GSC Biological and Pharmaceutical Sciences
eISSN: 2581-3250 CODEN (USA): GBPSC2
Cross Ref DOI: 10.30574/gscbps
Journal homepage: https://gsconlinepress.com/journals/gscbps/

(RESEARCH ARTICLE)

Check for updates

# Biologically relevant surrogates of coumarins: 2-phenyl H-isophosphinoline 2-oxides with antibacterial activity

Mina Hariri [1,2], Fatemeh Darvish [2], Karen-Pacelye Mengue Me Ndong [1,3], Rachida Babouri [1], Jacques Lebibi [3], Gabin Mwande-Maguene [3], Alexander R Burilov [4], Patricia Licznar-Fajardo [5], Jean-Luc Pirat [1,*], Tahar Ayad [1,*] and David Virieux [1,*]

[1] *ICGM, Univ. Montpellier, ENSCM, CNRS, Montpellier, France.*
[2] *Department of Chemistry, K. N. Toosi University of Technology, P.O. Box, 15875-4416 Tehran, Iran.*
[3] *Université des Sciences et Techniques de Masuku, Franceville, Gabon.*
[4] *Arbuzov Institute of Organic and Physical Chemistry, FRC Kazan Scientific Center of RAS, Arbuzov str. 8, Kazan 420088, Russian Federation.*
[5] *Hydro Sciences Montpellier, Univ Montpellier, CNRS, IRD, CHU Montpellier, Montpellier, France.*

GSC Biological and Pharmaceutical Sciences, 2021, 16(02), 283–296

Publication history: Received on 23 August 2021; revised on 26 August 2021; accepted on 28 August 2021

Article DOI: https://doi.org/10.30574/gscbps.2021.16.2.0252

## Abstract

The present study aims to investigate the *in vitro* antibacterial activities of several isophosphinoline-2-oxides that can be perceived as combined bio isosteres of coumarins and flavonoids. More specifically, antibacterial activity was evaluated against four bacterial strains, including the Gram-negative bacteria Escherichia coli and Pseudomonas aeruginosa and the Gram-positive bacteria Staphylococcus aureus and Enterococcus faecalis by using disk diffusion assay. Notably, isophosphinoline-2-oxide compounds showed promising and highly selective antimicrobial activity against S. aureus

**Keywords:** Isophosphinoline-2-oxides; Phosphacoumarins; Synthesis; Antibacterial activities

## 1. Introduction

With the advent of the antibiotic era, the overuse and inappropriate consumption of antibiotics have driven the rapid emergence of multidrug-resistant pathogens [1]. Among them, Gram-positive bacteria such as methicillin-resistant Staphylococcus aureus (MRSA) have become a major global healthcare problem in the 21st century. In India, for example, MRSA prevalence among invasive S. aureus isolates, increased from 29% in 2009 to 47% in 2014 [2]. In fact, the total deaths involving MRSA are now comparable to those caused by Human Immunodeficiency Virus (HIV). Consequently, barring any last-minute miracle, it is estimated by the year 2050, that at least 10 million people will die annually due to antimicrobial resistance—a death toll higher than all cancers combined [3]. At the same time, there is a reduced interest of the pharmaceutical industry for the development of new effective antibacterial drugs. The consequence is that the pipeline for potential new antibiotics in clinical development partially addresses the problem of critical WHO priority pathogens [4]. Moreover, the new potential molecules are derivatives of existing classes of antibiotics. As an illustration, among the 11 new FDA approved antibiotics from 2017 to 2019, only two represent a new class. Therefore, there is a clear demand for the discovery and development of new antibiotic agents with an alternate mode of action and new chemical structures to re-arming the current arsenal [5].

* Corresponding author: Jean-Luc Pirat, Tahar Ayad and David Virieux
ICGM, Univ. Montpellier, ENSCM, CNRS, Montpellier, France.

Heterocyclic compounds play a determinant role in present-day living organisms and their broad occurrence underline this significance in several fundamental biological pathways [6]. Coumarin derivatives and substituted flavonoids are natural compounds that show numerous therapeutic applications. The coumarin scaffold constitutes several marketed drugs. Most of them are oral anticoagulants and are used for the treatment of thromboembolic diseases [7] while others such as novobiocin were licensed for the treatment of human infections [8]. For instance, some breakthroughs were found for applications in photochemotherapy, antitumor and anti-HIV therapy, as central nervous system (CNS) stimulants, anti-inflammatory, anti-coagulant, dyes and antibacterials [9]. Furthermore, hydroxycoumarins, chain-breaking antioxidants can also prevent free radical injury by scavenging reactive oxygen species (ROS). Coumarin derivatives are extensively used as substrates for detecting enzymatic activity in cells and substituted ones have shown interesting antibacterial activity mostly against Gram-positive bacteria such as novobiocin (Figure 1) [10]. Phosphorus-based antibiotics and more specifically aminophosphonic and aminophosphinic derivatives represent an interesting group of antibacterial agents [11]. In spite of potency on one of the simplest naturally occurring phosphonate, namely Fosfomycin has to date reached the market [12]. It can be noticed that Fosfomycin irreversibly forms a covalent linkage with Cys 115 of the UDP-N-acetylglucosamine enolpyruvyl transferase (MurA).

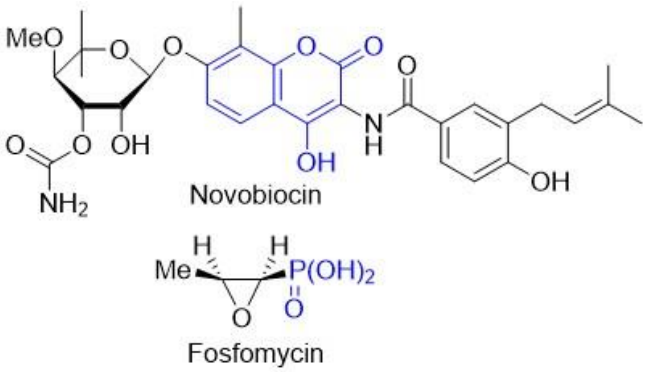



**Figure 1** Antibacterial coumarin derivative Novobiocin and naturally occurring phosphonate Fosfomycin

## 2. Material and methods

### 2.1. Chemistry

All experiments were carried out under nitrogen atmosphere unless otherwise stated. Unless specified, all of the commercially available reagents and starting materials purchased from commercial sources were used as received without further purification. Chromatography: Thin Layer Chromatography (TLC) was performed on precoated plates of silica gel 60 F254 Merck. Visualization was performed with UV light and sometimes with phosphomolybdic acid solution or permanganate solution followed by heating. Flash chromatography was performed manually with silica gel (60 Å, 35–70 µm SDS). $^{1}H$, $^{13}C$ and $^{31}P\{1H\}$ NMR spectroscopic data were recorded at 400, 100 MHz and 162 MHz respectively. The chemical shifts are reported in ppm, and the coupling constants (*J*) are reported in Hz. The chemical shift values are referenced against the residual proton in the deuterated solvents. In the $^{13}C\{1H\}$ NMR spectra, signals corresponding to C, CH, $CH_2$, or $CH_3$ were assigned from the JMOD sequence. The multiplicities are given as s (singlet), d (doublet), t (triplet), q (quadruplet), and m (multiplet). Low- and high-resolution mass spectra were recorded with a time-of-flight mass spectrometer using electrospray ionization (ESI). Melting points were measured with an automatic melting point apparatus SMP50 from Stuart. HRMS (Q-TOF) were performed on a JEOL JMS-DX300 spectrometer (3 keV, xenon) in a *m*-nitrobenzyl alcohol matrix. Microwave reactions (MW) were performed using CEM Discover apparatus in 10- and 35-mL sealed reactors forrespectively small- and large-scale synthesis. Reactions were performed by maintaining the temperature to the set point. For reactions that require heating, the heat source was “heat on” systems.

### 2.2. Biological tests

As recommended, colonies from an overnight culture were picked and suspended in saline solution. Density of micro-organisms was adjusted to 1-2 $10^8$ CFU/mL in saline solution. The suspension was used within 15 min of preparation. A sterile cotton wab dipped into the suspension was used to inoculate Mueller-Hinton agar plates. 20 µL of each preparation of phosphine oxide (10 mg dissolved in 500 µL of DCM, 0.4 mg) was applied on a sterile paper disc (Biomerieux). After evaporation of DCM, disks were aseptically placed on the inoculated plates. Negative control was done with 20 µL of DCM. Then, plates were incubated at 37°C for 18 to 24 h. The inhibition zones were measured in millimeters. The average of inhibition diameters was calculated to classify the phosphine oxide compounds as follows:

- not active (**0**) for a diameter < 6 mm,
- weak activity (**+**) for a diameter ≥ 6 mm and < 8 mm,
- moderate activity (**++**) for a diameter ≥ 8 mm and < 10 mm,
- very active (**+++**) for a diameter ≥ 10 mm.

