



Comparison of select analytes in aerosol from e-cigarettes with smoke from conventional cigarettes and with ambient air



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ABSTRACT

Leading commercial electronic cigarettes were tested to determine bulk composition. The e-cigarettes and conventional cigarettes were evaluated using machine-puffing to compare nicotine delivery and relative yields of chemical constituents. The e-liquids tested were found to contain humectants, glycerin and/or propylene glycol, ($\geq 75\%$ content); water ($< 20\%$); nicotine (approximately 2%); and flavor ($< 10\%$). The aerosol collected mass (ACM) of the e-cigarette samples was similar in composition to the e-liquids. Aerosol nicotine for the e-cigarette samples was 85% lower than nicotine yield for the conventional cigarettes. Analysis of the smoke from conventional cigarettes showed that the mainstream cigarette smoke delivered approximately 1500 times more harmful and potentially harmful constituents (HPHCs) tested when compared to e-cigarette aerosol or to puffing room air. The deliveries of HPHCs tested for these e-cigarette products were similar to the study air blanks rather than to deliveries from conventional cigarettes; no significant contribution of cigarette smoke HPHCs from any of the compound classes tested was found for the e-cigarettes. Thus, the results of this study support previous researchers' discussion of e-cigarette products' potential for reduced exposure compared to cigarette smoke.

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1. Introduction

Electronic cigarettes (e-cigarettes) are a relatively new consumer product. Unlike conventional cigarettes, e-cigarettes do not burn tobacco to deliver flavor. Instead, they contain a liquid-based flavorant (typically referred to as e-liquid or e-juice) that is thermally vaporized by an electric element. This liquid typically consists of a mixture of water, glycerin, and/or propylene glycol. The liquid also contains nicotine and flavor, although nicotine-free products are available.

While there are decades of characterization studies and numerous standardized analytical procedures for conventional cigarettes,

relatively little published analytical data exists for commercial e-cigarette products. Furthermore, no standardized test methods or reference products exist for e-cigarettes.

Electronic cigarettes are generally purported to provide reduced exposure to conventional cigarettes' chemical constituents because they deliver flavors and nicotine through vaporization rather than by burning tobacco. [Goniewicz et al. \(2014\)](#) reported low levels of select chemical constituents in select e-cigarette brands commercially available in Poland. A recent review of analyses from diverse e-cigarettes shows comparatively simple chemical composition relative to conventional cigarette smoke ([Burstyn, 2014](#)). However, limited published results exist for commercial products that represent a significant presence in the marketplace ([Cheng, 2014](#)).

The purpose of this study was to evaluate e-cigarette products with a significant presence in the marketplace for bulk composition, including nicotine, and for select constituents for comparison with conventional cigarette products. Three blu eCigs products (approximately 50% of the US market) and two SKYCIG products (approximately 30% of the UK market) were chosen for evaluation. Marlboro Gold Box (US), and Lambert & Butler Original and Menthol products (UK), with significant market share in their respective geographical areas, were included in the study for conventional cigarette comparisons.

Abbreviations: ACM, aerosol collected mass; HPHC, harmful and potentially harmful constituents; CO, carbon monoxide; TSNA, tobacco-specific nitrosamines; PAA, polyaromatic amines; PAH, polyaromatic hydrocarbons; LOQ, limit of quantitation; LOD, limit of detection; CAN, Health Canada Test Method T-115; blu CTD, Classic Tobacco Disposable; blu MMD, Magnificent Menthol Disposable; blu CCH, Cherry Crush, Premium, High Strength; SKYCIG CTB, Classic Tobacco Bold; SKYCIG CMB, Crown Menthol Bold; MGB, Marlboro Gold Box; L&B O, Lambert & Butler Original; L&B M, Lambert & Butler Menthol; TPM, total particulate matter; PG, propylene glycol.

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The products used in the study were evaluated for content and delivery of major ingredients (glycerin, propylene glycol, water, and nicotine) and for select constituents (carbon monoxide (CO), carbonyls, phenolics, volatile organic compounds (volatiles), metals, tobacco-specific nitrosamines (TSNAs), polyaromatic amines (PAAs), and polyaromatic hydrocarbons (PAHs)). Many of these constituents are included in cigarette industry guidance issued by the FDA that includes reporting obligations for harmful and potentially harmful constituents (HPHCs) in cigarette filler and smoke under section 904(a)(3) of the 2009 Family Smoking Prevention and Tobacco Control Act (FDA, 2012). For delivery studies, the conventional cigarettes were smoked under an intense puffing regime published by Health Canada (1999). The e-cigarettes were tested using minimal modifications to this smoking regime. Ninety-nine puffs were used to collect approximately the same aerosol mass as obtained from conventional cigarette testing. Ambient 'air' samples, empty port collections, were included as a negative control of aerosol testing for cigarette constituents (i.e. HPHC).

