Regulatory Toxicology and Pharmacology 66 (2013) 197–207

Contents lists available at SciVerse ScienceDirect

Regulatory Toxicology and Pharmacology

journal homepage: www.elsevier.com/locate/yrtph

TSNA levels in machine-generated mainstream cigarette smoke: 35 years of data

Scott Appleton *, Raquel M. Olegario, Peter J. Lipowicz

Altria Client Services Inc., 601 East Jackson Street, Richmond, VA 23219, USA

ARTICLE INFO

Article history:
Received 16 December 2012
Available online 2 April 2013

Keywords:
Cigarettes
Mainstream cigarette smoke
NNN
NNK
Tobacco products
Tobacco specific nitrosamines (TSNA)

ABSTRACT

This paper characterizes historical and current tobacco specific nitrosamine (TSNA) levels in mainstream (MS) cigarette smoke of US commercial cigarettes. To conduct this analysis, we gathered 35 years of published data of 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) and *N*-nitrosonornicotine (NNN) levels in MS cigarette smoke. We also assessed internal data of MS smoke NNK and NNN levels generated from various market monitoring initiatives and from control cigarettes used in a multi-year program for testing cigarette ingredients. In all, we analyzed machine smoking data from 401 cigarette samples representing a wide range of products and design characteristics from multiple manufacturers and market leaders. There was no indication that TSNA levels systematically increased in cigarette MS smoke over the 35-year analysis period. In particular, TSNA levels expressed as either per cigarette or normalized for tar suggest a downward trend in MS smoke over the past 10 years. The apparent downward trend in TSNA levels in MS smoke may reflect industry and agricultural community efforts to reduce levels of TSNAs in tobacco and cigarette smoke.

1. Introduction

Tobacco specific nitrosamines (TSNAs) are a class of cigarette smoke constituents believed to play a potential role in smoking related carcinogenesis (IARC, 2007). The TSNAs, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) and *N*-nitrosonornicotine (NNN), have received the greatest attention due to their carcinogenic activity in animal studies. Both are classified by the International Agency for Research on Cancer (IARC) as carcinogenic to humans (Group 1) and both are on the US Food and Drug Administration established list of harmful and potentially harmful constituents in tobacco products and tobacco smoke (IARC, 2007; USDHHS, 2012).

Concerns have been raised by the tobacco control community suggesting that little progress has been made by the tobacco industry to reduce TSNA levels in cigarette smoke (Stepanov et al., 2012). It has also been suggested that TSNA levels in cigarette smoke may have even increased over time (Burns et al., 2011a).

This report presents a compilation of mainstream (MS) cigarette smoke NNK and NNN levels reported in the published literature and augmented with internal, unpublished data from Philip Morris USA Inc. (PM USA), an Altria company. We gathered and analyzed available published and internal data on NNK and NNN levels in MS cigarette smoke with the following objectives:

1. To characterize historical and current TSNA levels in MS cigarette smoke
2. To conduct secondary analyses to gain insights into patterns of TSNA levels in MS cigarette smoke over time.

2. Methods

2.1. Published literature values

We gathered published literature values of NNK and NNN levels in MS cigarette smoke. We searched Chemical Abstracts Service (CAS) for the CAS Registry® number of NNK indexed to "smoke" or "both" (tobacco and smoke). Original research studies that included a description of analytical methods identified in this search were reviewed to determine if they met the following additional inclusion criteria:

- NNK and NNN data in MS cigarette smoke because MS smoke is the most proximate product to which the smoker is exposed to TSNAs
- TSNA data from cigarettes smoked under Federal Trade Commission (FTC) or International Organization for Standardization (ISO) conditions, which provide the largest data set available for evaluation

Abbreviations: MS, mainstream; NNK, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone; NNN, *N*-nitrosonornicotine; PM USA, Philip Morris USA Inc.; TSNA, tobacco specific nitrosamine.

* Corresponding author. Fax: +1 804 335 2095.
E-mail addresses: SciencePublications@altria.com, Scott.Appleton@altria.com (S. Appleton).

0273-2300/$ - see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.yrtph.2013.03.013

- Data from US commercial cigarettes in order to compare results with prior analyses of US commercial cigarettes by Hoffmann and Hoffmann (1997) and Stepanov et al. (2012)
- Peer reviewed published papers
- NNK and NNN levels presented as individual numeric values.

Published studies meeting the above inclusion criteria were included in this review.

Numeric values for NNK, NNN, and tar levels are presented as they appeared in data tables in the original published papers. The default approach for the year of a given data value was to use the year of the publication. If a publication specifically stated sample acquisition years that differed from the manuscript publication year, the sample acquisition year stated in the original manuscript was used.

2.2. Monitoring data

PM USA affiliated and contracted laboratories measured a range of smoke constituents in commercial cigarettes to help evaluate new or non-conventional cigarette products. Since 2002, both NNK and NNN levels in smoke were regularly measured from cigarettes produced by PM USA and from competitive US commercial cigarettes. The constituent analysis methods used were the same as, or equivalent to, those described in Counts et al. (2005) and Morton and Laffoon (2008).

2.3. Control cigarette data

PM USA produced control cigarettes as part of a multi-year ingredient testing program. From 1998 through 2007, PM USA manufactured control cigarettes to the same specifications over 60 times and measured the resulting tar, NNN, and NNK levels in the smoke, among other smoke constituents. The design and construction of control cigarettes were consistent with US commercial manufacturing, and their specifications can be found elsewhere (Gaworski et al., 2011). Although the amount of each component in the tobacco blend was the same throughout the period of ingredient testing, different crop years were used for the tobaccos due to depletion of available tobacco inventory. Therefore, in Table 1 we report the results for PM USA control cigarettes based on the year of manufacturing, even though the results were first published elsewhere (Coggins et al., 2011a,b,c,d,e,f,g,i).

2.4. Data analyses

The data were analyzed for trend with year by determination of Pearson's correlation coefficient, R, using standard statistical analysis software (IBM SPSS Statistics, Armonk, NY). All data points and all years were equally weighted. Three time periods were analyzed: 1978–2012, which is the complete data set; 1978–1995, which corresponds to Hoffmann and Hoffmann (1997); and 2002–2012, which corresponds to changes in flue-curing practices and burley seed selection to reduce NNK and NNN in tobacco leaf.

3. Results

3.1. TSNA levels in MS smoke: 35 years of data

We identified 14 published studies that met the data inclusion criteria (Adams et al., 1987; Brunnemann and Hoffmann, 1991; Brunnemann et al., 1996; Counts et al., 2005, 2006; Djordjevic et al., 1991; Hecht et al., 1979; Hoffmann et al., 1979, 1982, 1994; Patskan et al., 2008; Roemer et al., 2004; Stepanov et al., 2012; Swauger et al., 2002). Table 1 presents cigarette product identifications, levels of MS smoke tar (mg/cigarette), NNK (ng/cigarette), and NNN (ng/cigarette), and sources of the data presented in this review. The data in Table 1 are presented in chronological order according to either the year of the publication or, when reported, the year of sample acquisition (see Section 2). Because several papers reported sample acquisition years that were different from publication years, the publication years presented in Table 1 are not necessarily presented in strict chronological order. We were able to acquire data from a total of 401 cigarette samples analyzed for MS smoke NNK and NNN levels over a 35-year period. This includes data from published studies and internal data generated from PM USA-affiliated and contracted laboratories. Collectively, these data reflect a wide range of US commercial cigarette products from multiple manufacturers and include a wide range of design characteristics and market leaders.

Studies published prior to the 1990s were conducted primarily by scientists from the American Health Foundation, who typically analyzed small numbers of samples. Studies conducted from the mid-1990s to the present were primarily market-map studies conducted by the cigarette industry, which tended to analyze larger numbers of samples.

Fig. 1 presents a scatter plot of 35 years of data as the sum of NNK plus NNN expressed as ng/cigarette. Note higher and more variable levels in early years with a flattening of levels in later years.

TSNA levels in MS cigarette smoke can be influenced by several variables. The most prominent are the TSNA levels of the cigarette tobacco blend and the overall smoke yield of the cigarette. Because overall smoke yields can markedly influence TSNA levels in MS smoke of cigarettes with similar blends, it is possible that temporal trends in cigarette tar levels or sample selections could bias an assessment of TSNA levels in MS cigarette smoke over time. To correct for the effect of overall smoke yields, we normalized smoke TSNA data by dividing the levels of TSNAs by the amount of tar per cigarette. A scatter plot of these data as the sum of NNK plus NNN expressed as ng/mg tar is presented in Fig. 2.

