Synthesis of novel antiproliferative hybrid bis-(3-indolyl)methane phosphonate derivatives

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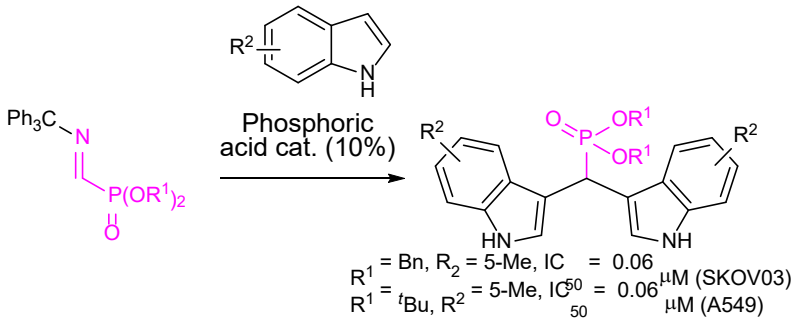
Keywords: Indole, bis-(3-indolyl)methanes (BIMs), Phosphonate, Antiproliferative effect.

Abstract:

An efficient synthetic methodology for the preparation of phosphorus substituted bis-(3-indolyl)methane through a double nucleophilic addition of indole derivatives to an *in situ* generated α-iminophosphonate is reported. In addition, bis-(3-indolyl)methane substrates showed *in vitro* cytotoxicity, inhibiting the growth of carcinoma human tumor cell lines A549 (carcinomic human alveolar basal epithelial cell) and SKOV03 (human ovarian carcinoma).



1. Introduction:

Due to the notable grown in the life expectancy during the last decades, cancer has become one of the leading causes of death worldwide [1]. The World Health Organization (WHO) reports 8.8 million people died of cancer globally in 2015, being the most common cause of cancer death the cancer of lung with 1,69 million (19,4%) of deaths [2]. Cancer treatment comprises, in most of the cases, a combination of surgery and chemotherapy [3] and here is where Drug Discovery can play a crucial role into this area. There is still a serious need to search for some newer and safer anticancer agents and, therefore, the discovery of new active compounds and the *in vitro* evaluation of their anticancer properties represents an important task in Medicinal Chemistry, in order to improve our toolbox for the treatment of cancer.

Indole framework holds a very high affinity to multiple receptors and enzymes and, accordingly, it is considered a privileged structure in many active medicine compounds for human health and represents a promising scaffold for drug development [4]. In particular, bis-indole family derivatives, are of extraordinary significance in Synthetic and Medicinal Chemistry due to their wide occurrence in nature and their assorted biological activity [5]. Simple bis-(3-indolyl)methane (BIM) **I** and their derivatives (BIMs) **II-VIII** are nitrogen-

containing heterocyclic compounds which structure can be found in many alkaloids isolated from natural sources (Figure 1) [6]. Alkaloids such as arundine (**I**, R = H) [7], vidrindole (**II**, R = Me) [8], antibiotic turbomycins B, C, D, E (**III**, **IV**; R = Ar)[9] or arsindoline B (**V**, R = ^{n}Pr) [10] are isolated from marine bacteria, while streptindole (**VI**, R = Me) [11] is found in human feces. Cytotoxic barakacin (**VII**) has been isolated from a ruminal Pseudomonas [12] and the antibacterial and anti-inflammatory dalesindole (**VIII**), a new alkaloid, was obtained by mycosynthesis [13]. In addition, numerous BIMs and their analogues show a wide range of biological and pharmacological activities [14], including growth inhibition of numerous tumor types [15].

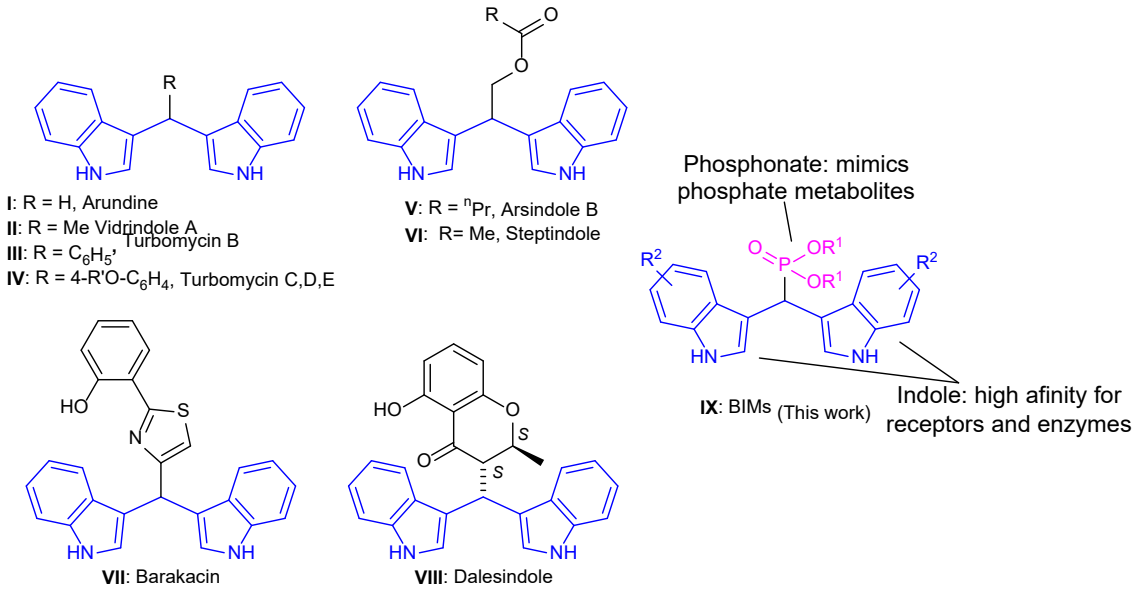


Figure 1. Structures of BIMs **I-VIII** and its phosphorated derivatives **IX**.

Among the innumerable amount of chemotherapeutic agents tested in vitro, we focused in organophosphorus compounds [16], particularly phosphoric acid esters. It is well known that structural modifications of active molecules involving the introduction of phosphorated functionalities, very often results in increased or new activities [17]. Due to the stability of its P-C bond and their chemical similitude to phosphate ester and anhydride metabolites, phosphonate derivatives show an assorted biological activity and, consequently, they have found numerous applications in medicine and agrochemistry [18]. Organophosphorus derivatives are interesting compounds from a biological point of view, since it is known that phosphorus substituents may affect the reactivity of heterocycles and regulate important biological functions [19]. Likewise, the development of new strategies for the preparation of aminophosphonates [20], phosphinated [21] or phosphorylated azaheterocycles [22] implies the incorporation of organophosphorus functionalities in simple synthons.

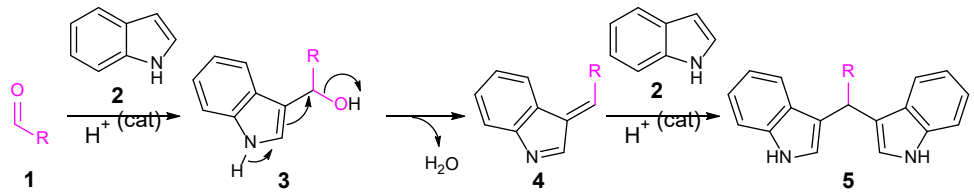
The hybrid anticancer drug approach is an innovative synthetic strategy for the discovery of new biologically active hybrid molecules [23]. It is believed that the presence of two or more pharmacophores in a single unit not only synergizes their biological effect but also upsurge their ability to inhibit more than one biological target. Recently, the molecular hybrid approach has resulted in several novel chemical entities with improved anticancer activity and selectivity with reduced side effects [24].

With these considerations in mind, we believed that the development of new hybrid molecules [25], such as phosphonate functionalized bis-indole derivatives **IX** (Figure 1), incorporating a phosphonate group in the BIMs structure (arundine, **I**, figure 1) may be privileged scaffolds for pharmaceuticals [26] and may improve the antiproliferative cytotoxic properties with respect to other biologically active structures. This represents an interesting challenge, due to the potential interest of these molecules not only in synthetic but also in medicinal chemistry. As far as we know, not only the synthesis but also the study of the biological activity of BIMs containing a phosphorated moiety have not been described.

2. Results and discussion:

2.1. Chemistry

The most straightforward method for the preparation of BIM derivatives, reported by Fischer in the late 19th century [27], is the acid catalyzed Friedel-Crafts reaction between aldehydes **1** and indoles **2**. After the first addition of electron rich indole **2** to electrophilic aldehyde **1**, the intermediate indole-3-carbinol **3** yields azafulvene **4** by means of elimination of a molecule of water. This new electrophilic species **4** undergoes a second acid catalyzed nucleophilic addition of indole **2** leading to the formation of BIM derivatives **5** [28] (Scheme 1).

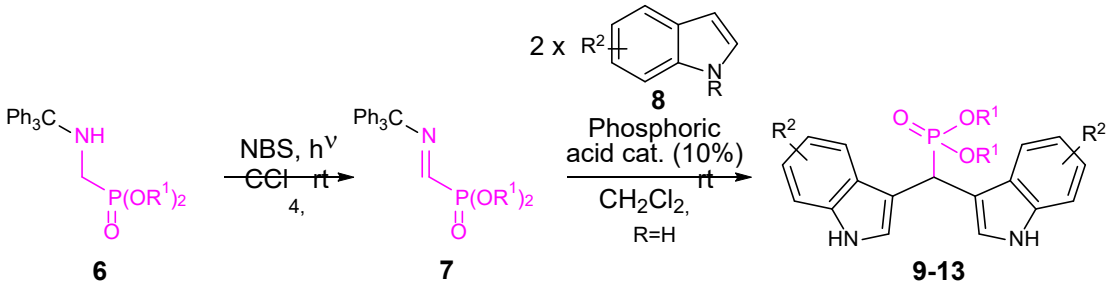


Scheme 1. Conventional synthesis of BIM derivatives **5**.

Following this approach, simply by using phosphorylated aldehydes **1** (R = P(O)OR$_2$) as starting substrates, our target molecules may be obtained in a single step. However, the use of formylphosphonates should be avoided since it suffers of numerous drawbacks. First, they are extremely moisture sensitive, affording readily acetal derivatives and, moreover, they have to be prepared by oxidation of diazomethylphosphonates using unstable Murray reagent [29]. For those reasons, we thought that α-phosphorylated aldimines would be much more suitable substrates for this reaction. Although less electrophilic, this aldimine substrates show slightly higher stability to moisture and, more importantly, they can be easily prepared by a formal

oxidation from α-aminophosphonates in a two-step protocol that comprises halogenation followed by β-elimination of hydrogen halide [30].