2. Materials and methods

2.1. Test products

Two disposable e-cigarette products and three rechargeable e-cigarette products were obtained from the manufacturers. Three conventional cigarette products were purchased through wholesale or retail sources for testing. Information for each of the products is listed in Table 1.

2.2. Methods overview

ISO 17025 accredited analytical methods were used to evaluate the cigarette samples for select HPHCs in mainstream smoke. Official methods are cited and other, internally validated, methods are briefly described for general understanding. Furthermore, because no standardized methods exist for e-cigarette analysis, the methods used to evaluate the conventional cigarettes were adapted to evaluate the e-cigarette products and the study blanks (room air). In an effort to maximize signal and lower methods' limits of quantitation, aerosol collection amounts were maximized (but maintained below breakthrough) and extraction solvent volumes were minimized. In some cases, alternative instrumentation was employed to improve detection. For example, mainstream smoke TSNAs were analyzed by GC-TEA while aerosol and air blank samples were analyzed by LC-MS/MS. Accuracy, precision, and method limits of quantitation and detection (LOQ and LOD) were verified for each method. On average, accuracy and method variability for the analytes tested were determined to be 98% and 3%, respectively. Analyte LOD and LOQ information is listed in Supplemental Appendix A Tables 1 and 2. Method resolution for low levels of analytes was influenced by background levels of select analytes in air control samples. These background levels are attributed to

instrument or smoking machine carry-over as evidenced in solvent or air blanks. In addition, the high concentration of glycerin and water in e-cigarette aerosol present challenges for volatile-based measurement systems (i.e. GC). Additional method refinements and dedicated e-cigarette puffing machines are two areas for consideration to improve e-cigarette aerosol method sensitivities. Method development and verification details for e-cigarette liquids and aerosols are the subject of a future publication.

2.3. Smoke and aerosol collection

Cigarette preparation and machine smoking for conventional cigarettes are described in Health Canada Test Method T-115 (CAN) (1999). Two to three cigarettes were smoked per replicate for conventional cigarettes and 99 puffs were taken from single e-cigarettes for no more than approximately 200 mg of particulates collected per pad. Three to five replicates were tested for each measurement. Prior to analysis, filter pads from cigarette smoke collection were visually inspected for overloading of particulates, as evidenced by brown spotting on the back of the filter pad. To ensure no overloading of particulates for aerosol collection, e-cigarette units were weighed before and after collection to verify that product weight change and filter pad weight change were comparable. Air blanks were prepared by puffing room air (99 puffs) through an empty smoking machine port to the indicated trapping media for an analysis method. These air blank samples were prepared and analyzed in the same manner and at the same time as the e-cigarette aerosol samples. Smoke and aerosol collection sections were conducted separately. Smoke and aerosol particulate was collected onto 44 mm glass fiber filter pads with >99% particulate trapping efficiency for each replicate analysis. For carbonyls, smoke/aerosol was collected directly by two impingers, in series. For smoke metals analysis, electrostatic precipitation was used. For volatiles and PAH determinations, single chilled impingers were placed in-line with the filter pads. e-Liquid glycerin and nicotine were quantitated using GC-FID and/or GC-MS using a method equivalent to ISO 10315 (ISO, 2000a). e-Liquid water was quantitated using Karl Fischer analysis. A reference e-liquid was developed and used as a testing monitor for ingredient determinations in the e-liquid samples. The reference e-liquid is composed primarily of glycerin, propylene glycol, and water with low levels of nicotine, menthol, and Tween 80. The Tween 80 is added to improve solubility of menthol in the solution. The reference is not meant to directly mimic an e-liquid used for consumption but merely used for analytical control charts. Three replicates were tested for each sample and the reference.

2.4. Analytical assays

Carbon monoxide was determined concurrently with aerosol and smoke collection for nicotine and water and analyzed by NDIR using ISO method 8454:2007 (ISO, 2007). Carbonyls were trapped using 2,4-dinitrophenylhydrazine as a derivatizing agent with

Table 1
List of cigarette and e-cigarette products tested.