For the entire data set 1978–2012 (NNK + NNN)/tar trended down with year ($R = -0.62$) and the trend was statistically significant ($p < 0.00001$). Comparable analyses for NNK + NNN on a per cigarette basis for the time period 1978–2012 showed similar results ($R = -0.54$, $p < 0.00001$). Removal from the 1978–2012 data set of Virginia blended cigarettes, which are known to have generally lower NNK and NNN, did not significantly alter the results on either (NNK + NNN)/tar ($R = -0.59$, $p < 0.00001$) or NNK + NNN per cigarette ($R = -0.52$, $p < 0.00001$).

3.2. Have TSNA levels increased over time?

Several papers published by Hoffmann and Hoffmann (1994, 1997) and Hoffmann et al. (1993) suggest that NNK levels in MS cigarette smoke have increased over time, specifically from 1979–1995. In all of these papers, the data that are presented appear to be from the same line graph and data from a single unidentified brand of cigarette referred to as "US leading NF cigarette." These sources are frequently cited by other researchers to support the suggestion that TSNA levels have generally increased in cigarette smoke over unspecified periods of time (Burns et al., 2011a,b). The reports in which Hoffmann suggests that TSNAs have increased over time are review articles where data are presented in the form of line graphs, but the original sources of the data in the graphs are not presented. Moreover, we were unable to identify a primary original source for these data. However, Hoffmann and others published a significant amount of data regarding cigarette smoke TSNA during 1978–1995.

We therefore, conducted a secondary analysis of data from published sources from the period of 1978–1995 to characterize NNK

Table 1
Machine smoking data from 401 cigarette samples representing a wide range of products and design characteristics from multiple manufacturers and market leaders.

Cigarette[a]	Tar (mg/cigarette)	NNK (ng/cigarette)	NNN (ng/cigarette)	Source
Commercial w/o filter	NR	110	240	Hoffmann et al. (1979)
Commercial with filter	NR	150	310	Hoffmann et al. (1979)
Commercial NF	NR	110	240	Hecht et al. (1979)
Commercial F	NR	190	310	Hecht et al. (1979)
Commercial D F	12.9	213	342	Hoffmann et al. (1982)
Commercial E F	13.4	249	513	Hoffmann et al. (1982)
Commercial B F	14.2	252	324	Hoffmann et al. (1982)
Commercial A F	13	360	458	Hoffmann et al. (1982)
Commercial C F	13.6	399	505	Hoffmann et al. (1982)
Commercial B NF	22.7	481	533	Hoffmann et al. (1982)
Commercial A NF	21.8	720	816	Hoffmann et al. (1982)
Commercial C NF	22.5	727	813	Hoffmann et al. (1982)
Commercial D NF	23.3	803	1110	Hoffmann et al. (1982)
Commercial E NF	21.8	868	1760	Hoffmann et al. (1982)
Commercial D (PF)	0.9	17.3	66.3	Adams et al. (1987)
Commercial C (F)	6.8	56.2	273	Adams et al. (1987)
Commercial B (F)	15.6	180	488	Adams et al. (1987)
Commercial A (NF)	20.1	425	1007	Adams et al. (1987)
US. blended NF 85 mm	NR	49	162	Djordjevic et al. (1991)
US F Ultra lights	NR	8.7	46	Brunnemann and Hoffmann (1991)
US F Lights	NR	47	188	Brunnemann and Hoffmann (1991)
US NF	NR	49	162	Brunnemann and Hoffmann (1991)
US F	NR	75	162	Brunnemann and Hoffmann (1991)
US F Ultra lights	NR	17	40	Hoffmann et al. (1994)
US F Lights	NR	106	122	Hoffmann et al. (1994)
US F	NR	156	209	Hoffmann et al. (1994)
US NF	NR	156	278	Hoffmann et al. (1994)
Merit ultima 85	0.7	13	22	Swauger et al. (2002)
Cambridge UL 85	1.3	16	18	Swauger et al. (2002)
Now UL 85	1	17	32	Swauger et al. (2002)
Carlton UL 85	1.1	18	25	Swauger et al. (2002)
GPC UL 100	4.7	46	46	Swauger et al. (2002)
Vantage UL 85	5.1	54	58	Swauger et al. (2002)
Kent LT 85	8.1	62	81	Swauger et al. (2002)
Doral LT 100 M	9	67	53	Swauger et al. (2002)
GPC LT 100	10.7	67	85	Swauger et al. (2002)
Winston UL 85	5.4	69	62	Swauger et al. (2002)
Kool milds 100	11.6	78	79	Swauger et al. (2002)
Marlboro LT 85	10.6	79	113	Swauger et al. (2002)
Capri SSL 100 M	8.6	81	91	Swauger et al. (2002)
Virginia slims UL 100 M	7.2	83	96	Swauger et al. (2002)
Vantage LT 85	8.8	86	66	Swauger et al. (2002)
Marlboro LT 100	11.6	97	142	Swauger et al. (2002)
Cambridge FF 100	14.9	99	111	Swauger et al. (2002)
Monarch FF 85	13.9	102	93	Swauger et al. (2002)
Basic FF 85 M	16.2	102	112	Swauger et al. (2002)
Camel LT Reg 85	10.8	115	76	Swauger et al. (2002)
Pall mall FF 100	12.9	117	181	Swauger et al. (2002)
Marlboro FF 85	16.5	119	175	Swauger et al. (2002)
Marlboro FF 100	14.8	120	191	Swauger et al. (2002)
B&H FF 100 M	15.8	128	168	Swauger et al. (2002)
Salem FF 100 M	17.2	144	168	Swauger et al. (2002)
GPC FF 100	15.2	151	147	Swauger et al. (2002)
Camel FF Reg 85	16.7	188	170	Swauger et al. (2002)
Winston FF 100	15.7	212	129	Swauger et al. (2002)
Commercial J F UL	NR	17	40	Brunnemann et al. (1996)
Commercial I F L	NR	87	138	Brunnemann et al. (1996)
Commercial H F L	NR	106	122	Brunnemann et al. (1996)
Commercial F F M	NR	126	151	Brunnemann et al. (1996)
Commercial E F M	NR	151	264	Brunnemann et al. (1996)
Commercial D F	NR	156	209	Brunnemann et al. (1996)
Commercial A NF	NR	156	278	Brunnemann et al. (1996)
Commercial B NF	NR	168	274	Brunnemann et al. (1996)
Commercial G F M	NR	173	250	Brunnemann et al. (1996)
Commercial C F	NR	194	287	Brunnemann et al. (1996)
Control	8.2	75.8	81.3	PM USA (1998)
Control	9.3	79.1	82.9	PM USA (1998)
Control	8.6	81.7	87.6	PM USA (1998)
Control	7.8	81.7	93.9	PM USA (1998)
Control	8.4	92.9	84.9	PM USA (1998)
Control	8.7	90.0	91.0	PM USA (1999)
Salem UL 85 M SP	5	52	64	Swauger et al. (2002)
Merit UL 85 M SP	4.2	55	96	Swauger et al. (2002)
GPC UL 85 SP	5	63	100	Swauger et al. (2002)

(continued on next page)

Table 1 (*continued*)

