Chapter 3
Assessment of Exposure to Secondhand Smoke

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Introduction

This chapter provides a review of key factors that determine exposures of people to secondhand smoke in indoor environments. The discussion describes (1) the dynamic movement of secondhand smoke throughout indoor environments, (2) the factors that determine secondhand smoke concentrations in these environments, (3) the atmospheric markers of secondhand smoke that are measured to assess concentrations, (4) the biomarkers that are measured to assess doses of tobacco smoke components, and (5) the models that can be used to describe patterns of human exposures. Chapter 4 (Prevalence of Exposure to Secondhand Smoke) reports on findings of studies on exposures to secondhand smoke that applied these methods with a focus on measurements of nicotine in the air and cotinine in biologic materials. The validity of nicotine as a marker for secondhand smoke concentrations supports the use of cotinine, a principal metabolite of nicotine, as an exposure biomarker.

As described earlier, the term secondhand smoke refers to a complex mixture of particulate (or solid) and gaseous components. The characteristics of secondhand smoke change over time, particularly the components of sidestream smoke that the smoldering cigarette releases. Sidestream smoke dilutes quickly and changes as the particles release volatile compounds and change in size and composition as they age. Although few studies have made measurements, available data indicate that the estimated median aerodynamic diameter of secondhand smoke particles is 0.4 micrometers (μm), a size range where particles tend to remain suspended in the air unless removed by diffusion to or impaction with a surface, or by air cleaning (Hiller et al. 1982; Jenkins et al. 2000).

The composition of secondhand smoke was addressed in the 1986 report of the Surgeon General, *The Health Consequences of Involuntary Smoking* (U.S. Department of Health and Human Services [USDHHS] 1986), and was the focus of a comprehensive monograph first published in 1992 and updated in 2000 (Guerin et al. 1992; Jenkins et al. 2000). The 1986 report commented on the richness of secondhand smoke as a mixture and its inherent variability over time and space as it moves through the air (USDHHS 1986). Nonetheless, the report concluded that secondhand smoke and mainstream smoke were qualitatively similar, a conclusion that subsequent research supports (U.S. Environmental Protection Agency [USEPA] 1992; Scientific Committee on Tobacco and Health 1998; International Agency for Research on Cancer [IARC] 2004).

People are exposed to secondhand smoke in multiple places where they spend varying amounts of time. The term "microenvironment" refers to places that have a fairly uniform concentration of a mixture of pollutants across the time that is spent there (National Research Council [NRC] 1991; Klepeis 1999a). In the microenvironmental model, total human exposure to an atmospheric contaminant, such as secondhand smoke, represents the time-integrated sum of the exposures in the multiple microenvironments where time is spent. The source of secondhand smoke—the burning cigarette—produces the resulting concentrations of secondhand smoke in the air of places where people spend time. The concentration depends on the intensity of smoking, dilution by ventilation, and other processes that remove smoke from the air. The consequent exposures lead ultimately to doses of secondhand smoke components that reach and harm target organs and manifest as adverse health effects. This conceptual framework, which is central to this chapter, makes clear distinctions between cigarette smoking as the source, secondhand smoke concentrations in the air (the amount of material present per unit volume), exposures to secondhand smoke (the time spent in contact with secondhand smoke at various concentrations), and the doses from secondhand smoke exposure (the amount of material entering the body). The strength of the source—cigarette smoking—depends on the number of smokers and the rate at which they are smoking. Total human exposure can be estimated by measuring secondhand smoke concentrations in key microenvironments and assessing the time spent in those environments. Concentrations are also determined by aspects of the design and operation of a building (NRC 1986, 1991).

The mass balance model is a conceptual approach that provides a framework for how the design and operation of a building may affect secondhand smoke concentrations within the building (Ott 1999). In this model, which is considered in more detail later in this chapter (see "Exposure Models"), the concentration of indoor air contaminants (such as secondhand smoke) is a function of the strength of the source(s) generating the contaminant, the dilution of the contaminant by the exchange of outdoor with indoor air, and the rate of removal of the contaminant by air cleaning and other processes.

Building Designs and Operations

Determinants of Secondhand Smoke Concentrations

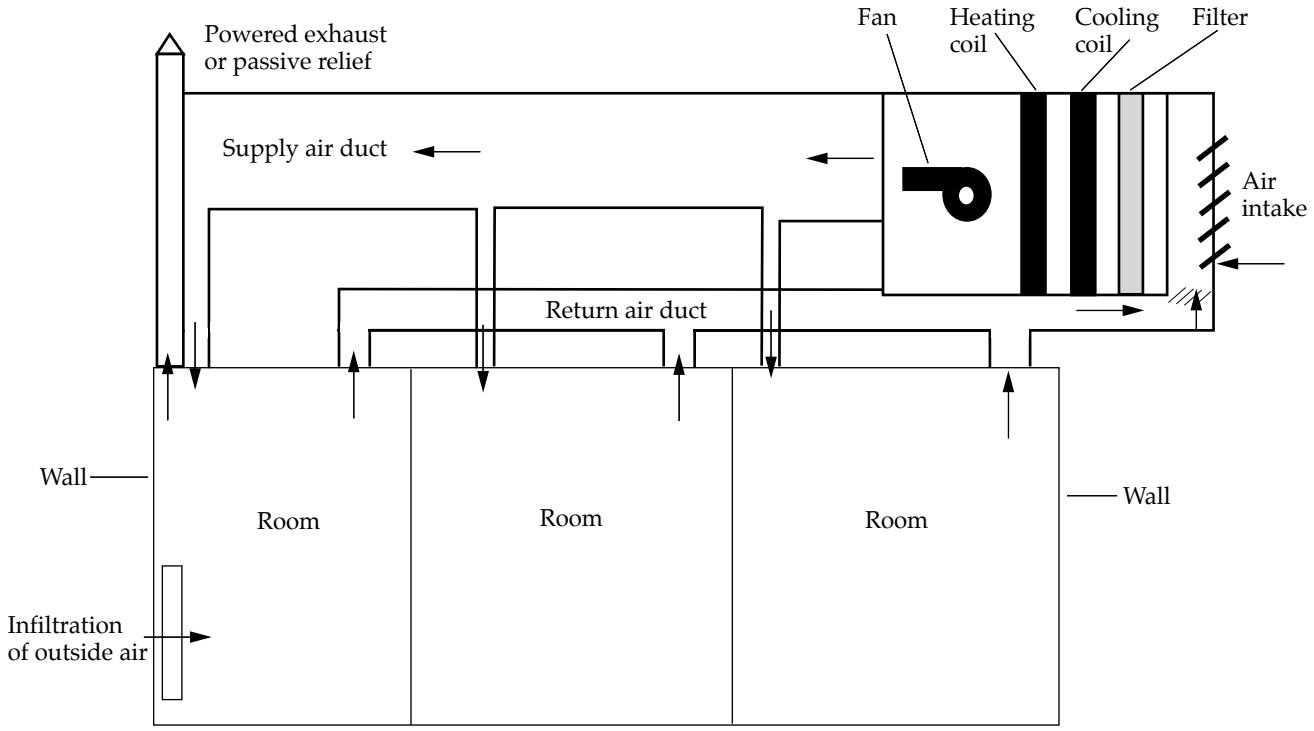
When people are exposed to secondhand smoke in indoor environments, the concentrations to which they are exposed depend not only on the number of cigarettes smoked, which determines the strength of the source, but on how air moves through buildings and at what rate indoor air is exchanged with outdoor air. The exchange of indoor with outdoor air is referred to as ventilation (American Society of Heating, Refrigerating and Air-Conditioning Engineers [ASHRAE] 1989). In general, the concentration of an indoor contaminant in a building or in a space within a building depends on the volume of the space and the rate at which the contaminant is generated and then removed. The removal may be by ventilation, air cleaning, or other processes such as chemical reactions or adsorption onto surfaces. This set of relationships is referred to as the mass balance model. It implies that concentrations of secondhand smoke components in a space (1) increase as the number of cigarettes smoked increases, (2) decrease with an increase in ventilation, and (3) decrease in proportion to the rate of cleaning or removal of secondhand smoke components from the air (Ott 1999). The cleaning or removal processes might include active air cleaning with a device, the naturally occurring passive deposition of particles onto surfaces, and the adsorption of gases onto materials.

The factors in the mass balance model vary across different kinds of buildings. Buildings can be ventilated using natural or mechanical methods. Air can be supplied naturally through windows, louvers, and leakages through building envelopes; air is supplied mechanically through a heating, ventilating, and air conditioning (HVAC) system that usually includes fans, duct work, and a system for delivering air in a controlled manner throughout a building (Figure 3.1). In most homes, ventilation occurs by a naturally occurring exchange of indoor with outdoor air. Commercial and public buildings generally have HVAC systems that move air through buildings to accomplish the exchange of indoor with outdoor air. Important considerations are variations in the range of surfaces and their characteristics across different kinds of buildings and microenvironments. For example, most HVAC systems incorporate a component for air cleaning that typically removes large particles but not the smaller particles or the gases found in secondhand smoke. The central air cleaning systems in homes and in many commercial buildings generally are not designed to remove smaller particles or gases (Spengler 1999).

Heating, Ventilating, and Air Conditioning Systems

For modern public and commercial buildings, often with sealed windows, air ventilation is required to provide a safe, functional, and comfortable environment for the occupants, and is defined as "outside air" delivered to or brought indoors. For many types of indoor environments, mechanical ventilation systems are used to control contaminant concentrations and to meet the comfort needs of occupants. Such systems are almost always used in hospitals, large office buildings, theaters, hospitality venues, schools, and many other larger buildings. This discussion addresses how these systems affect secondhand smoke concentrations in indoor environments and focuses on public and commercial buildings where HVAC units are generally in place. Mechanical systems are intended to provide thermally conditioned air, dissipate thermal loads, and dilute contaminants (Bearg 2001). These systems can also be used to maintain pressure differentials between areas when air is extracted and exhausted from specific spaces, or to clean and recirculate the air using filters, catalytic converters, and various sorbent beds. The efficiencies and costs for an entire ventilation system vary depending on specific requirements and settings (Liddament 2001). Although mechanical systems are widely used for general ventilation, their potential use as a control strategy for secondhand smoke requires a detailed understanding of the constituents to be controlled, the air distribution patterns within structures, the air cleaning or extraction techniques, and the requirements for ongoing operation and maintenance (Ludwig 2001). If not properly designed and maintained, mechanical systems can increase the risk of exposures by distributing pollutants (including secondhand smoke) throughout the building, by direct recirculation, or by poor pressure control.

Figure 3.1 Schematic of a typical air handling unit



Source: U.S. Environmental Protection Agency 1994, with modifications.

Complex and dynamic processes affect the characteristics and concentrations of secondhand smoke. As a foundation for considering ventilation systems commonly found in buildings, here is a description of the transport and fate of particles and gases released from a burning cigarette. In still air, the smoke plume from a cigarette is often observed rising intact as high as several meters above the burning tip. If plume gases remain concentrated, they are buoyant and have a temperature several degrees higher than the surrounding room air temperature. If the room air is not still, as in buildings with mechanical air handling systems, or if people move within the space, there will be some mixing that breaks up the plume and disperses "pockets" of smoke throughout the air space (Klepeis 1999b). Concentrations of secondhand smoke components are then reduced and, as the smoke spreads and ages, its components change as a result of condensation, evaporation, coagulation, and deposition to surfaces. The characteristics of secondhand smoke within a particular building thus depend, to an extent, on chemical and physical characteristics of spaces that vary among buildings. Volatile components such as nicotine are adsorbed and degassed by materials. As a consequence, the smell of cigarettes emanates from clothing, carpets, air conditioners, and other surfaces without the presence of active smoking, as previously deposited or adsorbed material is re-emitted by air currents (Klepeis 1999b).

Although interactions in the air and at surfaces modify the secondhand smoke mixture, under most circumstances concentrations within the original space will depend strongly on an exchange of air in the space with less contaminated air (Spengler 1999). Mechanically delivered air disperses secondhand smoke constituents through mixing (turbulence) and dilutes secondhand smoke by supplying less contaminated air. Generally, mechanical mixing is significantly more effective in reducing concentrations from a "point source" of pollution in a room, such as a burning cigarette, than is diffusion alone in still air. Air exchange and surface removal processes act together to lower secondhand smoke concentrations. Surface removal is enhanced if air is forced through

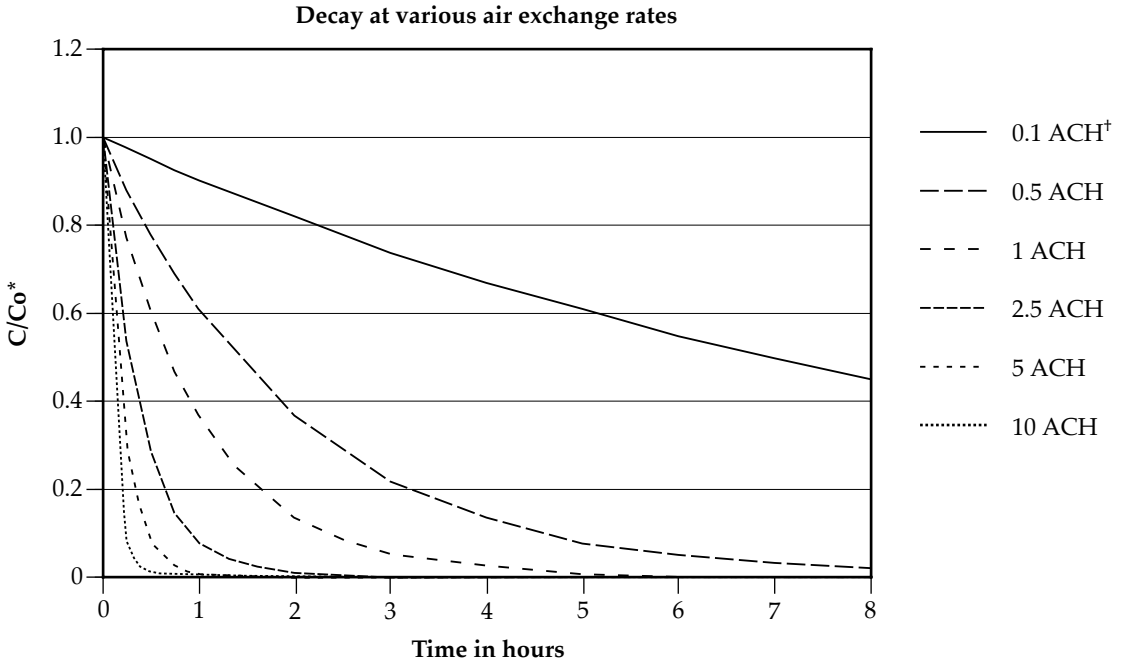
an air cleaning device and delivered back to the room with a reduced secondhand smoke concentration (McDonald and Ouyang 2001).