Product	Manufacturer	Product type	Nicotine information provided on packaging
Classic Tobacco Disposable (blu CTD)	blu eCigs	Disposable e-cigarette	Content: 24 mg/unit
Magnificent Menthol Disposable (blu MMD)	blu eCigs	Disposable e-cigarette	Content: 24 mg/unit
Cherry Crush, Premium, High Strength (blu CCH)	blu eCigs	Rechargeable e-cigarette	Content: 16 mg/unit
Classic Tobacco Bold (SKYCIG CTB)	SKYCIG	Rechargeable e-cigarette	Content: 18 mg/unit
Crown Menthol Bold (SKYCIG CMB)	SKYCIG	Rechargeable e-cigarette	Content: 18 mg/unit
Marlboro Gold Box (MGB)	Philip Morris USA	Conventional cigarette	-
Lambert & Butler Original (L&B O)	Imperial Tobacco	Conventional cigarette	Yield: 0.9 mg/cig (ISO)
Lambert & Butler Menthol (L&B M)	Imperial Tobacco	Conventional cigarette	Yield: 0.5 mg/cig (ISO)

subsequent analysis by UPLC–UV using CORESTA method 74 (CORESTA, 2013). For phenolics determination, filter pads were extracted with 20 mL of 1% acetic acid/2.5% methanol (MEOH) in water using 30 min of agitation. Extracts were analyzed by UPLC–fluorescence detection using a C18 column for separation. For volatiles analysis, filter pads and impinger solutions (20 mL MEOH) were combined. Extracts were analyzed by GC–MS in SIM mode using a WAX capillary column. For metals analysis, cigarette smoke was collected using an electrostatic precipitator while e-cigarette aerosol was collected on glass fiber filter pads. After smoking, the cigarette smoke condensate was rinsed from the electrostatic precipitation tube using methanol. The dried condensates were digested using hydrochloric (10% v/v), nitric acids (80% v/v), and heat and were diluted prior to analysis by ICP–MS. For aerosol samples, filter pads were extracted using 20 mL of a mixture of nitric (2% v/v) and hydrochloric acids (0.5% v/v) using wrist action shaker (20 min). Resultant extracts were analyzed by ICP–MS equipped with an octapole reaction cell.

For TSNA analysis of smoke, samples were extracted in nonpolar solvent, treated to an SPE clean-up, concentrated and analyzed by GC–TEA following CORESTA method 63 (CORESTA, 2005). For TSNA analysis of aerosol samples, filter pads were extracted with 20 mL of 5 mM aqueous ammonium with 15 min of shaking. Extracts were analyzed by LC–MS/MS with a C18 column. For PAA determinations, filter pads were extracted using 25 mL of 5% HCl (aq) and shaking (30 min) followed by solvent exchange and derivatization with pentafluoropropionic acid anhydride and trimethylamine. After an SPE clean-up step (Florisil® SEP-PAK), samples were analyzed by GC–MS in SIM mode using negative chemical ionization. PAH analysis was conducted by extraction in MEOH followed by SPE clean-up and analysis by GC–MS in SIM mode (Tarrant et al., 2009).

The results obtained from these analyses were tabulated as mean ± one standard deviation for levels of selected compounds in Supplementary Appendix A. In cases where quantifiable amounts of analyte were present in an e-cigarette aerosol sample above that of the associated air blanks, an Analysis of Variance (ANOVA) was used to compare the means for the cigarette smoke data with respective aerosol data. Statistical analyses were performed using JMP 10.0.0 (SAS Institute, Inc. Cary, NC, USA). The significance level was established as $p < 0.05$ for all comparisons.

3. Results and discussion

3.1. Collection of aerosol

Machine smoking of cigarettes under standardized regimes is for comparative purposes and is not intended to represent the

range of consumer smoking behaviors. Thus, standardized equipment, cigarette reference products, and methodology have been established to allow comparison of different products under a common set of controlled conditions. ISO 3308:2000E and Health Canada (CAN) methods are frequently used for standardized smoking of conventional cigarettes for the purposes of laboratory comparisons among products (ISO, 2000b; Health Canada, 1999). Following each of these methods, conventional cigarettes are smoked to a specified butt length using a fixed and specified puffing volume, duration, and interval.

