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Variation in tobacco and mainstream smoke toxicant yields from selected commercial cigarette products

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ABSTRACT

There is a drive toward the mandated lowering and reporting of selected toxicants in tobacco smoke. Several studies have quantified the mainstream cigarette emissions of toxicants, providing benchmark levels. Few, however, have examined how measured toxicant levels within a single product vary over time due to natural variation in the tobacco, manufacturing and measurement. In a single centre analysis, key toxicants were measured in the tobacco blend and smoke of 3R4F reference cigarette and three commercial products, each sampled monthly for 10 months. For most analytes, monthly variation was low (coefficient of variation <15%); but higher (⩾20%) for some compounds present at low (ppb) levels. Reporting toxicant emissions as a ratio to nicotine increased the monthly variation of the 9 analytes proposed for mandated lowering, by 1–2 percentage points. Variation in toxicant levels was generally 1.5–1.7-fold higher in commercial cigarettes compared with 3R4F over the 10-month period, but increased up to 3.5-fold for analytes measured at ppb level. The potential error (2CV) associated with single-point-in-time sampling averaged ~20%. Together, these data demonstrate that measurement of emissions from commercial cigarettes is associated with considerable variation for low-level toxicants. This variation would increase if the analyses were conducted in more than one laboratory.

1. Introduction

Since 2000, cigarette smoke toxicants have slowly developed into a global regulatory issue. Starting with the mandated measurement and reporting of toxicant emissions from cigarettes in Canada (Health Canada, 2000) and Brazil (Brazil Resolution, 2007), the requirement to measure and report emissions has spread to other countries. Regulatory reporting may also include measurement of specific compounds in the cigarette tobacco filler blend and reporting of cigarette physical attributes.

In the United States, the Food and Drug Administration (FDA) has published a list of 93 harmful and potentially harmful constituents (HPHCs) in tobacco products and tobacco smoke (FDA, 2012a) and issued draft guidance on the reporting of an abbreviated list of 24 HPHCs, 18 in mainstream cigarette smoke and 6 in the cigarette filler blend, for which analytical protocols are well established and widely available although currently not standardised (Table 1) (FDA, 2012b). The FDA has also introduced a pre-market approval process, wherein toxicant emissions from cigarettes are evaluated (among other information) before permission is granted to market new tobacco products. This legislation, embodied in the US Family Smoking Prevention and Tobacco Control Act, also empowers the FDA to enact toxicant reduction strategies, although they have yet to do so (US, 2009).

The World Health Organization (WHO) Study Group on Tobacco Product Regulation (TobReg), composed of leading public health scientists, has been working towards a scientific basis for tobacco product regulation (WHO, 2008). As summarised by Burns et al. (2008), TobReg concluded that chemical measurements of smoke produced by smoking machines is probably the most effective approach currently available for scientifically assessing differences between products for regulatory assessment of product toxicity. TobReg has proposed the measurement and reporting of selected smoke toxicants and some compounds in cigarette filler blends. It has taken the further step of proposing mandated ceilings on

Abbreviations: FDA, Food and Drug Administration; HPHCs, harmful and potentially harmful constituents; TNCO, tar, nicotine and carbon monoxide; TobReg, the WHO Study Group on Tobacco Product Regulation; TSNA, tobacco-specific nitrosamine; WHO, World Health Organization; HCI, Health Canada intense machine smoking regime as determined by Health Canada; dwb, dry weight basis.

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emissions for nine of these selected toxicants as a means of detoxifying cigarette smoke (Table 1) (WHO, 2008). These proposed ceilings are based on toxicant measurements determined under the intense machine smoking regime developed by Health Canada (HCI) when the levels are expressed as a ratio to the nicotine yield (Hammond et al., 2007). The developing WHO Framework Convention on Tobacco Control (FCTC) represents a mechanism whereby toxicant reporting and proposed ceiling regulations might spread worldwide.

Several studies have reported levels of toxicants in both cigarette smoke and the tobacco blend (Health Canada, 2004; Gregg et al., 2004; Australian DOH, 2002). For example, comprehensive data on mainstream smoke constituents of contemporary cigarettes, based on standardised machine-smoking methods, have been compiled by both Borgerding et al. (2000), who monitored 26 leading brands from the United States by FTC/ISO parameters for 44 constituents, and Counts et al. (2005), who analysed smoke and cigarette tobacco filler blends from 48 commercial cigarettes from international markets, smoked across 3 regimes, for tar and 44 constituents. However, the majority of data were compiled from a single sample of product (Borgerding et al., 2000; Counts et al., 2005; Gregg et al., 2004; Australian DOH, 2002), providing snapshots in time.

By contrast, fewer studies document how toxicant levels within a given product might vary over time. Natural variation in levels might be expected among product batches due to changes in both the tobacco sources for the blend and process fluctuations in cigarette manufacturing steps. Design alterations in commercial cigarette products also occur from time to time, which may impact on toxicant emission levels.

Another important source of variation is measurement uncertainty, that is, analytical variation. Many studies have estimated the variability of various smoke analyte measurements within a single laboratory (Rickert and Wright, 2002) and across several laboratories (Hyodo et al., 2006; Intorp et al., 2009; Teillet et al., 2013; Purkis and Intorp, 2014). These studies, using reference cigarettes, have concluded that smoke toxicant measurements are generally more variable as compared with measurements of tar, nicotine and carbon monoxide (TNCO), both within and among laboratories, and that measurements are more variable among laboratories than within a single laboratory. Morton and Laffoon (2008) described both temporal cigarette and testing variation, in their extension of a market-mapping approach to compare cigarette products using the puffing regime defined by the Massachusetts Department of Public Health. They noted that market maps and the associated prediction intervals calculated from single-point-in-time samples were likely to understate the true variability that would be expected over time. More recently, the long-term and short-term variability of toxicant emissions was compared for the 9 priority smoke toxicants identified by TobReg for several commercial cigarette products from the Japanese market (Minagawa et al., 2012). Statistically significant analytical variability was also observed in the measurement of most of the 96 HPHCs on the FDAs list, using single manufactured lots of samples of 20 commercial cigarette products determined at two time-points (Oldham et al., 2014). This paper highlights the need for standardised analytical methods with established repeatability

Table 1
Study toxicants with regulatory relevance.

Toxicant	Abbr.	Regulatory relevance		
		TobReg proposal for		FDA initial list
		Mandated lowering	Reporting	
Smoke				
4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone	NNK	√		√
N-nitrosonornicotine	NNN	√		√
Acetaldehyde		√		√
Acrolein		√		√
Acrylonitrile			√	√
4-Aminobiphenyl	4-ABP		√	√
1-Aminonaphthalene	1-AN			√
2-Aminonaphthalene	2-AN		√	√
Ammonia	NH_3			√
Benzene		√		√
Benzo[a]pyrene	B[*a*]P	√		√
1,3-Butadiene		√		√
Cadmium			√	
Carbon monoxide	CO	√		√
Catechol			√	
Crotonaldehyde			√	√
Formaldehyde		√		√
Hydrogen cyanide	HCN		√	
Hydroquinone			√	
Isoprene				√
Nicotine				√
Nitrogen oxides	NO*x*		√	
Toluene				√
Cigarette filler blend				
Ammonia	NH_3		√	√
4-(Methylnitrosamino)-1-(3-pyridyl)-1-butanone	NNK			√
N-nitrosonornicotine	NNN			√
Arsenic	As			√
Cadmium	Cd			√
Glycerol			√	
Nicotine (Total)			√	√
Propylene glycol	PG		√	
Triethylene glycol			√	

and reproducibility, ideally using certified reference materials for accuracy, and inter-laboratory proficiency testing studies to demonstrate laboratory capability.

An understanding of result variation is required in order to put toxicant measurement data into context. The aim of the present study was to provide data on the variability of smoke toxicant emissions and cigarette filler blend components for three high-volume commercial cigarette products in order to understand how representative a sample of a commercial cigarette product taken at a single point in time is to the general performance of a product over time (Table 1).

2. Materials and methods

2.1. Materials

Three high-volume, internationally available, commercial cigarette products were acquired from a single market: two British American Tobacco (BAT) products, one at 10 mg ISO tar (product A) and one at 4 mg ISO tar (product C), acquired directly from the BAT factory in Bayreuth, Germany; and one high-volume commercial product from another manufacturer at 10 mg ISO tar (product B), purchased from a nearby German retail outlet. All three products were king-size in length (~84 cm) and circumference (~24 mm), with a single sectioned cellulose acetate filter, non-banded cigarette paper and US blended style tobacco filler blend, typical of the German market. Product samples were collected monthly for 10 consecutive months from August 2010 to May 2011 inclusive. The BAT product did not undergo any product design changes over the course of this study; however, this aspect was unknown for the non-BAT product.

A laboratory monitor cigarette, the University of Kentucky (UKy) Reference Cigarette 3R4F (3R4F), was included in every batch of analysis. The UKy reference cigarettes are manufactured using a standard design that is specified by the University of Kentucky Tobacco Research Institute. They are provided to researchers worldwide, and are widely used, as consistent and uniform test items for research purposes [3R4F.com]. The 3R4F reference cigarette was manufactured under normal production tolerances over 11 consecutive production days, producing 60 million cigarettes; this amount was designed to last 10 years, with stock levels in 2013 at 25 million (Chambers et al., 2013). This is the third production run of this design of cigarette, subsequent production runs of this design will be designated 4R4F, 5R4F, and so on.

2.2. Study protocol

All analyses were conducted at a single, ISO 17025 accredited BAT laboratory. Forty-four toxicants in mainstream cigarette smoke were determined, covering the lists of toxicants currently required for regulatory reporting by Health Canada and ANVISA (Health Canada, 2000; Brazil Resolution, 2007). Thirty-four components in the cigarette tobacco blend were determined using the analytical methods in routine use by this laboratory, again covering the lists of blend components currently required for regulatory reporting by Health Canada and ANVISA (Health Canada, 2000; Brazil Resolution, 2007). The three commercial products were acquired and sent to the analytical laboratory each month over the 10-month study period and the analysis schedule was not controlled, i.e. a real-world approach to sample analysis was used.

Each sample of product was prepared for analysis according to ISO 8243 without further selection by, for example, weight or pressure drop. The three commercial product samples from a single month were generally analysed in a single analytical batch, which included a sample of 3R4F. In some cases, two monthly batches of product were combined into a single analysis batch, or the three products from one month were split over two analysis batches. This resulted in between 7 and 12 analytical batches per toxicant analysis, and correspondingly between 7 and 12 analytical samples of 3R4F. Five replicate analyses were conducted per monthly sample of product for most toxicants because analytical experience over the past 15 years has shown that this is more than sufficient to provide reproducible data. For TNCO, however, eight replicate analyses were routinely conducted in accordance with the ISO requirements for regulatory reporting (which stipulate that measurements should be from an average of 40 cigarettes, equating to 8 replicates on a linear smoking machine that smokes 5 cigarettes per replicate (ISO, 2013)). Other regulators stipulate seven replicates per analyte (FDA, 2012b; Health Canada, 2000).

2.3. Analysis methods

For mainstream smoke analyses, the products were smoked under two smoking regimes: ISO 3308 (ISO) (ISO, 2012) and Health Canada Intense (HCI) (Hammond et al., 2007). Because the toxicants were measured by different analytical procedures, the weight of the cigarettes used in each analysis, the puff number and, where available, the total particulate matter (TPM) were also recorded.

The mainstream smoke toxicants and blend components were analysed by standard methods that have been internally validated for repeatability and reproducibility (AOAC, 2002; ISO, 1994). Most of these methods follow, or are based on, internationally accredited protocols (i.e. ISO, CORESTA or official Health Canada methods). The methods are multi-analyte, whereby members of a group of toxicants (e.g. volatile organic compounds or carbonyls or aromatic amines, etc.) are analysed simultaneously from the same cigarettes. The details of the analytical methods and appropriate references are summarised in Supplementary Tables 1 and 2, and further details are available on request.

2.4. Data analysis

Statistical analysis was carried out using Minitab® v16.0 (Minitab Inc., USA) and SAS 9.3 (SAS Institute, Cary NC, USA) software.

As discussed above, toxicant emission and filler blend component levels were measured from product sampled each month using 5 or 8 replicate analyses. The reported mean result of replicate analyses from a single product sample (and the associated SD) is the result that is usually reported for regulatory purposes (Health Canada, 2000; FDA, 2012b). Summary statistics of these mean monthly product sample results were used to describe toxicant data per product across the study period (No. of monthly samples, mean, SD, CV). CV (%) was used to compare monthly variation within and between products. The ratio to nicotine of each smoke toxicant (toxicant emission/nicotine emission) was also calculated for each monthly product sample using the mean result (of the replicates for that monthly sample), and summary statistics determined for these ratio results (mean, SD, CV).

