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Cleavage of Diethyl Chromonyl α-Aminophosphonate with Nitrogen and Carbon Nucleophiles: A Synthetic Approach and Biological Evaluations of A Series of Novel Azoles, Azines and Azepines Containing α-Aminophosphonate and Phosphonate Groups

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ABSTRACT

A convenient synthetic approach leading to synthesize a series of novel substituted azoles, azines and azepines linked to α-aminophosphonate moiety was achieved. The methodology depends on ring-opening and ring-closure (RORC) of chromone ring of diethyl chromonyl α-aminophosphonate **1** *via* its reaction with nitrogen nucleophiles such as primary amines, 1,2-, 1,3- and 1,4-*bi*-nucleophiles in ethanolic sodium ethoxide. Also, treatment of compound **1** with some acyclic and cyclic active methylene compounds under the same reaction conditions afforded interesting novel isolated and fused pyridine systems bearing phosphonate groups at α-position. The screening of antimicrobial activity for the synthesized compounds indicates that connection of pyrazole, oxazepine and benzodiazepine rings with α-aminophosphonate moiety exhibited good antimicrobial effects. Also, evaluation of their antioxidant properties exemplifies that the compounds having 1,5-benzoxazepinyl and 1,5-benzodiazepinyl units in combination with α-amino-phosphonic diester moiety are the most powerful antioxidant agents.

KEYWORDS: Chromone, α-Aminophosphonate, Phosphonate, Antimicrobial, Antioxidant.

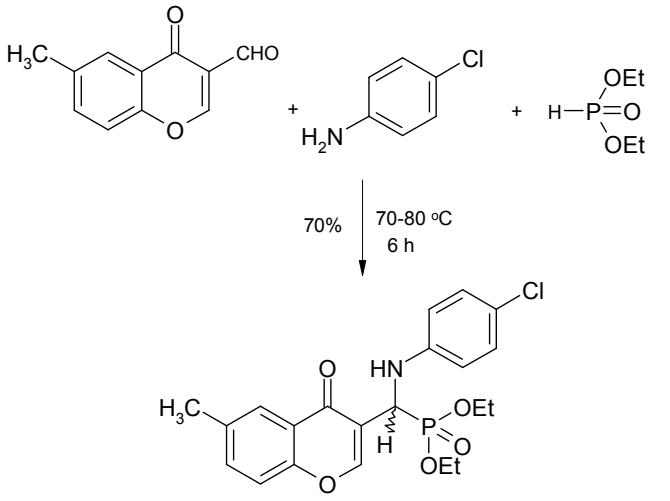
INTRODUCTION

Chromone compounds have attracted considerable attention as highly reactive compounds, which can serve as starting materials in synthesis of a whole series of heterocycles with useful properties due to two strong electrophilic centers (carbon atoms C−2 and C−4 of the chromone system).[1,2] The 3-heteroaryl chromones possess

a highly polarized C_2-C_3 π-bond and their reactions with *bi*-nucleophiles occur predominantly *via* a nucleophilic attack on the unsubstituted C−2 atom (1,4-addition) and are accompanied by ring-opening to form the β-carbonyl intermediate capable of undergoing intramolecular heterocyclization.[3,4] On the other hand, α-amino-phosphonates act as important family of organophosphorus compounds which possesses various important biological properties.[5,6] Some of these biological activities are enzyme inhibition,[7] antitumor,[8] antibiotics[9] and antiproliferative.[10] α-Aminophosphonates containing five- and six-membered heterocycles at α-position are known.[11-16] To the best our knowledge, α-aminophosphonates possess seven-membered heterocycles at α-position have not been reported hitherto. Moreover a basic difficulty in the synthesis of parent α-heterocyclic α-aminophosphonate is an applicability of known regular procedures for synthesis of the typical α-amino-phosphonates. Therefore, there is a need to search for new methods, which could be more useful in preparation of α-heterocyclic α-aminophosphonates. In continuation of our interest in the synthesis of new α-aminophosphonates containing different bioactive heterocyclic rings,[17−19] we report herein an efficient synthesis of novel α-aminophosphonates containing different nitrogen heterocycles and also α-pyridinyl phosphonates were achieved. The method depends on ring-opening and ring-closure (RORC) of chromone ring in diethyl [(4-chlorophenylamino)(6-methyl-4-oxo-4*H*-chromen-3-yl)methyl]phosphonate (**1**) *via* its reaction with nitrogen and carbon nucleophiles in ethanolic sodium ethoxide. The antimicrobial activities and antioxidant properties of the synthesized compounds were also evaluated.

RESULTS AND DISCUSSION

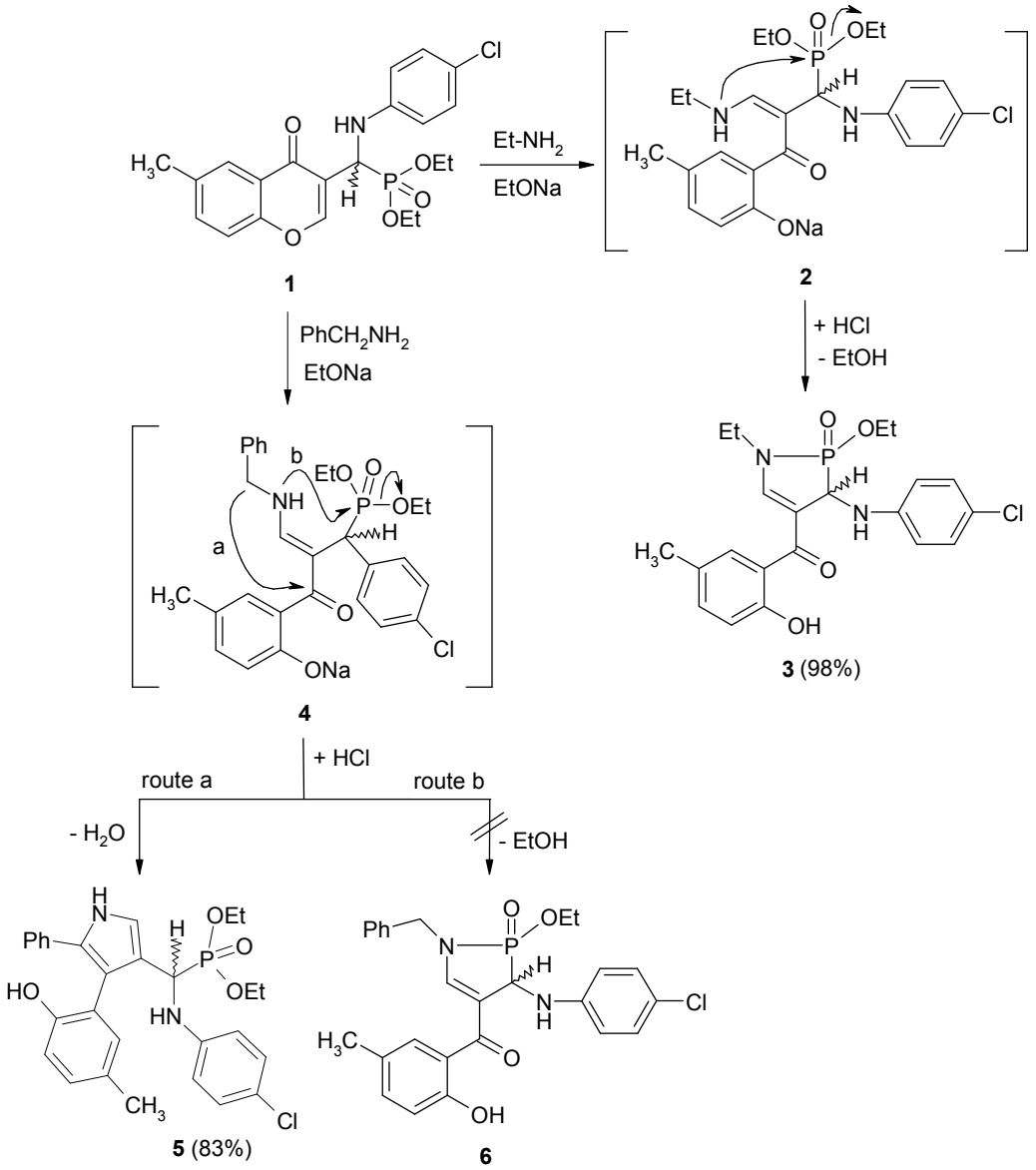
The starting material, diethyl [(4-chlorophenylamino)(6-methyl-4-oxo-4*H*-chromen-3-yl)methyl]phosphonate (**1**) used in this study, was prepared in our recent article [20] in a quantitative yield using a modified literature procedure [21] by fusion of 6-methyl-4-oxo-4*H*-chromen-3-carboxaldehyde, 4-chloroaniline and diethyl phosphite at 70−80 ^{o}C (Scheme 1). The chemical reactivity of diethyl chromonyl α-aminophosphonate **1** towards some nitrogen and carbon nucleophiles in ethanolic sodium ethoxide was investigated.



