ISSN 1070-4280, Russian Journal of Organic Chemistry, 2010, Vol. 46, No. 3, pp. 399–405. © Pleiades Publishing, Ltd., 2010.
Original Russian Text © I.A. Khalfina, N.V. Vasil'eva, I.G. Irtegova, L.A. Shundrin, V.A. Reznikov, 2010, published in Zhurnal Organicheskoi Khimii, 2010, Vol. 46, No. 3, pp. 405–411.

Synthesis and Electrochemical Reduction of 2,2′-Diaryl-5,5,5′,5′-tetramethyl-3,3′-bi(pyrrol-3-ylidene)-4,4′(5*H*,5′*H*)-dione 1,1′-Dioxides

I. A. Khalfina[a,b], N. V. Vasil'eva[a], I. G. Irtegova[a], L. A. Shundrin[a], and V. A. Reznikov[a,b]

[a] *Vorozhtsov Novosibirsk Institute of Organic Chemistry, Siberian Division, Russian Academy of Sciences, pr. Akademika Lavrent'eva 9, Novosibirsk, 630090 Russia*
e-mail: shundrin@nioch.nsc.ru

[b] *Novosibirsk State University, Novosibirsk, Russia*

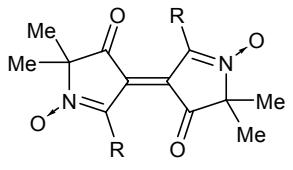
Received November 11, 2008

Abstract—2,2′-Diaryl-5,5,5′,5′-tetramethyl-3,3′-bi(pyrrol-3-ylidene)-4,4′(5*H*,5′*H*)-dione 1,1′-dioxides containing a carboxy, alkoxycarbonyl, or carbamoyl group in the *para* position of one or both benzene rings were synthesized. These compounds may be regarded as cyclic dinitrones with conjugated C=C bond. Mild aminolysis of carboxy groups in the title compounds may be used to introduce dinitrone fragments into oligonucleotide or polypeptide structures. Electrochemical reduction of the resulting amides involves reversible one-electron transfer in the first step at a near-zero potential, which makes it possible to use the title compounds as electrochemically active labels in applied bioorganic electrochemistry.

DOI: 10.1134/S1070428010030176

We previously reported [1, 2] on electrochemical behavior of 2,2′-substituted 5,5,5′,5′-tetramethyl-3,3′-bi(pyrrol-3-ylidene)-4,4′(5*H*,5′*H*)-dione 1,1′-dioxides **I** (R = Me, Ph, *t*-Bu, CF_3) which may be regarded as cyclic dinitrones with conjugated C=C bond. Electrochemical reduction of dinitrones in dimethylformamide and acetonitrile is an EE process including two reversible steps which give rise to long-lived (at room temperature) radical anions. The latter were characterized by ESR spectroscopy.

The potentials of the first one-electron reduction peaks of dinitrones **I** in aprotic solvents are very small in absolute value.* They become less negative as the concentration of water in MeCN–H_2O mixtures increases and are close to zero in water (relative to a saturated calomel electrode), and the first step of electrochemical reduction of dinitrones retains its reversible and one-electron character [3]. The potentials of electrochemical reduction of the examined dinitrones (except for trifluoromethyl-substituted derivative) approach the range typical of redox potentials of ferrocene derivatives that are widely used in the recent time as reporter groups** in oligonucleotides covalently immobilized on the surface of modified microelectrodes for electrochemical detection of DNA hybridization [4, 5], in particular while developing DNA microchips for gene diagnostics [6]. It is quite probable that dinitrones functionalized at one R substituent may be promising as reporter labels, which is important from the viewpoint of extension of a set of reporter groups with different characteristic potentials [7].



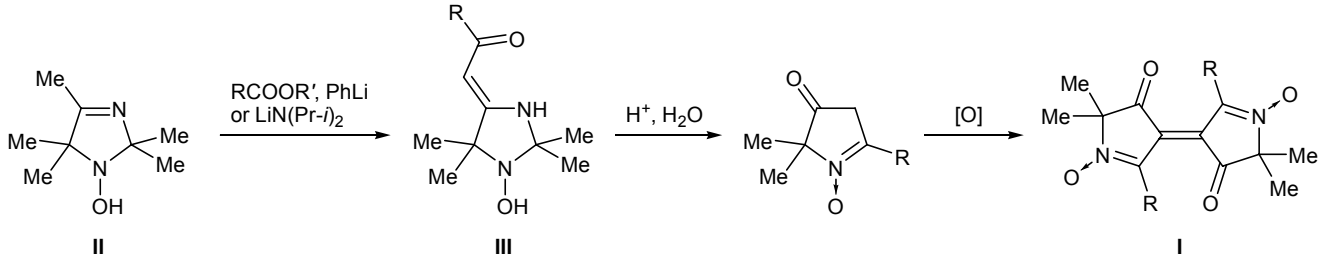
I

R = Me, Ph, *t*-Bu, F_3C, 4-$HOCOC_6H_4$, 4-$EtOCOC_6H_4$, 4-*i*-$PrNHCOC_6H_4$.

