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Synthesis of Novel Coumarin Derivatives Bearing Phosphor Ester Motifs and Evaluation of their Antioxidant Activities

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Abstract

phosphoramidate, phosphoranylidenetriazene, and azaphosphole derivatives can be respectively formed when the substituted azidocoumarin **1** reacted with trialkylphosphites (TAP), dialkylphosphonates (DAP), and tris(dialkylamino)phosphines. The known triazinedione was revealed to be a side product in the reactions of carbaldehyde **1** with TAP and tris(dialkylamino)-phosphines. In the same context, when the starting coumarin **16** was allowed to react with P (*III*) reagents, chromen phosphoramidates and chromen phosphoric triamide derivatives were obtained via Staudinger reaction. Contrary to that pathway, phosphoranylidenetriazen chromen derivative could be isolated and identified when coumarin **16** was allowed to react with tris(diethylamino)-phosphine. The antioxidant activities of 6-newly products were evaluated. The results showed that two from the six screened compounds exhibited good antioxidant activities.

***Keywords*:** Azidocoumarin; phosphoramidates; Azaphospholes; Chromen phosphoric triamide; Antioxidant activity

1. Introduction

Coumarin derivatives have great utility, since they present a wide range of biological properties [1-4] as antitumor [5], antimicrobial [6-9], anti-inflammatory [10], anticoagulant [11], and antioxidant agents [12, 13], in addition to their coronary vasodilating activity [14]. The coumarins have also used in treating infections of retroviral against HIV- protease [15]. Coumarin compounds (2H-1-benzopyran-2-one) contain a large category of phenolic moieties that exist in plants and consist of fused benzene and pyrone cycles [16]. Coumarins are also used as ingredients in cosmetics, perfumes, additives in food, pharmaceuticals and optical brighteners [17].

The attention in coumarin derivative chemistry and the biological activities of these compounds has increased through the previous five decades. In follow up to our lab research plan that pointed to the utility of phosphorus reagents in pharmaceuticals synthesis [18-29], we expand the previous work on phosphor reagents and coumarins [30, 31] to produce unprecedented coumarin derivatives of prospective biological activities, particularly as antioxidants. The methodology depends on the reaction of azidochromene carbaldehyde **1** and 4-azidocoumarin (**16**) with trialkyl phosphites **2a**–**c**, dialkyl phosphites **7a**–**c**, and hexaalkylphosphorus triamides **11a**,**b**.

2. Experimental

Electrothermal (variable heater) melting point apparatus was used to determine the Melting points via open capillary tube, Melting points were corrected. JEOL E.C.A-500 MHz spectrometer (JEOL, Japan, ^{1}H: 500.7 MHz, ^{13}C: 125.4 MHz, ^{31}P: 200.7 MHz) was used to measure NMR spectra. H_3PO_4 (85%) was used as external reference on recording the ^{31}P NMR spectra. TMSi was used as internal standard in DMSO-d_6 on recording ^{1}H and ^{13}C NMR spectra, while chemical shifts (δ) are given

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in ppm. IR spectra were recorded on a JASCO FT-IR 6100 using KBr disc (JASCO, Japan). MS-50 Kratos (A.E.I.) spectrometer which provided with a data system spectrometer (Kratos, UK) was used at recording the mass spectra (70 eV). Using the elementary Analysen-systeme GmbH-vario EL III Element Analyzer, Germany, Elemental analysis was executed at the Microanalysis Laboratory, Cairo University, Giza, Egypt. On the other hand, the purity of all new compounds was established by microchemical analysis (C/H/N) and the measured values were to be in good harmonization (± 0.2%) with the calculated values. Thin layer chromatography (TLC): (Merck), precoated silica gel 60 F245 aluminum plates. Solvents were dried by standard techniques. The proper precautions in dealing with the moisture-sensitive samples were followed.

2.1. Synthesis

Synthesis of phosphoramidates 5a–c

The 4-azido-2-oxo-2*H*-chromene-3-carbaldehyde **(1)** (3.7 mmol, 0.8 g) [32] and trialkyl phosphites **2a-c** (4 mmol) were stirred in 15 mL THF at r.t. for ~6–8 h (TLC). Under vacuum, excess of the volatile materials was extracted. The resulting residue was chromatographed on silica gel with n-hexane/$CHCl_3$ (7:3, v/v) to give 3*H*-chromeno[4,3-*d*][1,2,3]triazine-4,5-dione (**6**) as pale yellow crystals in ~11 % yield; m.p. 96 ºC (Ref. [30] 94-96 ºC). Elution with *n*-hexane/$CHCl_3$ (1:1, v/v) afforded **5a**–**c**.

Dimethyl methyl(3-formyl-2-oxo-2H-chromen-4-yl)phosphoramidate (5a)

Yield: 0.87 g (75.5%). Product **5a** was separated as yellow substance. m.p.: 209 ºC (MeCN); IR (KBr): ύ$_{max}$ = 1720 (br, 2C=O, lactone, aldehyde), 1320, 860 (PNC), 1245 (P=O), 1032 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 3.16 (d, $^3J_{PH}$ 8.1 Hz, 3H, N-*Me*), 3.79 (d, $^3J_{PH}$ 7.6 Hz, 6H, P(O*Me*)$_2$), 7.36-7.58 (m, 4H, *H*/Ar), 9.51(s, 1H, *H*/aldehyde) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 189.1 (*C*/aldehyde), 154.8, 151.6, 140.2, 131.1, 129.4, 127.2, 117.9, 114.9, 98.4 (*C*/chromenone), 55.3 (d, $^2J_{PC}$ 12.8 Hz, P(O*Me*)$_2$), 29.9 (d, $^2J_{PC}$ 12.5 Hz, N-*Me*) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 19.2 ppm; MS (70 eV): *m*/*z* (%) = 311 (35) [M$^+$]; Anal. Calcd. for $C_{13}H_{14}NO_6P$ (311.23) C, 50.17; H, 4.53; N, 4.50. Found: C, 50.31; H, 4.38; N, 4.36.

Diethyl ethyl(3-formyl-2-oxo-2H-chromen-4-yl)phosphoramidate (5b)

Yield: 0.97 g (73.9%). Product **5b** was separated as Straw yellow substance. m.p.: 179 ºC ($CHCl_3$); IR (KBr): ύ$_{max}$ = 1715 (br, 2C=O, lactone, aldehyde), 1329, 865 (P-NC), 1252 (P=O), 1050 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.16 (dt, $^3J_{HH}$ 5.3, $^4J_{PH}$ 4.2 Hz, 3H, NC*Me*), 1.30 (dt, $^3J_{HH}$ 6.8, $^4J_{PH}$ 4.8 Hz, 6H, P(OCH$_2$*Me*)$_2$), 3.14 (dq, $^3J_{HH}$ 5.3, $^3J_{PH}$ 8.4 Hz, 2H, NC*H*$_2$CH$_3$), 4.28 (dq, $^3J_{HH}$ 6.8, $^3J_{PH}$ 8.8 Hz, 4H, P(OC*H*$_2$Me)$_2$), 7.01- 7.64 (m, 4H, *H*/Ar), 9.56(s, 1H, *H*/aldehyde) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 188.8 (*C*/aldehyde), 155.2, 153.1, 138.8, 131.0, 129.1, 126.7, 118.1, 114.2, 98.9 (*C*/chromenone), 61.9 (d, $^2J_{PC}$ 13.8 Hz, O*C*H$_2$Me), 43.1 (d, $^2J_{PC}$ 13.7 Hz, N*C*H$_2$Me), 19.9 (d, $^3J_{PC}$ 8.4 Hz, POCH$_2$*Me*), 14.7 (d, $^3J_{PC}$ 7.9 Hz, NCH$_2$*Me*) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 20.9 ppm; MS (70 eV): *m*/*z* (%) = 353 (33) [M$^+$]; Anal. Calcd. for $C_{16}H_{20}NO_6P$ (353.31) C, 54.39; H, 5.71; N, 3.96. Found: C, 54.55; H, 5.56; N, 3.78.