Scheme 1

At first, we investigated reaction of compound **1** with ethylamine. This reaction was carried out in ethanolic sodium ethoxide under reflux to give 3-(4-chlorophenylamino)-2-ethoxy-1-ethyl-4-[(2-hydroxy-5-methylphenyl)carbonyl]-2-oxido-2,3-dihydro-1*H*-1,2-azaphosphole (**3**) as cyclic α-aminophosphonate in excellent yield (Scheme 2). We proposed the reaction mechanistic pathway for this reaction, took place *via* a nucleophilic attack of the ethylamine on chromone ring at carbon atom C−2. There was an opening of the chromone ring to give the nonisolable intermediate **2** which underwent a nucleophilic intramolecular cyclization on the phosphorus atom by NH of ethylamine moiety (Scheme 2).[22,23]

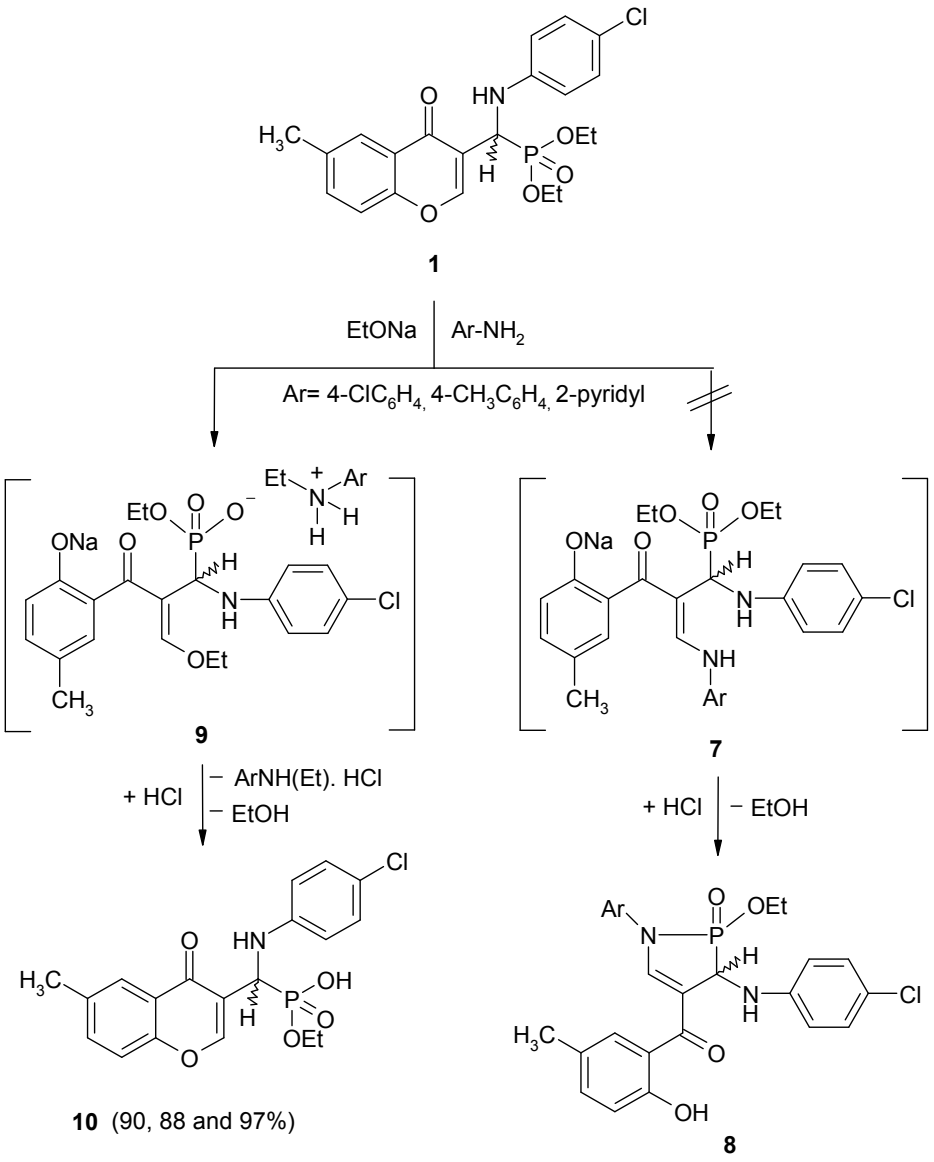
Similarly, treatment of compound **1** with benzylamine under the same reaction conditions gave the pyrrolyl α-aminophosphonate **5** as orange crystalline product in 83% yield (Scheme 2). The suggested reaction mechanism for this reaction is similar to formation of compound **3** to afford the nonisolable intermediate **4**, but the cyclization process is a result of condensation between the benzyl group and the carbonyl group that is more electrophilic center than phosphonate group (route a) (Scheme 2).



