Mechanistic rationale for ketene formation during vaping

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Abstract. Ketene is one of the most toxic vaping emissions identified to date. However, its high reactivity renders it relatively challenging to identify. In addition, certain theoretical studies show that realistic vaping temperature settings are too low to produce ketene. Each of these issues is addressed herein. First, an isotopically-labeled acetate precursor is used for the identification of ketene with enhanced rigor in vaped aerosols. Second, discrepancies between theoretical and experimental findings are explained by accounting for the effects of aerobic (experimental) versus anaerobic (simulated and theoretical) pyrolysis conditions. This finding is also relevant to explaining the relatively low temperature production of aerosol toxicants beyond ketene. Moreover, the study herein shows that ketene formation during vaping is not limited to molecules possessing a phenyl acetate substructure. This means that ketene emission during vaping, including from popular flavorants such as ethyl acetate, may be more prevalent than is currently known.

1. Introduction

Ketenes comprise a unique class of cumulene molecules with the general formula RR'C=C=O. Initially described by Wedekind as a reactive intermediate in 1901,[1] Staudinger reported the first synthesis and characterization of a ketene molecule, diphenyl ketene (R=R'=Ph), in 1905.[2] Since that time, despite their relative instability, ketenes have served as valuable reagents and intermediates in organic synthesis.[3] [4] Currently, however, the smallest homolog (R=R'=H, ketene, ethenone) of the series is receiving attention as a potentially significant public health hazard,[5] since it has been identified in aerosols generated by commercial vaping products.[6] [7]

The chemical and toxicological properties of ketene mirror those of phosgene ($Cl_2C=O$), a WW-I chemical warfare agent, as a reactive acylating agent and respiratory poison.[8] There is a lack of human toxicity data for ketene exposure. The available animal data was obtained mainly prior to 1950. Exposure to animals causes alveolar damage and a delayed onset of pulmonary toxicity leading to death by pulmonary edema.[8] Similar to phosgene, the delayed effects result from the non-enzymatic acylation of lung proteins, as opposed to direct irritation. The Acute Exposure Guideline Levels (AEGL)-3 (life-threatening levels) for ketene are 0.24 ppm for 10 min and 0.088 ppm for 8 h.[8]

Vitamin E acetate (VEA) has been linked to the 2019-2020 e-cigarette or vaping product use–associated lung injury (EVALI) epidemic, in large part due to VEA's prevalence in patient samples.[9] In 2020, Wu and O'Shea reported the formation of ketene when heating and vaping VEA,[7] indicating an additional possible link between ketene and EVALI. There is general agreement that EVALI is caused by chemical toxicant inhalation.[10] However, to date, neither ketene, VEA, nor any other specific chemical has been conclusively proven to be the causative agent of EVALI. Moreover, although there has been a significant decline since 2020, cases continue to be observed throughout the US.[10]

Regardless of whether VEA-derived ketene is a primary cause of EVALI, any source of exposure to ketene may put one at risk for significant lung injury. Recently, we reported that four acetylated cannabinoids (acetylated Δ^8- and Δ^9- THC, CBN, and CBD; **Figure 1**), produce ketene emissions under real-world vaping conditions, from either a vape pen or dab platform, including at levels in range of NIOSH (National Institute for Occupational Safety & Health) thresholds.[7] Interestingly, aerosolized THC products had been reported to cause acute respiratory syndromes prior to the EVALI outbreak.[11]

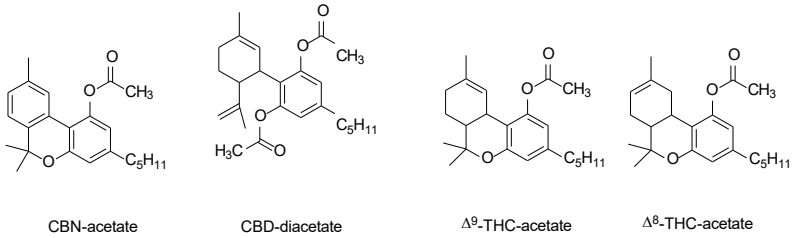
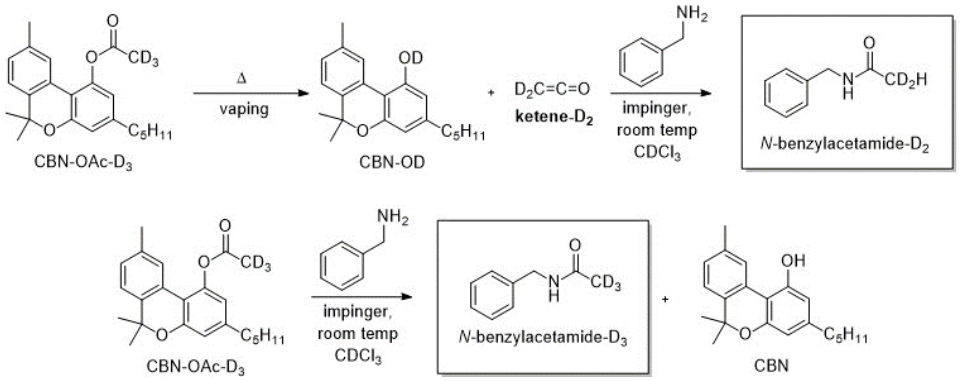


Figure 1. Cannabinoid acetates previously shown to produce ketene emissions under vaping conditions. The study described herein focuses on CBN-acetate because it produces the relatively cleanest aerosol analytical data due to the added stability of its second aryl ring.

The strong experimental evidence for ketene emissions arising from vaping cannabinoid acetates or VEA raises two issues. First, since ketene is too reactive and short-lived to be characterized as an intact molecule under common laboratory conditions, vaping studies to date have employed the well-known method of trapping ketene with a nucleophile (i.e., benzylamine) for characterization as the corresponding *N*-benzylacetamide. Since amines are relatively non-selective reagents, their use for ketene trapping and determination should ideally be limited to relatively well-defined systems that do not contain molecules that can react to form the same *N*-benzylacetamide product as ketene. However, manufacturers are not required to disclose most vaping product ingredients and, moreover, heating and vaping produces aerosols containing complex chemical mixtures.

