SYNTHESIS AND EVALUATION OF NOVEL PHOSPHONATED AND BISPHOSPHONATED METHACRYLATES WITH POSSIBLE ENVIRONMENTAL AND BIOMEDICAL APPLICATIONS

by

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To My Daughter & My Husband…

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ABSTRACT

SYNTHESIS AND EVALUATION OF NOVEL PHOSPHONATED AND BISPHOSPHONATED METHACRYLATES WITH POSSIBLE ENVIRONMENTAL AND BIOMEDICAL APPLICATIONS

In this study, a series of mostly alkyl α-hydroxymethacrylate (RHMA)-derived novel monomers in four groups, and polymers of some of them were synthesized for various purposes including dental, bone-targeting and metal-binding applications. The first-group monomers, containing bisphosphonate, were synthesized from reaction of RHMA-derived ethyl or *tert*-butyl α-bromomethacrylate (EBBr, TBBr) with tetraethyl 4-hydroxybutane-1,1-diyldiphosphonate (**A1**, **A2**). Their homo- and copolymerizations with poly(ethylene glycol) methyl ether methacrylate, hydrolysis of bisphosphonate groups of one of the polymers to give a polymer with binding ability to hydroxyapatite (HAP) indicated their potential to deliver an attached drug to bone tissues. The monomers of the second group contain either bisphosphonic (**B3**), or carboxylic acid (**B4**), or the last two together (**B5**). They were synthesized by the reactions of tetraethyl 1-hydroxyethane-1,1-diyldiphosphonate with EBBr and TBBr (**B1**, **B2**) followed by hydrolysis of their bisphosphonate and/or *tert*-butyl groups. Hydrolytic stability, copolymerizations with commercial dental monomers and HAP interactions make these monomers promising candidates for dental adhesives. Aminophosphonate-containing methacrylates making up the third group were synthesized by reactions of diethyl aminomethylphosphonate or diethyl 2-aminoethylphosphonate with EBBr (**C1**, **C2**) or TBBr (**C3**, **C4**); or with 3-(acryloyloxy)-2-hydroxypropyl methacrylate (AHM) (**C5**, **C6**). **C1**-**C4** gave crosslinked or soluble polymers depending on monomer structure and polymerization conditions. The fourth group contains eight water-soluble, zwitterionic monomers (**D1**-**D8**), synthesized by hydrolysis of the phosphonate or *tert*-butyl or both groups of **C1**-**C4**. The copolymerizations with diallyldimethylammonium chloride gave cyclic polymers which showed polyelectrolyte behaviour in water. Polymers obtained from these monomers may have potential applications in water treatment, coatings, and pharmaceutics.

ÖZET

OLASI ÇEVRE VE BİYOMEDİKAL UYGULAMALAR İÇİN YENİ FOSFONATLI VE BİSFOSFONATLI METAKRİLAT MONOMERLERİNİN SENTEZ VE DEĞERLENDİRİLMELERİ

Bu çalışmada, diş, kemik-hedefleme ve metal bağlayıcılığı uygulamaları gibi çeşitli amaçlar için, çoğunluğu alkil α-hidroksimetakrilat (RHMA) türevi olan dört yeni monomer grubu ile bunların bazılarının polimerleri sentezlenmiştir. Birinci gruptaki monomerler bisfosfonat içermekte olup, RHMA türevi olan etil ya da *tert*-butil α-bromometakrilat (EBBr, TBBr) ile tetraetil 4-hidroksibütan-1,1-diildifosfonatın tepkimesinden sentezlendi (**A1**, **A2**). Bu monomerlerin homopolimerizasyonu ve poli(etilenglikol) metil eter metakrilatla kopolimerizasyonları ve bu polimerlerden birinin bisfosfonat grubunun hidrolize edilerek hidroksiapatite (HAP) bağlanma özelliği kazanması, bu polimerlerin takılı bir ilacı kemiğe iletme potansiyeli olduğunu gösterdi. İkinci grup monomerler ya bisfosfonik asit (**B3**), ya karboksilik asit (**B4**) ya da ikisini birden içerir (**B5**). Bunlar tetraetil 1-hidroksietan-1,1diildifosfonatın EBBr ve TBBr ile tepkimelerinin ardından bisfosfonat ve/veya *tert*-butil gruplarının hidrolize edilmesiyle sentezlendiler. Hidrolize dayanıklılık, ticari diş monomerlerleri ile kopolimerleşme ve HAP etkileşimleri bu monomerlerin diş yapıştırıcaları için umut vadettiğini gösterir. Üçüncü grubu oluşturan aminofosfonat içeren metakrilatlar, dietil aminofosfonat ya da dietil 2-aminoetilfosfonatın EBBr (**C1**, **C2**), TBBr (**C3**, **C4**) veya 3-((akriloksi)-2-hidroksipropilmetakrilatla (**C5**, **C6**) tepkimesi sonucu sentezlendi. **C1-C4** monomer yapısına ve polimerleşme şartlarına bağlı olarak çapraz bağlı ya da çözünebilir polimerler verdiler. Dördüncü grup, **C1-C4** monomerlerinin fosfonat ya da *tert*-bütil gruplarının hidrolizi sonucunda sentezlenen sekiz tane suda çözünebilen zwiteriyonik monomer içermektedir (**D1-D8**). Bunların diallildimetilamonyum klorür ile kopolimerizasyonları suda polielektrolit davranış gösteren siklik polimerler vermiştir. Bu monomerlerden elde edilen polimerlerin su arıtma, kaplama ve ilaç alanında kullanılabilme potensiyeleri mevcuttur.

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LIST OF SYMBOLS

A1	Ethyl 2-((4,4-bis(diethoxyphosphoryl)butoxy)methyl)acrylate
A2	*Tert*-butyl 2-((4,4-bis(diethoxyphosphoryl)butoxymethyl)acrylate
B1	Ethyl 2-((1,1-bis(diethoxyphosphoryl)ethoxy)methyl)acrylate
B2	*Tert*-butyl 2-((1,1-bis(diethoxyphosphoryl)ethoxy)methyl)acrylate
B3	1-((2-(ethoxycarbonyl)allyl)oxy)ethane-1,1-diyl)diphosphonic acid
B4	2-((1,1-bis(diethoxyphosphoryl)ethoxy)methyl)acrylic acid
B5	2-((1,1-diphosphonoethoxy)methyl)acrylic acid
C1	Diethyl 2,2'-(((diethoxyphosphoryl)methyl)azanediyl)bis(methylene))diacrylate
C2	Di-*tert*-butyl2,2'-((((diethoxyphosphoryl)methyl)azanediyl)bis(methylene)) diacrylate
C3	Diethyl 2,2'-(((2-(diethoxyphosphoryl)ethyl)azanediyl)bis(methylene)) diacrylate
C4	Di-*tert*-butyl 2,2'-(((2-(diethoxyphosphoryl)ethyl)azanediyl)bis(methylene)) diacrylate
C5	3-((3-(((diethoxyphosphoryl)methyl)amino)propanoyl)oxy)-2-hydroxypropyl methacrylate
C6	3-((3-((2-(diethoxyphosphoryl)ethyl)amino)propanoyl)oxy)-2-hydroxypropyl methacrylate
D1	(Bis(2-(ethoxycarbonyl)allyl)amino)methylphosphonic acid
D2	((Bis(2-(tert-butoxycarbonyl)allyl)amino)methyl)phosphonic acid
D3	(2-(Bis(2-(ethoxycarbonyl)allyl)amino)ethyl)phosphonic acid
D4	(2-(Bis(2-(tert-butoxycarbonyl)allyl)amino)ethyl)phosphonic acid
D5	2,2'-((((diethoxyphosphoryl)methyl)azanediyl)bis(methylene))diacrylic acid
D6	2,2'-(((2-(diethoxyphosphoryl)ethyl)azanediyl)bis(methylene))diacrylic acid
D7	2,2'-(((phosphonomethyl)azanediyl)bis(methylene))diacrylic acid
D8	2,2'-(((2-phosphonoethyl)azanediyl)bis(methylene))diacrylic acid
R_p	Rate of polymerization
T_g	Glass transition temperature
V_{50}	2,2’-azobis(N,N-amidinopropane) dihydrochloride

LIST OF ACRONYMS/ABBREVIATIONS

AIBN	2,2-azobis(isobutyronitrile)
BAPO	Bis(2,4,6-trimethylbenzoyl)phenylphosphine oxide
BP	Benzophenone
DMPA	2,2'-dimethoxy-2-phenylacetophenone
AHM	3-(acryloyloxy)-2-hydroxypropyl methacrylate
DADMAC	Diallyldimethylammonium chloride
EBBr	Ethyl-α-bromomethyl acrylate
GDMA	Glycerol dimethacrylate
HAP	Hydroxyapatite
HEMA	2-Hydroxyethyl methacrylate
PEGMA	Poly(ethylene glycol) methyl ether methacrylate
TBBr	*tert*-Butyl-α-bromomethyl acrylate
TEA	Triethylamine
TFA	Trifluoroacetic acid
TMSBr	Trimethylsilyl bromide
DSC	Differential Scanning Calorimetry
ESEM	Environmental Scanning Electron Microscope
FT-IR	Fourier Transform Infrared Spectroscopy
GPC	Gel Permeation Chromatography
NMR	Nuclear Magnetic Resonance Spectroscopy
XRD	X-ray Diffraction

1. INTRODUCTION

Phosphorus-containing monomers and polymers have attracted considerable attention because of their broad variety of application areas.

The use of these materials made possible the development of nonhalogen flame retardants [1, 2]. Phosphorous-containing halogen-free retardant coatings can lead to a char which forms a protecting coating that cuts off oxygen transport toward the burning area. Polymeric materials with phosphonic acid groups were also investigated in proton-conducting fuel-cell membranes for alternative energy production [3, 4].

Phosphorus-containing monomers and polymers have also been used in biomedical field due to their properties such as biocompatibility, hemocompatibility, protein adsorption resistance and strong interactions with dental tissue and bones [5,6]. These materials are used in dentistry due to complex formation with calcium in hydroxyapatite (HAP) [7]. This property also leads to their usage in tissue engineering as scaffolds. They also play an important role as drug carriers for biologically active molecules [8, 9].

Polymers containing both amine and phosphonic acid groups or aminomethylphosphonic acid group are used as chelating resins for metals [10-12]. Monomeric and polymeric aminomethylphosphonic acids exhibit good complexation ability for the transition metal ions Cd^{+2} and Hg^{+2}, this selective extraction of the metal ions from aqueous solution can be used for the treatment of wastewater, groundwater and seawater [13, 14]. Besides their chelating abilities to metals, aminophosphonic acids which are structural analogues of amino acids are suitable for biological applications such as inhibition of enzymes, antibacterials, neuroactive compounds, anticancer drugs or pesticides [15, 16].

In this thesis, we will focus on two classes of phosphorus-containing materials. The first class of materials are designed to bind to dental and/or bone tissue and the second class to metals. We discuss these materials in more detail below.

1.1. Dental Materials

1.1.1. Self-etching Dental Adhesives

Dental adhesives are used to achieve a strong and durable bond between a filling composite and tooth tissue. In recent years, new generation one-bottle self-etching enamel-dentin adhesives were developed. These adhesives which are based on acidic monomers remove the weak smear layer on top of the dentin and create an etch pattern on the enamel in a clinically relevant period of time (i.e., 15–30 s). Diffusion of co-monomers into etched enamel and dentin followed by the polymerisation forms a hybrid layer which is responsible for strong bonds between the adhesive layer and the dental hard tissues [17-19].

The monomers used in commercial self-etching dental adhesives can be divided into three main groups according to their function: a) self-etching adhesive monomers; b) cross-linking monomers; and, c) monofunctional comonomers (Figure 1.1). All these monomers should have the following properties [20]:

- High rate of free-radical homo- or copolymerisation with the other monomers in the adhesive.
- Solubility in the adhesive composition. The monomers should be miscible both with aqueous solutions of acetone or ethanol used as solvents in commercial self-etching adhesives, and the other monomers.
- Sufficient stability of both the monomer and the formed polymer.
- Minimal water uptake and low swelling degree of the formed polymer and the monomers so that the mechanical strength of the adhesive layer is not impaired.
- Low oral toxicity and cytotoxicity.

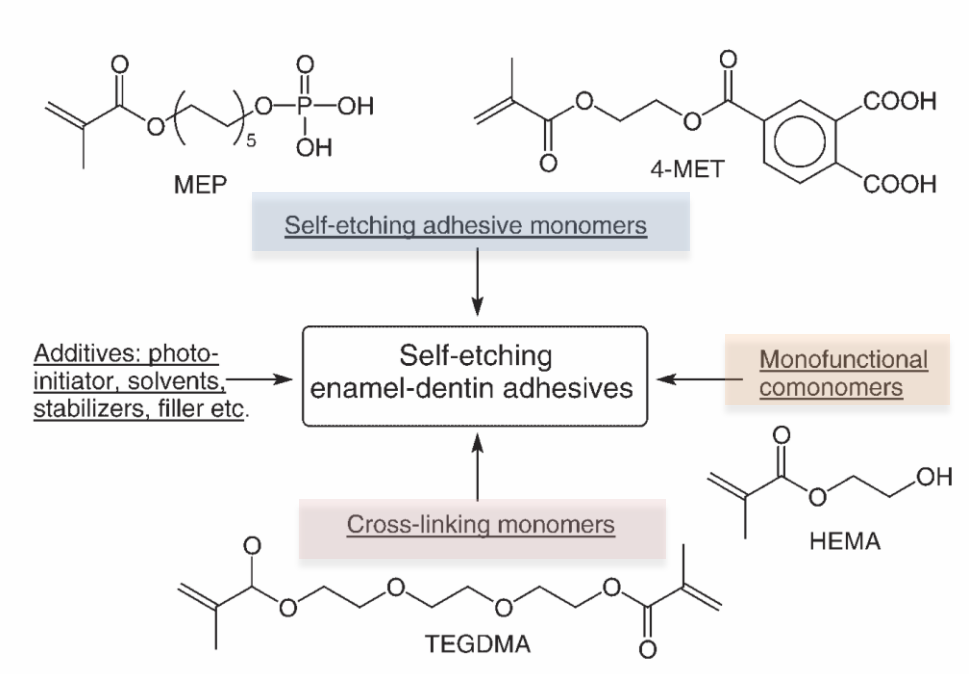


Figure 1.1. Components of self-etching dental adhesives [20].

In general, self-etching adhesive monomers are bifunctional molecules, containing a polymerizable group, an adhesive group AD, such as a strong acidic group and a spacer group R (Figure 1.2).

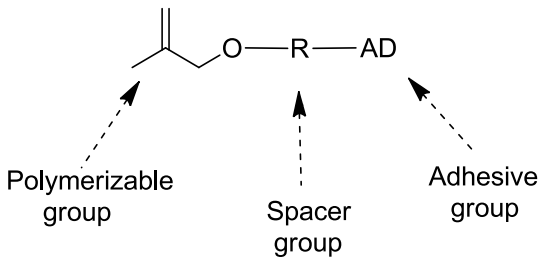


Figure 1.2. General structure of a self-etching adhesive monomer.

The polymerizable group reacts both with the other monomers of the adhesive and the restorative material by copolymerization. Usually, free-radically polymerizable methacrylate and methacrylamide functionalized monomers are used. Spacer groups have an effect on the adhesive monomer by changing solubility, flexibility, volatility and wetting properties. The adhesive group (AD) must be capable of both etching the dental hard tissues and interacting with the tooth substance, for example penetrating into the dentinal tubules. As a result, ionic bonds are formed by the acidic groups reacting with the

main inorganic component of the dental hard tissue, which is HAP. Suitable adhesive groups are acidic groups such as phosphonic acid and mono-or dihydrogenphosphate groups, which are stronger acids than the corresponding carboxylic acids. The acidity of the monomers increases in the order: carboxylic acids < phosphonic acids < acid phosphates < sulfonic acids.

Self-etching adhesive monomers are responsible for the interaction of the adhesive with the dental hard tissue. Therefore, they should have the following additional properties [20].

- Capability of etching the enamel surface in a relatively short time while forming a surface with increased roughness that enables micromechanical bonding of the adhesive on enamel.
- Optimal wetting and film-forming behaviour on the tooth surface and the capability of penetrating, for example, into the dentinal tubules.
- Fast ionic or covalent interaction with components of the dental hard tissue, (formation of low soluble calcium salts or covalent bonds with collagen).

Examples of commercial adhesive monomers are, 4-methacryloyloxyethyl trimellitic acid (4-MET), glycerol dimethacrylate ester of phosphoric acid (GDMP), 10-methacryloyloxydecyl dihydrogen phosphate (MDP), methacryloyloxyethyl phenyl hydrogen phosphate (MEP-P) and methacryloyloxyethyl dihydrogen phosphate (MEP, HEMA-phosphate) (Figure 1.3) [21].

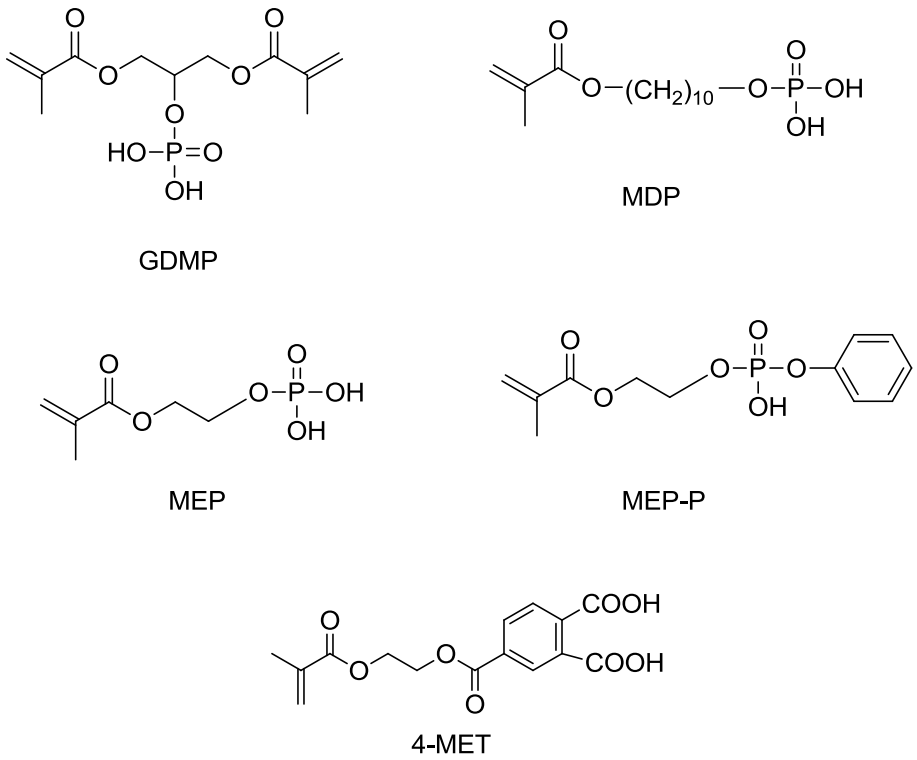


Figure 1.3. Examples of polymerizable acidic phosphates used in dental adhesives.

In the literature, synthesis and evaluations of many phosphonic, carboxylic and phosphoric acid-containing monomers were reported [22-33].

Inoue *et al*. reported that the long-term durability of adhesive-dentin bonds depends on the hydrolytic stability of the functional monomer and its chemical interaction potential with the dental tissue [34]. Therefore, extensive research has been conducted to develop new monomers with acidic functional groups, which may strongly bind to HAp. More recently bisphosphonic acid-containing monomers were investigated for self-etching dental adhesive applications [35-38] which facilitate adhesion of dental restoratives and orthodontic appliances to dental tissue (Figure 1.4). Moreover, it was also reported that bisphosphonates can inhibit enzymes (metalloproteinase) which degrade the collagen network [39-41].

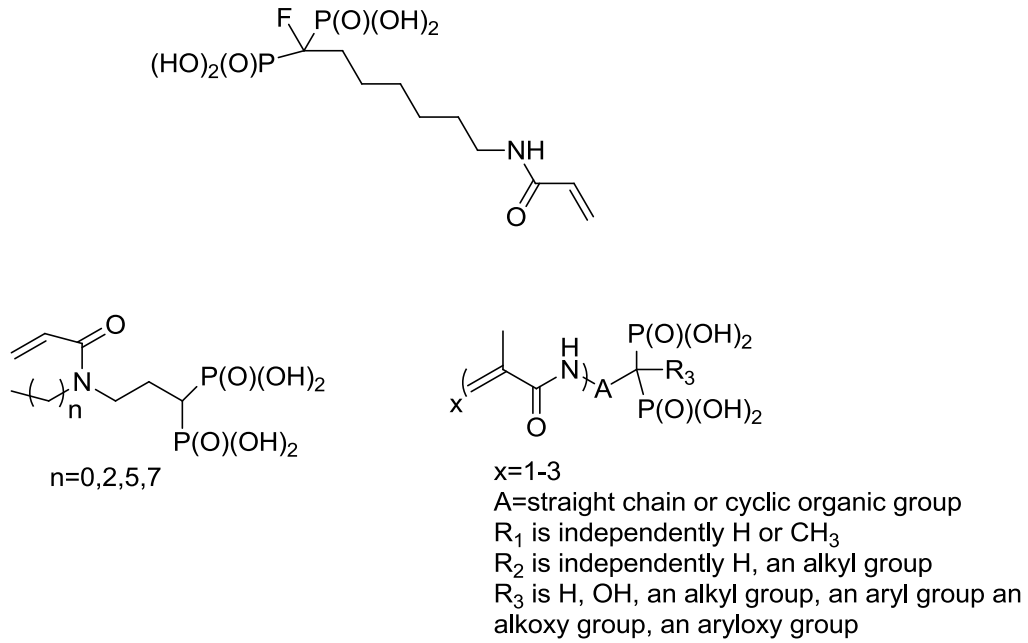


Figure 1.4. Examples of dental bisphosphonated monomers.

In a self-etching dental adhesive, usually water is used as a solvent or co-solvent. Thus, the methacrylates may undergo hydrolysis in the presence of strongly acidic adhesive monomers, which changes the chemical composition of the adhesive and thus impairs its performance (Figure 1.5). Therefore, new monomers with improved hydrolytic stability under acidic conditions are necessary.

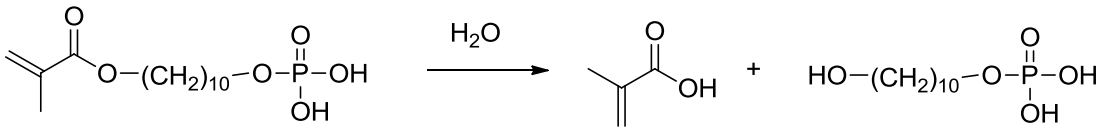


Figure 1.5. Hydrolysis of MDP.

The invention of more hydrolytically stable acrylic phosphonic acids has improved the self-etching adhesive systems remarkably [42-49]. Figure 1.6 shows the structures of some acrylic monomers containing phosphonic acids, synthesized by Ivoclar Vivadent Company [50-52].

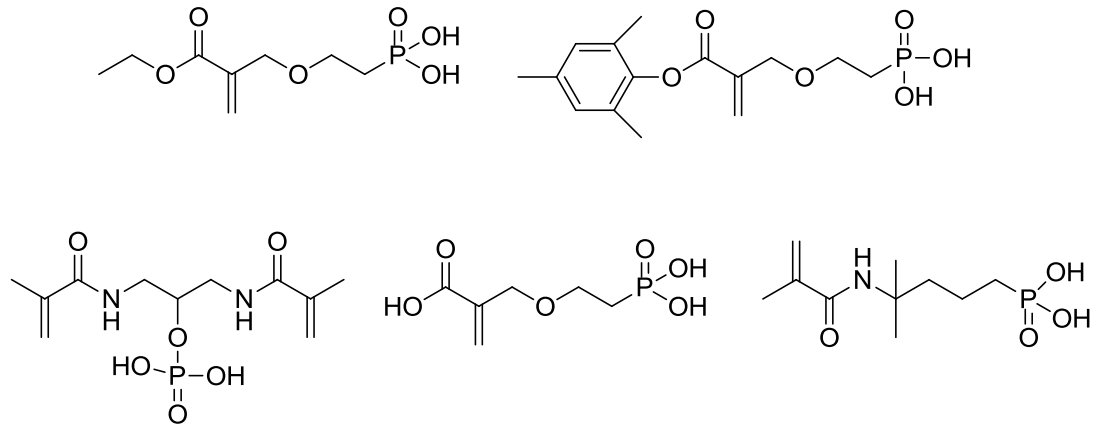


Figure 1.6. Hydrolytically stable self-etching monomers synthesized by Ivoclar Vivadent.

In Figure 1.7 there are some RHMA based hydrolytically stable monomers which were synthesized by our group [53-55].

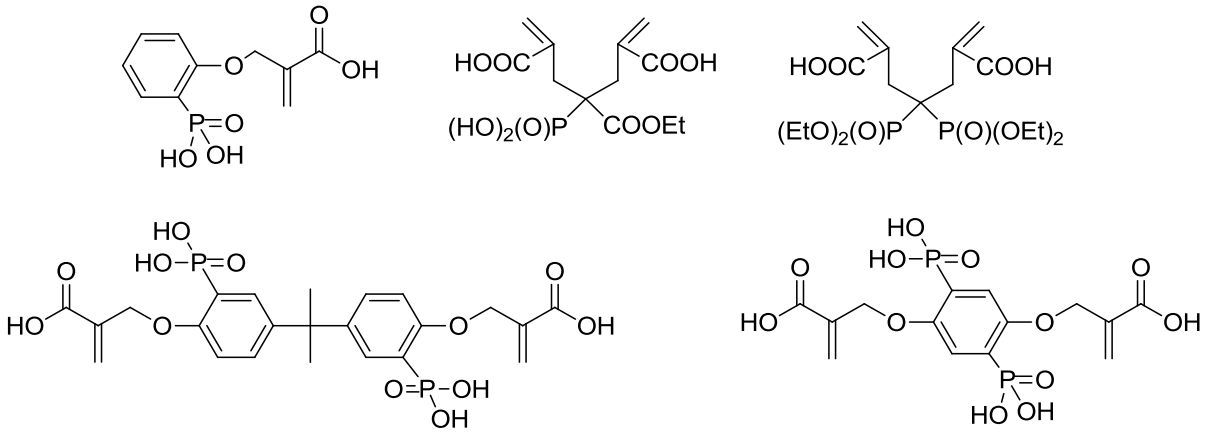


Figure 1.7. Hydrolytically stable self-etching monomers synthesized by our group.

Cross-linking dimethacrylates, such as triethylene glycol dimethacrylate (TEGDMA), 2,2-bis[4-(2-hydroxy-3-methacryloyloxypropyl)phenyl]propane (Bis-GMA), and glycerol dimethacrylate (GDMA) are used in self-etching enamel-dentin adhesives to increase both the polymerisation rate and the mechanical properties and to decrease water solubility of the polymer and the degree of swelling [20]. However, these dimethacrylates contain ester linkages in their structures and hydrolyze in acidic aqueous solutions to form the corresponding diols and methacrylic acid.

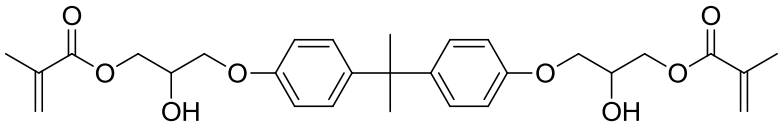
Hydrophilic monomethacrylates such as HEMA are used as comonomers in self-etching enamel-dentin adhesives to improve the miscibility and solubility of the polar and non-polar components of the adhesive and the wetting behaviour on dental tissue. It also stabilizes the collagen fibrils, improve permeability of dentin and monomer diffusion. But it is prone to hydrolysis under acidic aqueous conditions.

A final comment, applicable to self-etching adhesives is that in dental applications, it is very important to keep hydroxyapatite at the interface to protect collagen. Strong acid etching agents remove HAP completely so dentinal collagen becomes highly vulnerable to hydrolytic and enzymatic degradation. However, mild self-etch adhesives demineralize dentin only partially, leaving a substantial amount of HAP-crystals around the collagen fibrils. This residual HAP is utilized not only in protection of collagen, but also in chemically interaction with mild self adhesives [56-58].

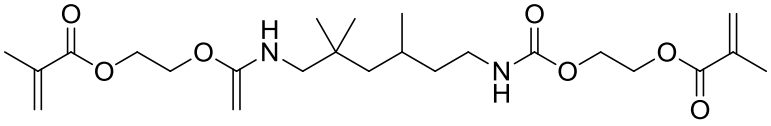
1.1.2. Self-adhesive Resin Cements

Self-adhesive resin cements were introduced within the past decade and gained importance rapidly. They are hybrid materials that combine advantages of composite resins (aesthetic, mechanical properties), self-etching adhesives (chemical bonding to tooth) and sometimes dental cements. They are designed to adhere to tooth structure without the requirement of a separate adhesive or etchant. Because of their simplicity of application, such materials have gained popularity day by day among dentists.

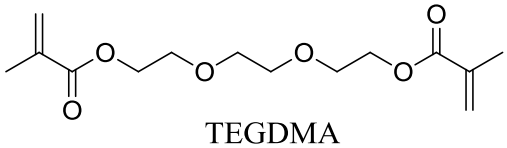
The self-adhesive resin cements are two-part materials which require generally hand-mixing. One component contains one of, or a mixture of some mono-, di- or multi-methacrylate monomers that are used in a variety of resin-based dental materials: Bis-GMA, urethane oligomers of 2,2-bis[4-(2-hydroxy-3-methacryloyloxy propyloxy) phenyl] propane (BisGMA), urethane dimethacrylate (UDMA), hydroxyethylmethacrylate (HEMA), glyceroldimethacrylate (GDMA), triethyleneglycol dimethacrylate (TEGDMA), etc. (Figure 1.8).



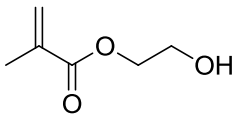
Bis-GMA



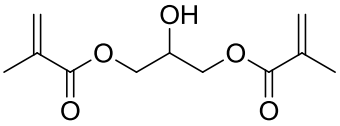
UDMA



TEGDMA



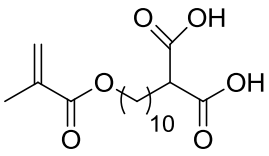
HEMA



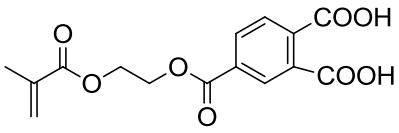
GDMA

Figure 1.8. Non-acidic methacrylate monomers in resin adhesive systems.

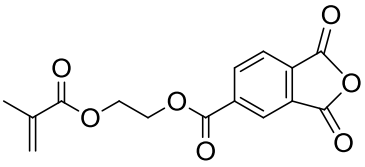
The other component contains an acid-functionalised monomer. Acidic monomers are used to demineralize the tooth tissue as well as form a strong adhesion with HAP. Moreover, they react with the filler (included in the first component) to release fluoride ions. These monomers are predominantly (meth)acrylate monomers containing either carboxylic acid groups (Figure 1.9), or phosphoric acid groups (Figure 1.10) [34, 58-60].



11-methacryloyloxy-1,1-undecanedicarboxylic acid (MAC-10)

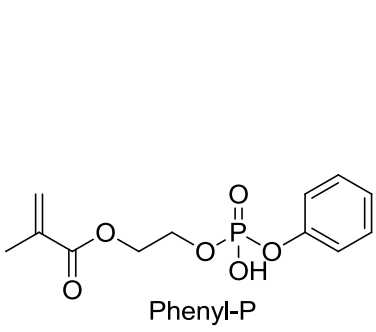


4-methacryloyloxyethyl trimellitic acid (4-MET)

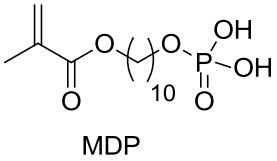


4-methacryloxyethyl trimellitic anhydride (4-META)

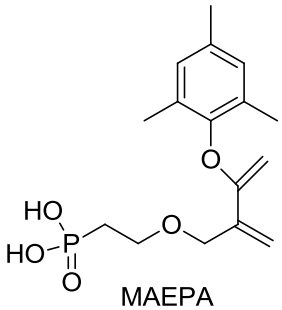
Figure 1.9. Carboxylic acid-containing monomers used in self-adhesive resin cements.



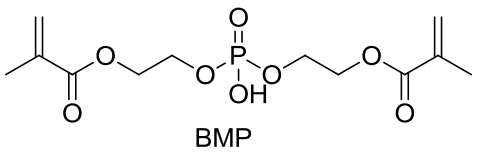
Phenyl-P



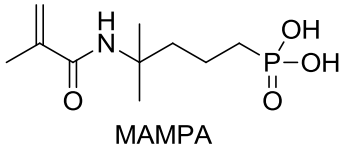
MDP



MAEPA



BMP



MAMPA

Figure1.10. Phosphoric and phosphonic acid-containing monomers in self-adhesive resin cements.

Catel *et al.* synthesized new phosphonic acid containing monomers and showed their potential to be used in such cements (Figure 1.11) [61].

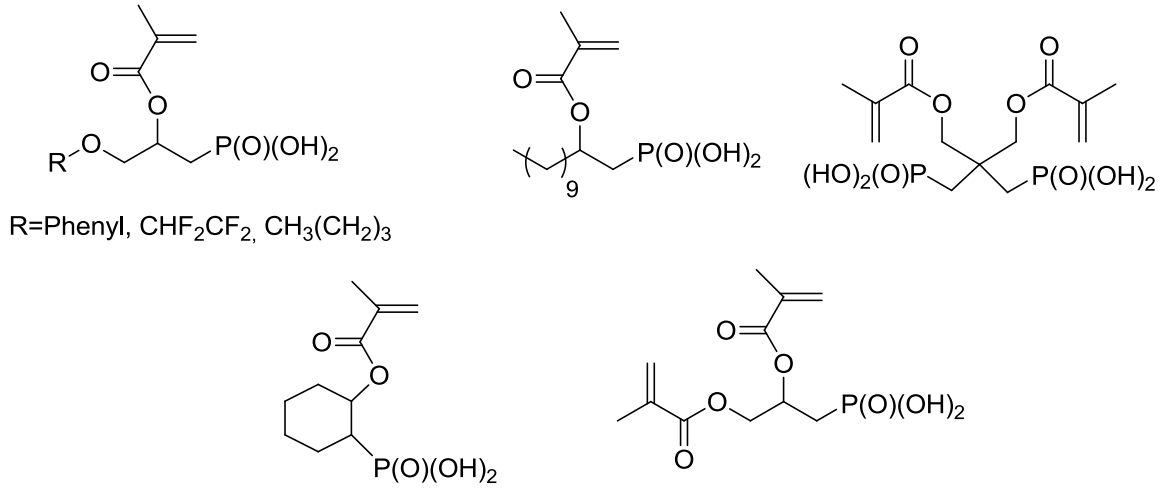


Figure 1.11. Acid monomers synthesized by Catel *et al*.

1.1.3. Interaction of Dental Materials with Tooth Tissues: Theory and Evaluation

1.1.3.1. Adhesion-decalcification model. The interaction of acids with human hard tissues such as tooth and bone is fundamental for dental medicinal processes [62]. For example, oral bacteria produce lactic acid which dissolves inorganic part of teeth resulting in dental caries [63]. On the treatment side, acids such as citric, maleic, or phosphoric have been used to pretreat dentin and enamel as a part of resin based adhesive systems.

Generally, the pH value of an acid is considered the major parameter for determining its interaction with mineralized tissue [64-66]. However, Yoshida and coworkers showed that an oxalic acid solution with a pH of 0.6 chemically bonds to HAP, but a maleic acid solution with a pH of 0.9 decalcifies it, so lower pH does not always mean more demineralization [67]. On the other hand, the weaker polyalkenoic or polycarboxylic acids (acid components in glass ionomer cements) decalcify tooth hardly at all, but rather bond to mineral tissue. To understand these sometimes counterintuitive interactions between the acid molecules and HAP-based tissue a model of "Adhesion-Decalcification" concept was proposed [57, 62, 67, 68].

According to this model, initially all acids ionically bond to the calcium of HAP. This first bonding phase goes together with the release of phosphate (PO_4^{3-}) and hydroxide (OH^-) ions from HAP into their own solution, such that the surface remains electroneutral

(Figure 1.12). Stability of formed Ca salt determines whether the molecule will remain bonded or not.

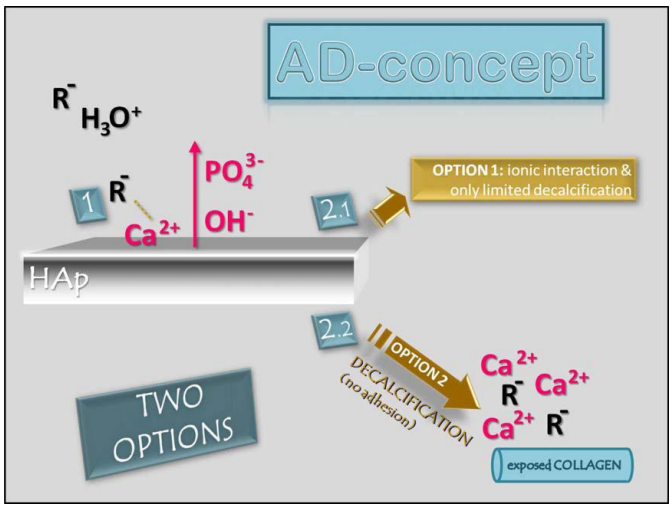


Figure 1.12. Schematic presentation of "Adhesion-Decalcification Concept".

1.1.3.2. Evaluation of bonding ability of monomers to hydroxyapatite or dentin. Fourier transform infrared spectrometer (FTIR) technique can be used to investigate chemical interaction of adhesive monomers with HAP and dentin. The monomer-HAP interaction can be observed by a decrease in the intensity of the peaks due to HAP at 564, 605, 1034 and 1093 cm^{-1} and appearance of new carbonyl peak around 1718 cm^{-1} (for 10-MDP and MAEPA) [69].

Scanning electron microscopy (SEM) can also be used to investigate the same interactions. For example, precipitation of Ca monomer salts can be observed for MAEPA and 10 MDP [69].

Another technique is X-ray diffraction (XRD) in which crystal phases are investigated to see formed layered structure. For example, Yoshihara *et al*. compares the HAP binding capability of 4-MET and Phenyl-P with the monomer 10-MDP which was studied earlier by Fukegawa *et al*. [70, 71]. In that study, the three detected new peaks for 10-MDP shows its crystalline phase constituted a layered structure similar to 10-MDP-Ca salt. Also within 24 h new peak at $2\theta = 11.8°$ was observed which is characteristic peak of $CaHPO_4.2H_2O$ (DCPD). 4-MET showed only DCPD formation in 1 week. Phenyl-P showed only formation of DCPD. Non-formation of Phenyl-P-Ca salt indicates the lack of

ionic interaction of HAP with this monomer (Figure 1.13). According to these results, three functional monomers interact differently with HAP; and the study proposes models for these monomers (Figure 1.14).

