The Changing Cigarette: Chemical Studies and Bioassays

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INTRODUCTION In 1950, the first large-scale epidemiological studies on smoking and lung cancer conducted by Wynder and Graham, in the United States, and Doll and Hill, in the United Kingdom, strongly supported the concept of a dose response between the number of cigarettes smoked and the risk for cancer of the lung (Wynder and Graham, 1950; Doll and Hill, 1950).

In 1953, the first successful induction of cancer in a laboratory animal with a tobacco product was reported with the application of cigarette tar[a] to mouse skin (Wynder *et al.*, 1953). The particulate matter of cigarette smoke generated by an automatic smoking machine was suspended in acetone (1:1) and painted onto the shaven backs of mice three times weekly for up to 24 months. A clear dose response was observed between the amount of tar applied to the skin of the mice and the percentage of skin papilloma- and carcinoma-bearing animals in the test group (Wynder *et al.*, 1957). Since then, mouse skin has been widely used as the primary bioassay method for estimating the carcinogenic potency of tobacco tar and its fractions, as well as of particulate matters of other combustion products (Wynder and Hoffmann, 1962, 1967; NCI, 1977a, 1977b, 1977c, 1980; Hoffmann and Wynder, 1977; IARC, 1986a). Intratracheal instillation in rats of the PAH-containing neutral subfraction of cigarette tar led to squamous cell carcinoma of the trachea and lung (Davis *et al.*, 1975). A cigarette tar suspension in acetone painted onto the inner ear of rabbits led to carcinoma with metastasis in thoracic organs (Graham *et al.*, 1957).

Dontenwill and colleagues (1973) developed a method that involved placing Syrian golden hamsters individually into plastic tubes and exposing them to cigarette smoke diluted with air (1:15) twice daily, 5 days a week, for up to 24 months (Dontenwill *et al.*, 1973). The method led to lesions primarily in the epithelial tissue of the outer larynx. Using an inbred strain of Syrian golden hamsters with increased susceptibility of the respiratory tract to carcinogens, long-term exposure to cigarette smoke produced a high tumor yield in the larynx (Bernfeld *et al.*, 1974). A dose response was recorded between the degree of smoke exposure and the induction of benign and malignant tumors in the larynges of the hamsters.

In general, inhalation studies have not found that tobacco smoke leads to squamous cell carcinoma of the lung (Wynder and Hoffmann, 1967; Mohr and Reznik, 1978; IARC, 1986a & b). Dalbey and associates from the National Laboratory in Oak Ridge, Tennessee, exposed female F344 rats to diluted smoke of up to 7 cigarettes daily, 5 times a week for up to 2.5 years. A high percentage of the smoke-exposed rats developed hyperplasia and

[a] Throughout the article, the term "tar" is only used as descriptive noun.

metaplasia in the epithelium of the nasal turbinates and in the larynx, and also some hyperplasia in the trachea. The sham-treated rats developed a small number of lesions in nasal and laryngeal epithelia but none in the trachea. Ten tumors of the respiratory system were observed in 7 out of 80 smoke-exposed rats. These were 1 adenocarcinoma, 1 squamous cell carcinoma in the nasal cavity, 5 adenomas of the lung, 2 alveologenic carcinomas, and 1 squamous cell carcinoma of the lung (Dalbey *et al*., 1980). In the control group of 93 sham-smoked rats, 1 developed an alveologenic carcinoma (Dalbey *et al*., 1980). In 1952, Essenberg reported that cigarette smoke induces an excessive number of pulmonary adenomas; whereas, the sham-exposed mice, as well as the untreated mice, developed significantly lower rates of pulmonary tumors (Essenberg, 1952). In the following years, the Leuchtenbergers repeatedly confirmed the findings by Essenberg. They also demonstrated that even the gas phase increased the occurrence of pulmonary tumors in mice (Leuchtenberger *et al*., 1958; Leuchtenberger and Leuchtenberger, 1970). Several additional studies demonstrated the induction of pulmonary tumors in several strains of mice exposed to diluted cigarette smoke (Mühlbock, 1958; Wynder and Hoffmann, 1967; Mohr and Reznik, 1978; IARC, 1986a & b). Otto exposed mice to diluted cigarette smoke for 60 minutes daily for up to 24 months. Of 30 mice, 4 developed lung adenomas and 1 an epidermoid carcinoma of the lung. In the untreated control group, 3 of 60 mice developed lung adenomas (Otto, 1963).

IDENTIFICATION OF CARCINOGENS, TUMOR PROMOTERS, AND CARCINOGENS IN TOBACCO SMOKE

Green and Rodgman (1996) estimated that there were about 4,800 compounds in tobacco smoke. In addition, additives out of a list of 599 compounds disclosed by tobacco companies (Doull *et al*., 1994) may be added to cigarette tobacco in the process of manufacturing a cigarette in the United States (Green and Rodgman, 1996; Doull *et al*., 1994). Tables 5-1 and 5-2 list the major constituents of the vapor phase (Table 5-1) and the particulate phase (Table 5-2) and their concentrations in the mainstream smoke (MS) of non-filter cigarettes (Hoffmann and Hecht, 1990; Ishiguro and Sugawara, 1980). The agricultural chemicals and pesticides, as well as their specific thermic degradation products, are omitted from the two tables because of the many variations in the nature and amount of these agents in tobacco from country to country and from year to year (Wittekindt, 1985). Table 5-3 lists the major toxic components in the MS of cigarettes (Hoffmann *et al*., 1995).

Development of highly sensitive analytical methods, as well as reproducible short-term and long-term assays, has led to the identification of 69 carcinogens (Table 5-4). Of these, 11 are known human carcinogens (Group I), 7 are probably carcinogenic in humans (Group 2A), and 49 of the animal carcinogens are possibly also carcinogenic to humans (Group 2B). This classification of the carcinogens is according to the International Agency for Research on Cancer (IARC, 1983, 1984, 1986b, 1987, 1988, 1991, 1992, 1994a–e, 1995a & b, 1996, 1999a & b). Two suspected carcinogens have yet to be evaluated by the IARC.

Table 5-1
Major Constituents of the Vapor Phase of the Mainstream Smoke of Non-Filter Cigarettes

Compound[a]	Concentration/Cigarette (% of Total Effluent)
Nitrogen	280-320 mg (56-64%)
Oxygen	50-70 mg (11-14%)
Carbon dioxide	45-65 mg (9-13%)
Carbon monoxide	14-23 mg (2.8-4.6%)
Water	7-12 mg (1.4-2.4%)
Argon	5 mg (1.0%)
Hydrogen	0.5-1.0 mg
Ammonia	10-130 µg
Nitrogen oxides (NO_x)	100-600 µg
Hydrogen cyanide	400-500 µg
Hydrogen sulfide	20-90 µg
Methane	1.0-2.0 mg
Other volatile aromatic alkanes (20)	1.0-1.6 mg[b]
Volatile alkenes (16)	0.4-0.5 mg
Isoprene	0.2-0.4 mg
Butadiene	25-40 µg
Acetylene	20-35 µg
Benzene	12-50 µg
Toluene	20-60 µg
Styrene	10 µg
Other volatile hydrocarbons (29)	15-30 µg
Formic acid	200-600 µg
Acetic acid	300-1,700 µg
Propionic acid	100-300 µg
Methyl formate	20-30 µg
Other volatile acids (6)	5-10 µg
Formaldehyde	20-100 µg
Acetaldehyde	400-1400 µg
Acrolein	60-140 µg
Other volatile aldehydes (6)	80-140 µg
Acetone	100-650 µg
Other volatile ketones (3)	50-100 µg
Methanol	80-180 µg
Other volatile alcohols (7)	10-30 µg
Acetonitrile	100-150 µg
Other volatile nitriles (10)	50-80 µg[b]
Furan	20-40 µg
Other volatile furans (4)	45-125 µg[b]
Pyndine	20-200 µg
Pyridine (3)	15-80 µg
3-Vinylpyridine	10-30 µg
Other volatile pyridines (25)	20-50 µg[b]
Pyrrole	0.1-10 µg
Pyrrolidine	10-18 µg
N-Methylpyrrolidine	2.0-3.0 µg
Volatile pyrazines (18)	3.0-8.0 µg
Methylamine	4-10 µg
Other aliphatic amines (32)	3-10 µg

[a]*Numbers in parentheses represent the individual compounds identified in a given group*
[b]*Estimate*

Table 5-2

Major Constituents of the Particulate Matter of the Mainstream Smoke of Non-Filter Cigarettes

Compound[a]	µg/Cigarette[b]
Nicotine	1.000-3.000
Nornicotine	50-150
Anatabine	5-15
Anabasine	5-12
Other tobacco alkaloids (17)	NA
Bipyridyls (4)	10-30
n-Hentriacontane (*n*-$C_{31}H_{64}$)[c]	100
Total nonvolatile hydrocarbons (45)[c]	300-400[c]
Naphthalene	2-4
Naphthalenes (23)	3-6[c]
Phenanthrenes (7)	0.2-0.4[c]
Anthracenes (5)	0.05-0.1[c]
Fluorenes (7)	0.6-1.0[c]
Pyrenes (6)	0.3-0.5[c]
Fluoranthenes	0.3-0.45[c]
Carcinogenic polynuclear aromatic hydrocarbons (11)[b]	0.1-0.25
Phenol	80-160
Other phenols (45)[c]	60-180[c]
Catechol	200-400
Other catechols (4)	100-200[c]
Other dihydroxybenzenes (10)	200-400[c]
Scopoletin	15-30
Other polyphenols (8)[c]	NA
Cyclotenes (10)[c]	40-70[c]
Quinones (7)	0.50
Solanesol	600-1,000
Neophytadienes (4)	200-350
Limonene	30-60
Other terpenes (200-250)[c]	NA
Palmitic acid	100-150
Stearic acid	50-75
Oleic acid	40-110
Linoleic acid	150-250
Linolenic acid	150-250
Lactic acid	60-80
Indole	10-15
Skatole	12-16
Other indoles (13)	NA
Quinolines (7)	2-4
Other aza-arenes (55)	NA
Benzofurans (4)	200-300
Other *0*-heterocyclic compounds (42)	NA
Stigmasterol	40-70
Sitosterol	30-40
Campesterol	20-30
Cholesterol	10-20
Aniline	0.36
Toluidines	0.23
Other aromatic amines (12)	0.25
Tobacco-specific *N*-nitrosamines (6)	0.34-2.7
Glycerol	120

[a]Numbers in parentheses represent individual compounds identified.
[b]For details, See Table 5-4
[c]Estimate. NA=Not available.

