CHAPTER-4

STUDIES ON SYNTHESIS OF BISPHOSPHONIC ACIDS AND THEIR SODIUM SALTS

4.1 INTRODUCTION

Based on the pioneering studies on inorganic pyrophosphate by Fleish and his colleagues more than three decades ago, the bisphosphonates have now become an important class of osteotropic drugs [146]. They have found extensive clinical use to inhibit the elevated bone resorption occurring in patients with tumor-induced hypercalcemia and Paget’s disease [147,148].

The present chapter describes the synthesis of some of the bisphosphates from the phosphorylation of carboxylic acid having the inhibition of bone resorption.

4.2 LITERATURE SURVEY

Bisphosphonates (65) or diphosphonates are having the P-C-P structure, which are the analogues of pyrophosphates (66) in which the oxygen in P-O-P has been replaced by a carbon, resulting in a metabolically stable structure [149].

![Chemical Structures]

(65)    (66)
Bisphosphonates make a multi-billion-dollar contribution to the global pharmaceutical market where they are used extensively in treating various bone diseases, such as osteoporosis, Paget’s disease [150], and hypercalcemia [147,148], due to malignancy [151]. They also have activity as herbicides [152,153], anticancer agents [154,155] and antiparasitics [156-158]. Fleisch, H et al [159] reported that bisphosphonates acted by directly modulating bone mineral dissolution.

In 19th century, the first bisphosphonates were synthesized. Initially they were used mainly as antiscaling and anticorrosive agents. Later their use was extended to act as complexing agents in the textile, fertilizer, and oil industries [160]. The actual mechanism of action on calcium metabolism of bisphosphonates was demonstrated [161] in 1990s only. Fleish and his co-workers have shown to impair the formation and dissolution of calcium phosphate crystals \textit{in vitro} [162,163].

The replacement of the oxygen atom between the two phosphonic acid moieties of pyrophosphate by a carbon atom opened up the possibility of attaching side chain. Preferably R\(^1\) is a hydroxyl group, which increases the affinity for calcium even further owing to the ability of such derivatives to act as tridentate ligands. The nature of R\(^2\) is key to the optimization of bisphosphonates as potent inhibitors of osteoclastic bone resorption.
There are two classes of bisphosphonates: the N-containing and non-N-containing bisphosphonates.

**N-containing bisphosphonates:-**

Some important compounds of this class and their structures are given below Table 4.1.

**Table 4.1**

<table>
<thead>
<tr>
<th>S.No</th>
<th>Name</th>
<th>Structure</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>1-hydroxy-2-(3-pyridyl)ethylidene bisphosphonic acid (Risedronic acid)</td>
<td><img src="image1" alt="Structure" /></td>
</tr>
<tr>
<td>2.</td>
<td>1-hydroxy-2-(2-pyridyl)ethylidene bisphosphonic acid</td>
<td><img src="image2" alt="Structure" /></td>
</tr>
<tr>
<td>3.</td>
<td>1-hydroxy-3-(methylpentylamino) propylidenebisphosphonicacid (Ibandronic acid)</td>
<td><img src="image3" alt="Structure" /></td>
</tr>
<tr>
<td>4.</td>
<td>1-hydroxy-3-(pentylamino) propyldienebisphosphonic acid</td>
<td></td>
</tr>
<tr>
<td>5.</td>
<td>1-hydroxy-2-(1-imidazolyl) ethyldienebisphosphonic acid <strong>(Zoledronic acid)</strong></td>
<td></td>
</tr>
<tr>
<td>6.</td>
<td>1-hydroxy-3-(1-imidazolyl) propyldienebisphosphonic acid</td>
<td></td>
</tr>
<tr>
<td>7.</td>
<td>(3-amino-1-hydroxypropyldiene)-bisphosphonic acid <strong>(Pamidronic acid)</strong></td>
<td></td>
</tr>
<tr>
<td>8.</td>
<td>(4-amino-1-hydroxybutylidene)-bisphosphonic acid <strong>(Alendronic acid)</strong></td>
<td></td>
</tr>
<tr>
<td>9.</td>
<td>(6-amino-1-hydroxyhexylidene)-bisphosphonic acid <strong>(Neridronic acid)</strong></td>
<td></td>
</tr>
</tbody>
</table>
The most useful commercial drugs in this class of compounds are risedronate, ibandronate, zoledronate, pamidronate and alendronate.