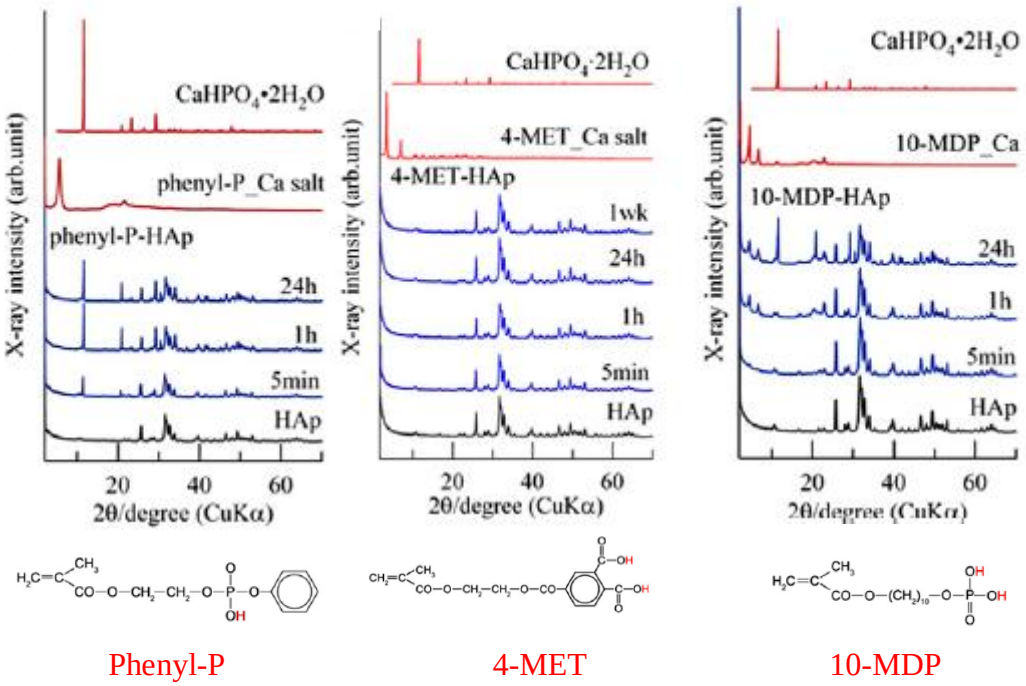


Figure 1.13. XRD analysis of Phenyl-P, 4-MET and 10-MDP.

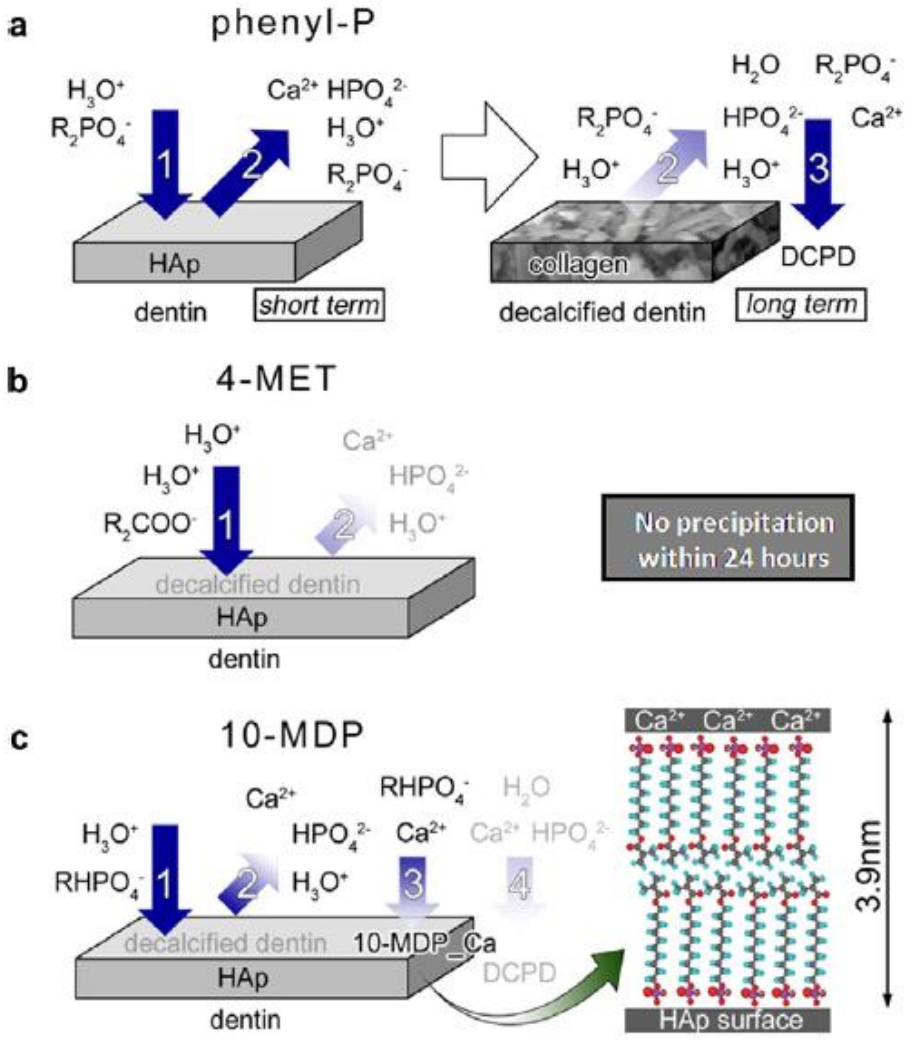


Figure 1.14. HAP interaction models for Phenyl-P, 4-MET and 10-MDP.

1.2. Bone-targeting Materials

Bone is a highly specified form of connective tissue, which provides an internal support system for the body [72]. The inorganic matrix of bone contains crystalline mineral salts and calcium, which is present in the form of hydroxyapatite. Seventy percent of bone is made up of this mineral with formula $Ca_{10}(PO_4)_6(OH)_2$. One of the most important molecules which can bind to HAP is bisphosphonate.

1.2.1. Bisphosphonates

Bisphosphonates (BPs) are structural analogues of naturally occurring pyrophosphate with increased chemical and enzymatic stability due to the replacement of the oxygen in P-O-P by a carbon, resulting in a P-C-P structure (Figure 1.15) [73].

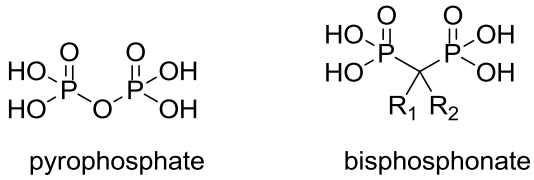


Figure 1.15. Structure of pyrophosphate versus general structure of a bisphosphonate.

This structure is the most important part of the bisphosphonates and is essential for biological activity. The $(HO)_2$-(O)P-C-P(O)-$(OH)_2$ group is used for chealation with Ca^{2+} ions in HAP so they bind to bone minerals and inhibit the resorption of living bone. The binding ability of BPs to bone mineral is enhanced by R_1 group (especially hydroxyl group). The structure and three-dimensional configuration of the R_2 side chain determines the cellular effects of BPs, and their potential as bone resorption inhibitors. Each BPs has different activity, determined by its unique side chain (Figure 1.16) [74].

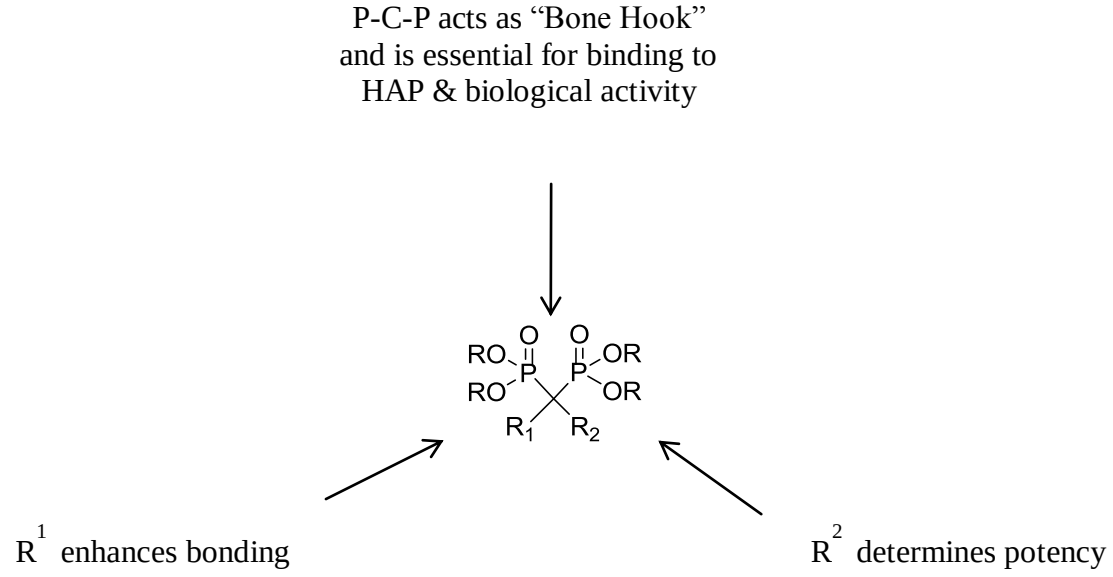


Figure 1.16. Structure-reactivity relationship of BPs.

These properties of BPs have been utilized to design and synthesize a vast array of bone-targeting compounds. The most commonly used bisphosphonates as anti-resorptive and bone-seeking agents are shown in Figure 1.17.

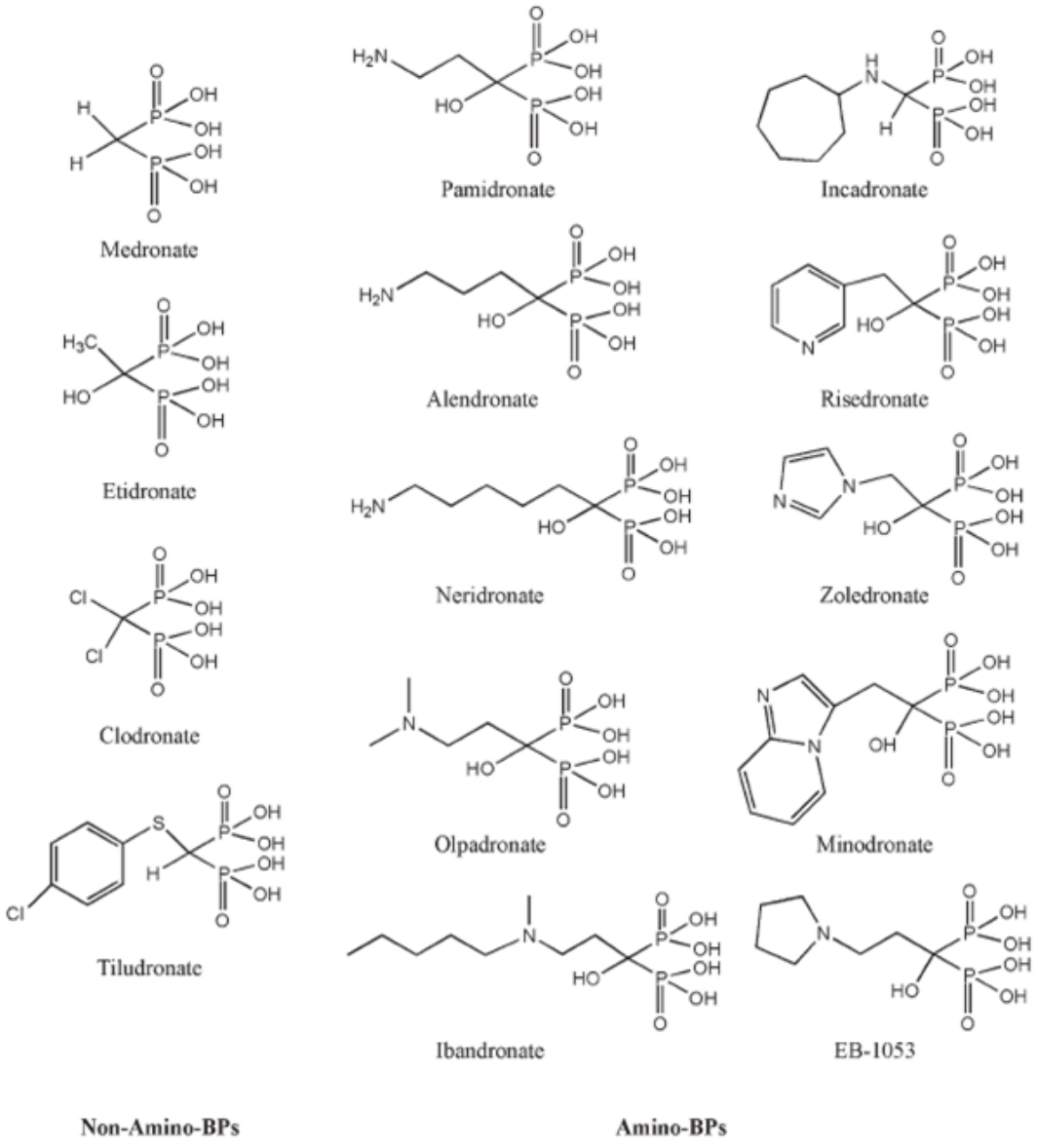


Figure 1.17. Examples of BP compounds.

Bisphosphonates have been used as drugs for bone diseases including osteoporosis, Paget's disease, bone cancer, etc [75]. They can inhibit bone resorption [76, 77], be used to deliver small molecule drugs [78-80], imaging agents [81, 82], peptides or proteins to the bone [83, 84].

1.2.2. Bone-targeted Drug Delivery

Although bisphosphonates are excellent therapeutic agents, polymers carrying high density of bisphosphonic acid groups (multimeric BPs) will obviously have advantages over small molecule BPs for bone targeting. The polymer-drug conjugates with bisphosphonic acid groups are expected to both improve efficiency of drugs by increasing half-life of drugs in circulation and solubility of drugs, and to decrease their toxicity (Figure 1.18) [85].

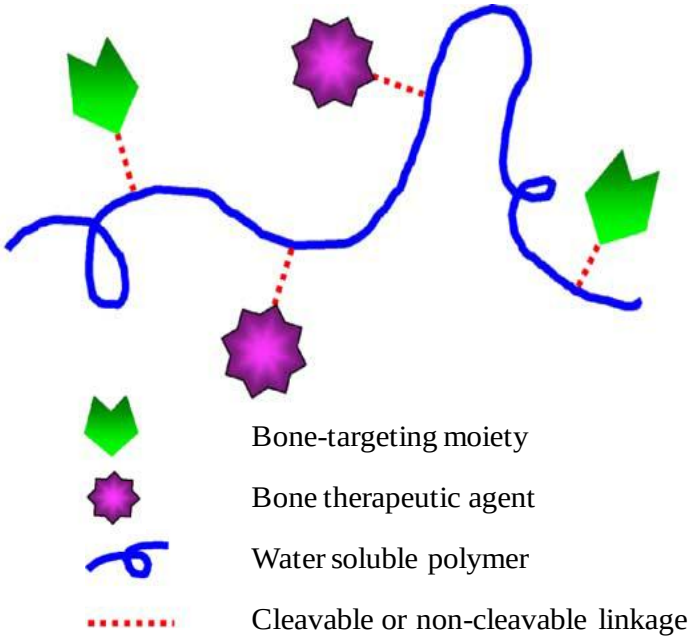


Figure 1.18. General structure of bone-targeting water-soluble polymeric drug delivery systems [85].

However, there are only a few reports about incorporation of bisphosphonates into polymers: Bone-targeting drug conjugates based on poly(ethylene glycol) (PEG) and poly[N-(2-hydroxypropyl)methacrylamide] (PHPMA) containing alendronate as bone targeting groups were tested (Figure 1.19 and Figure 1.20) [85-87].

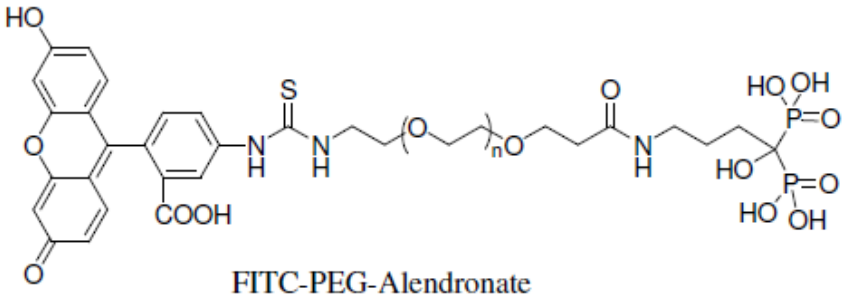


Figure 1.19. Structures of bone-targeting PEG conjugates.

Miller *et al.* used alendronate (ALN) targeted paclitaxel (PTX) containing [N-(2-hydroxypropyl)methacrylamide] (HPMA) copolymers to decrease migration and proliferation of human prostate adenocarcinoma (PC3) cells. They incorporated an enzymatically cleavable linker (GFLG) for both PTX and ALN. *In vitro* data demonstrated that the HPMA copolymer prevented migration and proliferation of the prosteate cancerous cell [87].

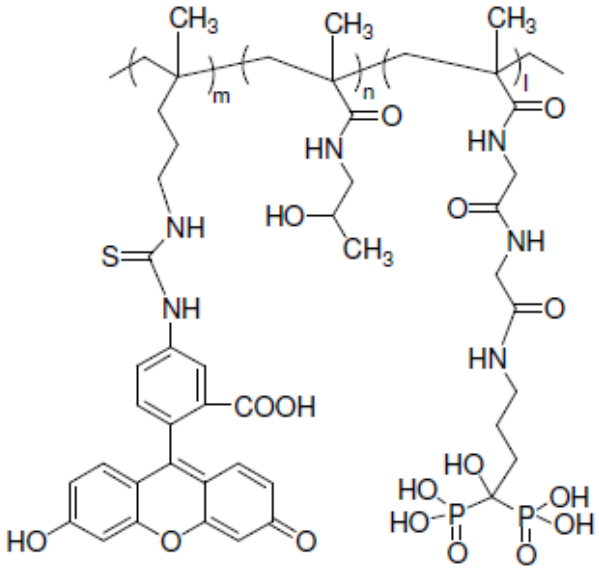
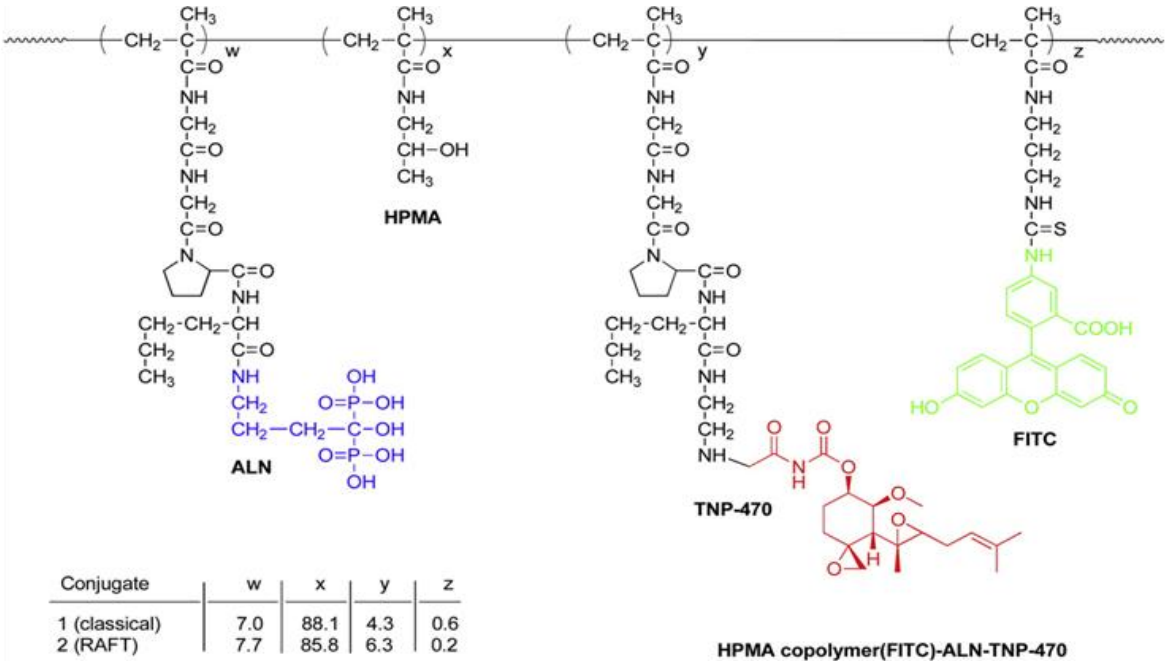


Figure 1.20. Structures of bone-targeting HPMA copolymer conjugate.

In another study Miller *et al*. showed the *in vivo* tumor inhibition and safety profiles on Balb/c mice. The PTX, ALN, HPMA copolymer demonstrated no significant toxicity while free PTX significantly reduced white blood cell (WBC) counts. Moreover,

copolymer of PTX, ALN HPMA or free PTX+ALN in combination were administrated, the polymer conjugate inhibited tumor growth by 60%, while free PTX+ALN only inhibited 37% as compared to controls [88].

Similar experiments were conducted using TNP-470 bound to HMPA copolymer conjugate targeted with ALN. TNP-470 is an anticancer agent which showed high efficacy in clinical trials but its side effects prevented clinical applications [89]. In vivo studies indicated (Figure 1.21) that not only do ALN and TNP-470 have synergism, but revealed that the HPMA copolymer induced a decrease in osteosarcoma growth by 96% compared to the control, as opposed to 45% with free ALN in combo with TNP- 470 [90, 91].



Conjugate	w	x	y	z
1 (classical)	7.0	88.1	4.3	0.6
2 (RAFT)	7.7	85.8	6.3	0.2

Figure 1.21. TNP-470- HPMA copolymer conjugation targeted with ALN.

Similar to the PTX, ALN, HPMA copolymer, TNP-470, ALN, HPMA copolymer's toxicity is low, as opposed to ALN+TNP-470, which caused in vivo weight loss, neurological dysfunction, and low WBC counts [92].While ALN facilitates the delivery of a drug to the bones, the conjugation with a polymer provides targeting to tumor tissue within the bones.

1.3. Bone-tissue Engineering

In recent years, biomaterials from polymers for applications such as total hip replacement, dental implants, and screws for fracture fixation have been designed to have better integration with bone than metals or alloys previously used for these purposes [93-95]. Composites with a dispersed synthetic biodegradable polymer in calcium phosphate were prepared.

In addition to composite preparation, many approaches using biomimetic principles to mineralize polymer scaffolds were developed. Natural bone consists of a matrix of collagen upon which carbonated apatite deposition occurs. Anionic proteins serve as nucleators and inhibitors, the concentration depending on bone type and age, control this deposition [96].

Another approach is the functionalization of biomaterials with negatively charged functional groups which provide nucleating sites to induce mineralization of polymer scaffolds [97-99]. The incorporation of anions such as carboxylate or oxyphosphorus (e.g. phosphate, phosphonate) groups into polymers is inspired from the nature and gives biomaterials with the calcification ability. Phosphorus-containing anionic functional groups are among the most efficient groups for mineralization.

Phosphorus-containing polymers have been shown to mineralize in vitro and vivo such that they have a radioopacity similar to that of bone [100]. Polymers containing phosphate and phosphonate groups mineralize faster and more completely than analogues lacking the phosphorus-containing groups, therefore vinyl monomers containing these groups were used for copolymerization [101-104]. Gemeinhart *et al*. developed a copolymer of vinyl phosphonic acid (VPA) and acrylamide and found that, at 30% VPA concentration, osteoblast-like cells exhibited better proliferation, adhesion, differentiation, and ability to mineralize the polymer surface than other monomer ratios of VPA and pure polyacrylamide [104]. Synthesis of phosphoester-containing poly(ethyleneglycol) (PEG-based) hydrogels were described (Figure 1.22) [105-107].

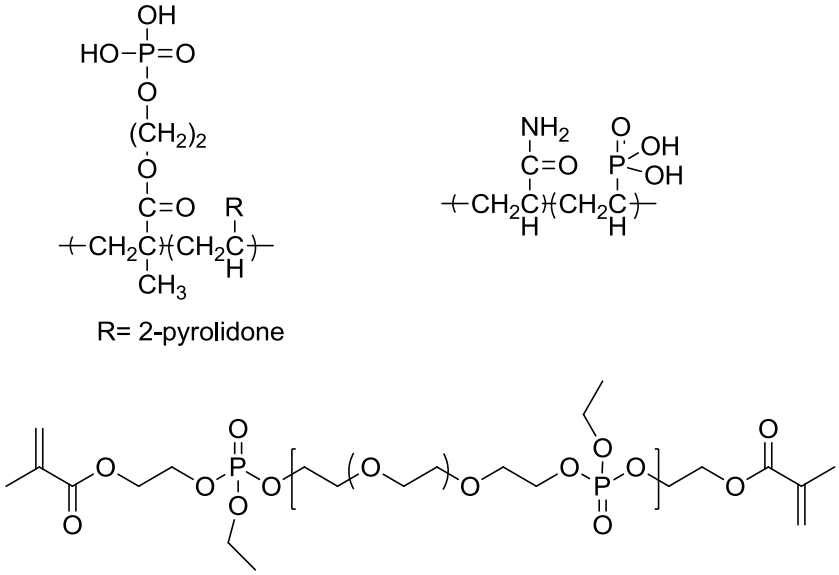


Figure 1.22. Structures of phosphorus-containing materials.

A hydrogel prepared from copolymer of N-acrylpamidronate and N-isopropylacrylamide was used as scaffold for mineralization of HAP (Figure 1.23) [108], bisphosphonate derivatives of cationic polymers such as poly(l-lysine) and poly(ethylenimine) were tested for affinity to HAP [109] and bisphosphonate-modified polyurethanes were prepared to resist calcification around implants [110]. Hybrid hydrogels of a hyaluronic acid derivative functionalized with BP ligands were prepared (Figure 1.24) [111].

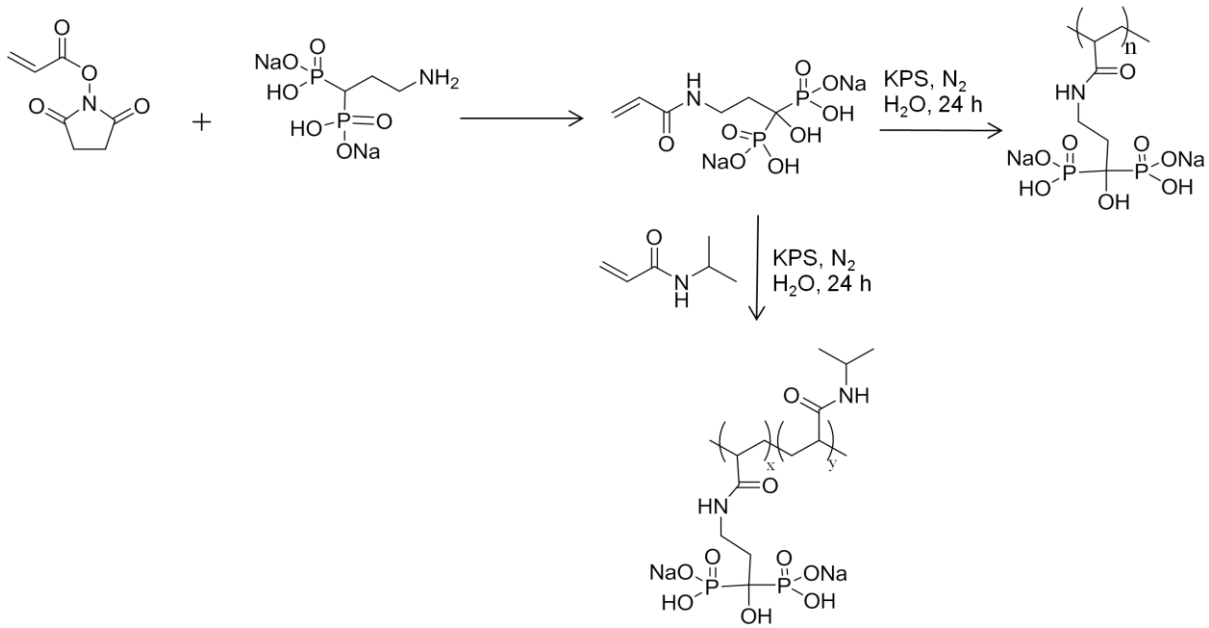


Figure 1.23. Synthesis and hydrogelation of polymers consisting of pamidronate [108].

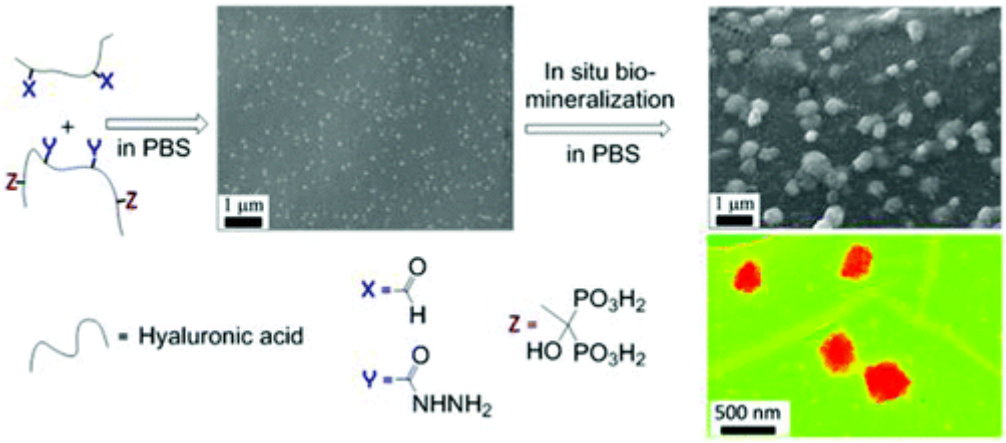


Figure 1.24. Hyaluronic acid hydrogels with bisphosphonate ligands [111].

1.4. Metal-binding Materials

Phosphonic acids and phosphonate esters form variety of monomeric and polymeric complexes by interacting with a large number of transition and nontransition metal salts. In most of these complexes, the oxygen atom of phosphonic acid group formed complexation with the metal ions [-C-P=O→M]. In the acidic pH range, the metal ion is not bound by four oxygen atoms, but only two oxygen atoms of two different phosphonic acid groups are used for complexation, because the second OH group of the phosphonic acid molecule is not dissociated (Figure 1.25) [112, 113].

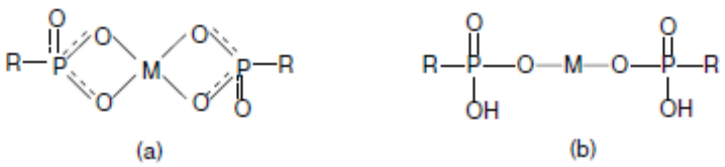


Figure 1.25. Suggested structure of the metal ion-monomer complex at different pH's, (a) in basic medium (b) in acidic medium.

Generally, bi-functional phosphonic acids, as the presence of a secondary functional group extends the versatility of metal phosphonates to new and more selective applications and can result in more porous materials [114, 115]. The presence of two different functional groups provides numerous chelation modes as shown in Figure 1.26 [116].

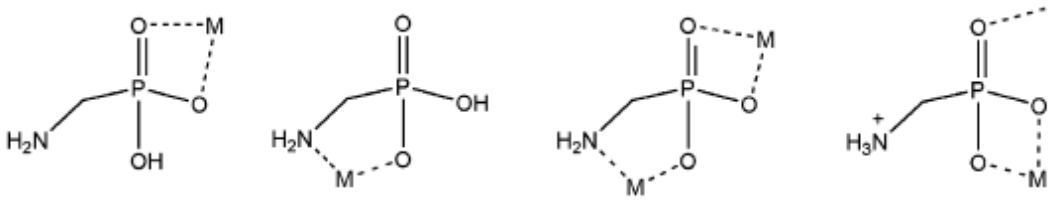


Figure 1.26. Possible coordination form of aminomethylphosphonic acid.

Samanamu *et al.* showed the chelating modes by exploring the chemical behavior of aminomethylphosphonic acid (ampa) with different metal salts such as zinc, cadmium, mercury, lead, silver and copper (Figure 1.27) [116].

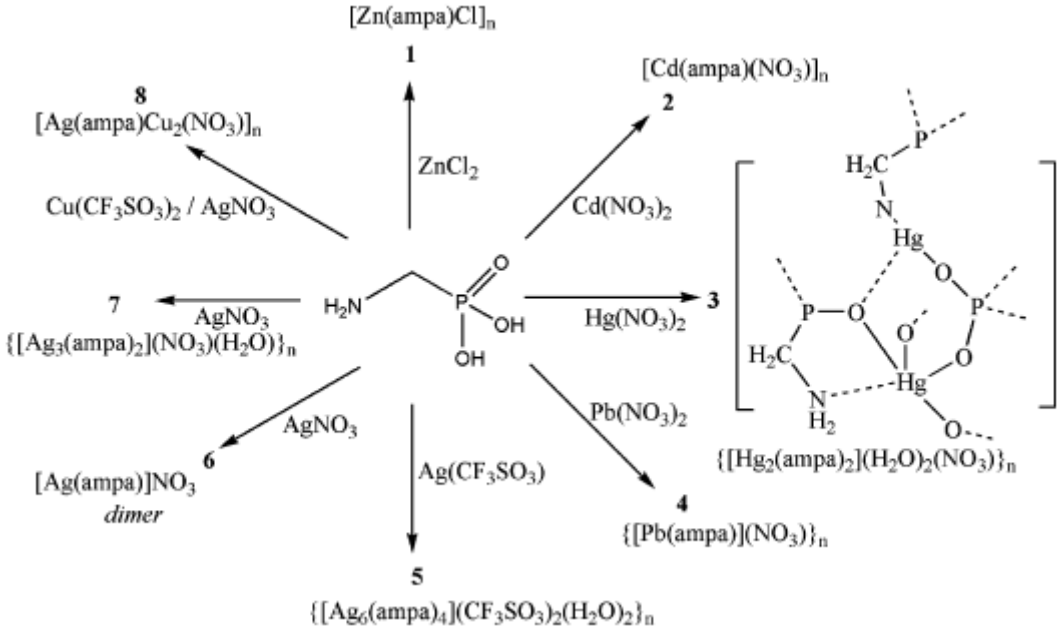


Figure 1.27. Reactions of aminomethylphosphonic acid with metal precursors [116].

Phosphonate-containing polymers are also considered to act as excellent ion exchange and adsorption resins for the removal of trace amount of toxic metal pollutants from potable water and waste water [112-117]. Riedelsberger and Jaeger showed that monomeric and polymeric aminomethylphosphonic acids exhibit good complexation ability for the transition metal ions Cd^{+2} and Hg^{+2} (Figure 1.28) [13].

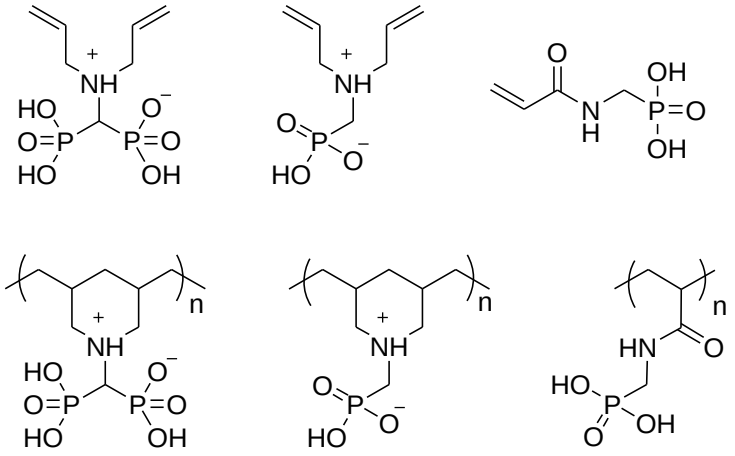


Figure 1.28. Monomeric and polymeric aminomethyl phosphonic acids.

Aminophosphonate monomers and polymers, in which cationic and anionic functional groups are located on the same monomer/repeat unit, are also known as phosphobetaines [118, 119]. Phosphobetaines are specific type of polybetaines in which the anionic group is phosphonate instead of carboxylate or sulfonate as in the cases of carboxybetaines or sulfobetaines [120-125].

The alpha-aminomethylene phosphonate betaines are useful in a variety of applications including chelating agents for corrosion inhibition, soil anti-redeposition agents in detergents, as crystal modifiers [124, 126-128].

Besides their chelating abilities to metals, aminophosphonic acids which are structural analogues of amino acids exhibits biological activities such as inhibition of enzymes, antibacterial activity, neuroative compounds, anticancer drugs or pesticides [129-132].

Moreover, phosphobetaine-based polymers or surfaces have been shown to greatly reduce nonspecific protein adsorption [133-139]. They are considered as biomimetic fouling-resistant materials, since they contain phosphorylcholine headgroups, which are found in the outside layer of cell membranes. Phosphobetaine monomers, such as 2-methacryloyloxyethyl phosphorylcholine (MPC) have very versatile usage due to its biocompatibility (Figure 1. 29) [6, 140-146].

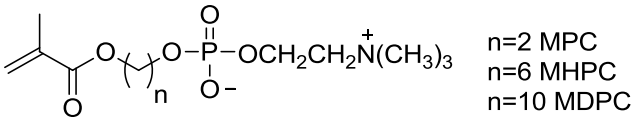


Figure 1.29. Structure MPC and its derivatives 6-methacryloyloxyethyl phosphorylcholine (MHPC) and 10-methacryloyloxydecyl phosphorylcholine (MDPC).

N. Nakabayashi and Y. Iwasaki showed that MPC copolymers can suppress adsorption of proteins and prevent their conformational change both on the surface and in the solution [147, 148].

Moreover, hydrogels containing MPC moieties were formed from aqueous solutions with water soluble MPC based random and block polymers because of hydrogen bonding formation. In Figure 1.30 there are samples of these copolymers; poly(2-methacryloyloxyethyl phosphorylcholine-co-methacrylic acid) and poly(2-methacryloyloxyethyl phosphorylcholine-co-butyl methacrylate). These copolymers acted as hydrogel physical cross-linkers [6, 149, 150].

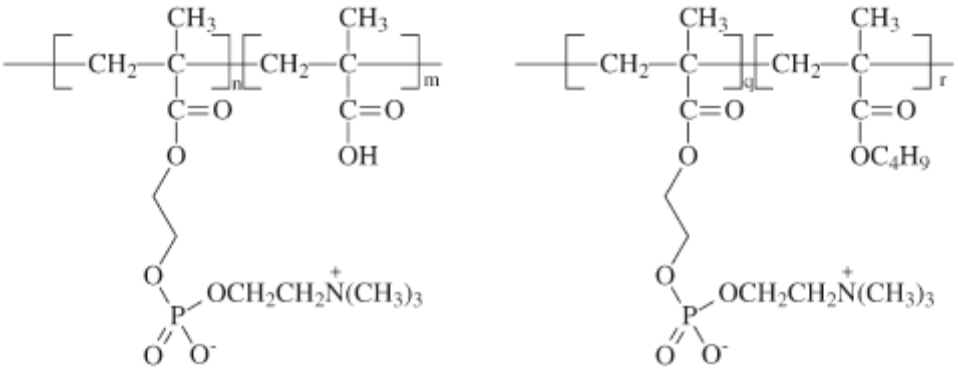


Figure 1.30. Chemical structure of copolymers leading to phospholipid polymer-hydrogel.

Phosphonated poymers are also used for artifical medical materials; to control the surface properties under different conditions (such as pH, ionic strength, etc.) and formation of hydrophilic polymer chains of PEG on these artifical materials are very crucial. Zoulalian *et al*. synthesized alkyl-phosphonated and PEG methacrylated polymer in the presence of a chain transfer agent. Figure 1.31 shows the resulting PEG-

poly(phosphonate)-terpolymer molecules self-assembled on TiO_2 surface to form a layered structure. They investigated the stability of layered structure at different pHs [151].