To address this issue, herein we describe the use of isotopically-labeled CBN-acetate to rigorously identify ketene formation during vaping. All experiments were performed using a commercially available device set at temperature settings consistent with normal user practices. The main hypothesis addressed via isotopic labeling is illustrated in **Scheme 1**: **(Top)** A characteristic dideuterated *N*-benzylacetamide product (*N*-benzylacetamide-D_2) would result if a ketene intermediate is formed from the trideuterated acetate methyl (i.e., CBN-OAc-D_3). **(Bottom)** Conversely, if ketene is not formed as a reaction intermediate, an alternative addition-elimination or related reaction would result in trideuterated *N*-benzylacetamide (*N*-benzylacetamide-D_3).



Scheme 1. Top: Ketene possesses a methylene carbon. Its formation from a trideuterated actetate will therefore result in *N*-benzylacetamide-D_2. Bottom: Alternatively, *N*-benzylacetamide-D_3 will form if a different (e.g., addition-elimination) mechanism not involving a ketene intermediate is relevant.

The second issue centers on concerns that real-world vaping power settings do not provide enough energy to produce ketene. Recent theoretical studies show that unrealistically high vaping temperatures, in excess of at least 700 ºC, are required for any significant levels of ketene to form from VEA or other acetates.[12,13] To address this issue, some have proposed that ineffective wicking (resulting in dry, overheated vaporizer coils, colloquially termed "dry puff") or catalysis are potential causes of ketene production at relatively low power settings.[12] Alternatively, herein we hypothesize that ketene formation at real world vaping temperatures can be explained if one accounts for the impact of oxygen. This hypothesis is based on prior studies reported by our group in 2017,[14] as well as in subsequent reports by us[15] and by others.[16,17]

2. Results

2.1. Synthesis and mass spectroscopic analysis of CBN-OAc-D_3

Since a large portion of the volatile thermal reaction products of Δ^8 and Δ^9 THC formed under vaping conditions derive from their cyclohexene ring, [18-20] we focused current experiments on CBN-acetate. CBN-acetate contains a second aryl, rather than a cyclohexene ring **(Figure 1,** see CBN-OAc-D_3**)** that renders it relatively stable, thereby reducing the analytical complexity of its corresponding aerosol samples. In addition, CBN and CBN-OAc are crystalline and thus easier to purify and handle compared to THC or CBD and their acetate derivatives.

CBN-OAc-D_3 was obtained in 85 % yield by heating CBN and acetic anhydride-D_6 under neat conditions at reflux for 2 h. Previously, Nishida et al. synthesized trideuterated phenyl acetate to obtain evidence for the thermolytic mechanism of ketene formation.[21] They

determined that a four membered ring transition state involving transfer of a hydrogen atom from the acetate methyl to the phenolic ester oxygen led to ketene. Consistent with this mechanism, we tentatively assigned the CBN-OD byproduct from deuterated ketene production shown in Scheme 1 and **Figure 2** with a deuterated phenolic oxygen.

Figure 2 shows the EIMS fragmentation pattern of CBN-OAc-D_3. The corresponding data for CBN-OAc is displayed for comparison in the Figure S2. Loss of a geminal dimethyl from the structure corresponding to the molecular ion peak affords a base peak at 340 amu. Loss of dideuterated ketene (44 amu) affords the peak corresponding to CBN-OD.

Two other prominent peaks appear at 238 amu, arising from the loss of a four-carbon subunit from the cannabinoid pentyl side chain, at 46 amu, corresponding to the trideuterated acylium ion. The appearance of the latter peak potentially serves as evidence for an alternative intermolecular pathway to ketene, for example, involving dedeuteration of the acylium ion. Although the mass spectrometric fragmentation pattern also shows the feasibility of loss of ketene (from the base peak), electron impact mass spectral fragmentation conditions may not be directly applicable to the analysis of products arising from heating and aerosolization as used in vaping.

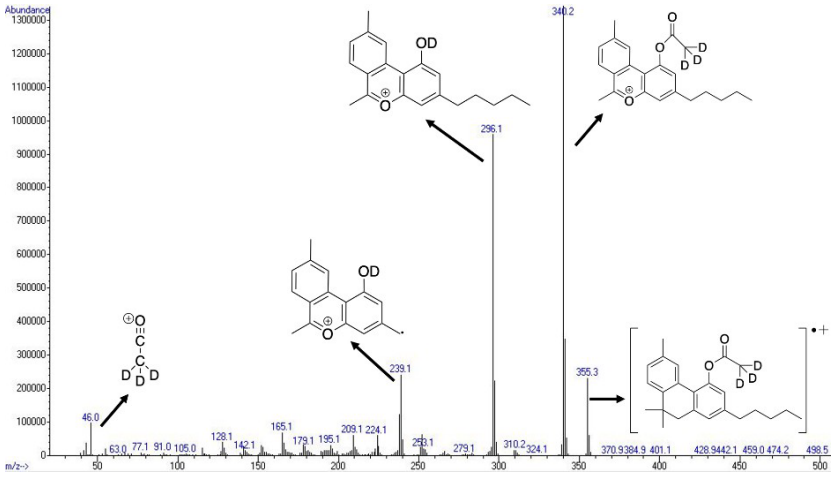


Figure 2. GC-EIMS (positive mode) of CBN-OAc-D3 showing fragmentation involving the loss of the ketene-D2 (44 amu) from the base peak. The analogous spectrum for unlabeled CBN-OAc is shown in the Supporting Information.

2.2. Aerosol generation and collection

In this and prior studies we used a dabbing platform to heat and aerosolize cannabinoids.[7,18-20] Dabbing is a type of cannabis vaping that entails flash vaporizing manufactured cannabis products (e.g, high THC- or CBD-potency extracts) on a hot surface

(colloquially referred to as a *nail*), with users often inhaling close to an entire lung volume in one puff. A national web survey of cannabis consumers found that 60% of respondents dabbed at least once, and that 38% endorsed its regular use.[22] Apart from embodying a popular means of cannabis vaping, using a dab platform has experimental advantages compared to vape pens. For example, the aerosols produced are not as prone to the influences of solvent, viscosity, or wicking that can significantly impact vaping chemistry and aerosolization efficiency.[19,20]

The specific aerosolization and collection method used herein has been described by us previously, with minor changes (see Supporting Information).[7] Briefly, the cannabinoid sample (~100 mg) is flash vaporized on a quartz dab platform surface and the generated aerosol pulled via a smoking machine into an in-line impinger. The impinger contains NMR solvent ($CDCl_3$) and the trapping agent (benzylamine) at room temperature. Aliquots of the impinger solution containing the dissolved aerosol contents generated by a single puff are directly analyzed by NMR and GC-MS. The yield of ketene (as *N*-benzylamide equivalents) formed from 40 mg CBN-OAc under these conditions is 0.078 ± 0.016 mg (n=4) at 378 °C.[7]