Building Ventilation Control

Mechanical HVAC systems that heat, ventilate, and air-condition indoor spaces achieve controlled building ventilation (Spengler 1999). The HVAC systems in buildings are composed of air handling units (AHUs) of various sizes and complexities that filter and condition air supplied to the building with varying degrees of effectiveness, depending upon need, design, and maintenance. Components of AHUs typically include fans, filters, cooling coils, and heat exchangers. Air ventilated by air conditioning (i.e., mechanical cooling) can be ducted to separate areas within a building and removed with an air return system that recirculates and/or exhausts the air. In Figure 3.1, a schematic demonstrates a typical AHU configured for general ventilation and pressure relationship control (USEPA 1994).

Three major categories are used for airborne contaminant control: general or dilution ventilation, displacement ventilation, and local exhaust ventilation. General or dilution ventilation requires mixing large volumes of outdoor air with room air. Although this ventilation system is the most commonly used method in buildings today for thermal comfort, it is not very efficient for controlling contaminant emissions from human activities such as smoking. Its effectiveness is highly dependent upon the number and location of emission sources (the smokers), the volume of air supply to the room, the capacity of materials and surfaces to remove various constituents of secondhand smoke, and the mixing efficiency of the room. Figure 3.2 demonstrates that the term "air exchange rate," when applied to dilution ventilation, is a misnomer. Mixing the supply air within the zone served by the AHU is often not uniform or complete. Even for a well-mixed space, one air change per hour (ACH) means that only 63.2 percent of the original air, including the corresponding airborne contaminants, is removed in one hour. So even though an amount

Figure 3.2 Anticipated changes in concentrations of airborne materials for various air exchange rates



*C/Co = Fraction of original concentration of contaminant at time t.
†ACH = Air changes per hour.

of air equivalent to the volume of the room is introduced during one hour, it does not completely replace all of the air occupying the space previously. Short-circuiting or moving air directly from inlets to the exhaust without mixing, obstructions to supply and exhaust air, and thermal gradients can reduce the mixing efficiency to much less than the theoretical limit. Thus, an air exchange rate greater than that made with simple calculations based on the volume of the space may be required to effect a meaningful reduction in airborne concentrations of various contaminants (Liddament 2001). Simple mass balance and volumetric calculations assume perfect mixing, no sink effects (the adsorption and possible re-emission of pollutants by materials acting as "sinks" [Sparks 2001]), and constant emission sources; these conditions generally are not met in real-world indoor environments. Any occupant of a space, particularly a space near a pollution source, may be exposed to much higher concentrations than estimated for the overall area.

Displacement ventilation, which is also referred to as piston or plug flow, conditions the space and removes contaminants by admitting air at one location and "sweeping" it across the space before exhausting it at the opposite "face." This design often uses low-velocity grills at or near floor level to admit cool supply air into the space that is then exhausted at ceiling level. For maximum effectiveness, displacement ventilation requires a more or less uniform and unidirectional flow. This flow structure might easily be disrupted by large numbers of people moving about a space, or through the use of ceiling fans or supplementary ventilation systems. Displacement ventilation often uses specific characteristics of the contaminant to aid in its capture. For example, a heated plume from a computer, copier, or cigarette develops convective (vertical) flows. If the displacement air is also moving vertically from floor to ceiling, pollutants and excess heat can be captured, treated, or exhausted from the ceiling. With this strategy, however, contaminants on their way to the exhaust stage can still pass through the breathing zones of both smokers and nonsmokers. Furthermore, vertical flows may be disrupted by furniture that is in the space, thus limiting the effectiveness of displacement ventilation.

Local exhaust ventilation extracts the air around a specific point source. It has been used for many decades to effectively control a variety of contaminants from specific activities or processes, often in industrial settings. Its effectiveness relies upon strict compliance with control measures that can include source enclosure, high air exhaust rates, and direct ducting to the outdoors that minimizes entrainment into outdoor air intakes. Restrictive compliance requirements limit its application to secondhand smoke in general indoor environments, except in separately exhausted smoking enclosures.

Operation of Ventilation Systems

Ventilation requirements for spaces such as office buildings, classrooms, and various hospitality venues are expressed as the volume of outside air per unit of time (e.g., liters per second, cubic feet per minute) per person, and/or volume flow rates of outdoor air per square foot of the area of the building. ASHRAE (1999) included the latter criterion in the revised Standard 62-1999 as a result of the recognition that air pollutants are also released by building sources—building materials, furnishings, and the HVAC equipment itself—and that to protect the occupants, ventilation standards should also apply to these sources as well as to the occupants. ASHRAE develops standards to guide building designs and operations that often become part of municipal codes (Chapter 10, Control of Secondhand Smoke Exposure). Consequently, ASHRAE standards are considered relevant to the control of secondhand smoke in the United States (Table 3.1). Building ventilation codes generally specify the total amount of air as well as a minimum percentage of outdoor air that should be supplied to occupied spaces. Minimum amounts between 10 and 20 percent are often specified, but in practice, outdoor air delivery into a building may vary from 0 to 100 percent over time. The variation depends on the design requirements of the space and operational characteristics of the ventilation system.

Ventilation systems are often quite complex and have multiple components. Controls are in place to modulate the air intake louvers, airflow, air temperature, and sometimes the humidity to meet specified thermal conditions (ASHRAE 1999). These control systems often consist of combinations of sensors, signal processors, computerized controllers, switches, dampers, valves, relays, and motors. The operating strategies for ventilation systems can have a major impact on the control of secondhand smoke within buildings. For example, many systems operate on economizer cycles that use the cooling or heating capacity of the outside air. During the economizer phase, the outside louvers open. Often, depending on the climate and season, a temperature range (generally between 50° and 65° F) will completely open the outside dampers (Spengler 1999; Bearg 2001). If ambient conditions become too warm and humid, the outside air vents

Table 3.1 Outdoor air requirements for ventilation*

Application	Estimated maximum† occupancy per 1,000 ft²‡ or 100 m²§	Outdoor air requirements		Comments
		cf/m/person∆	cf/m/ft²	
Food and beverage services				
Dining rooms	70	20	NR¶	
Cafeteria, fast food	100	20	NR	
Bars, cocktail lounges	100	30	NR	Supplementary smoke-removal equipment may be required
Kitchen (cooking)	20	15	NR	Make-up air for hood exhaust may require more ventilating air; the sum of the outdoor air and transfer air of acceptable quality from adjacent spaces shall be sufficient to provide an exhaust rate of not less than 1.5 cf/m/ft² (7.5 liters/second/m²)
Hotels, motels, resorts, dormitories			cf/m/room	
Bedrooms	NR	NR	30	
Lobbies	30	15	NR	
Conference rooms	50	20	NR	
Casinos	120	30	NR	Supplementary smoke-removal equipment may be required
Offices				
Office space	NR	20	NR	Some office equipment may require local exhaust
Public spaces				
Smoking lounge	70	60	NR	Normally supplied by transfer air; local mechanical exhaust with no recirculation is recommended

*This table prescribes supply rates of acceptable outdoor air required for acceptable indoor air quality. These values have been chosen to dilute human bioeffluents and other contaminants with adequate margins of safety and to account for health variations and varied activity levels among people.
†Net occupiable space.
‡ft² = Square feet.
§m² = Square meters.
∆cf/m/person = Cubic feet per minute per person.
¶NR = Data were not reported.
Source: American Society of Heating, Refrigerating and Air-Conditioning Engineers Standard 62-1999, Table 2.1 (1999).

will return to minimum or closed settings. To protect coils from freezing or to minimize heating, outside air vents might be closed or set at minimum openings during colder temperatures. Thus, contaminants such as secondhand smoke that are generated within a building are often subject to varying amounts of dilution air, and building occupants may face indoor air quality that varies during a day or over longer periods of time (Spengler 1999).

Most large, modern buildings use a building automation system (BAS) to provide direct digital control of ventilation through a central computer. Planned into the BAS is a sequence of operations for the HVAC system (USEPA 1998). Knowledge of routine activities related to building occupancy allow engineers to program HVAC systems through the central BAS to improve comfort and optimize energy efficiency.

However, a BAS is generally not programmed to control indoor air pollutants such as secondhand smoke.

Mechanical air handling systems exchange indoor air with outside air by pressure-driven flows through windows, doors, and cracks. Some buildings are not designed or constructed to be airtight; an estimated 40 percent of commercial buildings have operable windows, and natural ventilation is more common in older and smaller buildings (Liddament 2001). Pressure differentials across the building envelope are caused by wind and by indoor and outdoor temperature differences. The wind that flows around a building creates static positive pressures as well as negative pressures in the wake flow that is downstream of objects. Pressure differences across openings can force air into or out of a building. The HVAC system of pressurized ducts and building exhaust fans also creates an air exchange. Plumbing and electrical chases, elevator shafts, leaky air ducts, and cracks and openings between floors can become unplanned pathways for pressure-driven internal flows. Thus, contaminants such as secondhand smoke are not always controlled by HVAC airflows alone, and the HVAC ducts may transport and distribute secondhand smoke-contaminated air. Entrainment from doors, window cracks, or loading docks can bring tobacco smoke back into a building even when smokers are restricted to smoking outdoors. Even within buildings, secondhand smoke can move along unplanned or uncontrolled pathways to annoy and irritate occupants in other rooms or even on other floors far removed from the smoking areas.

Residential Ventilation

There are more than 100 million residential units in the United States. The most common types are single family (73 percent) followed by multifamily structures that include both low-rise and high-rise apartments (21 percent) and mobile homes (6 percent). The United States has a high rate of owner-occupied households (67 percent); 33 percent of households live in rental units (Diamond 2001).

The age and size of housing vary around the country. In general, older homes are smaller (<2,000 square feet of conditioned space) and are more common in the Northeast and Midwest. The average apartment unit is about half that size (approximately 1,000 square feet). Three million Americans live in public housing, most of which are two-bedroom units built in the 1950s and 1960s; the total size is typically 500 to 600 square feet (Diamond 2001). The south and southwestern regions of the United States continue to be the fastest growing areas and lead in new housing construction (Joint Center for Housing Studies of Harvard University 2002). Despite a decrease in the size of households, the size of single-family homes has increased with more square feet per person. Homes built in 1995 were 17 percent larger than those built just a decade earlier. During a 15-year period, new apartment units increased in average floor space by almost 10 percent (Diamond 2001).

Most houses and apartments have heating systems. Besides the size of the unit (i.e., volume), the type of heating, cooling, and exhaust system is an important factor in the dispersion, dilution, and removal of indoor-generated secondhand smoke across a room or throughout a residence. More than 50 percent of U.S. residences have central warm air furnaces. These systems include fan-forced directed air distributed to rooms with a gravity or ducted return back to the heat exchange unit of the furnace. Gravitational settling is not intended to remove the smaller particles found in secondhand smoke, nor is it efficient at removing them. Filters upstream of the blower serve to protect the mechanical parts from objects and large particles, but these filters also fail to remove the smaller secondhand smoke particles and gases.

Air conditioning can affect the distribution and concentration of secondhand smoke. Air conditioning systems are common in U.S. residences, including apartments. According to the Residential Energy Conservation Survey (U.S. Department of Energy 1999), 48 percent of residences were equipped with central air conditioning and 27 percent had window units. Forty-seven percent of the respondents with central systems versus only 18 percent with window units reported using their air conditioning "quite a bit" or "just about all summer." Similar to forced warm air mechanical systems, central air-cooling systems can rapidly mix secondhand smoke throughout the conditioned space. Doors and windows are generally closed when the air conditioner is in use and the system is usually set to recirculate the indoor air. These closed conditions tend to reduce the dilution of secondhand smoke.

Wallace (1996) comprehensively reviewed indoor air particle concentrations and sources and quantified the effect of air conditioning on the concentration of secondhand smoke. His review included studies that measured indoor and outdoor particulate matter 2.5 ($PM_{2.5}$) concentrations across six U.S. communities (Dockery and Spengler 1981; Spengler et al. 1981; Spengler and Thurston 1983; Letz et al. 1984; Neas et

al. 1994). Estimated concentrations of fine particles were 30 micrograms per cubic meter ($\mu g/m^3$) higher in homes with smokers than in homes without smokers. According to Wallace (1996), "A mass balance model was used to estimate the impact of cigarette smoking on indoor particles. Long-term mean infiltration of outdoor $PM_{2.5}$ was estimated to be 70% for homes without air conditioners, but only 30% for homes with air conditioners. An estimate of 0.88 $\mu g/m^3$ per cigarette (24-h average) was made for homes without air conditioning, while in homes with air conditioning the estimate increased to 1.23 $\mu g/m^3$ per cigarette" (p. 100). The greater estimate for air conditioning is consistent with lowered air exchange rates while the air conditioning is operating, and is supported by a 1994 study (Suh et al. 1994).

Air exchange rates in homes are usually determined by one of two methods: blower door pressurization or tracer gases. Blower door pressurization tests identify air leakage areas that are then used to estimate air exchange rates. Sherman and Matson (1997), who modeled the results of blower door tests, found that a typical single-family house constructed before 1990 has an estimated air exchange rate of 1.0 ACH. Homes built to meet more energy efficient building codes have estimated rates of 0.5 ACH.