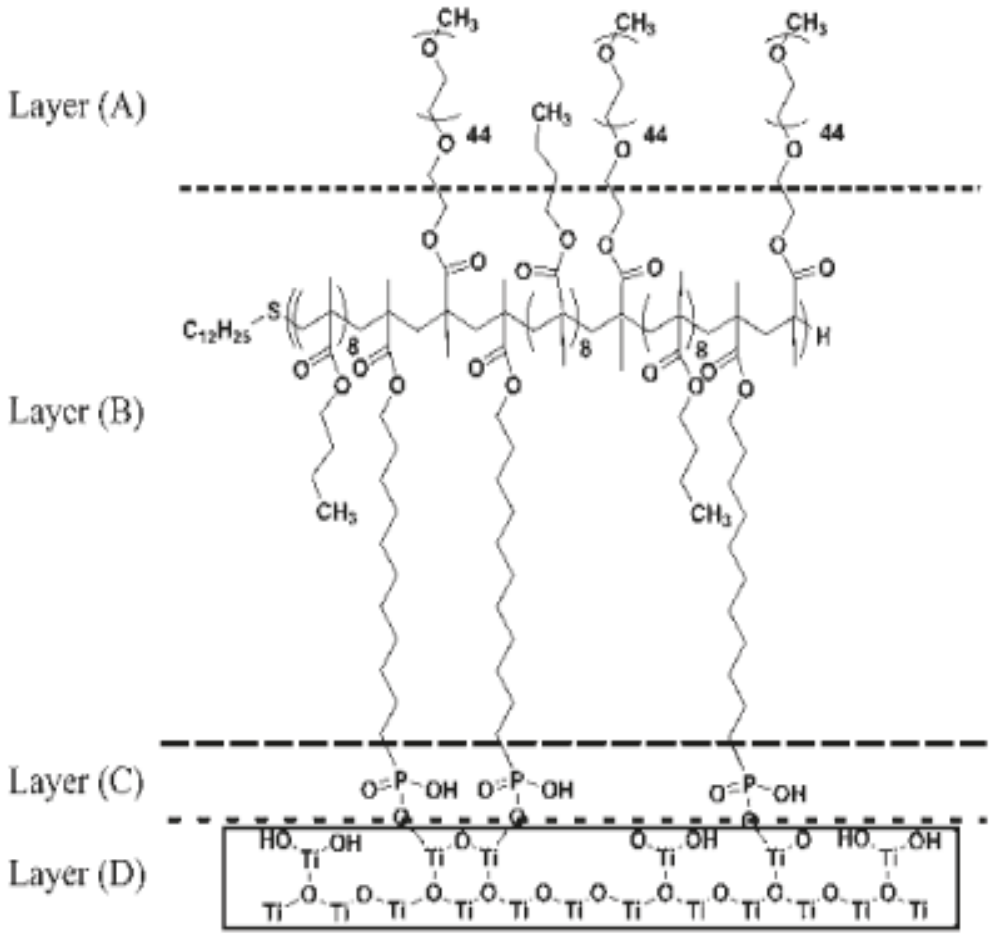


Figure 1.31. The PEG-poly(alkyl phosphonate) terpolymer on TiO_2 surface in a four-layer system.

2. OBJECTIVES

The purpose of this thesis is to design, synthesize, characterize and evaluate novel, highly reactive phosphonate, phosphonic acid, (bis)phosphonate and (bis)phosphonic acid-containing monomers derived mostly from alkyl α-hydroxy methacrylates (RHMA) for mainly biomedical and environmental applications.

The (bis)phosphonate- and (bis)phosphonic acid-containing monomers reported have potential as dental adhesives. They are designed for good binding to tooth material; and are expected to have improved shelf life and bonding reliability, as they will not hydrolyze in aqueous acidic conditions due to the presence of ether linkages in their structures.

Homopolymers and copolymers of the (bis)phosphonic acid-containing monomers reported here with biocompatible monomers are expected to have potential to be used in bone-targeting drug delivery applications.

The aminophosphonate-containing 1,6-heptadiene monomers reported offers a great variety of water soluble zwitterionic monomers and polymers for potential applications in water treatment.

Therefore the objectives of this thesis are (i) synthesis of α-amino phosphonates and gem-hydroxy-diphosphonates, (ii) attachment of these amines and alcohols to RHMA derivatives to obtain novel monomers desired, (iii) polymerization of the monomers and evaluation of their reactivities and polymer properties.

3. SYNTHESIS AND POLYMERIZATIONS OF NOVEL BISPHOSPHONATE-CONTAINING METHACRYLATES DERIVED FROM ALKYL α- HYDROXYMETHACRYLATES

3.1. Introduction

Bisphosphonates (BPs) are structural analogues of naturally existing pyrophosphate with increased chemical and enzymatic stability due to the replacement of the oxygen in P-O-P by a carbon, resulting in a P-C-P structure [73]. They have strong affinity for bone mineral, hydroxyapatite (HAP), enabling them to chelate calcium ions and prevent bone dissolution. Bisphosphonates have been used as drugs for bone diseases including osteoporosis, Paget's disease, bone cancer, etc [73]. They can inhibit bone resorption [75, 151], and can be used to deliver small molecule drugs [77-80], imaging agents [81, 82], peptides or proteins to bone [83, 84]. Although bisphosphonates are excellent therapeutic agents, polymers carrying a high density of bisphosphonic acid groups (multimeric BPs) will obviously have advantages over small molecule bisphosphonates for bone targeting. Polymer-drug conjugates with bisphosphonic acid groups are expected to both improve efficiency of drugs by increasing half-life of drugs in circulation and solubility of drugs, and to decrease drug toxicity.

However, there are only a few reports about incorporation of bisphosphonates into polymers: bone-targeting drug conjugates based on poly(ethylene glycol) and poly[*N*-(2-hydroxypropyl)methacrylamide] containing alendronate as bone targeting groups were tested [85,86], a hydrogel prepared from a copolymer of *N*-acrylpamidronate disodium salt and *N*-isopropylacrylamide was used as a scaffold for mineralization of HAP [108], bisphosphonate derivatives of cationic polymers such as poly($_{L}$-lysine) and poly(ethylenimine) were tested for affinity to HAP [109] and bisphosphonate-modified polyurethanes were prepared to resist calcification around implants [110]. More recently bisphosphonate-containing monomers were investigated for self-etching dental adhesive applications [35-37, 47] which facilitate adhesion of dental restoratives and orthodontic

appliances to dental tissue. They were also incorporated into adhesion/anticorrosive coatings [152].

In this work, design and synthesis of two new bisphosphonate (ethyl ester) containing monomers based on alkyl α-hydroxymethacrylates are reported. Their homo- and copolymerizations with poly(ethyleneglycol) methyl ether methacrylate (PEGMA) followed by hydrolysis of bisphosphonate groups to bisphosphonic acids leads to new polymeric systems with strong affinity to HAP.

3.2. Experimental

3.2.1. Materials and Apparatus

3.2.1.1. Materials. Ethyl α-bromomethacrylate (EBBr), *tert*-butyl α-bromomethacrylate (TBBr), tetraethyl 4- hydroxybutane-1,1-diyldiphosphonate were synthesized according to literature procedures [153-156]. Poly(ethylene glycol) methyl ether methacrylate (PEGMA) (M_n =950), 2,2'-azobis(isobutyronitrile) (AIBN), 2,2'-dimethoxy-2-phenyl acetophenone (DMPA), HAP, all other reagents and solvents were obtained from Aldrich Chemical Co. and used as recieved. Trimethylsilyl bromide (TMSBr; Aldrich, Taufkirchen, Germany) was distilled before use. Tetrahydrofuran (THF) was dried by heating under reflux over sodium in the presence of benzophenone as indicator.

3.2.1.2. Apparatus. Monomer characterization involved ^{1}H NMR, ^{13}C NMR and ^{31}P NMR spectroscopy (Varian Gemini 400 MHz) and Fourier transform infrared (FTIR) spectroscopy (T 380). The photopolymerizations were carried out on a TA Instruments Q100 differential photocalorimeter. Gel permeation chromatography was performed using a Viscotek apparatus equipped with a refractive index detector, using THF solvent and polystyrene standards. Elemental analysis was carried out using a Thermo Electron SpA FlashEA 1112 elemental analyser (CHNS separation column, polytetrafluoroethylene; 2 m, 6x5 mm). Mass spectrum was obtained with a Thermo Finnigan LCQ Advantage Max instrument in electrospray ionization positive, ESI (+), mode.

3.2.2. Synthesis of Monomers

3.2.2.1. Ethyl 2-((4,4-bis(diethoxyphosphoryl)butoxy)methyl)acrylate (**A1**). To an ice-cold mixture of tetraethyl 4- hydroxybutane-1,1-diyldiphosphonate (0.30 g, 0.87 mmol) and triethyl amine (0.176 g, 1.73 mmol) in dry THF (4 mL) under nitrogen, EBBr (0.25 g, 1.31 mmol) was added dropwise. The mixture was stirred at 60 °C for 24 h under nitrogen atmosphere. After removal of the solvent, the mixture was diluted with 5 mL of dichloromethane and extracted with water (3x3 mL). The organic layer was dried over anhydrous sodium sulfate, filtered and evaporated under reduced pressure to leave the crude product, which was purified by column chromatography on silica gel 60 (70-230 mesh) using hexane initially and gradually changing to ethyl acetate as eluent. The pure product was obtained as light yellow oil in 38 % yield.

^{1}H NMR (400 MHz, $CDCl_3$, δ): 1.32 (t, $^{3}J_{HH}$=7.1 Hz, 15H, CH_2CH_3), 1.88 (m, 2H, CH_2CHP), 1.99 (m, 2H, $CH_2CH_2CH_2$), 2.41 (tt, $^{2}J_{HP}$=24.1 Hz; $^{3}J_{HH}$=6.1 Hz, 1H, CHP_2), 3,50 (t, $^{3}J_{HH}$=6.1 Hz, 2H, CH_2CH_2O), 4.13 (m, 10H, OCH_2CH_3), 4.19 (m, 2H, CCH_2O), 5.84, 6.27 ppm (s, 2H, $C=CH_2$).

^{13}C NMR (400 MHz, $CDCl_3$, δ): 14.04 (CH_3CH_2OC), 16.29 (CH_3CH_2OP), 22.48 (CH_2CHP_2), 28.83 ($CH_2CH_2CH_2$), 36.32 (CH_2CHP_2), 60.54 (CH_2OC), 62.44 (CH_2OP), 68.67 (CH_2CH_2O), 70.23 (CCH_2O), 125.01 (CH_2=C), 137.48 (CH_2=C), 165.77 ppm (C=O).

^{31}P NMR ($CDCl_3$, δ): 24.22 ppm.

FTIR (ATR): ν = 2877 (C-H), 1714 (C=O), 1636 (C=C), 1258 (P=O), 1021 and 958 cm^{-1} (P-O-Et).

Calcd. for $C_{18}H_{36}O_9P_2$: C 47.16, H 7.92, O 31.41, P 13.51; found: C 46.95, H 8.01.
MS (m/z) 458.86 $(M+H)^+$.

3.2.2.2. *Tert*-butyl 2-((4,4-bis(diethoxyphosphoryl)butoxymethyl)acrylate (**A2**). Monomer **A2** was synthesized using the same procedure as for monomer **A1** using TBBr instead of EBBr. The pure product was obtained as yellow oil in 32 % yield.

^{1}H NMR (400 MHz, $CDCl_3$, δ): 1,34 (t, $^{3}J_{HH}$=7.1 Hz, 12H, OCH_2CH_3), 1.49 (s, 9H, $C(CH_3)_3$), 1.86 (m, 2H, CH_2CHP), 1.98 (m, 2H, $CH_2CH_2CH_2$), 2.33 (tt, $^{2}J_{HP}$=24.1 Hz; $^{3}J_{HH}$=6.1 Hz, 1H, CHP_2), 3.49 (t, $^{3}J_{HH}$=6.1 Hz, 2H, CH_2CH_2O), 4,09 (s, 2H, CH_2O), 4,15 (m, 8H, OCH_2CH_3), 5.75, 6.15 ppm (s, 2H, $C=CH_2$).

^{13}C NMR (400 MHz, $CDCl_3$, δ): 13.96 (CH_3CH_2O), 16.23 (CH_2CHP_2), 27.92 ($C(CH_3)_3$), 31.77 ($CH_2CH_2CH_2$), 35.06 (CH_2CHP_2), 62.37 (CH_3CH_2O), 68.76 (CH_2CH_2O), 70.19 (CCH_2O), 80.68 ($C(CH_3)_3$), 123.96 ($CH_2=C$), 138.76 ($CH_2=C$), 164.99 ppm (C=O).

^{31}P NMR ($CDCl_3$, δ): 23.78 ppm.

FTIR (ATR): ν = 2978 (C-H), 1711 (C=O), 1640 (C=C), 1252 (P=O), 1018 and 964 cm^{-1} (P-O-Et).

3.2.3. Polymerizations

3.2.3.1. Thermal polymerizations. The thermal homo- and copolymerizations were carried out with standard freeze-evacuate-thaw procedures:

(i) The homopolymerizations of monomers **A1** and **A2** were carried out in bulk at 65 °C with AIBN as initiator. The polymers were purified by precipitation into hexane in which monomers are soluble.

(ii) The copolymerizations of monomers with PEGMA in three different ratios were carried out (monomer: PEGMA, 10:90, 30:70 and 50:50) in methanol at 65 °C with AIBN as initiator. The copolymers were purified by precipitation into solvents where both monomers were soluble.

3.2.3.2. Photopolymerization. The photopolymerizations were carried out using a DSC equipped with a mercury arc lamp. The samples (3-4 mg) containing 2.0 mol % initiator

were irradiated for 10 min at 40 °C with an incident light intensity of 20 mW/ cm^2 under a nitrogen flow of 20 mL min^{-1}. The heat flux as a function of reaction time was monitored using DSC under isothermal conditions, and both the rate of polymerization (R_p) and degree of conversion were calculated as a function of time. The theoretical value used for the heat of reaction (ΔH_p) was 13.1 kcal/mol for methacrylate double bonds [157, 158]. Rates of polymerization were calculated according to the formula

$$Rate = \frac{\left({}^{Q}/_{s}\right)M}{n(\Delta Hp)m} \qquad (3.1)$$

where Q/s is heat flow per second, M the molar mass of the monomer, n the number of double bonds per monomer molecule, ΔH_p is the heat released per mole of double bonds reacted and m the mass of monomer in the sample.

3.2.4. Hydrolysis of Polymers

The selective hydrolysis of poly-**A1** was carried out using TMSBr:

TMSBr (0.696 g, 4.55 mmol) was added dropwise to a solution of poly-**A1** (0.1 g) in 1.0 mL dry dichloromethane in an ice bath and under nitrogen. After 4 h of reflux, the solvent was removed under reduced pressure. The residue was diluted with 3.0 mL of methanol and the solution was stirred at room temperature overnight. After evaporation of methanol, the polymer was obtained as oil in 60% yield.

3.3. Results and Discussion

3.3.1. Monomer Synthesis and Characterization

Two different bisphosphonate-containing monomers (**A1** and **A2**), where the bisphosphonate groups are attached to double bond through an ether link were synthesized in three steps: (i) monoalkylation of the sodium carbanion of tetraethyl

methylenebisphosphonate in dry THF with 2-(3-bromopropoxy)tetrahydro-2*H*-pyran to give the first intermediate (tetraethyl 4-(tetrahydro-2*H*-pyran-2yloxy)butane-1,1-diyldiphosphonate) in 62 % yield, (ii) deprotection of the tetrahydropyranyl group of the first intermediate using Amberlyst H-15 in methanol to afford the second intermediate (tetraethyl 4-hydroxybutane-1,1-diyldiphosphonate) in 85 % yield, (iii) reaction of the second intermediate with EBBr or TBBr in the presence of TEA in THF at 60 °C for 24 h to give monomers **A1** and **A2** (Figure 3.1). These monomers were obtained as light yellow liquids after purification by column chromatography with overall yields of ca 38 and 32 %, respectively. The monomers are soluble in hexane, acetone, THF, methylene chloride and methanol but insoluble in water (Table 3.1).

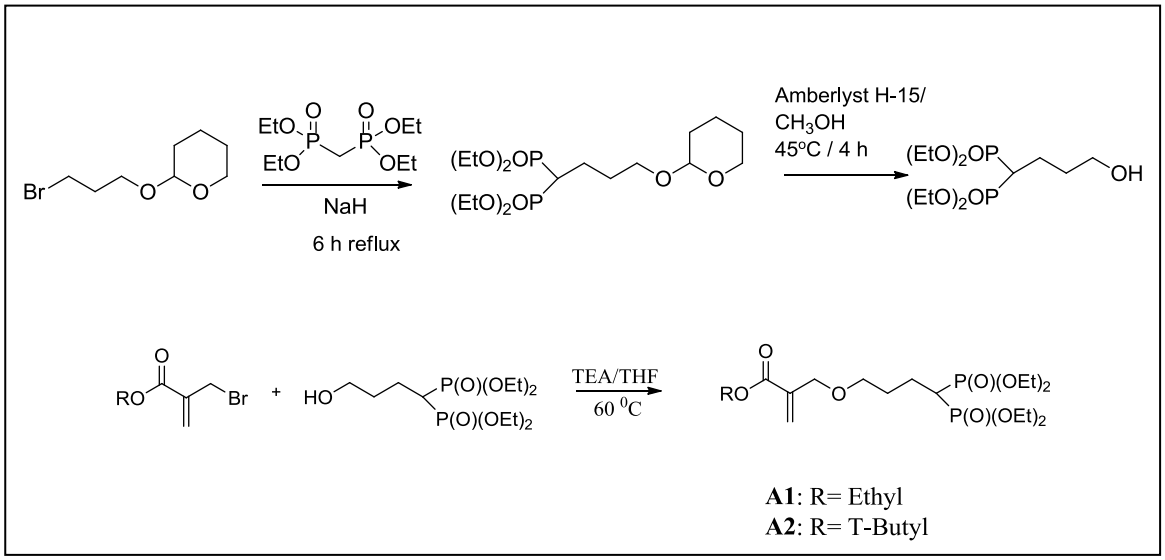


Figure 3.1. Synthesis of monomers **A1** and **A2**.

Table 3.1. Solubility of monomers and polymers in selected solvents.

Monomers/ Polymers	Water	MeOH	Acetone	CH_2Cl_2	THF	Ether	Hexane
A1	-	+	+	+	+	+	+
A2	-	+	+	+	+	+	+
Poly-**A1**	+	+	+	+	+	+	-
Poly-**A2**	+	+	+	+	+	+	-

The characterization of the novel monomers was carried out by ^{1}H NMR, ^{13}C NMR, ^{31}P NMR and FTIR spectroscopies and the data obtained are in agreement with the

expected monomer structures. For example, the ^{1}H NMR spectrum of monomer **A1** shows ethyl peaks at around 1.32 and 4.13 ppm, the methylene protons attached to oxygen at 4.19 ppm and the double-bond protons at 5.84 and 6.27 ppm. The proton attached to bisphosphonate carbon is confirmed by the presence of a triplet at 2.41 ppm (Figure 3.3). The peak of the phosphorus atom at 23.78 ppm in the ^{31}P NMR spectrum of **A2** is characteristic of the phosphonate group (Figure 3.2). In the FTIR spectrum of the same monomer, the bisphosphonate function is evidenced by the P=O band at 1258 cm^{-1} and P-O-C bands at 1021 and 958 cm^{-1} (Figure 3.7).

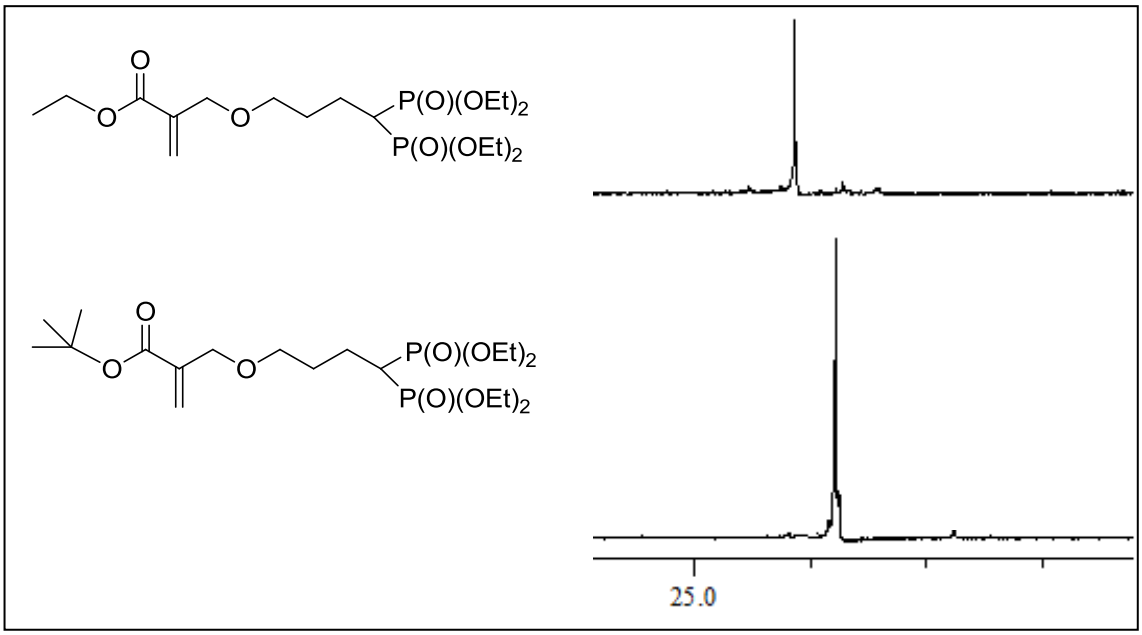


Figure 3.2. ^{31}P NMR spectra of **A1** and **A2**.

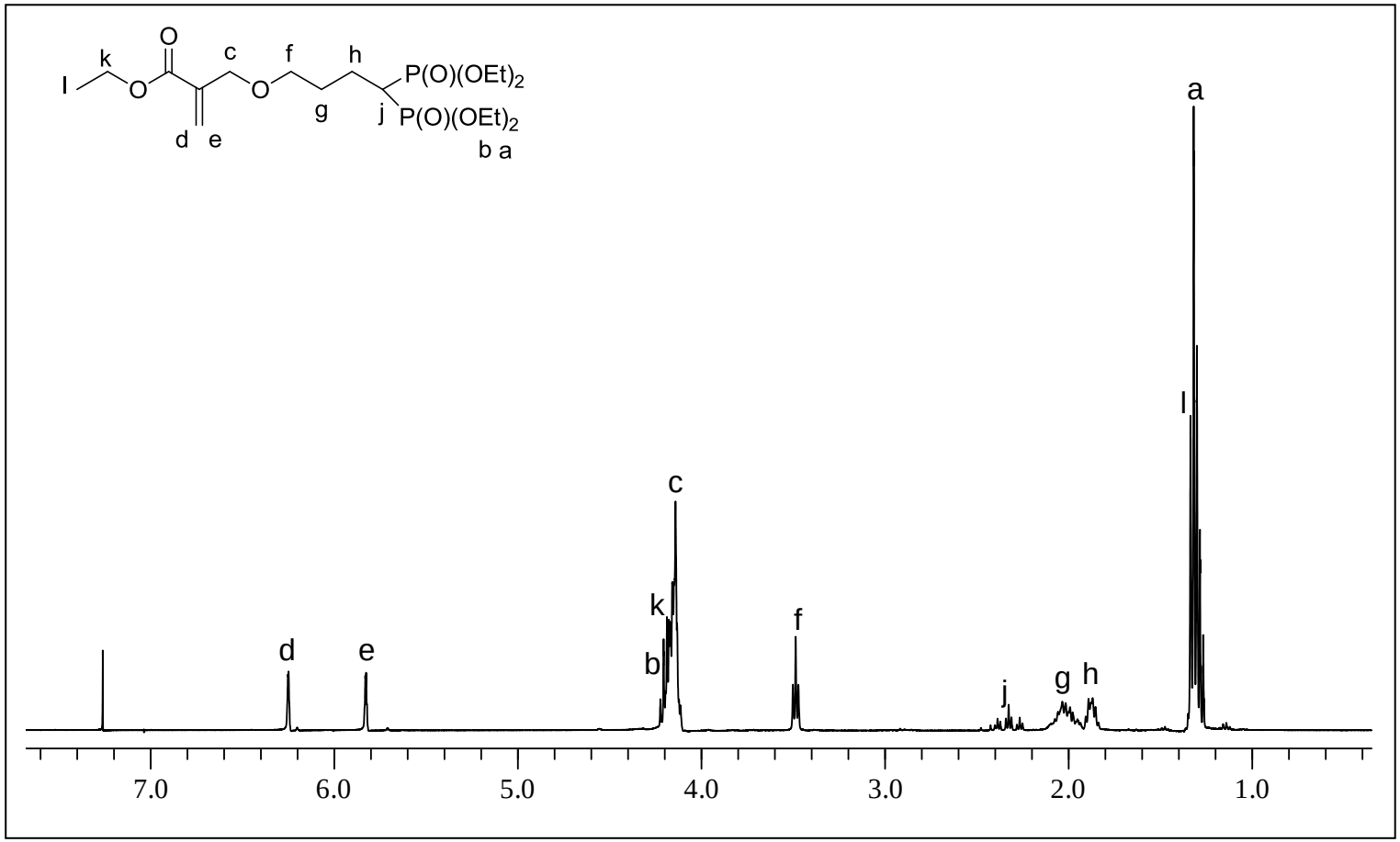


Figure 3.3. ^{1}H NMR spectrum of **A1**.

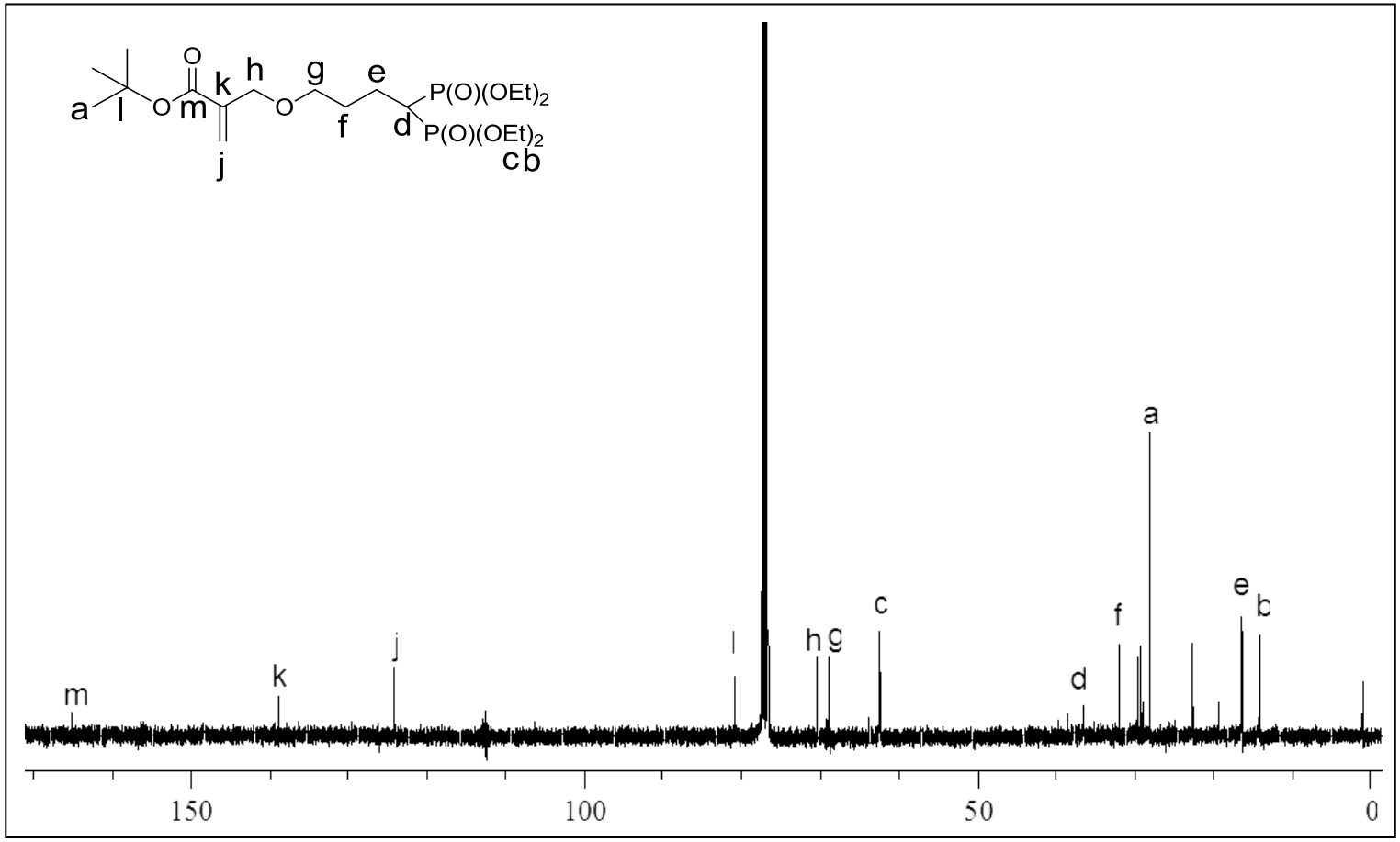


Figure 3.4. ^{13}C NMR spectrum of **A2**.

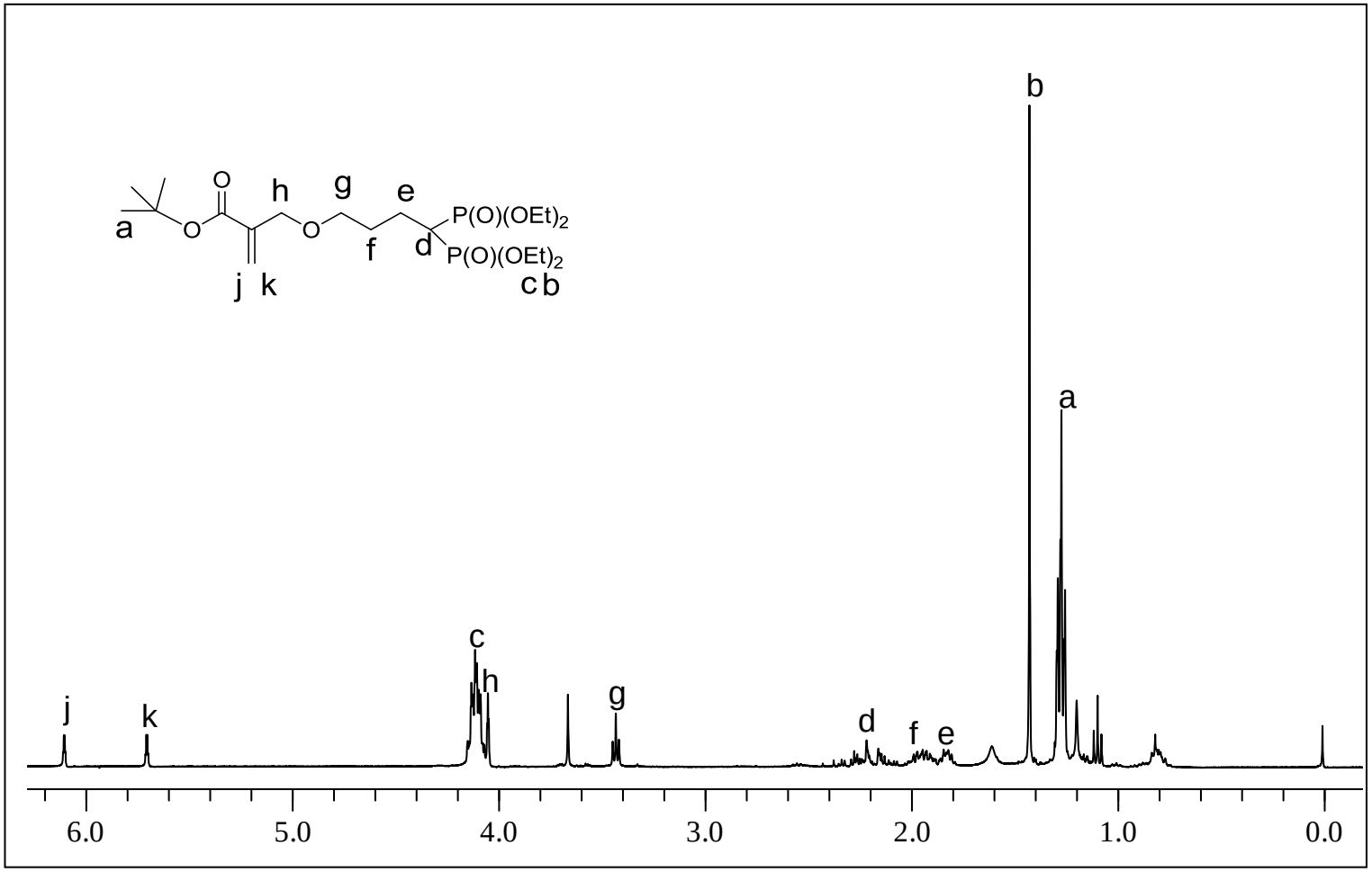


Figure 3.5. ^{1}H NMR spectrum of **A2**.

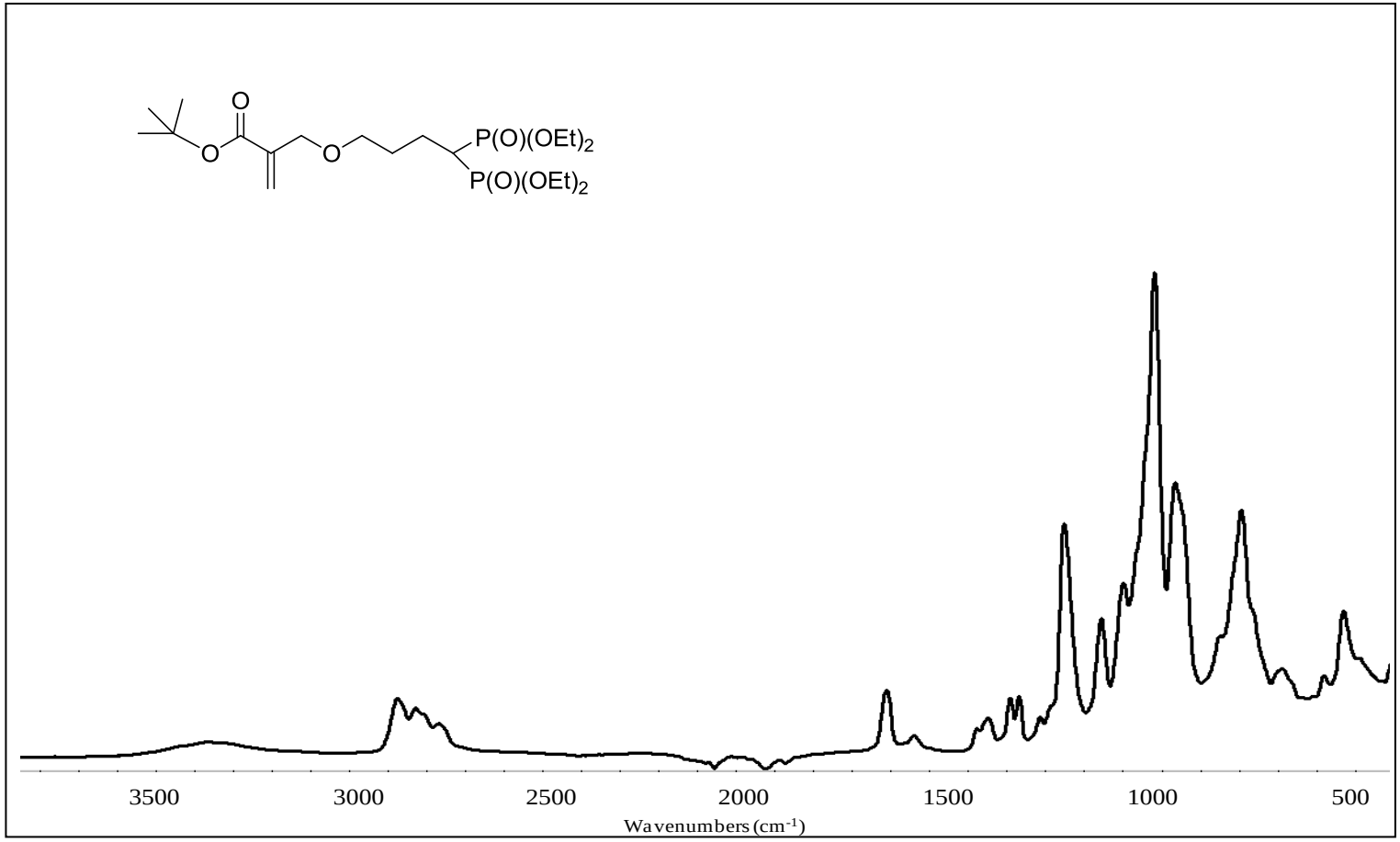


Figure 3.6. FTIR spectrum of **A2**.

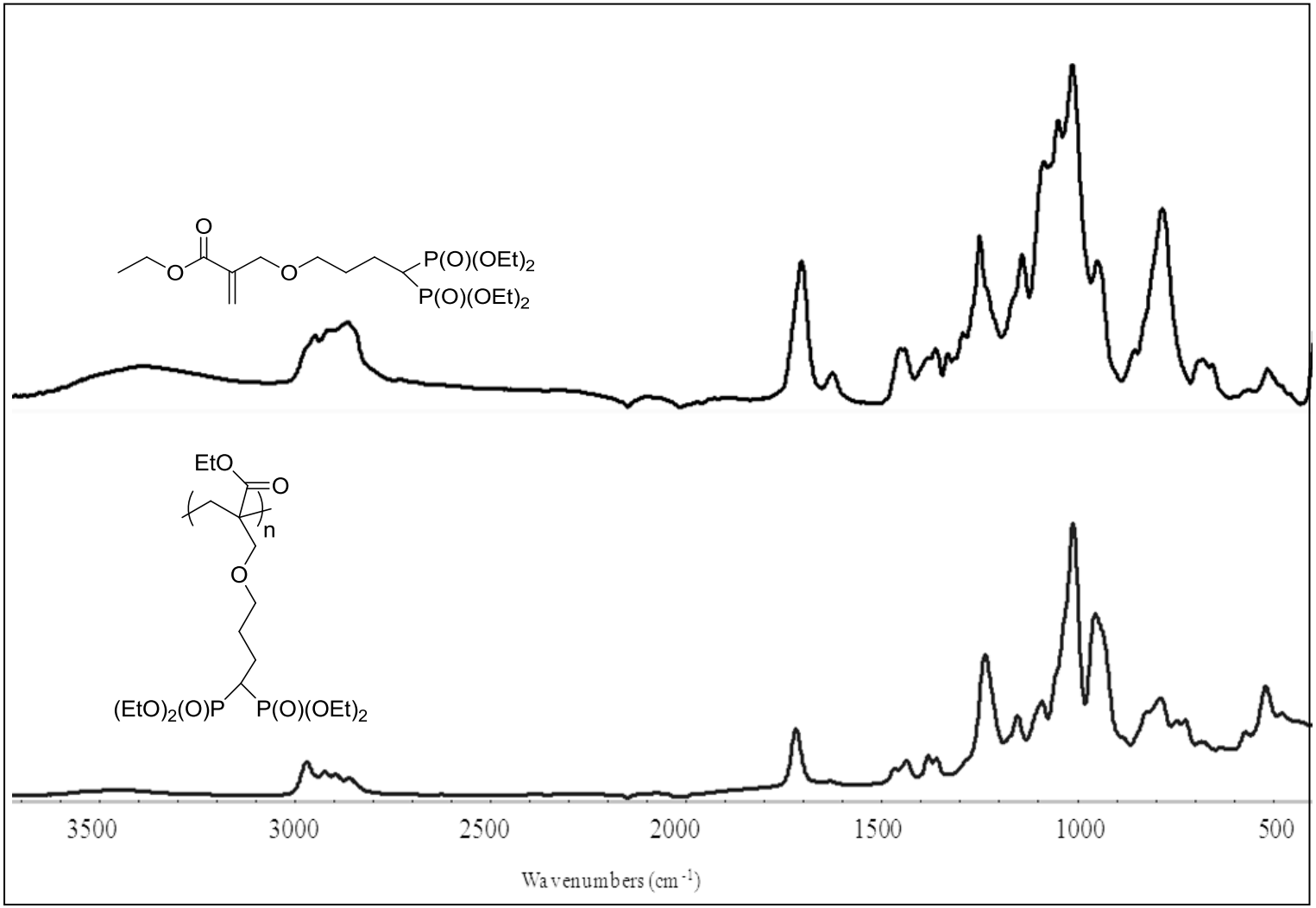


Figure 3.7. FTIR spectra of **A1** and poly-**A1**.

