

ISOLATION AND IDENTIFICATION OF TWO NEW  
SECO-CEMBRANOIDS FROM CIGARETTE SMOKE

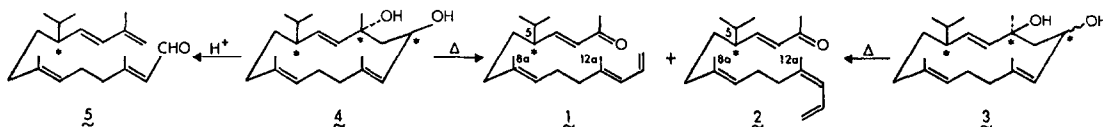
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ABSTRACT

The isolation and identification from cigarette smoke condensate of 5-isopropyl-8,12-dimethyl-3E,8E,12E,14-pentadecatetraen-2-one and its 12Z isomer are reported.

$\alpha$ -4,8,13-Duvatriene-1,3-diol, 3, and the  $\beta$ -diol, 4,<sup>1,2</sup> most likely derived from cembrene<sup>3</sup>, comprise 30-60% of the surface waxes<sup>4,5</sup> of immature tobacco leaves from duvane producing varieties of *Nicotiana tabacum*. These tobacco cembranoids<sup>6</sup> are drastically reduced during senescence and curing, coupled with a rapid increase in numerous organoleptically significant irregular isoprenoids.<sup>7</sup> The initial step in this series of degradative reactions is ring opening to form C<sub>20</sub> seco-cembranoids<sup>8</sup>, such as aldehyde 5, which can be formed from 4 by an acid catalyzed ring opening.<sup>9</sup>

We wish to report the isolation and identification of two new C<sub>20</sub> seco-cembranoids, 5-isopropyl-8,12-dimethyl-3E,8E,12E,14-pentadecatetraen-2-one, 1, and its 12Z isomer, 2, from cigarette smoke. Thermolysis of 3 or 4\*\* yields as major products both 1 and 2, significant as potential precursors of the complex of isoprenoids in cured tobacco.<sup>7,10</sup>



Isomers 1 and 2 were obtained by a separation scheme similar to that used to isolate solanoquinone.<sup>11</sup> First, smoke condensate was partitioned between methanol/water (9:1,v:v) and cyclohexane followed by partitioning of the cyclohexane residue between cyclohexane and nitromethane. Next, a fraction enriched in both isomers was obtained by silica gel (EM-60) column chromatography of the nitromethane residue with hexane, benzene and benzene/ethyl acetate (99:1,v:v). Preparative GLC (3% OV-22 on 100/120 Supelcoport) was used to isolate milligram quantities of 1 and 2.

The spectral data are consistent with structures 1 and 2. High resolution MS gave a molecular formula C<sub>20</sub>H<sub>32</sub>O [calculated for 1 and 2: 288.2453; found 288.2468 (1) and 288.2455 (2)].<sup>12</sup> The most significant data were the parent ion and ions showing losses of methyl, propyl and acetyl groups. Also present were unsaturated hydrocarbon ions and oxygen containing ions differing by CH, CH<sub>2</sub> or CH<sub>3</sub> units. A UV maximum at 230 nm (methanol) indicates the

\*\* Manuscript in preparation

$\alpha,\beta$ -unsaturated carbonyl group. IR bands ( $\text{cm}^{-1}$ ) indicate the following groups: butadiene (890), trans double bond (980), methyls (1360, 1370, 1387), methylene (1455),  $\alpha,\beta$ -unsaturated carbonyl (1680), isopropyl (2878, 2960) and a terminal methylene (3080).

