1,4-DIHYDROPYRIDINE DERIVATIVES AS DEACTIVATORS

OF SINGLET OXYGEN

G. D. Tirzit, I. M. Byteva,
K. I. Salokhiddinov, G. P. Gurinovich,
and G. Ya. Dubur

The constants of deactivation (k_q) of $^1O_2$ by 1,4-dihydropyridine (1,4-DHP) derivatives were determined by quenching of the luminescence of singlet oxygen ($^1O_2$). The k_q values for 1,4-DHP derivatives range from 10^6 to 10^7 liters-mole^{-1}-sec^{-1} and depend to a considerable extent on the nature of the substituents in the 1,4-DHP ring. The presence of a substituent in the 4 position decreases k_q, while conversion of the 1,4-DHP system to the corresponding pyridine system deprives the compound of its ability to deactivate $^1O_2$. As a result of tests of 12 1,4-DHP derivatives it was found that 2,6-dimethyl-3,5-di(phenylcarbamoyl)-1,4-dihydropyridine deactivates $^1O_2$ most effectively.

It is known [1-3] that oxygen in the singlet excited state, the so-called singlet oxygen ($^1O_2$), actively participates in many biological processes. It has been established that $^1O_2$ reacts at a high rate both with lipids, thereby triggering their peroxide oxidation, and with amino acids, thereby giving rise to their degradation and, as a result, damage to enzymes, structural proteins, and biomembranes [4, 5]. Hence the interest in substances that deactivate $^1O_2$ and thus protect biological systems from damage is understandable. The natural substances α-carotene and β-tocopherol are effective $^1O_2$ deactivators [6].

Very few tests have been devoted to heterocyclic compounds as $^1O_2$ deactivators. We have studied 1,4-dihydropyridine (1,4-DHP) derivatives, which constitute a relatively new class of antioxidants that protect lipids from peroxide oxidation [7-9], as $^1O_2$ deactivators. The literature contains virtually no information relative to the reaction of 1,4-DHP derivatives with $^1O_2$, except for a reference [10] to the fact that the photooxidation of NADPH$_2$ to NADP$^+$ in the presence of hematoporphyrin takes place with the participation of $^1O_2$ and our data [11] that 1,4-DHP derivatives in chloroform undergo sensitized photooxidation.

We determined the constants of deactivation (k_q) of $^1O_2$ by 1,4-DHP derivatives from the quenching of the luminescence of oxygen that corresponds to the $^1\Delta_g - ^3\Sigma_g$ transition in the O_2 molecule (λ_{max} 1.27 μ).

The results obtained (Table 1) show that 1,4-DHP derivatives are good $^1O_2$ deactivators. Although the investigated 1,4-DHP derivatives are inferior with respect to their effectiveness to the most active $^1O_2$ deactivator, viz., β-carotene, they have virtually the same degree of activity as 1,4-diazabicyclo[2.2.2]octane (the generally accepted standard synthetic $^1O_2$ deactivator) and the widely used antioxidant Ionol (2,6-di-tert-butyl-4-hydroxytoluene). The pyridine derivative corresponding to 1,4-DHP (see V and XIII in Table 1) does not deactivate $^1O_2$, and this indicates the definite role played by the dihydropyridine system in the reaction of the tested compounds with $^1O_2$.

Substitution in the 4 position of the 1,4-dihydropyridine ring by even an electron-donor substituent leads to a significant decrease in the deactivating capacity of 1,4-DHP (compare V and VI, as well as XI and XII). The presence of electron-donor groups in the 2 and 6 positions is also important. The introduction of aryl substituents in these positions (VII) deprives the dihydropyridine system of its ability to deactivate $^1O_2$.