3.3.2. Homopolymerizations

The free radical (co)polymerization of phosphonate vinyl monomers were investigated thoroughly [159]. Phosphonate-containing (meth)acrylic monomers were found to be the more reactive ones although their polymerizabilities are mainly effected by chain transfer reactions. However, for bisphosphonates, radical homopolymerizability seems not to have been investigated at all. Reports exist only of a few syntheses [35-37, 47, 152] and copolymerizations with commercial dental [37, 47] and other monomers [152]; although they have a variety of potential medical, dental and orthodontic applications.

Thermal bulk polymerizations of monomers **A1** and **A2** at 65 °C using AIBN and standard freeze-evacuate-thaw procedures were successful and gave soluble polymers. The polymerization conditions and the characteristics of the polymers are listed in Table 3.2. The results show that the conversions reached ranged from 22 to 34% within 3.5-4.0 h. These low conversions may be due to chain transfer reactions from the labile hydrogen between two electron withdrawing phosphonate groups in the monomer structure.

The homopolymer of **A1** was obtained as white solid, whereas the polymer of **A2** was colorless oil after precipitation and drying. The polymers are soluble in weakly polar organic solvents such as acetone, methylene chloride, THF and also soluble in polar solvents such as methanol and water (Table 3.1). Figure 3.8 shows ^{1}H NMR spectrum of poly-**A1** with no trace of unreacted monomer. Also, the occurrence of polymerizations is confirmed from the presence of the C=O peak around 1728 cm^{-1} due to saturated ethyl or *tert*-butyl ester in the FTIR spectra. The molecular weights (M_n) were found to be 56000 and 25000 $gmol^{-1}$ for poly-**A1** and poly-**A2**, respectively, indicating effect of the bulky *tert*-butyl group.

Table 3.2. Thermal homo- and copolymerizations of bisphosphonate monomers with PEGMA.

Monomer[a]	Bisphosphonate monomer in feed (mol %)	Bisphosphonate monomer in copolymers[b] (mol%)	Solvent	AIBN (wt %)	Time (min)	Yield (%)	M_n	PDI
A1	100	-	Bulk	1.5	240	34	56000	1.68
A1	50	24	MeOH	3	24h	65	16000	1.33
A1	30	-	MeOH	3	9	Crosslinked	-	-
A1	10	-	MeOH	3	6	Crosslinked	-	-
A2	100	-	Bulk	1.5	200	22	25000	1.75
A2	50	27	Bulk	2	90	60	28400	1.66
A2	30	-	MeOH	3	60	Crosslinked	-	-
A2	10	-	MeOH	3	20	Crosslinked	-	-

[a] [M]= 2.2 mol L^{-1}.

[b] Determined from ^{1}H NMR spectra.

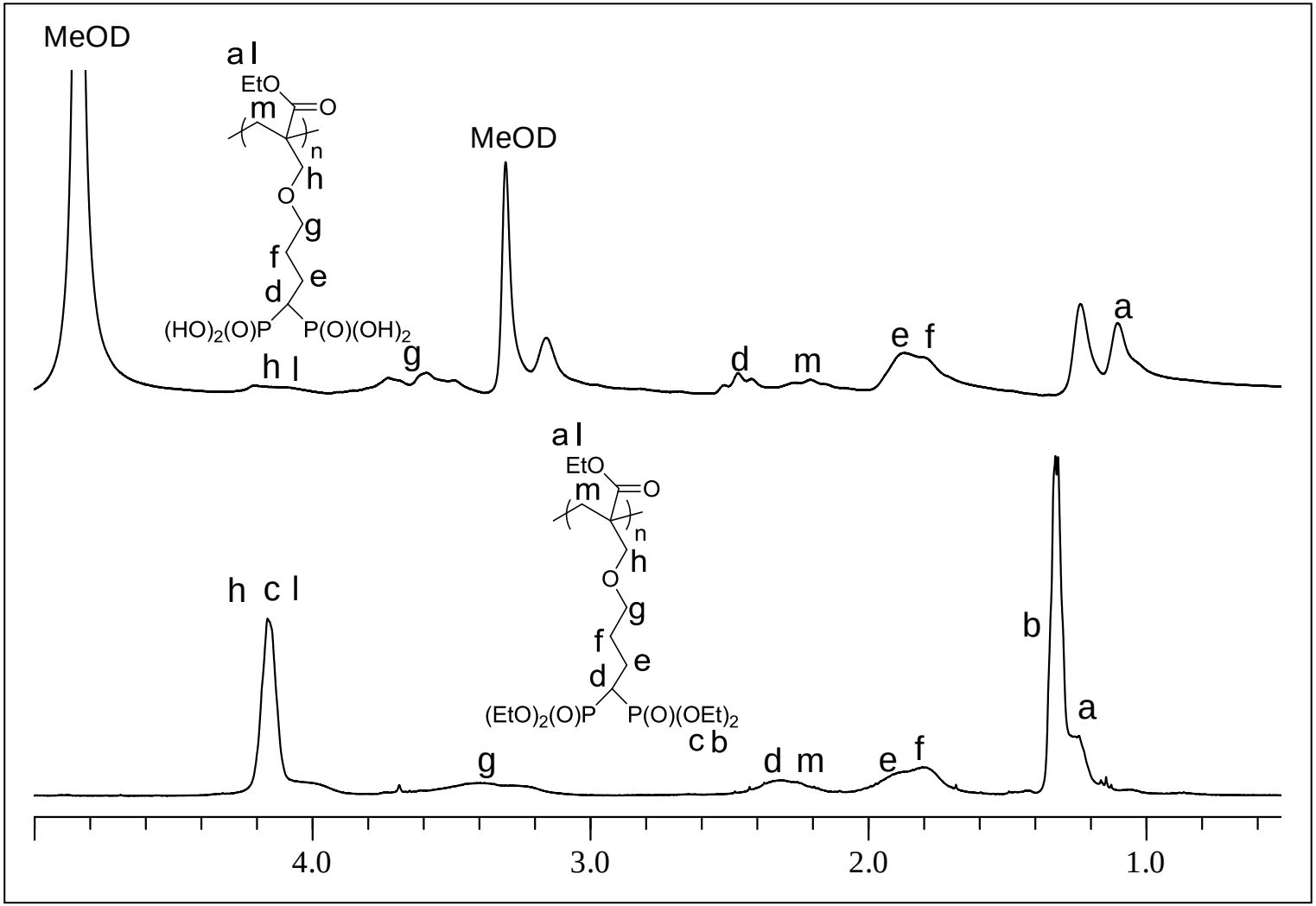


Figure 3.8. ^{1}H NMR spectra of poly-**A1** and hydrolyzed poly-**A1**.

The glass transition temperatures (T_g) of poly-**A1 (**15-20 °C) and poly-**A2 (**25-30 °C) are around room temperature. The flexible structures of these polymers due to longer methylene linkages decrease T_g of these polymers. The bulkiness of the *tert*-butyl groups seems to cause the T_g's of poly-**A2** to be higher than that of poly-**A1**.

To further evaluate the reactivities of these monomers in free-radical polymerizations, they were photopolymerized with 2 mol % DMPA using photo-DSC. All the polymerizations were performed under identical conditions of temperature, initiator concentration and light intensity. Figure 3.9 shows the time dependences of the polymerization rate and conversions for monomers. In earlier work, we observed that increasing the size of the ester group from ethyl to *tert*-butyl decreases the polymerization rate because of steric hindrance [160]. The behavior observed (Figure 3.9) is mostly compatible with this expectation such as **A1** has higher rate and conversion relative to **A2**. The conversions are 43 and 25 % for monomers **A1** and **A2**, respectively, and they are reached in about 6 min.

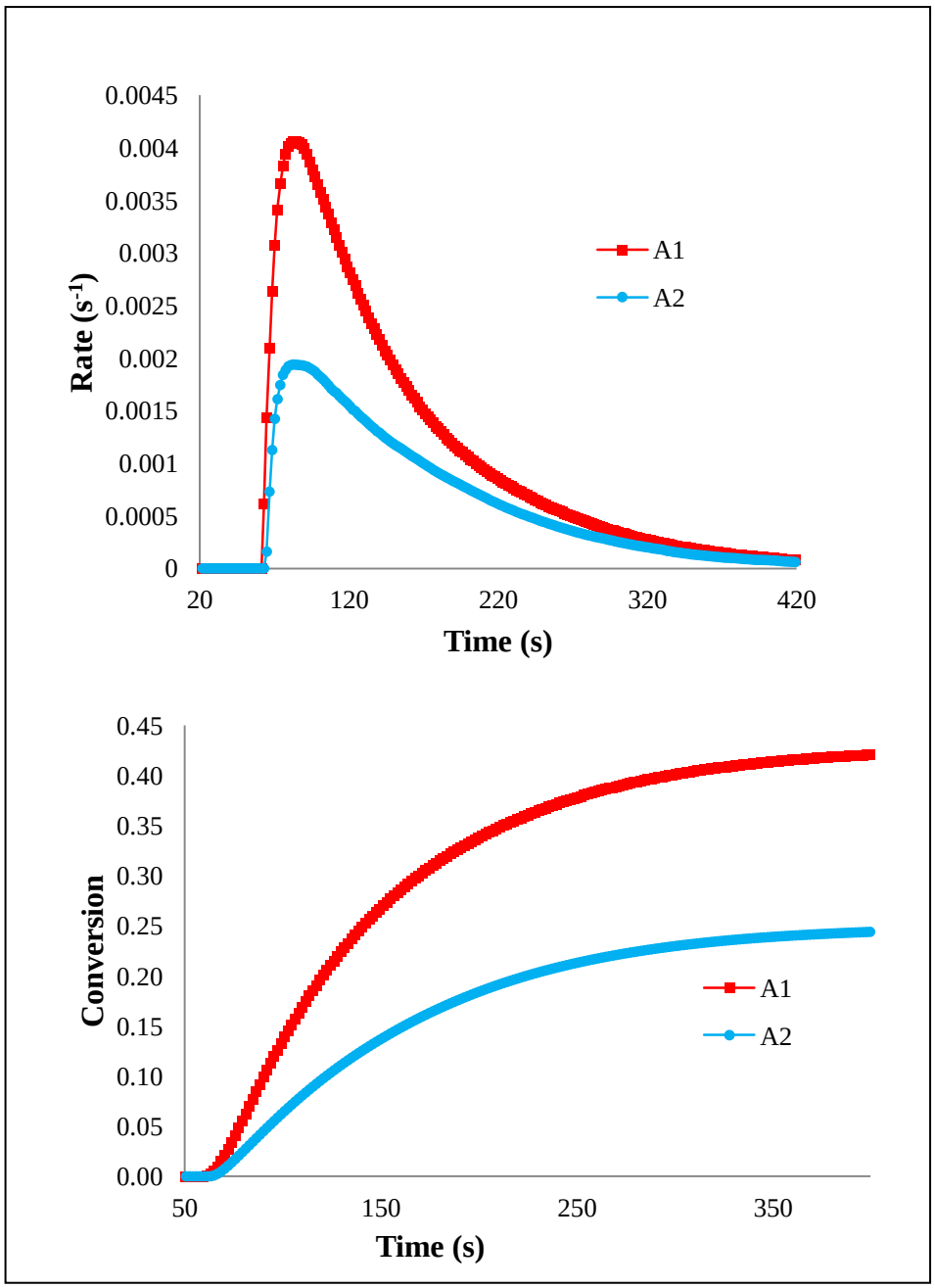


Figure 3.9. Rate-time and conversion-time curves for the polymerizations of **A1** and **A2**.

3.3.3. Copolymerizations

Water soluble copolymers of **A1** with PEGMA, a universal polymer used for drug delivery due to its biocompatible and nonimmunogenic nature, were prepared [161]. To get hints about the reactivity differences of PEGMA and the synthesized monomers, we first conducted photopolymerization of **A1** with PEGMA under the same conditions (Figure 3.10). It is obvious that PEGMA polymerizes much faster than **A1**, with its maximum rate of polymerization approximately four times higher than that of **A1.**

Furthermore, conversion reached for PEGMA was much higher than **A1**. Addition of 10 and 30 mol% of **A1** to PEGMA slightly decreased both its rate and conversion.

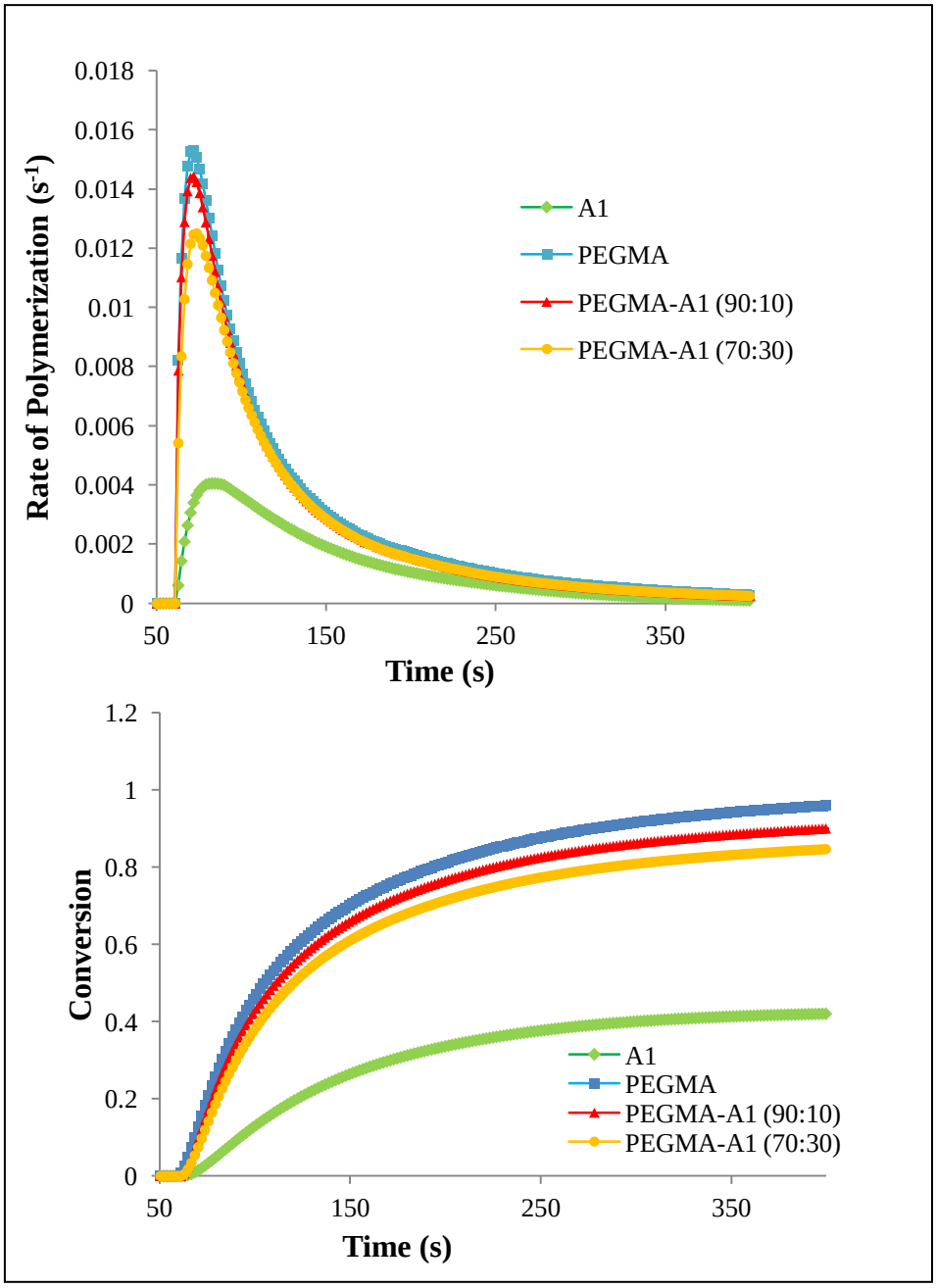


Figure 3.10. Rate-time and conversion-time curves in the copolymerizations of PEGMA with **A2** using DMPA at 40 ^{o}C.

Although several PEGMA derivatives with different molecular weights and end groups are available commercially, that with methoxy end-groups was used due to easy detection by ^{1}H NMR spectroscopy. The copolymerization of the synthesized monomers (**A1** and **A2**) with PEGMA (M_n =950) was performed at 65 °C using AIBN as initiator.

Bulk copolymerization studies mostly result in crosslinked polymers in a very short time, similar to homopolymerization of PEGMA. Therefore, solution polymerizations in methanol were carried out and the soluble polymers were purified by precipitation into cold ether, yielding white powders. The concentration of bisphosphonate monomer in the feed ratio was 10, 30 and 50 mol %. It is observed that as the concentration of PEGMA in feed ratio increases, the rate of polymerization and the crosslinking tendency of the copolymerization system increase (Table 3.2). During copolymerizations of **A1** with PEGMA soluble copolymer is obtained at 50:50 mol % feed ratios. This result is expected due to higher reactivity of PEGMA compared to bisphosphonate-containing monomers observed from photo-DSC experiments. To compare, PEGMA homopolymerization was carried out in methanol but at much lower monomer concentrations (0.6 mol L^{-1}) and a soluble polymer is obtained with M_n value of 235000. The lower molecular weight of the copolymers (16000-28400) is due to chain transfer ability of the synthesized monomers.

The copolymer compositions were determined from the integrated ^{1}H NMR spectra of the copolymers. Figure 3.11 shows the ^{1}H NMR spectrum of one of the copolymers (PEGMA:**A2**, 50:50 mol% in feed). The peaks at 3.38 and 3.60-3.90 ppm are due to the methoxy protons (-OCH_3) and methylene protons (-OCH_2CH_2-) of PEGMA. Methyl protons of PEGMA and **A2** and backbone protons overlap at 0.8-1.6 ppm. The peak at around 4.0 ppm is assigned to methylene protons of **A2** and PEGMA adjacent to oxygen. Integration of this peak relative to methoxy protons of PEGMA (3.38 ppm) showed PEGMA:**A2**, 63:27 mol %. The structures of the copolymers are also confirmed from the FTIR spectra. The DSC curves of PEGMA, and all of the copolymers obtained showed one melting endotherm which indicates the melting of PEG.

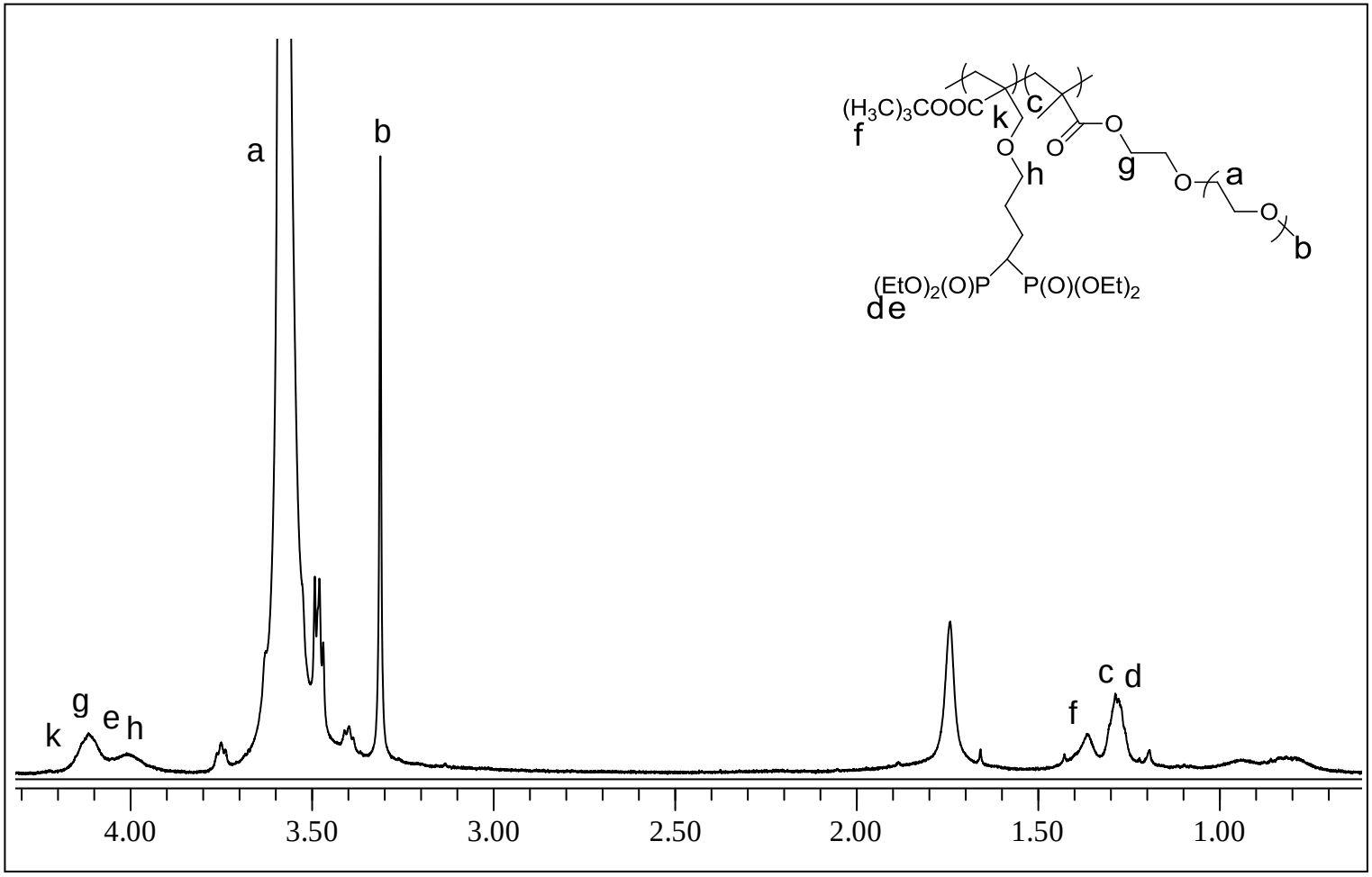


Figure 3.11. ^{1}H NMR spectra of PEGMA:**A2** (50:50 mol%) copolymer.

3.3.4. Hydrolysis of Polymers

The bisphosphonate groups of one of the homopolymers (poly-**A1**) were hydrolyzed using TMSBr. The integration of the ethyl peak of bisphosphonate ester at 1.22 ppm relative to that of carboxylic ester at 1.09 ppm indicates that approximately 80 % of the bisphosphonate groups are hydrolyzed in poly-**A1** (Figure 3.8). The FTIR spectrum of hydrolyzed poly-**A1** showed broad peaks in the region of 2600-3500 cm^{-1} due to OH stretching and peaks at around 1700 and 1210 cm^{-1} due to C=O and P=O stretchings. Also the bands at 990 and 910 cm^{-1} correspond to the symmetric and asymmetric vibration of P-O.

3.3.5. Acidity and Interactions with HAP

The pH value of aqueous solution of the hydrolyzed polymer **A1** (5 wt %) is found to be 1.39. To this solution 15 and 30 mg HAP, a model compound for bone, was added and the solution was stirred at 37 °C for 1 h. The FTIR spectra of the mixtures are quite different from that of the pure polymer: P-O (990 and 910 cm^{-1}) stretching vibrations present in the polymer show a sharp decrease in intensity, which is an indication of an interaction between the bisphosphonic acid functional groups in the polymer and the calcium ions of the HAP (Figure 3.12). The FTIR spectrum of HAP showed symmetric and antisymmetric P-O stretching modes between 1200 and 900 cm^{-1} and antisymmetric P-O bending modes in the 500-700 cm^{-1}. Polymer-HAP interaction is also observed on the FTIR spectrum of HAP particles after washing with ethanol, which shows an extra carbonyl peak in the 1600-1700 cm^{-1} region.

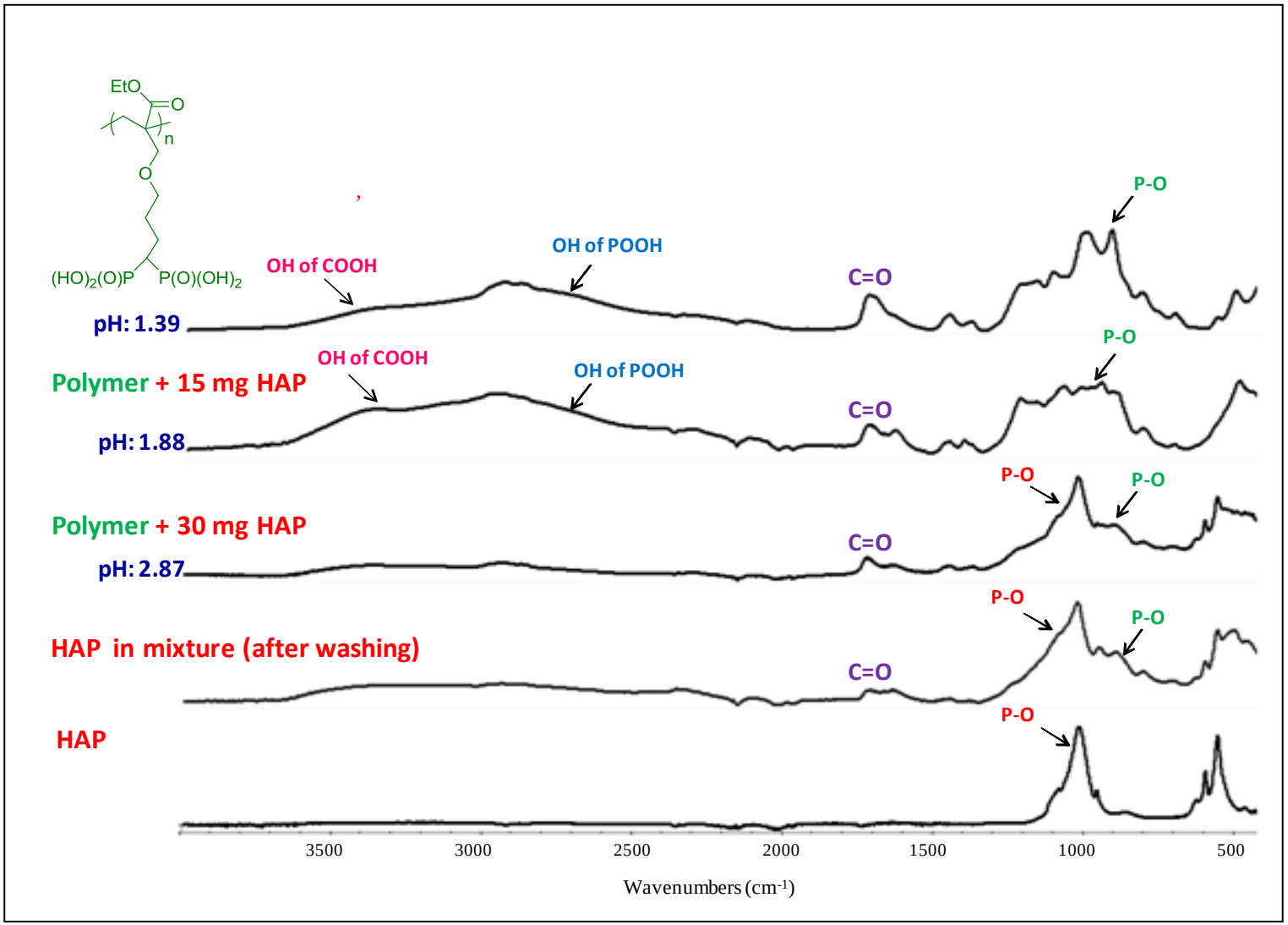


Figure 3.12. FTIR spectra of hydrolyzed poly-**A1**, hydrolyzed poly-**A1** with 15 mg and 30 mg HAP, HAP in mixture and HAP.

3.4. Conclusion

Two new alkyl α-hydroxymethacrylate derivatives carrying bisphosphonate moieties were successfully synthesized. These monomers have the bisphosphonate group attached to the double bond through an ether linkage. Radical homopolymerization and copolymerizations with PEGMA were carried out to obtain bisphosphonate-containing polymers. The results indicated (i) good homopolymerization tendencies (22-34 % in thermal and 25-43 % in photo-polymerizations), (ii) good copolymerization tendencies with PEGMA. Bisphosphonate groups of one of the homopolymers were hydrolyzed to give a new bisphosphonic acid-containing polymer. Interaction of the hydrolyzed polymer with HAP was confirmed, which makes these polymers good candidates to be used as biomaterials. These monomers can also generate a new class of self-etching dental adhesive monomers having both bisphosphonic and carboxylic acid functional groups after hydrolysis of the bisphosphonate and *tert*-butyl ester linkages. These monomers can be used to overcome the problem of hydrolysis in the aqueous environment which is the most common problem of self-etching adhesives.

4. NOVEL BISPHOSPHONATED-METHACRYLATES: SYNTHESIS, POLYMERIZATIONS AND INTERACTIONS WITH HYDROXYAPATITE

4.1. Introduction

The monomers functionalized with acid groups such as dihydrogen phosphates, carboxylic and phosphonic acids can be utilized in dental materials such as self-adhesive resin cements and self-etching adhesives [20, 42, 59, 162]. The currently used examples of acid monomers are 4-methacryloxyethyl trimellitic anhydride (4-META), 2-methacryloxyethyl phenyl hydrogen phosphate (Phenyl-P), 10-methacryloxydecyl dihydrogen phosphate (MDP) and 2,4,6-trimethylphenyl 2-[4-(dihydroxyphosphoryl)-2-oxabutyl]acrylate (MAEPA)]. Some of the properties desired for adhesive monomers are (i) high rate of free-radical homopolymerization or copolymerization with the other monomers in the adhesive, (ii) ability to form strong bonds with tooth tissue, (iii) sufficient stability both in storage and in the mouth. The first requirement can be fulfilled using variously designed methacrylates and methacrylamides with sufficient reactivity. The adhesion of monomers on tooth tissue are mainly due to chemical bonds, such as covalent or ionic bonds, resulting from reaction of acidic and chelating groups with HAP and physical bonds due to Van der Waals forces, London dispersion forces, hydrogen bonding or charge transfer complexes [57]. According to the 'Adhesion-Decalcification' concept of Yoshida *et al.*, the bonding performance of the adhesives depends on the chemical stability of the monomer-Ca salts formed with the interaction of the acid monomer and HAP [34, 62, 67, 69]. Therefore structure of the acid monomer is very important and small differences such as polarity in the monomer structures cause significant differences in their adhesive performances. For example, while 4-META and Phenyl-P deposit unstable calcium salts due to dissolution, MDP forms hydrolysis resistant calcium salts due to its hydrophobic decyl group. Therefore, extensive research has been conducted to develop new monomers with acid functional groups which may strongly bond to HAP. For example, bisphosphonates, structural analogues of naturally existing pyrophosphate with increased chemical and enzymatic stability and strong affinity for HAP, can be

incorporated into monomers [37, 61, 163]. Although many dental adhesives accomplish good binding to HAP, the main problem is hydrolysis of the ester groups in (meth)acrylates which these monomers are based on. Adhesive mixtures contain water and the mouth is an aqueous environment so ester based adhesives generally can hydrolyze both in storage and in the mouth which results in both inadequate shelf life of adhesives and undesirably short useful lifetime of the dental material. To overcome this disadvantage, hydrolytically stable ether and/or amide linked monomers were synthesized [20, 46-51, 164, 165].

In this study, five novel methacrylates were synthesized three of which are possible dental monomers with improved hydrolytic stability and interaction with HAP. These monomers are derivatives of alkyl α-hydroxymethyl acrylates (RHMAs) with bisphosphonic and carboxylic acid functional groups so they have high potential to binding to HAP. Also these monomers are expected to have improved shelf life and bonding reliability, as they will not hydrolyze in aqueous acidic conditions due to the presence of ether linkage in their structures.

4.2. Experimental

4.2.1. Materials and Apparatus

4.2.1.1. Materials. Ethyl α-bromomethacrylate (EBBr), *tert*-butyl α-bromomethacrylate (TBBr) and *gem*- hydroxy-diphosphonates (tetraethyl 1-hydroxyethane-1,1-diyldiphosphonate and tetraethyl hydroxy(phenyl)methylenediphosphonate) were synthesized according to the literature procedures [153-155, 166]. The following analytical-grade chemicals were obtained from commercial sources and used without further purification: Triethyl amine (TEA), triethyl phosphite, dibutyl amine, diethyl phospite, 2-hydroxyethyl methacrylate (HEMA), glycerol dimethacrylate (GDMA), HAP, 2,2'-azobis(2-methylpropionamidine)dihydrochloride (V-50), 2,2'-dimethoxy-2-phenyl acetophenone (DMPA) and bis(2,4,6-trimethylbenzoyl)phenylphosphine oxide (BAPO) were obtained from Aldrich; sodium sulphate, TFA from Merck. TMSBr (Aldrich) was freshly distilled before use and dichloromethane (CH_2Cl_2; Merck) and tetrahydrofuran

(THF; Merck) were dried over molecular sieves. Dentin samples were obtained from Marmara University Department of Prosthodontics.

4.2.1.2. Apparatus. Monomer characterization involved ^{1}H, ^{13}C and ^{31}P NMR spectroscopy (Varian Gemini, 400 MHz) and FTIR spectroscopy (T 380). The photopolymerizations were carried out using a TA Instruments Q100 differential photocalorimeter. The interactions of monomers with HAP or dentin were studied with Scanning Electron Microscopy ESEM-FEG (FEI-Philips XL30) and X-Ray Diffractometer (Rigaku D/max-2200/PC). Gel permeation chromatography (GPC) was performed on a Agilent 1100 GPC Instrument equipped with a refractive index detector, using water as solvent and polyacrylic acid sodium salt standards.

4.2.2. Synthesis of Monomers

4.2.2.1. Ethyl 2-((1,1-bis(diethoxyphosphoryl)ethoxy)methyl)acrylate (**B1**). To a mixture of tetraethyl 1-hydroxyethane-1,1-diyldiphosphonate (0.5 g, 1.57 mmol) and triethylamine (0.317 g, 3.14 mmol) in THF (3 mL) in an ice bath, EBBr (0.454 g, 2.36 mmol) was added dropwise under N_2. After stirring at 60 °C for 24 h, the solvent was removed under vacuum. CH_2Cl_2 (5 mL) was added and the solution was extracted with water (3x3 mL). The organic phases were dried with anhydrous Na_2SO_4, filtered and the solvent was evaporated. The crude product was purified by column chromatography using hexane initially and gradually changing to ethyl acetate as eluent. The pure product was obtained as colourless oil in 54 % yield.

^{1}H NMR ($CDCl_3$), δ =1.24 (t, $^{3}J_{HH}$= 7.2 Hz, 3H, $COCH_2CH_3$), 1.28 (td, $^{3}J_{HH}$= 7.1 Hz, $^{4}J_{HP}$= 2.9 Hz, 12H, $POCH_2CH_3$), 1.70 (t, $^{3}J_{HP}$= 16.3 Hz, 3H, CH_3), 4.17 (m, 10H, OCH_2CH_3), 4.52 (s, 2H, CCH_2O) 5.92, 6.23 ppm (s, 2H, $C=CH_2$).

^{13}C NMR ($CDCl_3$), δ = 14.25 (CH_3CH_2OC), 16.22 (CH_3CH_2OP), 16.58 (CH_3C), 60.75 (CH_3CH_2OC), 63.44 (CH_3CH_2OP), 63.88 (CCH_2O), 77.13 (t, $OC[P(O)(OEt)_2]_2$), 125.62 ($CH_2=C$), 137.58($CH_2=C$), 165.66 ppm (C=O).

^{31}P NMR ($CDCl_3$), δ = 18.82 ppm.

FTIR (ATR): 2983 (C-H), 1717 (C=O), 1638 (C=C), 1247 (P=O), 1013 and 966 cm^{-1}(P-O-Et).

4.2.2.2. *Tert*-butyl 2-((1,1-bis(diethoxyphosphoryl)ethoxy)methyl)acrylate (**B2**). Monomer **B2** was synthesized with the same procedure for monomer **B1** using TBBr instead of EBBr. The pure product was obtained as colourless oil in 52 % yield.
^{1}H NMR ($CDCl_3$), δ = 1.30 (td, $^{3}J_{HH}$= 7.1 Hz, $^{4}J_{HP}$= 2.9 Hz, 12H, CH_2CH_3), 1.45 (s, 9H, C-CH_3), 1.72 (t, $^{3}J_{HP}$= 16.3 Hz, 3H, CH_3), 4.19 (m, 8H, OCH_2CH_3), 4.57 (s, 2H, CCH_2O), 5.87, 6.17 ppm (s, 2H, $C=CH_2$).

^{13}C NMR ($CDCl_3$), δ = 16.43 (CH_3CH_2O), 16.66 (CH_3C), 27.96 [$(CH_3)_3C$], 63.31 (CH_3CH_2O), 63.72 (CCH_2O), 77.31 (t, $OC[P(O)(OEt)_2]_2$), 80.87 [$(CH_3)_3C$], 124.66 ($CH_2=C$), 138.62 ($CH_2=C$), 164.74 ppm (C=O).
^{31}P NMR ($CDCl_3$), δ = 18.87 ppm.

FTIR (ATR): 2979 (C-H), 1713 (C=O), 1637 (C=C), 1252 (P=O), 1016 and 968 cm^{-1} (P-O-Et).

4.2.2.3. (1-((2-(ethoxycarbonyl)allyl)oxy)ethane-1,1-diyl)diphosphonic acid (**B3**). TMSBr (0.455 g, 2.97 mmol) was added dropwise to a solution in of monomer **B1** (0.256 g, 0.59 mmol) in 0.2 mL of dry CH_2Cl_2 in an ice bath under N_2. After stirring for 100 min at 40 °C, the volatile components were removed under vacuum. Methanol (3 mL) was added and the mixture was stirred at room temperature overnight. After removal of the solvent, the orange-yellow residue was purified by reversed-phase flash chromatography on C18, eluting with water followed by lyophilisation of the eluted solutions. The pure product was obtained in 34 % yield as a waxy solid.