2.3. NMR and Mass Spectrometric Evidence for Ketene Formation

The production of *N*-benzylacetamide-D_2, the "trapped" ketene equivalent product shown in Scheme 1, was unambiguously confirmed. The observed ESI-HRMS peak at

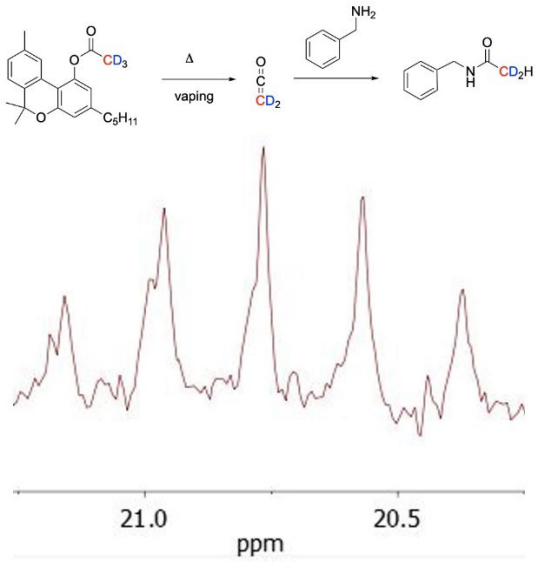


Figure 3. Expansion of the ^{13}C NMR spectrum ($CDCl_3$) of the aerosol generated under flash vaporization conditions (378 °C) on a quartz surface showing the anticipated quintet splitting pattern (J = 20 Hz) corresponding to the di-deuterated acetate methyl carbon of *N*- benzylamide-D_2. See also Figure S6.

150.0883 amu is within 0.7 ppm of the calculated value (150.0882 amu). Significantly, the proton-decoupled ^{13}C NMR spectrum shows a characteristic quintet splitting pattern centered at 20.8 ppm for - CD_2H, J = 20 Hz, (Figure 3).

2.4. Control experiments

Three additional experiments were conducted to provide added evidence for ketene formation under real-world conditions. The first involved demonstrating that unreacted CBN-OAc is not a competing substrate in the impinger for benzylamine (see Scheme 1, bottom). We stirred CBN-OAc for up to 8 h in a control impinger solution containing $CDCl_3$ and benzylamine. No *N*-benzylacetamide formation was observed in the ^1H NMR spectrum.

In contrast, in the vaping experiments, the ^1H NMR spectrum of the impinger solution showed the presence of *N*-benzylacetamide within minutes of aerosol generation and collection. Moreover, had CBN-OAc-D_3 (rather than ketene-D_2) directly reacted with benzylamine, a septet arising from -CD_3 splitting (from *N*-benzylacetamide-D_3) would have been observed in the ^{13}C NMR, rather than the quintet shown in Figure 3.

The purity of the CBN-OAc-D_3 was confirmed prior to the vaping experiments (Figure S3). Purification of CBN-OAc-D_3 was performed by dissolving the crude reaction mixture in an excess of hot ethanol, from which solvent the desired product crystallizes upon cooling. Acetic anhydride reacts rapidly and irreversibly with alcohols like ethanol, especially at elevated temperatures; upon solvation any residual acetic anhydride-D_6 forms trideuterated acetic acid (AcOH-D_3) and trideuterated ethyl acetate (EtOAc-D_3), which both join the mother liquor and wash cleanly away from the CBN-OAc crystals. However, if any residual acetic anhydride-D_6 remained in the sample and was responsible for the formation of *N*-benzylacetamide, the ^{13}C resonance from the resultant trideuterated acetate methyl carbon would split into a septet, rather than the observed quintet.

A second control study was performed to validate the feasibility of relatively lower vaping temperatures leading to the formation of ketene. Each of the dab platform experiments described above was initially run at 378 °C, in keeping with our prior studies.[7] During the course of this study, we collaborated on a peer-reviewed systematic survey and analysis of user-preferred THC-acetate dabbing temperatures.[23] Temperatures ≥ 378 °C were preferred by 8 % of respondents. For relevance to a wider range of users we thus investigated and found readily detectable (^1H NMR, Figure S1) levels of ketene as *N*-benzylacetamide equivalents when the aerosolization was performed at 287 °C, a temperature setting at or above which > 40 % of the survey respondents reported dabbing. [23]

To investigate the potential role of oxygen in ketene formation, we dabbed ethyl acetate (EtOAc, **Figure 4**) and geranyl acetate (Figure S5) at 378 °C in a glove bag in ambient air (~21% O_2) conditions and also under N_2 (<0.1% O_2). EtOAc or geranyl acetate (500 ul) were used in these experiments since they are liquids, and thus more amenable to glove bag conditions. The transformation of EtOAc to ketene had been simulated by others, with vaping temperatures > 840 ºC needed to afford enough ketene to be introduced into a user's lungs.[13] However, Figure 4 shows that, at 378 ºC, *N*-benzylacetamide derived from dabbing EtOAc is ~ 10-fold higher under ambient aerobic (0.025 mg) versus reduced O_2 conditions (0.0025 mg). Importantly, EtOAc is a prevalent flavorant molecule used in tobacco e-cigarettes. It was observed to be the fifth most frequently occurring flavor chemical in a study of 277 e-liquid products. [24] Geranyl acetate (Figure S5) exhibited analogous behavior, with ~ 20 fold more ketene generated under ambient aerobic conditions.

3. Discussion

The safety and regulation of vaping products are controversial issues, arguably among the most polarizing in the history of tobacco control.[25,26] Evaluation of tobacco as well as cannabis vaping products must include an understanding of their emissions, since significant chemical reactions occur upon both product storage as well as upon heating and vaping. Determining not only the identities of toxicant emissions, but also their origins, can support efforts towards improved product safety.

Ketene is one of the most toxic vaping emissions identified to date. To address exposure it is necessary to identify ketene at its source; otherwise its high reactivity renders its determination impractical *in vivo* or *in vitro*.[27] The use of an isotopically labeled acetate precursor (Scheme 1) enables rigorous ketene determination via ^{13}C NMR splitting patterns and high resolution mass spectrometry (Figure 3).

