Assignment of ^{13}C Nuclear Magnetic Resonance Signals in Fatty Compounds with Allylic Hydroxy Groups

G. Knothe* and M.O. Bagby
NCAUR, ARS, USDA, Peoria, Illinois 61604

ABSTRACT: ^{13}C Nuclear magnetic resonance (NMR) signals in several fatty compounds with allylic mono- and dihydroxy groups were assigned by comparing compounds with and without other functional groups (allylic hydroxy, carboxylic acid, respectively, methyl ester at C_1). The simple ^{13}C NMR spectra of hydroxylated compounds derived from symmetrical alkenes are particularly useful in making assignments. The compounds whose signals were partially assigned are 8-hydroxy-9(*E*)-octadecenoic acid, 11-hydroxy-9(*E*)-octadecenoic acid, 8,11-dihydroxy-9(*E*)-octadecenoic acid, 9(*E*)-octadecen-8-ol, and 9(*E*)-octadecene-8,11-diol. The present evaluation can be used for assigning signals in other fatty compounds.
JAOCS 73, 661–663 (1996).

KEY WORDS: Allylic hydroxy groups, carbon nuclear magnetic resonance, 2(*E*)-ene-1,4-diols, methylene envelope, symmetrical compounds.

Assignment of the ^{13}C nuclear magnetic resonance (NMR) signals of fatty compounds to specific carbon atoms has been the subject of numerous publications (1 and references therein). Gunstone *et al.* (1,2) provided the data for all isomeric octadecenoic acids. All signals in the ^{13}C NMR spectra were assigned to specific carbon atoms.

Recently, we synthesized numerous fatty compounds with allylic hydroxy and allylic dihydroxy [2(*E*-)ene-1,4-diols] groups (3–6). The evaluation of carbons in the methylene envelope of 28–30 ppm is intricate, and the assignments of some nonmethylene envelope carbons (olefinic and hydroxy-bearing) were not complete. With the availability of 1,4-diols, synthesized from symmetrical alkenes (6), it is now possible to assign these signals by comparison of spectra.

DISCUSSION

The number of ^{13}C NMR signals is halved in symmetrical straight-chain compounds, derived from symmetrical alkenes (6), compared to nonsymmetrical fatty compounds. Thus, the evaluation of their spectra is significantly simplified, and most signals could be readily assigned.

Part of the chain (containing terminal methyl) in nonsymmetrical compounds with carboxylic acid and methyl ester functionalities at C_1 is identical to the symmetrical alkenes and alkenols. Therefore, the signals that are not assignable by comparison with the hydroxy compounds derived from symmetrical alkenes are assigned to the "nonsymmetrical" part of the molecule. The hydrocarbon chainlengths of some of the symmetrical compounds coincide with the chainlengths of the terminal methyl group in nonsymmetrical, olefinic, C_1-functionalized compounds. Furthermore, it is assumed that the functional group at C_1 does not influence any methylene carbon signals beyond the allylic mono- or dihydroxy moiety.

Assigning signals of the hydroxy-bearing and unsaturated carbons in the allylic dihydroxy group. The symmetrical 2-ene-1,4-diols can be utilized in unambiguously assigning the ^{13}C NMR peaks of the fatty acids and esters that contain these structural groups. We did not unambiguously assign the signals of the olefinic and hydroxy-bearing carbons in previous work (3–5). Parra *et al.* (7) reported the ^{13}C NMR spectrum of 7,10-dihydroxy-8(*E*)-octadecenoic acid (in CD_3OD), but did not assign the signals of these carbons.

For the present evaluation, it is most convenient to select the diastereomers of 6,9-dihydroxy-7(*E*)-nonadecenoic acid (5). Here, the shift differences of the carbons in question are most pronounced because the 2-ene-1,4-diol group is closest to C_1 of all allylic dihydroxy acids. Fatty acids and esters with the allylic dihydroxy group closer to C_1 than $\Delta 7$ were not obtained due to lactonization (4,5). When the functional group is more remote from C_1 (approaches the position of shift equivalence; see Ref. 4), the shift differences become small (often <0.1 ppm).

In *threo*-6,9-dihydroxy-7(*E*)-nonadecenoic acid, the unsaturated carbons exhibit peaks at 133.87 and 133.48 ppm. The signals of the olefinic carbons in the *threo* isomers of the symmetrical 2-ene-1,4-diols are consistently in the range of 133.82–134.09 ppm. Therefore, the downfield olefinic carbon in the fatty compounds, containing the allylic dihydroxy group is on the side of the terminal methyl group [position II in previous terminology (4); position I is then the side of the functional group at C_1].

