ISSN 1070-3632, Russian Journal of General Chemistry, 2010, Vol. 80, No. 3, pp. 423–427. © Pleiades Publishing, Ltd., 2010.
Original Russian Text © E.E. Nifant'ev, M.S. Krymchak, T.S. Kukhareva, L.K. Vasyanina, M.V. Turbina, 2010, published in Zhurnal Obshchei Khimii, 2010, Vol. 80, No. 3, pp. 398–402.

Dihydroquercetin in the Reactions with Phosphorous Acid Hexaethyltriamide

E. E. Nifant'ev, M. S. Krymchak, T. S. Kukhareva, L. K. Vasyanina, and M. V. Turbina

Moscow State Pedagogical University, Nesvizhskii per. 3, Moscow, 119021 Russia
e-mail: chemdept@mtu-net.ru

Received July 8, 2009

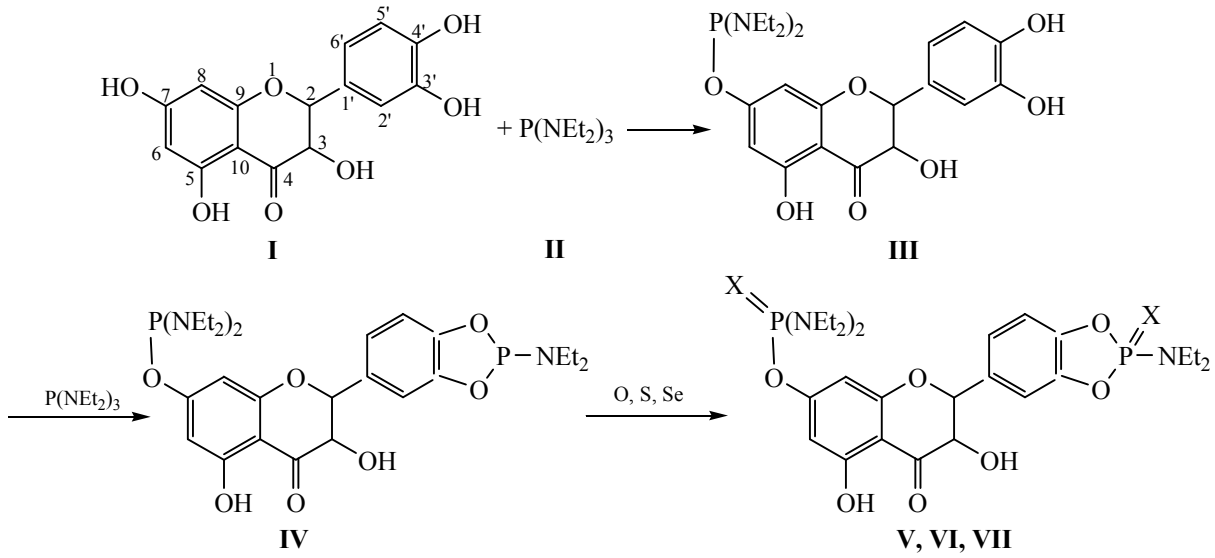
Abstract—The possibility was demonstrated and conditions were developed of a directional phosphorylation of dihydroquercetin flavonoid with phosphorous hexaethyltriamide. The chemical properties of the obtained compounds of trivalent phosphorus are currently under investigation.

DOI: 10.1134/S1070363210030084

Formerly the study was initiated in our laboratory of a reaction of dihydroquercetin flavonoid **I** with reagents of trivalent phosphorus [1]. Diamidophosphites were used as phosphorylating agents, which provide a soft introduction of amidophosphite group into the structure of the substrate [2]. These amidophosphites were brought in an oxidation processes that unabled us to obtain effective antitumor drugs.

The aim of this work was to develop further this investigation, in particular, to study phosphorylation of flavonoid **I** with phosphous hexaethyltriamide, i.e., with the complete amide **II**, which had synthetic advantages over previously studied amidophosphites.

We found that flavonoid **I** readily reacted at 20°C with equimolar amount of phosphorous amide **II** to afford diamidophosphite **III** at the hydroxy group in the position 7. A singlet in ^{31}P NMR spectrum (δ_P 132.3 ppm) of the reaction mixture showed that diamidophosphite **III** was the main product of the reaction. Immediately after completion of the initial phosphorylation a second equivalent of phosphorous amide **II** was added to the reactor. This resulted in the

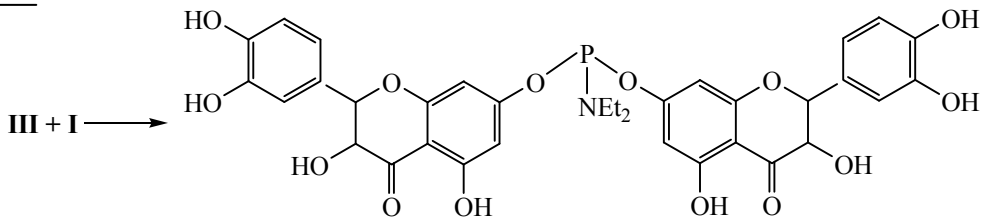


X = O (**V**), S (**VI**), Se (**VII**).

