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Antioxidant Effect of Tocopherolhydroquinone on the Oxidation of Ethylbenzene

V. V. Naumov

Emanuel Institute of Biochemical Physics, Russian Academy of Sciences, Moscow, 119991 Russia
e-mail: my9name@mail.ru

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Abstract—The antioxidation properties of tocopherolhydroquinone were studied by the chemiluminescence method in combination with mathematical simulation. The rate constant of the liquid-phase reaction of tocopherolhydroquinone with the α-ethylphenylperoxy radical was determined (2.4×10^6 l mol^{-1} s^{-1}). The rate constants of chain propagation in the reactions of the tocopherolhydroquinone radical with RH (≤0.07 l mol^{-1} s^{-1}) and O_2 (950 l mol^{-1} s^{-1}) were estimated.

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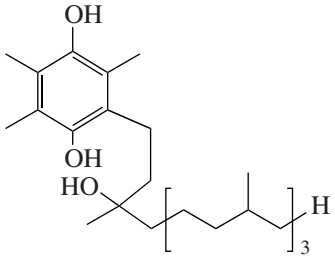
INTRODUCTION

Investigation of the antioxidation properties of tocopherolhydroquinone seems to be very important because this substance is the reduced form of tocopherolquinone, which is a metabolite of the natural antioxidant tocopherol (vitamin E). Tocopherolhydroquinone, along with tocopherol and tocopherolquinone, is abundant in many organs and tissues of animal organisms [1–3]. Tissue tocopherol is believed to serve as a tocopherolhydroquinone reservoir [4].



Tocopherolhydroquinone

The rate constant of the reaction of tocopherolhydroquinone with the free stable phenoxy radical galvinoxyl was estimated in an earlier study [5]. However, the reactivity of tocopherolhydroquinone toward peroxy radicals was estimated only qualitatively from the results of its competition with *N*,*N*-diphenyl-*p*-phenylenediamine [5].

Earlier, we carried out preliminary studies using the chemiluminescence method and showed that tocopherolhydroquinone is a highly reactive agent terminating free radical oxidation chains (chain-breaking antioxidant) through a reaction with peroxy radicals, whose rate constant is about 10^6 l mol^{-1} s^{-1} [6]. More precise quantitative estimation of the rate constant of this reaction from chemiluminescence data was impossible because of the absence of reliable rate constant data for chain propagation in oxidation by semiquinone radicals formed from tocopherolhydroquinone [7]. In the present work, we combined analytical calculations [7] with computer simulation [8–10]. This approach provides a way to surmount these difficulties.

The purpose of the work is to study, in detail, the kinetics of the tocopherolhydroquinone effect on the initiated oxidation of a model hydrocarbon by the chemiluminescence method and to obtain quantitative estimates of the rate constant for the reactions of tocopherolhydroquinone with peroxy radicals and for the chain propagation steps of oxidation by semiquinone radicals.

Ethylbenzene, whose oxidation is known in detail [11], was chosen as the model hydrocarbon. This substance is oxidized with molecular oxygen via a free radical mechanism. The rate constants of the elementary steps of this process are known and can be used in further calculations.

EXPERIMENTAL

Tocopherolhydroquinone was synthesized by the reduction of the corresponding quinone with sodium borohydride. The initiator 2,2'-azodiisobutyronitrile (AIBN) was used as the source of free radicals. Experiments were carried out at 50°C. The rate of free radical formation through AIBN decomposition (initiation rate w_i) was calculated from literature data [11]. The experimental procedure is detailed elsewhere [6, 7, 12]. The tocopherolhydroquinone concentration range from 0.4 to 15 μmol/l was examined. The free radical initiation rate (w_i) was varied in an interval of $(1–9) \times 10^{-8}$ mol l^{-1} s^{-1}.

Computer simulation was carried out using Mendes' approach [8–10]. This approach has successfully been

tested in the simulation of the multistep kinetics of the antioxidant effect of tocopherol [13].

Kinetic data were interpreted on the basis of the kinetic scheme presented in the table. The elementary steps of this scheme are numbered according to the rules accepted in the domestic literature.