## 3. Results and discussion

In medicinal chemistry, the bioisosterism concept proved to be a fruitful approach in the design of new drugs with potent activity and reduced risks through modulation of the metabolic and/or the pharmacokinetic properties [13]. In the quest for new biologically relevant substances, there is a significant predominance on non-classic bioisosteric relationships [14]. Among the variety of chemical fragments that have been studied by medicinal chemists over the last decades, the replacement of an atom, or group of atoms of a biologically active compound by a phosphorus-containing functional group has attracted renewed interest. Phosphorus-containing drugs that bear phosphate, phosphoramide, phosphonate group as bioisostere, have had commercial success [15]. As a contribution in phosphorus-based bioisosteres, we recently demonstrated that phosphinolactones can be considered as a potent surrogate of lactol group and can be used as an unprecedented scaffold for the elaboration of new anticancer and neurodegenerative candidate drugs such as phostines [16] and 1,4,2-oxazaphosphinane derivatives [17].

In the same time, the phosphine oxide functional group represents a structural scaffold that is, still underrepresented in today's drug discovery projects and, only few examples can be found amongst approved drugs. More specifically, phosphorus-based heterocycles containing a phosphine oxide functionality represent the poor sibling in the development of new drugs [18]. Taking all these together and in continuation with our research program dedicated to the synthesis of valuable new phosphorus based heterocycles featuring relevant biological properties, we report, herein, the first and promising antibacterial activities of 2-phenyl H-isophosphinoline 2-oxides and their precursors, against four bacterial species, including the Gram-negative bacteria (Escherichia coli and Pseudomonas aeruginosa) and the Gram-positive bacteria (Staphylococcus aureus and Enterococcus faecalis) using disk diffusion assay. From a structural point of view, we assumed that 2-phenyl H-isophosphinoline 2-oxides 4 can be seen as a hydrolytically stable surrogate of phosphacoumarins 3, a combined bioisostere of both coumarin and flavonoid natural compounds, which expressed a wide range of potent activities. Noteworthy, aryl phosphorus esters are known to be biologically reactive species, leading to the facile cleavage of the phosphorus-oxygen bond [19]. It should be emphasized that detailed studies on antibacterial activity of 2-phenyl H-isophosphinoline 2-oxide derivatives have to date not been reported in the literature. In another way, covalent inhibitors received increased attention and a number of functional groups have been now exploited to target specific amino acid residues. To the best of our knowledge, more than 50 approved covalent inhibitors are on the market [20]. We also hypothesized that the vinylphosphine oxide moiety in isophosphinoline **4** may act as a potential covalent inhibitor scaffold. Indeed, electrophilic character of vinylphosphine oxide group was illustrated in literature and it proved to react with a broad range of nucleophiles [21].

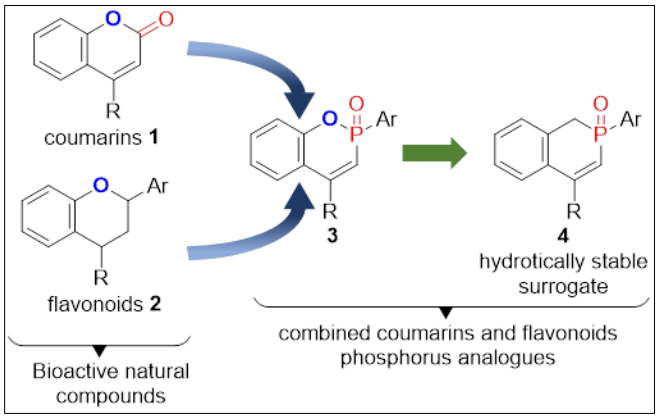
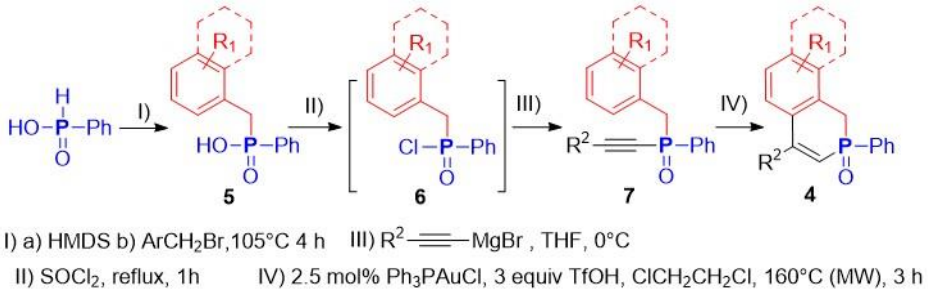



**Figure 2** The logical route to stable surrogates of coumarins

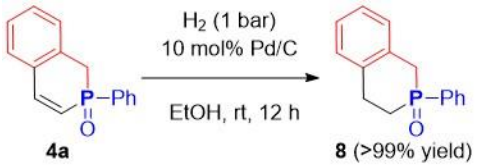
The synthetic route already described in JOC 2021 [22], for the preparation of 2-phenyl H-isophosphinoline 2-oxide derivatives **4**, is depicted in Scheme 1. The first step began with the preparation of diversely substituted phenyl(arylmethyl)phosphinic acids **5** easily obtained in good yields from the reaction of various arylmethyl halides and phenylphosphinic acid using hexamethyldisilazane as coreagent. The second step involved thionyl chloride

mediated-chlorination of acids **5** to afford the corresponding phosphinic chlorides **6** which were directly engaged in an alkynylation reaction with alkynylmagnesium bromides ($R^2$ = H, Me, Ph). Under optimized conditions, a wide range of phenyl(arylmethyl)(alkynyl)phosphine oxides **7** were isolated in good to excellent yields. The key step was an intramolecular hydroarylation reaction mediated by 2.5 mol% of $Ph_3PAuCl$ and 3 equivalents of triflic acid in dichloroethane at 160°C under microwave (MW) during 3 hours. 2-Phenyl H-isophosphinoline 2-oxide derivatives **4a-j** were obtained in high to quantitative yields (75%-99%), irrespective of the nature and position of the substituents attached to both the aromatic and alkyne partners.




**Scheme 1** Synthetic pathway of Isophosphinoline 2-oxides **4**

To decipher key interaction features of isophosphinoline oxides, the intracyclic alkene of **4a** was reduced by hydrogenation in the presence of 10% mol of Pd/C. The reaction afforded the trihydrophosphinoline **8** in almost quantitative yield depicted in Scheme 2.