^{1}H NMR (MeOD), δ = 1.10 (t, $^{3}J_{HH}$=7.1 Hz, 3H, CH_2CH_3), 1.55 (t, $^{3}J_{HP}$=15.8 Hz, 3H, CH_3), 4.03 (q, $^{3}J_{HH}$= 7.1 Hz, 2H, OCH_2CH_3), 4.37 (s, 2H, CCH_2O), 5.90, 6.08 (s, 2H, $C=CH_2$) ppm.

^{13}C NMR (MeOD), δ = 14.51 (CH_3CH_2O), 17.07 (CH_3C), 61.94 (CH_3CH_2O), 62.30 (CCH_2O), 75.25 (t, $OC[P(O)(OEt)_2]_2$), 126.76 ($CH_2=C$), 139.28 ($CH_2=C$), 167.51 (C=O).

^{31}P NMR (MeOD), δ = 18.67 ppm.

FTIR (ATR): 2500-3500 (O-H), 2984 (C-H), 1689 (C=O), 1629 (C=C), 1140 (P=O), 915 cm^{-1} (P-O).

4.2.2.4. 2-((1,1-bis(diethoxyphosphoryl)ethoxy)methyl)acrylic acid (**B4**). CF_3COOH (0.262 g, 2.3 mmol) was added to monomer **B2** (0.3 g, 0.65 mmol) dropwise in an ice-bath under nitrogen. The solution was stirred at room temperature for 24 hours. After removal of excess CF_3COOH, the crude product was obtained as a light yellow viscous liquid. The crude product was purified by reversed-phase flash chromatography on C18, eluting with water:acetonitrile (60:40) to give monomer **B4** as colourless viscous liquid in 38 % yield.

^{1}H NMR (MeOD), δ = 1.18 (t, $^3J_{HH}$= 7.1 Hz, $^4J_{HP}$= 2.7 Hz, 12H, CH_2CH_3), 1.59 (t, $^3J_{HP}$= 16.6, 3H, CH_3CP), 4.06 (m, 8H, OCH_2CH_3), 4.35 (s, 2H, CCH_2O), 5.79, 6.12 ppm (s, 2H, $C=CH_2$).

^{13}C NMR (MeOD), δ = 16.66 (CH_3C), 17.11 (CH_3CH_2O), 65.45 (CH_3CH_2O), 69.92, 71.74 (d, CCH_2O),78.67 (t, $OC[P(O)(OEt)_2]_2$), 126.85 ($CH_2=C$), 139.45 ($CH_2=C$), 168.76 ppm (C=O).

^{31}P NMR (MeOD), δ = 18.43 ppm.

FTIR (ATR): 2500-3500 (O-H), 2986 (C-H), 1711 (C=O), 1638 (C=C), 1224 (P=O), 1162 and 950 cm^{-1} (P-O-Et).

4.2.2.5. 2-((1,1-diphosphonoethoxy)methyl)acrylic acid (**B5**). TMSBr (1.198 g, 7.8 mmol) was added dropwise to a solution of monomer **B2** (0.718 g, 1.6 mmol) in 0.5 mL of CH_2Cl_2 in an ice bath under N_2. After stirring for 100 min at 40 °C, the volatile components were removed under vacuum. Methanol (5 mL) was added and the mixture was stirred at room temperature overnight. After removal of the solvent, CF_3COOH (0.76 g, 0.0068 mmol) was added dropwise in an ice-bath under N_2. The solution was stirred at room temperature overnight. After removal of excess CF_3COOH, the residue was purified by reversed-phase flash chromatography on C18, eluting with water followed by

lyophilisation of the eluted solutions to give monomer **B5** as a white solid in 20 % yield. It decomposed at around 130 °C before melting.

^{1}H NMR (MeOD), δ = 1.61 (s, 3H, CH_3CP), 4.42 (s, 2H, CCH_2O), 5.97, 6.17 (s, 2H, $C=CH_2$) ppm.

^{13}C NMR (MeOD), δ = 17.21 (CH_3C), 65.31 (CCH_2O), 81.35 (t, $OC[P(O)(OH)_2]_2$), 127.32 ($CH_2=C$), 139.40 ($CH_2=C$), 169.84 ppm (C=O).

^{31}P NMR (MeOD), δ = 18.67 ppm.

FTIR (ATR): 2500-3500 (O-H), 2926 (C-H), 1689 (C=O), 1628 (C=C), 1140 (P=O), 910 cm^{-1} (P-O).

4.2.3. Interactions of Monomers with Hydroxyapatite or Dentin

4.2.3.1. FTIR technique. HAP particles (0.2 g) were dispersed in 1.00 g of monomer/EtOH/H_2O (15/45/40 wt%) solution under stirring as described by Yoshihara *et al.* [69]. After 24 hrs, the HAP particles were separated by centrifugation and washed three times with water and then with ethanol and dried.

4.2.3.2. NMR technique. To measure the binding ability of monomers with HAP the method given by Nishiyama *et al.* was used [167]. The acidic monomer (0.27 mmol) was dissolved in 1.25 g of 20 wt % D_2O/H_2O solution. The pH value of the solution was measured and ^{13}C NMR spectrum was obtained. Deuteriated DMSO was used as an external standard. HAP (15 and 30 mg) was then added to this solution. After the suspension was stirred at 37 °C for 1 h, the pH values were measured and the ^{13}C NMR spectra were obtained. The chemical shift values of the α-methylene carbon attached to the phosphonic acid and carbonyl carbon assigned to carboxylic acid were determined. For monomer **B3** ^{31}P NMR shifts were also investigated before and after HAP addition.

4.2.3.3. XRD technique. The crystal phases on the monomer-coated HAP particles were identified by means of a powder XRD operated under 40 kV acceleration and 40 mA

current and scanning rate of 2°/min for 2θ/θ-scan. HAP sample 'Monomer-HAP-24h', was prepared as in the case of FTIR technique and washed with ethanol (x3) and water (x3) and dried at room temperature before analysis.

4.2.3.4. ESEM technique. Monomer 3/EtOH/H_2O (15/45/40 wt%) solutions were also applied to dentin disks of approximately 1.5 mm thickness by rubbing the surface with a Microbrush (Bisco Brush, Bisco Inc, Schaumburg, IL, USA). After 20 sec, the samples were strongly air dried and rinsed with ethanol (3-Dentin-EtOH).

4.2.4. Hydrolytic Stability

The hydrolytic stabilities of monomers **B3** and **B5** were studied by ^{1}H NMR measurements of 5 wt % solutions of the monomers in methanol-d_4/D_2O after storage at 37 °C for 1 month.

4.2.5. Polymerizations

4.2.5.1. Thermal polymerizations. Thermal homopolymerization of monomer **B3** was carried out in water ([M]= 0.398 mol L^{-1}) with V-50 ([M]= 0.024 mol L^{-1}) at 70 °C using the standard freeze-evacuate-thaw procedure. The polymer was purified by precipitation into isopropyl alcohol twice, filtered and dried under vacuum (40% yield). The attempted polymerizations of monomers **B4** and **B5** under similar conditions were not successful.

4.2.5.2. Photopolymerizations. The photopolymerizations were carried out using a DSC instrument equipped with a mercury arc lamp. Samples (3-4 mg) containing 2.0 mol% initiator were irradiated for 10 min at 40 °C with an incident light intensity of 20 mW/cm^2 under nitrogen flow. The heat flux as a function of reaction time was monitored under isothermal conditions, and both the rate of polymerization (R_p) and degree of conversion were calculated as a function of time. The theoretical value used for the heat of reaction (ΔH_p) was 13.1 kcal/mol for methacrylate double bonds [157, 158]. Rates of polymerization were calculated according to the following formula:

$$Rate = \frac{\left(Q/_s\right)M}{n(\Delta Hp)m} \qquad (4.1)$$

where *Q/s* is heat flow per second, *M* the molar mass of the monomer, *n* the number of double bonds per monomer molecule, ΔH_p the heat released per mole of double bonds reacted and *m* the mass of monomer in the sample.

4.3. Results and Discussion

4.3.1. Synthesis of Monomers

Two different *gem*-hydroxy-diphosphonates GDH1 and GDH2 (tetraethyl 1-hydroxyethane-1,1-diyldiphosphonate and tetraethyl hydroxy(phenyl)methylene diphosphonate) were prepared in two steps according to a literature procedure (Figure 4.1) [166]. In the first step, acetyl or benzoyl chloride was subjected to the Arbuzov reaction with triethyl phosphite to give diethyl acylphosphonates. The acylphosphonates were purified by vacuum distillation and identified by FTIR. In the second step, the diethyl acylphosphonates were converted to the corresponding *gem*-hydroxy-diphosphonates by treatment with diethyl phosphite in the presence of catalytic amount of base (n-dibutylamine- 5 mol %). It is known that, *gem*-phosphonate-phosphates are also produced in this reaction by rearrangement, especially at higher base concentration (Figure 4.1) [168,169]. These unwanted isomers are soluble in hexane, consistent with the less polar structure of the *gem*-phosphonate-phosphates. They were completely removed after

several washings and the pure products (GDH1 and GDH2) were obtained as colorless solids around 50 % yields. The GDH2 was found to show stronger tendency to rearrangement compared to GDH1.

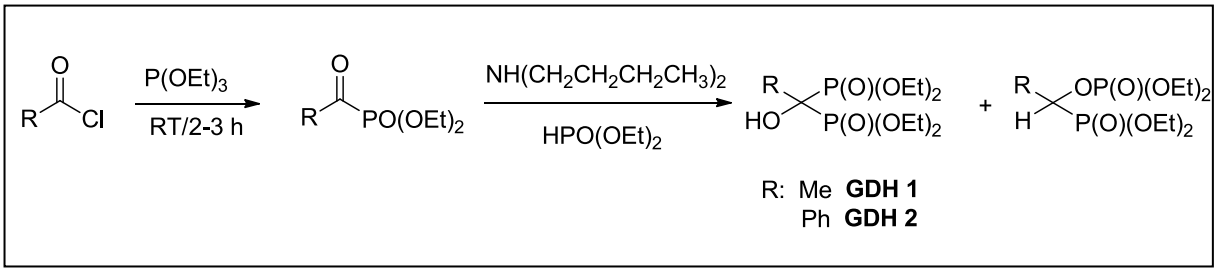


Figure 4.1. Synthesis of gem-hydroxy-diphosphonates.

The synthesized *gem*-hydroxy-diphosphonates (GDH1 and GDH2) were reacted with EBBr and TBBr in the presence of TEA catalyst at 60 °C. The reaction of tetraethyl 1-hydroxyethane-1,1-diyldiphosphonate (GDH1) with EBBr and TBBr was successful and gave two novel bisphosphonate-containing methacrylates (**B1** and **B2**) (Figure 4.2). The monomers **B1** and **B2** were obtained as colorless liquids with the yields of 54 and 52 % after column chromatography. These monomers are soluble in common organic solvents such as acetone, THF, methylene chloride and methanol but insoluble in water (Table 4.1).

Table 4.1. Solubility of the synthesized monomers in selected solvents.

Monomer	**H_2O**	**MeOH**	**Acetone**	**CH_2Cl_2**	**THF**	**Hexane**
B1	-	+	+	+	+	-
B2	-	+	+	+	+	+
B3	+	+	+	-	+	-
B4	+	+	+	-	+	-
B5	+	+	-	-	+	-

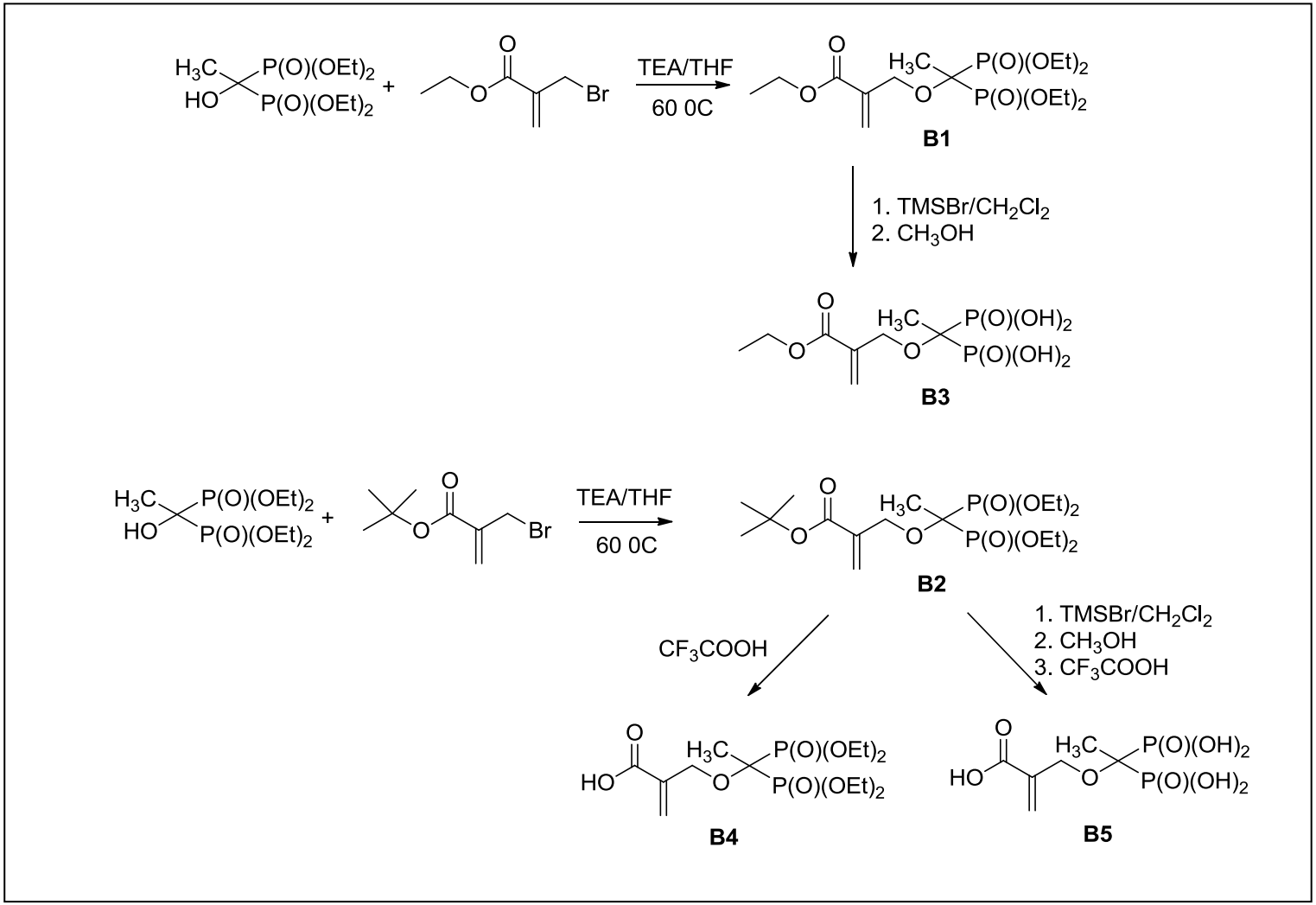


Figure 4.2. Synthesis of bisphosphonate and bisphosphonic acid-containing methacrylates.

The expected structures of monomers **B1** and **B2** were confirmed by ^{1}H, ^{13}C NMR, ^{31}P NMR and FTIR spectroscopies. Examination of the ^{1}H NMR spectra of both monomers revealed the characteristic triplet for CH_3 attached to the bisphosphonate group due to splitting effect of phosphorous and gave a coupling constant: $^{3}J_{H(d)P}$= 16.3 Hz. Moreover, CH_3 and CH_2 protons of the phosphonate esters gave triplets of doublet (*td*) ($^{3}J_{H(b)H(c)}$= 7.1 Hz, $^{4}J_{H(c)P}$= 2.9 Hz) and multiplet at around 4.19 ppm due to the same effect (Figure 4.3). In the ^{13}C NMR spectra, the triplet seen at around 77.31 ppm is due to the quaternary carbon attached to the phosphonate groups.

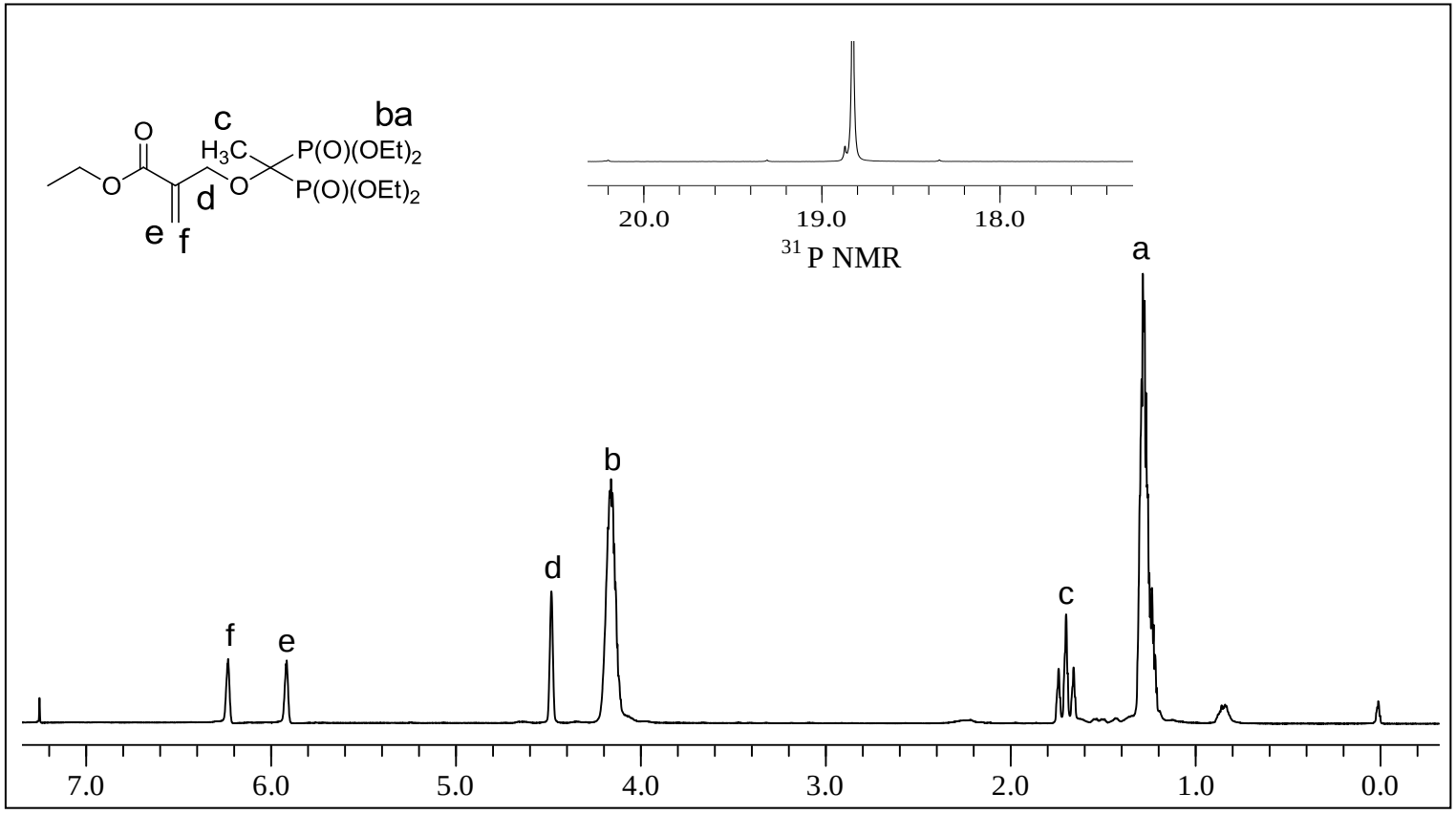


Figure 4.3. ^{1}H and ^{31}P NMR spectra of **B1**.

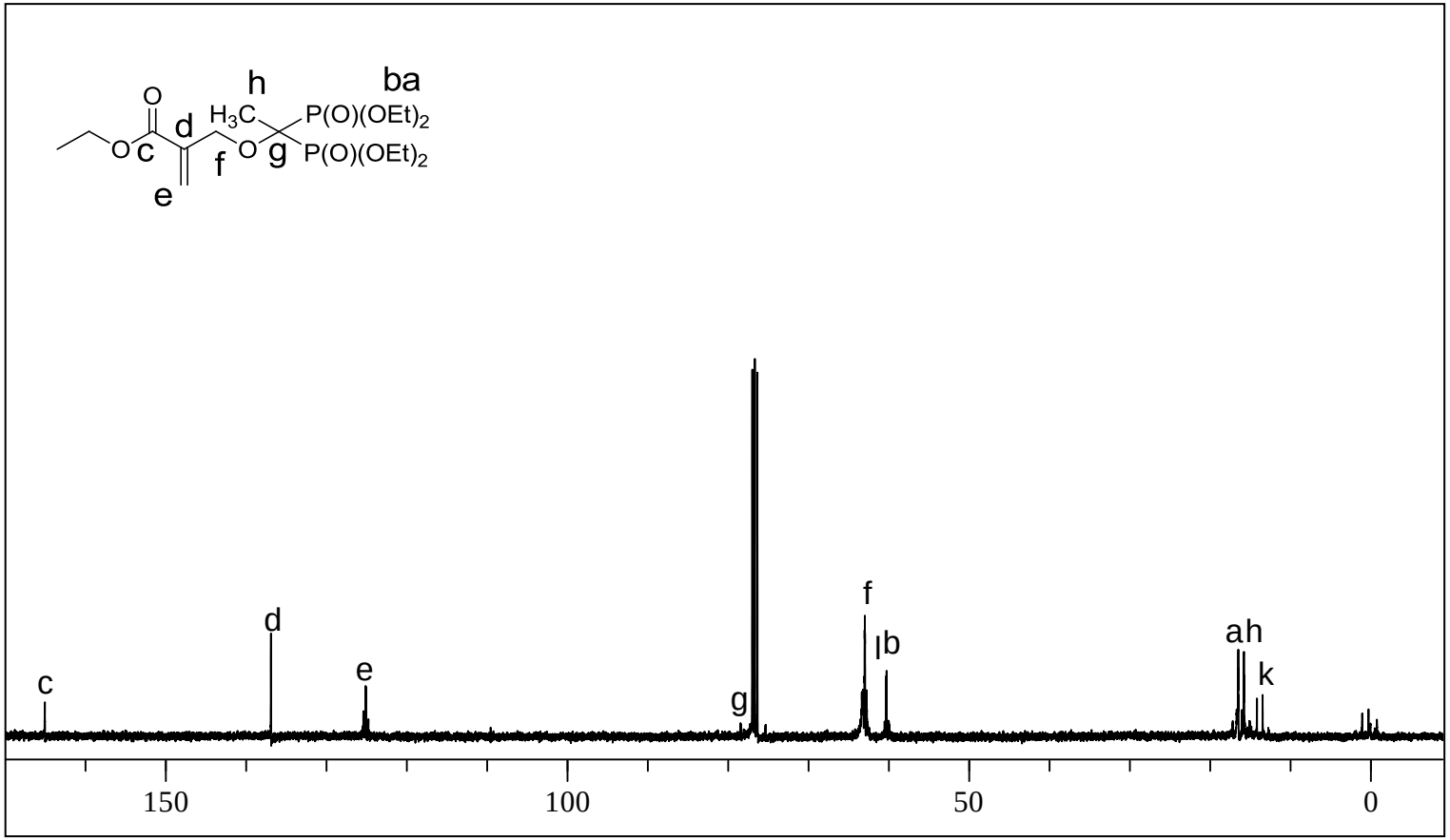


Figure 4.4. ^{13}C NMR spectrum of **B1**.

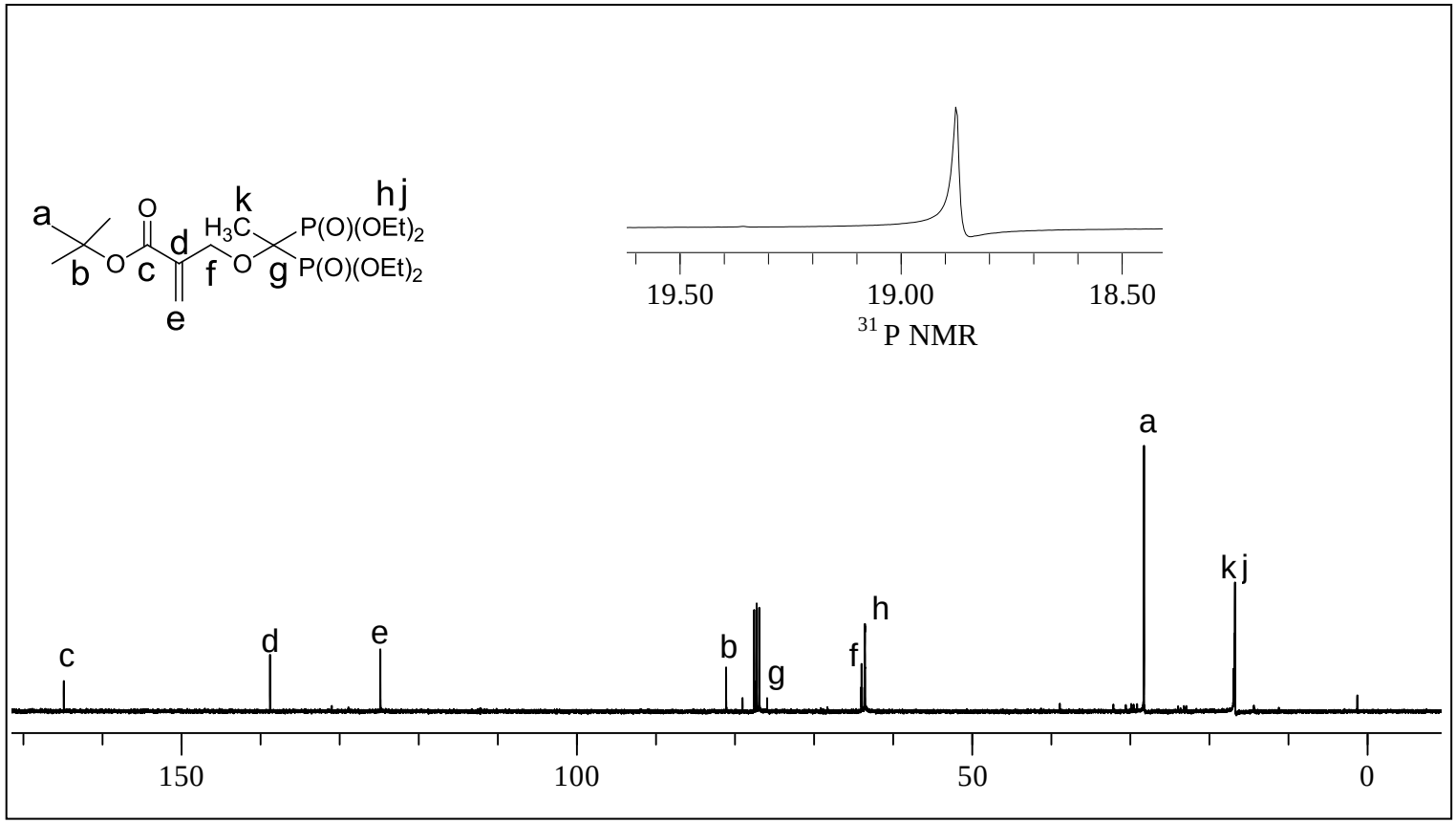


Figure 4.5. ^{13}C and ^{31}P NMR spectra of **B2**.

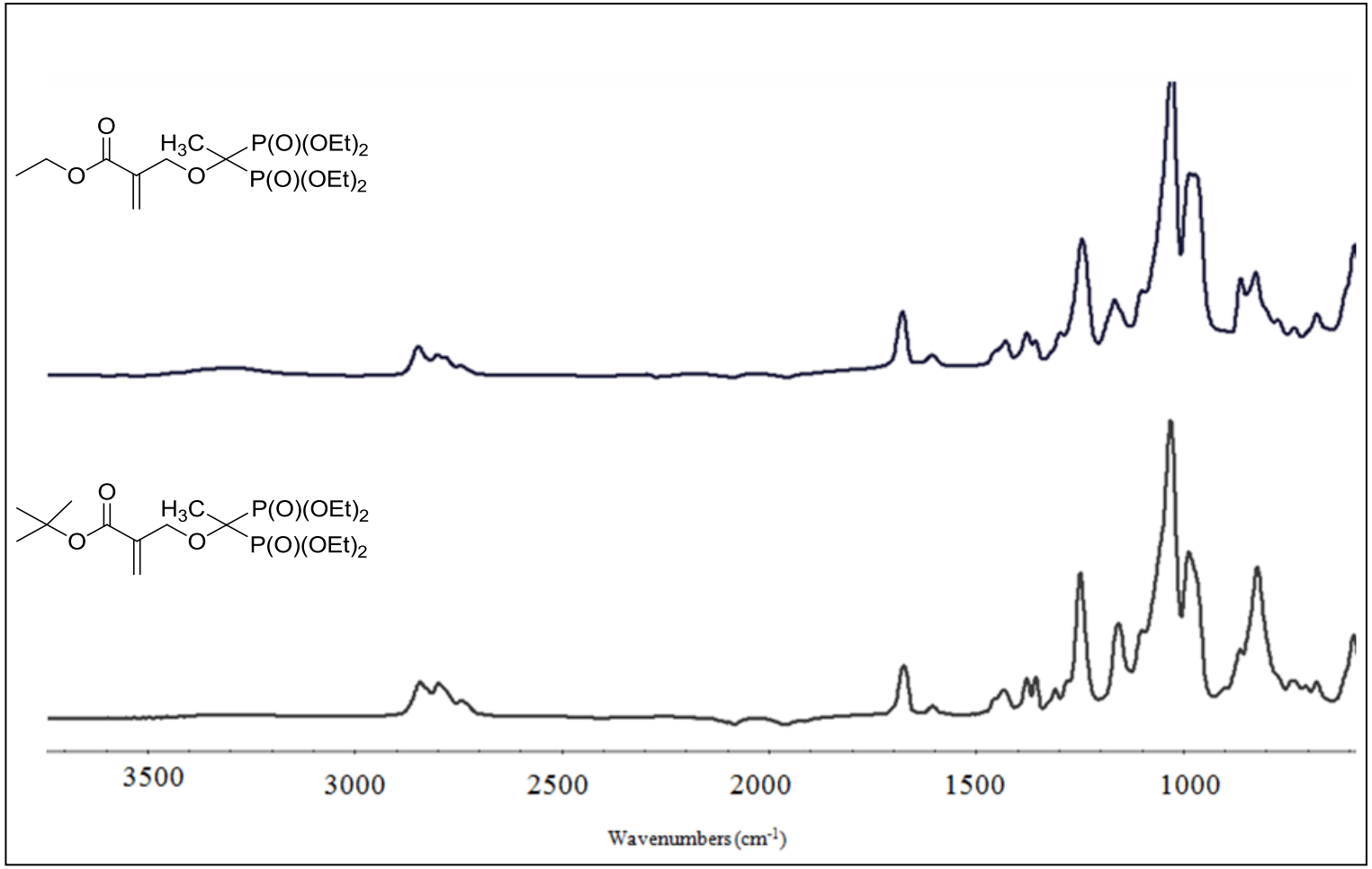


Figure 4.6. FTIR spectra of **B1** and **B2**.

GDH2 did not react with EBBr and TBBr in an analogous way, instead giving ethyl 2-((diethoxyphosphoryl)methyl)acrylate which is a phosphonated monomer synthesized by our group in an earlier work (Figure 4.7) [31]. This may be explained by rearrangement of GDH2 under reaction conditions followed by the reaction of the phosphate group of this isomer with EBBr. In the ^{1}H NMR spectrum, the absence of any phenyl proton proves formation of the predicted product.

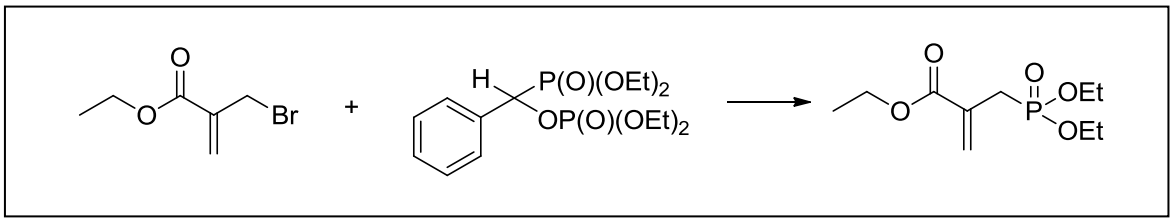


Figure 4.7. Formation of ethyl 2-((diethoxyphosphoryl)methyl)acrylate.

The phosphonate ester groups of monomer **B1** were hydrolyzed under mild conditions with TMSBr (Figure 4.2). The silylation of this monomer, followed by methanolysis, gave a bisphosphonic acid-containing monomer, **B3**, as a waxy solid in 34 % yield after purification with C18 reversed-phase flash chromatography. This monomer dissolves very well in water which is very important for dental applications. ^{1}H NMR spectrum of **B3** shows the complete disappearance of bisphosphonic ester peaks at 1.28 and 4.17 ppm (Figure 4.10). The *tert*-butyl groups of monomer **2** were hydrolysed using TFA to give monomer **B4**. The hydrolysis was confirmed by the disappearance of *tert*-butyl group at 1.45 ppm in the ^{1}H NMR spectrum of **B4** (Figure 4.8). The FTIR spectrum of this monomer shows broad peaks in the region of 3500-2500 cm^{-1} due to OH stretching and strong peaks at 1711, 1638, 1224, 1162 and 950 cm^{-1} due to C=O, C=C, P=O and P-O-Et stretchings.

Moreover, monomer **B2** was hydrolyzed with TMSBr and afterwards with TFA to obtain a new monomer **B5** with both bisphosphonic and carboxylic acid groups. It was purified by C18 reversed-phase flash chromatography using water and obtained as white solid in 20 % yield. The structure of this monomer was confirmed by the absence of ethyl and *tert*-butyl protons in its ^{1}H NMR and ^{13}C NMR spectra (Figure 4.8). This monomer was found to decompose around 120-130 °C and easily picks up water from the air due to

its hygroscopic structure. FTIR spectrum indicates disappearance of P-O-Et peak at 1016 and 968 cm^{-1} (Figure 4.11).

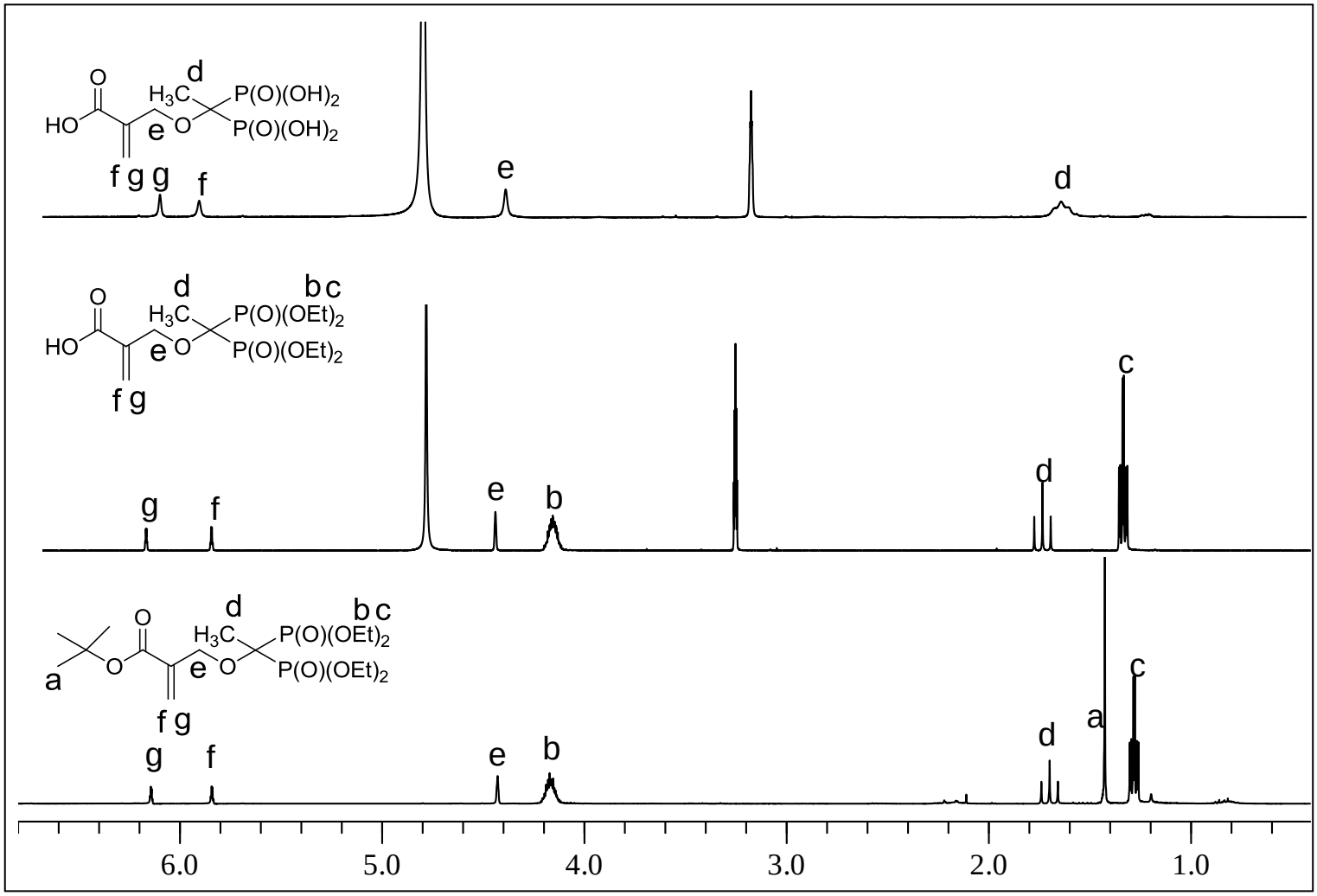


Figure 4.8. ^{1}H NMR spectra of **B2**, **B4** and **B5**.

