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Headspace analysis for screening of volatile organic compound profiles of electronic juice bulk material

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Abstract

The use of electronic nicotine delivery systems continues to gain popularity, and there is concern for potential health risks from inhalation of aerosol and vapor produced by these devices. An analytical method was developed that provided quantitative and qualitative chemical information for characterizing the volatile constituents of bulk electronic cigarette liquids (e-liquids) using a static headspace technique. Volatile organic compounds (VOCs) were screened from a convenience sample of 146 e-liquids by equilibrating 1 g of each e-liquid in amber vials for 24 h at room temperature. Headspace was transferred to an evacuated canister and quantitatively analyzed for 20 VOCs as well as tentatively identified compounds using a preconcentrator/gas chromatography/mass spectrometer system. The e-liquids were classified into flavor categories including brown, fruit, hybrid dairy, menthol, mint, none, tobacco, and other. 2,3-Butanedione was found at the highest concentration in brown flavor types, but was also found in fruit, hybrid dairy, and menthol flavor types. Benzene was observed at concentrations that are concerning given the carcinogenicity ofthis compound (max 1.6 ppm in a fruit flavortype). The proposed headspace analysis technique coupled with partition coefficients allows for a rapid and sensitive prediction of the volatile content in the liquid. The technique does not require onerous sample preparation, dilution with organic solvents, or sampling at elevated temperatures. Static headspace screening of e-liquids allows for the identification of volatile chemical constituents which is critical for identifying and controlling emission of potentially hazardous constituents in the workplace.

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Compliance with ethical standards

This study did not involve research on human participants or animals.

Conflict of interest The authors declare that they have no conflict of interest.

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Keywords

Volatile organic compounds; Headspace; Gas chromatography mass spectrometry; Electronic cigarette liquids; Partition coefficients

Introduction

Electronic nicotine delivery systems (ENDS) such as electronic cigarettes (e-cigarettes) are battery-powered devices that heat a liquid (also called an e-liquid or e-juice) without combustion to create an aerosol and vapor which the user inhales. The major ingredients of e-juices are propylene glycol (PG) and vegetable glycerin (VG) with water and nicotine in smaller proportions. In addition, e-juices contain numerous minor ingredients such as flavoring chemicals [1–3] as well as trace amounts of ingredients and contaminants from the manufacturing process [4].While use of e-cigarettes continues to gain popularity [5], there is concern for potential health risks from inhalation of aerosol and vapor produced by these devices [6, 7]. For example, Reidel et al. [8] reported that use of e-cigarettes is associated with alterations in the innate defense proteins in airway secretions. Constituents of concern in inhaled aerosol and vapor may include (1) those of the e-liquid itself such as PG, VG, and nicotine [9]; (2) contaminants that are released from the e-cigarette device into the e-juice such as metals [10]; and (3) reaction and thermal breakdown products of e-juice constituents, such as carbonyls [11]. These vapor constituents would depend on the combination of e-juice and e-cigarette device (construction materials, operating voltage, etc.) as well as vaping patterns and are therefore very complex and require considerable resources to characterize. In contrast to the vapor, the composition of e-juices is relatively less complex and characterization of their composition is therefore useful to screen for constituents of concern and correlate to aerosol measurements [12, 13]. The screening can also be used to monitor e-liquid formulation manufacturing processes for quality and to protect manufacturing and service workers from hazardous volatile constituents that may evaporate during production or handling.

From a health perspective, chemicals of concern that have been identified in e-juices include flavoring chemicals, aromatics, nicotine (and its alkaloids) and tobacco-specific nitrosamines (TSNAs), carbonyls, volatile organic compounds (VOCs), glycols, and metals [13]. As noted above, these chemicals may be present in e-juices as ingredients or contaminants from low-quality ingredients. A recent study found a total of 15,586 distinct flavors sold by websites - more than double the 7764 flavor labels found in 2013–2014 [14]. These flavored e-liquids may attract youth and young adults who would not otherwise choose to start using tobacco products [15, 16]. Given the rate at which new flavors of e-juices are entering the market, it is clear that detailed characterization of each product is not possible. Therefore, easy, rapid, and sensitive methods to screen e-juices for constituents of concern are necessary.

