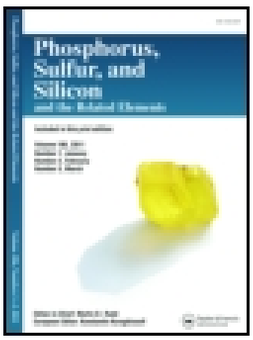

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New class of diethyl substituted phosphoramidimidates and phosphonimidates: synthesis, spectral characterization and antimicrobial activity

Mavallur Varalakshmi[a], Chamarthi Nagaraju[b], and Palaa Krishna[c]

[a]Basic Sciences & Humanities, Centre for Applied Sciences, Sree Vidyanikethan Engineering College, Tirupati, Andhra Pradesh, India; [b]Department of Chemistry, Sri Venkateswara University, Tirupat, Andhra Pradesh, India; [c]Department of Chemistry, Geethanjali Institute of Science and Technology, Gangavaram, Nellore, India

ABSTRACT

A series of new class of diethyl *N*-2-hydroxyethyl-*N*′-substituted phosphoramidimidates **6(a–e)** and diethyl *P*-morpholino-*N*-substituted phosphonimidates **6(f–j)** was synthesized. The precursor intermediates, diethyl substituted phosphoramidites **3(a–b)** were prepared initially by a reaction of various amines **1(a–b)** and diethyl phosphorochloridite **(2)** and then they were treated by *in situ* with aromatic/alkyl azides through Staudinger reaction to accomplish title products. Structures of all the synthesized compounds were characterized by spectroscopic data such as IR, NMR (^{1}H, 13C, ^{31}P), mass, and elemental analyses. The synthesized compounds were screened for their *in vitro* antimicrobial activity to understand their biological potency. The biological screening results disclosed that compounds **6b, 6c, 6e, 6g, 6h and 6j** having potent antimicrobial activity against all the tested pathogens.

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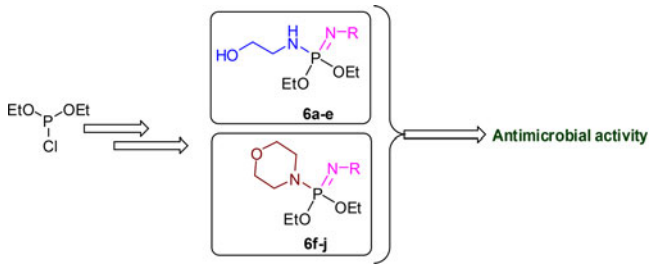
Iminophosphoranes; diethyl phosphorochloridite; Staudinger reaction; antimicrobial activity

GRAPHICAL ABSTRACT



Introduction

Phosphorus compounds includes trivalent and pentavalent states bonded with nitrogen species have become most attractive molecules in diverse fields.[1] They have been used as catalysts in numerous organic transformations[2] and ligands in asymmetric synthesis to achieve high enantioselectivity[3–5] as well as inhibitors for the treatment of various diseases.[6] Particularly, phosphoramidites belong to a family of amides of trivalent phosphorous acid, distinct from other trivalent phosphorous compounds, as they possess one P-N and two P-O bonds. These phosphoramidites are important intermediates in the synthesis of numerous libraries of organic molecules and can be acted as ligands in asymmetric synthesis because of their easy accessibility and rapid tuning of properties with the variation of substituents at the nitrogen atoms.[7]

Iminophosphoranes ($R_3P{=}NR$) are a class of organic compounds, also known as phosphazenes, phosphoranimines, or phosphine imides, where phosphorus atom is linked to a nitrogen atom by a double bond and to three other atoms or groups by single bonds.[8] These derivatives are useful in many applications like building blocks of polymers,[9,10] key components in organic synthesis,[11,12] super-bases (e.g. Schwesinger base) with nonmetallic, nonionic and low nucleophilic characteristics[13,14] and serves as key intermediates in the synthesis of various biologically active compounds[15] aplysinopsin,[16] leucettamine B,[17] Fascaplysin.[18] Staudinger reaction is the most widely employed protocol for the synthesis of iminophosphoranes. This reaction involves the right components of Staudinger reaction such as trivalent phosphorus and organic azide species leading to the formation of P=N linkages and it can be used to enables the straightforward synthesis of various phosphorous nitrogen compounds.[19] To the best of literature knowledge, there is no reports on the investigation of biological activity of phosphoramimidate derivatives.