TABLE 1. Constants of Deactivation (k_q) of Singlet Oxygen by 1,4-Dihydropyridine Derivatives

Compound	$R^{2,6}$	$R^{3,5}$	R^4	k_q, liters·mole^{-1}·sec^{-1}
I	H	CH$_3$	H	$4,7 \cdot 10^6$
II	CH$_3$	CH$_3$	H	$5,1 \cdot 10^6$
III	CH$_3$	C$_6$H$_5$	H	$3,8 \cdot 10^6$
IV	—CH$_2$C(CH$_3$)$_2$CH$_2$—		H	$2,0 \cdot 10^6$
V	CH$_3$	OC$_2$H$_5$	H	$3,4 \cdot 10^6$
VI	CH$_3$	OC$_2$H$_5$	CH$_3$	$<10^5$
VII	C$_6$H$_5$	OC$_2$H$_5$	H	$<10^5$
VIII	CH$_3$	OC$_3$H$_7$·iso	H	$6,6 \cdot 10^6$
IX	CH$_3$	OC$_6$H$_{13}$	H	$5,0 \cdot 10^6$
X	CH$_3$	OC$_6$H$_{11}$·cyclo	H	$6,0 \cdot 10^6$
XI	CH$_3$	NHC$_6$H$_5$	H	$7,5 \cdot 10^7$
XII	CH$_3$	NHC$_6$H$_5$	CH$_3$	$6,5 \cdot 10^6$
XIII	2,6-Dimethyl-3,5-diethoxycarbonylpyridine			$<10^5$
XIV	1,4-Diazabicyclo-2.2.2-octane			$2,0 \cdot 10^7$
XV	Ionol			$6,5 \cdot 10^6$
XVI	β-Carotene			$(1,1—2,7) \cdot 10^{10}$ [15]

The effectiveness of the deactivation of $^1O_2$ by the tested compounds depends to a great degree on the nature of the substituents in the 3 and 5 positions. The deactivation constant increases as the electron-acceptor nature of these substituents decreases by giving rise to an increase in the electron-donor capacity of the $R^{3,5}$ part of this substituent and, as expected, is greatest in the case of 3,5-di(phenylcarbamoyl) groups. Among 3,5-di(alkoxycarbonyl) derivatives the reactivity increases with respect to $^1O_2$ increases as the electron-donor nature of the alkyl group increases. On the other hand, spatial fixation of the 2,6 and 3,5 substituents in an orientation that is coplanar with the aminovinyl system, which reduces the electron-donor properties of the 1,4-DHP ring [12], also decreases k_q.

A comparison of the changes in the rates in sensitized photooxidation [11] and the constants of deactivation of $^1O_2$ by the corresponding 1,4-DHP derivatives shows that the effect of the substituents is symbatic in both cases. The mechanism of the reaction of 1,4-DHP derivatives with excited molecular oxygen evidently includes both a chemical reaction and physical inactivation of $^1O_2$ and can be represented by the general scheme

$$^1O_2 + 1,4\text{-DHP} \xrightarrow{A} [O_2 \ldots 1,4\text{-DHP}]^* \xrightarrow{B} \text{oxidation products}$$
$$\downarrow C$$
$$^3O_2 + 1,4\text{-DHP},$$

where A is the reaction involving the formation of the excited intermediate complex, B is the reaction involving deactivation of $^1O_2$ via chemical reaction, and C is the reaction involving deactivation of $^1O_2$ via a physical pathway.

The data obtained make it possible to assume that 1,4-DHP derivatives can serve as $^1O_2$ deactivators in both chemical and biological systems. It is possible that one of the mechanisms of the antioxidant effect of 1,4-DHP derivatives is deactivation or tying up of $^1O_2$, which initiates peroxide oxidation of lipids.

EXPERIMENTAL

The study was carried out with a pulse apparatus [13, 14]. The lifetime of the excited oxygen molecules was measured from the oscillograms of the extinction of the luminescence. The deactivation constants (k_q) were calculated from the Stern–Volmer equation $\tau_0/\tau = 1 + k_q \cdot C_T \cdot \tau_0$, where τ_0 and τ are the times required for extinction of the luminescence in systems with and without a quenching agent, respectively, and C_T is the 1,4-DHP concentration, which did not exceed $5 \cdot 10^{-3}$ mole-liter^{-1}. The mean-square error in k_q was ±20%. The measurements were made in air-saturated chloroform solutions; luminescence was not recorded under anaerobic conditions. In control experiments it was established that 1,4-DHP does not sensitize the formation of singlet oxygen.

