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Chemical Transformation of Vaping Emissions under Indoor Atmospheric Aging Processes

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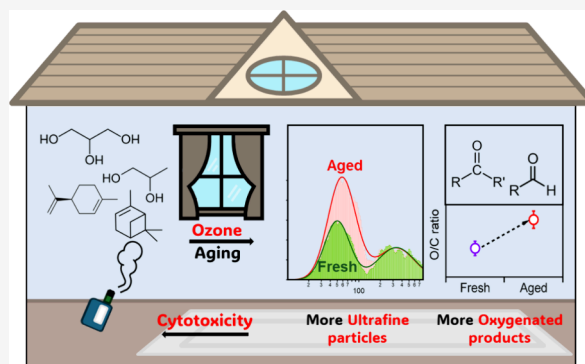


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ABSTRACT: E-cigarette emissions, which contain a variety of hazardous compounds, contribute significantly to indoor air pollution and raise concerns about secondhand exposure to vaping byproducts. Compared to fresh vape emissions, our understanding of chemically aged products in indoor environments remains incomplete. Terpenes are commonly used as flavoring agents in e-liquids, which have the ability to react with the dominant indoor oxidant ozone (O_3) to produce reactive oxygenated byproducts and result in new particle formation. In this study, mixtures of propylene glycol (PG), vegetable glycerin (VG), and terpenes as e-liquids were injected into a 2 m³ FEP chamber to simulate the indoor aging process. 100 ppbv O_3 was introduced into the chamber and allowed to react with the fresh vape emissions for 1 h. Complementary online and offline analytical techniques were used to



characterize the changes in the aerosol size distribution and chemical composition during the aging processes. We observed more ultrafine particles and a greater abundance of highly oxygenated species, such as carbonyls, in aged e-cigarette aerosols. Compared with their fresh counterparts, the aged emissions exhibited greater cytotoxic potential, which can be attributed to the formation of these highly oxygenated compounds that are not present in the fresh emissions. This work highlights the dynamic chemistry and toxicity of e-cigarette aerosols in the indoor environment as well as the indirect risks of secondhand exposure.

1. INTRODUCTION

The rise in cases of severe lung diseases and fatalities associated with their consumption has labeled vaping products, such as e-cigarettes (e-cigs), as a significant public health issue.^{1–3} The National Health Interview Survey data in 2021 revealed that 4.5% of adults aged 18 and over were actively using e-cigs.⁴ Given the increasing popularity of e-cigs and the absence of stringent emission regulations, these products are continuously developing and emerging as novel sources of indoor air contaminants. The vaping process transforms e-liquids into aerosols that users inhale, and the exhaled aerosols can persist in the surrounding atmosphere. Therefore, passive exposure to vaping aerosols may pose a significant threat to nonusers, particularly vulnerable populations.^{5,6}

The main components of e-liquids include PG, VG, nicotine, and different flavorings.^{7–10} Recently, terpene or terpene-like compounds have been commonly used in e-cig flavorings. Terpenes and terpene-like compounds are naturally abundant in hemp plants, which are widely found in Δ -tetrahydrocannabinol (THC) and bioactive cannabidiol (CBD) vaping products.^{11,12} In addition to cannabis vaping products, terpenes or terpenoids are also applied in e-juices and supplements to provide a variety of sensory experiences for users, such as α -pinene, limonene, geraniol, and linalool.^{11,13} Thermal degradation of these terpenes or terpenoids can generate many byproducts from dehydrogenation, epoxidation,

and allylic oxidation,¹⁴ effectively contributing to secondary organic aerosol (SOA) formation.^{15,16} In addition to thermal degradation, these terpenes or terpenoids typically have C=C double bonds or four-membered ring structures. These properties make them unstable and reactive to atmospheric oxidants such as O_3 ¹⁷ and OH radicals.^{18,19} Oxidation of terpenes is known to have a high yield of SOAs and may adversely impact indoor and outdoor air quality.

Ozonolysis is the most important pathway for the oxidation of terpenes in the atmosphere. Although the reaction rate between terpenes and OH radicals or NO_3 radicals is faster than ozonolysis,^{20,21} oxidation products formed from ozonolysis are produced in higher oxygenation degrees compared to OH or NO_3 -initiated compounds.²² Ozonolysis of limonene contributes 47% to its tropospheric degradation in daytime outdoor chemistry.²⁰ In indoor environments, O_3 is a major oxidant;²³ therefore, ozonolysis of terpenes may lead to greater particle formation and change their chemical composition

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during the aging process of vaping. Furthermore, ozonolysis-derived terpene products were identified as less volatile than OH-initiated compounds, contributing more to the SOA formation in the previous study.²⁴

Extensive research has been conducted on the chemical composition and potential health effects of active vaping emissions.¹⁷ A few recent studies indicate that the aging of e-cig aerosols can promote reactive oxygen species (ROS) generation.^{25–28} However, our understanding of the composition of aged e-cig aerosols remains incomplete. For example, how may the changes in the chemical characteristics of the fresh versus aged e-cig aerosols cause differential toxicity for active users versus bystanders? Given that O₃ is the primary oxidant in indoor environments, what are the major products from the reaction between O₃ and vaping emissions that are responsible for the elevated toxicity? Further, what are the main contributors to the aged aerosol formation: e-liquid solvents or flavorings? Addressing these questions will help to better understand the increasingly important secondary pollution from e-cigs in indoor environments and help assess the potential health effects of the passive vaping process.

In this study, we comparatively analyzed the chemical composition of freshly generated and chemically aged vaping emissions to assess the impacts of atmospheric processing on particle formation, including the particle size distribution, number, and mass concentrations, and their associated toxicological responses. We conducted a series of chamber experiments in the presence or absence of O₃ using various e-liquids. Several complementary online and offline measurements were carried out to gain a better understanding of the chemical composition and properties of e-cig aerosols. Overall, the findings of this study reveal that secondhand exposure to aged e-cig aerosols characterized with an increased oxygenation degree is associated with higher oxidative potential, which in turn poses certain health risks distinct from fresh vaping emissions.

2. METHODOLOGY

2.1. Materials. All analytical standards used in this study had purities greater than 95%. We purchased geraniol (99%) from Acros Organics, α -pinene (>99%) from Sigma-Aldrich, VG from Fisher Chemical, PG from TCI America, and methanol (99.9%) and acetonitrile (ACN, 99.95%) from Fisher Scientific. The commercial terpene-flavored e-liquids (GG#4) were purchased from Gold Coast, which consisted of α -pinene, α -terpinene, terpineol, limonene, β -pinene, 3-carene, linalool, terpinolene, ocimene, geraniol, β -caryophyllene, α -humulene, camphene, α -phellandrene, α -cedrene, cymene, myrcene, nerolidol, and pulegone. The e-cig device consisted of a battery (Silo, CCELL) that operated at 3.6 V and a 510-thread 0.5 mL glass silver cartridge (threaded white ceramic mouthpiece) and had a coil resistance of 1.4 Ω (CCELL).

2.2. Preparation of E-Liquids. In this study, a mixture of 30% PG and 70% VG was utilized as the diluent in our e-liquids. Three different types of e-liquids were prepared to conduct a comparative analysis, including (1) only PG/VG (30:70 in volume); (2) 3% terpene mixtures, specifically α -pinene or geraniol, dissolved in 30:70 PG/VG; and (3) a 3% mixture of GG#4 prepared by the same method (Table S1). Each cartridge was filled with 450 μ L of e-liquid and used only once to prevent potential coil material degradation affecting vaping emissions.

2.3. Chamber Experiments. All experiments were conducted in a 2 m³ fluorinated ethylene propylene (FEP) film chamber at room temperature (Figure S1A). Before each experiment, the chamber was flushed with zero air at a flow rate of 30 L min⁻¹ to ensure any potential contaminants were removed, and the cartridge was preconditioned by taking five puffs before e-cig aerosol generation. The chamber was filled with zero air before each fresh condition experiment and with 100 ppbv of O₃ in zero air before each aged condition experiment. O₃ was introduced into the chamber using an O₃ generator (A2Z Ozone 3GLAB) and stabilized at \sim 100 ppbv for 30 min before the reaction. The concentration of O₃ in the chamber was monitored in real time using an O₃ analyzer (Advanced Pollution Instrumentation, model 400A). Fresh vaping emissions, including gas- and particle-phase compounds, were injected into the chamber during each experiment. For the chamber experiments with filter collection, 30 puffs were injected into the chamber using an e-cig puffing machine to ensure sufficient mass loading (Figure S1B) (CSM-eSTEP, CH Technologies, Inc., Westwood, NJ, USA) with a positive pressure of 20 psi. An additional experiment with 5 puffs was performed to analyze particle size distribution and mass concentration using a scanning electrical mobility spectrometer to prevent overloading of analytical instruments, with further details provided in Section 2.4. The puff cycle followed the CORESTA recommended puffing topography with a puff period of 3 s, a puff interval of 30 s, and a puff volume of 55 mL during each cycle.²⁹

2.4. Aerosol Size and Volume Characterizations. A scanning electrical mobility spectrometer (SEMS, Brechtel Manufacturing Inc.) was used to monitor the particle size distributions and volume concentrations of e-cig aerosols in real time throughout the chamber experiments. The SEMS was set to scan from 10 to 800 nm, with 140 size bins. Aerosol mass concentrations were converted from the volume concentrations assuming a unity density.

