchangeably with abuse potential. Abuse liability traditionally refers to the likelihood of addiction to the product based on product characteristics (e.g., level and rate of free nicotine delivery, flavorants, and method of use). However, in the broader sense of the term, abuse liability can also refer to the population effects of the product and involves the interaction between the product and the user as well as the social and environmental context for its use (e.g., peer uptake, product marketing, and cost). For purposes of this review, the more narrow assessments of abuse liability will be addressed. Historically, abuse liability studies have been conducted to examine the potential for abuse of prescription or over-the-counter medications [e.g., sedative-hypnotics, barbiturates, pain medications, and nicotine replacement therapies (NRT)] or to examine the relative abuse potential of existing or emerging recreational drugs and medications. Several excellent supplemental journal issues have been written on methods for the assessment of drug abuse liability (13-15). Fewer human clinical trial studies have been conducted in assessing the abuse liability of PREPs.

One method to assess abuse liability is to measure the nicotine pharmacokinetics and pharmacodynamics of a product (refs. 17-22; see Table 2); the faster the absorption of nicotine and the greater the amount of initial nicotine delivery, the greater the subjective response and potential for abuse (20). By this measure, tobacco products with the highest potential for abuse are cigarettes, and those with the lowest potential for abuse would be nicotine patches (see Fig. 3). Even within products, such as smokeless tobacco, a significant variability in nicotine pharmacokinetics is observed (refs. 17, 21, 22). Typically, these studies use a within-subject, crossover design in which subjects are assigned to all products. Some of these studies have included medicinal nicotine as a comparison to the tobacco products (16, 17, 21, 22) or a non-nicotine tobacco-like product (e.g., ref. 17). Subjects are required to be abstinent overnight, report to the laboratory in the morning, where abstinence is verified (CO in the case of cigarette smoking or a reduced cotinine level in the case of smokeless tobacco), and a sample of the product is administered. Multiple blood samples are taken to determine time to maximum plasma nicotine concentration (T_{max}), maximum plasma nicotine concentration (C_{max}), area under the curve, as well as half-life ($t_{1/2}$) and clearance (CL). Another biomarker that has been used is carboxyhemoglobin, and in one study, the extracted dose of nicotine from the tobacco product was examined (22). Also during this time, vitals and/or skin temperature are assessed, as well as subjective responses to a product. Such subjective responses include withdrawal symptoms and craving; drug liking (e.g., whether they felt any good effects from the study product, how satisfying the product was, how much they liked the study product, how much they desired the study product, how strong the study product was), and drug effect (e.g., felt any bad effects from the study product; felt alert; felt relaxed; felt a head rush or high; felt a tremor in hands, arms, or face; felt light-headed/dizzy; felt drowsy; felt energetic or stimulated; felt jittery); and product evaluation (e.g., strength, smoothness, flavor quality, satisfaction, comparison to usual cigarettes, estimate of nicotine yield). In the case of a study conducted by Benowitz et al. (19), in which a major focus was to determine compensatory smoking behavior in cigarettes that differed in nicotine content but not tar yield, the measures included the ratio of nicotine intake/content and ratio of nicotine intake/Federal Trade Commission (FTC) nicotine yield.

Another method for assessing abuse liability is to examine the relative preference for the PREP relative to

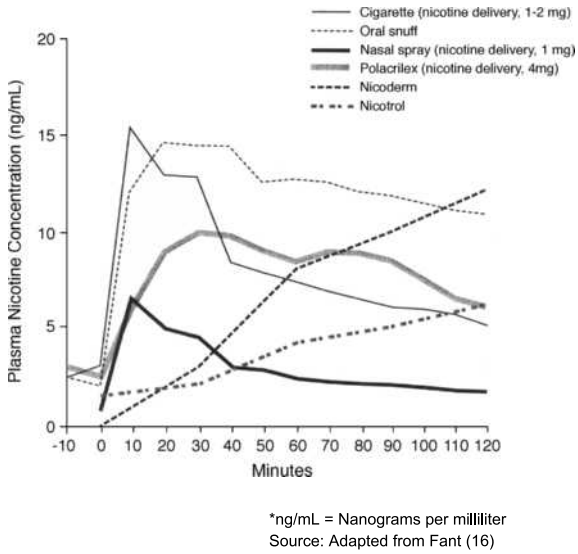

Figure 3. Venous blood concentrations of nicotine over time for various nicotine delivery systems (16).

other products or another PREP using a forced-choice paradigm (see Table 3). These studies typically involve sampling each of the products and then forcing the subject to choose one product over another (ref. 23; see below). The product sampling phase is conducted within a session (24) or over the course of several days and weeks (e.g., ref. 23). Another option is to allow the subject to have concurrent access to the products and allow the subject to choose any of the products over the course of this choice phase or even throughout the course of the study, where the number of choices made of each of the products would be calculated. Subjective responses of withdrawal, product liking, strength of effect, and product evaluation of other characteristics are also assessed. PREPs can be compared with conventional tobacco products (e.g., either high or low nicotine yield delivery), medicinal nicotine, or other PREPs. Therefore, these models can provide clues about the abuse potential or preference for a PREP compared to conventional highly addictive products, to medicinal nicotine (safer) products, and over another PREP. To date, few studies on PREPs have used this type of experimental paradigm.

Another paradigm that has been used to examine PREPs involves examining the extent to which a person would work to obtain a product and the extent to which a particular product substitutes for another product and at what cost (price elasticity). As an example, in one outpatient laboratory study, cigarette-deprived dependent smokers worked for standardized cigarette puffs by pulling on a plunger on a progressive-ratio schedule (increasing number of pulls for each puff) for either nicotinized or denicotinized cigarettes. These cigarettes were provided alone or concurrently with the opportunity to earn money (25). Another variation included using the same paradigm in which subjects earned standardized puffs on both types of cigarettes when provided alone, except in another phase, where the subjects chose between the two cigarette types (26).

The two cigarettes in both studies were compared on such measures as the breakpoints (the ratio at which subjects no longer worked for a puff), number of puffs earned per session, peak response rates, ratio producing peak response rates, and the demand elasticity for cigarette puffs across a range of prices (number of required plunger pulls). In addition, cigarettes were rated on subjective measures (e.g., taste, drug effect, smoothness, enjoyment, the amount subjects would pay per pack of each type of cigarette). In another study, nicotine-containing cigarettes were available at increasing unit price (increasing number of plunger pulls) with nicotine gum, denicotinized cigarettes, or both concurrently available at a fixed price (e.g., fixed number of plunger pulls; ref. 27). The outcome measure was cross-price elasticity (point at which smokers switched to the alternative product) for each alternative that was offered at a fixed priced. In this paradigm, the preference between two concurrently offered products can be determined as well as the reinforcing value of the alternative compared with usual brand cigarettes. In addition, withdrawal and smoking urges also are measured to determine if the products relieve both withdrawal and urges to smoke, and if these variables would affect behavioral responses. Similar to the forced-choice paradigm, only a few studies have used this type of experimental design to examine PREPs.

Other components for the assessment of abuse liability of a product include withdrawal relief from a product as a result of switching from usual brand to a PREP, withdrawal effects from the product, how much the product is used, occurrence of compensatory tobacco use behavior or dose escalation of the product over time, and dependence on the product. These studies can be conducted within a laboratory (as described below) or in short-term or intermediate-term studies.

Several groups have published in-laboratory clinical studies where subjects are tested using products in the laboratory to assess acute subjective and physiologic responses to a product and biomarkers of exposure (refs. 28-46; see Table 4). These studies vary in the number of PREPs tested, whether PREPs are used *ad libitum* or in a controlled manner, whether one or more products are provided within a laboratory session or across several sessions, and duration of the session. Eissenberg and his colleagues have conducted several of these laboratory studies. In their experimental designs, laboratory sessions are typically held after overnight abstinence and subjects would participate in a within-subject, crossover design involving a 2.5-hour session. Subjects were asked to complete an 8-puff smoking bout every 30 minutes, with each session involving one of three to four different products (28, 29, 35). A similar laboratory design (e.g., four 30-minute episodes of oral tobacco product use over a 4.5-hour session) has been used with smokeless tobacco users (44). Other within-subject laboratory studies have varied the way in which cigarettes are smoked. For example, studies have asked smokers to smoke two cigarettes with different nicotine yields either rapidly (up to nine cigarette puffs every 6 seconds) or at a normal pace (36), *ad libitum* during a 5-hour session (41), *ad libitum* at 30, 60, and 240 minutes during a 240-minute session (37), or every 30 minutes over 2 hours (34), or smoking one PREP consecutively in a standard way (e.g., taking large puffs every 30 seconds and inhaling as deeply as possible, with the subsequent PREP smoked 45 minutes later; ref. 33).

Other studies have had participants smoke two or three different cigarettes (e.g., denicotinized versus nicotinized cigarettes) during independent sessions without controlling for other variables (30) or smoke one cigarette *ad libitum* during independent sessions varying the length of abstinence before the session (30, 39, 43). Laboratory studies rarely provide a trial period for the product before the laboratory session; however, one study allowed 2 weeks of acclimation to the cigarette before laboratory testing (32). When comparing the instructions for product use on outcome measures across studies examining similar products, the direction of results tends to be the same whether the subjects smoked *ad libitum* or at a fixed rate (28, 34-36, 39). However, because this observation is made across studies, no quantitative comparisons could be made.

Another unique study asked subjects to smoke different combinations of five denicotinized and nicotine cigarettes (i.e., 0, 1, 2, 3, 4 or 5 denicotinized cigarettes out of 5 total cigarettes) during each study day (40). Other noncigarette studies have examined the effects of oral tobacco products given in increasing doses every 90 minutes (e.g., 1 Ariva, 2 Arivas 90 minutes later, followed by 3 Arivas 90 minutes later; ref. 45). Only one study was conducted with adolescents, where subjects were asked to smoke

Table 3. Abuse liability assessment: drug choice and self-administration

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Johnson (27)	To determine the substitutability of denicotinized cigarettes and/or nicotine gum for nicotine-containing cigarettes by implementing a behavioral economic substitution design	Two types of cigarettes produced by the University of Kentucky Tobacco and Health Research Program (nicotine-containing cigarettes with 1.16-mg nicotine and 15-mg tar and denicotinized cigarettes with 0.16-mg nicotine and 1.67-mg tar) and 4-mg nicotine gum	Computer-directed controlled smoking and nicotine gum chewing conditions	Outpatient laboratory study; nicotine containing cigarettes available at increasing unit prices (increase in progressive-ratio schedule) across sessions with nicotine gum or denicotinized cigarettes or both concurrently available at a constant unit price; exclusion criteria: current use of psychoactive medications, a recent history of medical problems, psychiatric disorders, or drug dependence, plans to quit smoking	Exhaled CO; cross-price elasticity; cigarette puffs; nicotine gum used; Questionnaire of Smoking Urges, Minnesota Nicotine Withdrawal Scale	$N = 6$; mean age, 31.3 y; mean CPD, 20.8; mean FTND score, 7.2
Mendoza-Baumgart (23)	To determine product preference and the effects of ST products compared with medicinal nicotine	Exalt ST or Ariva vs MNL	Complete substitution of these products for smoking; use of product at least every 2 h	Experimental crossover study design involving a sampling phase consisting of two 2-wk periods, where subjects sampled either the oral tobacco or medicinal product first and crossed over to the other product in randomized order and a 1-wk drug choice phase; recruitment through advertisements in local and university newspapers, flyers, and the radio; inclusion criteria: 18-65 y old, good physical and mental health, and regular smoker; exclusion criteria: currently using other types of tobacco products, using any methods for cutting down on tobacco use, pregnant or breast-feeding	Urine samples to analyze total cotinine, total nicotine, and total NNAL, WBC and hemoglobin; exhaled CO, heart rate, blood pressure; questionnaires and daily diaries to assess tobacco and nicotine use status, Minnesota Nicotine Withdrawal Scale, Drug Effects and Liking Visual Analogue, percent preference for a product during the drug choice phase	Study 1: $N = 39$ (19 MNL-Exalt, 20 Exalt-MNL); 18 males, 21 females; mean age, 38.7 y; mean CPD, 21.3; 34 White non-Hispanics; mean FTND score, 5.9; Study 2: $N = 26$ (12 MNL-Ariva, 14 Ariva-MNL); 10 males, 16 females; mean age, 35.7 y; mean CPD, 20.9; 18 White non-Hispanics; mean FTND score, 6.0

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Table 3. Abuse liability assessment: drug choice and self-administration (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Shahan (26)	To compare the reinforcing efficacy of denicotinized cigarettes and conventional cigarettes	Conventional (light and regular) and denicotinized research cigarettes	Controlled puffing by computer cue	A progressive-ratio schedule for the number of plunger pulls needed for cigarette puffs increased across sessions; responding for the two types of cigarettes was examined when each was available alone and when both were concurrently available; recruitment through local advertising; inclusion criteria: good health, no current psychiatric problems and related medications, drug or alcohol abuse	Measures of elasticity of demand across the range of prices, breakpoints, peak response rates, ratio producing peak response rates; number of puffs consumed; self-reports of taste, drug effect, smoothness, and enjoyment; maximum amount of money subjects would pay per pack for each type of cigarette	*N* = 8; 6 males, 2 females; age range, 19-48 y; mean CPD, 31.3; mean FTQ score, 7.6; mean expired CO, 32.9 ppm
Shahan (25)	To evaluate the self-administration of nicotine-containing and denicotinized cigarettes by assessing the effects of alternative nondrug reinforcement	Conventional (light and regular) and denicotinized research cigarettes	Puffs were earned by pulling on one or more of the brass plungers, with changing cigarettes each day; controlled puffing by computer cue	A progressive-ratio schedule for the number of plunger pulls needed for cigarette puffs increased across sessions; cigarettes were available alone or concurrently with money as an alternative reinforcer; recruitment through local advertising; inclusion criteria: good health, no current psychiatric problems and related medications, drug or alcohol abuse	Number of puffs for each cigarette type, number of puffs consumed per session as a function of unit price, breakpoints, peak response rates, ratio producing peak response rates, demand elasticity for cigarette puffs across a range of prices	*N* = 8; 6 males, 2 females; age range, 30-52 y; mean CPD, 28.1; mean FTQ score, 7.6; mean baseline expired CO, 36.3 ppm

Abbreviation: MNL, medicinal nicotine lozenge.

Table 4. In-laboratory studies

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Blank (45)	To determine the effects of Ariva in cigarette smokers	Ariva tablets	Participants allowed each dose to dissolve in their mouths according to package instructions	Single-session, clinical laboratory study of smokers given Ariva tablets (1, 2, and 3 tablets in ascending order), at 90-min intervals; inclusion criteria were healthy individuals, 18-50 y old, expired air CO ≤15 ppm, smoke at least 10 CPD for at least 1 y; exclusion criteria: chronic health problems, current pregnancy or breast-feeding, history of or active cardiovascular disease, and regular use of prescription medication	Plasma nicotine; heart rate, blood pressure; measures of nicotine/tobacco withdrawal symptoms, 10 VAS items that included known nicotine effects	*N* = 10; 5 males and 5 females; mean age, 32.8 y; mean CPD, 22.0; mean exhaled CO, 22.7 ppm
Breland (28)	To assess a clinical laboratory procedure for measuring the effects of PREPs	Usual brand, Accord, Eclipse, denicotinized cigarettes	Eight self-paced puffs every 30 min for each product	Four Latin-square ordered 2.5-h sessions with an 8-puff smoking bout every 30 min for 4 smoking bouts; recruitment through advertisements and word of mouth; inclusion criteria: 18-50 y old, expired air CO ≥15 ppm, king-sized, nonmentholated, light, and ultralight smokers smoking at least 15 CPD; exclusion criteria: current pregnancy or breast-feeding, history of active cardiovascular disease, previous use of Eclipse or Accord, and smoking cessation or reduction efforts	Puff topography; plasma nicotine; heart rate, skin temperature, exhaled CO; tobacco withdrawal symptoms, Questionnaire of Smoking Urges	*N* = 20; mean age, 21.6 y; mean CPD, 18.9; mean FTND score, 5.6; mean exhaled CO, 24.3 ppm; mean BMI, 26.4 (males), 23.4 (females)
Breland (29)	To examine the acute effects of Advance on biomarkers, subjective responses, and smoking topography	Usual brand, Advance (both with 100% ventilation hole blocking)	Eight self-paced puffs every 30 min for each product	Three Latin-square ordered 2.5-h sessions in which they completed an 8-puff smoking bout every 30 min, including sham cigarette; recruitment through advertisements; inclusion criteria: 18-50 y old, expired air CO ≥15 ppm, and king-sized, nonmentholated, light, and ultralight smokers smoking at least 15 CPD; exclusion criteria: current pregnancy or breast-feeding, history of or active cardiovascular disease, previous use of Advance, and smoking cessation or reduction efforts	Puff topography; plasma nicotine; heart rate, blood pressure, exhaled CO; VAS items described tobacco/nicotine withdrawal symptoms, Questionnaire of Smoking Urges	*N* = 20; 10 males and 10 females; mean age, 25.8 y; mean CPD, 20; mean FTND score, 5.3; mean CO, 25.1 ppm