Famele et al. [13] have reviewed the appropriateness of various analytical techniques for specific classes of chemicals in e-juices and ENDS vapor (e.g., use of high-pressure liquid chromatography (HPLC) analysis for nicotine). Our focus is on methods for understanding

the volatile chemical composition of e-juices. Methods for characterization of metals are beyond the scope of this article and the reader is referred to other reports for more information on that topic [10, 17, 18]. Additionally, aerosol constituents are beyond the scope of this article and the reader is referred to other literature for more details on analytical methods for characterization of aerosols [11, 19–26]. As summarized in Table 1, researchers are still working to optimize analytical methods for characterization of e-juices. Many analytical methods employed to date involve detailed and time-consuming sample preparation, use solvents that are not consistent with the need for “green chemistry” approaches, or require specialized instrumentation such as a nuclear magnetic resonance spectrometer. In contrast, a headspace sampling technique based on vapor pressures favors volatile components, is insensitive to PG and VG matrix effects [12, 18], does not require derivatization, and is easier and less time-consuming than many other methods, making it ideal for identifying and quantifying volatile constituents evolved from the bulk e-juice [29, 30]. To help fill this knowledge gap, we developed a simple (minimal sample preparation), solvent-free, quantitative screening tool for VOCs based on headspace analysis of bulk e-juices by gas chromatography-mass spectrometry (GC-MS) and results are reported herein for 20 VOC constituents including the following classes of compounds: aldehydes, alcohols, ketones, aromatics, alkanes, and alkenes.

Methods

A convenience sample of 146 e-juices obtained from vendors across the USAwas analyzed for VOCs present in the headspace above the liquid. Sample nicotine contents were no nicotine (*n*= 34), 3 mg/mL (*n*=20), 6 mg/mL (*n*=23), 12 mg/mL (*n*=39), 18 mg/mL (*n*=15), and 24 mg/mL (*n*=15). E-juices were categorized into flavor types brown (e.g., caramel, butterscotch, maple, coffee), fruit, hybrid dairy (e.g., butter pecan, strawberry cream, vanilla crème, chai), menthol, mint, none (no flavoring), tobacco, and Other. These flavor types were modified from a list of flavors that may contain diacetyl (2,3-butanedione) put forth by the Occupational Safety and Health Administration [40]. A similar classification scheme was also used in a flavoring chemical analysis of e-cigarette aerosol [2]. Samples were stored at room temperature (22ºC) prior to analysis. Headspace analysis was performed by placing approximately 1 g of e-juice in a preweighed 40 cm^3 amber volatile organic analysis (VOA) vial (actual volume approximately 42 cm^3). The vial plus e-juice was weighed again to determine mass of e-juice. The sample was allowed to equilibrate for 24 h at room temperature (23 °C). Then, 2 cm^3 of headspace was transferred using a 2.5 cm^3 gas-tight syringe to a 450 cm^3 fused-silica-lined evacuated canister. The canister was pressurized with ultra-high purity nitrogen to approximately 1.5 times atmospheric pressure equating to a dilution factor of 338. Minimum detectable concentrations (MDCs) in the vial headspace ranged from 98 ppb for methylene chloride (approximately 14 ng in the headspace of the vial) to 275 ppb for *d*-limonene (approximately 64 ng in the headspace of the vial) (Table 2). A 250 cm^3 gas aliquot was analyzed using a 7200/7032 preconcentrator/autosampler (Entech Instruments, Simi Valley, CA) attached to a 7890/5977 gas chromatograph/mass spectrometer (Agilent, Santa Clara, CA) with a Restek Rxi-1ms column 60 m×0.32 mm ×1 μm (Bellefonte, PA).

Twenty different analytes were quantified using internal standard relative responses. The quantified compounds were acetaldehyde[*,†], ethanol[‡], acetone[‡], 2-propanol, hexane, methylene chloride, chloroform, 2,3-butanedione (diacetyl)[†], 2,3-pentanedione[†], 2,3-hexanedione[†], methyl methacrylate[‡], benzene*, toluene*, ethylbenzene, *m,p*-xylene, *o*-xylene, acetonitrile, styrene, α-pinene, and *d*-limonene. Compounds denoted with an asterisk (*) are on the FDA abbreviated list of harmful and potentially harmful constituents for currently regulated tobacco products and are of interest for e-cigarettes [28]. Compounds denoted with a dagger (†) are high priority substances and compounds denoted with a double dagger (‡) are low priority substances on the Flavor and Extract Manufacturers Association (FEMA) list of substances that "may pose potential respiratory hazards when improperly handled" [41]. FEMA categorizes styrene as an "other flavoring substance" (i.e., not on the high or low priority list). The internal standards were bromochloromethane, 1,4difluorobenzene, and chlorobenzene-d5 at 25 ppb with 50 cm^3 added to the preconcentrator trap prior to gas aliquot transfer. Samples were screened for tentatively identified compounds using library search reports against the NIST11 mass spectral database (NIST, Gaithersburg, MD). A match confidence indicator, called a quality factor, of 75 or greater was reported. Data were analyzed using JMP 12 and SAS 9.3 (SAS Institute, Cary, NC). A standard least squares model in JMP was used to compare group means of measured headspace concentration for each analyte for nicotine content and flavor type groups. Four e-juices were diluted in 95% ethanol:5% water and analyzed by GC-MS to determine the 2,3-butanedionecontent for comparisonagainst calculatedcontent based on a theoretical partition coefficient.

