[Chem. Pharm. Bull. 31(2) 557—559 (1983)]

The Structures of 15-Dehydro-prostaglandin B_1 Dimers

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(Received August 16, 1982)

Prostaglandin Bx(PGBx), a complex mixture of closely related oligomers of Prostaglandin B_1 (PGB_1) or 15-dehydro-PGB_1 which has unique biological activities, is formed under basic conditions. For mechanistic studies of the oligomerization, two dimers of 15-dehydro-PGB_1 were prepared under mild conditions and their structures were determined by means of nuclear magnetic resonance (NMR) spectroscopic studies.

Keywords——prostaglandin Bx; prostaglandin B_1; 15-dehydro-prostaglandin B_1; 15-dehydro-prostaglandin B_1 dimer; prostaglandin B_1 dimer

Prostaglandin Bx (PGBx), an oligomeric derivative of prostaglandin B_1 (PGB_1) or 15-dehydro-PGB_1 which was first synthesized by Polis,[1] has unique properties; it affords protection against oxidative phosphorylation in isolated degraded rat river mitochondria *in vitro*[2] and has ionophoretic activity in the release of Ca^{2+} from heart mitochondria.[3] PGBx is a complex mixture of closely related oligomers, and the molecular weights of the most active fractions of PGBx fall between 2000 and 2600.[2]

In the previous paper we reported the structures of PGB_1 dimers.[4] During the course of mechanistic studies on base-promoted oligomerization of 15-dehydro-PGB_1, we succeeded in isolating 15-dehydro-PGB_1 dimers. In this paper we report the structures of these 15-dehyro-PGB_1 dimers.[5]

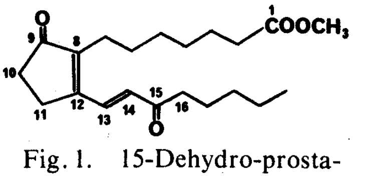


Fig. 1. 15-Dehydro-prostaglandin B_1 Methyl Ester

15-Dehyro-PGB_1 methyl ester was treated with K_2CO_3 in dry methanol at 0 °C for 1.5 h to afford dimer I and II, both in 5% yield (see "Experimental").

Dimer I showed λ (MeOH) 238 and 299 nm and conjugated double bond absorptions at 1635 and 1590 cm^{-1} ($CHCl_3$). ^1H-nuclear magnetic resonance (NMR) decoupling experiments in C_6D_6 (Table I) showed that the proton at 3.67 ppm (1H, ddd, $J=5.5$, 6.5, 10 Hz) and the proton at 3.24 ppm (1H, dd, $J=6.5$, 17.5 Hz),[6] which were coupled to each other, were adjacent to isolated methylene groups ($-CH_2-\overset{|}{C}H-\overset{|}{C}H-CH_2-$). These data indicate that the 13′,14′-double bond in 15-dehydro-PGB_1 was lost in dimer I and a new single bond was formed between C-13′ or C-14′ and C-10 or C-11 by Michael addition. Comparison[7] of the ^{13}C-NMR spectra of dimer I (Table II), 15-dehydro-PGB_1 and 13,14-dihydro-15-dehydro-PGB_1[5a] revealed that the signals of the carbons at C-13′ and C-10 were shifted downfield (*ca.* 10 ppm) to 37.4 ppm (doublet) and to 45.5 ppm (doublet),and the carbons at C-14′ and C-11 were also shifted slightly downfield (*ca.* 4 ppm) to 43.5 ppm (triplet) and 29.6 ppm (triplet), respectively. These data indicate that Michael addition had occurred between C-13′ and C-10, and thus the structure of dimer I is as shown in Fig. 2.