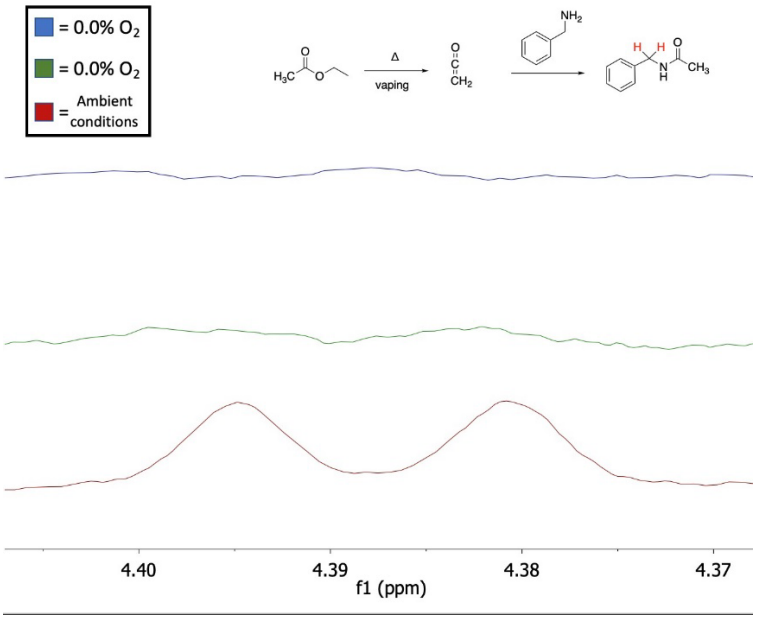


Figure 4. Expanded ^{1}H NMR spectra showing the methylene protons of *N*-benzylamide that was formed upon collection of the aerosol generated by ethyl acetate under ambient atmospheric conditions (red, bottom spectrum) along with two trials conducted in a 0.0% O2 (<LOD of instrument) atmosphere, displayed in the green (middle) and blue (top) spectra. qNMR analysis of the samples was conducted and shows that ambient atmospheric conditions generated ten times the amount of ketene (trapped as *N*-benzylacetamide) when compared to the ketene generated under anaerobic conditions.

3. Ketene is formed under real-world vaping temperatures

Ketene was detectable upon vaping CBN-acetate at vaping temperatures as low, to date, as 278 ºC using a dab platform (Figure S1). This is consistent with our prior work, wherein ketene was detectable using a pre-filled commercial vape cartridge and a low battery power setting.[7] However, according to theoretical simulations, ketene formation should not occur from vaping VEA,[12] cannabinoid[5] or other acetates[13] at real-world vaping temperature settings.

Importantly, ketene is not the only aerosol toxicant whose formation under realistic vaping power settings has been questioned. For example, findings showing that e-cigarette flavorant additives were linked to elevated toxic aldehyde emissions[28] were initially criticized by industry supporters as faulty.[29] This judgment was based on speculation that the researchers did not account for dry puffs and overheated e-liquid.[29] Dry puff was later discounted as relevant to aldehyde production from flavorant molecules.[30] It was also shown that, at least in the case of one specific additive (triacetin), that acrolein, acetaldehyde and formaldehyde formation was promoted by the catalytic reaction of flavorant-derived acetic acid with e-liquid solvent.[31]

An alternative to dry puff as an explanation for enhanced levels of aerosol toxicants is film boiling.[32] Film boiling occurs when the heat transfer between submerged metal heating coils and e-liquid is inhibited by formation of an insulating thin vapor film. The film forms

due to excess heat flux at temperatures above the e-liquid boiling point, thereby promoting toxicant formation from overheated e-liquid. Significantly, e-cigarette manufacturers' recommended device power levels were shown to afford heat fluxes at levels that can produce film boiling.[32] Film boiling thus can potentially explain why recommended safe power settings can lead to elevated aerosol toxicant emissions.In the studies described herein, a dab platform was used in conjunction with programmed temperatures and a quartz surface. There were no solvent or wicking effects involved as when using e-cigarettes, thus reducing the likelihood dry puff or film boiling as causes of ketene production. Another potential explanation for ketene as well as other toxicant emissions at relatively low temperatures is catalysis. For example, in 2018, Saliba et al. showed that toxic aldehydes, including methyl glyoxal, formaldehyde, and acetaldehyde, can form from the breakdown of the e-cigarette solvent propylene glycol at temperatures as low as ~ 80 ºC due to surface catalysis by metal heating element material (e.g., kanthal or nichrome).[33]

To investigate catalysis and ketene formation, a preliminary analysis of used cannabinoid EVALI patient vape cartridge components by Wu and O'Shea uncovered features including nickel and chromium filaments within a charred, oil-soaked, silica ceramic. [34] Catalysis could have potentially occurred via the filaments, although charring is evidence of elevated temperatures. To the best of our knowledge, quartz (as used herein) has also not (to date) been identified as a heating component involved in catalysis during vaping.

In addition to dry puff, solvent front and catalytic effects, there is another potential cause of ketene and other toxicant emissions at relatively low vaping power settings. We first reported the effect of aerobic conditions on vaping chemistry in 2017.[14] Upon examining the aerosol chemical profiles obtained from vaping propylene glycol and glycerol it became clear to us that oxidation reactions, particularly hydrogen atom abstractions, were prevalent during vaping.[14]

Diaz had demonstrated in 2010 that O_2 initiates the thermal degradation of propylene glycol and glycerol at significantly lower temperatures compared to anaerobic (i.e., pyrolysis) conditions.[35] For example, O_2-promoted hydrogen abstraction from propylene glycol was shown to occur at temperatures as low as 127 °C, leading to products derived from carbon-centered radicals.

In our earlier studies, to confirm that O_2 promotes toxic product formation under vaping conditions, we compared product yields obtained under ambient vs. reduced-O_2 conditions.[14] An obvious decrease in total decomposition products was observed when samples of aerosolized e-liquid were collected in a sealed glove-bag that had been flushed with N_2.