Diisopropyl isopropyl(3-formyl-2-oxo-2H-chromen-4-yl)amidophosphate (5c)

Yield: 1.1 g (76.3%). Product **5c** was separated as Pale yellow solid. m.p.: 193 ºC (EtOH); IR (KBr): ύ$_{max}$ = 1723 (br, 2C=O, lactone, aldehyde), 1325, 862 (P-NC), 1250 (P=O), 1045 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.27 (dd, $^3J_{HH}$ 4.6, $^4J_{PH}$ 3.8 Hz, 6H, NCH*Me*$_2$), 1.38 (dd, $^3J_{HH}$ 5.6, $^4J_{PH}$ 4.1 Hz, 12H, P(OCH*Me*$_2$)$_2$), 3.89 (m, 1H, NC*H*), 4.57 (m, 2H, P(OC*H*Me$_2$)$_2$), 7.00-7.66 (m, 4H, *H*/Ar), 9.47 (s, 1H, *H*/aldehyde) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 189.0 (*C*/aldehyde), 153.7, 152.1, 138.8, 131.5, 128.9, 125.7, 118.3, 113.2, 97.9 (*C*/chromenone), 73.6 (d, $^2J_{PC}$ 13.9 Hz, O*C*H Me$_2$), 50.9 (d, $^2J_{PC}$ 13.9 Hz, N*C*H Me$_2$), 23.9 (d, $^3J_{PC}$ 7.9 Hz, POCH*Me*$_2$), 21.1 (d, $^3J_{PC}$ 8.5 Hz, NCH*Me*$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 22.5 ppm; MS (70 eV): *m*/*z* (%) = 395 (27) [M$^+$]; Anal. Calcd. for $C_{19}H_{26}NO_6P$ (395.39) C, 57.72; H, 6.63; N, 3.54. Found: C, 57.90; H, 6.46; N, 3.38.

Synthesis of amidophosphates 10a–c

The azide **1** (0.8 g, 3.7 mmol) and dialkyl phosphite **7a-c** (4 mmol) in 15 mL THF were refluxed for 21–24 h (TLC). Under vacuum, the solvent was evaporated to dryness and the crude material was washed with cyclohexane. The product substances were crystallized from the suitable solvent to provide the corresponding amidophosphates **10a**–**c**.

Dimethyl (3-formyl-2-oxo-2H-chromen-4-yl)amidophosphate (10a)

Yield: 0.89 g (81%). Product **10a** was separated as yellow substance. m.p.: 169 ºC ($CHCl_3$); IR (KBr):

υ_{max} = 3335 (NH), 1705 (br, 2C=O, lactone, aldehyde), 1238 (P=O, bonded), 1134 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 3.82 (d, $^3J_{PH}$ 8.5 Hz, 6H, P(O*Me*)$_2$), 7.25- 7.41 (m, 4H, *H*/Ar), 9.67 (s, 1H, *H*/aldehyde), 10.45 (br, 1H, N*H*) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 188.1 (*C*/aldehyde), 155.8, 154.9, 139.7, 133.6, 129.8, 126.9, 120.8, 114.5, 95.8 (*C*/chromenone), 53.1 (d, $^2J_{PC}$ 13.6 Hz, P(O*Me*)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 18.5 ppm; MS (70 eV): *m*/*z* (%) = 297 (35) [M^+], 296 (40) [M^+ -1]; Anal. Calcd. for $C_{12}H_{12}NO_6P$ (297.20) C, 48.50; H, 4.07; N, 4.71. Found: C, 48.68; H, 3.90; N, 4.53.

Diethyl (3-formyl-2-oxo-2H-chromen-4-yl)amidophosphate (10b)

Yield: 0.96 g (79%). Product **10b** was separated as yellow substance. m.p.: 139 ºC (cyclohexane); IR (KBr): υ_{max} = 3340 (NH), 1715 (br, 2C=O, lactone, aldehyde), 1230 (P=O, bonded), 1135 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.02 (dt, $^3J_{HH}$ 6.8, $^4J_{PH}$ 3.6 Hz, 6H, P(OCH$_2$*Me*)$_2$), 4.09 (dq, $^3J_{HH}$ 6.8, $^3J_{PH}$ 5.2 Hz, 4H, P(OC*H*$_2$)$_2$), 7.24- 7.75 (m, 4H, *H*/Ar), 9.71 (s, 1H, *H*/aldehyde), 10.25 (br, 1H, N*H*) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 185.9 (*C*/aldehyde), 156.1, 154.9, 137.9, 132.5, 129.2, 126.5, 119.4, 112.5, 96.9 (*C*/chromenone), 62.6 (d, $^2J_{PC}$ 12.6 Hz, P(O*C*H$_2$)$_2$), 16.3 (d, $^3J_{PC}$ 9.6 Hz, P(OCH$_2$*Me*)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 20.1 ppm; MS (70 eV): *m*/*z* (%) = 325 (28) [M^+], 324 (32) [M^+-1]; Anal. Calcd. for $C_{14}H_{16}NO_6P$ (325.25) C, 51.70; H, 4.96; N, 4.31. Found: C, 51.88; H, 4.77; N, 4.15.

Diisopropyl (3-formyl-2-oxo-2H-chromen-4-yl)amidophosphate (10c)

Yield: 1.06 g (81%). Product **10c** was separated as pale yellow substance. m.p.: 155 ºC ($CHCl_3$); IR (KBr): υ_{max} = 3337 (NH), 1710 (br, 2C=O, lactone, aldehyde), 1240 (P=O, bonded), 1138 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.44 (dd, $^3J_{HH}$ 6.5, $^4J_{PH}$ 4.2 Hz, 12H, P(OCH*Me*$_2$)$_2$), 4.79 (m, 2H, P(OC*H*)$_2$), 7.44- 7.82 (m, 4H, *H*/Ar), 9.68 (s, 1H, *H*/aldehyde), 10.30 (br, 1H, N*H*) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ= 187.9 (*C*/aldehyde), 155.9, 1542, 139.2, 133.2, 129.0, 126.9, 119.2, 114.7, 95.6 (*C*/chromenone), 72.4 (d, $^2J_{PC}$ 12.8 Hz, P(O*C*H)$_2$), 24.1 (d, $^3J_{PC}$ 7.8 Hz, P(OCH*Me*$_2$)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 19.2 ppm; MS (70 eV): *m*/*z* (%) = 353 (36) [M^+], 352 (44) [M^+ - 1]; Anal. Calcd. for $C_{16}H_{20}NO_6P$ (353.31) C, 54.39; H, 5.71; N, 3.96. Found: C, 54.55; H, 5.58; N, 3.79.

Synthesis of triazenylidene-phosphoranes 12a,b

Hexalkylphosphorus triamide (**11a**,**b**, 4 mmol) in THF (5 mL) was added dropwise to azide **1** (3.7 mmol, 0.8 g) in THF (10 mL). At room temperature, the reaction mixture was stirred (~15 h, TLC). Under vacuum, the solvent was evaporated to dryness. The resulting residue was chromatographed on silica gel and eluted with *n*-hexane/$CHCl_3$ (7:3, v/v) to give chromenotriazine **6** in ~9 % yield. **12a**,**b** were obtained by using *n*-hexane/$CHCl_3$ (1:1, v/v) as eluent.