* Electrochemical potentials of first reduction peaks of dinitrones **I** (R = Me, Ph, *t*-Bu, CF_3) are, respectively, –0.58, –0.55, –0.52, and –0.18 V (relative to a saturated calomel electrode) at a platinum electrode in a 0.1 M solution of Et_4NClO_4, v = 0.1 V/s in MeCN [2] and –0.14, –0.09, –0.08, and 0.19 V in a 0.1 M solution of $LiClO_4$, v = 0.1 V/s in H_2O [3].

** Introduction of reporter groups (labels) into oligonucleotides makes it possible to study them by spectrophotometric, electrochemical, and other methods.

Scheme 1.



Symmetric structure of dinitrones functionalized at both R substituents could give rise to dimeric structures (e.g., polypeptide) with a central group capable of transferring an electron in a reversible mode. On this basis, artificial biologically important compounds ensuring reversible electron transport may be designed in the future.

A standard procedure for the introduction of electrochemically active groups into modified oligonucleotide*** or polypeptide structures is based on aminolysis of the corresponding carboxylic acid esters. Therefore, the goal of the present work was to synthesize symmetrical and unsymmetrical dinitrones with R substituents having a carboxy or alkoxycarbonyl group, convert them into the corresponding amides (this process should simulate covalent binding of dinitrone to terminal amino groups in linkers of modified oligonucleotides or polypeptides), and examine the ability of the compounds thus obtained to undergo reversible one-electron transfer in the first step of electrochemical reduction. As subjects for study we selected dinitrone derivatives **I** containing the above substituents in the *para* position of the benzene ring.

Dinitrones **I** are generally synthesized via oxidative dimerization of dihydropyrrolones formed by recyclization of enamino ketones [8]. The latter are prepared by condensation of 2,2,4,5,5-pentamethyl-2,5-dihydro-1*H*-imidazol-1-ol (**II**) with carboxylic acid ester in the presence of phenyllithium or lithium diisopropylamide (Scheme 1) [9]. It is seen from Scheme 1 that substituted carboxylic acid esters may be used to obtain dinitrones having functional groups. As shown in [10], the reaction of compound **II** with dimethyl terephthalate in the presence of lithium diisopropylamide gave the corresponding condensation product at one ester group in a poor yield (10%). The yield of enamino ketone **III** may be increased to 55%, and the subsequent reaction of **III** with $(i\text{-Pr})_2NH$ (leading to the corresponding diisopropylamide [10]) may be avoided by replacing dimethyl terephthalate by diethyl terephthalate which is readily soluble in diethyl ether.

When enamino ketone **III** (Scheme 2) was heated in aqueous–alcoholic solution of HCl, it underwent recyclization to 1-hydroxy-4,5-dihydropyrrol-4-one **IV**, while the ester group in the *para* position of the benzene ring remained unchanged. Acid **V** was obtained by hydrolysis of **IV** in the presence of a base and subsequent neutralization of the reaction mixture.