Cigarette[a]	Tar (mg/cigarette)	NNK (ng/cigarette)	NNN (ng/cigarette)	Source
Camel LT 85 SP	10.1	64	100	Swauger et al. (2002)
Merit LT 85 SP	7.8	70	152	Swauger et al. (2002)
Montclair LT 100 SP	10.1	73	173	Swauger et al. (2002)
Merit UL 83 CP	5.1	75	164	Swauger et al. (2002)
Kent Gold LT 85 SP	7.8	81	119	Swauger et al. (2002)
Camel LT 85 CP	9.6	85	133	Swauger et al. (2002)
Newport LT 85 M SP	8.9	86	149	Swauger et al. (2002)
Capri LT 100 CP	8.1	87	112	Swauger et al. (2002)
L&M FF 85 SP	12.9	87	178	Swauger et al. (2002)
Kamel red LT 83 CP	10.5	90	135	Swauger et al. (2002)
Doral FF 85 CP	13.1	90	155	Swauger et al. (2002)
Winston select LT 83 CP	8.4	97	116	Swauger et al. (2002)
Doral FF 85 SP	12.1	97	134	Swauger et al. (2002)
Salem LT 85 M SP	9.8	100	138	Swauger et al. (2002)
Parliament LT 85 SP	8.3	101	185	Swauger et al. (2002)
GPC FF 100 SP	12.7	104	162	Swauger et al. (2002)
Marlboro LT 85 SP	11.1	105	160	Swauger et al. (2002)
Basic LT 100 SP	9.5	110	175	Swauger et al. (2002)
B&H LT 100 M SP	10	111	163	Swauger et al. (2002)
Basic LT 85 M SP	10.4	112	178	Swauger et al. (2002)
Marlboro LT 85 CP	10.3	117	194	Swauger et al. (2002)
Winston FF 85 SP	13.1	119	160	Swauger et al. (2002)
More FF 120 SP	14.8	122	164	Swauger et al. (2002)
Marlboro FF 100 SP	13.8	136	208	Swauger et al. (2002)
GPC FF 83 CP	15.1	137	191	Swauger et al. (2002)
Camel wides FF 80 CP	17	142	185	Swauger et al. (2002)
Richland FF 85 M SP	16.7	143	189	Swauger et al. (2002)
Marlboro LT 85 M SP	9.3	144	225	Swauger et al. (2002)
Marlboro FF 85 SP	15.6	146	246	Swauger et al. (2002)
Newport FF 83 M CP	15.8	147	218	Swauger et al. (2002)
Kool FF 85 M SP	15.1	151	224	Swauger et al. (2002)
Marlboro FF 85 CP	14.2	153	247	Swauger et al. (2002)
Virginia slims FF 100 SP	15.5	154	222	Swauger et al. (2002)
Virginia slims FF 100 M SP	13.4	158	269	Swauger et al. (2002)
Salem FF 100 M SP	17.7	159	259	Swauger et al. (2002)
B&H FF 100 M CP	15.1	168	280	Swauger et al. (2002)
Carlton UL 100 CP	0.4	20	31	Swauger et al. (2002)
Carlton 100 SP	0.9	13	26	Swauger et al. (2002)
Doral UL 100 SP	5	28	46	Swauger et al. (2002)
Pyramid UL 100 SP	5.3	43	55	Swauger et al. (2002)
Salem UL 85 M SP	5	46	54	Swauger et al. (2002)
Doral LT 100 SP	10.5	58	91	Swauger et al. (2002)
Basic UL 100 M CP	5.5	58	97	Swauger et al. (2002)
Winston UL 85 SP	5.2	59	82	Swauger et al. (2002)
Vantage 84 SP	8.4	61	74	Swauger et al. (2002)
Marlboro UL 83 CP	5.8	61	87	Swauger et al. (2002)
Kool LT 84 M SP	7.2	67	79	Swauger et al. (2002)
Salem LT 85 M SP	10.2	73	108	Swauger et al. (2002)
Newport LT 80 M CP	8.8	77	96	Swauger et al. (2002)
Bailey 100 SP	12.7	78	56	Swauger et al. (2002)
Old Gold LT 84 SP	9.2	79	96	Swauger et al. (2002)
Capri 120 M CP	12.3	84	93	Swauger et al. (2002)
Kool FF 85 M SP	15.8	85	110	Swauger et al. (2002)
Camel LT 83 CP	10	86	113	Swauger et al. (2002)
Camel LT 85 SP	11.5	88	117	Swauger et al. (2002)
Marlboro LT 84 SP	11.3	89	123	Swauger et al. (2002)
Winston LT 85 SP	9.3	92	110	Swauger et al. (2002)
GPC 100 SP	13.4	98	146	Swauger et al. (2002)
Basic 99 SP	10.6	100	143	Swauger et al. (2002)
Salem 85 M SP	17.4	102	165	Swauger et al. (2002)
Marlboro LT 84 CP	10.8	103	144	Swauger et al. (2002)
Monarch 100 SP	13.1	105	163	Swauger et al. (2002)
Newport 120 M CP	12	108	160	Swauger et al. (2002)
Winston 84 SP	15.2	110	154	Swauger et al. (2002)
Newport 84 M SP	16.8	111	160	Swauger et al. (2002)
Marlboro FF 100 CP	15	119	177	Swauger et al. (2002)
Marlboro Med 83 CP	12	123	165	Swauger et al. (2002)
Newport 80 M CP	16	124	173	Swauger et al. (2002)
Marlboro 80 CP	14.7	124	186	Swauger et al. (2002)
Camel 83 CP	15.3	131	184	Swauger et al. (2002)
Marlboro King 84 SP	16.5	133	196	Swauger et al. (2002)
Newport 100 M CP	18.7	149	211	Swauger et al. (2002)
USA 85 SP	12.5	185	328	Swauger et al. (2002)
Basic NF 85 SP	28.1	262	343	Swauger et al. (2002)
Merit KS F SP Ultima	1.3	19.2	38	Counts et al. (2005)

Table 1 (*continued*)

Cigarette[a]	Tar (mg/cigarette)	NNK (ng/cigarette)	NNN (ng/cigarette)	Source
Virginia slims 100 F HP ULt Men	5.1	53.4	78.6	Counts et al. (2005)
Merit KS F SP ULt	4.9	53.9	103.5	Counts et al. (2005)
Marlboro KS F HP ULt Men	5.9	55.1	95.1	Counts et al. (2005)
Marlboro 100 F HP Lt	9.6	87.4	126.9	Counts et al. (2005)
Parliament 100 F SP Lt	11.6	101.3	170.8	Counts et al. (2005)
Marlboro KS F SP	14.2	107.8	157	Counts et al. (2005)
Control	7.5	74.9	105.0	PM USA (2001)
Control	7.5	86.5	99.8	PM USA (2001)
Control	7.7	91.1	104.3	PM USA (2001)
Control	7.3	93.9	127.8	PM USA (2001)
Control	7.4	93.9	94.9	PM USA (2001)
Control	7.2	96.4	106.2	PM USA (2001)
Control	7.5	116.3	138.3	PM USA (2001)
Control	7.4	116.8	152.5	PM USA (2001)
Control	7.7	133.5	150.8	PM USA (2001)
Control	8.6	134.0	166.0	PM USA (2001)
Now KS SP	1.5	23	64	Counts et al. (2006)
Merit ultima 100 HP	2.7	27	55	Counts et al. (2006)
Monarch ULT KS SP	3.6	38	97	Counts et al. (2006)
Liggett select LT 100 SP	9.6	51	88	Counts et al. (2006)
Kent golden lights KS HP	8.1	51	102	Counts et al. (2006)
Merit ULT KS HP	4.9	56	106	Counts et al. (2006)
Virginia slims ULT 100 HP	5.1	58	94	Counts et al. (2006)
Newport LT KS HP	9.5	58	107	Counts et al. (2006)
Kool lights KS SP	8.5	60	96	Counts et al. (2006)
Winston LT KS HP	8.5	61	95	Counts et al. (2006)
Winston Evo LT tin	8.9	61	149	Counts et al. (2006)
Marlboro ULT KS HP	6.2	63	89	Counts et al. (2006)
GPC LT KS HP	9.1	63	96	Counts et al. (2006)
Basic ULT KS HP	5.9	64	88	Counts et al. (2006)
Doral LT KS HP	10.1	71	169	Counts et al. (2006)
Camel LT KS HP	10	74	104	Counts et al. (2006)
Winston KS HP	12.5	75	189	Counts et al. (2006)
Merit LT KS HP	8.2	80	126	Counts et al. (2006)
Doral KS HP	14.3	91	210	Counts et al. (2006)
Basic LT KS HP	10	94	125	Counts et al. (2006)
Marlboro LT KS HP	10.5	96	139	Counts et al. (2006)
Marlboro KS HP	15	104	165	Counts et al. (2006)
Kool KS HP Men	17.1	105	156	Counts et al. (2006)
Salem KS SP Men	16.9	105	234	Counts et al. (2006)
Parliament LT 100 HP	11.4	106	176	Counts et al. (2006)
Basic KS HP	15	124	180	Counts et al. (2006)
Marlboro ultra lights KS HP	5.9	65.0	94.0	PM USA (2002)
Marlboro lights KS HP	9.9	95.0	143.0	PM USA (2002)
Marlboro LS HP	15.0	106.0	170.0	PM USA (2002)
Basic KS SP	14.7	124.0	180.0	PM USA (2002)
Control	8.1	97.1	130.5	PM USA (2002)
Control	7.3	98.2	110.9	PM USA (2002)
Control	8.4	98.7	145.0	PM USA (2002)
Control	7.9	99.0	143.4	PM USA (2002)
Control	7.9	102.0	136.0	PM USA (2002)
Control	7.5	108.8	132.8	PM USA (2002)
Control	7.1	112.3	144.8	PM USA (2002)
Control	7.6	115.8	145.3	PM USA (2002)
Control	7.8	118.0	123.1	PM USA (2002)
Control	8.3	121.0	152.0	PM USA (2002)
Control	7.0	133.5	146.3	PM USA (2002)
Control	7.3	158.8	189.3	PM USA (2002)
Control	7.7	160.0	160.0	PM USA (2002)
Marlboro ultra lights KS HP	6.2	54.0	96.0	PM USA (2003)
Marlboro ultra lights KS HP	6.4	62.0	110.0	PM USA (2003)
Marlboro lights KS HP	10.6	85.0	138.0	PM USA (2003)
Marlboro lights KS HP	10.8	92.0	148.0	PM USA (2003)
Marlboro blend 27 HP	12.2	93.0	140.0	PM USA (2003)
Marlboro blend 27 HP	12.3	108.0	150.0	PM USA (2003)
Basic KS SP	14.9	127.0	169.0	PM USA (2003)
Basic KS SP	15.5	128.0	178.0	PM USA (2003)
Control	8.4	92.3	167.4	PM USA (2003)
Control	7.8	105.9	152.6	PM USA (2003)
Control	7.2	110.0	157.0	PM USA (2003)
Control	8.1	112.0	147.0	PM USA (2003)
Control	7.3	112.0	169.8	PM USA (2003)
Control	7.6	115.0	155.0	PM USA (2003)
Control	7.9	117.0	165.0	PM USA (2003)
Control	7.5	118.0	171.0	PM USA (2003)