Regarding e-cigarette experimentation, there is no generally accepted standard e-cigarette puffing regime at this time. Topography studies are limited but anecdotal information indicates e-cigarette usage depends greatly on the individual consumer and product design and capabilities. For the purposes of this study, our objective was to collect sufficient aerosol to be able to detect, if present, select HPHCs. A wide range of parameters would be adequate to accomplish this. Given the objectives of this study, use of collection parameters which are compatible with conventional and electronic cigarettes was essential for facilitating comparisons between cigarette smoke and e-cigarette aerosol. The more intense of the standard regimes used with cigarettes, CAN, which requires 55 mL puffs taken twice a minute, was adapted for this investigation. The key difference required for testing e-cigarettes with the CAN method is that a fixed puff count (rather than ‘butt length’) is necessary for aerosol collection. A standard of 99 puffs was adopted for all e-cigarette and air blank analyses. This puff count provides similar total particulate collection per pad between the e-cigarette samples and the conventional cigarette testing. This also represents approximately 11 times more puffs than are typically observed for a conventional cigarette. Marlboro Gold Box, L&B O, and L&B M averaged 9.1, 8.2, and 7.2 puffs per cigarette, respectively, when machine-smoked to the standard butt length. If more aggressive puffing parameters had been chosen for the study, the puff count specification would have been lowered to maintain the target level of ACM collected. Note that the range of puffs collected in-use may vary widely depending on product design, battery strength, and user puffing preferences. Thus, the 99 puffs collection in this study is not intended to represent a life time use yield for any of the analytes tested.

3.2. Aerosol and smoke characterization – reference information

Traditional cigarette testing incorporates the use of monitor or reference cigarettes that serve as positive controls and provide quality metrics for standardized analytical methods. Key examples are Kentucky Reference cigarettes and CORESTA monitor cigarettes (CORESTA, 2009; ISO, 2003; University of Kentucky, 2014). Each of

Table 2
Percent composition of e-liquid and aerosol.

	Glycerin (%)	Propylene glycol (%)	Water (%)	Nicotine (%)	Flavor ^a (%)
<i>e-Liquid composition</i>					
blu Classic Tobacco Disposable	82	–	9	2	7
blu Magnificent Menthol Disposable	75	–	18	2	5
blu Cherry Crush High Premium	77	–	14	2	7
SKYCIG Classic Tobacco Bold	24	67	6	2	1
SKYCIG Crown Menthol Bold	21	66	7	2	4
<i>e-Cigarette aerosol composition^b</i>					
blu Classic Tobacco Disposable	73	–	15	1	11
blu Magnificent Menthol Disposable	80	–	18	2	–
blu Cherry Crush High Premium	70	–	19	1	10
SKYCIG Classic Tobacco Bold	24	61	10.4	1.4	3
SKYCIG Crown Menthol Bold	21	59	12	2	6

^a Flavor content is estimated by difference.

^b Aerosol % composition calculated based on the ACM delivery as analyte yield (mg)/ACM (mg) × 100.