Table 5-3
Major Toxic Agents in Cigarette Smoke[a]

Agent	Concentration/ Non-Filter Cigarette	Toxicity
Carbon monoxide	10-23 mg	Binds to hemoglobin, inhibits respiration
Ammonia	10-130 µg	Irritation of respiratory tract
Nitrogen oxide (NO_x)	100-600 µg	Inflamation of the lung
Hydrogen cyanide	400-500 µg	Highly ciliatoxic, inhibits lung clearance
Hydrogen sulfide	10-90 µg	Irritation of respiratory tract
Acrolein	60-140 µg	Ciliatoxic, inhibits lung clearance
Methanol	100-250 µg	Toxic upon inhalation and ingestion
Pyridine	16-40 µg	Irritates respiratory tract
Nicotine[b]	1.0-3.0 mg	Induces dependence, affects cardiovascular and endocrine systems
Phenol	80-160 µg	Tumor promoter in laboratory animals
Catechol	200-400 µg	Cocarcinogen in laboratory animals
Aniline	360-655 µg	Forms methemoglobin, and this affects respiration
Maleic hydrazide	1.16 µg	Mutagenic agent

[a]*This is an incomplete list.*
[b]*Toxicity: oral/rat, LD_{50} free nicotine 50 mg/kg, nicotine bitartrate 65 mg/kg.*
Source: Hoffmann et al., 1998.

Table 5-4
Carcinogens in Cigarette Smoke

Agent	Conc./Non-filter Cigarette	IARC Evaluation Evidence of Carcinogenicity in Lab Animals	in Humans	Group[a]
PAH				
Benz(*a*)anthracene	20-70 ng	Sufficient		2A
Benzo(*b*)fluoranthene	4-22 ng	Sufficient		2B
Benzo(*j*)fluoranthene	6-21 ng	Sufficient		2B
Benzo(*k*)fluoranthene	6-12 ng	Sufficient		2B
Benzo(*a*)pyrene	20-40 ng	Sufficient	Probable	2A
Dibenz(*a,h*)anthracene	4 ng	Sufficient		2A
Dibenzo(*a,l*)pyrene	1.7-3.2 ng	Sufficient		2B
Dibenzo(*a,e*)pyrene	Present	Sufficient		2B
Indeno(1,2,3-*cd*)pyrene	4-20 ng	Sufficient		2B
5-Methylchrysene	0.6 ng	Sufficient		2B
Heterocyclic Compounds				
Quinoline[b]	1-2 ng			
Dibenz(*a,h*)acridine	0.1 ng	Sufficient		2B
Dibenz(*a,j*)acridine	3-10 ng	Sufficient		2B
Dibenzo(*c,g*)carbazole	0.7 ng	Sufficient		2B
Benzo(*b*)furan	Present	Sufficient		2B
Furan	18-37 ng	Sufficient		2B

Table 5-4 (continued)

Agent	Conc./Non-filter Cigarette	IARC Evaluation Evidence of Carcinogenicity in Lab Animals	in Humans	Group[a]
***N* -Nitrosamines**				
N -Nitrosodimethylamine	2-180 ng	Sufficient		2A
N -Nitrosoethylmethylamine	3-13 ng	Sufficient		2B
N -Nitrosodiethylamine	ND-2.8 ng	Sufficient		2A
N -Nitrosodi-*n*-propylamine	ND-1.0 ng	Sufficient		2B
N -Nitroso-di-*n*-butylamine	ND-30 ng	Sufficient		2B
N -Nitrosopyrrolidine	3-110 ng	Sufficient		2B
N -Nitrosopiperidine	ND-9 ng	Sufficient		2B
N -Nitrosodiethanolamine	ND-68 ng	Sufficient		2B
N -Nitrosonornicotine	120-3,700 ng	Sufficient		2B
4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone	80-770 ng	Sufficient		2B
Aromatic Amines				
2-Toluidine	30-337 ng	Sufficient		2B
2,6-Dimethylaniline	4-50 µg	Sufficient		2B
2-Naphthylamine	1-334 ng	Sufficient	Sufficient	1
4-Aminobiphenyl	2-5.6 ng	Sufficient	Sufficient	1
***N* -Heterocyclic Amines**				
AaC	25-260 ng	Sufficient		2B
IQ	0.3 ng	Sufficient		2B
Trp-P-1	0.3-0.5 ng	Sufficient		2B
Trp-P-2	0.8-1.1 ng	Sufficient		2B
Glu-P-1	0.37-0.89 ng	Sufficient		2B
Glu-P-2	0.25-0.88 ng	Sufficient		2B
PhIP	11-23 ng	Sufficient	Possible	2A
Aldehydes				
Formaldehyde	70-100 µg	Sufficient	Limited	2A
Acetaldehyde	500-1,400 µg	Sufficient	Insufficient	2B
Volatile Hydrocarbons				
1,3-Butadiene	20-75 µg	Sufficient	Insufficient	2B
Isoprene	450-1,000 µg	Sufficient		2B
Benzene	20-70 µg	Sufficient	Sufficient	1
Styrene	10 µg	Limited		2B
Misc. Organic Compounds[c]				
Acetamide	38-56 µg	Sufficient		2B
Acrylamide	Present	Sufficient		2B
Acrylonitrile	3-15 µg	Sufficient	Limited	2A
Vinyl chloride	11-15 ng	Sufficient	Sufficient	1
DDT	800-1,200 µg	Sufficient	Probable	2B
DDE	200-370 µg	Sufficient		2B
Catechol	100-360 µg	Sufficient		2B
Caffeic acid	< 3 µg	Sufficient		2B
1,1-Dimethylhydrazine	Present	Sufficient		2B
Nitromethane	0.3-0.6 µg	Sufficient		2B

Table 5-4 (continued)

Agent	Conc./Non-filter Cigarette	IARC Evaluation Evidence of Carcinogenicity in Lab Animals	in Humans	Group[a]
2-Nitropropane	0.7-1.2 µg	Sufficient		2B
Nitrobenzene	25 µg	Sufficient		2B
Ethyl carbamate	20-38 µg	Sufficient		2B
Ethylene oxide	7 µg	Sufficient	Sufficient	1
Propylene oxide	12-100 ng	Sufficient		2B
Methyleugenol	20 ng			
Inorganic Compounds				
Hydrazine	24-43 ng	Sufficient	Inadequate	2B
Arsenic	40-120 µg	Inadequate	Sufficient	1
Beryllium	0.5 ng	Sufficient	Sufficient	1
Nickel	ND-600 ng	Sufficient	Sufficient	1
Chromium (only hexavalent)	4-70 ng	Sufficient	Sufficient	1
Cadmium	7-350 ng	Sufficient	Sufficient	1
Cobalt	0.13-0.2 ng	Sufficient	Inadequate	2B
Lead	34-85 ng	Sufficient	Inadequate	2B
Polonium-210	0.03-1.0 pCi	Sufficient	Sufficient	1

*Abbreviations: ND, not detected; PAH, polynuclar aromatic hydrocarbons; AaC, 2-amino-9*H*-pyrido[2,3-*b*]indole; IQ, 2-amino-3-methylimidazo[4,5-*b*]quinoline; Trp-P-1, 3-amino-1,4-dimethyl-5H-pyrido[4,3-*b*]indole; Trp-2, 3-amino-1-methyl-5H-pyrido[4,3-*b*]indole; Glue-P-1, 2-amino-6-methyl[1,2-*a*:3',2"-*d] imidazole; Glu-P-2, 2-aminodipyrido[1,2-*a*:3'2"-*d*]imidazole; PhIP, 2-amino-1-methyl-6-phenylimidazo [4,5-*b*]pyridine.*

[a]IARC Monographs on the Evaluation of Carcinogenic Risks. *Volume 1 and Supplements 1-8, 1972-1999. (1) Human carcinogens; (2A) Probably carcinogenic in humans; (2B) Possibly carcinogenic to humans; (3) Not classifiable as to their carcinogenicity to humans.*

[b]*Unassigned carcinogenicity status by IARC at this time*

[c]*In 1982, the IARC assigned di(2-ethylhexyl)phthalate as sufficient to Group 2B. However, in 2000, re-evaluation of the carcinogenicity was classified as not carcinogenic (IARC 1982, 2000). We cited di(2-ethylhexyl)phthalate as a 2B carcinogen (Hoffmann and Hoffmann, 1997) and in this article, it is deleted from Table 5-4, "Carcinogenicity in Cigarette Smoke."*

Sources: International Agency for Research on Cancer, 1982, 2000.

SMOKING CONDITIONS

In 1936, the American Tobacco Company began using standard machine smoking conditions, which, to some extent, reflected the smoking habits of cigarette smokers at that time. The estimated sales-weighted average nicotine yields of the cigarettes smoked at that time were around 2.8 mg (Bradford *et al.*, 1936). In agreement with the U.S. tobacco industry, the Federal Trade Commission (FTC) adapted the 1936 standard method in 1969 with only slight modifications. Since then, machine-smoking conditions are one puff/minute with a volume of 35 ml drawn during 2 seconds, leaving a butt length of 23 mm for a non-filter (plain) cigarette, and length of the filter and overwrap plus 3 mm for a filter cigarette (Pillsbury *et al.*, 1969). In Canada and the United Kingdom, the standard smoking conditions of the International Standards Organization (ISO) have been accepted since 1991 (ISO, 1991). In other European countries, the standard smoking conditions for cigarettes are those developed by CORESTA (Centre De Cooperation Pour Les Recherches Scientifiques Relative Au Tabac), which are similar to the FTC standard smoking conditions (CORESTA, 1991). In Japan, the FTC standard smoking conditions are employed for the machine smoking of cigarettes (Pillsbury *et al.*, 1969). The FTC method defines tar as smoke particulates minus water and nicotine, whereas CORESTA defines tar as total particulates minus water (Pillsbury *et al.*, 1969; ISO, 1991; CORESTA, 1991). The standard conditions for machine

smoking of tobacco products used by the different testing protocols are presented in Table 5-5. Using the FTC method, the sales-weighted average tar and nicotine yields of U. S. cigarettes decreased from about 37 mg and 2.7 mg in 1954 to 12 mg and 0.85 mg in 1993 (Figure 5-1).