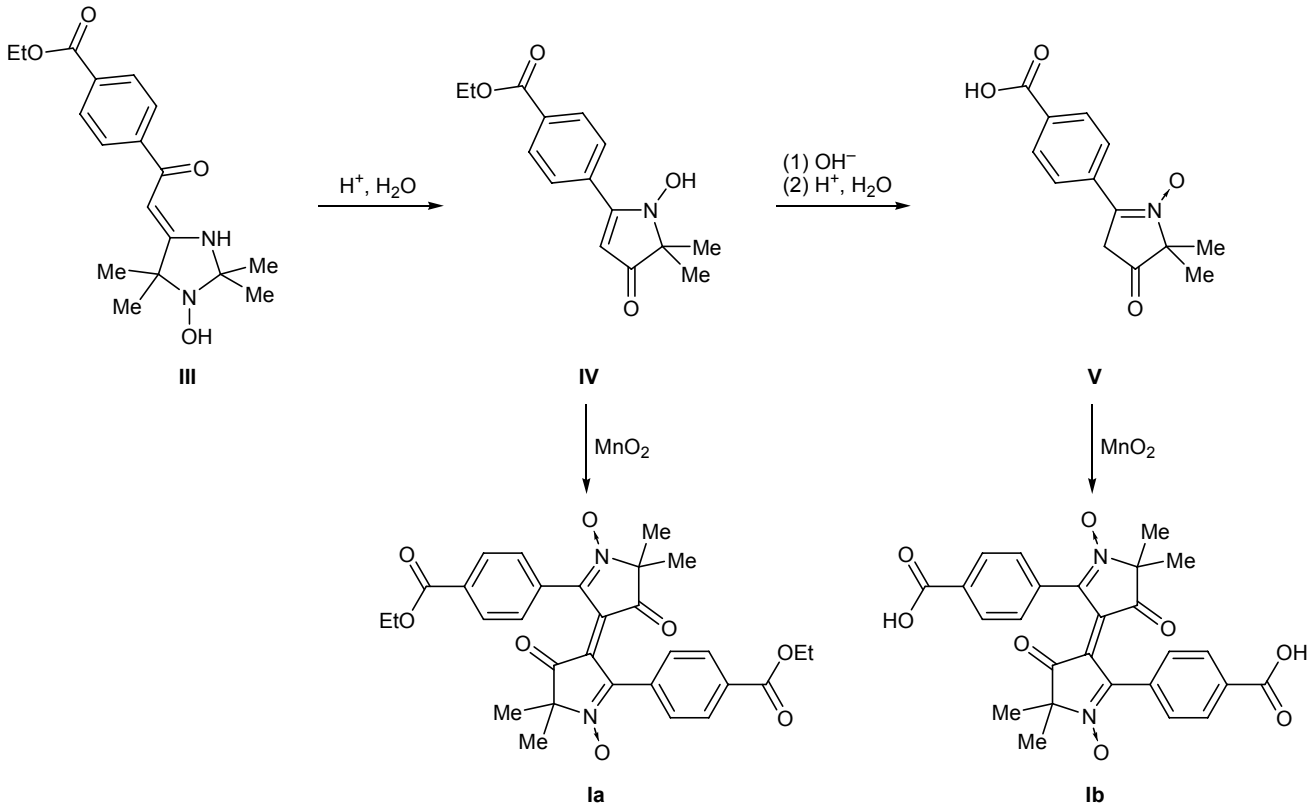
Oxidative dimerization of dihydropyrrolones **IV** and **V** by the action of manganese(IV) oxide in acetonitrile gave dinitrones **Ia** and **Ib**, respectively. The reaction of ester **Ia** with isopropylamine under the conditions described in [11] resulted in the formation of a complex mixture of products; therefore, this reaction is hardly suitable for the introduction of dinitrone fragment into oligonucleotide or polypeptide molecules.

In order to obtain dinitrone **Ic** having one carboxy group, a mixture of compound **V** and 2,2-dimethyl-5-phenyl-3,4-dihydro-2*H*-pyrrol-3-one 1-oxide (**VI**) was subjected to oxidation with MnO_2 (Scheme 3). In this case the yield of unsymmetrical dinitrone **Ic** was 27%, i.e., almost twice as low as that reported in [8] for nitrones with similar reactivities.

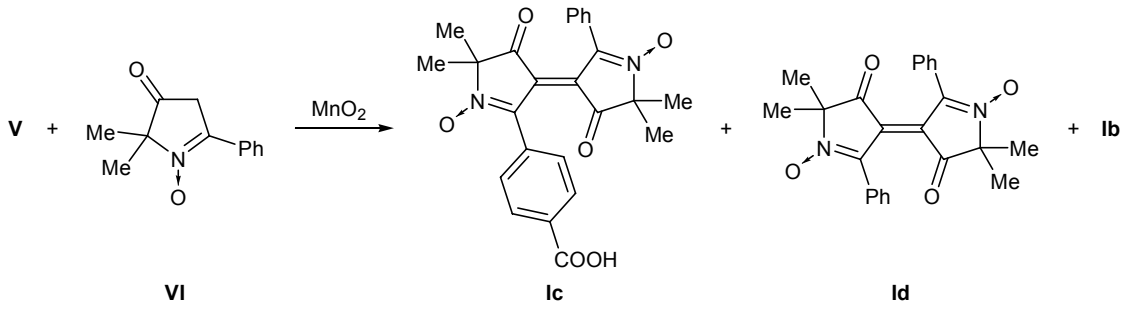
One of the most widely used methods for the introduction of reporter groups into oligonucleotide structures is based on the activation of the corresponding carboxylic acids with *N*-hydroxysuccinimide and subsequent reaction of reactive ester derivative with amino group in the linker of modified oligonucleotide at pH ~7. A necessary condition is conservation of electrochemical properties of the reporter group incorporated into oligonucleotide chain. With a view to simulate such process we synthesized diamide **Ie** and amide **If** by reaction of compounds **Ib** and **Ic**, respectively, with *N*-hydroxysuccinimide in the presence of *N*,*N*′-dicyclohexylcarbodiimide (DCC), followed by

*** Standard modification of oligonucleotides is performed at the 3′- or 5′-terminal groups with aminohexyl linkers (e.g., in automated phosphamidite synthesis).

Scheme 2.



Scheme 3.