Results

Ethanol was the most prevalent quantified compound in ejuice headspace at 95% (Table 2). Acetaldehyde was the second most frequently identified compound at 61% followed by *d*-limonene at 54%. 2,3-Butanedione was identified in 46% of e-juices tested and 2,3-pentanedione was identified in 19% of e-juices tested. α-Pinene was identified in 38% of e-liquids tested.

OSHA developed a categorization framework for flavoring formulations that may contain 2,3-butanedione which was adapted for e-liquids to produce the following list: brown, fruit, hybrid dairy, menthol, mint, none, tobacco, and other [40]. Using this framework, the frequencies of quantified compounds varied by flavor type (Table 3). Note that the frequencies of quantified compounds depend upon the number of e-liquids evaluated and this denominator varied by flavor type. The nicotine content was also variable by flavor type because this was a convenience sample of e-liquids, some of which were purchased based on staff and customer usage during site visit investigations of vape shops in California while others were purchased with varying levels of nicotine content for a given e-liquid sample. Even when stratified by flavor type, ethanol was still the most frequently identified compound within a given flavor type. The frequencies of other chemicals were more widely varied. For example, 2,3butanedione was more prevalent in brown (70%), fruit (44%), and hybrid dairy (64%) flavor types.

Nicotine content and flavor type group means of measured headspace concentration were statistically different depending on analyte. Nicotine content group means were statistically different for 2,3-butanedione ($p = 0.03$), 2,3-pentanedione ($p = 0.03$), and methylene chloride ($p = 0.02$) due to large group means (e.g., 4 ppm least square mean for 2,3pentanedione at 3 mg/mL nicotine content) or small group means (e.g., 0.06 ppm least square mean for 2,3-butanedione at no nicotine content) for a particular nicotine content compared to the other groups. Nicotine content group means were not statistically different ($p = 0.73$) when analyte was included in the model asa fixed effect, meaning nicotinecontent did not likely affect measured headspace concentration of volatile organic compounds. Flavor type only had a statistically significant effect for 2,3-butanedione ($p \leq 0.01$), 2,3-pentanedione ($p = 0.04$), α-pinene ($p = 0.02$), and ethanol ($p \leq 0.01$).

Among all quantified compounds, ethanol was present at the highest concentration in the headspace of most samples and exceeded 20,000 ppm in a single sample (Fig. 1a). At this concentration, ethanol is 29% of the saturation concentration (69,000 ppm at 23 °C using a vapor pressure of 52.5 mmHg calculated from Antoine equation). *d*-Limonene was the next highest with a concentration that exceeded 25 ppm in a single measured e-liquid (Fig. 1b). Looking at specific categories of flavor types, 2,3-butanedione was found at the highest concentration in brown flavor types, but also had considerable concentrations in fruit, hybrid dairy, and menthol flavor types (Fig. 1c). Both 2,3-butanedione and its common substitute 2,3-pentanedione (Fig. 1d) were measured with high frequency in brown flavor types. 2,3-Butanedione was also measured in the none flavor type in two samples at 291 and 246 ppb. While these concentrations were above the MDC, they were below the minimum quantifiable concentration (i.e., 3.33 times MDC) which is an inherently more variable region of quantitation. Benzene was found at concentrations that are concerning given the carcinogenicity of this compound (max 1.6 ppm in a fruit flavor type) (Fig. 1e). Benzene was also found in most flavor types except for the none flavor type. Acetaldehyde had a max concentration of 160 ppm in a fruit flavor type (Fig. 1f). Box plots of headspace concentrations for the remaining 14 target compounds are presented in Figures S1-S7 (Supplementary Material).

