Original Investigation

ITC “spit and butts” pilot study: The feasibility of collecting saliva and cigarette butt samples from smokers to evaluate policy

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Abstract

Introduction: Large-scale epidemiological surveys have frequently relied upon clinic-based sample collection to incorporate biological data, which can be costly and result in nonrepresentative data. Collecting samples in a nonclinical setting (i.e., through postal mail or at the subject's home) offers an alternative option that is minimally invasive and can be incorporated into large population-based studies.

Objectives: (a) To assess the feasibility of collecting biological data from a cohort of smokers in the International Tobacco Control (ITC) study, through the mail and in the home; (b) to examine whether participants are representative of the population under consideration; and (c) to evaluate how the added burden of providing biomarker samples might impact subsequent participation in a follow-up survey.

Methods: Participants were asked to provide a saliva sample and five cigarette butts from cigarettes smoked on a single day, using standardized procedures. Sample collection kits were mailed to a random sample of 400 daily cigarette smokers who were involved in the 2006 annual ITC Four Country (United Kingdom, United States, Canada, and Australia) telephone survey and agreed to participate in sample collection. A random sample of 179 daily smokers who participated in a face-to-face ITC survey in Mexico and Uruguay and agreed to participate in sample collection were also asked to provide samples.

Results: Samples were collected from 96% of invited participants in the face-to-face surveys and 52% of participants in the

telephone survey. The added burden of the sample collection did not reduce survey retention rates. Participants who initially agreed to participate in the sample collection were more likely to participate in the subsequent survey than participants who were not asked or declined to participate (odds ratio [OR] = 1.28; 95% CI = 1.01–1.62, $p = .021$). Further, those who provided samples were also more likely to participate in the subsequent survey than those who did not (OR = 2.78; 95% CI = 1.71–4.52, $p < .001$).

Discussion: Collecting saliva and cigarette butt samples from a group of smokers is feasible, yields a representative sample, and the added participant burden does not reduce subsequent survey response rates.

Introduction

The U.S. National Institutes of Health have promoted an interdisciplinary approach to health behavior research that integrates biological measures with behavioral sciences (McDade, Williams, and Snodgrass, 2007). A trend toward the acceptability of collecting biological samples in a nonclinical setting (National Research Council, 2001) offers health behavior researchers the opportunity to utilize biomarkers to supplement self-report survey data, traditionally used in behavioral research.

Biomarker data can be utilized in several different aspects of health behavioral research, such as confirming the accuracy of self-reported survey data, supplementing survey data, and providing a piece of more objective data to policy makers (McDade et al., 2007).

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Large-scale epidemiological surveys seeking to incorporate biological data might consider collecting samples in clinical settings. While this offers a wider range of data collection options and ensures high sample quality, this method can be costly and is limiting in terms of collecting data from more representative samples of a population. Collecting biological data through a postal mail or face-to-face in a nonclinical setting may offer an alternative option that is minimally invasive and can often be incorporated into large population-based studies. However, there are concerns about response rates, the quality of the samples, and the potential impact this added data collection effort might have on subsequent epidemiological data collection efforts.

The development of relatively feasible and minimally invasive methods for biological sample collection might be particularly useful in low- and middle-income countries, which may lack the laboratory resources to analyze the biological samples. However, logistical concerns associated with collecting biological samples from population-based research participants in low- and middle-income countries may be an obstacle to integrating biological data into self-report data (Boerma, Holt, and Black, 2001). Additionally, it is unknown how the added burden will impact response rates when a biological sample collection is introduced to a cohort of survey participants.

The data source for this study is the International Tobacco Control (ITC) Policy Evaluation Survey Project. The objective of the ITC Project was to evaluate the psychosocial and behavioral effects of national-level and subnational tobacco control policies (Fong et al., 2006).

The ITC Project is a prospective cohort study consisting of surveys in multiple countries. The first such survey was the ITC Four Country Survey, which has been conducted in Australia, Canada, the United Kingdom, and the United States since 2002. The ITC Project has since expanded so that at present there are or have been ITC surveys conducted in 17 countries, inhabited by 50% of the world's population, 60% of the world's smokers, and 70% of the world's tobacco users. The ITC survey asks participants questions regarding their smoking behavior, attempts at cessation, and attitudes and beliefs about tobacco products as well as questions pertaining to each of the demand reduction policies of the Framework Convention on Tobacco Control (e.g., warning labels, smoke-free laws, advertising/promotion, price/taxation) and a set of important psychosocial mediators and moderators of tobacco use and of cessation (e.g., perceived risk, quit intentions, time perspective).