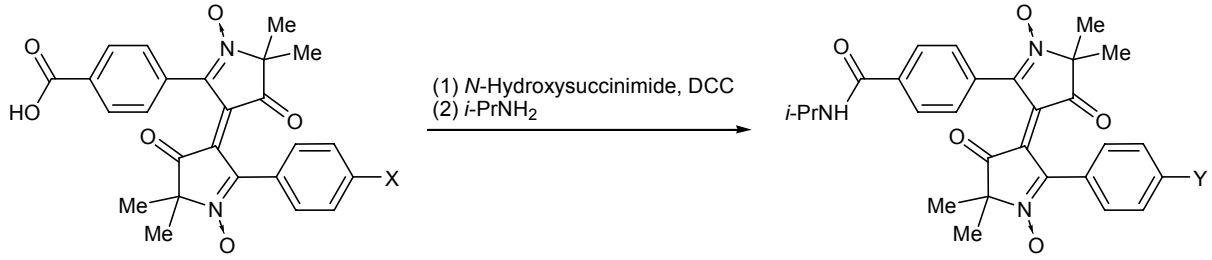


treatment with isopropylamine in a 1:1 mixture of acetonitrile with aqueous phosphate buffer (pH 7.01) (Scheme 4).

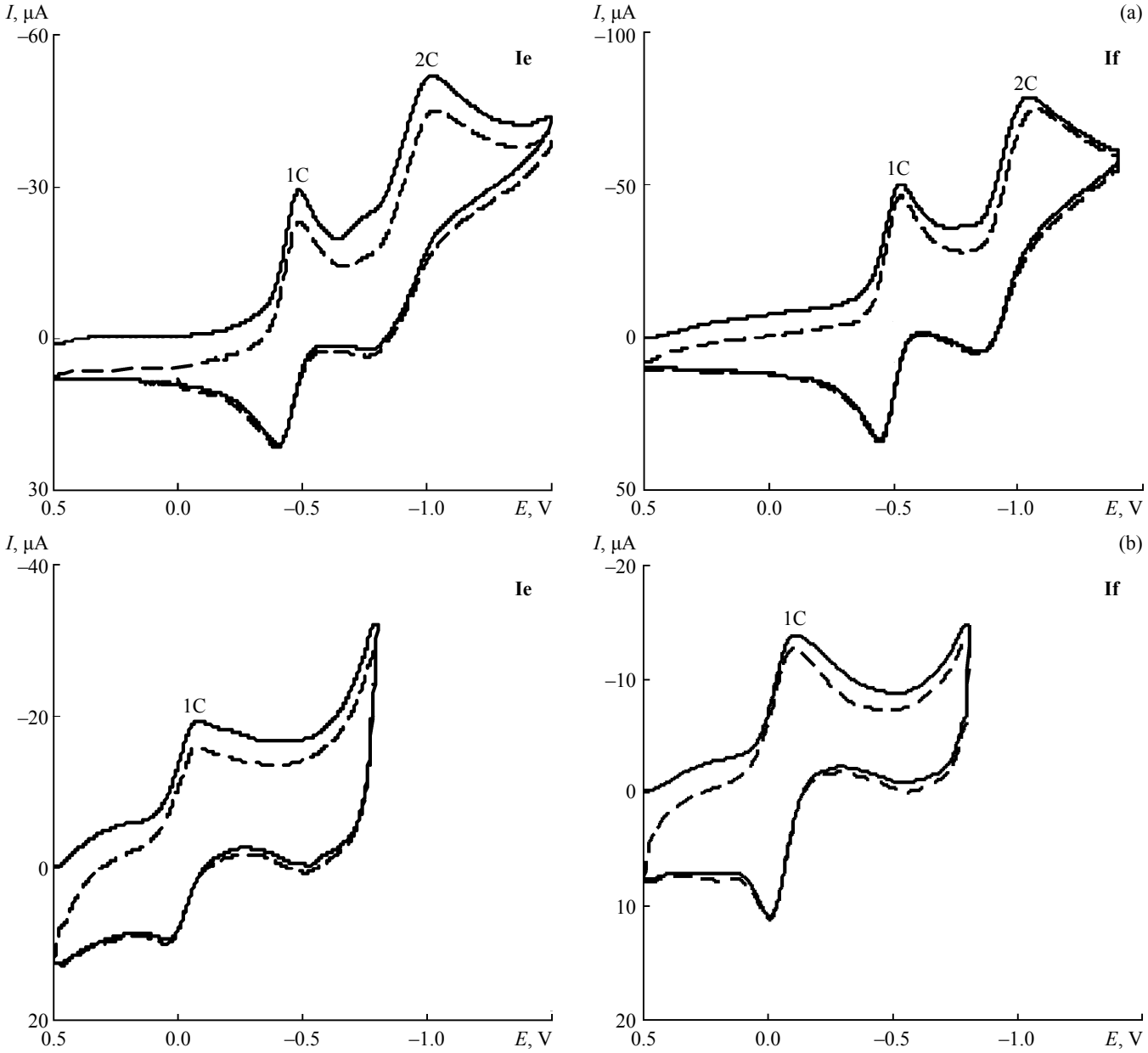
Electrochemical reduction of dinitrones **Ia**, **Ie**, and **If** was studied by cyclic voltammetry in acetonitrile and water. The reduction of carboxy derivatives **Ib** and **Ic** was not studied. Electrochemical reduction of diester **Ia** in acetonitrile was an EE process including two reversible one-electron transfer steps. In the reduction of amides **Ie** and **If** the first reduction peaks (1C) were one-electron, reversible ($E_r^{1A} - E_r^{1C} = 0.06$ V), and diffusion-controlled ($i_p v^{-1/2}$ = const, where i_p is the peak current, and v is the potential sweep rate). The second peak corresponding to the formation of dianion is quasi-reversible for monoamide **If** and irreversible for diamide **If**, and their electrochemical reduction is an EEC process (see figure). The reduction peak potentials are given in Table 1.