The influence of aerobic conditions on the formation of toxic degradation products via hydrogen atom abstraction is consistent with the studies of Son[36] and Zhao [37]who showed that hydroxyl radicals are produced during vaping. Moreover, subsequent studies by other researchers confirmed that O_2 is a major factor in promoting aerosol toxicant formation in the presence or absence of metal catalysts, and at relatively mild vaping temperatures. [16,17]

The results shown in Figure 4 embody strong evidence that ketene emissions are O_2-dependent. Dabbing EtOAc as well as geranyl acetate showed clear reductions in ketene yields under anaerobic versus aerobic conditions. As noted above, theoretical studies had shown that ketene is not expected to form, from VEA or EtOAc, for example, at vaping temperatures < 700 – 850 ºC, respectively. The simulated reaction pathways had activation energies of > 50 kcal/mol, which is associated with temperatures much higher than normal vaping conditions. However, the simulations were done under anaerobic (pyrolysis) conditions, which therefore can account for the discrepancies between the theoretical and experimentally observed temperatures.

In using EtOAc and geranyl acetate to compare ketene production under anaerobic and aerobic environments, the results (Figure 4) showed that acetates besides phenyl acetate-containing VEA and cannabinoids, can serve as ketene precursors. This is significant since ester flavorants are the most common class of tobacco e-cigarette flavorants. Exposure to ketene emissions may thus be more prevalent than currently known. This finding suggests the need for further studies on vaping and ketene emissions as well as ketene toxicology.

4. Conclusion

Two main issues were addressed during the studies reported herein. First, rigorous evidence for ketene formation during vaping was obtained via the use of an isotopically-labeled acetate precursor. Second, ketene formation was shown to be feasible at common vaping temperature settings. Moreover, under anaerobic conditions, ketene formation was inhibited, demonstrating that the role of oxygen and hydroxyl radicals should be accounted for when investigating the mechanism of ketene formation. In addition, we found that ketene formation was not limited to molecules containing a phenyl acetate substructure. These findings show that theoretical models based on anaerobic pyrolysis studies may provide an insufficient estimate of the thermal oxidation chemistry that is possible under real world vaping and dabbing conditions. Further investigations into the scope of ketene formation from additional ester-containing flavorant additives is ongoing in our laboratories and will be reported in due course.

Supporting Information

Supporting Information is available.

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Supporting Information

Mechanistic rationale for ketene formation during vaping

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Experimental Section

Materials and Methods. All NMR experiments described herein were run on a Bruker Avance III 400 MHz NMR spectrometer. All ^{13}C NMR experiments (100 MHz) were run with 1024 scans. HPLC-MS analysis was conducted on a ThermoFisher Scientific Q Exactive high resolution time of flight mass spectrometer (HR-TOF-MS) using electrospray ionization. Gas Chromatography Mass Spectrometry (GCMS) was performed on a capillary GC column mounted in an Agilent (Santa Clara, CA) 7890A GC interfaced to an Agilent 5975C MS, and operated in electron impact ionization mode. Mass fragmentation patterns were compared to the NIST database to generate a percent match based on the observed fragmentation pattern. CBN was obtained from FloraWorks at 99% purity. All other chemicals were received from Sigma-Aldrich and used without purification.

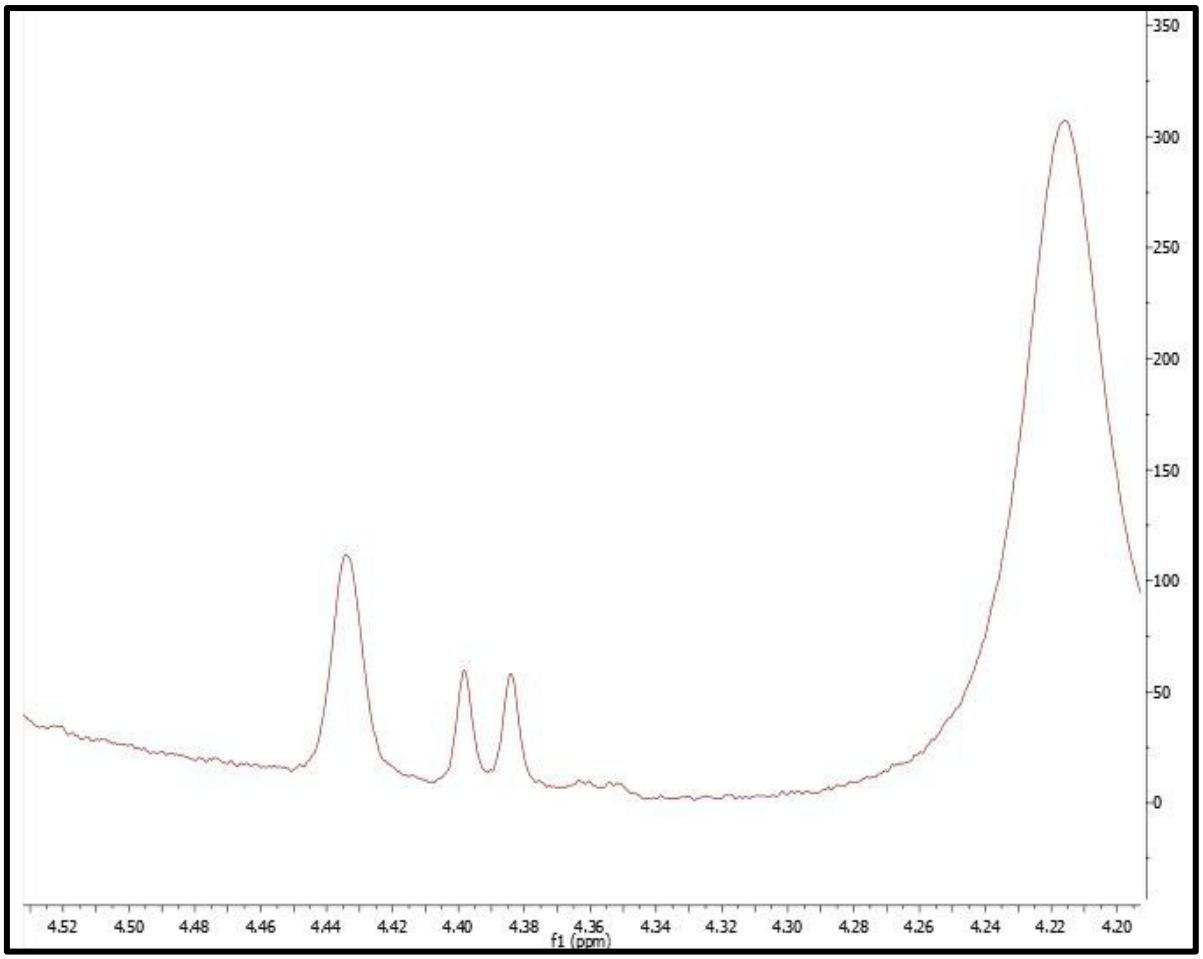
Synthesis of CBN-OAc-D$_3$. CBN (3.0 g) was heated at reflux with 1.5 ml acetic anhydride-D_6 for 2 h at 200 °C. While the solution was still warm, 15 ml of 70 °C EtOH was added and the mixture was cooled for 24 h. The CBN-OAc-D_3 crystallized and was filtered and dried under vacuum. A second volume equivalent of EtOH was added to the filtered crystals and heated at 70 ºC until complete dissolution at which time the solution was cooled for 24 h at rt affording recrystallized CBN-OAc-D_3. The crystals were filtered, dried under vacuum and weighed (3.0 g, 88 % yield, 99% purity by GC-MS). The ^1H NMR spectrum was identical to that of CBN-OAc synthesized previously[1] (except for the deuterated acetate methyl proton peaks).