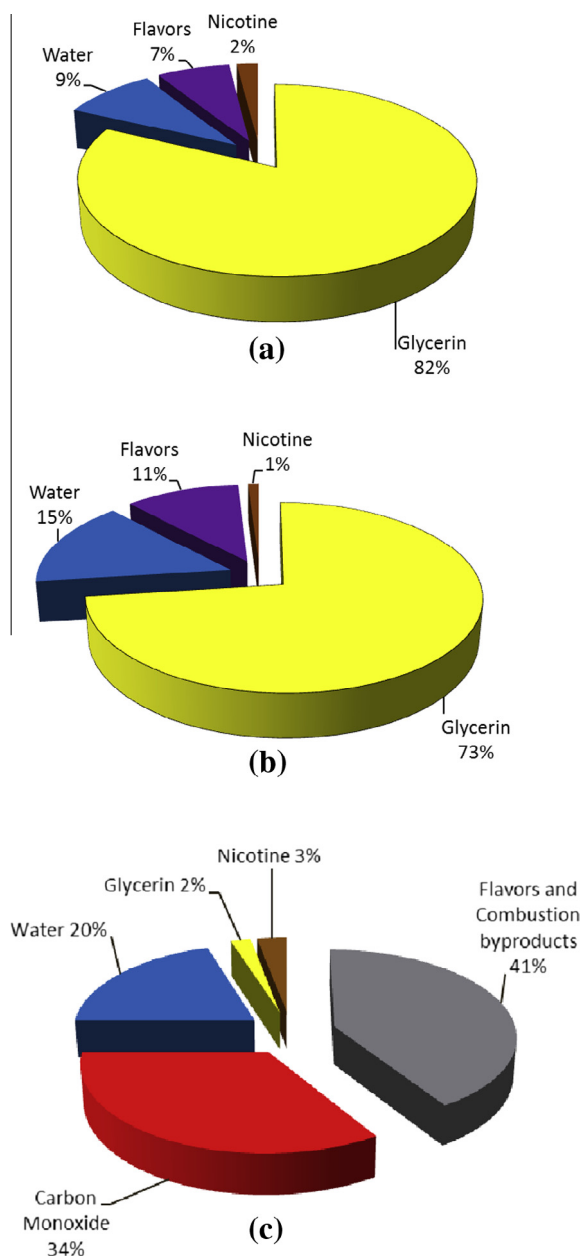


Fig. 1. Percent composition comparison for e-liquid, e-cigarette aerosol, and cigarette smoke: (a) Classic Tobacco Disposable e-liquid Composition. (b) Classic Tobacco Disposable Aerosol Composition (99 puffs, CAN). (c) Marlboro Gold Box Smoke Composition (9 puffs, CAN).

these reference cigarettes can serve as a single positive control and an indicator of method variability within and among laboratories for all analytes of interest. The manufacture, design, and function of these reference products are similar to those of commercial cigarettes. Currently reference products are not available for e-cigarette testing. Given the range of e-cigarette designs, development of a consensus strategy to produce positive controls or monitors for e-cigarette testing is needed.

In the absence of standardized e-cigarette references, measures were taken to ensure experimental robustness. For example, aerosol collected mass (ACM) results for the e-cigarette samples were compared across methods as an indicator of puffing consistency for a given product among the machine-puffing sessions required to conduct the battery of tests. Thus, if a sample set yielded ACM outside of a specified ranged deemed typical for a given product,

the sample set was repeated. This range was determined for each product based on collection of 20 or more replicates across the product lot using CAN parameters.

Also, because results from initial analyses indicated low or no measurable levels of many of the analytes, blank samples were included to verify any contribution of analyte from the laboratory environment, sample preparation, and/or analyses for each HPHC test method. The air blank results are listed with the samples' results in Tables 4 and 5. There were instances for which solvent blank and air blank samples had measurable levels of an analyte. This is due to the ubiquitous nature of some of the analytes, such as formaldehyde, or to carry-over. Laugesen reported similar findings (2009). These observations serve as a cautionary note regarding the measurement of extremely low levels of constituents with highly sensitive instrumentation.

3.3. Main ingredients

e-Liquid expressed from the individual products was tested for reported e-cigarette ingredients to compare the percent compositions of the e-liquids and the aerosols. Percent composition calculations of the ingredients are shown in Table 2 for each sample and in Fig. 1 for blu CTD, as this product's comparative results were exemplary of the samples. The primary ingredients in the e-cigarette samples were glycerin and/or propylene glycol ($\geq 75\%$). Water ($\leq 18\%$) and nicotine ($\sim 2\%$) were also present. Based on a mass balance, other ingredients, presumed to be flavorants, were present at less than 7%. Note that this calculation would also include method uncertainty and any possible HPHCs, if present. The composition of the aerosol was calculated based on the ACM delivery as analyte yield (mg)/ACM (mg) $\times 100$. The bulk composition of the delivered aerosol was similar to the bulk composition of the e-liquid.

By comparison, the total particulate matter (TPM) of the conventional cigarettes tested is 30% water and $<5\%$ nicotine. The essential difference between the ACM composition of the e-cigarettes tested and the TPM of the conventional cigarettes is that the remaining 65% of the TPM of the conventional cigarette is predominantly combustion byproducts. There was no detectable carbon monoxide in the emitted aerosol of the e-cigarette samples. The conventional cigarettes, on the other hand, delivered more than 20 mg/cig of CO. Smoke composition for Marlboro Gold Box, exemplary of the conventional cigarettes tested, is shown in Fig. 1 in contrast to the e-liquid and aerosol results for blu CTD.

While the percent composition of the nicotine in the ACM and TPM are relatively similar, it should be noted that the actual deliveries of nicotine are markedly lower for the e-cigarettes tested than the conventional cigarettes. The nicotine yields ranged from 8 $\mu\text{g}/\text{puff}$ to 33 $\mu\text{g}/\text{puff}$ for the e-cigarette samples which was 85% lower than the 194–232 $\mu\text{g}/\text{puff}$ for the conventional cigarettes. These results are presented in Table 3.

3.4. Aerosol and smoke HPHC testing

For cigarette smoke analysis, the conventional cigarettes were machine smoked by established cigarette smoking procedures. Approximately 7–9 puffs per cigarette were collected. For the e-cigarette samples and air blanks, 99 puffs were collected. Results were compared on an 'as tested' basis; i.e. yields for a single cigarette of 7–9 puffs compared to yields from 99 puffs of an e-cigarette as displayed in Table 4. Additionally, in order to simplify making comparisons between the cigarette and e-cigarette samples, all values were converted to yield per puff. These results are summarized by class in Table 5. Results for individual analytes are tabulated as mean \pm one standard deviation in Supplemental Appendix A Tables 1 and 2.