In the electrochemical reduction of dinitrones **Ia**, **Ie**, and **If** in acetonitrile, the first peak potential corresponds to the formation of radical anions which were characterized by ESR (Table 2). In all cases, both ^{14}N nuclei in the N-oxide groups are spectrally equivalent, and the hyperfine structure (HFS) is represented by a quintet with an intensity ratio of 1:2:3:2:1 and is analogous to HFS of the ESR spectrum of radical anion derived from diphenyl derivative **Id** [1, 2]. Symmetry violation in going to amide **If** radical anion

Scheme 4.



X = COOH (**b**), H (**c**); Y = CONHPr-*i* (**e**), H (**f**).



Cyclic voltammograms for the reduction of dinitrones **Ie** and **If** in (a) acetonitrile and (b) water [v = 0.2 V/s; for dinitrone **Ie** in H_2O, v = 1 V/s]; the second potential sweep cycle is shown with dashed line.

Table 1. Peak potentials for electrochemical reduction of dinitrones **Ia**, **Id**, **Ie**, and **If** in acetonitrile and water[a]

Compound no.	R	Solvent	E_r^{1C}, [b] V	E_r^{1A}, V	E_r^{2C}, V	E_r^{2A}, V
Ia	C_6H_4COOEt-4	MeCN	–0.48	–0.42	–0.98	–0.88
Ie	$C_6H_4CONHPr$-*i*-4[b]	MeCN	–0.49	–0.43	–1.03	–
		H_2O	–0.07	–0.01	–	–
If	$C_6H_4CONHPr$-*i*, Ph[b]	MeCN	–0.53	–0.46	–1.01	–0.90
		H_2O	–0.08	–0.02	–	–
Id	Ph	MeCN [2]	–0.55	–0.49	–1.05	–0.95
		H_2O [3]	–0.09	–0.03	–	–

[a] Potential sweep rate v = 0.1 V/s; supporting electrolyte Et_4NClO_4 (0.1 M) for MeCN or $LiClO_4$ (0.1 M) for H_2O; saturated calomel electrode as reference.
[b] See figure.

does not affect spectral equivalence of the N→O nitrogen atoms, and no hyperfine coupling with protons in the benzene rings and nitrogen nuclei in the amide groups is observed due to essential noncoplanarity of the benzene and pyrrole rings contributing mainly to the singly occupied molecular orbital of the radical anion (cf. [1]); in this case, benzene rings may be regarded as linkers.

Cyclic voltammograms for the reduction of dinitrones **Ie** and **If** in water (see figure) in the potential sweep range from 0.5 to –0.8 V display a single reversible one-electron peak with a near-zero potential (Table 1). Introduction of a carbamoyl group into the *para* position of one or two benzene rings does not affect the potential of the first reduction peak to an appreciable extent. The first reduction step of compounds **Ie** and **If** retains its reversible and one-electron character both in acetonitrile and in water, whereas low reduction potentials make it possible to use dinitrones **Ie** and **If** as electrochemically active groups in applied bioorganic electrochemistry.

The synthesis of monoamide derivatives **If** from the corresponding carboxylic acid through intermediate *N*-hydroxysuccinimide ester may be regarded as a method for the introduction of a dinitrone fragment into molecules of modified oligonucleotides with a terminal amino group with a view to obtain DNA markers containing a novel electrochemically active reporter group.

EXPERIMENTAL

The IR spectra were recorded in KBr on a Bruker Vector 22 spectrometer. The ^{1}H NMR spectra were measured on a Bruker AV-300 instrument from solutions in $CDCl_3$. The elemental compositions were determined at the Microanalysis Laboratory, Novosibirsk Institute of Organic Chemistry, Siberian Division, Russian Academy of Sciences. Silica gel (5–100 μm) manufactured at the Novosibirsk Institute of Organic Chemistry was used for column and preparative thin-layer chromatography. 2,2,4,5,5-Pentamethyl-2,5-dihydro-1*H*-imidazol-1-ol (**II**) was synthesized according to the procedure described in [12].