Double bond positions were determined by ozonolysis in ethyl acetate at  $-78^\circ\text{C}$  followed by triphenylphosphine reduction of the nitrogen degassed reaction mixture. 5-Isopropyl-3-E-nonen-2,8-dione<sup>13</sup>, 5-oxo-2-isopropylhexanal<sup>14</sup>, levulinialdehyde<sup>15</sup> and a MW126 compound\* were identified by GC/EIMS and GC/CIMS analyses after ozonolysis of 1. Since 1 and 2 and their ozonolysis products were not isolated in sufficient quantities for optical rotation measurements, it was not possible to determine the configuration of these compounds. However, it is reasonable to assume their configuration is identical to (5S)-5-isopropyl-3-E-nonen-2,8-dione and to other northunberganoids isolated previously.<sup>16</sup>

The  $^1\text{H}$  NMR spectra (Table I) clearly show that the isopropyl group is attached to an asymmetric carbon. Proton resonances of three other methyls and seven olefinics are also present. Furthermore, the all trans geometry of 1, the trans, trans, cis geometry<sup>17</sup> of 2 and the butadiene group are indicated by analysis of the 360 MHz  $^1\text{H}$  olefinic resonances and comparison with model compounds<sup>18</sup>.

TABLE I  
 $^1\text{H}$  NMR DATA<sup>a</sup> FOR ISOMERS 1 and 2

Position (ppm)		Assignment	Multiplicity	Isomer <u>1</u> <sup>c</sup>
<u>1</u>	<u>2</u>			Coupling Constant (Hz)
0.73 <sup>b</sup>	0.73	isopropyl methyl	Doublet of Doublets	$J_{16, 16a}$ (19)
1.48	1.50	<u>trans</u> CH <sub>3</sub> at C-8	Singlet	--
1.59	1.68	<u>trans</u> CH <sub>3</sub> at C-12 in <u>1</u> ; <u>cis</u> CH <sub>3</sub> at C-12 in <u>2</u>	Singlet	--
1.91	1.91	ketone methyl	Singlet	--
2.02	2.15	-CH <sub>2</sub> CH <sub>2</sub> CH-	Multiplet	--
4.94	4.95	<u>cis</u> H at C-15	Broadened Doublet	$J_{14, 15}$ (12) $J_{15, 15}$ (2)
5.10	5.08	<u>trans</u> H at C-15 and <u>trans</u> H at C-9	Multiplet	--
5.90	5.90	<u>trans</u> H at C-13	Doublet	$J_{13, 14}$ (12)
6.00	6.00	<u>trans</u> H at C-3	Doublet	$J_{3, 4}$ (16)
6.35	6.35	<u>trans</u> H at C-4	Doublet of Doublets	$J_{3, 4}$ (16) $J_{4, 5}$ (10)
6.58	6.58	<u>trans</u> H at C-14	Multiplet (Six Lines)	$J_{13, 14}$ (12) $J_{14, 15E}$ (17) $J_{14, 15Z}$ (12)

a 80 MHz spectra in benzene- $d_6$ , chemical shifts are in ppm from benzene- $d_5$  (7.15 ppm).

b Center of both doublets.

c Coupling constants assigned for isomer 1 by analysis of its 360 MHz NMR spectrum.

The stereochemistry of 1 and 2 were confirmed by  $^{13}\text{C}$  NMR. The  $^{13}\text{C}$  NMR resonances were assigned (Table II) by comparison with model compounds having carbons in almost identical structural and/or stereochemical environments as those in 1 and 2. The  $^{13}\text{C}$ -NMR of each isomer is consistent with eight olefinic carbons, eleven aliphatic carbons and one carbonyl

\*The MS data are consistent with 3-methyl-2-hexen-1,6-dialdehyde (not in the literature).

carbon that absorbs in the range expected for simple  $\alpha,\beta$ -unsaturated ketones<sup>19a</sup>. The presence of trans and cis 4-methyl-1,3-butadiene moieties in 1 and 2 is shown by the differences in chemical shifts<sup>20</sup> at carbons 11 and 12a for these isomers. Also, 8,9 olefinic carbons and 8a methyl carbons absorb at positions clearly showing the trans geometry<sup>21a,b</sup> of the 8,9 carbons in these isomers. The 3,4 carbons absorb at positions consistent with  $\alpha,\beta$ -unsaturated ketones<sup>19b</sup>. As expected, the isopropyl methyl carbons 16a and 16b are non-equivalent because the C-5 isopropyl group is attached to an asymmetric carbon atom. A difference between the <sup>13</sup>C-NMR chemical shift of a model compound and the C-6 shifts of 1 and 2 is likely due to the conformational differences between these molecules.