For data that were below the limit of detection (<LOD) or below the limit of quantitation (<LOQ), values of ½LOD or ½(LOD + LOQ), respectively, were used. If the majority of the replicates were <LOQ, then SD and CV were not reported.

The empirical rule that 2× SD gives an approximation of the 95% confidence intervals around the mean for a normal distribution was used to estimate the within-laboratory 'tolerance' around a single measured value. All replicate results for each product were visually screened to check for normality (histogram of all replicates, plus comparison of 95% confidence intervals of mean and median results). All results were approximately normal, with a few exceptions where results were near or below LOQ (nitrite, myosmine, triacetin in the filler blend plus some of the metals in

both filler blend and smoke). The data were judged to be sufficiently normally distributed to apply this empirical rule.

3. Results

3.1. Monthly variation in filler blend components

Over the 10-month study period, the levels of 34 blend components were measured for three commercial products and the monitor cigarette Kentucky Reference 3R4F. The mean ± SD levels of these blend components were used to determine the monthly variation (Table 2). Levels of nicotine, nitrate, ammonia and TSNAs in the cigarette filler blends were generally consistent with previously reported levels (Counts et al., 2005).

Variation in the levels of blend components ranged from under 2% CV for total alkaloids and total sugar levels in 3R4F, to over 20% for B[*a*]P, caffeic acid and cadmium in the three commercial products. Overall, however, variation in blend component levels was generally low (<10% CV).

The variation in toxicant levels in 3R4F (which was produced at effectively a single point in time) was compared with that in the commercial products (which were obtained at monthly intervals) to give an indication of month-to-month manufacturing batch variability among the commercial products. Thus, the ratio of the average CV for the three commercial products to the CV for 3R4F was determined for each blend component (Table 2). Across all blend measurements, the CV ratio averaged 1.7, ranging from less than 1 for nicotine and glycerol, to greater than 2 for total and reducing sugars, NNN, chlorogenic acid and cadmium (Table 2). So on average, variation was 70% higher for a repeatedly made product than for a single batch product, and more than twice as high for total and reducing sugars, NNN, chlorogenic acid and cadmium.

Estimates of the within-laboratory 'tolerance' around a single measured value were made for each blend component using 2SD and the associated 2CV to give percentage values. For the commercial products, 2CV values ranged from <10%, for total alkaloids, chloride, total nitrogen, protein nitrogen and nicotine, to ~50% for B[*a*]P, caffeic acid and cadmium. For 3R4F, by contrast, 2CV values ranged from <10% for total alkaloids, total and reducing sugar, chloride, total and protein nitrogen, nitrate, chlorogenic acid and rutin, to a maximum of 34.2% for B[*a*]P.

3.2. Monthly variation in smoke toxicant emission levels

Over the 10-month study period, the emission levels of 44 smoke toxicants were measured for three commercial products and the monitor cigarette Kentucky Reference 3R4F under both the ISO and HCI regimes. The mean ± SD levels of toxicant emissions from these cigarette samples were generally consistent with previously reported levels (Gregg et al., 2004; Counts et al., 2005), and were used to determine the monthly variation (Table 3).

Variation in toxicant emissions ranged from less than 5% CV for TNCO under both ISO and HCI regimes and acetaldehyde under the HCI regime, to >20% for 1,3-butadiene under the HCI regime for all four products, resorcinol under the ISO regime and arsenic under the HCI regime for the commercial products.

Overall, however, variation was generally low with CV values often less than 10%. Under both regimes, TSNAs, phenols, semi-volatiles and HCN analyses tended to give more variable results for each product (CV 10–20%) as well as a few individual analytes including: water determination under ISO; formaldehyde and crotonaldehyde emissions under either regime; several of the aromatic amines under either regime and Cadmium under either regime (Table 3). As mentioned above, 1,3-butadiene under HCI, resorcinol under ISO and arsenic under HCI produced results with the highest variation (CV > 20%).

As above, the CV ratio (average CV for the three commercial products/CV for 3R4F) was calculated to evaluate the month-to-month effect of manufacturing batch for each toxicant (Table 3). The average CV ratio across all smoke toxicants was 1.7 under ISO and 1.5 under HCI, i.e. 70% and 50% more variable for a repeatedly made product. The CV ratio ranged from 0.7 for 1,3-butadiene under HCI and toluene under ISO, to more than 2.0 for: nicotine under ISO (driven by a high CV of 14% for product C); NNK, NNN and NAT under ISO; 2-AN and 4-ABP under HCI; o-cresol, m-cresol and hydroquinone under ISO; phenol, o-cresol and p-cresol under HCI; HCN under ISO; arsenic under HCI, and cadmium under ISO. Butyraldehyde under HCI had a notably high CV ratio (5.8) driven by an atypically low CV for 3R4F (2.6%).

Estimates of a within-laboratory 'tolerance' around a single measured value were made for each smoke toxicant using 2SD and the associated 2CV to give percentage values. For the commercial products, 2CV values ranged from <10% for TNCO analyses under both regimes (although the lower, 4 mg ISO tar product C was more variable under ISO at 13–14%); to greater than 40% for: 1,3-butadiene under HCI; resorcinol under ISO (commercial products only); NNK under both regimes for product A (products B and C, 20–23% and 27–29%, respectively); quinoline under ISO for product C (products A and B, 32% and 36% respectively); and arsenic under HCI for products A and B (product C under HCI and all commercial products under ISO were below LOQ). For 3R4F, by contrast, 2CV values ranged from <10%, for TNCO analyses under both regimes, to a maximum of 57% for 1,3-butadiene under HCI (Table 3).

The variation in toxicant emissions generated under the two regimes was contrasted by calculating the ratio of CV under HCI to CV under ISO (Table 3). These ratios averaged 0.9 over all the smoke toxicants with 72% of results less than 1.0, indicating that results obtained under ISO smoking conditions were generally slightly more variable than results obtained under HCI smoking conditions.

3.3. Monthly variation in toxicant emission levels as a ratio to nicotine

In keeping with TobReg recommendations to establish levels for selected toxicant emissions per mg of nicotine under HCI conditions (WHO, 2008), the variation in toxicant emissions was calculated as a ratio to nicotine for 23 toxicants of current regulatory interest (Table 4), including 9 toxicants proposed for mandated lowering by TobReg, 9 proposed for measurement and reporting by TobReg, and 5 other toxicants on the FDA's initial list that are not on the TobReg list.

When expressed as a ratio to nicotine, the CVs of toxicant emission measurements (Table 4) were similar to the CVs for absolute toxicant emission measurements under HCI conditions (Table 3) with increased variation of the order of −2.9 to 3.4 percentage points in analytes for each product (Table 4). The average difference between the ratio to nicotine CV and the absolute emission level CV was 1.4 percentage points.

Regarding the 9 toxicants proposed for mandated lowering by TobReg, the CV of the ratio to nicotine data was typically 1–2 percentage points higher than the CV determined for absolute measurements. By contrast, the CV as a ratio to nicotine for aromatic amines, phenol and some cadmium emission data tended to be lower than that for the corresponding absolute measurements.

4. Discussion

The present single-centre analysis has measured the monthly variation in key toxicants in both the tobacco blend and smoke of a monitor cigarette (3R4F) and three commercial products sampled over 10 months. In general, the month-to-month

Table 2
Ten-month variation in levels of cigarette filler components.

Analysis	Analyte	Units[c]	Product	Cigarette filler blend components						
				Sample No.	Mean	SD	CV (%)	2SD	2CV (%)	CV ratio[a]
Blend chemistry	Total alkaloids	%dwb	3R4F	12	1.84	0.04	1.9	0.07	3.8	2.0
			A	10	1.86	0.08	4.6	0.17	9.1	
			B	10	1.75	0.06	3.6	0.13	7.3	
			C	10	1.93	0.06	3.1	0.12	6.2	
	Total sugar	%dwb	3R4F	12	11.8	0.2	1.9	0.4	3.7	3.1
			A	10	10.7	0.6	5.9	1.3	11.8	
			B	10	13.7	0.6	4.7	1.3	9.4	
			C	10	10.3	0.7	7.1	1.5	14.1	
	Reducing sugars	%dwb	3R4F	12	10.2	0.3	2.6	0.5	5.2	2.6
			A	10	7.9	0.6	7.7	1.2	15.4	
			B	10	8.6	0.5	5.3	0.9	10.6	
			C	10	7.6	0.6	7.7	1.2	15.4	
	B[*a*]P[b]	ng/g dwb	3R4F	11	10.0	1.7	17.1	3.43	34.2	1.5
			A	10	22.2	5.8	26.1	11.55	52.1	
			B	10	19.9	5.3	26.5	10.56	53.1	
			C	10	36.7	8.3	22.5	16.53	45.0	
	Chloride	%dwb	3R4F	12	0.89	0.03	3.3	0.06	6.6	1.4
			A	10	0.84	0.06	6.7	0.11	13.3	
			B	10	0.75	0.03	3.6	0.05	7.3	
			C	10	0.85	0.03	3.3	0.06	6.7	
Nitrogenous compounds	Total nitrogen	%dwb	3R4F	12	2.51	0.05	2.1	0.10	4.2	1.7
			A	10	2.85	0.11	4.0	0.23	7.9	
			B	10	2.62	0.09	3.2	0.17	6.5	
			C	10	2.89	0.09	3.2	0.18	6.3	
	Protein nitrogen	%dwb	3R4F	11	1.21	0.03	2.7	0.07	5.5	1.7
			A	10	1.40	0.06	4.4	0.12	8.9	
			B	10	1.29	0.06	4.9	0.13	9.9	
			C	10	1.38	0.06	4.2	0.12	8.4	
	Ammonium[b]	µg/g dwb	3R4F	12	1055	56	5.3	112	10.7	1.5
			A	10	1354	136	10.1	273	20.2	
			B	10	1239	62	5.0	125	10.1	
			C	10	1483	124	8.4	248	16.7	
	Nitrate ion	%dwb	3R4F	12	1.06	0.03	3.2	0.07	6.5	1.9
			A	10	0.73	0.06	8.7	0.13	17.5	
			B	10	0.74	0.03	3.9	0.06	7.7	
			C	10	0.74	0.05	6.1	0.09	12.3	
	Nitrite ion	µg/g dwb	3R4F	12	<1.00					
			A	10	<1.00					
			B	10	<1.00					
			C	10	<1.00					
Individual alkaloids	Nicotine[b]	%dwb	3R4F	12	2.25	0.12	5.1	0.23	10.3	0.8
			A	10	2.23	0.08	3.4	0.15	6.9	
			B	10	2.13	0.09	4.1	0.17	8.1	
			C	10	2.31	0.10	4.2	0.19	8.3	
	Nornicotine	%dwb	3R4F	12	0.091	0.010	10.6	0.019	21.2	1.2
			A	10	0.155	0.023	14.5	0.045	29.1	
			B	10	0.109	0.013	12.1	0.026	24.1	
			C	10	0.157	0.020	12.6	0.040	25.3	
	Anabasine	%dwb	3R4F	12	0.010					
			A	10	0.017	0.003	16.6	0.006	33.1	
			B	10	0.016	0.002	13.8	0.004	27.6	
			C	10	0.017	0.003	15.7	0.005	31.5	
	Anatabine	%dwb	3R4F	12	0.092	0.006	6.6	0.012	13.2	1.1
			A	10	0.109	0.008	7.7	0.017	15.3	
			B	10	0.101	0.006	5.6	0.011	11.1	
			C	10	0.114	0.010	9.1	0.021	18.3	
	Myosmine	%dwb	3R4F	12	<0.010					
			A	10	<0.010					
			B	10	<0.010					
			C	10	<0.010					
Polyphenols	Chlorogenic acid	mg/g dwb	3R4F	12	3.36	0.07	2.2	0.15	4.4	3.3
			A	10	3.33	0.21	6.3	0.42	12.6	
			B	10	3.98	0.24	6.1	0.49	12.2	
			C	10	3.51	0.32	9.2	0.65	18.4	
	Caffeic acid	mg/g dwb	3R4F	12	0.10	0.01	13.9	0.03	27.9	2.0
			A	10	0.16	0.04	26.9	0.09	53.8	
			B	10	0.12	0.04	35.1	0.08	70.1	
			C	10	0.17	0.04	23.4	0.08	46.7	
	Rutin	mg/g dwb	3R4F	12	2.29	0.07	3.1	0.14	6.1	1.9
			A	10	3.50	0.20	5.6	0.40	11.3	
			B	10	3.42	0.17	4.9	0.33	9.8	