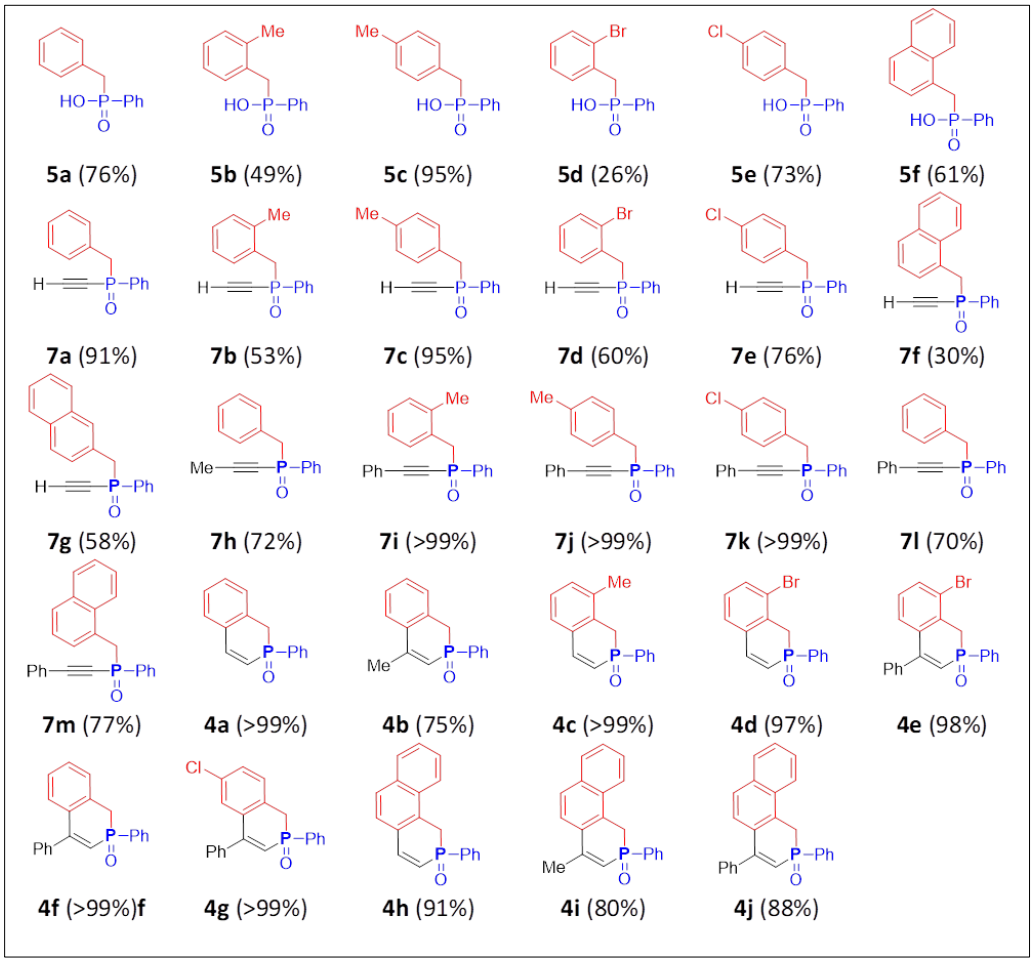

**Scheme 2** Reduction of Isophosphinoline 2-oxides **4a** into trihydrophosphinoline **8**

To determine an antibacterial effect, the disk diffusion method was used [23]. All experiments were done in duplicate. A first screening assay was performed on four strains *P. aeruginosa* ATCC9027, *E. faecalis* ATCC2921, *E. coli* ATCC8739 and *S. aureus* ATCC6538 and a selection of 16 compounds. Interestingly, only *S. aureus* strain was sensitive while no inhibitory activity was detected against the other ones. This first experiment demonstrated two key strengths of this series of compounds. Firstly, there were active on a specific gram-positive bacterium. Infections caused by Gram-positive bacteria have gained a worldwide notoriety as they continue to pose significant treatment challenges particularly with multi-resistant strains such as *S. Aureus* [24]. Second, the narrow spectrum of activity almost excluded the behavior of the designed compounds as simple cytotoxic agents. Then, the antibacterial activity of the 24 compounds listed in Table 2 was evaluated consecutively against the Gram-positive bacteria *S. aureus* using disk diffusion assay. Isophosphinolines oxides 4a-j expressed the full range of activities and the activity cannot be associated to the simple electrophilic character of the vinylphosphine oxide moiety. Indeed the naked isophosphinoline **4a** only expressed a weak efficacy on *S. aureus*. Substitution of alkene function by a 4-methyl or a 4-phenyl group generally resulted in an increased activity (**4a** *vs* **4b** and **4f**) while formerly the electrophilic character of the alkene was drastically reduced due to increased steric hindrance and conjugation. Modification of the fused-phenyl ring induced contrasted effects on the activity. Then, substitution of a hydrogen by a 8-methyl residue resulted in equivalent inhibition on *S. aureus* (**4a** *vs* **4c**) while the activity was increased when replacing the phenyl ring by an extended naphthyl ring (**4a** *vs* **4h**). In this series, the best activity was observed when a bromine was introduced in position 8 (**4a** *vs* **4d**). The situation was far more complex when both modifications on alkene and fused aryl groups were combined. 4-Methyl and 4-phenyl substituted benzoisophosphinolines **4i** and **4j** were less efficient than their corresponding related compounds, respectively **4b** and **4f**.

We also took the benefit to evaluate the activity of alkyne intermediates **7a-m** as potential covalent inhibitors. Indeed, nonactivated terminal alkynes are prone to react irreversibly as latent electrophiles with endogenous thiols and are considered as a golden standard to develop cysteine-reactive warheads [25]. Surprisingly electron-deficient alkynes were less cited while the electrophilic character persisted in such compounds. Antibacterial activity remained high for

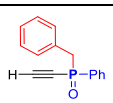
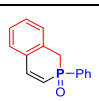
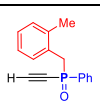
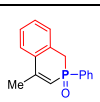
the unsubstituted alkynes **7a-e**. Then, compounds **7a-7e** showed to be very active while naphthyl derivatives **7f** and **7g** presented moderate to weak activity. This observation could be attributed to the poor aqueous solubility of such derivatives. By contrast, with the isophosphinolines **4**, alkynyl substituted phosphine oxides **7h-7l** were mostly inactive or weakly active.

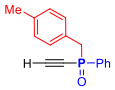
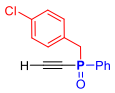
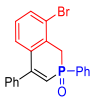
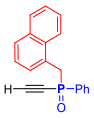
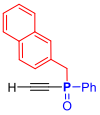
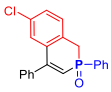
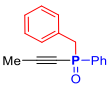
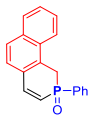
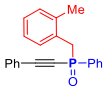
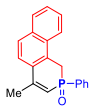
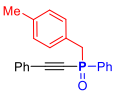
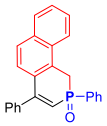
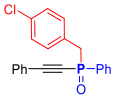
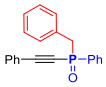
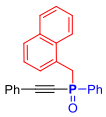
**Table 1** Arylmethyl(phenyl)phosphinic acids 5, (Arylmethyl)(alkynyl)phenylphosphine oxides 7a-m and Isophosphinoline 2-oxides 4a-j




To validate the nature of the pharmacophore, activity of the saturated trihydroisophosphinoline **8** was also determined in the same conditions. As expected, the lack of the alkene resulted in the disappearance of any effect on *S. Aureus* strain (Table 2 compound **8**).

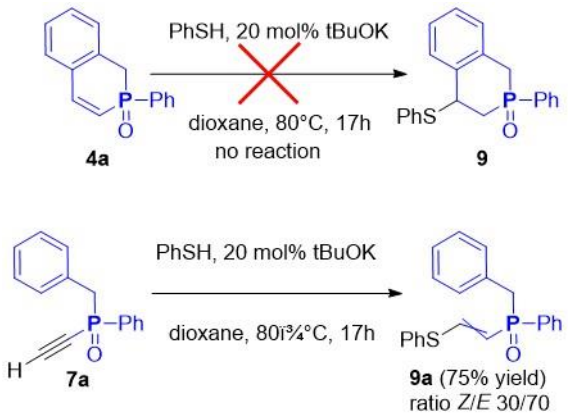
**Table 2** Activity on disk diffusion assay against *S. aureus*

| Compound | structure | *Activity against S. aureus*[1] | Compound | structure | *Activity against S. aureus*[1] |
|---|---|---|---|---|---|
| **7a** |  | +++ | **4a** |  | + |
| **7b** |  | +++ | **4b** |  | +++ |

| | | | | | |
|---|---|---|---|---|---|
| **7c** |  | +++ | **4c** |  | + |
| **7d** |  | +++ | **4d** |  | +++ |
| **7e** |  | +++ | **4e** |  | + |
| **7f** |  | ++ | **4f** |  | +++ |
| **7g** |  | + | **4g** |  | ++ |
| **7h** |  | + | **4h** |  | ++ |
| **7i** |  | + | **4i** |  | 0 |
| **7j** |  | 0 | **4j** |  | ++ |
| **7k** |  | 0 | **8** |  | 0 |
| **7l** |  | 0 | | | |
| **7m** |  | ++ | | | |

[1] not active (0) for a diameter < 6 mm, weak activity (+) for a diameter ≥ 6 mm and < 8 mm, moderate activity (++) for a diameter ≥ 8 mm and < 10 mm, very active (+++) for a diameter ≥ 10 mm.

Targeted covalent inhibitor mechanism of action involves the formation of covalent bonding with protein nucleophilic residues, mostly cysteines [26]. Consequently, formation of such covalent linkage was simulated through a model reaction. Benzyl(ethynyl)phenylphosphine oxide **7a** and dihydrophosphinoline **4a** were reacted with thiophenol in dioxane using catalytic amounts of a base (20 mol% *t*BuOK) (Scheme 3). The alkene bond of isophosphinoline **4a** appeared not reactive enough and no nucleophilic addition was observed even at 80°C heating. By contrast, ethynylphosphine oxide **7a** afforded smoothly the corresponding adduct 10 isolated in 75% yield as a mixture of *Z*/*E* 30/70. If alkenes bearing electron-withdrawing groups were classically considered as good Michael acceptor, conjugation with fused-phenyl ring in association with poor orbital overlap between carbon and phosphorus atoms in isophosphinoline could explain this non-reactivity. It is therefore possible that, although structurally related, the mode of action of isophosphinolines **4** and alkynylphosphines **7** are different. These results also help to explain the selectivity observed between the different bacterial strains.