Tracer gases are emitted into a home and measured over time to calculate short-term (decay rate) or long-term (mass balance method) air exchange rates. Murray and Burmaster (1995) examined the Brookhaven National Laboratory tracer gas data that included almost 3,000 households. The analysis derived best-fit, log-normal distributions from data classified by four regions or by heating degree days (a measurement used to relate a day's temperature to the demand for fuel to heat buildings: a 65° average daily temperature = the number of heating degree days), and by the four seasons. In general, air exchange rates are higher for homes that are in warmer climates. Air exchange rates across all regions are higher during the summer months followed by spring, fall, and winter. The summer mean air exchange rate is 1.5 h^{-1} (air changes per hour) versus 0.41 h^{-1} for the fall.

Other characteristics of air exchange rates derived from blower door and tracer gas methods indicate that apartment units and multifamily structures with shared interior walls have less external surface area, less unplanned air leakage, and typically lower air exchange rates compared with single-family detached houses.

Conclusions

1. Current heating, ventilating, and air conditioning systems alone cannot control exposure to secondhand smoke.

2. The operation of a heating, ventilating, and air conditioning system can distribute secondhand smoke throughout a building.

Implications

These conclusions suggest that control strategies for indoor exposure to secondhand smoke cannot use approaches based on HVAC system design and operation. The benefits from HVAC systems include a number of critical functions that help to maintain a healthful and comfortable indoor environment. This review of their functioning shows, however, that current HVAC systems cannot fully control exposures to secondhand smoke unless a complete smoking ban is enforced. Furthermore, unless carefully controlled, HVAC operations can distribute air that has been contaminated with secondhand smoke throughout a building. Simple predictions cannot be made about the consequences of these operations because they vary with the building and with the HVAC characteristics. However, to develop models that assess the effects of indoor secondhand tobacco smoke exposures, it is necessary to first develop an understanding of HVAC systems and their effectiveness in a particular structure. However, this review indicates that a complete ban on indoor smoking is the most efficient and effective approach to control exposures to secondhand smoke. Additional implications of these findings are considered in Chapter 10, Control of Secondhand Smoke Exposure.

Atmospheric Markers of Secondhand Smoke

Concepts and Interpretations of Exposure Markers

Secondhand smoke is a dynamic mixture that contains thousands of compounds in its vapor and particle phases. Some of these components are specific to secondhand smoke, such as nicotine, but others have additional sources and are not specific to secondhand smoke, as in the case of carbon monoxide (CO). Some of the more specific markers can be useful indicators of secondhand smoke concentrations, but no particular marker will be predictive of the full range of risks from exposures to secondhand smoke. Additionally, some components of particular interest for disease risk, such as the tobacco-specific nitrosamines, are not easily measured at typical indoor air concentrations (Hecht 1999). Nonetheless, some components of secondhand smoke can be quantified in indoor air. This quantification enables researchers to estimate exposures to secondhand smoke for research purposes and for tracking population exposures. In 1986, the NRC report on involuntary smoking proposed useful atmospheric markers that are believed to be unique to tobacco smoke or that are believed to have cigarette smoking as their primary source in most environments; the mass that is emitted is believed to be similar across cigarette brands (NRC 1986). Subsequent studies have evaluated some of the markers used to detect secondhand smoke in indoor environments (Guerin et al. 1992; Daisey 1999; Jenkins et al. 2000).

Researchers need sensitive and specific markers of secondhand smoke for exposure surveillance and potentially for enforcement of regulations. For research and for population risk assessments, measurements of marker compounds can be used with microenvironmental models to estimate exposures to secondhand smoke (Jaakkola and Samet 1999). Researchers can also estimate the relative contributions of different environments to these exposures and the potential consequences of exposure levels. Furthermore, the concentration of one marker may be used to predict concentrations of other constituents if the concentration ratios between the marker and the other constituents of interest are known.

Evaluation of Specific Markers

Concentrations of secondhand smoke components in indoor air have multiple determinants: the rate of smoking, the volume of the room or space, the air exchange rate, the exchange of volatile components between vapor and particle phases, deposition rates on surfaces, rates of re-emission from the surfaces, and chemical transformations (Daisey 1999). Although studies have measured concentrations of some of these chemicals in laboratory conditions, the behaviors of only a few of these compounds as tracers have been characterized in field settings. Studies document that each component under consideration has potential limitations as a marker. These limitations may be the result of photodegradation, variable partitioning between the particle and vapor phases, or adsorption/re-emission rates that differ from those of other compounds of concern. No single compound or component has been identified as a completely valid marker for every constituent found in secondhand smoke. On the other hand, several useful markers have a sufficient specificity for secondhand smoke and they can be used to characterize exposures of the public in general or of particular groups. Of these markers, nicotine is highly specific and is considered a valid marker of the PM component of secondhand smoke across a wide range of concentrations in indoor environments (Daisey 1999).

Researchers have studied secondhand smoke characteristics in chambers, with different cigarette brands as the source. In these studies, many different brands generated similar steady-state concentrations of both vapor phase nicotine and respirable particles, and the relationship between these two markers was similar among brands (Leaderer and Hammond 1991; Daisey et al. 1998). Sources other than smoking also contribute to background concentrations of particles found indoors, such as cooking and particles that have infiltrated from the outdoors (Leaderer and Hammond 1991). Thus, the models for estimating the relationship between nicotine and respirable particle concentrations involve regression approaches that estimate increases in nicotine concentrations

with increases in particle concentrations. In such linear regression models, the intercept estimates the background concentration of particles and the slope describes the relationship between concentrations of nicotine and secondhand smoke particles. In most environments where people spend time, secondhand smoke concentrations are usually much lower than in laboratory chambers, so background particles represent a significant fraction of the particle concentration. The relationship between concentrations of nicotine and respirable particles in indoor air has been consistent across field studies in 47 homes (Leaderer and Hammond 1991), in 44 office samples (Schenker et al. 1990), and in 14 other workplaces (Miesner et al. 1989). The range of slopes for the increase of respirable particulate matter (RPM) concentration with nicotine concentration is narrow: 8.6 to 9.8 μg of RPM per μg of nicotine. Daisey (1999) calculated a slope of 10.9 μg of RPM per μg of nicotine using personal sampling data that Jenkins and colleagues (1996) had compiled from more than 1,500 people in the United States. Thus, for each microgram of atmospheric nicotine in the various environments where people spend time, there is an estimated increase of about 10 μg in secondhand smoke particle concentrations.

Until recently, most studies incorporated either respirable particles or nicotine as markers for secondhand smoke, and they remain the most commonly used markers. The literature on the concentrations of these markers is now substantial. In an early study carried out in the late 1970s, Repace and Lowrey (1980) evaluated secondhand smoke levels by contrasting the concentration of particles measured during a bingo game in a church with the concentration measured during a church service with a similar number of people present who were not smoking. The particle levels were much higher during the bingo game (279 $\mu g/m^3$) compared with during the service (30 $\mu g/m^3$). Similarly, studies in the early 1980s of respirable particles in homes found that concentrations in the homes of smokers were substantially higher than concentrations in the homes of nonsmokers (approximately 74 $\mu g/m^3$ versus 28 $\mu g/m^3$, respectively) (Spengler et al. 1985). However, the high levels of respirable particles from other sources and the variability in the concentrations of these particles make it difficult to use the respirable particle concentration as an indicator of secondhand smoke, particularly if secondhand smoke concentrations are low.

In most environments where the public spends time, nicotine in the air comes only from tobacco smoke, so there is no background concentration to be considered. This very high specificity, in combination with the development of inexpensive, sensitive, and passive methods to measure nicotine concentrations in real-world environments, has led to the widespread use of nicotine as a marker for secondhand smoke (Jenkins et al. 2000). A 1999 review concluded that nicotine was a suitable marker for secondhand smoke (Daisey 1999).

Findings from initial secondhand smoke chamber studies that used nicotine as a marker provide evidence supporting its use (Hammond et al. 1987; Leaderer and Hammond 1991). The ambient concentrations of both nicotine and respirable particles were similar when human volunteers smoked 12 brands of cigarettes in separate tests. Nicotine and tar yields varied in mainstream smoke over an order of magnitude (0.1 milligram [mg] of nicotine per cigarette for ultra-low nicotine cigarettes to 1.3 mg per cigarette for regular cigarettes). Subsequent studies showed that nicotine decay in chambers did not follow first-order kinetics (where the speed of a chemical reaction is proportional to the concentrations of the reactants), and short-term measurements in chambers indicated varying ratios of nicotine when compared with other secondhand smoke constituents (Eatough et al. 1989a; Nelson et al. 1992; Van Loy et al. 1998). However, further investigations showed that these findings were artifacts of the chambers themselves. In real-world settings with longer sampling times, nicotine concentrations closely tracked levels of other secondhand smoke constituents (Van Loy et al. 1998; Daisey 1999; LaKind et al. 1999a).

Concentrations of eight possible tracers for secondhand smoke (nicotine, 3-ethenyl pyridine, myosmine, solanesol, scopoletin, RPM, ultraviolet-absorbing particulate matter [UVPM], and fluorescing particulate matter [FPM]) were measured in 469 personal samples collected in workplaces where smoking was allowed (LaKind et al. 1999a). The first three chemicals were in the gas phase, while the latter five were in the particle phase. Concentrations of the three gas phase markers (nicotine, 3-ethenyl pyridine, and myosmine) were highly correlated (r^2 >0.8, where r^2 = the coefficient of determination describing the strength of the model), as were those for three of the particle phase markers (UVPM, FPM, and solanesol) (Table 3.2). Scopoletin was also correlated with UVPM, but only at higher concentrations. Respirable particle concentrations were not strongly correlated with concentrations of UVPM or of nicotine, probably because respirable particles were present in the workplaces from sources other than smoking. Nicotine concentrations in the gas phase correlated with concentrations of the particle phase marker UVPM and with the other particle phase markers that were correlated with UVPM: FPM, solanesol, and scopoletin.

Table 3.2 Correlations between various secondhand smoke constituents as selective markers of exposures

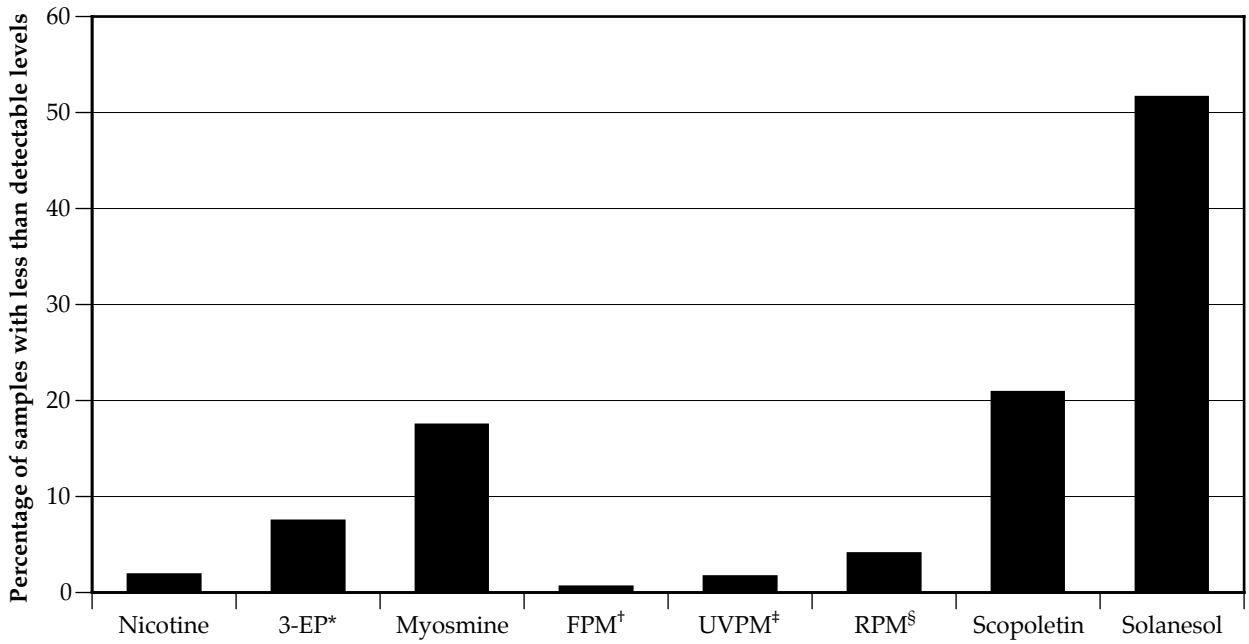
Secondhand smoke constituent	Secondhand smoke exposure marker	R^2*
Nicotine	3-EP†	.83
	Myosmine	.88
	UVPM‡	.63
UVPM	FPM§	.96
	Solanesol	.84
	Scopoletin >1	.73
	Scopoletin <1	.10

Note: 469 personal samples collected from workplaces that permitted smoking.
*R^2 = The coefficient of determination describing the strength of the model.
†EP = Ethenyl pyridine.
‡UVPM = Ultraviolet-absorbing particulate matter.
§FPM = Fluorescing particulate matter.
Source: LaKind et al. 1999b (from the 16 Cities Study).

Several studies examined concentrations of some of the toxic compounds that cigarette smoking emits into the air. Two studies found that different brands of cigarettes released very similar amounts of two nitrosamines, *N*-nitrosodimethylamine and *N*-nitrosopyrrolidine (Mahanama and Daisey 1996). Other toxic volatile organic compounds in secondhand smoke, including benzene, formaldehyde, 1,3-butadiene, and styrene, also exhibited little variation among brands (Daisey et al. 1998). This consistency in emissions among several different brands indicates that changes in the concentration of a particular marker imply proportional changes in the concentrations of other airborne toxic chemicals that are in secondhand smoke.