CONTACT Mavallur Varalakshmi varalakshmi.mavallur@gmail.com
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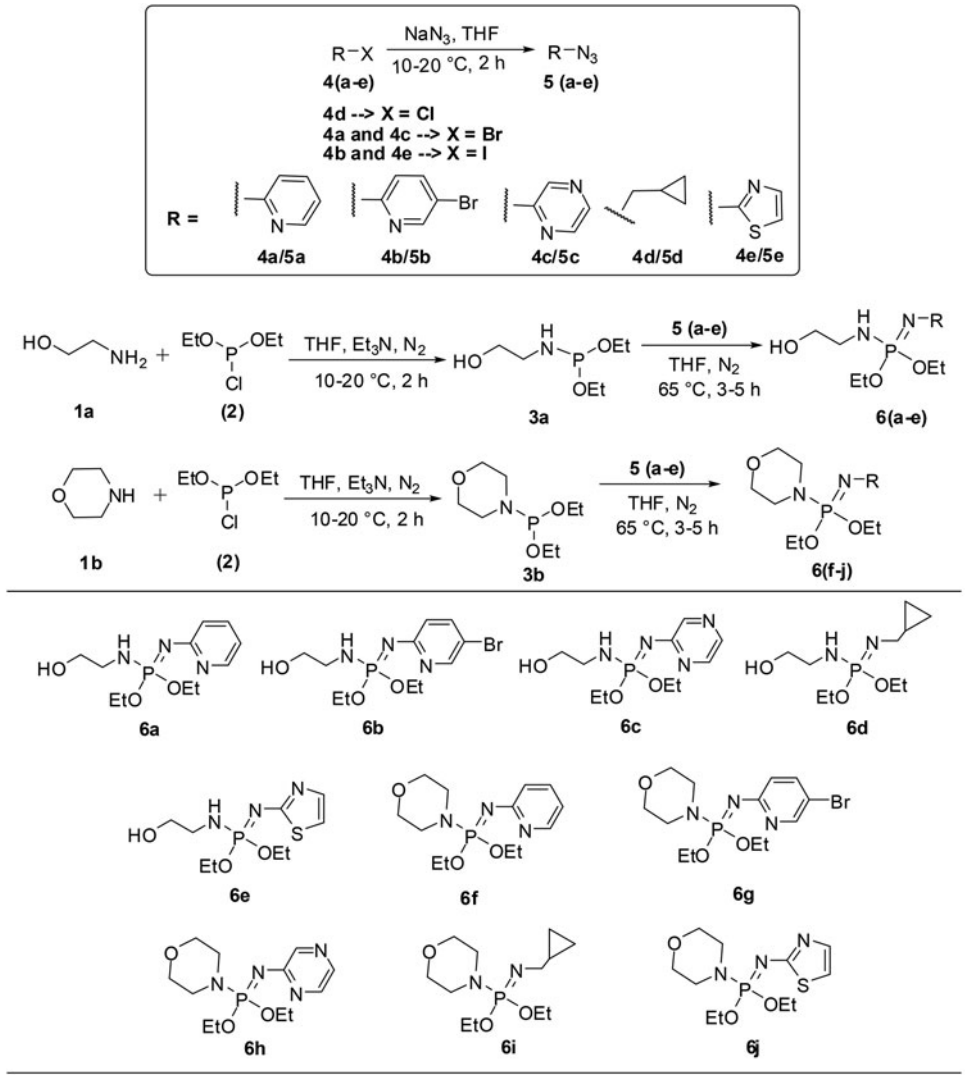


Scheme 1. Synthesis of diethyl substituted phosphoramidimidates and phosphonimidates **6(a–j)**.

Considering the overview facts, we herein report the synthesis of new class of diethyl substituted phosphoramidimidate/phosphonimidate derivatives through Staudinger reaction using substituted phosphoramidites and heterocyclic/alkyl azides. The antimicrobial activity was evaluated to understand their biological potency.

Results and discussion

The synthesis of new iminophosphorane derivatives, diethyl *N*-2-hydroxyethyl-*N*'-substituted phosphoramidimidates **6(a–e)** and diethyl *P*-morpholino-*N*-substituted phosphonimidates **6(f–j)** was depicted in Scheme 1. All the title compounds were purified by column chromatography and stored in nitrogen atmospheric conditions.