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Table 4. In-laboratory studies (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Buchhalter (34)	To evaluate the effects of Accord on subjective and physiologic measures	Usual brand, Accord	Fixed interval between cigarettes and then *ad libitum*	2 sessions, during which subjects smoked their own brand or Accord; order of the sessions was counterbalanced across subjects; subjects smoked single cigarettes at 30-min intervals for 2 h and then *ad libitum* for 1 h; recruitment through advertisements and word of mouth; inclusion criteria: light or ultralight smokers, smoking 10 or more CPD, 18-60 y old, exhaled CO >7 ppm; exclusion criteria: previous Accord use, past or current cardiovascular disorders, current pregnancy or breast-feeding	Puff topography; heart rate, skin temperature, blood pressure, exhaled CO; thirteen VAS items to assess tobacco withdrawal, Questionnaire of Smoking Urges	*N* = 10; 7 females and 3 males; mean age, 23.2 y; mean CPD, 15.3; 2 Blacks and 8 Whites; mean FTND score, 4.0; mean exhaled CO, 14.6 ppm
Buchhalter (35)	To examine the acute effects of Accord compared with own brand, ultralight cigarettes, and denicotinized cigarettes	Usual brand, Merit Ultra Light, Accord, and denicotinized cigarettes	Eight self-paced puffs every 30 min for each product	Smokers abstinent for at least 8 h participated in 4 Latin-square ordered 2.5-h sessions in which they completed an 8-puff smoking bout every 30 min; recruitment through advertisements and word of mouth; inclusion criteria: light or ultralight smokers, smoking 10 or more king-sized, nonmentholated CPD, 18-60 y old, exhaled CO >7 ppm; exclusion criteria: previous Accord use or Merit Ultra Light smokers, past or current cardiovascular disorders, current pregnancy or breast-feeding, current attempt to quit or reduce smoking	Puff topography; heart rate, skin temperature, blood pressure, exhaled CO; tobacco withdrawal symptoms, Questionnaire of Smoking Urges	*N* = 32; 16 females and 16 males; mean age, 25.9 y; mean CPD, 20.6; 3 non-White; mean FTQ score, 4.7; mean CO, 21.1 ppm; mean BMI, 23.5 (females), 25.7 (males)
Butschky (40)	To assess tobacco withdrawal and liking after smoking varying numbers of denicotinized and regular cigarettes	Regular conventional cigarettes, Next, and a lettuce cigarette	Controlled puffing conditions	Five cigarettes were smoked per day (nicotinized or denicotinized cigarettes), varying cigarette dose (5 lettuce cigarettes, 4 lettuce leaf cigarettes and 1 nicotinized or denicotinized cigarette, 3 lettuce leaf cigarettes and 2 nicotinized or denicotinized cigarettes, etc.); recruitment through existing subjects from an addiction research center; inclusion criteria: regular smoker of at least one pack per day	Plasma nicotine and cotinine; blood pressure, pulse, oral and skin temperature, pupil diameter, exhaled carbon monoxide; tobacco withdrawal symptoms, ratings of cigarettes	*N* = 7; all males; mean age, 35.7 y; mean CPD, 26.4; mean nicotine yield, 1.15 mg; mean FTND score, 8.43

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Table 4. In-laboratory studies (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Dallery (36)	To compare craving and the effects of rapid smoking using nicotinized and denicotinized cigarettes	Usual brand, research nicotinized and denicotinized cigarettes	Up to 9 cigarettes, with rapid and self-paced conditions	Within-subject design with one session each of rapid smoking (up to 9 cigarettes with puffs every 6 s) and normal-paced smoking with nicotinized and denicotinized cigarettes; during the 3-h post-intervention period, participants were allowed to smoke every 15 min of usual brand cigarettes at a self-regulated pace; inclusion criteria: >15 CPD, exhaled CO >15 ppm, 18-55 y old; exclusion criteria: medical or psychiatric illness that would significantly interfere with the study, abnormal ECG, pregnancy, and drug abuse	Puff topography; plasma nicotine; heart rate, blood pressure, exhaled CO; Withdrawal Symptoms Questionnaire, Nicotine Effects VAS, Desire to Smoke VAS, Questionnaire of Smoking Urges, Shiffman-Jarvik Smoking Withdrawal Questionnaire, Cigarette Effect Questionnaire	*N* = 15; 8 females and 7 males; mean age, 35 y; mean CPD, 25; and mean exhaled CO, 25 ppm
Eid (37)	To assess the effects of recentness of smoking and nicotine delivery on subjective and physiologic measures	Research conventional and denicotinized cigarettes	Smoked *ad libitum* every 30, 60 or 240 min up to 240 min	Within-subject design with 6 sessions over 240 min, where the time intervals and research cigarettes varied (3 × 2 design); recruitment through newspaper and word of mouth; inclusion criterion: good health	Heart rate, exhaled CO; four-item visual analogue questionnaire on cigarette craving, the short form (10 items) of the Questionnaire of Smoking Urges	*N* = 8; 4 males, 4 females; mean age, 35.8 y; mean CPD, 30.6; mean nicotine yield, 1.2 mg; 5 Blacks and 3 Whites; mean FTND score, 7.0
Gross (30)	To compare the sensory and reinforcing effects of denicotinized and nicotinized cigarettes	Usual brand, denicotinized, and commercial light cigarettes	Controlled smoking	Within-subject design with 3 experimental sessions; subjects had a morning session smoking their own brand cigarette and afternoon session smoking a total of 20 puffs (4 puffs/cigarette, 5 cigarettes) from one of three cigarettes; recruitment through local community-based advertising; inclusion criterion: not interested in quitting	Puff topography; plasma nicotine; heart rate, exhaled CO; 3-item craving/satisfaction measure using a computerized VAS, an 8-item cigarette characteristics measure to rate the sensory and reinforcing properties of the cigarettes, craving or urge for a cigarette using a 4-item measure, tobacco withdrawal scale	*N* = 10; 7 males and 3 females; mean age, 38.2 y; mean CPD, 28.3; mean nicotine yield, 1.07 mg; mean FTND score, 8.3

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Table 4. In-laboratory studies (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Kassel (46)	To determine topography in young smokers using denicotinized cigarettes	Research high nicotine yield and denicotinized cigarettes	*Ad libitum*	Between-subject design in which each participant was given the opportunity to smoke one research cigarette and was blind to the nicotine content of the cigarette; recruited subset from larger ongoing study; inclusion criteria: 15-18 y old, smoking for at least 4 wk, 1 cigarette a week, but no more than five cigarettes a day on average	Smoking topography; exhaled CO; mean FTQ, craving subscale of the Shiffman-Jarvik Smoking Withdrawal Questionnaire, cigarette ratings, smoking behavior	*N* = 35 (19 denicotinized, 16 nicotinized); 18 females and 17 males; mean CPD, 3.6; mean age, 17.5 y; 43% Caucasians, 25% Asia-Pacific Islanders, 17% Hispanics, 6% African Americans and 9% others; mean FTQ score, 2.2
O'Connor (38)	To determine topography and evaluate characteristics of flavored and unflavored cigarettes in college students	Commercial light cigarettes and flavored cigarettes of their choice	*Ad libitum*	Within-subject design, 60-min laboratory session; subjects smoked the first cigarette (light or flavored cigarette first); after a 30-min washout period, subjects repeated the procedure with the other cigarette brand; recruitment from local college; inclusion criteria: 18-30 y old, having smoked at least 100 lifetime cigarettes, currently smoking a nonmenthol brand for at least 6 mo, not trying to quit	Puff topography; CO boost; cigarette characteristics	*N* = 20; 10 male, 10 females; mean CPD, 11.5
Pickworth (39)	To examine the pharmacologic effects of denicotinized and conventional cigarettes as it relates to nicotine and other tobacco smoke components	Four types of research cigarettes: reduced-tar and full-tar standard cigarettes and reduced-tar and full-tar denicotinized cigarettes	*Ad libitum*	Double-blind outpatient laboratory study with 4 experimental sessions; subjects randomly assigned to either smoke full-tar conventional and denicotinized cigarettes or smoke reduced-tar conventional and denicotinized cigarettes; on 2 of the experimental days, subjects were abstinent for 3 h before the session; for the other 2 experimental days, subjects were abstinent overnight; exclusion criteria: any chronic physical or mental health conditions requiring medication, current drug or alcohol addiction, use of other tobacco products, current treatment for smoking cessation	Smoking topography; plasma nicotine and cotinine; blood pressure, heart rate; exhaled CO; Minnesota Nicotine Withdrawal Scale, Questionnaire on Smoking Urges (short version), drug liking question, cigarette characteristics	*N* = 20 (10 in each condition); mean age, 34.1 y; mean CPD, 31; mean nicotine yield, 1.2 mg; mean FTND score, 8.0

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Table 4. In-laboratory studies (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Rose (31)	To examine smoking topography when subjects smoked a low-nicotine cigarette	Low nicotine cigarettes and commercial highly ventilated cigarettes	*Ad libitum*	During two 8-h sessions, subjects smoked one of each type of cigarette in a counterbalanced order; recruitment through the community by newspaper and radio advertisements and by word of mouth; inclusion criteria: 18-65 y old and smoked at least 15 CPD of a brand of cigarette having an FTC nicotine yield of at least 0.5 mg.; exclusion criteria: major medical problems based on physical examination, ECG, serum chemistries, complete blood count, and urinalysis, a specific intention to quit within the next 6 mo; assessment of compliance with overnight smoking abstinence by CO and nicotine	Smoking topography; plasma nicotine and cotinine concentrations; heart rate, blood pressure, exhaled CO; Shiffman-Jarvik questionnaire for craving, negative affect, and arousal, cigarette evaluation questionnaire, sensory questionnaire; signs of vent blocking	*N* = 16; 8 males, 8 females; mean age, 36 y; mean CPD, 25; mean nicotine yield, 0.74 mg; 8 Whites, 8 Blacks; mean FTND score, 6.4; mean baseline CO, 27 ppm
Russell (41)	To examine the role of nicotine yield as a determinant of the amount of cigarettes smoked	One high-nicotine cigarette (Capstan Full Strength) and one low-nicotine cigarette (Silk Cut Extra Mild)	*Ad libitum*	Within-subject design; cigarette consumption was studied over four 5-h periods, on 4 separate days; 2 consecutive days of 1 wk and the same 2 d of the following week; on the first day of each week's dyad, the subjects smoked their usual brand of cigarette, and on the second day, they smoked either a high- or a low-nicotine cigarette; order of the high- or low-nicotine cigarette was randomized; inclusion criterion: regular cigarette smokers who inhaled deeply	Pre- and post-session blood was taken after smoking 1 cigarette; carboxyhemoglobin; VAS for comparing the different brands of cigarette on subjective ratings of satisfaction, strength and taste evaluation, number of cigarettes smoked	*N* = 10; 6 females, 4 males; mean age, 30 y; mean CPD, 27.2; mean nicotine yield, 1.34 mg
Schuh (42)	To examine the effects of two non-nicotine cigarettes with differing amounts of tar	Two types of nonmenthol, non-nicotine cigarettes	Controlled	Within-subject design; 1-h abstinence before session; subjects then smoked cigarettes during a sampling phase in a random, counterbalanced order so they smoked both the high- and low-tar cigarettes; after the sampling phase, half of the subjects smoked the low-tar cigarette and the other half of the subjects smoked the high-tar cigarette; subjects were asked which cigarette they thought they had smoked (high- or low-tar) at 5, 30, 60, 300, and 900 s after the first puff; recruitment through a clinical trial examining fluoxetine for smoking cessation	Heart rate, blood pressure, CO; VAS to provide information on drug effects and taste qualities of the cigarettes adapted from previous research	*N* = 18; 10 females and 8 males; mean age, 40.2 y; mean CPD, 21.8; mean nicotine yield, 0.83 mg; 8 African Americans, 10 Caucasians

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Table 4. In-laboratory studies (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Sepkovic (32)	To assess the smoking behavior of subjects who switched to a higher nicotine content cigarette	Usual brand and a higher-yield brand	*Ad libitum*	Three laboratory sessions (2 wk after *ad libitum* smoking of usual brand, after the introduction of the higher-nicotine cigarette, and 2 wk after acclimation to the higher-nicotine cigarette); subjects smoked their usual brand for 2 wk; they then switched to a cigarette with an average increase in nicotine of 0.34 mg for 2 wk; all subjects abstained from smoking for 3 h before reporting to the laboratory; inclusion criteria: good health with no history of diabetes, hypertension or respiratory tract disorders, and no interest in smoking reduction	Plasma cotinine, nicotine and thiocyanate; blood carboxyhemoglobin; blood pressure, pulse	*N* = 7; mean age, 25.7; mean CPD, 33
Stapleton (33)	To measure nicotine levels obtained from Eclipse cigarettes	Eclipse	Controlled	After at least 12 h of abstinence, subjects smoked an Eclipse cigarette, every 30 s taking large puffs and inhaling as deeply as possible until it went out; 45 min after finishing the cigarette, a second Eclipse was smoked	Blood nicotine levels, blood nicotine boost per cigarette, CO boost, heart rate; subjective effects experienced before and after smoking	*N* = 4; all males
Strasser (43)	To determine if compensatory smoking occurred with a low-nicotine cigarette	Quest cigarettes (nicotine content 0.6, 0.3, and 0.05 mg)	*Ad libitum*	Within-subject laboratory study; subjects smoked the 3 different Quest cigarettes in randomized order in the same session, counterbalanced across subjects, double-blinded; cigarettes were smoked *ad libitum* with 30-min intervals between cigarettes; recruitment through community based flyers; inclusion criteria: >18 y old, at least 10 CPD, 5 y of minimum smoking, not currently trying to quit smoking, reported inhaling when smoking; exclusion criteria: reported trying to quit smoking, including current use of NRT, reported consuming more than 25 alcohol drinks/wk, use of Quest	Smoking topography; CO boost; rating scale of cigarette features; speculation of which color cigarette had the most and least amount of nicotine; FTND, rating scale of cigarette features	*N* = 50; 54% male, 46% female; mean CPD, 21.3; mean age, 44.5 y; mean nicotine yield, 0.94; 72% White, 24% Black, 2% Native American, 2% Asian American, 4% Hispanic; mean FTND score, 5.5; mean BMI, 26.7

Abbreviations: ECG, electrocardiogram; VAS, visual analogue scale.

one of two cigarettes differing in nicotine yields in a between-subject study design (46).

Measures. A variety of measures are typically used in abuse liability studies. Subjective measures have included (*a*) nicotine withdrawal typically using a modified Minnesota Nicotine Withdrawal Scale (28-30, 34-36, 39, 45) or Shiffman-Jarvik Scale (31, 36); (*b*) smoking urges or desire to smoke using the Questionnaire of Smoking Urges or other measures (28-30, 34-37, 39); (*c*) subjective responses to the product using such scales as the Nicotine Effects Visual Analogue Scale [nausea, clammy skin, dizziness, light headed, burning throat, tingling sensations, and heart racing; Houtsmuller and Stitzer (1999), cited in a study by Dallery et al. (36)], the Cigarette Effect Questionnaire [pleasant, unpleasant, like taste, dislike taste, smoke versus air (anchored with mostly smoke to mostly air), harsh, strength, high in nicotine, like drug effect, dislike drug effect, satisfying, more awake, more calm, easier to concentrate, and less irritable; Gross et al. (1997), cited in a study by Dallery et al. (36)], or Cigarette Evaluation Scale (satisfaction, psychological reward, nausea or dizziness, craving relief, and enjoyment or airway sensation; ref. 31); (*d*) Sensory Questionnaire (estimated nicotine delivery, similarity to usual brand, and perceived strength on the tongue, nose, back of mouth and throat, windpipe, and chest; ref. 31); or (*e*) other scales that measure variables such as strength, mildness, taste, satisfaction, pleasantness, harshness, heat, smell, ease of draw, similarity to own brand of cigarettes, good effects, and bad effects (30, 38-42, 46).

Physiologic measures used in prior studies included heart rate, blood pressure, pupil diameter, and/or skin temperature (28-30, 32, 34-37, 39, 42, 45).

Smoking topography assessments included measures such as puff volume, duration, interpuff interval, maximum flow rate velocity (28, 29, 36, 46), number of puffs in those studies that did not control for this variable (31, 34, 35, 38, 39, 46), or signs of vent blocking (31).

Recruitment Method. Advertisements for these prior studies were mostly through various media, flyers, and word of mouth. Two studies recruited subjects from other studies (40, 42).

Subject Characteristics. The majority of the abuse liability studies recruited subjects who were physically and mentally healthy, currently not taking psychiatric medications or taking medications or products that would interact with the product tested, and who were not dependent on or misusing other substances of abuse. Some studies stated that they excluded pregnant smokers (19) or smokers who had plans to quit smoking (27). The number of subjects for these studies typically ranged from 8 to 12, although one study had a number as high as 39 (20). The subject characteristics varied across studies from a relatively young, less dependent population (e.g., ref. 16) to a relatively young but more dependent population (27).

For in-laboratory clinical studies, the inclusion criteria for adult subjects included being in general good health and, for some studies, a specified age range (18 to up to 65; refs. 28, 29, 31, 34-36, 38, 45); a specified number of cigarettes smoked, ranging from at least 10 or 15 cigarettes per day (28, 29, 31, 34-36, 43, 45) or at least 100 lifetime cigarettes (38); a specific type of cigarette smoked, such as nonmenthol, light, or ultralight cigarettes depending on the study product being examined (28, 29, 35, 38); a specified FTC nicotine yield such as at least 0.5 mg (31); or a specific cutoff for CO of at least 7 to 15 ppm or higher (28, 29, 34-36, 45). Subjects were excluded in some studies if they have had previous experience with the product being tested (28, 29, 34, 35), were pregnant or breast-feeding (28, 29, 34-36, 45), engaged in current attempts at smoking cessation or reduction (28, 29, 35, 38, 43), or had intentions to quit in the next 6 months (31). Other criteria required for participation included being in good mental health or no chronic mental condition requiring medication and with no active drug abuse (36, 39) or excessive alcohol use (43). Whereas the rationale might be apparent in some cases, some of the above inclusion criteria seemed arbitrary and may affect research results; however, this has not been studied.