2-Oxo-4-{(1Z)-3-[tris(dimethylamino)phosphoranylidene]triaz-1-en-1-yl}-2H-chromene-3-carbaldehyde (12a)

Yield: 1.13 g (80.6%). Product **12a** was separated as yellow material. m.p.: 173 ºC (EtOH); IR (KBr): υ_{max} = 1722, 1662 (2C=O, aldehyde, lactone), 1355 (P=N), 1338,860 (P(NMe$_2$)$_3$) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 2.49, 2.81 (2d, $^3J_{PH}$ 8.9 Hz, 18H, P(N*Me*$_2$)$_3$), 7.12- 7.65 (m, 4H, *H*/Ar), 10.21 (s (br), 1H, *H*C=O) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 184.3 (*C*=O, aldehyde), 155.8, 154.6, 134.8, 128.1, 127.2, 119.1, 110.6, 107.6 (*C*/chromenone),153.8 (*C*=O, lactone), 38.2 (d, $^2J_{PC}$ 14.1 Hz, P(N*Me*$_2$)$_3$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 39.2 ppm; MS (70 eV): *m*/*z* (%) = 378 (28) [M^+], 350 (42) [M^+ - N_2]; Anal. Calcd. for $C_{16}H_{23}N_6O_3P$ (378.37) C, 50.79; H, 6.13; N, 22.21. Found: C, 50.97; H, 5.98; N, 22.05.

2-Oxo-4-{(1Z)-3-[tris(diethylamino)phosphoranylidene]triaz-1-en-1-yl}-2H-chromene-3-carbaldehyde (12b)

Yield: 1.39 g (80.8%). Product **12b** was separated as yellow substance. m.p.: 159 ºC ($CHCl_3$); IR (KBr): υ_{max} = 1725, 1660 (2C=O, aldehyde, lactone), 1352 (P=N), 1325, 855 (P(NEt$_2$)$_3$) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 0.95, 1.39 (2dt, $^3J_{HH}$ 8.5, $^4J_{PH}$ 4.8 Hz, 18H, P(N(C*Me*)$_2$)$_3$), 2.91, 3.18 (2dq, $^3J_{HH}$ 8.5, $^3J_{PH}$ 9.2 Hz, 12H, P(N(C*H*$_2$Me)$_2$)$_3$), 7.10- 7.57 (m, 4H, *H*/Ar), 10.19 (s (br), 1H, *H*C=O) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 186.4 (*C*=O, aldehyde), 155.8, 154.1, 134.5, 128.7, 127.3, 118.4, 109.7, 106.0 (*C*/chromenone),153.7 (*C*=O, lactone), 39.6 (d, $^2J_{PC}$ 13.9 Hz, P(N(*C*H$_2$Me)$_2$)$_3$), 14.6 (d, $^3J_{PC}$ 8.4 Hz, P(N(C*Me*)$_2$)$_3$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 39.9 ppm; MS (70 eV): *m*/*z* (%) = 462 (23) [M^+], 434 (40) [M^+ - N_2]; Anal. Calcd. for $C_{22}H_{35}N_6O_3P$ (462.53) C, 57.13; H, 7.63; N, 18.17. Found: C, 57.31; H, 7.45; N, 17.97.

Synthesis of compounds 15a,b

Aminophosphines **11a**,**b** (4 mmol) in THF (10 mL) was added in one portion to a solution of the azide **1** (3.7 mmol, 0.8 g) and H_2O (2 mL) in THF (10 mL). The reaction mixture was refluxed for ~5 h (TLC), and then evaporation of solvent to dryness was taken place. Further, Chromatography process for the crude product was occurred on silica gel. chromenotriazine **6** (~6 % yield) was separated when *n*-hexane/$CHCl_3$

(7:3, v/v) was used as eluent. While **15a**,**b** were obtained by elution with *n*-hexane/$CHCl_3$ (1:1, v/v).

2-(Dimethylamino)-1,2-dihydrochromeno[3,4-d][1,2]azaphosphole-3,4-dione 2-oxide (15a)

Yield: 0.78 g (75.4%). Product **15a** was separated as Pale yellow substance. m.p.: 129 ºC (cyclohexane); IR (KBr): ύ$_{max}$ = 3410 (NH), 1690, 1633 (2C=O, ketone, lactone), 1224 (P=O, bonded), 1330, 855 (PNMe) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 2.82 (d, $^3J_{PH}$ 10.4 Hz, 6H, PN*Me*$_2$), 7.02- 7.47 (m, 4H, *H*/Ar), 9.48 (s (br), 1H, N*H*, D_2O exch) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 186.2 (d, $^1J_{PC}$ 136.6 Hz, *C*=O, ketone), 158.8 (d, $^3J_{PC}$ 8.8 Hz, *C*=O, lactone), 154.2, 144.1, 130.8, 127.7, 126.1, 118.6, 113.1, 105.8 (*C*/chromenone), 37.1 (d, $^2J_{PC}$ 14.6 Hz, PN*Me*$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 14.8 ppm; MS (70 eV): *m*/*z* (%) = 278 (45) [M$^+$], 277 (49) [M$^+$ - 1]; Anal. Calcd. for $C_{12}H_{11}N_2O_4P$ (278.20) C, 51.81; H, 3.99; N, 10.07. Found: C, 51.97; H, 3.80; N, 9.91.

2-(Diethylamino)-1,2-dihydrochromeno[3,4-d][1,2]azaphosphole-3,4-dione 2-oxide (15b)

Yield: 0.86 g (75.9%). Product **15b** was separated as yellowish grey powder. m.p.: 114 ºC (cyclohexane); IR (KBr): ύ$_{max}$ = 3415 (NH), 1695, 1630 (2C=O, ketone, lactone), 1235 (P=O, bonded),1332, 859 (P-NEt) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 0.98 (dt, $^3J_{HH}$ 7.4, $^4J_{PH}$ 5.6 Hz, 6H, PN(C*Me*)$_2$), 2.89 (dq, $^3J_{HH}$ 7.4, $^3J_{PH}$ 10.1 Hz, 4H, PN(C*H*$_2$Me)$_2$), 7.00- 7.49 (m, 4H, *H*/Ar), 9.51 (s (br), 1H, N*H*, D_2O exch) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 188.3 (d, $^1J_{PC}$ 137.9 Hz, *C*=O, ketone), 159.9 (d, $^3J_{PC}$ 8.6 Hz, *C*=O, lactone), 154.8, 145.7, 132.1, 127.9, 126.4, 118.2, 114.0, 104.7 (*C*/chromenone), 38.1 (d, $^2J_{PC}$ 14.9 Hz, PN(*C*H$_2$)$_2$), 14.1 (d, $^3J_{PC}$ 8.2 Hz, PN(C*Me*)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 15.2 ppm; MS (70 eV): *m*/*z* (%) = 306 (40) [M$^+$], 305 (47) [M$^+$ - 1]; Anal. Calcd. for $C_{14}H_{15}N_2O_4P$ (306.25) C, 54.91; H, 4.94; N, 9.15. Found: C, 55.10; H, 4.76; N, 9.00.

Synthesis of dialkyl alkyl(2-oxo-2H-chromen-4-yl)phosphoramidates 18a–c

A mixture of coumarin derivative **16** [33] (0.8 g, 4.3 mmol) and trialkyl phosphites **2a-c** (4.6 mmol) was stirred in THF (15 mL) at r.t. (~15 h, TLC). Under vacuum, the volatile materials were removed, and the crude material was washed with cyclohexane. The product substances were crystallized from the suitable solvent to provide the corresponding amidophosphates **18a**–**c**.