The rate constants of the elementary reactions used in our calculations were borrowed from [11, 14–16] and were converted to 50°C. When no literature data were available, we used estimates based on analogy with other processes and reactivity data for the reacting species [11, 16].

RESULTS AND DISCUSSION

The chemiluminescence accompanying the initiated oxidation of ethylbenzene is known to result from the interaction of peroxy radicals according to the scheme

$$\mathrm{ROO^\bullet + ROO^\bullet} \longrightarrow \mathrm{ROH + R^1{=}O^* + O_2} \longrightarrow \mathrm{R^1{=}O} + h\nu,$$

where $R^1{=}O^*$ is the resulting ketone (acetophenone) in the excited state. Therefore, $I \sim [ROO^\bullet]^2$, where I is the luminescence intensity. The inhibitor of free radical oxidation (antioxidant), which is a trap of free radicals, reacts with the peroxy radicals in reactions (VII) and (VIII) (table) and thus reduces their concentration and, accordingly, the chemiluminescence intensity.

The model system was ethylbenzene being oxidized with continuously bubbling atmospheric oxygen in the presence of an initiator (AIBN) and a luminescence activator (dibromoanthracene) [17]. The process took place as a steady-state reaction and was accompanied by chemiluminescence, whose intensity remained at a constant level of I_0. The chemiluminescence intensity decreased upon the addition of tocopherolhydroquinone, indicating a decrease in the peroxy radical concentration. The kinetic curves shown in Fig. 1 have an S-like shape typical of highly reactive antioxidants [6, 12–14, 18–22]. The chemiluminescence intensity decreases sharply upon the addition of the antioxidant owing to the decreasing peroxy radical concentration. The concentration of free radicals increases as the inhibitor (antioxidant) is consumed, and the luminescence intensity returns to its initial steady-state level.

The stoichiometric inhibition factor f was used to evaluate the antioxidant effect. The f factor is the average number of oxidation chain termination events per inhibitor molecule (InH) [14, 23]. The calculation was performed using the formula

$$f = \frac{w_\mathrm{i}}{[\mathrm{InH}]_0}\int_0^\infty\left(1 - \frac{I}{I_0}\right)\mathrm{d}t,$$

where the integral $\int_0^\infty\left(1 - \frac{I}{I_0}\right)\mathrm{d}t$ is proportional to the number of oxidation chains terminated on the molecules of the antioxidant or its transformation products [14]. The f values derived from the slopes of the straight lines representing the relationships

$$\int_0^\infty\left(1 - \frac{I}{I_0}\right)\mathrm{d}t \div [\mathrm{InH}]_0 \quad \text{and} \quad \int_0^\infty\left(1 - \frac{I}{I_0}\right)\mathrm{d}t \div \frac{1}{w_\mathrm{i}},$$

range from 0.5 (for low w_i) to 0.6. These low f values, which are also observed for ubiquinols [12, 21], indi-

Kinetic scheme of ethylbenzene (RH) oxidation in the presence of tocopherolhydroquinone (TQH_2)

Reaction		Rate constant, $l\ mol^{-1}\ s^{-1}$
$R^\bullet + O_2 \longrightarrow ROO^\bullet$	(I)	1.7×10^6
$ROO^\bullet + RH \longrightarrow ROOH + R^\bullet$	(II)	1.7
$ROO^\bullet + ROO^\bullet \longrightarrow NRP$	(VI)	10^7
$ROO^\bullet + TQH_2 \longrightarrow ROOH + TQH^\bullet$	(VII)	$(1.4–3.0) \times 10^6$
$ROO^\bullet + TQH^\bullet \longrightarrow ROOH$	(VIII)	10^8
$TQH^\bullet + TQH^\bullet \longrightarrow TQH_2 + TQ$	(IX)	5×10^8
$TQH^\bullet + RH \longrightarrow TQH_2 + ROO^\bullet$	(X)	0–10
$TQH^\bullet + O_2 \longrightarrow TQ + HOO^\bullet\ (ROO^\bullet)$	(X')	$0–10^3$

Note: NRP is the nonreactive product, $TQH^\bullet$ is tocopherolsemiquinone, and TQ is tocopherolquinone.