(*continued on next page*)

Table 1 (*continued*)

Cigarette[a]	Tar (mg/cigarette)	NNK (ng/cigarette)	NNN (ng/cigarette)	Source
Control	7.6	124.0	165.0	PM USA (2003)
Control	7.7	125.3	157.0	PM USA (2003)
Control	8.3	126.0	177.0	PM USA (2003)
Control	7.7	126.8	164.2	PM USA (2003)
Marlboro ultra lights KS HP	6.0	49.0	92.0	PM USA (2004)
Marlboro ultra lights KS HP	6.1	51.0	93.0	PM USA (2004)
Marlboro ultra lights KS HP	6.2	53.0	93.0	PM USA (2004)
Marlboro ultra lights KS HP	6.0	58.0	99.0	PM USA (2004)
Marlboro lights KS HP	10.6	74.0	132.0	PM USA (2004)
Marlboro lights KS HP	10.6	77.0	137.0	PM USA (2004)
Marlboro lights KS HP	10.5	77.0	137.0	PM USA (2004)
Marlboro lights KS HP	10.6	85.0	136.0	PM USA (2004)
Marlboro blend 27 HP	12.6	89.0	131.0	PM USA (2004)
Marlboro blend 27 HP	12.7	91.0	139.0	PM USA (2004)
Marlboro LS HP	15.3	94.0	176.0	PM USA (2004)
Marlboro LS HP	15.5	95.0	177.0	PM USA (2004)
Marlboro blend 27 HP	12.4	100.0	136.0	PM USA (2004)
Marlboro blend 27 HP	13.1	100.0	143.0	PM USA (2004)
Basic KS SP	15.3	116.0	156.0	PM USA (2004)
Basic KS SP	15.5	118.0	168.0	PM USA (2004)
Basic KS SP	15.3	120.0	161.0	PM USA (2004)
Basic KS SP	15.0	130.0	175.0	PM USA (2004)
Control	8.0	77.6	116.0	PM USA (2004)
Control	7.6	82.0	116.0	PM USA (2004)
Control	8.5	87.4	122.0	PM USA (2004)
Control	8.7	87.8	124.0	PM USA (2004)
Control	8.2	89.4	121.0	PM USA (2004)
Control	7.7	89.5	133.0	PM USA (2004)
Control	7.9	96.4	146.0	PM USA (2004)
Control	8.1	102.0	128.0	PM USA (2004)
Control	6.7	110.0	132.0	PM USA (2004)
Control	7.0	122.0	143.0	PM USA (2004)
Control	8.3	122.0	152.0	PM USA (2004)
Merit ultima	1.63	16.4	42	Roemer et al. (2004)
Virginia slims superslim	4.88	51.2	66	Roemer et al. (2004)
Virginia slims ultra lights	5.47	56.4	83	Roemer et al. (2004)
Marlboro lights	10	94.2	137	Roemer et al. (2004)
Benson & hedges 100 lights	11.23	99.2	139	Roemer et al. (2004)
Marlboro	14.51	128.1	196	Roemer et al. (2004)
Parliament lights 100	11.8	131.2	196	Roemer et al. (2004)
Basic	23.05	223.5	270	Roemer et al. (2004)
Marlboro ultra lights KS HP	6.2	43.0	83.0	PM USA (2005)
Marlboro ultra lights KS HP	6.1	43.0	87.0	PM USA (2005)
Marlboro ultra lights KS HP	5.9	46.0	85.0	PM USA (2005)
Marlboro ultra lights KS HP	6.1	48.0	89.0	PM USA (2005)
Marlboro lights KS HP	11.3	67.0	132.0	PM USA (2005)
Marlboro lights KS HP	11.2	68.0	126.0	PM USA (2005)
Marlboro lights KS HP	10.9	70.0	123.0	PM USA (2005)
Marlboro lights KS HP	11.0	73.0	131.0	PM USA (2005)
Marlboro Blend 27 HP	12.4	78.0	134.0	PM USA (2005)
Marlboro LS HP	15.2	81.0	161.0	PM USA (2005)
Marlboro Blend 27 HP	12.4	82.0	127.0	PM USA (2005)
Marlboro LS HP	15.4	83.0	160.0	PM USA (2005)
Marlboro Blend 27 HP	13.3	84.0	132.0	PM USA, 2005
Marlboro LS HP	14.9	85.0	155.0	PM USA (2005)
Marlboro Blend 27 HP	12.0	89.0	139.0	PM USA (2005)
Marlboro LS HP	15.2	92.0	165.0	PM USA (2005)
Basic KS SP	15.7	97.0	139.0	PM USA (2005)
Basic KS SP	14.8	103.0	141.0	PM USA (2005)
Basic KS SP	15.3	104.0	134.0	PM USA (2005)
Basic KS SP	15.7	118.0	155.0	PM USA (2005)
Control	7.4	72.5	112.0	PM USA (2005)
Control	8.5	74.7	125.4	PM USA (2005)
Control	7.9	75.8	137.0	PM USA (2005)
Control	7.8	78.7	146.0	PM USA (2005)
Control	8.0	83.4	118.0	PM USA (2005)
Control	8.5	85.0	155.2	PM USA (2005)
Control	8.3	89.2	140.2	PM USA (2005)
Control	8.1	89.7	145.2	PM USA (2005)
Marlboro ultra lights KS HP	5.6	45.0	80.0	PM USA (2006)
Marlboro ultra lights KS HP	5.6	46.0	81.0	PM USA (2006)
Marlboro lights KS HP	10.7	62.0	120.0	PM USA (2006)
Marlboro lights KS HP	10.7	72.0	124.0	PM USA (2006)
Marlboro blend 27 HP	12.0	75.0	124.0	PM USA (2006)
Marlboro LS HP	14.6	78.0	151.0	PM USA (2006)

Table 1 (*continued*)