Aerosol generation and collection. An EKS REX-C100 temperature controller was used for heating 500 mg of acetate on the quartz nail during dabbing experiments. The impinger solution proportions were adjusted slightly from the prior studies to improve the resolution of the *N*-benzylacetamide methylene peak and avoid overlap with the benzylamine peak.[1] The solution in the impinger consisted of 950 ul $CDCl_3$ (950 ul) and benzylamine (50 ul). This ratio was found through analysis of multiple samples to optimize the reaction of benzylamine and ketene. Once the impinger was

prepared, the nail was heated to 378 °C (or 287 ºC), within the range of temperatures at which cannabis consumers typically dab. All flow rate values and puff durations were the same as previously described by us.[1]

After aerosol generation and collection over ~10 min using a CSM-STEP smoking machine, the solution was removed from the impinger and transferred to an NMR tube and immediately run on the Bruker Avance III 400 MHz NMR spectrometer. Additional trials were performed, and samples were run on the high-resolution quadrupole mass spectrometer for analysis. The samples were diluted with MeOH (1.5 mL) and analyzed via direct injection with a solvent mix of 50%/50% MeOH/H_2O and a 2 min total run time: HR ESI-TOF Mass (negative mode) *m/z*= 150.0883 (calc for $C_9H_9D_2NO$ = 150.0882 [*M*]-) for *N*-benzylacetamide.

Anaerobic dabbing experimental setup. A Cole-Palmer glove bag (37”x37”x25”) was used for all anaerobic experiments. The glove bag contained the electronic nail used in previous dabbing experiments, the dabbing glassware, and the compound of interest (for this experiment ethyl acetate or geranyl acetate was used). Three different lines were connected and sealed to the glove bag. One end of the bag was connected to the fume hood air line as well as another connection to an ultra-pure N_2 gas cylinder so switchover could occur without breaking the seal on the system. The other side of the glove bag was connected to the fume hood vacuum. Initial pressurization of the bag was achieved by closing the front zipper on the bag and taping the gas lines to ensure a proper seal. Air was allowed to flow into the bag at 2 PSI until the bag was inflated to a suitable working size. After a 5 min equilibration period the e-nail was turned on and allowed to heat to 378 °C. Once the desired temperature had been reached by the quartz surface (e-nail), 500 ul of ethyl acetate was administered onto the nail. The aerosol was pulled into an impinger containing the same solvent ratio as described in the section above. After 5 min the impinger was disconnected from the system and the solvent transferred to an NMR tube for analysis. After the initial samples were collected under normal atmospheric conditions, the air line was turned off and the ultra-pure nitrogen line was opened up and allowed to flow into the bag at 2 PSI while the residual air was pulled out via the fume hood vacuum at the same rate. The system was allowed to normalize to anaerobic conditions (0.0% O_2 as measured by the Vzmcov 4 in 1 gas detector). Once optimal conditions were obtained the vacuum was turned off and the same dabbing experiment was again conducted under N_2. Analysis of the impinger solvent system via NMR was again performed.



<u>Figure S1:</u> NMR spectrum showing *N*-benzylamide formation upon vaping CBN-OAc at lower temperature. An additional experiment to the one outlined in the methodology section was performed on CBN-OAc with the only change being a lowered e-nail temperature (from 378 °C) of 287 °C. Upon analysis of the impinger solution, a doublet was observed to have formed centered at 4.39 ppm, indicating that the benzylamine trapping agent had reacted with the generated ketene to form *N*-benzylacetamide.

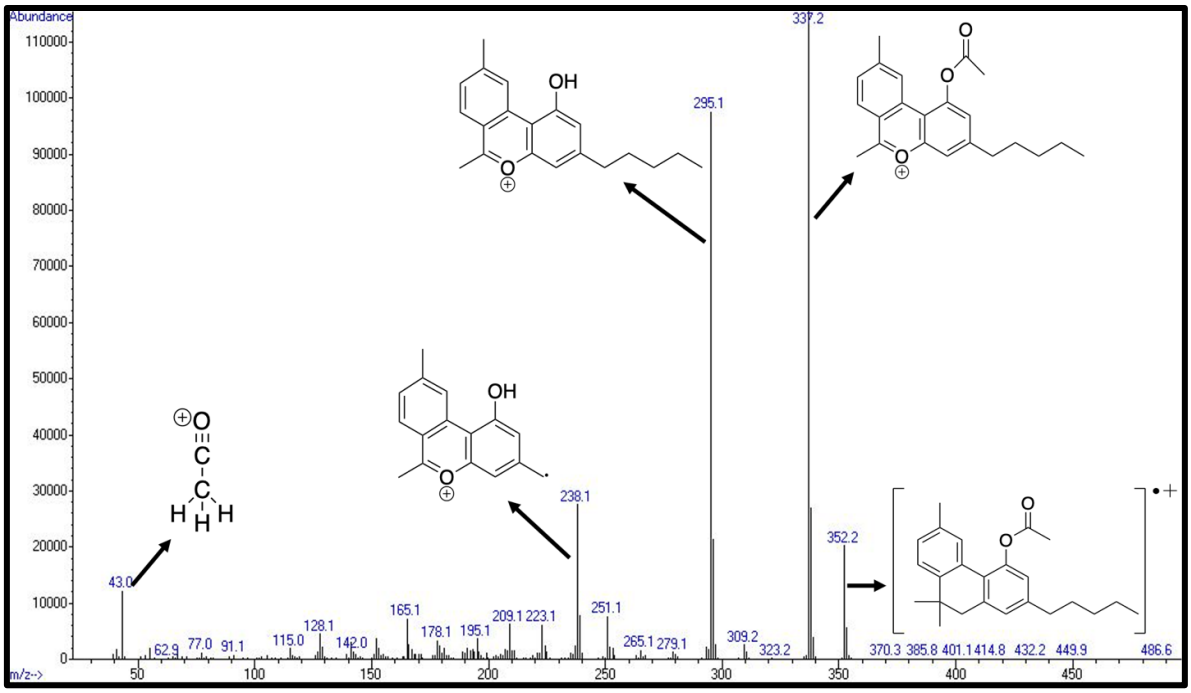


Figure S2: GCMS fragmentation of CBN-OAc starting material, along with assigned fragments. Note the loss of ketene (42 amu) from the base peak.