The structures of all the newly synthesized compounds **6(a–j)** were characterized by IR, NMR (^{1}H, 13C & ^{31}P), mass spectroscopy and CHN analyses and the details provided in the experimental section and supporting information. In IR spectrum of compound **6c,** the absorption bands appeared in the regions of 3472, 3267 and 1342 cm^{-1}corresponding to the functional groups, –OH, -NH and P = N, respectively. In ^{1}H NMR spectra, the chemical shifts in the regions of δ 1.39–1.42 ppm as a triplet and δ 4.13–4.18 ppm as a quartet confirmed CH_3 (H-16 and H-18) and $-OCH_2$ (H-15 and H-17) protons present in ethoxy groups, respectively. The –OH (H-1) and –NH (H-4) protons resonated as singlets at δ 4.76 and δ 3.73 ppm respectively. The remaining aliphatic protons appeared in the regions of δ 2.05–3.17 ppm (H-2 and H-3). In 13C NMR spectra, the chemical shift values at δ 65.5 and δ 154.5 ppm are corresponding to CH_2OH (C_{15} and C_{17}) and –P=N–C (C_9) carbons, respectively. The remaining aliphatic carbons were observed in the range of δ 19.3–51.2 ppm (C_2, C_3, C_{16} and C_{18}). The ^{31}P NMR signal of compound **6c** was observed as a singlet at δ 12.6 ppm.

Biological activity

In vitro antimicrobial activity of the newly synthesized phosphoramidimidate/phosphonimidate derivatives **6(a–j)** was screened to understand their biological potency. The bacterial strains such as *Staphylococcus aureus (S. aureus)*, *Lactobacillus acidophillus* (*L. acidophillus*) (Gram positive)

and *Escherichia coli* (*E. coli*) and *Vibrio cholerae* (*V. cholerae*) (Gram negative), and four fungal strains like *Trichoderma longibrachiatum* (*T. longibrachiatum*), *Clostridium tetani* (*C. tetani*), *Aspergillus niger* (*A. niger*) and *Aspergillus fumigates* (*A. fumigates*) were selected to investigate the activity. The agar diffusion method and poison plate technique were used to screen the antibacterial and antifungal activities, respectively. Tetracycline in antibacterial activity and Fluconazole in antifungal activity were used as the standards. The bacterial/fungal growths of zone of inhibition exhibited by the title compounds at the concentration of 200 µg/mL were measured in millimeters. The experiments were conducted in duplicate and average value was taken as final result. The results of antibacterial and antifungal activities were summarized in **Table S1** and **Table S2**, respectively **(Supplemental Materials)**.

As can be seen in **Table S1** and **Table S2**, all the compounds showed moderate to potential antimicrobial activity at the concentration of 200 µg/mL except compound **6f** and **6i** against fungal strains. Whereas, 2-hydroxy ethylamine substituted phosphoramidimidate derivatives **6b** linked with 4-bromo-2-pyridine, **6c** bonded with cyclopropyl methyl and **6e** bearing thiazol-2-yl, and morpholine substituted phosphonimidate derivatives **6g** linked with 4-bromo-2-pyridine, **6h** bonded with cyclopropyl methyl and **6j** bearing thiazol-2-yl showed potential activity which are almost closer activity to the standards. The substituents, 4-bromo-2-pyridine, cyclopropyl methyl and thiazol-2-yl on the title compounds could be reason to exhibited potential activity. Future research by the synthesis of library compounds through altering different substituted amines and organic azides followed by screening of biological activity is required to understand clearly the structural-activity relationship.

Experimental

The analytical grade chemicals and GC-grade solvents used in the reactions were purchased from Sigma Aldrich and S. D. fine chem (India) and used without further purification. The progress of reactions was monitored by TLC on Merck silica plates. All the reactions were carried out under nitrogen atmospheric conditions. Neutral alumina was used as a solid phase and moisture free solvents ethyl acetate and *n*-hexane was used as eluents in the column chromatography to purify the title compounds. Melting points were determined in open capillaries on Guna melting point apparatus and are uncorrected. IR spectra were recorded on JASCO FT-IR 5300 using KBr discs. Nuclear magnetic resonance (NMR) spectroscopic data were recorded on Bruker AV-400 spectrometer. Tetramethylsilane (TMS) and 85% H_3PO_4 were used as an internal and external standards in the recording of ^{1}H NMR, 13C NMR and ^{31}P NMR spectra, respectively. CHN analysis was recorded on Thermo Finnigan Flash 1112 instrument. Results are presented as, chemical shift δ in ppm, multiplicity, J values in Hertz (Hz). The Supplemental Materials contains complete characterization data for products 6, and select ^{1}H, ^{13}C and ^{31}P NMR spectra for the products (Figures S1–S10).