Cohort members who are lost to follow-up are replaced with newly recruited participants to preserve the overall sample size from wave to wave. More recently, the ITC survey has been implemented utilizing a face-to-face survey methodology in countries where telephone survey methods may not have sufficient reach, including China, Malaysia, Thailand, Mexico, and Uruguay.

One question raised by the ITC investigative team was whether it was feasible to obtain biospecimens from a representative sample of survey participants in varying geographic locations and if this information could be used to help evaluate tobacco control policies. Previously published literature indi-

cates that research teams have been able to collect biospecimens using different methods of collection and incentives (Bauer et al., 2004; Etter, Perneger, and Ronchi, 1998; Kozlowski et al., 2002).

The principal objective of this pilot study was to develop methods of collecting saliva and cigarette butt samples from survey respondents in varying geographic locations. In this study, feasibility was defined as the ability to add a biomarker sample collection onto the existing survey protocol at a reasonable cost while achieving a reasonable rate of returned samples from a group of subjects who are representative of the population under consideration.

A secondary objective was to evaluate the potential impact of implementing a biomarker sample collection on future response rates of cohort members of the ITC study. The addition of this extra component presented an additional burden to participants, and it was important to understand better whether the additional burden placed on participants by this type of data collection would impact response rates to subsequent survey waves.

Methods

Two types of data collection were undertaken for this study—saliva samples and discarded cigarette butts. Saliva samples can be used to test for levels of exposure to nicotine and are a validated marker of smoke exposure (Benowitz, 1999). Spent, or smoked, cigarette butts can be examined for their tar staining patterns to obtain information about filter vent blocking and puff topography or how intensely the smoker smoked a particular cigarette (O'Connor, Stitt, & Kozlowski, 2005, 2007). The results of saliva and cigarette filter assays will be reported in a separate publication.

Samples were collected either through postal mail or face-to-face. The purpose of utilizing two different modalities of collection was not to compare the two methods. Rather, the method of collection chosen reflected the method of survey administration, which was based on the available resources in each country. For example, mail system reliability prohibited the use of a centralized mail-based collection method in Uruguay and Mexico. In these two countries, the face-to-face collection method was utilized to overcome these challenges.

Saliva sample collection was conducted via postal mail because of the geographic distribution of the ITC participants residing in Australia, Canada, the United Kingdom, and the United States. Saliva and cigarette butt sample kits were mailed to a random sample of 100 daily smokers of factory-made cigarettes from each of the four countries in the ITC survey. Briefly, smokers who were newly recruited to the ITC Four Country Project in 2006 were eligible to be selected for this study. Upon completing the phone survey, eligible subjects were asked whether or not they would be willing to participate in this saliva and cigarette butt collection pilot study. If a participant initially agreed to provide saliva and cigarette samples, their contact information was exported to a separate contact database that was used to mail the kits and track whether a participant had returned a kit. It was not noted whether a participant was not asked or declined to participate. The participants who initially agreed to take part in the saliva and cigarette butt collection were then mailed a letter to inform them that they would be receiving a kit to provide saliva and cigarette butt samples. Kits were mailed within 2 weeks of this initial contact letter. Participants who agreed to return completed kits by mail received the equivalent of \$15

USD. Those who did not initially send back a kit received reminder phone calls and another letter.

The kits for the mail-based portion of this study included the following: a cover letter, instructions for providing a sample, a Salivette cotton wool swab saliva sample collection device (Sarstedt, Inc., Nümbrecht, Germany, catalog number 51.1534), a plastic container in which the Salivette was to be sealed, a set of five cigarette butt collection containers, a secondary storage bag (e.g., ziplock type) marked with the biohazard symbol, informed consent forms, return mailing supplies, and an incentive equivalent to \$15 USD to compensate them for their time. Participants residing in Australia, Canada, and United States received a check, while the U.K. participants received a voucher that could be redeemed for merchandise at a retail chain having outlets throughout the country. The Salivette saliva sample collection device is a standardized means of collecting saliva samples. Subjects were to simply place the swab under their tongue for a few minutes, allowing the swab to absorb their saliva. For the cigarette butt samples, participants were instructed to provide the butt from the first cigarette smoked and any four others from the same day.