Dimethyl methyl(2-oxo-2H-chromen-4-yl)phosphoramidate (18a)

Yield: 0.96 g (79.4%). Product **18a** was separated as yellow solid. m.p.: 189 ºC (MeCN); IR (KBr): ύ$_{max}$ = 1725 (C=O, lactone), 1320, 860 (P-NC), 1255 (P=O), 1080 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 2.93 (d, $^3J_{PH}$ 9.8 Hz, 3H, N*Me*), 3.81 (d, $^3J_{PH}$ 10.1 Hz, 6H, P(O*Me*)$_2$), 5.58 (d, $^4J_{PH}$ 3.8 Hz, 1H, *H*C(3)/Ar), 7.08- 7.31 (m, 4H, *H*/Ar) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 161.9 (*C*=O, lactone), 154.1, 146.5, 131.7, 128.8, 126.5, 118.8, 116.7, 89.1 (*C*/chromenone), 53.1 (d, $^2J_{PC}$ 14.5 Hz, P(O*Me*)$_2$), 30.4 (d, $^2J_{PC}$ 14.4 Hz, N*Me*) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 17.6 ppm; MS (70 eV): *m*/*z* (%) = 283 (33) [M$^+$]; Anal. Calcd. for $C_{12}H_{14}NO_5P$ (283.22) C, 50.89; H, 4.98; N, 4.95. Found: C, 51.03; H, 4.79; N, 4.78.

Diethyl ethyl(2-oxo-2H-chromen-4-yl)phosphoramidate (18b)

Yield: 1.1 g (79.0%). Product **18b** was separated as yellowish solid. m.p.: 175 ºC (EtOH); IR (KBr): ύ$_{max}$ = 1720 (C=O, lactone), 1323, 865 (P-NC), 1247 (P=O), 1074 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.21–1.32 (m, 9H, NC*Me* & P(OCH$_2$*Me*)$_2$), 2.92 (dq, $^3J_{HH}$ 6.7, $^3J_{PH}$ 8.3 Hz, 2H, NC*H*$_2$), 4.09 (m, 4H, P(OC*H*$_2$)$_2$), 5.56 (d, $^4J_{PH}$ 3.9 Hz, 1H, *H*C(3)/Ar), 7.13–7.39 (m, 4H, *H*/Ar) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 162.2 (*C*=O, lactone), 153.7, 146.1, 131.5, 129.1, 126.1, 119.1, 117.0, 88.8 (*C*/chromenone), 62.7 (d, $^2J_{PC}$ 12.8 Hz, P(O*C*H$_2$)$_2$), 42.5 (d, $^2J_{PC}$ 11.9 Hz, N*C*H$_2$), 16.3 (d, $^3J_{PC}$ 9.6 Hz, P(OC*Me*)$_2$), 14.1 (d, $^3J_{PC}$ 9.0 Hz, NC*Me*) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 18.2 ppm; MS (70 eV): *m*/*z* (%) = 325 (30) [M$^+$]; Anal. Calcd. for $C_{15}H_{20}NO_5P$ (325.30) C, 55.38; H, 6.20; N, 4.31. Found: C, 55.56; H, 6.02; N, 4.13.

Diisopropyl isopropyl(2-oxo-2H-chromen-4-yl)phosphoramidate (18c)

Yield: 1.25 g (79.8%). Product **18c** was separated as yellow solid. m.p.: 183 ºC ($CHCl_3$); IR (KBr): ύ$_{max}$ = 1726 (C=O, lactone), 1325, 859 (P-NC), 1245 (P=O), 1070 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.26 (dd, $^3J_{HH}$ 4.6, $^4J_{PH}$ 3.8 Hz, 6H, NC*Me*$_2$), 1.35 (dd, $^3J_{HH}$ 6.7, $^4J_{PH}$ 4.1 Hz, 12H, P(OC*Me*$_2$)$_2$), 3.65 (m, 1H, NC*H*), 4.70 (m, 2H, P(OC*H*)$_2$), 5.54 (d, $^4J_{PH}$ 3.6 Hz, 1H, *H*C(3)/Ar), 7.10–7.35 (m, 4H, *H*/Ar) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 162.6 (*C*=O, lactone), 153.5, 144.3, 131.2, 129.3, 125.8, 118.8, 117.2, 88.7 (*C*/chromenone), 72.8 (d, $^2J_{PC}$ 13.7 Hz, P(O*C*H)$_2$), 52.6 (d, $^2J_{PC}$ 12.6 Hz, N*C*H), 24.4 (d, $^3J_{PC}$ 7.9 Hz, P(OC*Me*$_2$)$_2$), 23.1 (d, $^3J_{PC}$ 8.5 Hz, NC*Me*$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 17.8 ppm; MS (70 eV): *m*/*z* (%) = 367 (27) [M$^+$]; Anal.

Calcd. for $C_{18}H_{26}NO_5P$ (367.38) C, 58.85; H, 7.13; N, 3.81. Found: C, 58.96; H, 6.98; N, 3.65.

Synthesis of chromen phosphoramidates 20a–c

A solution of the coumarin **16** (4.3 mmol, 0.8 g) and dialkyl phosphite **7a-c** (4.5 mmol) in THF (15 mL) was refluxed (24–26 h, TLC). Under vacuum, the solvent was evaporated to dryness, and the residue was washed with cyclohexane. The product substance was crystallized from the proper solvent to provide the corresponding amidophosphates **20a**–**c**.

Dimethyl (2-oxo-2H-chromen-4-yl)phosphoramidate (20a)

Yield: 0.99 g (86.4%). Product **20a** was separated as yellow substance. m.p.: 196 ºC (MeCN); IR (KBr): υ_{max} = 3325 (NH), 1730 (C=O, lactone), 1235 (P=O, bonded), 1070 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 3.55 (d, $^3J_{PH}$ 11.5 Hz, 6H, P(O*Me*)$_2$), 5.18 (d, $^4J_{PH}$ 3.6 Hz, 1H, *H*C(3)/Ar), 7.02- 7.31 (m, 4H, *H*/Ar), 8.35 (s (br), 1H, *H*N, D_2O exch) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 163.0 (*C*=O, lactone), 153.8, 145.9, 131.6, 129.4, 126.6, 119.3, 116.9, 88.8 (*C*/chromenone), 52.6 (d, $^2J_{PC}$ 12.9 Hz, P(O*Me*)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 18.9 ppm; MS (70 eV): *m*/*z* (%) = 269 (48) [M^+], 268 (50) [M^+ - 1]; Anal. Calcd. for $C_{11}H_{12}NO_5P$ (269.19) C, 49.08; H, 4.49; N, 5.20. Found: C, 49.25; H, 4.34; N, 5.02.

Diethyl (2-oxo-2H-chromen-4-yl)phosphoramidate (20b)

Yield: 1.08 g (85.2%). Product **20b** was separated as yellow substance. m.p.: 180 ºC ($CHCl_3$); IR (KBr): υ_{max} = 3322 (NH), 1732 (C=O, lactone), 1229 (P=O, bonded), 1066 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.32 (dt, $^3J_{HH}$ 6.7, $^4J_{PH}$ 4.4 Hz, 6H, P(OC*Me*)$_2$), 4.09 (dq, $^3J_{HH}$ 6.7, $^3J_{PH}$ 10.6 Hz, 4H, P(OC*H*$_2$)$_2$), 5.17 (d, $^4J_{PH}$ 3.8 Hz, 1H, *H*C(3)/Ar), 7.11–7.37 (m, 4H, *H*/Ar)), 8.30 (s (br), 1H, *H*N, D_2O exch) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 163.4 (*C*=O, lactone), 154.4, 145.7, 131.5, 129.4, 126.3, 119.4, 116.6, 88.6 (*C*/chromenone), 62.9 (d, $^2J_{PC}$ 13.5 Hz, P(O*C*H$_2$)$_2$), 16.1 (d, $^3J_{PC}$ 8.2 Hz, P(OC*Me*)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 19.7 ppm; MS (70 eV): *m*/*z* (%) = 297 (44) [M^+], 296 (49) [M^+ - 1]; Anal. Calcd. for $C_{13}H_{16}NO_5P$ (297.24) C, 52.53; H, 5.43; N, 4.71. Found: C, 52.38; H, 5.26; N, 4.56.