Procedure for the synthesis of 2-azidopyrazine 5c

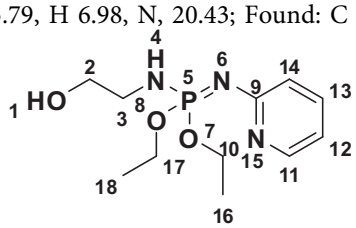
The mixture of 2-chloropyrazine **(4c)** (235.42 mg, 1.5 mmol) and sodium azide (9 mg, 1.5 mmol) in tetrahydrofuran (THF) (10 mL) was stirred for 2 h at 40–45 °C. After completion of the reaction as checked by TLC, the reaction mass was filter at ambient temperature to remove sodium chloride as residue and then the filtrate was concentrated under vacuum at 45 °C to afford 2-azidopyrazine **(5c)**. It was used directly without purification in the Staudinger reaction. The same procedure was adopted to prepare remaining organic azide derivatives used in the present study.

Procedure for the synthesis of compound 6c

Diethyl phosphorochloridite (**2**) (0.21 mL, 1.5 mmol) solution in THF (5 mL) was added slowly in drop wise to the cold solution of ethanolamine (**1a**) (0.09 mL, 1.5 mmol) and triethylamine (TEA) (1.5 mmol) in THF (10 mL) at 0–5 °C for 45 min under nitrogen atmospheric conditions. The organics were stirred for 2.0 h at 15–20 °C to give precursor intermediate, diethyl 2-hydroxyethylphosphoramidite (**3a**). The reaction completion was judged by TLC. The solution of 2-azidopyrazine **(5c)** (1.5 mmol) in THF (5 mL) was added to the above reaction mass at 15–20 °C for 30 min. The resulting organics were agitated for 5.0 h at 60-65 °C and TLC confirmed the completion of reaction. The reaction mass was filtered under nitrogen atmospheric conditions to remove the salt, triethyl ammonium hydrochloride. The filtrate was concentrated under vacuum to obtain the crude compound. It was subjected to column chromatography using ethyl acetate and n-hexane (3:7) as a mobile phase to obtain diethyl *N*-2-hydroxyethyl-*N*'-(pyrazin-2-yl)phosphoramidimidate (**6c**). The same experimental procedure was followed for the synthesis of remaining title compounds **6a, 6b & 6(d–j)**.

Diethyl N-2-hydroxyethyl-N'-(pyrazin-2-yl)phosphoramidimidate (6c)

Semi-solid, Yield:78%; IR (KBr, cm^{-1}): 1342 (-P=N, str), 3267 (-P-NH, str), 3472 (-OH, str); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 1.39 (t, J = 7.2 Hz, 6H, H-16, H-18), 2.05-2.27 (q, 2H, H-3), 3.15-3.17 (q, 2H, H-2), 3.73 (s, 1H, H-4), 4.76 (s, 1H, H-1), 4.13-4.18 (q, 4H, H-15, H-17), 7.79 (s, 1H, H-10), 8.10 (d, J = 8.8 Hz, 1H, H-12), 8.47 (d, J = 5.2 Hz, 1H, H-13); 13C NMR ($CDCl_3$, 100.6 MHz): δ 19.3 (C_{16}, C_{18}), 39.9 (C_3), 51.2 (C_{15}, C_{17}), 65.5 (C_2), 140.8 (C_{12}), 147.2 (C_{13}), 150.5 (C_{10}), 154.5 (C_9). ^{31}P NMR ($CDCl_3$, 161.9 MHz): δ 12.6. MS (m/z): 273 (M-H$^+$). Anal.Calcd.for$C_{10}H_{19}N_4O_3P$: C 43.79, H 6.98, N, 20.43; Found: C 43.82, H 6.87, N 20.45.