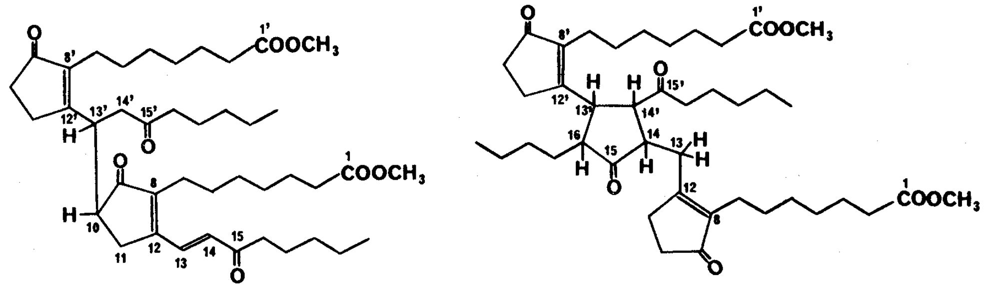
Dimer II showed λ (MeOH) 238 nm, and its infrared (IR) spectrum showed a conjugated double bond absorption at 1640 cm^{-1} (two 13, 14-double bonds were lost in dimer II). ^1H-NMR decoupling experiments of dimer II (Table I) established the partial structure -C(13)H_2-C(14)H-C(14′)H-C(13′)H-C(16)H-CH_2-, and on the basis of J-values, the protons (H-13′, H-14′, H-14 and H-16) were assigned as ring protons with *trans* relationships. An INDOR experment

TABLE I. ^{1}H-NMR Data for Dimer I and Dimer II

	Dimer I (δ in C_6D_6)	Dimer II (δ in C_6D_6)
H-10	3.24 (dd, J=6.5, 17.5 Hz)	
H-11	*ca.* 2.1	
H-13	6.30 (d, J=16 Hz)	2.42 (dd, J=5, 14 Hz), 2.78 (dd, J=10, 14 Hz)
H-14	7.69 (d, J=16 Hz)	3.10 (ddd, J=5, 10, 11 Hz)
H-16		2.08 (dt, J=12, 5 Hz)
H-13′	3.67 (ddd, J=5.5, 6.5, 10 Hz)	3.16 (dd, J=11, 12 Hz)
H-14′	2.06, 2.14	2.62 (t, J=11 Hz)

TABLE II. ^{13}C-NMR Data for Dimer I and Dimer II

Dimer I (δ in $CDCl_3$)				Dimer II (δ in C_6D_6)			
C- 9′	209.7^s	C-13	133.4^d	C-15	213.9^s	C-14′	56.3^d
C-15′	208.8^s	C-14	130.8^d	C- 9	208.6^s	C-16	52.5^d
C- 9	207.8^s	C-10	45.5^d	C- 9′	207.3^s	C-14	50.9^d
C-15	200.2^s	C-14′	43.5^t	C-15′	206.6^s	C-13′	47.0^d
C-12′	172.0^s	C-16′	43.1^t	C-12	168.0^s	C-16′	45.2^t
C-12	158.0^s	C-16	42.1^t	C-12′	166.1^s	C-13	31.1^t
C- 8	147.5^s	C-13′	37.4^d	C- 8	144.7^s		
C- 8′	142.1^s	C-11	29.6^t	C- 8′	142.1^s		



dimer I dimer II

Fig. 2. The Structures of 15-Dehydro-prostaglandin B_1 Dimers

confirmed that the signal of H-16 appeared at 2.08 ppm as a doublet of triplets, $J_{13',16}$= 12Hz and $J_{16,17}$=5Hz. The 5Hz J-value indicates that C-16 is connected to a freely rotating methylene group. The ^{13}C-NMR spectrum of dimer II (Table II) showed the presence of four methine carbons (C-13′, C-14′, C-14 and C-16) along with a carbonyl carbon at 213.9 ppm indicating the formation of two C-C bonds leading to a cyclopentanone ring. The signal at 45.2 ppm (triplet), assigned to C-16 in 15-dehydro-PGB$_1$, was moved downfield to 52.5 ppm (doublet). These data indicate that the initial Michael addition had occured between C-16 and C-13′ or C-14′ and then another C-C bond was formed. The signals at 47.0, 50.9 and 56.3 ppm[8] were assigned to the carbons at C-13′, C-14 and C-14′, respectively, on the basis of selective proton irradiation experiments. These spectroscopic studies indicate the strcture of dimer II to be as shown in Fig. 2.