The level of sensitivity is another key characteristic of a potential marker for secondhand smoke. High sensitivity enables markers to detect low levels of secondhand smoke, which is a necessary quality for evaluating control programs and for surveillance. Some markers have this necessary degree of sensitivity. In the 16 Cities Study conducted by Jenkins and colleagues (1996), researchers collected 469 samples of these eight markers during one workday at worksites where smoking was allowed. Three markers were quite sensitive: nicotine, FPM, and UVPM; less than 2 percent of the samples had concentrations below the limit of detection. More than 10 percent of the samples fell below the limit of detection for myosmine, scopoletin, and solanesol (Figure 3.3). In fact, less than half of the samples collected in workplaces where smoking was allowed had detectable levels of solanesol.

Figure 3.3 Sensitivity of markers for secondhand smoke exposure



Note: 469 personal samples from workplaces that permitted smoking.
*EP = Ethenyl pyridine.
†FPM = Fluorescing particulate matter.
‡UVPM = Ultraviolet-absorbing particulate matter.
§RPM = Respirable particulate matter.
Source: Calculated from data in LaKind et al. 1999a.

Exposure Models

Models and mathematical representations can also be used to estimate human exposures to secondhand smoke (Ott 1999) because they are useful for predicting secondhand smoke concentrations with different patterns of cigarette smoking and for comparing control measures. The microenvironmental model is a tool that can estimate population exposures to secondhand smoke when there is information on the places where people spend time and whether people are smoking. Secondhand smoke concentrations can be inferred from models that characterize contamination of indoor spaces from smoking or from measurements made in the various microenvironments.

Standard techniques that are used to model concentrations of air contaminants indoors, based on the mass balance model, typically include terms that account for the volume of the room, the generation rate, and the removal rate. For secondhand smoke, the generation rate is the number of cigarettes smoked, and the removal rate may include terms such as the air exchange rate, the rate of deposition on surfaces, and

terms for chemical transformations. In some cases, the rate of re-emission from surfaces may also be important. Van Loy and colleagues (1998) have written one such equation:

$$\frac{dC_i}{dt} = \frac{E_i(t)}{V} - ACH * C_i - \frac{1}{V}\sum_{j=1}^{g} S_j \frac{dM_{ij}}{dt}$$

where C_i is the concentration of airborne chemical i, $E_i(t)$ is the emission rate of i, V is the volume of the room, ACH is the air exchange rate, S_j is the area of surface j, and M_{ij} is the mass of i deposited on surface j. The term

$$\frac{dC_i}{dt}$$

gives the rate of change of the concentration. The first term on the right is the emissions rate per volume, the second is the loss of concentration due to air exchange, and the third is the loss to surfaces.

Adapted to secondhand smoke, the model implies that secondhand smoke concentrations depend on the number of smokers and their rate of smoking corresponding to $E_i(t)$ and the space, air exchange rate, and surface deposition—the factors that determine the net removal of secondhand smoke. Ott (1999) has more specifically formulated this model for secondhand smoke, as have others (Daisey et al. 1998; Klepeis 1999a).

$$\overline{C(t)} = \frac{n_{ave} g_{cig}}{Q} - \frac{\Delta C}{(ACH)t}$$

The average secondhand smoke concentration at some time ($\overline{C}(t)$) depends on two terms. The first term

$$\frac{n_{ave} g_{cig}}{Q}$$

has the source strength as its numerator: n_{ave} is the number of smokers, and g_{cig} is the emission rate from the cigarette as mass multiplied by time. The denominator is the air flow rate, with higher air flows leading to lower concentrations. The second term

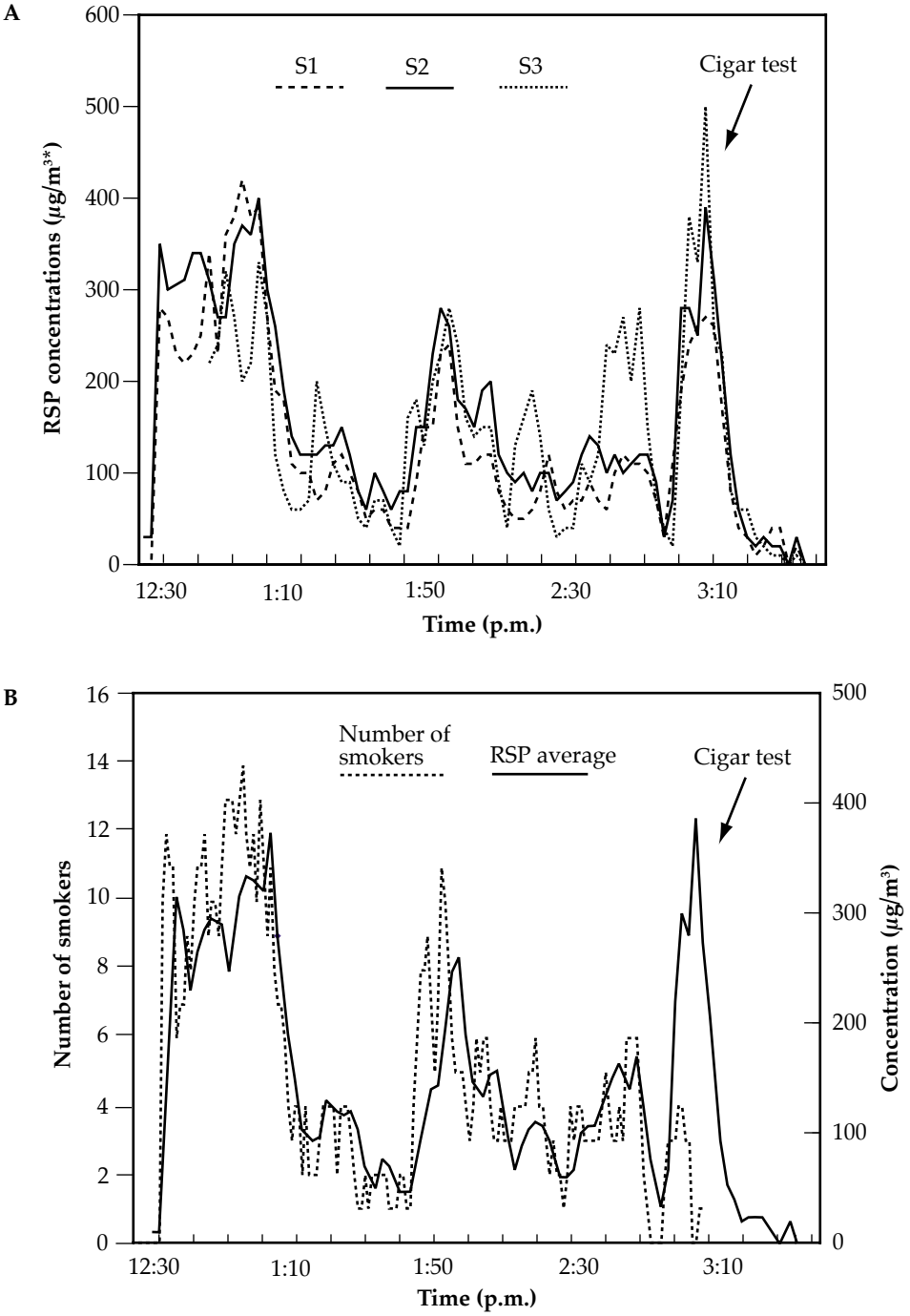
$$\frac{\Delta C}{(ACH)t}$$

captures changes in concentrations over the time of observation (ΔC), the air exchange rate (ACH), and the time of observation t. Thus, the average concentration is determined by source strength (the first term) and loss rate (the second term). If conditions are stable, then $\Delta C = 0$, and the secondhand smoke concentration depends only on source strength ($n_{ave} g_{cig}$) and dilution rate (Q). This model assumes a uniform mixing of the smoke throughout the space.

Klepeis and colleagues (1996) applied this multismoker model to data collected from observations of respirable particle and CO measurements in smoking lounges in two airports. During 10 visits, the authors carefully tracked the number of cigarettes smoked and measured continuous particle and CO concentrations. A test with a cigar (several cigars at a time) generated substantial concentrations of CO and RPM that were then tracked as they decayed exponentially. Because CO does not react with surfaces, its decay rate was used to determine the mechanical air exchange rate. Calculating the difference between the CO and RPM decay rates provided estimates of the effective decay rate, which takes into account physical and chemical reactions that affect particle concentrations in addition to removal (dilution) by the mechanical ventilation system. The report documented that the removal of RPM by surface deposition and chemical reaction in both lounges was about 19 to 20 percent of the ventilatory removal. Air exchange rates for these airport smoking lounges were high, approximately 11 and 13 ACH. Mechanically induced turbulence will increase particle removal by surface deposition, but if the number of air changes is similar to that found in office buildings (1 to 3 ACH) and homes (0.3 to 3 ACH), the removal of RPM by deposition, evaporation, and agglomeration would be a more substantial fraction of the overall effective ventilation rate.

Surface adsorption also removes gaseous constituents of secondhand smoke. Because different physical and chemical processes are involved, different decay rates are expected for different components. Sorption, or the uptake and release of gaseous components of secondhand smoke, is a complex phenomenon involving physical and chemical processes on surfaces. Coverage of this topic is beyond the scope of this chapter. The model developed by Ott and colleagues (1992) and validated by Klepeis and colleagues (1996) provided realistic estimates of time-varying concentrations of respirable suspended particles associated with secondhand smoke (Figure 3.4) (Klepeis 1999a). The estimated RPM from cigarettes (11.4 mg per cigarette) was similar to the value derived independently by Özkaynak and colleagues (1996), who used a mass balance regression

Figure 3.4 Estimates of time-varying respirable suspended particle (RSP) concentrations associated with secondhand smoke



Note: Figure A shows RSP concentration time series measured by piezobalances (labeled S1, S2, and S3) at three widely spaced locations in the smoking lounge taken at the San Jose International Airport (SJC5) fifth study visit. The large decay c

Figure B shows the cigarette count time series and the mean RSP concentration time series from the three piezobalances taken at the SJC5 study visit.

*$\mu g/m^3$ = Micrograms per cubic meter.

Source: Klepeis et al. 1996. Reprinted with permission.

model and indoor $PM_{2.5}$ data from the Particle Total Exposure Assessment Study. The model predicted CO emissions per cigarette similar to the values presented by Owens and Rosanno (1969).

The model for RPM exposures from secondhand smoke that Ott and colleagues (1992) developed is a useful tool for estimating short-term concentrations in settings where the smoking rates and ventilation rates are known. The model could also be used to advance exposure assessment studies and as a design aid for designated smoking areas within buildings. Mass-based models also successfully predict the concentration of nicotine. Repace and colleagues (1998) used a similar model to predict nicotine from secondhand smoke in office air and in salivary cotinine among office workers exposed only in the office; the agreement between the predicted concentrations and the levels observed in field studies was excellent: the mean-predicted concentration was 13.8 $\mu g/m^3$ and the observed mean of 61 samples in nine offices was 15.8 $\mu g/m^3$; the median-predicted salivary cotinine was 0.49 nanograms (ng)/m compared with an observed median of 0.5 ng/milliliter (mL) in 89 nonsmoking office workers who had not been exposed at home.

Both chamber and field studies have validated these models. Experimental chambers differ from many real-world environments such as homes, restaurants, and workplaces in several important aspects. For example, chambers typically have much greater surface to volume ratios, which increase the opportunity for adsorption onto those surfaces, and the air exchange rates are carefully controlled and often kept low to maintain high concentrations. Thus, adsorption onto and desorption from surfaces may have a greater impact in chamber studies than in the field. In fact, the adsorption and desorption of secondhand smoke chemicals onto surfaces have been studied in chambers, and concerns have been raised about the different rates of adsorption and desorption with different markers. However, this phenomenon was less important in field studies than in chamber studies. Thus, the concentrations of secondhand smoke marker chemicals measured in the workplace are well correlated with one another (Table 3.2).

Summary of Atmospheric Markers and Exposure Models

Researchers have suggested several markers for measuring the concentration of secondhand smoke (USDHHS 1986). Of the gas phase markers that researchers have most often used (nicotine, 3-ethenyl pyridine, and myosmine), concentrations were highly correlated in various real-world environments and were correlated with particle phase markers when these markers were detectable (Jenkins et al. 1996). Nicotine, FPM, and UVPM were the most sensitive of these gas and particle phase markers, detecting low levels of secondhand smoke when levels of other markers were below the limit of detection (LaKind et al. 1999b).

Conclusions

1. Atmospheric concentration of nicotine is a sensitive and specific indicator for secondhand smoke.

2. Smoking increases indoor particle concentrations.

3. Models can be used to estimate concentrations of secondhand smoke.

Implications

A set of approaches is available for documenting the exposures of people to secondhand smoke in indoor environments. The atmospheric concentration of nicotine can be readily measured, offering a valid quantitative indicator of the presence of secondhand smoke in the indoor air. Smoking increases levels of other contaminants, including particles. Measurements of nicotine can be used for both research and surveillance purposes. Models have also been developed to estimate concentrations of secondhand smoke in indoor spaces. These models can be used to assess the consequences of various scenarios of controlling for secondhand smoke.