More than 20 years ago, M.A.H. Russell in the United Kingdom and N.L. Benowitz in the United States reported that long-term smokers of cigarettes with lower nicotine yields took more than one puff per minute, drew puff volumes exceeding 35 ml, and inhaled the smoke more deeply than smokers of higher yield cigarettes (Russell, 1976, 1980; Benowitz *et al.*, 1983).

Table 5-6 presents the smoking characteristics of 56 volunteer smokers who regularly consumed low-yield cigarettes (≤ 0.8 mg nicotine/cigarette according to the FTC smoking machine method) and of 77 volunteer smokers regularly consuming medium-nicotine cigarettes (FTC, 0.9–1.2 mg/cigarette). These two ranges of nicotine yield constituted more than 73.4 percent of all cigarettes smoked in the U.S. in 1993 (FTC, 1995). The results of this study clearly indicated that the majority of U.S. smokers smoked their cigarettes much more intensely to satisfy their acquired need for nicotine. Comparing the yields of the same cigarettes smoked under FTC standard machine smoking conditions with the smoke inhaled by the consumers of cigarettes with low- and medium-nicotine content revealed that smokers inhaled 2.5 and 2.2 times more nicotine/cigarette, 2.6 and 1.9 times more tar, 1.8 and 1.5 times more carbon monoxide, 1.8 and 1.6 times more BaP, and 1.7 and 1.7 times more NNK than is generated by the FTC machine-smoking method (Table 5-6; Djordjevic *et al.,* 2000).

The discrepancy in exposure assessment between recent measurements and former interpretations of machine-smoking data has led to criticism of the FTC standard machine smoking method for consumer guidance. The suggestion that there is a meaningful quantitative relationship between the FTC-measured yields and actual intake (by the cigarette smoker) is misleading (Benowitz, 1996). In view of these concerns, it appears "that the time has come for meaningful information on the yields of cigarettes" (Wilkenfeld *et al.*, 2000a & b). The FTC agrees, in principle, that a better and more comprehensive test program for cigarettes is needed (Peeler and Butters, 2000).

CHANGES IN CIGARETTE SMOKE COMPOSITION WITH VARIOUS DESIGN CHANGES

Filter Tips

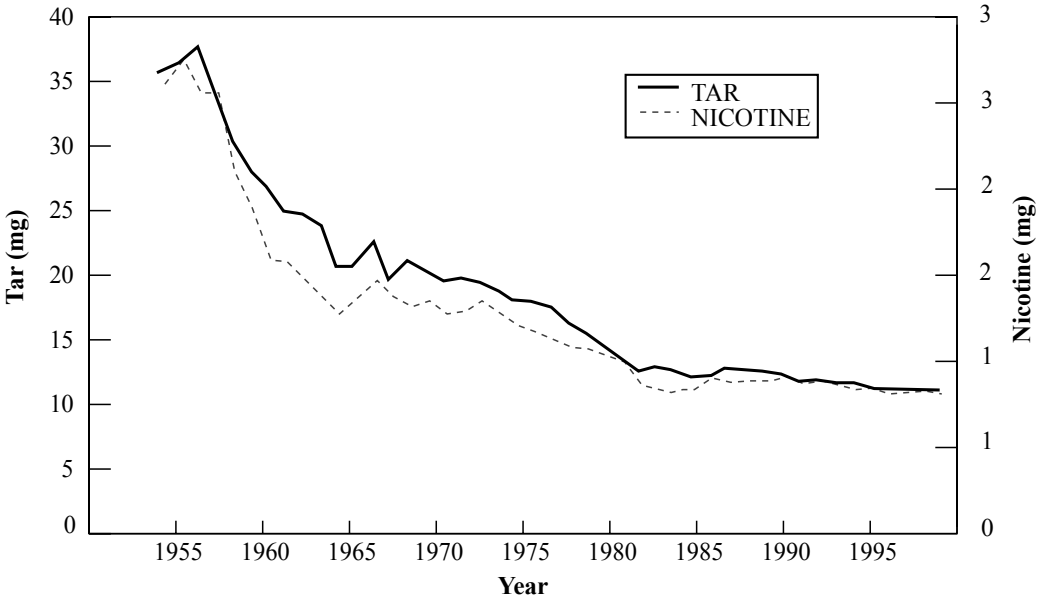
In 1959, Haag *et al*. reported the selective reduction of volatile smoke constituents by filtration through charcoal filter tips (Haag *et al.*, 1959). Several of the compounds that are selectively removed from mainstream smoke (MS) in this fashion are major ciliatoxic agents, such as hydrogen cyanide, formaldehyde, acrolein, and acetaldehyde. Charcoal filters reduce the MS levels of these agents by up to 66 percent (Kensler and Battista, 1966; Tiggelbeck, 1976; Battista, 1976). However, for tar reduction, charcoal filters are less efficient than cellulose acetate filters. Several types of combination filters are in use. The early charcoal-activated dual and triple filter tips were cellulose acetate filters with embedded charcoal powder or granulated char-

Table 5-5
Standard Conditions for Machine Smoking of Tobacco Product

Parameters	Cigarettes		Bidis	Little Cigars	Small Cigars	Cigars	Premium	Pipes
	FTC	CORESTA		FTC	CORESTA	CORESTA	CORESTA	CORESTA
Weight (g)	0.8-1.1	0.8-1.1	0.55-0.80	0.9-1.3	1.3-2.5	5-17	6-20	*
Puff								
Frequency (sec)	60.0	60.0	30.0	60.0	40.0	40.0	40.0	20.0
Duration (sec)	2.0	2.0	2.0	2.0	1.5	1.5	1.5	2.0
Volume (ml)	35.0	35.0	35.0	35.0	40.0	40.0	40.0	50.0
Butt length (mm)								
Non-filter	23.0	23.0	23.0	23.0	33.0	33.0	33.0	
Filter	F&OW+3	F+8	F&OW+3					

Abbreviations: FTC = Federal Trade Commission method; CORESTA = Centre de Cooperation Pour Les Reherches Scientifiques Relative au Tabac Method; F = filter tip; OW = overwrap.
**One gram of pipe tobacco smoked.*
Sources: Hoffmann et al., 1974; International Committee for Cigar Smoking, 1974; Miller, 1963.

Figure 5-1
Sales-Weighted Tar and Nicotine Values for U.S. Cigarettes as Measured by Machine Using the FTC Method, 1954*-1998



**Values before 1968 are estimated from available data.*

coal sandwiched between cellulose acetate segments. These filters have been improved by innovative filter designs, incorporating cellulose acetate, charcoal, and cigarette filter paper (Shepherd, 1994). In the United States, however, cigarettes with charcoal filters have accounted for only about 1 percent of all cigarette sales over the past 15 years. In most developed coun-

Table 5-6
A Comparison of Smoke Data for Two Low-Yield U.S. Filter Cigarettes Smoked According to the FTC-Method and by Smokers

Parameters	FTC Machine Smoking	Cigarette Smokers FTC 0.6-0.8 Nicotine	Cigarette Smokers FTC 0.9-1.2 Nicotine
Puff			
Volume (ml)	35.0	48.6 (45.2-52.3)[a]	44.1 (40.8-46.8)[b]
Interval (sec)	58.0	21.3 (19.0-23.8)[a]	18.5 (16.5-20.6)[b]
Duration (sec)	2.0	1.5 (1.4-1.7)[a]	1.5 (1.4-1.6)[b]
Nicotine (mg/cig)	0.7 (0.6-0.8)	1.74 (1.54-1.98)[c]	
	0.1 (1.09-1.13)		2.39 (2.20-2.60)[d]
Tar (mg/cig)	8.5 (7.7-9.5)	22.3 (18.8-26.5)[e]	
	15.4 (14.2-14.9)		29.0 (25.8-32.5)[f]
CO (mg/cig)	9.7 (9.0-10.4)	17.3 (15.0-20.1)[g]	
	14.6 (14.2-14.9)		22.5 (20.3-25.0)[h]
Ba P (ng/cig)	10 (8.2-12.3)	17.9 (15.3-20.9)[i]	
	14 (10.1-19.4)		21.4 (19.2-23.7)[j]
NNK (ng/cig)	112.9 (96.6-113.0)	186.5 (158.3-219.7)[i]	
	146.2 (132.5-165.5)		250.9 (222.7-282.7)[j]

Test Groups: [a]56 smokers; [b]71 smokers; [c]30 smokers; [d]42 smokers; [e]18 smokers; [f]19 smokers; [g]15 smokers; [h]16 smokers; [i]6 smokers; [j]3 smokers.
Source: Djordjevic et al.*, 2000.*

tries, charcoal filter cigarettes have accounted for, at most, a small percentage of the open cigarette market, exceptions are Japan, South Korea, Venezuela and Hungary, where at least 90 percent of the cigarettes have charcoal filter tips (John, 1996; Fisher, 2000).

Cellulose acetate filter cigarettes first became popular in Switzerland, during the early 1950s, and soon thereafter in Germany. Their popularity spread to the U.S., the UK and Japan, and finally to France. In 1956, the market share of filter cigarettes in Switzerland was 57.2 percent, in Germany 16.7 percent, and in the USA 29.6 percent, with only a few percent in Japan, England, and France. By 1965, the filter cigarette market share in these countries had risen to about 82 percent, 80 percent, 63 percent, 50 percent , 52 percent , and 21 percent, respectively. At this time, cellulose acetate filter cigarettes accounted for at least 95 percent of the cigarette markets in all of the developed countries, except France, where filter cigarettes remained at 85 percent of all cigarette sales (Wynder and Hoffmann, 1994; Waltz and Häusermann, 1963; Hoffmann and Hoffmann, 1997).