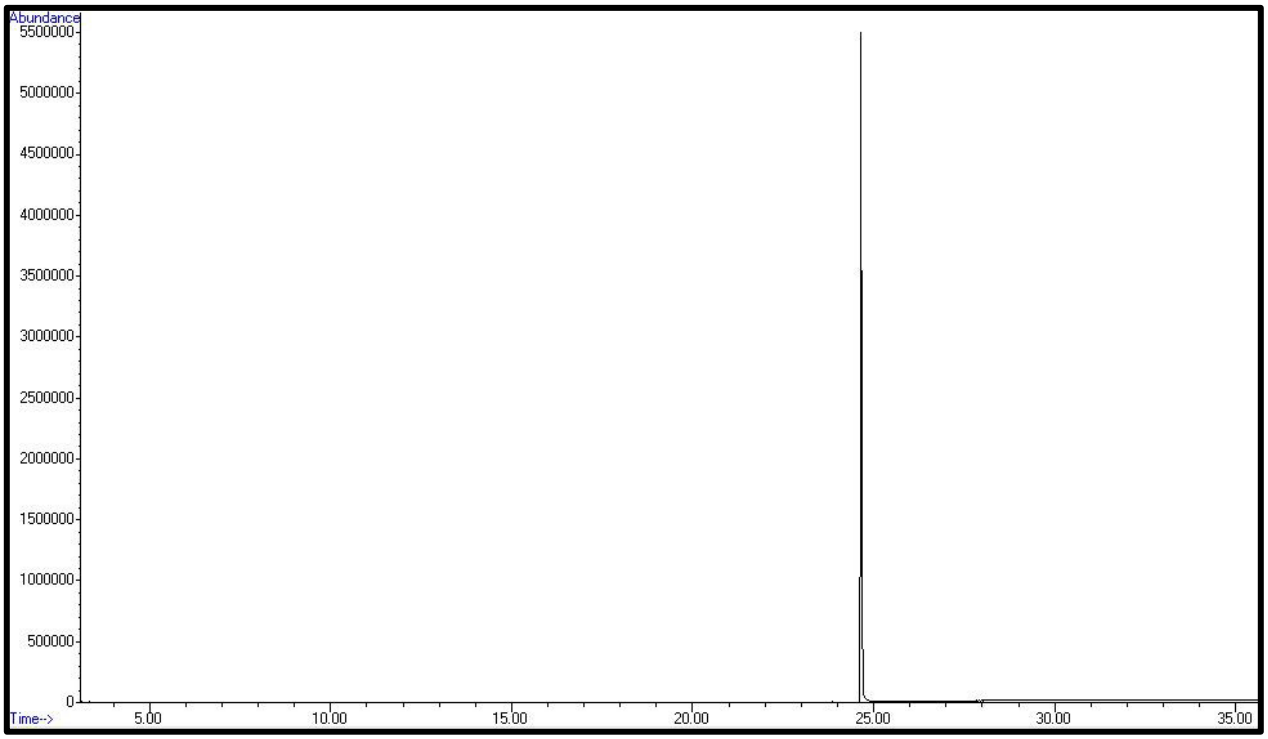
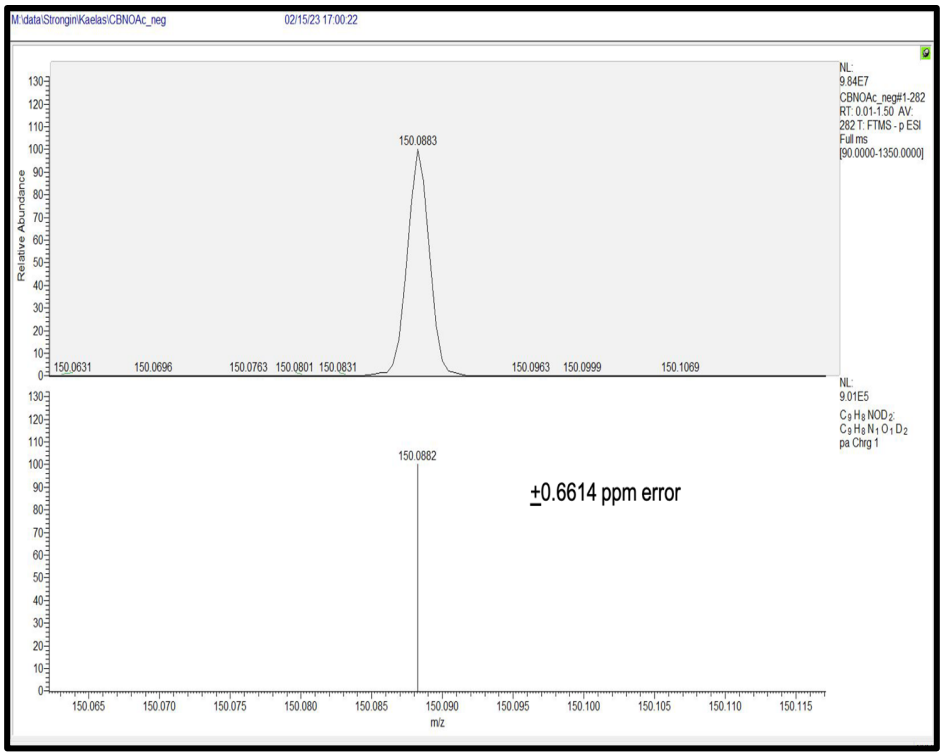


Figure S3: GCMS chromatogram of synthesized CBN-OAc-D_3. The corresponding mass fragmentation obtained from this sample is shown in the main text (Figure 2).