2.5. Chemical Composition of Gaseous Vaping Emission Products. The molecular formula of gas-phase e-cig aerosol constituents was determined in real time using an online iodide-adduct time-of-flight chemical ion mass spectrometer coupled with a Filter Inlet for Gases and AEROSols system (FIGAERO-ToF-CIMS, Aerodyne Research Inc.). Reagent ions were generated by flowing a mixture of CH₃I and humidified N₂ through a polonium-210 ionizer (NRD; model P-2021).³⁰ For the collection of gaseous compounds, the air was directly sampled from the chamber at a flow rate of \sim 2.0 L min⁻¹. The iodide ion chemistry has been known to be affected by the water vapor pressure inside the IMR owing to the difference in thermodynamics between I⁻ and IH₂O⁻ to analyte compounds.³¹ Therefore, the addition of a continuous flow of humidified N₂ (1 L min⁻¹) through a bubbler mentioned above minimized changes in the water vapor pressure inside the IMR. In addition, all signals detected were normalized to the sum of reagent ions, such as I⁻, H₂OI⁻, and I₃⁻, to avoid changes in sensitivity influenced by reagent ions in the ion–molecule reaction (IMR) chamber across experiments. The data were analyzed using Tofware v3.2.5.

2.6. Chemical Composition of Oxygenated Compounds in E-Cigarette Aerosols. Polytetrafluoroethylene (PTFE) membrane filters (Zefluor, Pall Laboratory, 25 mm, 1.0 μ m pore size) were used to collect the aerosol samples at a flow rate of 10 L min⁻¹ for 15 min. The collected aerosol samples were stored at -20 °C until analysis. Upon analysis,

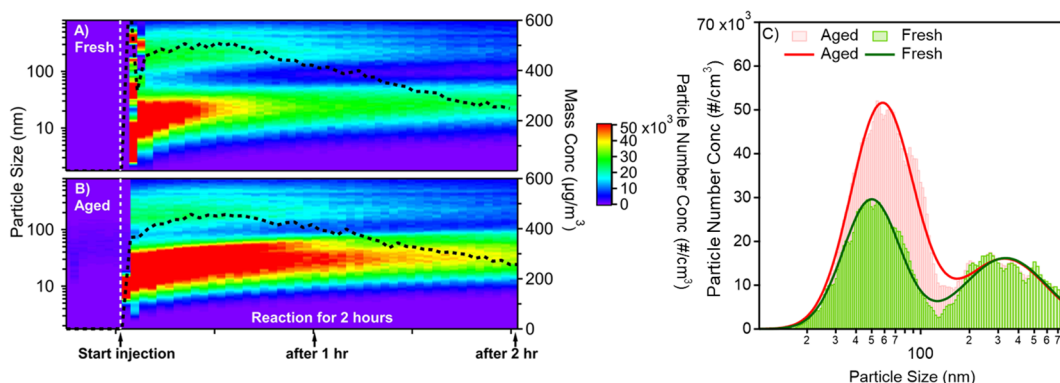


Figure 1. Aerosol growth and particle size distribution during the vaping process under different conditions. E-cig aerosol of PG/VG + GG#4 growth under (A) fresh and (B) aged conditions, with the black dashed line indicating the mass concentrations and the heat map indicating the number concentrations; (C) comparison of particle size distribution between fresh (green) and aged (red) e-cig aerosols (PG/VG + GG#4) measured 30 min after injection.

the filter samples were thermally desorbed on the FIGAERO inlet and directly analyzed by ToF-CIMS, following previous studies.³² The temperature program for FIGAERO thermal desorption was set as follows: (1) temperature ramping from room temperature (~ 25 °C) to 200 °C in 15 min (11.6 °C min^{-1}), (2) a 10 min soaking period (200 °C) to allow signals to return to background levels, and (3) a cooling period to decrease the temperature from 200 to 25 °C in 10 min. The data were analyzed using Tofware v3.2.5.

2.7. Characterization of Carbonyls in E-Cigarette Aerosols. 2,4-Dinitrophenylhydrazine (2,4-DNPH) was used to derivatize the vaping aerosols collected on filters to identify carbonyls. The 2,4-DNPH derivatization method used was modified from previously published studies.^{33,34} Vaping emissions (total 50 puffs) from both fresh and aged conditions were collected directly after injection onto 25 mm PTFE filters (Zefluor, Pall Laboratory, 1 μm pore size) at a flow rate of 20 L min^{-1} for 30 min. The filters were directly extracted with 1 mL of 6.5 mM 2,4-DNPH solution (acidified with HCl in acetonitrile) for 90 min at 50 °C. After that, samples were aliquoted into autosampler vials and stored at 4 °C prior to LC/ESI(-)-QToF-MS analysis, as described in the previous study and Supporting Information (SI, Text S1).³⁴

2.8. Cell Culture and Exposure. Human bronchial epithelial cells (BEAS-2B) were purchased from the American Type Culture Collection (ATCC). Cells were cultured in 75 cm^2 tissue culture flasks (Fisher Scientific) with LHC-9 medium (Gibco). Cells were incubated at 37 °C with 5% CO_2 . The growth media was replaced every 2–3 days, and the cells were subpassaged once 70–80% confluent. Once confluent, the cells were transferred onto 96-well plates (Corning) with a seeding density of 1×10^4 cells per well. The cells were allowed 24 h for attachment before vaping aerosol exposures. After 24 h, the cells were exposed to various concentrations of fresh and aged GG#4 aerosols. The aerosols were collected onto 47 mm PTFE filters at a flow rate of 20 LPM to obtain a mass loading of 10–12 mg. The filters were extracted by vortexing with cell media. Once exposed, the cells were incubated for 24 h at 37 °C and 5% CO_2 . Each condition had four replicates, and untreated cells were used as a negative control, while cells treated with 0.1% Triton X-100 acted as a positive control for downstream assays. After 24 h of incubation, the cell media was collected, and the cells were washed once with DPBS (Corning).

2.9. Cell Viability and Cytotoxicity Assays. Cell viability was determined by an XTT cell proliferation kit (Roche) according to the manufacturer's protocol. The absorbance was measured at 470 nm with a SpectraMax iD5 microplate reader. Cell viability was expressed relative to the negative and positive control, where the untreated group was assigned 100% cell viability, while the cells treated with 0.1% Triton X-100 were assigned 0% cell viability. Cytotoxicity was assessed by the amount of lactate dehydrogenase (LDH) release during exposure as an indicator of increased cell membrane permeability using an LDH cytotoxicity detection kit (Roche) according to the manufacturer's protocol. The absorbance was measured at 490 nm with a reference wavelength of 620 nm with a SpectraMax iD5 microplate reader. LDH release was expressed relative to the negative and positive control, where the untreated group was assigned 0% LDH release, while the cells treated with 0.1% Triton X-100 were assigned 100% LDH release.

3. RESULT AND DISCUSSION

3.1. Dynamic Characteristics of E-Cig Aerosols. Figure 1 shows the dynamic evolution of aerosols produced by five puffs of e-cigs. As depicted in Figure 1A, the total mass concentration of aerosols increases immediately following the vaping process, peaking at $500 \mu\text{g m}^{-3}$, and then gradually declining to a steady-state concentration of approximately $250 \mu\text{g m}^{-3}$. The time series of the average mass concentrations of e-cig particles (30 puffs with three replicates) is detailed in Figure S2 and Table S2. Analysis of the size distribution of fresh aerosol particles reveals their segregation into two distinct categories: one predominantly consisting of particles within the size range of 10–150 nm and another comprising larger particles ranging from 150 to 800 nm. The temporal analysis of the smaller particles indicates a maximum number concentration of 5×10^4 particles cm^{-3} , observed concomitantly with the aerosol generation. Following the generation of e-cig aerosols, a decline in the concentrations of small and large particles was observed, suggesting an absence of further particle formation. This phenomenon could be attributed to the absence of extra oxidants under fresh conditions, preventing the nascent aerosols from undergoing additional oxidation processes and relegating them to partitioning or condensation.³⁵