In the early 1960s, investigators found that cellulose acetate filter tips retained up to 80 percent of the volatile phenols from the smoke. Reduction of the emissions of volatile phenols from cigarettes was desirable because their tumor promoting activity had been demonstrated in carcinogenesis assays (Roe *et al*., 1959; Wynder and Hoffmann, 1961; Hoffmann and Wynder, 1971). When tested on a gram-to-gram basis, the tar from cellulose acetate-filtered smoke is somewhat more toxic, but less carcinogenic, than tars obtained from charcoal-filtered smoke or from the smoke of non-

filter cigarettes (Hoffmann and Wynder, 1963; Spears, 1963; Wynder and Mann, 1957; Bock *et al.*, 1962; NCI, 1977c). Cellulose acetate filter tips also selectively remove up to 75 percent of the carcinogenic volatile *N*-nitrosamines (VNA), whereas charcoal-filter tips are much less effective in removing VNA (Brunnemann *et al.*, 1977). Exposure of Syrian golden hamsters twice daily, 5 days per week, for over 60 weeks to the diluted smoke from two different cellulose acetate filter cigarettes elicited a significantly lower incidence of carcinoma of the larynx than exposure to the diluted smoke from the non-filter cigarette ($p<0.01$). In contrast, the incidence rate of carcinoma of the larynx of hamsters exposed to diluted smoke from charcoal-filter cigarettes did not differ significantly from that of larynx carcinoma in hamsters exposed to diluted smoke from the non-filter cigarette (Dontenwill *et al.*, 1973).

Filter perforation allows air dilution of smoke during puff drawing. The velocity of airflow through the burning cone of cigarettes with perforated filters is slowed down because the negative pressure generated by drawing a puff is reduced by drawing air through the filter perforations, and the pressure drop across the tobacco rod is reduced, thus slowing the flow of smoke through the rod. This results in more complete combustion of the tobacco and a higher retention of particulate matter by cellulose acetate in the filter tip (Hoffmann and Hoffmann, 1997; Norman *et al.*, 1984; Durocher, 1984; Baker, 1984). Presently, more than 50 percent of all cigarettes have perforated filter tips. Table 5-7 compares smoke yields of cigarettes without filter tips, cigarettes with cellulose acetate filter tips, and cigarettes with cellulose acetate filter tips that are perforated. The filling tobaccos of these experimental cigarettes were made of an identical blend. The conventional filter tip of cellulose acetate retains more tar, nicotine, and phenol, but releases more CO and ciliatoxic agents, hydrogen cyanide, acetaldehyde, and acrolein than does the cigarette with the perforated filter tip (NCI, 1977c). In mouse skin assays, the tars from both types of filter cigarettes have comparable tumorigenic activity. However, one needs to bear in mind that (a) these comparative data are generated with tars obtained by the standardized machine smoking method, with a 35-ml puff, taken once a minute over 2 seconds; (b) more than 60 percent of today's smokers in the United States and in many developed countries smoke cigarettes with nicotine yields of only 1.2 mg or less (according to FTC standards of smoking); and (c) most of these smokers compensate for the low nicotine delivery.

Compensation and greater smoke intake is governed by the smoker's acquired need for nicotine and, in essence, negates the intended benefits of reducing smoke yields by technical means (Russell, 1976, 1980; Benowitz *et al.*, 1983; Benowitz and Henningfield, 1994; Schultz and Seehofer, 1978; Moody, 1980; Herning *et al.*, 1981; Gritz *et al.*, 1983; Nil *et al.*, 1986; Djordjevic *et al.*, 1995, 2000; see Chapter 2).

Paper Porosity Since about 1960, higher cigarette paper porosity and treatment of paper with citrate have significantly contributed to the reduction of smoke yields of several smoke components. During and in between puff drawing, porous paper enhances the outward diffusion through the paper of hydrogen, NO, CO, CO_2, methane, ethane, and ethylene. On the other hand, it

Table 5-7
Comparison of Experimental Cigarettes (Yield/Cigarette)[a,b]

Smoke Components	Unit of Measurement	Non-Filter Cigarette	Cellulose Acetate Filter Cigarette	Cellulose Acetate Filter w/Perforation	Cellulose Acetate Filter w/Perforation & Highly Porous Paper
Carbon monoxide	ml	16.2	19.2	8.62	6.66
Hydrogen cyanide	µg	368	296	201	109
Nitrogen oxides-NOx	µg	406	438	364	224
Formaldehyde	µg	36.0	20.9	31.7	21.4
Acetaldehyde	µg	1,040	1,290	608	550
Acrolein	µg	105	104	58.6	48.6
Tar	mg	27.0	14.7	19.2	19.5
Nicotine	mg	1.8	0.94	1.31	1.5
Phenol	µg	161	61.7	122	129
Benz(a)anthracene	µg	40.6 [1.40]	35.3 [2.25]	38.5 [1.88]	40.1 [1.91]
Benzo(a)pyrene	ng	29.9 [1.09]	19.6 [1.25]	29.2 [1.13]	23.9 [1.14]

[a]*The composition of the cigarette tobacco is identical in all four experimentsal cigarettes*
[b]*Numbers in square brackets = µg/dry tar*
Source: National Cancer Institute, 1977c.

accelerates the diffusion of O_2 and N_2 into the tobacco column; this, in turn, causes more rapid smoldering during puff intervals (Hoffmann and Hoffmann, 1997; Owens, 1998). Porous cigarette paper causes a significant decrease of CO, hydrogen cyanide, nitrogen oxides, volatile aldehydes; yet, it hardly changes the yields of tar, nicotine, benz(*a*)anthracene (BaA), and BaP. Importantly, the significant reduction of nitrogen oxides in the smoke of these cigarettes reduces the formation and, thus, significantly lowers the yields of volatile and tobacco-specific *N*-nitrosamines (TSNA) (Owens, 1998; Brunnemann *et al.*, 1994).

Cigarette Construction Smoke yields of cigarettes are also dependent on physical parameters, such as length and circumference of the cigarette, and the width of the cut (number of cuts per inch) of the tobacco filler. Extending the cigarette length from 50 mm to 130 mm produces an increase in the level of oxygen in the mainstream smoke, while the absolute levels of hydrogen, carbon monoxide, methane, ethane, and ethylene decrease. The major reason for this lies in the diffusion of oxygen through the paper into the smoke stream (Terrell and Schmeltz, 1970). This phenomenon is also reflected in an increased CO delivery with ascending number of puffs because the available surface area of the paper diminishes as the cigarette is smoked. With increasing length of the cigarette, the overall yields of tar, nicotine, PAH, and other particulate components increase (DeBardeleben *et al.*, 1978). A circumference of cigarettes smaller than the regular 24.8–25.5 mm (*e.g.*, 23 mm or less) translates into less tobacco being burned and a greater volume of oxygen available during combustion. Thus, the smoke yields of tar, nicotine, and other particulate components are lowered (DeBardeleben *et al.*, 1978; Lewis, 1992; Brunnemann *et al.*, 1994; Hoffmann and Hoffmann, 1997). Cigarettes with small circumference also have a lower ignition propensity toward inflammable materials than ciga-

rettes that have a 24.8- to 25.5-mm circumference. It has been estimated that in all fire deaths in the U.S. in 1997, 30% were caused by smoking, and worldwide, 10% (Lecstikoo *et al*., 2000).

The number of cuts per inch (width of tobacco strands) applied to the filler tobacco of cigarettes has an impact on smoke yields and/or on the carcinogenicity of the tars. The first investigation on the importance of tobacco cuts per inch, with regard to smoke yields and tumorigenicity of the resulting tars, was published in 1965 (Wynder and Hoffmann, 1965). It compared the smoke yields of tar and BaP when 8, 30, 50, and 60 cuts per inch of tobacco were applied. Tar yields per cigarette decreased from 29.1 to 23.0 mg and B*a* P from 37 to 21 ng. The tumorigenicities of tars derived from cigarettes made with 8, 30, or 50 cuts per inch of tobacco declined from 27 percent to 16 percent and 13 percent of tumor-bearing mice. In a large-scale study of cigarettes filled with an identical blend, cut 20 and 60 times per inch, the smoke yields per cigarette of tar, nicotine, volatile aldehydes, BaA, and BaP were significantly reduced for the fine-cut. However, hydrogen cyanide was insignificantly increased. Gram-to-gram comparison of tumorigenicities of both tars on mouse skin revealed statistically insignificant differences (NCI, 1977a). As the large-scale bioassay was repeated twice, one has to conclude that, in terms of mouse skin carcinogenicity, activities of tars obtained from coarse-cut and fine-cut tobaccos are comparable.

Tobacco Types The botanical genus *Nicotiana* has two major subgenera: *N. rustica* and *N. tabacum*. *Nicotiana rustica* is primarily grown in Russia, the Ukraine, and other East European countries, including Georgia, Moldavia, and Poland. It is also grown in South America and, to a limited extent, in India. In the rest of the world, *Nicotiana tabacum* is grown as the major tobacco crop; it is classified into flue-cured type (often called bright, blond, Virginia or Maryland tobacco), air-cured type (often called burley tobacco; light air-cured tobacco grown in Kentucky, and dark air-cured type grown in parts of Tennessee and Kentucky, South America, Italy, and France) and sun-cured type (often called oriental tobacco; primarily grown in Greece and Turkey). In addition, there are special classes of air-cured tobaccos for cigars, chewing tobacco, and snuff (Tso, 1990).

Prior to the last two decades, flue-cured tobaccos were used exclusively for cigarettes in the United Kingdom and in Finland; they were also the predominate type used in Canada, Japan, China, and Australia. Air-cured tobaccos are preferred for cigarettes in France, southern Italy, some parts of Switzerland and Germany, and South America. Cigarettes made exclusively from sun-cured tobaccos are popular in Greece and Turkey. In the rest of Western Europe and in the United States, cigarettes contain blends of flue-cured and air-cured tobaccos as major components. Today, in many countries, such as the United Kingdom, France, and other developed nations, the U.S. blended cigarette is gaining market share. In the United States, the composition of the cigarette blend has undergone gradual changes. In the 1960s and early 1970s, 45–50 percent of the cigarette blend were flue-cured (Virginia) tobaccos, 35 percent were air-cured (burley) tobaccos, and a few

percent were Maryland air-cured and oriental tobaccos. By 1980, the average blend was composed of 38 percent flue-cured, 33 percent air-cured, and a few percent each of Maryland and oriental tobaccos. In the early 1990s, these proportions were about 35 percent, 30 percent, and, again, a few percent of Maryland and oriental tobaccos (Hoffmann and Hoffmann, 1997; Spears and Jones, 1981). The blended cigarette is preferred in many countries, in part because each of the three major *N. tabacum* types adds a certain aroma to the smoke. Some isoprenoids, and a relatively high number of agents with carboxyl content, are associated with the aroma of flue-cured tobacco. Other isoprenoids, and especially the composition of the acidic fraction, are related to the special aroma of air-cured tobaccos (Roberts and Rowland, 1962; Enzell, 1976; Spears and Jones, 1981; Tso, 1990). 3-Methylbutanoic acid (isovaleric acid) is considered to impart the most important flavor characteristic to oriental tobacco (Stedman *et al.*, 1963; Schumacher, 1970).

