

1 **Characterization of polar organosulfates in secondary organic aerosol from**
2 **the green leaf volatile 3-Z-hexenal**

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12

13 **ABSTRACT**

14 Evidence is provided that the green leaf volatile 3-Z-hexenal serves as a precursor for biogenic
15 secondary organic aerosol through formation of polar organosulfates (OSs) with molecular
16 weight (MW) 226. The MW 226 C₆-OSs were chemically elucidated, along with structurally
17 similar MW 212 C₅-OSs, whose biogenic precursor is likely related to 3-Z-hexenal but still
18 remains unknown. The MW 226 and 212 OSs have a substantial abundance in ambient fine
19 aerosol from K-pusztá, Hungary, which is comparable to that of the isoprene-related MW 216
20 OSs, known to be formed through sulfation of C₅-epoxydiols, second-generation gas-phase
21 photooxidation products of isoprene. Using detailed interpretation of negative ion electrospray
22 ionization mass spectral data, the MW 226 compounds are assigned to isomeric sulfate esters of
23 3,4-dihydroxyhex-5-enoic acid with the sulfate group located at the C-3 or C-4 position. Two

24 MW 212 compounds present in ambient fine aerosol are attributed to isomeric sulfate esters of
25 2,3-dihydroxypent-4-enoic acid, of which two are sulfated at C-3 and one is sulfated at C-2. The
26 formation of the MW 226 OSs is tentatively explained through photooxidation of 3-Z-hexenal in
27 the gas phase resulting in an alkoxy radical, followed by a rearrangement, and subsequent
28 sulfation of the epoxy group in the particle phase.

29 30 INTRODUCTION

31 Much information is available about secondary organic aerosol (SOA) formation from terpenes,
32 including mono- and sesquiterpenes, and isoprene [for a review, see (1)]. However, information
33 about SOA formation from green leaf volatiles (GLVs), an important class of biogenic volatile
34 organic compounds (BVOCs), is very scarce. To our knowledge, only SOA formation from the
35 GLVs 3-Z-hexen-1-ol and 3-Z-hexenyl acetate has been examined (2-4). In a previous study, we
36 presented evidence that a polar organosulfate (OS) related to the C₅ plant volatile, 2-pentenal,
37 which is a photolysis product of 3-Z-hexenal (5), occurs in ambient fine aerosol from a forested
38 site, i.e., K-pusztá, Hungary; more specifically, a sulfate ester of 2,3-dihydroxypentanoic acid
39 could be identified using liquid chromatography combined with negative ion electrospray
40 ionization mass spectrometry [LC/(-)ESI-MS] and detailed interpretation of the MS data (6).
41 Here, we provide evidence that the unsaturated aldehydic GLV, 3-Z-hexenal, is a potential
42 precursor for biogenic SOA through formation of organosulfates.

43 A small number of VOCs are naturally released from plants such as isoprene and terpenes.
44 However, many more VOCs are emitted when plants are wounded or attacked by insects. The
45 chemical characteristics of these VOCs differ with the plant species and with the herbivorous
46 insect species, and both parasitic and predatory insects, natural enemies of herbs, are attracted by
47 these volatiles (7, 8). GLVs also have a potential role in tropospheric chemistry as they may

48 serve as precursors for ozone and SOA (9). 3-Z-hexenal is an important GLV formed in green
49 leaves, from the unsaturated fatty acid α -linolenic acid in cell membranes, by the combined
50 reaction of lipoxygenase and hydroperoxide lyase enzymes (10). It is known to preferentially
51 react in the atmosphere during daytime through reaction with the OH radical and to a lesser
52 extent with ozone (11, 12).

53 Organosulfate formation has been well documented for SOA from the photooxidation of
54 isoprene (6, 13-16) and proceeds in the particle phase via sulfation of intermediary epoxy
55 derivatives (15, 17, 18) or reaction of first-generation oxidation products (i.e., methacrolein and
56 methyl vinyl ketone) with the sulfate anion radical (19-22). The most abundant isoprene-related
57 OSs in ambient fine aerosol from forested sites during summer are sulfate esters of the 2-
58 methyltetrols (14, 23). Similar to the polar OSs related to SOA from the photooxidation of
59 isoprene, those originating from the photooxidation of unsaturated aldehydes may also be
60 hydrophilic and substantially contribute to the total SOA budget. Polar OSs are of climatic
61 interest due to their capability to enhance the hydrophilic properties of the aerosol, and, hence,
62 their cloud-forming properties (24). Furthermore, the novel OSs are potential marker compounds
63 for SOA formation occurring under acidic conditions by particle-phase reactions with sulfuric
64 acid (25), formed by oxidation of sulfur dioxide, which is mainly from anthropogenic origin in
65 continental regions of the globe (26).

66 In the present study, we have investigated organosulfate formation from 3-Z-hexenal by
67 conducting smog chamber experiments in the presence of acidic ammonium sulfate seed aerosol
68 and have focused on the structural characterization of MW 226 OSs, which are also present at a
69 substantial relative abundance in ambient fine aerosol. In addition, attention was given to the
70 chemical characterization of abundant MW 212 OSs, which are lower homologs of the MW 226

71 3-Z-hexenal-related OSs but whose biogenic precursor(s) remain(s) to be identified. Abundant
72 MW 212 OSs have been noted in earlier studies dealing with the chemical characterization of
73 OSs or biogenic SOA but are largely unknown so far (6, 14, 23, 27). Both MW 226 and 212 OSs
74 have been quantified in fine aerosol (PM_{2.5}) samples from Brasschaat, Belgium, and their median
75 concentrations were estimated at 6.5 ng m⁻³ and 4.6 ng m⁻³, comparing quite favorably with that
76 of 6.4 ng m⁻³ determined for the isoprene-related MW 216 OSs and thus suggesting that they
77 have a similar magnitude (23). Two different LC techniques were employed to separate the polar
78 OSs: the first technique uses a reversed-phase trifunctionally bonded C₁₈ stationary phase (16),
79 whereas the second one is based on ion-pairing C₁₈ LC using dibutylammonium acetate as ion-
80 pairing reagent. The latter technique has recently been applied to polar isoprene SOA-related
81 OSs and has been shown to provide an improved chromatographic separation for isomeric and
82 isobaric compounds compared to the first technique using a trifunctionally bonded C₁₈ phase
83 (28).

