ON THE PHOTOISOMERISATION OF 13-DESMETHYL-RETINAL

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Abstract: All mono-cis-isomers of 13-desmethyl-retinal have been prepared from the all-
trans-compound by photoisomerisation. The various isomers were separated by HPLC and
identified by their 400 MHz-^{1}H-NMR-spectra.

In our studies on retinal-protein interaction we were interested in 13-desmethyl-retinal (1)
since it offers a unique structural change at a point in the retinal molecule which is possibly
involved in the proton pumping function of bacteriorhodopsin[2]. Further, it has been demon-
strated that upon illumination of 1 only the 7-cis-, 9-cis-, and 11-cis-isomers are found[3-6].
In this note we present evidence that the hitherto unknown 13-cis-isomer is also present to
the extent of 7% in a photochemical equilibrium mixture of 1 in hexane.

All-trans-13-desmethyl-retinal (1) was prepared analogously to a published procedure[7]: Con-
densation of ß-ionone (EGA-Chemie) with diethylphosphonato acetonitrile led to the C-15 ni-
trile 2. Treatment of this compound with diisobutylaluminiumhydride in benzene gave the cor-
responding aldehyde which by condensation with methyl 4-diethylphosphonato crotonate led to
methyl 13-desmethyl-retinoate. This was reduced with lithiumaluminiumhydride to produce
13-desmethyl-retinol which was oxidised over manganese dioxide to yield 1, which along with
the intermediate C-15-compounds were purified by HPLC using an ALTEX model 110 A pump
and Kontron Uvikon LCD 725 UV-detector set at 380 nm. Separations were obtained on a
Knauer 30 x 1.6 cm column packed with LiChrosorb 60 and eluted with 10% ether in petroleum
ether at a flow rate of 9 ml/min. Areas under peaks as detected at 380 nm were integrated
using a Hewlett Packard 3380 A integrator, no corrections were made for individual isomer
response. Solutions of all-trans-1 (10^{-3} - 10^{-4} m) were placed 30 cm in front of a white light
source, obtained from a 1200 Watt high pressure Xenon lamp whose light was filtered through
a 400 nm cut-off filter (Schott GG 400), and were stirred throughout the irradiation for periods
up to 10 min, during which time the temperature rose from 18 to 45°C.

The all-trans-13-desmethyl-retinal was taken in one of several solvents for photoisomerisation;
Table 1 shows the isomer composition found after 5 min illumination. Typical analytical HPLC
chromatograms are reproduced in figure 1. Other isomers, appearing as small shoulders,
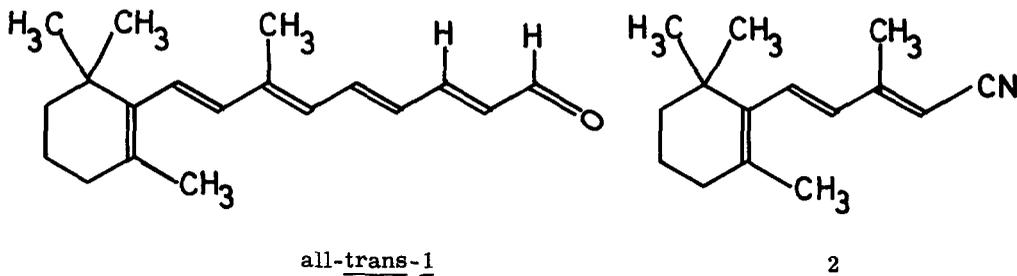were detected but not identified.

all-trans-1   2

Table 1: Photoisomerisation of all-trans-1 in solvents of different polarity

Solvent	Dielectric constant	Percentage isomer composition				
		trans	7-cis	9-cis	13-cis	11-cis
Hexane	1.9	77.1	–	12.6	6.9	3.5
Toluene	2.4	48.2	–	34.4	4.1	12.1
Isopropanol	18.3	43.4	1.5	25.6	2.3	21.0
Methanol	32.6	40.3	2.9	29.8	1.2	25.8
Acetonitrile	37.5	40.0	7.0	33.7	–	19.1

Figure 1: HPLC chromatograms of 13-desmethyl-retinal (1) after irradiation in acetonitrile and in hexane (authentic traces)
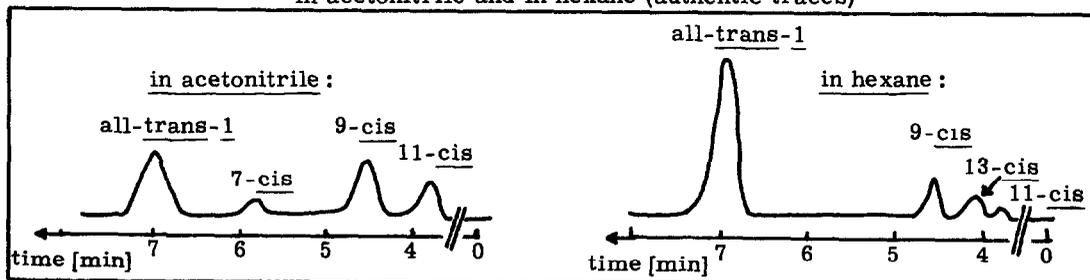
The assignment of the chromatographic bands to the isomeric structures was performed in deuteriochloroform using 400- and 500 MHz-^{1}H-NMR-spectroscopy. In all cases the multiplets of the olefinic protons were sufficiently separated such that by decoupling experiments the protons at carbon atoms 7, 8, 10, 11, 12, 13, and 14 could be unambiguously assigned (Table 2). The cis- and trans-configuration of the double bonds were confirmed by measurement of the ^{3}J-coupling constants. In the case of the 9, 10-double bond, where no vicinal coupling is present, there is a remarkable downfield shift of H-8 for the cis-configuration, as is found with retinal[8].