**Scheme 3** Model of thiolation between ethynylphosphine oxide 7a or dihydrophosphinoline 4a and thiophenol

## 4. Experimental Section

### 4.1. General procedure

*4.1.1. 2-Phenyl-1,3,4-trihydroisophosphinoline 2-oxide **8**.*

In a Schlenk tube were successively introduced 2-phenyl-1H-isophosphinoline 2-oxide (0.1 g, 0.4 mmol, 1 eq) **4a**, palladium on carbon 10% (20 mg, 0.05 mol%) and ethanol (2 mL). Vacuum (until solvent bubbling) then hydrogen, at atmosphere pressure, was introduce on the system. After 12 h of stirring, the reaction mixture was filtered on celite and solvent was removed under vacuum to give colorless yellow oil 8, 0.1 g (>99% yield).

$^{1}$H NMR (400 MHz, Chloroform-d) δ 7.59 – 7.33 (m, 6H), 7.31 – 7.18 (m, 3H), 7.12 (d, *J* = 7.2 Hz, 1H), 3.51 – 3.25 (m, 2H), 3.09 (ddt, *J* = 29.6, 15.0, 5.3 Hz, 1H), 2.40 (ddt, *J* = 17.2, 15.0, 5.1 Hz, 1H), 2.29 – 2.06 (m, 1H). $^{13}$C NMR (101 MHz, Chloroform-d) δ 138.6 (d, *J* = 9.1 Hz), 134.1 (d, *J* = 95.6 Hz), 131.8 (d, *J* = 3.0 Hz), 131.6 (d, *J* = 6.4 Hz), 130.4 (d, *J* = 8.9 Hz), 130.2 (d, *J* = 9.1 Hz), 128.6 (d, *J* = 11.6 Hz), 128.4 (d, *J* = 2.7 Hz), 127.7 (d, *J* = 1.8 Hz), 127.7, 34.8 (d, *J* = 62.4 Hz), 28.4 (d, *J* = 72.4 Hz), 28.2 (d, *J* = 5.9 Hz). $^{31}$P NMR (162 MHz, $CDCl_3$) δ 35.48 (s). HRMS: m/z calcd for $C_{15}H_{16}OP$ 243.0939 $[M + H]^+$, Found 243.0949.

*4.1.2. Typical procedure for the thiolation of benzyl(ethynyl)(phenyl) phosphine oxide **9a** (**Z** and **E**).*

Z/E-benzyl(phenyl)(2-(phenylthio)vinyl)phosphine oxide **9a**

Under $N_2$, in a Schlenk tube, were successively introduced **7a** (0.1 g, 0.416 mmol, 1 eq), thiophenol (47 µL, 0.45 mmol, 1.1 eq), *t*-BuOK (9 mg, 20%) and 1,2 dioane (3 mL). The reaction was heating at 80°C for 17h. Then, at room temperature, after addition of MeOH (1mL), 5 mL $HCl_{aq}$ (1N) extraction with DCM, and concentration under vacuum, the solid was purified by flash column chromatography eluting with EtOAC (110 mg, mixture of Z/E 30/70, 72% yield).

$^{1}$H NMR (400 MHz, Chloroform-d) δ 8.01 – 6.85 (m, 15H), 5.98 (dd, J = 21.0, 12.3 Hz, 0.3H), 5.87 (dd, J = 22.5, 16.5 Hz, 0.7H), 3.51 (dd, J = 15.3, 3.7 Hz, 0.6H), 3.43 – 3.24 (m, 1.3H). $^{31}$P NMR (162 MHz, Chloroform-d) δ 26.82 (30%), 26.41 (70%). HRMS: m/z calcd for $C_{21}H_{20}OPS$ 351.0971 $[M + H]^+$, found 351.0967.