The subject numbers in these in-laboratory clinical studies also tended to be small, typically under 20, with a range of up to 32 subjects (35) and 50 subjects (43). Subjects in the studies conducted in Eissenberg's laboratory tended to be younger (age ranges from 22 to 33 years), smoked fewer cigarettes (range of 15-22 cigarettes per day), and showed less dependence on the Fagerström test for nicotine dependence (FTND)/Fagerström tolerance questionnaire (FTQ; scores of 4.0-5.6) than the other studies. Similarly, O'Connor et al. (38) recruited only college students with a low rate of smoking (11.5 cigarettes per day). With the exception of the study conducted with adolescents (46), the other studies tended to have smokers over the age of 30 years (ranging from 34 to 45 years) who smoked more than 20 cigarettes per day (21-31 cigarettes per day), and had higher FTND scores (5.5-8.4). Likewise, the Eissenberg research group enrolled smokers who smoked fewer number of years (4-7 years; refs. 29, 35, 45), whereas in other studies, smokers smoked for at least 18 years (30, 31, 33, 42, 43). Most of the population was White, although a few studies had 50% or more as minorities (31, 37). Most studies were evenly split between males and females, with some studies having all males (30, 33). Some of the studies reported FTC determined nicotine yield of the cigarettes (30, 31, 37, 39, 40, 43) and body mass index (BMI; refs. 28, 35, 43) or weight (40). How PREP use might differ and affect outcomes, including biomarkers, physiologic response, delay for quitting, etc., has not been studied for different age, gender, and racial groups.

Product Compliance. Product use was not a significant issue because most of these studies were conducted in the laboratory.

Summary. Abuse liability and in-laboratory clinical studies can be valuable in providing information on nicotine delivery of a product, acute toxicant exposure using biomarkers with very short-half lives, acute physiologic and subjective responses to the product, and the potential for use or abuse of the product. The best methods to measure these outcomes and whether the responses observed in the laboratory generalize to actual product use and risk is unclear. For example, short-term in-laboratory studies differed in the method by which products were tested. Some involved standardized methods of product administration (i.e., established number of puffs) whereas other

studies allowed *ad libitum* use. Whereas the use of controlled smoking conditions might be important to determine how products compare against each other in exposure and subjective responses to the product, the extent to which these values are similar to those observed when smokers are allowed to use the product *ad libitum*, or how either of these methods reflects how the product will be used or the extent of exposure in the real world, is unknown. Furthermore, many studies involved first-time exposure to the product in the laboratory without time for adaptation. Whether or not responses to a product change when the subject is allowed to adapt to the product is also unknown. Additionally, the number and the duration of product use that is required for adaptation are unclear and may be dependent on the product and on the individual.

Clinical Trials Workshop Recommendations. For the assessment of the abuse liability of PREPs, the Clinical Trials Workshop participants emphasized the importance of examining the weight of evidence based on the results from multiple studies. Furthermore, in interpreting the data from these multiple studies, the emphasis to place on each component of a comprehensive battery to assess abuse potential (e.g., reinforcing effects versus withdrawal relief versus dependence) must be carefully considered. Workshop participants also raised the issue of comparison products and suggested that a subject's own or preferred brand should be used to anchor the high end of the abuse potential continuum and the nicotine replacement product to anchor the low end.

The critical questions that need to be addressed for these types of studies include the following: (*a*) how valid are these methods in predicting abuse liability and adverse effect of tobacco products; (*b*) what types of studies are required to determine weight of evidence and how much valence would be assigned to each type study; (*c*) how do responses to a tobacco product differ when an individual has had some exposure to the product compared with no exposure before the laboratory session; (*d*) how do responses differ when subjects are asked to use a product *ad libitum* compared with when they are asked to use the product in a prescribed manner?

Short-Term Clinical Trials

Study Design. Short-term clinical trials, that is, product use of less than 2 weeks where subjects use the product throughout the day, have been conducted in the natural environment and in the residential unit (see Table 5). Most of the studies that have been conducted on PREPs have focused on examining the toxicant exposure and biological effect of the product when compared with usual brand cigarettes and in some cases, to medicinal nicotine products or cessation, reference cigarettes or to marketed "low-tar" cigarettes.

Most of the short-term studies conducted in the natural environment have used crossover designs such as (*a*) assessments during use of usual brand cigarettes for 1 week, use of PREP for 1 week and then use of usual brand cigarettes for 1 week (47, 48); (*b*) own brand for 5 days, 1 or 2 PREP(s) for 5 days each, no smoking for 5 days, with laboratory sessions on the first and last days of each condition (44, 49, 50); and (*c*) usual brand for 1 day, another PREP for 1 day, and abstinent for 1 day, then a reversal in order (51). In a study examining the effects of progressive reduction of nicotine content of cigarettes, participants were required to smoke each of the different yields of cigarettes for 1 week, after which time subjects could quit or return to smoking (52). Other natural environment studies have used a between-subject design where subjects are typically assessed while smoking usual brand cigarettes and then are randomly assigned to intervention conditions or continued use of usual brand for 1 week or less (53-55).

Some short-term clinical trials have controlled subjects' diets, where food is provided in a cafeteria during the day or given as take-home for weekends (56). This procedure aids in reducing dietary confounders for biomarker analysis. Compliance to diet, however, is difficult to verify.

Residential studies have also been conducted. The advantage of a residential setting is the strict control over diet and assurance of compliance with the use of the assigned products. One residential study involved a forced-switching, parallel-group design where subjects were randomized to one of five treatment conditions for a period of 8 days after a 2-day acclimation phase: own brand (Marlboro Light), one of two EHCSS, Marlboro ultralight, or no smoking condition (57, 58). A similarly designed residential study randomized subjects to one of five conditions except that instead of the two different EHCSS, this study examined EHCSS under controlled versus uncontrolled smoking conditions (59). Another inpatient study compared three conditions over a 10-day period of time: no smoking, denicotinized cigarettes, and nicotinized cigarettes (60). Sarkar and colleagues (61) conducted two inpatient studies that involved randomized, controlled, open-label, parallel-group, switching design. Following an acclimation and baseline day where the subjects smoked conventional cigarettes, they were randomized to continue smoking conventional cigarettes (6-mg tar for one study and 11-mg tar for the other study) or test cigarettes (containing carbon filters) or stopping smoking for 8 days in a confined clinic setting. Cigarettes were smoked in a controlled fashion. Subjects in the conventional or test cigarette groups then had the option to participate in a long-term study with these products.

As described above, PREPs have been compared with own brand cigarettes (47-49, 52, 55, 57-59, 62), nicotinized cigarettes that were not the subject's own brand (60), no smoking (49, 51, 53, 57-60), another PREP (50), ultralight cigarettes (57-59), or, reduced smoking (53). Most studies allowed *ad libitum* smoking of the product although some studies required smoking a specified number of cigarettes by either maintaining baseline frequency of cigarettes smoked per day (53), smoking no more than baseline frequency of cigarettes or, smoking no more than 20% above baseline smoking (57-59) and at predetermined smoking times (58, 59).

Measures. The measures in short-term continuous-use studies vary and have included (*a*) amount of product use and pattern of product use; (*b*) smoking topography (e.g., determined in the laboratory or with a portable device, measuring number of puffs per cigarette, puff volume, puff duration, and interpuff interval); (*c*) nicotine and its metabolites, alveolar carbon monoxide and carboxyhemoglobin; (*d*) nicotine and carboxyhemoglobin boost; (*e*) carcinogen biomarkers of exposure, for example,

Table 5. Short-term clinical trials

Reference	Goals	Product	Product instructions	Study methods	Measures	Subjects
Baldinger (51)	To examine the responses to low-nicotine cigarette	Usual brand and low-nicotine research cigarette	*Ad libitum*	Crossover design, outpatient study with 2 experimental phases; usual brand one day, low-nicotine cigarette for one day and abstinence for one day; then order was reversed; recruitment through newspaper advertisements; inclusion criteria: smoke at least 15 CPD, nicotine yield of at least 0.7 mg	Saliva cotinine; exhaled CO, heart rate, ECG; physical activity; Stroop task performance, cigarette ratings and withdrawal symptoms, CPD	*N* = 12; mean age, 30 y; all females; mean CPD 21.2
Benowitz (47)	To assess the effects of switching from regular to light cigarettes	Usual brand and a commercial light cigarette that has a nicotine yield 50% of usual brand	*Ad libitum*	3-wk crossover outpatient study; subjects smoked their usual cigarettes during the first and 3rd weeks, and a light test cigarette during the 2nd week; recruitment through newspaper advertising	Plasma nicotine, plasma cotinine, blood carboxyhemoglobin, carboxyhemoglobin boost, nicotine boost; NNAL; 1-HOP, 1- and 2-naphthols, hydroxyphenanthrenes, hydroxyfluorenes; Minnesota Nicotine Withdrawal Scale, Profile of Mood Scale, CESD scale, cigarette acceptability, CPD	*N* = 16; 9 females, 7 males; mean age, 36 y; mean CPD, 19; mean FTND score, 6.0
Benowitz (52)	To examine the relationship of gradual reduction in nicotine and exposure to tobacco smoke toxins and assessed biomarkers	Reduced nicotine content research cigarettes provided by cigarette manufacturer; the target nicotine content per cigarette was 12, 8, 4, 2 and 1 mg	*Ad libitum*	Ten-week, unblinded, outpatient study; after 1-wk baseline, cigarettes smoked progressively decreased in nicotine content each week; then for the next 4 wk, subjects smoked the cigarettes of their choice or quit; recruitment through advertising; inclusion criteria: healthy, no plans to quit within 6 mo	Plasma cotinine, plasma nicotine, hemoglobin, high-density lipoprotein cholesterol, WBC count, C-reactive protein, fibrinogen, interleukin-6, sICAM, and P-selectin, urinary NNAL, metabolites of polycyclic aromatic hydrocarbons; body weight, blood pressure, heart rate; smoking behavior, Profile of Mood Scale, Minnesota Nicotine Withdrawal Scale, CESD scale, FTND, a self-efficacy for resisting smoking questionnaire and a cigarette acceptance questionnaire, CPD	*N* = 20; mean age, 29 y; mean CPD, 20.1; mean FTND score, 4.3

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Table 5. Short-term clinical trials (Cont'd)

Reference	Goals	Product	Product instructions	Study methods	Measures	Subjects
Bowman (48)	To determine the effect of switching to Eclipse cigarettes on urine mutagenicity	Usual brand, Eclipse cigarettes	*Ad libitum*	Three trials with smokers of ultra-low-tar, full-flavor low-tar, and full-flavor tar cigarettes and nonsmokers (controls) who ate a low-mutagen diet; subjects smoked their usual brand for at least 1 wk, switched to Eclipse cigarettes for 1 wk and then went back to their usual brand for at least 1 wk; subjects served as their own controls; inclusion criteria: smoked at least 20 CPD (or nonsmoker); exclusion criteria: smokers of Eclipse, occasional smokers, use of other tobacco products	Urine concentrates were tested for mutagenic activity; salivary cotinine; daily questions about exposure to occupational or environmental substances, use of medications, vitamins or antioxidants, exercise levels, dietary compliance, CPD	*N* = 67 smokers (11 ultra-low-tar, 41 full-flavor low-tar, 15 full-flavor tar cigarettes) and 31 nonsmokers; 27 males, 40 females (smokers); 15 males, 16 females (nonsmokers)
Breland (49)	To develop a method for evaluating the carcinogen delivery of PREPs such as when using a product like Advance	Usual brand or Advance	*Ad libitum*	Outpatient crossover design, Latin-square ordered, 3 conditions; subjects smoked own brand, Advance, or no cigarettes for 5 d; additionally, on days 1 and 5 of each condition, subjects smoked one cigarette in the laboratory; inclusion criteria: 18-50 y old, exhaled CO ≥15, ≥15 CPD (light or ultralight cigarettes); exclusion criteria: past or current cardiovascular disorders and current pregnancy, breast-feeding, smoking cessation or reduction efforts	Smoking topography; urine cotinine and NNAL; heart rate, skin temperature, exhaled CO; tobacco/nicotine withdrawal measure, Questionnaire of Smoking Urges	*N* = 12; 8 females, 4 males; mean age, 24 y; mean CO, 25.3 ppm; mean CPD, 18.8; mean FTND score, 4.9
Breland (50)	To develop clinical laboratory methods to examine the effects of PREPs	Advance, Eclipse, and own brand	*Ad libitum*	Outpatient, crossover design, Latin-square ordered, 4 conditions (Advance, Eclipse, own brand, or no smoking) for 5 d; laboratory measures on days 1 and 5; inclusion criteria: healthy, 18-50 y old, exhaled CO ≥15 ppm, smoking ≥15 CPD (light or ultralight, nonmentholated, king size) for ≥1 y; exclusion criteria: history of chronic health problems, current pregnancy or breast-feeding, active menopause, at least 15 d of past-month marijuana use, or self-reported previous Advance or Eclipse experience (i.e., >1 pack); menstruating women participated on days 2-16 of their cycles	Smoking behavior (puff number, volume, duration, and interpuff interval); cotinine, NNAL and its glucuronide conjugate, NNAL-glucuronide (total NNAL), 1-HOP; exhaled CO, blood pressure, heart rate; Smoking Withdrawal Questionnaire, Questionnaire of Smoking Urges, The Direct Effects of Smoking Questionnaire consisting of 15 VAS items from studies of cigarette effects; butts were collected and counted on days 2-5	*N* = 35; 27 males, 8 females; mean age, 22.4 y; 2 non-White; mean CPD, 21.0

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Table 5. Short-term clinical trials (Cont'd)

Reference	Goals	Product	Product instructions	Study methods	Measures	Subjects
Donny (60)	To examine the effects of smoking denicotinized cigarettes over an extended period	Nicotinized and denicotinized cigarettes	*Ad libitum* and controlled conditions	13-d inpatient study, between-subject, double-blind design; subjects were randomly assigned to smoke denicotinized cigarettes, nicotinized cigarettes or no smoking; recruitment through advertising; inclusion criteria: age 18-65, ≥10 CPD, no intention to quit in the next 3 mo, inhale while smoking, drug-free urine at screening; exclusion: significant medical illness, major psychiatric illness, pregnancy, breast-feeding, current drug abuse treatment, drug dependence, and diagnosed sleep disorder	Smoking topography; heart rate, blood pressure, exhaled CO; weight of unused cigarettes; perception and sensory, Shiffman-Jarvik Withdrawal Scale, Questionnaire on Smoking Urges, Schuh-Stitzer VAS Craving Scale, Profile of Mood States, Positive and Negative Affect Scale, St. Mary's Sleep Questionnaire, smoking behavior, Cigarette Effects Questionnaire	*N* = 30 (10 per group); 53% female; mean age, 37.8 y; mean CPD, 21.4; 72% African American; mean FTND score, 5.3
Feng (57)	To evaluate biomarkers after switching to PREPs, conventional low-yield cigarettes, or stopping smoking	Marlboro lights and ultralights and two types of EHCSS	Controlled for CPD	10-d inpatient, open-label, randomized, forced-switching, parallel-group study in which adult smokers of a conventional cigarette brand were randomly assigned to one of five study groups—2 types of Marlboros, 2 types of EHCSS, or no smoking; subjects were limited to their maximum daily CPD; inclusion criterion: smoked Marlboro lights for at least 1 y before the start of the study	1-HOP in urine, *S*-PMA in urine, *trans,trans*-muconic acid in urine, 3-methyladenine and 3-ethyladenine in urine, 8-hydroxy-2′-deoxyguanosine in urine, thioethers in urine	*N* = 110 (20 in each product condition, 30 in no smoking condition; 55 males, 55 females); mean age, 31.4 y; mean CPD, 16.0; 99 Caucasian, 11 Non-Caucasian; mean BMI, 24.5

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Table 5. Short-term clinical trials (Cont'd)

Reference	Goals	Product	Product instructions	Study methods	Measures	Subjects
Gray (44)	To determine biomarkers and the effects of using PREPs in smokeless tobacco users.	Own brand, Bacc-off, Stonewall, General snus, and no smokeless tobacco	*Ad libitum*	Study 1: 4 Latin-square ordered, 4-h conditions; each condition was separated by at least 48 h; subject used smokeless tobacco products (own brand, Bacc-off, Stonewall or General snus) *ad libitum* for 30 min with 30 min between uses; Study 2: 4 Latin-square ordered, 5-d conditions; subjects used smokeless tobacco products (own brand, Stonewall, General snus, or No ST); each condition was separated by at least 72 h; on days 1-5, participants attended a 30-min laboratory session; recruitment through advertising and word of mouth; inclusion criteria: 18-50 y old, generally healthy by self-report, reported using five or fewer smoked tobacco products in the last 6 mo, and reported current use of smokeless tobacco on a daily basis for the last 12 mo; exclusion criteria: history of chronic health or psychiatric conditions, history of or active cardiovascular disease, current pregnancy, current breast-feeding, low or high blood pressure, seizures, or regular use of prescription medication (other than vitamins or birth control)	Study 1: Plasma nicotine; heart rate, blood pressure, expired air CO; measures of withdrawal and Direct Effects scale; Questionnaire of Smoking Urges; Study 2: NNK, total NNAL; urinary cotinine; heart rate, blood pressure, expired air CO; the same subjective measures as in Study 1	Study 1: N = 13; 12 males, 1 female; all White; mean age, 29.2 y; mean uses per day of smokeless tobacco, 4.6; Study 2: N = 19; all males; 1 non-White; mean age, 24 y; mean uses per day of smokeless tobacco, 5.2
Hammond (55)	To assess smoking topography outside a laboratory setting and switching from regular to low-yield cigarette brands	Usual brand and Matinee Extra Mild	*Ad libitum*	Within and between-subject design consisting of three 1-wk trials (first 2 trials were usual brand, third trial subjects were randomly assigned to lower-yield cigarette or usual brand), using smoking topography device at home; participants were recruited through a random-digit dial telephone survey; inclusion criteria: ≥5 CPD, no intention to quit smoking in the next 3 mo, and smoked a brand with ISO tar yields between 10 and 14 mg	Smoking topography; saliva cotinine, brand elasticity (examines the increase in nicotine delivery compared with increases in puff volume); daily diary for smoking	N = 59; 30 males, 29 females; mean age, 37.1 y; mean CPD, 19.3

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Table 5. Short-term clinical trials (Cont'd)