Scheme 2

Analogue reaction of the diethyl chromonyl α-aminophosphonate **1** with aromatic amines such as 4-chloroaniline, *p*-toluidine and 2-aminopyridine did not lead to construction of products which are similar to that formed in case of the used aliphatic amines. However, all the used aromatic amines gave only one product identified with ethyl {(4-chlorophenylamino)(6-methyl-4-oxo-4*H*-chromen-3-yl) methyl}phosphonate (**10**) (Scheme 3). The aromatic amines reacted exclusively with the diester group of the phosphonate forming ammonium salts of phosphonic acid monoester **9** as intermediate and the ring opening occurred spontaneously as a result

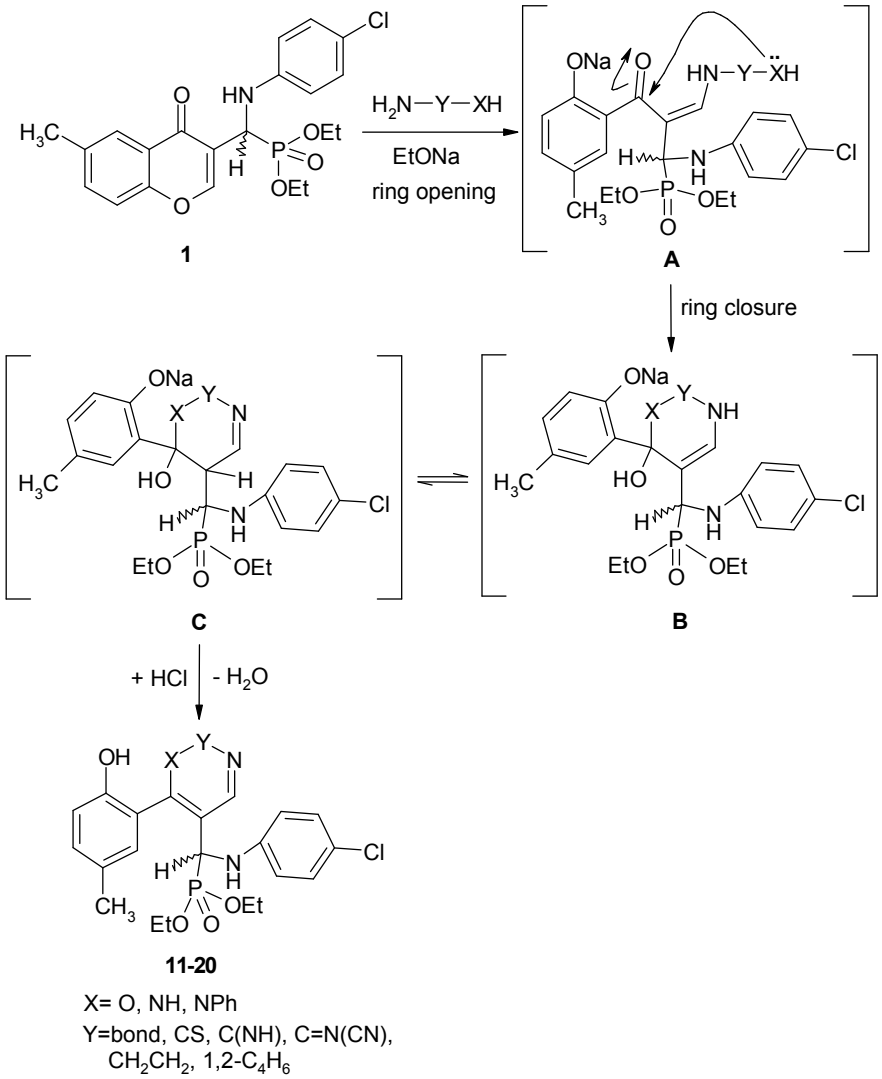
of effect of sodium ethoxide. The neutralization of the reaction mixtures with diluted hydrochloric acid (5%) underwent ring closure into chromone ring and elimination of amine hydrochloride salt to afford the final product **10** (Scheme 3). Such transfer of an ethyl ester group from α-aminophosphonic ester to aromatic amines was exhaustively described in several studies.[24,25] Compound **10** gave negative result with alcoholic $FeCl_3$ confirming the absence of phenolic group obtained by γ-pyrone ring opening.



Scheme 3

The synthetic utilities of diethyl chromonyl α-aminophosphonate **1** are derived from its reaction with 1,2-, 1,3- and 1,4-*bi*-nucleophiles that start predominantly from

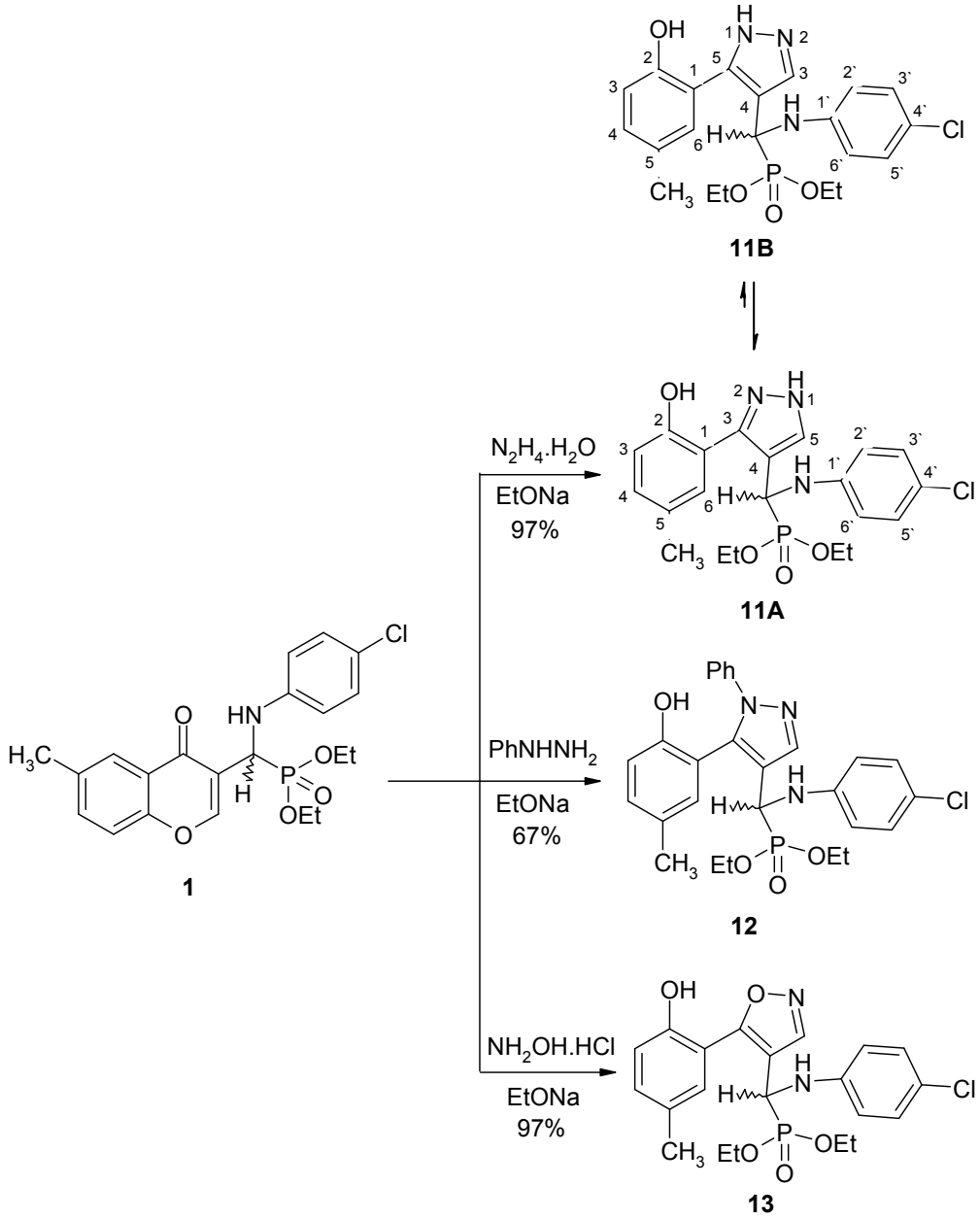
a nucleophilic attack at the unsubstituted C−2 atom followed by γ-pyrone ring opening especially in ethanolic sodium ethoxide. Subsequent ring closure to the carbonyl group at the aromatic ring forms a whole series of azaheterocycles after losing of water molecule (Scheme 4).[2,26]