*4.1.3. $^{1}H$, $^{13}C$, $^{31}P$ NMR spectra and HRMS of compound **8***

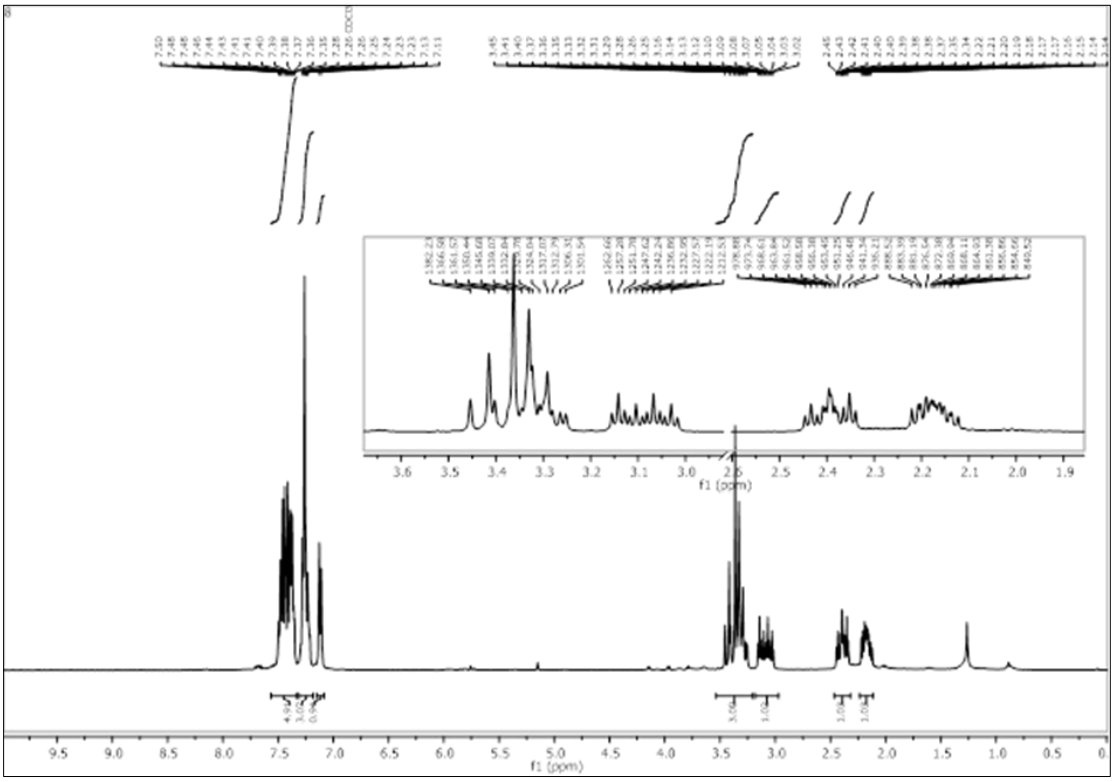


**Figure 3** $^{1}H$ NMR spectra of compound **8**

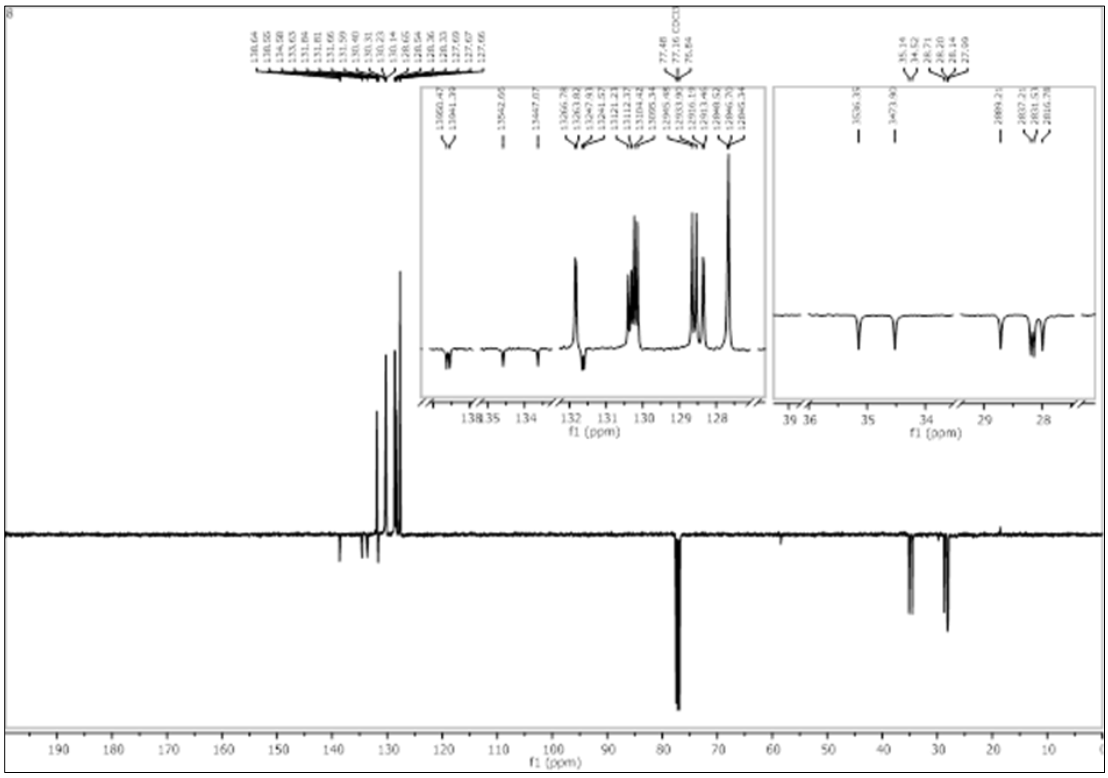


**Figure 4** $^{13}C$ NMR spectra of compound **8**

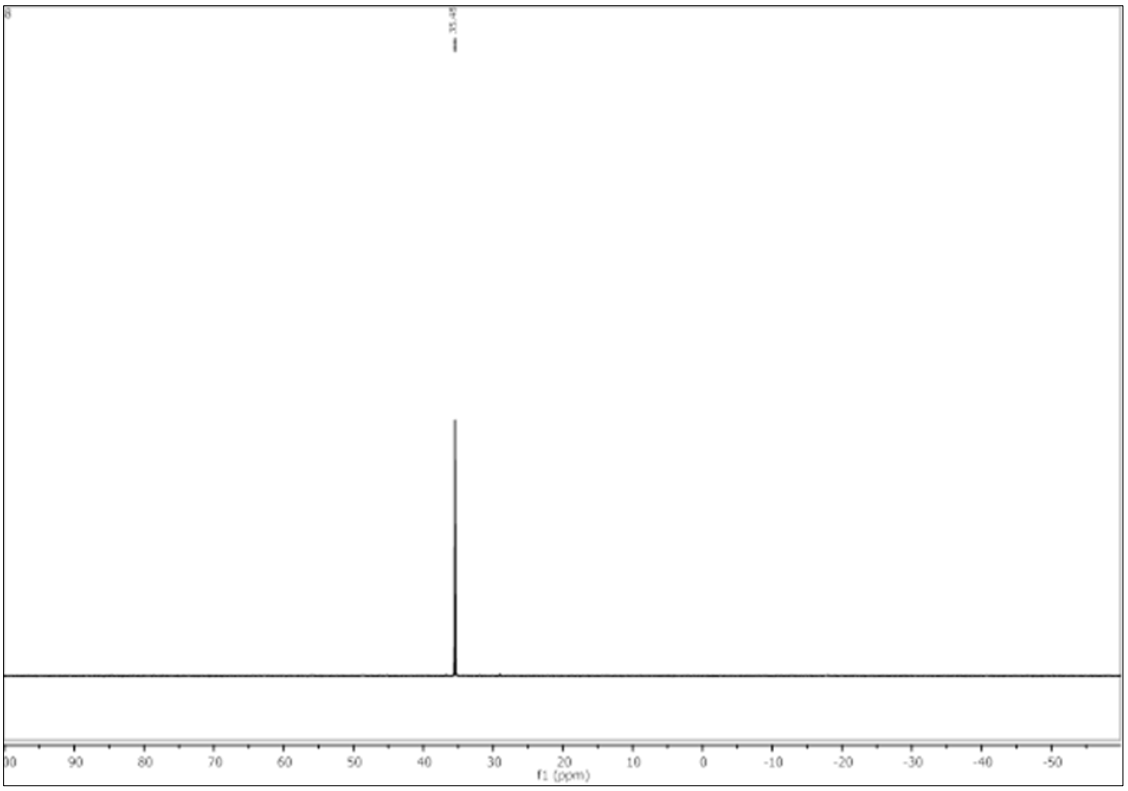


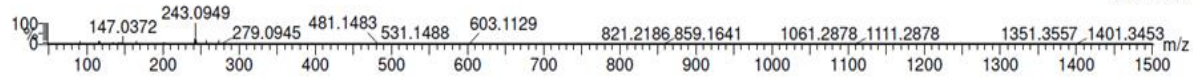

**Figure 5** $^{31}P$ NMR spectra of compound **8**

**Elemental Composition Report** **Page 1**

**Single Mass Analysis**
Tolerance = 2.0 mDa / DBE: min = -10.0, max = 50.0
Element prediction: Off
Number of isotope peaks used for i-FIT = 4

Monoisotopic Mass, Even Electron Ions
150 formula(e) evaluated with 1 results within limits (up to 50 best isotopic matches for each mass)
Elements Used:
C: 0-100 H: 0-100 O: 0-20 P: 0-2

Synapt G2S UEB205 MH5a 02-Nov-2020
PIR-2 5 (0.228) Cm (3:7) 1: TOF MS ES+
8.67e+006




| Mass | Calc. Mass | mDa | PPM | DBE | i-FIT | Norm | Conf(%) | Formula |
|---|---|---|---|---|---|---|---|---|
| Minimum: | | | | -10.0 | | | | |
| Maximum: | | 2.0 | 1.0 | 50.0 | | | | |
| 243.0949 | 243.0939 | 1.0 | 4.1 | 8.5 | 1345.3 | n/a | n/a | C15 H16 O P |

**Figure 6** HRMS of compound **8**

*4.1.4. $^{1}H$, $^{13}C$, $^{31}P$ NMR spectra and HRMS of compound **9a** (**Z** and **E**)*