**Non-N-containing bisphosphonated:**

Some of the important drugs under this class are Etidronate and clodronate shown in Table 4.2

**Table 4.2**

<table>
<thead>
<tr>
<th>S.No</th>
<th>Name</th>
<th>Structure</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>1-hydroxyethylidene bisphosphonic acid</td>
<td><img src="#" alt="Structure1" /></td>
</tr>
<tr>
<td></td>
<td>(Etidronate)</td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>Dichloromethylene bisphosphonic acid</td>
<td><img src="#" alt="Structure2" /></td>
</tr>
<tr>
<td></td>
<td>(Clodronate)</td>
<td></td>
</tr>
</tbody>
</table>

**General preparations of Bisphosphonic acids:**

Kieczykowski et al.[164] reported that carboxylic acids (67) on treatment with phosphorus trichloride followed by hydrolysis with water gave 1-hydroxy-1,1-bisphosphonic acid (68) (Scheme 4.1).
Lecouvey et al [165] synthesized the bisphosphonic acids (68) from acid chlorides (69) (Scheme 4.2)

Mikhalin et al [166] reported the synthesis of 1-hydroxy-1,1-bisphosphonic acids are formed from carboxylic acids (67) (Scheme 4.3)
on treatment with a mixture of phosphorous acid and phosphorus trichloride, followed by hydrolysis.

Allen et al [167] described a method for synthesis of 1,2-bisphosphonates 71 from aryl alkynes 70.
The aminomethylene bisphosphonates (73) were synthesized starting with a primary amine 72, on treatment with triethylorthoformate and 2 equiv. of diethylphosphite by Martin et.al. [168] (Scheme 4.5).

Hutchinson and Thornton reported [169] that generation of aminoethylene bisphosphonates (74) from amines 72 (Scheme 4.6)
1-Aminobisphosphonates (76) were prepared by Widler et al. [170] starting from aminonitrile compounds (75) using phosphoric acid and phosphorus tribromide. (Scheme 4.7).

\[
\begin{align*}
\text{H}_3\text{C}_\text{N}-\text{CN} & \xrightarrow{\text{H}_3\text{PO}_4, \text{PBr}_3} \text{H}_3\text{C}-\text{N}=\text{O} & \text{OH} \\
\text{CH}_3 & & \text{O}=\text{P}-\text{OH} \\
\end{align*}
\]

Scheme 4.7

Several methods are reported [171-173] in literature for the synthesis of important class of 1-hydroxyalkylidene bisphosphonates, useful for the treatment of calcium metabolism. 1-Hydroxyalkylidene bisphosphonates and their pharmaceutically active salts were prepared from the corresponding carboxylic acids 67 with phosphorous acid and phosphorous trichloride, in solvents such as chlorobenzene [171], methanesulphonic acid [172], sulfolane [173], ionic liquids [174] and diphenyl ether [175].

4.3 PRESENT WORK

The present chapter describes the design and preparation of various bisphosphates from the phosphorylation of carboxylic acid and is summarized in a schematic way (Scheme 4.8).
4.4 DOCKING STUDIES

Molecular docking is a key tool in structural molecular biology and computer-assisted drug design. The goal of ligand—protein docking is to predict the predominant binding model(s) of a ligand with a protein of known three-dimensional structure. In this study, we have used several molecular docking programs such as LIGANDFIT (ACCELERYS), AUTODOCK Vina as to screen of Ibandronate and Zoledronate analogues.
Materials and Methods

Data set: A set of designed molecules was taken for docking studies.

Computational Details

Ligandfit of Discovery Studio 2.1[176] and Autodock Vina [177,178] was used to generate grids, calculate score and binding affinity. The structure of compounds was drawn by using symyx 3.1.