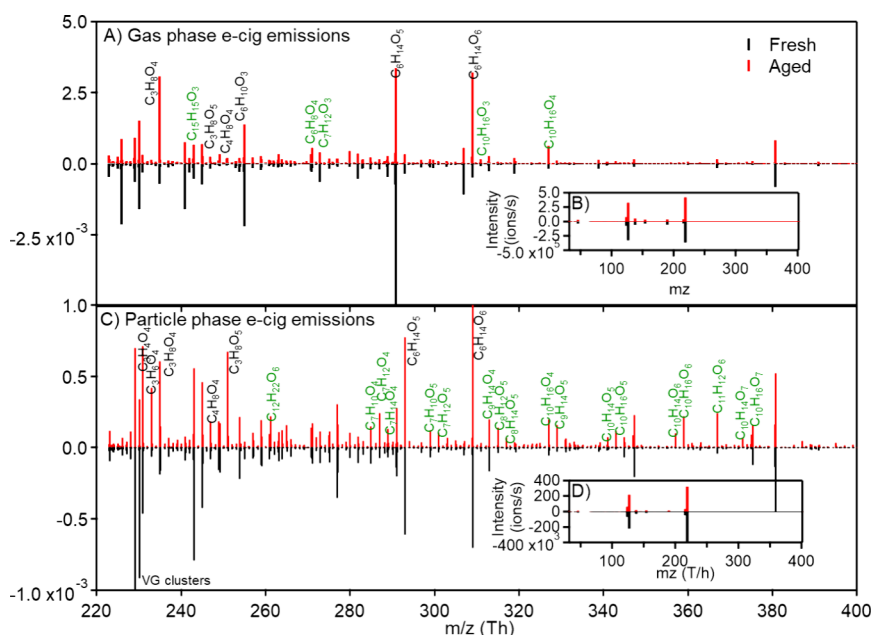


Figure 2. Difference in chemical composition of fresh and aged e-cig emissions: (A) signal of gas-phase e-cig emissions normalized to the intensity of VG ($C_3H_8O_3I^-$), (B) original CIMS intensity of gas-phase e-cig emissions, (C) signal of particle-phase e-cig emissions normalized to the intensity of VG signal ($C_3H_8O_3I^-$), and (D) original CIMS intensity of particle-phase e-cig emissions. Terpene-like oxidation products are labeled in green, while common compounds directly from PG/VG are shown in black.

Figure 1B shows the temporal evolution of aged e-cig aerosols, where the total mass concentration of large particles increases immediately after vaping and then decreases. This trend is similar to that of fresh e-cig aerosols, suggesting their primary formation occurs during vaping, with no further generation afterward. However, a notable difference is observed in the behavior of the smaller particles between the two aerosol types. After the injection, the small particles of aged aerosols sharply increased, and the number concentrations remained constant at 5×10^4 particles cm^{-3} within the next 2 h. Besides the difference in the temporal trend, we also observed varying size distributions of ultrafine particles (UFPs). For fresh aerosols, the mean diameter of UFPs was 45 nm, which increased to 60 nm for aged aerosols. This phenomenon was also observed in aged PG/VG + α -pinene geraniol and aged PG/VG + geraniol e-cig aerosols (Figures S3 and S4). The larger size and greater concentration of UFPs in aged aerosols indicate further oxidation in the presence of O_3 , which decreased around 30 ppbv during the aging processes (Figure S5). Through ozonolysis, the chemical transformation of e-cig aerosols is likely to happen.

We performed aging experiments with pure PG/VG aerosols as control experiments to investigate the effects of terpene flavor additives during the vaping process. Figure S6 shows the temporal evolution of aged PG/VG aerosols, which exhibit a total mass concentration comparable to that of aged commercial e-cig aerosols. However, in the presence of O_3 , the number concentrations of aged PG/VG aerosols decreased significantly after injection, particularly for small particles. In addition, the sizes of UFPs for pure PG/VG e-cig aerosols were similar under fresh and aged conditions (Figure S6B and S6c). This finding suggests that the oxidation of flavorings, specifically terpenes in this case, promoted the formation of UFPs. Previous studies have reported that the ozonolysis of terpenes can contribute to the formation of new particles and SOAs.^{36–38} Our study found that while the mass fraction of

terpenes in vaping emissions was much lower than that of PG/VG, the ozonolysis of terpenes was the main cause of the substantial rise in both the particle size and the number concentrations of UFPs. These results highlight the crucial role of terpenes in vaping emissions, their complex chemistry in the indoor environment, and the potential health effects of secondhand or third hand exposure.³⁹

3.2. Changes in Chemical Composition in the Gas Phase

The gas-phase emissions of the e-cigs were analyzed in real time using iodide-CIMS after being injected into the chamber. The gaseous emissions mainly consisted of volatile compounds with a relatively high vapor pressure. After 1 h, around 300 peaks were identified as oxidized products resulting from the aging process of the commercial e-liquid (GG#4) (Figure 2A,B). Compared with their aged counterparts, the number and intensity of identified peaks in fresh emissions were significantly lower. This is due to the absence of extra oxidants under fresh conditions compared to the aging experiments, which in turn restricts further oxidation. However, iodide-CIMS can only detect substances with a relatively high degree of oxygenation.³¹ Strong signals of $C_3H_8O_2$ and $C_3H_8O_3$, as well as $C_6H_{14}O_5$ and $C_6H_{14}O_6$, are present in both fresh and aged e-cig emissions, which primarily originate from unreacted PG/VG, the dimer products of PG/VG, and the cluster of PG/VG, respectively.⁴⁰ This can be attributed to the high PG/VG ratio in the e-liquids, which is approximately 30 times greater than that of other flavorings. Some oxygenated products, including $C_4H_8O_4$, $C_3H_8O_4$, and $C_3H_8O_5$, can also be observed in both conditions. However, due to the high viscosity of the e-liquids, it is unclear whether these compounds are actual PG/VG oxidation products or lower oxygenated species that have formed clusters with H_2OI^- or O_2 during the chemical ionization process.⁴¹ When the relative fractions of these compounds are compared, it is evident that they contribute to a greater percentage in aged aerosols, which is five times higher than in fresh aerosols. This

suggests that both the thermal reaction during vaping and the further oxidation process influence their formation.

Furthermore, the aged conditions reveal additional peaks that are likely the result of the ozonolysis products of the flavorings. These products primarily consist of 8–12 carbons with oxygen numbers ranging from 2 to 6 and a double bond equivalent (DBE) ($\text{DBE} = (\text{number of carbon} \times 2 - \text{number of hydrogen} + 2)/2$) value between 0 and 2. Certain peaks have identical formulas to the oxidation products of terpenes reported in recent studies, such as $\text{C}_7\text{H}_{12}\text{O}_3$, $\text{C}_{10}\text{H}_{16}\text{O}_3$, and $\text{C}_{10}\text{H}_{16}\text{O}_4$.^{42–44} The presence of O_3 triggers the initiation of ozonolysis of these terpenes, which starts with the formation of primary ozonide and proceeds via Criegee intermediates, further reactions, and the release of an OH radical.⁴⁵ This intermediate then forms RO_2 radicals, which undergo either multistep oxidation or autoxidation. Ultimately, this process results in closed-shell products through various termination pathways, including self-termination.⁴⁶ In the absence of NO_x , the RO_2/HO_2 reaction is the main termination step, which promotes the generation of non-nitrogen-containing species like organic hydroperoxides, carbonyls, and alcohols.⁴⁷ Due to the technical constraints of the iodide-CIMS, it is only capable of determining molecular formulas based on accurate mass fittings. Therefore, additional validation using alternative analytical methods is necessary to confirm the structural and functional group information on gas-phase e-cig emissions.

3.3. Changes in Chemical Composition in the Particle Phase. Figure 2C,D shows the differences in particle-phase chemical composition between fresh and aged e-cig aerosols. Overall, the compositions of e-cig particles are mainly VG, accounting for a larger fraction (Figure 2D) due to its lower vapor pressure compared to PG.³² When the fresh and aged particles are compared, the unreacted PG and VG show greater fractions in fresh particles, indicating that the e-liquid solvent may participate in further reactions. While PG/VG cannot directly react with O_3 , the presence of intermediate radicals or byproducts may promote their oxidation.^{48,49} For example, when Criegee intermediates interact with chemicals that have alcohol and carboxyl functional groups, it can lead to the formation of multifunctional and highly oxygenated species.^{50–52} We focused on the peaks identified within the m/z 200 to 400 range, which are primarily ionized by forming an I^- adduct (Figure 2C). The main species detected in the particle phase include $\text{C}_3\text{H}_4\text{O}_4$, $\text{C}_3\text{H}_6\text{O}_3$, and $\text{C}_3\text{H}_6\text{O}_4$, which are oxidation products of PG/VG.^{53,54}

In comparison to those in the gas phase, products observed in the particle phase exhibit a greater degree of oxygenation, particularly under aged conditions. Several major species detected in this study have been previously reported through various techniques, such as ESI-MS/MS, which include compounds with molecular formulas like $\text{C}_7\text{H}_{10-12}\text{O}_{4-5}$, $\text{C}_8\text{H}_{12-14}\text{O}_{4-5}$, $\text{C}_9\text{H}_{14-16}\text{O}_{4-6}$, and $\text{C}_{10}\text{H}_{16-18}\text{O}_{4-7}$.⁵⁵ The presence of homologous + CH_2 or + O compounds is more prominent in aged e-cig aerosols, as shown in Figure S7, suggesting that the generation of these aerosols is influenced by multistep oxidation or autoxidation processes.^{56,57} The aerosols composed with unsaturated products ($\text{DBE} > 0$) primarily consists of carbonyls, organic acids, and hydroperoxides.²⁵