Diisopropyl (2-oxo-2H-chromen-4-yl)phosphoramidate (20c)

Yield: 1.19 g (85.9%). Product **20c** was separated as Pale yellow substance. m.p.: 188 ºC ($CHCl_3$); IR (KBr): υ_{max} = 3329 (NH), 1735 (C=O, lactone), 1231 (P=O, bonded), 1061 (P-O-C) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.51 (dd, $^3J_{HH}$ 6.6, $^4J_{PH}$ 4.7 Hz, 12H, P(OC*Me*$_2$)$_2$), 4.70 (m, 2H, P(OC*H*)$_2$), 5.59 (d, $^4J_{PH}$ 3.4 Hz, 1H, *H*C(3)/Ar), 7.01–7.34 (m, 4H, *H*/Ar), 8.28 (s (br), 1H, *H*N, D_2O exch) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 163.7 (*C*=O, lactone), 154.3, 145.7, 131.6, 129.4, 126.4, 119.4, 117.7, 88.6 (*C*/chromenone), 72.2 (d, $^2J_{PC}$ 14.2 Hz, P(O*C*H)$_2$), 24.4 (d, $^3J_{PC}$ 8.7 Hz, P(OC*Me*$_2$)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 19.4 ppm; MS (70 eV): *m*/*z* (%) = 325 (37) [M^+], 324 (44) [M^+ - 1]; Anal. Calcd. for $C_{15}H_{20}NO_5P$ (325.30) C, 55.38; H, 6.20; N, 4.31. Found: C, 55.54; H, 6.01; N, 4.14.

Synthesis of phosphoranylidenetriazen chromen 21

To a solution of coumarin **16** (0.8 g, 4.3 mmol) in THF (10 mL) hexaethylphosphorus triamide (**11b**, 4.5 mmol) in THF (5 mL) was added dropwisly. The previous mixture was stirred at r.t. (10 h). The precipitated compound was filtered, collected, and washed many times with light petroleum (40–60°C) to produce **21**. The purity of compound **21** is sufficient for the spectroscopic analyses to carry out. Compound **21** is stable for three days.

4-{(1Z)-3-[Tris(diethylamino)phosphoranylidene]triaz-1-en-1-yl}-2H-chromen-2-one (21)

Yield: 1.52 g (82.0%). Product **21** was separated as orange substance. m.p.: 136 ºC (EtOH); IR (KBr): υ_{max} = 1732 (C=O, lactone),1350 (P=N), 1340, 865 (P(NEt_2)$_3$) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 0.96, 1.09 (2dt, $^3J_{HH}$ 8.6, $^4J_{PH}$ 6.5 Hz, 18H, P(N(C*Me*)$_2$)$_3$), 2.86, 3.26 (2dq, $^3J_{HH}$ 8.6, $^3J_{PH}$ 9.1 Hz, 12H, P(N(C*H*$_2$)$_2$)$_3$), 5.54 (s, 1H, *H*C(3)/Ar), 7.10–7.33 (m, 4H, *H*/Ar) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 163.9 (*C*=O, lactone), 163.2, 154.8, 132.7, 129.0, 126.6, 119.0, 112.6, 94.5 (*C*/chromenone), 39.5 (d, $^2J_{PC}$ 13.8 Hz, P(N(*C*H$_2$)$_2$)$_3$), 14.5 (d, $^3J_{PC}$ 8.5 Hz, P(N(C*Me*)$_2$)$_3$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 41.9 ppm; MS (70 eV): *m*/*z* (%) = 434 (25) [M^+], 406 (37), [M^+ - N_2] ; Anal. Calcd. for $C_{21}H_{35}N_6O_2P$ (434.52) C, 58.05; H, 8.12; N, 19.34. Found: C, 57.92; H, 7.95; N, 19.18.

Reaction of 16 with tris(diethylamino)phosphine (11b) in the presence of a protonating agent

Synthesis of compound 23

In one portion, Aminophosphine **11b** (4.5 mmol) solution in THF (10 mL) was added to a mixture of the coumarin **16** (0.8 g, 4.3 mmol) and H_2O (2 mL) in THF (10 mL). The previous mixture was refluxed (4 h). The excess solvent was vaporized till drying. The crude material was washed many times with light petroleum (40–60 ºC), and crystallized from cyclohexane to give phosphoric triamide **23** in 79.4 % yield.

***N,N,N',N'*-Tetraethyl-*N''*-(2-oxo-2H-chromen-4-yl)phosphoric triamide (23)**

Yield: 1.19 g (79.4%). Product **23** was separated as yellow substance. m.p.: 124 ºC (cyclohexane); IR (KBr): ύ$_{max}$ = 3330 (NH), 1740 (C=O, lactone), 1340, 865 (P(NEt$_2$)$_2$) 1232 (P=O, bonded) cm^{-1}; ^{1}H NMR (500 MHz, DMSO-d_6): δ = 1.11 (dt, $^3J_{HH}$ 7.7, $^4J_{PH}$ 5.4 Hz, 12H, P(N(C*Me*)$_2$)$_2$), 2.94 (dq, $^3J_{HH}$ 7.7, $^3J_{PH}$ 9.4 Hz, 8H, P(N(C*H$_2$*)$_2$)$_2$), 5.43 (d, $^4J_{PH}$ 3.4 Hz, 1H, *H*C(3)/Ar), 7.00- 7.38 (m, 4H, *H*/Ar), 9.02 (s (br), 1H, N*H*, D$_2$O exch) ppm; ^{13}C NMR (125.7 MHz, DMSO-d_6): δ = 163.7 (*C*=O, lactone), 154.0, 145.2, 131.4, 128.4, 126.2, 119.3, 118.0, 83.9 (*C*/chromenone), 41.1 (d, $^2J_{PC}$ 13.5 Hz, P(N(*C*H$_2$)$_2$)$_2$), 14.9 (d, $^3J_{PC}$ 8.2 Hz, P(N(C*Me*)$_2$)$_2$) ppm; ^{31}P NMR (202.4 MHz, DMSO-d_6): δ = 34.2 ppm; MS (70 eV): *m/z* (%) = 351 (30) [M$^+$], 350 (39) [M$^+$ - 1]; Anal. Calcd. for $C_{17}H_{26}N_3O_3P$ (351.38) C, 58.11; H, 7.46; N, 11.96. Found: C, 58.28; H, 7.29; N, 11.81.

2.2. In vitro antioxidant activity

1- DPPH• (2, 2´-diphenylpicrylhydrazyl) scavenging activity

The selected compounds **5b**, **10b**, **15b**, **18b**, **20b** and **23** were screened for antioxidant potency by using DPPH at concentration 0.5 mg/mL in 96-well plate as preliminary screening to find the most active compounds. Compounds **20b** and **23** were the most active compounds in this test. Other compounds show lowered antioxidant activity such as **5b**, **10b**, **15b**, and **18b**. Therefore, antioxidant screening was carried out on compounds (**20b** and **23**) at concentrations ranged from 0.4- 0.05 mg/mL.

Antioxidant activity by DPPH as scavenging was determined by applying the bleaching of the purple colored methanol solution of DPPH$^•$ and methanol was applied as blank, while the ascorbic acid (0.001-0.02 mg/mL) was applied as reference compound as cited in Mossa and Nawwar [34], with some modification.

Different concentration of compounds **20b** and **23** were prepared and 250 µL of each were transfer to test tube and the same volume of DPPH (0.1 mM) in methanol was used. The tubes were mixed will and stand in dark at r.t. (0.5 h). The absorbance of DPPH in methanol was measured at 517 nm and use as control while methanol was used as blank.

Radical scavenging activity of DPPH$^•$ was expressed as % of inhibition and calculated from the following equation:

$$I\ (\%) = [(A_C - A_S)/A_C] \times 100$$

Where: A_C and A_S are the absorbance of the control and compound, respectively.

The value of IC_{50} is the concentration of compound that caused 50% inhibition.

2- Scavenging activity by using ABTS$^{•+}$ (2, 2'azinobis- (3-ethylbenzthiazoline -6-sulphonic acid))

The radical scavenging efficiency by ABTS$^{•+}$ test was studied on compounds **20b** and **23** depending on the method mentioned by Mossa et al. [35] with some modification.