Protein Preparation

The X-ray structure of Crystal structure of human FPPS in complex with ibandronate (PDB ID –2f94) [179] and Crystal structure of human FPPS in complex with Zoledronate and Zn\textsuperscript{2+} (PDB ID –2f9K) [180] was used for docking studies.

Validation of the Molecular Docking method

To ensure that the ligand orientation and the position obtained from the docking studies were likely to represent valid and reasonable binding modes of the inhibitors, docking parameters was first validated for the crystal structure used PDB ID –2F94 and 2F9K.

The ligand Ibandronate and Zoledronate found in the crystal structure, was extracted and docked back to the corresponding binding pocket, to determine the ability of docking method to reproduce the orientation and position of the inhibitor observed in the crystal structure.

Molecular Docking Studies

Molecular docking of Ibandronate and Zoledronate analogues to the active site of human FPPS was carried out using modern docking
engine Ligandfit available with Discovery Studio 2.1 (http://www.accelrys.com) and Autodock Vina.

As shown in docking diagrams the designed analogues were docked in the same binding site, by using mentioned docking procedure.

**Results:** The docking results of both Ibandronate and Zoledronate are given in the scoring table. According to docking studies which is based on binding affinity, H-bond interaction and scoring value [181], it was revealed that out of three docked analogues for Ibandronate 79h is best fit in terms of dockings and scoring followed by 79g.

And in case of Zolendronic acid 79a is best fit in the given five molecules. To our big surprise we got same docking results with both docking program Autodock Vina and Ligandfit, which make our work very easy to select the best fit in the given set.

**Scoring Table for Ibandronate Analogues**

**Scoring Table:**

**Ligandfit**

<table>
<thead>
<tr>
<th>Molecule</th>
<th>PLP1</th>
<th>PLP2</th>
<th>Dock</th>
<th>Lig2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ibandronate</td>
<td>102.37</td>
<td>95.71</td>
<td>94.03</td>
<td>6.37</td>
</tr>
<tr>
<td>79g</td>
<td>78.03</td>
<td>69.71</td>
<td>70.55</td>
<td>6.03</td>
</tr>
<tr>
<td>79f</td>
<td>60.93</td>
<td>51.8</td>
<td>74.88</td>
<td>6.03</td>
</tr>
<tr>
<td><strong>79h</strong></td>
<td><strong>100.88</strong></td>
<td><strong>94.08</strong></td>
<td><strong>100.61</strong></td>
<td><strong>6.35</strong></td>
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</tbody>
</table>
II- Autodock Vina.

<table>
<thead>
<tr>
<th>Molecule</th>
<th>Binding Affinity</th>
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</thead>
<tbody>
<tr>
<td>Ibandronate</td>
<td>-8.5</td>
</tr>
<tr>
<td>79g</td>
<td>-8.0</td>
</tr>
<tr>
<td>79f</td>
<td>-7.8</td>
</tr>
<tr>
<td>79h</td>
<td>-8.2</td>
</tr>
</tbody>
</table>

Scoring Table for Zolendronic Acid Analogues

Scoring Table:

**Ligandfit**

<table>
<thead>
<tr>
<th>Molecule</th>
<th>PLP1</th>
<th>PLP2</th>
<th>Dock</th>
<th>Lig2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zoledronic acid</td>
<td>97.8</td>
<td>78.2</td>
<td>92.78</td>
<td>5.24</td>
</tr>
<tr>
<td>79a</td>
<td>113.71</td>
<td>92.35</td>
<td>106.83</td>
<td>3.64</td>
</tr>
<tr>
<td>79b</td>
<td>108.3</td>
<td>89.54</td>
<td>100.11</td>
<td>2.53</td>
</tr>
<tr>
<td>79c</td>
<td>102.4</td>
<td>90.31</td>
<td>99.34</td>
<td>2.93</td>
</tr>
<tr>
<td>79d</td>
<td>101.79</td>
<td>86.83</td>
<td>93.82</td>
<td>0.62</td>
</tr>
<tr>
<td>79e</td>
<td>103.01</td>
<td>80.45</td>
<td>92.45</td>
<td>1.83</td>
</tr>
</tbody>
</table>