Biological activity

The antibacterial activity of the newly synthesized compounds **6(a–j)** was carried out using agar diffusion

method[20] against the bacterial strains, *Staphylococcus aureus*, *Lactobacillus acidophillus* (Gram positive) and *Escherichia coli* and *Vibrio cholerae* (Gram negative). The results are presented in **Table S1**. The antifungal activity was screened using poison plate technique[21] against fungal strains such as *Trichoderma longibrachiatum, Clostridium tetani, Aspergillus niger* and *Aspergillus fumigates* and the results summarized in **Table S2**. The detailed procedure has been provided in the supporting information.

Conclusion

We have synthesized a new series of iminophosphorane derivatives, diethyl *N*-2-hydroxyethyl-*N*'-substituted phosphoramidimidates **6(a–e)** and diethyl *P*-morpholino-*N*-substituted phosphonimidates **6(f–j)** through Staudinger reaction. The newly synthesized title compounds were investigated their antimicrobial activity to understand their biological potency. The biological activity results disclosed that most of the compounds exhibited potent to moderate antimicrobial activity. Whereas, the compounds **6b, 6c**, **6e, 6g, 6h** and **6j** possessing 4-bromo-2-pyridine, cyclopropyl methyl and thiazol-2-yl as substituents showed potent antibacterial and antifungal activities. In addition, we are unable to understand unambiguously the structural-activity relationship with the presented results; therefore further comprehensive research is to be warranted.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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References

[1] Greenwood, N.; Earnshaw, A. *Chemistry of the Elements*. Oxford: Pergamon, **1984**.

[2] Rueping, M.; Nachtsheim, B. J.; Ieawsuwan, W.; Atodiresei, I. Modulating the Acidity: Highly Acidic Bronsted Acids in Asymmetric Catalysis. *Angew. Chem. Int. Ed.* **2011**, *50*, 6706–6720. DOI: 10.1002/anie.201100169.

[3] Szulc, I.; Kołodziuk, R.; Kryczka, B.; Zawisza, A. New Phosphine-Imine Ligands Derived from *d*-Gluco- and *d*-Galactose in Pd-Catalysed Asymmetric Allylic Alkylation. *Tetrahedron Lett.* **2015**, *56*, 4740–4743. DOI: 10.1016/j.tetlet.2015.06.031.

[4] (a) Knowles, W. S.; Sabacky, M. J.; Vineyard, B. D. Catalytic Asymmetric Hydrogenation. *Chem. Commun.* **1972**, *0*, 10–11. DOI: 10.1039/C39720000010. (b) Lega, M.; Margalef, J.; Ruffo, F.; Pamies, O.; Dieguez, M. Application of pyranoside phosphite-pyridine ligands to enantioselective metal-catalyzed allylic substitutions and conjugate 1,4-additions. *Tetrahedron: Asymmetry.* **2013**, *24*, 995–1000. DOI: 10.1016/j.tetasy.1013.06.011.

[5] Boiteau, J. G.; Imbos, R.; Minnard, A. J.; Feringa, B. L. Rhodium-Catalyzed Asymmetric Conjugate Additions of Boronic Acids Using Monodentate Phosphoramidite Ligands. *Org. Lett.* **2003**, *5*, 681–684. DOI: 10.1021/ol027465.

[6] Mucha, A.; Kunert, A.; Grembecka, J.; Pawełczak, M.; Kafarski, P. A Phosphonamidate Containing Aromatic *N*-Terminal Amino Group as Inhibitor of Leucine Aminopeptidase-Design, Synthesis and Stability. *Eur. J. Med. Chem.* **2006**, *41*, 768–772. DOI: 10.1016/j.ejmech.2006.03.023.

[7] Madhurima, H.; Kyalo, S. K.; Takanori, S. Enantioselective Synthesis of Planar-Chiral 1,11-Dioxa[11]Paracyclophane-Derived Phosphoramidities and Their Use as Chiral Ligands. *Tetrahedron: Asymmetry*. **2016**, *27*, 1081–1087. DOI: 10.1016/j.tetasy.2016.08.015.

[8] Fang, J.-K.; Xu, Z.; Sun, T.; Fang, Y.; Yin, Z.; Wang, S. Synthesis and Spectral Properties of Novel Iminophosphoranes. *Phosphorus Sulfur Silicon*. **2016**, *191*, 1229–1234. DOI: 10.1080/10426507.2016.1165677.