Production of ABTS$^{•+}$ cation radical was taken place by the reaction of ABTS (7 mM) with potassium persulfate (2.45 mM) in water at r.t. (12 h) in the dark. Before antioxidant activity test the ABTS$^{•+}$ solution was diluted to obtain an absorbance of 0.70 ± 0.02 at 734 nm by 0.1 M phosphate buffer at pH 7.4. The mixture content (250 µL) of several concentrations for each sample and the same volume of ABTS and ascorbic acid (1-20 µg/mL) was used as standard. The tubes were mixed well and stand in dark at room temperature (0.5 h). The absorbance of ABTS was misread at 734 nm and use as control while water was used as blank. ABTS radical scavenging activity of ABTS$^{•+}$ was expressed as % of inhibition.

The scavenging potency of ABTS$^{•+}$ radical was determined by application of the following formula:

$$ABTS^{•+}\ Scavenging\ effect\ (\%) = [(A_{control} - A_{compound})/A_{control})] \times 100$$

Where $A_{control}$ is the absorbance of the control and $A_{compound}$ is the absorbance of the compound. The IC_{50} value is the concentration of sample that caused 50% inhibition was calculated.

3- Scavenging activity of hydrogen peroxide (H_2O_2)

Scavenging activity of compounds **20b** and **23** by hydrogen peroxide was determined and carried out following the way cited by Ozsoy et al. [36]. The test is depended on the decrease in absorbance of H_2O_2 upon oxidation of H_2O_2 by antioxidant compound. H_2O_2 solution (40 mM) was prepared in phosphate buffer (0.1 M pH 7.4). Furthermore 300 µL of H_2O_2 solution was transferred to test tube contain 300-µL different concentration of each compounds (0.6-0.1 mg/mL). The final volume was completed to 3 mL by 40 mM phosphate buffer (*pH* 7.4). The absorbance of mixture and H_2O_2 were measured after 10 min at 230 nm against phosphate buffer solution as a blank solution. The reference compound used in this test was ascorbic acid (0.005-0.05 mg/mL).

The percentage of scavenged [H_2O_2] = [(A_c-As)/A_c] x 100

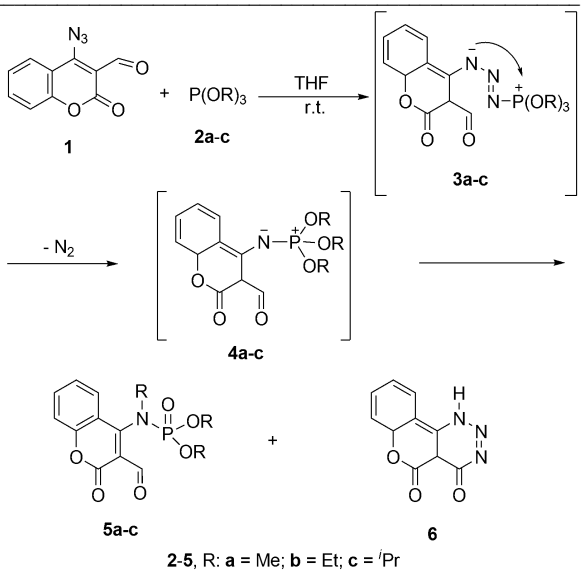
Where A_c is the absorbance of the control while, As is the absorbance in the presence of compound. The IC_{50} value is the concentration of the sample, which caused 50% inhibition was calculated.

Results and Discussion

3.1. Chemistry

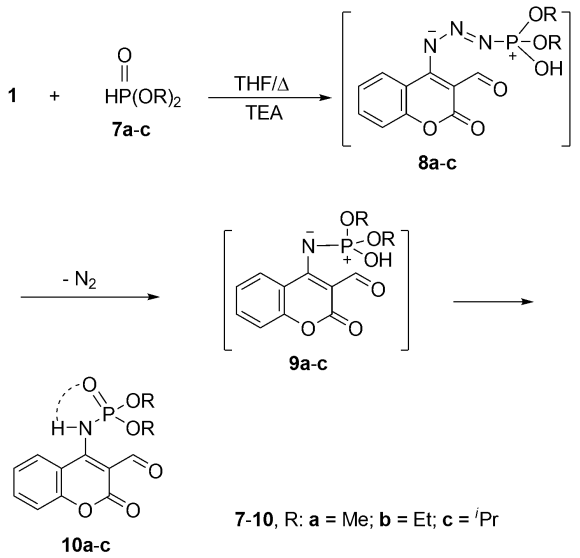
Treatment of the bifunctional compound **1** by trialkyl phosphites **2a**–**c** in dry tetrahydrofuran (THF) at room temperature in 6–8 h led to the formation of the corresponding phosphoramidates **5a**–**c** as major products in ~ 75% yields, along with the known triazinedione **6** [30] in ~ 11% yields (Scheme 1). Compounds **5a**–**c** were recognized from their elemental analyses and spectroscopic data. The ^{31}P NMR spectrum of **5a** showed signal at δ_P 19.2 ppm, while, the ^{1}H NMR spectrum of **5a** displayed at δ_H 3.79 ppm a doublet (d, $^3J_{PH}$ 7.6 Hz) indicated the phosphonate moiety (MeOP). The methyl group attached to nitrogen (NMe) was assigned by a doublet at 3.16 (3H, $^3J_{PH}$ 8.1 Hz). In the IR spectrum of **5a**, the absorption bands at 1245 and 1032 cm^{-1} related to P=O and P–O–C groups were observed. Meanwhile, the azide stretching vibration band in the IR chart of **1** at 2095 cm^{-1} has been lost in the IR chart of **5a**.

A plausible explanation for the mechanism of the reaction of azidochromene carbaldehyde **1** with trialkyl phosphites **2a**–**c** is proposed in Scheme 1. Compounds **5a-c** could be formed by the application of TAP **2a-c** to the azide **1** to form phosphorimidates **3a-c** which follows Staudinger reaction and loss of nitrogen molecule to form the corresponding phosphorimidates **4** [37, 38]. Further the phosphorimidates **4** were been underwent intramolecular group translocation of an alkyl moiety to form the phosphoramidates **5a**–**c**, this in turn, promotes the stability of the resulting compound **5a–c** [21, 29, 39-43]. Whereas the known triazinedione **6** was formed through cyclization of azide **1** due to the presence of the trialkyl phosphites **2a**–**c** that acted as Lewis base and facilitate formation of the triazine cyclic form **6** [30] (Scheme 1).



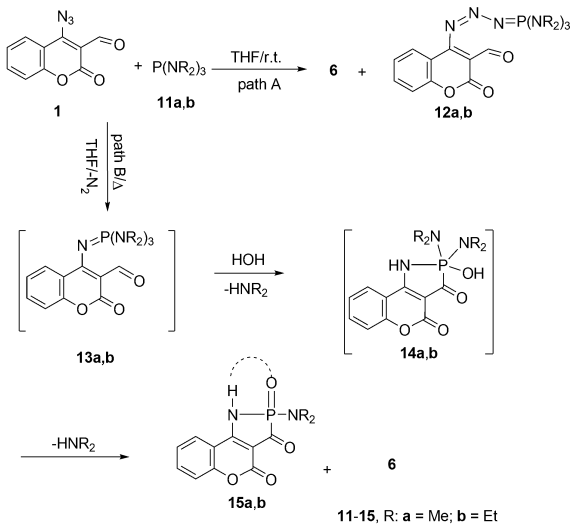
Scheme 1. Synthesis of chromen phosphoramidates 5a–c & triazinedione 6.