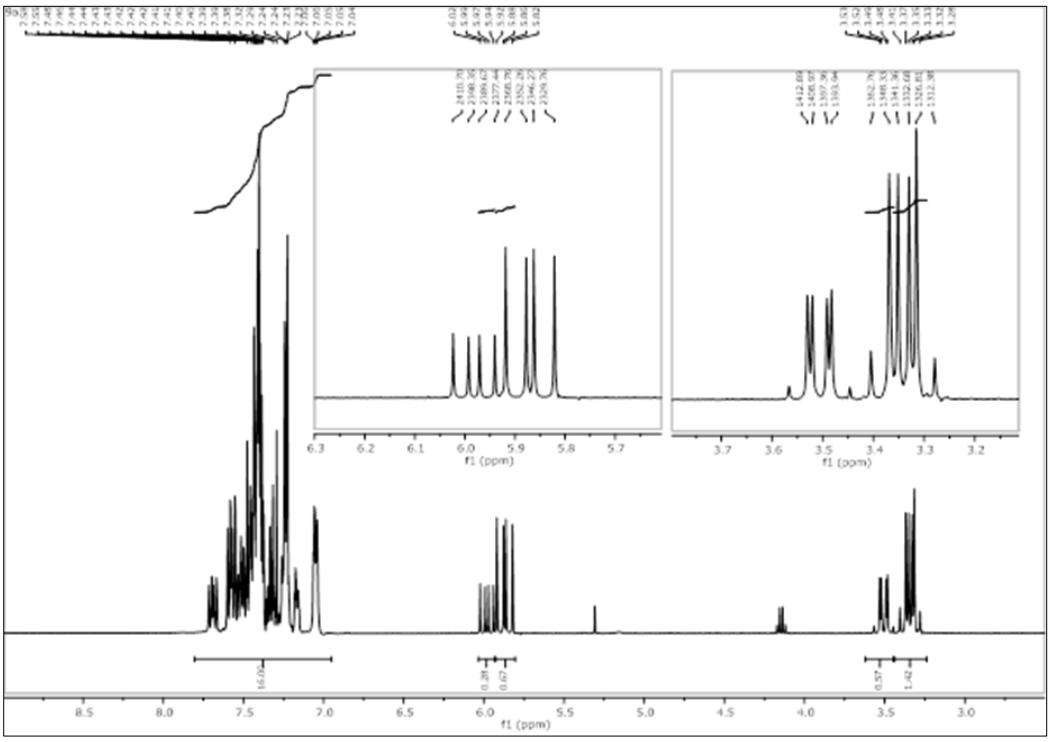


**Figure 7** $^{1}H$ NMR spectra of compound **9a**

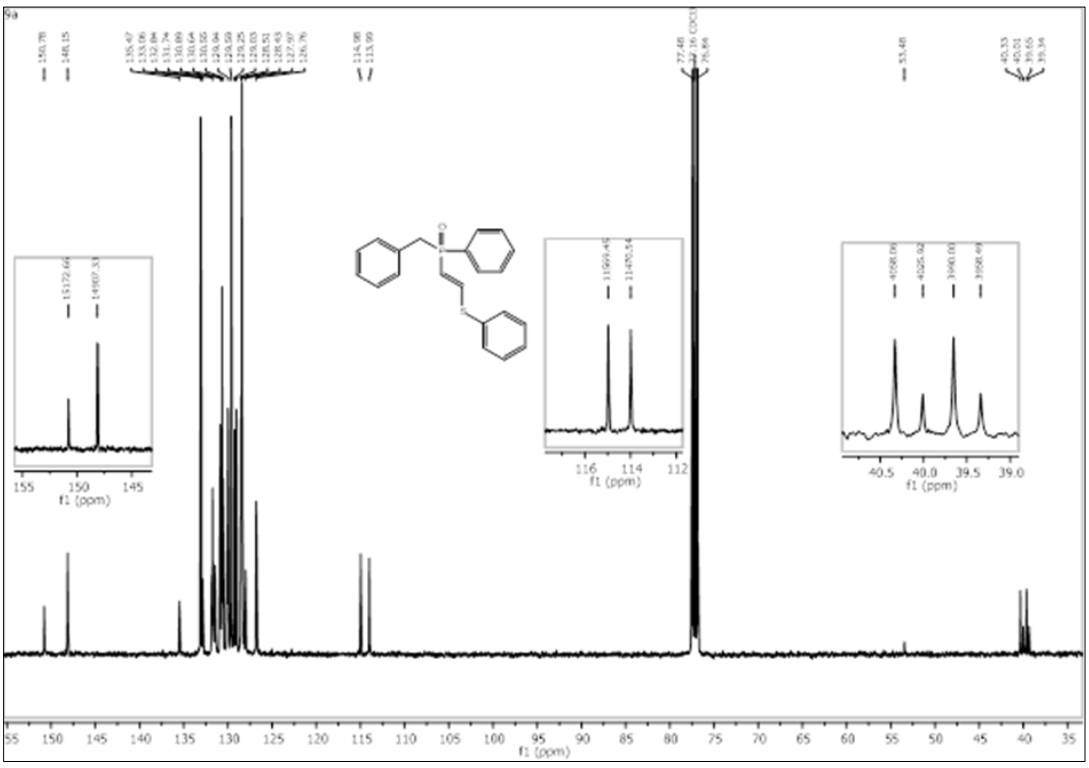


**Figure 8** $^{13}C$ NMR spectra of compound **9a**

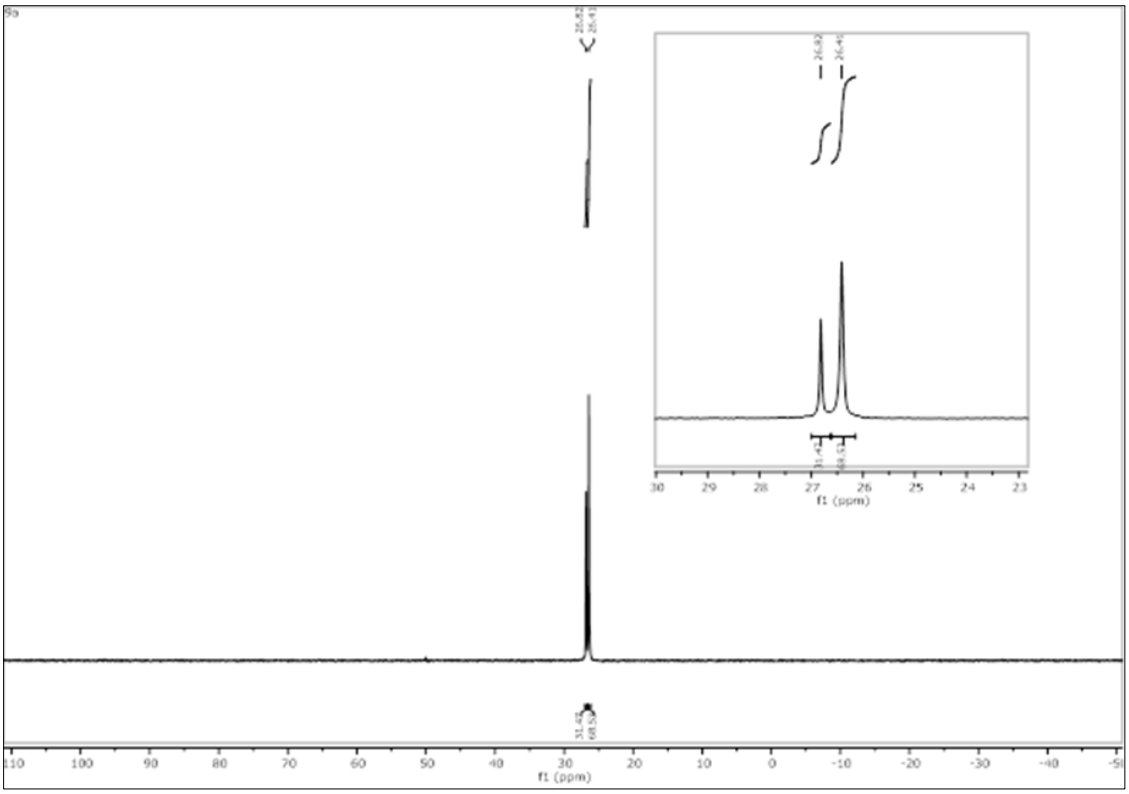



**Figure 9** $^{31}P$ NMR spectra of compound **9a**

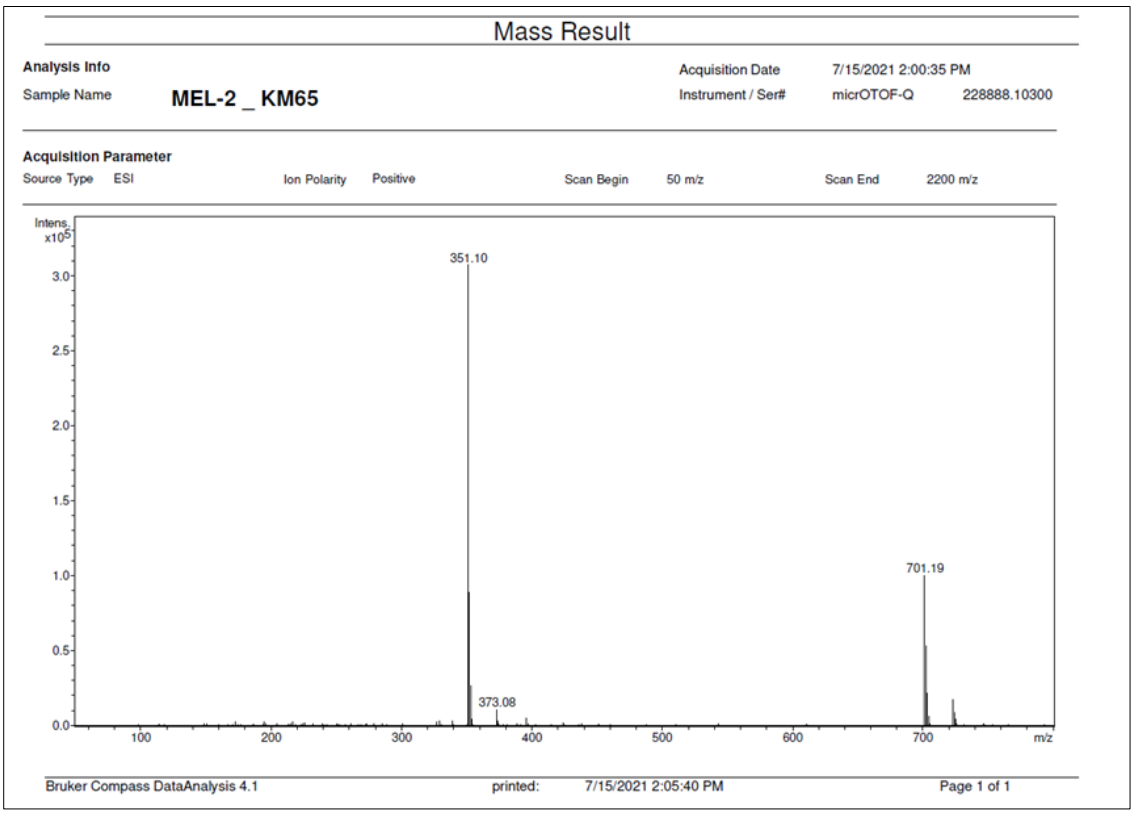



**Figure 10** HRMS of compound **9a**

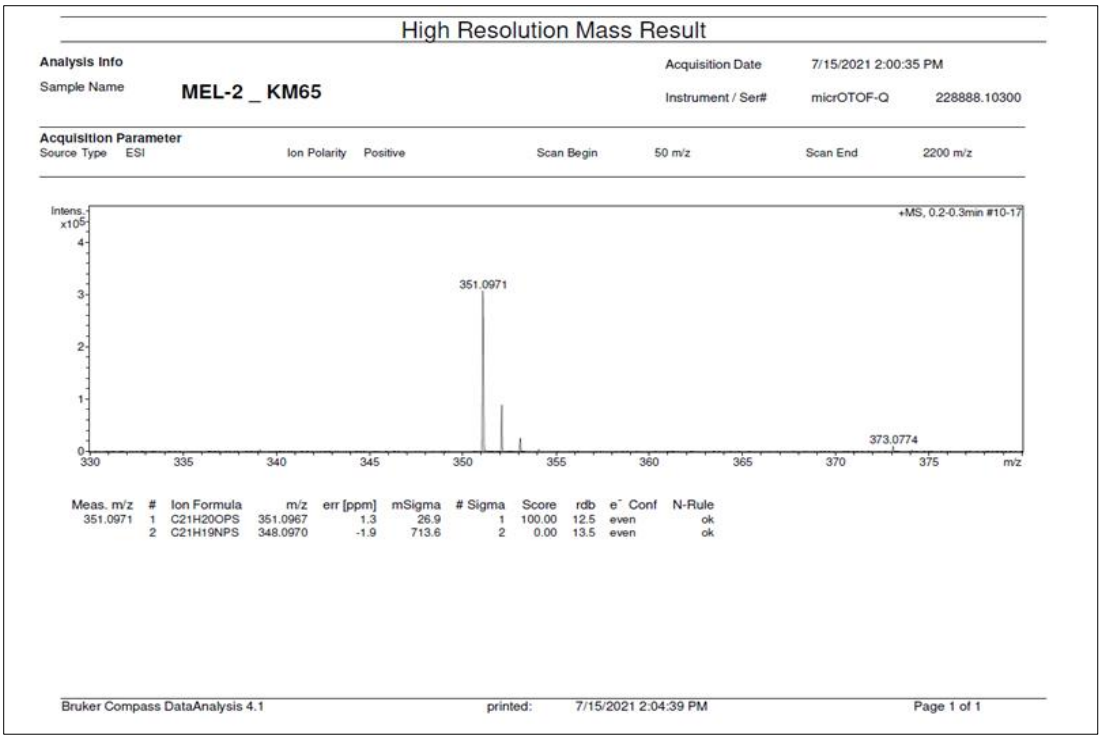



**Figure 11** HRMS of compound **9a**

## 5. Conclusion

In conclusion, hydrolytically stable surrogates of coumarins, 2-phenyl H-isophosphinoline 2-oxides **4** were easily obtained from a gold-mediated cyclization of P-arylmethyl alkynylphosphine oxides **5**. This intramolecular hydroarylation is highly selective and led to isophosphinoline oxides **4** generally in high yields. Consecutively, isophosphinoline oxides **4a-j** and alkynylphosphine oxides **5a-5m** were screened in vitro for their potential antibacterial activity against four Gram-negative and Gram-positive bacterial strains. Interestingly, antibacterial activity was only observed on Gram-positive S. aureus strain whereas Gram-negative bacteria and gram-positive E. faecalis were not sensitive. Isophosphinolines **4b**, **4d** and **4f** which shared the common heterocyclic core, showed the best activity. Alkynes intermediates **5a-e** also demonstrated potency in such antibacterial tests. On the contrary to isophosphinolines **4**, the best activity of alkynylphosphine oxides **7a-e** was observed for terminal alkyne derivatives which can be directly associated to the electrophilic character of such compounds. Structure activity relationship (SAR) for isophosphinolines is more complex. If the presence of the intracyclic alkene appeared essential for activity, a correct balanced electrophilic character was required. Finally, our results highlighted the effectiveness of alkenyl and alkynylphosphine oxides as potent motifs for the inhibition of bacterial growth. Future work will focus on understanding the mode of action of these compounds and identifying their respective biological target.

## Compliance with ethical standards

### *Acknowledgments*

The research leading to these results received funding from the French Embassies in Iran, Gabon and Russia.

The Ministry of Science, Research and Technology of the Islamic Republic of Iran is gratefully acknowledged for the financial support provided to Mina Hariri (PhD student at the K. N. Toosi University of Technology of Tehran, Iran).

We are grateful to the National Graduate School of Chemistry of Montpellier and to the Institut Charles Gerhardt of Montpellier that offered to Victor Mesnil the possibility to participate in this project.

### *Disclosure of conflict of interest*

There is no Conflict of interest.

*Statement of ethical approval*