84 MATERIALS AND METHODS

85 *Chemicals*

86 Methanol (ULC/MS grade) used for sample preparation and as LC mobile phase and acetonitrile
87 (HPLC supra-gradient grade) were purchased from Biosolve NV (Valkenswaard, The
88 Netherlands); the ion-pairing (IP) reagent dibutylammonium acetate (DBAA, 0.5 mol/L in water;
89 for LC/MS) was from TCI (Tokyo, Japan); and *cis*-pinonic acid (purity: 98%; for MS signal
90 optimization) was from Sigma-Aldrich (St. Louis, MI, USA). High-purity water (resistivity, 18.2
91 MΩ cm; total organic carbon, 2 ppb) used for redissolving aerosol extracts and preparing the
92 aqueous LC mobile phase was prepared with a Milli-Q water purification system (Millipore,

93 Bedford, MA, USA). 3-Z-hexenal (50% solution in triacetin) used for chamber experiments was
94 from Sigma-Aldrich.

95 *Aerosol samples*

96 Laboratory SOA samples: The experimental conditions for the 3-Z-hexenal chamber experiments
97 used in the present study are summarized in Table 1. The experiments were conducted in a 14.5
98 m³ stainless-steel chamber with 40 µm Teflon-coated walls (29). The temperature, aerosol size
99 distribution, and relative humidity, as well as the O₃, nitric oxide (NO) and NO_x concentrations
100 were continuously measured. OH radicals were generated from the NO_x-mediated photochemical
101 chain reactions. Different seed aerosols were used to create different acidity levels in the
102 chamber, either sulfuric acid (SA) alone or a mixture of sulfuric acid and ammonium sulfate
103 (AS) (1/1; w/w), which were introduced by nebulization from aqueous solutions using a TSI
104 Model 9302 nebulizer (TSI Incorporated, Shoreview, MN, USA). Experiments were conducted
105 in the dynamic mode (i.e., continuously stirred tank reactor) with a residence time of
106 approximately 4 hr. NO was continuously added from a high-pressure cylinder to the reaction
107 chamber through a mixing manifold. 3-Z-hexenal was introduced by air flow through the neat
108 liquid in a temperature-controlled bath. The steady-state nature of the chamber operation allows
109 for filter sampling for extended periods, providing sufficient aerosol mass for determining the
110 composition of the resultant SOA. Once steady-state conditions were attained (24 h), samples for
111 determining the composition of the SOA were collected on Teflon-impregnated glass fiber filters
112 (Pallflex Fiberfilm, Pall Corporation, Port Washington, NY, USA). The filters were extracted
113 three times with 10 mL of methanol in an ultrasonic bath. The extracts were combined and
114 concentrated in a rotary evaporator at 35 °C and 200 mbar to approximately 1 mL, filtered
115 through a Teflon filter (0.45 µm), and evaporated to dryness under a nitrogen stream. The residue

116 was redissolved in 1 mL of methanol and divided in two portions, which were dried under
117 nitrogen, and one of the portions was stored at $-20\text{ }^{\circ}\text{C}$ for possible later use. One portion was
118 redissolved in 250 μL of methanol/water (1:1; v/v) by first dissolving it in 125 μL of methanol
119 and then adding 125 μL of water. In the case of ion-pairing LC/MS, the IP reagent was placed in
120 the injection solution by adding 10 μL of 0.5 mol L^{-1} DBAA to 90 μL of the above redissolved
121 sample. Afterwards, the sample was analyzed with LC/(-)ESI-MS using an aliquot of 5 μL .

122 [Table 1]

123 Ambient fine aerosol: Archived $\text{PM}_{2.5}$ (particulate matter with an aerodynamic diameter ≤ 2.5
124 μm) aerosol samples collected from K-pusztá, Hungary, during the BIOSOL (Formation
125 mechanisms, marker compounds, and source apportionment for biogenic atmospheric aerosols)
126 campaign between 22 May and 29 June 2006 were used. Information about the site and the
127 sampling campaign can be found elsewhere (30). Day- and night-time samples were collected on
128 quartz fiber filters using a high-volume dichotomous sampler providing two size fractions, a fine
129 ($\text{PM}_{2.5}$) and a coarse size fraction (with aerodynamic diameter $> 2.5\text{ }\mu\text{m}$). Sections of the $\text{PM}_{2.5}$
130 filter samples from different days and/or nights of the warm period of the campaign were
131 extracted three times for 30 min in an ultrasonic bath with 20 mL of methanol. The extracts were
132 combined and concentrated in a rotary evaporator at $35\text{ }^{\circ}\text{C}$ and 200 mbar to approximately 1 mL
133 and filtered through a Teflon filter (0.45 μm), and evaporated to dryness under a nitrogen stream.
134 The residue was redissolved in 1 mL of methanol and divided in five equal portions, which were
135 dried under nitrogen, and the residues were stored at $-20\text{ }^{\circ}\text{C}$ until use on different experimental
136 days. Before each series of experiments, a portion containing 570 μg of organic carbon, as
137 determined with a thermal-optical method (31), was redissolved in 1.5 mL of methanol/water
138 (1:4; v/v) by first dissolving it in 0.3 mL of methanol and then adding 1.2 mL of water. In the

139 case of ion-pairing LC/MS, the IP reagent was placed in the injection solution by adding 10 μL
140 of 0.5 mol L^{-1} DBAA to 90 μL of the redissolved sample. Afterwards, the sample was analyzed
141 with LC/(-)ESI-MS using an aliquot of 5 μL .