Because the investigative teams in Mexico and Uruguay conducted the ITC survey face-to-face, rather than via telephone, this method was also utilized to collect saliva and cigarette butt samples. The subjects were randomly sampled using a unique identification number prior to completing the interview. After completing the ITC survey, eligible participants were invited to participate in this onetime biological sample collection. Participants were asked to submit a saliva sample immediately following their participation in the survey using the same methods as described above. Interviewers then scheduled an appointment to return to the home of each participant to collect the cigarette butt samples a day or two after the survey. In Mexico, 79 individuals were sampled, while in Uruguay, 100 smokers were sampled (total sample size across six countries = 579).

Analysis

Data were analyzed using SPSS 15.0 (SPSS, Inc., Chicago, IL). Statistical analyses include descriptive statistics, multivariate logistic regression, and chi-square tests. The primary outcome was whether or not a respondent invited to provide saliva and butt samples did so. Upon sample receipt, the Salivettes were visually inspected to determine whether saliva had been absorbed into the swab. If the swab had absorbed liquid, it expanded noticeably from its unsaturated size. This was considered adequate. In order to perform the salivary cotinine assay, 0.5 ml of saliva is required. A complete returned sample collection kit was defined as the provision of a saliva sample and all five cigarette butt samples. Sample return rates are reported overall, by country and by mode of data collection using descriptive statistics and chi-square tests.

The characteristics of participants were also examined, including an analysis of responders and nonresponders, using a logistic regression model in which the outcome was whether the participant completed the biospecimen data collection request, and independent variables include country, mode of data collection, demographics, and past smoking behavior. Descriptive statistics and chi-square tests were utilized in order to characterize differences between participants who returned a sample collection kit and those who did not. This analysis was restricted to those who agreed to participate in the saliva and cigarette butt

collection. The purpose was to determine whether subjects who returned a kit were different from those who did not return a kit.

An additional outcome of interest was whether those who initially agreed to participate in the biological specimen data collection and returned a sample would be more or less likely to complete the subsequent ITC survey wave. Here, the key question we assessed was whether this additional burden would impact future survey response rates. The analyses used to measure this impact compared two groups: those who assumed the additional burden of providing saliva and cigarette butt samples and those who did not. The latter group includes subjects who were not asked to participate and those who were initially asked to participate but declined to do so.

Two multivariate logistic regression models were constructed in order to examine correlates of the likelihood of responding to the Wave 6 survey. Main predictor variables included whether a participant initially agreed to provide a biomarker sample (model 1) and whether a participant actually returned a sample collection kit (model 2). Covariates used in both models included country of residence, age, gender, and Heaviness of Smoking Index (HSI).

Results

Sample collection feasibility

At the conclusion of the sample collection phase of this pilot study, saliva and cigarette butts were collected from 96% of participants in Mexico ($n = 77$) and Uruguay ($n = 97$), where a face-to-face method of sample collection was utilized. However, in Australia, Canada, the United Kingdom, and the United States, where a mail-based method of collection was employed, 52% of participants who initially agreed to participate in the biomarker collection pilot actually returned a saliva and cigarette butt sample. These results are presented in Table 1.

In the four countries utilizing the mail-based method of saliva and cigarette butt sample collection, follow-up telephone calls were made to participants who failed to return a sample within 10 days after the kits were mailed out. As shown in Table 1, fewer than half of the participants (30% of all who were invited to participate) who returned a saliva and cigarette butt sample did so on their own, with no follow-up telephone call to prompt them. Of the 279 participants who were reminded to return their samples during the follow-up telephone call, 31% did so.

Of the 400 smokers sent data collection kits, 207 returned a completed kit and 181 provided usable saliva samples (87.4% of collected kits); results and demographic characteristics by country are shown in Table 1. Insufficient quantity of saliva in the Salivette was the primary reason for samples not being usable.

