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^{13}C NMR SPECTRA OF 5- AND 7-BROMO-6-KETOSTEROIDS AND RELATED COMPOUNDS

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^{13}C NMR spectra have been studied and signals of C atoms have been assigned for 5- and 7-bromo-6-ketosteroids, which are intermediates in the synthesis of ecdysteroids.

Key words: ^{13}C NMR spectra, 6-ketosteroids, 5- and 7-bromo-6-ketosteroids.

Steroidal 5α- and 7α-bromo-6-ketones are well known as important intermediates in the synthesis of ecdysteroid insect hormones [1, 2]. The 5β-hydroxy or 7-double bond, which are necessary structural elements for producing high biological activity in ecdysteroids, are easily introduced if they are used. We used previously namely these α-bromo-6-ketosteroids to synthesize ecdysteroids and their structural analogs [3-7]. The importance of 5α- and 7α-bromo-6-ketosteroids prompted us to study their ^{13}C NMR spectra. We hoped that the results would be useful for establishing the structures of new compounds of this group. This research was also necessary because the scientific literature was limited to a study of the ^{13}C NMR spectrum of only one 5α-bromo-6-ketosteroid, 3β-acetoxy-5-bromo-5α-cholestan-6-one.

Table 1 lists the ^{13}C NMR spectra of **1**-**11**. Besides 5-bromo-6-ketosteroids **2** and **3** and 7-bromo-6-ketosteroids **4**-**6** and **9**-**11**, Table 1 also contains spectra of compounds of similar structure, 6-ketones **1** and **8a**-**b**, 6-alcohol **7a**, and formate **7b**. Signals in the ^{13}C NMR spectra were assigned to resonances of particular atoms using mainly the chemical shift of the signals for the C atoms and their multiplicity determined using the DEPT method. It should be noted that the studied compounds were prepared from cholesterol, β-sitosterol, or stigmasterol. Therefore, signals of C atoms in rings *C* and *D* and the side chains were assigned by comparing their spectra with those of steroids of analogous structure that we studied earlier [10-12]. Signals of C atoms in the side chain of **5**, which contains a 22,23-double bond, were assigned by comparing the position of analogous signals in the spectrum of (22S,23S)-28-homobrassinolide [13].

Table 1 shows that the signals for C-6 atoms in spectra of saturated α-bromo-6-ketosteroids appear at δ 203-204 ppm. Thus, they are shifted to strong field compared with the analogous signals at δ 210-212 ppm in spectra of 6-ketosteroids **1** and **8a** and **b**, which have no α-Br relative to the ketone.

Definite conclusions about the influence of an additional Br atom on the chemical shifts of proximal C atoms can be made by comparing the spectra of **1** and **2**. This situation shifts the signal for C-5 in the spectrum of **2** to weak field by 23.1 ppm compared with its position in the spectrum of **1**. The observed shift is surely caused by the α-effect of the electronegative Br atom. Also, the β-effect of the 5α-Br shifts the signals of C-4 and C-10 in the spectrum of **2** to weak field by 8.7 and 1.8 ppm, respectively, compared with their positions in the spectrum of **1**. The presence in **2** of an axial 5α-Br also causes a significant shift to strong field of the signals for C-1, C-7, and C-9 by 6.0, 6.2, and 6.6 ppm, respectively, compared with their positions in the spectrum of **1** owing to a γ-gauche effect. However, the signal for C-3, which is also oriented γ-gauche with respect to the Br is shifted to weak field in the spectrum of **2** by only 1.8 ppm. It should also be mentioned that the signal for C-19 in the spectrum of **2** is observed at δ 14.5 ppm and is shifted by 1.5 ppm to weak field compared with its position in the spectrum of **1**. This phenomenon is due to a γ-anti-periplanar effect of the Br on the 19-methyl [8, 14].

A comparison of the spectra of **2** and **3** shows that the signals for C-6, C-7, C-8, C-9, C-10, and C-19 have practically the same chemical shifts in them. However, the chemical shifts of C-1 to C-5 depend on the nature of the C-3 substituent. The position of the signal for C-5 changes insignificantly. Assigning the signal for C-5 in ^{13}C NMR spectra is usually simple.

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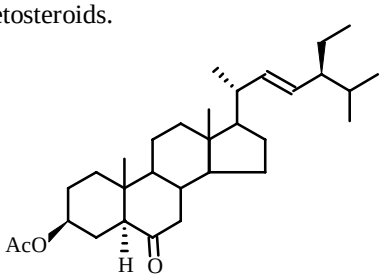
Therefore, the presence at weak field of a signal near 80 ppm provides a reliable criterion for proving the structure of 5α-bromo-6-ketosteroids.