Terpenes, such as α -pinene, limonene, and geraniol, are the primary components of GG#4 flavoring, as described in the methodology. To investigate the effects of terpenes on the formation of e-cig aerosols under both fresh and aged

conditions, we compared the compositions of e-cig aerosols from four e-liquid formulations, including PG/VG + 3% GG#4, PG/VG + 3% α -pinene, PG/VG + 3% geraniol, and PG/VG only. Figure 3 displays the normalized mass spectra of

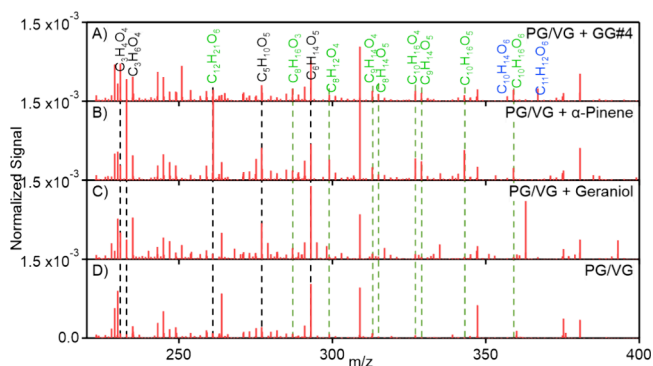


Figure 3. Chemical composition of particle-phase aged e-cig emissions generated from four e-liquids: (A) PG/VG + 3% GG#4, (B) PG/VG + 3% α -pinene, (C) PG/VG + 3% geraniol, and (D) PG/VG. Spectra are signals normalized to the intensity of VG ($\text{C}_3\text{H}_8\text{O}_3\text{I}^-$). Terpene-like oxidation products are labeled in green, unique products detected only in PG/VG + 3% GG#4 in blue, while common compounds directly from PG/VG are shown in black.

aged e-cig aerosols generated from these four e-liquids. Some common peaks shown across all four e-liquid conditions, such as $\text{C}_3\text{H}_6\text{O}_4$, $\text{C}_3\text{H}_4\text{O}_4$, $\text{C}_5\text{H}_{10}\text{O}_5$, and $\text{C}_6\text{H}_{14}\text{O}_5$, are likely PG/VG oxidation products, whereas terpene oxidation products, such as $\text{C}_8\text{H}_{16}\text{O}_3$, $\text{C}_8\text{H}_{12}\text{O}_4$, $\text{C}_9\text{H}_{14}\text{O}_4$, and $\text{C}_{10}\text{H}_{16}\text{O}_{4-5}$, show relatively higher intensity in GG#4 and α -pinene-containing e-liquids.^{58–60} The five common peaks were also present in geraniol-containing e-liquids, though at significantly lower intensities. However, the oxidation products of geraniol, such as $\text{C}_{10}\text{H}_{20}\text{O}_6$ and $\text{C}_8\text{H}_{16}\text{O}_6$, were not detected in GG#4 e-liquid vaping emissions. Despite geraniol undergoing more rapid ozonolysis compared to α -pinene, the higher concentration of α -pinene in the GG#4 e-liquid formulation relative to geraniol resulted in a greater contribution of oxidation products from α -pinene rather than geraniol. In addition to the common products from the oxidation of terpenes, unique peaks, such as $\text{C}_{10}\text{H}_{14}\text{O}_6$ and $\text{C}_{11}\text{H}_{12}\text{O}_6$, were observed exclusively in GG#4. These highly oxygenated species with high DBE values (formulas colored with blue in Figure 3; $\text{DBE} = 4$ for $\text{C}_{10}\text{H}_{14}\text{O}_6$ and $\text{DBE} = 6$ for $\text{C}_{11}\text{H}_{12}\text{O}_6$) are likely generated from interactions between different terpenes or between terpenes and PG/VG. Previous studies on e-cig aerosols have also identified products with high DBE values related to PAHs.^{61–63} Some studies indicated that the dehydration and cyclization of PG/VG would lead to the formation of aromatics,⁶⁴ but the detailed mechanisms remain unclear, which warrants further investigation in future studies.

3.4. Formation of Carbonyls during the Aging Processes. As mentioned in the previous section, the main products from the ozonolysis of terpenes are organic peroxides, carbonyls, and organic acids. Previous studies have extensively discussed the formation of carbonyls during the vaping processes, including both the gas and particle phases;^{34,53,65,66} however, less focus has been given to the carbonyl formation during the aging processes. In this study, we performed DNPH-assisted LC-MS to compare the carbonyl compounds present in fresh and aged e-cig aerosols produced from the

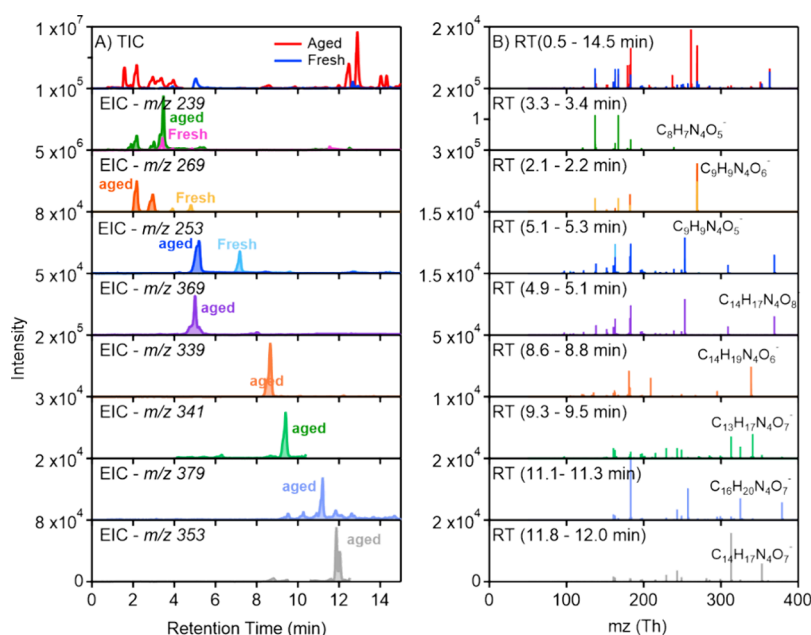


Figure 4. Detected carbonyls from the particle-phase e-cig emissions under aged and fresh conditions. (A) TICs of the e-cigarette aerosol sample extracts and EICs for nine deprotonated molecular ions $[M-H]^-$ of selected carbonyl-DNPH hydrazones and (B) the averaged mass spectra for peaks eluted at their specified retention times.

PG/VG + 3% GG#4 e-liquid. Given that no extra nitrogen-containing species, such as NO_x or N-containing organic compounds, were added to the chamber, the DNPH-carbonyls or DNPH-dicarbonyls in this study should contain only four or eight nitrogen atoms. With this in mind, we performed a nontarget analysis on the LC-MS high-resolution mass spectral data and fitted eight formulas with a mass tolerance of 2 ppm, as detailed in Table S3 and Figure 4. In addition to these 8 confirmed formulas, 20 probable carbonyl products were observed exclusively in the aged e-cig aerosols, as shown in Table S4. Figure 4A presents the total ion chromatograms (TICs) and extracted ion chromatograms (EICs) of eight carbonyl-DNPH compounds, while Figure 4B displays the extracted mass spectra under the specified retention time. The comparison of TICs for DNPH-assisted e-cig aerosols reveals a significant difference, with more peaks and greater intensity attributed to aged e-cig aerosols. The specific peak observed in the fresh e-cig aerosols (RT = 4.95 min) corresponds to the unreacted DNPH reagent.

Upon analysis of the common formulas in both fresh and aged e-cig aerosols, such as $\text{C}_2\text{H}_4\text{O}_2$, $\text{C}_3\text{H}_6\text{O}_3$, and $\text{C}_3\text{H}_6\text{O}_2$, we observed that only $\text{C}_2\text{H}_4\text{O}_2$ has the same retention times under both conditions, suggesting that it is the same compound. In contrast, the varying retention times of $\text{C}_3\text{H}_6\text{O}_3$ indicate the formation of distinct isomers under fresh and aged conditions, even though they share the same molecular formula. Although these three carbonyl compounds have been observed in previous studies and primarily attributed to the thermal degradation products of PG and VG,^{67–70} the presence of different isomers in fresh and aged e-cig aerosols remains unexplored. The differences observed between the fresh and aged e-cig aerosols suggest that the presence of O_3 could lead to varying oxidation mechanisms for PG/VG in contrast to thermal degradation processes. It is important to note that certain carbonyls, including formaldehyde, were not observed in this study due to their higher vapor pressure, which makes them difficult to detect in the particle phase.