Table 3
Nicotine content and yield comparison between e-cigarettes and conventional cigarettes (mean ± standard deviation).

	Nicotine content (µg/unit)	Nicotine yield (µg/puff)
blu Classic Tobacco Disposable	20,600 ± 1500	33 ± 12
blu Magnificent Menthol Disposable	20,000 ± 300	25 ± 4
blu Cherry Crush High Premium	11,700 ± 300	8 ± 3
SKYCIG Classic Tobacco Bold	12,750 ± 295	29 ± 4
SKYCIG Crown Menthol Bold	13,027 ± 280	33 ± 6
Marlboro Gold Box	11,431 ± 80	226 ± 2
L&B Original	12,941 ± 26	232 ± 5
L&B Menthol	12,131 ± 24	194 ± 10

Number of replicates = 3–5.

Table 4
Analytical characterization of commercial e-cigarettes and conventional cigarettes collected using CAN parameters – select cigarette HPHC methodology (mg/total puffs collected) summary by analyte classes.

	CO	Carbonyls ^a	Phenolics ^b	Volatiles ^c	Metals ^d	TSNAs ^e	PAA ^f	PAH ^g	Sum
Marlboro Gold Box (mg/cig)	27	1.92	0.204	1.430	<0.00020	0.000550	0.000024	0.00222	<30.6 mg
L&B Original (mg/cig)	22	1.89	0.26	1.02	<0.0002	0.000238	0.000019	0.00219	<25.2
L&B Menthol (mg/cig)	20	1.81	0.17	0.94	<0.0003	0.000185	0.000017	0.00153	<22.9
blu CTD (mg/99 puffs)	<0.1	<0.07	<0.001	<0.001	<0.00004	<0.00002	<0.000004	<0.00016	<0.17
blu MMD (mg/99 puffs)	<0.1	<0.08	<0.001	<0.001	<0.00004	<0.00002	<0.000004	<0.00016	<0.18
blu CCHP (mg/99 puffs)	<0.1	<0.05	<0.003	<0.0004	<0.00004	<0.00002	<0.000004	<0.00014	<0.15
SKYCIG CTB (mg/99 puffs)	<0.1	<0.06	<0.0010	<0.008	<0.00006	<0.000013	<0.000014	<0.00004	<0.17
SKYCIG CMB (mg/99 puffs)	<0.1	<0.09	<0.0014	<0.008	<0.00006	<0.000030	<0.000014	<0.00004	<0.20
Air Blank (blu Set) (mg/99 puffs)	<0.1	<0.06	<0.001	<0.0004	<0.00004	<0.00002	<0.000004	<0.00015	<0.16
Air Blank (SKYCIG Set) (mg/99 puffs)	<0.1	<0.05	<0.0009	<0.008	<0.00006	<0.000013	<0.000014	<0.00006	<0.16

< Indicates some or all values were below method limits of quantitation or detection, number of replicates = 3–5.

^a Formaldehyde, acetaldehyde, acrolein propionaldehyde, crotonaldehyde, MEK, butyraldehyde.

^b Hydroquinone, resorcinol, catechol, phenol, m-+p-cresol, o-cresol.

^c 1,3-Butadiene, isoprene, acrylonitrile, benzene, toluene, styrene.

^d Beryllium, cadmium, chromium, cobalt, lead, manganese, mercury, nickel, selenium, tin.

^e NNN, NAT, NAB, NNK.

^f 1-Aminonaphthalene, 2-aminonaphthalene, 3-aminobiphenyl, 4-aminobiphenyl.

^g Naphthalene, acenaphthylene, acenaphthene, fluorine, phenanthrene, anthracene, fluoranthene, pyrene, benzanthracene, chrysene, benzo(b)fluoranthene, benzo(k)fluoranthene, B(a)P, indeno[1,2,3-cd]pyrene, benzo(g,h,i)perylene.

Table 5
Analytical characterization of commercial e-cigarettes and conventional cigarettes collected using CAN parameters – select cigarette HPHC methodology (µg/puff) summary by analyte classes.