In addition to the quantified compounds,many other VOCs were identified in the samples. Comparison of sample mass spectra with the NIST11 mass spectral library produced 123 tentatively identified compounds. When ranked by decreasing frequency of identification, 32 observations have a frequency greater than or equal to 4%; this is approximately the upper quartile of the frequency distribution (Table 4). Ethyl acetate (78%) and ethyl butanoate (62%) were the most frequently identified compounds during screening. Ethyl propionate was found in 40% of samples and has a fruity smell. Benzaldehyde was found in 12% of samples and has an almond odor. Flavor typeaffectedthe composition oftentatively identified compounds (Table S1). For fruit flavor type, ethyl butanoate was identified in 88% of samples and ethyl acetate was present in 86% of samples. For mint flavor type, the most commonly identified compounds were terpenes: γ-terpinene (86%) and β-pinene (71%).

Discussion

Ethanol may be present in residual amounts from solvent extraction of plant material [42] to create extracts (aka concrete) or preparation of absolutes (an alcohol-soluble volatile concentrate) from concretes or as a dilution agent for the flavoring additives. Ethanol has been frequently identified in e-liquids [29, 43, 44]. Acetaldehyde is used as a flavoring agent [45]. Acetaldehyde has been identified as an oxidation product when propylene glycol is heated [46]. Acetaldehyde is not likely formed during headspace sample preparation as the technique is conducted at room temperature. Identification and quantification of acetaldehyde from the bulk e-liquid material before heating is an important finding given its carcinogenic potential.

Terpenes, including α-pinene and d-limonene, react with ambient ozone to produce oxidation products. This may occur during e-liquid production or steeping (the process of aging the e-liquid for hours to days sealed or open to allow contact with air to develop the flavor profile). Rohr et al. [47] showed upper airway irritation and airflow limitation caused by d-limonene and α-pinene oxidation products in mice; these adverse effects were not immediately reversible. Benzene may be present due to chemical reactions and transformations from other constituents including benzoic acid [48] and due to impurities in bulk material used in e-liquid compounding. Ethylbenzene is a high priority substance and ethyl acetate, isobutyl acetate, and isoamyl acetate are low priority substances on the FEMA list of substances that “may pose potential respiratory hazards when improperly handled” [41]. FEMA categorizes 2-heptanone as an “other flavoring substance.” None of the compounds listed in Table 4 are on the FDA abbreviated list of harmful and potentially harmful constituents for currently regulated tobacco products that are of interest for e-cigarettes [28]. Ethyl acetate has a sweet fruity aroma and is used as a flavor enhancer in fruit flavors such as peaches, pineapple, bananas, and strawberries. Ethyl acetate may also be present in the e-liquid as a residual solvent from extraction of plant material [42]. Ethyl butanoate also has a sweet smell commonly used in pineapple flavors. Benzaldehyde has also been found by other researchers in cherry-flavored e-liquid and tobacco products [1, 49].

A debilitating respiratory disease, obliterative bronchiolitis, may be caused by exposure to 2,3-butanedione and 2,3-pentanedione [50]. Note that 2,3-pentanedione is a poor choice as a substitute because it has similar lung toxicity to 2,3-butanedione [50]. The frequency of 2,3butanedione in e-liquids observed here (46%) is low compared to 76.5% observed by Allen et al. [2] from aerosolized e-liquids and to 69.2% observed by Farsalinos et al. [51]. This discrepancy is not surprising since the studies were conducted on convenience samples and the flavor type categories were different. In our study, sample sizes for the flavor types may have been smaller due to the addition of mint, none, tobacco, and other.

An added value to the use of a simple static headspace concentration screening strategy is that measurement data may be used to rapidly estimate the chemical composition of volatile constituents in the liquid. Partition coefficients are required to convert the concentration in the gas to the concentration in the liquid; however, these coefficients are currently unavailable for chemicals in e-liquid formulations. Partition coefficients can be estimated

from chemical properties: vapor pressure of the pure liquid, assumed activity coefficients, and average molecular weight of the e-liquid [52]. Pankow calculates the distribution of chemicals between the gas and particle phase of a liquid aerosol [52]. The partitioning of chemicals between gas and bulk liquid (not aerosolized) will follow the same relationship:

$$K_i = \frac{RT}{10^6 \overline{MW} \gamma_i p_i^o} \quad (1)$$

where K_i refers to the gas/liquid partition coefficient of chemical i ($m^3/\mu g$), R refers to the gas constant (8.2×10^{-5} m^3-atm/ mol-K); T refers to the temperature (K); $\overline{MW}$ refers to the mole-average molecular weight of liquid (g/mol); γ_i refers to the mole-fraction-scale activity coefficient of chemical *i* (dimensionless); and p^o_i refers to the vapor pressure of pure chemical *i* (atm). The activity coefficient is dependent on the temperature, the chemical, and the interactions of the chemical with the e-liquid matrix, which is dominated by PG and VG.