However, in regard to the toxicity and carcinogenicity of tobacco and tobacco smoke, the difference in the nitrate content of the tobaccos is of primary significance. Flue-cured tobacco can contain up to 0.9 percent of nitrate; yet, as it is used for regular cigarettes, it contains less than 0.5 percent of NO_3. In oriental tobaccos, one finds up to 0.6 percent of NO_3, in air-cured tobaccos between 0.9 percent and 5.0 percent, but generally below 3 percent in commercial cigarettes. The highest concentration of nitrate is present in the ribs, and the lowest concentration is in the laminae, especially in the laminae harvested from the top stalk positions of the tobacco plant (Neurath and Ehmke, 1964; Tso *et al.*, 1982). With the utilization of a greater proportion of air-cured tobacco in the U.S. cigarette tobacco blend, the nitrate content of the blended U.S. cigarette tobacco has risen from about 0.5 percent in the 1950s to 1.2–1.5 percent in the late 1980s (U.S. DHHS, 1989).

The concentrations of nitrogen oxides (NO_x) and methyl nitrite in smoke depend primarily on the nitrate concentrations in the tobacco, even though a portion of the nitrogen oxides is also formed during smoking from amino acids and certain proteins (Philippe and Hackney, 1959; Sims *et al.*, 1975; Norman *et al.*, 1983). Cigarettes made with flue-cured tobaccos deliver up to 200 µg of NO_x and 20 µg methyl nitrite in the smoke. Smoking U.S. blended cigarettes produces up to 500 µg NO_2 and 200 µg methyl nitrite, and the smoke of air-cured tobacco cigarettes contains up to 700 µg NO_x and 400 µg methyl nitrite. The major source of nitrate is air-cured tobacco and, thus, the major source of NO_x in its smoke is the nitrogen fertilizers (Sims *et al.*, 1975). The stems of air-cured tobaccos are especially rich in nitrate (≤6.8 percent). Consequently, stems, as components of expanded and reconstituted tobaccos, contribute in a major way to NO_x in the smoke (Brunnemann *et al.*, 1983).

Freshly generated smoke, as it leaves the mouthpiece of a cigarette, contains NO_x virtually only in the form of nitric oxide (NO), and contains practically no nitrogen dioxide (NO_2). However, nitrogen dioxide is quickly formed upon aging of the smoke. It has been estimated that, within 500 seconds half of the NO in undiluted smoke is oxidized to NO_2 (Neurath,

1972). Of major importance is the high reactivity of NO_x upon its formation in the burning cone and in the hot zones of a cigarette. The thermically activated nitrogen oxides serve as scavengers of C,H- radicals, whereby they inhibit the pyrosynthesis of carcinogenic polynuclear aromatic hydrocarbons. Table 5-8 presents data on the smoke yields of tar, nicotine, phenol, and BaP, and the tumorigenicities of the tars on mouse skin (Wynder and Hoffmann, 1963).

Freshly generated nitrogen oxides also react with secondary and tertiary amines to form volatile *N*-nitrosamines (VNA) and several *N*-nitrosamines from amino acids, as well as from additives. The NO_x also form tobacco-specific *N*-nitrosamines (TSNA) by *N*-nitrosation of nicotine and of the minor tobacco alkaloids (Brunnemann *et al*., 1977; Brunnemann and Hoffmann, 1981; Tsuda and Kurashima, 1991; Hoffmann *et al*., 1994). BaP declined while NNK increased in the smoke of a leading U.S. non-filter cigarette between 1974 and 1997. Both trends correlate with the use of tobaccos with higher nitrate content. Recently, it was suggested that the formation of tobacco-specific nitrosamines in flue-cured tobacco in the United States is, in part, due to the use of propane gas heaters in the curing process. Oxides of nitrogen generated during the burning of the liquid propane react with nicotine in the tobacco leaf to form TSNA. This change in the curing method, introduced in the mid 1960s, is a likely contributor to the increase of TSNA levels in cigarette tobacco. Other important factors are the proportionally greater use of air-cured tobacco and the use of reconstituted tobaccos in the cigarette tobacco blend (Neurath and Ehmke, 1964; Brunnemann *et al*., 1983; Peel *et al*., 2001). Increased amounts of TSNA in tobacco compound the carcinogenic potency of the resulting cigarette smoke (Hoffmann *et al*., 1994) and are considered to contribute to the rise of adenocarcinoma, which has become the dominant form of lung cancer in both male and female smokers during the last three decades (Vincent *et al*., 1977; Cox and Yesner, 1979; el-Torkey *et al*., 1990; Devesa *et al*., 1991; Stellman *et al*., 1997). Increasing concentrations of nitrate in tobacco have also led to an

Table 5-8
Smoke Yields and Tumorigenicity of the Tars from the Four Major *N. tabacum* Varieties

Factors	Flue-Cured Tobacco	Sun-Cured Tobacco	Air-Cured Tobacco	
			Kentucky[a]	Maryland
A: Yields/Cigarette				
Tar (mg)	33.4	31.5	25.6	21.2
Nicotine (mg)	2.4	1.9	1.2	1.1
Phenol (µg)	95	120	60	43
Benzo(*a*)pyrene (ng)	53 (1.6)[b]	44 (1.4)[b]	24 (0.94)[b]	18 (0.85)[b]
B: Tumorigenicity[c]				
Percentage of mice with skin tumors	34	35	23	18

[a]*Low-nicotine, air-cured tobacco (Kentucky)*
[b]*Number in parentheses = µg BaP/g dry tar*
[c]*Bioassayed on a gram-to-gram basis of tar*
Source: Wynder and Hoffmann, 1963.

increase in cigarette smoke of the human bladder carcinogens 2-naphthylamine and 4-aminobiphenyl and of other aromatic amines (Patrianakos and Hoffmann, 1979; Grimmer *et al.*, 1995).

An important aspect relative to the toxicology of cigarette smoke is the correlation between the nitrate content of tobacco and the pH of cigarette smoke. Even though the different processes used to flue-cure and air-cure tobaccos have a significant impact on the smoke composition of the major types of tobacco, the role of nitrate is of major importance in determining the pH of the smoke. Whereas flue-cured tobacco and U.S. cigarette tobacco blends deliver weakly acidic smoke (pH 5.8–6.3), the smoke of cigarettes made from air-cured tobacco delivers neutral to weakly alkaline smoke (pH 6.5–7.5). A major reason for the range of pH values encountered in the smoke of the two major tobacco types is the concentration of ammonia in the smoke, which is directly tied to the concentration of nitrate in the tobacco. When pH levels of the smoke rise to greater than 6.0, the percentage of free, unprotonated nicotine increases to about 30 percent at pH 7.4 and to about 60 percent at pH 7.8 (Brunnemann and Hoffmann, 1974). Protonated nicotine is only slowly absorbed in the oral cavity; yet, unprotonated nicotine, which is partially present in the vapor phase of the smoke, is quickly absorbed through the mucosal membranes of the mouth (Armitage and Turner, 1970). The pH of cigar smoke rises with increasing puff numbers from pH 6.5 to 8.5; consequently, the rapid oral absorption of the free nicotine in the vapor phase gives a primary cigar smoker immediate nicotine stimulation so that he has no need for inhaling the smoke. Similarly, the smoker of black, air-cured cigarettes tends not to inhale the smoke at all, or only minimally (Armitage and Turner, 1970; NCI, 1998).

In 1963, the first comparative study on the tumorigenicity on mouse skin of tars from the four major types of *N. tabacum* revealed the highest activity for tars from flue-cured and sun-cured tobaccos, and the lowest for the two varieties of air-cured tobaccos (Table 5-8; Wynder and Hoffmann, 1963). The concentration of BaP, as an indicator of the concentrations of all carcinogenic PAH, is correlated with the tumor initiation potential of the tars. Upon topical application to mouse skin and human epithelia, carcinogenic PAH induces papilloma and carcinoma. In inhalation studies with Syrian golden hamsters, the smoke of a cigarette made with a particular tobacco blend was significantly more active in inducing carcinoma of the larynx than was the smoke of a cigarette with air-cured (black) tobacco (Dontenwill *et al.*, 1973).

To verify whether a reduction of carcinogenic PAH in the smoke due to the presence of high levels of nitrate in tobacco leads to reduced mouse skin tumorigenicity of the tar, sodium nitrate (8.3 percent) was added to the standard tobacco blend. On a gram-to-gram basis, the tar from the cigarette with added nitrate (0.6 µg BaP per gram tar) induced skin tumors in only 2 of 50 mice, whereas the tar from the control cigarette (without the addition of nitrate; 1.05 µg BaP per gram tar) induced skin tumors in 25 of 100 mice (Hoffmann and Wynder, 1967). In inhalation experiments with Syrian golden hamsters, smoke from the control cigarette plus 8.0 percent of sodium

nitrate induced laryngeal carcinomas in only 25 of 160 animals (15.6 percent), compared to this type of neoplasm in 60 of 200 animals (30 percent) in assays with the control cigarette (Dontenwill *et al.*, 1973). Thus, all of these bioassays on the skin of mice and the inhalation studies with hamsters support the concept that increased nitrate content of the tobacco inhibits the pyrosynthesis of the carcinogenic PAH and that the tars of these cigarettes, and their smoke as a whole, have a reduced potential for inducing benign and malignant tumors in epithelial tissues when compared to the tar or whole smoke of cigarettes with tobacco that is low in nitrate.