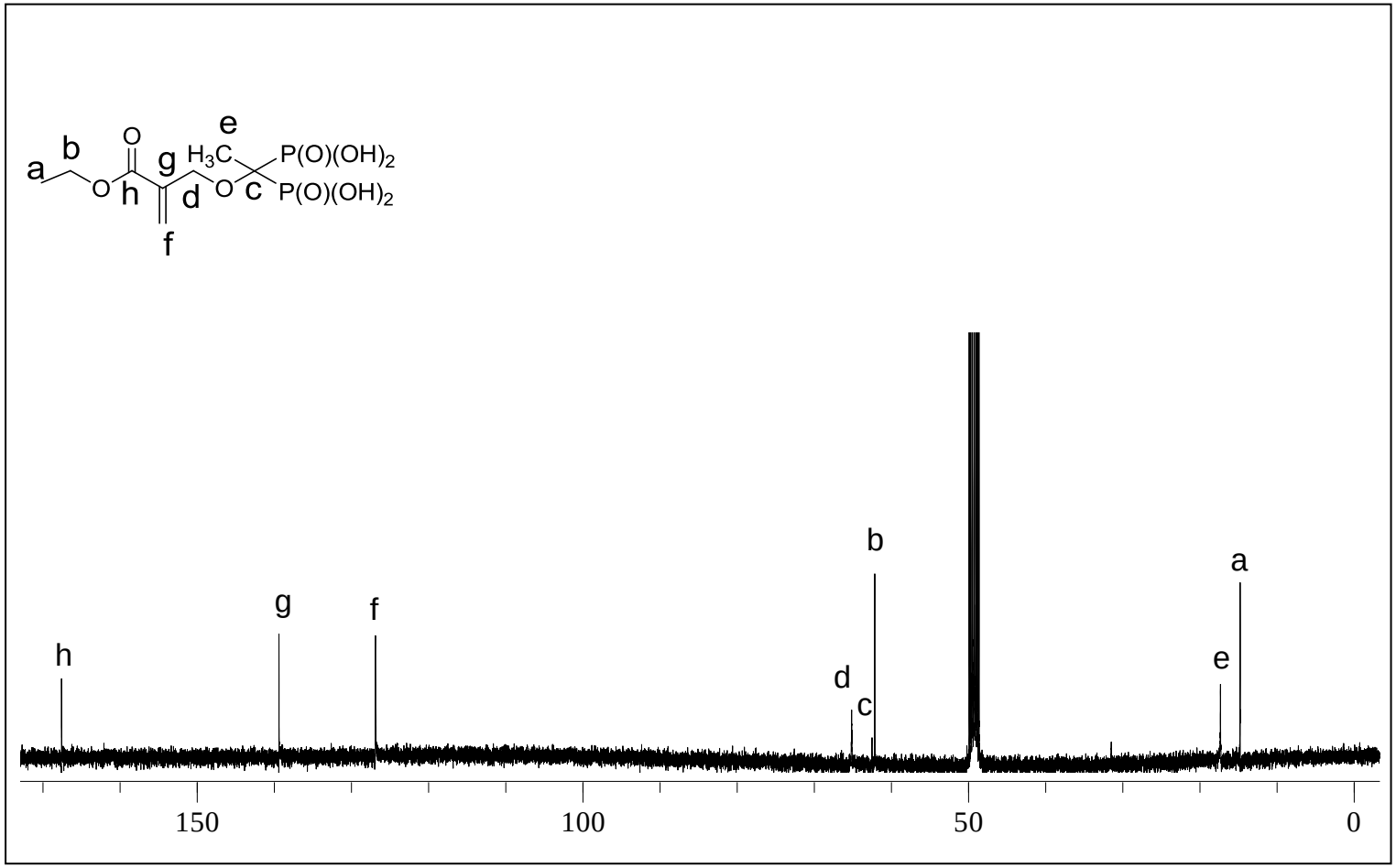


Figure 4.9. ^{13}C NMR spectrum of **B3**.

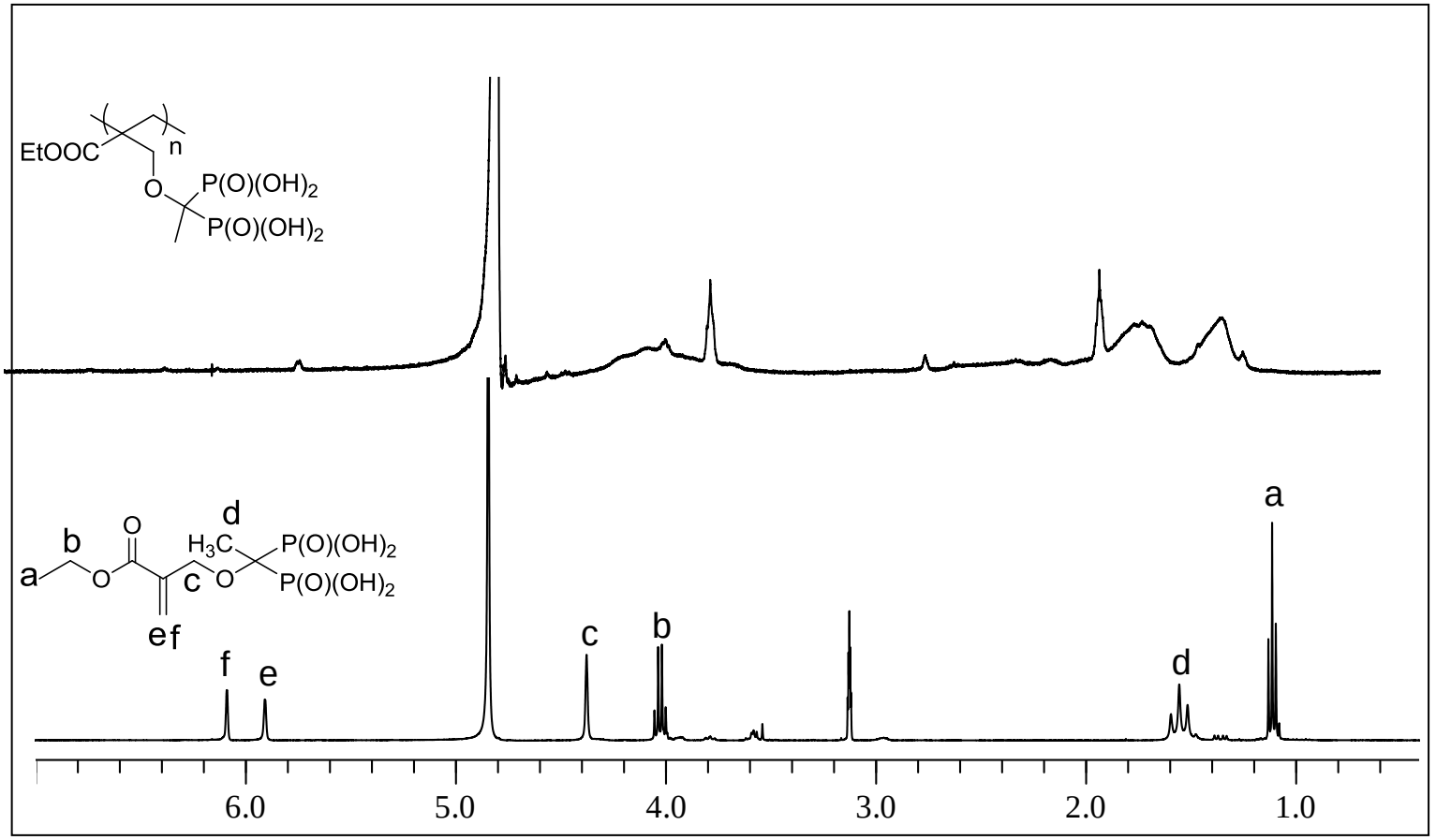


Figure 4.10. ^{1}H NMR spectra of **B3** and poly-**B3**.

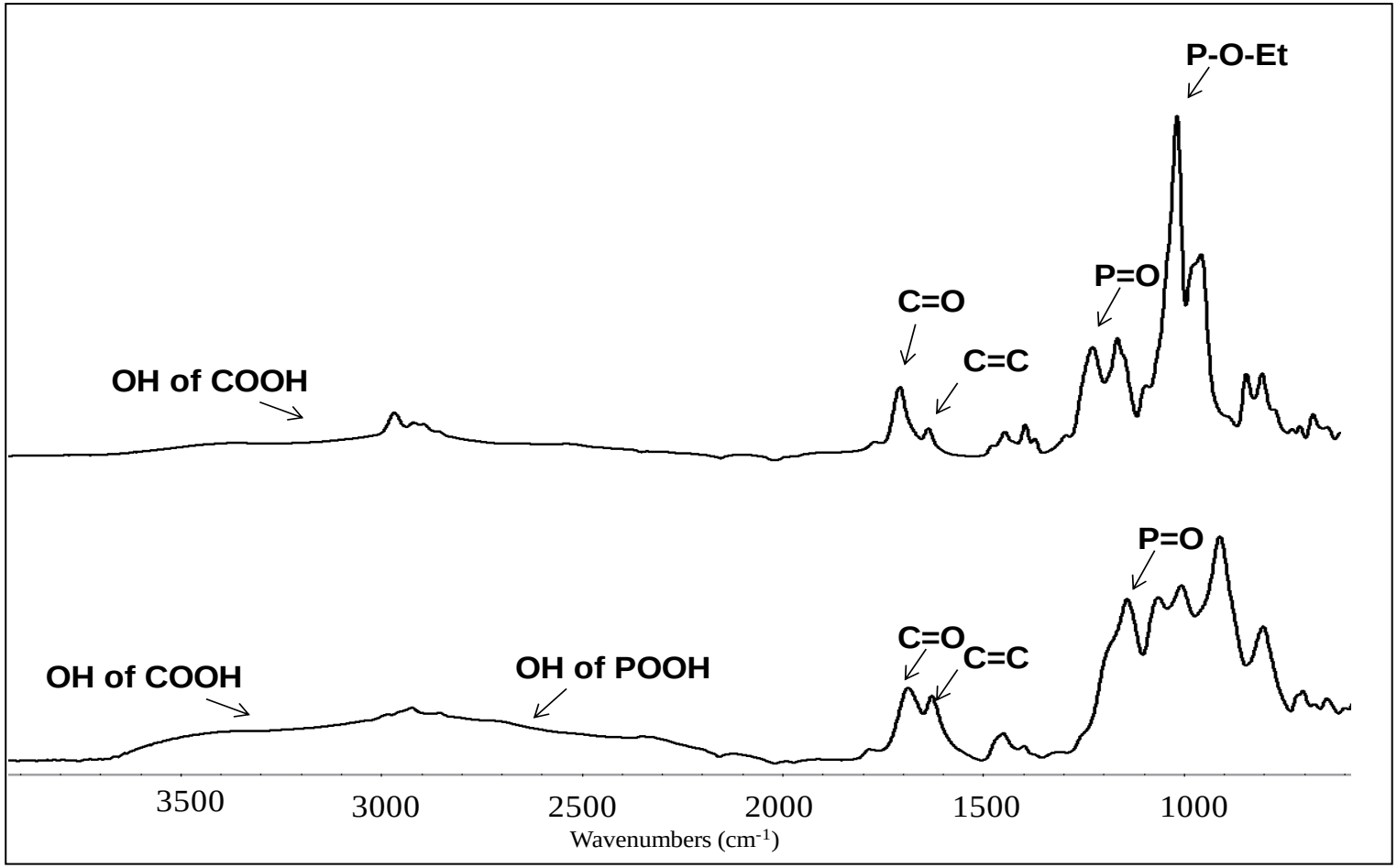


Figure 4.11. FTIR spectra of **B4** and **B5**.

4.3.2. Interactions of Monomers with Hydroxyapatite and Dentin

To determine the interaction of the synthesized monomers with HAP, we used ^{13}C and ^{31}P NMR, FTIR, SEM and XRD techniques. FTIR spectra of HAP particles mixed with monomer **B3**/EtOH/H_2O mixture for 5 min or 24 h and washed with water and ethanol were recorded. For the 5 min sample, the carbonyl peak observed on HAP disappeared after washing. But for the 24 h sample, the monomer stuck to and remained on HAP even after washing with ethanol and water (Figure 4.12). FTIR spectra of the solution part also confirmed the interaction of this monomer with HAP. The peaks at 920 and 999 cm^{-1} corresponding to the symmetric and asymmetric vibration of P-O in monomer **B3** decreased and a new peak appeared around 1142 cm^{-1}.

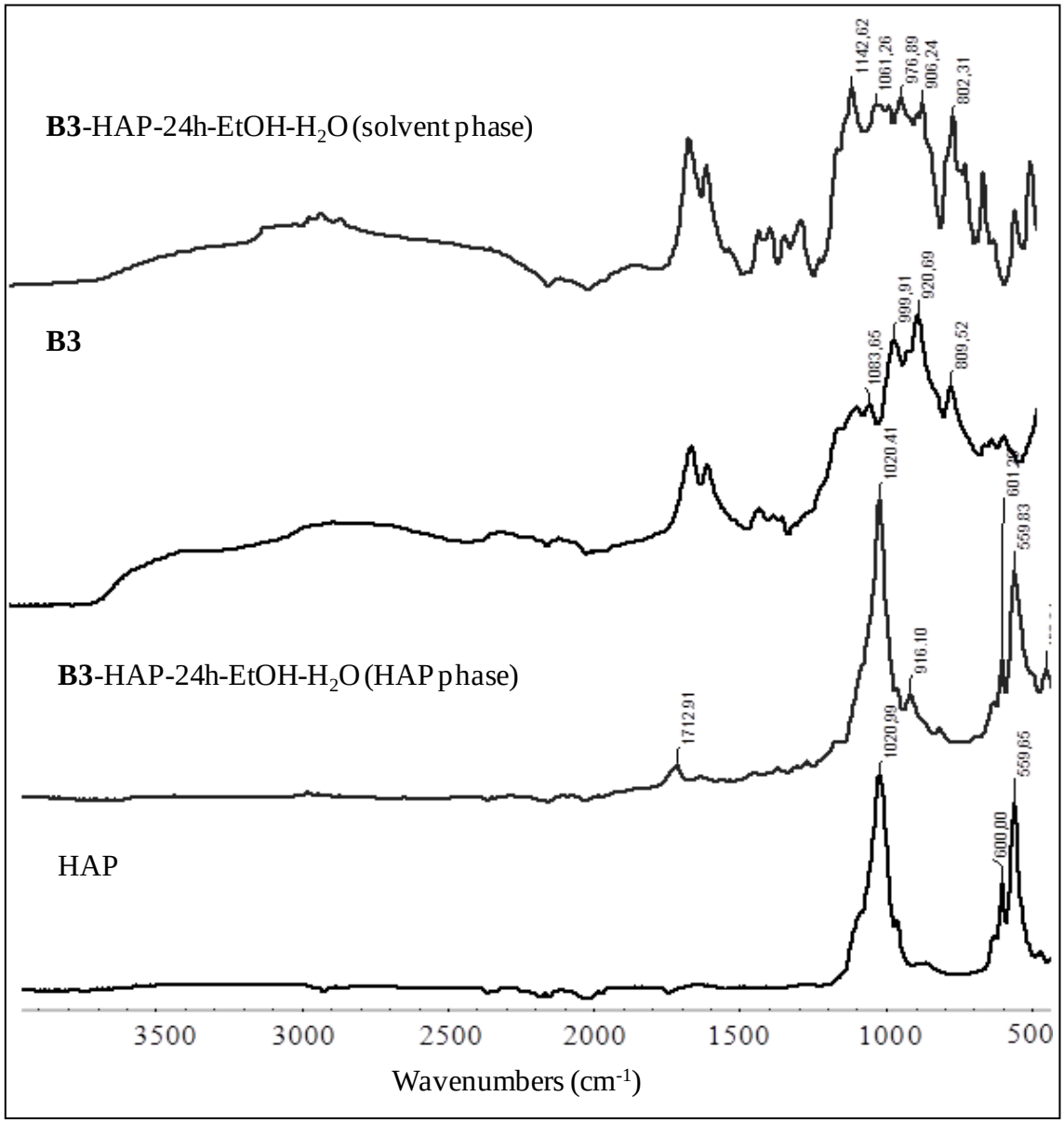


Figure 4.12. FTIR spectra of HAP, **B3**-HAP-24h-EtOH (HAP), **B3**, **B3**-HAP-24h-EtOH (solution).

SEM images of dentin samples treated with monomer **B3** showed complete coating of the surface by the monomer so that no dentin tubules were visible. After washing with ethanol, most of the monomer washed out but some was seen to have remained (Figure 4.13).

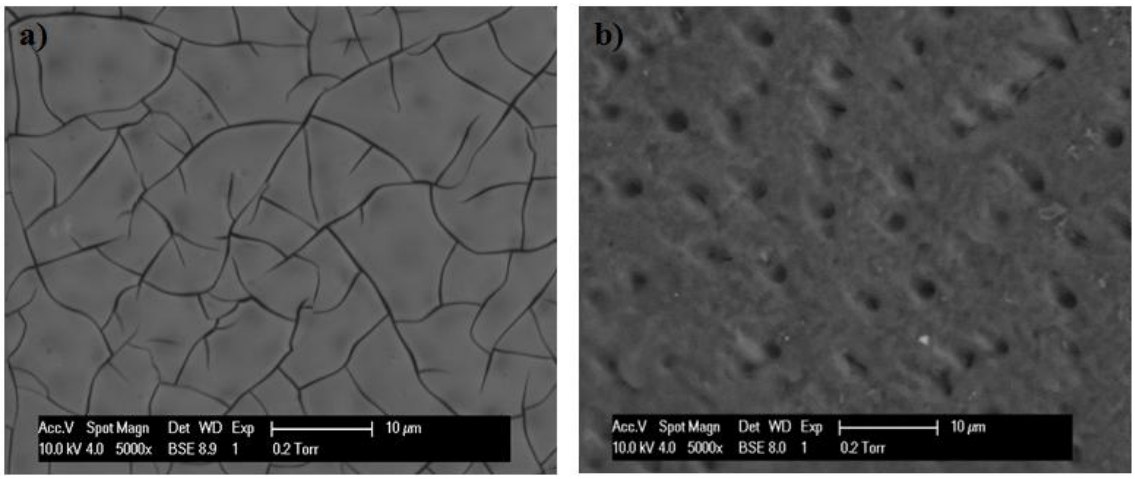


Figure 4.13. SEM images of dentin treated with **B3** a) before; b) after rinsing with EtOH.

Our XRD results can be interpreted within the framework of the mechanism proposed by Yoshida *et al.* [67]. According to this mechanism, first the decalcification of HAP is induced by monomer adsorption in the aqueous solution. Then, calcium salts of monomers, ($CaMHP_2$) and $CaHPO_4.2H_2O$ (DCPD), are deposited on HAP depending on their solubilities in water-ethanol solution. XRD spectra of HAP treated with **B3**/EtOH/H_2O solutions showed a new peak around $2\theta=5.2°$ which is assigned to Ca salts of monomers (Figure 4.14). These salts remained attached to HAP even after being washed with ethanol and water, indicating their stabilities and bonding performances. However, no peak was detected at $2\theta=11.6°$ due to deposited DCPD indicating absence of demineralization. We further analyzed the interaction of monomer **B3** with dentin. XRD pattern of dentin treated with **B3**/EtOH/H_2O solution showed no peak due to Ca salt of monomer **B3** and DCPD. This result may be explained by short application time of 20 s compared to 24 h application time of HAP (Figure 4.14).

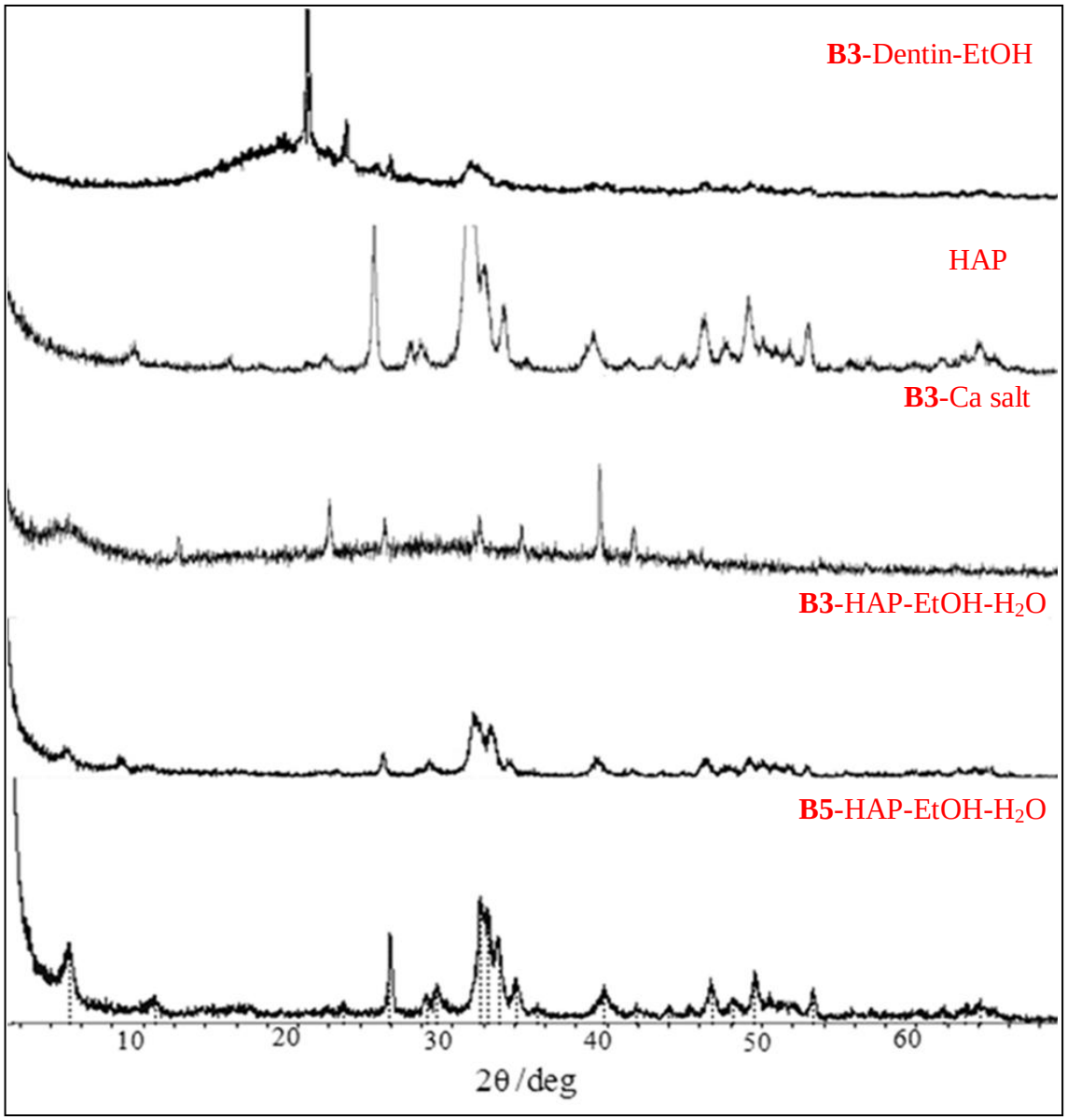


Figure 4.14. XRD patterns of **B3**-Dentin-EtOH, HAP, **B3**-Ca salt, **B3**-HAP-EtOH-H_2O and **B5**-HAP-EtOH-H_2O samples.

The interactions of monomers **B3** and **B5** with HAP were also studied by ^{13}C NMR and ^{31}P NMR techniques. The pH values of the aqueous solutions of **B3** and **B5** (7 wt %) were 1.87 and 1.64, respectively, and increased with the amount of HAP added, due to the acid-base reaction (Table 4.2). The ^{13}C NMR spectra showed that the quaternary carbon peak attached to the phosphonic acid groups and carbonyl carbon peak shifted to a lower field indicating the interaction of both monomers with HAP. ^{13}C NMR samples were also

analyzed with ^{31}P NMR. It was observed that the ^{31}P peak of monomer **B3** shifted to the left and the peak intensity decreased after mixing with HAP (Figure 4.15).

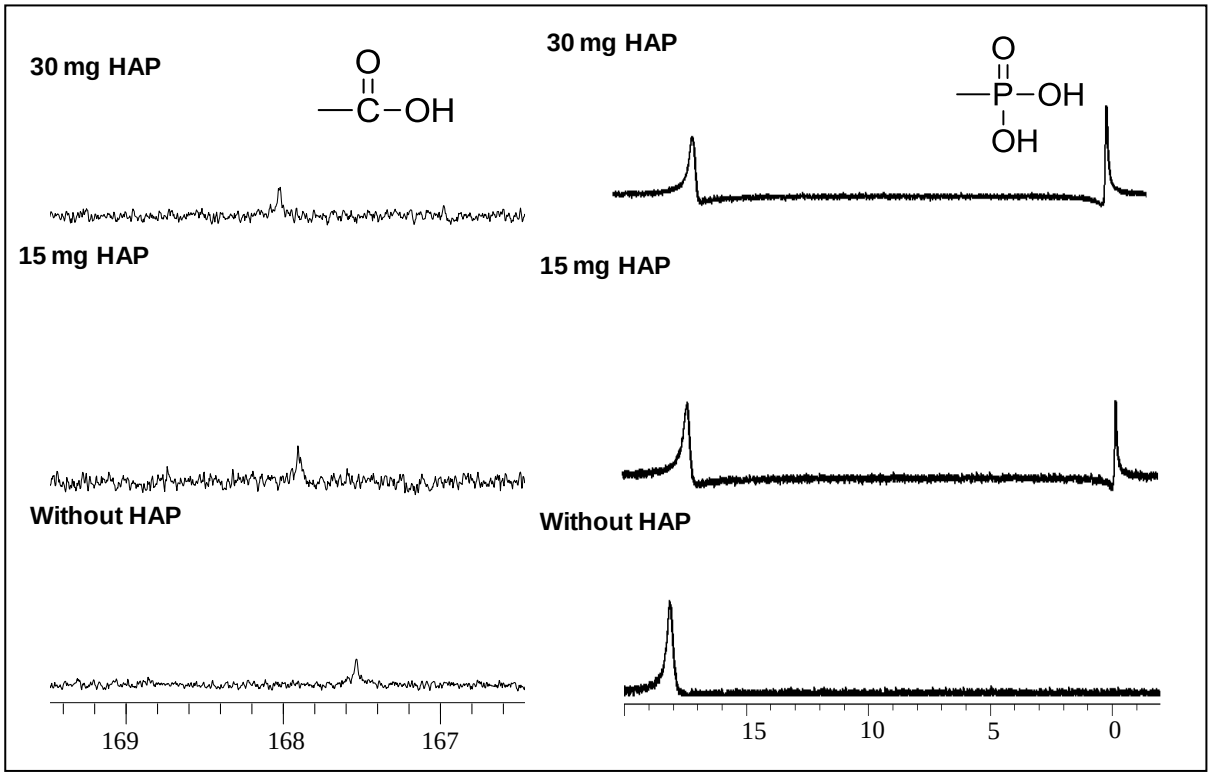


Figure 4.15. a) Expanded ^{13}C NMR spectra of the carbonyl region of **B5**; b) ^{31} P NMR spectra of **B3** treated with 15 and 30 mg HAP.

Table 4.2. Interactions of monomers **B3** and **B5** with HAP.

Monomer	**pH**[a]	**HAP**[b] **(mg)**	**pH**[c]	**-C-P(O)(OH)$_2$ SD**[d]	**-COOH SD**[e]
B3	1.87	15	2.79	+0.7	-
		30	3.88	+1.0	-
B5	1.64	15	2.53	+0.4	+0.4
		30	3.20	+0.6	+0.5

[a] pH of 7 wt % of monomer in water.

[b] Amount of HAP added to monomer solution.

[c] pH values of monomers after the addition of HAP.

[d] Chemical shift differences for the α-methylene carbon next to phosphorus in **B3** and **B5** after addition of HAP.

[e] Chemical shift differences for the carbonyl carbon of the carboxylic acid in **B5** after addition of HAP.

4.3.3. Hydrolytic Stability

Monomer **B3** and **B5** have bisphosphonic acid groups attached to double bond through ether linkage so these monomers were expected to be resistant to hydrolysis. This expectation was confirmed by ^{1}H NMR spectra of the monomers in aqueous methanol at 37 °C taken 30 days apart. No hydrolysis was observed for monomer **B5**. The only change was for the ethyl ester group of monomer **B3** which is susceptible to hydrolysis in acidic environment: At the end of 30 days it had hydrolysed by about 28 % (Figure 4.16). This hydrolysis reaction results in a carboxylic acid group, in other words, it converts monomer **B3** to monomer **B5**. The carboxylic acid group is one of those suitable for dental adhesive monomers, so this hydrolysis is not a problem.

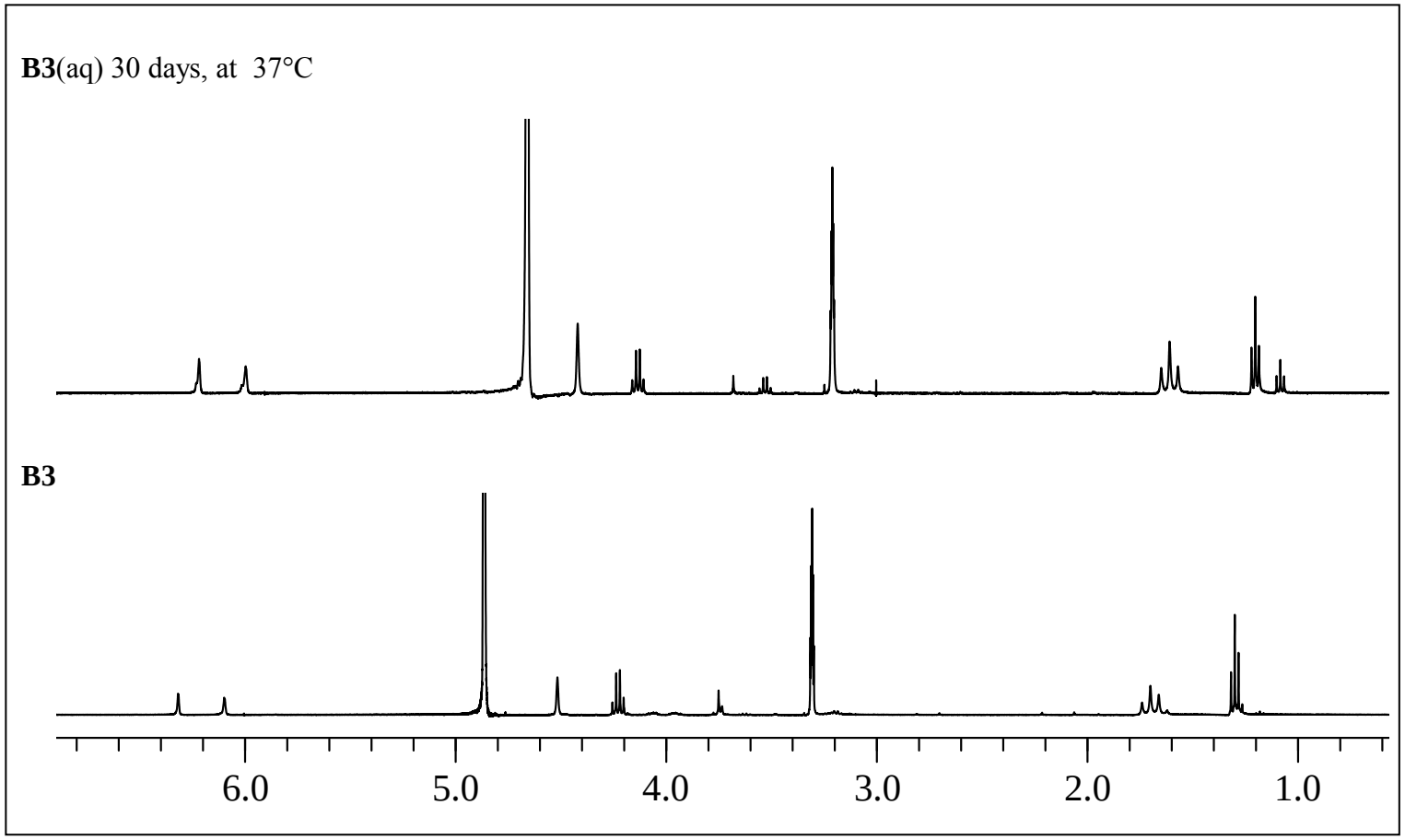


Figure 4.16. ^{1}H NMR spectrum of **B3** before and after 30 days in aqueous methanol at 37°C.

4.3.4. Polymerizations

In order to determine the reactivity of the synthesized monomers, they were photopolymerized using photodifferential scanning calorimetry. First, monomers **B1** and **B2** were homo- and copolymerized with HEMA at 40 °C using DMPA (2 mol %). The results indicated very low maximum rate of polymerization and conversion of these monomers. Addition of 10 and 50 mol% of these monomers to HEMA decreased both its rate and conversion (Table 4.3).

Table 4.3. Photopolymerization results of **B1** and **B2** at 40 °C using DMPA.

Monomer	**R_p (s^{-1})**	**Conversion (%)**
B1	0.001	23
B2	0.001	14
HEMA	0.032	83
B1/HEMA (10/90 mol %)	0.029	88
B1/HEMA (50/50 mol %)	0.012	47
B2/HEMA (10/90 mol %)	0.028	90
B2/HEMA (50/50 mol %)	0.012	44

Then, using BAPO (2 mol %) as catalyst, the acid monomers **B3** and **B4** were homopolymerized at 40 °C in water; but not monomer **B5**. This monomer becomes solid during photopolymerization; probably it precipitates due to its lower solubility, when some of the water evaporates during the process. In order to test the potential of the bisphosphonic acid monomers to be used in self-etching dental adhesives, their copolymerization kinetics with HEMA were also investigated. Various formulations consisting of mixtures of HEMA and water (60/40 wt %); HEMA, **B3** or **B5** and water (at different ratios) were photopolymerized (Table 4.4). The results show that addition of 5 wt% of **B3** or **B5** to HEMA increases its rate of polymerization slightly without changing

the conversion. Increasing monomer **B3** to 20 wt% in the mixture increases the rate significantly but decreases the conversion. A similar rate increase was not observed for monomer **B5** since it did not dissolve completely in the mixture. Both monomers **B3** and **B5**, when they replace some of HEMA in the formulation the conversion decreases, as can be seen in the Table 4.4. This is probably due to the presence in **B3** and **B5** of polar groups such as phosphonic acids, which increase the intermolecular interactions in the system and hence decrease the flexibility of the system. However, it was not possible to prepare HEMA, **B4** and water mixtures at the same ratios due to low solubility of **B4** in this mixture.

Table 4.4. Photopolymerization results of acid monomers at 40 °C using BAPO.

Monomer compositions (wt %)	**R_p (s^{-1})**	**Conversion (%)**
HEMA/H_2O (60/40)	0.031	91
B3/H_2O (60/40)	0.007	42
B3/HEMA/H_2O (5/55/40)	0.035	89
B3/HEMA/H_2O (20/40/40)	0.052	73
B4/H_2O (60/40)	0.007	54
B5/HEMA/H_2O (5/55/40)	0.034	92
B5/HEMA/H_2O (20/40/40)	0.038	80

Bulk copolymerization of the acid monomers with a mixture of HEMA and GDMA were studied in the presence of DMPA. The mixtures of HEMA/monomer/GDMA (5/2/3 mol/mol/mol) and HEMA/GDMA (7/3 mol/mol) were photopolymerized at 40 °C. Figure 4.17 shows photopolymerization results such as time to reach the maximum polymerization rate (t_{max}), maximum rate of polymerization (R_{pmax}) and conversion obtained, using photo-DSC. It was clearly seen that the mixture containing the acid monomer **B3** was more reactive than HEMA/GDMA (7/3 mol/mol) mixture with improved

t_{max} (4.8 s), R_{pmax} (0.061 s^{-1}) values. The mixtures containing acidic monomers **B4** and **B5** also showed lower t_{max} values compared to HEMA/GDMA (7/3 mol/mol) mixture but comparable R_{pmax} values.

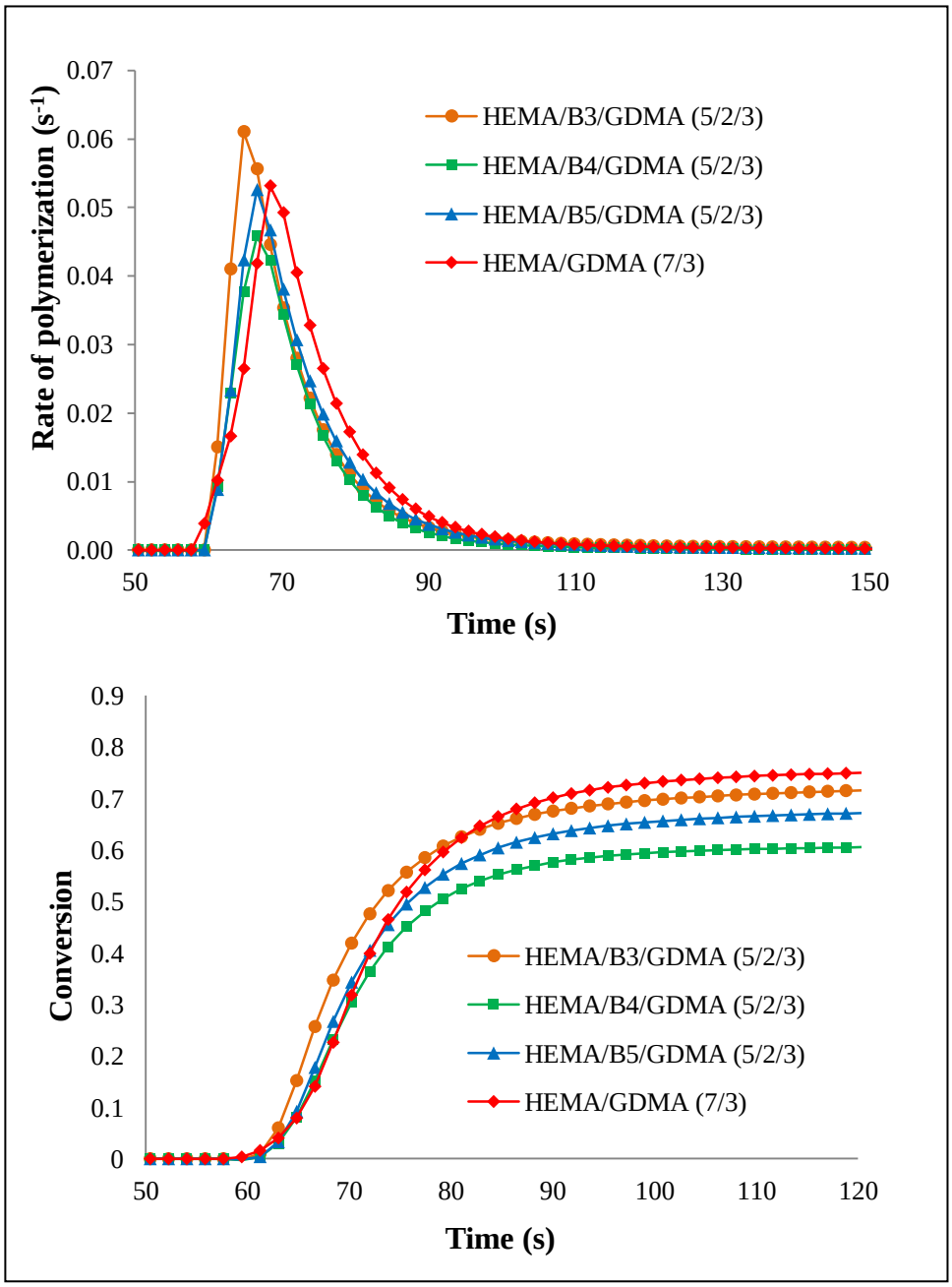


Figure 4.17. Rate-time and conversion-time curves for copolymerizations of **B3, B4** and **B5** with GDMA and HEMA.

Thermal homopolymerization of monomer **B3** in water with V-50 gave soluble polymers in 40% yield. This polymer was soluble in methanol and water but insoluble in isopropyl alcohol and THF. ^{1}H NMR of this polymer indicated disappearance of double

bond peaks at 5.90 and 6.08 ppm after polymerization (Figure 4.10). The number average molecular weight (M_n) for this polymer changed between 1000-6000 as estimated by GPC, indicating the importance of chain transfer reactions due to bulky structure of this monomer. The DSC analysis showed a T_g value at around 75-80 °C. No results are reported for poly- **B4** or poly-**B5** since did not polymerize under the same conditions.