Scheme 4

One would expect that the reaction of diethyl chromonyl α-aminophosphonate **1** with 1,2-*bi*-nucleophilic reagents would lead to novel azolyl α-aminophosphonates. Thus, compound **1** reacted with hydrazine hydrate in ethanolic sodium ethoxide under reflux to afford the corresponding pyrazolyl α-aminophosphonates **11A**,**B**. The latter compound existed in two tautomeric forms **11A** and **11B** in ratio 5:1 as a result of possible prototropism (Scheme 5).[27,28]

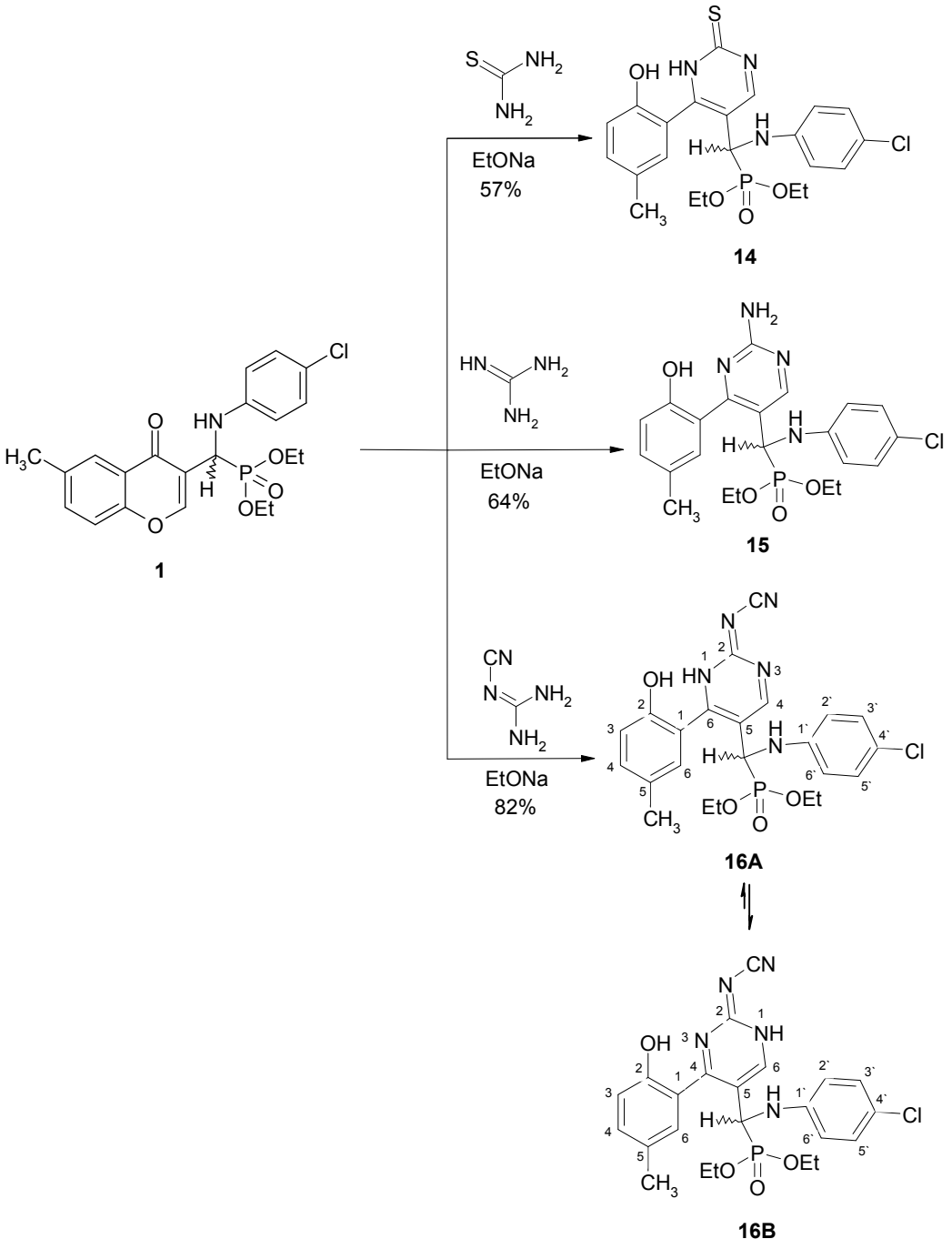
Similarly, treatment of compound **1** with phenylhydrazine and hydroxylamine hydrochloride under the same reaction conditions gave the corresponding pyrazolyl α-aminophosphonate **12** and isoxazolyl α-aminophosphonate **13**, respectively (Scheme 5).



Scheme 5

Pyrimidines containing α-aminophosphonate skeleton are of great interest according to their herbicidal activities.[16] Thus, diethyl chromonyl α-amino-phosphonate **1** was reacted with thiourea, guanidinium carbonate and cyanoguanidine

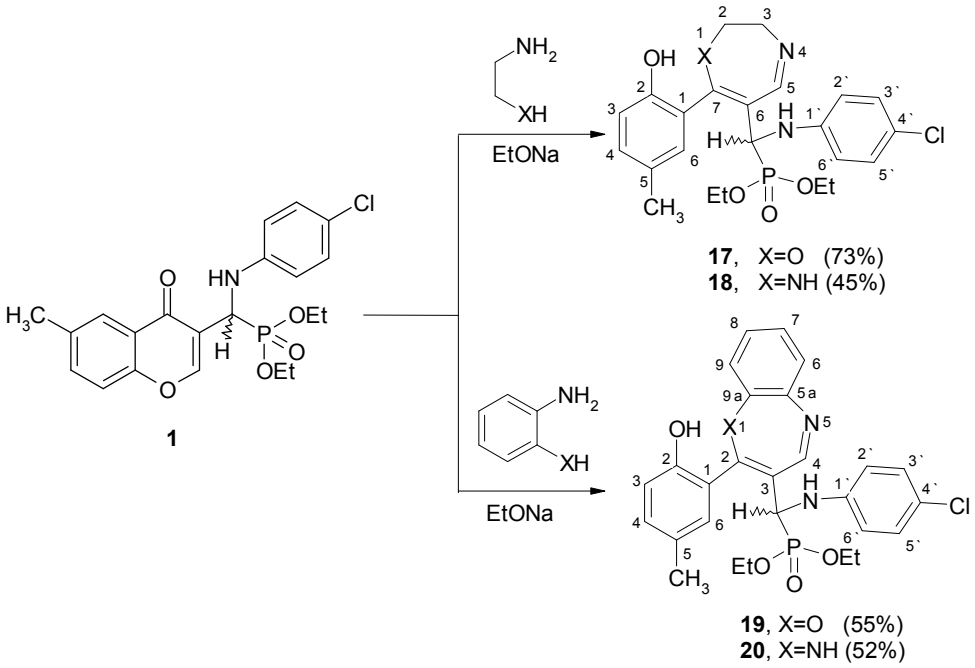
as 1,3-*bi*-nucleophiles in ethanolic sodium ethoxide to yield the corresponding pyrimidinyl α-aminophosphonates **14**−**16**, respectively (Scheme 6).