142 *Liquid Chromatography/Mass Spectrometry*

143 Liquid Chromatography: The LC system consisted of a Surveyor Plus system (pump and
144 autosampler) (Thermo Scientific, San Jose, USA). Two types of columns were employed, an
145 Atlantis C18 T3 column (3 μm ; 2.1 x 150 mm) (Waters, Milford, USA), containing
146 trifunctionally-bonded C18 alkyl chains, for regular reversed-phase (RP) LC, and an Hypersil
147 C18 Gold column (3 μm ; 2.1 x 150 mm) (Thermo Scientific, PA, USA) for ion-pair RP-LC. The
148 mobile phases for regular RP-LC consisted of 50 mM ammonium formate buffer pH 3 (A) and
149 methanol (B). The applied 60-min gradient elution program was as follows: the concentration of
150 eluent B was kept at 3% for 5 min, then increased to 95% in 15 min, kept at 95% for 25 min,
151 then decreased to 3% in 10 min, and kept at 3% for 10 min. The mobile phases for ion-pair RP-
152 LC consisted of 50 mM DBAA in water (A), acetonitrile (B), and water (C). The applied 70-min
153 gradient elution program was as follows: the concentration of eluent A was kept at 10% during
154 the whole 70 min program; the concentration of water (C) was kept at 3% for 10 min, then
155 increased to 87% in 15 min, kept at 87% for 25 min, then decreased to 3% in 10 min, and kept at
156 3% for 10 min. The injection volume and flow rate were 5 μL and 0.2 mL min^{-1} . The advantages
157 of both chromatographic techniques have been reported in earlier studies (6, 16, 28). However, it
158 should be cautioned that the ion-pairing technique rapidly contaminates the electrospray
159 ionization source.

160 Mass Spectrometry: A linear ion-trap mass spectrometer (LXQ, Thermo Scientific) was operated
161 under the following conditions: sheath gas flow (nitrogen), 50 arbitrary units; auxiliary gas flow

162 (nitrogen), 5 arbitrary units; source voltage, -4.5 kV; capillary temperature, 350 °C; and
163 maximum ion injection time, 200 ms. For MS^2 and MS^3 experiments, an isolation width of 2 m/z
164 units and a normalized collision energy of 35% were applied. The $[M - H]^-$ signal optimization
165 was done by introducing a 50 $\mu\text{g mL}^{-1}$ *cis*-pinonic acid standard solution. Data were acquired
166 and processed using Xcalibur 2.0 software.

167 Accurate mass measurements were carried out using an LTQ-Orbitrap mass spectrometer
168 (Thermo Scientific), equipped with a Waters Alliance 2695 HPLC system (Waters, Milford,
169 Massachusetts, USA). The LTQ Orbitrap mass spectrometer was equipped with an ESI source
170 operated in the negative ion mode under the same conditions as described above for the ion-trap
171 experiments. The source parameters were tuned for maximum sensitivity using a 50 $\mu\text{g mL}^{-1}$
172 malic acid standard solution. The mass resolution was set at $100,000$ in the MS^1 mode and $7,500$
173 in the MS^2 and MS^3 modes. For MS^2 and MS^3 experiments, an isolation width of 5 m/z units and
174 a normalized collision energy level of 35% were applied. The accurate mass measurements were
175 obtained using external calibration, providing a mass accuracy better than 1 mDa. The accurate
176 mass measurements were only carried out for K-pusztá $PM_{2.5}$ aerosol analyzed with reversed-
177 phase LC for selected peaks. Data were acquired and processed using Xcalibur 2.0 software.

178 RESULTS AND DISCUSSION

179 Figure 1 shows selected LC/MS chromatographic data using regular RP-LC obtained for K-
180 pusztá fine aerosol, including a base peak chromatogram (BPC) and extracted ion
181 chromatograms (EICs), which correspond to the deprotonated forms of OSs related to isoprene
182 (m/z 215), (an) unknown BVOC precursor(s) (m/z 211) and 3-Z-hexenal (m/z 225). Of these, the
183 m/z 215 isoprene SOA-related OSs have been identified in previous studies as sulfate esters of 2-
184 methyltetrols (6, 13-15), which are formed by sulfation of intermediary C_5 -epoxydiols (15). The

185 LC/MS chromatographic data reveal that the targeted OSs at m/z 225 and 211 have a relative
186 abundance (based on signal intensities) comparable to the m/z 215 compounds. It has already
187 been demonstrated in a previous study (28) that the m/z 215 isoprene-related OSs could be
188 resolved in two peaks with the ion-pair RP-LC technique; with the latter technique an improved
189 separation could also be obtained for the m/z 225 and 211 OSs, as will be illustrated in the
190 following two sections. Figure 1 also shows selected LC/MS chromatographic data for 3-Z-
191 hexenal SOA [sample ER627-GF10 (Table 1)] using the RP-LC technique; comparable results
192 were obtained for sample ER627-GF6 (results not shown).

193 [Figure 1]

194 **Structural characterization of MW 226 organosulfates related to 3-Z-hexenal**

195 It can be seen from Figure 1 (m/z 225 EIC) that there are several unresolved peaks eluting
196 between 2 and 6 min of which the major one ($C_6H_9O_7S$; measured mass: 225.00809; error: +0.6
197 mDa; K-pusztá fine aerosol) eluting at a retention time (RT) of 5.3 min is present in 3-Z-hexenal
198 SOA (RT 5.4 min); hence, the m/z 225 OSs can be related to 3-Z-hexenal. Selected MS data
199 (MS^2 and MS^3) for the major peak are given in Figure 2(A-D). Additional MS data for the early-
200 eluting m/z 225 compounds are presented in Figure 3. Figure 2(E, F) illustrates that the ion-pair
201 RP-LC technique provides a separation of the m/z 225 isomers. It can be noted that there is a
202 difference in the chromatographic profiles: ambient fine aerosol reveals a more complex pattern
203 with at least six isomers, whereas 3-Z-hexenal shows four distinct isomers with corresponding
204 RTs. A possible explanation for the more complex m/z 225 isomeric pattern of ambient fine
205 aerosol could be the presence of isomers originating from 2-Z-hexenal, which is a commonly
206 occurring plant leaf volatile (10). The differences in the relative abundances of characteristic ions
207 in the spectra for the early-eluting m/z 225 isomers in RP-LC (Fig. 3) are also likely due to a