Five carbonyl compounds ($\text{C}_8\text{H}_{14}\text{O}_5$, $\text{C}_8\text{H}_{16}\text{O}_3$, $\text{C}_7\text{H}_{14}\text{O}_4$, $\text{C}_8\text{H}_{14}\text{O}_4$, and $\text{C}_5\text{H}_{10}\text{O}_4$) were observed solely in aged e-cig aerosols, all of which are probably products of terpene oxidation with 7–10 carbon atoms. Previous studies have reported the formation of carbonyls as a result of the thermal oxidative degradation of terpenes.¹⁴ However, these carbonyls from thermal reactions tend to have a lower degree of oxygenation and greater volatility, hindering their partitioning into the particle phase. This likely explains the absence of terpene-related carbonyls in the fresh aerosols. From another perspective, these terpene-derived carbonyls in our aged e-cig aerosols underwent multistep oxidation only in the presence of O_3 . Table S4 shows that there are 20 different potential carbonyls in the aged e-cig aerosols, some of which are highly oxygenated, indicating that the interaction of both terpenes as flavorings and O_3 leads to particle formation. Enhanced oxygenation in aerosols may potentially elevate the cytotoxicity of e-cig aerosols due to the increased presence of ROS²⁵ and electrophilic carbonyl compounds, which can cause oxidative stress and damage to cellular components such as lipids, proteins, and DNA.⁵⁵

3.5. Cytotoxicity of Fresh and Aged E-Cig Aerosols.

Following exposure to fresh and aged GG#4 vaping emissions, BEAS-2B cells exhibited differential responses in terms of cell viability and LDH release. As shown in Figure 5, the aged emissions induced a drastic reduction in the cell viability in a concentration-dependent manner. Furthermore, the aged emissions were able to cause LDH release, indicating the aerosol's ability to induce cell membrane permeability or cell lysis. When exposed to the fresh emissions, BEAS-2B cells did not exhibit any significant reductions in cell viability nor increases in LDH release. Although the aged emissions were able to enhance LDH release, the increase was modest and not as pronounced as the reduction of cell viability, which was determined by an XTT assay. The tested concentration of 2.5 mg mL^{-1} led to a percent cell viability that was near zero. Considering the principle of the XTT assay, this indicates that

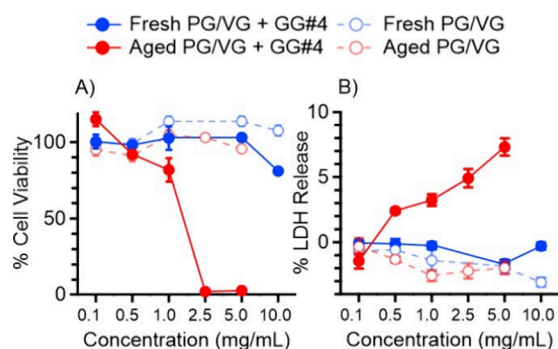


Figure 5. Cell viability and LDH release curves for BEAS-2B cells treated with aged and fresh e-cig aerosols generated from PG/VG + GG #4 and PG/VG only e-liquids, as measured by (A) XTT and (B) LDH assays, respectively. The results were normalized to the negative (unexposed) and positive (cells treated with 0.1% v/v Triton X-100) controls and expressed as the percentage of cell viability and LDH release. Each point represents the mean \pm standard error of the mean (SEM) of four biological replicates ($n = 4$).

the aged emissions can reduce mitochondrial respiration and bring about apoptosis.⁷¹ In contrast, no detectable cytotoxic effects were observed following exposure to control PG/VG only e-cig aerosols under fresh and aged conditions (Figure 5). Neither reductions in cell viability nor increases in LDH release were detected, suggesting that the cytotoxicity observed in the PG/VG + GG#4 aerosols was likely attributed to terpene oxidation products during aging rather than to the PG/VG vehicle itself.

Even at the highest concentration of 10 mg mL⁻¹, the fresh GG#4 vaping emissions did not exhibit any significant cytotoxicity. Particle-phase constituents found in the fresh vaping emissions are mainly composed of PG, VG, and unreacted terpenes, which are generally considered to be fairly benign substances regarding inhalation toxicity.⁷² Thermal degradation products generated during the vaping process, such as formaldehyde and acetaldehyde, are too volatile to be localized in the particle phase of vaping emissions; therefore, the cells in this study were not able to be exposed to significant amounts of those VOCs, which are considered as more potent toxins.⁷³ Thus, these results provide evidence that highly oxygenated compounds, which are produced from the indoor aging process of vaping emissions, have their own unique cytotoxic mechanisms that allow them to be much more harmful than their precursors. Although there is a lack of structural information in our work, there is a possibility that some of the carbonyls formed from the aging process of e-cig aerosols are aldehydes and α,β -unsaturated carbonyls that exhibit electrophilic properties.^{34,74} These classes of carbonyls are known to induce intracellular damage to biological targets that are responsible for proper cellular function and homeostasis.⁷⁵ Accumulation of this carbonyl-induced damage may eventually lead to cell death if left unrepaired, which corresponds well with the results shown in Figure 5.

Due to the potential for aged vaping emissions to be significantly more toxic than their fresh counterparts, vaping indoors, especially in urban environments polluted with ground-level O₃, poses a risk of secondhand exposure. While secondhand exposure may expose a bystander to a lower aerosol mass concentration compared to that of the primary e-cig user, the aged vaping emissions exhibit a lower threshold concentration for *in vitro* toxicity. Additionally, the *in vitro*

toxicity of SOAs formed from terpene oxidation is well characterized by their ability to induce oxidative stress.⁵⁵ As a result, chronic exposure to aged vaping emissions could increase the risk of developing respiratory conditions caused by excessive pulmonary oxidative stress.⁷⁶

4. IMPLICATIONS

This study investigated the particle properties, chemical composition, and *in vitro* cytotoxicity of aged e-cig aerosols. Our findings highlight that more UFPs were generated in the presence of O₃ compared to the zero-air condition. Vaping emissions containing terpene flavor additives were shown to interact with O₃ and generate these UFPs, which consist of oxygenated and functionalized products such as organic hydroperoxides,²⁵ carbonyls, and organic acids. These chemical transformations increase the concentration and size of aerosol particles and enhance their atmospheric lifetime and oxidative potential. These byproducts are also shown to result in greater oxidative stress²⁵ and respiratory issues.⁷⁷ Such changes exacerbate indoor air pollution and elevate health risks associated with passive vaping. These emissions persist when exhaled in indoor environments, posing secondary exposure risks to nonusers.

Our findings also indicate a need for implementing measures that limit passive exposure, such as designated vaping areas and enhanced ventilation requirements in public and private spaces. Furthermore, research should investigate the mechanisms underlying the formation of harmful byproducts in aged aerosols as well as their potential interactions with other indoor pollutants. To more accurately define the composition and behavior of e-cig aerosols under various environmental conditions, the further development and improvement of analytical techniques are essential.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at <https://pubs.acs.org/doi/10.1021/acs.chemrestox.4c00402>.

LC-MS measurement of DNPH-derived carbonyls (Text S1); formulations of e-liquids used in this study (Table S1); mass concentrations of e-cig particles from PG/VG + GG#4 and PG/VG under two conditions (Table S2); carbonyls characterized by DNPH-assisted LC-MS in Figure 4 (Table S3); other possible carbonyls characterized by DNPH-assisted LC-MS (Table S4); design of the chamber experiment e-cigarette aerosols; (A) schematic of the experimental setup for e-cigarette chamber aerosol introduction and collection and (B) picture of the cigarette smoking machine CSM-STEP (Figure S1); time series of mass concentrations of e-cigarette aerosols for (A) PG/VG + GG#4 and (B) PG/VG under fresh and aged conditions (Figure S2); aerosol growth and particle size distribution during the vaping process under different conditions (Figure S3); aerosol growth and particle size distribution during the vaping process under different conditions (Figure S4); ozone consumption during the chemical aging processes (Figure S5); aerosol growth and particle size distribution during the vaping process under different conditions (Figure S6); mass defect (MD) plot of aged e-cigarette aerosols, color-coded by the number of oxygen atoms

and symbol size scaled by the normalized signal (Figure S7) (PDF)

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Notes

The authors declare no competing financial interest.