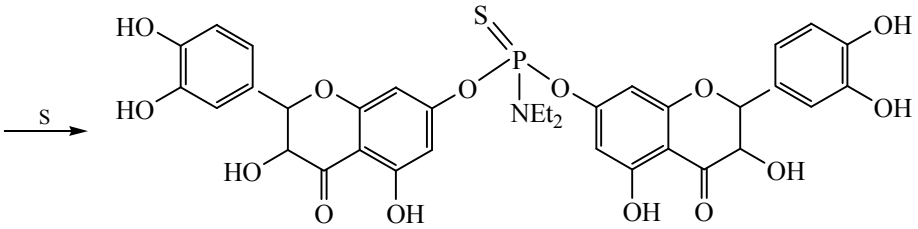
appearance of a new singlet in ^{31}P NMR spectrum (δ_P 152.1 ppm) corresponding to the pyrocatecholamidophosphite site. Thus, we performed the second act of phosphorylation of the flavonoid and obtained diphosphite **IV**.

The reaction mixture containing amidophosphite product **IV** was involved into reactions of oxidation, sulfurization and seleniztion. Therewith the respective phosphate **V**, thiophosphate **VI**, and selenophosphate **VII** were obtained.

Unfortunately, the yields of these first obtained phosphoflavonoid systems are modest (10–20%) at the present stage of work. The reason of this are side processes. For example, at the primary phosphorylation of flavonoids **I** into respective amidophosphite **III** a parallel synthesis of diorganyl phosphite **VIII** takes place:



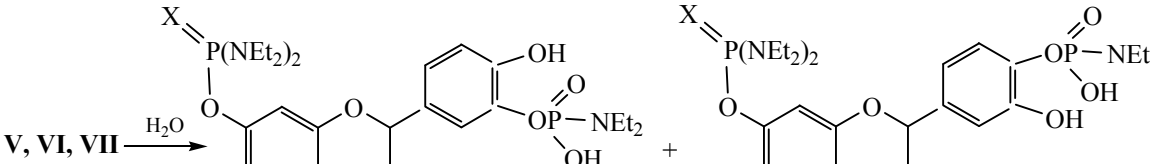
VIII



IX

This side reaction is confirmed by the accumulation in the reaction system after the primary phosphorylation of a compound giving a singlet signal in ^{31}P NMR spectrum, δ_P 149 ppm, corresponding to structure **VIII** which is identified after sulfurization by mass spectrometry (MALDI–TOF). However, the attempted isolation of this compound in pure form failed.

The final part of the work was devoted to opening of rings of cyclophosphorylated pyrocatechols **V–VII**. For this purpose we carried out hydrolysis of these compounds:



X = O (**Va, Vb**), S (**VIa, VIb**), Se (**VIIa, VIIb**).

Note that the opening of the ring leads to the formation of two isomers, whose spectral characteristics are similar to each other. We were unable to separate the isomeric products.

The result of this work is the development of the routes to the synthesis of original complex derivatives of the dihydroquercetin flavonoid by the methods of modern chemistry of organophosphorus compounds.

EXPERIMENTAL

^1H NMR spectra were obtained on a spectrometer JEOL JNM-ECX 400 (400 MHz). ^{31}P NMR spectra

were obtained on a spectrometer JNM-ECX 400 (161.83 MHz). Chemical shifts are given relative to 85% phosphoric acid.

Mass spectral studies were performed on a Bruker Ultra Flex device with time-of-flight detector (TOF) by the method of matrix-activated laser desorption and ionization (MALDI) (λ 337 nm) using as a matrix trihydroxyanthracene.

For elemental analysis a Perkin-Elmer Analyzer 2400 was used.

For the syntheses a microwave reactor CEM "Discover" was used. All syntheses involving compounds of trivalent phosphorus were carried out in dry nitrogen atmosphere in anhydrous solvents dried by standard methods.

TLC analysis was carried out on the Silica gel 60 F 254 plates using the system benzene–dioxane, 3:1 (A), acetonitrile–ethanol, 3:1 (B), and chloroform–ethanol, 1:5 (C). The development was carried out by iodine vapor and by calciation.