208 different isomeric pattern between ambient fine aerosol and 3-Z-hexenal SOA. Detailed
209 interpretation of the MS data allows the assignment of the m/z 225 OSs to isomeric forms of
210 sulfate esters of 3,4-dihydroxyhex-5-enoic acid, with the sulfate group located at the C-3 or C-4
211 position [Scheme 1(A)]. The presence of the bisulfate [HSO_4^-] ion (m/z 97) in both the MS^2 and
212 MS^3 spectra (Fig. 2) is consistent with an organosulfate (6, 13-16, 32, 33), whereas the loss of
213 CO_2 (44 u) upon fragmentation of the precursor ion (m/z 225), affording m/z 181, indicates a
214 carboxyl group. Other diagnostic fragmentations include the loss of 44 u ($\text{C}_2\text{H}_4\text{O}$; m/z 137) from
215 one of the m/z 181 isomeric forms [Scheme 1(A), right], pointing to a terminal 1-hydroxyethyl
216 group (16), as well as the further loss of 28 u (C_2H_4 ; m/z 153) from both m/z 181 isomeric forms,
217 consistent with a terminal ethylene group. It is worth noting that the m/z 225 \rightarrow m/z 181 MS^3
218 spectrum obtained for ambient fine aerosol shows an abundant m/z 181 precursor ion, whereas
219 the latter ion is completely fragmented upon MS^3 in the case of 3-Z-hexenal SOA, consistent
220 with the observation that ambient fine aerosol shows a more complex m/z 225 EIC profile upon
221 ion-pairing RP-LC. The detection of multiple isomers in 3-Z-hexenal SOA upon ion-pairing RP-
222 LC is in agreement with the structural proposal as the two positional isomers with the sulfate
223 group positioned at C-3 or C-4 each can occur as diastereoisomeric pairs.

224 [Figure 2]

225 [Figure 3]

226 [Scheme 1]

227 **Structural characterization of MW 212 organosulfates related to (an) unknown BVOC** 228 **precursor(s)**

229 Based on accurate mass measurement of the m/z 211 compounds (RT 3.5 min) in K-pusztá fine
230 aerosol ($\text{C}_5\text{H}_7\text{O}_7\text{S}$; measured mass: 210.99252; error: +0.7 mDa), it can be concluded that these

231 compounds are lower homologs of the 3-Z-hexenal SOA-related MW 226 OSs ($C_6H_9O_7S$). As
232 can be seen in Figure 4(A), four fully resolved peaks with comparable signal intensity could be
233 obtained for ambient fine aerosol with the ion-pairing RP-LC technique. However, none of these
234 peaks were found to correspond to m/z 211 compounds present in 3-Z-hexenal SOA. Hence, the
235 MW 212 compounds from ambient fine aerosol do not originate from 3-Z-hexenal SOA. Here,
236 we only discuss the data for the ambient fine aerosol and show that one of the MW 212
237 compounds is structurally related to the 3-Z-hexenal SOA-derived MW 226 OSs. The presence
238 of the bisulfate [HSO_4^-] ion (m/z 97) in the m/z 211 MS^2 spectra for the four compounds [Fig.
239 4(B, C, D and E)] is in agreement with organosulfates; in addition, the m/z 211 MS^2 spectrum for
240 the second-eluting isomer [Fig. 4(C)] shows an ion at m/z 113, due to the combined loss of water
241 (m/z 193) and SO_3 (80 u), of which the latter supports an organosulfate (6, 32, 33). Detailed
242 interpretation of the MS data [Fig. 4(C, F)] allowed the assignment of the second-eluting MW
243 212 compound to a sulfate ester of 2,3-dihydroxy-4-pentenoic acid with the sulfate group located
244 at C-3 [Scheme 1(B)]. A minor corresponding diastereoisomer with a similar fragmentation
245 pattern was present at RT 11.1 min (results not shown). The third-eluting MW 212 compound
246 (RT 8.3 min) only showed limited fragmentation through loss of CO_2 (m/z 167) and formation of
247 m/z 97 [Fig. 4(D, G)]; hence, its attribution to a positional isomer, i.e., a sulfate ester of 2,3-
248 dihydroxy-4-pentenoic acid with the sulfate group located at C-2 should be regarded as tentative
249 [Scheme 1(B, right)].

250 [Figure 4]

251 **Formation pathways for the organosulfates related to 3-Z-hexenal SOA**

252 The MW 226 compounds could be assigned to OSs of 3,4-dihydroxyhex-5-enoic acid with the
253 sulfate group positioned at C-3 or C-4. A possible formation pathway leading to these isomeric

254 organosulfates is presented in Scheme 2. The proposed pathway involves the formation of a
255 radical species with the radical located at the C-3 position [species (1)], followed by reaction
256 with oxygen and NO, resulting in a C₆ alkoxy radical [species (2)]. The latter species may
257 undergo a rearrangement, resulting in an unsaturated C₆-epoxy carboxylic acid [species (3)].
258 Subsequent sulfation of the epoxy group in the particle phase leads to isomeric organosulfates of
259 3,4-dihydroxyhex-2-enoic acid with the sulfate group positioned at C-3 or C-4. The formation of
260 the intermediate radical species (1) is proposed to proceed through OH abstraction of the
261 aldehydic hydrogen atom, reaction with oxygen and an intramolecular hydrogen rearrangement
262 reaction. With regard to hydrogen abstraction from the aldehydic group, it is known that the OH
263 reaction with 2-*E*-hexenal leads to both OH addition to the double bond and hydrogen
264 abstraction from the aldehydic group (11); however, such data are not available for 3-*Z*-hexenal.
265 An alternative mechanistic route leading to the formation of OSs from 3-*Z*-hexenal that was
266 considered and does not require epoxide formation is reactive uptake in the particle phase,
267 followed by reaction with the sulfate radical anion (19, 20). This pathway was proposed for the
268 formation of OSs from the isoprene gas-phase oxidation products methacrolein and methyl vinyl
269 ketone (20-22). However, it results in saturated OSs and thus does not allow to explain the
270 formation of the unsaturated OSs found for 3-*Z*-hexenal in the current study.