For our synthetic approach, we chose *N*-trityl substituted α-aminophosphonates **6** as starting substrates. Those substrates are readily obtained by a hydrophosphorylation reaction of dialkylphosphites and imines and, due to the bulkiness expected in the nitrogen atom, we envisioned the elimination of tritylamine to be favored in order to drive the process to the formation of azafulvene intermediate **4** (Scheme 1). We were delighted to discover that the radical bromination of α-aminophosphonate **6** using *N*-bromosuccinimide as bromine source in carbon tetrachloride led to the direct formation of a clean solution of α-iminophosphonate **7** by a subsequent spontaneous β-elimination of hydrogen bromide (Scheme 2). The generated succinimide together with the remaining *N*-bromosuccinimide are insoluble in carbon tetrachloride and they can be quickly removed by a simple filtration under inert atmosphere, in order to prevent degradation of α-iminophosphonate **7**.

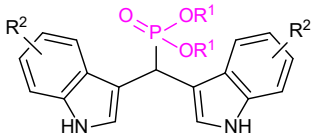


Scheme 2. Synthesis of bis-(3-indolyl)methyl phosphonate derivatives **9-13**.

Next, as we expected, the reaction of α-iminophosphonates **7** with two equivalents of indole derivative **8** in the presence of a catalytic amount of a phosphoric acid catalyst afforded the target BIM derivatives **9-13** in good yields (Chart 1, Scheme 2).

The reaction using simple indole **8a** (R^2 = R = H) was initially applied to the preparation of different phosphonates such as methylphosphonate **9** (R^1 = Me, Table 1, Entry 1), ethylphosphonate **10** (R^1 = Et, Table 1, Entry 2), *iso*-propylphosphonate **11a** (R^1 = ^{i}Pr, Table 1, Entry 3), *tert*-butylphosphonate **12a** (R^1 = ^{t}Bu, Table 1, Entry 7) and benzylphosphonate **13a** (R^1 = CH_2Ph = Bn, Table 1, Entry 10). Moreover, this methodology allows a wide range of electron-donating and electron-withdrawing substituents at the heteroaromatic substrate. According to this, the reaction was successfully extended to several indole substrates bearing strong (Table 1, Entries 15-17) and weak electron donating substituents (Table 1, Entries 6, 9, 18-20) and to 5- and 6-fluorine substituted indoles (Table 1, Entries 4-5, 8, 11-12). Finally, even some deactivated trifluoromethyl substituted [31] indoles were used with success in the reaction (Table 1, Entries 13-14).

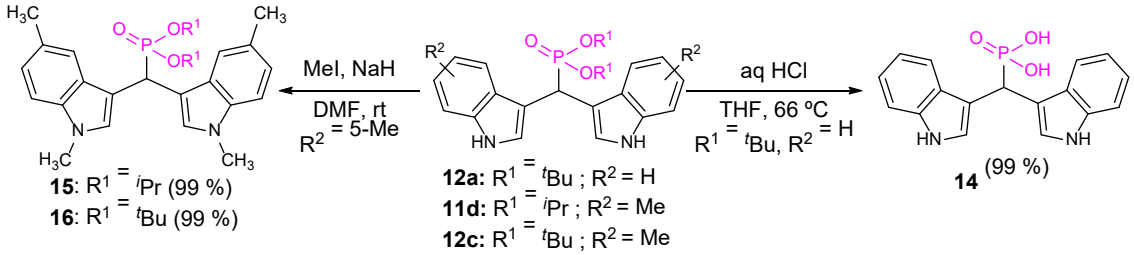
Table 1. BIM derivatives **9-13** obtained



Entry	Comp.	R^1	R^2	Yield (%)[a]
1	**9**	Me	H	59
2	**10**	Et	H	61
3	**11a**	^{i}Pr	H	60
4	**11b**	^{i}Pr	6-F	69
5	**11c**	^{i}Pr	5-F	67
6	**11d**	^{i}Pr	5-Me	62
7	**12a**	^{t}Bu	H	68
8	**12b**	^{t}Bu	5-F	69
9	**12c**	^{t}Bu	5-Me	72
10	**13a**	Bn	H	55
11	**13b**	Bn	6-F	65
12	**13c**	Bn	5-F	67
13	**13d**	Bn	5-CF_3	54
14	**13e**	Bn	6-CF_3	53
15	**13f**	Bn	5-OMe	57
16	**13g**	Bn	6-OMe	61
17	**13h**	Bn	5,6-OMe	58
18	**13i**	Bn	6-Me	64
19	**13j**	Bn	5-Me	65
20	**13k**	Bn	2-Me	63

[a] Yield after purification

In order to extend the diversity of substituents in our substrates, the acidic hydrolysis of dimethylphosphonate **12a** was performed using an aqueous solution of hydrochloric acid in refluxing tetrahydrofurane (THF). Work up of the reaction afforded phosphonic acid derivative **14** in very good yield (Scheme 3).



Scheme 3. Hydrolysis of dimethylphosphonate **12a** and *N*-methylation of **11d** and **12c**.

For the same purpose, the alkylation of indolic nitrogen of *iso*-propyl and *tert*-butyl phosphonate derivatives **11d** and **12c** was performed in the presence of methyl iodide in dimethylformamide, using sodium hydride as a base to yield bis-(3-*N*-methylindolyl) derivatives **15-16** in almost quantitative yields (Scheme 3). It should be noted that the direct synthesis of *N*-methylated BIM derivatives **15-16** using our reported protocol with *N*-

methylindole (**8** R=Me, Scheme 2) is not feasible, which may suggest a key role of NH group in the phosphoric acid catalyzed nucleophilic addition of indole species to α-iminophosphonates.

This methodology represents an easy protocol for the synthesis of BIM derivatives that avoids the use of unstable formylphosphonates and, as far as we know, this strategy is the first example of the preparation of BIMs with a phosphorus containing moiety.

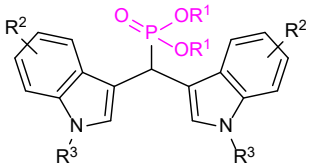
Next, with this collection of promising structures in our hands, we considered the study of their biological activity. Therefore, the behavior of prepared compounds as antiproliferative agents was investigated.

2.2. Biological results

In vitro cytotoxicity of our novel BIM derivatives was evaluated by testing their antiproliferative activities against two human cancer cell lines: A549 (carcinomic human alveolar basal epithelial cell) and SKOV03 (human ovarian carcinoma). Cell counting kit (CCK-8) assay was used in order to evaluate growth inhibition. Cell proliferation inhibitory activities of the bis-indol derivatives **9-16** and chemotherapeutic doxorubicin are shown in Table 2 as IC_{50} values. Moreover, MRC-5 non-malignant lung fibroblasts were tested for studying selective toxicity [32] and in contrast, none of the synthesized phosphorated compounds or doxorubicin exhibited any toxicity toward MRC-5 cells.

Regarding the effect of the substitution at the phosphonate ester group in BIM derivatives **9-16** into their cytotoxicity against SKOV03 cell line *in vitro*, it was evidenced that the presence of a bulky group, in general, resulted in an increased activity. Considering only substrates holding non-substituted indole moieties, while IC_{50} values higher than 50 μM were observed for methyl or ethyl esters **9** and **10** (Table 2, entries 1, 2), those values were dropped to 11.43±0.83 μM for *iso*-propyl ester **11a** (entry 3) and, interestingly, lower values of 1.06±0.30 μM and 1.26±0.13 μM were obtained for *tert*-butyl and benzyl esters **12a** and **13a** respectively (entries 7, 10). Although practically the same pattern was observed for A549 cell line, in this case the best result was observed for *iso*-propyl ester **11a** with IC_{50} values of 1.06±0.30 μM (entry 3). In contrast to this tendency, phosphonic acid derivative **14** did not show any cytotoxicity in any of the cell lines tested. Moreover, simple bis-(3-indolyl)methane Arundine (**I**, Figure 1) did not show cytotoxic activity *in vitro* against A549 cell line.[33]

Table 2. Antiproliferative activity of BIM derivatives **9-16** against lung and ovarian cancer cell lines.



Entry	Comp.	R^1	R^2	R^3	IC_{50} ovarian (μM)[a] (SKOV03)	IC_{50} lung (μM)[a] (A549)
1	**9**	Me	H	H	>50	>50
2	**10**	Et	H	H	>50	10.48±1.01
3	**11a**	^{i}Pr	H	H	11.43±0.83	1.06±0.30
4	**11b**	^{i}Pr	6-F	H	6.30±0.24	0.85±0.00
5	**11c**	^{i}Pr	5-F	H	3.52±0.37	0.58±0.09
6	**11d**	^{i}Pr	5-Me	H	7.78±0.47	0.41±0.13
7	**12a**	^{t}Bu	H	H	1.06±0.30	15.90±0.74
8	**12b**	^{t}Bu	5-F	H	0.65±0.05	11.30±0.08
9	**12c**	^{t}Bu	5-Me	H	9.01±0.58	**0.06±0.04**
10	**13a**	Bn	H	H	1.26±0.13	10.53±0.39
11	**13b**	Bn	6-F	H	7.78±0.47	0.22±0.15
12	**13c**	Bn	5-F	H	7.18±1.20	0.63±0.16
13	**13d**	Bn	5-CF_3	H	5.98±0.32	6.10±0.23
14	**13e**	Bn	6-CF_3	H	11.99±1.60	8.90±0.92
15	**13f**	Bn	5-OMe	H	6.77±1.03	1.05±0.58
16	**13g**	Bn	6-OMe	H	(>50)[b]	7.45±0.35
17	**13h**	Bn	5,6-OMe	H	>50	(≈10)[b]
18	**13i**	Bn	6-Me	H	4.90±0.36	7.16±0.89
19	**13j**	Bn	5-Me	H	**0.06±0.02**	0.93±0.13
20	**13k**	Bn	2-Me	H	>50	10.79±2.52
21	**14**	H	H	H	>50	>50
22	**15**	^{i}Pr	5-Me	Me	11.91±1.84	>50
23	**16**	^{t}Bu	5-Me	Me	>50	>50
24	**Doxorubicin**	-	-	-	0.00184±0.00022	0.48±0.017

[a] Concentration corresponding to 50% growth inhibition. [b] Approximate value due to systematic error in experimental.