Cigarette[a]	Tar (mg/cigarette)	NNK (ng/cigarette)	NNN (ng/cigarette)	Source
Marlboro blend 27 HP	12.0	84.0	124.0	PM USA (2006)
Marlboro LS HP	14.6	85.0	149.0	PM USA (2006)
Basic KS SP	14.6	91.0	133.0	PM USA (2006)
Basic KS SP	14.6	92.0	137.0	PM USA (2006)
Control	7.9	69.7	108.0	PM USA (2006)
Control	8.5	76.8	122.0	PM USA (2006)
Control	9.0	81.6	152.2	PM USA (2006)
Control	8.0	91.7	133.9	PM USA (2006)
Control	8.1	77.6	108.0	PM USA (2007)
Marlboro ultra lights	5.77	49	79	Patskan et al. (2008)
Marlboro lights	9.58	82	122	Patskan et al. (2008)
Marlboro full flavor	13.5	104	151	Patskan et al. (2008)
Natural American spirit	19.3	18.7	18.5	PM USA (2010)
Marlboro 72s silver pack	6.2	35.8	82.2	PM USA (2010)
Marlboro silver pack KS	6.7	36.6	59.5	PM USA (2010)
Camel blue KS	10.4	38.8	47.5	PM USA (2010)
Marlboro virginia blend KS	11.7	41.3	22.9	PM USA (2010)
Merit gold pack KS	7.4	45.0	70.2	PM USA (2010)
Newport menthol box	15.5	47.0	125.1	PM USA (2010)
Kool menthol filter KS box	15.8	48.2	69.2	PM USA (2010)
Marlboro menthol gold pack KS	10.7	49.7	71.2	PM USA (2010)
Marlboro gold pack KS	11.1	53.5	81.2	PM USA (2010)
Pall mall red 100	14.6	66.8	87.3	PM USA (2010)
Basic box KS	14.7	89.7	125.0	PM USA (2010)
Marlboro virginia blend	NR	25.5	19.5	Stepanov et al. (2012)
Camel No. 9 menthol	NR	43.4	75.2	Stepanov et al. (2012)
Camel No. 9	NR	44.4	102.8	Stepanov et al. (2012)
Camel silver	NR	45.6	100.3	Stepanov et al. (2012)
Camel crush	NR	48.1	96.9	Stepanov et al. (2012)
Kool filter kings	NR	63	135.8	Stepanov et al. (2012)
Newport menthol	NR	65.6	151.8	Stepanov et al. (2012)
Camel full flavor	NR	67.5	120.2	Stepanov et al. (2012)
Pall mall full flavor	NR	72.8	114.3	Stepanov et al. (2012)
Marlboro special blend	NR	77.3	141.6	Stepanov et al. (2012)
Winston full flavor	NR	78.5	172.8	Stepanov et al. (2012)
Marlboro smooth menthol	NR	86.4	164.2	Stepanov et al. (2012)
Marlboro full flavor	NR	90.3	171	Stepanov et al. (2012)
Marlboro blend No. 27	NR	91.2	145.2	Stepanov et al. (2012)
Doral full flavor	NR	100.4	225.9	Stepanov et al. (2012)
Marlboro blend No. 54	NR	133.7	232.1	Stepanov et al. (2012)
Basic full flavor	NR	146.1	207.1	Stepanov et al. (2012)
Marlboro virginia blend KS	11.7	19.6	18.9	PM USA (2011)
Natural American spirit (Blue Pack)	15.1	22.2	18.6	PM USA (2011)
Marlboro virginia blend KS	11.4	22.2	20.9	PM USA (2011)
Marlboro virginia blend KS	10.8	25.1	26.2	PM USA (2011)
Natural American spirit (Blue Pack)	19.1	25.6	20.4	PM USA (2011)
Natural American spirit	17.9	31.5	30.0	PM USA (2011)
Marlboro 72s silver pack	6.3	33.3	72.93	PM USA (2011)
Marlboro 72s silver pack	6.9	35.6	77.3	PM USA (2011)
Marlboro Silver Pack KS	6.0	36.0	56.5	PM USA (2011)
Marlboro 72s silver pack	5.8	36.1	78.9	PM USA (2011)
Marlboro virginia blend KS	11.6	37.3	32.0	PM USA (2011)
Marlboro silver pack KS	6.4	39.1	64.9	PM USA (2011)
Natural American spirit (Blue Pack)	18.6	39.6	23.7	PM USA (2011)
Marlboro silver pack KS	6.3	40.1	62.3	PM USA (2011)
Marlboro 72s silver pack	6.5	40.1	76.2	PM USA (2011)
Marlboro silver pack KS	5.9	42.8	67.4	PM USA (2011)
Camel Blue KS	10.4	43.3	59.2	PM USA (2011)
Newport Menthol Box	14.8	44.7	97.5	PM USA (2011)
Merit Gold Pack KS	7.3	45.0	82.6	PM USA (2011)
Camel Blue KS	10.7	45.0	71.0	PM USA (2011)
Newport Menthol Box	14.0	47.4	98.5	PM USA (2011)
Merit Gold Pack KS	7.5	47.8	73.6	PM USA (2011)
Camel Blue KS	11.2	48.6	64.1	PM USA (2011)
Merit Gold Pack KS	7.1	49.6	72.4	PM USA (2011)
Newport Menthol Box	14.6	53.8	130.0	PM USA (2011)
Camel Blue KS	12.0	54.0	85.0	PM USA (2011)
Merit Gold Pack KS	7.3	55.1	86.2	PM USA (2011)
Marlboro Gold Pack KS	10.0	57.4	86.8	PM USA (2011)
Newport Menthol Box	14.4	57.4	127.0	PM USA (2011)
Marlboro Menthol Gold Pack KS	9.5	57.6	70.7	PM USA (2011)
Marlboro Gold Pack KS	10.3	58.2	88	PM USA (2011)
Marlboro Menthol Gold Pack KS	9.4	59.1	77.3	PM USA (2011)
Marlboro Menthol Gold Pack KS	9.0	60.4	83.5	PM USA (2011)
Kool Menthol Filter KS Box	15.1	63.5	83.4	PM USA (2011)

(*continued on next page*)

Table 1 (*continued*)

Cigarette[a]	Tar (mg/cigarette)	NNK (ng/cigarette)	NNN (ng/cigarette)	Source
Marlboro Gold Pack KS	10.9	63.6	96.3	PM USA (2011)
Marlboro Menthol Gold Pack KS	9.1	66.4	95.4	PM USA (2011)
Marlboro Gold Pack KS	10.4	67.7	99.7	PM USA (2011)
Kool Menthol Filter KS Box	15.6	69.6	113.0	PM USA (2011)
Pall Mall Red	13.1	70.0	103.5	PM USA (2011)
Pall Mall Red KS	14.6	70.6	76.2	PM USA (2011)
Kool Menthol Filter KS Box	16.3	73.7	87.4	PM USA (2011)
Basic Box KS	11.4	73.8	108.0	PM USA (2011)
Pall Mall Red 100	13.3	79.3	103.0	PM USA (2011)
Kool Menthol Filter KS Box	15.4	79.4	101.0	PM USA (2011)
Doral Red 100's	13.3	83.4	95.4	PM USA (2011)
Pall Mall Red KS	13.8	85.5	82.9	PM USA (2011)
Basic Box KS	14.2	85.6	126.0	PM USA (2011)
Basic Box KS	14.1	86.8	112	PM USA (2011)
Basic Box KS	14.7	98.5	139.7	PM USA (2011)
Marlboro Virginia Blend KS	11.4	19.6	18.4	PM USA (2012)
Natural American Spirit (Blue Pack)	18.6	25.1	23.3	PM USA (2012)
Marlboro 72s silver pack	5.6	36.1	64.7	PM USA (2012)
Marlboro silver pack KS	6.3	38.2	55.9	PM USA (2012)
Camel blue KS	10.6	48.5	67.1	PM USA (2012)
Merit gold pack KS	6.2	48.9	67.8	PM USA (2012)
Newport menthol box	14.9	53.1	88.2	PM USA (2012)
Marlboro menthol gold pack KS	10.5	63.0	78.2	PM USA (2012)
Marlboro gold pack KS	10.6	63.9	88.2	PM USA (2012)
Kool menthol filter KS Box	15.2	66.1	91.3	PM USA (2012)
Pall mall red KS	13.1	69.1	81.5	PM USA (2012)
Basic box KS	14.8	85.7	103.4	PM USA (2012)

NNK = 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone; NNN = *N*-nitrosonornicotine; NR = not reported; PM USA = Philip Morris USA.
[a] See referenced sources for cigarette sample descriptions. For PM USA data, abbreviations are: HP = hard pack; KS = king size; LS = long size; SP = soft pack.

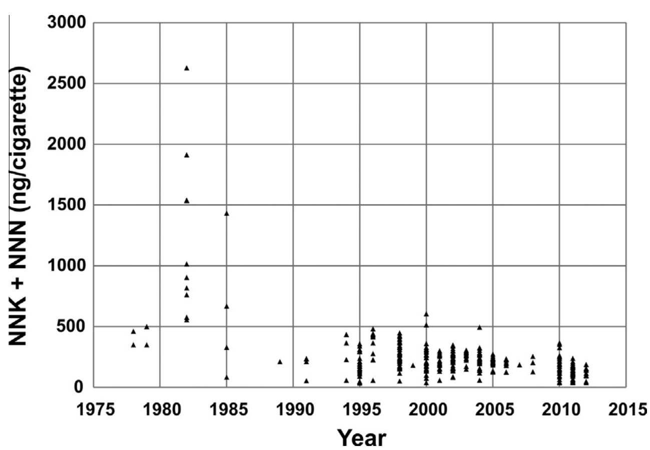

Fig. 1. Scatter plot of 35 years of data as the sum of NNK plus NNN in mainstream smoke, expressed as ng/cigarette.