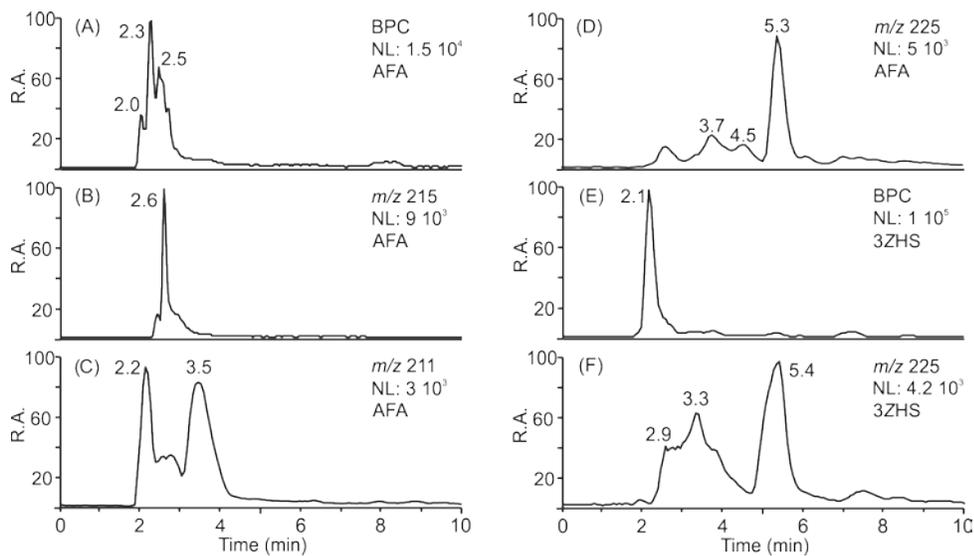
271 [Scheme 2]

272 **Atmospheric implications**

273 In the present study, we have demonstrated that the plant volatile 3-*Z*-hexenal serves as a
274 precursor for polar organosulfates with MW 226, which occur in ambient fine aerosol at
275 concentrations comparable to those of the isoprene SOA-related 2-methyltetrol OSs. The source
276 of the MW 212 OSs, which have a substantial abundance in ambient fine aerosol and comprise

277 one isomer that is structurally related to the MW 226 OSs, remains to be established but is likely
278 a plant volatile related to 3-Z-hexenal. The formation of the MW 226 OSs from 3-Z-hexenal is
279 tentatively explained through a rearrangement of an alkoxy radical into an epoxy carboxylic acid
280 in the gas phase and subsequent sulfation of the epoxy group with sulfuric acid in the particle
281 phase. Further research is warranted to explore this novel SOA formation pathway for BVOCs.
282 The formation of OSs through epoxy intermediates is in line with a previously suggested
283 pathway for the formation of OSs that are related to isoprene [i.e., the 2-methyltetrols (*15*) and 2-
284 methylglyceric acid (*17*)], β -pinene [i.e., β -pinanediol (*34*)], and 2-methyl-3-buten-2-ol [i.e., 2,3-
285 dihydroxyisopentanol (*35*)]. We speculate that, in addition to 3-Z-hexenal, other plant and floral
286 volatiles serve as precursors for polar organosulfates. In this context, it would be worthwhile to
287 examine commonly occurring green leaf and floral volatiles as biogenic sources for polar
288 organosulfates that are present in ambient fine aerosol and have not yet been elucidated.
289