Reconstituted Tobacco and Expanded Tobacco

In the early 1940s, the technology for making reconstituted tobacco (RT) was developed. Manufacturing RT enables the utilization of tobacco fines, ribs, and stems in cigarette tobacco blends (Halter and Ito, 1979). Prior to this technology, tobacco fines and stems had been discarded. With the utilization of RT as part of the tobacco blend, less top quality tobacco is needed and, thereby, the cost of making cigarettes has been reduced. Laboratory studies (Wynder and Hoffmann, 1967) have shown that cigarettes made entirely of RT deliver a smoke with significantly reduced levels of tar, nicotine, volatile phenols, and carcinogenic PAHs.

The two major technologies for making RT for cigarettes are the slurry process and the paper process. Either process leads to RT with low density. The advantage of RT lies in the creation of a high degree of aeration of the tobacco which enhances combustibility. Most of the tested tars from reconstituted tobaccos had significantly reduced carcinogenic activity on mouse skin (Wynder and Hoffmann, 1965; NCI, 1977a). In inhalation assays with Syrian golden hamsters, diluted smoke from cigarettes made of reconstituted tobacco induced significantly fewer carcinomas in the larynx (19/160) than the diluted smoke from control cigarettes (60/200). The cigarette with RT gave only 7 puffs per cigarette and yielded 20.8 mg tar and 16 ng of BaP compared to 10 puffs, 33.7 mg tar, and 35.4 ng BaP for the control cigarette (Dontenwill *et al.*, 1973). This result supports the concept that, at least in the experimental setting, the carcinogenic PAH, with BaP as a surrogate, are correlated with the induction of papilloma and carcinoma in epithelial tissues. The procarcinogenic TSNA, on the other hand, are not activated by enzymes to their reactive species in epithelial tissues; thus, they induce few, if any, tumors in such tissues. Tobacco ribs and stems, the major components of RT, are richer in nitrate (and this applies especially to the ribs and stems of air-cured tobaccos) than the laminae of tobacco (Neurath and Ehmke, 1964; Brunnemann *et al.*, 1983; Brunnemann and Hoffmann, 1991; Burton *et al.*, 1992). Therefore, in general, the nitrate content of today's blended U.S. cigarette, which may contain 20–30 percent RT, is at 1.2–1.5 percent—much higher than the nitrate level in cigarettes during the fifties and sixties when it was ≤0.5 percent (U.S. DHHS, 1989; Spears, 1974). Cigarettes with RT emit in their smoke significantly greater amounts of TSNA than cigarettes of the past. These TSNA include the adenocarcinoma-inducing NNK, which is metabolically activated to carcinogenic species in target tissues like the lungs (Hoffmann *et al.*, 1994). One major U.S. cigarette manufacturer was awarded a patent in December 1978 for developing

a process that reduces more than 90 percent of the nitrate content of the RT made from ribs and stems (Kite *et al.*, 1978; Gellatly and Uhl, 1978). It is unclear to which extent this patented method has been applied to the RT manufacture for U.S. commercial cigarettes.

There are at least three methods for expanding tobacco by freeze-drying (NCI, 1977b). As a result of freeze-drying, expanded tobacco has greater filling power than natural tobacco, meaning that less tobacco is needed to fill a cigarette. An 85-mm filter cigarette, filled entirely with expanded tobacco, requires 630 mg tobacco; while a regular non-filter control cigarette of the same dimensions requires 920 mg tobacco. The tar yields in the smoke of both types of cigarettes amounted to 12.4 mg and 22.1 mg, respectively (NCI, 1977b, 1980). In 1982, incorporation of all possible modifications in the makeup of the cigarette required only 785 mg leaf tobacco; in contrast, in 1950, the blended U.S. cigarette required 1,230 mg leaf tobacco (Spears, 1974). Table 5-9 presents analytical data for the smoke of experimental cigarettes filled with puffed tobacco, expanded or freeze-dried tobacco, and a control cigarette. Levels of most components measured in the smoke of cigarettes with puffed tobacco, expanded tobacco, or freeze-dried tobacco were reduced, compared with data for the control cigarette (NCI, 1977b, 1980).

The changes that have occurred between 1950 and 1995 in the makeup of U.S. cigarettes, have significantly altered smoke composition. Table 5-10 compares data for individual components in the smoke of U.S. blended cigarettes of the 1950s with corresponding data for the cigarette smoke composition profiles that have been established between 1988 and 1995. All of these cigarettes were smoked using the FTC method (Pillsbury *et al.*, 1969).

Additives

Humectants

Humectants serve to retain moisture and plasticity in cigarette and pipe tobaccos. They prevent the drying of tobacco, which would lead to a harsh tasting smoke; importantly, they also preserve those compounds that impart flavor to the smoke. Today, the principal humectants in cigarette tobacco are glycerol (propane-1,2,3-triol) and propylene glycol (PG; propane-1-2-diol); of lesser importance are diethylene glycol (2.2'-di[hydroxyethyl]ether) and sorbitol (Voges, 1984). In the past, ethylene glycol (ethane-1,2,-diol) has been used as a humectant for cigarette tobacco. However, because this compound leads to the formation of ethylene oxide, which is carcinogenic to both animals and humans, its use has been prohibited (IARC, 1994a). In 1972, Binder and Lindner reported the presence of 20 µg ethylene oxide per cigarette in the smoke of the untreated tobacco of one cigarette brand (Binder and Lindner, 1972). In this context, it is noteworthy that Törnqvist and colleagues (1986) found significant levels of the *N*-hydroxyethylvaline moiety of hemoglobin in the blood of smokers ranging between 217 and 690 pmol/g Hb, averaging 389 ± 138 pmol/g, while levels in nonsmokers' blood ranged between 27 and 106 pmol/g Hb and averaged 58 ± 25 pmol/g Hb. The authors suggest that most of the ethylene oxide in the hemoglobin adduct is derived from endogenous oxidation of ethene in cigarette smoke (50–250 µg/cigarette) (Törnqvist *et al.*, 1986).

Humectants may comprise up to 5 percent of the weight of cigarette

Table 5-9
Smoke Analyses of Cigarettes Made from Puffed, Expanded, and Freeze-Dried Tobacco and from a Control Cigarette

Smoke Component	Puffed Tobacco	Expanded Tobacco	Freeze-dried Tobacco	Expanded Stems	Control
CO (mg)	9.33	11.8	12.3	23.1	18.0
Nitrogen oxides (μg)	247.0	293.0	235.0	349.0	269.0
HCN (μg)	199.0	287.0	234.0	248.0	413.0
Formaldehyde (μg)	20.7	21.7	33.4	58.0	31.7
Acetaldehyde (μg)	814.0	720.0	968.0	803.0	986.0
Acrolein (μg)	105.0	87.7	92.4	93.0	128.0
Tar (mg)	16	18	16	23	37
Nicotine (mg)	0.8	0.7	0.8	0.4	2.6
BaA (ng)	13.7	11.8	15.3	19.5	37.1
BaP (ng)	11.8	8.2	9.2	16.2	28.7

Abbreviations: CO=carbon monoxide; HCN=hydrogen cyanide; BaA=banz(a)anthracene; BaP=benzo(a)pyrene.
Source: National Cancer Institute, 1980.

Table 5-10
Changes in the Yields of Selected Toxic Agents in the Smoke of U.S. Cigarettes (FTC Smoking Conditions)

	Earlier Cigarettes[a]		Current Cigarettes[a]	
Smoke Component	**Year**	**Concentration**	**Year**	**Concentration**
Carbon monoxide (CO)	1953	33-38 mg (NF)	1994	11 mg (F)
Nitrogen oxides (HNO_x)	1965	330 μg (NF)	1994	500 μg (NF)
Benzene	1962	30 μg (NF)	1988	48 μg (NF)
	1962	25-30 μg (F)	1990	42 μg (F)
Acetaldehyde	1960	1,000 μg (NF)	1992	400 μg (F)
NDMA	1976	43 ng (NF)	1989	65 ng (NF)
Tar	1953	38 mg (NF)	1994	12 mg (F)
Nicotine	1953	2.7 mg (NF)	1994	0.85 mg (F)
	1959	1.7 mg (F)	1994	1.1 mg (F)
Phenol	1960	100 μg (NF)	1994	70 μg (NF)
	1960	46 μg (F)	1994	35 μg (F)
Catechol	1965	390 μg (NF)	1994	
	1976	790 μg (F)	1994	140 μg (F)
2-Naphthylamine	1968	22 ng (NF)	1985	35 ng (F)
BaP	1959	50 ng (NF)	1995	19 ng (NF)
	1959	27 ng (F)	1995	8 ng (F)
NNN	1978	220 ng (NF)	1995	300 ng (NF)
	1978	240 ng (F)	1995	280 ng (F)
NNK	1978	110 ng (NF)	1995	190 ng (NF)
	1978	100 ng (F)	1995	144 ng (F)

[a]*Abbreviations: NF=non-filter; F=filter; NDMA=N-nitrosodimethylamine; BaP=benzo(a)pyrene; NNN=N'-nitrosonornicotine; NNK=4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone*
Source: Pillsbury et al.*, 1969.*

tobacco. In a 1964 study, 18 U.S. cigarette tobacco blends that were analyzed for humectants contained between 1.7 and 3.15 percent of glycerol, which is to some extent decomposed to the ciliatoxic acrolein, and between 0.46 and 2.24 percent of PG (Cundiff *et al.*, 1964). The smoke of four American cigarettes contained between 0.34 and 0.96 mg/cigarette of PG (Lyerly, 1967). However, PG may be thermically degrading to yield propylene oxide. This would be of concern because propylene oxide is regarded as possibly carcinogenic to humans (IARC, 1994b). Four U.S. cigarettes contained between 0.34 and 0.96 mg per cigarette (Lyerly, 1967). In 1999, between 12 and 100 ng of propylene oxide were detected in the smoke of cigarettes filled with PG treated tobacco. Several commercial samples of PG, used as a humectant for cigarette tobacco, already contained traces of propylene oxide (Kagan *et al.*, 1999).

Flavor Additives Natural tobacco is composed of a wide spectrum of components that, upon heating, release agents, which contribute to the flavor of the smoke. These include tobacco-specific terpenoids, pyrroles, and pyrazines among others (Roberts and Rowland, 1962; Gutcho, 1972; Senkus, 1976; Leffingwell, 1987; Roberts, 1988). The effective reduction of smoke yields by filter tips and by the incorporation of reconstituted tobacco also brought about a reduction of flavor components in the smoke. To counteract this loss of smoke flavor, the tobacco blends are treated with additives that are essentially precursors to smoke flavors. They include natural agents contributing to minty, spicy, woody, fruity and flowery flavors. In some instances, such additives also include synthetic agents as flavor enhancers. While most of the flavor enhancers are chosen indiscriminately, it is realized that some of them may contribute to toxicity or carcinogenicity of cigarette smoke. A case in point was the cessation of the use of deer tongue extract which contained several percent of the animal carcinogen coumarin (Voges, 1984). It has been suggested that additives to cigarettes are used to reduce the perception of environmental tobacco smoke (ETS; Connolly *et al.*, 2001).