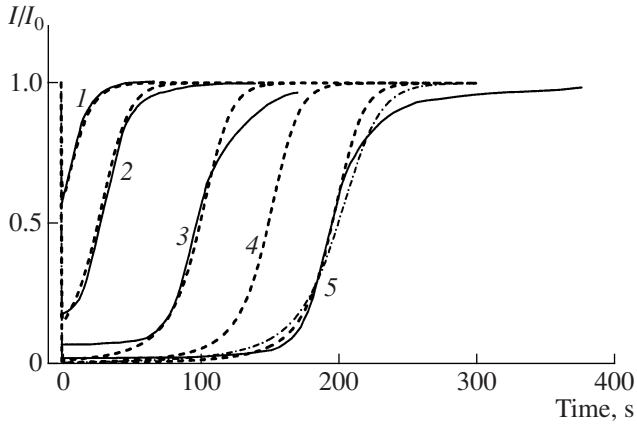


Fig. 1. Chemiluminescence curves obtained upon the addition of tocopherolhydroquinone to the system. Tocopherolhydroquinone concentrations: (*1*) 0.4, (*2*) 1.8, (*3*) 7.2, (*4*) 11.2, and (*5*) 15 μmol/l. The initiation rate is 4.4×10^{-8} mol l^{-1} s^{-1}. The solid lines represent experimental data, and the dashed lines represent the results of mathematical simulation at $k_7 = 2.4 \times 10^6$ l mol^{-1} s^{-1}, $k_{10} = 0.07$ l mol^{-1} s^{-1}, and $k_{10'} = 948$ l mol^{-1} s^{-1}. The dot-and-dash line shows the result of the mathematical simulation at $k_7 = 1.7 \times 10^6$ l mol^{-1} s^{-1}, $k_{10} = 0$, and $k_{10'} = 948$ l mol^{-1} s^{-1}.

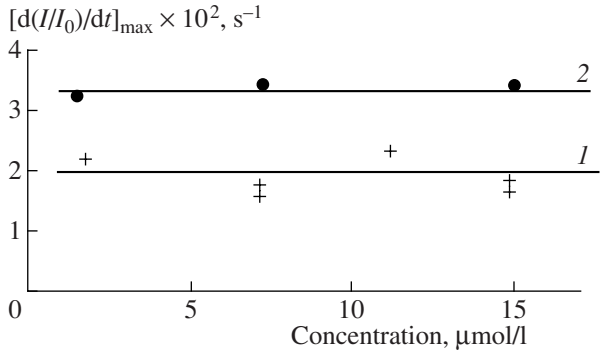


Fig. 2. Plots of $\left[\frac{d(I/I_0)}{dt}\right]_{max}$ versus the tocopherolhydroquinone concentration at initiation rates of (*1*) 4.4×10^{-8} and (*2*) 8.9×10^{-8} mol l^{-1} s^{-1}.

cate that the probability of reaction (X') is high.

The maximum slope of the kinetic curve, $\left[\frac{d(I/I_0)}{dt}\right]_{max}$, is commonly used [7, 12, 14, 18, 20, 21] in the estimation of the rate constants of free radical oxidation inhibitors by the chemiluminescence method.

In an earlier work [7], we reported the expression

$$\left[\frac{d(I/I_0)}{dt}\right]_{max} = \frac{lk_7\sqrt{w_i}\left(\sqrt{w_i} + \frac{mk_{10'}[RH]}{\sqrt{k_9}}\right)}{\sqrt{k_6}\left(\sqrt{w_i} + \frac{n(k_{10} + k_{10'})[RH]}{\sqrt{k_9}}\right)}, \quad (1)$$