	CO	Carbonyls ^a	Phenolics ^b	Volatiles ^c	Metals ^d	TSNAs ^e	PAA ^f	PAH ^g	Sum
Marlboro Gold Box	2967	211	22	157	<0.026	0.0604	0.00264	0.244	<3357 µg
L&B Original	2683	230	32	124	<0.024	0.0290	0.00232	0.267	<3069
L&B Menthol	2778	251	24	130	<0.042	0.0257	0.00236	0.213	<3183
blu Classic Tobacco Disposable	<1.0	<0.7	<0.01	<0.01	<0.0004	<0.0002	<0.000004	<0.002	<1.7
blu Magnificent Menthol Disposable	<1.0	<0.8	<0.01	<0.01	<0.0004	<0.0002	<0.000004	<0.002	<1.8
blu Cherry Crush High Premium	<1.0	<0.5	<0.03	<0.004	<0.0004	<0.0002	<0.000004	<0.001	<1.5
SKYCIG Classic Tobacco Bold	<1.0	<0.6	<0.01	<0.08	<0.0006	<0.0001	<0.00014	<0.0004	<1.7
SKYCIG Crown Menthol Bold	<1.0	<0.9	<0.01	<0.08	<0.0006	<0.0003	<0.00014	<0.0004	<2.0
Air Blank (blu Set)	<1.0	<0.6	<0.01	<0.004	<0.0004	<0.0002	<0.000004	<0.002	<1.6
Air Blank (SKYCIG Set)	<1.0	<0.5	<0.01	<0.08	<0.0006	<0.0001	<0.00014	<0.001	<1.6

< Indicates some or all values were below method limits of quantitation or detection, number of replicates = 3–5.

^a Formaldehyde, acetaldehyde, acrolein propionaldehyde, crotonaldehyde, MEK, butyraldehyde.

^b Hydroquinone, resorcinol, catechol, phenol, m-+p-cresol, o-cresol.

^c 1,3-Butadiene, isoprene, acrylonitrile, benzene, toluene, styrene.

^d Beryllium, cadmium, chromium, cobalt, lead, manganese, mercury, nickel, selenium, tin.

^e NNN, NAT, NAB, NNK.

^f 1-Aminonaphthalene, 2-aminonaphthalene, 3-aminobiphenyl, 4-aminobiphenyl.

^g Naphthalene, acenaphthylene, acenaphthene, fluorine, phenanthrene, anthracene, fluoranthene, pyrene, benzanthracene, chrysene, benzo(b)fluoranthene, benzo(k)fluoranthene, B(a)P, indeno[1,2,3-cd]pyrene, benzo(g,h,i)perylene.

Table 6

Per puff comparisons of quantifiable analytes for blu eCigs products from CAN puffing – yields and ratios to conventional product yields.

	Marlboro Gold Box µg/puff	blu MMD µg/puff	MGB/blu MMD
Acrolein	16.4 ± 0.2	0.19 ± 0.06	86
Phenol	1.53 ± 0.16	0.0017 ^a	900

^a Fewer than three replicates were quantifiable; no standard deviation is listed.**Table 7**

Per puff comparisons of quantifiable analytes for SKYCIG products from CAN puffing – yields and ratios to conventional product yields.

	L&B average µg/puff	SKYCIG CTB µg/puff	SKYCIG CMB µg/puff	L&B average/SKYCIG CTB	L&B average/SKYCIG CMB
Acetaldehyde	174	–	0.32 ^a	–	544
Acrolein	17	0.15 ± 0.02	–	113	–
Propionaldehyde	12	–	0.11 ± 0.05	–	109
N-Nitrosoanatabine	0.010	–	0.0002 ± 0.0001	–	50

^a Fewer than three replicates were quantifiable; no standard deviation is listed.

All analytes tested were present in the cigarette smoke at quantifiable levels except for select metals. These results are consistent with internal historical results for commercial cigarettes tested under the CAN smoking regime. For the cigarette samples, the total yield range was 3069–3350 µg/puff of HPHCs tested.

Of the 55 HPHCs tested in aerosol, 5 were quantifiable in an e-cigarette sample but not the associated air blank. The quantifiable results for aerosol are listed in Tables 6 and 7 in contrast with the conventional cigarettes from the same geographical region. The five analytes which were quantifiable were statistically different ($p < 0.05$) at levels 50–900 times lower than the cigarette smoke samples. Phenol was quantified in one e-cigarette product at 900 times lower than cigarette smoke. N-Nitrosoanatabine was quantified in one product at 50 times lower than cigarette smoke. Three carbonyls (acrolein, acetaldehyde, and propionaldehyde) were quantified at 86–544 times lower than cigarette smoke.