With the purpose to improve further the activity of the synthesized BIM substrates, next we studied the effect of the substitution at the indole ring into their cytotoxicity against both cell lines. Although generally, the effect of fluorine on the biological activity of organic compounds is rather difficult to predict, it is well known that the introduction of fluorine substituents in bioactive molecules very often leads to increased activities [34]. For this reason, first we tested the *in vitro* cytotoxicity of fluorine substituted BIM derivatives against SKOV03 and A549 cell lines. In the case of SKOV03 cell line, high cytotoxicity is observed for 6- and 5-fluoro substituted indole moieties in *iso*-propyl esters **11b**, **11c**, *tert*-butyl esters **12b** and benzyl ester derivatives **13b** and **13c** (entries 4, 5, 8, 11 and 12). Alternatively, in the case of A549 cell lines, substitution of indole ring with fluorine atoms has a higher cytotoxic effect since IC_{50} values of 0.85±0.007 μM and 0.58±0.09 μM were revealed for *iso*-propyl ester derivatives **11b** and **11c**

(entries 4, 5), for *tert*-butyl ester derivative **12b** with an IC_{50} value of 11.30±0.08 μM (entry 8) and the values for benzyl ester derivatives **13b** and **13c** dropped to 0.22±0.15 μM and 0.63±.16 μM (entries 11,12). Surprisingly, the introduction of a trifluoromethyl electron withdrawing group in the indole ring had a low effect into the cytotoxicity of BIMs either on SKOV03 cell line nor when they were tested in A549 cell line (entries 13, 14).

Methoxy group is a strong electron donating group, when it is present in aromatic rings, that it is known to be a widespread motif in drugs and natural products. For that reason, next we studied the introduction of methoxy groups in the skeleton of indole ring in the benzyl ester BIM derivatives. Respect to the antiproliferative activity *in vitro* in SKOV03 cell line, while 5-methoxy substituted BIM substrate **13f** showed an IC_{50} value of 6.77±1.03μM (entry 15), substitution in the 6-position of the indole ring was affected with a complete loss of the cytotoxic properties for 6-methoxy and 5,6-dimethoxy BIMs **13g** and **13h** (entries 16, 17). Whereas, when the cytotoxicity of those methoxy-substituted BIM substrates was tested in A549 cell line, a slight increase in the activity was observed for 5-methoxy and 6-methoxy substituted BIMPs **13f** and **13g** with IC_{50} values of 1.05±0.58 μM and 7.45±0.35 μM.

The introduction of methyl groups into a bioactive structure makes it more lipophilic [35]. In general, the methyl group in the indole ring of our BIM substrates **11d**, **12c**, **13i-k** (entries 6, 9, 18-20) showed interesting cytotoxic effects and substitution with methyl group in 5-possition had a very significant impact into the cytotoxicity of BIM derivatives. For instance, *tert*-butyl ester derivative **12c** showed an IC_{50} value of 9.01±0.58 μM in SKOV03 cell line and an improved value of 0.06±0.04 μM was obtained in A549 cell line (entry 9). Consistently, benzyl ester derivative **13j** with a methyl group in 5-position showed an improved cytotoxicity in the nanomolar range when tested against the A549 cell line, with an IC_{50} value of 0.06±0.02 μM and a significant effect in the SKOV03 cell line with an IC_{50} value of 0.93±0.13 μM (entry 19).

Finally, in order to determine the importance of NH group in indole moiety, *N*-methylindole derivatives **15** and **16** were tested as antiproliferative agents. In this case, an increase of IC_{50} values to 11.91±1.84 μM and 11.41±2.00 μM was observed in SKOV03 cell line for *iso*-propyl ester derivative **15** and an absolute lack of toxicity in both cell lines was observed for *tert*-butyl ester derivative **16**. This result suggests a significant role of NH group in the cytotoxic activity of BIMs.

In conclusion, the new synthetic methodology proves to be very efficient for the preparation for the first time of BIMs containing a phosphonate substituent and, in addition, the use of undesirable formylphosphonates is avoided. This strategy allows the possibility of assorted structural diversity in the resultant scaffold depending on the starting phosphonate and commercial indole substrates. Moreover, obtained BIMs showed *in vitro* cytotoxicity inhibiting the growth of human tumor cell lines A549 (carcinomic human alveolar basal epithelial cell) and SKOV03 (human ovarian carcinoma) and none of the synthesized phosphorated compounds

exhibited any toxicity toward MRC-5 non-malignant lung fibroblasts. Substrates **13j** and **12c** presented very promising IC50 values of about 0.06 μM.

3. Experimental section

3.1 Chemistry

3.1.1. General experimental information

Solvents for extraction and chromatography were technical grade. All solvents used in reactions were freshly distilled from appropriate drying agents before use. All other reagents were recrystallized or distilled as necessary. All reactions were performed under an atmosphere of dry nitrogen. Analytical TLC was performed with silica gel 60 F_{254} plates. Visualization was accomplished by UV light. ^{1}H, ^{13}C, ^{31}P and ^{19}F NMR spectra were recorded on a Varian Unity Plus (at 300 MHz, 75 MHz, 120 MHz and 282 MHz respectively) and on a Bruker Avance 400 (at 400 MHz for ^{1}H, and 100 MHz for ^{13}C). Chemical shifts (δ) are reported in ppm relative to residual $CHCl_3$ (δ = 7.26 ppm for ^{1}H and δ = 77.16 ppm for ^{13}C NMR) and using phosphoric acid (50 %) as external reference (δ = 0.0 ppm) for ^{31}P NMR spectra. Coupling constants *(J)* are reported in Hertz. Data for ^{1}H NMR spectra are reported as follows: chemical shift, multiplicity, coupling constant, integration). Multiplicity abbreviations are as follows: s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet). ^{13}C NMR peak assignments were supported by Distortionless Enhanced Polarization Transfer (DEPT). High resolution mass spectra (HRMS) were obtained by positive-ion electrospray ionization (ESI). Data are reported in the form *m/z* (intensity relative to base = 100). Infrared spectra (IR) were taken in a Nicolet iS10 Termo Scientific spectrometer as neat solids. Peaks are reported in cm^{-1}.

3.1.2. Compounds Purity Analysis

All synthesized *compounds* were analyzed by HPLC to determine their purity. The analyses were performed on Agilent 1260 infinity HPLC system (C-18 column, Hypersil, BDS, 5μm, 0.4 mm×25 mm) at room temperature. All the tested compounds were dissolved in dichloromethane, and 5μL of the sample was loaded onto the column. Ethanol and heptane were used as mobile phase, and the flow rate was set at 1.0 mL/min. The maximal absorbance at the range of 190−400 nm was used as the detection wavelength. The purity of all the tested BIMP substrates **9-16** is >95%, which meets the purity requirement by the Journal.

*3.1.3. Experimental procedure and characterization data for compounds **6** and **9-16**.*

General procedure for the synthesis of starting α-aminophosphonates 6:

According to the previously described procedure [36], aqueous formaldehyde (37 %, 0.81 mL, 10 mmol) was added to a solution of tritylamine (1.30 g, 5 mmol) in toluene (20 mL) and the reaction mixture was stirred for 20 h at room temperature. Then, water was removed using a Dean-Stark and the corresponding hydrogen phosphate (6 mmol) and triethylamine (69.6 μL, 0.5 mmol) were added. The reaction was stirred under reflux in toluene overnight and the volatiles were distilled off at reduced pressure to yield the crude product as clear liquids which can be crystallized from toluene-pentane**.**

Dimethyl ((tritylamino)methyl)phosphonate 6a.[36] The general procedure was followed affording 1.81 g (95%) of **6a** as a white solid. M.p. (Toluene-pentane) = 200-204 ºC (Literature [36] 210-211°C ($CHCl_3$-MeOH)). ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.48 (d, $^{3}J_{HH}$ = 7.7 Hz, 6H), 7.34 – 7.26 (m, 6H), 7.26 – 7.17 (m, 3H), 3.86 (s, 3H), 3.83 (s, 3H), 2.54 (d, $^{2}J_{PH}$ = 13.9 Hz, 2H), 2.05 (broad s, 1H, NH). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 144.8 (C_{quat}), 128.6 (CH),

128.2 (CH), 126.7 (CH), 71.6 (d, $^{3}J_{PC}$ = 19.1 Hz, C_{quat}), 53.1 (d, $^{2}J_{PC}$ = 6.6 Hz, CH_3), 39.3 (d, $^{1}J_{PC}$ = 160.1 Hz, CH_2). ^{31}P NMR (120 MHz, $CDCl_3$) δ 30.9.

Diethyl ((tritylamino)methyl)phosphonate 6b.[36] The general procedure was followed affording 1.88 g (92%) of **6b** as a white solid. M.p. (Toluene-pentane) = 116-117 °C (Literature [36] 115-117 °C ($CHCl_3$-MeOH)). ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.49 (d, $^{3}J_{HH}$ = 8.0 Hz, 6H), 7.30 (m, 6H), 7.21 (m, 3H), 4.38 – 4.05 (m, 4H), 2.52 (d, $^{2}J_{PH}$ = 13.8 Hz, 2H), 2.04 (broad s, 1H, NH), 1.38 (t, $^{3}J_{HH}$ = 7.0 Hz, 6H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$) δ 145.0 (C_{quat}), 128.7 (CH), 128.1 (CH), 126.7 (CH), 71.5 (d, $^{3}J_{PC}$ = 19.1 Hz, C_{quat}), 62.3 (d, $^{2}J_{PC}$ = 6.6 Hz, CH_2), 39.9 (d, $^{1}J_{PC}$ = 160.2 Hz, CH_2), 16.7 (d, $^{3}J_{PC}$ = 5.9 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 28.4.

Di-iso-propyl ((tritylamino)methyl)phosphonate 6c. The general procedure was followed affording 1.99 g (91%) of **6c** as a white solid. M.p. (Toluene-pentane) = 74-76 °C. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.50 (d, $^{3}J_{HH}$ = 7.6 Hz, 6H), 7.37 – 7.26 (m, 6H), 7.26 – 7.16 (m, 3H), 4.86 – 4.70 (m, 2H), 2.46 (d, $^{2}J_{PH}$ = 14.0 Hz, 2H), 2.06 (broad s, 1H), 1.37 (d, $^{3}J_{HH}$ = 6.2 Hz, 12H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 145.1 (C_{quat}), 128.7 (CH), 128.1 (CH), 126.6 (CH), 71.5 (d, $^{3}J_{PC}$ = 19.2 Hz, C_{quat}), 70.8 (d, $^{2}J_{PC}$ = 6.8 Hz, CH), 40.7 (d, $^{1}J_{PC}$ = 161.3 Hz, CH_2), 24.3 (d, $^{3}J_{PC}$ = 5.3 Hz, CH_3), 24.2 (d, $^{3}J_{PC}$ = 6.3 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 26.4.