**Autodock Vina.**

<table>
<thead>
<tr>
<th>Molecule</th>
<th>Binding Affinity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zoledronic acid</td>
<td>-8.7</td>
</tr>
<tr>
<td>79a</td>
<td>-9.6</td>
</tr>
<tr>
<td>79b</td>
<td>-6.7</td>
</tr>
<tr>
<td>79c</td>
<td>-5.3</td>
</tr>
<tr>
<td>79d</td>
<td>-5.5</td>
</tr>
<tr>
<td>79e</td>
<td>-5.0</td>
</tr>
</tbody>
</table>
Docking Pictures of Zolendronic acid (PDB ID –2f9k)

Superimposed structure of 79a (grey) with Zolendronic acid (yellow)

H-bond interaction of 79a (grey) represented as green dots.
H-bond interaction of 79a (grey) with water molecule present in binding sites of protein represented as green dots.

Docking Pictures of Ibandronate (PDB ID –2f94)

Superimposed structure of 79h (grey) with Ibandronate (yellow)
H-bond interaction of 79h (grey) represented as green dots.

H-bond interaction of 79h (grey) with water molecule present in binding sites of protein represented as green dots.
4.5 RESULTS AND DISCUSSION

Process for the preparation of the key intermediate \textit{77e} starts from benzimidazole (80). Compound 80 was reacted with methylacrylate (81) in methanol at reflux temperature to afford methyl-3-(1H-benzimidazole)-propionate (82) (Scheme 4.9), and was characterized on the basis of IR and \textit{1}H NMR. Its IR (neat) (Fig 4.1) showed strong and sharp ester carbonyl peak at 1736cm$^{-1}$. Aromatic C=C str peaks are observed at 1439, 1459 and 1496 cm$^{-1}$. In \textit{1}H NMR (CDCl$_3$, 400MHz) (Fig 4.2) methylene (-CH$_2$-) peaks are noticed at 2.88(t, 2H) and 4.515(t, 2H). Methyl protons are observed at 3.67(s, 3H) and aromatic protons are observed at 7.2 to 8.1

![Scheme 4.9](image)

\textbf{Scheme 4.9}

The resulting ester 82 was hydrolyzed in the presence of aq. hydrochloride at reflux to yield 3-(1H-benzimidazole)-propionic acid hydrochloride (77e) (Scheme 4.10). This compound was characterized on the basis of IR (Fig 4.3) exhibiting acid carbonyl peak at 1718 cm$^{-1}$ and broad OH peak at about 3000cm$^{-1}$. 
Phosphonylation of 3-(1H-benzimidazole)-propionic acid hydrochloride (77e) (Scheme 4.11) with orthophosphoric acid/phosphorous acid and phosphorus trichloride in chlorobenzene as solvent gave 1-hydroxy-3-(1H-benzimidazole)-propane–1,1-bisphosphonic acid (78e), followed by alkylation in situ with alkali solution afforded 1-hydroxy-3-(1H-benzimidazole)-propane–1,1-bisphosphonate sodium (79e). Its $^1$H NMR (Fig 4.4) showed a peak at 2.38-2.48 (m, 2H) assignable to methylene group in α (Alfa) position. The peak at 4.605 (t, 2H), is assignable to methylene (-CH$_2$-) group attached to nitrogen atom in imidazole. Peaks observed at 7.3-7.4 (m, 2H) and 7.68- 7.7 (m, 2H) are representing the aromatic protons. The peak at 8.4 (s, 1H) is assignable to N-CH=N proton. Its $^{13}$C NMR (Fig 4.5) spectrum exhibited peaks at 34.15, 41.77 are assignable to two methylene carbons (2x-CH$_2$) in alkane chain. The carbon atom attached to the phosphorous atoms has been split into triplet and is observed at 72.68ppm. Aromatic carbons are observed in the region 111 to 143 ppm. Its $^{31}$P NMR (Fig 4.6) spectrum recorded in D$_2$O showed a signal at δ 17.29 ppm, assignable to the two
phosphorous atoms, which are chemically and magnetically equivalent. Mass spectrum of 79c (Fig 4.7) showed its m/z 357 corresponding to its base value [M+1]+.