The same evaluation holds for the hydroxy-bearing carbons and the α methylene carbons, where the downfield carbons are also in position II. For example, in *threo*-6,9-dihydroxy-7(*E*)-nonadecenoic acid, the hydroxy-bearing carbons

*To whom correspondence should be addressed at NCAUR, 1815 N. University St., Peoria, IL 61604.

resonate at 72.31 and 71.91 ppm (6). In the *threo* symmetrical compounds, the signals of the hydroxy-bearing carbons are observed at 72.29–72.54 ppm [excluding the shorter C_8 compounds; (6)].

Assigning signals of the hydroxy-bearing and α carbons in saturated 1,4-diols. Generally, this evaluation holds for the saturated 1,4-diols. The downfield signals are assigned to the position II carbons by comparison of the saturated symmetrical compounds (6) and saturated fatty acids, esters, and alcohols (4). The previous assignment of the downfield signals to position II (4) is confirmed by this comparison. This assignment also holds for those made by Gunstone (1) for 7,10-dihydroxyoctadecanoic acid [in this compound the original, reverse assignment for the hydroxy-bearing carbons (8) is incorrect].

Assigning carbons within the methylene envelope: monohydroxy compounds. The methylene carbons that exhibit signals around 29 ppm have been termed the methylene envelope by Gunstone (1). Table 1 contains shift data on methylene envelope carbons of the 9(*E*)-octadecene-8,11-diols, 9(*E*)-octadecen-8-ol, 8-hydroxy-9(*E*)-octadecenoic acid, and 11-hydroxy-9(*E*)-octadecenoic acid. A comparison of the methylene envelope shift values of these compounds leads to the evaluations given in Table 1. The key to the assignments is the observation that the two symmetrical octadecenediols only possess position II methylene carbons because of the molecular symmetry and the methyl terminus. With this key, the signals were assigned to carbons in either position I or position II. For two carbon signals of 8-hydroxy-9(*E*)-octadecenoic acid, an unambiguous assignment of the signals to specific carbons of positions I or II is not possible. Note that, because there is no functional group at C_1 in 9(*E*)-octadecen-8-ol, the hydroxy-bearing side of the double bond is considered position I here.

TABLE 1
Partial Assignment of Methylene Envelope Carbons in Fatty Compounds with Allylic Hydroxy Groups

Compound	Signals[a]				
m-9(*E*)-Octadecene-8,11-diol[b]	29.49	29.23[c]			
t-9(*E*)-Octadecene-8,11-diol[b]	29.46	29.24[c]			
9(*E*)-Octadecen-8-ol[d]	29.50	29.44	29.27[e]	29.27	29.16
	I	II	I	II	II
8-OH-9(*E*)-octadecenoic acid	29.44	29.28	29.18	29.14	28.99
	II	II	I/II[f]	I/II	I
11-OH-9(*E*)-octadecenoic acid	29.52	29.27	29.06	28.98	28.83
	II	II	I	I	I

[a]Values for the signals taken from Reference 6 and the supplemental publication to Reference 4. The assignments are indicated by "I" or "II," referring to the side of the functional group the signal is assigned to [see text for explanation of I and II. Despite the differences in C_1 (methyl or acid), the designations of I or II do not change].
[b]*m* and *t* are abbreviations for *meso* and *threo*, respectively.
[c]These signals are assigned to the carbon atom pairs 4, 15, and 5, 14.
[d]The hydroxy-bearing side of this compound is considered position I despite the lack of a functional group at C_1 (see text).
[e]The signal at 29.27 ppm is caused by two carbon atoms (see Ref. 6) and therefore is attributed to one position I and one position II carbon.
[f]I or II.

Another example where methylene envelope carbons can be assigned to a double bond side is 10-hydroxy-11(*E*)-octadecenoic acid, which exhibits signals at 29.42, 29.32, 29.26, 29.14, and 29.00 ppm (supplementary publication to Ref. 4). Here a comparison with 7(*E*)-tetradecen-6-ol, which displays signals at 29.10 and 28.77 ppm (6), is helpful. Because the diastereomers of 7(*E*)-tetradecene-6,9-diol do not exhibit methylene envelope carbons, the two previously mentioned signals of 7(*E*)-tetradecen-6-ol are assigned to the nonhydroxylated side of the double bond. The nonhydroxylated side of 7(*E*)-tetradecen-6-ol corresponds to the position II side of 10-hydroxy-11(*E*)-octadecenoic acid. Therefore, the signals at 29.14 and 29.00 in 10-hydroxy-11(*E*)-octadecenoic acid, their shifts being closest to those in 7(*E*)-tetradecen-6-ol, are likely in position II. Similarly, two methylene carbons at 28.98 and 28.80 ppm in 8-hydroxy-9(*E*)-octadecenoic acid (supplementary publication to Ref. 4) are assigned to position II, and the signal at 29.12 ppm is likely in position I.