<u>Figure S4:</u> LCMS chromatogram of the trapping (impinger) solution after vaping CBN-OAc-(D_3). The theoretical product upon reaction with ketene should yield *N*-benzylacetamide-D_2H, M/Z=150.0882 (Bottom spectrum). The top spectrum corresponds to the LCMS analysis of the sample in question M/Z=150.0883.

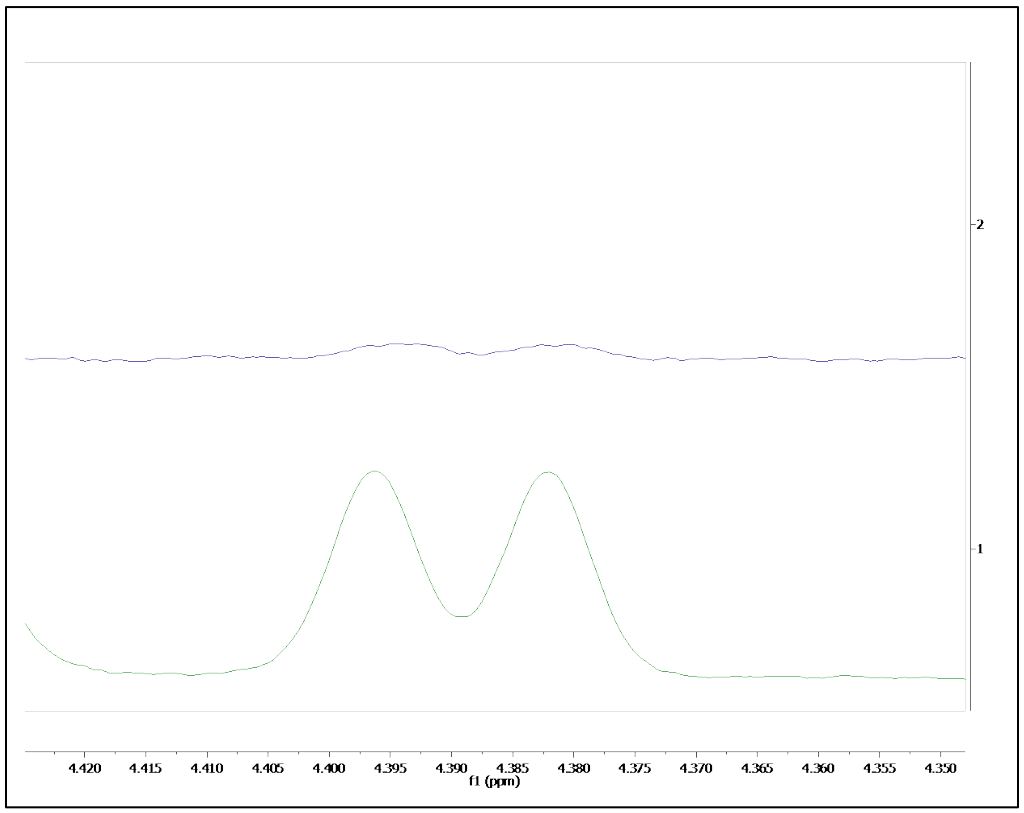


Figure S5: The top (purple) ^{1}H NMR spectrum shows *N*-benzylacetamide generated from dabbed geranyl acetate at 378 °C under an anaerobic environment containing O_2 levels below LOD (0.0% O_2 reported from Vzmcov sensor). The bottom (green) ^{1}H NMR spectrum is of dabbed geranyl acetate at 378 °C under ambient atmospheric and environmental conditions (20.9% O_2 reported from Vzmcov sensor). QNMR analysis of both samples (500 ul geranyl acetate) yielded 0.0045 mg ketene under anaerobic/N_2 conditions and 0.089 mg of ketene formed under ambient environmental conditions.

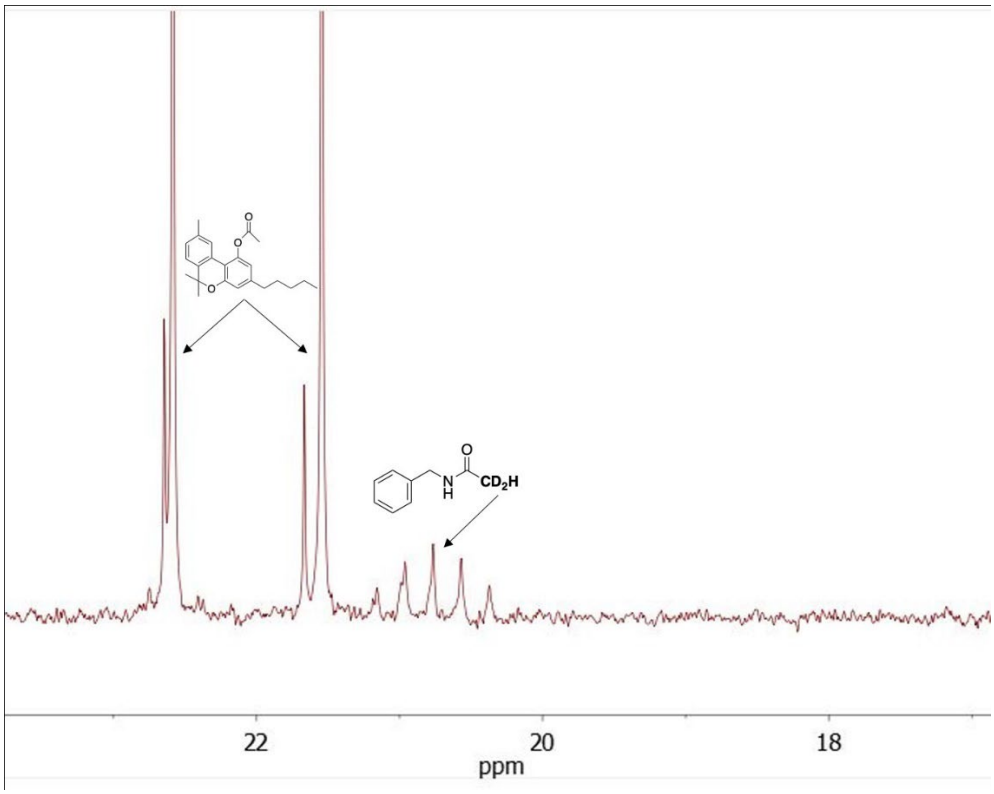


Figure S6: Spectrum of the ^{13}C NMR spectrum (CDCl3) of the quintet shown in text Figure 3.