![Scheme 4.11](image)

**Scheme 4.11**

The above reaction was found to be a general one and has been observed to be facile with a variety of carboxylic acids 77 as shown in **Scheme 4.8**. The structures of the bisphosphonic acids 78 and their sodium salts 79, thus obtained were confirmed on the basis of spectral and analytical data (Experimental Section).
General preparation for the key starting materials 77 (a, b, c and f):
Substituted benzimidazoles / cyclopropyl amine are reacted with methylchloroacetate (84) in presence of potassium carbonate in DMF at 0-5 °C followed by hydrolysis with hydrochloric acid at 85-95 °C (Scheme 4.12) to afford the corresponding carboxylic acid hydrochloride (77).
**Scheme 4.12**

\[
R-H + \text{Cl} \overset{\text{DMF, K}_2\text{CO}_3}{\longrightarrow} \text{R-(CH}_2\text{-COOCH}_3 \overset{\text{HCl}}{\longrightarrow} \text{R-(CH}_2\text{-COOH} . \text{HCl}
\]

(83)  (84)  (85)  (77) (a,b,c and f)

**General preparation for the key starting materials 77 (d, e, g and h):**

Substituted benzimidazoles / cyclopropyl amine derivatives are reacted with methylchloroacetate (86) in methanol at room temperature followed by hydrolysis with hydrochloric acid at 85-95 °C (Scheme 4.13) to afford the corresponding carboxylic acid hydrochloride.

\[
R-H + \text{CH}_3\text{COOCH}_3 \overset{\text{CH}_3\text{OH}}{\overset{50-60^\circ\text{C}}{\longrightarrow}} \text{R-} \overset{\text{HCl}}{\longrightarrow} \text{R-} \overset{\text{COOH} . \text{HCl}}{\text{COOH}}
\]

(83)  (86)  (87)  (77)(d,e,g and h)

**Scheme 4.13**

(d)  (e)  (g)  (h)
4.6 EXPERIMENTAL SECTION

**General procedure for the preparation of starting materials 77 (a, b, c and f):** Compound 83 (0.04mole), potassium carbonate (0.03mole) and DMF (20ml) were taken in a 250ml 4N RB flask. Reaction mass was cooled to 0-5 °C. Methyl chloroacetate (84) (0.06mole) was added over a period of 1h. Aliquots were followed by TLC. Water (60ml) was added to the reaction mass and the product was extracted into toluene (2x 30ml). Combined toluene layer was dried over sodium sulphate and toluene was distilled off under vacuum to yield the ester 85. Compound 85 was refluxed with conc. HCl (20ml) and water (20ml) for 3-4h. Water was distilled off completely under vacuum at 85-90 °C and the residue was diluted with acetone (75ml) to afford 77 (a, b, c and f).

**General procedure for the preparation of starting materials 77 (d, e, g and h):** Compound 83 (0.1mole), methanol (50-75ml) were taken in a 250ml 4N RB flask. Methyl acrylate (86) (0.1mole) was added at 20-25 °C. The temperature of the reaction mass was raised to reflux and maintained for 6-7h. Aliquots were followed by TLC. Distilled of solvent completely under vacuum afforded 87 as oil. Compound 87 was refluxed with conc. HCl (20ml) and water (20ml) for 3-4h. Water was distilled off completely under vacuum at 85-90 °C and the residue was diluted with acetone (200ml) to afford 77 (d, e, g and h).
79a: (77a) (6.1g, 0.03mole), phosphoric acid (7.4g, 0.07mole) and chlorobenzene (20ml) were charged in a 250ml 4N RB flask. The temperature of the reaction mass was raised to 70-75 °C. To the resulting mixture was added phosphorus trichloride (11.7g) at 75-100 °C. The reaction mass was maintained at 100-110 °C for 3-4h. The solvent was decanted off from the thick residue and stirred under reflux with conc. hydrochloric acid (10ml) for 7-9h. The mass was treated with carbon and diluted the filtrate with 3.5L of acetone, filtered and dried. After recrystallising twice from water/methanol 78a was obtained (7.1g, 65% of theory) as an analytically pure product in the form of free acid. Compound 78a was diluted with water (15ml) and pH was adjusted to 4.5 with 50% aq. NaOH. This reaction mass was added to methanol (100ml) and the resulting mass was filtered and dried at 60-65 °C to afford 79a (1.6g, 25% yield).