(*continued on next page*)

Table 2 (*continued*)

Analysis	Analyte	Units[c]	Product	Cigarette filler blend components						
				Sample No.	Mean	SD	CV (%)	2SD	2CV (%)	CV ratio[a]
			C	10	3.55	0.24	6.9	0.49	13.8	
	Scopoletin	mg/g dwb	3R4F	12	0.28	0.04	14.8	0.08	29.6	1.2
			A	10	0.31	0.06	19.6	0.12	39.1	
			B	10	0.23	0.03	11.7	0.05	23.5	
			C	10	0.33	0.07	20.2	0.13	40.3	
Tobacco specific nitrosamines	NNN[b]	μg/g dwb	3R4F	10	2.40	0.15	6.4	0.31	12.9	2.2
			A	10	2.08	0.38	18.4	0.76	36.7	
			B	10	1.04	0.09	8.5	0.18	16.9	
			C	10	1.77	0.26	14.8	0.53	29.6	
	NAB	μg/g dwb	3R4F	10	0.13	0.02	15.2	0.04	30.4	1.0
			A	10	0.11	0.01	14.0	0.03	28.1	
			B	10	0.09	0.02	23.1	0.04	46.3	
			C	10	0.10	0.01	7.7	0.02	15.4	
	NAT	μg/g dwb	3R4F	10	2.00	0.12	6.1	0.24	12.2	1.7
			A	10	1.14	0.17	15.2	0.34	30.3	
			B	10	0.98	0.08	7.7	0.15	15.4	
			C	10	0.96	0.07	7.6	0.15	15.1	
	NNK[b]	μg/g dwb	3R4F	10	0.71	0.06	7.8	0.11	15.7	1.5
			A	10	0.30	0.04	12.9	0.08	25.8	
			B	10	0.34	0.04	13.3	0.09	26.6	
			C	10	0.28	0.03	10.0	0.06	20.0	
	Total TSNA	μg/g dwb	3R4F	10	5.24	0.34	6.4	0.67	12.8	1.8
			A	10	3.63	0.58	15.9	1.15	31.7	
			B	10	2.45	0.19	7.8	0.38	15.5	
			C	10	3.11	0.31	10.0	0.62	20.1	
Metals	Lead	μg/g dwb	3R4F	12	0.425	0.033	7.8	0.066	15.6	1.8
			A	10	0.522	0.088	16.8	0.175	33.6	
			B	10	0.472	0.064	13.6	0.128	27.1	
			C	10	0.620	0.076	12.3	0.152	24.5	
	Chromium	μg/g dwb	3R4F	12	0.831	0.092	11.1	0.185	22.3	1.4
			A	10	1.306	0.147	11.2	0.293	22.5	
			B	10	1.211	0.259	21.4	0.518	42.8	
			C	10	0.960	0.139	14.5	0.278	29.0	
	Nickel	μg/g dwb	3R4F	12	1.477	0.095	6.4	0.189	12.8	1.5
			A	10	2.046	0.193	9.5	0.387	18.9	
			B	10	1.959	0.244	12.5	0.488	24.9	
			C	10	1.579	0.113	7.2	0.227	14.4	
	Cadmium[b]	μg/g dwb	3R4F	12	0.988	0.098	9.9	0.196	19.9	2.3
			A	10	0.789	0.174	22.0	0.347	44.0	
			B	10	0.712	0.158	22.2	0.317	44.5	
			C	10	0.971	0.242	24.9	0.483	49.8	
	Mercury	μg/g dwb	3R4F	12	<0.031					
			A	10	<0.031					
			B	10	<0.031					
			C	10	<0.031					
	Arsenic[b]	μg/g dwb	3R4F	12	0.251	0.023	9.2	0.046	18.3	1.3
			A	10	0.211	0.029	13.6	0.057	27.2	
			B	10	0.207	0.021	10.3	0.042	20.5	
			C	10	0.181	0.024	13.2	0.048	26.4	
	Selenium	μg/g dwb	3R4F	12	<0.130					
			A	10	<0.130					
			B	10	<0.130					
			C	10	<0.130					
Humectants	Glycerol[b]	mg/g dwb	3R4F	12	23.44	2.04	8.7	4.08	17.4	0.7
			A	10	21.12	1.56	7.4	3.12	14.8	
			B	10	17.04	0.72	4.2	1.44	8.5	
			C	10	19.27	1.49	7.7	2.98	15.5	
	Propylene glycol[b]	mg/g dwb	3R4F	12	<0.75					
			A	10	12.63	0.80	6.3	1.59	12.6	
			B	10	12.06	0.42	3.5	0.84	7.0	
			C	10	9.62	0.50	5.2	1.00	10.4	
	Triethylene glycol[b]	mg/g dwb	3R4F	12	<0.75					
			A	10	<0.75					
			B	10	<0.75					
			C	10	<0.75					
	Triacetin	mg/g dwb	3R4F	12	<0.75					
			A	10	0.87	0.01	0.7	0.01	1.3	
			B	10	<0.75					
			C	10	<0.75					

[a] CV ratio = mean CV commercial product/CV 3R4F.
[b] Blend filler components of regulatory relevance (Table 1).
[c] dwb: dry weight basis.

Table 3
Ten-month variation in emission levels of mainstream smoke toxicants from four products smoked under ISO and HCI conditions.

Analysis	Analyte	Units	Product	ISO smoking regime							HCI smoking regime							HCI CV/ISO CV
				No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	
Tar, nicotine and CO	Cigarette weight	mg/cig	3R4F	10	1029	3	0.3	6	0.6	2.3	7	1027	5	0.4	9	0.9	1.6	
			A	10	912	8	0.9	17	1.8		10	908	6	0.7	13	1.4		
			B	10	886	6	0.7	13	1.4		10	884	6	0.7	13	1.5		
			C	10	835	4	0.5	8	1.0		10	832	6	0.8	13	1.6		
	Puff No.		3R4F	10	8.2	0.1	1.5	0.2	2.9	1.6	7	10.4	0.2	1.7	0.3	3.4	1.3	
			A	10	8.3	0.1	1.2	0.2	2.3		10	10.7	0.2	1.5	0.3	3.0		
			B	10	7.8	0.2	2.7	0.4	5.4		10	10.0	0.3	3.4	0.7	6.8		
			C	10	7.5	0.2	2.9	0.4	5.8		10	8.7	0.1	1.7	0.3	3.4		
	TPM	mg/cig	3R4F	10	9.5	0.4	3.7	0.7	7.4	1.3	7	44.1	1.4	3.2	2.8	6.4	0.9	
			A	10	13.2	0.5	4.1	1.1	8.3		10	51.8	0.9	1.7	1.8	3.4		
			B	10	11.9	0.4	3.5	0.8	7.0		10	48.2	1.8	3.7	3.6	7.5		
			C	10	4.9	0.3	6.4	0.6	12.8		10	35.3	1.2	3.3	2.3	6.6		
	Tar	mg/cig	3R4F	10	7.9	0.3	3.9	0.6	7.7	1.2	7	26.3	0.7	2.6	1.4	5.3	0.9	
			A	10	10.8	0.4	3.9	0.8	7.9		10	31.4	0.5	1.7	1.1	3.4		
			B	10	9.7	0.3	2.6	0.5	5.2		10	29.0	0.9	3.2	1.8	6.3		
			C	10	4.1	0.3	6.8	0.6	13.6		10	20.8	0.5	2.6	1.1	5.2		
	Nicotine[b]	mg/cig	3R4F	10	0.73	0.02	2.3	0.03	4.6	2.2	7	1.97	0.04	2.3	0.09	4.6	1.6	1.0
			A	10	0.95	0.04	4.6	0.09	9.2		10	2.33	0.08	3.2	0.15	6.4		0.7
			B	10	0.85	0.03	3.0	0.05	6.1		10	2.13	0.08	3.7	0.16	7.5		1.2
			C	10	0.43	0.03	7.2	0.06	14.4		10	1.56	0.07	4.2	0.13	8.5		0.6
	Water	mg/cig	3R4F	10	0.9	0.1	11.7	0.2	23.3	1.0	7	15.9	0.7	4.7	1.5	9.3	1.0	0.4
			A	10	1.5	0.2	15.9	0.5	31.7		10	18.0	0.6	3.5	1.3	7.1		0.2
			B	10	1.3	0.2	11.7	0.3	23.5		10	17.1	1.0	5.7	1.9	11.4		0.5
			C	10	0.3	0.0	6.6	0.0	13.2		10	12.9	0.7	5.3	1.4	10.7		0.8
	CO[b]	mg/cig	3R4F	10	10.7	0.4	4.0	0.9	8.1	1.2	7	31.2	1.0	3.1	1.9	6.2	0.9	0.8
			A	10	9.6	0.4	4.1	0.8	8.2		10	26.8	0.7	2.7	1.5	5.4		0.7
			B	10	8.9	0.3	3.7	0.7	7.4		10	26.1	0.8	3.1	1.6	6.1		0.8
			C	10	5.0	0.3	6.6	0.7	13.1		10	23.0	0.6	2.8	1.3	5.6		0.4
TSNA	Cigarette weight	mg/cig	3R4F	8	1037	3	0.3	6	0.6	3.1	9	1079	9	0.9	18	1.7	1.2	
			A	10	923	8	0.9	16	1.8		10	961	10	1.0	20	2.0		
			B	10	898	7	0.7	13	1.5		10	936	9	1.0	18	2.0		
			C	10	847	9	1.1	18	2.2		10	886	11	1.2	21	2.4		
	Puff No.		3R4F	8	7.9	0.2	2.1	0.3	4.1	1.4	9	10.6	0.2	1.4	0.3	2.9	1.6	
			A	10	7.9	0.2	2.9	0.5	5.8		10	10.9	0.3	2.8	0.6	5.5		
			B	10	7.5	0.2	3.1	0.5	6.1		10	10.2	0.2	2.0	0.4	4.1		
			C	10	7.2	0.2	2.6	0.4	5.3		10	8.9	0.2	2.2	0.4	4.4		
	TPM	mg/cig	3R4F	8	9.9	0.4	4.3	0.9	8.7	1.5	9	44.1	1.3	2.9	2.5	5.8	1.0	
			A	10	13.4	0.9	6.8	1.8	13.6		10	50.9	1.6	3.1	3.2	6.2		
			B	10	12.2	0.7	5.5	1.3	11.1		10	49.8	0.9	1.9	1.8	3.7		
			C	10	5.6	0.4	7.8	0.9	15.6		10	36.6	1.5	4.0	2.9	8.0		
	NNK[b]	ng/cig	3R4F	8	103.6	6.8	6.6	13.6	13.1	2.4	9	309.6	27.6	8.9	55.2	17.8	1.7	1.4
			A	10	50.5	11.1	21.9	22.1	43.8		10	131.0	27.2	20.8	54.4	41.5		0.9
			B	10	56.5	8.1	14.3	16.2	28.7		10	151.3	20.3	13.4	40.6	26.8		0.9
			C	10	21.4	2.4	11.3	4.8	22.6		10	78.1	7.8	10.0	15.5	19.9		0.9
	NNN[b]	ng/cig	3R4F	8	113.5	5.3	4.6	10.5	9.3	3.2	9	311.6	20.6	6.6	41.1	13.2	2.0	1.4
			A	10	138.7	26.7	19.2	53.3	38.5		10	329.1	55.8	17.0	111.6	33.9		0.9
			B	10	67.2	7.6	11.3	15.2	22.7		10	163.5	13.6	8.3	27.3	16.7		0.7
			C	10	60.4	8.1	13.4	16.2	26.8		10	215.1	30.8	14.3	61.6	28.6		1.1
	NAB	ng/cig	3R4F	8	14.1	0.8	5.4	1.5	10.8	1.9	9	37.0	3.0	8.1	6.0	16.2	1.2	1.5
			A	10	12.7	1.4	10.8	2.7	21.7		10	29.8	1.7	5.8	3.5	11.6		0.5
			B	10	9.9	0.8	8.1	1.6	16.2		10	23.1	2.4	10.5	4.9	21.0		1.3
			C	10	6.0	0.7	11.6	1.4	23.1		10	20.5	2.8	13.9	5.7	27.7		1.2

(*continued on next page*)

Table 3 (*continued*)