Di-tert-butyl ((tritylamino)methyl)phosphonate 6d. The general procedure was followed affording 2.26 g (97%) of **6d** as a colorless liquid. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.49 (d, $^{3}J_{HH}$ = 7.7 Hz, 6H), 7.31 – 7.23 (m, 6H), 7.17 (t, $^{3}J_{HH}$ = 7.2 Hz, 3H), 2.35 (dd, $^{3}J_{PH}$ = 13.5, $^{3}J_{HH}$ 8.1 Hz, 2H), 1.95 (broad s, 1H), 1.49 (s, 18H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 145.5 (C_{quat}), 128.8 (CH), 128.0 (CH), 126.5 (CH), 82.2 (d, $^{2}J_{PC}$ = 8.8 Hz, C_{quat}), 71.4 (d, $^{3}J_{PC}$ = 18.8 Hz, C_{quat}), 43.1 (d, $^{1}J_{PC}$ = 163.2 Hz, CH_2), 30.6 (d, $^{3}J_{PC}$ = 4.0 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 19.3.

Dibenzyl ((tritylamino)methyl)phosphonate 6e. The general procedure was followed affording 2.53 g (95%) of **6e** as a white solid. M.p. (Toluene-pentane) = 104-105 °C. ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.55 – 7.02 (m, 25H), 5.37 – 4.88 (m, 4H), 3.00 (m, 1H), 2.51 (d, $^{2}J_{PH}$ = 13.5 Hz, 2H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 144.9 (C_{quat}), 136.5 (d, $^{3}J_{PC}$ = 5.9 Hz, C_{quat}), 128.8 – 128.5 (CH), 128.1 (CH), 126.7 (CH), 71.5 (d, $^{3}J_{PC}$ = 19.4 Hz, C_{quat}), 67.9 (d, $^{2}J_{PC}$ = 6.7 Hz, CH_2), 40.3 (d, $^{1}J_{PC}$ = 160.5 Hz, CH_2). ^{31}P NMR (120 MHz, $CDCl_3$): δ 29.4.

General procedure for the synthesis of BIM substrates 9-13:

N-Bromosuccinimide (178 mg, 1 mmol) was added on a solution of corresponding ((tritylamino)methyl)phosphonate (1 mmol) in CCl_4 (3 mL) The mixture was stirred in quartz flask under UV light until observing the disappearance of starting aminomethylphosphonate by ^{31}P-NMR. Then, the corresponding indole (2 mmol) and phosphoric acid catalyst (0.1 mmol) in CH_2Cl_2 (10 mL) were added at room temperature and the reaction mixture was stirred for 36 h at room temperature. After that, 20 mL of water were added and the organic phase was extracted with CH_2Cl_2 (2x10 mL) and dried of in $MgSO_4$. The volatiles were distilled off at reduced pressure to yield the crude product, which was purified by column chromatography (AcOEt / Hexanes).

Dimethyl (di(1*H*-indol-3-yl)methyl)phosphonate 9. The general procedure was followed affording 209 mg (59%) of **9** as a white solid. M.p. (Et_2O) = 239-240°C. ^{1}H NMR (400 MHz, MeOH-d_4): δ 7.61 (dt, $^{3}J_{HH}$ = 8.0 Hz, $^{4}J_{HH}$ = 1.0 Hz, 2H), 7.39 (d, $^{3}J_{HH}$ = 2.7 Hz, 2H), 7.34 (dt, $^{3}J_{HH}$ = 8.1 Hz, $^{4}J_{HH}$ = 1.0 Hz, 2H), 7.08 (ddd, $^{3}J_{HH}$ = 8.1 Hz, $^{3}J_{HH}$ = 7.0 Hz, $^{4}J_{HH}$ = 1.1 Hz, 2H), 7.00 (ddd, $^{3}J_{HH}$ = 8.0Hz , $^{3}J_{HH}$ = 7.0 Hz, $^{4}J_{HH}$ = 1.0 Hz, 2H), 5.16 (d, $^{2}J_{PH}$ = 25.3 Hz, 1H), 3.52 (d, $^{3}J_{PH}$ = 10.6 Hz, 6H). ^{13}C {^{1}H} NMR (75 MHz, MeOH-d_4): δ 137.8 (C_{quat}), 128.3 (d, $^{2}J_{PC}$ = 8.7 Hz, C_{quat}), 125.4 (d, $^{3}J_{PC}$ = 6.4 Hz, CH), 122.5 (CH), 120.0 (CH), 119.6 (CH), 112.3 (CH), 111.2 (d, $^{3}J_{PC}$ = 6.1 Hz, C_{quat}), 53.9 (d, $^{2}J_{PC}$ = 7.4 Hz, CH_3), 32.8 (d, $^{1}J_{PC}$ = 143.7 Hz, CH). ^{31}P

NMR (120 MHz, $CDCl_3$:) δ 30.0. FTIR (neat) ν_{max} : 3378 (N-H), 3177 (C-H_{Ar}), 1209 (P=O), 1059 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{19}H_{19}N_2O_3P$ 354,1133, found 354,1144.

Diethyl (di(1*H*-indol-3-yl)methyl)phosphonate 10. The general procedure was followed affording 233 mg (61%) of **10** as a white solid. M.p. (Et_2O) = 144-146 °C. ^1H NMR (300 MHz, $CDCl_3$): δ 8.65 (s, 2H), 7.65 (d, $^3J_{HH}$ = 7.8 Hz, 2H), 7.39 – 7.20 (m, 4H), 7.20 – 6.93 (m, 4H), 5.07 (d, $^2J_{PH}$ = 24.9 Hz, 1H), 4.04 – 3.84 (m, 2H), 3.84 – 3.61 (m, 2H), 1.04 (t, $^3J_{HH}$ = 7.0 Hz, 6H). ^{13}C {^1H} NMR (75 MHz, $CDCl_3$): δ 136.1 (C_{quat}), 127.1 (d, $^2J_{PC}$ = 8.6 Hz, C_{quat}), 124.6 (d, $^3J_{PC}$ = 6.2 Hz, CH), 121.9 (CH), 119.4 (CH), 119.1 (CH), 111.4 (CH), 111.2 (d, $^3J_{PC}$ = 5.8 Hz, C_{quat}), 62.8 (d, $^2J_{PC}$ = 7.3 Hz, CH_2), 32.1 (d, $^1J_{PC}$ = 143.6 Hz, CH), 16.4 (d, $^3J_{PC}$ = 5.7 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$) δ 28.1. FTIR (neat) ν_{max} : 3406 (N-H), 3248 (C-H_{Ar}), 1206 (P=O), 1076 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{21}H_{23}N_2O_3P$ 382,1446, found 382,1456.

Di-*iso*-propyl (di(1*H*-indol-3-yl)methyl)phosphonate 11a. The general procedure was followed affording 246 mg (60%) of **10** as a white solid. M.p. (Et_2O) = 162-164 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 8.96 (s, 2H), 7.68 (d, $^3J_{HH}$ = 7.7 Hz, 2H), 7.31 – 7.21 (m, 4H), 7.15 – 7.04 (m, 4H), 5.04 (d, $^2J_{PH}$ = 25.2 Hz, 1H), 4.58 – 4.27 (m, 2H), 1.20 (d, $^3J_{HH}$ = 6.2 Hz, 6H), 0.72 (d, $^3J_{HH}$ = 6.2 Hz, 6H). ^{13}C {^1H} NMR (100 MHz, $CDCl_3$): δ 136.0 (C_{quat}), 128.4 (CH), 128.1 (CH), 127.3 (d, $^2J_{PC}$ = 8.7 Hz, C_{quat}), 124.6 (d, $^3J_{PC}$ = 6.0 Hz, CH), 121.6 (CH), 119.1 (CH), 111.2 (d, $^3J_{PC}$ = 5.6 Hz, C_{quat}), 71.5 (d, $^2J_{PC}$ = 7.8 Hz, CH), 32.4 (d, $^1J_{PC}$ = 145.5 Hz, CH), 24.3 (d, $^3J_{PC}$ = 3.2 Hz, CH_3), 23.2 (d, $^3J_{PC}$ = 5.3 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 26.3. FTIR (neat) ν_{max} : 3404 (N-H), 3180 (C-H_{Ar}), 1216 (P=O), 1099 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{23}H_{27}N_2O_3P$ 410,1759, found 410,1778.

Di-iso-propyl (bis(6-fluoro-1*H*-indol-3-yl)methyl)phosphonate 11b. The general procedure was followed affording 322 mg (69%) of **10** as a pale brown solid. M.p. (Et_2O) = 85-86 °C. ^1H NMR (400 MHz, $CDCl_3$): δ 8.89 (s, 2H), 7.53 (dd, $^3J_{HH}$ = 8.8 Hz, $^4J_{FH}$ = 5.3 Hz, 2H), 7.29-7.26 (m, 2H), 7.00 (dd, $^3J_{FH}$ = 9.7 Hz, $^4J_{HH}$ =2.3 Hz, 2H), 6.81 (ddd, $^3J_{FH}$ = 9.6 Hz, $^3J_{HH}$ = 8.8 Hz, $^4J_{HH}$ = 2.3 Hz, 2H), 4.91 (d, $^2J_{PH}$ = 25.4 Hz, 1H), 4.55 – 4.43 (m, 2H), 1.22 (d, $^3J_{HH}$ = 6.1 Hz, 6H), 0.77 (d, $^3J_{HH}$ = 6.2 Hz, 6H). ^{13}C {^1H} NMR (100 MHz, $CDCl_3$): δ 160.0 (d, $^1J_{FC}$ = 237.3 Hz, 2x C_{quat}), 136.08 (d, $^3J_{FC}$ = 12.7 Hz, C_{quat}), 124.56 (dd, $^3J_{PC}$ = 6.4 Hz, $^5J_{FC}$ = 3.57 Hz, CH), 124.00 (d, $^2J_{PC}$ = 8.5 Hz, C_{quat}), 120.0 (d, $^3J_{FC}$ = 10.2 Hz, CH), 111.6 (d, $^3J_{PC}$ = 5.5 Hz, C_{quat}), 108.1 (d, $^2J_{FC}$ = 24.6 Hz, CH), 97.6 (d, $^2J_{FC}$ = 26.0 Hz, CH), 71.6 (d, $^2J_{PC}$ = 7.8 Hz, CH), 32.8 (d, $^1J_{PC}$ = 146.1 Hz, CH), 24.4 (d, $^3J_{PC}$ = 3.3 Hz, CH_3), 23.4 (d, $^3J_{PC}$ = 5.3 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 24.9. ^{19}F NMR (282 MHz, $CDCl_3$): δ -121.9. FTIR (neat) ν_{max} : 3398 (N-H), 3216 (C-H_{Ar}), 1218 (P=O), 1100 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 446,1571, found 466,1573.