Ethical approval was obtained from concerned institutions.

*Statement of informed consent*

Informed consent was obtained from all individual participants included in the study.

## References


[1] (a)Neu HC. Science 1992; 257, 1064. https://doi:10.1126/science.257.5073.1064. (b) Martens E.; Demain A. L. Jpn. J. Antibiot. 2017, 70, 520. https://doi:10.1038/ja.2017.30. (c) Bell B. G.; Schellevis F.; Stobberingh E.; Goossens H.; Pringle M. BMC Infect. Dis. 2014, 14. https://doi.org/10.1186/1471-2334-14-13.

[2] (a) Lee AS, de Lencastre H, Garau J, Kluytmans J, Malhotra-Kumar S, Peschel A, Harbarth S. Nat Rev Dis Prim. 2018; 4: 18033. https://doi:10.1038/nrdp.2018.33. (b) Lakhundi S, Zhang K. Clin Microbiol Rev. 2018; 31: 1. https://doi:10.1128/CMR.00020-18. (c) Kulkarni AP, Nagvekar VC, Veeraraghavan B, Warrier AR TS D, Ahdal J, Jain R. Hindawi Interdisciplinary Perspectives on Infectious Diseases 2019; Article ID 7601847, 1. https://doi.org/10.1155/2019/7601847.

[3] de Kraker ME, Stewardson AJ, Harbarth S. PLoS Med. 2016; 13: 11.

[4] (a) 2020 Antibacterial agents in clinical and preclinical development: an overview and analysis. Geneva: World Health Organization. 2021; ISBN 978-92-4-002130-3. (b) Bell BG, Schellevis F, Stobberingh E, Goossens H, Pringle M. BMC Infect Dis. 2014; 14: 13. https://doi:10.1186/1471-2334-14-13. (c) Rocha-Granados MC, Zenick B, Englander HE, Mok WWK. Cell Signal. 2020; 75: 109750.

[5] (a) Smith PA, Romesberg FE. Nat Chem Biol. 2007; 3: 549. https://doi:10.1038/nchembio.2007.27. (b) Feature N. Nat Rev Drug Discov. 2007; 6: 8. https://doi:10.1038/nrd2225.

[6] Fesatidou M, Petrou A, Athina G. Curr Pharm Des. 2020; 26: 867.

[7] (a) Petersen P, Godtfredsen J, Boysen G, Andersen E, Andersen B. Lancet. 1989; 333: 175. https://doi:10.1016/S0140-6736(89)91200-2. Levine MN, Hirsh J, Gent M, Turpie AG, Weitz J, Ginsberg J, Geerts W, LeClerc J, Neemeh J. Thromb. Haemost. 1995; 74: 606.

[8] (a) Walsh TJ, Standiford HC, Reboli AC, John F, Mulligan ME, Ribner BS, Montgomerie JZ, Goetz MB, Mayhall CG, Rimland D. Antimicrob Agents Chemother. 1993; 37: 1334. https://doi:10.1128/AAC.37.6.1334. (b) Raad II, Hachem RY, Abi-Said D, Kenneth VI, Rolston MD, Whimbey E, Buzaid EC, Legha S. Cancer 1998; 82: 403. https://doi:10.1002/(SICI)1097-0142(19980115)82:2<412::AID-CNCR22>3.0.CO;2-0.

[9] (a) Patel G, Banerjee S, Curr Org Chem. 2020; 24: 2566. https://doi:10.2174/1385272824999200709125717. (b) Calcio Gaudino E, Tagliapietra S, Martina K, Palmisano G, Cravotto G. RSC Adv. 2016; 6: 46394. https://doi:10.1039/C6RA07071J. (c) Vazquez-Rodriguez S, Joao Matos M, Borges F, Uriarte E, Santana L, Curr Top Med Chem. 2015; 15: 1755. https://doi:10.2174/1568026615666150427125916. (d) Riveiro ME, De Kimpe N, Moglioni A, Vazquez R, Monczor F, Shayo C, Davio C. Curr Med Chem. 2010; 17: 1325. https://doi:10.2174/092986710790936284.