Analysis	Analyte	Units	Product	ISO smoking regime							HCI smoking regime							HCI CV/ISO CV
				No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	
	NAT	ng/cig	3R4F	8	125.4	5.4	4.3	10.8	8.6	2.8	9	323.0	22.9	7.1	45.9	14.2	1.1	1.7
			A	10	99.9	13.5	13.5	27.1	27.1		10	228.8	22.3	9.7	44.6	19.5		0.7
			B	10	85.0	9.1	10.8	18.3	21.5		10	198.2	16.2	8.2	32.5	16.4		0.8
			C	10	47.3	5.5	11.6	11.0	23.2		10	156.8	9.0	5.8	18.1	11.5		0.5
Carbonyls	Cigarette weight	mg/cig	3R4F	9	1041	8	0.8	16	1.6	1.1	10	1081	11	1.0	22	2.0	1.4	
			A	10	924	8	0.9	17	1.8		10	963	20	2.0	39	4.1		
			B	10	899	11	1.2	21	2.3		10	943	12	1.3	24	2.5		
			C	10	842	4	0.4	8	0.9		10	886	8	0.9	16	1.8		
	Puff No.		3R4F	9	8.7	0.1	1.3	0.2	2.6	2.0	10	10.6	0.3	3.0	0.6	5.9	1.0	
			A	10	8.6	0.2	2.5	0.4	4.9		10	11.0	0.3	2.5	0.5	5.0		
			B	10	8.2	0.2	2.5	0.4	5.1		10	10.2	0.2	2.4	0.5	4.8		
			C	10	7.7	0.2	2.7	0.4	5.5		10	9.0	0.3	3.9	0.7	7.7		
	Formaldehyde[b]	µg/cig	3R4F	9	25.0	1.6	6.3	3.2	12.7	1.7	10	72.2	5.8	8.0	11.5	16.0	1.5	1.3
			A	10	25.0	2.5	10.2	5.1	20.3		10	65.6	6.4	9.7	12.8	19.5		1.0
			B	10	34.3	4.3	12.4	8.5	24.8		10	92.7	11.0	11.9	22.0	23.8		1.0
			C	10	9.0	0.9	10.0	1.8	20.0		10	46.8	7.2	15.4	14.4	30.8		1.5
	Acetaldehyde[b]	µg/cig	3R4F	9	469	20	4.3	40	8.5	1.6	10	1235	39	3.1	77	6.3	1.6	0.7
			A	10	428	32	7.4	64	14.9		10	1090	49	4.5	97	8.9		0.6
			B	10	416	27	6.5	54	12.9		10	1103	39	3.5	78	7.1		0.5
			C	10	243	15	6.4	31	12.8		10	973	71	7.3	143	14.7		1.2
	Acetone	µg/cig	3R4F	9	199	7	3.7	15	7.4	2.0	10	576	33	5.8	67	11.6	1.0	1.6
			A	10	186	13	7.1	26	14.1		10	514	29	5.6	58	11.3		0.8
			B	10	177	12	7.0	25	14.0		10	508	20	4.0	40	8.0		0.6
			C	10	103	8	7.6	16	15.2		10	447	38	8.6	77	17.2		1.1
	Acrolein[b]	µg/cig	3R4F	9	52.2	3.2	6.1	6.3	12.1	1.2	10	124.9	8.5	6.8	17.0	13.6	1.0	1.1
			A	10	48.5	3.8	7.9	7.7	15.9		10	113.7	8.1	7.1	16.2	14.3		0.9
			B	10	48.5	4.0	8.2	8.0	16.5		10	120.7	6.1	5.0	12.2	10.1		0.6
			C	10	24.9	1.4	5.8	2.9	11.6		10	100.0	8.5	8.5	16.9	16.9		1.5
	Propionaldehyde	µg/cig	3R4F	9	45.0	2.5	5.6	5.1	11.3	1.3	10	129.7	6.0	4.6	11.9	9.2	1.4	0.8
			A	10	44.9	3.5	7.8	7.0	15.6		10	127.2	8.3	6.6	16.7	13.1		0.8
			B	10	43.7	3.3	7.6	6.6	15.1		10	129.8	7.0	5.4	14.0	10.8		0.7
			C	10	24.2	1.5	6.3	3.0	12.5		10	105.2	8.3	7.9	16.6	15.8		1.3
	MEK	µg/cig	3R4F	9	52.4	3.5	6.7	7.1	13.5	1.2	10	158.7	8.0	5.0	16.0	10.1	1.2	0.7
			A	10	48.3	3.3	6.9	6.6	13.7		10	140.2	8.5	6.1	17.0	12.2		0.9
			B	10	47.2	4.0	8.6	8.1	17.1		10	138.7	5.2	3.8	10.4	7.5		0.4
			C	10	26.0	2.3	8.8	4.6	17.6		10	119.6	10.7	9.0	21.5	17.9		1.0
	Crotonaldehyde[b]	µg/cig	3R4F	9	10.7	0.9	8.1	1.7	16.2	1.5	10	46.4	3.6	7.8	7.3	15.6	1.1	1.0
			A	10	12.1	1.5	12.0	2.9	24.0		10	46.4	3.6	7.7	7.1	15.4		0.6
			B	10	12.0	1.4	11.4	2.7	22.9		10	47.1	3.5	7.5	7.0	14.9		0.7
			C	10	4.8	0.7	13.9	1.3	27.8		10	37.9	4.3	11.2	8.5	22.5		0.8
	Butyraldehyde	µg/cig	3R4F	9	33.6	1.9	5.7	3.8	11.3	1.6	10	86.8	1.1	1.3	2.2	2.6	5.8	0.2
			A	10	37.6	3.7	9.8	7.4	19.6		10	106.2	7.2	6.7	14.3	13.5		0.7
			B	10	34.2	3.1	9.2	6.3	18.4		10	99.5	7.7	7.8	15.5	15.6		0.8
			C	10	20.5	1.6	8.0	3.3	15.9		10	90.4	7.2	8.0	14.5	16.0		1.0
Miscellaneous organic compounds	Cigarette weight	mg/cig	3R4F	9	1038	4	0.4	7	0.7	2.0	8	1053	16	1.5	32	3.0	0.7	
			A	10	919	8	0.9	17	1.8		10	959	10	1.0	19	2.0		
			B	10	893	6	0.7	13	1.4		10	934	7	0.8	15	1.6		
			C	10	839	4	0.5	9	1.1		10	887	11	1.2	21	2.4		
	Puff No.		3R4F	9	8.4	0.1	1.1	0.2	2.3	1.7	8	10.5	0.5	4.5	1.0	9.1	0.7	
			A	10	8.3	0.2	2.7	0.5	5.5		10	10.8	0.3	2.9	0.6	5.7		
			B	10	8.0	0.1	1.6	0.3	3.2		10	10.0	0.3	3.2	0.6	6.3		
			C	10	7.6	0.1	1.4	0.2	2.8		10	8.9	0.3	3.9	0.7	7.8		

	TPM	mg/cig	3R4F	9	10.1	0.5	4.6	0.9	9.2	1.0	8	43.6	1.2	2.8	2.4	5.6	1.6	
			A	10	13.7	0.5	3.9	1.1	7.8		10	51.8	2.1	4.1	4.2	8.2		
			B	10	12.4	0.7	5.7	1.4	11.3		10	48.3	1.9	3.9	3.8	7.9		
			C	10	5.4	0.2	4.0	0.4	7.9		10	34.9	1.9	5.4	3.8	10.8		
	1,3-Butadiene[b]	µg/cig	3R4F	9	30.5	2.6	8.7	5.3	17.3	0.8	8	90.5	25.8	28.6	51.7	57.1	0.7	3.3
			A	10	35.1	2.1	6.1	4.3	12.2		10	90.3	19.0	21.0	38.0	42.0		3.5
			B	10	30.6	2.8	9.3	5.7	18.5		10	80.9	17.3	21.4	34.6	42.8		2.3
			C	10	20.2	1.3	6.4	2.6	12.8		10	80.9	16.4	20.3	32.8	40.6		3.2
	Isoprene[b]	µg/cig	3R4F	9	347	22	6.3	43	12.5	1.1	8	1042	76	7.3	151	14.5	0.9	1.2
			A	10	373	28	7.5	56	15.0		10	1013	70	6.9	140	13.8		0.9
			B	10	329	29	8.9	59	17.9		10	922	52	5.6	104	11.2		0.6
			C	10	207	9	4.2	17	8.3		10	870	59	6.8	118	13.6		1.6
	Acrylonitrile[b]	µg/cig	3R4F	9	10.7	0.5	4.7	1.0	9.3	2.0	8	33.7	1.5	4.4	3.0	8.9	1.7	0.9
			A	10	12.6	0.8	6.5	1.6	13.0		10	37.4	2.3	6.1	4.6	12.2		0.9
			B	10	11.0	1.0	8.7	1.9	17.4		10	32.4	2.2	6.8	4.4	13.6		0.8
			C	10	6.1	0.8	12.9	1.6	25.7		10	32.8	3.2	9.7	6.4	19.4		0.8
	Benzene[b]	µg/cig	3R4F	9	43.4	1.6	3.7	3.2	7.3	1.8	8	99.2	5.6	5.6	11.2	11.3	1.1	1.5
			A	10	45.5	2.2	4.8	4.4	9.6		10	98.5	5.3	5.3	10.5	10.7		1.1
			B	10	40.1	2.8	6.9	5.5	13.7		10	86.5	4.5	5.2	9.0	10.5		0.8
			C	10	27.9	2.2	7.9	4.4	15.8		10	83.5	6.5	7.7	12.9	15.5		1.0
	Toluene[b]	µg/cig	3R4F	9	64.5	6.1	9.4	12.1	18.8	0.7	8	164.0	8.4	5.1	16.9	10.3	1.0	0.5
			A	10	70.1	5.9	8.5	11.9	16.9		10	166.1	9.5	5.7	18.9	11.4		0.7
			B	10	61.1	3.1	5.1	6.2	10.1		10	142.2	4.7	3.3	9.4	6.6		0.6
			C	10	38.0	1.9	5.0	3.8	9.9		10	129.5	8.0	6.2	16.1	12.4		1.2
Aromatic amines	Cigarette weight	mg/cig	3R4F	9	1037	4	0.4	8	0.8	2.3	7	1075	14	1.3	28	2.6	0.9	
			A	10	921	8	0.9	17	1.8		10	960	12	1.2	23	2.4		
			B	10	896	7	0.7	13	1.5		10	936	9	0.9	18	1.9		
			C	10	839	9	1.0	18	2.1		10	884	11	1.2	21	2.4		
	Puff No.		3R4F	9	7.9	0.1	0.8	0.1	1.5	2.7	7	10.7	0.2	1.6	0.3	3.1	1.5	
			A	10	7.9	0.1	1.5	0.2	3.1		10	10.8	0.3	2.6	0.6	5.2		
			B	10	7.5	0.2	2.2	0.3	4.5		10	10.2	0.2	1.6	0.3	3.2		
			C	10	7.1	0.2	2.4	0.3	4.8		10	8.8	0.2	2.7	0.5	5.3		
	TPM	mg/cig	3R4F	9	10.3	0.5	4.7	1.0	9.5	1.0	7	45.9	2.3	5.1	4.6	10.1	0.9	
			A	10	13.5	0.7	4.9	1.3	9.7		10	52.4	2.0	3.9	4.1	7.7		
			B	10	12.2	0.5	4.1	1.0	8.2		10	50.4	2.2	4.3	4.3	8.6		
			C	10	5.6	0.3	5.6	0.6	11.2		10	36.6	2.1	5.8	4.3	11.6		
	1-AN[b]	ng/cig	3R4F	9	13.6	1.3	9.7	2.6	19.3	0.9	7	22.9	1.5	6.3	2.9	12.7	1.4	0.7
			A	10	17.5	1.7	9.7	3.4	19.5		10	29.5	2.3	7.8	4.6	15.6		0.8
			B	10	15.5	1.2	7.8	2.4	15.5		10	26.5	3.0	11.3	6.0	22.7		1.5
			C	10	8.3	0.8	10.0	1.6	19.9		10	18.8	1.5	7.9	3.0	15.9		0.8
	2-AN[b]	ng/cig	3R4F	9	7.7	0.5	6.3	1.0	12.5	1.1	7	13.4	0.5	3.7	1.0	7.3	2.1	0.6
			A	10	9.9	0.9	8.6	1.7	17.2		10	16.6	1.1	6.8	2.3	13.6		0.8
			B	10	8.5	0.4	4.8	0.8	9.7		10	14.7	1.5	10.1	3.0	20.3		2.1
			C	10	4.7	0.4	7.9	0.8	15.8		10	10.9	0.6	5.8	1.3	11.5		0.7
	3-ABP	ng/cig	3R4F	9	1.93	0.18	9.5	0.37	19.1	1.3	7	4.51	0.47	10.3	0.93	20.7	1.3	1.1
			A	10	2.56	0.36	14.0	0.72	28.0		10	5.62	0.69	12.3	1.39	24.7		0.9
			B	10	2.14	0.22	10.4	0.45	20.9		10	4.82	0.73	15.1	1.46	30.2		1.4
			C	10	1.27	0.17	13.2	0.34	26.5		10	3.83	0.48	12.6	0.96	25.1		1.0
	4-ABP[b]	ng/cig	3R4F	9	1.15	0.07	6.4	0.15	12.8	0.9	7	2.66	0.11	4.3	0.23	8.6	2.2	0.7
			A	10	1.61	0.11	6.5	0.21	13.1		10	3.64	0.30	8.2	0.60	16.4		1.3
			B	10	1.31	0.06	4.4	0.11	8.7		10	3.04	0.33	10.9	0.66	21.8		2.5
			C	10	0.83	0.05	6.1	0.10	12.2		10	2.60	0.23	8.9	0.46	17.7		1.4
Phenols	Cigarette weight	mg/cig	3R4F	9	1036	6	0.6	12	1.1	1.8	8	1042	9	0.9	18	1.7	1.8	
			A	10	916	11	1.2	22	2.4		10	962	11	1.2	23	2.4		
			B	10	895	7	0.7	13	1.5		10	936	16	1.7	32	3.4		
			C	10	845	9	1.1	18	2.1		10	883	15	1.7	30	3.4		
	Puff No.		3R4F	9	8.4	0.1	1.7	0.3	3.5	1.7	8	11.0	0.5	4.7	1.0	9.4	0.7	
			A	10	8.3	0.2	2.5	0.4	5.0		10	10.8	0.3	2.8	0.6	5.7		