Di-iso-propyl (bis(5-fluoro-1*H*-indol-3-yl)methyl)phosphonate 11c. The general procedure was followed affording 299 mg (67%) of **10** as a pale brown solid. M.p. (Et_2O) = 88-89 °C (dec.). ^1H NMR (400 MHz, $CDCl_3$): δ 9.11 (d, $^3J_{HH}$ = 1.7 Hz, 2H), 7.30 – 7.24 (m, 4H,), 7.15 (dd, $^3J_{HH}$ = 8.8 Hz, $^4J_{FH}$ = 4.4 Hz, 2H), 6.85 (td, $^3J_{FH}$ = $^3J_{HH}$ = 9.1 Hz, $^4J_{HH}$ = 2.5 Hz, 2H), 4.82 (d, $^2J_{PH}$ = 25.3 Hz, 1H), 4.57 – 4.46 (m, 2H), 1.22 (d, $^3J_{HH}$ = 6.2 Hz, 6H), 0.79 (d, $^3J_{HH}$ = 6.2 Hz, 6H). ^{13}C {^1H} NMR (100 MHz, $CDCl_3$): δ 157.9 (d, $^1J_{FC}$ = 234.3 Hz, C_{quat}), 132.7 (C_{quat}), 127.7 (m, C_{quat}),126.1 (d, $^3J_{PC}$ = 6.0 Hz, CH), 112.0 (d, $^3J_{FC}$ = 9.5 Hz, CH), 111.7 (m, C_{quat}), 110.5 (d, $^2J_{FC}$ = 26.3 Hz, CH), 104.3 (d, $^2J_{FC}$ = 23.8 Hz, CH), 71.7 (d, $^2J_{PC}$ = 7.7 Hz, CH), 33.0 (d, $^1J_{PC}$ = 145.9 Hz, CH), 24.3 (d, $^3J_{PC}$ = 3.3 Hz, CH_3), 23.4 (d, $^3J_{PC}$ = 5.1 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 24.8. ^{19}F NMR (282 MHz, $CDCl_3$): δ -125.1. FTIR (neat) ν_{max} : 3419 (N-H), 3213 (C-H_{Ar}), 1218 (P=O), 1101 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 446,1571, found 446,1573.

Di-iso-propyl (bis(5-methyl-1*H*-indol-3-yl)methyl)phosphonate 11d. The general procedure was followed affording 272 mg (62%) of **10** as a brown solid. M.p. (Et_2O) = 92-94 °C (dec.). ^1H

NMR (300 MHz, $CDCl_3$): δ 8.34 (s, 2H), 7.48 (s, 2H), 7.34-7.29 (m, 2H), 7.18 (d, $^3J_{HH}$ = 8.3 Hz, 2H), 6.96 (dd, $^3J_{HH}$ = 8.1, 1.1 Hz, 2H), 4.94 (d, $^2J_{PH}$ = 25.2 Hz, 1H), 4.57 – 4.42 (m, 2H), 2.42 (s, 6H), 1.22 (d, $^3J_{HH}$ = 6.2 Hz, 6H), 0.77 (d, $^3J_{HH}$ = 6.2 Hz, 6H). ^{13}C {^1H} NMR (75 MHz, $CDCl_3$): δ 134.5 (C_{quat}), 128.4 (C_{quat}), 127.7 (d, $^2J_{PC}$ = 8.5 Hz, C_{quat}), 124.5 (d, $^3J_{PC}$ = 5.9 Hz, CH), 123.5 (CH), 119.1 (CH), 111.5 (d, $^3J_{PC}$ = 5.6 Hz, C_{quat}), 110.8 (CH), 71.2 (d, $^2J_{PC}$ = 7.7 Hz, CH), 32.6 (d, $^1J_{PC}$ = 145.3 Hz, CH), 24.5 (d, $^3J_{PC}$ = 2.8 Hz, CH_3), 23.4 (d, $^3J_{PC}$ = 5.3 Hz, CH_3), 21.7 (CH_3). ^{31}P NMR (120 MHz, $CDCl_3$) δ 25.5. FTIR (neat) ν_{max} : 3401 (N-H), 3245 (C-H_{Ar}), 1380 (C-H_{Me}), 1218 (P=O), 1097 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 438,2072, found 438,2067.

Di-*tert*-butyl (di(1*H*-indol-3-yl)methyl)phosphonate 12a. The general procedure was followed affording 298 mg (68%) of **12a** as a white solid. M.p. (Et_2O) = 162-164 °C (dec.). ^1H NMR (300 MHz, $CDCl_3$): δ 8.66 (s, 2H), 7.66 (d, $^3J_{HH}$ = 7.8 Hz, 2H), 7.39 – 7.28 (m, 4H), 7.18 – 6.98 (m, 4H), 4.95 (d, $^2J_{PH}$ = 25.6 Hz, 1H), 1.20 (s, 18H). ^{13}C {^1H} NMR (75 MHz, $CDCl_3$): δ 136.0 (C_{quat}), 127.8 (d, $^2J_{PC}$ = 8.4 Hz, C_{quat}), 124.6 (d, $^3J_{PC}$ = 6.1 Hz, CH), 121.7 (CH), 119.5 (CH), 119.2 (CH), 112.6 (d, $^3J_{PC}$ = 5.6 Hz, C_{quat}), 111.2 (CH), 82.7 (d, $^2J_{PC}$ = 10.5 Hz, C_{quat}), 35.1 (d, $^1J_{PC}$ = 149.4 Hz, CH), 30.3 (d, $^3J_{PC}$ = 3.8 Hz, CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 18.7. FTIR (neat) ν_{max} : 3416 (N-H), 3219 (C-H_{Ar}), 1224 (P=O), 1168 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{25}H_{31}N_2O_3P$ 438,2072, found 438,2090.

Di-tert-butyl (bis(5-fluoro-1*H*-indol-3-yl)methyl)phosphonate 12b. The general procedure was followed affording 327 mg (69%) of **12b** as a pale brown solid. M.p. (Et_2O) = 145-147 °C (dec.). ^1H NMR (300 MHz, Acetone-d_6): δ 10.39 (s, 2H), 7.60 (t, $^3J_{HH}$ = 2.4 Hz, 2H,), 7.42 (dd, $^3J_{FH}$ = 10.3, $^4J_{HH}$ = 2.5 Hz, 2H), 7.34 (dd, $^3J_{HH}$ = 8.8 Hz, $^4J_{FH}$ = 4.5 Hz, 2H), 6.83 (td, $^3J_{FH}$ = $^3J_{HH}$ = 9.1 Hz, $^4J_{HH}$ = 2.5 Hz, 2H), 4.88 (d, $^2J_{PH}$ = 25.7 Hz, 1H), 1.28 (s, 18H). ^{13}C {^1H} NMR (75 MHz, Acetone-d_6): δ 158.2 (d, $^1J_{FC}$ = 231.1 Hz, C_{quat}), 133.9 (C_{quat}), 129.1 (d, $^2J_{PC}$ = 10.1 Hz, C_{quat}), 127.5 (dd, $^3J_{PC}$ = 6.3 Hz, $^5J_{FC}$ = 3.2 Hz, CH), 113.2 (m, C_{quat}), 112.9 (d, $^3J_{FC}$ = 9.6 Hz, CH), 110.0 (d, $^2J_{FC}$ = 26.4 Hz, CH), 105.1 (d, $^2J_{FC}$ = 23.9 Hz, CH), 82.5 (d, $^2J_{PC}$ = 9.9 Hz, C_{quat}), 36.9 (d, $^1J_{PC}$ = 149.7 Hz, CH), 30.6 (d, $^3J_{PC}$ = 3.8 Hz, CH_3). ^{31}P NMR (120 MHz, Acetone-d_6): δ 18.9. ^{19}F NMR (282 MHz, Acetone-d_6): δ -127.5. FTIR (neat) ν_{max} : 3412 (N-H), 1217 (P=O), 1103 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{25}H_{29}F_2N_2O_3P$ 474,1884, found 474,1886.

Di-tert-butyl (bis(5-methyl-1*H*-indol-3-yl)methyl)phosphonate 12c. The general procedure was followed affording 336 mg (72%) of **12c** as a white solid. M.p. (Et_2O) = 194-195 °C (dec.). ^1H NMR (300 MHz, Acetone-d_6): δ 10.25 (s, 2H), 7.59 – 7.52 (m, 2H), 7.47 (t, $^3J_{HH}$ = 2.7 Hz, 2H), 7.27 (d, $^3J_{HH}$ = 8.2 Hz, 2H), 6.92 (dd, $^3J_{HH}$ = 8.3 Hz, $^3J_{HH}$ = 1.5 Hz, 2H), 4.96 (d, $^2J_{PH}$ = 26.0 Hz, 1H), 2.41 (s, 6H), 1.27 (s, 18H). ^{13}C {^1H} NMR (75 MHz, Acetone-d_6): δ 135.5 (C_{quat}), 128.9 (d, $^2J_{PC}$ = 8.0 Hz, C_{quat}), 127.80 (C_{quat}), 125.6 (d, $^3J_{PC}$ = 6.2 Hz, CH), 123.4 (CH), 119.7 (CH), 112.4 (d, $^3J_{PC}$ = 5.7 Hz, C_{quat}), 111.7 (CH), 82.4 (d, $^2J_{PC}$ = 10.3 Hz, C_{quat}), 36.1 (d, $^1J_{PC}$ = 149.7 Hz, CH), 30.5 (d, $^3J_{PC}$ = 3.8 Hz, CH_3), 21.8 (CH_3). ^{31}P NMR (120 MHz, Acetone-d_6): δ 19.5. FTIR (neat) ν_{max}: 3407 (N-H), 1442 (C-H_{Me}), 1227 (P=O), 1106 (P-O-C) cm^{-1}. HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 466,2385, found 466,2392.