Reference	Goals	Product	Product instructions	Study methods	Measures	Subjects
Hatsukami (53)	To compare acute tobacco withdrawal symptoms between total cigarette cessation, smoking reduction, and nicotine yield reduction	Carlton 100's	Controlled for CPD	Smokers were asked to smoke *ad libitum* for 3 d, and for the next 5 d, they were randomly assigned to no smoking, 50% reduction of number of cigarettes, or reduction of nicotine yield of cigarettes; recruitment through newspaper advertisements; inclusion criteria: >18 y old, ≥20 CPD, nicotine yields >0.5 mg, must have made a previous attempt to quit smoking and experienced at least one episode of tobacco withdrawal symptoms; exclusion criteria: history of alcohol or drug abuse, emotional or physical health problems, taking medications, or obtaining nicotine in forms other than smoking cigarettes	Heart rate, body weight; Profile of Mood States, Stanford Sleep Scale and a 100-mm visual analogue craving line for tobacco, Minnesota Nicotine Withdrawal Scale and observer withdrawal ratings	$N = 32$; $N = 11$ (total cessation), $N = 11$ (50% reduction), $N = 10$ (reduction of nicotine yield in cigarettes); 17 males and 15 females; mean age, 26.1 y old; mean CPD, 26.1
Jacober (54)	To investigate the effects of switching to ultralight cigarettes in and out of the laboratory	Usual brand and commercial ultralights	*Ad libitum*	Subjects completed 4 (2 × 2) measurement days and came to the laboratory three times (on the day before the first measurement day and the days after the 2nd and 4th measurement days); during 2 d, subjects smoked usual brand, for the other 2 d smoked 1 brand of the 6 commercially available ultralights; the order of smoking was balanced; recruitment through newspaper advertisements; inclusion criteria: ≥15 CPD with a minimal nicotine delivery of 0.7 mg	Urine cotinine; heart rate, blood pressure, exhaled CO; heart rate, blood pressure at home; diet; subjective well-being parameters	$N = 48$ mean age, 28.3 y; mean CPD, 27.7
Roethig (58)	To evaluate first-generation EHCSS compared with low-tar conventional cigarettes, usual brand, and no smoking	Accord, Oasis, Marlboro Lights (usual brand), and Marlboro Ultra	Controlled	10-d inpatient, open-label, randomized, forced-switching, controlled, parallel-group design study; subjects were stratified for gender and cigarette consumption and randomized to one of five groups—Accord, Oasis, Marlboro Lights (usual brand), Marlboro Ultra, or no smoking and used the product for 8 d; subjects were permitted to smoke between 7 a.m. and 11 p.m. at set times (i.e., every 32 min); subjects were not forced to smoke and could reduce or quit smoking, but were expected to smoke their daily allotment throughout the day; inclusion criterion: smoke between 5-25 Marlboro Lights daily; exclusion criteria: use of oral antidiabetic medication, insulin therapy, and bronchodilator medications	Urine nicotine and five of its metabolites (nicotine-*N*-glucuronide, cotinine, cotinine-*N*-glucuronide, *trans*-3′-hydroxycotinine, and *trans*-3′-hydroxycotinine-*O*-glucuronide), urine mutagenicity; carboxyhemoglobin; exhaled CO; environmental tobacco smoke; FTND, product assessment; cigarette butts collected	$N = 110$ (20 in each product group, 30 in no-smoking group); 55 male and 55 female; mean age, 31 y; mean CPD, 16.3; mean FTND score, 3.5; 11 non-Caucasian, 99 Caucasian

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Table 5. Short-term clinical trials (Cont'd)

Reference	Goals	Product	Product instructions	Study methods	Measures	Subjects
Roethig (59)	To examine the effect of Accord compared with very-low-tar conventional cigarettes or to no smoking	Accord, Marlboro lights, and Merit Ultima	Controlled or uncontrolled	10-d inpatient, randomized, controlled, forced-switching, open-label, parallel-group study in smokers of conventional cigarettes randomized to one of five conditions (Accord controlled smoking, Accord uncontrolled smoking, Marlboro Lights, Merit Ultima, or no smoking) and used the product for 8 d; in the controlled condition, subjects were permitted to smoke between 7 a.m. and 11 p.m. at set times (i.e., every 32 min) with a maximum number of cigarettes per day equal to the number smoked on the acclimation day; in the uncontrolled condition, subjects could smoke anytime between 7 a.m. and 11 p.m. with a maximum of 60 cigarettes per day; recruitment through advertisements; inclusion criteria: 21-65 y old, good general health, smoked 10-30 conventional cigarettes with 7-12 mg of tar delivery daily for at least the 12 mo preceding the study, smoked Marlboro Lights for at least the 4 wk preceding the study, no use of tobacco or nicotine-containing products other than manufactured cigarettes for at least the 3 mo before the start of the study; exclusion criteria: clinically significant renal, liver, metabolic, cardiac, and pulmonary disease and illicit drug use, pregnancy, breast-feeding or intention to become pregnant during the study, use of antidiabetic and bronchodilator medications	Smoking topography; urine nicotine, plasma cotinine, 1-HOP, NNAL, urine mutagenicity and 3-HPMA and *S*-PMA	*N* = 100 (20 in each group); 50 males and 50 female; mean age, 34 y; 3 non-White, 97 White; mean BMI, 24.6
Smith (62)	To determine the effects of Eclipse compared with usual brand	Eclipse prototype and usual brand	*Ad libitum*	4-wk, crossover study, with each smoker consuming test cigarettes *ad libitum* for a week and their usual brand of tobacco-burning cigarettes for the other 3 wk; a control group of nonsmokers was included; inclusion criteria: healthy based on a physician's review of a medical questionnaire	Salivary cotinine levels, urine nicotine, urine cotinine, urine mutagenicity; cigarette consumption	*N* = 34 (20 smokers and 14 nonsmokers); all males

NOTE: Majority of trials assess for amount of product(s) used.
Abbreviations: 1-HOP, 1-hydroxypyrene; 3-HPMA, 3-hydroxypropylmercapturic acid; CESD, Center for Epidemiologic Studies-Depression; sICAM, soluble intercellular adhesion molecule; *S*-PMA, *S*-phenylmercapturic acid.

NNAL and its glucuronides (total NNAL) for NNK exposure, 1-hydroxypyrene for pyrene exposure, 3-hydroxypropylmercapturic acid for acrolein exposure, *S*-phenylmercapturic acid for benzene exposure, monohydroxybutenyl mercapturic acid for 1,3-butadiene exposure, *trans,trans*-muconic acid, 3-methyladenine, 3-ethyladenine, 8-hydroxy-2′-deoxyguanosine, thioethers, and urine mutagenicity; (*f*) biomarkers of effect (inflammatory response, endothelial function, platelet activation, C-reactive protein, fibrinogen, interleukin-8, soluble intercellular adhesion molecule, and P-selectin); (*g*) weight, skin temperature, and vitals (blood pressure and heart rate); (*h*) physical activity or diet; (*i*) subjective responses such as withdrawal symptoms and craving, desire to smoke; (*j*) product evaluation or product acceptance (e.g., strength/mildness of product, smoothness/harshness, quality of flavor, overall cigarette quality, and satisfaction) or comparison of nicotine yield of study tobacco products to conventional products on the market; (*k*) moods [e.g., Profile of Mood States (63), Positive and Negative Affect Scale (64), depression (Center for Epidemiologic Studies-Depression; ref. 65), subjective well being]; (*l*) self-efficacy for resisting smoking usual brand cigarettes in high-risk situations; (*m*) dependence (e.g., FTND; ref. 66); (*n*) performance tasks (Stroops task; ref. 67); and (*o*) sleep quality. One study described collecting questions on exposure to occupational and environmental substances, medications, vitamins, or antioxidants and exercise levels to assess for factors that might affect outcome measures (48).

Recruitment Method. Subjects were recruited by advertisements (47, 51-54, 60, 61), random-dial telephone survey (55), or the method of recruitment was not reported (48, 49, 57, 58). Many of the studies, including the studies that did not report recruitment methods, did not describe the content of the advertisements (47, 51, 52, 61).

Subject Characteristics. Most studies that described inclusion and exclusion criteria indicated subjects needed to be in good mental and physical health and not pregnant or breast-feeding. Other inclusion criteria included limitations on the amount smoked (e.g., ranging from at least 5 to 25 cigarettes per day or no more than 30 cigarettes per day; refs. 48, 51, 54, 55, 58-61), specific CO levels (≥15 ppm; refs. 49, 50), specific nicotine or tar yields of cigarettes or type of cigarette smoked such as light or ultralights (e.g., refs. 49-51, 53-55, 58, 59, 61), and not planning on quitting (49, 52, 55, 60). Other studies excluded smokers who are using or have used the product being tested (48), using other tobacco products (50), or using other nicotine-containing products (53). Some studies excluded subjects who were currently taking medications (53), who reported at least 15 days of past-month marijuana use (50), or were drug dependent (60). Another study excluded women who were in active menopause (50). Other studies did not report inclusion criteria (47, 57).

Study sample sizes ranged from 12 to 110 with generally 10 to 20 subjects in each condition, with higher sample sizes in tobacco industry conducted studies (57-59). Most of the subjects who participated in the study tended to be young (mean age ranged from 24 to 38 years). The samples were typically evenly divided between males and females, although some studies were either all females (51) or were predominantly female (49) or male (50). The range of mean cigarette intake was from 16 to 28 cigarettes per day. Some studies indicated the nicotine yield of cigarettes (47, 50, 51) or the FTND scores (47, 49, 60) or intentions to quit (55).

Compliance with Product Use. Compliance to product use was maximized in the following ways: providing free products (all cited studies); payment contingent on compliance (although no biochemical verification was obtained for product use; refs. 51, 53); payment contingent on verification of abstinence conditions via biochemical verification (49, 50); or use of a bogus pipeline (60). Other studies did not address the issues of compliance (48, 52, 55, 62). Some studies were conducted in a residential unit where use of the products was monitored (57-59).

Typically, subjects are requested to not use other tobacco products. However, for PREPs, which are tobacco products, this cannot be verified. For persons who switch to NRT, this can be assessed by measuring urinary anatabine, which is a tobacco alkaloid (68), or total NNAL (69). For persons who report cessation, urinary or serum cotinine levels can be measured.

Intermediate-Term Clinical Trials

Study Design. Intermediate-term trials are defined herein as being conducted for longer than 2 weeks but no longer than around 52 weeks in the natural environment (see Table 6). Product is used throughout the day, and in most studies, product use is *ad libitum*. These studies used a between-subject design comparing different products (56, 61, 70-78), a within-subject design taking assessments during usual brand cigarette smoking and after switching to a product (79-81), or a within-subject crossover design with different products (23, 82-85). Subjects were required to use a specific product (61, 70, 72-83, 85-89) or were given a choice after sampling the products (56, 71). Some of these studies included additional or different experimental design features. The study conducted by Mendoza-Baumgart et al. (23) required use of one of the two assigned products for 2 weeks and then a crossover to the other product, during which time biomarkers for exposure were assessed, and a choice of products for the final week, where no biomarker assessments were made. Another study involved smoking the product for 2 weeks and then a test session after this 2-week period as well as during usual brand use (80). One study provided smokers of "medium tar yield" cigarette with commonly smoked medium tar yield brand of cigarettes in unmarked boxes. Subjects were then unknowingly switched to low-tar cigarettes or continued on "medium-tar cigarettes" (84), therefore blinding the subject to the switch in cigarettes. One study allowed a 2-week acclimatization time with higher nicotine yield cigarettes compared with usual brand cigarettes and then made biomarker assessment after another 2 weeks of use. The values for thiocyanate and carbon monoxide were higher after the 2-week acclimatization period, indicating a period of time to stabilize to product use may be warranted for some products (70).

Unique designs have been used in studies conducted by a tobacco company. One study involved having all subjects switch to the reference conventional cigarette, then random assignment to continued use of the reference cigarette or switching to an EHCSS. Subjects were assessed in a residential clinic for 36 hours during baseline

smoking and at the end of the first week of using the randomly assigned product (EHCSS versus reference cigarette), and then during continued use of the assigned product over the course of 12 weeks (74). In another study, as previously described, subjects underwent a short-term residential phase (similar to ref. 55), and then continued with the assigned product for 24 weeks in the natural environment (73). In one longer-term study, all subjects underwent a 2-week trial period of a PREP (EHCSS) before enrolling in a 12-month-long study (75). Baseline and visits at 2 weeks and monthly thereafter were conducted in a controlled, confined clinic setting from 07:00 to 07:00 the next day.

Published intermediate-term studies have control arms such as medicinal nicotine (23, 56, 72, 82, 83), nonsmokers (86), smokers using their usual brand of cigarettes (79-81, 87), or a control group with conventional cigarettes similar to usual brand (61, 70, 74-76, 78, 84, 89). Some of these studies examined the effects of cigarettes with different nicotine or tar yields (70, 71, 73, 76-78, 84, 85, 87-89) or different amounts of product use (83).

Three intermediate-term studies allowed concurrent use of their usual brand cigarettes with the PREP (56, 82, 83). In the studies conducted by Fagerström et al. (56, 82), subjects were instructed to smoke as few cigarettes of their own brand as possible without discomfort and instead use as much of the treatment product (nicotine inhaler or Eclipse) as needed. In the study conducted by Hughes and Keely (83), subjects were asked to use a specified number of Accord cigarettes per day (5, 10, or 15). Other studies allowed *ad libitum* product use and, typically, no other nicotine-containing products (73-75, 79, 81, 86). Another study allowed *ad libitum* product use and excluded subjects if more than 5% of their total daily cigarettes smoked were nonstudy cigarettes (61). Hatsukami et al. (72) and Mendoza-Baumgart et al. (23) required a specified amount of use (the same amount as usual brand or use every 2 hours, respectively).

Measures. The measures across the published intermediate-term studies have included (*a*) amount of product used and in some studies, when the products were used; (*b*) extent of compensatory smoking (as measured by smoking topography, cotinine, and/or CO); (*c*) the number of usual brand cigarettes per day and use of other tobacco products; (*d*) biomarkers of exposure such as carbon monoxide, total nicotine equivalents or cotinine, thiocyanate, total NNAL, 1-hydroxypyrene, 3-hydroxypropylmercapturic acid, monohydroxybutenyl mercapturic acid, *S*-phenylmercapturic acid, and 4-aminobiphenyl hemoglobin adducts reflecting exposure to aromatic amines; (*e*) biomarkers of effect such as pulmonary function tests, measures of lower respiratory tract or airway inflammation, goblet cell metaplasia, peripheral blood measures, ^{99m}technicium-diethylenetriaminepentaacetic acid clearance, blood leukocyte activation, reactive oxygen species, WBC count and hemoglobin levels, and respiratory symptoms; (*f*) cardiovascular risk factors such as hemoglobin, hematocrit, RBC, WBC count, fibrinogen, lipoproteins, triglycerides, high-sensitivity C-reactive protein, bilirubin, von Willebrand factor, 11-dehydrothromboxane B_2, 8-*epi*-prostaglandin $F_{2\alpha}$, and microalbumin; (*g*) weight and vitals (blood pressure and heart rate); (*h*) subjective measure of withdrawal and craving; (*i*) drug effects and liking, sensory ratings, and product evaluation (odor, strength, draw resistance, taste, embarrassment regarding use, and liking); and (*j*) intention or motivation to quit.

Recruitment Methods. The advertisements used to recruit subjects varied in content. Some advertisements described the study as one that was testing new products that may reduce the risk of smoking or may be safer (56, 81) and/or with no second-hand smoke exposure (56, 83). Other advertisements called for smokers or tobacco users who were interested in participating in studies that compared new tobacco products with nicotine replacements (72). One study sent potential subjects a questionnaire asking for details of their smoking habits to determine their eligibility and willingness to participate in a trial requiring subjects to switch to low-tar cigarettes (71). In another study, subjects were recruited among a group of "acceptors" of a new cigarette (Eclipse) that was in test marketing (86). These acceptors had smoked at least 75% of two cartons of Eclipse cigarettes and expressed future purchase intent. Most of these studies seemed to recruit subjects that were interested in trying a new product.

Subject Characteristics. The inclusion criteria in intermediate-term studies generally specified that subjects had to be in good current physical and/or good mental health with no clinically significant diseases (56, 72-75, 79, 82, 86, 89), with some studies reporting a specified level of lung function (79) or no history of any respiratory and/or cardiovascular disease (76, 86, 89) or diabetes mellitus and hypertension (89); no regular medications (76); a specified age range (e.g., ages 18, 20, 21, or 25 to 50 or 65 years old; refs. 23, 56, 61, 72, 74, 75, 82, 86) or at least 18 years old with no upper limit on age (76, 81, 83); a minimum amount of cigarettes smoked per day, ranging from at least 5 cigarettes to 20 per day (56, 61, 72-74, 76, 81-83, 85) or even as high as 40 cigarettes per day (79); smoking a specified type of cigarettes (e.g., full flavor, light, or ultralight) or a tar range for cigarettes (e.g., light or ultralight; refs. 73, 74, 83) or a minimum tar yield (77, 86, 87) or minimum nicotine yield; not using smoking cessation or reduction methods (72) or intending to quit (79) or to reduce cigarette use (89); wanting to switch to low-tar/nicotine cigarettes and perhaps quit (76); and no experience with a PREP that is similar to the study product (83) or use of any other non-cigarette or nicotine product (72, 83, 86) or any product other than the reference conventional cigarette, which was smoked 4 weeks preceding the start of the study (74). Most studies reported eliminating pregnant or breast-feeding women.