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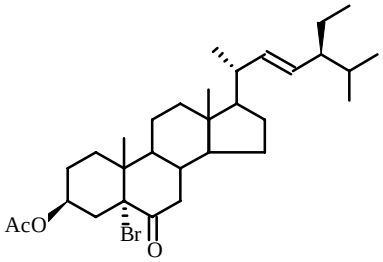

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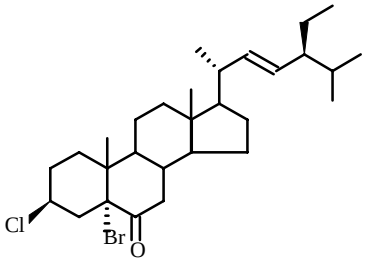

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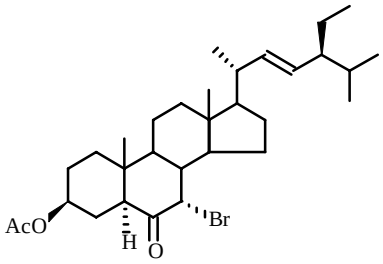

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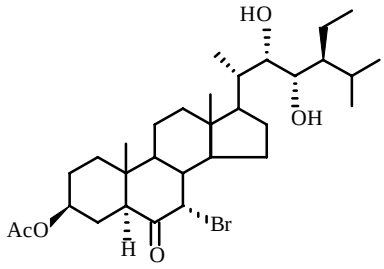

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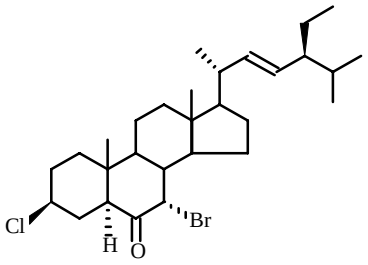

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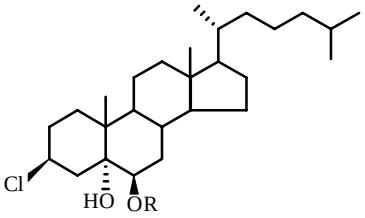
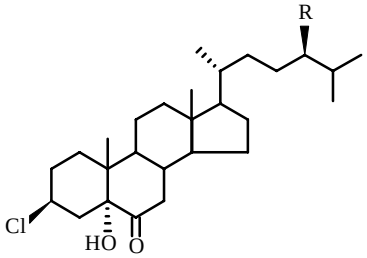
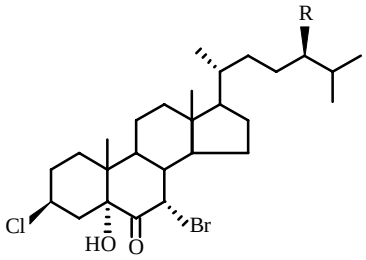


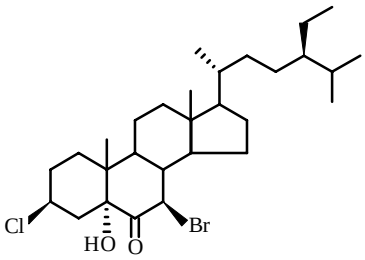
7 a: R=H
b: R=OCHO



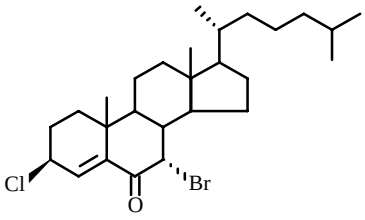
8 a: R=H
b: R=Et



9 a: R=H
b: R=Et



10



11

The position of the Br in **4**-**6** can be rather simply defined from the ^{13}C NMR spectra listed in Table 1. Thus, the signal for C-7 (δ 58.3-58.6 ppm) shifts significantly to weak field compared with its position in spectra of unsubstituted **1** (δ 46.6 ppm) and **2**-**3** (δ 40.4-40.5 ppm). Furthermore, DEPT spectra suggest that C-7 in **4**-**6** is a methine. This also confirms that a Br atom is bonded to it. Comparing ^{13}C NMR spectra of **1** and **4** reveals another series of changes in the chemical shifts of certain atoms that are caused by the presence in **4** of an additional Br atom in the 7α-position. Thus, the signal for C-8 in the spectrum of **4** shifts to weak field by 2.3 ppm compared with its position in the spectrum of **1** as a result of the β-effect of the Br. The axial (i.e., α) orientation of the Br in **4** is confirmed by the significant shift to strong field of the signals of the γ-gauche oriented C-5 (by 6.6 ppm), C-9 (by 7.9 ppm), and C-14 (by 4.4 ppm) atoms compared with their positions in the spectrum of **1**. Because the chemical shifts of the signals for C-5, C-7, C-8, C-9, and C-14 in the spectra of **4**-**6** are practically the same, it can be concluded that this is a general rule that can be used to establish the structures of new compounds of this series.