Dibenzyl (di(1*H*-indol-3-yl)methyl)phosphonate 13a. The general procedure was followed affording 273 mg (55%) of **13a** as a white solid. M.p. (Et_2O) = 131-132 °C (dec.). ^1H NMR (400 MHz, $CDCl_3$): δ 8.24 (s, 2H), 7.64 (d, $^3J_{HH}$ = 8.0 Hz, 2H), 7.41 (t, $^3J_{HH}$ = 2.4 Hz, 2H), 7.34 (d, $^3J_{HH}$ = 8.1 Hz, 2H), 7.25 – 7.14 (m, 8H), 7.10 – 7.01 (m, 6H), 5.15 (d, $^2J_{PH}$ = 25.2 Hz, 1H), 4.88 (dd, $^2J_{HH}$ = 11.8 Hz, $^3J_{PH}$ = 7.3 Hz, 2H), 4.62 (dd, $^2J_{HH}$ = 11.8 Hz, $^3J_{PH}$ = 8.7 Hz, 2H). ^{13}C {^1H} NMR (100 MHz, $CDCl_3$): δ 136.6 (d, $^3J_{PC}$ = 5.8 Hz, C_{quat}), 136.1 (C_{quat}), 128.4 (CH), 128.2 (CH), 128.0 (CH), 127.1 (d, $^2J_{PC}$ = 8.9 Hz, C_{quat}), 124.4 (d, $^3J_{PC}$ = 6.4 Hz, CH), 122.2 (CH), 119.7 (CH), 119.3 (CH), 111.5 (d, $^3J_{PC}$ = 5.4 Hz, C_{quat}), 111.3 (CH), 68.2 (d, $^2J_{PC}$ = 7.3 Hz, CH_2), 32.6 (d, $^1J_{PC}$ = 143.5 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 28.9. FTIR (neat) ν_{max}:

3399 (N-H), 3172 (C-H_{Ar}), 1218 (P=O), 1092 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{27}N_2O_3P$ 506,1759, found 506,1758.

Dibenzyl (bis(6-fluoro-1*H*-indol-3-yl)methyl)phosphonate 13b. The general procedure was followed affording 352 mg (65%) of **13b** as a pale orange solid. M.p. (Et_2O) = 60-62 °C (dec.). ^{1}H NMR (300 MHz, $CDCl_3$): δ 9.00 (s, 2H), 7.47 (dd, $^3J_{FH}$ = 8.6 Hz, $^3J_{HH}$ =5.3 Hz, 2H), 7.30 – 7.11 (m, 8H,), 7.01 (d, $^3J_{HH}$ = 7.5 Hz, 4H), 6.94 (d, $^3J_{HH}$ = 9.6 Hz, 2H), 6.83-6.73 (m, 2H), 5.06 (d, $^2J_{PH}$ = 25.1 Hz, 1H), 4.84 (dd, $^2J_{HH}$ = 11.6 Hz, $^3J_{PH}$ = 7.4 Hz, 2H), 4.63 (dd, $^2J_{HH}$ = 11.6 Hz, $^3J_{PH}$ = 9.1 Hz, 2H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 160.0 (d, $^1J_{FC}$ = 237.4 Hz, C_{quat}), 136.1 (d, $^3J_{PC}$ = 8.5 Hz, C_{quat}), 136.0 (d, $^3J_{FC}$ = 9.8 Hz, C_{quat}), 128.5 (m, CH), 128.3 (m, CH), 127.9 (m, CH), 124.9 (dd, $^3J_{PC}$ = 6.5 Hz, $^5J_{FC}$ = 3.3 Hz, CH), 123.5 (d, $^3J_{PC}$ = 8.4 Hz, C_{quat}), 119.8 (d, $^3J_{PC}$ = 10.0 Hz, CH), 110.6 (d, $^3J_{PC}$ = 6.0 Hz, C_{quat}), 108.3 (d, $^2J_{FC}$ = 24.5 Hz, CH), 97.8 (d, $^2J_{FC}$ = 25.8 Hz, CH), 68.4 (d, $^2J_{PC}$ = 7.4 Hz, CH_2), 32.7 (d, $^1J_{PC}$ = 143.3 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 27.6. ^{19}F NMR (282 MHz, $CDCl_3$): δ -121.6. FTIR (neat) ν_{max}: 3431 (N-H), 1224 (P=O), 1098 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 542,1571, found 542,1561.

Dibenzyl (bis(5-fluoro-1H-indol-3-yl)methyl)phosphonate 13c. The general procedure was followed affording 363 mg (67%) of **13c** as a pale orange solid. M.p. (Et_2O) = 87-89 °C (dec.). ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.84 (s, 2H), 7.54 – 7.04 (m, 12H), 6.96 (d, $^3J_{HH}$ = 6.9 Hz, 4H), 6.90-6.79 (m, 2H), 4.91 (d, $^2J_{PH}$ = 25.0 Hz, 1H), 4.73 (dd, $^2J_{HH}$ = 11.3 Hz, $^3J_{PH}$ = 7.4 Hz, 2H), 4.51 (m, 2H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 157.9 (d, $^1J_{FC}$ = 234.6 Hz, C_{quat}), 143.8 (C_{quat}), 135.9 (d, $^3J_{PC}$ = 5.9 Hz, C_{quat}), 132.5 (C_{quat}), 128.5 (CH), 128.4 (CH), 127.9 (CH), 127.2 (m, C_{quat}), 126.6 (d, $^3J_{PC}$ = 5.9 Hz, CH), 112.3 (d, $^3J_{FC}$ = 9.2 Hz, CH), 110.5 (d, $^2J_{FC}$ = 26.6 Hz, CH), 103.8 (d, $^2J_{FC}$ = 23.7 Hz, CH), 68.4 (d, $^2J_{PC}$ = 7.4 Hz, CH_2), 32.4 (d, $^1J_{PC}$ = 143.3 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 27.0. ^{19}F NMR (282 MHz, $CDCl_3$): δ -124.8. FTIR (neat) ν_{max}: 3432 (N-H), 1212 (P=O), 1105 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 542,1571, found 542,1561.

Dibenzyl (bis(5-(trifluoromethyl)-1*H*-indol-3-yl)methyl)phosphonate 13d. The general procedure was followed affording 347 mg (54%) of **13d** as a pale orange solid. M.p. (Et_2O) = 107-108 °C (dec.). ^{1}H NMR (400 MHz, $CDCl_3$): δ 9.27 (s, 2H), 7.91 (s, 2H), 7.40 – 7.13 (m, 12H), 7.00 (d, $^3J_{HH}$ = 6.9 Hz, 4H), 5.07 (d, $^2J_{PH}$ = 24.9 Hz, 1H), 4.87 (dd, $^2J_{HH}$ = 11.4 Hz, $^3J_{PH}$ = 8.2 Hz, 2H), 4.71 – 4.63 (m, 2H). ^{13}C {^{1}H} NMR (100 MHz, $CDCl_3$): δ 137.5 (C_{quat}), 135.7 (d, $^3J_{PC}$ = 5.4 Hz, C_{quat}), 128.3 (CH), 128.1 (CH), 128.0 (CH) , 126.3 (d, $^3J_{PC}$ = 6.4 Hz, CH), 126.1 (d, $^2J_{PC}$ = 8.0 Hz, C_{quat}), 125.4 (q, $^1J_{FC}$ = 271.5 Hz, C_{quat}), 122.1 (q, $^2J_{FC}$ = 31.8 Hz, C_{quat}), 119.0 (q, $^3J_{FC}$ = 3.3 Hz, CH), 116.8 (q, $^3J_{FC}$ = 4.5 Hz, CH), 111.9 (CH), 111.2 (d, $^3J_{PC}$ = 6.0 Hz, C_{quat}), 68.7 (d, $^2J_{PC}$ = 7.5 Hz, CH_2), 32.6 (d, $^1J_{PC}$ = 142.4 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 26.5. ^{19}F NMR (282 MHz, $CDCl_3$): δ -60.6. FTIR (neat) ν_{max}: 3423 (N-H), 1256 (P=O), 1101 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{33}H_{25}F_6N_2O_3P$ 642,1507, found 642,1487.

Dibenzyl (bis(6-(trifluoromethyl)-1*H*-indol-3-yl)methyl)phosphonate 13e. The general procedure was followed affording 340 mg (53%) of **13e** as a pale orange solid. M.p. (Et_2O) = 96-97 °C (dec.). ^{1}H NMR (400 MHz, $CDCl_3$): δ 9.30 (s, 2H), 7.63 – 7.59 (m, 4H), 7.35 – 7.22 (m, 4H), 7.21 – 7.13 (m, 6H), 7.03 – 6.97 (d, $^3J_{HH}$ = 6.7 Hz, 4H), 5.11 (d, $^2J_{PH}$ = 25.2 Hz, 1H), 4.87 (dd, $^2J_{HH}$ = 11.7 Hz, $^3J_{PH}$ = 7.7 Hz, 2H), 4.71 (dd, $^2J_{HH}$ = 11.7 Hz, $^3J_{PH}$ = 9.3 Hz, 2H). ^{13}C {^{1}H} NMR (100 MHz, $CDCl_3$): δ 135.8 (d, $^3J_{PC}$ = 5.7 Hz, C_{quat}), 135.0 (C_{quat}), 129.0 (d, $^2J_{PC}$ = 9.3 Hz, C_{quat}), 128.3 (CH), 128.1 (CH), 128.0 (CH), 127.1 (d, $^3J_{PC}$ = 6.2 Hz, CH), 125.3 (q, $^1J_{FC}$ = 271.6 Hz, C_{quat}), 124.3 (q, $^2J_{FC}$ = 31.9 Hz, C_{quat}), 119.5 (CH), 116.4 (q, $^3J_{FC}$ = 3.5 Hz, CH), 110.9 (d, $^3J_{PC}$ = 5.8 Hz, C_{quat}), 109.2 (q, $^3J_{FC}$ = 4.4 Hz, CH), 68.6 (d, $^2J_{PC}$ = 7.5 Hz, CH_2), 32.6 (d, $^1J_{PC}$ = 143.9 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$) δ 26.9. ^{19}F NMR (282 MHz, $CDCl_3$) δ -

61.0. FTIR (neat) ν_{max}: 3419 (N-H), 1260 (P=O), 1097 (P-O-C) cm^{-1}. HRMS (Q-TOF) m/z calcd for $C_{33}H_{25}F_6N_2O_3P$ 642,1507, found 642,1496.