[10] (a) de Souza SM, Delle Monache F, Smânia Jr A, Z. Naturforsch. 1999; 54c: 169. (c) Smyth T, Ramachandran VN, Smyth WF. International Journal of Antimicrobial Agents. 2009; 33: 421. (d) Yang L, Ding W, Xu Y, Wu D, Li S, Chen J, Guo B. Molecules 2016; 21: 468. https://doi.org/10.1016/j.ijantimicag.2008.10.022.

[11] Kafarski P, Lejczak B. The Biological Activity of Phosphono- and Phosphinopeptides in: Aminophosphonic and aminophosphinic acids. John Wiley & Sons, LTD. 2000; 407-442.

[12] Chekan JR, Cogan DP, Nair SK. Medchemcomm. 2016; 7: 28. https://doi:10.1039/c5md00351b.

[13] (a) Patani GA, LaVoie EJ. Chem Rev. 1996; 96: 3147. https://doi:10.1021/cr950066q. (b) Wermuth CG, Ciapetti P, Giethlen B, Bazzini P, Bioisosterism. In: Comprehensive Medicinal Chemistry II. Vol 2. Elsevier; 2007; 649-711. https://doi:10.1016/B0-08-045044-X/00051-1.

[14] Lima L, Barreiro E. Curr Med Chem. 2005; 12: 23. https://doi:10.2174/0929867053363540.

[15] (a) Smith BR, Eastman CM, Njardarson JT, Beyond CJ, Med Chem. 2014; 57: 9764. https://doi:10.1021/jm501105n. (b) Finkbeiner P, Hehn JP, Gnamm CJ. Med Chem. 2020; 63: 7081. https://doi:10.1021/acs.jmedchem.0c00407. (c) Volle JN, Guillon R, Bancel F, Bekro YA, Pirat JL, Virieux D.

Phosphono- and Phosphinolactones in the Life Sciences. Advances in Heterocyclic Chemistry. 2016; 118: 129. https://doi.org/10.1016/bs.aihch.2015.10.004. (d) Brewster R, Vandergeten MC, Montel F Eur. J. Org. Chem. 2014; 905. https://doi.org/10.1002/ejoc.201301299. (e) Mucha A, Kafarski P, Berlicki LJ. Med. Chem. 2011; 54: 5955. https://doi.org/10.1021/acs.jmedchem.0c00407. (f) Moonen K, Laureyn I, Stevens CV. Chem. Rev. 2004; 104: 6177.

[16] (a) Clarion L, Jacquard C, Sainte-Catherine O, Loiseau S, Filippini D, Hirlemann MH, Volle JN, Virieux D, Lecouvey M, Pirat JL, Bakalara N. J Med Chem. 2012; 55: 2196. https://doi:10.1021/jm201428a. (b) Clarion L, Jacquard C, Sainte-Catherine O, Decoux M, Loiseau S, Rolland M, Lecouvey M, Hugnot JP, Volle JN, Virieux D, Pirat JL Bakalara N. J Med Chem. 2014; 8293. https://doi:10.1021/jm500522y. (c) Babouri R, Rolland M, Sainte-Catherine O, Kabouche Z, Lecouvey M, Bakalara N, Volle JN, Virieux D, Pirat JL, Eur J Med Chem. 2015; 104: 33. https://doi:10.1016/j.ejmech.2015.09.027. (d) Hassani Z, Saleh A, Turpault S, Khiati S, Morelle W, Vignon J, Hugnot JP, Uro-Coste E, Legrand P, Delaforge M, Loiseau S, Clarion L, Lecouvey M, Volle JN, Virieux D, Pirat JL, Duffau H, Bakalara N. Mol Cancer Res. 2017; 15: 1376. https://doi:10.1158/1541-7786.MCR-17-0120. (e) Bousseau S, Marchand M, Soleti R. FASEB J. 2019; 33: 5864. https://doi:10.1096/fj.201801450RRR.

[17] (a) Volle JN, Filippini D, Krawczy B, Kaloyanov N, Van der Lee A, Maurice T, Pirat JL, Virieux D, Org Biomol Chem. 2010; 8: 1438. https://doi:10.1039/b919345f. (b) Maurice T, Volle JN, Strehaiano Crouzier L, Pereira C, Kaloyanov N, Virieux D, Pirat JL. Pharmacol Res. 2019; 144: 315. https://doi:10.1016/j.phrs.2019.04.026.

[18] (a) Summy JM, Trevino JG, Lesslie DP, Justin M. Summy, Trevino JG, Lesslie DP, Baker CH, Shakespeare WC, Wang Y, Sundaramoorthi R, Metcalf C. A. III; Keats JA, Sawyer TK, Gallick GE, Mol Cancer Ther. 2005; 4: 1900. https://doi:10.1158/1535-7163.MCT-05-0171. (b) Huang WS, Liu S, Zou D, Wang Y, Zhou T, Romero J, Kohlmann A, Li F, Qi J, Cai L, Dwight TA, Xu Y, Xu R, Dodd R. Toms A, Parillon L, Lu X, Anjum R, Zhang S, Wang F, Keats J, Wardwell SD, Ning Y, Xu Q, Moran LE, Mohemmad QK, Gyung Jang H, Clackson T, Narasimhan NI, Rivera VM, Zhu X, Dalgarno D, Shakespeare WCJ. Med. Chem. 2016; 59: 4948. https://doi:10.1021/acs.jmedchem.6b00306. (c) Fedyk A, Slobodyanyuk EY, Stotska O, Vashchenko BV, Volochnyuk DM, Sibgatulin DA, Tolmachev AA, Grygorenko OO. Eur J Org Chem. 2021; 1. https://doi:10.1002/ejoc.202100581.

[19] (a) Belyaev A, Zhang X, Augustyns K, J Med Chem. 1999; 42: 1041. https://doi:10.1021/jm981033g. (b) Foust B. J, Poe MM, Lentini NA, Hsiao CHC, Wiemer AJ, Wiemer DF. ACS Med Chem Lett. 2017; 8: 914. https://doi:10.1021/acsmedchemlett.7b00245.

[20] Sutanto F, Konstantinidou M, Dömling A. RSC Med Chem. 2020; 1: 876. https://doi:10.1039/D0MD00154F.

[21] a) Wallis CJ, Virieux D, Cristau HJ. Diphenyl(vinyl)- phosphine Oxide. Encyclopedia of Reagents for Organic Synthesis; John Wiley & Sons, Ltd: Chichester, U.K. 2005; 1: 1. (b) Rahman MS, Steed JW, Hii KK. Synthesis 2000; 1320. https://doi:10.1055/s-2000-6422. (c) Scherner C, Ergüden JK, Adiwidjaja G, Schaumann E. Synthesis; 2014; 46: 2506. (d) Gonzalez-Nogal AM, Cuadrado P, Sarmentero MA. Tetrahedron. 2010; 66: 9610. https://doi:10.1016/j.tet.2010.10.016.

[22] Hariri M, Darvish F, Mengue Me Ndong KP, Sechet N, Chacktas G, Boosaliki H, Tran Do ML, Mwande-Maguene G, Lebibi J, Burilov AR, Ayad T, Virieux D, Pirat JL. JOC. 2021; 86: 7813. https://doi.org/10.1021/acs.joc.1c00648.

[23] https://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Disk_test_documents/2021_manuals/Manual_v_9.0_EUCAST_Disk_Test_2021.pdf

[24] (a) Banwan K, Senok AC, Rotimi VOJ. Infect Public Health. 2009; 2: 62. https://doi:10.1016/j.jiph.2009.04.003. (b) Munita JM, Bayer AS, Arias CA. Clin Infect Dis. 2015; 61: S48. https://doi:10.1093/cid/civ523. (c) Abbas M, Paul M, Huttner A. Clin Microbiol Infect. 2017; 23: 697. https://doi:10.1016/j.cmi.2017.06.010.

[25] (a) Gehringer M Laufer SAJ. Med Chem. 2019; 62: 5673. https://doi:10.1021/acs.jmedchem.8b01153. (b) Mons E, Jansen IDC, Loboda J, van Doodewaerd BR, Hermans J, Verdoes M, van Boeckel CAA, van Veelen PA, Turk B, Turk D, Ovaa H. J Am Chem Soc. 2019; 141: 3507. https://doi:10.1021/jacs.8b11027. (c) Mons E, Kim RQ, van Doodewaerd BR, van Veelen PA, Mulder MPC, Ovaa H. J Am Chem Soc. 2021; 143: 64: 23.