Scheme 6

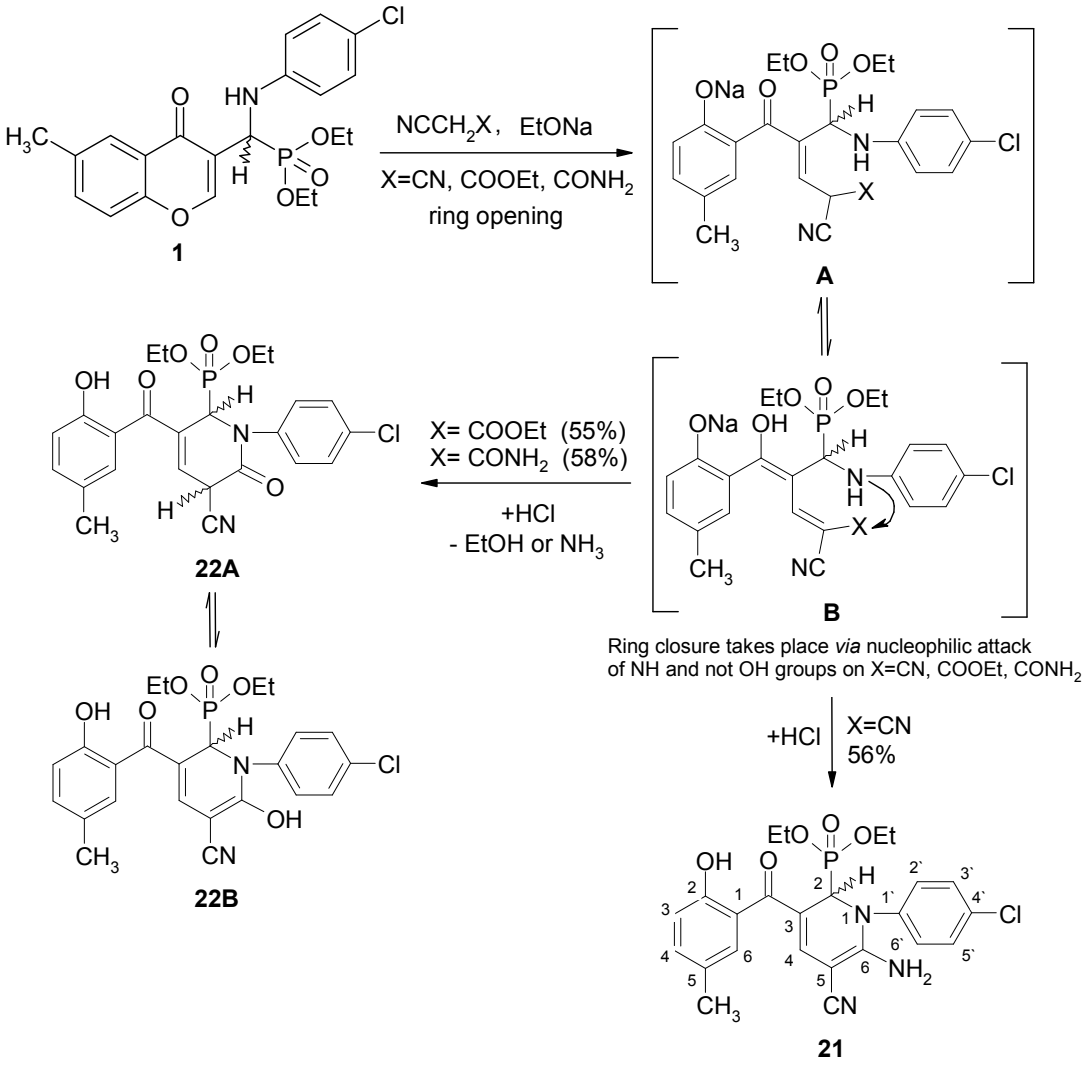
To the best our knowledge, α-aminophosphonates possess seven-membered rings are unknown. Thus, treatment of diethyl chromonyl α-aminophosphonate **1** with some selected 1,4-*bi*-nucleophiles such as ethanolamine, ethylenediamine,

2-aminophenol and 1,2-phenylenediamine in ethanolic sodium ethoxide led to the formation of interesting novel diethyl α-aminophosphonates containing dihydroxazepine and dihydrodiazepine rings **17** and **18**, and their benzo analogues **19** and **20**, respectively, in moderate yields (Scheme 7).



Scheme 7

In recent years, much attention was focused on the synthesis of phosphonate esters of *N*-heterocyclic systems (pyridine or quinoline) and their metal complexes, because of their potential applications and significant antitumor activities.[29,30] The foregoing results prompted us to investigate the applicability and synthetic potency of compound **1** towards active acyclic methylene compounds such as malononitrile, ethyl cyanoacetate and cyanoacetamide. Thus, refluxing of compound **1** with malononitrile in ethanolic sodium ethoxide afforded diethyl {6-amino-1-(4-chlorophenyl)-5-cyano-3-[(2-hydroxy-5-methylphenyl)carbonyl]-1,2-dihydro-pyridin-2-yl]}phosphonate (**21**) (Scheme 8). On the basis of spectral data of compound **21**, the formation of this product may be attributed to the intermediacy of the nonisolable adduct **A** or **B** which formed *via* a nucleophilic attack of active methylene carbanion at C−2 of chromone ring. The intermediate **A** or **B** underwent cyclization *via* addition of NH (more nucleophilic than OH) on the nitrile group (Scheme 8).