4.4. Conclusion

Five novel ether-linked bisphosphonate, carboxylic acid and bisphosphonic acid-containing methacrylates were successfully synthesized starting from a *gem*-hydroxy-diphosphonate. They were evaluated for potential performance as dental materials. The photopolymerization reactivities of the bisphosphonate-containing ones were found to be very low; carboxylic acid containing one had low solubility in dental formulations prepared. However, the bisphosphonic acid-containing monomers showed good performance in terms of solubility, acidity and copolymerizability with HEMA in water and HEMA/GDMA in bulk. Interactions of these monomers with HAP were found to result in the formation of hydrolytically stable monomer Ca salts. One of these monomers is hydrolytically stable, the other one turns into the former upon hydrolysis, showing promise of good shelf life and bonding reliability. In summary, our investigations indicate that these two bisphosphonic acid-containing methacrylates constitute a new class of monomers for potential use in self-etching adhesives and self-adhesive resin cements.

5. SYNTHESIS AND POLYMERIZATIONS OF SIX AMINOPHOSPHONATE-CONTAINING METHACRYLATES

5.1. Introduction

Phosphorus-containing monomers and polymers have been have been the subject of much research due to their broad variety of application areas. Generally, polyphosphonates and polyphosphates are known as flame retardants [1, 2]. They have also been used in biomedical field as adhesion promoters to dental tissue, bone and metals. For example, monomers with phosphonic and phosphoric acid functional groups have been used in dental composites and self-etching dental adhesives [20, 27, 44, 170]. Bisphosphonate-containing polymers are used in bone-targeted drug delivery applications [171]. Polymeric bisphosphonates inhibit bone resorption due to their chelating ability with calcium ions of bone [108, 110].

Polymers containing both amine and phosphonic acid groups and aminomethylphosphonic acid group are used as chelating resins for metals [113, 172-174]. Monomeric and polymeric aminomethylphosphonic acids exhibit good complexation ability for the transition metal ions Cd^{+2} and Hg^{+2}, this selective extraction of the metal ions from aqueous solution can be used for the treatment of wastewater, groundwater and seawater [112]. Besides their chelating abilities to metals, aminophosphonic acids which are structural analogues of amino acids are suitable for biological applications such as inhibition of enzymes, antibacterials, neuroactive compounds, anticancer drugs or pesticides [175].

In recent years, we synthesized phosphorus-containing monomers based on alkyl α-hydroxymethacrylates, methacryloyl chloride and glycidyl methacrylate via different routes to be used in dental materials [30, 31, 53, 54, 176]. In this work, we report synthesis of new monomers based on alkyl α-hydroxymethacrylates (RHMA) and 3-(acryloyloxy)-2-hydroxypropyl methacrylate (AHM) and containing aminophosphonate groups and investigation of their homopolymerization behavior using thermal or photopolymerization techniques to determine structure-reactivity relationships of these monomers.

5.2. Experimental

5.2.1. Materials and Apparatus

5.2.1.1. Materials. Ethyl α-bromomethacryate (EBBr), *tert*-butyl α-bromomethacryate (TBBr), diethyl aminomethylphosphonate and diethyl 2-aminoethylphosphonate were synthesized according to literature procedures [153-155, 177, 178]. The initiators, 2,2'-azobis(isobutyronitrile) (AIBN), benzophenone (BP) and 2,2'-dimethoxy-2-phenyl acetophenone (Irgacure 651), solvents and starting materials were obtained from Aldrich and used as received.

5.2.1.2. Apparatus. The monomer characterization involved ^{1}H, ^{13}C and ^{31}P NMR spectroscopy (Varian Gemini 400 MHz) and Fourier transform infrared (FTIR) spectroscopy (T 380). The photopolymerizations were carried out on a TA Instruments Q100 differential photocalorimeter (DPC). Gel permeation chromatography (Viscotek) was carried out with THF solvent using polystyrene standards. Thermogravimetric analysis was done with a TA Instrument Q50. Elemental analyses were obtained fromThermo Electron SpA FlashEA 1112 elemental analyser (CHNS separation column, PTFE; 2 m; 6 x5 mm).

5.2.2. Synthesis of Monomers

5.2.2.1. Diethyl 2,2'-(((diethoxyphosphoryl)methyl)azanediyl)bis(methylene))diacrylate **(C1)**. To a mixture of diethyl aminomethylphosphonate (2.67 g, 15.97 mmol) and K_2CO_3 (4.74 g, 34.34 mmol) in DMF (6.5 mL) and THF (18.6 mL) in an ice bath, EBBr (7.71 g, 39.95 mmol) was added dropwise. The mixture was stirred at 80 °C for 4 h. After removal of the solvent, the residue was washed with 15 mL of water and the aqueous phase was extracted with CH_2Cl_2 (6 mLx3). The organic phases were combined and dried with Na_2SO_4, filtered and the solvent was evaporated. The crude product was purified by column chromatography using hexane initially and gradually changing to ethyl acetate as eluent. The pure product was obtained as colorless oil in 39 % yield.

^{1}H NMR (400 MHz,$CDCl_3$, δ): 1.24 (t, 6H, C*H*$_3$CH$_2$OC), 1.26 (t, 6H, C*H*$_3$CH$_2$OP), 2.87, 2.90 (d, 2H, CH_2-P), 3.47 (s, 4H, CH_2-N), 4.05 (m, 4H, CH_2-O), 4.12 (m, 4H, CH_2-OP), 5.86 (s, 2H, CH_2=), 6.23 ppm (s, 2H, CH_2=).

^{13}C NMR (400 MHz, $CDCl_3$, δ):13.73 (*C*H$_3$CH$_2$OC), 16.07 (*C*H$_3$CH$_2$OP), 47.65, 49.21 (CH_2-P), 55.11 (CH_2-N), 60.12 (CH$_3$*C*H$_2$OC), 61.29 (CH$_3$*C*H$_2$OP), 126.00 (*C*H$_2$=C), 137.17 (CH_2=*C*), 166.06 ppm (C=O).

^{31}P NMR (400 MHz, $CDCl_3$, δ): 25.12 ppm.

FTIR(ATR): *v* = 2982 (m, C-H), 1712 (vs, C=O), 1634 (s, C=C), 1259 (vs, P=O), 1021 and 953 cm^{-1} (vs, P-O-Et).

5.2.2.2. Di-*tert*-butyl2,2'-((((diethoxyphosphoryl)methyl)azanediyl)bis(methylene)) diacrylate (**C2**). To a mixture of diethyl aminomethylphosphonate (1.00 g, 5.98 mmol) and K_2CO_3 (1.77 g, 12.86 mmol) in DMF (2.2 mL) and THF (7.0 mL) in an ice bath, TBBr (4.63 g, 20.95 mmol) was added dropwise. The mixture was stirred at 65 °C for 24 h. After removal of the solvent, the residue was washed with 12 mL of water and the aqueous phase was extracted with CH_2Cl_2 (5 mLx3). The organic phases were combined, dried with Na_2SO_4, filtered and the solvent was evaporated. The crude product was purified by column chromatography using hexane initially and gradually changing to ethyl acetate as eluent. The pure product was obtained as colorless oil in 77 % yield.

^{1}H NMR (400 MHz,$CDCl_3$, δ): 1.25 (t, 6H, C*H*$_3$CH$_2$), 1.43 [s, 18H, (CH$_3$)$_3$C], 2.88 (d, 2H, CH_2-P), 3.42 (s, 4H, CH_2-N), 4.06 (q, 4H, CH_2-O), 5.80 (s, 2H, CH_2=), 6.14 ppm (s, 2H, CH_2=).

^{13}C NMR (400 MHz, $CDCl_3$, δ): 16.32 (*C*H$_3$CH$_2$O), 27.90 [(*C*H$_3$)$_3$C], 48.13, 49.70 (CH_2-P), 55.43 (*C*H$_2$-N), 61.68 (CH$_3$*C*H$_2$O), 80.53 [(CH$_3$)$_3$*C*], 125.26 (*C*H$_2$=C), 138.67 (CH_2=*C*), 165.78 ppm (C=O).

FTIR (neat): *v* = 2978 (m, C-H), 1710 (vs, C=O), 1635 (s, C=C), 1250 (vs, P=O), 1025 and 953 cm^{-1} (vs, P-O-Et).

Calcd. for $C_{21}H_{38}NPO_7$: C 56.36, H 8.56, N 3.13, O 25.03, P 6.92; found: C 56.36, H 9.26, N 3.04.

5.2.2.3. Diethyl 2,2'-(((2-(diethoxyphosphoryl)ethyl)azanediyl)bis(methylene)) diacrylate (**C3**). Monomer **C3** was synthesized with the same procedure for monomer **C1** using diethyl 2-aminoethylphosphonate instead of diethyl aminomethylphosphonate. The pure product was obtained as colorless oil in 36 % yield.

^{1}H NMR (400 MHz, $CDCl_3$, δ):1.25 (t, 6H, C*H*$_3$CH$_2$OC), 1.26 (t, 6H, C*H*$_3$CH$_2$OP), 1.95 (m, 2H, CH$_2$C*H*$_2$P), 2.75 (m, 2H, C*H*$_2$CH$_2$-P), 3.24 (s, 4H, CH$_2$-N), 4.04 (m, 4H, CH$_3$*C*H$_2$OC), 4.16 (q, 4H, CH$_3$*C*H$_2$OP), 5.75 (s, 2H, CH$_2$=), 6.19 ppm (s, 2H, CH$_2$=).

^{13}C NMR (400 MHz, $CDCl_3$, δ): 14.06 (*C*H$_3$CH$_2$O), 16.35 (*C*H$_3$CH$_2$O), 22.80, 24.16 (CH$_2$*C*H$_2$P), 47.09 (*C*H$_2$CH$_2$P), 53.77 (CH$_2$N), 60.50 (CH$_3$*C*H$_2$O), 61.43 (CH$_3$*C*H$_2$O), 125.92 (*C*H$_2$=C), 137.90 (CH$_2$=*C*), 166.60 ppm (C=O).

^{31}P NMR (400 MHz, $CDCl_3$, δ): 25.16 ppm.

FTIR(neat): *v* = 2983 (m, C-H), 1714 (vs, C=O), 1635 (s, C=C), 1250 (vs, P=O), 1021 and 955 cm^{-1} (vs, P-O-Et).

5.2.2.4. Di-*tert*-butyl 2,2'-(((2-(diethoxyphosphoryl)ethyl)azanediyl)bis(methylene)) diacrylate (**C4**). Monomer **C4** was synthesized with the same procedure for monomer **C2** using diethyl 2-aminoethylphosphonate instead of diethyl aminomethylphosphonate. The pure product was obtained as colorless oil in 20 % yield.

^{1}H NMR (400 MHz, $CDCl_3$, δ):1.29 (t, 6H, C*H*$_3$CH$_2$), 1.46 [s, 18H, (CH$_3$)$_3$C], 2.02 (t, 2H, CH$_2$C*H*$_2$P), 2.77 (m, 2H, C*H*$_2$CH$_2$P), 3.22 (s, 4H, CH$_2$-N), 4.07 (q, 4H, CH$_3$*C*H$_2$O), 5.70 (s, 2H, CH$_2$=), 6.11 ppm (s, 2H, CH$_2$=).

^{13}C NMR (400 MHz, $CDCl_3$, δ): 16.44 (*C*H$_3$CH$_2$O), 23.02, 24.38 (CH$_2$-P), 28.04 [(*C*H$_3$)$_3$C], 47.28 (CH$_2$-N), 53.88 (*C*H$_2$C=), 61.49 (CH$_3$*C*H$_2$0), 80.64 [(CH$_3$)$_3$*C*], 124.98 (*C*H$_2$=C), 139.34 (CH$_2$=*C*), 166.05 ppm (C=O).

^{31}P NMR (400 MHz, $CDCl_3$, δ): 30.45 ppm.

FTIR (neat): *v* = 2987 (m, C-H), 1710 (vs, C=O), 1635 (s, C-C), 1246 (vs, P=O), 1027 and 951 cm^{-1} (vs, P-O-Et).

5.2.2.5. 3-((3-(((diethoxyphosphoryl)methyl)amino)propanoyl)oxy)-2-hydroxypropyl methacrylate (**C5**). Equimolar amounts of AHM (0.285 g, 1.33 mmol) and diethyl aminomethylphosphonate (0.22 g, 1.33 mmol) were stirred at room temperature for 24 h. The product was obtained as colorless oil.

^{1}H NMR (400 MHz, $CDCl_3$, δ): 1.30 (t, 6H, C*H*$_3$CH$_2$), 1.92 (s, 3H, CH_3C), 2.54 (m, 2H, CH_2-C=O), 2.95 (m, 2H, CH_2CH_2N), 2.99 (m, 2H, NCH_2P), 4.10 (q, 4H, CH_3CH_2O), 4.19 (m, 2H, CH_2O), 4.23 (m, 2H, C*H*$_2$CHOH), 4.24 (m, 1H, CH_2CHOH), 5.57 (s, 1H, CH_2=), 6.11 ppm (s, 1H, CH_2=).

^{13}C NMR (400 MHz, $CDCl_3$, δ):16.19 (*C*H$_3$CH$_2$O), 18.04 (*C*H$_3$C), 34.30 (*C*H$_2$C=O), 43.66, 46.04 (CH_2P), 45.19 (CH_2-N) 62.01 (CH_3CH_2O), 65.01 ($CHCH_2O$), 65.46 (CH_2CHOH), 67.25 ($CHCH_2O$), 125.89 (*C*H$_2$=C), 135.66 (CH_2=*C*), 167.95 (C=O), 171.97 ppm (C=O).

^{31}P NMR (400 MHz, $CDCl_3$, δ): 25.78 ppm.

FTIR (neat): *v* = 3332 (br, m, O-H), 2982 (m, C-H), 1718 (vs, C=O), 1636 (s, C=C), 1295 (s, P=O), 1021 and 958 cm^{-1} (vs, P-O-Et).

5.2.2.6. 3-((3-((2-(diethoxyphosphoryl)ethyl)amino)propanoyl)oxy)-2-hydroxypropyl methacrylate (**C6**). Monomer **C6** was synthesized with the same procedure for monomer **C5** using diethyl 2-aminoethylphosphonate instead of diethyl aminomethylphosphonate. The product was obtained as colorless oil.

^{1}H NMR (400 MHz, $CDCl_3$, δ): 1.31 (t, 6H, C*H*$_3$CH$_2$), 1.94 (s, 3H, CH_3C), 1.98 (m, 2H, CH_2-P), 2.55 (m, 2H, CH_2-C=O), 2.86 (m, 2H, PCH_2CH_2N), 2.91 (m, 2H, $O=CCH_2CH_2N$),

4.09 (q, 4H, CH_3CH_2O), 4.21 (m, 2H, CH_2CHOH), 4.29 (m, 2H, CH_2CHOH), 4.30 (m, 1H, CH_2CHOH), 5.58 (s, 1H, CH_2=), 6.13 ppm (s, 1H, CH_2=).

^{13}C NMR (400 MHz, $CDCl_3$, δ): 16.24 (CH_3CH_2O), 18.15 (CH_3C), 25.00, 26.39 (CH_2P), 34.66 (CH_2C=O), 42.82 (CH_2-N), 44.84 (CH_2-N), 61.69 (CH_3CH_2O), 65.04 ($CHCH_2O$), 65.57 (CH_2CHOH), 67.28 ($CHCH_2O$), 125.97 (CH_2=C), 135.78 (CH_2=C), 167.07 (C=O), 172.29 ppm (C=O).

^{31}P NMR (400 MHz, $CDCl_3$, δ): 30.35 ppm.

FTIR (neat): ν = 3372 (br, m, OH), 2983 (m, CH), 1717 (vs, C=O), 1636 (s, C=C), 1296 (s, P=O), 1021 and 952 cm^{-1} (vs, P-O-Et).

5.2.3. Polymerizations

5.2.3.l. Thermal polymerizations. All thermal polymerizations were run in septum-sealed glass tubes that were subjected to freeze-evacuate-thaw procedures and placed in 60-80 °C oil baths. After a selected period, polymers were dissolved in CH_2Cl_2, polymers of **C1** and **C3** were precipitated into methanol: water mixture (1:1) twice while polymers of **C2** and **C4** were precipitated into hexane.

5.2.3.2. Photopolymerizations. Approximately 3.0 or 4.0 mg of sample was placed in an aluminium DSC pan. The photoinitiator which was dissolved in CH_2CI_2 was added with a micro-syringe to give a final concentration in the monomer of 2.0 mol per cent after evaporation of the solvent. The sample and the reference pans were placed in the DSC chamber, the system was purged with nitrogen flow to remove air and CH_2Cl_2 for 10 min before polymerization and purging was continued during polymerization. Heats of photoreactions were measured using a DPC equipped with a mercury arc lamp. The samples were irradiated for 10 min at 40 °C with an incident light density of 20 mW/cm^2. The heat flux as a function of reaction time was monitored using DSC under isothermal conditions and both the rate of polymerization (R_p) and conversion were calculated as a function of time. The theoretical value used for the heats of reaction (ΔH_p) was 13.1

kcal/mol for methacrylate double bonds [157, 158]. Rates of polymerization were calculated according to the following formula:

$$Rate = \frac{\left(Q/s\right)M}{n(\Delta Hp)m} \quad (5.1)$$

where Q/s is heat flow per second, M the molar mass of the monomer, n the number of double bonds per monomer molecule, ΔH_p is the heat released per mole of double bonds reacted and m the mass of monomer in the sample.

5.3. Results and Discussion

5.3.1. Synthesis of Monomers

The first group of aminophosphonate-containing monomers (**C1-C4**) were synthesized by reaction of diethyl aminomethylphosphonate or diethyl 2-aminoethylphosphonate with EBBr or TBBr (Figure 5.1). Monomer to amine ratio used was 2.5 to 3.5 to minimize the formation of monoadducts (secondary amines). Reaction times for the *tert*-butyl ester derivatives were longer (24 h at 65°C) compared to those of ethyl ester derivatives (4 h at 80 °C) due to steric effect of bulky *tert*-butyl groups. Monomers were obtained as colorless liquids after purification using column chromatography with overall yields of 39, 77, 36 and 20 % respectively, as reported above. Satisfactory microanalysis results were obtained for representative monomer **C2**. The monomers were soluble in acetone, THF, methylene chloride and methanol but insoluble in water; while **C1** and **C3** were also insoluble in hexane, monomers **C2** and **C4** were soluble (Table 5.1).

The expected structures of the monomers were confirmed by NMR and FTIR spectroscopies. The ^{13}C NMR spectrum of monomer **C1** showed characteristic peaks for methyl carbons at 13.73 and 16.07 ppm, methylene carbon attached to phosphorus at 47.65

and 49.21 ppm, methylene carbon attached to nitrogen at 55.11 ppm, methylene carbons attached to oxygen at 60.12 and 61.29 ppm, double bond carbons at 126.00 and 137.17 ppm and a carbonyl carbon at 166.06 ppm (Figure 5.2). In the ^{31}P NMR spectrum, one peak at 25.12 ppm confirmed the purity of this monomer (Figure 5.2). The ^{1}H NMR of monomer **C2** is shown in Figure 5.5. The doublet seen at 2.88 ppm is due to methylene carbon attached to phosphorus. The FTIR spectra of these monomers are similar and shows CH, C=O, C=C, P=O and P-O-C peaks around 2978, 1710, 1635, 1250, 1025 and 953 cm^{1}.

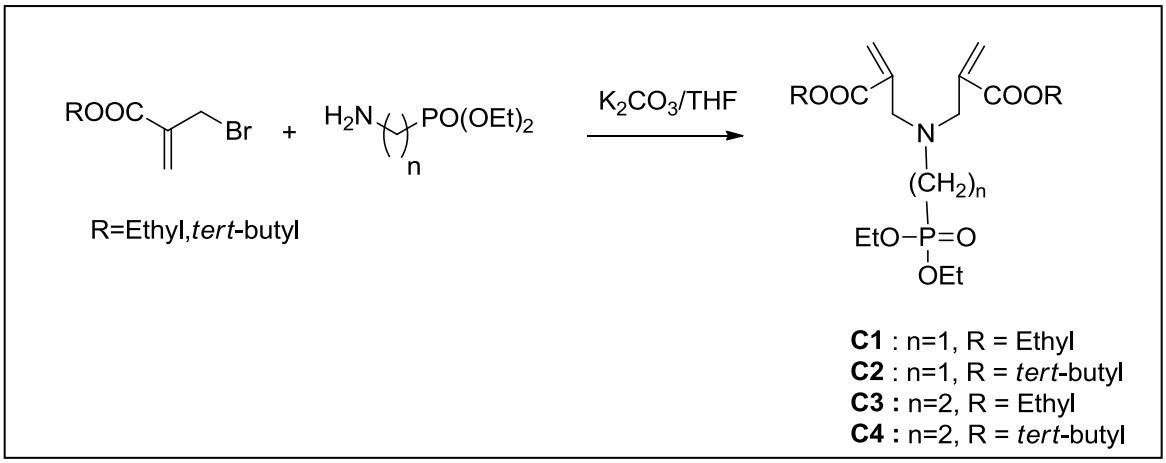


Figure 5.1. Synthesis of monomers **C1-C4.**

Table 5.1 Solubility of monomers and polymers in selected solvents.

Monomers	**Hexane**	**THF**	**Acetone**	**CH_2Cl_2**	**MeOH**	**H_2O**
C1	-	+	+	+	+	-
C2	+	+	+	+	+	-
C3	-	+	+	+	+	-
C4	+	+	+	+	+	-
C5	-	+	+	+	+	+
C6	-	+	+	+	+	+
Poly-**C1**	-	+	+	+	+	-
Poly-**C2**	-	+	+	+	+	-
Poly-**C3**	-	+	+	+	+	-
Poly-**C4**	-	+	+	+	+	-
Poly-**C5**	-	-	-	-	-	-
Poly-**C6**	-	-	-	-	-	-

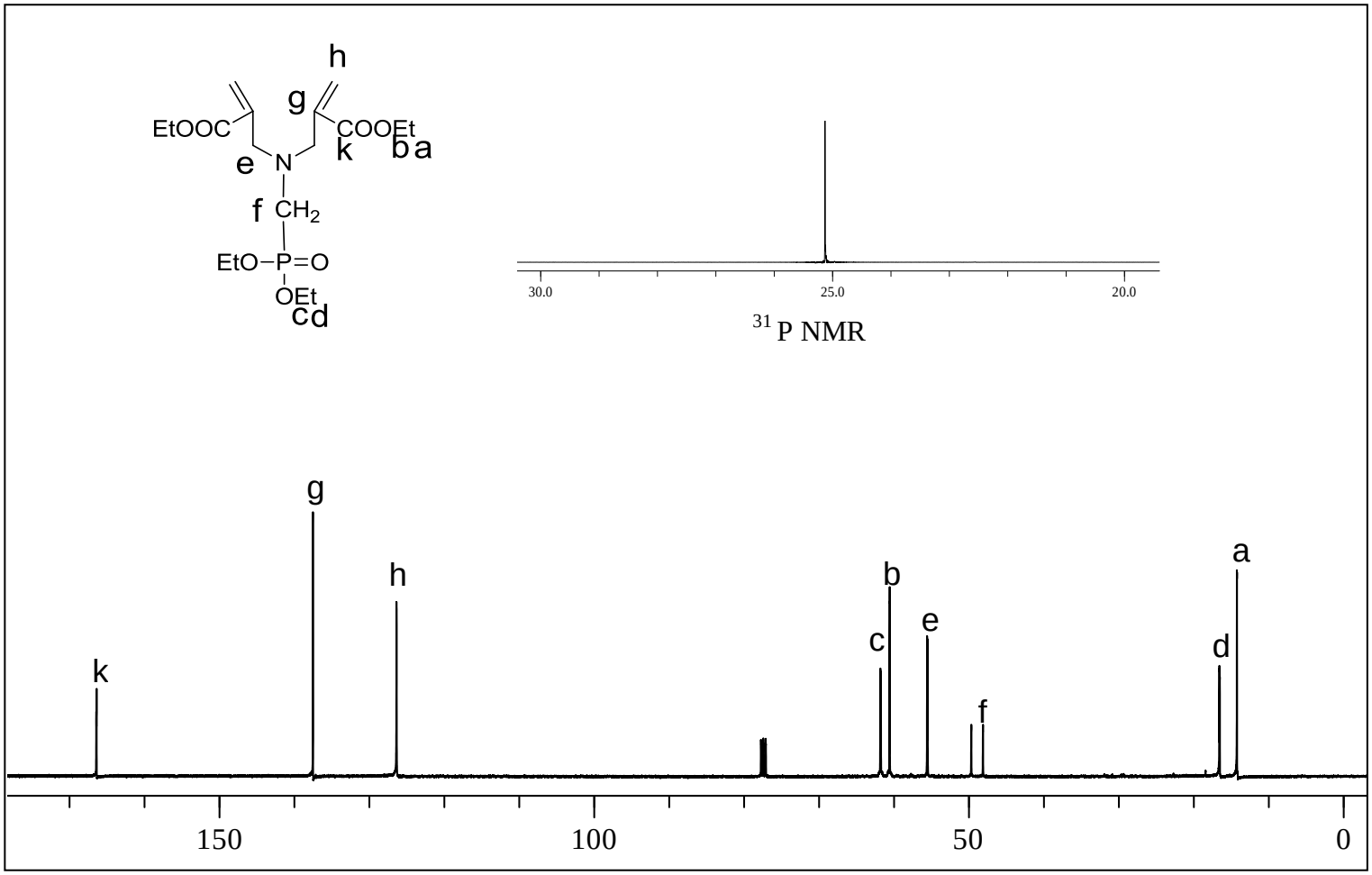


Figure 5.2 . ^{13}C and ^{31}P NMR spectra of monomer **C1**.

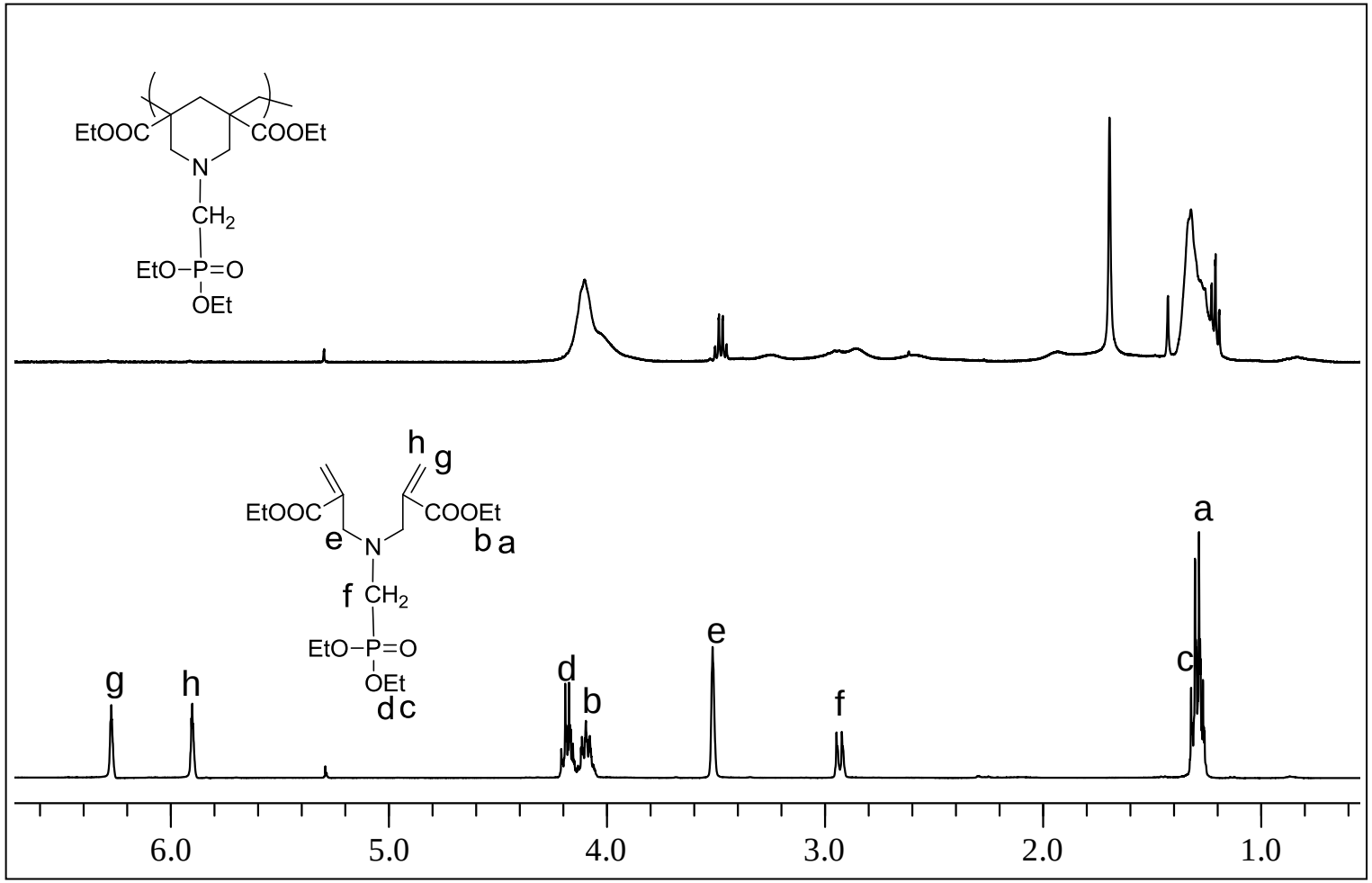


Figure 5.3. ^{1}H NMR spectra of **C1** and poly-**C1**.

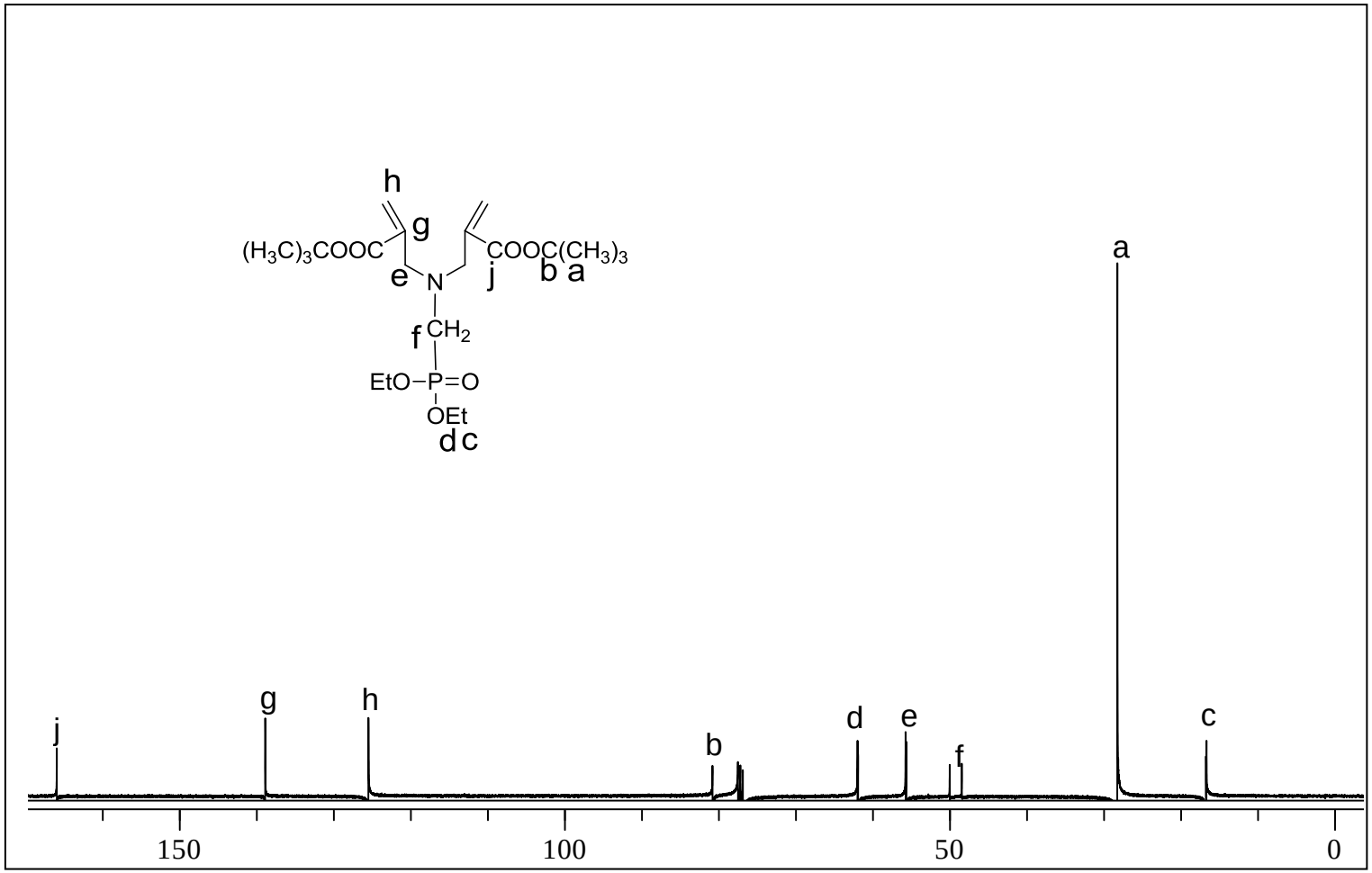


Figure 5.4. ^{13}C NMR spectrum of **C2**.

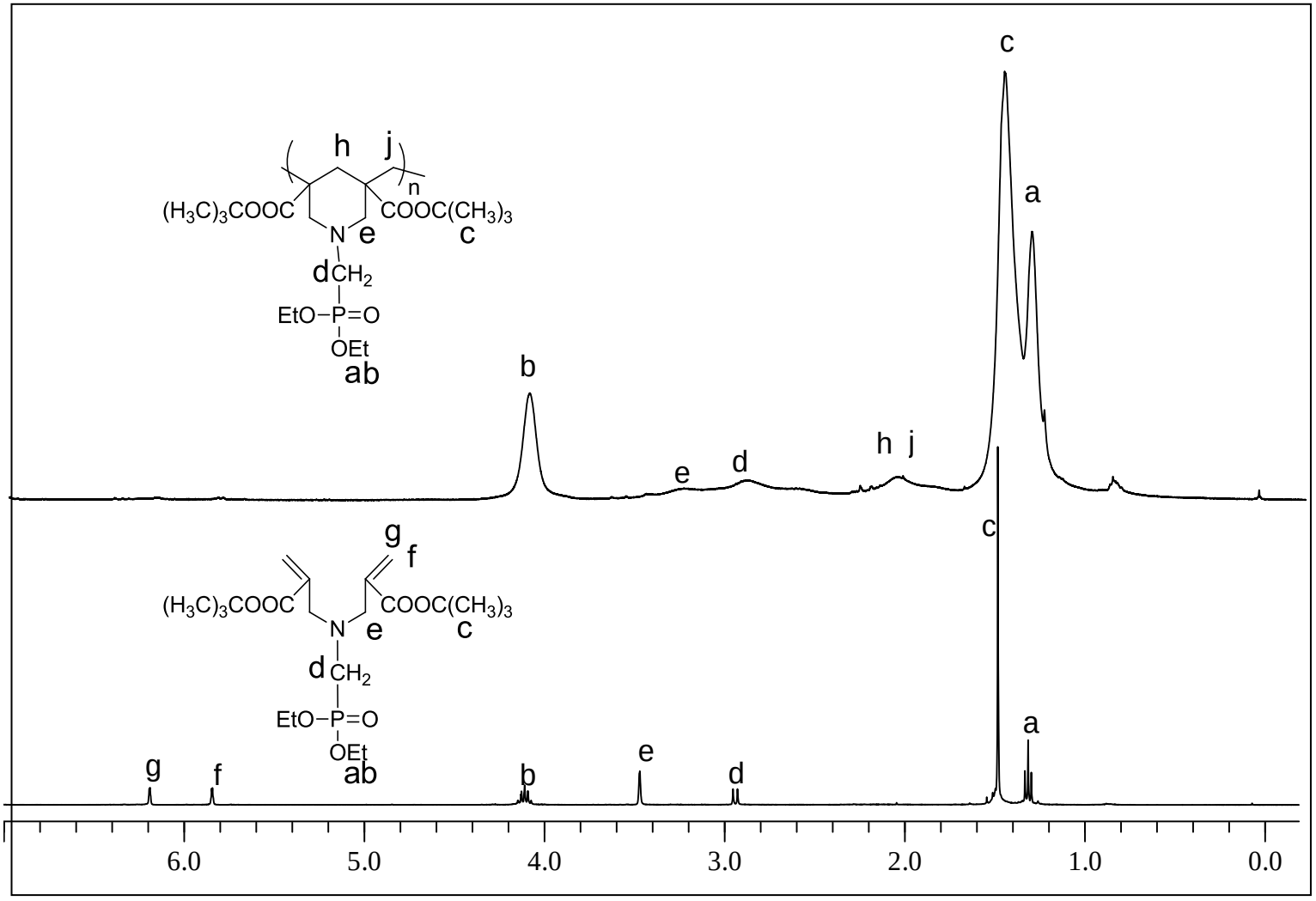


Figure 5.5. ^{1}H NMR spectra of **C2** and poly-**C2**.

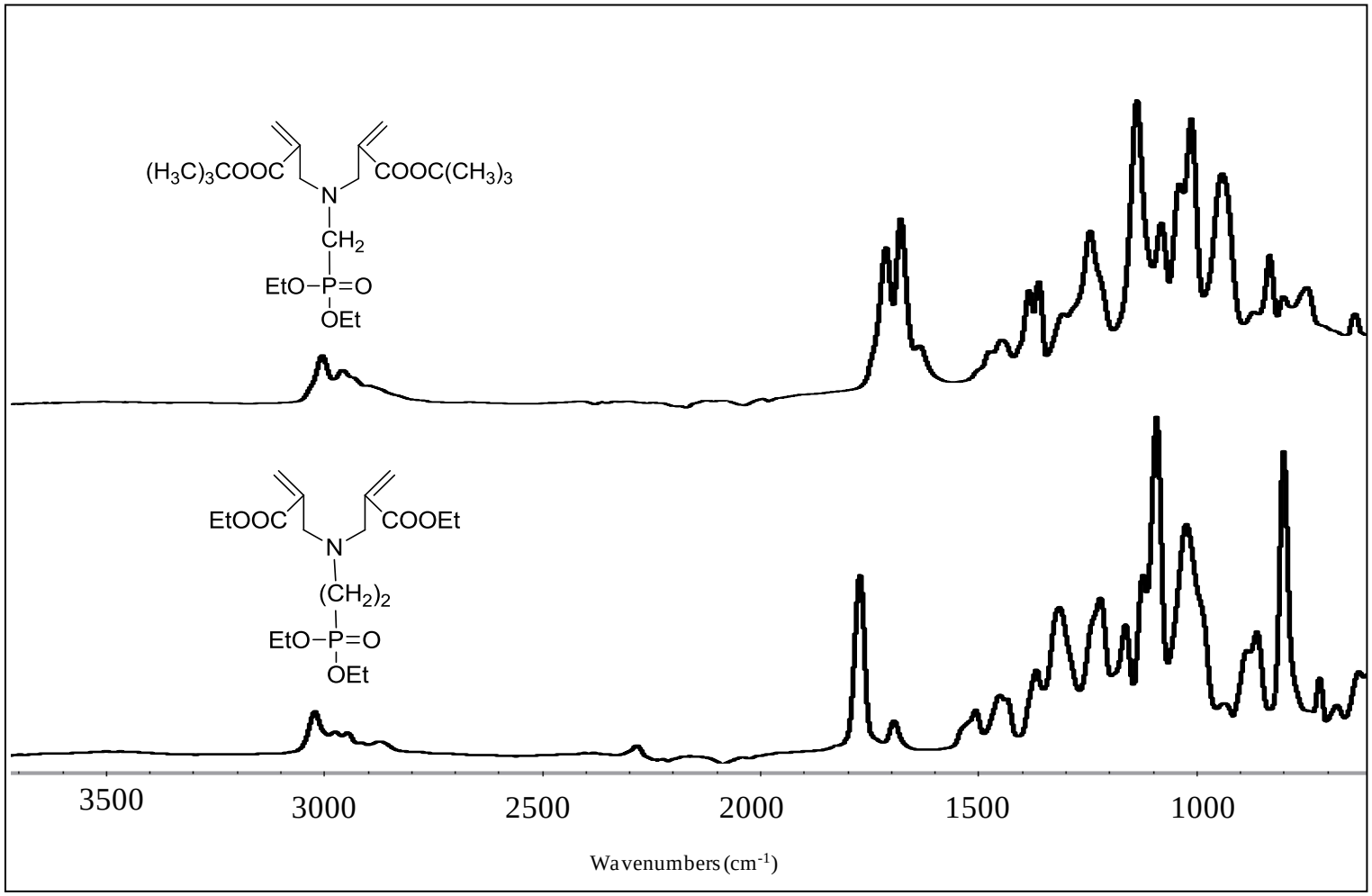


Figure 5.6. FTIR spectra of **C2** and **C3**.