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ABBREVIATIONS

2,4-DNPH, 2,4-Dinitrophenylhydrazine
CBD, Cannabidiol
CIMS, Chemical ionization mass spectrometry
DBE, Double bond equivalent
E-cig, E-cigarette
EIC, Extracted ion chromatograms
FEP, Fluorinated ethylene propylene
FIGAERO, Filter inlet for gases and aerosols system
LC/ESI, Liquid chromatography and electrospray ionization
LDH, Lactate dehydrogenase
PAH, Polycyclic aromatic hydrocarbons
PG, Propylene glycol
PTFE, Polytetrafluoroethylene
QToF-MS, Quadrupole time-of-flight mass spectrometry
ROS, Reactive oxygen species
RT, Retention time
SEMS, Scanning electrical mobility spectrometer
SOA, Secondary organic aerosol
THC, Δ -Tetrahydrocannabinol
TIC, Total ion chromatograms
ToF, Time of flight
UFP, Ultra fine particle
VG, Vegetable glycerin

REFERENCES

- (1) Cullen, K. A.; Ambrose, B. K.; Gentzke, A. S.; Apelberg, B. J.; Jamal, A.; King, B. A. Notes from the Field: Use of Electronic Cigarettes and Any Tobacco Product Among Middle and High School Students — United States, 2011–2018. *MMWR. Morb. Mortal. Wkly. Rep.* **2018**, *67* (45), 1276–1277.
- (2) Breland, A.; Soule, E.; Lopez, A.; Ramôa, C.; El-Hellani, A.; Eissenberg, T. Electronic Cigarettes: What Are They and What Do They Do? *Ann. N.Y. Acad. Sci.* **2017**, *1394* (1), 5–30.
- (3) Layden, J. E.; Ghinai, I.; Pray, I.; Kimball, A.; Layer, M.; Tenforde, M. W.; Navon, L.; Hoots, B.; Salvatore, P. P.; Elderbrook, M.; Haupt, T.; Kanne, J.; Patel, M. T.; Saathoff-Huber, L.; King, B. A.; Schier, J. G.; Mikosz, C. A.; Meiman, J. Pulmonary Illness Related to E-Cigarette Use in Illinois and Wisconsin — Final Report. *N. Engl. J. Med.* **2020**, *382* (10), 903–916.
- (4) Kramarow, E. A.; Elgaddal, N. Current Electronic Cigarette Use Among Adults Aged 18 and Over: United States, 2021. *NCHS Data Brief* **2023**, *475*, 1–8.
- (5) Giroud, C.; de Cesare, M.; Berthet, A.; Varlet, V.; Concha-Lozano, N.; Favrat, B. E-Cigarettes: A Review of New Trends in Cannabis Use. *Int. J. Environ. Res. Public Health* **2015**, *12* (8), 9988–10008.
- (6) Ballbè, M.; Martínez-Sánchez, J. M.; Sureda, X.; Fu, M.; Pérez-Ortuño, R.; Pascual, J. A.; Saltó, E.; Fernández, E. Cigarettes vs. e-Cigarettes: Passive Exposure at Home Measured by Means of Airborne Marker and Biomarkers. *Environ. Res.* **2014**, *135*, 76–80.
- (7) Jiang, H.; Ahmed, C. M. S.; Martin, T. J.; Canchola, A.; Oswald, I. W. H.; Garcia, J. A.; Chen, J. Y.; Koby, K. A.; Buchanan, A. J.; Zhao, Z.; Zhang, H.; Chen, K.; Lin, Y. H. Chemical and Toxicological Characterization of Vaping Emission Products from Commonly Used Vape Juice Diluents. *Chem. Res. Toxicol.* **2020**, *33* (8), 2157–2163.
- (8) Ogunwale, M. A.; Li, M.; Ramakrishnam Raju, M. V.; Chen, Y.; Nantz, M. H.; Conklin, D. J.; Fu, X. A. Aldehyde Detection in Electronic Cigarette Aerosols. *ACS Omega* **2017**, *2* (3), 1207–1214.
- (9) Khlystov, A.; Samburova, V. Flavoring Compounds Dominate Toxic Aldehyde Production during E-cigarette Vaping. *Environ. Sci. Technol.* **2016**, *50* (23), 13080–13085.
- (10) Gillman, I. G.; Pennington, A. S. C.; Humphries, K. E.; Oldham, M. J. Determining the Impact of Flavored E-Liquids on Aldehyde Production during Vaping. *Regul. Toxicol. Pharmacol.* **2020**, *112*, No. 104588.
- (11) Kocis, P. T.; Vrana, K. E. Terpenes/Terpenoids in Cannabis: Are They Important? *Med. Cannabis Cannabinoids* **2020**, *3* (1), 61–73.
- (12) Chen, C.; Pan, Z. Cannabidiol and Terpenes from Hemp — Ingredients for Future Foods and Processing Technologies. *J. Futur. Foods* **2021**, *1* (2), 113–127.
- (13) Nguyen, T. D.; Riordan-Short, S.; Dang, T. T. T.; O'Brien, R.; Noestheden, M. Quantitation of Select Terpenes/Terpenoids and Nicotine Using Gas Chromatography-Mass Spectrometry with High-Temperature Headspace Sampling. *ACS Omega* **2020**, *5* (10), 5565–5573.
- (14) McGraw, G. W.; Hemingway, R. W.; Ingram, L. L.; Canady, C. S.; McGraw, W. B. Thermal Degradation of Terpenes: Camphene, Δ 3-Carene, Limonene, and α -Terpinene. *Environ. Sci. Technol.* **1999**, *33* (22), 4029–4033.
- (15) Pathak, R. K.; Salo, K.; Emanuelsson, E. U.; Cai, C.; Lutz, A.; Hallquist, Å. M.; Hallquist, M. Influence of Ozone and Radical Chemistry on Limonene Organic Aerosol Production and Thermal Characteristics. *Environ. Sci. Technol.* **2012**, *46* (21), 11660–11669.
- (16) Inuma, Y.; Böge, O.; Miao, Y.; Sierau, B.; Gnauk, T.; Herrmann, H. Laboratory Studies on Secondary Organic Aerosol Formation from Terpenes. *Faraday Discuss.* **2005**, *130*, 279–294.
- (17) Eshraghian, E. A.; Al-Delaimy, W. K. A Review of Constituents Identified in E-Cigarette Liquids and Aerosols. *Tob. Prev. Cessat.* **2021**, *7*, 1–15.
- (18) Orlando, J. J.; Nozière, B.; Tyndall, G. S.; Orzechowska, G. E.; Paulson, S. E.; Rudich, Y. Product Studies of the OH- and Ozone-