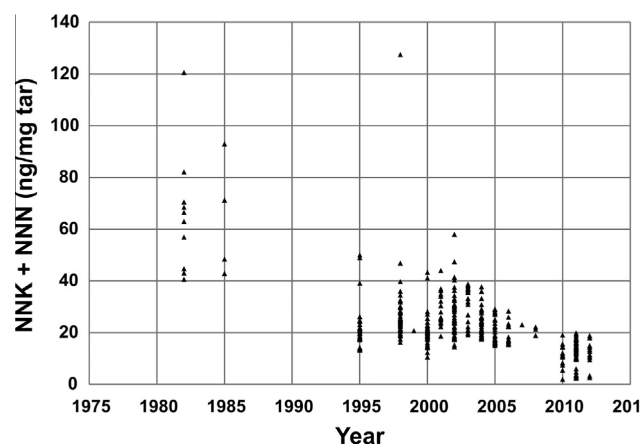

Fig. 2. Scatter plot of 35 years of data as the sum of NNK plus NNN in mainstream smoke, expressed as ng/mg tar.

levels in cigarette smoke over that time period. A scatter plot of these data is presented in Fig. 3.

These data show a scatter of a wide range of values for NNK per cigarette from 1978–1995. Values for smoke NNK levels presented in the Hoffmann papers as evidence of an increasing trend in TSNA levels fall within the overall range of scatter plot values. NNK per cigarette trended down from 1978–1995 ($R = -0.57$) and the trend was statistically significant ($p < 0.00001$). This result is in contrast with the conclusions of Hoffmann and Hoffmann (1997).

3.3. About analytical methods for measuring MS smoke TSNA levels

A variety of methods have been used to quantify MS smoke TSNA levels over the past three decades. Prior to the mid-1990s, the methods were essentially research methods with little published documentation related to method validation and standardization across various laboratories. Figs. 1 and 2 show that TSNA levels in cigarette smoke within a given year are variable, even when normalized by tar levels. Reported TSNA levels appeared to be higher and more variable prior to the mid-1990s compared with levels reported in post-2000 studies. With the information available, it is not possible to determine the relative contributions of analytical variation and other factors to total variability.

To address the issue of uncertain validation or standardization of methods across various laboratories for smoke TSNA levels over 35 years, we assessed data generated by PM USA and its contracted laboratories over the past 10 years. We selected this time period for several reasons. Analytical methods used in determining TSNAs in MS cigarette smoke have been harmonized among these laboratories starting in the late 1990s. PM USA and its contracted laboratories have undergone laboratory quality systems accreditation, such as ISO 17025, and have participated in collaborative studies

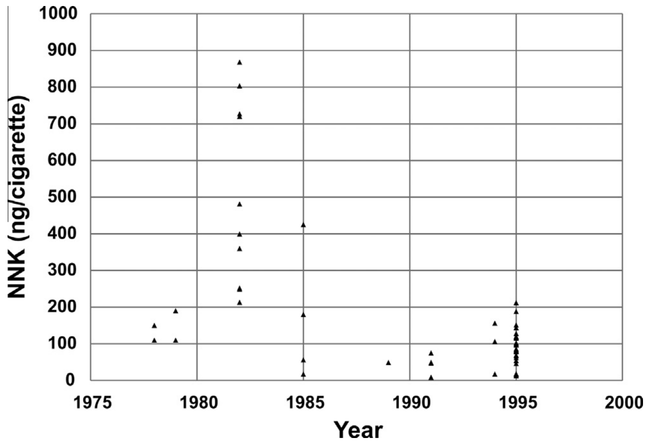

Fig. 3. Scatter plot of data reported for the years 1979–1995 for NNK in mainstream smoke, expressed as ng/cigarette.

to establish recommended methodology for MS cigarette smoke TSNAs. These include CORESTA Methods No. 63 and No. 75 (CORESTA, 2005, 2012). In addition, it is routine practice among these laboratories to measure MS smoke TSNA levels for reference and monitor cigarettes, which allows for method quality control and comparison across laboratories.

Except for data from Stepanov et al. (2012), all of the data presented from 2002–2012 were from PM USA affiliated and contracted laboratories. Because Stepanov et al. (2012) did not present tar levels in their report, these data cannot be compared with other TSNA data expressed on a per mg tar basis. Therefore, all of the data for the years 2002–2012 presented in Fig. 2 were from PM USA affiliated laboratories. This data set includes 219 cigarette samples of various designs and from various manufacturers.

Finally, implementation of various TSNA reduction efforts by the tobacco industry and agricultural community began around the year 2000. If such efforts resulted in reductions in MS smoke TSNA levels, they should be detected in this time period.

Fig. 2 suggests an overall downward trend of MS smoke TSNA levels over the past decade. For the period 2002–2012, (NNK + NNN)/tar trended down over time ($R = -0.70$) and the trend was statistically significant ($p < 0.00001$).

4. Discussion

The US Surgeon General and others state that the role of specific smoke constituents in smoking related disease is currently not known (Burns et al., 2008; USDHHS, 2010). Likewise, it is not known whether reduction of the levels of TSNAs in MS cigarette smoke would result in a reduction in the risk of smoking-related disease.

For more than a decade, PM USA along with other tobacco companies and the agricultural community have actively researched the fundamental science related to TSNA formation, TSNA levels in tobacco and tobacco smoke, and methods to reduce these levels. One outcome of this research was the retrofitting of commercial curing barns, in 2000, from direct-fired to heat-exchanger systems. Prior to that year, Virginia tobacco was cured in barns equipped with direct-fired propane or liquid petroleum gas burners. In 1999, it was recognized that substantial reductions in TSNAs occurred in flue-cured tobacco that was cured in barns equipped with heat-exchangers as compared with tobacco that was cured using the direct-fire method (Nestor et al., 2003; Peele et al., 1999). An industry-wide retrofitting of US commercial curing barns from direct-fired to the heat-exchanger system started in 2000 with farmers' expenses partially reimbursed by an industry-supported cost-share program (Reed, 2009). A similar effort was undertaken in Canada (Rickert et al., 2008). Because the effect of heat exchangers in reducing TSNAs is effective only for Virginia tobaccos, its impact in reducing TSNA levels in cigarette smoke is dependent on the amount of Virginia tobacco in the final tobacco blend. Additional measures have also been implemented to further reduce TSNA levels in burley tobacco. For example, tobacco seed screening for low conversion of nicotine to nornicotine, a precursor to NNN formation in burley tobacco, is now widely implemented (Jack et al., 2007). Additionally, PM USA requires that US tobacco growers adhere to other production practices designed to minimize TSNA for Virginia and burley tobaccos similar to those defined by tobacco production guides issued by several agricultural authorities (North Carolina State University Cooperative Extension, 2011; University of Tennessee Institute of Agriculture and University of Kentucky College of Agriculture, 2011; Virginia Cooperative Extension and Virginia Polytechnic Institute and Virginia State University, 2011).

Evidence suggests that efforts by the tobacco industry and agricultural community to reduce TSNA levels in cigarette tobacco have been making progress. Counts measured tobacco filler TSNA levels in 26 US. commercial cigarette brands that were sampled in 2002 (Counts et al., 2006). The average level of NNK plus NNN in tobacco filler from these 26 brands was 4.4 μg/g. Morton and Laffoon measured tobacco filler TSNA levels in 23 US. commercial cigarette brands that were sampled in 2004–2005 (Morton and Laffoon, 2008). The average level of NNK plus NNN in tobacco filler from these 23 brands was 3.9 μg/g. Stepanov measured tobacco filler TSNA levels in 17 US. commercial cigarette brands that were sampled in 2010 (Stepanov et al., 2012). The average level of NNK plus NNN (adjusted for moisture) of the 17 commercial cigarette samples measured by Stepanov et al. was 2.9 μg/g. This represents a reduction in tobacco filler NNK plus NNN of 35% and 27% from 2002 to 2010 and 2004–2005 to 2010, respectively. These trends are consistent with the trends in smoke NNK plus NNN reported in our work.