Sample representativeness from the ITC Four Country Survey

Descriptive statistics and chi-square tests were utilized to determine whether the population of smokers who initially agreed to participate in the biomarker collection was representative of the population under consideration (ITC Four Country Survey participants). Chi-square tests indicated no differences in demographic characteristics such as age ($p = .803$) and gender ($p = .228$) and measures of nicotine dependence such as HSI

Table 1. Description of biomarker pilot study methods

Country (<i>n</i> = 100)	Sample collection method	Incentive amount ^a	% Subjects returned samples overall	% Subjects returned samples without a follow-up telephone call	% Male	Mean age (years)	% TTF within 30 min
United States	Mail based	\$15 USD	56	29	68	49	73
United Kingdom	Mail based	£8 GBP “Boots” voucher	36	27	47	47	78
Canada	Mail based	\$15 CAD	60	24	56	50	75
Australia	Mail based	\$15 AUD	55	41	58	46	69
Mexico	Face-to-face	\$50 MXN telephone card	97	N/A	N/A	N/A	N/A
Uruguay	Face-to-face	\$200 UYU telephone card	97	N/A	N/A	N/A	N/A

Note. TTF = time to first cigarette after waking; N/A = not applicable.

^aIncentive amounts converted to USD: £8 GBP = ~\$16 USD, \$15 CAD = ~\$14 USD, \$15 AUD = ~\$13 USD, \$50 MXN = ~\$5 USD, and \$200 UYU = ~\$8 USD.

(*p* = .506) between participants and nonparticipants in the biomarker collection.

Factors significantly associated with nonresponse were U.K. residency and being aged 18–24 years (*p* < .002); nicotine dependence (cigarettes per day or time to first cigarette) was not significantly associated with kit return. When examined in a multivariate logistic regression model, non-U.K. residents were 2.61 times more likely (95% *CI* = 1.58–4.34) to have returned a kit. Because of the lower response among U.K. participants, U.K. nonresponders were compared with nonresponders residing in the other three countries in order to characterize any differences in the level of dependence among the groups of smokers who did not return a sample collection kit. No differences were observed in measures of cigarettes per day and time to first cigarette after waking, suggesting that the U.K. nonresponders were not more or less nicotine dependent than nonresponders in the other countries.

Impact on response to future surveys from the ITC Four Country Survey

Results are presented in Figures 1 and 2. Of the 2,635 participants who completed the ITC Wave 5 replenishment survey, 400 subjects (396 of whom provided usable survey data) initially agreed to participate in the biomarker sample collection. Of those who initially agreed to participate in the collection, 67.7% responded to the Wave 6 survey. Of those who did not initially agree or were not asked to participate in the biomarker sample collection, 61.6% completed a Wave 6 interview (*p* = .021). Among participants who initially agreed to participate in the biomarker sample collection, 79.8% of those who returned a kit (*n* = 207) participated in the Wave 6 survey. This is in contrast to the 54.3% of participants who initially agreed to participate in the biomarker collection pilot but failed to return a collection kit (*n* = 188), yet completed a Wave 6 interview (*p* < .001).

Participants who initially agreed to receive a biomarker sample collection kit were more likely to have participated in the Wave 6 survey than those who did not agree or were not asked to provide a biomarker sample (*OR* = 1.28; 95% *CI* = 1.01–1.62, *p* = .021). Additionally, participants who returned a sample collection kit were more likely to participate in the subsequent ITC survey than participants who initially agreed to but did not return a sample (*OR* = 2.78; 95% *CI* = 1.71–4.52, *p* < .001).

Discussion

The results observed in this study suggest that collecting biomarker samples from population-based samples is feasible, able to yield a representative sample, and the added burden placed on the participant does not reduce subsequent survey response rates.

One limitation was that fewer follow-up calls were made to participants residing in Australia and the United Kingdom than to those residing in Canada and the United States. Follow-up calls were made from a centralized location in the United States, and time differences restricted the timeframe in which follow-up telephone calls could be made. Future biomarker collections may want to consider utilizing a more decentralized approach to the kit follow-up process, if there is staff and infrastructure available in other participating countries.

Figure 1. % Responding to Wave 6 survey when initially agreeing to provide a biomarker sample. Chi-square test comparing response to the Wave 6 survey among subjects asked to provide a biomarker sample and subjects who were not asked ($p = .021$). When adjusted for country of residence, age, gender, and Heaviness of Smoking Index, those who were asked to provide a biomarker sample were 1.28 times (odds ratio) more likely (95% CI = 1.01–1.62) to have participated in the Wave 6 survey.