Biomarkers of Exposure to Secondhand Smoke

A biomarker of exposure has been defined by the NRC (1989) as "…an exogenous substance or its metabolite or the product of an interaction between a xenobiotic agent (an external, biologically active agent) and some target molecule or cell that is measured in a compartment within an organism" (p. 12). Thus, measuring specific biomarkers in people can provide evidence that exposure of the individual to secondhand smoke has actually occurred. For some agents, measurements of biomarkers that have interacted with a target site in the body may indicate the biologically effective dose (Sampson et al. 1994; Perera 2000). However, biomarkers do not provide direct information on exposure microenvironments and are therefore complementary to environmental and personal monitoring (NRC 1991). In 1992, the EPA listed several criteria that a biomarker of exposure for a specific air contaminant should meet (USEPA 1992). Based on those criteria, the ideal biomarker of exposure to secondhand smoke should (1) be specific for involuntary smoking, (2) have an appropriate half-life in the body, (3) be measurable with high sensitivity and precision, (4) be measurable in samples collected by noninvasive techniques, (5) be inexpensive to assay, (6) be either an agent associated with health effects or strongly and consistently associated with such an agent, and (7) be related quantitatively to a prior exposure to secondhand smoke. Several biomarkers have been used to assess involuntary smoking, but each has had limitations when matched against these criteria. Nevertheless, these biomarkers have provided information for tracking population exposures to secondhand smoke. There are several published reviews of biomarkers of secondhand smoke exposure (Benowitz 1996, 1999; Jaakkola and Jaakkola 1997; Scherer and Richter 1997; National Cancer Institute 1999; Woodward and Al-Delaimy 1999).

Compounds that have been used as biomarkers for involuntary smoking include CO in exhaled air, carboxyhemoglobin (the complex form of CO found in the blood), thiocyanate, nicotine and its primary metabolite cotinine, polycyclic aromatic hydrocarbon (PAH) adducts in leukocyte DNA or plasma albumin, and hemoglobin (Hb) adducts of tobacco-related aromatic amines such as 3-aminobiphenyl (3AB) and 4AB. A relationship between urinary concentrations of hydroxyproline, an indicator of collagen degradation (a marker of effect), and exposure to secondhand smoke has been proposed (Yanagisawa et al. 1986) but has not been confirmed by other investigators (Adlkofer et al. 1984; Verplanke et al. 1987; Scherer and Richter 1997), and hydroxyproline analyses have not been used in more recent studies. The tobacco-specific nitrosamine 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol (NNAL) may prove to be quite useful as an exposure marker in the future (Hecht et al. 1993b), although relatively few studies have been conducted of NNAL levels in nonsmokers (Hecht et al. 1993b, 2001; Parsons et al. 1998; Meger et al. 2000; Anderson et al. 2001). Levels of other compounds present in tobacco smoke such as benzene, 2,5-dimethylfuran, and benzo[*a*]pyrene (B[*a*]P) may be significantly higher among smokers compared with nonsmokers, but such compounds are of limited value as biomarkers of involuntary smoking because they are not specific to tobacco smoke. Thus, although some of these compounds may be of value in classifying active smokers and nonsmokers, only those compounds with the highest specificity and sensitivity are potentially useful for assessing variations in exposure to secondhand smoke. Feasibility and cost are additional considerations. The biomarkers most commonly proposed for this purpose have been CO, thiocyanate, and nicotine or its metabolites.

Carbon Monoxide and Thiocyanate

The compound CO is present in both mainstream and sidestream smoke and can be measured in people as either expired breath CO or as carboxyhemoglobin. Such measurements may be useful in confirming the absence of active smoking, but they are of limited value as markers of exposure to secondhand smoke because of a relatively short half-life and because of the nonspecificity of CO as a marker for exposure to tobacco smoke. In addition to tobacco combustion, CO has both indoor and outdoor sources, including vehicle exhaust and incomplete combustion in furnaces, space heaters, and other similar devices. The human body's own metabolic processes also produce CO, and nonsmokers have a typical carboxyhemoglobin concentration of about 1 percent. The half-life of CO in the body is about two to four hours (Castleden and Cole 1974). Therefore, although this time period varies with individual activity levels,

CO is only useful as an indicator of recent exposures. Both expired breath CO and blood level carboxyhemoglobin measurements have been used in studies of exposure to secondhand smoke. In general, however, a definite increase in these markers has only been noted immediately following substantial exposures (Table 3.3). Thus, levels of CO in exhaled breath or in carboxyhemoglobin in blood are of limited value as routine markers of involuntary smoking.

Cigarette smoke also contains significant amounts of hydrogen cyanide, which is detoxified in the body by conversion to thiocyanate. As a marker, thiocyanate is easily measured in serum, urine, or saliva by manual or automated colorimetric methods. Thiocyanate has an estimated half-life of about one week—a period of time that is a fairly long interval for the integration of an exposure (Junge 1985). However, thiocyanate lacks specificity as a marker of involuntary smoking primarily because of dietary contributions from cyanide-containing foods, such as almonds, or from the presence of thiocyanate itself in certain cruciferous vegetables such as cabbage, broccoli, and cauliflower. This lack of specificity restricts the usefulness of thiocyanate in assessing exposure to tobacco smoke. Although some studies have reported significantly increased levels of thiocyanate among nonsmokers exposed to secondhand smoke (Table 3.3), two rather large studies with more than 1,000 persons apiece found no significant difference in serum thiocyanate levels between nonsmokers with and those without reported exposure to secondhand smoke (Table 3.3) (Foss and Lund-Larsen 1986; Woodward et al. 1991). Both expired breath CO and serum thiocyanate levels may be useful as confirmatory markers in smoking cessation studies because no interference from nicotine replacement therapy occurs, but the lack of specificity of these markers limits their application in studies of involuntary smoking.

Nicotine and Cotinine

Nicotine is a highly tobacco-specific component of cigarette smoke that is present in abundant amounts (approximately 7 to 8 mg per cigarette) (IARC 2004). Nicotine can be readily measured in both active and involuntary smokers in a number of biologic materials including serum, urine, and saliva. Most of the nicotine emitted from a cigarette is found in sidestream smoke (NRC 1986), which is the major contributor to secondhand smoke. Nonsmokers inhale nicotine, which is present as a gas, during involuntary smoking. Some of the absorbed nicotine is excreted in urine, but on average, about 90 percent of the nicotine is further metabolized (Benowitz and Jacob 1994). Of this nicotine, about 70 to 80 percent is metabolized to cotinine (range: 60 to 90 percent). Cotinine is the major proximate metabolite of nicotine and the predominant nicotine metabolite present in the blood; cotinine is further metabolized to other chemicals, such as hydroxycotinine and cotinine glucuronide. Nicotine can be measured in physiologic fluids as an exposure biomarker, but its short half-life in the body of approximately one to three hours limits its utility as a marker of chronic exposure (Scherer et al. 1988; Benowitz et al. 1991). Consequently, cotinine, the primary metabolite of nicotine with a substantially longer half-life, is regarded as the biomarker of choice for exposure to secondhand smoke (Jarvis et al. 1987; Watts et al. 1990; Benowitz 1999). Participants in a workshop convened to discuss analytical approaches suitable for assessing involuntary smoking among people concluded with a general consensus "…that the nicotine metabolite, cotinine, has the prerequisites of specificity, retention time in the body, and detectable concentration levels that make it the analyte of choice for quantifying exposures" (Watts et al. 1990, p. 173).

The estimated half-life of cotinine in serum, urine, or saliva averages about 16 to 18 hours (Table 3.4) (Jarvis et al. 1988). Some investigators have reported that the cotinine half-life in nonsmokers may be significantly longer than in smokers, whereas other studies have found a similar half-life in both groups (Table 3.4). Kyerematen and colleagues (1982) used a relatively low dose of nicotine (less than 0.2 mg based on an assumed mean body weight of 70 kilograms) and found a statistical, but small, difference in the half-life of labeled cotinine between smokers and nonsmokers. However, Sepkovic and colleagues (1986) and Haley and colleagues (1989) reported a much longer half-life of cotinine in nonsmokers than in smokers. Both studies used a radioimmunoassay (RIA) for their analyses, and the cross-reactivity or limited sensitivity of their assays during the terminal elimination phase when cotinine concentrations would be low may have contributed to their results. Benowitz (1996) pointed out that more recent data indicate similar cotinine clearance rates for both smokers and nonsmokers. Benowitz (1996) suggested that any increase in the apparent half-life for nonsmokers at low nicotine concentrations may represent residual tissue storage of nicotine with continued release over time. This notion would be consistent with the finding that the mean half-life for the elimination of cotinine derived from labeled nicotine among nonsmokers was slightly

Table 3.3 Expired air carbon monoxide (CO), carboxyhemoglobin, and thiocyanate levels following exposure to secondhand smoke

			Findings		
Study	**Analysis**	**Method**	**Unexposed**	**Exposed**	**Difference**
Russell et al. 1973	Carboxy-hemoglobin	CO oximeter	1.6% ± 0.6	2.6% ± 0.7	p <0.001
Jarvis et al. 1983	Expired air CO	Data were not reported	4.7 ppm	10.6 ppm	p <0.001
Poulton et al. 1984	Serum thiocyanate	Colorimetric	54.2 ± 11.3 μmol/L[†] n = 10	97.3 ± 45.3 μmol/L n = 14	p <0.002
Foss and Lund-Larsen 1986	Serum thiocyanate	Colorimetric	Men 29.7 ± 14.2 μmol/L n = 248 Women 30.2 ± 13.6 μmol/L n = 366	 30.9 ± 13.5 μmol/L n = 328 31.9 ± 15.8 μmol/L n = 229	 NS[‡] NS
Husgafvel-Pursiainen et al. 1987	Carboxy-hemoglobin	CO oximeter	0.6% ± 0.2 n = 20	0.7% ± 0.3 n = 27	NS
	Plasma thiocyanate	Colorimetric	46 ± 16 μmol/L n = 20	58 ± 18 μmol/L n = 27	p <0.01
Robertson et al. 1987	Serum thiocyanate	Colorimetric	44.8 ± 21.2 μmol/L n = 57	Group A 44.1 ± 18.5 μmol/L n = 69 Group B 49.6 ± 27.3 μmol/L n = 21	 NS NS
Chen et al. 1990	Serum thiocyanate	Colorimetric	26.9 (9.3–40.9) μmol/L n = 20	35.8 (14.8–78.2) μmol/L n = 26	p <0.05
Woodward et al. 1991	Expired air CO	Ecolyser	Men 2 ppm n = 519 Women 2 ppm n = 817	 3 ppm n = 259 2 ppm n = 461	 NS NS
	Serum thiocyanate	Colorimetric	Men 37 μmol/L n = 455 Women 40 μmol/L n = 702	 35 μmol/L n = 244 39 μmol/L n = 401	 NS NS
Otsuka et al. 2001	Carboxy-hemoglobin	Spectrophotometry	0.24% ± 0.18	1.57% ± 0.32	p <0.001

*ppm = Parts per million.
[†]μmol/L = Micromoles per liter.
[‡]NS = Not significant.

Comments
12 nonsmoking volunteers assayed before (unexposed) and immediately after remaining in a smoke-filled room for an average of 79 minutes; mean CO in the room was about 38 ppm*
7 nonsmokers assayed before (unexposed) and after 2 hours of exposure to secondhand smoke in a bar; peak ambient CO in the bar was 13 ppm
24 children or adolescents (mean age 7.6 years), with 14 living in homes with ≥1 smoker in the immediate family (exposed)
Nonsmokers in Norway with self-reported exposures to secondhand smoke at home or at work
Office workers with no reported exposure (unexposed) and restaurant employees exposed an average of 40 hours per week
Nonsmoking office workers who reported no exposure to secondhand smoke; exposure to secondhand smoke only at work (Group A); or exposure to secondhand smoke both at home and at work (Group B)
Median and range of serum levels among infants in the Chang-Ning Epidemiological Study who lived in nonsmoking homes (unexposed) or in homes where ≥20 cigarettes/day were smoked
Nonsmokers in the Scottish Heart Health Study self-reported either "none" or "a lot" of exposure to secondhand smoke
15 healthy nonsmokers assayed before (unexposed) and immediately after remaining in a room for 30 minutes with people who were smoking; the mean CO level in the room was approximately 6 ppm

longer (21 ± 4.6 hours) (Benowitz and Jacob 1993) than the mean half-life measured in nonsmokers (17 ± 3.9 hours) in a separate study that used labeled cotinine (Benowitz and Jacob 1994). Zevin and colleagues (1997) compared labeled nicotine with labeled cotinine and reported similar results. However, a small increase in the effective half-life resulting from tissue distribution effects would not be expected to influence estimates of secondhand smoke exposure based on cotinine measurements made under steady-state conditions. Collier and colleagues (1990) reported a significantly longer cotinine half-life in neonates and children, but a more recent evaluation found a similar half-life in both newborns and adults (Dempsey et al. 2000).

Besides possible differences in the effective half-life of cotinine among smokers and nonsmokers, research suggests that differences based on gender, race, and ethnicity may exist. Two studies found higher levels of serum cotinine per cigarette smoked in Black smokers than in White smokers—a finding that may reflect differences in nicotine metabolism or in the way that cigarettes are smoked (Wagenknecht et al. 1990; Caraballo et al. 1998). Total and nonrenal clearance of cotinine were significantly lower among Black smokers, and the metabolism of nicotine, cotinine, and *N*-glucuronidation activities were slower among Black smokers than among White smokers (Pérez-Stable et al. 1998; Benowitz et al. 1999). The mean half-life of cotinine among Black smokers (18 hours) was 12.5 percent longer than that found among White smokers (16 hours). One report also suggests that in comparisons with either Latinos or Whites, Chinese Americans metabolized nicotine more slowly; the mean increase in the cotinine half-life among Chinese American smokers was about 14 percent (Benowitz et al. 2002). Although Lynch (1984) found no gender differences in the cotinine half-life, Benowitz and colleagues (1999) found a significantly shorter cotinine half-life in women (14.5 hours) than in men (18.5 hours), a difference that the researchers attributed to a smaller volume of cotinine distribution in women. The same group reported higher metabolic clearance rates and a substantially shorter half-life (about nine hours) for cotinine in pregnant women (Dempsey et al. 2002), a finding that may require a slight revision of classification cutoff levels when assessing active smokers and women exposed to secondhand smoke during pregnancy.