15-Dehydro-PGB$_1$ dimer I was smoothly converted to PGBx on treatment with 1 N NaOH in 50% EtOH at room temperature, but oligomerization of dimer II under the same conditions was very slow.

Experimental

NMR spectra were recorded on Varian XL-200 and Nicolet NT-360 spectrometers, and signals are given in δ units downfield from tetramethylsilane (TMS) as an internal standard. IR, ultraviolet (UV) and mass spectra

(MS) were measured on Hitachi 260-30, Hitachi 124 and JMS-OISG spectrometers, respectively.

15-Dehydro-PGB$_1$ Dimer I and Dimer II——A mixture of 15-dehydro-PGB$_1$ methyl ester (1.0g) and K_2CO_3 (100 mg) in dry MeOH (20 ml) was stirred at 0°C for 1.5 h then diluted with a large amount of AcOEt. The resulting mixture was washed with water, dried over Na_2SO_4 and concentrated *in vacuo* to afford a yellow oil. The oil was separated by gel-filtration chromatography on Sephadex LH-20 (Pharmacia Fine Chemicals) with MeOH to afford monomer fractions[9)] (20% yield), dimer fraction[10)] (25% yield) and trimer fractions[10)] (10% yield).

The dimer fraction was separated by preparative thin-layer chromatographies (TLC) on Kieselgel$_{60}$ (Merck, 0.2 mm thick) with EtOAc–cyclohexane (1:1) and then with CH_2Cl_2–MeOH (30:1) to give dimer I (5% yield), dimer II (5% yield) and dimer III[11)] (8% yield). Dimer I: *Rf* 0.77 (CH_2Cl_2–MeOH/30:1). UV λ_{max}^{MeOH} nm(ϵ): 238 (19300) and 299 (19000). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 2940, 860, 1725, 1695, 1635 and 1590. MS: Calcd for $C_{42}H_{64}O_8$, *m/z* 696.46009. Found *m/z* 696.45571. Dimer II: *Rf* 0.67 (CH_2Cl_2–MeOH/30:1). UV λ_{max}^{MeOH} nm(ϵ): 238 (26100). IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 2900, 2870, 1725, 1695 and 1640. MS: Calcd for $C_{42}H_{64}O_8$, *m/z* 696.46009. Found *m/z* 696.45963.

References and Notes

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5) Attempts by other research groups to resolve the complex mixture of PGBx have been reported. 5*a*) B. D. Polis, E. Polis and S. Kwang, *Physiol. Chem. & Physics,* **13**, 111 (1981); 5*b*) B.D. Polis, E. Polis and S. Kwang, *Physiol. Chem. & Physics,* **13**, 531 (1981); 5*c*) G.L. Nelson and G.L. Verdine, *Tetrahedron Lett.,* **23**, 1967 (1982).
6) The coupling constant between the proton at 3.24 ppm and one of the adjacent methylene protons is *ca.* zero.
7) The signals of the carbons at C-13′ and C-14′ were compared with those of C-13 and C-14 of 15-dehydro-PGB$_1$ and those of the carbons at C-10 and C-11 were compared with those of C-10 and C-11 of 13,14-dihydro-15-dehydro-PGB$_1$.
8) It seems reasonable to assign the signal at 56.3 ppm to the carbon (C-14) adjacent to the carbonyl group,not to the carbon (C-13′) adjacent to the double bond.
9) Monomer fraction mainly contained 15-dehydro-PGB$_1$ methyl ester and 13-methoxy-13,14-dihydro-PGB$_1$ methyl ester.
10) Mass spectra of the dimer fraction (mixture) and trimer fraction (mixture) showed molecular ion peaks at *m/e* 696 and *m/e* 1044, respectively.
11) Dimer III (*Rf*=0.65, CH_2Cl_2–MeOH/30:1) is an inseparable mixture of two dimeric compounds, λ (MeOH) 238 and 299 nm.