Melting points were determined in sealed capillaries while heating at a rate of 1 deg min^{-1}.

Phosphorous hexaethyltriamide **II** was prepared by the method of [3], the constants coincided with the literature data.

2,3-Dihydroquercetin 7-tetraethyldiamidophosphite (III). To a solution of 0.304 g of dihydroquercetin in 100 ml of dioxane at room temperature under stirring was added 0.247 g of phosphorous hexaethyltriamide, and the reaction mixture was maintained for 1 h at room temperature. The product was not isolated from the reaction mixture. The ^{31}P NMR spectrum (dioxane), δ, ppm: 132.3 s. R_f 0.55 (A).

2,3-Dihydroquercetin 7-tetraethyldiamidophosphite-3',4'-diethylamidocyclophosphite (IV). To a solution of 0.478 g of compound **III** in 100 ml of dioxane at room temperature under stirring was added 0.247 g of phosphorous hexaethyltriamide, and the reaction mixture was left for 3 h at room temperature. The product of the reaction was not isolated. The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 132.3 s and 152.1 s. R_f 0.85 (A).

2,3-Dihydroquercetin 7-tetraethyldiamidophosphate-3',4'-diethylamidocyclophosphate (V). To 0.579 g of compound **IV** without isolation from the reaction mixture was added 0.188 g of the complex of urea and hydrogen peroxide, and the mixture was stirred at room temperature for 2 h. The reaction mixture was filtered, the solvent was removed in a vacuum, the dry residue was dissolved in benzene. The product of reaction **V** was purified by chromatography on a column with silica gel, eluent 50 ml of hexane–dioxane, 3:1. The compound obtained was dried for 4 h (25°C, 1 mm Hg). The resulting compound is a yellow powder. Yield of compound **V** 0.085 g (14%), mp 51–53°C, R_f 0.6 (B). ^1H NMR spectrum (DMSO-d_6), δ, ppm: 1.11 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.1 Hz), 3.11 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.1 Hz, $^3J_{HP}$ 11.5 Hz), 4.62 d (1H, C^3H, $^3J_{HH}$ 11.7 Hz), 4.92 d (1H, C^2H, $^3J_{HH}$ 11.7 Hz), 6.1 br.s (1H, C^3OH), 6.39 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.81 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.57 s (2H, $C^{5'}H$ and $C^{6'}H$), 7.81 s (1H, $C^{2'}H$), 13.4 in (1H, $C^{5'}OH$). The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 21.9 s and 15.6 s. Found, %: C 52.8, H 6.35; N 6.91; P 10.41. $C_{27}H_{39}N_3O_9P_2$. Calculated, %: C 53.03, H 6.43; N 6.87; P 10.13.

2,3-Dihydroquercetin 7-tetraethyldiamidothiophosphate-3',4'-diethylamidocyclothiophosphate (VI). To 0.579 g of compound **IV** without isolation from the reaction mixture was added 0.064 g of sulfur, and the mixture was stirred at 50°C for 2 h. The solvent was removed in a vacuum. The dry residue was dissolved in benzene. The product of reaction **VI** was purified by chromatography on a column packed with silica gel, elution with 50 ml of hexane–dioxane, 3:1. The substance obtained was dried for 4 h (25°C, 1 mm Hg). The resulting compound is a yellow powder. Yield 0.064 g (10%). mp 68–70°C. R_f 0.8 (A). ^1H NMR spectrum (DMSO-d_6), δ, ppm: 1.07 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz), 3.16 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz, $^3J_{HP}$ 11.4 Hz), 4.76 d (1H, C^3H, $^3J_{HH}$ 11.4 Hz), 5.25 d (1H, C^2H, $^3J_{HH}$ 11.4 Hz), 6.19 br.s (1H, C^3OH), 6.25 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.50 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.97 s (2H, $C^{5'}H$ and $C^{6'}H$), 8.05 s (1H, $C^{2'}H$), 12.3 in (1H, C^5OH). The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 76.2 s and 88.5 s. Found, %: C 50.41, H 6.09; N 6.48; P 9.67. $C_{27}H_{39}N_3O_7P_2S_2$. Calculated, %: C 50.38, H 6.11; N 6.52; P 9.62.