Table 3.4 Half-life of cotinine in smokers and nonsmokers from several studies

Study	Exposure	Assay	Cotinine half-life in hours (mean ± SD*)	Comments
Kyerematen et al. 1982	Intravenous dose of ^{14}C-labeled nicotine at 2.7 μg/kg†	LC‡ separation; then measured radiolabeled metabolite	10.3 ± 2.3 n = 6	6 male smokers; overnight abstention before dosing and throughout the study; plasma assays
		Same	13.3 ± 2.2 n = 6	6 male nonsmokers
Benowitz et al. 1983	Intravenous cotinine infusion	GLC/NPD§	15.8 ± 4 n = 8	5 male and 3 female smokers; plasma assays
	Cotinine washout during 3 days of smoking abstention	GLC/NPD	19.7 ± 6.5 n = 12	8 male and 4 female smokers
Lynch 1984	Cotinine washout during 24 hours of smoking abstention	GLC/NPD	14.6 (men) 15.1 (women)	Averages from 47 male and 41 female smokers; cotinine half-life was calculated from 2-point data only; plasma assays
	Cotinine washout during 3 days of smoking abstention	GLC/NPD	15.4 (men) 15.7 (women)	8 male and 11 female smokers in a smoking cessation program; assayed once/day for 3 days
Sepkovic et al. 1986	Smokers abstained for 7 days	RIA∆	18.5 (plasma) 21.9 (urine)	10 smokers were followed during 7 days of smoking abstention
	Nonsmokers exposed to secondhand smoke in a chamber	RIA	49.7 (plasma) 32.7 (urine)	4 nonsmokers were exposed to secondhand smoke for 80 minutes/day for 4 days, then followed for an additional 7 days
De Schepper et al. 1987	Oral dose of cotinine at 10 and 20 mg¶ concentrations	GC–MS**	12.3 ± 2.6 n = 4	4 male nonsmokers; cotinine half-life was independent of dose, so both doses were averaged per person; the same results were obtained with infused cotinine; plasma assays
Jarvis et al. 1988	Oral dose of nicotine at 28 mg/day for 5 days before analysis	GLC/NPD 2 labs performed each assay	16.6 ± 3.4 n = 5	3 male and 2 female nonsmokers; plasma cotinine assays
			15.9 ± 3.1 n = 5	Salivary cotinine assays
			18.0 ± 4.0 n = 9	Urine cotinine assays

Table 3.4 Continued

Study	Exposure	Assay	Cotinine half-life in hours (mean ± SD*)	Comments
Scherer et al. 1988	Cotinine intravenous infusion	GLC/NPD	17.1 ± 4.4 n = 6	6 smokers; 5 days of smoking abstention before infusion; serum assays
Haley et al. 1989	Cotinine washout during 5 days of smoking abstention	RIA	16.6 ± 3.4 n = 9	9 smokers were followed for 5 days beginning with smoking cessation; urine assays
	Nonsmokers exposed to secondhand smoke in a chamber	RIA	27.3 ± 5.9 n = 10	10 nonsmokers were exposed to secondhand smoke for 8 minutes/day for 2 days, then followed for 4 additional days; urine assays
Curvall et al. 1990b	Oral dose of cotinine at indicated amount Followed for 4 days	GLC/NPD	14.9 ± 4.1 n = 3	7 male and 2 female nonsmokers; plasma cotinine assays following 5 mg dose
			15.6 ± 3.7 n = 9	Plasma cotinine assays following 10 mg dose
			14.9 ± 4.3 n = 9	Plasma cotinine assays following 20 mg dose
			16.3 ± 1.9 n = 3	Salivary cotinine assays following 5 mg dose
			15.7 ± 2.9 n = 9	Salivary cotinine assays following 10 mg dose
			14.9 ± 3.7 n = 9	Salivary cotinine assays following 20 mg dose
Benowitz and Jacob 1994	Native and isotopically labeled intravenous cotinine infusion	GC–MS	16.3 ± 4.4 n = 6	3 male and 3 female nonsmokers dosed with an average of 4.4 mg cotinine over 30 minutes (2 μg/minute/kg body weight); plasma half-life was measured for native cotinine
			16.9 ± 4.3 n = 6	Plasma half-life was measured for dideuterated cotinine
			17.2 ± 3.9 n = 6	Plasma half-life was measured for tetradeuterated cotinine

*SD = Standard deviation.
†μg/kg = Micrograms per kilogram.
‡LC = Liquid chromatography.
§GLC/NPD = Gas-liquid chromatography with nitrogen-phosphorus–specific detectors.
ΔRIA = Radioimmunoassay.
¶mg = Milligram.
**GC–MS = Gas chromatography with mass spectrometry.

Cotinine Analytical Procedures

Cotinine can be measured by a variety of techniques, but for application to studies of involuntary exposure, methods of high specificity and sensitivity are needed. The most commonly used methods have included RIAs and enzyme-linked immunoassays, gas-liquid chromatography (GLC) with nitrogen-phosphorus–specific detectors (NPD) or coupled to a mass spectrometer, and high-performance liquid chromatography (HPLC) using either ultraviolet (UV) or mass spectrometric detection. With the development of suitable antibodies (Langone et al. 1973; Knight et al. 1985), RIAs were made available for relatively sensitive and rapid analyses of nicotine and cotinine in biologic matrices. Enzyme-linked immunosorbent assays that use monoclonal antibodies have also been developed (Bjercke et al. 1986) that obviate radioactive reagents and provide a consistent antibody source. Immunoassays are well suited for screening large numbers of samples in epidemiologic investigations, but may be subject to cross-reactivity from other compounds that can limit the specificity. Even the more sensitive immunoassays for serum cotinine provide reliable results only for more heavily exposed nonsmokers who have serum cotinine concentrations of approximately 0.3 to 1 ng/mL or greater (Coultas et al. 1988; Emmons et al. 1996).

Chromatographic procedures for nicotine and cotinine measurements have commonly involved either HPLC with UV detection (Machacek and Jiang 1986; Hariharan et al. 1988; Oddoze et al. 1998), or capillary GLC/NPD (Jacob et al. 1981; Davis 1986; Teeuwen et al. 1989; Feyerabend and Russell 1990). The sensitive GLC/NPD methods of Feyerabend and Russell (1990) and of Jacob and colleagues (1981), with reported detection limits of about 0.1 ng/mL, have been used in support of several studies of exposure to secondhand smoke. There has been a more recent increase in the use of mass spectrometry for these analyses (Daenens et al. 1985; Norbury 1987; Jacob et al. 1991; McAdams and Cordeiro 1993; James et al. 1998). Gas chromatography (GC) with mass spectrometric detection provides a sensitive analytical method with inherently high specificity and enables the optimal use of stable isotopically labeled forms of the analyte as internal standards. This type of analysis is particularly well suited for sensitive cotinine measurements in complex biologic matrices. The recent availability of instrumentation combining HPLC with atmospheric pressure ionization tandem mass spectrometry has enabled the development of methods that provide high sensitivity and analytical specificity. These methods are also well suited for application to epidemiologic studies that analyze large numbers of samples (Bernert et al. 1997; Bentley et al. 1999; Tuomi et al. 1999). Benowitz (1996) has compared the relative sensitivity, specificity, and costs of these analytic procedures (Table 3.5).

Table 3.5 Analytical methods for measuring cotinine in nonsmokers

Study	Method	Sensitivity	Specificity	Cost
Langone et al. 1973; Haley et al. 1983; Knight et al. 1985	Radioimmunoassay	1–2 nanograms/ milliliter (ng/mL)	Variable (poorest in urine)	Low
Jacob et al. 1981; Feyerabend et al. 1986	Gas chromatography	0.1–0.2 ng/mL	Good	Moderate
Hariharan and VanNoord 1991	High-performance liquid chromatography	±1 ng/mL	Good	Moderate
Jacob et al. 1991	Gas chromatography–mass spectrometry	0.1–0.2 ng/mL	Excellent	High
Bernert et al. 1997	Liquid chromatography/atmospheric pressure ionization tandem mass spectrometry	<0.05 ng/mL	Excellent	Extremely high

Source: Benowitz 1996.

Analytical Matrices for Cotinine Measurements

Nicotine and cotinine have been measured in a wide variety of physiologic matrices, including amniotic fluid (Lähdetie et al. 1993; Jauniaux et al. 1999), meconium (Ostrea et al. 1994; Dempsey et al. 1999; Nuesslein et al. 1999), cervical lavage (Jones et al. 1991), seminal plasma (Shen et al. 1997), breast milk (Luck and Nau 1984; Becker et al. 1999), sweat (Balabanova et al. 1992), and pericardial fluid (Milerad et al. 1994). However, most investigations of exposure to secondhand smoke have involved assays of cotinine in blood, urine, or saliva, or of nicotine or cotinine in hair. Nicotine is metabolized to cotinine mainly in the liver, but also in the lungs and kidneys; cotinine then enters the bloodstream. When an individual is subjected to involuntary smoking on a regular basis, a steady-state condition may be achieved in which blood cotinine levels remain fairly constant during the day (Benowitz 1996). Because of this stability in concentration levels, in conjunction with the reliable and well-defined composition of blood samples, blood serum or plasma has been considered the matrix of choice for quantitative cotinine assays (Watts et al. 1990; Benowitz 1996). Thus, in the past few years, plasma or serum cotinine measurements have been used in several large epidemiologic investigations of secondhand smoke exposure (Tunstall-Pedoe et al. 1991; Wagenknecht et al. 1993; Pirkle et al. 1996).

Despite a preference for blood plasma or serum as the matrix for cotinine assays, obtaining a blood sample is invasive, and collecting samples from younger children may be difficult. Consequently, saliva cotinine has been suggested as a useful alternative in many cases (Jarvis et al. 1987; Curvall et al. 1990a; Etzel 1990). Saliva is secreted into the oral cavity primarily by the parotid, sublingual, and submandibular glands. These glands typically produce between 18 and 30 mL of unstimulated saliva per hour (Sreebny and Broich 1987); the flow of stimulated saliva is three to six times greater. Oral fluids are a mixture derived from the individual salivary glandular secretions and oral mucosal transudates (gingival crevicular fluid), which are filtrates of plasma. Specific secretions may be recovered, but mixed or "whole" saliva is most commonly collected for cotinine analysis either by direct collection in an appropriate vessel or by adsorption onto commercially available collection pads (Sreebny and Broich 1987).

Many lipophilic drugs may pass from blood into saliva by simple diffusion through the lipid membranes of acinar cells. Because cotinine is a small, relatively lipophilic molecule with little protein binding (Benowitz et al. 1983), its concentration in saliva tends to closely parallel its concentration in blood. Several investigators have found a linear relationship between blood and saliva cotinine concentrations, with saliva levels typically about 1.1 to 1.5 times higher than the corresponding serum concentrations (Jarvis et al. 1988; Curvall et al. 1990a; Rose et al. 1993; Bernert et al. 2000). Schneider and colleagues (1997) compared cotinine levels in saliva samples that were obtained by using either sugar or paraffin wax to stimulate flow—unstimulated saliva samples were collected from the same persons. The researchers concluded that the significantly lower levels found in stimulated samples resulted from higher salivary flow rates. Other investigators, however, concluded that salivary flow rates did not influence cotinine concentrations in their samples (Van Vunakis et al. 1989; Curvall et al. 1990a), and the use of stimulated saliva with a somewhat higher and more uniform pH may reduce both the interindividual and intraindividual variability in the saliva-plasma ratio of a weak base such as cotinine (Knott 1989). Saliva cotinine assays have proven to be a quite useful noninvasive approach for assessing exposures to secondhand smoke, although a greater consistency in salivary collection methods among studies may facilitate subsequent comparisons of the results (Schneider et al. 1997).

Urine can also be readily obtained. Urine cotinine assays have several additional advantages over blood or saliva assays, such as the availability of the large volumes that can usually be collected, and typical cotinine concentration levels that average about five to six times higher than serum levels for unconjugated cotinine (Jarvis et al. 1984; Benowitz 1996). Besides nicotine and cotinine, urine samples may also contain significant amounts of the cotinine metabolite *trans*-3′-hydroxycotinine (Dagne and Castagnoli 1972; Neurath and Pein 1987) as well as several additional minor metabolites including nicotine-1′-*N*-oxide, cotinine-*N*-oxide, nornicotine, and norcotinine (Beckett et al. 1971; Jacob et al. 1986; Zhang et al. 1990; Benowitz et al. 1994). Two additional metabolites that were described more recently are 4-oxo-4-(3-pyridyl)butanoic acid and 4-hydroxy-4-(3-pyridyl)butanoic acid, which possibly arise from 2′-hydroxylation of nicotine and represent up to 14 percent of the nicotine dose (Hecht et al. 1999b, 2000). Nicotine, cotinine, and hydroxycotinine predominate in urine and are present in both an unconjugated form and as their glucuronides (Byrd et al. 1992), with nicotine and cotinine forming *N*-glucuronides and hydroxycotinine forming an O-glucuronide (Byrd et al. 1994; Benowitz et al. 1999).

Hydroxycotinine is often the most abundant nicotine metabolite present in urine, with a half-life of approximately six hours in adults when given alone, which is much shorter than that of cotinine (Scherer et al. 1988; Benowitz and Jacob 2001). In the presence of cotinine, however, the elimination half-life of 3′-hydroxycotinine is similar to that of continine (Dempsey et al. 2004). Consequently, cotinine is the most commonly used biomarker in urine samples. However, this half-life differential may not be present in newborns in whom the half-life is about the same for cotinine and 3′-hydroxycotinine (Dempsey et al. 2000). As with saliva, urine cotinine concentrations are also highly correlated ($r \pm 0.8$) with blood concentrations (Jarvis et al. 1984; Thompson et al. 1990; Benowitz 1996). Measuring a range of nicotine metabolites rather than cotinine alone may also be useful in some circumstances, and for such analyses, urine would often be the matrix of choice.