In theory, 2,3-butanedione should have an activity coefficient of 1 since the vapor pressure is high, relative to PG and VG and the solution is an ideal dilute solution (usually < 0.1%), meaning the probability of intermolecular interactions between 2,3-butanedione and other chemicals is low (i.e., most of the interactions are presumably between 2,3butanedione and the e-liquid matrix). For 2,3-butanedione, K_i is calculated as 4.15×10^{-9} using the following assumptions: an activity coefficient of 1.0, a vapor pressure of 0.069 atm, and a mole-average molecular weight of 84 g/ mol for 50:50 PG:VG with minor other constituents (nicotine, flavoring chemicals, etc.). A 50:50 PG:VG mix was chosen based on Pankow [52] but the average molecular weight will only change no more than ±10% from 100:0 to 0:100. Therefore, the estimate of K_i will change by no more than ± 10% depending on the humectant composition of PG:VG. Applying this K_i to our data set shows a range of 2,3butanedione content from zero to 106 μg 2,3-butanedione/g e-liquid (upper quartile 2.9 μg/g), which equates to zero to 0.0106% by weight (upper quartile 0.00029%). The ratios of measured theoretical 2,3-butanedione content for the four ejuices diluted were 0.9, 1.1, 1.3, and 2.7. The discrepancies may be due to differences in humectant formulation (i.e., the ratio of PG to VG in the e-juice or water content), which can affect the activity coefficient and average MW used in the theoretical partition coefficient. In order to estimate the volatile compound content in solution, the humectant formulation (including water content) should be known so that average MW can be adjusted and the activity coefficient should be determined empirically. For ethanol, K_i is calculated as 4.9×10^{-9}. Fruit flavor types had the highest percent content by weight for ethanol in e-liquid (max 19%) using a calculated K_i of 4.9×10^{-9}. The max of 19% ethanol by weight (15% ethanol by volume = 19%* density of 0.789 g/mL) seems high, but within the range of alcohol content (23.5% alcohol (presumably ethanol by volume) to 0.4% alcohol) used by a researcher to look at the effects of inhaled alcohol from ecigarette use [53]. When a constituent increases considerably beyond 0.1% content in the e-liquid, as is the case with ethanol, the assumption of an ideal dilute solution begins to degrade and other mechanisms such as direct quantitative analysis of the solution should be considered.

The proposed headspace analysis technique coupled with partition coefficients may allow for a rapid and sensitive prediction of the content of volatile chemicals in the liquid. The technique does not require laborious sample preparation, dilution with organic solvents, or sampling at elevated temperatures. Other researchers have sampled VOCs from e-juice headspace at elevated temperatures (50 °C) using solidphase microextraction [27, 39], which has the benefit of concentrating analytes on the fiber prior to analysis but requires strict adherence to sample preparation and optimized extraction conditions. Barhdadi et al. [30] used a headspace autosampler that incubated and agitated the sample at 85 °C for 15 min prior to a gas transfer, but only looked at 2,3butanedione and 2,3-pentanedione and used dimethyl sulfoxide as a dilution solvent.

The headspace technique outlined in this paper could also allow for estimating emission potential due to open e-liquid containers or spills. Determination of the volatile constituents of bulk e-juice allows for a better understanding of the chemical emission potential due to evaporation. This information also provides an expected chemical profile when aerosolized during vaping due to direct emission, as oxidation products, or as toxic thermal degradation products (e.g., propylene glycol and glycerin can thermally degrade to formaldehyde, acetaldehyde, and acrolein). By analyzing the headspace with minimal sample preparation and at room temperature, which is a typical temperature of storage, the emitted volatile constituents assessed by this technique will realistically mimic those seen in the field. An advantage of this technique is that it does not require organic solvents.