[9] Baumgartner, T.; Reau, R. Organophosphorus π-Conjugated Materials. *Chem. Rev.* **2006**, *106*, 4681–4727. DOI: 10.1021/cr040179m.

[10] Blackstone, V.; Lough, A. J.; Murray, M.; Manners, I. Probing the Mechanism of the PCl_5—Initiated Living Cationic Polymerization of the Phosphoranimine $Cl_3P{=}NSiMe_3$ Using Model Compound Chemistry. *J. Am. Chem. Soc.* **2009**, *131*, 3658–3667. DOI: 10.1021/ja808517d.

[11] Foster, R. S.; Jakobi, H.; Harrity, J. P. A General and Regioselective Synthesis of 5-Trifluoromethyl-Pyrazoles. *Org. Lett.* **2012**, *14*, 4858–4861. DOI: 10.1021/ol3021918.

[12] Monnereau, L.; Sémeril, D.; Matt, D. Calixarene-Derived Mono-Iminophosphoranes: Highly Efficient Ligands for Palladium- and Nickel-Catalysed Cross-Coupling. *Adv. Synth. Catal.* **2013**, *355*, 1351–1360. DOI: 10.1002/adsc.201300091.

[13] Kanazawa, C.; Terada, M. Dichotomous Control of *E*/*Z*-Geometry in Intramolecular Cyclization of *o*-Alkynylbenzamide Derivatives Catalyzed by Organic Superbase P4-*t*Bu in the Presence/Absence of Water. *Chem. Asian J.* **2009**, *4*, 1668–1672. DOI: 10.1002/asia.200900342.

[14] Goldys, A. M.; Nunez, M. G.; Dixon, D. J. Creation through Immobilization: A New Family of High Performance Heterogeneous Bifunctional Iminophosphorane (BIMP) Superbase Organocatalysts. *Org. Lett.* **2014**, *16*, 6294–6297. DOI: 10.1021/ol5029942.

[15] Molina, P.; Murcia, F.; Fresneda, P. M. A Straight Forward and Practical Formal Synthesis of Lavendamycin Ethyl Ester. *Tetrahedron Lett* **1994**, *35*, 1453–1456. DOI: 10.1016/S0040-4039(00)76244-7.

[16] Molina, P.; Almendros, P.; Fresneda, P. M. Iminophosphorane-Mediated Imidazole Ring Formation: A New and General Entry to Aplysinopsin-Type Akaloids of Marine Origin. *Tetrahedron*. **1994**, *50*, 2241–2254. DOI: 10.1016/S0040-4039(00)78515-7.

[17] Molina, P.; Almendros, P.; Fresneda, P. M. An Iminophosphorane-Mediated Efficient Synthesis of the Alkaloid Leucettamine B of Marine Origin. *Tetrahedron Lett.* **1994**, *35*, 2235–2236. DOI: 10.1016/S0040-4039(00)76806-7.

[18] Molina, P.; Fresneda, P. M.; García-Zafra, S.; Almendros, P. Iminophosphorane-Mediated Syntheses of the Fascaplysin Alkaloid of Marine Origin and Nitramarine. *Tetrahedron Lett.* **1994**, *35*, 8851–8854. DOI: 10.1016/S0040-4039(00)78515-7.

[19] Staudinger, H.; Meyer, J. Uberneue Organische Phosphorverbindungen III. Phosphinmethylenderivate and Phosphinimine. *Hca.* **1919**, *2*, 635–646. DOI: 10.1002/hlca.19190020164.

[20] Varalakshmi, M.; Naga Raju, C. Nucleoside Substituted Perhydro-1λ5-[1,3,2]Diazaphospholo[1,5-a]Pyridine-1-Thione Analogues: Synthesis and Evaluation of Antiviral and Antimicrobial Activities. *Orgcommun*. **2018**, *11*, 35–45. DOI: 10.25135/.acg.oc.35.17.12.055.

[21] Varalakshmi, M.; Srinivasulu, D.; Venkata Kotakadi, S. Nano-$BF_3.SiO_2$ Catalyst-Promoted Michaelis-Arbuzov Reaction: Solvent-Free Synthesis and Antimicrobial Evaluation. *Phosphorus, Sulfur, Silicon Relat. Elem*. **2015**, *190*, 1518–1524. DOI: 10.1080/10426507.2014.996643.