2,3-Dihydroquercetin 7-tetraethyldiamidoselenophosphate-3',4'-diethylamidocycloselenophosphate (VII). To 0.579 g of compound **IV** without isolation from the reaction mixture was added 0.158 g of selenium, and the mixture was stirred at 50°C for 2 h. The solvent was removed in a vacuum. The dry residue was dissolved in benzene. The product of reaction **VII** was purified by chromatography on a column packed with silica gel, elution with 50 ml of hexane–dioxane,

3:1. Substance was dried for 4 h (25°C, 1 mm Hg). The resulting compound is a yellow powder. Yield 0.126 g (17%). mp 70–72 ° C. R_f 0.75 (A). ^{1}H NMR spectrum (DMSO-d_6), δ, ppm: 1.16 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz), 3.31 m (12H, CH_3 CH_2 N, $^3J_{HH}$ 7.3 Hz, $^3J_{HP}$ 11.3 Hz), 4.5 d (1H, C^3H, $^3J_{HH}$ 11.9 Hz), 5.07 d (1H, C^2H, $^3J_{HH}$ 11.9 Hz), 5.9 br.s (1H, C^3OH), 6.24 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.49 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.53 s (2H, $C^{5'}H$ and $C^{6'}H$), 8.06 s (1H, $C^{2'}H$), 11.6 in (1H, C^5OH). The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 77.4 and from 91.7 to. Found, %: C 43.75, H 5.20; N 5.68; P 8.52. $C_{27}H_{39}N_3O_7P_2Se_2$. Calculated, %: C 43.97, H 5.33; N 5.70; P 8.40.

2,3-Dihydroquercetin 7-tetraethyldiamidophosphate-3'-diethylamidophosphate (Va) and 2,3-dihydroquercetin 7-tetraethyldiamidophosphate-4'-diethylamidophosphate (Vb). To 0.611 g of compound **V** was added while stirring at room temperature 1.8 ml of water. The reaction mixture was left for 4 h at room temperature. The reaction product precipitated spontaneously from the reaction mixture. The substance was filtered off, washed with hexane, and dried for 3 h in a vacuum (25°C, 1 mm Hg). The resulting compound is a yellow powder. Yield 0.232 g (38%). R_f 0.75 (B). The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 14.2 s and 12.7 s. Found, %: C 51.03, H 6.48; N 6.34; P 9.72. $C_{27}H_{41}N_3O_{10}P_2$. Calculated, %: C 51.51, H 6.56; N 6.67; P 9.84.

Compound Va. ^{1}H NMR spectrum (DMSO-d_6), δ, ppm: 1.14 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.1 Hz), 3.15 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.1 Hz, $^3J_{HP}$ 11.5 Hz), 4.73 d (1H, C^3H, $^3J_{HH}$ 11.7 Hz), 5.16 d (1H, C^2H, $^3J_{HH}$ 11.7 Hz), 6.09 br.s (1H, C^3OH), 6.44 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.84 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.42 s (2H, $C^{5'}H$ and $C^{6'}H$), 7.77 s (1H, $C^{2'}H$), 9.20 s (1H, $C^{4'}OH$), 12.90 s (1H, C^5OH).

Compound Vb. ^{1}H NMR spectrum (DMSO-d_6), δ, ppm: 1.10 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.1 Hz), 3.10 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.1 Hz, $^3J_{HP}$ 11.5 Hz), 4.71 d (1H, C^3H, $^3J_{HH}$ 11.7 Hz), 5.10 d (1H, C^2H, $^3J_{HH}$ 11.7 Hz), 6.09 br.s (1H, C^3OH), 6.40 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.82 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.40 s (2H, $C^{5'}H$ and $C^{6'}H$), 7.75 s (1H, $C^{2'}$ H), 8.96 s (1H, $C^{3'}OH$), 12.90 s (1H, C^5OH).

2,3-Dihydroquercetin 7-tetraethyldiamidothiophosphate-3'-diethylamidophosphate (VIa) and 2,3-dihydroquercetin 7-tetraethyldiamidothiophosphate-4'-diethylamidophosphate (VIb). To 0.643 g of compound **VI** was added with stirring at room temperature 1.8 ml of water. The reaction mixture was maintained for 3 h at 40°C in a microwave reactor CEM "Discover." The reaction product spontaneously precipitated from the reaction mixture. The substance formed was filtered off, washed with hexane, and dried for 3 h in a vacuum (25°C, 1 mm Hg). The resulting compound is a yellow powder. Yield 0.286 g (40%). R_f 0.7 (B). The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 75.06 s and 0.42 s. Found, %: C 49.8, H 6.79; N 6.44; P 9.18. $C_{27}H_{41}N_3O_9P_2S_2$. Calculated, %: C 50.23, H 6.40; N 6.51; P 9.59.