Higher cotinine concentrations present in urine can enhance sensitivity in an analysis of secondhand smoke exposure. However, urine assays have the disadvantage of being subject to variability that results from hydration differences among participants at the time of collection, because 24-hour urine samples are rarely available and random samples are most often used. Many investigators have attempted to circumvent this limitation by measuring both cotinine and creatinine in the sample and expressing the results as simple cotinine-creatinine ratios (NRC 1986), or by normalizing to a standardized creatinine concentration based on a regression between cotinine and creatinine in urine (Thompson et al. 1990). However, although daily urinary creatinine excretion is rather uniform within individuals, creatinine production is also directly related to muscle mass and varies by age and gender. Despite these potential limitations, creatinine adjustments of cotinine measurements are often used to provide an index of exposure to secondhand smoke from spot urine samples (NRC 1986).

Nicotine and Cotinine in Hair

One of the primary limitations of blood, urine, or saliva cotinine as a biomarker of exposure is the short exposure period that is represented. Assuming that substances such as nicotine are incorporated into the growing hair shaft over time, the use of hair as an analytical matrix has been suggested as an enhanced index of exposure to secondhand smoke covering a period of several months rather than just a few days. Ishiyama and colleagues (1983) first proposed using hair as a matrix for nicotine analyses, and several investigators have subsequently evaluated both nicotine and cotinine in hair. Unlike other matrices, the concentration of nicotine in hair is greater than that of cotinine (Haley and Hoffmann 1985; Kintz 1992; Koren et al. 1992). Because both concentrations are assumed to be stable once they have been deposited into the hair shaft, many hair analyses have included nicotine measurements or assays of both nicotine and cotinine. Studies of adult nonsmokers have reported a significant increase in hair nicotine concentrations with an increase in self-reported exposures to secondhand smoke (Eliopoulos et al. 1994; Dimich-Ward et al. 1997; Al-Delaimy et al. 2001; Jaakkola et al. 2001). Studies of infants and children have documented similar findings (Nafstad et al. 1995; Pichini et al. 1997; Al-Delaimy et al. 2000). Nafstad and colleagues (1998), however, found no significant differences in hair nicotine levels in a study of 68 nonsmoking women with no known exposure to secondhand smoke and 54 nonsmoking women with reported exposures. Some studies also found that hair nicotine levels for those most heavily exposed to secondhand smoke tended to overlap substantially with levels found in active smokers (Dimich-Ward et al. 1997; Al-Delaimy et al. 2001).

At this point, significant uncertainties remain concerning the use of hair analyses for either nicotine or cotinine to assess exposure to secondhand smoke, including the influence of variations in hair growth rates and in hair treatments such as bleaching or permanents. The mechanism of deposition and the influence of pigmentation are questions that also need to be addressed. The rate of hair growth, which varies among individuals, normally averages about one centimeter per month (Wennig 2000). Selecting nonrepresentative telogen stage (resting phase) hairs is a risk when only a few strands are selected for analysis (Uematsu 1993). Researchers believe that the systemic incorporation of nicotine or cotinine involves the passive diffusion of the substance from the blood into the hair follicle, and then into the growing hair shaft. Findings from studies that administered nicotine to animals are consistent with the systemic incorporation of both nicotine and cotinine into hair in this manner (Gerstenberg et al. 1995; Stout and Ruth 1999). In addition, Gwent and colleagues (1995) administered a single dose of nicotine (Nicorette Plus chewing gum) to six nonsmoking volunteers and demonstrated the incorporation of cotinine (but not nicotine) into beard hair. Cotinine levels peaked on the third day following the exposure. However, drugs may also be deposited

in the hair from contact with apocrine and sebaceous gland secretions, as well as directly into the hair shaft from the environment (Henderson 1993). Nicotine is present in apocrine and eccrine sweat (Balabanova et al. 1992), and studies have clearly demonstrated the adsorption of nicotine into hair from the environment (Nilsen et al. 1994; Zahlsen et al. 1996). Thus, multiple sources may contribute to the presence and levels of nicotine found in hair. Although each of these routes still reflects exposure of the nonsmoker to secondhand smoke, the proper interpretation of the results requires a better understanding of the relative contributions of these various factors. Direct environmental adsorption represents a form of personal air monitoring rather than a biomarker assessment. Because the adsorption of cotinine directly from the environment is expected to be quite low (Eatough et al. 1989b), the analysis of cotinine in hair would seem to provide an advantage in minimizing contributions directly from the environment. However, studies have found cotinine hair measurements to be generally less useful than nicotine hair measurements in assessing differences in exposure to secondhand smoke (Kintz 1992; Dimich-Ward et al. 1997; Al-Delaimy et al. 2000).

An additional concern with hair analyses is the influence of hair pigmentation on nicotine incorporation. Studies have documented a significantly greater systemic accumulation of nicotine in pigmented versus unpigmented hair in rodents (Gerstenberg et al. 1995; Stout and Ruth 1999), and in black hairs compared with white hairs from the same persons (Mizuno et al. 1993; Uematsu et al. 1995). This difference presumably reflects the strong binding of nicotine to melanin (Stout and Ruth 1999; Dehn et al. 2001), which is a relevant issue because differences in deposition as a function of either pigmentation or hair structure could lead to a differential sensitivity of detection or exposure classification among participants, including persons of differing ethnicity. This concern may be specific to nicotine deposition, however, because a similar differential response was not seen in a study of hair cotinine levels among children with either light or dark hair (Knight et al. 1996). Although the analysis of nicotine or cotinine in hair is potentially useful in assessing a longer-term exposure to secondhand smoke, this approach needs additional work.

Dietary Sources of Nicotine

Researchers consider the presence of nicotine or its metabolites in the body to be a specific indicator of prior exposures to tobacco smoke. This consideration thus provides an important rationale for the use of nicotine or its metabolites as biomarkers for secondhand smoke exposure. However, researchers have suggested that nicotine could be detected in some samples of tea and in certain vegetables, including potatoes and tomatoes, that belong to the same family (*Solanaceae*) as tobacco (Castro and Monji 1986; Sheen 1988). Idle (1990) subsequently referenced Sheen's (1988) results and suggested that cotinine measurements might be influenced by the ingestion of significant amounts of nicotine from these or other foodstuffs. Idle (1990) hypothesized that the uptake of dietary nicotine would be similar to the nicotine that is absorbed from the vapor phase in the lungs. However, Svensson (1987) proposed that at the acid pH of the stomach, nicotine would be protonated and not readily absorbed. Using direct measurements, Ivey and Triggs (1978) found essentially no absorption of nicotine from the human stomach at pH 1 and an approximate 8 percent absorption at pH 7.4. Even under moderately alkaline conditions (pH 9.8), the mean absorption was less than 20 percent. However, extensive intestinal absorption of nicotine does occur. Benowitz and colleagues (1991) found that the oral bioavailability of encapsulated nicotine administered to 10 smokers averaged about 44 percent. Bioavailability is low because of first-pass metabolism, which is when nicotine is converted to cotinine and other metabolites.

On the basis of their measurements and projections of dietary intake, Davis and colleagues (1991) proposed that from 9 μg to nearly 100 μg of nicotine per day might be ingested from food. However, this projection was based on maximum intakes of each of the foods of interest including large quantities of tea; actual intakes at that level would be unlikely (Benowitz 1999). In contrast, Repace (1994) used the food-nicotine concentrations reported by Domino and colleagues (1993) as well as a more realistic average consumption quantity of potatoes and tomatoes in the diet. The estimated daily nicotine intake from these foods was approximately 0.7 μg/day. Furthermore, more recent analyses of nicotine content in foodstuffs by specific mass spectrometric procedures found values that were somewhat lower than the earlier estimates. Siegmund and colleagues (1999a) developed a validated method for the extraction and recovery of nicotine from foods using capillary GC–mass spectrometry analysis. This method was subsequently applied to an analysis of a variety of foodstuffs including solanaceous vegetables and tea (Siegmund et al. 1999b). The estimated daily intake of nicotine from all

dietary sources for 14 countries, including the United States, was about 1.4 μg/day, with an estimated 2.25 μg/day at the 95th percentile. These values, which were derived from a Monte Carlo simulation that used mean daily consumption and measured nicotine contents of the foods, are well below the earlier estimates made by Davis and colleagues (1991) but are closer to those reported by Repace (1994).

Calculations of dietary nicotine contributions are necessarily imprecise. Direct evaluations of dietary intake should be more meaningful, and these measurements tended to produce lower results. For example, the dietary intake of nicotine estimated by Davis and colleagues (1991) included an important contribution from tea. Researchers assessed the contribution from tea in more than 1,800 nonsmokers, including many customary tea drinkers, in the Scottish Heart Health Study; no consistent relationship was found between serum cotinine levels and a daily tea intake of up to 10 cups (Tunstall-Pedoe et al. 1991). Those who consumed 10 or more cups per day had a slight increase in serum cotinine, but the effect of tea was noted to be inconsistent. In a large, national epidemiologic survey conducted in the United States, Pirkle and colleagues (1996) used a 24-hour food recall diary, which was completed by each study participant, to compare the dietary intake of potatoes, tomatoes, eggplants, cauliflowers, green peppers, and both instant and brewed tea with serum cotinine levels. Using regression models, these food items explained less than 2 percent of the variance in serum cotinine levels.

Benowitz and Jacob (1994) proposed a conversion factor between nicotine and serum cotinine and suggested that it can be used to estimate nicotine exposure under steady-state conditions. For example, using the most recent estimate from Siegmund and colleagues (1999b) of 1.4 μg of nicotine per day in the average diet, and assuming that 71.3 percent of the dietary nicotine is absorbed in the same manner as vapor phase nicotine from secondhand smoke (Iwase et al. 1991), applying this conversion factor would result in a predicted mean serum cotinine concentration of no more than 0.013 ng/mL; at the 95th percentile of dietary nicotine intake, the estimate would be 0.020 ng/mL. These estimates are consistent with the results of Pirkle and colleagues (1996) and indicate a minimal dietary contribution to serum cotinine measurements. Thus, trace amounts of nicotine may be consumed in the diet, but any contribution from this source is likely to be quite small for most people compared with the amount of nicotine absorbed from secondhand smoke exposure. Additionally, comparisons of cotinine within individuals over time, such as before and after an intervention, would probably be unaffected by diet.

Cotinine Measurements as an Index of Nicotine Exposure

Although the potential for overlap of levels always exists between nonsmokers with an extensive exposure to secondhand smoke and occasional or currently abstinent smokers, the use of cotinine measurements to separate smokers from nonsmokers provides a generally valid approach. Benowitz and colleagues (1983) originally proposed 10 ng/mL as a reasonable cutoff level for cotinine in serum to distinguish between smokers and nonsmokers. Consistent with that proposal, Repace and Lowrey (1993) estimated median serum cotinine levels to be about 1 ng/mL for U.S. adult nonsmokers and about 10 ng/mL for the most heavily exposed nonsmokers. In a study of 211 people in London, England, a plasma cutoff of 13.7 ng/mL provided an optimal classification with 94 percent sensitivity and 81 percent specificity based on self-reported exposure levels (Jarvis et al. 1987). The authors attributed the relatively poor specificity to "deception" in the self-reports of some participants with high serum cotinine levels. When the investigators reclassified those believed to be deceptive as smokers, sensitivities were 96 to 97 percent and specificities were 99 to 100 percent using plasma, saliva, or urine cotinine as the biomarker for comparison. The optimal cutoff values in this study were 14.2 ng/mL in saliva and 49.7 ng/mL in urine (Jarvis et al. 1987).

Pirkle and colleagues (1996) used a serum cotinine cutoff level of 15 ng/mL in a large U.S. epidemiologic study. They found a strong agreement with the self-reported nonsmoking status of the participants: those with serum cotinine levels above 15 ng/mL who claimed no tobacco use comprised only about 1.3 percent of the adult participants and 2.6 percent of the adolescents. Caraballo and colleagues (2001) examined the participants in this study aged 17 years and older in detail and used the same nominal cutoff of 15 ng/mL. There was a 92.5 percent agreement between serum cotinine concentrations and self-reported active smoking status and a 98.5 percent agreement among self-reported nonsmokers. The researchers regarded the infrequent or low rate of cigarette use as an explanation for the disagreement with serum cotinine levels among self-reported smokers in most cases. However, there may have been some deception in the 1.5 percent with discrepant results between their serum cotinine levels and self-reported

status as nonsmokers, particularly among those with relatively high concentrations of serum cotinine. Wagenknecht and colleagues (1992) found similar results in the Coronary Artery Risk Development in (Young) Adults Study, which had a serum cotinine cutoff value of 15 ng/mL that produced a sensitivity of 94.5 percent and a specificity of 96 percent. In general, self-reports of smoking status validated with biomarker assays were accurate in most studies (Patrick et al. 1994), although small adjustments to customary cutoff values between smokers and nonsmokers may be needed based on gender and race for both males and females and for pregnant women. The accuracy of questionnaire reports in determining the extent of exposure may be higher in population contexts than in clinical studies, particularly in investigations of smoking cessation.

The objective in many studies is not only to identify nonsmokers exposed to secondhand smoke, but also to estimate the relative extent of their exposure. If a quantitative relationship exists between exposure to nicotine in secondhand smoke and cotinine biomarker concentrations, then investigators should be able to estimate the average nicotine exposure of groups of individuals from their biomarker levels. Repace and Lowrey (1993) developed a model that related nicotine exposure to cotinine levels measured in both the plasma and urine of nonsmokers. Subsequent comparisons of the model predictions with data from 10 epidemiologic studies were consistent within 10 to 15 percent for median and peak levels of cotinine. Using the fractional conversion of nicotine to cotinine and estimated cotinine clearances in active smokers, Benowitz and Jacob (1994) proposed a factor (K = 0.08 with a coefficient of variation ±22 percent) that could be used to estimate daily nicotine intake (in milligrams of nicotine) from the steady-state plasma cotinine concentration in ng/mL. The validity of this factor is supported by the data from Galeazzi and colleagues (1985). They administered measured doses of nicotine intravenously to six volunteers on four consecutive days and assessed serum cotinine levels on the fourth day, when steady-state conditions had been reached. The results indicate that plasma cotinine concentrations could be directly and linearly related to daily nicotine intake. Predicted nicotine intake calculations, based on the factor proposed by Benowitz and Jacob (1994), demonstrated a close agreement in all cases with the actual exposures (Table 3.6).