Ethyl 4-[2-(1-hydroxy-2,2,5,5-tetramethylimidazolidin-4-ylidene)acetyl]benzoate (III). A solution of 1 g (6.4 mmol) of *N*-hydroxydihydroimidazole **II** in 20 ml of anhydrous diethyl ether was added with stirring under argon to a solution of phenyllithium prepared from 2 ml (19.2 mmol) of bromobenzene and 0.29 g (38.4 mmol) of metallic lithium in 30 ml of anhydrous diethyl ether. The mixture was stirred for 30 min, cooled to –10°C, a solution of 3.5 g (15.8 mmol) of diethyl terephthalate in 15 ml of anhydrous diethyl ether was added in one portion, and the mixture was stirred first for 30 min at –5°C and then for 1.5 h at 20°C. The mixture was treated with 10 ml of water, the organic phase was separated, and the aqueous phase was extracted with diethyl ether (3× 30 ml). The extracts were combined with the organic phase and dried over $MgSO_4$, the solvent was distilled off, the residue was diluted with 10 ml of hexane, and the precipitate of **III** was filtered off and purified by

Table 2. Isotropic hyperfine coupling constants for radical anions derived from dinitrones **Ia**, **Ie**, and **If** in acetonitrile

Compound no.	R	a_H, G
Ia	C_6H_4COOEt-4	4.320
Ie	$C_6H_4CONHPr$-*i*-4	4.320
If	$C_6H_4CONHPr$-*i*-4, Ph	4.275

column chromatography using chloroform as eluent. Yield 55%, mp 171–173°C. IR spectrum, ν, cm^{-1}: 3200–3500 (OH), 3303 (NH), 1703 (C=O, ester), 1607 (C=O). ^{1}H NMR spectrum, δ, ppm: 1.39 t (3H, CH_2CH_3, J = 7.1 Hz), 1.42 s (6H, 2-CH_3), 1.48 s (6H, 5-CH_3), 4.38 q (2H, CH_2CH_3, J = 7.1 Hz), 4.86 br.s (OH), 5.66 s (1H, =CH) 7.89 d (2H, *o*-H, J = 8.3 Hz), 8.06 d (2H, *m*-H, J = 8.3 Hz). Found, %: C 65.01; H 7.25; N 8.49. $C_{18}H_{24}N_2O_4$. Calculated, %: C 65.06; H 7.23; N 8.43.

Ethyl 4-(1-hydroxy-5,5-dimethyl-4-oxo-4,5-dihydro-1*H*-pyrrol-2-yl)benzoate (IV). A suspension of 0.3 g (1.1 mmol) of enamino ketone **III** in a mixture of 5.5 ml of ethanol and 5.5 ml of 10% hydrochloric acid was kept for 72 h at 20°C (until the precipitate dissolved completely). The solvent was distilled off under reduced pressure (2 mm) at room temperature, the residue was diluted with 3 ml of hexane, and the precipitate was filtered off and purified by preparative thin-layer chromatography using chloroform–ethyl acetate (1:1) as eluent. Yield 87%, mp 120–122°C. IR spectrum, ν, cm^{-1}: 1760, 1563 (C=O, C=N), 1714 (C=O, ester). ^{1}H NMR spectrum, δ, ppm: 1.37 t (3H, CH_2CH_3, J = 7.0 Hz), 1.60 s (6H, CH_3), 4.37 q (2H, CH_2CH_3, J = 7.0 Hz), 6.16 s (1H, 3-H), 8.11 br.s (4H, H_{arom}). Found, %: C 65.45; H 6.20; N 4.99. $C_{15}H_{17}NO_4$. Calculated, %: C 65.45; H 6.18; N 5.09.

4-(2,2-Dimethyl-1-oxido-3-oxo-3,4-dihydro-2*H*-pyrrol-5-yl)benzoic acid (V). A solution of 0.012 g (0.05 mmol) of compound **IV** in 0.2 ml of 30% aqueous sodium hydroxide was stirred for 2 h at 20°C. The mixture was acidified with 20% hydrochloric acid to pH 3, and the precipitate was filtered off and purified by preparative thin-layer chromatography using acetonitrile as eluent. Yield 80%, decomposes above 150°C. IR spectrum, ν, cm^{-1}: 3400 (OH), 1766, 1532 (C=O, C=N), 1700 (COOH). ^{1}H NMR spectrum, δ, ppm: 1.56 s (6H, CH_3), 3.80 s (2H, 4-H), 7.67 d (2H, *o*-H, J = 8.5 Hz), 8.42 d (2H, *m*-H, J = 8.5 Hz). Found, %: C 63.11; H 5.30; N 5.62. $C_{13}H_{13}NO_4$. Calculated, %: C 63.16; H 5.26; N 5.67.