Table 2: ^{1}H-NMR Data of 13-desmethyl retinal isomers in deuteriochloroform

Chemical shift (ppm)	all-trans	7-cis	9-cis	11-cis	13-cis
7-H	6.38	6.03	6.37	6.40	6.37
8-H	6.17	6.15	6.67	6.24	6.18
10-H	6.19	6.26	5.99	6.62	6.22
11-H	7.07	7.01	7.16	6.84	6.96
12-H	6.46	6.43	6.40	6.24	7.20

Table 2 (cont'd.)

13-H	7.21	7.20	7.20	7.68	7.04
14-H	6.14	6.16	6.04	6.17	5.83
15-H	9.56	9.57	9.54	9.63	10.19
$1-CH_3$	1.04	1.04	1.05	1.05	1.05
$5-CH_3$	1.72	1.51 (?)	1.75	1.74	1.74
$9-CH_3$	2.02	1.92	2.03	2.02	2.04
Coupling constants (Hz)					
J(7, 8)	16	12.5	16	16	16
J(10, 11)	11.5	11.5	12	12	12
J(11, 12)	14.5	14.5	14.5	11	14
J(12, 13)	11.5	11.5	11.5	11.5	12
J(13, 14)	15	15	15	15	11
J(14, 15)	8	8	8	8	8

We prepared a photochemical equilibrium mixture of 1 in hexane having an isomer composition (HPLC analysis) of all-trans : 9-cis : 13-cis : 11-cis = 69 : 19 : 7 : 3. When subjected to ^{1}H-NMR-spectroscopy at 500 MHz a doublet could be seen at 10.2 ppm, due to the 13-cis-isomer[6]. The ^{1}H-NMR spectrum of the aldehydic region of the mixture is reproduced in Figure 2: at least eight isomers can be detected.

Figure 2: ^{1}H-NMR spectrum (500 MHz) of a photochemical equilibrium mixture of 1 showing the aldehydic region
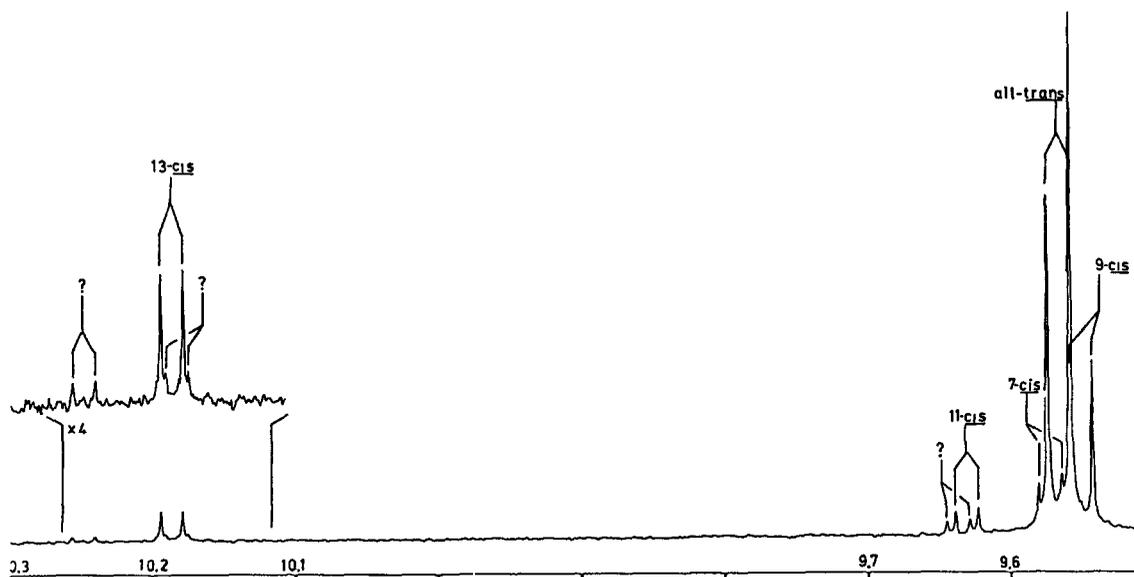
Previous workers had not detected the 13-cis-isomer[5], no doubt due to performing the photo-isomerisation in polar solvents. In the extensively studied retinal photochemistry[9, 10, 11] it appears that irradiation of that molecule in solvents of low dielectric constant results in only

non-sterically hindered isomers, viz. 9-cis and 13-cis, whereas solvents of high dielectric constant additionally yield sterically hindered isomers (7-cis and 11-cis). In the case of 1 only the 7-cis-isomer is sterically hindered. Thus irradiation in hexane results in the appear-ance of 9-cis, 11-cis and 13-cis-isomers, and when using acetonitrile we find that 7-cis ac-companies them. The reason for a predominance of 9-cis in the cis-isomer composition of 1 upon illumination in hexane is unknown; the photoequilibrium changed slightly upon illumi-nation at 360 nm, and possibly the yield of 13-cis could be further optimised although the 9-cis isomer would not doubt still predominate.

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