TABLE 1. Chemical Shifts of C Atoms (δ, ppm) in ^{13}C NMR Spectra of **1**-**11**

Atom	**1**	**2**	**3**	**4**	**5**	**6**	**7a**	**7b**	**8a**	**8b**	**9a**	**9b**	**10**	**11**
C-1	36.4	30.4	31.9	36.2	36.1	31.1	34.8	33.4	37.9	37.9	38.3	38.3	38.8	36.0
C-2	26.9	26.0	31.5	26.8	26.7	32.3	33.9	32.1	31.9	32.0	31.7	31.8	31.9	29.5
C-3	72.8	71.0	56.3	72.4	72.4	58.3	57.0	56.2	56.1	56.0	55.6	55.6	55.4	54.0
C-4	26.1	34.8	39.7	25.9	25.8	37.9	42.4	41.9	31.1	31.1	30.5	30.6	31.7	134.0
C-5	56.5	79.6	80.6	49.9	49.9	51.2	76.3	75.7	80.7	80.7	82.2	82.2	81.0	144.0
C-6	210.3	204.0	203.6	203.9	203.4	203.4	75.9	75.2	211.8	211.9	202.9	202.9	203.0	195.4
C-7	46.6	40.4	40.5	58.5	58.3	58.6	32.3	31.5	41.9	41.9	53.9	54.0	60.7	58.8
C-8	37.9	36.2	36.2	40.2	40.2	40.1	30.1	30.5	37.3	37.3	39.1	39.1	47.0	37.7
C-9	53.9	47.3	47.4	46.0	45.9	45.9	45.9	45.3	44.5	44.5	37.8	37.9	48.0	43.6
C-10	40.9	42.7	42.5	41.3	41.2	41.1	38.2	38.2	42.3	42.3	42.1	42.1	41.6	38.6
C-11	21.5	21.7	21.6	20.9	20.9	20.8	21.1	20.9	21.3	21.3	20.7	20.8	22.2	20.4
C-12	39.3	39.1	39.2	38.6	38.6	38.5	39.9	39.7	39.5	39.5	38.6	38.6	39.9	38.5
C-13	42.9	42.9	42.9	42.6	43.2	42.5	42.8	42.7	43.1	43.1	42.8	42.9	44.7	42.3
C-14	56.8	55.7	55.8	52.4	52.3	52.4	55.9	55.6	56.4	56.4	52.6	52.7	56.7	52.2
C-15	24.0	23.8	23.8	22.9	23.0	22.8	24.1	24.0	23.9	23.9	23.0	23.2	27.2	22.8
C-16	28.7	28.7	28.7	28.5	27.6	28.5	28.2	28.1	28.0	28.1	27.8	27.9	28.5	27.8
C-17	56.6	56.3	56.3	55.6	51.9	55.5	56.3	56.1	56.1	56.0	55.5	55.6	55.4	55.7
C-18	12.2	12.3	12.3	12.6	12.3	12.6	12.2	12.1	12.0	12.0	12.5	12.1	12.0	12.3
C-19	13.0	14.5	14.6	12.6	12.6	12.9	16.8	16.4	14.0	14.0	14.1	14.1	15.4	20.0
C-20	40.4	40.4	40.4	40.5	42.2	40.4	35.8	35.7	35.7	36.1	35.9	36.1	36.1	35.9
C-21	21.1	21.1	21.1	21.1	14.6	21.1	18.7	18.6	18.6	18.7	18.7	18.8	19.0	18.6
C-22	138.0	137.9	137.9	137.9	72.1	137.8	36.2	36.1	36.1	33.9	36.1	33.9	33.8	36.0
C-23	129.6	129.6	129.7	129.8	70.6	129.8	23.9	23.8	23.9	26.1	23.8	26.2	26.1	23.8
C-24	51.2	51.2	51.3	51.3	49.5	51.4	39.5	39.5	39.5	45.8	39.5	45.9	45.8	39.5
C-25	31.9	31.9	31.9	31.9	26.9	31.8	28.0	28.0	28.0	29.2	28.0	29.2	29.2	28.0
C-26	21.3	21.3	21.2	21.2	21.7	21.2	22.6	22.5	22.6	19.0	22.6	19.1	19.8	22.5
C-27	19.0	19.0	19.0	19.0	17.7	19.0	22.8	22.8	22.8	19.8	22.8	19.9	20.2	22.8
C-28	25.4	25.4	25.4	25.4	18.6	25.4				23.1		23.1	23.1	
C-29	12.2	12.3	12.3	12.3	14.2	12.3				12.0		12.6	12.4	
CH_3CO	21.2	21.2		21.1	21.3									
CH_3CO	170.5	170.3		170.5	170.6									
HCO								160.3						