Some studies had small sample sizes per product condition (N = 8-15, e.g., refs. 56, 79, 81, 86, 88, 89) or moderate sample sizes per condition (e.g., N = 25-75; refs. 72-74, 80, 87). One study had about 145 subjects per product condition (71). Not all studies reported the subject characteristics of the sample that was recruited. In general, the study groups tended to be older in age (mean age of 35 to 48 years; refs. 23, 73-75, 81-83); more than 50% of the population was female (23, 74, 75, 81-83, 87) with the exception of a few studies where subjects were predominantly male (73, 76, 77, 85, 86); predominantly White (23, 61, 72-75, 81); and tended to be heavy smokers (mean of 20 to 29 cigarettes per day; refs. 23,

Table 6. Intermediate-term clinical trials

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Armitage (85)	To determine the effects of switching to low-tar/nicotine similar to usual brand or to conventional low-tar/low-nicotine cigarettes	Lower-tar cigarette with amounts of nicotine similar to a conventional middle-tar cigarette (maintained nicotine), conventional low-tar/low-nicotine cigarettes and middle-tar cigarettes with amounts of nicotine similar to a conventional middle-tar cigarette	*Ad libitum*	A randomized, balanced, crossover design (double 3 × 3 Latin square) study; subjects were randomly assigned to one of six orders of product use and used the 3 products; each product was smoked exclusively for 2 wk; a laboratory visit occurred at end of every 2 wk, during which 1 cigarette was smoked; recruitment through being chosen from a clinical studies volunteer panel; inclusion criteria: smoked at least 10 filter-tipped middle-tar cigarettes per day	Puff topography; nicotine in plasma and urine, cotinine in plasma, saliva, and urine, nicotine-l′-*N*-oxide in urine; carboxyhemoglobin; exhaled CO; derived estimates of tar intake; smoking behavior, product acceptance questionnaires	*N* = 24; all males; mean age, 26 y
Fagerström (82)	To assess Eclipse on cigarette smoking behavior and biomarkers compared with a nicotine inhaler	Eclipse and nicotine inhaler	*Ad libitum* use of Eclipse and nicotine inhaler, smoking as few cigarettes as possible	Crossover design (2 × 2) study; smokers were randomized to either Eclipse or nicotine inhaler for 2 wk and then switched to the other product for another 2 wk; smokers were instructed to smoke as few cigarettes as possible and use as much of the treatment product as needed; recruitment through advertising in newspapers that stated that they were looking at a new product to reduce the risk of smoking; inclusion criteria: 20-65 y old, in good general health, good ability to read and understand Swedish, smoke at least 5 CPD; exclusion criteria: severe or symptomatic cardiovascular disease, pregnancy, breast-feeding, regular psychotropic medication use, abuse of alcohol or any other drug, or use of smokeless tobacco or NRT	Plasma carboxyhemoglobin, nicotine and cotinine concentrations in saliva and blood; exhaled CO, heart rate; withdrawal symptoms, attitudes toward smoking, motivation to quit, FTND; amount of product use, CPD	*N* = 50; 67% females; mean age, 49.2 y; mean CPD, 20.2; mean FTND score, 5.4

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Fagerström (56)	To determine the effects of long-term use of Eclipse	Eclipse and nicotine inhaler	*Ad libitum* use of Eclipse and nicotine inhaler, smoking as few cigarettes as possible	Between-subject, open-label, product choice design; before subjects entered this study, they had participated in a 4-wk crossover trial of Eclipse and the inhaler (82); after the crossover trial was completed, there was a 2-wk washout period in which subjects returned to their regular smoking and then subjects chose Eclipse, the inhaler, or their preferred brand to use for another 8 wk; recruitment through a prior 4-wk crossover trial of Eclipse and the inhaler (82); inclusion criteria: 20-65 y old, in good general health, good ability to read and understand Swedish, smoke at least 5 CPD; exclusion criteria: severe or symptomatic cardiovascular disease, pregnancy, breast-feeding, regular psychotropic medication use, abuse of alcohol or any other drug, or use of smokeless tobacco or NRT	Carboxyhemoglobin, plasma cotinine and nicotine; exhaled CO; amount of product used, cigarettes per day, withdrawal symptoms, evaluation of product, motivation to quit, FTND, amount of product use, CPD	N = 39 (15 inhaler, 10 Eclipse, 13 regular cigarette brand, 1 noncompleter); mean FTND score, 5.6 (inhaler); 4.9 (Eclipse)
Frost (71)	To quantify compensatory smoking in a long-term randomized trial after smokers switched to low-tar cigarettes	Choice of brand within a given range of tar yield	*Ad libitum*	Randomized, between-subject, controlled, forced-switching, open-label design; subjects switched to cigarettes with a tar yield that was about 10% lower than their usual brand for 2 mo to identify compliant subjects; subjects were then followed for 6 mo, during which they were assigned randomly to switch immediately to a tar yield cigarette about half that of their usual brand, a group that was forced to reduce to the same level in steps during several months or a control group that continued to smoke a cigarette with an ~10% lower tar yield than usual brand; recruitment through 33,800 British government workers enrolled in a cohort study; inclusion criteria: smokers of relatively high tar yield cigarettes	Carboxyhemoglobin, serum cotinine levels; exhaled CO; relative intake (measures the strength of inhaling); compensation	N = 434 (144 control group, N = 145 (fast reduction group), 145 (slow reduction group)

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Frost-Pineda (74)	To assess changes in biomarkers of exposure of EHCSS compared with Marlboro Ultra Lights cigarettes	Marlboro Ultra Lights cigarettes, EHCSS (series K)	Uncontrolled (for residential component) and unrestricted during 12 wk. (for non-residential component)	Randomized, controlled, open-label, forced-switching, parallel-group design; baseline biomarker levels were measured, then participants were randomly assigned to switch to an EHCSS (series K) or to continue smoking the reference conventional cigarette of similar tar yield for 12 wk; two 36-h clinic-confined sessions held at baseline and after 1 wk; recruitment through local advertising; inclusion criteria: healthy, 21-65 y old, smoked 10-30 manufactured, nonmenthol cigarettes with 3-6 mg of tar delivery (FTC) daily for at least the preceding 12 mo and Marlboro Ultra Lights cigarettes (the reference conventional cigarette) for at least 4 wk preceding the start of the study; exclusion criteria: clinically significant renal, liver, metabolic, cardiac, and pulmonary disease, alcohol or drug abuse, use of any tobacco- or nicotine-containing products other than the reference conventional cigarette, pregnant, lactating or intended to get pregnant during the study, antidiabetic and bronchodilator medications	Smoking topography; nicotine, nicotine-*N*-glucuronide, *trans*-3′-hydroxycotinine (total), cotinine (total), NNAL and its glucuronides (total NNAL), 1-HOP and its glucuronide and sulfate (total 1-HOP), carboxyhemoglobin, 3-HPMA, *S*-PMA; exhaled CO; daily diary	*N* = 90 (60 EHCSS, 30 conventional cigarette), 31 males, 59 females; 96.7% White, 1.1% Black, 1.1% Hispanic, 1.1% mixed; mean age, 35.1 y; mean CPD, 22.7
Guyatt (87)	To determine smoking behavior after switching to cigarettes with lower tar and nicotine yields	Own brand and choice of a brand at least 3 mg less tar than own brand	*Ad libitum*	Within-subject, forced-switching, open-label design; subjects smoked their usual brand for 5 mo, then switched to cigarettes with lower tar and nicotine machine yields for 9 mo; lab sessions were held every month during which usual brand cigarettes were smoked and then every 6 wk after switching where subjects smoked 1 cigarette; recruitment through local television, radio, and newspapers, promotions in public places, a recruitment agency, and word of mouth; inclusion criterion: cigarette tar yields >10 mg	Smoking topography, FEV1 and FVC; plasma cotinine, carboxyhemoglobin; exhaled CO; butt length, draw resistance; Medical Research Council questionnaire on respiratory symptoms, questionnaire on smoking habits	*N* = 151 (28 in study group, 123 in default group, subjects who attended on at least one occasion but either did not complete the study or did not fulfill the study criteria); 64 males and 87 females; mean age, 40.9 y; mean CPD, 23.6

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Hatsukami (72)	To determine the effect of PREPs compared with NRT on biomarkers	Omni, Snus, and nicotine patch	*Ad libitum*, except maintain CPD	Randomized, controlled, open-label, between-subject, parallel-arm, forced-switching design, 4-wk trial; ST users and cigarette smokers were randomly assigned to either use test products (Swedish snus for users of smokeless tobacco or Omni cigarettes for smokers) or quit while using the nicotine patch; recruitment through advertising for subjects to compare new tobacco and nicotine replacement products; inclusion criteria: 21-65 y old, good physical and mental health, ≥15 CPD or ≥1 tin snuff/wk for a minimum of 1 y, not using other tobacco products, and not quitting	Urinary cotinine, total NNAL, 1-HOP; tobacco and nicotine patch use	*N* = 79 (41 ST, 38 smokers); all males; mean age, 31.4 y (ST), 40.9 y (smokers); mean CPD, 21.8; mean tins/wk of smokeless tobacco, 3; 39 White (ST), 36 White (smokers); secondary analysis (due to excluding noncompliant subjects): *N* = 34 (15 nicotine patch, 19 ST)
Heinonen (88)	To determine levels of thioethers in the urine of low-tar and medium-tar cigarette smokers and nonsmokers	Commercial medium-tar and low-tar cigarettes	*Ad libitum*	Crossover, forced switching design, subjects smoked low-tar cigarettes for 3 wk and switched to medium-tar cigarettes for 3 wk or in reverse order; 4 urine samples were collected; the nonsmokers' urine samples were taken at the same time periods as the smokers' urine samples; recruitment of men in military service; inclusion criteria: active smokers of medium- or low-tar cigarettes or nonsmokers	CPD; thioethers	*N* = 37 (11 nonsmokers, 13 medium tar, 13 low tar), all males; mean age, 19.0 y; CPD varied from 10 to 25
Hughes (81)	To determine the effects of switching to Omni on smoking behavior, liking of the product, and biomarkers	Omni and own brand	*Ad libitum*	12-wk randomized, crossover design; subjects smoked their own brand for 6 wk and then switched to Omni for 6 wk or in reverse order; 3 biweekly lab visits of each 6 wk period; inclusion criteria: ≥18 y old; smoke ≥10 CPD for ≥1 y; rate themselves as <7 on a 1-10 scale (1, definitely do not intend to quit in the next month; 10, definitely intend to quit in the next month); not pregnant, breast-feeding, or planning to become pregnant and have a negative pregnancy test at the onset of the study	Smoking topography; urinary cotinine plus its glucuronide (total cotinine), nicotine plus its glucuronide (total nicotine), and carcinogen metabolites 4-(methylnitrosamino)-1-(3-pyridyl)-1 butanol plus its glucuronides and 1-HOP; expired air CO; Tiffany craving scale, Minnesota Nicotine Withdrawal Scale, FTND	*N* = 34; 59% men; mean age, 48 y; minorities 3%; mean CPD, 29; mean FTND score, 6.4; 53% pre-contemplators, 47% contemplators

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Hughes (83)	To assess the effect of Accord on ongoing cigarette smoking and biomarkers	Accord and nicotine gum	*Ad libitum*	Within-subject A-B-A study of 3 wk of *ad libitum* smoking, 6 wk of using at least 5 Accords per day plus *ad libitum* smoking, and then 3 wk of *ad libitum* smoking; this was followed by a second study using at least 10 and then at least 15 Accord per day and finally at least six pieces of 4-mg nicotine gum, for 2 wk each, separated by 1 wk of baseline; recruitment through advertising for smokers who wanted to smoke a smokeless cigarette; inclusion criteria: ≥18 y old, ≥10 CPD for 1 y, smoked light or ultralight cigarettes, never smoked Accord, Eclipse, or Premier cigarettes; exclusion criteria: pregnancy, use of any medication for smoking cessation in the past month, a change of at least 25% in CPD in the past 2 mo, use of non-cigarette tobacco in the past month, asthma, and significant cardiovascular disease in the past 6 mo	Urine cotinine; exhaled CO; FTND, motivation to quit smoking, CPD, nicotine withdrawal, toxicity symptoms	*N* = 11; 6 females, 5 males; mean age, 42.5 y; mean CPD, 24.4; mean FTND score, 6.8; all pre-contemplators
Mendes (73)	To assess changes in biomarkers of exposure in smokers switching to light or ultralight cigarettes	Full-flavor Marlboro, Marlboro Lights and Marlboro Ultra Lights cigarettes	Controlled during short-term phase; unrestricted during long-term phase	8-d inpatient, randomized, controlled, forced-switching study with a 24-wk follow-up; subjects smoked full-flavor Marlboro cigarettes and were randomly assigned to smoke Marlboro Lights or Marlboro Ultra Lights cigarettes; recruitment through newspaper ads; inclusion criteria: healthy male and female adults who reported smoking between 10 and 30 full-flavor (15-mg tar) cigarettes daily; exclusion criteria: subjects with clinically significant diseases, health conditions or abnormal laboratory results, pregnant or lactating females and subjects who required antidiabetic or insulin therapy, bronchodilators, or antibiotic therapy for an acute infection	Smoking topography; biomarkers of exposure to nicotine [urinary nicotine and metabolites, 1-HOP, 3-HPMA, *S*-PMA and plasma cotinine, urinary total NNAL, exhaled CO, benzene, acrolein, and carboxyhemoglobin]; smoking history, FTND	*N* = 225 (77 full flavor, 73 light, 75 ultralight); 62 females, 163 males; mean age, 35.0 y; 5 Black, 200 White, 17 Hispanic, 3 other; mean CPD, 19.9; mean FTND score, 5.5

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Minty (84)	To determine effects on switching to low-tar cigarettes and low-CO cigarettes on lung clearance	Usual brand, commercial low-tar cigarettes, and research low-CO cigarettes	*Ad libitum*	Study 1: Between-subject design, controlled, forced-switching study; subjects all smoked usual brand cigarettes for 2 wk and were then assigned to continuing on or switch to low-tar cigarettes for 2 wk; Study 2: double-blind, crossover study; subjects smoked the mid-carbon monoxide cigarette for 3 wk and then switched to the low-carbon monoxide cigarette for 3 wk or in reverse order; subjects were not informed when the change-over occurred; inclusion criteria: smoked middle-tar cigarettes	Lung clearance rate by ^{99m}Tc-DTPA and carboxyhemoglobin; cigarette butts were measured to determine cigarette consumption	Study 1: *N* = 20; all males; mean age, 30 y; Study 2: *N* = 15; mean age, 38 y
Ossip-Klein (76)	To examine the effects of smoking cigarettes with lower tar and nicotine compared with subjects' usual brand on alveolar carbon monoxide	Own brand, 30%, 60%, 90% less tar/nicotine than own brand (in the 90% brand fade menthol users used Cambridge Regular or Carlton Menthol)	*Ad libitum*	Between-subject design; for 5 wk, subjects were assigned to either a brand-reduction treatment (brand-fading) or to a delayed-brand fading control group; recruitment through newspaper, poster, television, and radio advertisements; inclusion criteria: smoking at least 20 CPD with at least 0.8-mg nicotine for at least 2 y, no cardiovascular or pulmonary disease, no regular medications, desire to switch to low-tar/nicotine cigarettes with optional cessation	Smoking topography; alveolar CO, resting CO body burden and CO uptake per cigarette, smoking rate; CPD	*N* = 40 (brand fading, 19; delayed treatment, 21); 23 males and 17 females; mean age, 38 y; mean CPD, 37.5
Peach (77)	To evaluate the effect of a low- versus a middle-tar cigarette on respiratory symptoms	Middle-tar cigarette and low-tar cigarette	*Ad libitum*	Double-blind, randomized controlled design; smokers of middle-tar cigarettes smoked their usual brand for 1 wk; they were then randomly assigned to smoke either low-tar or middle-tar cigarettes for 5 wk; the cigarettes were sold to them at three different reduced prices; recruitment through a smoking questionnaire sent to 19,366 households; inclusion criteria: men 20-44 y old, middle-tar cigarette smokers	Nicotine metabolites; cigarette butts counted and weighed, tar yield, depth of inhalation, CPD; Medical Research Council respiratory symptoms questionnaire	*N* = 183 (95 middle tar, 88 low tar); all males

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Rennard (79)	To determine the effect on the lungs after switching to Eclipse	Eclipse, usual brand	*Ad libitum*	Controlled, within-subject, open-label design; smokers and nonsmokers had paired bronchoscopies, bronchoalveolar lavages and endobronchial biopsies at baseline and after 8 wk of smoking Eclipse in smokers; recruitment through advertising; inclusion criteria: smoking at least 40 CPD, not currently interested in quitting, free of any significant medical condition, not taking medications regularly and a normal forced expiratory volume in 1 s by spirometry; exclusion criteria: prior diagnosis of chronic bronchitis	Nicotine and cotinine levels; exhaled CO, pulmonary function testing, weight, pulse, blood pressure, bronchoscopy and bronchoalveolar lavage, peripheral blood counts, respiratory symptoms	$N = 26$ (18 smokers and 8 nonsmokers)
Robinson (70)	To test if switching to low yield cigarettes affected biomarkers	Usual brand and commercial low-tar cigarettes	*Ad libitum*	Randomized, controlled, open-label, between-subject design; during the first 2 wk, all smokers used usual brand; the control group then smoked cigarettes with similar nicotine yields of their usual brand and the treatment group switched to cigarettes with nicotine yields that were slowly lowered over 6 wk; inclusion criteria: ≥20 CPD and cigarettes with nominal nicotine delivery between 0.8 and 1.1 mg per cigarette	Urine cotinine, thiocyanate, and carboxyhemoglobin	$N = 22$ (16 treatment, 6 control)
Robinson (78)	To determine if switching to cigarettes with lower nicotine yields affects exposure to three biologically active smoke constituents	Nicotine yields within ±0.1 mg of usual brand (controls) and reduced yield brands (treatment group; first stage 33% reduction; second stage 61% reduction)	*Ad libitum*	Randomized, controlled, open-label, between-subject design; subjects were switched to lower nicotine yield cigarettes in two stages over an 8-wk period; the control group switched to cigarettes with nicotine yields similar to their usual brand; inclusion criteria: smoked daily more than 20 high-nicotine (0.8-1.1 mg) Canadian cigarettes	Urine cotinine, plasma thiocyanate and carboxyhemoglobin, brand satisfaction	$N = 22$ (16 treatment, 6 control)