79b: (77b) (5.4g, 0.03mole), phosphoric acid (7.4g, 0.07mole) and chlorobenzene (20ml) were charged into a 250ml 4N RB flask. The temperature of the reaction mass was raised to 70-75 °C. To the resulting mixture was added phosphorus trichloride (11.7g) at 75-100 °C. The reaction mass was maintained at 100-110 °C for 3-4h. The solvent was decanted from the thick residue and stirred under reflux with conc. hydrochloric acid (10ml) for 7-9h. The mass was treated with carbon and diluted the filtrate with 3.5L of acetone, filtered and dried. 10g of crude product was obtained. After recrystallised twice from water/methanol
14b was obtained (4.3g, 47.2 % of theory) of analytically pure product in the form of free acid. Compound 78b was diluted with water (20ml) and pH was adjusted to 4.5 with 50% aq. NaOH. This reaction mass was added to methanol (125ml). Mass was filtered and dried at 60-65 °C afforded 79b.

m.p: 275.2 °C. \(^1\)H NMR (D\(_2\)O, 400MHz) \(\delta\): 3.06(s, 3H, -CH\(_3\)), 4.99(t, 2H, -CH\(_2\)-), 7.62(bs, 2H, aromatic H), 7.75(d, J=4.2, 1H, aromatic H), 8.12 (d, J= 4.2, 1H, aromatic H);

78c: In manner analogues to that described in 78a, from 2-(5-methyl,1H-Benzimidazole)acetic acid hydrochloride there is obtained the corresponding diphosphonic acid (78c) in a yield of 48 % of theory.

79d: 77d (3.5g, 0.014mole), phosphoric acid (4.34g, 0.036mole) and chlorobenzene (20ml) were charged in a 250ml 4N RB flask. The temperature of the reaction mass was raised to 70-75 °C. To the resulting mixture was added phosphorus trichloride (6.02g, 0.043mole) at 75-100 °C. The reaction mass was maintained at 100-110 °C for 3-4h. The solvent was decanted from the thick residue and stirred under reflux with 50% aq. hydrochloric acid (10ml) for 7-9h. Reaction mass was filtered. Obtained product (14d, 1.8g) was diluted with water (5ml) and pH was adjusted to 4.5 with 50% aq. NaOH. Reaction mass was filtered and dried afforded 79d (1.7g, 28.2 %yield) \(^1\)H NMR (D\(_2\)O, 400MHz) \(\delta\): 2.28-2.32 (bs, 2H, -CH\(_2\)-), 2.64 (s, 3H, -CH\(_3\)), 4.52 (bs, 2H, -CH\(_2\)-), 7.31-7.36 (m complex, 2H, aromatic), 7.56 (d, J=5.6, 1H,aromatic), 7.69 (d,
J=4.8, 1H, aromatic); $^{13}$C NMR (D$_2$O, 400 MHz) $\delta$ppm: 12.02, 33.58, 40.44, 72.5(t), 111.04, 115.95, 123.24, 133.28, 137.04, 152.42; $^{31}$P NMR (D$_2$O, 400 MHz) $\delta$: 17.29; Mass (m/z): 371 [M+1]$^+$, 349 [M-Na+1]$^+$.