Dibenzyl (bis(5-methoxy-1*H*-indol-3-yl)methyl)phosphonate 13f. The general procedure was followed affording 323 mg (57%) of **13f** as a brown solid. M.p. (Et_2O) = 140-142 °C (dec.). ^1H NMR (400 MHz, $CDCl_3$): δ 8.32 (s, 2H), 7.41 – 7.34 (m, 2H), 7.24 – 7.10 (m, 8H), 7.08 – 6.96 (m, 6H), 6.82 (dd, $^3J_{HH}$ = 8.7 Hz, $^4J_{HH}$ = 2.5 Hz, 2H), 5.03 (d, $^2J_{PH}$ = 25.1 Hz, 1H), 4.86 (dd, $^2J_{HH}$ = 11.6 Hz, $^3J_{PH}$ = 7.0 Hz, 2H), 4.60 (dd, $^2J_{HH}$ = 11.6 Hz, $^3J_{PH}$ = 8.4 Hz, 2H), 3.70 (s, 6H). ^{13}C {^1H} NMR (75 MHz, $CDCl_3$): δ 154.1 (C_{quat}), 136.4 (d, $^3J_{PC}$ = 6.3 Hz, C_{quat}), 131.3 (C_{quat}), 128.4 (CH), 128.2 (CH), 128.0 (CH), 127.5 (d, $^2J_{PC}$ = 8.4 Hz, C_{quat}), 125.4 (d, $^3J_{PC}$ = 6.1 Hz, CH), 112.4 (CH), 112.2 (CH), 110.7 (d, $^3J_{PC}$ = 5.8 Hz, C_{quat}), 101.0 (CH), 68.3 (d, $^2J_{PC}$ = 7.3 Hz, CH_2), 55.9 (CH_3), 32.1 (d, $^1J_{PC}$ = 142.7 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 27.7. FTIR (neat) ν_{max}: 3425 (N-H), 3189 (C-H_{Ar}), 1452 (C-H_{OMe}), 1218 (P=O). HRMS (Q-TOF) m/z calcd for $C_{33}H_{31}N_2O_5P$ 566,1971, found 566,1965.

Dibenzyl (bis(6-methoxy-1*H*-indol-3-yl)methyl)phosphonate 13g. The general procedure was followed affording 345 mg (61%) of **13g** as a brown solid. M.p. (Et_2O) = 109-110 °C (dec.). ^1H NMR (400 MHz, $CDCl_3$): δ 8.37 (s, 2H), 7.47 (d, $^3J_{HH}$ = 9.28 Hz, 2H), 7.29 – 7.26 (m, 4H), 7.21 – 7.17 (m, 4H), 7.03 – 6.98 (m, 4H), 6.72 – 6.68 (m, 4H), 5.04 (d, $^2J_{PH}$ = 25.1 Hz, 1H), 4.82 (dd, $^2J_{HH}$ = 11.7 Hz, $^3J_{PH}$ = 7.1 Hz, 2H), 4.56 (dd, $^2J_{HH}$ = 11.7 Hz, $^3J_{PH}$ = 8.6 Hz, 2H), 3.75 (s, 6H). ^{13}C {^1H} NMR (101 MHz, $CDCl_3$): δ 156.5 (C_{quat}), 136.8 (C_{quat}), 136.4 (d, $^3J_{PC}$ = 6.0 Hz, C_{quat}), 128.4 (CH), 128.1 (CH), 128.0 (CH), 123.5 (d, $^3J_{PC}$ = 6.6 Hz, CH), 121.5 (d, $^2J_{PC}$ = 8.7 Hz, C_{quat}), 119.7 (CH), 110.8 (d, $^3J_{PC}$ = 6.1 Hz, C_{quat}), 109.7 (CH), 94.7 (CH), 68.2 (d, $^2J_{PC}$ = 7.3 Hz, CH_2), 55.7 (CH_3), 32.7 (d, $^1J_{PC}$ = 142.7 Hz, CHP). ^{31}P NMR (120 MHz, $CDCl_3$): δ 27.9. FTIR (neat) ν_{max}: 3422 (N-H), 3245 (C-H_{Ar}), 1448 (C-H_{OMe}), 1194 (P=O), 1059 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{33}H_{31}N_2O_5P$ 566,1971, found 566,1968.

Dibenzyl (bis(5,6-dimethoxy-1*H*-indol-3-yl)methyl)phosphonate 13h. The general procedure was followed affording 363 mg (58%) of **13h** as a brown solid. M.p. (Et_2O) = 134-136 °C (dec.). ^1H NMR (400 MHz, DMSO-d_6): δ 10.66 (s, 2H), 7.37 – 7.29 (m, 2H), 7.27 – 7.20 (m, 4H), 7.14 – 7.04 (m, 8H), 6.86 (s, 2H), 5.19 (d, $^2J_{PH}$ = 25.3 Hz, 1H), 4.87 (dd, $^2J_{HH}$ = 12.1 Hz, $^3J_{PH}$ = 6.5 Hz, 2H), 4.70 (dd, $^2J_{HH}$ = 12.1 Hz, $^3J_{PH}$ = 7.8 Hz, 2H), 3.73 (s, 6H), 3.65 (s, 6H). ^{13}C {^1H} NMR (75 MHz, $CDCl_3$): δ 147.1 (C_{quat}), 145.0 (C_{quat}), 136.2 (d, $^3J_{PC}$ = 6.4 Hz, C_{quat}), 130.2 (C_{quat}), 128.6 (CH), 128.4 (CH), 127.9 (CH), 123.7 (d, $^3J_{PC}$ = 5.9 Hz, CH), 119.7 (d, $^2J_{PC}$ = 7.9 Hz, C_{quat}), 110.0 (d, $^3J_{PC}$ = 6.0 Hz, C_{quat}), 100.6 (CH), 94.7 (CH), 68.3 (d, $^2J_{PC}$ = 7.3 Hz, CH_2), 56.3 (CH_3), 56.1 (CH_3), 32.6 (d, $^1J_{PC}$ = 142.2 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 27.4. FTIR (neat) ν_{max}: 3401 (N-H), 1445(C-H_{OMe}), 1203 (P=O), 1150 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{35}H_{35}N_2O_7P$ 626,2182, found 626,2168.

Dibenzyl (bis(6-methyl-1*H*-indol-3-yl)methyl)phosphonate 13i. The general procedure was followed affording 342 mg (64%) of **13i** as a pale brown solid. M.p. (Et_2O) = 81-83 °C (dec.). ^1H NMR (300 MHz, $CDCl_3$): δ 8.23 (s, 2H), 7.49 (d, $^3J_{HH}$ = 8.1 Hz, 2H), 7.29 – 7.13 (m, 8H), 7.10 – 6.98 (m, 6H), 6.87 (d, $^3J_{HH}$ = 8.1 Hz, 2H), 5.09 (d, $^2J_{PH}$ = 25.1 Hz, 1H), 4.86 (dd, $^2J_{HH}$ = 11.7 Hz, $^3J_{PH}$ = 7.1 Hz, 2H), 4.60 (dd, $^2J_{HH}$ = 11.7 Hz, $^3J_{PH}$ = 8.6 Hz, 2H), 2.42 (s, 6H). ^{13}C {^1H} NMR (75 MHz, $CDCl_3$): δ 136.6 (C_{quat}), 131.8 (C_{quat}), 128.4 (CH), 128.1 (CH), 128.0 (CH), 125.0 (d, $^2J_{PC}$ = 8.8 Hz, C_{quat}), 123.9 (d, $^3J_{PC}$ = 6.3 Hz, CH), 121.4 (CH), 118.9 (CH), 111.3 (CH), 111.0 (d, $^3J_{PC}$ = 5.8 Hz, C_{quat}), 68.2 (d, $^2J_{PC}$ = 6.9 Hz, CH_2), 32.7 (d, $^1J_{PC}$ = 142.8 Hz, CH), 21.8 (CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 27.9. FTIR (neat) ν_{max}: 3430 (N-H), 1454 (C-H_{Me}), 1222 (P=O). HRMS (Q-TOF) m/z calcd for $C_{33}H_{31}N_2O_3P$ 534,2072, found 534,2072.

Dibenzyl (bis(5-methyl-1*H*-indol-3-yl)methyl)phosphonate 13j. The general procedure was followed affording 347 mg (65%) of **13j** as a white solid. M.p. (Et_2O) = 185-187 °C (dec.). ^1H NMR (300 MHz, $CDCl_3$): δ 8.43 (s, 2H), 7.43 (s, 2H), 7.32 – 7.27 (m, 2H), 7.25 – 7.13 (m, 8H),

7.06 – 6.94 (m, 6H), 5.11 (d, $^{2}J_{PH}$ = 25.1 Hz, 1H), 4.89 (dd, $^{2}J_{HH}$ = 11.7 Hz, $^{3}J_{PH}$ = 7.0 Hz, 2H), 4.59 (dd, $^{2}J_{HH}$ = 11.7 Hz, $^{3}J_{PH}$ = 8.1 Hz, 2H), 2.38 (s, 6H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 136.6 (d, $^{3}J_{PC}$ = 6.3 Hz, C_{quat}), 134.5 (C_{quat}), 128.7 (C_{quat}), 128.4 (CH), 128.1 (CH), 127.97 (CH), 127.30 (d, $^{2}J_{PC}$ = 8.4 Hz, C_{quat}), 124.81 (d, $^{3}J_{PC}$ = 6.2 Hz, CH), 123.72 (CH), 118.84 (CH), 111.05 (CH), 110.52 (d, $^{3}J_{PC}$ = 5.9 Hz, C_{quat}), 68.20 (d, $^{2}J_{PC}$ = 7.2 Hz, CH_2), 32.47 (d, $^{1}J_{PC}$ = 142.7 Hz, CH), 21.65 (CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 28.0. FTIR (neat) ν_{max}: 3431 (N-H), 1451 (C-H_{Me}), 1221 (P=O). HRMS (Q-TOF) m/z calcd for $C_{33}H_{31}N_2O_3P$ 534,2072, found 534,2068.