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Roethig (75)	To assess changes in biomarkers and cardiovascular risk factors after switching to a second-generation EHCSS	EHCSS and conventional cigarettes	*Ad libitum*	Randomized, controlled, forced-switching, open-label, parallel design; subjects were switched to EHCSS or continued smoking conventional cigarettes for 1 y; 24 hr clinic-confined laboratory visits were made throughout the study duration; recruitment through local ads by 2 clinical study centers; inclusion criteria: good general health, 25-65 y old, smoked 10 to 40 manufactured, nonmenthol cigarettes with 1- to 7-mg tar as measured by the FTC method daily for at least 10 y; exclusion criteria: clinically significant renal, hepatic, metabolic, cardiac, and pulmonary disease, illicit drug use, use of nicotine-containing products other than manufactured cigarettes, unwillingness to use the EHCSS exclusively; women were excluded if they were pregnant, lactating, or intended to get pregnant during the study period	Urine nicotine and metabolites, total NNAL, 3-HPMA, 1-HOP and its glucuronide and sulfate, urine mutagenicity, 4-aminobiphenyl hemoglobin adducts, carboxyhemoglobin, urinary 11-dehydrothromboxane B2, and 8-*epi*-prostaglandin $F_{2\alpha}$; low-density lipoprotein cholesterol and high-density lipoprotein cholesterol, triglycerides, bilirubin	N = 97 (64 EHCSS, 33 conventional cigarettes), 52 females, 42 male; mean CPD, 23.8; mean age, 41.8 y; 1% Black, 96% White, 3% Hispanic; mean BMI, 25.6
Sarkar (61)	To evaluate exposure to selected gas-phase constituents when smokers switched to cigarettes with a highly activated carbon filter	Conventional cigarettes (either a 6-mg or a 11-mg FTC tar product), or test cigarettes containing carbon filters (comparable tar levels)	Controlled in short-term studies; *ad libitum* in long-term studies	Randomized, controlled, open-label, parallel-group, forced-switching design; smokers in two separate studies were randomized to continue to smoke conventional cigarettes (a 6-mg or a 11-mg FTC tar product for each of the studies, respectively), to smoke test cigarettes containing carbon filters (comparable tar levels), or to stop smoking; after completing 8 d in controlled smoking conditions (short-term studies), smokers had the option to continue in 24-wk long-term ambulatory studies with unrestricted smoking using assigned product; subjects were dropped from the study if >5% of their daily cigarettes were nonstudy cigarettes; recruitment through local ads by a clinical study center; inclusion criteria: 21-65 y old, in good general health, smoked Marlboro Lights or Ultra Lights for at least 4 wk before the start of the study, had smoked 10 to 30 manufactured nonmenthol cigarettes daily for at least the preceding 12 mo	Smoking topography; urinary excretion of mercapturic acid metabolites of 1,3-butadiene, acrolein, and benzene; nicotine and five of its metabolites, total NNAL, 1-HOP, and creatinine; several biomarkers of inflammation, oxidative stress, and cardiovascular risk (urine microalbumin, urinary 11-dehydrothromboxane B2 and 8-*epi*-prostaglandin $F_{2\alpha}$, blood fibrinogen, von Willebrand factor and highly sensitive C-reactive protein, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, and triglycerides)	N = 160 (randomized to short term); mean age, 32.8 y; 79 males, 81 females; mean CPD, 18.2; N = 76 (completed long term)

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Table 6. Intermediate-term clinical trials (Cont'd)

References	Goals	Product	Product instructions	Study methods	Measures	Subjects
Sepkovic (89)	To determine levels of nicotine after switching to a lower- or higher-nicotine brand and when subjects returned to their usual brand	Standard brand, 0.4 mg higher-nicotine brand and 0.4 mg lower-nicotine brand	*Ad libitum*	Between-subject design; subjects were assigned to either continue to smoke their standard brand or to smoke cigarettes that were 0.4 mg higher or lower in nicotine, and after 4 wk, subjects switched back to their usual brand; inclusion criteria: good health with no history of diabetes mellitus, hypertension or respiratory tract disorders, no interest in smoking reduction	Blood carboxyhemoglobin, plasma nicotine, cotinine and thiocyanate; blood pressure, heart rate	*N* = 8 (4 per group); mean age, 28.5 y; mean CPD, 23
Stewart (86)	To examine the effects of switching to Eclipse on pulmonary and blood biomarkers	Eclipse	*Ad libitum*	Within-subject, forced-switching, open-label design; assessed smokers at baseline and at 2 and 4 wk after switching to Eclipse; recruitment through a group of smokers not interested in quitting; they were also identified as "acceptors" of Eclipse in another study; had smoked at least 75% of 2 cartons of Eclipse and expressed desire to purchase them in the future; inclusion criteria: 21-50 y old, healthy, and ≥20 CPD containing ≥5 mg of tar for a minimum of 3 y; exclusion criteria: pipe smoking, tobacco chewing, pregnancy, more than 14 alcoholic drinks/wk, chronic respiratory disease, ischemic heart disease, abnormal liver or renal function	Pulmonary function, sputum cells, ^{99m}Tc-DTPA, and blood leukocyte activation and production of reactive oxygen species; respiratory symptoms	*N* = 10; mean age, 35 y; mean pack-years, 19
Stiles (80)	To study Eclipse compared with subjects' regular brand	Eclipse and usual brand	*Ad libitum*	Within-subject, open-label design with 2 groups of subjects conducted about 6 mo apart; subjects smoked one of their usual brand cigarettes in the first test session after reporting the number of cigarettes they had already smoked that day; smokers were then sent home with a supply of Eclipse cigarette for 2 wk, then returned to the laboratory for their next test session	Smoking topography; blood carboxyhemoglobin, and serum nicotine; cigarette sensory attribute ratings	*N* = 52 (26 in each group)

NOTE: Majority of trials assess for amount of product(s) used.
Abbreviations: FEV1, forced expired volume in 1 second; FVC, forced vital capacity; ^{99m}Tc-DTPA, ^{99m}technicium-diethylenetriaminepentaacetic acid.

72-76, 81-83, 87, 89) and heavily dependent on tobacco (mean FTND of 5.4 to 6.8; refs. 23, 73, 81-83). Studies that did not specifically recruit for cigarette type observed that most smokers smoked low-tar cigarettes (54%; ref. 23) or were evenly split between light and regular cigarettes (39%, respectively; ref. 72). Two studies described the stages of change, with one study describing all of its subjects as pre-contemplators (83) or 53% pre-contemplators and 47% contemplators (81).

Compliance with Product Use. The majority of the intermediate-term studies that described compensation for participating in the study reported that subjects were paid for their time (56, 61, 74, 81, 82, 86). Two studies reported monetary compensation for time and for compliance with assigned product use (23, 72). In one study, subjects purchased cigarettes at reduced prices (77). Another study required subjects to deposit money at the start of the study and a portion of the money was returned each week for attendance and product compliance (76). Compliance to product use could only be determined by measuring abstinence from smoking, using CO if smokers were assigned to noncombustible tobacco containing PREPs (oral tobacco), using CO and a tobacco alkaloid such as anatabine or total NNAL if smokers are assigned to nicotine replacements (e.g., refs. 23, 72), or using cotinine if one of the conditions involved no smoking and no product use. One study examined spot urines to be analyzed for total NNAL as a compliance check for use of tobacco products other than the PREP, with levels above a specified level considered to be an indication of noncompliance (74). Otherwise, compliance was determined by self-report (89), comparing the amount of dispensed products with return of unused products or packaging (70, 71, 73, 74, 76, 81, 83-85, 90), or returning used products (77, 83). Self-reported number of cigarettes was determined either by written daily diaries (61, 76) at the weekly clinic visit or in two studies by calling an answering machine every night before bed time (83) or by recording date, time, and brand of each cigarette smoked using an electronic diary (74). Interestingly, the results from the Frost-Pineda et al. (74) study showed that the daily diaries underestimated report of product use compared with the pharmacy logs.

Summary for Short-Term and Intermediate-Term Clinical Trials. Short-term clinical trials allow for an assessment of a broader range of exposure biomarkers and some adaptation to product use than laboratory studies. These trials may have the advantage of potentially greater compliance and less dropouts than longer trials. Furthermore, short-term trials would allow studies to be conducted in a residential unit, which allows for a greater control over product use, protocol compliance, and control over confounding factors that may affect exposure biomarkers such as diet. However, these residential studies, which are typically 1 week in duration, may be too short in duration for using biomarkers with longer half-lives, which may lead to the necessity to make adjustments for residual effects for these biomarkers (59). Another drawback of these residential studies is the unnatural environment, which may affect the pattern of PREP use and which may not reflect product use in a more naturalistic setting. For example, in one study, subjects underwent a short-term residential phase involving controlled smoking of assigned cigarettes. This phase was followed by a 24-week period of unrestricted smoking of the assigned products in the natural environment. The results from the residential phase showed similar decreases in biomarkers as during the 24-week follow-up phase; however, the extent of reduction was greater during the follow-up phase. The authors attributed this difference to more restrictions on smoking in the subject's natural environment (73). On the other hand, in another study, subjects were confined for 36 hours during baseline and 1 week after assigned cigarette use, with instructions for unrestricted product use. The biomarker results showed greater change during confinement than observed at the end of a 12-week phase of product use in the natural environment (74). Thus, although both studies suggest that short-term residential studies show similar trends in results as the longer nonresidential studies, the extent of change in exposure may differ.

The strengths of the intermediate-term studies include greater time for stabilization of use, examination of more naturalistic use, and the ability to use biomarkers with longer half-lives. The limitations depend on the goal of the study. If the focus of the study is to determine toxicant exposure during long-term use, compliance with use is a major concern. However, if the goal is to examine the naturalistic pattern of use over time, then compliance is not as significant. One concern over some of the long-term studies is the potentially high dropout rate, which can be as high as 60% (73).

Several short-term and intermediate-term studies had design features that are useful in the examination of effects of products on exposure biomarkers. For example, as described previously, some studies have combined both short-term laboratory or residential phase with longer-term phase conducted in the natural environment (74). The advantages of these studies include assessing products while under greater control, yet assessing products for a longer period of time in a naturalistic setting. As another example, although within-subject crossover design studies tended to be smaller in subject size than a between-subject study design study, the within-subject design minimizes the effects of intersubject variability for biomarkers of exposure (e.g., how a subject metabolizes nicotine or carcinogens) and other biologically responses to a product. These types of studies, however, also tend to be shorter-term and may not be conducive to longer-term evaluation of a tobacco product because subject retention may be an issue. Also, because of the small sample size, generalizability of results may be a concern. The shorter-duration trials may be conducive to examining dose-response curves to determine if the effects on measures are an actual result of the product. For example, in the Hughes and Keely study (83), subjects were required to use 5, 10, and 15 Accord devices per day and then 4 mg of gum for 2 weeks each. In between sessions, smokers were required to resume using solely their usual brand cigarette. Although the focus of this study was to determine how the amount and type of product use affect the amount of usual brand cigarettes smoked, this study design can be easily adapted to determine the dose-response effect of sole use of a product.

Differences existed in instructions for product use across studies. In some studies, the amount of PREP use was controlled, whereas other studies allowed *ad libitum*

use. Requiring smokers to use a specified amount of the product may provide some insight into the toxicant exposure and effect from the product, although it would be difficult to control for topography of product use. On the other hand, *ad libitum* use might more accurately reflect how the product might be used. Few studies have compared the results between these two different instructions for product use. In the residential study conducted by Roethig et al. (59), one group of smokers was randomized to an EHCSS in which smoking opportunities were given every 32 minutes between 07:00 and 23:00 and use did not exceed rates observed during the acclimation period of usual brand use. The other group of smokers was allowed to smoke EHCSS at any time between 07:00 and 23:00 with a cap of 60 cigarettes. With the exception of greater number of EHCSS smoked per day but less puff volume observed in the unrestricted smoking condition compared with the controlled smoking condition, no significant differences were found in any of the biomarkers of exposure. It should be noted that the EHCSS only allows for a maximum of 8 puffs per cigarette. The impact of these different instructions for product use outside of a residential setting is unknown.

Short-term studies generally require subjects to use only the study product and disallow dual use (e.g., use of concurrent conventional tobacco products), although compliance cannot be verified. Some intermediate-term studies also encourage subjects to only use the PREP, whereas other studies allowed use of the subjects' usual brand of cigarettes. Both study designs are valuable—the former to determine the toxicity of the specific PREP and the latter studies to determine real-world pattern of use and resultant toxicant level. One study required sole use of the study product and then undertook a post hoc analysis that examined subjects who reported only using the assigned product versus subjects who reported using both the assigned product and conventional cigarettes, with higher exposures observed in the analysis among dual product users (74).

The measures in both the intermediate-term and short-term studies were somewhat similar, with some variations based on the intent of the study. However, most studies, especially intermediate-term ones, did not consider potential confounding factors that may influence the biomarker results, such as diet, other environmental exposures, and alcohol use. Studies on PREPs would benefit if similar measures across studies and a broader panel of biomarkers within and across disease states were used.

Short-term and intermediate-term studies include control groups, but the types of control conditions vary. It would seem that the most valuable comparison groups would include abstinence, with or without use of medicinal nicotine, to compare the product to an intervention with known reductions in health risks. Another option would be to use ultralight cigarettes (i.e., <1 mg tar yield as measured on a smoking machine) as a comparison to marketed products considered to have relatively low toxicant yield levels (although this option has not been adequately tested). In one intermediate-term study, subjects were required to have smoked an ultralight cigarette, as the reference conventional cigarette, for at least 4 weeks before the start of the study (74).

Clinical Trials Workshop Recommendations for Short-Term and Intermediate-Term Clinical Trials. The Clinical Trials Workshop participants recommended that a study design include multiple arms and both negative and positive comparators while varying the level of adherence to the product. For example, these arms would include (*a*) smokers who use usual brand; (*b*) smokers who decrease intake of usual brand (which can include different levels of decreased use based on claims); (*c*) controlled (i.e., fixed amount) use of the PREP product only; (*d*) *ad libitum* use of PREP product only; and (*e*) *ad libitum* use of PREP plus concurrent use of usual brand. Furthermore, nonsmokers, but particularly smokers who quit, were considered be a valuable comparison group depending on the specific aims of the study. A no-smoking group would compare PREP effects to the ideal case (cessation) while also showing the sensitivity of the design and outcome measures.

Other critical features of the study design would include being of sufficient duration to achieve stabilization of tobacco use behavior. Workshop participants reported that in prior studies, this duration has been less than 4 weeks for conventional cigarettes. Additionally, the length of study should be sufficient to achieve "steady state" of the biomarker, and when using within-subject study design, the half-life of a biomarker must be taken into consideration to ensure no carry-over effects. If disease outcome is to be assessed in the study, the study length has to be long enough (e.g., in some cases, months or years) to measure changes in disease occurrence.

The following critical questions need to be addressed for these types of trials: 1) how do responses differ across instructions for product use (*ad libitum* use versus use of specific amounts, concurrent product use with usual tobacco products versus product use only); 2) what is the length of time it takes for product use to stabilize and is stabilization under clinical trial conditions different from that under natural use conditions; 3) how do we determine if the exposure is due to the product, the way the product is used, or characteristics of the individual; and 4) how do results from switchers differ from those who have already chosen to use the products?

Cross-sectional Studies. Cross-sectional studies have been conducted to compare self-selected product users who use different brands or types of products (see Table 7). For example, past studies have recruited smokers with differing nicotine or tar yields to determine biomarker levels of cotinine, thiocyanate, carbon monoxide, tobacco specific nitrosamines, or cardiovascular or lung cancer risk factors across the different types of cigarettes (e.g., refs. 91-99). One study investigated the concentrations of urinary biomarkers in relation to the concentrations of selected toxicants in mainstream cigarette smoke as determined by machine smoking of cigarettes in a manner that mimics an individual's smoking behavior (100). In these cross-sectional studies, subjects have been recruited from general population surveys (92, 93, 96, 98), smoking cessation clinics, treatment trials or experimental studies (94, 95, 99), or recruited specifically for the study (91, 97, 100). The restrictiveness of the inclusion and exclusion criteria has varied across cross-sectional studies, ranging from just being an active smoker (91) to smokers who smoke within a specific range of number of cigarettes, use no other nicotine-containing products, or are in good general health with no current

mental health problems nor using of psychotropic medications (100). Very stringent inclusion criteria occur when recruiting from treatment studies (90). Other studies have also examined the association with disease status (or risk) across different yields of cigarettes using epidemiologic data (see ref. 101 for reviews; refs. 102, 103).

Cross-sectional studies have also compared different types of smokeless tobacco products on carcinogen biomarkers (see Table 7; e.g., refs. 104, 105). Snuff and snus users have been compared to smokers on cardiovascular or diabetes risk factors using population-based samples (e.g., refs. 106-108), on carcinogen exposure biomarkers (109), and on the enzyme aldehyde dehydrogenase (110) using a clinical sample, and on differences in the prevalence of actual disease (111-117). One study recruited relatively healthy smokers, smokeless tobacco users, NRT users, and healthy controls to measure serum immunoglobulin levels (118). Recruitment of subjects for these smokeless tobacco studies has either been specifically for the type of tobacco user of interest, as above, through another study sample (104, 109) or random sampling from a population (106-108). Some of these studies had specific criteria for recruitment such as gender, age, and ethnic/racial group (107); exclusion of specific medical conditions because biomarkers of effect were related to those conditions (107, 118); or specific criteria that would classify the sample as current users of moist snuff or cigarettes, ex-smokers who used nicotine replacement, or a control group of ex-tobacco users or never users (118).