**79e:** 77e (25g, 0.1mole), phosphoric acid (33g, 0.27mole) and chlorobenzene (75ml) were charged in a 250ml 4N RB flask. The temperature of the reaction mass was raised to 70-75 °C. To the resulting mixture phosphorus trichloride (45.6g, 0.3mole) was added at 75-100 °C. The reaction mass was maintained at 100-110 °C for 3-4h. The solvent was decanted from the thick syrupy mass and stirred the residue under reflux with 50% aq. hydrochloric acid (150ml) for 7-9h. Reaction mass was filtered and wet cake (41.5g) was taken into a water (40ml) and was added 50% aq. NaOH. The temperature of the reaction mass was raised to 80-85 °C. Filtered the mass and filtrate was diluted with methanol (300ml). Filtered the resulting solid and dried at 60-65 °C yielded 79e (35g, 77% yield) Meltingrange: 246-50 °C. $^1$H NMR (D$_2$O, 400MHz) $\delta$: 2.38-2.48 (m, 2H, -CH$_2$-); 4.58-4.62 (t, 2H, -CH$_2$-), 7.32-7.41 (m, 2H, aromatic Hs), 7.68-7.75 (dd, 2H, aromatic), 8.43 ( s, -CH<); $^{13}$C NMR (D$_2$O, 400 MHz) $\delta$ ppm: 34.15, 41.77, 72.68(t), 111.33,117.87, 123.10,123.57, 132.86, 139.93, 143.59; $^{31}$P NMR (D$_2$O, 400 MHz)$\delta$: 17.29; Mass (m/z): 357 [M+1]$^+$, 335 [M-Na+1]$^+$.

**78f:** 77f (10.0g, 0.066mole), phosphorous acid (6.8g, 0.08mole) were charged into 100ml RB flask and raised the temperature to 70-75 °C. To the resulting mixture phosphorus trichloride (18.1g, 0.13mole) was
added. The reaction mass was maintained at 100-110 °C for 3-4h. Water (20ml) was added to the mass at 20-25 °C and then stirred under reflux for 7-9h. The reaction mass was treated with activated carbon and the reaction mass was diluted with isopropanol (370ml), and kept aside for 7-10days. White crystalline solid formed, mass was filtered and dried afforded **78f** (2.6g, 14.6% yield). m.p. 178 °C; $^1$H NMR (D$_2$O, 400MHz) δ: 0.831 (bs, 4H, 2x-CH$_2$-), 2.72-2.76 (m complex, 1H, >CH-), 3.54 (t, 2H, -CH$_2$-); $^{13}$C NMR (D$_2$O, 400 MHz) δ ppm: 4.11, 31.66, 51.50, 70.78(t); $^{31}$P NMR (D$_2$O, 400 MHz) δ: 13.29;

**78g**: **77g** (140g, 1.0mole), phosphoric acid (326g, 2.7mole) and chlorobenzene (300ml) were charged in a 2.0L 4N RB flask. The temperature of the reaction mass was raised to 60 °C. To the resulting mixture phosphorus trichloride (450g, 3.2mole) was added at 60-100 °C over a period of 2h. The temperature of the reaction mass was maintained at 100-110 °C for 3-4h. Chlorobenzene was decanted from the reaction mass, conc. hydrochloric acid (330ml) was added to the residue and stirred under reflux for 7-9h. The reaction mass was treated with carbon and filtrate was diluted with acetone (3.5L). Filtered the reaction mass and dried yielded **78g** (160g) as crude product. After recrystallising twice from water/acetone, there are obtained **78 g** (148g, 63.6%yield); m.p: 205.8 °C; $^1$H NMR (D$_2$O, 400MHz) δ: 0.78-0.80(bs, 4H, -2x -CH$_2$-), 2.24-2.27(m, complex, 2H, -CH$_2$-), 2.65(bs, 1H, >CH-), 3.40 (bs, 2H, -CH$_2$-); $^{13}$C NMR (D$_2$O, 400 MHz) δ ppm: 2.86, 29.18, 29.81,
$30.08, 44.14, 71.75(t)$; $^{31}$P NMR (D$_2$O, 400 MHz) $\delta$: 17.37; Mass (m/z): 276[M+1]$^+$.  

**78h**: In manner analogues to that described in preparation of 78f, from 77h. hydrochloride there is obtained the corresponding diphophonate in a yield of 25% of theory; m.p 178-184 °C.