Although Benowitz and Jacob (1994) had derived their factor from smokers, the clearance of cotinine was similar for smokers and nonsmokers (Zevin et al. 1997), and Benowitz (1996) noted that the factor for nicotine exposure among nonsmokers should also be similar. The results obtained by Curvall and colleagues (1990b) with short-term exposures and nonsteady-state correlations are in general agreement with that expectation. After administering various low doses of nicotine intravenously to nonsmokers, the researchers concluded that the average intake of nicotine among their participants could be estimated from the following relationship:

Cotinine concentration (ng/mL) ~ 0.5 * [nicotine infusion rate in μg/min] * [absorption time in hours]

where 0.5 represents the somewhat lower fraction of nicotine metabolized to cotinine among nonsmokers as Curvall and colleagues (1990b) had reported. A comparison of this expression with that of Benowitz and Jacob (1994) suggests that both should generate similar results, with the main difference between them reflecting the lower fractional conversion of nicotine to cotinine among nonsmokers as Curvall and colleagues (1990b) had estimated. Curvall and colleagues (1990b) noted that this conversion may represent a true difference, or may have resulted from differences in the experimental setups between the two studies. Zevin and colleagues (1997) reported that the mean conversion of nicotine to cotinine is approximately the same

Table 3.6 Calculation of nicotine dosage from plasma cotinine concentrations

Nicotine administered* (milligrams [mg]/day)	Mean plasma cotinine† (nanograms/ milliliter)	Calculated dose† (mg/day)
7.3	92	7.4
14.6	185	14.8
22.0	278	22.2
29.3	381	30.5

*From the dosage and plasma cotinine concentrations given in Galeazzi et al. 1985 (Table 1). Doses were adjusted to mg/day based on the reported mean weight of the participants (61 kilograms, n = 6).
†Calculated from plasma cotinine multiplied by 0.08.
Sources: Galeazzi et al. 1985; Benowitz and Jacob 1994.

for nonsmokers as for smokers. If that conclusion is correct, then the factor derived by Benowitz and Jacob (1994) should be applicable to both groups.

These estimates are based on studies in which nicotine was infused into people, often at greater concentrations than would result from involuntary smoking. However, the estimates are consistent with a linear relationship between nicotine exposure and mean serum cotinine concentrations when measured under steady-state conditions. These findings suggest that at least an approximate quantitative estimate of nicotine exposures within population groups might be derived from their plasma cotinine concentrations. Because cotinine levels in an individual reflect not only exposure variations but also individual differences in metabolism and excretion, the value of a single measurement within an individual may be limited. However, the application of cotinine measurements in epidemiologic studies that involve large numbers of individuals may provide reliable estimates of average group exposures to nicotine in secondhand smoke (Benowitz 1999).

Protein and DNA Adducts

Measurements of DNA or protein adducts of carcinogens in secondhand smoke may indicate both the exposure (internal dose) and the interaction of the carcinogen or its metabolite with the host tissue, thus reflecting the biologically effective dose. Furthermore, if the adduct is stable, this approach can determine time-integrated exposures over the lifetime of the modified biopolymer. In the case of protein adducts, this exposure interval corresponds to the lifetime of the red cell (approximately 127 days) for Hb adducts and to the 21-day half-life of serum albumin adducts. Based on continuing daily exposures, this integration over time can lead to an approximate 60-fold amplification in Hb adduct levels and to a 30-fold amplification for serum albumin adduct levels (Skipper and Tannenbaum 1990). DNA adducts in human target tissue, such as the lung, are of particular interest because they may be directly relevant to carcinogenesis, but such tissue is available only by surgery or biopsy. Thus, many analyses have used white blood cell DNA adducts as surrogate markers. Many investigators prefer to analyze adducts in lymphocytes because of their significantly longer lifetimes (up to several years) than the lifetime of less than one day that monocytes and granulocytes have (Kriek et al. 1998). However, these assays are limited by the small amount of DNA that is available in peripheral blood, by the low rates of base modification typically observed, and by the removal of adducts through DNA repair mechanisms. Consequently, studies of adducts in response to the exposure of humans to secondhand smoke have largely focused on the use of protein adducts as surrogate markers because they are more abundant and are not subject to repair mechanisms.

Maclure and colleagues (1989) found that concentrations of both 4AB–Hb and 3AB–Hb adducts were significantly higher in nonsmokers with confirmed exposures to secondhand smoke (based on plasma cotinine concentrations) than in unexposed nonsmokers. The same investigators had previously demonstrated that concentrations of 4AB–Hb were significantly higher in smokers than in nonsmokers, and that the concentrations declined during smoking cessation to levels found in nonsmokers (Bryant et al. 1987; Skipper and Tannenbaum 1990). Hammond and colleagues (1993) found a dose-response relationship for 4AB–Hb concentrations in nonsmokers who were categorized into three levels of exposure to secondhand smoke based on their personal monitoring of nicotine exposure. These authors found that 4AB–Hb concentrations in nonsmokers exposed to secondhand smoke were about 14 percent of those found in smokers, whereas cotinine levels in nonsmokers were about 1 percent of those in smokers. These relative biomarker concentrations are consistent with the higher concentrations of 4AB–Hb and nicotine in sidestream versus mainstream smoke of about 31-fold and 2-fold, respectively (NRC 1986). These results implicate secondhand smoke exposure as a contributing factor to the amount of 4AB adducted to Hb. However, detectable background levels of 4AB–Hb adducts are commonly observed among nonsmokers with no known sources of exposure to secondhand smoke, although they were possibly exposed to other combustion emissions (Bryant et al. 1987; Maclure et al. 1990). As a consequence, the distributions of adduct levels in nonsmokers exposed to secondhand smoke and in those who have no known exposure may not be sharply separated. Additionally, at the time of these studies, secondhand smoke exposure may have been so ubiquitous that few persons were truly unexposed.

In a study of 109 children, 4AB–Hb and PAH–albumin adducts were higher in children whose mothers smoked and in children from households with a smoker other than the mother, compared with children unexposed to secondhand smoke (Crawford et al. 1994; Tang et al. 1999). Cotinine levels also increased with exposure and there were significant

differences among the groups for both biomarkers. After adjusting for the exposure group, the researchers found that these markers were higher among African American children than among Hispanic children. Conversely, in a study of 107 nonsmoking women, Autrup and colleagues (1995) found no significant difference in PAH–albumin levels of those exposed and those unexposed to secondhand smoke. Although serum cotinine measurements confirmed the status of the nonsmokers, the researchers did not compare cotinine and PAH–albumin levels of the participating smokers and nonsmokers. Scherer and colleagues (2000) also found no difference in B[*a*]P adducts of either Hb or albumin in a study of 19 nonsmokers exposed to secondhand smoke and 23 unexposed nonsmokers. This study measured nicotine from personal samplers on individual participants and cotinine levels in both plasma and urine. Cotinine levels were significantly higher among those exposed to secondhand smoke; this finding confirmed the differences in exposure. Additional work may be needed to resolve these findings for the PAH adducts.

Tobacco-Specific Nitrosamines

Tobacco-specific nitrosamines (TSNAs) are of considerable interest as biomarkers of exposure to secondhand smoke because they combine both high specificity for tobacco exposure and additional relevancy as presumed carcinogens. The formation, metabolism, and role of these nitrosamines as significant carcinogens in tobacco smoke were discussed in detail in Chapter 2 (Toxicology of Secondhand Smoke). Several recent studies demonstrated that NNAL and its glucuronide can be measured in the urine of nonsmokers exposed to secondhand smoke (Hecht et al. 1993b; Parsons et al. 1998; Meger et al. 2000; Anderson et al. 2001). There were significant correlations with urine cotinine levels (Hecht et al. 1993b; Parsons et al. 1998) and with nicotine exposures measured with personal samplers (Meger et al. 2000). An additional advantage of NNAL and NNAL-glucuronide as biomarkers is that they are reportedly eliminated more slowly than either nicotine or cotinine in smokers following smoking cessation (Hecht et al. 1999a). Hecht and colleagues (1999a) estimated that the elimination half-life of NNAL was 45 days compared with 40 days for NNAL-glucuronide. If a similar extended half-life can be confirmed in nonsmokers, then these markers may offer the promise of monitoring a longer period of exposure than is possible with either nicotine or cotinine. The main limitation of NNAL measurements is that the concentrations are quite low, even among active smokers, and relatively large urine sample volumes combined with extensive cleanup and sensitive analytical procedures are needed for assays of nonsmokers.

Besides forming urinary metabolites, both 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) and another TSNA, *N'*-nitrosonornicotine, may also form adducts with Hb and DNA that release 4-hydroxy-1-(3-pyridyl)-1-butanone (HPB) on hydrolysis (Hecht et al. 1994). However, the HPB yield has been surprisingly low and was significantly elevated in only a minority of active smokers and in very few nonsmokers. There was also a substantial overlap in values from the samples of both groups. The reason for this finding is unclear; it may reflect individual metabolic differences in Hb alkylation (Hecht et al. 1993a) or limitations in the analytical procedures. If such limitations could be identified and resolved, the analysis of TSNA adducts might offer considerable promise. However, measurements of NNAL and NNAL-glucuronide in urine appear to be the best approach for monitoring exposures to NNK among people exposed to secondhand smoke.

Evidence Synthesis

Biomarkers are valuable for providing an objective index of the internal dose of a component or its metabolite from secondhand smoke following exposure. Biomarkers can be particularly useful in verifying self-reports of exposure to secondhand smoke because individuals may differ in their awareness of the extent and duration of such exposures. Thus, the use of sensitive biomarker measurements may permit the identification of previously unrecognized exposures within nominal control or unexposed groups, and thereby improve the reliability of classifications. However, biomarkers are also limited by interindividual and intraindividual variability, analytical constraints, and limitations on the exposure timeframe that can be monitored.

For example, as tobacco smoke ages and decays, the physical and chemical composition of secondhand smoke changes (NRC 1986), and the ratio of a marker compound such as nicotine to other components of interest may also change. Temporal variations in the ratio of a biomarker to other hazardous compounds in tobacco smoke could thus complicate the interpretation of exposure based on the measurement of that marker. However, as Benowitz (1999) noted, when ratios of nicotine to other constituents such as

respirable suspended particulates are averaged over exposure-time intervals of hours or days, as is typical of a human exposure, the ratios remain consistent. This consistency suggests that biomarkers such as nicotine or its cotinine metabolite should provide a valid assessment of exposure to other toxic constituents in secondhand smoke. Nevertheless, the continual changes in composition during aging will complicate the assessment of tobacco smoke exposure based on one specific marker such as nicotine.

Cotinine measurements in blood or other matrices provide the most useful biomarker for assessing exposure to secondhand smoke because these measurements combine high levels of specificity and sensitivity for exposure. However, as noted above, cotinine measurements reflect an exposure only to nicotine; they are limited to monitoring an exposure over the previous few days unless hair cotinine is measured, and are susceptible to short-term fluctuations that reflect metabolic variations. Even regular smokers may display diurnal variations in plasma cotinine that average 30 percent from peak to trough, with higher concentrations occurring later in the day (Benowitz and Jacob 1994); similar fluctuations may be expected in nonsmokers regularly exposed to secondhand smoke. Cotinine may also reflect an exposure to nicotine previously adsorbed onto dust or emitted from room surfaces rather than a direct exposure to secondhand smoke (Hein et al. 1991), although the extent of this indirect mode of exposure is believed to be trivial (Hein et al. 1991; Benowitz 1999). The interpretation of a result from a single cotinine measurement for an individual is difficult, but multiple measurements over time and mean values from groups within a population may provide useful indices of typical exposure levels. As Benowitz (1999) noted, current evidence "…indicates that cotinine levels provide valid and quantitative measures of average ongoing human ETS [environmental tobacco smoke] exposure over time" (p. 353).

Besides cotinine, other promising biomarkers of involuntary smoking include the tobacco-specific nitrosamine NNAL, the 4AB–Hb adduct, and perhaps hair analysis for nicotine. Each of these markers has the potential to provide an index of exposure over a period of at least several weeks rather than the few days afforded by cotinine, and both NNAL and Hb adducts of aromatic amines are directly relevant as indicators of potential adverse health risks.

Conclusions

1. Biomarkers suitable for assessing recent exposures to secondhand smoke are available.

2. At this time, cotinine, the primary proximate metabolite of nicotine, remains the biomarker of choice for assessing secondhand smoke exposure.

3. Individual biomarkers of exposure to secondhand smoke represent only one component of a complex mixture, and measurements of one marker may not wholly reflect an exposure to other components of concern as a result of involuntary smoking.

Implications

There is a need to refine the methodology used to measure biomarkers to increase their sensitivity and for research into their validity as predictors of population risk. There remains a need for a biomarker capable of reliably indicating past exposures over an extended time period. Until such a marker can be identified, long-term exposures to secondhand smoke can only be assessed through the use of questionnaires and similar approaches.

Conclusions

Building Designs and Operations

1. Current heating, ventilating, and air conditioning systems alone cannot control exposure to secondhand smoke.

2. The operation of a heating, ventilating, and air conditioning system can distribute secondhand smoke throughout a building.

Exposure Models

3. Atmospheric concentration of nicotine is a sensitive and specific indicator for secondhand smoke.

4. Smoking increases indoor particle concentrations.

5. Models can be used to estimate concentrations of secondhand smoke.

Biomarkers of Exposure to Secondhand Smoke

6. Biomarkers suitable for assessing recent exposures to secondhand smoke are available.

7. At this time, cotinine, the primary proximate metabolite of nicotine, remains the biomarker of choice for assessing secondhand smoke exposure.

8. Individual biomarkers of exposure to secondhand smoke represent only one component of a complex mixture, and measurements of one marker may not wholly reflect an exposure to other components of concern as a result of involuntary smoking.

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