The advantage of the cross-sectional study is that these individuals have chosen to use the product, and therefore the results reflect values in a population most interested in using the product. Population-based sampling might provide the additional benefit of obtaining a sample somewhat representative of the general population of users for that product. In addition, in this population-based sample, the product for some of the subjects has been used for a sufficiently long enough duration that the pattern of use has stabilized. The disadvantage is that a sufficient population base of users is necessary so that the product can be evaluated and other factors associated with the product, such as product design, content, and marketing, may change over time.

Clinical Trials Workshop Recommendations. Clinical Trials Workshop participants suggested that cross-sectional studies be conducted to compare new, experienced, and long-term switchers to PREPs and to describe differences in subject characteristics and outcomes of these populations.

Clinical Trials Workshop Recommendations for Other Issues. Whereas Workshop recommendations for study design have been addressed in each of the study type subsections, the following provides a summary and recommendations on issues that are relevant to all types of clinical trial studies.

Subject Recruitment, Characteristics, and Retention. The issue that cuts across all study designs involves recruitment (how should subjects be recruited; what should they be told in advertising and consents; and who should be recruited). Studies differ in how they advertise for subjects, with some studies recruiting specifically for smokers interested in using new tobacco products that may be "safer" or with reduced tobacco toxins. How and what is conveyed to the subject may have major impact on how the subject perceives and uses the products.

In addition, most studies have very restrictive inclusion and exclusion criteria that are not unlike the criteria used in pharmacologic trials. The looming question, which also is relevant to pharmaceutical trials, is whether the products are being tested in a population that is representative of the typical user, and if not, whether the public can afford to determine the major effects of the product during post-marketing surveillance rather than during the clinical trials. For example, many of the laboratory studies tended to study populations that were young, healthy, and with lower levels of dependence, which may be quite unlike the population interested in using PREPs.

Although these criteria may not be problematic in the initial testing of the products, studies need to be conducted that use populations of smokers similar to the ones that are likely to use the products. Just as it is important to carefully examine who is being recruited, it is also important to characterize who drops out of these studies and why, and who persists in using these products. Only some of the studies captured this information (e.g., refs. 23, 72, 79, 82, 83).

Advertising and recruitment information was considered to be important by the Clinical Trials Workshop participants in determining a subject's likelihood of participating and subsequent behavior in the study. They believed that information and instructions should be communicated to the subject in a way that would avoid subject response biases due to subject expectations. To avoid bias, it was considered best if the wording was nondirective, such as advertising for "smokers interested in testing a new tobacco product" or specifically for "smokers not ready to quit smoking but interested in testing a product that may or may not reduce exposure to cancer-causing chemicals" depending on the targeted population. Regardless of the content of advertisement, it was recommended that the consent form should provide toxicity information to the potential study subject.

The Workshop participants thought newspaper or notice board advertisements were the most useful and accessible strategy for subject recruitment. Another potential recruitment source, which has been untapped by the majority of studies, are participants of national surveys. Standardized questions about PREP knowledge, interest, or use could be included in these surveys. Users of PREP products or those tobacco users meeting specific study criteria, such as those interested in trying PREP, could be invited to participate in subsequent research for a monetary incentive.

The criteria for inclusion in the study or subject characteristics were also discussed by the Clinical Trials Workshop participants. For abuse potential studies, the characteristics of the study participants would be dependent on the specific study design. Regular smokers are appropriate for acute effects and short-term self-administration studies, whereas heavier smokers would be more appropriate for cross-dependence and compensatory smoking studies. Tobacco users interested in quitting would be most appropriate for cessation studies and to gauge the effect of the product on quitting. In general, however, the Workshop participants believed that people

Table 7. Cross-sectional studies for smoking and smokeless tobacco use

Reference	Goals	Product	Study methods	Measures	Subjects
Smoking studies					
Bernert (91)	To examine biomarkers in smokers of either light or regular cigarettes	Commercial regular and light cigarettes	Subjects completed a brief questionnaire about smoking behavior; blood and untimed (spot) urine samples collected; inclusion criteria: current, active smokers	Serum cotinine, NNAL and NNAL-glucuronide, adducts of 4-aminobiphenyl hemoglobin, urinary creatinine	*N* = 150; 109 males, 41 females; mean age, 34.7 y; mean CPD, 17.1; mean years smoked, 18.7; 80 Blacks, 70 Whites
Blackford (92)	To examine salivary cotinine and addiction among smokers	Commercial cigarettes	Multi-country; Brazil: multistage random sampling; China: convenient sampling; Mexico: convenient sampling; Poland: random-route method; cotinine concentration was measured using a saliva sample from each participant; its relationship with numbers and types of cigarettes smoked was quantified by applying regression techniques; inclusion criteria: smoked 1-60 cigarettes in the previous 24 h, regular smoker, did not smoke cigars, did not use any NRT in the past 3 d, did not smoke hand-rolled cigarettes; exclusion criteria: ratio of cotinine concentration to number of cigarettes smoked, >35 ng/mL per cigarette	Salivary cotinine; height, weight, BMI; smoking behavior, FTND, American Thoracic Society adult respiratory questionnaire	Brazil: *N* = 360, China: *N* = 490, Mexico: *N* = 1,006, Poland *N* = 517
Borland (93)	To determine the relationship between levels of carbon monoxide in cigarettes and cardiovascular disease, lung disease, and mortality	Commercial cigarettes	Each subject completed a questionnaire about smoking behavior; inclusion criteria: participants in the Whitehall study of men that examined 18,403 civil servants in 1967-1969; exclusion criteria: carbon monoxide values of cigarettes that were unknown	Spirometry; smoking consumption and cardiovascular disease questions	*N* = 4,910; all males; 40-64 y old

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Groman (97)	To examine differences in CO concentrations in the expired air of smokers who smoked light cigarette brands versus smokers who smoked regular brands	Commercial cigarettes	Smokers were divided into two groups: those who smoked a light cigarette brand and those who smoked a regular cigarette brand; recruitment through first visit clients at publicized information meetings held by the Nicotine Institute, Vienna, during a 3-wk period	Exhaled CO; FTND	*N* = 178 (63 light cigarette brand smokers, 115 regular cigarette brand smokers); 83 males, 95 females; mean age, 49.1 y
Harris (98)	To determine the association of smoking medium-tar filter cigarettes versus low-tar or very-low-tar filter cigarettes and mortality from lung cancer	Commercial very-low-tar filter, low-tar filter, medium-tar filter, and high-tar filter cigarettes	Data from participants in the Cancer Prevention Study II were analyzed for cigarette brand smoked between 1982 and 1988 and the risk of lung cancer; inclusion criteria: ≥30 y old who had either never smoked, were former smokers, or were currently smoking a specific brand of cigarette when they were enrolled in the cancer prevention study, smokers of their current brand for at least 5 or 10 y; exclusion criteria: history of cancer other than nonmelanoma skin cancer, or emphysema, reported any smoking related condition (emphysema, chronic bronchitis, heart disease, use of heart drugs, stroke, diabetes, claudication, currently sick); men who ever smoked pipes or cigars or chewed tobacco; and men and women whose current smoking status could not be ascertained	Race, educational level, marital status, blue collar employment, occupational exposure to asbestos, intake of vegetables, citrus fruits, and vitamins; analyses of current and former smokers for age when they started to smoke and number of cigarettes smoked per day; death from cancer of the trachea, bronchus, or lung as the underlying cause, coded from the death certificate	*N* = 940,774; 364,239; males 576,535 females

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Hecht (94)	To evaluate levels of two urinary biomarkers of lung carcinogen uptake in smokers of different tar yield cigarettes	Commercial regular, light, and ultralight cigarettes	Subjects completed a tobacco use questionnaire stating their current brand of cigarettes as regular, light, or ultralight; urine samples were collected; recruitment in Study 1 through advertisements; Study 2 through invitation letters and advertisements; subjects were participants in two studies examining the effects of smoking reduction on levels of carcinogen biomarkers; inclusion criteria: (Study 1) cigarette smokers 18 to 70 y old and interested in reducing cigarette use but not quitting within the next 30 d, smoking 15 to 45 CPD for the past year, good physical health, no contraindications for nicotine replacement use, good mental health, not using other tobacco or nicotine products and not pregnant or nursing; (Study 2): 18 to 80 y old who also had heart disease and were interested in reducing cigarette use but not quitting within the next 30 d, smoking $\geq$ 15 CPD, having coronary artery disease, arrhythmia, congestive heart failure, peripheral vascular disease, or history of a cerebrovascular event; no unstable angina within the past 2 wk; no unstable psychiatric or substance use diagnoses; and no contraindications to NRT (including pregnancy or intention to become pregnant)	1-HOP, total NNAL plus its glucuronides, total cotinine (cotinine plus its glucuronides)	N = 115 subjects in Study 1 and N = 60 in Study 2; pooled results: smoking regulars, 26.9%, lights, 45.7%, ultralights, 27.4%; mean age: smokers of regulars, 50.5 y; lights, 49.1 y; ultralights, 51.4 y; mean CPD: smoking regulars, 27.9; lights, 24.1; ultralights, 26.1

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Melikian (100)	To examine the relationships between delivered dosages of smoke constituents (e.g., nicotine and select carcinogens) determined by using actual human smoking conditions with levels of corresponding urinary metabolites in smokers	Commercial cigarettes	Single visit; subjects collected butts 4 d before visit; administered comprehensive questionnaire about smoking history, gave urine sample after smoking 3-4 cigarettes during smoking topography measurements; recruitment through newspaper advertisement; inclusion criteria: 18 and 59 y old, smoked ≥10 CPD for 1 y, in good general health, no history of any tobacco-related disease, no unstable medical condition, no psychotropic medications, and no psychiatric diagnosis at the time of study; exclusion criteria: using any tobacco or nicotine-containing products other than cigarettes for at least 3 mo before the study, pregnant and nursing women	Quantified nicotine, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK), and benzo(*a*)pyrene in the mainstream smoke condensate generated by machine smoking of each individual's cigarettes under conditions that reflect that individual's smoking pattern; urinary cotinine, NNAL, 1-HOP; the brand of cigarettes smoked, type of pack (hard or soft), mentholated or nonmentholated cigarettes; occupational exposure, family medical history, diet and other lifestyle factors, FTND	*N* = 257; 129 females, 128 males; mean age: females, 33.1; males, 35.0; mean CPD: females, 15.9; males, 16.8; mean FTND score: females, 4.1; males, 4.6; mean BMI: females, 25.5; males, 26.8
Russell (95)	To determine differences in biomarkers and smoking behavior for different tar yield cigarettes	Commercial cigarettes	Single visit; subjects smoked one of their usual cigarettes and a venous blood sample was taken 2 min later; recruitment through Maudsley Hospital smokers' clinic or from experimental studies on smoking at the addiction research unit; exclusion criteria: cigars or hand-rolled cigarettes	Carboxyhemoglobin, plasma nicotine	*N* = 330; 124 males, 206 females; mean CPD: males, 36.2; females, 32.6
Russell (99)	To estimate the tar intake of low-tar smokers compared with smokers of other brands	Usual brand (middle, low to middle, and low tar)	Subjects attended in afternoon, smoked one of their cigarettes, and a venous blood sample was taken 2 min later; the tar, nicotine, and CO yields of the cigarettes were obtained from the Health Departments of the United Kingdom; exclusion criteria: use of cigars or hand-rolled cigarettes in the "middle to high" tar category (23-28 mg/cigarette)	Blood nicotine, cotinine, carboxyhemoglobin; CO, tar intake derived from the measured intake of a marker (e.g., blood nicotine), and the ratio of the tar to marker yields of the cigarette (e.g., tar intake = plasma nicotine × tar/nicotine yield ratio)	*N* = 392; 255 females, 137 males; mean age: females, 38.4 y; males, 40.4 y; mean CPD: females, 29.0; males, 31.8

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Woodward (96)	To determine differences in biomarkers and smoking behavior for different tar yield cigarettes	Commercial cigarettes	Single visit where subjects gave blood and CO samples and completed a smoking history questionnaire; recruitment through a baseline population survey of the Scottish Heart Health Study; inclusion criterion: current cigarette smokers; exclusion criteria: cigars and/or pipes and subjects who smoked cigarette brands that were not reported by the Government Chemist or rolled their own cigarettes	Serum thiocyanate, serum cotinine; exhaled CO	*N* = 127 low tar males, 405 low tar females; 540 middle tar males, 675 middle tar females; 466 high tar males, 541 high tar females
Smokeless tobacco studies					
Andersson (104)	To investigate the uptake and metabolism of nicotine by ST users and effects on oral mucosa	Loose snus, portion-bag snus, and chewing tobacco	Subjects attended the dental clinic for a thorough oral examination; subjects used their usual brand *ad libitum* and kept track of amount used for 7 d; on day 6, urine samples were collected for 24 h; on the same day, the ST users saved all used portions of ST; on day 7, one saliva sample was collected 30 min after using a pinch of snus or a piece of chewing tobacco; after 30 min, subjects rinsed their mouth with water and a whole mixed saliva sample was collected; recruitment from a previous study consisting of 252 healthy men with a regular snus habit for at least the previous 3 mo and with no other current tobacco use; the users of chewing tobacco were selected from another study of 20 healthy men with no other tobacco habit and who were living in the area; inclusion criteria: equal daily consumption and usage of the same tobacco brand	Urine nicotine and cotinine, glucuronic acid conjugates of nicotine and cotinine, *trans*-3′-hydroxycotinine and nicotine-*N*′-(1)-oxide and cotinine-*N*-(1)-oxide; salivary cotinine; analysis of chemical constituents of smokeless tobacco products: nicotine and tobacco-specific nitrosamines [*N*′-nitrosonornicotine, *N*′-nitrosoanatabine, *N*′-nitrosoanabasine, and 4-(*N*′-methyl-*N*′-nitrosoamino)-1-(3-pyridyl)-1-butanone]; clinical exam to record lesions in the oral mucosa; questions for ST use, general health, medication, previous tobacco habits, and alcohol consumption	*N* = 54 (22 loose snus users, 23 portioned snus users, 9 chewers); mean age range, 38.8-50.4 y

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Bolinder (111)	To examine whether long-term use of smokeless tobacco is associated with mortality from cardiovascular disease compared with nonusers and cigarette smokers	Smokeless tobacco and cigarettes	Construction industry workers received a health examination through the Swedish Construction Industry's Organization for Working Environment Safety and Health and the cause of their mortality during a 12-y period was determined; recruitment through invitation; exclusion criteria: women	Heart rate, blood pressure, weight, height; past and current health, medication use; cause of death; tobacco use	*N* = 135,036; all males; age range, 35-65 y; 6,297 smokeless tobacco users, 14,983 smokers of <15 cigarettes per day, 13,518 smokers of ≥15 cigarettes per day, 17,437 ex-smokers, 50,255 "other" tobacco users, and 32,546 nonusers
Eliasson (106)	To assess the relationship between cigarette smoking and snuff use and biomarkers	Usual brand cigarettes and snuff	Single visit; subjects (regular smokers, ex-smokers, snuff dippers, and nontobacco users) fasted overnight (12 h) and underwent a 75-g oral glucose tolerance test; recruitment through choosing subjects at random from a cardiovascular disease study; inclusion criterion: 25-64 y old	Blood lipids, plasma glucose, serum insulin, plasma fibrinogen, tissue plasminogen activity and plasminogen activator inhibitor type 1 activity, glucose and insulin levels; plasma nicotine, cotinine; BMI, physiologic measures	*N* = 1,266 (581 nontobacco users, 238 ex-smokers, 317 smokers, 92 snuff dippers analyzed, 38 snuff and cigarettes, but no other type of tobacco analyzed); 604 males, 662 females
Gyllen (118)	To determine if using smokeless tobacco or nicotine replacement affects serum immunoglobulin levels	Oral moist snuff and NRT	Cross-sectional, parallel-group study; subjects gave blood and urine samples and tobacco use history; healthy subjects with no exposure to nicotine served as a control group; recruitment through advertisements in 2 newspapers; inclusion criteria: 18-75 y old, former smokers in any group had quit smoking at least 6 mo before study entry; exclusion criteria: diseases or medical treatments that influence serum immunoglobulin levels, pregnancy; chronic liver or renal disease, diabetes, severe cardiac failure, severe chronic lung disease, any known immunodeficiency or rheumatologic disease, history of bronchial asthma, allergy or atopy; symptoms of infectious disease with pyrexia or any use of antibiotics, antihistamines or N-acetylcysteine during 4 wk before the investigation; use of steroids including estrogen hormone substitution or any other immunomodulating treatment or vaccination during 2 mo before the investigation	Urinary cotinine, immunoglobulin class, and IgG subclass data	*N* = 77; 48 ST users, 29 NRT users; 35 males, 42 females; mean age, 44 y; healthy controls: *N* = 44; 20 males, 24 females; mean age, 43 y

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Hecht (109)	To determine biomarkers among ST users and smokers	Commercial cigarette users and smokeless tobacco	Baseline data from three studies involving smokers and three studies involving smokeless tobacco users who were seeking treatment for tobacco use reduction; recruitment through local advertising; inclusion criteria (Study 1): 18-70 y old, interested in reducing cigarette use but not quitting within the next 30 d, smoking 15-45 CPD, in good physical health, no contraindications for nicotine replacement use, good mental health, not using other tobacco or nicotine products, and not pregnant or nursing; (Study 2): 18-80 y old who also had heart disease and were interested in reducing cigarette use but not quitting within the next 30 d, smoking ≥15 CPD, no unstable angina within the past 2 wk, no unstable psychiatric or substance use diagnoses, and no contraindications to NRT (including pregnancy or intention to become pregnant); (Study 3): smoking ≥15 CPD, an unsuccessful quit attempt in the past year, no specific plan to quit in the next 30 d and willing to attempt smoking reduction as a short-term goal, used other tobacco products three or fewer times in the past week, no current use of NRT, no use of Zyban in the past 2 wk, not pregnant, and no treatment for alcohol or drug abuse in the past year; (Studies 4-6): 18-70 y old, interested in reducing smokeless tobacco use but not quitting, using ST daily for the past 6 mo, in good physical health and good mental health	Urine NNAL and cotinine	Smokers: pooled (Studies 1-3): N = 420; 62% male; mean age, 49.5 y; mean CPD, 25.8; 80% White; smokeless tobacco users: pooled (Studies 4-6): N = 182; all males; mean age, 32.9 y; mean use of ST, 4.2 tins/wk; 99% White