Ethyl 4-{3-[2-(4-ethoxycarbonylphenyl)-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-3-ylidene]-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-2-yl}benzoate (Ia). Manganese(IV) oxide, 0.01 g, was added to a solution of 0.006 g (0.02 mmol) of compound **IV** in 1 ml of chloroform, and the mixture was stirred for 1 h at 20°C. The precipitate (manganese oxides) was filtered off and washed with chloroform (2×1 ml), the filtrate was evaporated under reduced pressure (2 mm) at room temperature, and the residue was purified by preparative thin-layer chromatography using ethyl acetate as eluent. Yield 82%, mp 230–235°C. IR spectrum, ν, cm^{-1}: 1720 (C=O, ester; C=C–C=O). ^{1}H NMR spectrum, δ, ppm: 1.39 t (6H, CH_2CH_3, J = 7.2 Hz), 1.40 s (12H, CH_3), 4.39 q (4H, CH_2CH_3, J = 7.2 Hz), 7.67 br.s (4H, *o*-H), 8.11 br.s (4H, *m*-H). Found, %: C 65.89; H 5.51; N 5.21. $C_{30}H_{30}N_2O_8$. Calculated, %: C 65.93; H 5.49; N 5.13.

4-{3-[2-(4-Carboxyphenyl)-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-3-ylidene]-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-2-yl}benzoic acid (Ib) was synthesized in a similar way by oxidation of compound **V**. Yield 80%, decomposes above 270°C. IR spectrum, ν, cm^{-1}: 3400 (OH), 1720 (C=C–C=O), 1700 (COOH). ^{1}H NMR spectrum, δ, ppm: 1.38 s (12H, CH_3), 7.83 br.s (8H, H_{arom}). Found, %: C 63.71; H 4.48; N 5.68. $C_{26}H_{22}N_2O_8$. Calculated, %: C 63.67; H 4.49; N 5.71.

4-[3-(5,5-Dimethyl-1-oxido-4-oxo-2-phenyl-4,5-dihydro-3*H*-pyrrol-3-ylidene)-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-2-yl]benzoic acid (Ic). Manganese(IV) oxide, 0.12 g, was added to a solution of 0.08 g (0.4 mmol) of compound **V** and 0.1 g (0.4 mmol) of 2,2-dimethyl-5-phenyl-3,4-dihydro-2*H*-pyrrol-3-one 1-oxide (**VI**) in 5 ml of acetonitrile, and the mixture was stirred for 1 h at 20°C. The precipitate of manganese oxides was filtered off and washed with acetonitrile (2×1 ml), and the filtrate was evaporated under reduced pressure (2 mm) at room temperature. Yield 27%, mp 117–120°C. IR spectrum, ν, cm^{-1}: 3440 (OH), 1718 (COOH), 1613 (C=C–C=O). ^{1}H NMR spectrum, δ, ppm: 1.45 s (12H, CH_3), 7.45 br.s (2H, *o*-H), 7.61 br.s (5H, C_6H_5), 8.20 br.s (2H, *m*-H). Found, %: C 67.19; H 4.95; N 6.33. $C_{25}H_{22}N_2O_6$. Calculated, %: C 67.26; H 4.93; N 6.28.

Dinitrones **Ib**–**Id** were isolated by preparative thin-layer chromatography using ethyl acetate as eluent. The yields of **Ib** and **Id** were 53 and 60%, respectively.

***N*-Isopropyl-4-{3-[2-(4-isopropylcarbamoylphenyl)-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-3-ylidene]-5,5-dimethyl-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-2-yl}benzamide (Ie).** A solution of 0.045 g (0.09 mmol) of dinitrone **Ib**, 0.04 g (0.2 mmol) of *N*,*N*′-dicyclohexylcarbodiimide, and 0.03 g (0.25 mmol) of *N*-hydroxysuccinimide in 5 ml of acetonitrile was stirred for 12 h at 20°C. A solution of 0.015 ml (0.37 mmol) of isopropylamine in a mixture of 1.5 ml of acetonitrile and 1.5 ml of phosphate buffer (pH 7.01) was then added, and the mixture was stirred for 4 h at 20°C. The solvent was distilled off at 20°C under reduced pressure (2 mm), and the residue

was purified by preparative thin-layer chromatography using acetone as eluent. Yield 21%, mp 145–148°C. IR spectrum, ν, cm^{-1}: 3325 (NH), 1740, 1635, 1539 (C=C–C=O; C=O, amide). ^1H NMR spectrum, δ, ppm: 1.29 d [12H, $CH(CH_3)_2$, J = 6.7 Hz], 1.43 br.s (12H, CH_3), 4.29 m [2H, $CH(CH_3)_2$], 6.00 br.s (2H, NH), 7.68 br.s (4H, *o*-H), 7.80 br.s (4H, *m*-H). Found, %: C 67.10; H 6.30; N 9.84. $C_{32}H_{36}N_4O_6$. Calculated, %: C 67.13; H 6.29; N 9.79.