In 1993 and 1994, the tobacco industry convened an expert panel of toxicologists to screen agents that were in use, or considered for use, as tobacco additives. The panel established a list of 599 agents that were generally regarded as safe (GRAS), whereby the term 'safe' applied to each of the additives as such without consideration of the fate and reactivity of these agents during and after combustion (Doull *et al.*, 1994). An exception was menthol, which was known to transfer into the smoke without yielding appreciable amounts of carcinogenic hydrocarbons (Jenkins *et al.*, 1970). A recent toxicologic evaluation of flavor ingredients dealt with 170 such agents that are commonly used in the manufacture of American blended cigarettes, and examined their effects in four sub-chronic, nose-only smoke inhalation studies in rats compared to effects of the smoke of tobacco blends without additives. Control animals were exposed to filtered air (Gaworski *et al.*, 1998). Smoke exposure was monitored with internal dose markers, including carboxyhemoglobin, serum nicotine, and serum cotinine. The mainstream smoke (MS) of flavored and nonflavored cigarette types caused essentially the same responses in the respiratory tracts of the rats; specifically hyperplasia and metaplasia in the nose and larynx. As this

study involved maximally 65 hours of exposure (while induction of tumors would not be expected until animals reach half their life span), one cannot deduce with certainty that the addition of these flavoring agents to tobacco blends has no impact on the development of tumors.

New Types of Cigarettes

The tobacco companies have undertaken a substantial research effort to develop new types of nicotine delivery devices. These devices were intended to generate an aerosol with nicotine in the range of the levels present in conventional cigarettes but with very low emissions of tar and other toxic agents. Toward the end of the 1980s, the first prototype of these new types of cigarettes was on the test market, a product named "Premier." It was a cigarette that "heats rather than burns tobacco" (R. J. Reynolds Tobacco Company, 1988; DeBethizy *et al.*, 1990; Borgerding *et al.*, 1990a & b, 1998). This 80-mm cigarette is comprised of three sections. The first 40-mm section of this cigarette is made with compressed charcoal, which is immediately linked to an inner aluminum tube containing tobacco, flavor additives, and glycerol. This tube is embedded in tobacco. Section 2 (~10 mm) is a cellulose acetate filter dusted with charcoal powder. The third section (~30 mm) is a cellulose acetate filter tip. Under FTC standard machine smoking conditions, the "Premier" delivers smoke containing 0.3 mg nicotine, 6.3 mg water, 4.6 mg glycerol, 0.4 mg propylene glycol, and 0.7 mg tar. Compared with the reference (conventional) cigarette, and disregarding nicotine, the majority of the known toxic and carcinogenic agents in the smoke are reduced by more than 90 percent. Known exceptions are carbon monoxide (CO) (+3.5 percent), ammonia (–5.6 percent), formaldehyde (–35.3 percent), resorcinol (–73.3 percent), quinoline (–56.6 percent), and acetamide (–18.2 percent). This new type of cigarette did not gain consumer acceptance, possibly because of difficulty in igniting the "Premier," the need for frequent puffing to ensure continuous burning, the lack of flavor, and the low nicotine delivery (0.3 mg/cigarette). Nicotine emission was below the level that would satisfy most smokers' acquired need for this agent even with compensatory smoking.

In 1996, a modified "Premier" came on the market. In the United States, it is known as "Eclipse"; in Germany it is called "HiQ," and in Sweden, it goes by the name "Inside." The "Eclipse" consists of four sections. Section 1, the heat source, is a specially prepared charcoal; section 2 consists of tobacco plus glycerol; section 3 contains finely shredded tobacco; and section 4 is a filter tip. Upon ignition, the special charcoal heats the air stream during puff drawing. The heated air stream enters the tobacco sections and vaporizes glycerol, as well as the volatile and semi-volatile tobacco components, including nicotine. Under FTC smoking conditions, the "Eclipse" delivers 8 mg CO (low-tar filter cigarette: 6–12 mg), 150 µg acetaldehyde (700 µg), 30 µg NO_x (200–300 µg), 180 µg hydrogen cyanide (300–400 µg), 5.1 mg tar (11–12 mg), and 0.2–0.4 mg nicotine (0.7–1.0 mg). The remainder of the smoke particulates consists of 33 percent water, 47 percent glycerol, and 17 percent of various other compounds. The concentrations of the major carcinogens, such as BaP, 2-aminonaphthalene, 4-aminobiphenyl, and the TSNA are lowered by 85–95 percent (Rose and Levin, 1996; Smith *et al.*, 1996). Currently, the "Eclipse" is being test marketed and it appears that

response is somewhat more favorable than it was to its predecessor, the "Premier." The products labeled, "Eclipse Full Flavor," "Eclipse Mild," and "Eclipse Menthol" produce FTC-standardized smoke yields of 0.2, 0.1, and 0.2 mg nicotine and 3, 2, and 3 mg tar per cigarette. Regular cigarette smokers were asked to switch for 2 weeks to "Eclipse." There were four study groups, each composed of 26–30 volunteers, for a total of 109 smokers. Smoking of "Eclipse" resulted in about a 30 percent larger puff volume, about 50 percent more puffs, which added up to a total puff volume per cigarette that was more than twice that of the total volume drawn from the control cigarettes (Stiles *et al.*, 1999). These data suggest that the volunteers smoked "Eclipse" more intensely than their non-filter cigarettes. This observation is also supported in the uptake of nicotine (Benowitz *et al.*, 1997). The mutagenic activities of the urine of smokers of four types of "Eclipse" were assayed on two bacterial strains and were reduced by 72% to 100%, compared with the mutagenic activities of the urine of the same volunteers after smoking their regular cigarettes (Smith *et al.*, 1996).

An Expert Committee from the Institute of Medicine of the National Academy of Sciences studied the scientific basis for a possible reduction of the "harm" induced by "Eclipse" relative to the "harm" induced by smoking conventional cigarettes. On the basis of the available data, the Committee came to the following conclusions: "Eclipse" offers the committed smoker an option that is currently not available. "Eclipse" does not add to the inherent biological activity of smoke from the range of cigarettes currently on the market. The elevated COHb levels should be regarded as a potential risk factor for cardiovascular diseases. The magnitude of this risk remains to be determined (Gardner, 2000).

The high concentration of glycerol in the "Eclipse" aerosol led to bioassays of glycerol in 2-week (1.0, 1.93, and 3.91 mg/L) and 13-week (0.033, 0.167, and 0.662 mg/L) "nose only" inhalation studies with Sprague-Dawley rats, testing for toxicity and especially for irritating effects. The investigators detected metaplasia of the lining of the epiglottis (Gardner, 2000). The 13-week inhalation studies with rats and hamsters had also resulted in some early histopathological changes in the upper respiratory tract in both laboratory animals. These observations signal the need for lifetime inhalation assays with the smoke of "Eclipse" in rats, preferably Fisher 344 rats, or better yet, in Syrian golden hamsters, possibly with an inbred strain of hamsters susceptible to carcinogens in the respiratory tract (Bernfeld *et al.*, 1974). Pauly *et al.*, from the Roswell Park Cancer Institute, Buffalo, New York, caution that harmful glass fibers have been found to migrate into the filter tip of the "Eclipse" and may be inhaled during puffing (Pauly *et al.*, 1998).

The Health Department of Massachusetts and the Society for Research on Nicotine and Tobacco disputed the claims made for "Eclipse." They requested that the FTC and the FDA institute regulatory procedures to ensure that insufficiently documented health claims are not made for tobacco products. Declaring "Eclipse" the "next best choice," or calling TSNA-reduced tobacco products "safer tobacco" (Anonymous, 2000; Society

for Research on Nicotine and Tobacco, 2000) is deceiving.

In 1998, Philip Morris USA released a new type of cigarette (EHC) that is heated electrically to release an aerosol. On the basis of chemical analyses and short-term bioassays, it has significantly lower toxicity and mutagenicity than the smoke of the Kentucky reference filter cigarette, 1R4F. The prototype, containing a tobacco filler wrapped in a tobacco mat, is kept in constant contact with eight electrical heater blades in a microprocessor-controlled lighter. This cigarette contains about half the amount of the tobacco of a conventional cigarette. Under FTC-standardized smoking conditions, the cigarette delivers, with an average of 8 puffs, about 1 mg of nicotine, whereas all other smoke constituents analyzed were significantly lower than those in the smoke of the low-yield Kentucky reference cigarette, 1R4F (Terpstra *et al.*, 1998). However, formaldehyde yields were significantly higher in the smoke of the EHC and emissions of glycerol and 2-nitropropane were comparable to those recorded in the smoke of the 1R4F cigarette. Per gram of tar, the smoke of the EHC had significantly lower mutagenic activity than the smoke of the 1R4F reference cigarette in TA98 and TA100 tester strains with metabolic activation (Terpstra *et al.*, 1998).

OBSERVATIONS ON CIGARETTE SMOKERS

In mice, rats, and hamsters, NNK induces adenomas and adenocarcinomas (AC) in the peripheral lung. This effect is independent of route and form of application (Hoffmann *et al.*, 1994). NNK is metabolically activated primarily to the unstable 4-(hydroxymethylnitrosamino)-1-(3-pyridyl) -1- butanone and to 4-(α-hydroxymethylene)-1-(3-pyridyl)-1-butanol, which decomposes into methane diazohydroxide and 4-keto-4-(3-pyridyl)butane diazohydroxide, respectively. The diazohydroxides react with DNA bases to form 7-methyl guanine, O^6-methyl guanine and O^4-methyl thymidine, respectively, and also form a pyridyloxobutyl adduct of presently unknown structure. Upon acid hydrolysis, this adduct releases 4-hydroxy-1-(3-pyridyl)-1-butanone. These adducts have been found in the lungs of mice and rats following treatment with NNK, and they have also been identified in human lungs. The origin of 7-methyl guanine in DNA from human lungs is unclear; conceivably, in addition to TSNA, nitroso compounds such as *N*-nitrosodimethylamine may also have been a source for this DNA-methylation. However, it is clear that higher levels of 7-methyl guanine have been found in the lung of smokers than in the lung of nonsmokers, thus strengthening the evidence that NNK is a major contributor to the methylation of the lung DNA of smokers (Hecht, 1998).