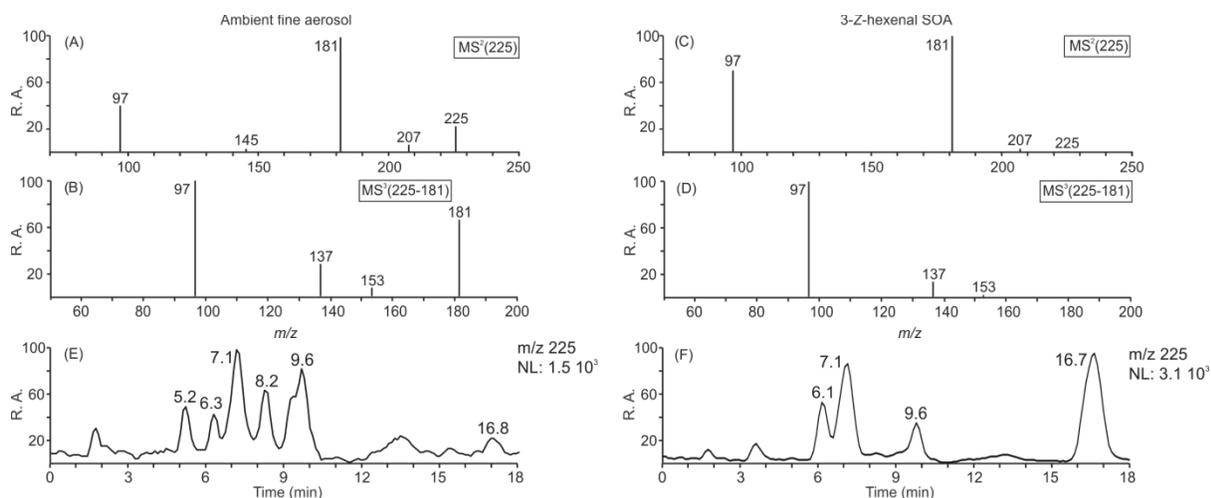
290 FIGURES



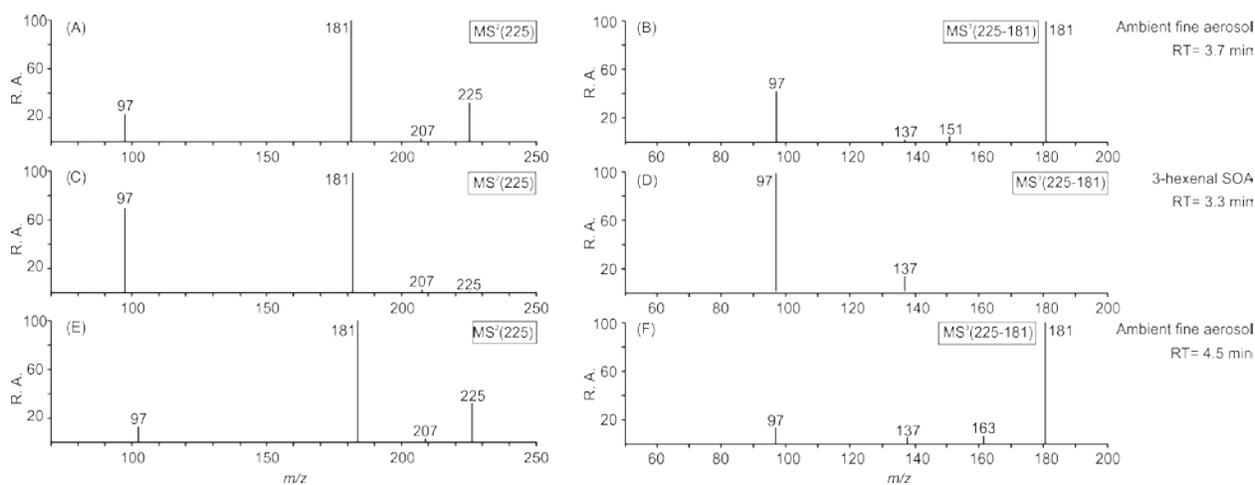
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292 **Figure 1.** Selected LC/MS chromatographic data using regular RP-LC obtained for ambient fine
 293 aerosol (A-D) and 3-Z-hexenal SOA (E,F), including a base peak chromatogram (BPC) and
 294 extracted ion chromatogram (EICs), corresponding to the deprotonated forms of OSs related to
 295 isoprene (m/z 215), 3-Z-hexenal (m/z 225) and (an) unknown biogenic volatile organic
 296 compound precursor(s) (m/z 211). The m/z 211 EIC for 3-Z-hexenal is not included because no
 297 peaks corresponding with those observed for ambient fine aerosol could be seen with the ion-
 298 pairing RP-LC technique. Abbreviations: AFA, ambient fine aerosol; NL, normalization level;
 299 3ZHS, 3-Z-hexenal SOA.

300



301
 302 **Figure 2.** Selected MS data (MS^2 or MS^3 product ion spectra) obtained with the regular RP-LC
 303 technique for (A, B) the compounds eluting at 5.3 min for ambient fine aerosol and (C, D) the
 304 compounds eluting at 5.4 min in 3-Z-hexenal SOA [Fig. 1; m/z 225 EIC]. LC/MS
 305 chromatographic data (m/z 225 EICs) obtained with the ion-pairing RP-LC technique for (E)
 306 ambient aerosol and (F) 3-Z-hexenal SOA. Abbreviation: NL, normalization level.
 307



308
 309 **Figure 3.** MS^2 and MS^3 product ion data obtained for ambient fine aerosol and 3-Z-hexenal SOA
 310 using the regular RP-LC technique [Fig. 1(D and F)] for the m/z 225 compounds eluting at RTs
 311 3.3, 3.7 and 4.5 min before the major peak at 5.3 and 5.4 min, respectively. Abbreviation: RT,
 312 retention time.