The presence of an additional 5α-hydroxyl in **9a** and **b** produces chemical shifts for C atoms in rings *A* and *B* that are significantly different from those for the analogous atoms in the spectrum of the structurally similar **6**. It should be noted that effects due to a 5α-hydroxyl on the position of signals for proximal C atoms is well known [9, 14]. Signals for C atoms in ring *A* in spectra of **7a** and **b** and **8a** and **b** were assigned taking this into account. Because the Br in **9a** and **b** is located far from the ring *A* atoms, their signals in the ^{13}C NMR spectra have approximately the same chemical shifts as those in the spectra of **8a** and **b**. On the other hand, the situation with the position of the signals for ring *B* atoms is more complicated because they are influenced simultaneously by the axial 5α-hydroxyl and the Br on C-7. Thus, the 5α-hydroxyl and Br atom exert opposing effects on the chemical shifts of C-5 and C-7. The effect of the hydroxyl dominates over that of the Br because of the greater electronegativity of the O atom. However, both the 5α-hydroxyl and Br in **9a** and **b** have a γ-gauche effect on C-9. This significantly shifts the signal for C-9 to strong field compared with its position in the spectra of unsubstituted 6-ketosteroids, for example, **1**, and 5α-hydroxy-6-ketosteroids **8a** and **b** and 7α-bromo-6-ketosteroids **4**-**6**.

One of the characteristic structural features of **10** is the Br atom situated in the plane of the steroid. Therefore, several C atoms immediately fall under its direct influence. Thus, signals for C-7, C-8, C-9, and C-14 and even C-15 shift significantly to weak field in the spectrum of **10** compared with their positions in the spectrum of isomeric **9b**. These differences in the

chemical shifts for the signals of these atoms are so typical that 5α-hydroxy-7β-bromo-6-ketosteroids can be unambiguously distinguished from their 7α-bromo isomers in the ^{13}C NMR spectra.

According to the ^{13}C NMR spectra, the presence of a 7α-bromo-6-ketone in the studied compounds can be reliably proved in the presence of not only a 5α-hydroxyl but also a 4-double bond, as in **11**. The double bond itself is reliably identified by the presence in the spectrum of signals for the two vinyl C atoms C-4 and C-5 with δ 134.0 and 144.0 ppm, respectively. The signal for C-7 in the spectrum of **11** has a characteristic chemical shift of δ 58.8 ppm. This value practically coincides with that of analogous ones in spectra of **4**-**6**. The slight shift to strong field by 2.3-2.5 ppm of the signals for C-8, C-9, and C-10 compared with their positions in spectra of **4**-**6** is also interesting. However, signals for C-14 in spectra of all 7α-bromo-6-ketosteroids **4**-**6** and **11** characteristically have the same chemical shifts. The presence of a 4-double bond causes certain changes in the position of the signals for ring *A* C atoms in the spectrum of **11** compared with their positions in the spectrum of the saturated analog **6**. The chemical shifts of C-19 at δ 20.0 and 12.9 ppm, respectively, differ most. This provides yet more proof of the presence in **11** of an additional double bond.

Thus, our results show that ^{13}C NMR spectra can be used to determine reliably the principal structural elements of 5- and 7-bromo-6-ketosteroids.

EXPERIMENTAL

^{13}C NMR spectra were recorded in $CDCl_3$ on a Bruker AC-200 NMR spectrometer at working frequency 50.32 MHz with 20-50 mg in 0.5 mL. Chemical shifts on the δ scale were determined relative to TMS internal standard. Details of the spectral experiments have been published previously [12].

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