In a presentation to the 2012 CORESTA Congress, Gunduz et al. reported that between 2000 and 2010, the average NNN content of tobacco filler from over 250 international cigarette brands was reduced by approximately 30% while the average NNK content was reduced by approximately 50%. These findings are consistent with the reductions in tobacco filler NNK plus NNN in US. commercial cigarettes indicated by comparing the results reported by Counts et al., Morton and Laffoon, and Stepanov et al. noted above. Gunduz et al. also reported substantial reductions in NNK and NNN levels in Virginia and burley tobacco lots produced in the US from 1999 to 2011 (Gunduz et al., 2012).

Contrary to the reductions noted above, Stepanov et al. reported that there has been no change in TSNA levels in cigarette tobacco filler over the past 30 years (Stepanov et al., 2012). The lack of agreement between the previously noted findings and conclusions of Stepanov et al. may be due to the relatively small number of tobacco filler samples used by Stepanov et al. to infer a trend in TSNA levels over three decades. The investigators relied on one sample from an unidentified cigarette in 1979, five samples from unidentified cigarettes in 1995, and 17 samples from identified cigarettes in 2010.

Based on the analysis presented here, TSNA levels in cigarette smoke appear to be on a downward trend and have been for a number of years. This is evident in all scatter plots, but it is particularly evident in plots of TSNA levels that are normalized for tar levels. Likewise, this trend is particularly evident in cigarettes analyzed within the past 10 years. This latter observation is consistent with the timing of implementation of various efforts by the tobacco industry and agricultural community to reduce TSNAs in tobacco.

Our analysis does not support the suggestion that TSNA levels in cigarette smoke have increased over time. Therefore, the conclusion by Hoffmann and Hoffmann (1997) that NNK levels have increased in smoke of commercially available cigarettes from 1979–1995 is not supported by data published during that time period. Likewise, our analysis is contrary to the contention by Stepanov et al. (2012) that there is no apparent significant reduction in smoke TSNA levels over the past decade.

The analyses discussed here should be considered in light of several limitations. The ideal study to address the question of trends in TSNA levels in MS smoke of cigarettes over time would be a longitudinal study spanning many years, employing a single cigarette design manufactured over many tobacco crop years, and using the same validated and standardized methods to analyze all samples over the course of the study. Ideally, such a study would include a reference cigarette that was manufactured at a single point in time and tested on multiple occasions over the course of the study as a test of stability of analytical results over time. To our knowledge, no such study has ever been conducted.

The data that are available come from a variety of sources published over a period of 35 years. These data comprise a wide range of cigarette designs from multiple manufacturers analyzed by various laboratories using a variety of analytical methods which were unlikely to have been standardized across laboratories. Data on variability of the measurements such as standard deviation are not available for some of the data set. With the information available, it is not possible to determine the relative contributions of analytical variation and other factors to total variability.

The samples selected for testing by various investigators may or may not have been representative of the range of all products in the market at a given point in time. Many of the samples were described only as commercial filter or non-filter cigarettes. Despite these limitations, the data set that does exist is relatively large (several hundred cigarette samples) and includes a wide variety of cigarette designs and market leaders. It is therefore reasonable to assume that the data can inform some general inferences about time trends of TSNA levels in cigarette smoke, particularly within time frames narrower than 35 years.

Smoke TSNA data presented in this manuscript was generated by machines smoking cigarettes under FTC/ISO smoking conditions. It is unlikely that smoke TSNA data generated under more intense smoking conditions, such as the Health Canada regimen, would result in a different outcome than the one reported in the present work. While a more intense smoking regimen would result in different absolute levels of smoke TSNA, there is no evidence that such a difference would change over time in a way that would reverse a trend identified by FTC/ISO smoking regimens. In their presentation to the 2012 CORESTA Congress, Gunduz et al. reported that between 2000 and 2010 the average NNK and NNN levels in mainstream cigarette smoke were reduced by approximately 13% and 23% respectively in a study testing a large number of international cigarette brands. This is consistent with our findings despite the fact that smoke TSNA data reported by Gunduz et al. were obtained using Health Canada smoking conditions.

In conclusion, this review of data collected over the past 35 years does not support the suggestion that TSNA levels in MS smoke of US commercial cigarettes have increased over time. On the contrary, the general impression of this data set is that TSNA levels in MS cigarette smoke appear to be generally decreasing. The statistical analyses are consistent with this impression. This trend is particularly evident over the past 10 years. The apparent downward trend in TSNA levels in MS smoke of US commercial cigarettes is consistent with efforts implemented by the tobacco industry and the agricultural community to reduce levels of TSNAs in tobacco and cigarette smoke.

Conflict of interest

The authors are employees of Altria Client Services Inc.

Funding sources

None.

Acknowledgment

The authors acknowledge the editorial assistance of Eileen Y. Ivasauskas of Accuwrit Inc.

References

Adams, J.D., O'Mara-Adams, K.J., Hoffmann, D., 1987. Toxic and carcinogenic agents in undiluted mainstream smoke and sidestream smoke of different types of cigarettes. Carcinogenesis 8, 729–731.

Brunnemann, K.D., Hoffmann, D., 1991. Analytical studies on tobacco-specific N-nitrosamines in tobacco and tobacco smoke. Crit. Rev. Toxicol. 21, 235–240.

Brunnemann, K.D., Prokopczyk, B., Djordjevic, M.V., Hoffmann, D., 1996. Formation and analysis of tobacco-specific N-nitrosamines. Crit. Rev. Toxicol. 26, 121–137.

Burns, D.M., Anderson, C.M., Gray, N., 2011a. Do changes in cigarette design influence the rise in adenocarcinoma of the lung? Cancer Causes Control 22, 13–22.

Burns, D.M., Anderson, C.M., Gray, N., 2011b. Has the lung cancer risk from smoking increased over the last fifty years? Cancer Causes Control 22, 389–397.

Burns, D.M., Dybing, E., Gray, N., Hecht, S., Anderson, C., Sanner, T., O'Connor, R., Djordjevic, M., Dresler, C., Hainaut, P., Jarvis, M., Opperhuizen, A., Straif, K., 2008. Mandated lowering of toxicants in cigarette smoke: a description of the World Health Organization TobReg proposal. Tob. Control 17, 132–141.

Coggins, C.R., Edmiston, J.S., Jerome, A.M., Langston, T.B., Sena, E.J., Smith, D.C., Oldham, M.J., 2011a. A comprehensive evaluation of the toxicology of cigarette ingredients: essential oils and resins. Inhal. Toxicol. 23 (Suppl. 1), 41–69.

Coggins, C.R., Fisher, M.T., Smith, D.C., Oldham, M.J., 2011b. A comprehensive evaluation of the toxicology of cigarette ingredients: cocoa-derived ingredients. Inhal. Toxicol. 23 (Suppl. 1), 70–83.

Coggins, C.R., Frost-Pineda, K., Smith, D.C., Oldham, M.J., 2011c. A comprehensive evaluation of the toxicology of cigarette ingredients: aromatic and aliphatic alcohol compounds. Inhal. Toxicol. 23 (Suppl. 1), 141–156.

Coggins, C.R., Jerome, A.M., Edmiston, J.S., Oldham, M.J., 2011d. A comprehensive evaluation of the toxicology of cigarette ingredients: aliphatic carbonyl compounds. Inhal. Toxicol. 23 (Suppl. 1), 102–118.

Coggins, C.R., Liu, J., Merski, J.A., Werley, M.S., Oldham, M.J., 2011e. A comprehensive evaluation of the toxicology of cigarette ingredients: aliphatic and aromatic carboxylic acids. Inhal. Toxicol. 23 (Suppl. 1), 119–140.

Coggins, C.R., Merski, J.A., Oldham, M.J., 2011f. A comprehensive evaluation of the toxicology of cigarette ingredients: heterocyclic nitrogen compounds. Inhal. Toxicol. 23 (Suppl. 1), 84–89.

Coggins, C.R., Sena, E.J., Langston, T.B., Oldham, M.J., 2011g. A comprehensive evaluation of the toxicology of cigarette ingredients: aromatic carbonyl compounds. Inhal. Toxicol. 23 (Suppl 1), 90–101.

Coggins, C.R., Sena, E.J., Oldham, M.J., 2011h. A comprehensive evaluation of the toxicology of cigarette ingredients: inorganic compounds. Inhal. Toxicol. 23 (Suppl. 1), 157–171.