(*continued on next page*)

Table 3 (*continued*)

Analysis	Analyte	Units	Product	ISO smoking regime							HCI smoking regime							HCI CV/ISO CV
				No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	
			B	10	8.0	0.2	3.1	0.5	6.2		10	10.3	0.5	4.7	1.0	9.3		
			C	10	7.7	0.3	3.5	0.5	7.0		10	9.0	0.3	3.1	0.6	6.1		
	TPM	mg/cig	3R4F	9	9.5	0.8	7.9	1.5	15.8	2.5	8	42.3	3.7	8.8	7.5	17.7	0.7	
			A	10	13.0	2.5	19.3	5.0	38.5		10	49.5	2.8	5.7	5.7	11.4		
			B	10	11.6	1.0	9.0	2.1	18.0		10	48.9	3.8	7.7	7.5	15.4		
			C	10	5.6	1.8	31.4	3.5	62.8		10	36.1	1.6	4.5	3.2	9.0		
	Phenol	µg/cig	3R4F	9	7.6	0.6	7.9	1.2	15.8	1.9	8	14.7	0.6	4.1	1.2	8.2	2.7	0.5
			A	10	14.0	2.4	17.4	4.9	34.8		10	28.5	3.3	11.6	6.6	23.2		0.7
			B	10	13.1	1.8	13.5	3.6	27.1		10	27.7	3.3	11.7	6.5	23.5		0.9
			C	10	3.8	0.5	13.8	1.1	27.7		10	12.6	1.3	10.1	2.5	20.1		0.7
	o-Cresol	µg/cig	3R4F	9	2.49	0.18	7.2	0.36	14.4	2.2	8	4.71	0.16	3.4	0.32	6.8	2.9	0.5
			A	10	3.71	0.71	19.2	1.43	38.5		10	7.50	0.78	10.4	1.56	20.8		0.5
			B	10	3.73	0.58	15.5	1.15	31.0		10	7.64	0.67	8.8	1.34	17.5		0.6
			C	10	1.18	0.15	12.7	0.30	25.4		10	3.34	0.35	10.4	0.69	20.8		0.8
	m-Cresol	µg/cig	3R4F	9	2.09	0.11	5.0	0.21	10.0	3.5	8	3.92	0.30	7.7	0.60	15.4	1.9	1.5
			A	10	2.99	0.58	19.4	1.16	38.7		10	6.15	0.96	15.6	1.91	31.1		0.8
			B	10	2.93	0.51	17.4	1.02	34.9		10	6.20	0.91	14.7	1.82	29.4		0.8
			C	10	1.02	0.16	15.8	0.32	31.6		10	2.89	0.40	13.9	0.80	27.8		0.9
	p-Cresol	µg/cig	3R4F	9	4.69	0.45	9.6	0.90	19.3	1.8	8	9.40	0.62	6.6	1.23	13.1	2.1	0.7
			A	10	7.57	1.42	18.8	2.84	37.6		10	16.07	2.33	14.5	4.65	29.0		0.8
			B	10	6.85	1.20	17.4	2.39	34.9		10	14.97	1.98	13.2	3.97	26.5		0.8
			C	10	2.41	0.38	15.8	0.76	31.6		10	7.50	0.98	13.1	1.96	26.2		0.8
	Catechol[b]	µg/cig	3R4F	9	38.9	4.3	11.1	8.6	22.1	1.0	8	97.7	5.4	5.6	10.9	11.2	1.8	0.5
			A	10	54.8	8.7	15.9	17.4	31.7		10	137.0	15.3	11.2	30.6	22.3		0.7
			B	10	50.1	4.3	8.6	8.6	17.3		10	124.7	11.6	9.3	23.3	18.7		1.1
			C	10	25.0	2.6	10.2	5.1	20.5		10	86.3	8.4	9.7	16.8	19.4		0.9
	Resorcinol	µg/cig	3R4F	9	0.78	0.11	14.0	0.22	28.0	1.8	8	2.25	0.37	16.3	0.73	32.6	1.0	1.2
			A	10	1.39	0.34	24.4	0.68	48.9		10	3.84	0.63	16.4	1.26	32.8		0.7
			B	10	1.20	0.31	25.5	0.61	51.0		10	3.45	0.61	17.8	1.23	35.6		0.7
			C	10	0.51	0.13	26.1	0.27	52.3		10	2.31	0.36	15.5	0.72	31.0		0.6
	Hydroquinone[b]	µg/cig	3R4F	9	31.1	1.9	6.3	3.9	12.5	2.1	8	86.7	4.9	5.6	9.8	11.3	1.7	0.9
			A	10	49.2	8.9	18.0	17.7	36.1		10	125.8	12.3	9.8	24.7	19.6		0.5
			B	10	46.3	4.0	8.7	8.0	17.3		10	122.1	13.0	10.6	26.0	21.3		1.2
			C	10	21.6	2.6	12.2	5.3	24.3		10	85.2	6.6	7.7	13.2	15.4		0.6
B[*a*]P	Cigarette weight	mg/cig	3R4F	9	1037	4	0.4	9	0.8	2.1	7	1075	14	1.3	28	2.6	0.9	
			A	10	921	8	0.9	17	1.8		10	960	12	1.2	23	2.4		
			B	10	896	7	0.7	13	1.5		10	936	10	1.1	20	2.1		
			C	10	839	9	1.0	18	2.1		10	884	12	1.3	23	2.6		
	Puff No.		3R4F	9	7.9	0.0	0.6	0.1	1.2	3.4	7	10.7	0.2	1.7	0.4	3.4	1.4	
			A	10	7.9	0.1	1.5	0.2	3.1		10	10.8	0.3	2.6	0.6	5.2		
			B	10	7.5	0.2	2.2	0.3	4.5		10	10.2	0.2	1.9	0.4	3.8		
			C	10	7.1	0.2	2.4	0.3	4.8		10	8.8	0.3	2.8	0.5	5.7		
	TPM	mg/cig	3R4F	9	10.3	0.5	4.7	1.0	9.5	1.0	7	45.7	2.2	4.9	4.4	9.7	1.0	
			A	10	13.5	0.7	4.9	1.3	9.7		10	52.4	2.0	3.9	4.1	7.7		
			B	10	12.2	0.5	4.1	1.0	8.2		10	50.4	2.2	4.3	4.3	8.6		
			C	10	5.6	0.3	5.6	0.6	11.2		10	36.4	2.4	6.7	4.9	13.4		
	B[*a*]P[b]	ng/cig	3R4F	9	6.22	0.29	4.6	0.57	9.2	1.5	7	13.59	0.69	5.1	1.38	10.1	1.0	1.1
			A	10	8.03	0.62	7.8	1.25	15.5		10	17.27	1.02	5.9	2.05	11.8		0.8
			B	10	7.00	0.45	6.4	0.90	12.9		10	15.73	0.81	5.2	1.62	10.3		0.8
			C	10	4.43	0.30	6.8	0.61	13.7		10	13.04	0.61	4.7	1.23	9.4		0.7
HCN	Cigarette weight	mg/cig	3R4F	9	1036	5	0.5	10	0.9	2.7	10	1073	7	0.6	14	1.3	2.2	
			A	10	919	11	1.2	21	2.3		10	963	18	1.9	37	3.8		

			B	10	891	14	1.6	28	3.1		10	936	8	0.8	16	1.7		
			C	10	844	8	1.0	17	2.0		10	888	13	1.5	27	3.0		
	Puff No.		3R4F	9	8.8	0.1	1.4	0.2	2.8	2.0	10	10.7	0.3	2.4	0.5	4.7	1.1	
			A	10	8.7	0.2	2.5	0.4	4.9		10	11.1	0.4	3.9	0.9	7.8		
			B	10	8.2	0.2	2.7	0.4	5.5		10	10.3	0.3	2.5	0.5	5.0		
			C	10	7.8	0.2	3.1	0.5	6.3		10	8.9	0.1	1.7	0.3	3.3		
	HCN[b]	µg/cig	3R4F	9	104.7	7.1	6.8	14.3	13.7	2.4	10	392.3	19.9	5.1	39.8	10.1	1.6	0.7
			A	10	106.9	16.1	15.0	32.2	30.1		10	381.1	42.4	11.1	84.8	22.2		0.7
			B	10	86.9	13.6	15.7	27.2	31.3		10	328.7	19.1	5.8	38.3	11.6		0.4
			C	10	37.4	6.9	18.3	13.7	36.6		10	338.9	26.4	7.8	52.8	15.6		0.4
Ammonium ion	Cigarette weight	mg/cig	3R4F	9	1037	4	0.4	8	0.8	1.9	10	1074	12	1.1	24	2.2	1.1	
			A	10	922	10	1.1	21	2.3		10	960	10	1.1	21	2.1		
			B	10	897	6	0.7	13	1.4		10	935	12	1.3	24	2.6		
			C	10	840	4	0.5	8	1.0		10	884	10	1.1	20	2.3		
	Puff No.		3R4F	9	8.4	0.2	2.2	0.4	4.4	1.3	10	10.5	0.2	2.1	0.4	4.2	1.6	
			A	10	8.3	0.2	2.7	0.4	5.4		10	10.7	0.4	4.0	0.9	8.1		
			B	10	7.9	0.2	2.8	0.4	5.5		10	10.0	0.3	3.2	0.6	6.5		
			C	10	7.5	0.2	3.1	0.5	6.2		10	8.9	0.3	3.0	0.5	6.1		
	Ammonium ion[b]		3R4F	9	7.9	0.4	5.5	0.9	11.0	1.1	10	29.8	0.9	3.1	1.9	6.3	2.2	0.6
			A	10	10.2	0.6	6.0	1.2	12.0		10	42.8	4.1	9.6	8.2	19.1		1.6
			B	10	9.2	0.6	6.5	1.2	13.0		10	36.0	1.9	5.2	3.7	10.4		0.8
			C	10	4.2	0.2	5.2	0.4	10.4		10	28.0	1.7	6.2	3.5	12.4		1.2
Semi volatiles	Cigarette weight	mg/cig	3R4F	9	1038	4	0.4	8	0.7	2.0	8	1053	16	1.5	32	3.0	0.7	
			A	10	919	8	0.9	17	1.8		10	959	10	1.0	19	2.0		
			B	10	893	6	0.7	13	1.4		10	934	7	0.8	15	1.6		
			C	10	839	4	0.5	9	1.1		10	887	11	1.2	21	2.4		
	Puff No.		3R4F	9	8.3	0.2	2.0	0.3	4.1	0.9	8	10.5	0.5	4.5	1.0	9.1	0.7	
			A	10	8.3	0.2	2.7	0.5	5.5		10	10.8	0.3	2.9	0.6	5.7		
			B	10	8.0	0.1	1.6	0.3	3.2		10	10.0	0.3	3.2	0.6	6.3		
			C	10	7.6	0.1	1.4	0.2	2.8		10	8.9	0.3	3.9	0.7	7.8		
	TPM	mg/cig	3R4F	9	10.0	0.5	4.9	1.0	9.9	0.9	8	43.6	1.2	2.8	2.4	5.6	1.6	
			A	10	13.7	0.5	3.9	1.1	7.8		10	51.8	2.1	4.1	4.2	8.2		
			B	10	12.4	0.7	5.7	1.4	11.3		10	48.3	1.9	3.9	3.8	7.9		
			C	10	5.4	0.2	4.0	0.4	7.9		10	34.9	1.9	5.4	3.8	10.8		
	Pyridine	µg/cig	3R4F	9	4.5	0.6	13.9	1.3	27.9	1.2	8	32.5	4.3	13.1	8.5	26.2	1.0	0.9
			A	10	8.0	1.3	16.6	2.6	33.2		10	40.2	4.6	11.4	9.2	22.8		0.7
			B	10	7.3	1.2	16.6	2.4	33.1		10	33.2	3.5	10.5	6.9	20.9		0.6
			C	10	2.9	0.5	18.4	1.1	36.8		10	25.4	4.0	15.8	8.0	31.6		0.9
	Quinoline	µg/cig	3R4F	9	0.27	0.03	12.5	0.07	25.0	1.5	8	0.47	0.06	12.2	0.11	24.4	0.8	1.0
			A	10	0.43	0.07	15.9	0.14	31.8		10	0.75	0.06	8.6	0.13	17.3		0.5
			B	10	0.44	0.08	18.1	0.16	36.3		10	0.71	0.06	8.8	0.13	17.7		0.5
			C	10	0.15	0.04	22.7	0.07	45.3		10	0.43	0.05	12.0	0.10	24.0		0.5
	Styrene	µg/cig	3R4F	9	4.2	0.5	12.5	1.1	25.0	1.0	8	16.7	1.7	10.1	3.4	20.2	1.2	0.8
			A	10	6.0	0.7	12.2	1.5	24.5		10	20.0	1.9	9.6	3.9	19.3		0.8
			B	10	5.4	0.5	8.9	1.0	17.7		10	17.1	2.0	11.6	4.0	23.2		1.3
			C	10	2.7	0.4	15.0	0.8	30.0		10	15.0	2.2	14.7	4.4	29.4		1.0
Metals	Cigarette weight	mg/cig	3R4F	10	1042	3	0.3	7	0.6	2.4	12	1086	5	0.5	11	1.0	2.7	
			A	10	926	9	0.9	18	1.9		10	966	10	1.0	19	2.0		
			B	10	900	9	1.0	18	2.0		10	941	13	1.4	26	2.8		
			C	10	846	3	0.4	7	0.8		10	889	15	1.7	30	3.3		
	Puff No.		3R4F	10	8.2	0.1	1.4	0.2	2.8	2.1	12	10.6	0.2	1.5	0.3	3.0	1.5	
			A	10	8.1	0.2	2.5	0.4	5.0		10	10.7	0.3	3.1	0.7	6.2		
			B	10	7.9	0.2	2.0	0.3	4.1		10	10.2	0.2	2.1	0.4	4.1		
			C	10	7.5	0.3	4.0	0.6	8.0		10	8.6	0.1	1.7	0.3	3.5		
	Arsenic	ng/cig	3R4F	10	<2.7						12	12.2	1.1	9.0	2.2	18.0	3.7	
			A	10	2.7						10	9.6	3.6	38.0	7.3	75.9		
			B	10	2.8						10	<8.8	2.5	28.9	5.0	57.8		