313

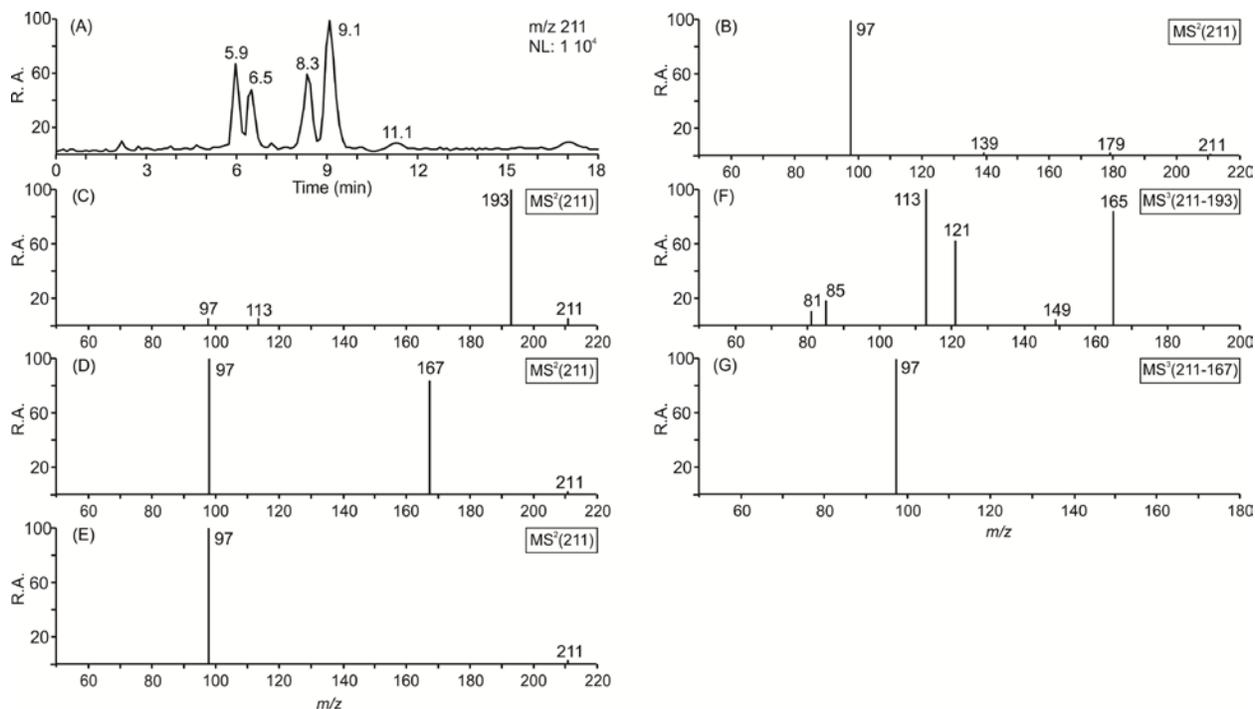
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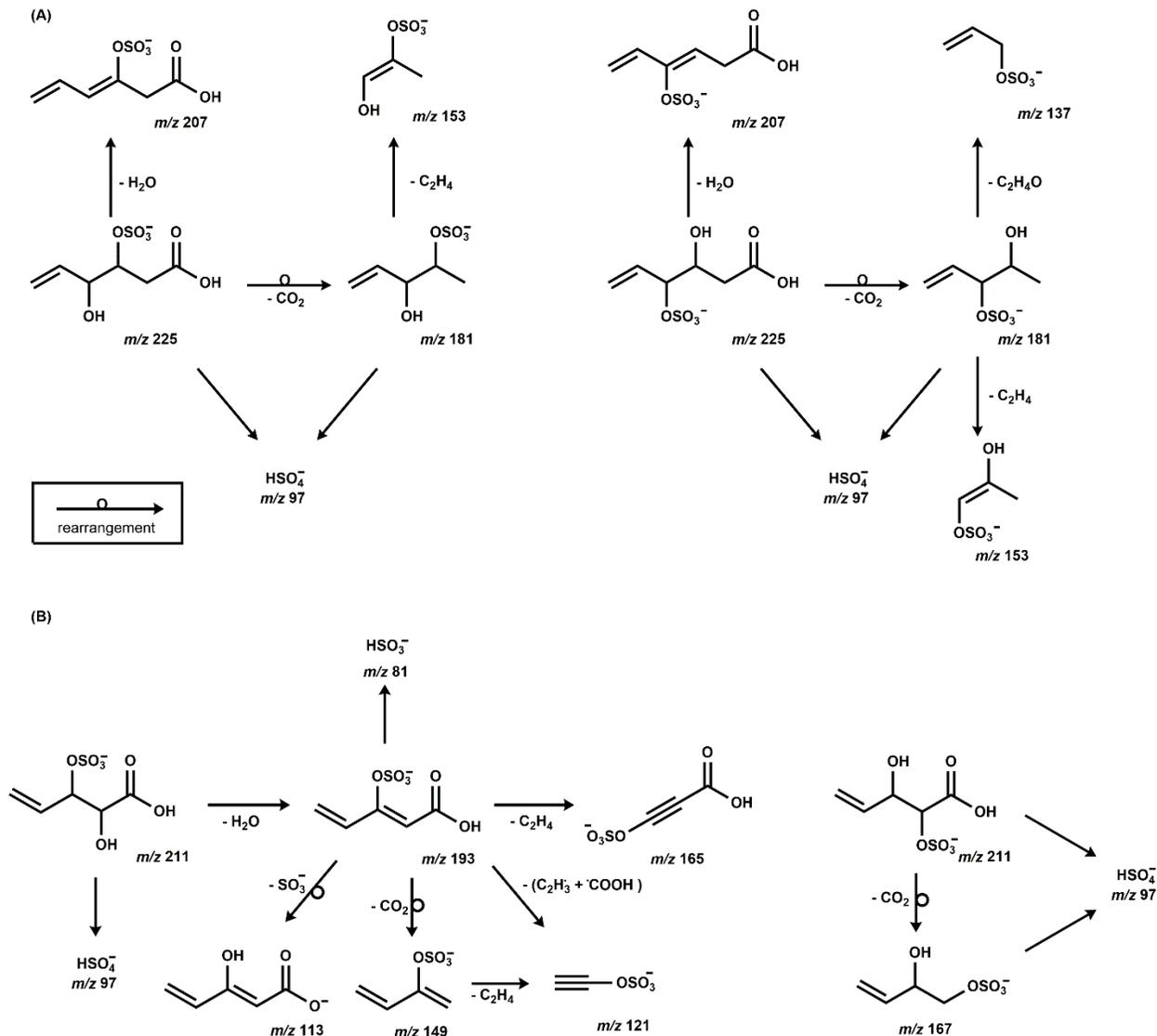
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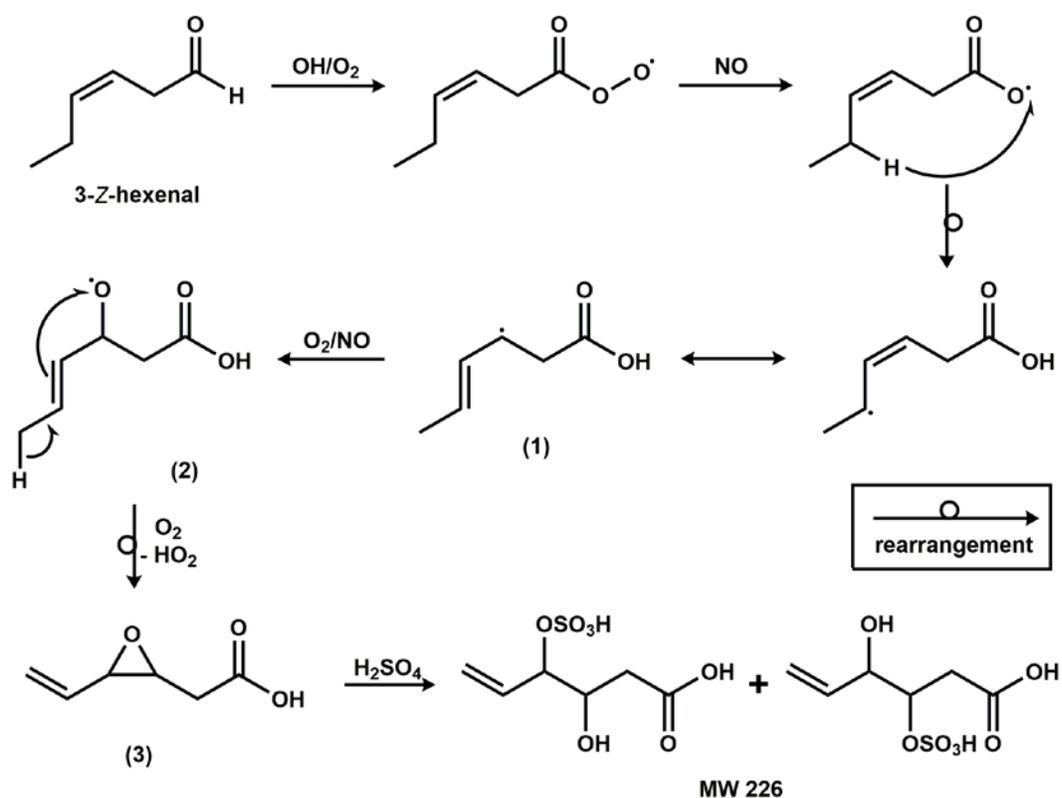
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320 **Figure 4.** Selected LC/MS chromatographic data (m/z 211 EIC) obtained with the ion-pairing
321 RP-LC technique for (A) ambient fine aerosol and selected MS data (MS² and MS³ product ion
322 spectra) for peaks eluting at (B) 5.9 min, (C, F) 6.5 min, (D, G) 8.3 min, and (E) 9.1 min.
323 Abbreviation: NL, normalization level.
324