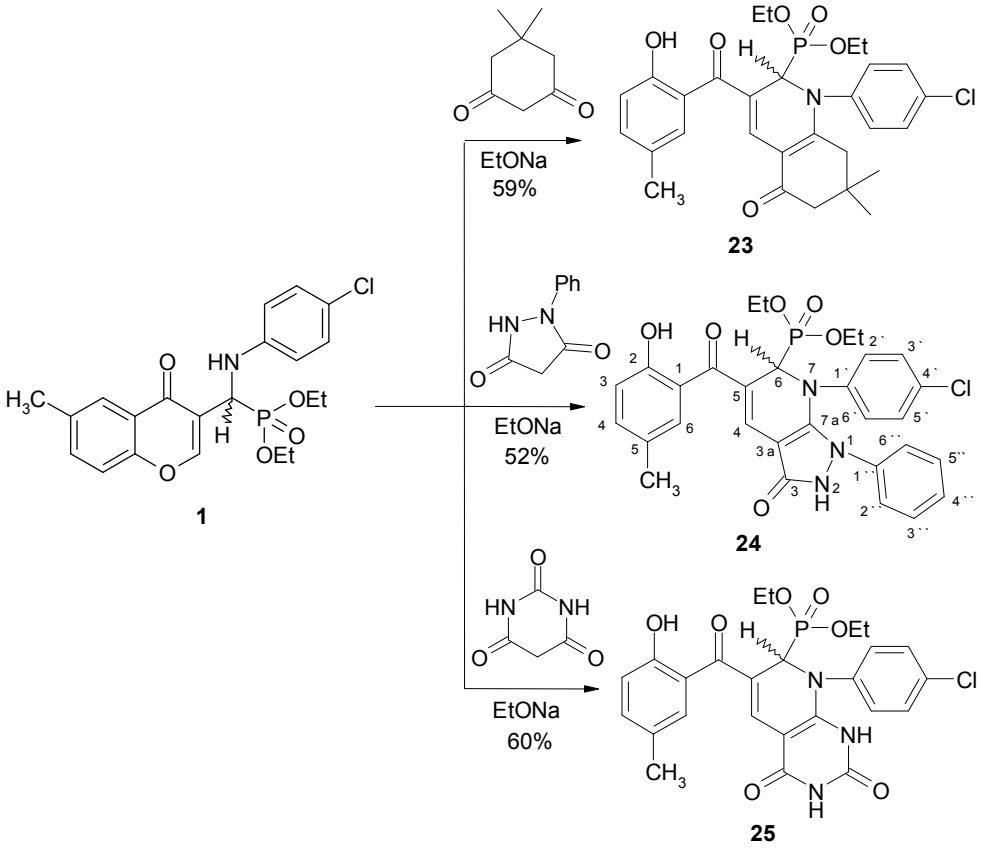


Scheme 8

Similarly, treatment of compound **1** with ethyl cyanoacetate or cyanoacetamide under the same basic conditions gave one product, existing in two tautomeric forms namely, diethyl{1-(4-chlorophenyl)-5-cyano-3-[(2-hydroxy-5-methylphenyl)carbonyl] -6-oxo-1,2,5,6-tetrahydropyridin-2-yl}phosphonate (**22A**, *keto form*) and diethyl {1-(4-chlorophenyl)-5-cyano-6-hydroxy-3-[(2-hydroxy-5-methylphenyl)carbonyl]-1,2-dihydropyridin-2-yl}phosphonate (**22B**, *enol form*) (Scheme 8). The mechanism pathway for formation of product **22** is similar to formation of compound **21**, but the cyclization process took place *via* addition of NH group on the ester and amide groups to lose ethanol and ammonia molecules, respectively (Scheme 8).

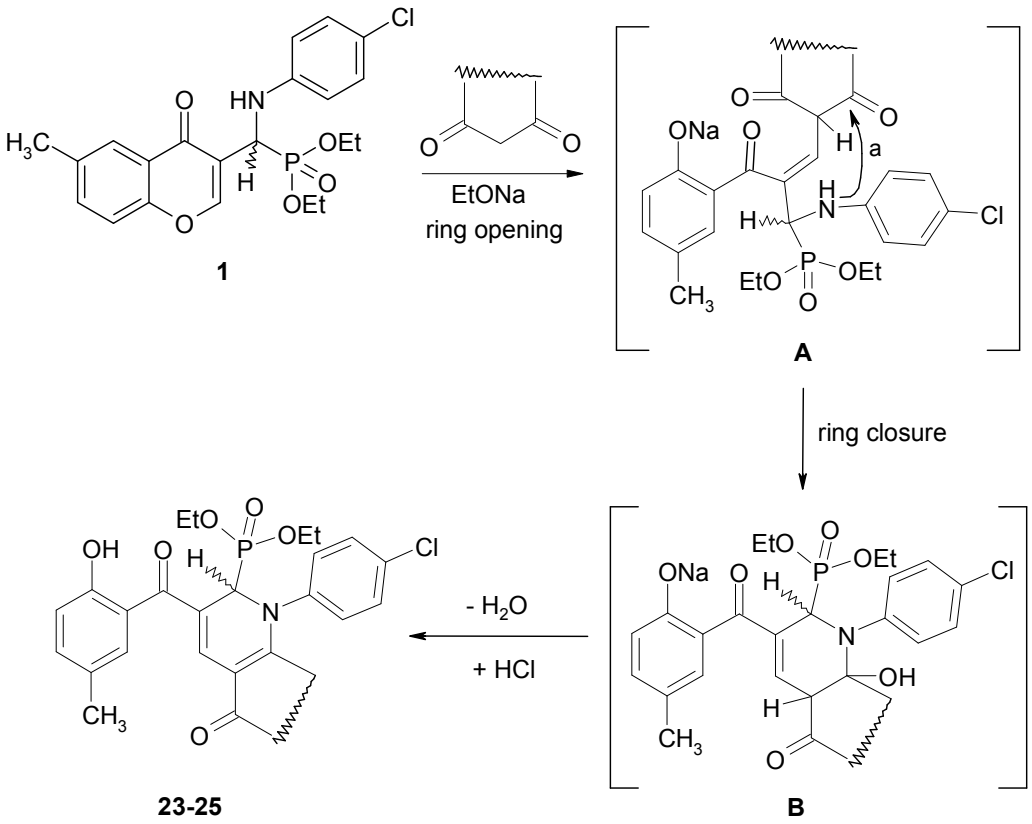
In contribution of this work, the present study has been focused on synthesis of interesting novel fused pyridine systems bearing phosphonate groups. Thus, diethyl

chromonyl α-aminophosphonate **1** reacted with active cyclic methylene compounds such as dimedone, 1-phenylpyrazolidin-3,5-dione and barbituric acid in ethanolic sodium ethoxide to afford the corresponding α-pyridinyl phosphonates **23**−**25**, respectively (Scheme 9).



Scheme 9

According to Scheme 10, the first step of formation of compounds **23**−**25** is a nucleophilic attack of the carbanion species of cyclic methylene compounds at C−2 of chromone ring affording the intermediate **A**. This intermediate underwent intramolecular cyclization *via* a nucleophilic attack of NH group on C=O group of methylene species under elimination of a molecule of water.



Scheme 10

BIOLOGICAL EVALUATION

Antimicrobial activity

All the newly synthesized compounds were evaluated in *vitro* for their antibacterial activities against *Staphylococcus aureus* (ATCC 25923) and *Bacillus subtilis* (ATCC 6635), as representatives of Gram–positive bacteria and *Escherichia coli* (ATCC 25922) and *Salmonella typhimurium* (ATCC 14028) as examples of Gram–negative bacteria. They were also examined against *Candida albicans* (ATCC 10231) as yeast and *Aspergillus fumigatus* as fungus. Agar-diffusion technique was used for the determination of the preliminary antibacterial and antifungal activities.[31,32] The minimum inhibitory concentration (MIC, μg/ml) for the most active compounds against the same microorganism used in the preliminary screening, was carried out using the tube dilution technique.[33] The obtained results on the antimicrobial activities of the compounds and control drugs are given in Table 1. In general, the prepared compounds recorded variable antimicrobial activities towards

the used microorganisms. The most compounds did not record any noticeable inhibitory effects towards Gram-negative bacteria. Similarly, only compounds **5**, **11**, **17**, **20**, **23** and **24** exhibited moderate activities against Gram-positive bacteria, especially compounds **17**, **23** and **24** that recorded their MIC value at 250 μg/ml. All the compounds except **10**, **12**, **15** and **18** exhibited relatively moderate to high

inhibitory activities against *Candida albicans*. Furthermore, the compounds **3**, **5**, **17**, **20**, **23-25** recorded remarkable inhibitory effects against *Aspergillus fumigatus*. The MIC values of the latter compounds against *Candida albicans* and *Aspergillus fumigatus* were at 62.5–125 μg/ml. From the above results, it is clear that connection of pyrazole, oxazepine and benzodiazepine rings with α-aminophosphonate moiety can exhibit good antimicrobial effects. Also, the presence of α-pyridinyl phosphonate (as cyclic α-aminophosphonate) fused with other heterocycles may enhance the antimicrobial properties. These results may help the chemists to make any structural modifications to improve the antimicrobial activities for these compounds.