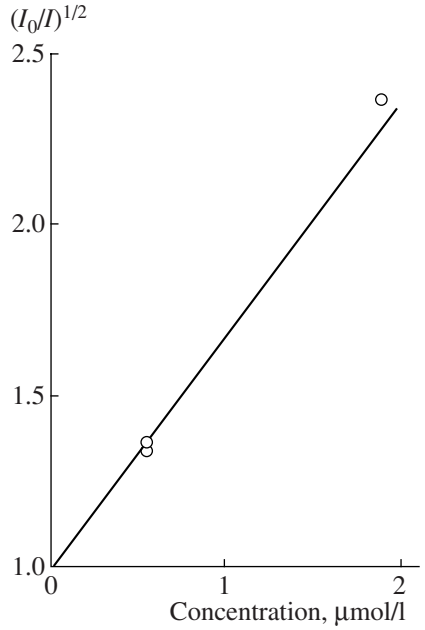


Fig. 3. Plot of the luminescence intensity versus the tocopherolhydroquinone concentration at an initiation rate of 4.4×10^{-8} mol l^{-1} s^{-1}.

where l, m, and n are constants varying in narrow ranges and depending on the ratios of the k_6, k_8, k_9, k_{10}, and $k_{10'}$ values. It follows from Eq. (1) that $\left[\frac{d(I/I_0)}{dt}\right]_{max}$ is independent of the concentration of the inhibitor (antioxidant) introduced. Indeed, in the tocopherolhydroquinone concentration range examined, this quantity remains almost invariable (Fig. 2), confirming that the above kinetic scheme is valid. Another confirmation of the accepted kinetic scheme [7, 14] is the nearly linear concentration dependence of the parameter $\sqrt{\frac{I_0}{I}}$ (Fig. 3).

The coefficient l in Eq. (1) was shown to take values of 0.12 to 0.24 [7]. For this reason, an intermediate value of $l = 0.18$ was used in the preliminary estimation of k_7 from experimental $\left[\frac{d(I/I_0)}{dt}\right]_{max}$ data by means of formula (1). For the sake of simplicity, at this stage, we also accepted that $k_{10} = k_{10'} = 0$. As a result, we obtained $k_7 = 1.7 \times 10^6$ l mol^{-1} s^{-1}. However, the substitution of this value into the set of differential equations used in mathematical simulation demonstrated that this value is underestimated: the calculated slope of the luminescence curves, $\left[\frac{d(I/I_0)}{dt}\right]_{max}$, was far below the experimental value (Fig. 1). The curves obtained at $k_7 = 2.4 \times 10^6$ l mol^{-1} s^{-1} are in best agreement with the experimental data (Fig. 1).

Next, using the mathematical model, we studied the effect of the rate constants k_{10} and $k_{10'}$ on the shape of the chemiluminescence curves. The model kinetic

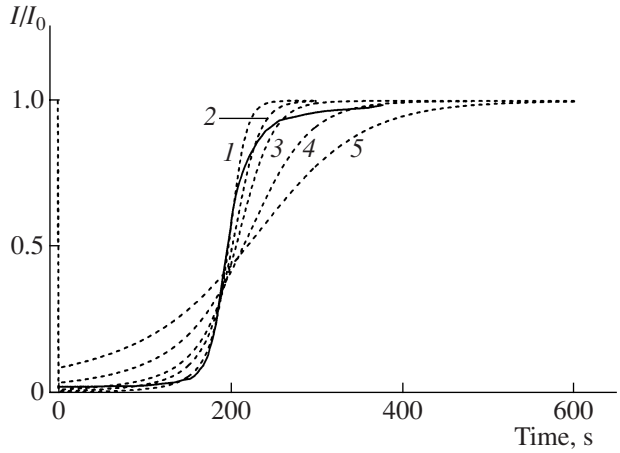


Fig. 4. Simulation of the kinetics of the antioxidant effect of tocopherolhydroquinone (15 μmol/l) at different values of k_{10}. The solid line represents experimental data, and the dashed lines represent the results of kinetic simulation at $k_7 = 2.4 \times 10^6$ l mol^{-1} s^{-1}, $k_{10'} = 948$ l mol^{-1} s^{-1}, and $k_{10} \leq$ (*1*) 0.07, (*2*) 1.0, (*3*) 2.0, (*4*) 5.0, and (*5*) 10 l mol^{-1} s^{-1}.