(*continued on next page*)

Table 3 (*continued*)

Analysis	Analyte	Units	Product	ISO smoking regime							HCI smoking regime							HCI CV/ISO CV
				No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	No. samples	Mean	SD	CV (%)	2 SD	2 CV (%)	CV ratio[a]	
			C	10	<2.7						10	<8.8						
	Cadmium[b]	ng/cig	3R4F	10	29.8	1.7	5.8	3.5	11.6	2.2	12	108.6	5.9	5.4	11.8	10.9	1.9	0.9
			A	10	30.0	5.1	16.9	10.2	33.9		10	96.7	11.3	11.7	22.7	23.4		0.7
			B	10	29.1	2.5	8.6	5.0	17.2		10	92.9	6.7	7.2	13.4	14.5		0.8
			C	10	13.2	1.6	12.0	3.2	24.0		10	77.8	9.7	12.5	19.4	24.9		1.0
	Chromium	ng/cig	3R4F	10	<3.7						12	<14.0						
			A	10	<3.7						10	<14.0						
			B	10	<3.7						10	<14.0						
			C	10	<3.7						10	<14.0						
	Mercury	ng/cig	3R4F	10	<1.1						12	5.2	0.3	5.6	0.6	11.2		
			A	10	<1.1						10	<4.2						
			B	10	<1.1						10	<4.2						
			C	10	<1.1						10	<4.2						
	Nickel	ng/cig	3R4F	10	<30.9						12	<25.5						
			A	10	<30.9						10	<25.5						
			B	10	<30.9						10	<25.5						
			C	10	<30.9						10	<25.5						
	Lead	ng/cig	3R4F	10	<10.3						12	34.4	4.3	12.6	8.7	25.2	0.9	
			A	10	11.0						10	33.3	3.5	10.5	7.0	20.9		
			B	10	12.7						10	35.2	3.2	9.0	6.3	18.0		
			C	10	<10.3						10	26.3	3.8	14.3	7.5	28.7		
	Selenium	ng/cig	3R4F	10	<2.3						12	<15.6						
			A	10	<2.3						10	<15.6						
			B	10	<2.3						10	<15.6						
			C	10	<2.3						10	<15.6						
Nitrogen oxides	Puff No.		3R4F	9	8.2	0.2	1.9	0.3	3.7	1.3	9	10.3	0.2	2.3	0.5	4.6	0.9	
			A	10	8.2	0.2	1.9	0.3	3.8		10	10.7	0.2	2.2	0.5	4.3		
			B	10	8.0	0.1	1.4	0.2	2.8		10	10.2	0.3	2.5	0.5	5.0		
			C	10	7.4	0.3	3.7	0.6	7.5		10	8.7	0.1	1.3	0.2	2.5		
	NO	µg/cig	3R4F	9	206	17	8.0	33	16.0	1.1	9	540	20	3.7	40	7.3	1.7	0.5
			A	10	153	17	11.0	34	22.0		10	375	28	7.4	56	14.8		0.7
			B	10	135	12	9.1	24	18.1		10	351	15	4.3	30	8.7		0.5
			C	10	90	6	6.8	12	13.6		10	328	22	6.6	43	13.2		1.0
	NOx[b]	µg/cig	3R4F	9	228	12	5.5	25	10.9	1.3	9	608	28	4.5	55	9.1	1.3	0.8
			A	10	169	12	7.3	25	14.6		10	433	33	7.6	66	15.3		1.0
			B	10	151	10	6.9	21	13.7		10	414	18	4.3	35	8.5		0.6
			C	10	99	7	6.7	13	13.5		10	384	21	5.5	42	10.9		0.8

[a] CV ratio = mean CV commercial product/CV 3R4F.
[b] Smoke toxicants of regulatory relevance (Table 1).

Table 4
Monthly variation in emission levels of mainstream smoke toxicants as a ratio to nicotine.

Toxicant	Unit per mg nicotine	Product	HCI data as a ratio to nicotine					
			Sample No.	Mean	SD	CV (%)	2CV (%)	CV ratio[a]
TPM	mg	3R4F	7	22.3	0.6	2.5	5.0	1.5
		A	10	22.2	0.6	2.7	5.5	
		B	10	22.7	0.8	3.5	7.0	
		C	10	22.7	1.1	5.0	10.0	
NNK	ng	3R4F	9	157.6	14.7	9.3	18.6	1.8
		A	10	56.4	12.4	22.0	44.1	
		B	10	71.5	11.7	16.4	32.8	
		C	10	50.2	6.0	12.0	24.1	
NNN	ng	3R4F	9	158.7	12.0	7.6	15.1	2.1
		A	10	141.7	27.2	19.2	38.4	
		B	10	77.1	8.8	11.3	22.7	
		C	10	138.7	23.7	17.1	34.2	
Acetaldehyde	µg	3R4F	10	627	28	4.5	9.1	1.6
		A	10	468	28	6.0	12.0	
		B	10	520	30	5.8	11.6	
		C	10	626	62	9.9	19.7	
Acrolein	µg	3R4F	10	63.5	4.8	7.6	15.1	1.0
		A	10	48.8	3.6	7.4	14.8	
		B	10	56.8	3.7	6.5	12.9	
		C	10	64.3	6.2	9.7	19.3	
Acrylonitrile	µg	3R4F	8	17.1	0.9	5.3	10.7	1.8
		A	10	16.1	1.2	7.6	15.2	
		B	10	15.2	1.4	9.1	18.2	
		C	10	21.1	2.4	11.3	22.7	
4-ABP	ng	3R4F	7	1.4	0.1	3.9	7.9	2.2
		A	10	1.6	0.1	7.9	15.7	
		B	10	1.4	0.1	8.1	16.3	
		C	10	1.7	0.2	9.9	19.9	
1-AN	ng	3R4F	7	11.6	0.6	4.9	9.8	1.5
		A	10	12.7	1.0	7.7	15.5	
		B	10	12.5	1.1	8.8	17.6	
		C	10	12.1	0.7	5.9	11.8	
2-AN	ng	3R4F	7	6.8	0.3	3.9	7.7	1.6
		A	10	7.1	0.5	6.5	13.0	
		B	10	6.9	0.5	7.2	14.5	
		C	10	7.0	0.4	5.0	10.0	
Ammonium ion	µg	3R4F	10	15.2	0.7	4.6	9.3	1.6
		A	10	18.4	1.6	8.8	17.6	
		B	10	16.9	0.9	5.4	10.7	
		C	10	18.0	1.4	7.6	15.1	
Benzene	µg	3R4F	8	50.4	3.6	7.1	14.2	1.2
		A	10	42.3	3.2	7.6	15.2	
		B	10	40.8	3.2	7.9	15.7	
		C	10	53.7	5.3	9.9	19.7	
B[*a*]P	ng	3R4F	7	6.89	0.44	6.4	12.7	0.9
		A	10	7.42	0.50	6.8	13.5	
		B	10	7.40	0.42	5.7	11.4	
		C	10	8.37	0.43	5.2	10.3	
1,3-Butadiene	µg	3R4F	8	46.0	13.3	28.8	57.7	0.8
		A	10	38.9	9.2	23.5	47.1	
		B	10	38.1	8.6	22.7	45.4	
		C	10	52.1	11.8	22.7	45.4	
Cadmium	ng	3R4F	12	55.3	3.0	5.4	10.7	2.0
		A	10	41.4	3.9	9.4	18.8	
		B	10	43.7	3.6	8.2	16.4	
		C	10	50.0	6.9	13.9	27.8	
CO	mg	3R4F	7	15.8	0.5	3.3	6.7	1.3
		A	10	11.5	0.4	3.5	6.9	
		B	10	12.3	0.4	3.1	6.2	
		C	10	14.8	0.9	6.2	12.3	
Catechol	µg	3R4F	8	49.7	3.2	6.5	13.0	1.4
		A	10	58.7	5.7	9.7	19.5	
		B	10	58.7	5.3	9.0	17.9	
		C	10	55.4	5.0	9.1	18.2	
Crotonaldehyde	µg	3R4F	10	23.6	2.0	8.5	17.1	1.2
		A	10	19.9	1.8	9.2	18.5	

(*continued on next page*)

Table 4 (*continued*)

Toxicant	Unit per mg nicotine	Product	HCI data as a ratio to nicotine					
			Sample No.	Mean	SD	CV (%)	2CV (%)	CV ratio[a]
		B	10	22.2	1.8	7.9	15.8	
		C	10	24.4	3.1	12.8	25.5	
Formaldehyde	μg	3R4F	10	36.7	3.3	9.1	18.2	1.5
		A	10	28.2	2.9	10.3	20.6	
		B	10	43.7	6.0	13.7	27.5	
		C	10	30.1	4.9	16.2	32.4	
HCN	μg	3R4F	10	199	8	4.1	8.2	2.5
		A	10	164	21	12.6	25.2	
		B	10	155	12	7.7	15.4	
		C	10	218	24	11.0	22.0	
Hydroquinone	μg	3R4F	8	44.1	2.6	6.0	12.0	1.4
		A	10	54.0	4.8	9.0	18.0	
		B	10	57.3	4.9	8.6	17.2	
		C	10	54.7	4.1	7.5	15.1	
Isoprene	μg	3R4F	8	530	45	8.6	17.1	0.9
		A	10	435	37	8.5	17.0	
		B	10	434	32	7.4	14.7	
		C	10	559	43	7.6	15.2	
NO*x*	μg	3R4F	9	308	16	5.2	10.5	1.5
		A	10	186	15	8.1	16.1	
		B	10	195	12	5.9	11.9	
		C	10	247	22	8.9	17.7	
Toluene	μg	3R4F	8	83.3	4.5	5.4	10.8	1.0
		A	10	71.3	4.5	6.2	12.5	
		B	10	66.9	2.5	3.7	7.4	
		C	10	83.1	5.4	6.5	13.1	

[a] CV ratio = mean CV commercial product/CV 3R4F.

variation was found to be less than 15% for most toxicants. For a few toxicants present at the ng/g or ng/cigarette (i.e. ppb) level, however, variation was 20% or even higher. The sources of this variation may be analytical in origin or may derive from the products themselves. These two sources will contribute to every measurement, but the relative magnitudes remain to be established.