When azide **1** was reacted with dialkyl phosphites **7a-c** in refluxed THF for 21–24 h, the reaction moved in the same way, to yield the phosphoramidates **10a-c** (~80 % yield, Scheme 2). The formation of phosphoramidates **10** is assumed to take place *via* the premier attack of nucleophilic phosphorus on the terminal nitrogen atom of azide to afford the dipolar species **8a-c**. Following Staudinger reaction, the dipolar species **8a-c** underwent loss of a nitrogen molecule to produce **9a-c**, further rearrangement led to the formation of the corresponding phosphoramidates **10a-c** [43, 44].



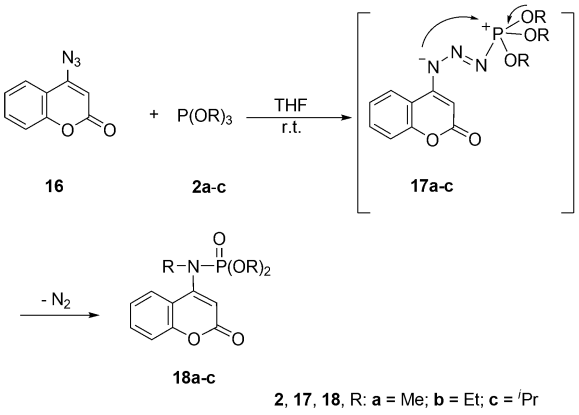
Scheme 2. Synthesis of phosphoramidates 10a–c.

After that, the azidocoumarin **1** was reacted with hexaalkylphosphorus triamides **11a**,**b** in tetrahydrofuran at r.t. (~15 h), phosphoranylidenetriazenes **12a**,**b** could be separated and identified (~80 % yield) [38] along with the cyclic trizine **6** (~9 % yield) (Scheme 3 path (A)). The ^{31}P NMR spectrum of **12a** showed one signal at δ_P 39.5. Compounds **12a**,**b** were formed *via* the reaction of the trivalent phosphorous reagents **11** and the terminal *N*- atom of the azide **1** [38]. The stability of compounds **12** under strict conditions were attributed to the presence of hexaalkylphosphorane triamides which are strong donor moiety [45]. On applying of a protonating agent (e.g., 2 mL of H_2O) to the reaction medium, the reaction is clearly accelerated producing the target azaphospholes **15a**,**b** (~75%). Compounds **15a**,**b** were obtained *via* the formation of the imines **13a**,**b** according to the Staudinger mechanism [29, 40, 41, 43, 46] accompanied by reacting with a molecule of H_2O to afford the intermediates **14** *via* extrusion of dialkylamine (HNR_2) moiety. Stabilization of **14** proceed through extrusion of one more molecule of dialkylamine leading to synthesis of azaphospholes **15a**,**b** (Scheme 3 path (B)). The ^{31}P NMR spectrum of **15a**,**b** showed signals around δ_P= 15 ppm. The cyclic compound **6** was also isolated along with azaphospholes **15a**,**b** [29, 40, 41].



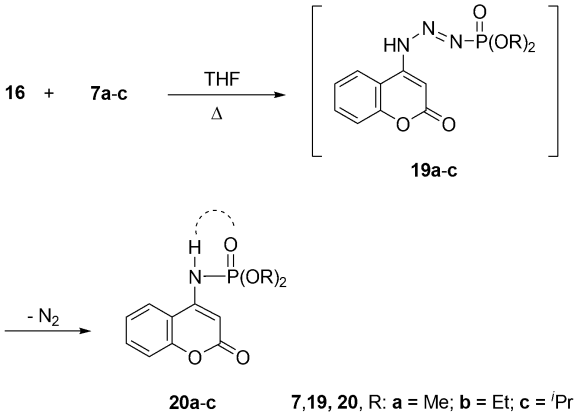
Scheme 3. Synthesis of triazenylidene-phosphoranes 12a,b & azaphospholes 15a,b & 6.

Next, we studied the behavior of another coumarin derivative **16** [33] against the phosphorus reagents used before, **2a-c**, **7a-c**, and **11b**. The reaction between trialkyl phosphites **2a-c** and coumarin derivative **16** proceeded in presence of THF at ambient temperature, leading to the corresponding chromen phosphoramidates **18a-c** (~ 79% yield) (the mechanism was described in Scheme 4). The ^{31}P NMR spectrum of **18a** showed one signal at δ_P 17.6, and ^{1}H NMR spectrum displayed a doublet (6H, $^3J_{PH}$ 10.1 Hz) at δ_H 3.81 correlated to the phosphonate moiety [$(MeO)_2P$], in addition to another doublet at δ_H 2.93 ($^3J_{PH}$ 9.8 Hz, 3H) due to CH_3 protons linked to nitrogen atom (NMe). On the other hand, The IR spectrum of **18a** detects the loss of stretching vibration band at 2170 (N_3), instead, they showed bands at 1255 (P=O) and 1080 (P-O-CH_3).The formation of amidophosphates **18a-c** progressed through nucleophilic addition reaction of the phosphorus reagents **2a-c** to the terminal *N*- atom of the azide to give the intermediate **17a-c** and then expulsion of nitrogen molecule (Staudinger reaction), followed by intramolecular phosphono-alkyl group translocation to afford the amidophosphate products **18a-c** [21, 40-45].



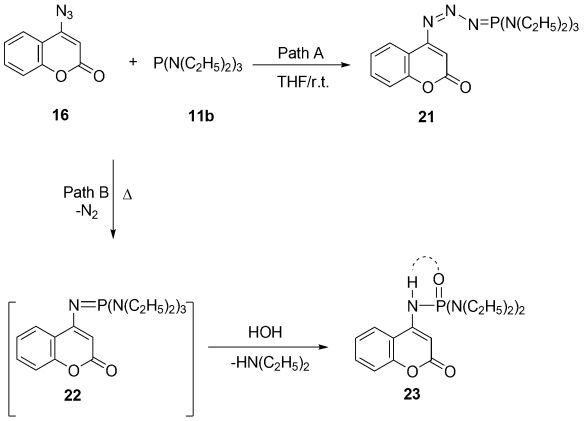
Scheme 4 Synthesis of phosphoramidates 18a–c

In addition, the chromenphosphoramidates **20a-c** (~85% yield) were easily produced *via* the reaction of coumarin derivative **16** and DAP **7a-c** in refluxed tetrahydrofuran. clearly, the first nucleophilic attack of the phosphite-phosphorus upon the N- atom of the aza group in compound **16** produced the triazenylphosphonates **19a-c**, followed by expulsion of nitrogen (Staudinger reaction) to give the phosphoramidate derivatives **20a-c** (Scheme 5) [43, 44, 46].



scheme 5. Synthesis of chromen phosphoramidates 20a–c.

Finally, treating the coumarin derivative **16** with tris(diethylamino)phosphine (**11b**) in tetrahydrofuran at ambient temperature (10h) afforded the phosphoranylidenetriazen chromen **21** as displayed in Scheme 6, Path (A). Triazen chromen **21** was powder compound, sensitive to water and stable in the desiccator for three days. The use of 2 mL of H_2O as protonating agent in the reaction medium, led to the acceleration of the reaction and formation of chromen phosphoric triamide **23** [33, 41, 44-47] (82%) (Scheme 6, Path (B)). The structure and purity of compounds **21** and **23** were established by ^{1}H-, ^{13}C-, and ^{31}P NMR spectroscopy, mass spectroscopy (EI), elemental analyses.



scheme 6. Synthesis of phosphoranylidenetriazen 21 & phosphoric triamide 23.

3.2. Biological evaluation

Antioxidant activity

Organophosphorus compounds, in particular P-heterocycles have been known as antioxidant drugs [18, 48]. Therefore, the activity of some new synthesized coumarin derivatives (diethyl chromen phosphoramidate **5b**, diethyl chromen amidophosphate **10b**, diethyl aminochromeno azaphosphole **15b**, diethyl ethyl chromen amidophosphate **18b**, diethyl chromen amidophosphate **20b**, and tetraethyl chromen phosphoric triamide **23**) were evaluated in *vitro* using DPPH preliminary screening test for antioxidant activity at concentration 0.5 mg/mL. Results showed that, the most active compounds were **20b** and **23** at the concentration (500 µg/mL) (Fig. 1).