PAH induce squamous cell carcinoma of the lung in laboratory animals and in workers with exposures to aerosols that are high in PAH. NNK metabolites induce primarily AC of the lung in laboratory animals. Reactive PAH metabolites bind to DNA in epithelial tissues. In laboratory animals, metabolically activated forms of NNK react with the DNA of Clara cells in the peripheral lung (Belinsky *et al.*, 1990) to form methylguanine and methylthymidine, as well as pyridyloxobutylated adducts. 7-Methylguanine has been found in smokers' lungs at higher levels than in the lungs of nonsmokers.

Additional support for the observation that adenocarcinoma of the lung

among cigarette smokers has increased relative to squamous cell carcinoma during the past 25 years, and for the concept that lung cancer risk of smokers of low-nicotine filter cigarettes is similar to that of smokers of non-filter cigarettes, comes from biochemical studies. In the mouse, the O^6-methylguanine pathway of metabolically activated NNK is clearly the major route for induction of lung tumors; this conclusion is consistent with the high percentage of GGT→GAT mutations in the K-*ras* oncogene induced by NNK (Hecht, 1998; Singer and Essigmann, 1991). A study from the Netherlands has shown that mutations on codon 12 of the K-*ras* oncogene are present in 24–50 percent of human primary adenocarcinoma. These mutations occur more frequently in AC of the lung in smokers than in nonsmokers. Twenty percent of the mutations in codon 12 involve GGT→GAT conversions, which supports the concept that NNK plays a role in the induction of AC of the lung in smokers. Histochemical examination of human lung cancer showed cyclooxygenase (COX)-2 expression in 70 percent of invasive carcinoma cases (Hida *et al.*, 1998). COX-2 expression was also identified in adenocarcinoma of the lung in rats treated with NNK (el-Bayoumy *et al.*, 1999). It is anticipated that future studies in molecular biology will fully elucidate the significance of TSNA, especially of NNK, and of the carcinogenic PAH in the induction of lung cancer in tobacco smokers.

SUMMARY Major modifications in the makeup of the commercial cigarette were introduced between 1950 and 1975. Since then, there have been no substantive changes toward a further reduction of the toxic and carcinogenic potential of cigarette smoke beyond reducing MS yields of tar, nicotine, and carbon monoxide. Some of these modifications have also resulted in diminished yields of several toxic and carcinogenic smoke constituents.

Cigarettes with charcoal filter tips deliver MS with significantly lower concentrations of the major ciliatoxic agents, such as hydrogen cyanide and volatile aldehydes. However, except in Japan, South Korea, Venezuela, and Hungary, cigarettes with charcoal filter tips account for less than one percent (USA) and at most for a small percentage of all cigarettes sold worldwide (Fisher, 2000).

Cellulose acetate filters with or without perforation have the capacity for selective reduction of smoke yields of volatile *N*-nitrosamines and semivolatile phenols. The latter are major tumor promoters in cigarette tar. In contrast to cigarettes manufactured in the 1950s, most of the cigarettes on the market today use a highly porous wrapper of paper treated with agents that enhance the burning, thus, contributing to the reduction of machine-measured yields of carbon monoxide, hydrogen cyanide, volatile aldehydes, volatile *N*-nitrosamines, PAH, and TSNA.

Reconstituted tobacco and expanded tobacco today amount to between 25 and 30 percent of the cigarette tobacco blend. Reconstituted tobacco reduces the yields of smoke components such as tar and CO. The tar from cigarettes made entirely of reconstituted tobacco is less carcinogenic on mouse skin and the smoke of these cigarettes reduces significantly the induction of carcinoma in the larynx of hamsters compared to the smoke of reference cigarettes made of natural tobacco. Reconstituted tobaccos and

expanded tobaccos have a significantly greater filling power than natural tobacco. An 85-mm filter cigarette that is filled entirely with expanded tobacco requires 363 mg tobacco while a regular filter-tipped cigarette requires 667 mg tobacco. The smoke of cigarettes made of expanded tobacco has significantly lower MS yields of tar, nicotine, CO, hydrogen cyanide, PAH, and TSNA. On the basis of weight-to-weight comparisons, the tar from these cigarettes is significantly less tumorigenic on mouse skin than the tar of a reference cigarette made of the corresponding natural tobacco.

Since 1959, each year the levels of tar, nicotine, and benzo(*a*)pyrene in the mainstream smoke of a leading U.S. non-filter cigarette have been monitored. Beginning in 1977, the MS was also analyzed for NNK and in 1981, the determinations of CO in the mainstream smoke were added. For all of these analyses, the MS was generated with the standardized machine smoking parameters that are mandated by the Federal Trade Commission. Table 5-11 documents the decline of tar levels from 29.8 mg to 24.3 mg in the years between 1959 and 1984, while nicotine levels fell from 2.4 mg to 1.6 mg between 1959 and 1977. Since then, the smoke yields of tar and nicotine for this non-filter brand have not changed. Carbon monoxide remained stable at 16 to 18 mg per cigarette since it was first reported in 1981. By 1997, it was clear that significant changes in the smoke yields of the major lung carcinogens BaP and NNK have occurred since 1977 in that BaP levels declined from 49 ng to 19 ng, but NNK increased from 120 ng to 195 ng per non-filter cigarette.

It is important to note that we are lacking analytical data regarding the levels of these major carcinogens and toxins in the mainstream smoke of leading cellulose acetate filter-tipped cigarettes with and without filter perforation, as well as in the MS of charcoal filter cigarettes. These cellulose acetate filter cigarettes were actually the ones dominating the U.S. cigarette market as the use of non-filter cigarettes faded over the years and charcoal filter cigarettes had only a modest market share. Most importantly, we are also lacking data on biological activities of the tars of leading brands of filter cigarettes produced since the 1960s because tumorigenicity and carcinogenicity of tars have not been monitored on a regular basis. There is now also an urgent need for analytical profiles of the toxic and carcinogenic mainstream smoke constituents that are generated under conditions reflecting the puff drawing profiles actually exhibited by humans who smoke these cigarettes that give lower yields as per FTC measurements. Such analytical data would have to be established for major U.S. cigarette brands manufactured since 1960. They would serve as the scientific basis in support of epidemiological observations regarding the risk of cancer of the lung and upper aerodigestive tract for smokers who have exclusively smoked filter-tipped brands as compared to the risk for smokers who used non-filter cigarettes.

Changes in the agricultural, curing, and manufacturing processes of cigarettes have resulted in an increase in tobacco-specific nitrosamines in cigarette smoke that may have contributed to the increase in adenocarcinoma of the lung observed over the past several decades.

Table 5-11
Tar, Nicotine, CO, B*a*P, and NNK in the Mainstream Smoke of a Leading U.S. NF Cigarette, 1959-1997[a]

Year	Tar (mg)	Nicotine (mg)	Carbon Monoxide[b] (mg)	Ba P (ng)	NNK[b] (ng)
1959	29.8	2.4		40	
1967	27.2	1.6		49	
1971	29.0	1.8		22	
1977	26.0	1.59		19	120
1981	24.3	1.52	16.7	19	130
1988	24	1.5	16	19	140
1991	25	1.7	16	18	190
1997	26	1.7	18	19	195

Abbreviations: NNK=4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone; BaP= benzo(a)pyrene.
[a]The analytical data were generated by smoking the leading U.S. NF cigarette according to the FTC-mandated standard machine smoking method (Pillsbury et al.*, 1969).*
[b]The open fields document the lack of analytical data for the years 1959, 1967, 1971, and 1977 for CO and 1959, 1967, and 1977 for NNK
Sources: Wynder and Hoffman, 1960; Federal Trade Commission, 1971, 1977, 1981, 1988, 1991, 1997; Hoffmann and Hoffmann, 1997.

CONCLUSIONS

1. Major modifications in the makeup of the commercial cigarette were introduced between 1950 and 1975, but since that time there have been few substantive changes toward a further reduction of the toxic and carcinogenic potential of cigarette smoke.

2. A variety of changes in cigarette design and filtration have resulted in chemical changes in cigarette smoke, some of which have also demonstrated decreased toxicity in animal assays. Toxicity or carcinogenicity in animal assays has not been monitored to allow evaluation of changes over time that have occurred for cigarette smoke produced by commercial brands of cigarettes.

3. Changes in the agricultural, curing, and manufacturing processes of cigarettes have resulted in an increase over the last several decades in the amounts of tobacco-specific nitrosamines in cigarette smoke. These changes are considered to have contributed to the increase in adenocarcinoma of the lung observed over the past several decades.

4. On the basis of the standard machine smoking method for cigarettes that has been mandated by the FTC, the sales-weighted average nicotine yields of U.S. cigarettes decreased gradually from 2.7 mg per cigarette in 1953 to 0.85 mg by the mid 1990s. Today, the smoker of filter cigarettes will greatly increase his/her smoking intensity to satisfy an acquired need for nicotine. Thus, the inhaled smoke of one cigarette contains 2 to 3 times the amount of tar, nicotine, and carbon monoxide and 1.6 to 1.8 times the level of biomarkers for the major lung carcinogens BaP, and NNK, compared to amounts in the smoke generated by the FTC method.

Appendix

Abbreviations

AC	Adenocarcinoma
BaA	Benz(a)anthracene
BaP	Benzo(a)pyrene
FTC	Federal Trade Commission
Hb	Hemoglobin
IARC	International Agency for Research on Cancer
MS	Mainstream Smoke
NNK	4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone
NNN	*N*′-Nitrosonornicotine
NO_x	Nitrogen oxides (NO, NO_2, and N_2O)
PAH	Polynuclear Aromatic Hydrocarbons
RT	Reconstituted Tobacco
SCC	Squamous Cell Carcinoma
TSNA	Tobacco Specific *N*-Nitrosamines
VNA	Volatile *N*-Nitrosamines

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