4.1. Analytical variation including sample preparation

Factors affecting the variability of an analytical method include preparation of the sample, the amount of analyte measured, and the complexity of the analysis method. The effects of all of these factors together are reflected in the finding that nicotine and CO emissions, which were measured at milligram per cigarette (mg/cig) levels after a simple sample extraction, showed much less variation (mean CV $\leqslant$ 7%) as compared with tobacco-specific nitrosamine (TSNA) emissions, which were determined at nanogram per cigarette (ng/cig) levels and required a more complex sample clean-up procedure (mean CV < 30%). This is in keeping with the observations of Horwitz et al. (1980) who demonstrated that the relative standard deviation for an analyte increases exponentially as its concentration decreases, regardless of the analyte being measured.

In the present study, most of the toxicants were measured by multi-analyte methods. Comparison of the variation among toxicants within a single analysis highlighted some with relatively high variation as compared with others within the same analysis. For example, in the analysis of carbonyl emissions, both crotonaldehyde (12.5%) and formaldehyde (10.9%) showed notably more variation than acetaldehyde (6.8%) under ISO conditions, and it is likely that the 10-fold higher level of acetaldehyde measured contributes significantly to this lower level of variation.

4.1.1. Variation due to smoke sample preparation

For each toxicant analysis, the weight of the cigarette used, the puff number and the TPM were also recorded to enable an estimation of the variation due to preparation of the smoke sample itself. These data document the build-up of variation from the product through the smoking process to the final preparation for toxicant analysis. In the analysis of NNK in mainstream smoke, for example, the variation in product is represented by the cigarette weight CV, the variation in the smoking process by the puff number CV, and the variation in the final preparation for toxicant analysis by the TPM CV. Fig. 1 shows that CV increased steadily across these attributes, giving a practical lower limit of variation for a relatively controlled analysis. Fig. 1 also shows the variation in levels of NNK in the cigarette filler blend and demonstrates that the variation in the filler blend is also a significant driver of the variation of NNK emissions in the smoke. This is true for other toxicants present in the filler blend, including NNN, B[*a*]P and cadmium (see Supplementary Figures, which also includes a figure for benzene for comparison as an analyte which is not present in the cigarette filler blend).

We would not expect the variation observed in the final toxicant analysis to be less than that observed in the final preparation of smoke for toxicant analysis; therefore, we would not expect variation in smoke emissions from a single product to be less than the variation in TPM. Variation in TPM measurements in the present study were typically 3–6% CV under both ISO and HCI regimes, indicating the lower limit of practical variability. Furthermore, if a toxicant is present in the filler blend and can readily distil to the smoke, then variation in its levels in the filler blend will contribute to the overall variation observed in the emissions of that toxicant. This also holds if the precursor of a toxicant varies in the filler blend.

4.1.2. Variation due to smoking regime

Current and proposed regulations for measurement and reporting or mandated lowering of smoke toxicant levels require data generated at various regimes, for example: in Brazil, ANVISA require measurement and reporting under the ISO smoking regime (Brazil Resolution, 2007); in the US, the FDA require measurement and reporting under both ISO and an intense (HCI) regime (FDA, 2012b), whilst the TobReg propose mandated lowering of toxicants when determined as a ratio to nicotine under HCI regime (WHO, 2008).

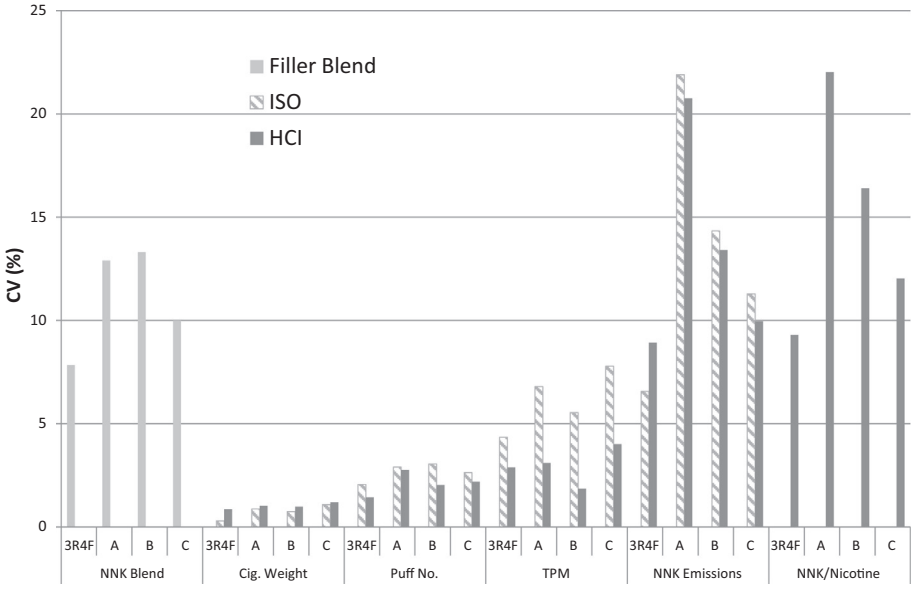

Fig. 1. Coefficients of variation for various aspects of NNK analysis.

The primary reason given by the TobReg for proposing mandated toxicant ceilings under the HCI regime is that variation (CV) for the determination of NNK and NNN is less when these toxicant emissions are determined under HCI than when determined under ISO conditions. In this study, the CV for NNK determined under HCI conditions was 10% lower than when determined under ISO conditions for all 3 commercial products. However for NNN the results were mixed (Table 3). B[*a*]P and CO variation was also less for all three commercial products determined under HCI compared to ISO, whilst variation in nicotine, formaldehyde, acetaldehyde, acrolein and benzene under the two regimes were mixed. Results for 1,3-butadiene determined under HCI were noticeably more variable than when determined under ISO due to analytical variability, as discussed in Section 4.1.3.

4.1.3. Variation due to analytical method

In the analysis of volatile compound emissions from products smoked under HCI conditions, benzene and 1,3-butadiene had average CVs of 6.1% and 20.9%, respectively. Both toxicants were measured in the same analysis and similar levels of each toxicant (~100 μg/cig) were found in smoke (Fig. 2). This is an example of an issue with the analytical method whereby the 1,3-butadiene results showed a synchronous pattern of variation across all four products (the three commercial cigarettes and 3R4F). Due to the different manufacturing histories of the four cigarettes, these temporal trends clearly indicate that the additional variability in 1,3-butadiene results, when determined under HCI conditions, was analytical in origin. This is potentially due to the higher volatility of 1,3-butadiene compared to benzene and suggests that there may be scope for improvement in the analytical method for this analyte in this laboratory.

This was the most striking example of the influence of analytical variation. Less pronounced examples of synchronous trends were observed, particularly where results were around the quantitation limits of the method, for example some of the minor nicotine alkaloids.

4.2. Product variation

Variations in the absolute levels of toxicants would be expected among products due to differences in product design and blend composition. It was known that the design and blend recipe of the BAT products (A and C) did not change during the course of this study (this aspect was unknown for product B, although our

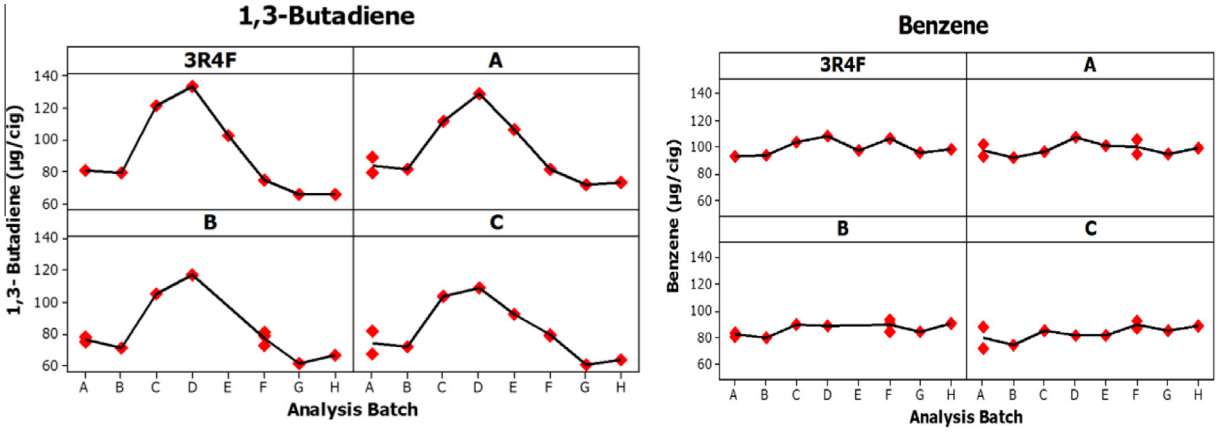

Fig. 2. Variation in monthly sample results for two volatile toxicants (1,3-butadiene and benzene) under HCI conditions. For this analysis, there were 8 rather than 10 analytical batches because more than one monthly sample of product was analysed in a single analytical batch on two occasions (analysis batch A and F). Also one monthly sample of product B was analysed in a different batch (batch F) to products A and C (Batch E).

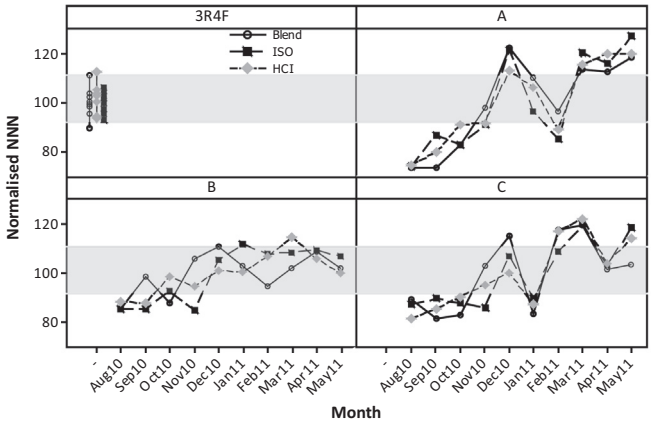

Fig. 3. Product variation in filler blend content and emission of NNN, showing variation related to month of sampling. Data for 3R4F data are plotted at a single undefined time-point, reflecting the provenance of these reference samples. The shaded area indicates the maximum variation in 3R4F data; data lying outside this region indicate that variation in the commercial products exceeds that in the 3R4F monitor cigarette.

analyses did not indicate any step change in product performance and levels of variability were similar to products A and C); nevertheless, variation might arise from factors such as the blending of tobacco grades that make up the cigarette filler blend, from the multiple sources of these grades over time, and from the multiple manufacturing operations, as well as from the inherent variation in the tobacco leaves themselves, tobacco being a natural product.

As an example, the blend contents and emission data for NNN, a TSNA that is readily transferred from the tobacco blend to the smoke during the combustion process, over the 10-month study period are shown for the four products in Fig. 3. To aid comparison, the NNN levels were normalised to the mean for each product to achieve a uniform scale across the three sets of data (blend, ISO and HCI). For the individual commercial products, the blend, ISO and HCI data showed similar trends over time: these trends demonstrate the variability of each commercial product over time. Notably, products A and C showed variation in the NNN levels of the monthly product samples that was greater than that of the 3R4F monitor (Fig. 3, shaded region).

The additional variation in commercial products for each analyte, due to manufacturing and compositional variation, can be quantified by calculating the CV ratio (average CV of the commercial products to CV of 3R4F), based on the view that the variation in 3R4F is a measure of the repeatability of the method (i.e. the smallest level of reference variability that can be obtained with a cigarette product). A ratio of greater than 1 indicates that the variability in the commercial products over time is greater than the repeatability of the method.