- initiated Oxidation of Some Monoterpenes. *J. Geophys. Res. Atmos.* **2000**, *105* (D9), 11561–11572.
- (19) Xu, L.; Møller, K. H.; Crouse, J. D.; Otkjær, R. V.; Kjaergaard, H. G.; Wennberg, P. O. Unimolecular Reactions of Peroxy Radicals Formed in the Oxidation of α -Pinene and β -Pinene by Hydroxyl Radicals. *J. Phys. Chem. A* **2019**, *123* (8), 1661–1674.
- (20) Ziemann, P. J.; Atkinson, R. Kinetics, Products, and Mechanisms of Secondary Organic Aerosol Formation. *Chem. Soc. Rev.* **2012**, *41* (19), 6582.
- (21) Atkinson, R.; Arey, J. Gas-Phase Tropospheric Chemistry of Biogenic Volatile Organic Compounds: A Review. *Atmos. Environ.* **2003**, *37*, 197–219.
- (22) Sbai, S. E.; Farida, B. Photochemical Aging and Secondary Organic Aerosols Generated from Limonene in an Oxidation Flow Reactor. *Environ. Sci. Pollut. Res.* **2019**, *26* (18), 18411–18420.
- (23) Wolkoff, P.; Clausen, P. A.; Jensen, B.; Nielsen, G. D.; Wilkins, C. K. Are We Measuring the Relevant Indoor Pollutants? *Indoor Air* **1997**, *7* (2), 92–106.
- (24) Jokinen, T.; Berndt, T.; Makkonen, R.; Kerminen, V.-M.; Junninen, H.; Paasonen, P.; Stratmann, F.; Herrmann, H.; Guenther, A. B.; Worsnop, D. R.; Kulmala, M.; Ehn, M.; Sipilä, M. Production of Extremely Low Volatile Organic Compounds from Biogenic Emissions: Measured Yields and Atmospheric Implications. *Proc. Natl. Acad. Sci. U. S. A.* **2015**, *112* (23), 7123–7128.
- (25) Woo, W.; Tian, L.; Lum, M.; Canchola, A.; Chen, K.; Lin, Y. Ozonolysis of Terpene Flavor Additives in Vaping Emissions: Elevated Production of Reactive Oxygen Species and Oxidative Stress. *Chem. Res. Toxicol.* **2024**, *37* (6), 981–990.
- (26) Zhao, T.; Nguyen, C.; Lin, C. H.; Middlekauff, H. R.; Peters, K.; Mohemani, R.; Guo, Q.; Zhu, Y. Characteristics of Secondhand Electronic Cigarette Aerosols from Active Human Use. *Aerosol Sci. Technol.* **2017**, *51* (12), 1368–1376.
- (27) Amalia, B.; Fu, M.; Tigova, O.; Ballbè, M.; Paniello-Castillo, B.; Castellano, Y.; Vyzikidou, V. K.; O'Donnell, R.; Dobson, R.; Lugo, A.; Veronese, C.; Pérez-Ortuño, R.; Pascual, J. A.; Cortés, N.; Gil, F.; Olmedo, P.; Soriano, J. B.; Boffi, R.; Ruprecht, A.; Ancochea, J.; López, M. J.; Gallus, S.; Vardavas, C.; Semple, S.; Fernández, E. Exposure to Secondhand Aerosol from Electronic Cigarettes at Homes: A Real-Life Study in Four European Countries. *Sci. Total Environ.* **2023**, *854*, No. 158668.
- (28) Alnajem, A.; Redha, A.; Alroumi, D.; Alshammasi, A.; Ali, M.; Alhussaini, M.; Almutairi, W.; Esmaeil, A.; Ziyab, A. H. Use of Electronic Cigarettes and Secondhand Exposure to Their Aerosols Are Associated with Asthma Symptoms among Adolescents: A Cross-Sectional Study. *Respir. Res.* **2020**, *21* (1), 1–9.
- (29) Tayyarah, R. 2014 Electronic Cigarette Aerosol Parameters Study; 2015. <https://www.coresta.org/2014-electronic-cigarette-aerosol-parameters-study-29232.html>.
- (30) Wang, Y.; Takeuchi, M.; Wang, S.; Nizkorodov, S. A.; France, S.; Eris, G.; Ng, N. L. Photolysis of Gas-Phase Atmospherically Relevant Monoterpene-Derived Organic Nitrates. *J. Phys. Chem. A* **2023**, *127* (4), 987–999.
- (31) Lee, B. H.; Lopez-Hilfiker, F. D.; Mohr, C.; Kurtén, T.; Worsnop, D. R.; Thornton, J. A. An Iodide-Adduct High-Resolution Time-of-Flight Chemical-Ionization Mass Spectrometer: Application to Atmospheric Inorganic and Organic Compounds. *Environ. Sci. Technol.* **2014**, *48* (11), 6309–6317.
- (32) Tian, L.; Woo, W.; Canchola, A.; Chen, K.; Lin, Y.-H. Correlation Gas Chromatography and Two-Dimensional Volatility Basis Methods to Predict Gas-Particle Partitioning for e-Cigarette Aerosols. *Aerosol Sci. Technol.* **2024**, *58* (6), 630–643.
- (33) Chen, J. Y.; Jiang, H.; Chen, S. J.; Cullen, C.; Ahmed, C. M. S.; Lin, Y. H. Characterization of Electrophilicity and Oxidative Potential of Atmospheric Carbonyls. *Environ. Sci. Process. Impacts* **2019**, *21* (5), 856–866.
- (34) Chen, J. Y.; Canchola, A.; Lin, Y.-H. Carbonyl Composition and Electrophilicity in Vaping Emissions of Flavored and Unflavored E-Liquids. *Toxics* **2021**, *9* (12), 345.
- (35) Srivastava, D.; Vu, T. V.; Tong, S.; Shi, Z.; Harrison, R. M. Formation of Secondary Organic Aerosols from Anthropogenic Precursors in Laboratory Studies. *npj Clim. Atmos. Sci.* **2022**, *5* (1), 22.
- (36) Koch, S.; Winterhalter, R.; Uherek, E.; Koloff, A.; Neeb, P.; Moortgat, G. K. Formation of New Particles in the Gas-Phase Ozonolysis of Monoterpenes. *Atmos. Environ.* **2000**, *34* (23), 4031–4042.
- (37) Youssefi, S.; Waring, M. S. Predicting Secondary Organic Aerosol Formation from Terpenoid Ozonolysis with Varying Yields in Indoor Environments. *Indoor Air* **2012**, *22* (5), 415–426.
- (38) Bonn, B.; Moortgat, G. K. New Particle Formation during α - and β -Pinene Oxidation by O₃, OH and NO₃, and the Influence of Water Vapour: Particle Size Distribution Studies. *Atmos. Chem. Phys.* **2002**, *2* (3), 183–196.
- (39) Rosales, C. M. F.; Jiang, J.; Lahib, A.; Bottorff, B. P.; Reidy, E. K.; Kumar, V.; Tasoglou, A.; Huber, H.; Dusanter, S.; Tomas, A.; Boor, B. E.; Stevens, P. S. Chemistry and Human Exposure Implications of Secondary Organic Aerosol Production from Indoor Terpene Ozonolysis. *Sci. Adv.* **2022**, *8* (8), 1–16.
- (40) Forbes, T. P.; Krauss, S. T. Confined DART-MS for Rapid Chemical Analysis of Electronic Cigarette Aerosols and Spiked Drugs. *J. Am. Soc. Mass Spectrom.* **2021**, *32* (8), 2274–2280.
- (41) Robinson, M. A.; Neuman, J. A.; Huey, L. G.; Roberts, J. M.; Brown, S. S.; Veres, P. R. Temperature-Dependent Sensitivity of Iodide Chemical Ionization Mass Spectrometers. *Atmos. Meas. Technol.* **2022**, *15* (14), 4295–4305.
- (42) Yan, C.; Nie, W.; Äijälä, M.; Rissanen, M. P.; Canagaratna, M. R.; Massoli, P.; Junninen, H.; Jokinen, T.; Sarnela, N.; Häme, S. A. K.; Schobesberger, S.; Canonaco, F.; Yao, L.; Prévôt, A. S. H.; Petäjä, T.; Kulmala, M.; Sipilä, M.; Worsnop, D. R.; Ehn, M. Source Characterization of Highly Oxidized Multifunctional Compounds in a Boreal Forest Environment Using Positive Matrix Factorization. *Atmos. Chem. Phys.* **2016**, *16* (19), 12715–12731.
- (43) Zhang, Y.; Peräkylä, O.; Yan, C.; Heikkinen, L.; Äijälä, M.; Daellenbach, K. R.; Zha, Q.; Riva, M.; Garmash, O.; Junninen, H.; Paatero, P.; Worsnop, D.; Ehn, M. Insights into Atmospheric Oxidation Processes by Performing Factor Analyses on Subranges of Mass Spectra. *Atmos. Chem. Phys.* **2020**, *20* (10), 5945–5961.
- (44) Schwantes, R. H.; Emmons, L. K.; Orlando, J. J.; Barth, M. C.; Tyndall, G. S.; Hall, S. R.; Ullmann, K.; St. Clair, J. M.; Blake, D. R.; Wisthaler, A.; Bui, T. P. V. Comprehensive Isoprene and Terpene Gas-Phase Chemistry Improves Simulated Surface Ozone in the Southeastern US. *Atmos. Chem. Phys.* **2020**, *20* (6), 3739–3776.
- (45) Berndt, T.; Richters, S.; Kaethner, R.; Voigtländer, J.; Stratmann, F.; Sipilä, M.; Kulmala, M.; Herrmann, H. Gas-Phase Ozonolysis of Cycloalkenes: Formation of Highly Oxidized RO₂ Radicals and Their Reactions with NO, NO₂, SO₂, and Other RO₂ Radicals. *J. Phys. Chem. A* **2015**, *119* (41), 10336–10348.
- (46) Calogirou, A.; Larsen, B. R.; Kotzias, D. Gas-Phase Terpene Oxidation Products: A Review. *Atmos. Environ.* **1999**, *33* (9), 1423–1439.
- (47) Presto, A. A.; Huff Hartz, K. E.; Donahue, N. M. Secondary Organic Aerosol Production from Terpene Ozonolysis. 2. Effect of NO_x Concentration. *Environ. Sci. Technol.* **2005**, *39* (18), 7046–7054.
- (48) Alam, M. S.; Rickard, A. R.; Camredon, M.; Wyche, K. P.; Carr, T.; Hornsby, K. E.; Monks, P. S.; Bloss, W. J. Radical Product Yields from the Ozonolysis of Short Chain Alkenes under Atmospheric Boundary Layer Conditions. *J. Phys. Chem. A* **2013**, *117* (47), 12468–12483.
- (49) Son, Y.; Mishin, V.; Laskin, J. D.; Mainelis, G.; Wackowski, O. A.; Delnevo, C.; Schwander, S.; Khlystov, A.; Samburova, V.; Meng, Q. Hydroxyl Radicals in E-Cigarette Vapor and E-Vapor Oxidative Potentials under Different Vaping Patterns. *Chem. Res. Toxicol.* **2019**, *32* (6), 1087–1095.
- (50) Hu, M.; Chen, K.; Qiu, J.; Lin, Y. H.; Tonokura, K.; Enami, S. Decomposition Mechanism of α -Alkoxyalkyl-Hydroperoxides in the Liquid Phase: Temperature Dependent Kinetics and Theoretical Calculations. *Environ. Sci. Atmos.* **2022**, *2* (2), 241–251.