Coggins, C.R., Wagner, K.A., Werley, M.S., Oldham, M.J., 2011i. A comprehensive evaluation of the toxicology of cigarette ingredients: carbohydrates and natural products. Inhal. Toxicol. 23 (Suppl. 1), 13–40.

CORESTA, 2005. Recommended Method No. 63: determination of tobacco specific nitrosamines in cigarette mainstream smoke – GC-TEA method. <http://www.coresta.org/Recommended_Methods/CRM_63.pdf> (accessed December 2012).

CORESTA, 2012. Recommended Method No. 75: determination of tobacco specific nitrosamines in mainstream cigarette smoke by LC-MS/MS. <http://www.coresta.org/Recommended_Methods/CRM_75.pdf> (accessed December 2012).

Counts, M.E., Morton, M.J., Laffoon, S.W., Cox, R.H., Lipowicz, P.J., 2005. Smoke composition and predicting relationships for international commercial cigarettes smoked with three machine-smoking conditions. Regul. Toxicol. Pharmacol. 41, 185–227.

Counts, M.E., Hsu, F.S., Tewes, F.J., 2006. Development of a commercial cigarette "market map" comparison methodology for evaluating new or non-conventional cigarettes. Regul. Toxicol. Pharmacol. 46, 225–242.

Djordjevic, M.V., Sigountos, C.W., Brunnemann, K.D., Hoffmann, D., 1991. Formation of 4-(methylnitrosamino)-4-(3-pyridyl)butyric acid in vitro and in mainstream cigarette smoke. J. Agric. Food Chem. 39, 209–213.

Gaworski, C.L., Oldham, M.J., Wagner, K.A., Coggins, C.R., Patskan, G.J., 2011. An evaluation of the toxicity of 95 ingredients added individually to experimental cigarettes: approach and methods. Inhal. Toxicol. 23 (Suppl. 1), 1–12.

Gunduz, I., Kondylis, A., Hofer, R., Jaccard, G., Ruffieux, L., Gadani, F., 2012. Levels of tobacco-specific N-nitrosamines NNN and NNK in commercial cigarettes sampled in 2008–2010. A review of analytical data. In: CORESTA Congress, Sapporo 23–27 September 2012, Plenary Session, Abstract IG02. <http://www.coresta.org/Meetings/Past_Abstracts/Sapporo2012-PlenarySessions.pdf> (accessed March 2013).

Hecht, S.S., Chen, C.H.B., Hoffmann, D., 1979. Tobacco-specific nitrosamines occurence, formation, carcinogenicity, and metabolism. Acc. Chem. Res. 12, 92–98.

Hoffmann, D., Adams, J.D., Brunnemann, K.D., Hecht, S.S., 1979. Assessment of tobacco-specific N-nitrosamines in tobacco products. Cancer Res. 39, 2505–2509.

Hoffmann, D., Adams, J.D., Brunnemann, K.D., Rivenson, A., Hecht, S.S., 1982. Tobacco specific N-nitrosamines: occurrence and bioassays. IARC Sci. Publ., 309–318.

Hoffmann, D., Rivenson, A., Murphy, S.E., Chung, F.L., Amin, S., Hecht, S.S., 1993. Cigarette smoking and adenocarcinoma of the lung: the relevance of nicotine-derived N-nitrosamines. J. Smoking-Related Dis. 4, 165–189.

Hoffmann, D., Brunnemann, K.D., Prokopczyk, B., Djordjevic, M.V., 1994. Tobacco-specific N-nitrosamines and Areca-derived N-nitrosamines: chemistry, biochemistry, carcinogenicity, and relevance to humans. J. Toxicol. Environ. Health 41, 1–52.

Hoffmann, D., Hoffmann, I., 1994. Recent developments in smoking-related cancer research. J. Smoking-Related Dis. 5, 77–93.

Hoffmann, D., Hoffmann, I., 1997. The changing cigarette, 1950–1995. J. Toxicol. Environ. Health 50, 307–364.

IARC, 2007. IARC monographs on the evaluation of carcinogenic risks to humans. Volume 89. Smokeless tobacco and some tobacco-specific nitrosamines. <http://monographs.iarc.fr/ENG/Monographs/vol89/mono89.pdf> (accessed December 2012).

Jack, A., Fannin, N., Bush, L., 2007. Implications of reducing nornicotine accumulation in burley tobacco. In: Jelesko, Nelson, Wanna (Eds.). Recent Advances in Tobacco Science: Frontiers in Tobacco Biotechnology. Tobacco Science Research Conference, Charlotte, NC, pp. 39–91.

Morton, M.J., Laffoon, S.W., 2008. Cigarette smoke chemistry market maps under Massachusetts Department of Public Health smoking conditions. Regul. Toxicol. Pharmacol. 51, 1–30.

Nestor, T.B., Gentry, J.S., Peele, D.M., Riddick, M.G., Conner, B.T., Edwards, M.E., 2003. Role of oxides of nitrogen in tobacco-specific nitrosamine formation in flue-cured tobacco. Beitr. Tabakforsch. 20, 467–475.

North Carolina State University Cooperative Extension, 2011. 2011 Flue-cured tobacco guide. <http://ipm.ncsu.edu/Production_Guides/Flue-cured/flue_cured.pdf> (accessed December 2012).

Patskan, G.J., Podraza, K.F., Meurrens, K., Coggins, C.R., Friedrichs, B., Gerstenberg, B., Gomm, W., Schnell, P., Stabbert, R., Veltel, D., Weber, S., Terpstra, P., 2008. Toxicological comparisons of three styles of a commercial US. cigarette Marlboro with the 1R4F reference cigarette. Inhal. Toxicol. 20, 695–721.

Peele, D.M., Riddick, M.G., Eisenbarth, J., Gentry, J.S., Nestor, T.B., 1999. Formation of tobacco specific nitrosamines in flue-cured tobacco. Joint Meeting of the CORESTA Agronomy and Phytopathology Study Groups, Suzhou, China, 14 October 1999.

Reed, D.T., 2009. Curing tobacco. In: 2008 Flue-Cured Tobacco Production Guide. Publication 436–048. Virginia Cooperative Extension and Virginia Polytechnic Institute and State University, Blacksburg, VA, pp. 61–63.

Rickert, W.S., Joza, P.J., Sharifi, M., Wu, J., Lauterbach, J.H., 2008. Reductions in the tobacco specific nitrosamine (TSNA) content of tobaccos taken from commercial Canadian cigarettes and corresponding reductions in TSNA deliveries in mainstream smoke from such cigarettes. Regul. Toxicol. Pharmacol. 51, 306–310.

Roemer, E., Stabbert, R., Rustemeier, K., Veltel, D.J., Meisgen, T.J., Reininghaus, W., Carchman, R.A., Gaworski, C.L., Podraza, K.F., 2004. Chemical composition, cytotoxicity and mutagenicity of smoke from US commercial and reference cigarettes smoked under two sets of machine smoking conditions. Toxicology 195, 31–52.

Stepanov, I., Knezevich, A., Zhang, L., Watson, C.H., Hatsukami, D.K., Hecht, S.S., 2012. Carcinogenic tobacco-specific N-nitrosamines in US cigarettes: three decades of remarkable neglect by the tobacco industry. Tob. Control 21, 44–48.

Swauger, J.E., Steichen, T.J., Murphy, P.A., Kinsler, S., 2002. An analysis of the mainstream smoke chemistry of samples of the US. Cigarette market acquired between 1995 and 2000. Regul. Toxicol. Pharmacol. 35, 142–156.

University of Tennessee Institute of Agriculture and University of Kentucky College of Agriculture, 2011. 2011–2012 Kentucky & Tennessee tobacco production guide. <http://www.ca.uky.edu/agc/pubs/id/id160/id160.pdf> (accessed December 2012).

USDHHS, 2010. A report of the Surgeon General: How tobacco smoke causes disease, 2010. The biology and behavioral basis for smoking-attributable disease. <http://www.surgeongeneral.gov/library/reports/tobaccosmoke/report-index.html> (accessed December 2012).

USDHHS, 2012. US Food and Drug Administration. Harmful and potentially harmful constituents in tobacco products and tobacco smoke: established list. <http://www.fda.gov/TobaccoProducts/GuidanceComplianceRegulatoryInformation/ucm297786.htm> (accessed December 2012).

Virginia Cooperative Extension and Virginia Polytechnic Institute and Virginia State University, 2011. 2012 Flue-cured tobacco production guide. Publication 436-048. <http://pubs.ext.vt.edu/AREC/AREC-14/AREC-14-pdf.pdf> (accessed December 2012).