The diethyl ester of mesoporphyrin IX and its complex with Pd(II) served as sensitizers for the formation of $^1O_2$; the sensitizer concentration was $5 \cdot 10^{-6}$ mole-liter^{-1}.

LITERATURE CITED

1. I. R. Politzer, G. W. Griffin, and J. L. Laseter, Chemical-Biological Interaction, <u>3</u>, 73 (1971).
2. N. J. Krinsky, Trends in Biochem. Sci., <u>2</u>, 35 (1977).
3. J. Bland, J. Chem. Educ., <u>53</u>, 274 (1976).
4. F. H. Doleiden, S. R. Fahrenholtz, A. A. Lamola, and A. M. Trozzolo, Photochem. Photobiol., <u>20</u>, 519 (1974).
5. J. B. C. Matheson and J. Lee, Photochem. Photobiol., <u>29</u>, 879 (1979).
6. V. Ya. Shlyapintokh and V. B. Ivanov, Usp. Khim., <u>45</u>, 202 (1976).
7. G. D. Tirzit and G. Ya. Dubur, Khim. Geterotsikl. Soedin., No. 1, 133 (1972).
8. G. Ya. Dubur, Yu. A. Zilber, A. Kh. Velena, A. O. Kumerova, and G. D. Tirzit, Izv. Akad. Nauk Latv. SSR, No. 1, 65 (1975).
9. G. D. Tirzit and G. Ya. Dubur, The 12th World Congress of the International Society for Fat Research, Milan, Italy, Sept. 2-7, 1974, Abstracts, p. 102.
10. R. S. Bodaness and P. C. Chan, J. Biol. Chem., <u>252</u>, 8554 (1977).
11. G. D. Tirzit and G. Ya. Dubur, in: Reactivities of Azines [in Russian], Novosibirsk (1979), p. 110.
12. G. Ya. Dubur, Doctoral Dissertation, Riga (1978).
13. I. M. Byteva, Zh. Prikl. Spektrosk., <u>31</u>, 333 (1979).
14. K. I. Salokhiddinov, I. M. Byteva, and B. M. Dzhagarov, Opt. Spektrosk., <u>47</u>, 881 (1979).
15. L. V. Stopolyanskaya and I. M. Byteva, Biofizika, <u>24</u>, 945 (1979).

CALCULATION OF THE ELECTRONIC STRUCTURE AND DIPOLE MOMENTS
OF 4-SUBSTITUTED TETRABROMOPYRIDINES

A. A. Kisilenko, A. A. Zeikan',
S. I. Vdovenko, and V. P. Kukhar'

UDC 541.67:543.52:547.822.5

It is demonstrated from the results of a quantum-chemical calculation by the CNDO/2 (complete neglect of differential overlap/2) method and the experimental dipole moments for a number of 4-substituted tetrabromopyridines that the character of the intramolecular interactions in the investigated compounds differs little from that observed for 4-substituted nonhalogenated pyridines. A linear relationship between the charge on the heteroatom and the σ_p constants for the substituents in the 4 position was observed. A similar relationship was obtained for the experimental dipole moments and the substituent constants.

In the present research we examined a number of 4-substituted tetrabromopyridines, for which we made a calculation of the electronic structure by means of the standard CNDO/2 (complete neglect of differential overlap/2) program [1] in order to study the distribution of the electron density and the transmission of the electronic effects in the investigated molecules. Similar data were previously obtained within the CNDO/2 approximation for fluorine- and chlorine-containing pyridines [2].

The geometrical parameters for the calculation were selected from the fragments of similar molecules [3-6]. The pentabromopyridine molecule was assumed to be planar. Since