- (51) Zhao, R.; Kenseth, C. M.; Huang, Y.; Dalleska, N. F.; Kuang, X. M.; Chen, J.; Paulson, S. E.; Seinfeld, J. H. Rapid Aqueous-Phase Hydrolysis of Ester Hydroperoxides Arising from Criegee Intermediates and Organic Acids. *J. Phys. Chem. A* **2018**, *122* (23), 5190–5201.
- (52) Lin, Y. H.; Yin, C.; Lin, W. H.; Li, Y. L.; Takahashi, K.; Lin, J. J. M. Criegee Intermediate Reaction with Alcohol Is Enhanced by a Single Water Molecule. *J. Phys. Chem. Lett.* **2018**, *9* (24), 7040–7044.
- (53) Li, Y.; Burns, A. E.; Tran, L. N.; Abellar, K. A.; Poindexter, M.; Li, X.; Madl, A. K.; Pinkerton, K. E.; Nguyen, T. B. Impact of E-Liquid Composition, Coil Temperature, and Puff Topography on the Aerosol Chemistry of Electronic Cigarettes. *Chem. Res. Toxicol.* **2021**, *34* (6), 1640–1654.
- (54) Bühler, W.; Dinjus, E.; Ederer, H. J.; Kruse, A.; Mas, C. Ionic Reactions and Pyrolysis of Glycerol as Competing Reaction Pathways in Near- and Supercritical Water. *J. Supercrit. Fluids* **2002**, *22* (1), 37–53.
- (55) Khan, F.; Kwapiszewska, K.; Zhang, Y.; Chen, Y.; Lambe, A. T.; Kołodziejczyk, A.; Jalal, N.; Rudzinski, K.; Martínez-Romero, A.; Fry, R. C.; Surratt, J. D.; Szmigielski, R. Toxicological Responses of α -Pinene-Derived Secondary Organic Aerosol and Its Molecular Tracers in Human Lung Cell Lines. *Chem. Res. Toxicol.* **2021**, *34* (3), 817–832.
- (56) Waring, M. S.; Wells, J. R.; Siegel, J. A. Secondary Organic Aerosol Formation from Ozone Reactions with Single Terpenoids and Terpenoid Mixtures. *Atmos. Environ.* **2011**, *45* (25), 4235–4242.
- (57) Coleman, B. K.; Lunden, M. M.; Destailats, H.; Nazaroff, W. W. Secondary Organic Aerosol from Ozone-Initiated Reactions with Terpene-Rich Household Products. *Atmos. Environ.* **2008**, *42* (35), 8234–8245.
- (58) Atkinson, R.; Arey, J. Atmospheric Degradation of Volatile Organic Compounds. *Chem. Rev.* **2003**, *103* (12), 4605–4638.
- (59) Forester, C. D.; Ham, J. E.; Wells, J. R. Geraniol (2,6-Dimethyl-2,6-Octadien-8-Ol) Reactions with Ozone and OH Radical: Rate Constants and Gas-Phase Products. *Atmos. Environ.* **2007**, *41* (6), 1188–1199.
- (60) Atkinson, R.; Winer, A. M.; Pitts, J. N. Rate Constants for the Gas Phase Reactions of O₃ with the Natural Hydrocarbons Isoprene and α - and β -Pinene. *Atmos. Environ.* **1982**, *16* (5), 1017–1020.
- (61) Belushkin, M.; Tabin Djoko, D.; Esposito, M.; Korneliou, A.; Jeannet, C.; Lazzarini, M.; Jaccard, G. Selected Harmful and Potentially Harmful Constituents Levels in Commercial E-Cigarettes. *Chem. Res. Toxicol.* **2020**, *33* (2), 657–668.
- (62) Beauval, N.; Verrièle, M.; Garat, A.; Fronval, I.; Dusautoir, R.; Anthérieu, S.; Garçon, G.; Lo-Guidice, J. M.; Allorge, D.; Locoge, N. Influence of Puffing Conditions on the Carbonyl Composition of E-Cigarette Aerosols. *Int. J. Hyg. Environ. Health* **2019**, *222* (1), 136–146.
- (63) Dusautoir, R.; Zarccone, G.; Verrièle, M.; Garçon, G.; Fronval, I.; Beauval, N.; Allorge, D.; Riffault, V.; Locoge, N.; Lo-Guidice, J.-M.; Anthérieu, S. Comparison of the Chemical Composition of Aerosols from Heated Tobacco Products, Electronic Cigarettes and Tobacco Cigarettes and Their Toxic Impacts on the Human Bronchial Epithelial BEAS-2B Cells. *J. Hazard. Mater.* **2021**, *401*, No. 123417.
- (64) Pankow, J. F.; Kim, K.; McWhirter, K. J.; Luo, W.; Escobedo, J. O.; Strongin, R. M.; Duell, A. K.; Peyton, D. H.; Niaura, R. Benzene Formation in Electronic Cigarettes. *PLoS One* **2017**, *12* (3), No. e0173055.
- (65) Li, Y.; Burns, A. E.; Burke, G. J. P.; Poindexter, M. E.; Madl, A. K.; Pinkerton, K. E.; Nguyen, T. B. Application of High-Resolution Mass Spectrometry and a Theoretical Model to the Quantification of Multifunctional Carbonyls and Organic Acids in e-Cigarette Aerosol. *Environ. Sci. Technol.* **2020**, *54* (9), 5640–5650.
- (66) Farsalinos, K. E.; Gillman, G. Carbonyl Emissions in E-Cigarette Aerosol: A Systematic Review and Methodological Considerations. *Front. Physiol.* **2018**, *8*, 1–14.
- (67) Seifter, J. L. Integration of Acid–Base and Electrolyte Disorders. *N. Engl. J. Med.* **2015**, *372* (4), 389–392.
- (68) Jensen, R. P.; Strongin, R. M.; Peyton, D. H. Solvent Chemistry in the Electronic Cigarette Reaction Vessel. *Sci. Rep.* **2017**, *7* (1), 42549.
- (69) Lee, C.; Park, C. Bacterial Responses to Glyoxal and Methylglyoxal: Reactive Electrophilic Species. *Int. J. Mol. Sci.* **2017**, *18* (1), 169.
- (70) Laino, T.; Tuma, C.; Moor, P.; Martin, E.; Stolz, S.; Curioni, A. Mechanisms of Propylene Glycol and Triacetin Pyrolysis. *J. Phys. Chem. A* **2012**, *116* (18), 4602–4609.
- (71) Kari, S.; Subramanian, K.; Altomonte, I. A.; Murugesan, A.; Yli-Harja, O.; Kandhavelu, M. Programmed Cell Death Detection Methods: A Systematic Review and a Categorical Comparison. *Apoptosis* **2022**, *27* (7–8), 482–508.
- (72) Phillips, B.; Titz, B.; Kogel, U.; Sharma, D.; Leroy, P.; Xiang, Y.; Vuillaume, G.; Lebrun, S.; Sciuscio, D.; Ho, J.; Nury, C.; Guedj, E.; Elamin, A.; Esposito, M.; Krishnan, S.; Schlage, W. K.; Veljkovic, E.; Ivanov, N. V.; Martin, F.; Peitsch, M. C.; Hoeng, J.; Vanscheeuwijck, P. Toxicity of the Main Electronic Cigarette Components, Propylene Glycol, Glycerin, and Nicotine, in Sprague-Dawley Rats in a 90-Day OECD Inhalation Study Complemented by Molecular Endpoints. *Food Chem. Toxicol.* **2017**, *109*, 315–332.
- (73) Lorkiewicz, P.; Keith, R.; Lynch, J.; Jin, L.; Theis, W.; Krivokhizhina, T.; Riggs, D.; Bhatnagar, A.; Srivastava, S.; Conklin, D. J. Electronic Cigarette Solvents, JUUL E-Liquids, and Biomarkers of Exposure: In Vivo Evidence for Acrolein and Glycidol in E-Cig-Derived Aerosols. *Chem. Res. Toxicol.* **2022**, *35* (2), 283–292.
- (74) Koleva, Y. K.; Madden, J. C.; Cronin, M. T. D. Formation of Categories from Structure-Activity Relationships to Allow Read-across for Risk Assessment: Toxicity of α,β -Unsaturated Carbonyl Compounds. *Chem. Res. Toxicol.* **2008**, *21* (12), 2300–2312.
- (75) Liu, Y. Y.; Lin, T. C.; Wang, Y. J.; Ho, W. L. Carbonyl Compounds and Toxicity Assessments of Emissions from a Diesel Engine Running on Biodiesels. *J. Air Waste Manag. Assoc.* **2009**, *59* (2), 163–171.
- (76) Sahiner, U. M.; Birben, E.; Erzurum, S.; Sackesen, C.; Kalayci, O. Oxidative Stress in Asthma. *World Allergy Organ. J.* **2011**, *4* (10), 151–158.
- (77) Gotts, J. E.; Jordt, S. E.; McConnell, R.; Tarran, R. What Are the Respiratory Effects of E-Cigarettes? *BMJ.* **2019**, *366*, l5275.