Conclusion

To characterize bulk e-liquids, we developed a static headspace VOC screening technique which provides quantitative and qualitative chemical information without the use of organic solvents or onerous sample preparation. Static headspace screening of e-liquids allows for the rapid identification of volatile chemical constituents which is critical for identifying potentially hazardous constituents and targeting e-juice products for more detailed characterization. Additionally, the identified constituents can then be targeted for industrial hygiene air sampling in occupational environments or used as precursors or input material for modeling exposures due to vaping. Coupled with partition coefficients based on constituent physical properties and empirically derived activity coefficients, the headspace results may be used to estimate volatile constituent content in the bulk e-liquid. Suppliers of flavors used in e-liquids and manufacturers of e-liquids may want to quantitatively assess known hazardous components, like 2,3butanedione and 2,3-pentanedione, regardless of their presence or absence from safety data sheets.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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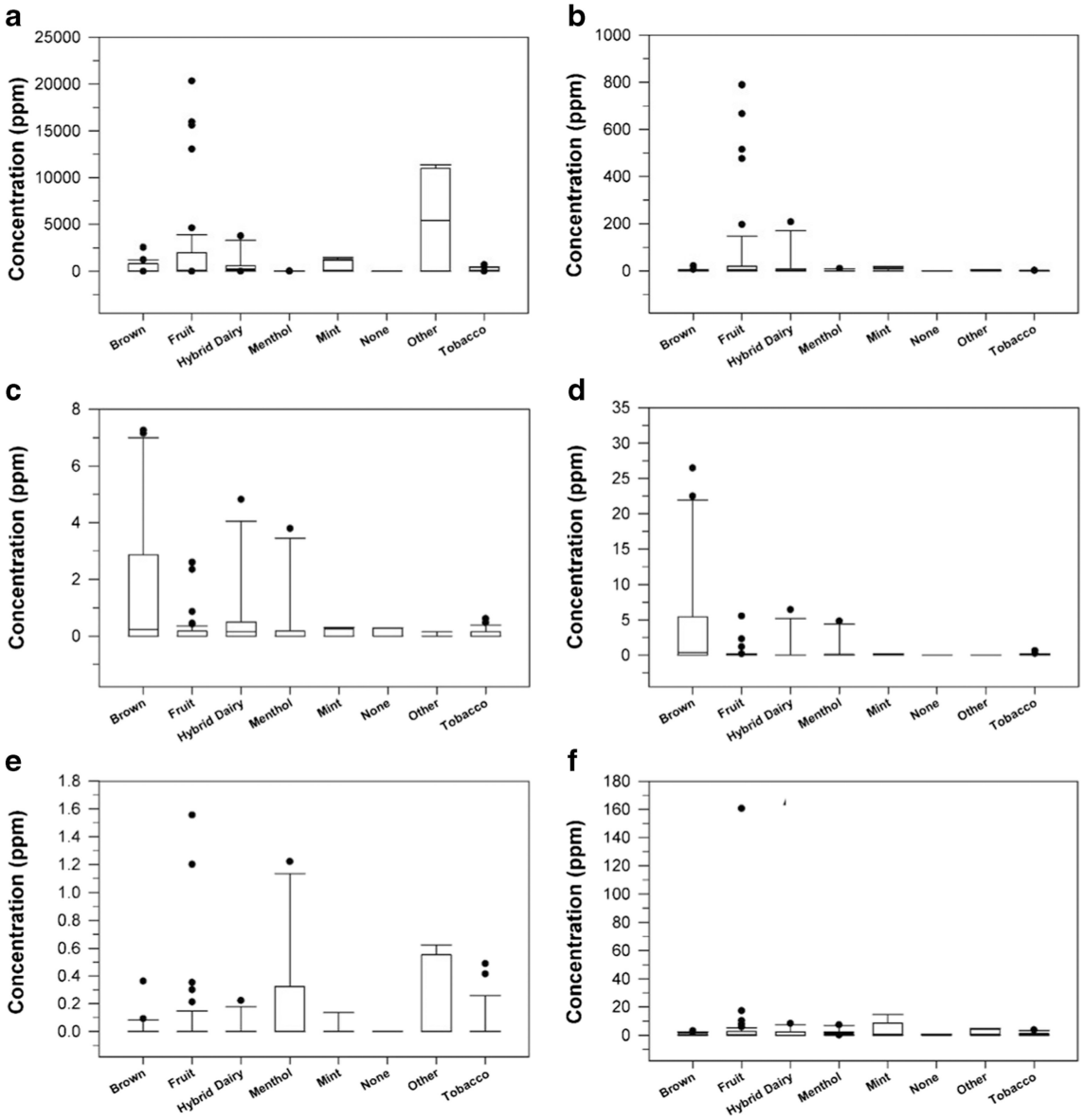

Fig. 1.