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Helander (110)	To determine levels of ALDH activity in Swedish moist snuff users	Swedish moist snuff and cigarettes	Blood samples collected between 9:00 and 12:00 a.m. from cigarette smokers, moist snuff users, and nontobacco controls were analyzed for biomarkers; recruitment through donors at a blood center	ALDH activity (whole blood, erythrocytes and leukocytes), plasma nicotine, cotinine	*N* = 66 (24 smokers, 17 moist snuff users, 25 nontobacco users); 46 males, 20 females
Huhtasaari (112)	To assess the risk of myocardial infarction among snuff users, cigarette smokers, and nontobacco users	Snuff and cigarettes	Case-control study in Northern Sweden; male patients with a myocardial infarction were compared with those without a myocardial infarction from a population survey of cardiovascular risk factors (WHO Multinational Monitoring of Trend and Determinants in Cardiovascular Disease Project); inclusion criteria: male, 35-64 y old	Serum samples for lipid concentrations and total cholesterol concentration; blood pressure; tobacco consumption (regular snuff dipping, regular cigarette smoking, nontobacco use); questionnaire with items on tobacco habits, social background, medical history, and drugs taken	*N* = 1,174 (585 myocardial infarction, 589 no myocardial infarction); all males; 169 cigarette smokers (myocardial infarction), 114 cigarette smokers (no myocardial infarction), 59 snuff dippers (myocardial infarction), 87 snuff dippers (no myocardial infarction)
Huhtasaari (113)	To determine if snuff use affects the risk of myocardial infarction	Snuff and cigarettes	Population-based study associated with the Northern Sweden center of the WHO Multinational Monitoring of Trend and Determinants in Cardiovascular Disease (WHO MONICA) Project; patients with a fatal or nonfatal myocardial infarction were compared with men without myocardial infarction from the MONICA project who were matched for age and place of living with regards to their tobacco habits; recruitment through population registers for referents and through hospital records, general practitioner reports and death certificates for cases; inclusion criteria: male, 25-64 y old	ECG and cardiac enzymes (in hospitalized patients); questionnaire about social conditions, risk factors, tobacco use	*N* = 1,374 (687 acute myocardial infarction, 687 no myocardial infarction); all males; 248 cigarette smokers (acute myocardial infarction), 99 cigarette smokers (no myocardial infarction), 59 snuff dippers (acute myocardial infarction), 90 snuff dippers (no myocardial infarction)

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Johansson (114)	To examine the association between cigarette and snuff use and coronary heart disease	Snuff and cigarettes	A follow-up study; in 1988-1989, men in a Swedish national survey were interviewed and were followed up until 2000; inclusion criteria: male, healthy, 30-74 y old; exclusion criteria: poor self-rated health, a coronary heart disease hospitalization 2 y before the start of the study, participants who were interviewed with the aid of relatives, participants who lacked information about weight or height	BMI; the time to first hospitalization for fatal or nonfatal coronary heart disease event was classified according to ICD-9 and ICD-10; socioeconomic status, tobacco habits, leisure time activities, health questions	*N* = 3,120; all males; 1,036 never smokers, 854, former smokers, 793 daily smokers, 107 daily snuffers and never smokers, 245 daily snuffers and former smokers, 85 daily snuffers and smokers; mean age, 45.7 y
Kresty (105)	To establish the levels of urinary biomarkers in smokeless tobacco users and smokers	Oral snuff and oral chew	Subjects received an oral cavity examination; 24-h urine samples were collected and biomarkers were determined; recruited through advertisements at the Ohio State University campus and surrounding area; inclusion criteria: male, nonsmokers, tobacco chewers and snuff dippers had been regular users of smokeless tobacco for ≥ 1 y	NNAL and NNAL-glucuronide, cotinine, creatinine; gross appearance of the lips, oral mucosa, palate, tongue, mouth floor, oropharynx, and teeth; general oral hygiene and the presence of gingivitis and leukoplakia	*N* = 47 (23 snuff dippers, 13 tobacco chewers, 3 users of both, and 8 nonusers); all males; mean age, 27 y (snuff dippers), 25 y (tobacco chewers); 92% Caucasian
Luo (115)	To assess the risks associated with Swedish moist snuff for cancer of the oral cavity, lung, and pancreas	Swedish moist snuff	Workers in the Swedish building industry were given a health exam during 1978-1992 and were followed until end of 2004 by links with population and health registers; data from never smokers was also collected; inclusion criteria: snus user status (never, previous, or current), grams of snus per day (<10 or ≥10 g), smoking status (never, previous, or current), grams of smoking tobacco per day (continuous), and BMI (<25, 25-29, or ≥30); exclusion criteria: records with incorrect National Registration Numbers, men with a death or emigration date before entry, men with cancer before entry, men with incomplete tobacco exposure data	Incident cancers of the oral cavity, lung, and pancreas	*N* = 279,897; all males; mean age, 35 y; current or previous snuff users, 31%

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Persson (108)	To examine the relationship between cigarette smoking and use of oral moist snuff and impaired glucose tolerance and type 2 diabetes	Cigarettes and snuff	A population-based cross-sectional study conducted during 1992-1994: 52% of subjects who had a family history of diabetes compared with a random sample of men without a family history of diabetes; information was also collected about tobacco use; recruitment through mailing a short questionnaire; exclusion criteria: men who did not have a strong family history of diabetes or men without diabetes in the family, men who were unable to provide complete answers on the presence of diabetes in relatives, men who were born outside Sweden, men who had diabetes known to themselves	Health examination included a standardized 75-g oral glucose tolerance test according to WHO 1985, weight, height, and waist/hip ratios when wearing light indoor clothes without shoes, blood pressure; detailed questionnaire on tobacco use, dietary habits, physical activity, and psychosocial conditions	*N* = 3,128; all males; age range, 35-56 y
Rosenquist (116)	To determine the association between Swedish moist snuff and OOSCC	Swedish moist snuff and cigarettes	Population-based, case-controlled study; during September 2000 and January 2004, subjects diagnosed with OOSCC and matched controls were interviewed and examined; inclusion criteria (cases): individuals with OOSCC, born in Sweden and without a previous cancer diagnosis, except for skin cancer; recruitment through the two university hospitals in the region where almost all oral cancer cases are treated; inclusion criteria (controls): persons born in Sweden with no previous cancer diagnosis with the exception of skin cancer and who were living in the Southern Healthcare Region of Sweden were selected from the Swedish Population Register through stratified random sampling matching for age, sex, and county to cases	Cell samples from the oral cavity were collected for HPV DNA analysis; a thorough investigation of the individual's oral hygiene, dental status, and oral mucosa was done; a general assessment of the marginal bone level and the periapical status was made from panoramic radiographs; in current snuff users, mucosal changes at the site(s) where the snuff quid was regularly placed were recorded and classified according to the degree of clinical severity using a four-point scale; questions about medical history, medication, reactivated herpes labialis infection, oral sexual habits, use of tobacco, and alcohol consumption	*N* = 452; 132 cases; 320 controls; gender, 91 males, 41 females (cases), 215 males, 105 females (controls)

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Table 7. Cross-sectional studies for smoking and smokeless tobacco use (Cont'd)

Reference	Goals	Product	Study methods	Measures	Subjects
Teo (117)	To evaluate the risks associated with tobacco use and second-hand tobacco smoke	Cigarettes, beedies, pipes or cigars, chewing tobacco, paan, snuff, sheesha or water pipe, and other forms of smoked or nonsmoked tobacco	Standardized case-control study of AMI among subjects in 52 countries; administered a standardized questionnaire and examination; inclusion criteria (cases): first AMI presenting within 24 h of symptom onset, no cardiogenic shock or history of major chronic diseases; inclusion criteria (controls): age-matched (plus or minus 5 y) and sex-matched control without a history of heart disease or exertional chest pain	Concentrations of apolipoproteins B and A1 in serum; height, weight, waist and hip circumferences, blood pressure, heart rate; tobacco use, second-hand tobacco smoke exposure; information on dietary patterns, physical activity, alcohol consumption, education, income, psychosocial factors, personal and family history of cardiovascular disease, and risk factors (hypertension, diabetes mellitus)	*N* = 27,098; 12,461 cases, 14,637 controls; 9,456 males, 3,005 females (cases), 10,851 males, 3,786 females (controls); mean age, 58.1 y (cases), 56.9 y (controls); current smokers, 45.2% (cases), 26.8% (controls)
Wallenfeldt (107)	To evaluate the association of tobacco use and cardiovascular risk factors	Cigarettes and oral moist snuff	Population-based study; all measurements conducted during the morning; recruitment through being invited by mail to a screening examination; inclusion criteria were age 58 y, male, Swedish ancestry; exclusion criteria: cardiovascular or other clinically overt disease, treatment with cardiovascular drugs for ischemic heart disease, heart failure, hypertension, diabetes mellitus, and hyperlipidemia, unwillingness to participate	C-reactive protein; intima-media thickness in the carotid bulb, the common carotid artery, and the common femoral artery and plaque occurrence were measured by ultrasound; cholesterol and triglyceride levels; blood glucose; plasma insulin; blood pressure, body weight, height, waist and hip circumference; BMI and waist-hip ratio; information on general health and tobacco habits	*N* = 143 never smokers, 152 ex-smokers, 96 current smokers; 310 never snuff user; 33 ex-snuff user; and 48 current snuff user; all males; all 58 y old

Abbreviations: ALDH, aldehyde dehydrogenase; AMI, acute myocardial infarction; HPV, human papillomavirus; ICD, International Classification of Diseases; OOSCC, oral and oropharyngeal squamous cell carcinoma.

who enter trials are unique and enter into the trial for a variety of reasons, ranging from interest of the product to financial incentives. A group of persons enrolled in a study may or may not be representative of persons who would naturally select a product. Unfortunately, the representativeness of the sample cannot be easily assessed. The issue is only important if there are some selection criteria or other factors that result in different biological outcomes. That is, generalizability of the results will only be affected if biologically or mechanistically the association between use of a PREP and outcome (e.g., preference, withdrawal, biomarker) is modified by a factor such as sex, age, ethnicity, and/or genetic makeup. Therefore, examining the effects of these factors on outcomes may be necessary. Short of this examination, some Workshop participants recommended that the study population should try to be representative of the population of smokers or tobacco users with respect to (*a*) gender; (*b*) ethnicity; (*c*) age; (*d*) socioeconomic status; and (*e*) in larger, non-laboratory trials, type of tobacco user (inveterate smoker, those interested in quitting, comorbid smoker). Workshop participants believed that it is important to keep in mind that clinical trials are not intended to, and cannot, simulate population effects, and therefore are not valid for extrapolating results beyond the assessment of clinical effects (e.g., biomarker, physiologic, and behavioral responses). Similarly, there can be no inferences about why a person stays in a trial or how this might relate to the general population acceptance of the product because people staying in trials might do so because of the implied contract the subject has with the study. It should be recognized that switching trials is only one tool to understand the effects of a PREP on smokers. Other designs are needed such as cross-sectional and prospective studies, and post-marketing surveillance.

Few studies describe recruitment methods, content for recruitment, and inclusion and exclusion criteria, and few studies describe the characteristics of subjects who dropped out of the study. Uniform reporting of these study design features across all studies is important. Critical questions to address on this topic include (*a*) what factors moderate the outcome variables (e.g., biomarkers, see "Predictors of Response"), and (*b*) do different methods of recruitment and types of studies (laboratory versus intermediate-term clinical trials) attract different types of smokers and if so, do these differences affect outcome.

Compliance. Another major issue that is relevant to the different types of clinical trials is compliance with product use, particularly if the outcome criteria are effects of a product on biomarkers of exposure and health risks. It is a challenge to determine if subjects are not dually using the test product and their usual products. Short of having smokers stay in a residential setting for 3 to 6 months, this issue may never be resolved unless a biomarker can be developed to determine exposure solely to the PREP and no other tobacco products. In prior studies, subjects have been asked to keep daily diaries of tobacco product use, to return used and unused tobacco products, and were paid for complying with use of only PREPs or, conversely, not penalized for noncompliance. Additionally, studies have emphasized to the subject the importance of accurate reporting of both assigned and not assigned products.

In many studies, subjects were paid for their participation, and although payment is critical for retaining subjects, it biases the population toward those who may be primarily interested in the money rather than using the product. This issue cannot be avoided in laboratory studies but may be particularly important in the long-term clinical trials. On the other hand, it would be difficult to not pay subjects for all the testing that is required of them for the study. No specific recommendations were made by the Workshop participants on the issues of compliance and subject payment other than the importance of being sensitive to these issues and using methods to maximize honest reporting.

Predictors of Response. Very few studies examined what predicts a subject's response to products (e.g., the amount of product use, dropouts, slipping back to usual brand use, compensatory tobacco use behavior, and extent of exposure to biomarkers). This is a critical area of inquiry that is neglected and should include assessment of sex, age, ethnicity, dependence, duration of tobacco use, type of smoker, comorbid psychiatric history, and, possibly, genetic makeup. For example, females may respond differently to changes in sensory aspects of smoking or nicotine content of cigarettes compared with males (56, 119-121). African Americans may metabolize nicotine (122-125) and toxicants such as carcinogens (126) differently than Whites. Expectations for quitting may also be related to outcome (56).

One particular area that was raised in the Clinical Trials Workshop was the role of consumer perception in the amount and pattern of tobacco use. In prior clinical trials, subjective effects of PREPs have been assessed, which typically fall into consumer perception measures of liking, sensory effects (e.g., harshness, irritation, smoothness, and aftertaste), and withdrawal suppression/craving reduction. These data may indicate which products are likely to catch on with consumers. Knowing such information from laboratory or short-term studies may guide selections of a PREP for long-term clinical trial. However, in the context of a clinical trial, it is very difficult to directly assess consumer perception of a product prior to use and its effect on use, presumably because smokers are paid to participate and will use PREP as part of an implied contract, regardless of perception. The best approach to examine the influence of consumer perception on use is to experimentally manipulate the information provided to the consumer (e.g., presence or absence of relative toxicant level, presence or absence of health warnings, use in situations where the consumer cannot smoke, or to reduce exposure to toxicants). These manipulations can then be tied to use of the product. The research gaps that still need to be addressed are (*a*) how people's perception of the product affects its use in a trial; (*b*) how product perception affects abuse liability of the product; and (*c*) conversely, how use of the product affects consumer perception.

Summary and Main Conclusions

The need for PREP assessments is rapidly growing due to increased marketing of such products by tobacco companies worldwide. The primary goal for PREP assessment is to determine their effect on morbidity and mortality

relative to not having PREPs on the market. The main way to make this determination is to conduct long-term epidemiology studies and intervention trials. However, because of the length of time required to conduct these studies, the post-marketing nature of this type of assessment, and the large population base that is required, laboratory, short-term, and intermediate-term trials are the most expeditious, albeit with limitations, to infer potential harm or benefit from PREPs.

To move the science forward in PREP assessment, we need validated methods of product evaluation. Toward this end, we need to develop a battery of valid measures (i.e., subjective, behavioral biomarkers) that would uniformly be used across clinical trials. Furthermore, all clinical trials need to describe methods for recruitment; inclusion and exclusion criteria; and characteristics of subjects calling into a trial, enrolling in a trial, dropping out, and completing the trial. Finally, we need a systematic examination of the critical methodologic questions that were raised in this review, with the primary intent of determining the generalizability of our results in helping us understand both individual and population effects. Based on the review of the literature and the deliberations of the Workshop, the following is a summary of the recommendations that were made with the caveat that very few PREP method validation studies have been conducted:

The focus of switching studies should primarily be on examining the extent of exposure to PREP and patterns of use. To this end, the following questions would be important to address: (*a*) controlled use (specific instructions for amount of product use) versus *ad libitum* use (no specific instructions for amount of product use); (*b*) concurrent use of PREP with use of own brand (partial substitution for usual brand) versus sole use of PREP (complete substitution for usual brand); and (*c*) controlled use varying the dose of the PREP. These questions need to be addressed in laboratory, short-term, and intermediate-term studies.

The control groups should include usual brand and no smoking (with or without medicinal products). Another potential control group can be the usual brand user who decreases intake.

Short-term residential and nonresidential studies and intermediate-term studies have different strengths and limitations. The evaluation of PREPs needs to entail all these different designs for cross-validation purposes and to answer specific questions (toxicant exposure associated with the product versus how products are used in the naturalistic environment).

Durations of the trial will depend on the half-life of a biomarker and the stabilization of product use behavior. Longitudinal trials may be needed to determine stabilization of PREP use behavior.

Advertisements for the recruitment of subjects should state that the study involves testing a new product or a product that may reduce exposure or risk.

It is critical to assess sex, race/ethnicity, smoking history, degree of dependence, stage of change, socioeconomic status, and genetic makeup (e.g., rate of nicotine metabolism) of the study pool to allow comparisons to a general population of smokers and of smokers who are interested in trying PREPs.

It is not necessary to assess consumer perception of the product in the context of a clinical trial that assesses toxicity of PREPs. Examination of the relationship between consumer perception and behavior should be determined in an independent trial.

Abuse liability is an important component in assessing a PREP. The abuse potential for a PREP should be compared with usual brand at one end and with nicotine replacement therapies at the other end. The best methods to assess the abuse liability of a tobacco product and the predictive validity of these methods are unclear; however, multiple methods are likely to be necessary.

Disclosure of Potential Conflicts of Interest

D.K. Hatsukami: Commercial research grant, Nabi Biopharmaceuticals. P.G. Shields serves as an expert witness in tobacco litigation cases on behalf of plaintiffs.

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