Antioxidant activity

All the synthesized compounds were tested for antioxidant property by DPPH method.[34,35] The observed data on the antioxidant activities of the compounds and control are shown in Table 2 and illustrated in Figure 1. The results of scavenging the stable DPPH radical recorded variable antioxidant activities towards the synthesized compounds at the different concentrations 150, 300 and 450 μmol L^{-1}. The compounds **10**, **13** and **25** showed moderate antioxidant activities. In the meantime, the compounds **11**, **12**, **15**–**18** and **21**–**24** displayed good antioxidant activities. On the other hand, the compounds **3**, **5**, **14**, **19** and **20** proved to exhibit potent antioxidative properties. The structure-activity relationships of the tested compounds demonstrated that all the synthesized compounds recorded remarkable inhibition activities in range 48–72% at the different concentrations due to the presence of 4-methylphenol group in all the compounds except compound **10** which has another OH group in α-amino-phosphonic monoester moiety. The presence of acyclic α-aminophosphonic diester moiety in the synthesized compounds **5**, **10**–**20** enhanced the antioxidative properties more than the cyclic α-aminophosphonic diester moiety in compounds **21**–**25**. This may due to the presence of free NH groups which can scavenge the DPPH radical. The appearance of pyrazole units in compound **11** and **12** exhibited greater activities

than those having isoxazole unit in compound **13**. Similarly, the thioxopyrimidinyl α-aminophosphonate **14** was more slightly active than the other amino/cyano-iminopyrmidinyl α-aminophosphonates **15** and **16**. Amongst compounds having seven-membered rings **17**–**20**, the benzo derivatives **19** and **20** exhibited higher inhibition activities when compared with the non-benzo derivatives **17** and **18**. On the other hand, the pyridine systems **21**–**25** recorded similar antioxidative properties due to the presence of NH groups. In this study, the systems **3**, **5**, **19** and **20** displayed the higher scavenging activities. However, the result exemplified that the compounds **19** and **20** having benzoxazepinyl and benzodiazepinyl units in combination with α-aminophosphonic diester moiety are the most powerful antioxidant agents.

Table 1: In *vitro* antimicrobial activities of the synthesized compounds **3-25** at 500 and 1000 μg/ml and the MIC values for some selected compounds.

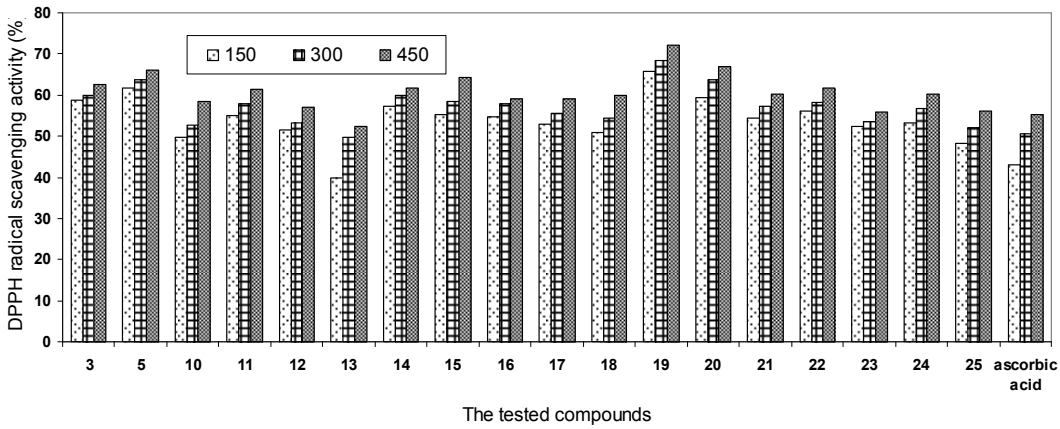
Compd.	Conc. (μg/ml)	Zone of inhibition in mm* and (MIC values in μg/ml)					
		Bacteria Gram (+) ve		Bacteria Gram (−) ve		Yeast	Fungi
		S. aureus	*B. subtilis*	*S. typhimurium*	*E. coli*	*C. albicans*	*A. fumigatus*
3	500	-	7	-	-	19 (62.5)	17 (125)
	1000	-	7	-	-	25	24
5	500	15 (> 250)	16 (> 250)	-	-	19 (62.5)	19 (125)
	1000	19	20	-	-	26	22
10	500	-	-	-	-	7	-
	1000	-	-	-	-	8	-
11	500	7 (> 250)	16 (250)	8	8	13 (250)	7
	1000	12	18	11	12	18	10
12	500	-	-	-	-	8	-
	1000	-	-	-	-	10	-
13	500	-	-	-	-	11	-
	1000	-	-	-	-	15	-
14	500	-	-	-	-	13	-
	1000	-	-	-	-	16	-
15	500	-	-	-	-	8	-
	1000	-	-	-	-	11	-
16	500	-	7	-	-	15	-
	1000	-	7	-	-	21	-
17	500	14 (250)	14 (250)	-	-	20 (125)	20 (62.5)
	1000	17	18	-	-	23	30
18	500	-	8	8	7	7	-
	1000	-	11	11	8	8	-
19	500	-	-	-	-	17	-
	1000	-	-	-	-	20	-
20	500	12 (> 250)	14 (250)	-	-	26 (62.5)	17 (125)
	1000	16	18	-	-	30	26
21	500	-	-	-	-	12	-
	1000	-	-	-	-	16	-
22	500	-	-	-	-	13	-
	1000	-	-	-	-	16	-
23	500	15 (250)	13 (250)	7	7	18 (125)	17 (62.5)
	1000	18	19	11	8	21	24
24	500	15 (250)	16 (250)	8	7	17 (62.5)	18 (62.5)
	1000	19	21	10	8	26	22
25	500	6	8	7	9	15 (250)	11 (> 250)
	1000	12	9	8	12	18	19
Standard drug	500	26	25	28	27	28	26
	1000	35	35	36	38	35	37

* Low active: 6–12 mm; moderately active: 13–19 mm; highly active: 20–30 mm; –: No inhibition or inhibition less than 5 mm.

Table 2: The DPPH radical scavenging activities of the synthesized compounds **3-25** at 150, 300 and 450 μmol L^{-1}.