As shown in Tables 2 and 3, the CV ratio averaged 1.7 for blend components, 1.5 for smoke emissions determined under the HCI regime, and 1.7 for smoke emissions determined under the ISO regime. Some toxicants such as 1,3-butadiene had a ratio of less than 1.0 (0.8 under ISO conditions, 0.7 under HCI conditions), indicating that analytical variability was greater than the variability induced by multiple manufacturing batches for this toxicant.

For several toxicants, the CV ratio was higher, between 2.0 and 3.5, indicating a greater degree of monthly variation in the commercial product as compared with 3R4F. This was true for total and reducing sugars, chlorogenic acid and cadmium in filler blend; nicotine, NNN, NNK, NAT, o- and p-cresol, hydroquinone, HCN and cadmium emissions under ISO conditions; 2-AN, 4-ABP, phenol, o- and p-cresol, arsenic and butyraldehyde under HCI conditions; and NNN, 4-ABP and HCN when determined as a ratio to nicotine under HCI conditions. All of these toxicants, with the exception of total sugars, reducing sugars and chlorogenic acid, were present at levels below 1000 ppm (Fig. 4), which may in part contribute to the greater variation. 'Total' and 'reducing' sugars are a collective term for a range of species determined colorimetrically by the current methods, rather than a specific chemical reaction, and therefore the results are likely to be more variable than for other more specific analyses. However the CV ratio demonstrates greater variability for total and reducing sugars in the commercial products, compared to 3R4F, which can only be due to product variability i.e. a combination of fluctuations in levels of sugars in the tobacco grades and the effects of curing on those initial sugar levels, similarly for chlorogenic acid. The high butyraldehyde CV ratio (5.8 under the HCI regime) is likely to be due to the atypically low variation in 3R4F (CV 2.6%) because the average CV for the commercial products was similar to that achieved under ISO smoking conditions.

4.3. Variation in toxicant emission levels as a ratio to nicotine

The TobReg recommendations include establishing levels for selected toxicant emissions per mg of nicotine, measured under

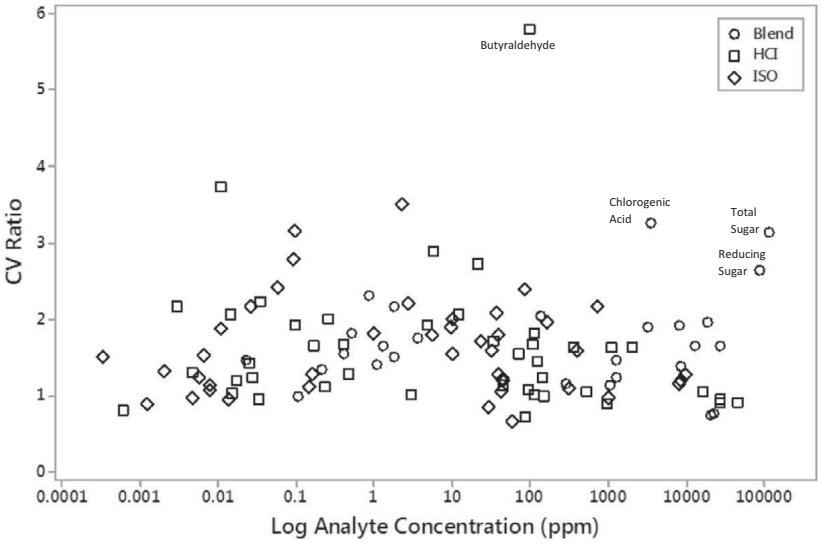

Fig. 4. CV ratio of all results versus analyte concentration.

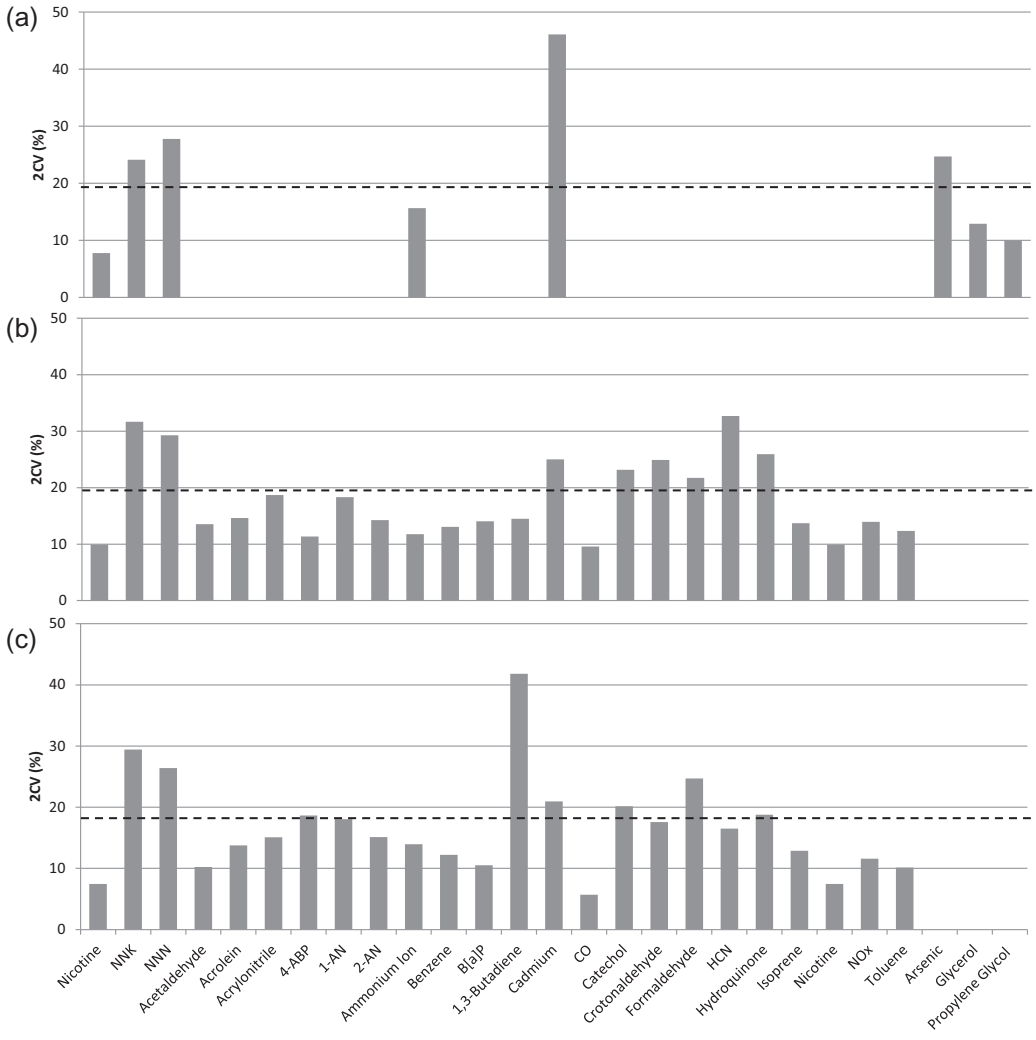

Fig. 5. Potential error associated with single-point-in-time analysis of toxicants of regulatory interest for commercial, high-volume cigarette products: (a) filler blend components, (b) toxicant emissions under ISO regime, (c) toxicant emissions under HCI regime. The dotted black line shows the median value for each of the 3 categories.

HCI conditions, and prohibiting cigarette products with yields above these levels (WHO, 2008). As summarised by Burns et al. (2008), the aim of comparing toxicant levels to nicotine is to shift the interpretation of the measurement away from the amount of smoke generated per cigarette and towards characterisation of smoke toxicity generated under standardised conditions.

Importantly, toxicant emission levels can be reported as a ratio to nicotine only via calculation based on the mean data per sample (i.e. toxicant mean divided by nicotine mean) because analysis of nicotine is done separately (i.e. using a different cigarette, and a different number of replicates) from that of most toxicants, CO being the exception. As a result, the calculation cannot take into account the variation in the individual replicates within each sample result.

When the current data were expressed as a ratio to nicotine, the variation in the data for the 9 toxicants mandated for lowering by TobReg tended to increase by 1–2 percentage points as compared with the absolute values (Tables 3 and 4), although for a few of the 23 toxicants (aromatic amines, phenols and cadmium), there was a small decrease in variability. This is not surprising because there is inherent product and analytical variability in each of the two measurements (toxicant and nicotine emission), and combining the data should in theory lead to an increase in the variability observed as the measurement error for each compound is combined. Even analysis of CO, which was performed on the same cigarette as nicotine, tended to show an increase in variation between the absolute values and the CO/nicotine ratio (from 0.0% to 4.2%), owing to the combined errors in analytical measurement. The TPM to nicotine data, calculated using the TNCO TPM, which was subsequently analysed for nicotine emissions, illustrates the potential decrease in variation achievable when a paired analysis is conducted (Tables 3 and 4).

Related to this, Cahours et al. (2013) noted that, owing to use of different (unpaired) cigarettes for nicotine analysis versus other toxicant analyses, ISO methods could not provide a robust estimate of repeatability and reproducibility for nicotine ratio analysis (ISO, 1994), thus questioning the accuracy of this method of reporting emissions data.

4.4. Tolerance levels and indications for product regulation

For an analyte, the value of 2SD can be calculated as an approximation of 95% confidence intervals, thereby indicating the range of values that might be reported for a single large-volume commercial product over time when measured in a single laboratory. Stated as a percentage (2CV), this kind of variability or "tolerance" has been recognised in the regulatory reporting of TNCO when determined under the ISO regime, where a tolerance of

±15% for tar and nicotine reporting and ±20% for CO reporting are applicable with repeated sampling (ISO, 2013). The maximum 2CV values for TNCO determined in this single-centre study under ISO were 14.4%. If closer to 100% coverage of data were required, then an interval of 3SD (99.7%, empirically) would be more appropriate; and, if measurements were carried out at more than one laboratory, then inter-laboratory variation would also need to be taken into account, again increasing the interval substantially (Intorp et al., 2009; Teillet et al., 2013).

Considering all of the results for the commercial products in the present study (i.e. tobacco blend, and smoke emissions determined under either smoking regime and when expressed as a ratio to nicotine), 2CV values averaged ~20% (Tables 2–4). However, the 2CV value reached in excess of 50% for some low-level toxicants, where the levels were near to the quantitation limits of the analytical method.

These 2CV values demonstrate the uncertainty and therefore the potential error associated with a single-point-in-time sample of a high-volume commercial cigarette product in a single laboratory. Fig. 5 summarises these findings for the toxicants of regulatory interest defined in Table 1 and shows the median 2CV value for all toxicant measurements in each category: 19.4% for filler blend components, 19.5% for toxicant emissions under ISO, 17.8% for toxicant emissions under HCI.

This suggests that tolerances higher than those currently applicable to TNCO analysis under the ISO regime would be advisable for many of these smoke toxicants, should proposed ceilings be enacted.

5. Conclusions

Cigarette smoke toxicants have become a global regulatory issue, with mandatory reporting of smoke toxicant emission levels and cigarette filler blend components in several countries, and proposals for further regulation including toxicant ceilings for emissions and/or filler blend components. Against this background, an understanding of variation is essential.

Here, the monthly variation in toxicant emissions from smoke and blend toxicants evaluated in a single laboratory was found to be less than 15% CV for many toxicants, but considerably higher for others, particularly those at the nanogram per gram (ppb) level. For some toxicants such as 1,3-butadiene (CV < 28%), the source of the variation was clearly analytical, indicating that it might be necessary to improve the accuracy of the analytical methods before standardised regulatory reporting might be achieved. As compared with the 3R4F reference cigarette, the 10-month variability in blend components, ISO and HCI toxicant emissions was 50–70% higher for the commercial cigarettes. Because the 3R4F reference was manufactured in a single batch, these values provide an estimate of the long-term variation due to natural fluctuations in the tobacco blend and adjustments in the manufacturing process.

This study has demonstrated the uncertainty around measured levels of toxicants in three high-volume commercial cigarettes and therefore the potential error which might be associated with single-point-in-time sampling of these products. Defined as 2CV, this potential error averaged ~20% but was approaching 50% for some low level toxicants.

It is important to stress that these values are for a single laboratory. Repeated measures across more than one laboratory will increase the variability substantially (Gregg et al., 2004. Intorp et al., 2009). Taken together, the present results demonstrate the importance of taking all sources of variation into account when mandating maximum product levels of toxicants and analytical tolerance limits.

Conflict of interest

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.yrtph.2015.01.006.

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