Figure 1. DPPH test as preliminary screening for tested products at 0.5 mg/mL concentration in 96-well plate.

Therefore, compounds **20b** and **23** were selected for further studies of antioxidant activity using DPPH, ABTS, and H_2O_2 and compared with natural antioxidant, ascorbic acid. Results showed that both compounds had moderate to good scavenging activity on DPPH and ABTS (Figs. 2, 3) and compound **20b** has the highest activity.

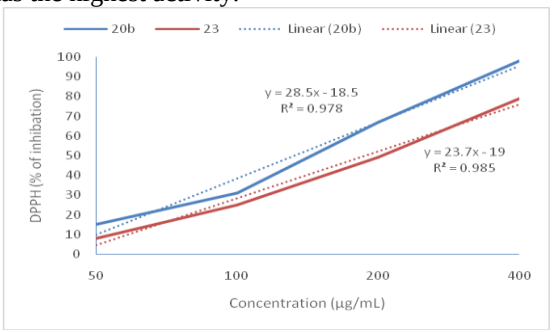


Figure 2. DPPH scavenging activity of compounds 20b and 23.

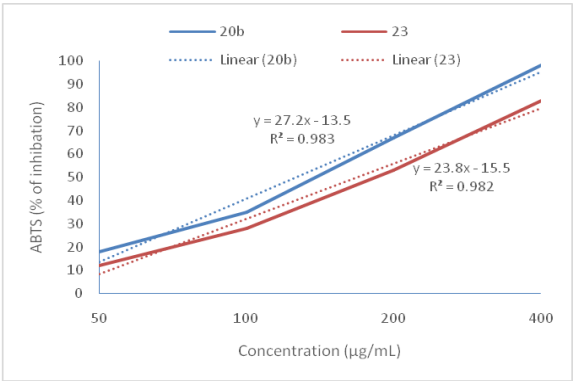
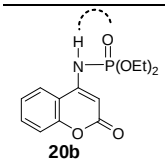
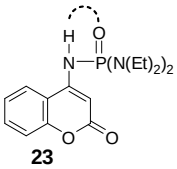


Figure 3. ABTS scavenging activity of compound 20b and 23.

The IC_{50} values of compounds **20b** and **23** were 0.161 mg/mL and 0.23436 mg/mL of DPPH and 0.15595 mg/mL and 0.2215 mg/mL of ABTS compared to 0.0065 mg/mL and 0.0059 mg/mL of ascorbic acid, respectively as shown in table 1.

Table 1. Scavenging activity of compounds 20b, 23 and ascorbic acid using DPPH, ABTS and hydrogen peroxide (H_2O_2)

Compound	IC_{50} (mg/mL)		
	DPPH	ABTS	H_2O_2
 **20b**	0.161	0.15595	0.29856
 **23**	0.23436	0.2215	0.36724
Ascorbic acid	0.0065	0.0059	0.0085

*IC_{50} was the value that represented the concentration of the sample, which caused 50% inhibition.

Result of hydrogen peroxide test proved that the chromen-phosphoramidate **20b** showed high radical scavenging activity on hydrogen peroxide (H_2O_2) with IC_{50} value 0.29856 mg/mL while that of compound **23** is 0.36724 compared to 0.0085 mg/mL of ascorbic acid (Table 1 and Fig. 4).

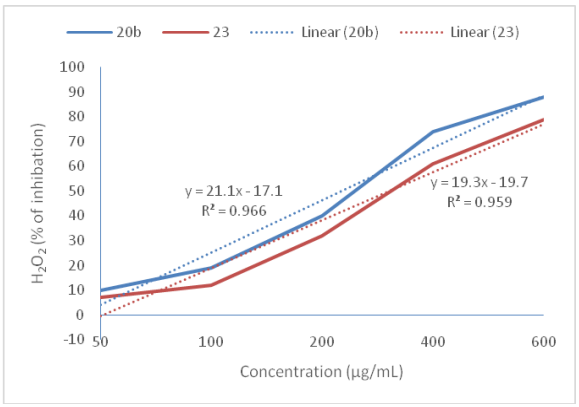


Figure 4. H_2O_2 scavenging activity of compound 20b and 23

Hydrogen peroxide can deactivate a few enzymes because it considers as soft oxidizing compound, ordinarily by oxidation effect on essential thiol groups (-SH). H_2O_2 can pass through cell membranes speedly and when inside the cell it may be reacted with Fe_2 and probably Cu_2 to form OH radicals, that may be the source of many toxic effects as shown in Eq. (1) [49]. Therefore, getting rid of hydrogen peroxide is very valuable for antioxidant protection in the cell.

Eq. (1)

$$Fe^{2+} + H_2O_2 \longrightarrow Fe^{3+} + OH^{\bullet} + OH^{-}$$

Eq. (1)

The strongest antioxidant properties observed in compounds **20b** and **23** is attributed to the presence of phosphoramidate moiety and -NH moiety. This could be depicted as the easy release of nitrogen–H to obtain radical with stable molecular structure that will stop the chain reaction.

3. Conclusions

In summary, we were successful in preparing two series of phosphorylated coumarin systems (bicyclic and tricyclic) in good to excellent yields. The new compounds were achieved by using P (*III*), trialkyl phosphites **2a-c**, dialkylphosphonates **7a-c**, and tris(dialkylamino)phosphines **11a**,**b** with 4-azido-2-oxo-2*H*-chromene-3-carbaldehyde (**1**) and 4-azidocoumarin (**16**). The proposed procedures of this work have different advantages, like good yields, suitable reaction conditions, and simple workup method. The differences and similarities in the behavior of azidocoumarins **1** and **16** toward the same phosphorus reagents were discussed along the Schemes 1-6. Finally, 6 out of the 18 synthesized derivatives were tested for their antioxidant potency. Results showed that compound **20b** > compound **23** had the most scavenging properties on DPPH and ABTS evaluation test for antioxidant activity.

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تخليق مشتقات كومارين جديدة تحمل إستر الفوسفور وتقييم أنشطة هذه المركبات كالمضادات للأكسدة

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تعرف مشتقات الكرومون والكرومين انها ذات فوائد إقتصادية (عقاقير) حيث أنه من المعروف أنها ذات فوائد فارماكولجية عالية ولهذا عنى هذا البحث بدراسة مقارنه لسلوك نمطان من مشتقات الازيد كومارين تجاه مجموعة من كواشف الفسفور مثل كواشف ثنائى- وثلاثى الكيل الفوسفيت وكذا كواشف سداسى ألكيل أمينوالفوسفينات. تم أجراء التفاعلات فى وجود مذيب THF وقد تم الحصول على مشتقات الفسفوراميدات و مشتقات الفسفورانيلدين ترايزين و بالاضافة الى مشتقات ازافسفول المقابلة من تفاعل البادئ الاول الدهيد ازيدو الكومارين مع كواشف الفسفور المستخدمة.

وفى الحالة الثانية تم الحصول على مشتقات من كرومين فسفوراميدات ومن كرومين فسفوريك ترای اميد المقابلة نتيجة من تفاعل البادئ الاخر الازيدو كومارين مع ثنائى وثلاثى الكيل الفوسفيت عن طريق تفاعل شتودجر. بينما بتفاعل الازيدو كومارين مع سداسى ألكيل أمينوالفوسفينات ينتج مشتق الفسفورانيليدين ترايزين كومارين.

- تم عمل فحص وتقييم لأنشطة ستة من المركبات المحضرة كمضادات للأكسدة. وقد اظهرت نتيجة التقييم انه يوجد مركبين من المركبات المختبرة لهم نشاط جيد كمضادات للأكسدة.