4-[5,5-Dimethyl-3-(5,5-dimethyl-1-oxido-4-oxo-2-phenyl-4,5-dihydro-3*H*-pyrrol-3-ylidene)-1-oxido-4-oxo-4,5-dihydro-3*H*-pyrrol-2-yl]-*N*-isopropylbenzamide (If) was synthesized in a similar way from dinitrone **Ic.** Yield 25%, mp 121–123°C. IR spectrum, ν, cm^{-1}: 3339 (NH), 1721, 1644, 1535 (C=C–C=O; C=O, amide). ^1H NMR spectrum, δ, ppm: 1.27 d [6H, $CH(CH_3)_2$, J = 6.8 Hz], 1.40 s (12H, CH_3), 4.28 m [1H, $CH(CH_3)_2$], 6.03 br.s (1H, NH), 7.44 br.s (2H, *o*-H), 7.60 br.s (5H, C_6H_5), 7.80 br.s (2H, *m*-H). Found, %: C 68.95; H 6.00; N 8.71. $C_{28}H_{29}N_3O_5$. Calculated, %: C 68.99; H 5.95; N 8.62.

Cyclic voltammetry. Cyclic voltammograms of dinitrones **Ia**, **Ie**, and **If** were obtained using an SVA-1BM electrochemical system (Bulgaria) equipped with a LAB-MASTER polyfunctional interface (Institute of Nuclear Physics, Novosibirsk, Russia) which allowed total digital control over the system. Measurements were performed in the symmetric triangular pulse mode at a potential sweep rate ν of 0.1–1 V/s. A 5-ml electrochemical cell was connected to the system through a three-electrode scheme. A 8-mm^2 stationary cylindrical platinum electrode was used as working one, a platinum helix was an auxiliary electrode, and an aqueous saturated calomel electrode was used as reference. Electrochemical data were obtained for 1×10^{-3} M solutions in acetonitrile with a 0.1 M solution of tetraethylammonium perchlorate as supporting electrolyte. A 0.1 M solution of lithium perchlorate was used as supporting electrolyte in electrochemical reduction in water (by analogy with the procedure described in [3]). The concentration of depolarizer corresponded to a saturated solution in water at 298 K. Working solutions were deoxygenated by purging with argon. Acetonitrile was dehydrated by distillation over potassium permanganate, followed by double distillation over phosphoric anhydride.

The ESR spectra of radical anions derived from dinitrones **Ia**, **Ie**, and **If** were measured on a Bruker ESP-300 radiospectrometer equipped with a double resonator. Electrochemical reduction of **Ia**, **Ie**, and **If** in combionation with ESR spectroscopy was performed under oxygen-free conditions at a potential corresponding to the first reduction peak (298 K) in a standard three-electrode cell with a platinum cathode for measurement of ESR spectra of electrochemically generated paramagnetic species. The working part (cathode) of the cell was placed into the probe of ESR spectrometer. Numerical simulation of the ESR spectra of radical anions was performed using Winsim 2002 program [13] with SIMPLEX multiparameter optimization algorithm.

This study was performed under financial support by the Russian Foundation for Basic Research (project no. 06-03-32859-a).

REFERENCES

1. Shundrin, L.A., Reznikov, V.A., Irtegova, I.G., and Starichenko, V.F., *Izv. Ross. Akad. Nauk, Ser. Khim*., 2003, p. 892.
2. Shundrin, L.A., Irtegova, I.G., Rogachev, A.D., and Reznikov, V.A., *Izv. Ross. Akad. Nauk, Ser. Khim*., 2005, p. 1148.
3. Shundrin, L.A., Vasil'eva, N.V., Irtegova, I.G., and Reznikov, V.A., *Izv. Ross. Akad. Nauk, Ser. Khim*., 2007, p. 1227.
4. Zatsepin, T.S., Andreev, S.Yu., Gianik, T., and Oretskaya, T.S., *Usp. Khim*., 2003, vol. 72, p. 602.
5. Anne, A., Bouchardon, A., and Moiroux, J., *J. Am. Chem. Soc*., 2003, vol. 125, p. 1112.
6. Abramowitz, S., *J. Biomed. Microdev*., 1999, vol. 1, p. 107.
7. Di Giusto, D.A., Wlassoff, W.A., Giesebrecht, S., Gooding, J.J., and King, G.C., *J. Am. Chem. Soc*., 2004, vol. 126, p. 4120.
8. Reznikov, V.A., Martin, V.V., and Volodarskii, L.B., *Izv. Akad. Nauk SSSR, Ser. Khim*., 1990, p. 1398.
9. Reznikov, V.A., Vishnevetskaya, L.V., and Volodarskii, L.B., *Izv. Ross. Akad. Nauk, Ser. Khim*., 1993, p. 1612.
10. Reznikov, V.A. and Volodarskii, L.B., *Khim. Geterotsikl. Soedin*., 1990, p. 921.
11. Cline, G.W. and Hanna, S.B., *J. Org. Chem*., 1988, vol. 53, p. 3583.
12. Sevast'yanova, T.K. and Volodarskii, L.B., *Izv. Akad. Nauk SSSR, Ser. Khim*., 1972, p. 1687.
13. Duling, D.R., *J. Magn. Reson., Ser. B*, 1994, vol. 104, p. 105.