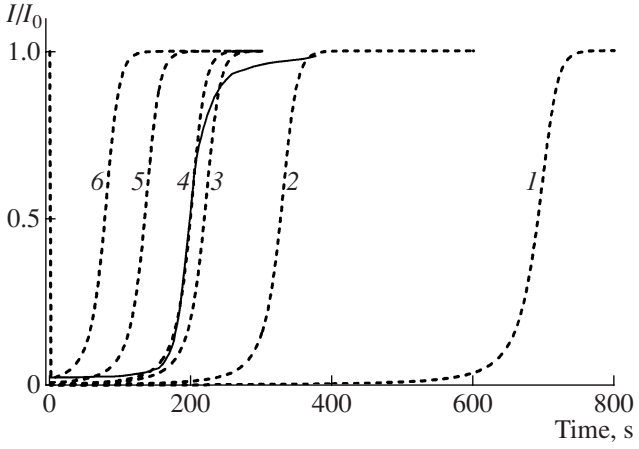


Fig. 5. Simulation of the kinetics of the antioxidant effect of tocopherolhydroquinone (15 μmol/l) at different values of $k_{10'}$. The solid line represents experimental data, and the dashed lines represent the results of kinetic simulation at $k_7 = 2.4 \times 10^6$ l mol^{-1} s^{-1}, $k_{10} \le 0.07$ l mol^{-1} s^{-1}, and $k_{10'} =$ (*1*) 0, (*2*) 410, (*3*) 820, (*4*) 948, (*5*) 1640, and (*6*) 3270 l mol^{-1} s^{-1}.

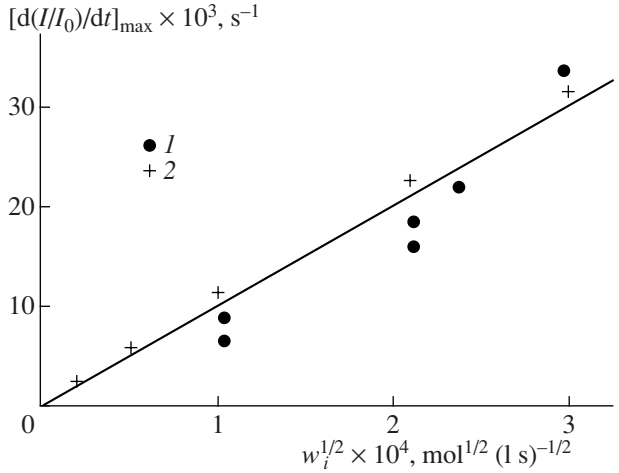


Fig. 6. Plot of $\left[\frac{d(I/I_0)}{dt}\right]_{max}$ versus the initiation rate in the presence of tocopherolhydroquinone (15 μmol/l): (*1*) experimental and (*2*) calculated data.

curves obtained for $k_{10} \le 0.07$ l mol^{-1} s^{-1} almost coincide (Figs. 4, 5). The constant k_{10} affects only the maximum slope of the curves and does not change the induction period (Fig. 4). In essence, reaction (X) smoothens the sharp transition from the inhibited process to the uninhibited process, being a backward step in the binding of the reactive peroxy radicals. Conversely, the constant $k_{10'}$ shortens the induction period, leaving unchanged the maximum slope of the chemiluminescence curves (Fig. 5). This is explained by the fact that, in this route, the peroxy radical returns to the system without regenerating an antioxidant molecule.

The best fit between the experimental and calculated kinetic curves is achieved at $k_7 \pm$ SD = $(2.4 \pm 0.1) \times 10^6$ l mol^{-1} s^{-1}, $k_{10} \le 0.07$ l mol^{-1} s^{-1}, and $k_{10'} = 950 \pm 14$ l mol^{-1} s^{-1} (SD is the standard deviation). At these values of the constants, the plot of $\left[\frac{d(I/I_0)}{dt}\right]_{max}$ versus the initiation rate presented in Fig. 6 also fits the calculated plot.

Thus, tocopherolhydroquinone shows a high antiradical activity (rate constant k_7) comparable with the activities of tocopherol [18, 20] and substituted pyrocatechols [19]. The set of the rate constants of elementary reactions reported here can be used in the description and simulation of the kinetics of the antioxidant effect of tocopherolhydroquinone.

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