Box plots of headspace concentrations (ppm) for six representative compounds: ethanol (**a**), *d*-limonene (**b**), 2,3-butanedione (**c**), 2,3-pentanedione (**d**), benzene (**e**), and acetaldehyde (**f**)

Table 1

Summary of analytical methods development literature for e-juices

Analyte(s)[a]	Technique[b]	Comment	Reference
Aldehydes	HS-SPME-GC-MS	+ Sensitive, specific − Sample preparation and derivatization	[27]
Ammonia	IC-CD	+ Sensitive, specific − Sample preparation with acid	[28]
Carbonyls	HPLC	+ Sensitive, specific − Sample preparation and derivatization	[25]
Ethanol	GC-FID	+ Sensitive, specific − Limited to e-juices	[29]
Flavoring chemicals	HS-GC-MS	+ Minimal sample preparation − Limited to diacetyl and acetylpropionyl	[30]
Gly, Nic	NMR	+ Broad range of constituents − Limited commercial availability	[4]
Gly, Nic	NMR	+ Broad range of constituents − Limited commercial availability	[31]
Nic	HPLC-MS/MS	+ Sensitive, rapid − Limited to Nic	[32]
Nic, TSNAs, flavoring chemicals	LC-MS	+ Good precision and accuracy − Dissimilar internal standard chosen	[33]
Nic	GC-MS	+ Free and protonated forms − Limited to Nic	[34]
Nic	SERS	+ Portable method for field use − Matrix and detection limit issues	[35]
Nic, PG, VG	GC-FID	+ Sensitive, accurate − Limited to major constituents	[26]
Nic, alkaloids	LC-MS/MS	+ Sensitive, minimal sample preparation − Limited to Nic and alkaloids	[36]
Nic, alkaloids	UHPLC-MS	+ Good precision and accuracy − Sample preparation with solvents	[18]
PG, VG, PAHs	GC-MS	+ PAHs not expected or detected in bulk − No internal standard used	[33]
Ph, Gly, flavoring chemicals	GC-MS	+ Analyte content calculated − Sample preparation, mass/volume units	[37]
TSNAs	LC-MS/MS	+ Sensitive, specific − Limited range of constituents	[38]
TSNAs, Nic, alkaloids	LC-MS/MS	+ Isotopically labeled internal standards − Sample preparation with solvents	[28]
VOCs[c]	GC-MS	+ Broad range of constituents − Low recoveries for some VOCs	[12]
VOCs	GC-MS	+ Purge and Trap sensitivity − Misses very light volatiles	[18]
VOCs	HS-SPME-GC-MS	+ Sensitive, specific − Extensive optimization of SPME fibers	[39]

[a] *Nic*, nicotine; *Ph*, phthalates; *PAHs*, polycyclic aromatic hydrocarbons; *TSNA*, tobacco-specific nitrosamines; *VOCs*, volatile organic compounds; *Gly*, glycols

[b] *CD*, conductivity detector; *FID*, flame ionization detector; *GC*, gas chromatography; *HPLC*, high-pressure liquid chromatography; *HS*, headspace; *IC*, ion chromatography; *LC*, liquid chromatography; *MS*, mass spectrometry; *NMR*, nuclear magnetic resonance spectrometry; *SERS*, surface-enhanced Raman spectroscopy; *SPME*, solid-phase microextraction; *UHPLC*, ultra-high-pressure liquid chromatography

[c] VOCs refers to aldehydes, ketones, esters, alcohols, aromatics, carboxyls, and phenols

Table 2

Frequency and minimum detectable concentrations of quantified compounds (n = 146 e-juices

Compound	Number and frequency (%)	Minimum detectable concentrations (ppb)[a]
Ethanol	139 (95)	255
Acetaldehyde	89 (61)	106
d-Limonene	79 (54)	275
Isopropyl alcohol	75 (51)	189
Acetone	74 (51)	275
2,3-Butanedione	67 (46)	102
α-Pinene	56 (38)	161
2,3-Pentanedione	28 (19)	141
Benzene	20 (14)	102
m,p-Xylene	16 (11)	114
Toluene	13 (8.9)	126
o-Xylene	6 (4.1)	102
2,3-Hexanedione	4 (2.7)	251
Methylene chloride	4 (2.7)	98
Ethylbenzene	3 (2.1)	138
Methyl methacrylate	3 (2.1)	98
n-Hexane	2 (1.4)	169
Acetonitrile	1 (0.7)	134
Styrene	1 (0.7)	181
Chloroform	0 (0)	102