Besides these assignments, a comparison of the ^{13}C NMR of allylic monohydroxy compounds derived from symmetrical alkenes (6) shows that for split signals of terminal methyl carbons and penultimate carbons, the carbons on the side of the hydroxy group are assigned to the upfield signal.

Assigning carbons within the methylene envelope: dihydroxy compounds. The ^{13}C NMR spectra of the diastereomers of 8,11-dihydroxy-9(*E*)-octadecene exhibit two peaks within the methylene envelope, 29.49 and 29.23 ppm for *meso* (*meso* corresponds to *erythro* in nonsymmetrical compounds) and 29.46 and 29.24 ppm for *threo* (see Table 1). The ^{13}C NMR spectra of *erythro*- and *threo*-8,11-dihydroxy-9(*E*)-octadecenoic acid contains four signals (one carbon each) in the methylene envelope (supplemental publication for Ref. 4). These are 29.50, 29.25, 28.97, and 28.89 ppm for the *erythro* diastereomer and 29.35, 29.12, 28.90, and 28.77 ppm for the *threo* congener. A comparison of the spectra of the diastereomers of 8,11-dihydroxy-9(*E*)-octadecenoic acid with the simple spectra of the symmetrical compounds therefore assigns the signals >29 ppm to methylene carbons on the terminal methyl side of the molecule and signals <29 ppm to methylene carbons between C_1 and the allylic dihydroxy group. As for monohydroxy compounds, further assignments to the specific carbons (besides position I or II) are not possible with this evaluation.

The present assignments and a comparison of shifts with those of unsubstituted octadecenoic compounds (2) confirm the validity of the assumption made earlier in this discussion that the functional group at C_1 has little or no influence on methylene carbon signals beyond the allylic mono- or dihydroxy group.

In conclusion, the simple ^{13}C NMR spectra of allylic mono- and dihydroxy compounds derived from symmetrical alkenes can be used to assign signals in the ^{13}C NMR spectra of fatty acids and esters that contain allylic mono- and dihy-

droxy groups. The method is based on the observation that parts of the symmetrical alkene-derived molecules are identical to parts of the fatty acid-derived compounds. This method also confirms some previous assignments.

REFERENCES

1. Gunstone, F.D., High Resolution ^{13}C NMR Spectroscopy of Lipids, in *Advances in Lipid Methodology—Two*, edited by W.W. Christie, The Oily Press, Dundee, 1993, pp. 1–68.
2. Gunstone, F.D., M.R. Pollard, C.M. Scrimgeour, and H.S. Vedanayagam, Fatty Acids. Part 50. ^{13}C NMR Nuclear Magnetic Resonance Studies of Olefinic Fatty Acids and Esters, *Chem. Phys. Lipids 18*:115–129 (1977).
3. Knothe, G., D. Weisleder, M.O. Bagby, and R.E. Peterson, Hydroxy Fatty Acids Through Hydroxylation of Oleic Acid with Selenium Dioxide/*tert.*-Butylhydroperoxide, *J. Am. Oil Chem. Soc. 70*:401–404 (1993).
4. Knothe, G., M.O. Bagby, D. Weisleder, and R.E. Peterson, Allylic Mono- and Dihydroxylation of Isolated Double Bonds with Selenium Dioxide-*tert.*-Butylhydroperoxide. NMR Characterization of Long-Chain Enols, Allylic and Saturated 1,4-Diols, and Enones, *J. Chem. Soc., Perkin Trans. 2*:1661–1669 (1994) (the supplement to this publication may be ordered from the *J. Chem. Soc., Perkin Trans.*).
5. Knothe, G., M.O. Bagby, D. Weisleder, and R.E. Peterson, Allylic Hydroxy Fatty Compounds with Δ5-, Δ7-, Δ8-, and Δ10-Unsaturation, *J. Am. Oil Chem. Soc. 72*:703–706 (1995).
6. Knothe, G., M.O. Bagby, and D. Weisleder, Fatty Alcohols Through Hydroxylation of Symmetrical Alkenes with Selenium Dioxide/*tert.*-Butylhydroperoxide, *Ibid. 72*:1021–1026 (1995).
7. Parra, J.L., J. Pastor, F. Comelles, M.A. Manresa, and M.P. Bosch, Studies of Biosurfactants Obtained from Olive Oil, *Tenside Surfactants, Deterg. 27*:302–306 (1990).
8. Knothe, G., M.O. Bagby, R.E. Peterson, and C.T. Hou, 7,10-Dihydroxy-8(*E*)-Octadecenoic Acid: Stereochemistry and a Novel Derivative, 7,10-Dihydroxyoctadecanoic Acid, *J. Am. Oil Chem. Soc.* 69:367–371 (1992).

[Received September 29, 1995; accepted February 2, 1996]