TABLE II  
ASSIGNED <sup>13</sup>C-NMR CHEMICAL SHIFTS\* FOR ISOMERS 1 and 2

Carbon No.	<u>1</u>	<u>2</u>	Model Compounds
1 <sup>±</sup>	27.1	27.1	(27.0) <sup>a</sup> , (26.8) <sup>b</sup>
2	196.0	196.1	(199.1) <sup>a</sup> , (197.4) <sup>b</sup>
3	132.7	132.7	(133.4) <sup>a</sup> , (131.5) <sup>b</sup>
4	149.1	149.1	(150.7) <sup>a</sup> , (151.6) <sup>b</sup>
5	49.0	49.1	(49.4) <sup>a</sup>
6	30.2	30.2	(25.5) <sup>a</sup>
7	37.9	38.0	(39.8) <sup>e</sup> , (40.0) <sup>f</sup>
8	135.0	135.3	(134.9) <sup>e</sup> , (135.1) <sup>e</sup>
8a	16.2	16.2	(15.9) <sup>e</sup> , (16.0) <sup>e</sup> , (16.0) <sup>f</sup>
9	124.7	124.5	(124.3) <sup>e</sup> , (124.5) <sup>e</sup> , (124.6) <sup>f</sup>
10 <sup>±</sup>	26.8	27.1	(26.8) <sup>c</sup> , (27.0) <sup>d</sup>
11	40.2	32.8	(40.1) <sup>c</sup> , (32.6) <sup>d</sup>
12	138.8	139.0	(139.1) <sup>c</sup> , (139.3) <sup>d</sup>
12a	16.8	23.9	(16.7) <sup>c</sup> , (23.7) <sup>d</sup>
13	126.4	127.4	(125.8) <sup>c</sup> , (126.7) <sup>d</sup>
14	133.8	133.6	(133.5) <sup>c</sup> , (133.2) <sup>d</sup> , (133.8) <sup>h</sup>
15	114.9	114.8	(114.4) <sup>c</sup> , (114.3) <sup>d</sup> , (114.3) <sup>h</sup>
16	32.0	32.1	(32.1) <sup>a</sup>
16a <sup>±</sup>	19.5	19.4	(19.4) <sup>a</sup> , (19.4) <sup>g</sup>
16b <sup>±</sup>	20.9	20.9	(20.7) <sup>a</sup> , (20.5) <sup>g</sup>

\* Spectra in benzene-d<sub>6</sub>, chemical shifts in ppm downfield from TMS, C-1 and C-10 peak in Isomer 2 overlap.

a Norsolanadione, this laboratory.

b 3E-5-Ethyl-3-hepten-2-one, Sadtler C-13 NMR No. 1174C.

c 3E-4,8-Dimethylnona-1,3,7-triene, ref. 20.

d 3Z-4,8-Dimethylnona-1,3,7-triene, ref. 20.

e Presqualene esters (18) and (19), see ref. 21a, p. 914.

f trans-1,4-Polyisoprene, ref. 21b.

g  $\beta$ -4,8,13-Duvatrien-1,3-diol, ref. 2 and this laboratory.

h 4-Methyl-1,3-pentadiene, this laboratory.

± Assignments for carbons 1, 10 and 16a, 16b may be reversed.

## ACKNOWLEDGEMENTS

The authors gratefully acknowledge Dr. John Lephardt for his expertise in FTIR measurements, Dr. Jerry F. Whidby and Mr. Ronald L. Bassfield for their skillful assistance in  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR spectroscopy and Dr. William B. Edwards, III for a synthetic sample of norsolanadione.

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(Received in USA 30 March 1981)