Although the reasons for the lower response rate observed among the U.K. participants remain unclear, it might be speculated that a higher response rate may have been observed in the United Kingdom if more follow-up callbacks could have been made during a wider range of hours. Of the 73% of U.K. participants who required a follow-up telephone call, only 12% returned a sample collection kit. A similar trend was observed in Australia, where just 10% of the participants who were followed up via telephone returned a kit. In Canada, where the range of hours available to place follow-up telephone calls was considerably wider, 60% of the participants who received a telephone call returned a saliva and cigarette butt sample kit.

The follow-up telephone calls had similar rates of success in Australia and the United Kingdom, while the final return rates were drastically different (55% in Australia vs. 36% in the United Kingdom). It can be hypothesized that the difference in the percentage of participants who returned sample collection kits was due to the type of incentive offered to participants in each of these countries. A voucher to a specific store may not have been as appealing to prospective participants as the offer of \$15 cash.

There is some uncertainty as to the reasons why the overall response rate in the United Kingdom was lower than that in the other three countries. The limited follow-up telephone call timeframe and the offer of a voucher as opposed to cash are two plausible reasons for this difference but are by no means definitive answers. Comparisons between participants who returned a sample collection kit and those who did not (between country and within country) indicated no significant differences between the two groups. Although we have explored several hypotheses as to why the overall response rate was lower in the United Kingdom, the reasons behind the low response rate might be a combination of these factors or some entirely different reason that remains unclear.

The addition of the biomarker sample collection presented an additional burden to participants, and it was important to be certain that this type of data collection would not decrease response rates to subsequent telephone survey waves. Our findings suggest that being asked to participate in the biomarker sample collection did not have a negative impact on the subsequent survey wave. A limitation of this aspect of the study is that

Figure 2. % Responding to Wave 6 survey when returning a biomarker sample. Chi-square test comparing response to the Wave 6 survey between subjects who returned a biomarker sample and subjects who did not return a sample ($p < .001$). When adjusted for country of residence, age, gender, and Heaviness of Smoking Index, those who returned a biomarker sample were 2.78 (odds ratio) times more likely (95% CI = 1.71–4.52) to have participated in the Wave 6 survey.

we cannot completely measure the impact of how simply being asked to participate in this biomarker sample collection might affect response rates to future surveys because of limitations in how the data were collected. It was not recorded whether a survey participant was initially asked to provide a sample but declined to do so. Rather, our main goal in this analysis compared those who initially agreed to provide a biomarker sample with those who were not asked and those who were asked but declined.

Further, participants who actually returned a sample collection kit were nearly three times more likely, when adjusted for demographic characteristics and smoking behavior, to have responded to the next ITC telephone survey wave than participants who initially agreed to participate in the biomarker collection but failed to return a sample collection kit. A similar trend was observed in Mexico and Uruguay. In terms of the face-to-face sample collection in Mexico, 94.8% of subjects who provided a saliva sample participated in the subsequent survey. This is in contrast with 72.6% of subjects who did not participate in the biomarker collection. In Uruguay, where the follow-up survey occurred 2 years after the biomarker collection, 76.2% of those who provided a saliva sample participated in the follow-up survey compared with 56.6% of those who did not provide a saliva sample.

These results strengthen the case that collecting biomarker samples from population-based samples is, indeed, feasible and that the added participant burden will not negatively influence response rates to the ITC telephone survey. In fact, among a subset, the additional data collection may engage them sufficiently to retain them in the ongoing cohort. As survey response rates continue to decline, cohort retention will become more crucial. The observation that additional relevant data collection of a different modality (which one would assume increases burden and would decrease participation) appears to enhance retention deserves further study. If simply asking for participation in an additional study component leads to continued participation in future telephone survey, it might be concluded that contact with the participants among survey waves may increase response rates to successive surveys. An area warranting further exploration is the development of methods to keep participants interested in the survey and willing to participate between telephone interview waves. Future studies should consider an analysis of how simply asking a survey respondent to participate in an additional study component might affect survey participation.

In conclusion, the results of this pilot study demonstrate that collecting biomarker samples from a representative sample of a population of survey participants is, indeed, feasible and able to yield a representative sample. Additionally, the inclusion of this type of sample collection does not seem to be an unreasonable burden on survey participants. The results presented here indicate that the additional burden of the saliva and cigarette butt collection did not reduce response rates to a subsequent telephone survey. It is unclear whether these results will generalize to the collection of other types of biomarker samples and other populations. Researchers should use discretion with regard to the generalizability of these findings, given the small sample size and the fact that this was a pilot study among a population of smokers.

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Declaration of Interests

None declared.

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