Compound VIa. ^{1}H NMR spectrum (DMSO-d_6), δ, ppm: 1.07 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz), 3.09 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz, $^3J_{HP}$ 11.4 Hz), 4.77 d (1H, C^3H, $^3J_{HH}$ 11.4 Hz), 4.95 d (1H, C^2H, $^3J_{HH}$ 11.4 Hz), 5.7 br.s (1H, C^3OH), 6.33 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.58 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.64 s (2H, $C^{5'}H$ and $C^{6'}H$), 8.08 s (1H, $C^{2'}H$), 8.98 s (1H, $C^{4'}OH$), 12.3 s (1H, C^5OH).

Compound VIb. ^{1}H NMR spectrum (DMSO-d_6), δ, ppm: 1.03 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz), 2.9 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz, $^3J_{HP}$ 11.4 Hz), 4.76 d (1H, C^3H, $^3J_{HH}$ 11.4 Hz), 4.93 d (1H, C^2H, $^3J_{HH}$ 11.4 Hz), 5.7 br.s (1H, C^3OH), 6.30 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.55 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.60 s (2H, $C^{5'}H$ and $C^{6'}H$), 8.05 s (1H, $C^{2'}$ H), 8.89 s (1H, $C^{3'}OH$), 12.3 s (1H, C^5OH).

2,3-Dihydroquercetin 7-tetraethyldiamidoselenophosphate-3'-diethylamidophosphate (VIIa) and 2,3-dihydroquercetin 7-tetraethyldiamidoselenophosphate-4'-diethylamidophosphate (VIIb). To 0.737 g of compound **VII** was added with stirring at room temperature 1.8 ml of water. The reaction mixture was maintained for 3 h at 40°C in a microwave reactor CEM "Discover." The reaction product spontaneously precipitated from the reaction mixture. The substance was filtered off, washed with hexane, and dried for 3 h in a vacuum (25°C, 1 mm Hg). The resulting compound is a yellow powder. Yield 0.34g (46%). R_f 0.65 (B). The ^{31}P NMR spectrum (dioxane), δ_P, ppm: 76.9 s and 2.14 s. Found, %: C 46.95, H 5.63; N 6.55; P 8.21. $C_{27}H_{41}N_3O_9P_2Se_2$. Calculated, %: C 46.83, H 5.97; N 6.07; P 8.95.

Compound VIIa. ^{1}H NMR spectrum (DMSO-d_6), δ, ppm: 1.19 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz), 3.24 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz, $^3J_{HP}$ 11.3 Hz), 4.52 d (1H, C^3H, $^3J_{HH}$ 11.9 Hz), 5.16 d (1H, C^2H, $^3J_{HH}$ 11.9 Hz), 6.07 br.s (1H, C^3OH), 6.46 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.79 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.25 s (2H,

$C^{5'}H$ and $C^{6'}H$), 7.70 s (1H, $C^{2'}H$), 9.30 s (1H, $C^{4'}OH$), 11.60 s (1H, $C^{5}OH$).

Compound VIIb. ^1H NMR spectrum (DMSO-d_6), δ, ppm: 1.15 t (18H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz), 3.17 m (12H, CH_3CH_2N, $^3J_{HH}$ 7.3 Hz, $^3J_{HP}$ 11.3 Hz), 4.48 d (1H, C^3H, $^3J_{HH}$ 11.9 Hz), 5.10 d (1H, C^2H, $^3J_{HH}$ 11.9 Hz), 6.07 br.s (1H, C^3OH), 6.44 s (1H, C^8H, $^4J_{HH}$ 1.8 Hz), 6.74 s (1H, C^6H, $^4J_{HH}$ 1.8 Hz), 7.25 s (2H, $C^{5'}H$ and $C^{6'}H$), 7.69 s (1H, $C^{2'}H$), 9.10 s (1H, $C^{3'}OH$), 11.60 with a (1H, C^5OH).

ACKNOWLEDGMENTS

This work was carried out with the financial support of the Program of the President of Russian Federation for support of the leading scientific schools of Russia (grant no. NSh-582.2008.3).

REFERENCES

1. Nifant'ev, E.E., Kukhareva, T.S., Koroteev, M.P., Dzgoeva, Z.M, Kaziev, G.Z., and Vasyanina, L.K., *Bioorganic Chemistry*, 2001, vol. 27, no. 4, p. 314 .
2. Nifant'ev, E.E., Kukhareva, T.S., Dzgoeva, Z.M., Vasyanina, L.K., Koroteev, M.P., and Kaziev, G.Z., *Heteroatom Chem.,* 2003, vol. 14, no. 5, p. 399.
3. Nifant'ev, E.E. and Zavalishina, A.I., *Khimiya elementoorganicheskikh soedinenii* (Chemistry Organo-element Compounds), Moscow: Lenin Mosk. Gos. Ped. Inst., 1980.