All other analytes were not quantifiable above the air blanks in aerosol samples. The e-cigarettes and air blanks total yields for analytes were <2 µg/puff which is 99% less than the approximately 3000 µg/puff quantified for the cigarette smoke samples. Thus, the results support the premise of potentially reduced exposure to HPHCs for the e-cigarette products compared to conventional cigarette smoke.

4. Conclusions

The purpose of this study was to determine content and delivery of e-cigarette ingredients and to compare e-cigarette aerosol to conventional cigarettes with respect to select HPHCs for which conventional cigarette smoke is routinely tested. Routine analytical methods were adapted and verified for e-cigarette testing. Aerosol collection was conducted using conventional smoking machines and an intense puffing regime. As machine puffing cannot, and is not intended to, mimic human puffing, results of this study are limited to the scope of the comparisons made between the e-cigarette and conventional cigarette products tested.

The main ingredients for the e-cigarettes tested were consistent with disclosed ingredients: glycerin and/or propylene glycol ($\geq 75\%$), water ($\leq 18\%$), and nicotine ($\sim 2\%$). Machine-puffing of these products under a standardized intense regime indicated a direct transfer of these ingredients to the aerosol while maintaining an aerosol composition similar to the e-liquid. Nicotine yields to the aerosol were approximately 30 µg/puff or less for the e-cig-

arette samples and were 85% lower than the approximately 200 µg/puff from the conventional cigarettes tested.

Testing of the e-cigarette aerosol indicates little or no detectable levels of the HPHC constituents tested. Overall the cigarettes yielded approximately 3000 µg/puff of the HPHCs tested while the e-cigarettes and the air blanks yielded <2 µg. Small but measurable quantities of 5 of the 55 HPHCs tested were found in three of the e-cigarette aerosol samples at 50–900 times lower levels than measurable in the cigarette smoke samples. Overall, the deliveries of HPHCs tested for the e-cigarette products tested were more like the study air blanks than the deliveries for the conventional cigarettes tested. Though products tested, collection parameters, and analytical methods are not in common between this study and others, the results are very consistent. Researchers have reported that most or all of the HPHCs tested were not detected or were at trace levels. [Burstyn \(2014\)](#) used data from approximately 50 studies to estimate e-cigarette exposures compared to workplace threshold limit values (TLV) based on 150 puffs taken over 8 h. The vast majority of the analytes were estimated as $\ll 1\%$ of TLV and select carbonyls were estimated as <5% of TLV. [Cheng \(2014\)](#) reviewed 29 publications reporting no to very low levels of select HPHCs relative to combustible cigarettes, while noting that some of the tested products exhibited considerable variability in their composition and yield. [Goniewicz et al. \(2014\)](#) tested a range of commercial products and reported quantifiable levels for select HPHCs in e-cigarette aerosols at 9- to 450-fold lower levels than those in cigarette smoke that in some instances were on the order of levels determined for the study reference (a medicinal nicotine inhaler). [Laugesen \(2009\)](#) and [Theophilus et al. \(2014\)](#) have presented results for commercial e-cigarette product liquids and aerosols having no quantifiable levels of tested HPHCs, or extremely low levels of measurable constituents relative to cigarette smoke. Additionally, findings from several recent studies indicate that short-term use of e-cigarettes by adult smokers is generally well-tolerated, with significant adverse events reported relatively rarely ([Etter, 2010](#); [Polosa et al., 2011, 2014](#); [Caponnetto et al., 2013](#); [Dawkins and Corcoran, 2014](#); [Hajek et al., 2014](#)). Thus, the results obtained in the aforementioned studies and in the present work broadly support the potential for e-cigarette products to provide markedly reduced exposures to hazardous and potentially hazardous smoke constituents in smokers who use such products as an alternative to cigarettes.

Additional research related to e-cigarette aerosol characterization is warranted. For example, continued characterization of

major components and flavors is needed. Establishment of standardized puffing regimes and reference products would greatly aid sharing of knowledge between researchers. Continued methods' refinement may be necessary for improved accuracy for quantitation of analytes at the low levels determined in this study. To that end, it is critical that negative controls and steps to avoid sample contamination be included when characterizing e-cigarette aerosol since analytes are on the order of what has been measured in the background levels of a laboratory setting. Though researchers have reported quantification of select analytes, great care must be taken when interpreting results at such trace levels.

Conflicts of interest

The company for which the study authors work and the companies that manufacture the e-cigarettes tested for this study are owned by the same parent company.

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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.2014.10.010>.

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