325 SCHEMES
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330 **Scheme 1.** Proposed fragmentation routes for deprotonated (A) MW 226 compounds, related to
331 3-Z-hexenal SOA, which are assigned to sulfate esters of 3,4-dihydroxyhex-5-enoic acid with the
332 sulfate group at C-3 (left) and C-4 (right), and (B) MW 212 compounds present in ambient
333 aerosol, which are assigned to sulfate esters of 2,3-dihydroxy-4-pentenoic acid with the sulfate
334 group at C-3 (left) and positional isomers with the sulfate group located at C-2 (right).
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338 **Scheme 2.** Proposed formation pathway for the MW 226 organosulfates related to 3-Z-hexenal
 339 SOA, assigned to sulfate esters of 3,4-dihydroxyhex-5-enoic acid.

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343 TABLE

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345 **Table 1.** Experimental chamber conditions for experiments used for the structural
346 characterization of unknown organosulfates related to 3-Z-hexenal SOA. Abbreviations: SA,
347 sulfuric acid; AS, ammonium sulfate; RH, relative humidity.

Sample code		Hydrocarbon Conc.	NO _x * Conc.	O ₃ Conc.	OC Conc.	Seed type Conc.	Other Conditions
ER627 – GF6	Initial:	2.3 ppmC	290 ppb	-	-	SA 30 µg m ⁻³	4 light banks 1079 nmol m ⁻³ [H ⁺]air RH: 29%
	Steady State:	0.02 ppmC	117 ppb	265 ppb	9 µg m ⁻³		
ER627 – GF10	Initial:	2.2 ppmC	290 ppb	-	-	½ AS + ½ SA 30 µg m ⁻³	4 light banks 439 nmol m ⁻³ [H ⁺]air RH: 29%
	Steady state:	0.02 ppmC	200 ppb	280 ppb	5 µg m ⁻³		

348 *All the initial NO_x was NO.

349

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356 **Notes**

357 The authors declare no competing financial interest.

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359 ACKNOWLEDGMENTS

360 Research at the University of Antwerp was supported by the Belgian Federal Science Policy

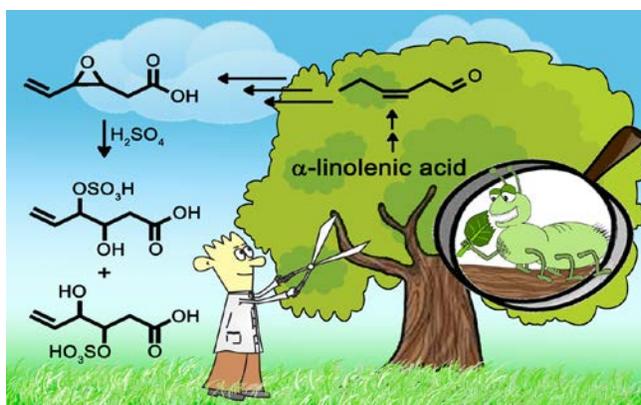
361 Office through the network project “Biogenic Influences on Oxidants and Secondary Organic

362 Aerosol: theoretical, laboratory and modelling investigations (BIOSOA)” and by the Research

363 Foundation – Flanders (FWO) through the project “Secondary Organic Aerosol formation from

364 monoterpenes: GAPS in our current understanding (SOAGAPS)”. The U.S. Environmental
365 Protection Agency through its Office of Research and Development partially funded and
366 collaborated in the research described here under contract EP-D-10-070 to Alion Science and
367 Technology. The manuscript has been subjected to Agency review and has been cleared for
368 publication. Mention of trade names or commercial products does not constitute an endorsement
369 or recommendation for use.

370 TOC GRAPHIC



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