[a]In the headspace vial

Table 3

Top seven frequencies of quantified compounds for each flavor type

Compound	Flavor type							
	Brown *n* = 20	**Fruit *n* = 57**	**Hybrid dairy *n* = 11**	**Menthol *n* = 10**	**Mint *n* = 7**	**None *n* = 5**	**Tobacco *n* = 28**	**Other *n* = 7**
Acetaldehyde	13 (65%)	34 (60%)	5 (45%)	9 (90%)	5 (71%)	2 (40%)	17 (61%)	4 (57%)
Acetone	9 (45%)	27 (47%)	3 (27%)	9 (90%)	6 (86%)	3 (60%)	15 (54%)	2 (29%)
Benzene	–	–	–	–	–	–	5 (18%)	2 (29%)[b]
2,3-Butanedione	14 (70%)	25 (44%)	7 (64%)	3 (30%)[a]	3 (43%)	2 (40%)	11 (39%)	–
2,3-Pentanedione	11 (55%)	–	–	–	–	0 (0%)	–	–
Ethanol	20 (100%)	53 (93%)	10 (91%)	10 (100%)	7 (100%)	5 (100%)	27 (96%)	6 (86%)
Isopropyl alcohol	12 (60%)	27 (47%)	3 (27%)	8 (80%)	3 (43%)	4 (80%)	16 (57%)	
d-Limonene	6 (30%)	39 (68%)	7 (64%)	4 (40%)	7 (100%)	2 (40%)	10 (36%)	3 (43%)
α-Pinene	–	27 (47%)	7 (64%)	4 (40%)	6 (86%)	–	–	5 (71%)
Toluene	–	–	–	–	–	–	–	3 (43%)

[a] Benzene and toluene also quantified at the same frequency

[b] Isopropyl alcohol and *m,p*-xylene also quantified in 2 of 7 samples or 29%

Table 4

Approximate upper quartile for frequency distribution of identified compounds (n = 146 e-juices)

Identified compound	CAS no.	Number and frequency (%)
Ethyl acetate[b]	141–78-6	114 (78)
Ethyl butanoate	105–54-4	91 (62)
Ethyl propionate	105–37-3	58 (40)
Ethyl 2-methylbutanoate	7452–79-1	53 (36)
Isoamyl acetate[b]	123–92-2	41 (28)
β-Pinene	127–91-3	34 (23)
2-Methylbutyl acetate	624–41-9	29 (20)
Isopentyl isovalerate	659–70-1	28 (19)
Isobutyl acetate[b]	110–19-0	26 (18)
p-Cymene	99–87-6	26 (18)
Ethyl 3-methylbutanoate	108–64-5	23 (16)
γ-Terpinene	99–85-4	23 (16)
1,3-Dioxolane, 2,2,4-trimethyl	1193–11-9	22 (15)
Ethyl hexanoate	123–66-0	19 (13)
Benzaldehyde[a]	100–52-7	18 (12)
Hexyl acetate	142–92-7	15 (10)
Isobutyl isovalerate	589–59-3	15 (10)
Pentyl acetate	628–63-7	13 (9)
Cyclohexanone, 5-methyl-2-(1-methylethyl)-, (2R-*cis*)	1196–31-2	12 (8)
Camphene	79–92-5	12 (8)
β-Ocimene	13877–91-3	10 (7)
α-Thujene	2867–05-2	9 (6)
1,1-Diethoxyethane	105–57-7	7 (5)
Terpinolene	586–62-9	7 (5)
4-Hexen-1-ol, acetate	7223736–6	7 (5)
(Z)-Ocimene	3338–55-4	7 (5)
2-Heptanone	110–43-0	7 (5)
Butyl isovalerate	109–19-3	7 (5)
Methylcyclopentane	96–37-7	7 (5)
Phellandral	21391–98-0	6 (4)
1,8-Cineole	470–82-6	6 (4)

[a]High priority substance or compound on the Flavor and Extract Manufacturers Association (FEMA) list of substances that “may pose potential respiratory hazards when improperly handled” (FEMA, 2012)

[b]Low priority substances on the FEMA list