Dibenzyl (bis(2-methyl-1*H*-indol-3-yl)methyl)phosphonate 13k. The general procedure was followed affording 337 mg (63%) of **13k** as a pale brown solid. M.p. (Et_2O) = 212-213 °C (dec.). ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.87 (m, 4H), 7.32 – 7.13 (m, 8H), 7.12 – 6.97 (m, 8H), 5.11 (d, $^{2}J_{PH}$ = 30.0 Hz, 1H), 4.85 (dd, $^{2}J_{HH}$ = 11.6 Hz, $^{3}J_{PH}$ = 7.5 Hz, 2H), 4.72-4.61 (m, 2H), 2.19 (s, 3H). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 136.6 (d, $^{3}J_{PC}$ = 5.9 Hz, C_{quat}), 134.9 (C_{quat}), 133.6 (d, $^{3}J_{PC}$ = 10.1 Hz, C_{quat}), 128.6 (d, $^{2}J_{PC}$ = 6.7 Hz, C_{quat}), 128.4 (CH), 128.2 (CH), 128.1 (CH), 120.9 (CH), 119.55 (CH), 119.5 (CH), 110.3 (CH), 106.6 (d, $^{3}J_{PC}$ = 3.6 Hz, C_{quat}), 67.9 (d, $^{2}J_{PC}$ = 7.3 Hz, CH_2), 33.2 (d, $^{1}J_{PC}$ = 146.2 Hz, CH), 12.8 (CH_3). ^{31}P NMR (120 MHz, $CDCl_3$): δ 28.4. FTIR (neat) ν_{max}: 3396 (N-H), 1451 (C-H_{Me}), 1227 (P=O). HRMS (Q-TOF) m/z calcd for $C_{33}H_{31}N_2O_3P$ 534,2072, found 534,2073.

Synthesis of (di(1*H*-indol-3-yl)methyl)phosphonic acid 14. Di-tert-butyl ((tritylamino)methyl)phosphonate (438.5 mg, 1 mmol) was diluted in THF (3 mL) and HCl 2M (3 mL) were added. The reaction mixture was stirred at room temperature until the starting material was consumed (monitored by TLC). The mixture was diluted in CH_2Cl_2 and the combined organic phases were collected, dried with anhydrous $MgSO_4$ and concentrated at reduced pressure to yield the crude product which was crystallized in $CHCl_3$-MeOH to afford 323 mg (99%) as a pink solid. M.p. ($CHCl_3$-MeOH) = 217-219 °C (dec.). ^{1}H NMR (400 MHz, MeOH-d_4): δ 7.52 (d, $^{3}J_{HH}$ = 7.9 Hz, 2H), 7.33 (d, $^{3}J_{HH}$ = 2.2 Hz, 2H), 7.26 (d, $^{3}J_{HH}$ = 8.1 Hz, 1H), 7.00 (ddd, $^{3}J_{HH}$ = 8.1 Hz, $^{3}J_{HH}$ =7.1 Hz, $^{4}J_{HH}$ = 1.1 Hz, 2H), 6.90 (ddd, $^{3}J_{HH}$ = 7.9 Hz, $^{3}J_{HH}$ = 7.1 Hz, $^{4}J_{HH}$ =1.0 Hz, 2H), 4.95 (d, $^{1}J_{PH}$ = 24.8 Hz, 1H). ^{13}C {^{1}H} NMR (75 MHz, MeOH-d_4): δ 137.8 (C_{quat}), 128.8 (d, $^{2}J_{PC}$ = 8.5 Hz, C_{quat}), 125.1 (d, $^{3}J_{PC}$ = 5.9 Hz, CH), 122.2 (CH), 119.9 (CH), 119.6 (CH), 113.2 (d, $^{3}J_{PC}$ = 5.0 Hz, C_{quat}), 112.1 (CH), 34.7 (d, $^{1}J_{PC}$ = 141.5 Hz, CH). ^{31}P NMR (120 MHz, $CDCl_3$): δ 26.4. FTIR (neat) ν_{max}: 3431 (N-H), 1216 (P=O). HRMS (Q-TOF) m/z calcd for $C_{17}H_{15}N_2O_3P$ 326,0820, found 326,0811.

General procedure for the synthesis *N*-methylated substrates 15, 16:

The corresponding dialkyl bis(1*H*-indol-3-yl)methyl phosphonate (0.5 mmol) was diluted in freshly distilled DMF (3 mL) under nitrogen atmosphere and the reaction mixture was cooled to 0ºC. NaH (36.0 mg, 1.5 mmol) were added and the reaction was stirred at room temperature for 1h. Then, the reaction was cooled to 0ºC, methyl iodide (0.12 mL, 2 mmol) was added and stirring was continued at room temperature for 4h. The reaction mixture was quenched with saturated NH_4Cl solution (15 mL) and diluted in 10 mL of Et_2O. DMF was removed by multiple washing with saturated NH_4Cl solution (10x15 mL). The organic phase was dried of in $MgSO_4$ and concentrated at reduced pressure to yield the pure product in quantitative yield.

Di-iso-propyl (bis(1,5-dimethyl-1*H*-indol-3-yl)methyl)phosphonate 15. The general procedure was followed affording 231 mg (99%) of **15** as a pale brown solid. M.p. (Et_2O) = 78-80 °C (dec.). ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.50 – 7.46 (m, 2H), 7.34 – 7.22 (m, 2H), 7.14 (d, $^{3}J_{HH}$ = 8.3 Hz, 2H), 7.00 (dd, $^{3}J_{HH}$ = 8.3 Hz, $^{4}J_{HH}$ = 1.6 Hz, 2H), 4.91 (d, $^{2}J_{PH}$ = 25.2 Hz, 1H), 4.55 – 4.44 (m, 2H), 3.70 (s, 6H), 2.44 (s, 6H), 1.22 (d, $^{3}J_{HH}$ = 6.2 Hz, 6H), 0.78 (d, $^{3}J_{HH}$ = 6.2 Hz, 6H). ^{13}C {^{1}H} NMR (100 MHz, $CDCl_3$): δ 135.3 (C_{quat}), 128.8 (d, $^{3}J_{PC}$ = 6.1 Hz, CH), 128.1 (d, $^{2}J_{PC}$ = 8.7 Hz, C_{quat}), 128.0 (C_{quat}), 123.2 (CH), 119.3 (CH), 110.2 (d, $3^{2}J_{PC}$ = 5.4 Hz, C_{quat}),

108.8 (CH), 71.1 (d, $^{2}J_{PC}$ = 7.5 Hz, CH), 33.0 (CH_3), 32.4 (d, $^{1}J_{PC}$ = 145.2 Hz, CH), 24.5 (d, $^{3}J_{PC}$ = 2.7 Hz, CH_3), 23.4 (d, $^{3}J_{PC}$ = 5.5 Hz, CH_3), 21.7 (CH_3). ^{31}P NMR (120 MHz, $CDCl_3$) δ 25.5. FTIR (neat) ν_{max}: 1448 (C-H_{Me}), 1222 (P=O), 1098 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{31}H_{25}F_2N_2O_3P$ 466,2385, found 466,2390.

Di-tert-butyl (bis(1,5-dimethyl-1*H*-indol-3-yl)methyl)phosphonate 16. The general procedure was followed affording 245 mg (99%) of **16** as a pale brown solid. M.p. (Et_2O) = 82-85 °C (dec.). ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.45 (s, 2H, 2x indol-H2), 7.28 – 7.26 (m, 2H, 2xCH_{Ar}), 7.14 (d, $^{3}J_{HH}$ = 8.3 Hz, 2H, 2xCH_{Ar}), 7.00 (dd, $^{3}J_{HH}$ = 8.3 Hz, $^{4}J_{HH}$ = 1.3 Hz, 2H, 2x indol-H6), 4.84 (d, $^{2}J_{PH}$ = 25.5 Hz, 1H, CHP), 3.71 (s, 6H, 2xNCH_3), 2.44 (s, 6H, 2xCH_3), 1.18 (s, 18H, 6xCH_3). ^{13}C {^{1}H} NMR (75 MHz, $CDCl_3$): δ 135.2 (C_{quat}), 129.1 (d, $^{3}J_{PC}$ = 6.0 Hz, CH), 128.44 (d, $^{2}J_{PC}$ = 8.5 Hz, C_{quat}), 127.8 (C_{quat}), 123.0 (CH), 119.3 (CH), 111.1 (d, $^{3}J_{PC}$ = 5.4 Hz, C_{quat}), 108.7 (CH), 82.3 (d, $^{2}J_{PC}$ = 10.5 Hz, C_{quat}), 34.7 (d, $^{1}J_{PC}$ = 149.4 Hz, CH), 32.9 (CH_3), 30.3 (d, $^{3}J_{PC}$ = 3.8 Hz, CH_3), 21.7 (CH_3). ^{31}P NMR (120 MHz, $CDCl_3$) δ 18.1. FTIR (neat) ν_{max}: 1452 (C-H_{Me}), 1227 (P=O), 1101 (P-O-C). HRMS (Q-TOF) m/z calcd for $C_{29}H_{39}N_2O_3P$ 494,2698, found 494,2709.

3.2 Biology

3.2.1. Materials

Reagents and solvents were used as purchased without further purification. All stock solutions of the investigated compounds were prepared by dissolving the powered materials in appropriate amounts of DMSO. The final concentration of DMSO never exceeded 10% (v/v) in reactions. The stock solution was stored at 5ºC until it was used.

3.2.2. Cytotoxicity assays.

Cells were cultured according to the supplier´s instructions. Cells were seeded in 96-well plates at a density of 2-4 x 103 cells per well and incubated overnight in 0.1 mL of media supplied with 10% Fetal Bovine Serum (Lonza) in 5% CO_2 incubator at 37 ºC. On day 2, drugs were added and samples were incubated for 48 hours. After treatment, 10 µL of cell counting kit-8 was added into each well for additional 2 hours incubation at 37 ºC. The absorbance of each well was determined by an Automatic Elisa Reader System at 450 nm wavelength. Camptothecin was purchased from Sigma-Aldrich and used as positive control.

4. Acknowledgments

Financial support by Ministerio de Economía, Industria y Competividad (MINECO, CTQ-2015-67871R) and Gobierno Vasco (GV, IT 992-16) is gratefully acknowledged. We also thank SGIker (UPV/EHU) technical support for NMR spectra (MINECO, GV/EJ, and European Social Found).

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