Compd. No.	DPPH % inhibition antioxidant ± SD		
	150 μmol L^{-1}	**300 μmol L^{-1}**	**450 μmol L^{-1}**
3	58.67 ± 0.06	59.86 ± 0.06	62.56 ± 0.18
5	61.75 ± 0.12	63.65 ± 0.18	66.01 ± 0.12
10	49.89 ± 0.06	52.72 ± 0.24	58.34 ± 0.18
11	54.90 ± 0.12	57.85 ± 0.13	61.49 ± 0.13
12	51.47 ± 0.38	53.29 ± 0.06	56.94 ± 0.06
13	39.75 ± 0.12	49.85 ± 0.18	52.28 ± 0.06
14	57.21± 0.18	59.98± 0.12	61.54 ± 0.18
15	55.23 ± 0.24	58.45 ± 0.12	64.28 ± 0.25
16	54.73 ± 0.06	57.90 ± 0.38	59.07 ± 0.38
17	53.09 ± 0.12	55.59 ± 0.18	59.07 ± 0.06
18	51.03 ± 0.18	54.34 ± 0.13	59.85 ± 0.18
19	65.64 ± 0.18	68.45 ± 0.18	72.12 ± 0.13
20	59.23 ± 0.18	63.78 ± 0.06	66.85 ± 0.24
21	54.34 ± 0.18	57.25 ± 0.06	60.28 ± 0.13
22	56.16 ± 0.06	58.06 ± 0.24	61.53 ± 0.13
23	52.45 ± 0.12	53.65 ± 0.13	55.74 ± 0.06
24	53.29 ± 0.06	56.68 ± 0.06	60.20 ± 0.06
25	48.36 ± 0.12	52.16 ± 0.30	56.04 ± 0.18
Ascorbic acid	43.00	50.70	55.20

Figure 1: The DPPH radical scavenging activities (%) of the synthesized compounds **3-25** at 150, 300 and 450 μmol L^{-1}.



EXPERIMENTAL

The melting point was determined in an open capillary tube on a digital Stuart SMP-3 apparatus. Infrared spectra were measured on FT-IR (Nicolet IS10) spectrophotometer using KBr disks. ^{1}H-NMR spectra were measured on Gemini-300BB spectrometer (300 MHz), using DMSO-d_6 as a solvent and TMS (δ) as an internal standard. ^{13}C-NMR spectra were measured on Mercury-300BB (75 MHz), using DMSO-d_6 as a solvent and TMS (δ) as an internal standard. ^{31}P-NMR spectra were registered on a Bruker (242 MHz) spectrometer at room temperature using DMSO-d_6 as a solvent and TMS as an internal standard and 85% H_3PO_4 as external reference. Mass spectra were recorded on a Gas Chromatographic GCMSqp 1000 ex Shimadzu instrument at 70 ev. Elemental microanalyses were performed Perkin-Elmer 2400II at the Chemical War department, Ministry of Defense. The purity of the synthesized compounds was checked by thin layer chromatography (TLC) and elemental microanalyses.

Synthesis of diethyl [(4-chlorophenylamino)(6-methyl-4-oxo-4*H*-chromen-3-yl) methyl]phosphonate (1)

A mixture of 6-methyl-3-formylchromone (5 mmol, 0.94 g), 4-chloroaniline (5 mmol, 0.64 g) and diethyl phosphite (10 mmol, 1.38 ml) was heated under reflux at 70−80 ^{o}C for 6 h. The reaction mixture was cooled then poured into ice and left for complete precipitation. The precipitate formed was filtered off, dried and crystallized from ethanol to give pale yellow crystals. Yield 70%. M.p.: 196–198 ^{o}C,[20] (Lit.[21] 199–201 ^{o}C).

General procedure for the preparation of target compounds 3-25

A mixture of diethyl [(4-chlorophenylamino)(6-methyl-4-oxo-4*H*-chromen-3-yl)methyl]phosphonate (**1**) (2.30 mmol, 1 g) and nucleophile (2.30 mmol) in ethanolic sodium ethoxide solution (4.35 mmol, 0.10 g of sodium metal in 20 ml of absolute ethanol) was refluxed for 6−10 hours. The reaction mixture was cooled then poured into ice, neutralized with diluted hydrochloric acid (5%) and left for complete

precipitation. The precipitate formed was filtered off, dried and crystallized from the proper solvent.

BIOLOGICAL ASSAY

Antimicrobial evaluation

All the newly synthesized compounds were evaluated in *vitro* for their antibacterial activities against *Staphylococcus aureus* (ATCC 25923) and *Bacillus subtilis* (ATCC 6635), as representatives of Gram–positive bacteria and *Escherichia coli* (ATCC 25922) and *Salmonella typhimurium* (ATCC 14028) as examples of Gram–negative bacteria. They were also examined against *Candida albicans* (ATCC 10231) as yeast and *Aspergillus fumigatus* as fungus. Agar-diffusion technique was used for the determination of the preliminary antibacterial and antifungal activities. The test was performed on medium potato dextrose agar (PDA) which contained infusion of 200 g potatoes, 6 g dextrose and 15 g agar. Uniform size filter paper disks (3 disks per compound) were impregnated by equal volume (10 µl) from the concentrations of 500 and 1000 µg/ml dissolved compounds in dimethylformamide (DMF) and carefully placed on inoculated agar surface. After incubation for 36 h at 27 °C in the case of bacteria and for 48 h at 24°C in the case of fungi, the antimicrobial activities were determined by measuring the inhibition zones. Cephalothin, Chloramphenicol and Cycloheximide were used as reference drugs (30 µg/ml) for Gram–positive bacteria, Gram–negative bacteria and fungi, respectively. The minimum inhibitory concentration (MIC, µg/ml) for some selected compounds against some species of microbes was also determined. The tube dilution technique was applied for the determination of MIC of the tested compounds against microbes. Dilution series were set up with 250, 125, 62.5……3.25 µg/ml of nutrient broth medium to each tube, 100 ml of standardized suspension of the test microbes (107 cell/ml) were added and incubated at 37 ^{o}C for 24 h.

Antioxidant activity

The nitrogen centered stable free radical 1,1-diphenyl-2-picrylhydrazyl (DPPH) has often been used to characterize antioxidants. It is reversibly reduced and the odd electron in the DPPH free radical gives a strong absorption maximum at λ

517 nm, which is purple in color. This property makes it suitable for spectrophotometric studies. A radical scavenging antioxidant reacts with DPPH stable free radical and converts into 1,1-diphenyl-2-picrylhydrazine. The resulting decolorization is stoichiometric with respect to the number of electrons captured. The change in the absorbance produced in this reaction has been used to measure antioxidant properties. The solutions of tested compounds (150, 300 and 450 μmol L^{-1}) were added to DPPH (100 μmol L^{-1}) in DMSO/ethanol. The tubes were kept at an ambient temperature for 20 minutes and the absorbance was measured at λ 517 nm. The difference between the test and the control experiments was taken and expressed as the percent scavenging of the DPPH radical using the following formula % inhibition = ($AB-AA/AB$) x 100 where AB = absorption of blank and AA = Absorption of the tested compound. The radical scavenging activity of ascorbic acid was also measured and compared with that of the different synthesized compounds.

CONCLUSION

A facile and convenient synthetic method has been described to attain novel substituted azoles, azines and azepines linked to α-aminophosphonate and so α-pyridinyl phosphonates. Compared to the previously reported methodologies, the present article offers procedure for synthesis of parent heterocyclic α-amino-phosphonate and phosphonates which are an inapplicability of known *via* the regular procedures. The methodology depends on ring opening and ring closure of chromone ring *via* different nitrogen and carbon nucleophiles. The screening of antimicrobial activity for the synthesized compounds indicates that connection of pyrazole, oxazepine and benzodiazepine rings with α-aminophosphonate moiety exhibited good antimicrobial effects. Also, evaluation of their antioxidant properties exemplifies that the compounds having 1,5-benzoxazepinyl and 1,5-benzodiazepinyl units in combination with α-aminophosphonic diester moiety are the most powerful antioxidant agents.

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