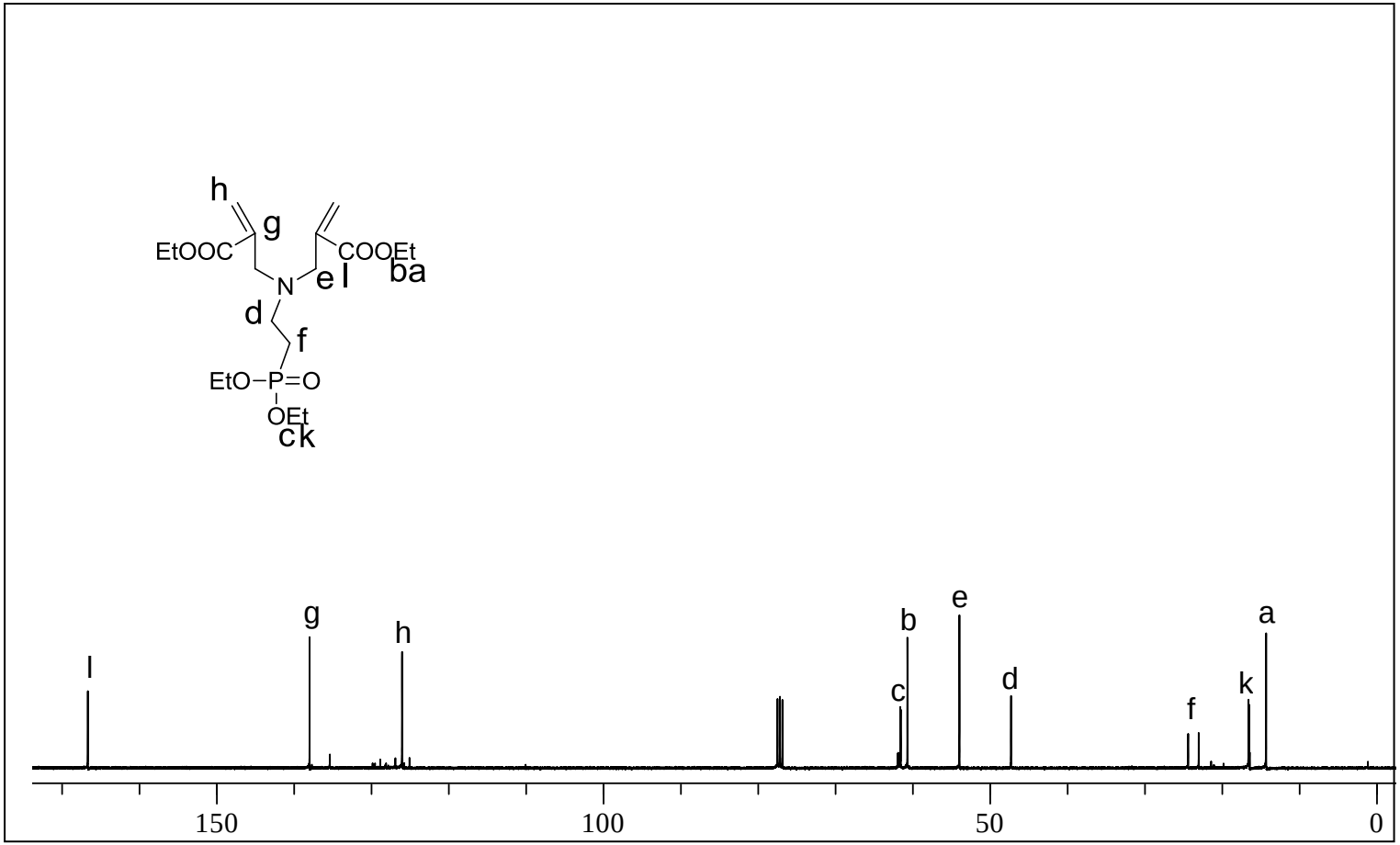


Figure 5.7. ^{13}C NMR spectrum of **C3**.

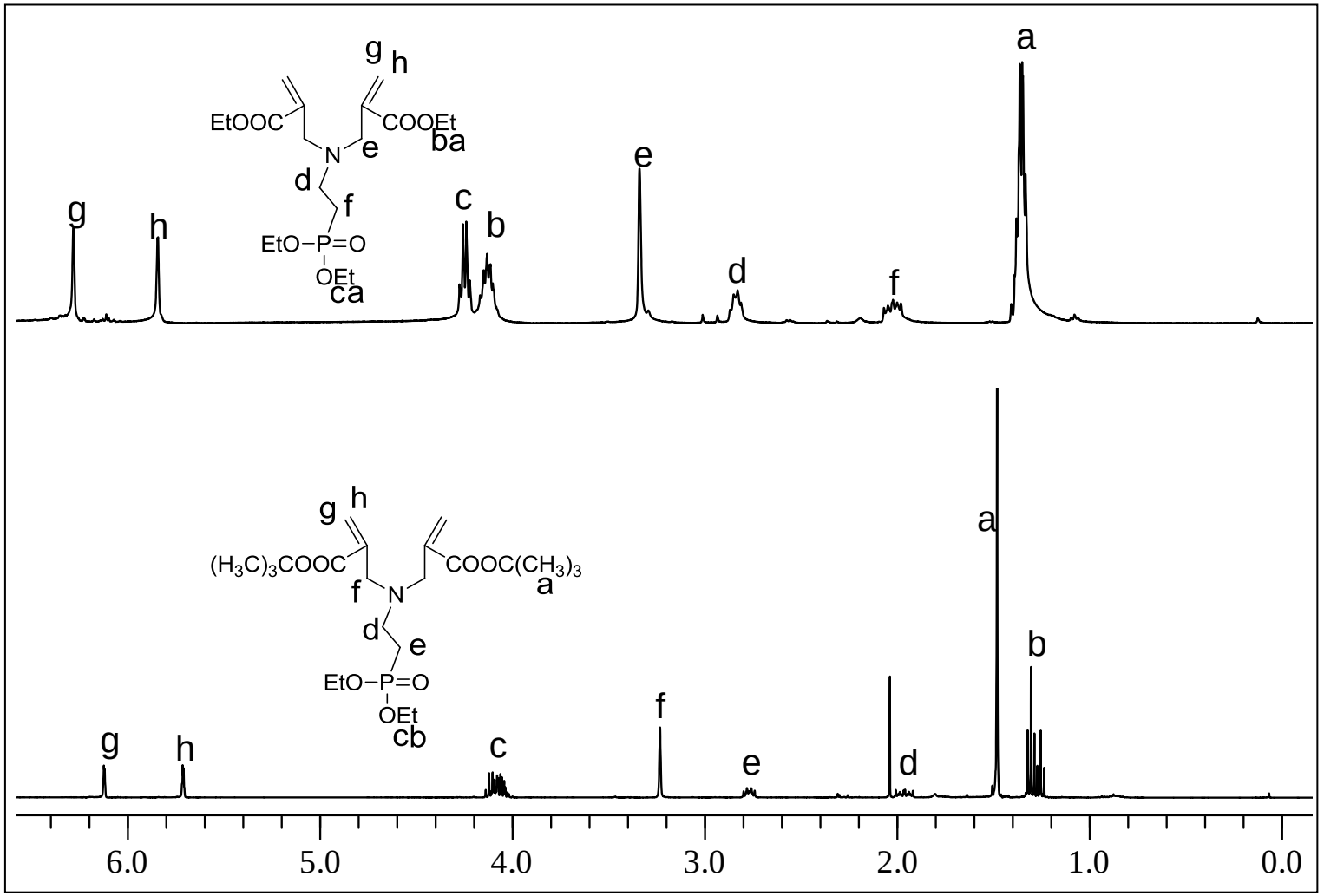


Figure 5.8. ^{1}H NMR spectra of **C3** and **C4**.

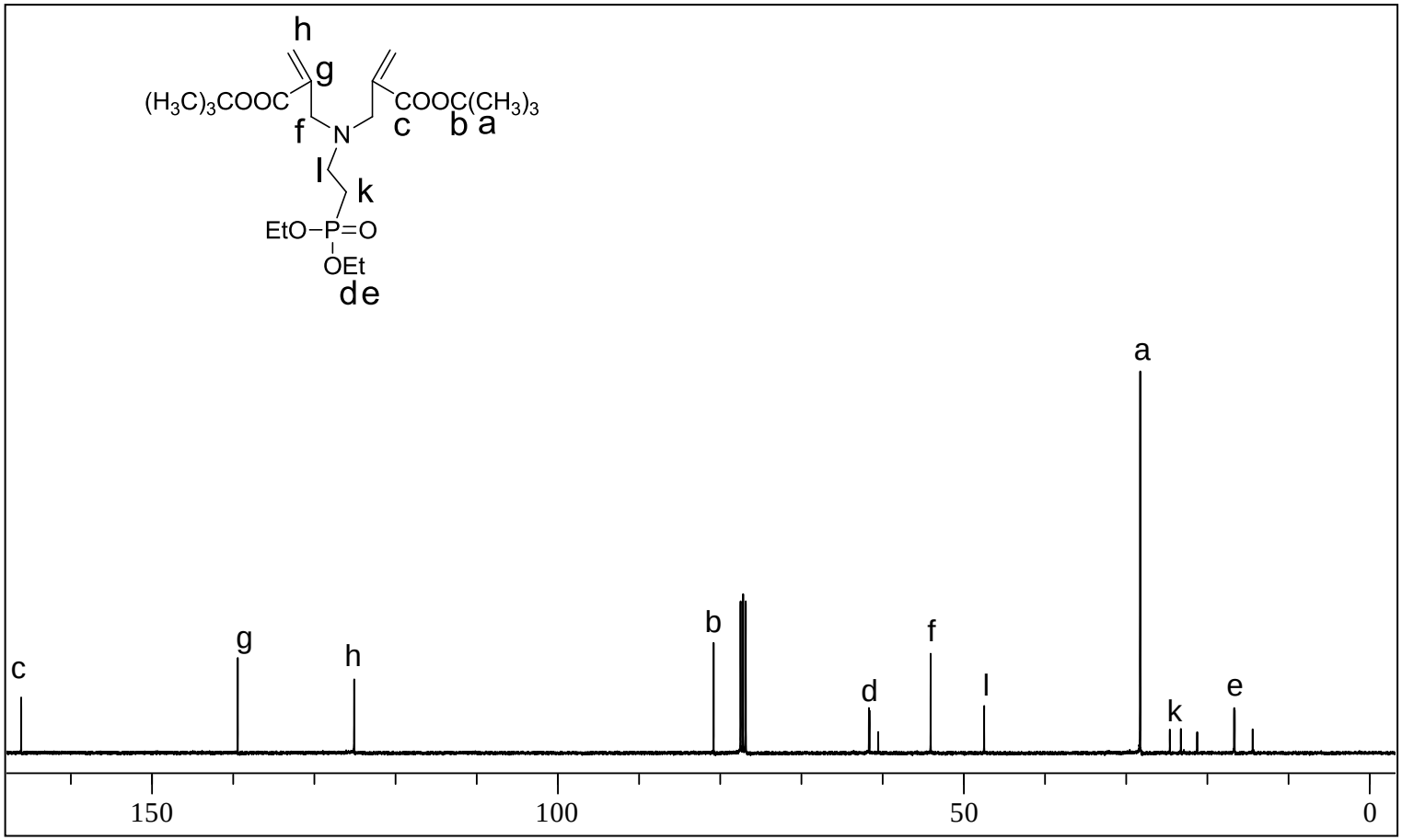


Figure 5.9. ^{13}C NMR spectrum of **C4**.

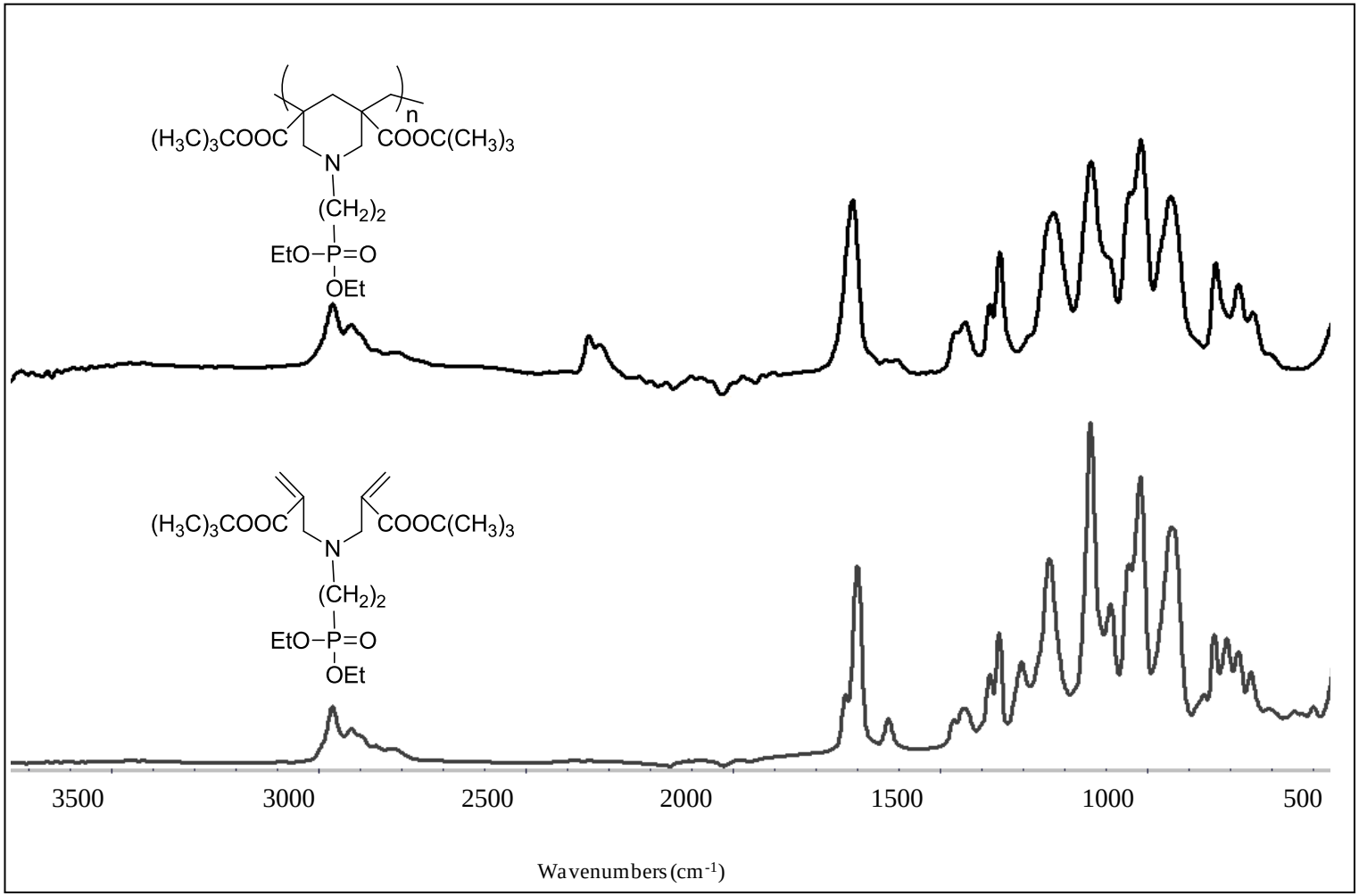


Figure 5.10. FTIR spectra of **C4** and poly-**C4**.

The second group of aminophosphonate-containing monomers (**C5** and **C6**) was synthesized according to the general route shown in Figure 5.11, by the Michael addition of diethyl aminomethylphosphonate and diethyl 2-aminoethylphosphonate to AHM which is a mixed acrylate/methacrylate monomer. AHM is a commercial monomer synthesized from reaction of glycidyl methacrylate and acrylic acid and contains a small amount of isomer due to the attack of acrylic acid to the more sterically hindered carbon of the epoxy ring of glycidyl methacrylate [179]. Michael addition of various amines to AHM and ethylene glycol acrylate methacrylate and ethylene glycol bisacrylate were reported by Mathias [179], Muh *et al*. [180] and us [181].

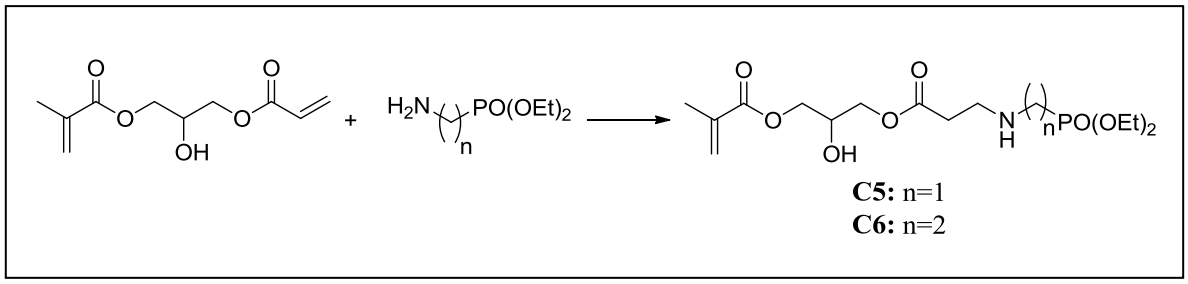


Figure 5.11. Synthesis of monomers **C5** and **C6**.

The reactions were carried out in bulk at room temperature using 1:1 ratio of AHM and amine to generate the secondary amine-based monomers (mono adducts). The disappearance of peaks of the acrylate double bonds at 5.8 and 6.4 ppm was monitored using ^{1}H NMR during the reactions (Figure 5.12). The small peaks around 3.7 and 5.2 ppm are due to the other isomer which also undergoes Michael addition reaction. The products were clear, colorless and very viscous liquids, soluble in common polar organic solvents and water. Synthesis of the bis adducts using 2:1 ratio of AHM to amine in bulk or in $CHCl_3$ at room temperature for 72 h was not successful. The unreactivity of monoadduct may be due to its steric hindrance or electron withdrawing effect of phosphonate groups.

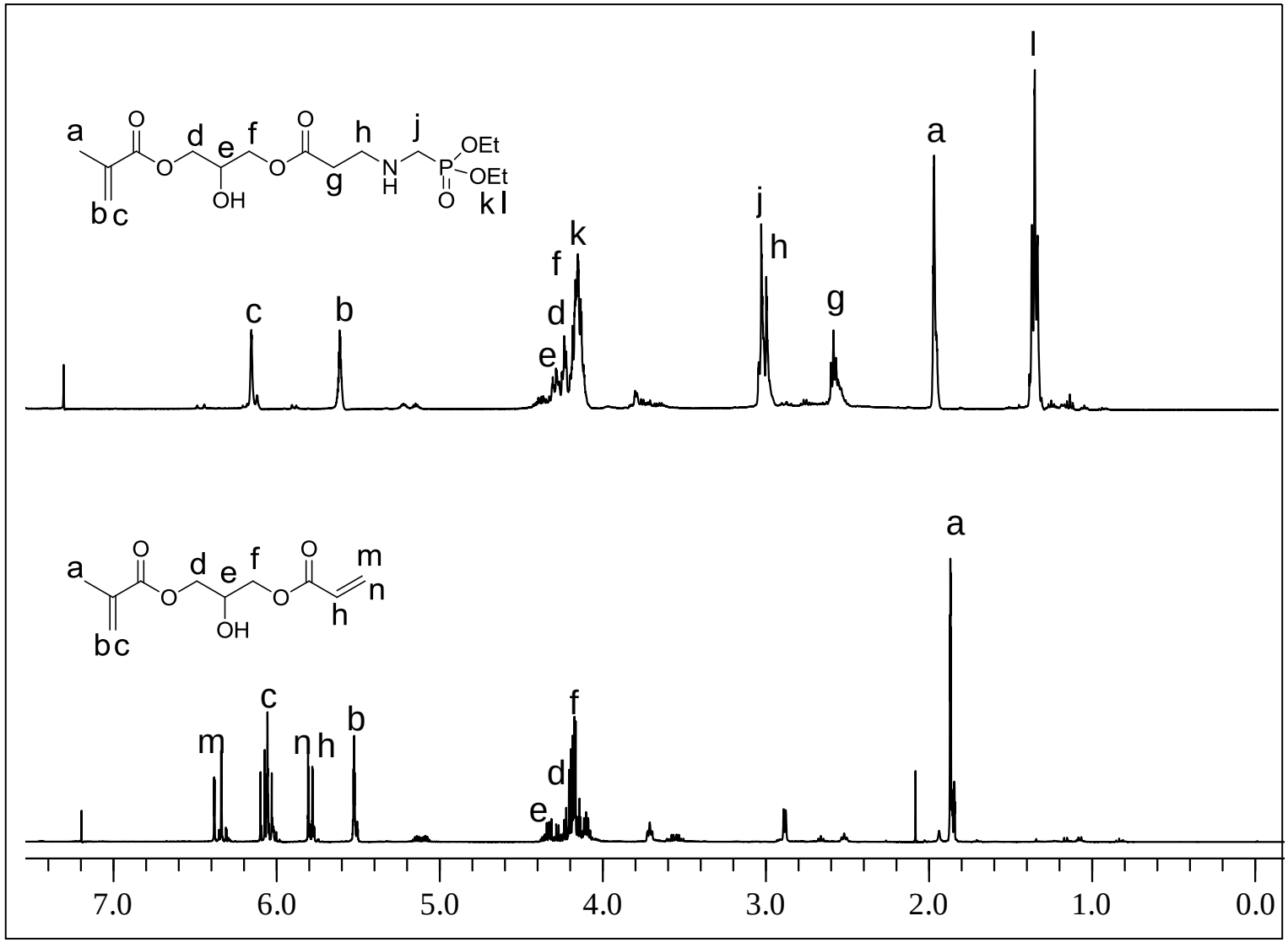


Figure 5.12. ^{1}H NMR spectra of AHM and **C5**.

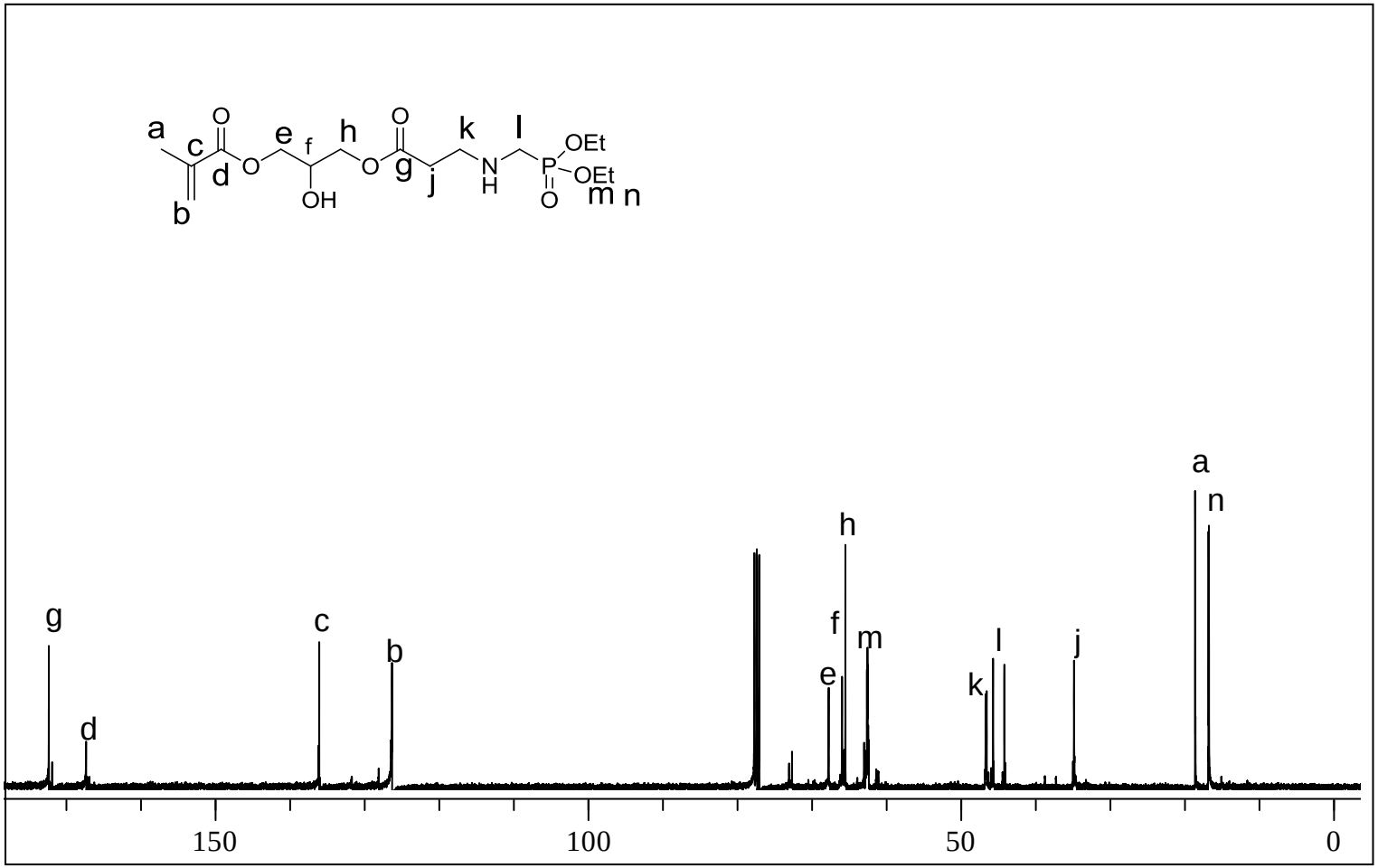


Figure 5.13. ^{13}C NMR spectrum of **C5**.

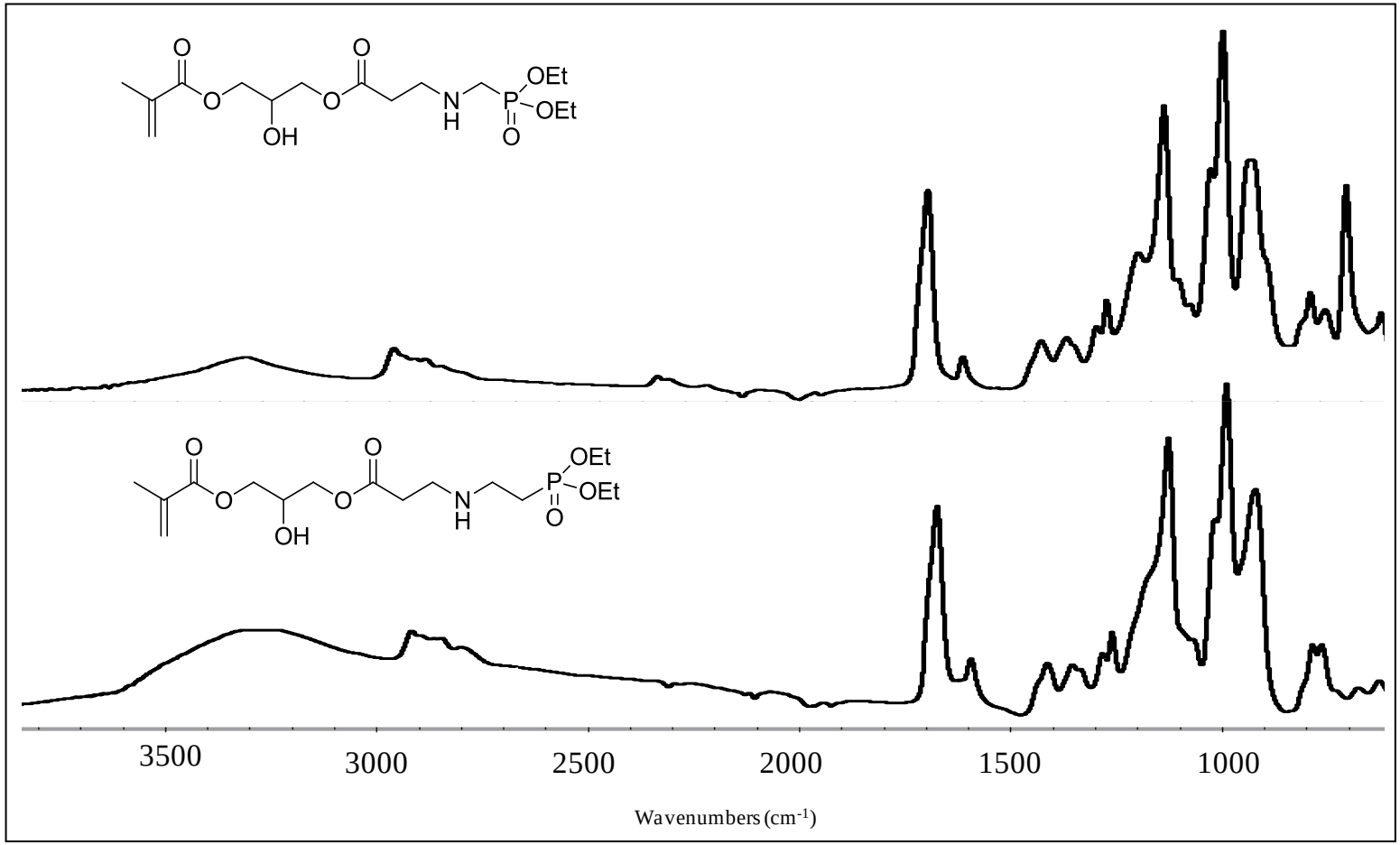


Figure 5.14. FTIR spectra of **C5** and **C6**.

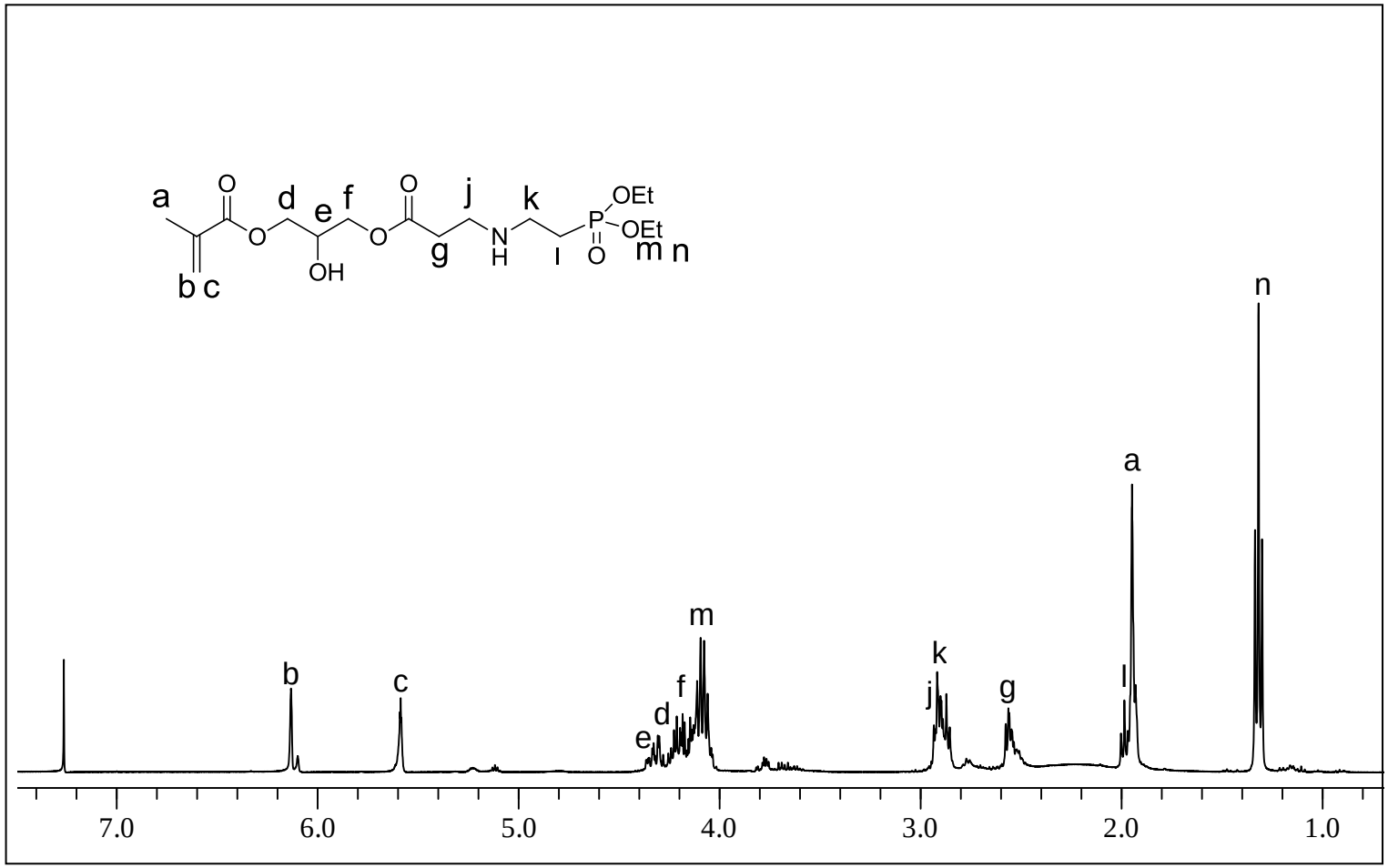


Figure 5.15. ^{1}H NMR spectrum of **C6**.

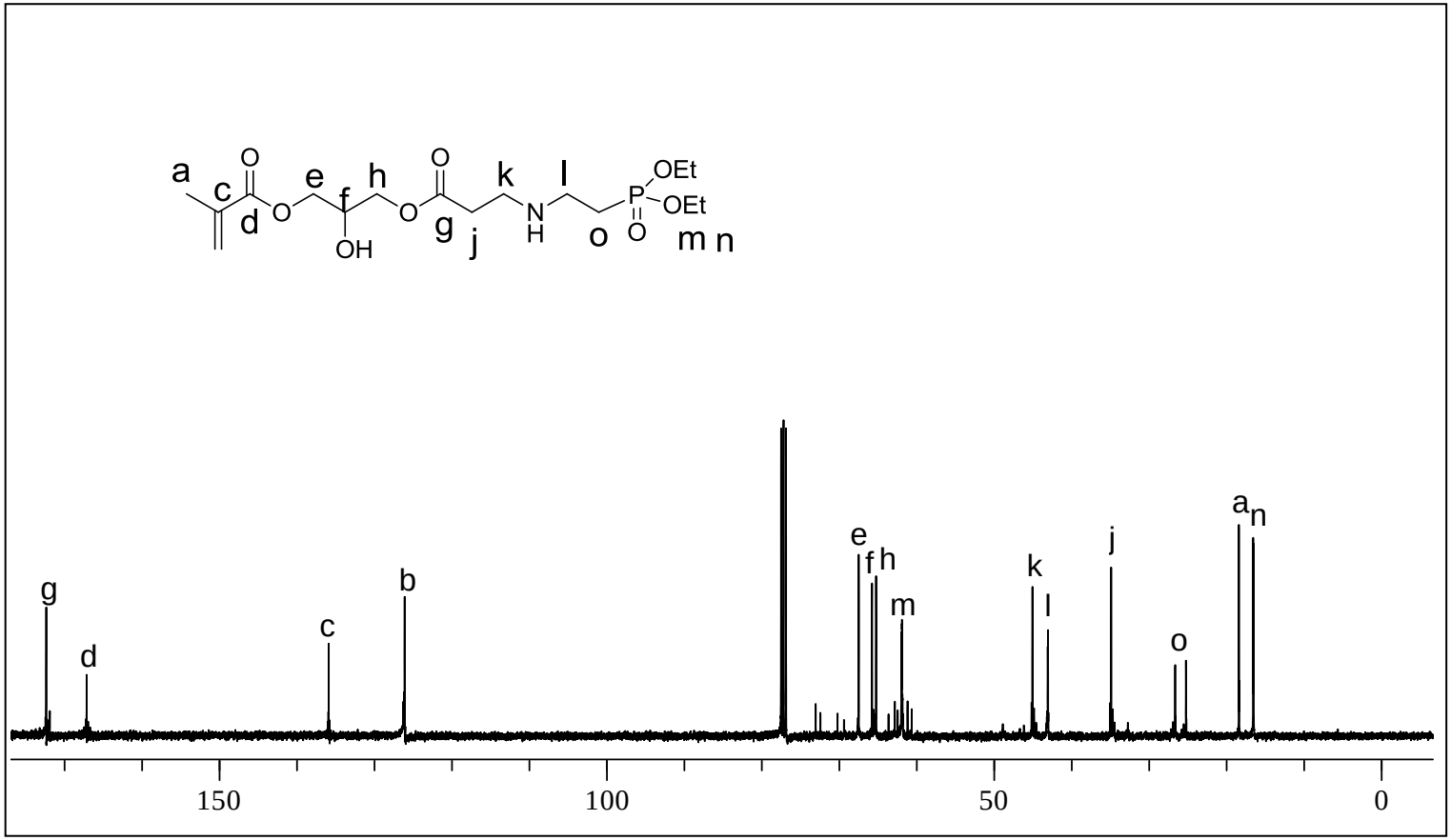


Figure 5.16. ^{13}C NMR spectrum of **C6**.

5.3.2. Synthesis of Polymers

It is known that the polymerization of 2,6-disubstituted 1,6-heptadienes give soluble polymers with cyclic units at the backbone under appropriate conditions [182]. Much attention has been given to the understanding of the factors enhancing cyclization efficiency to prevent pendent-group unsaturation which leads to crosslinking. The steric effects of the substituents were found to play an important role by inhibiting intermolecular addition much more than intramolecular cyclization. For example, ether dimers of *tert*-butyl and adamantyl α-hydroxymethacrylate showed very high cyclization efficiencies. The cyclopolymerization of some amine linked alkyl α-hydroxymethacrylates was investigated firstly by Choi *et al*. [183], who synthesized a methyl amine derivative from methyl α-chloromethacrylate and cyclopolymerized by group-transfer polymerization to give low molecular weight polymer (M_n=4.600-8.900). The aniline, adamantyl amine, *tert*-butyl amine, cyanamide, 4-tetradecylaniline and p-hydroxyaniline derivatives of both ethyl α-chloromethacrylate and *tert*-butyl α-chloromethacrylate were synthesized by us [184]. All monomers except cyanamide and p-hydroxyaniline derivatives displayed both high polymerization rate and cyclization tendencies to give polymers (M_n= 6.000-43.000).

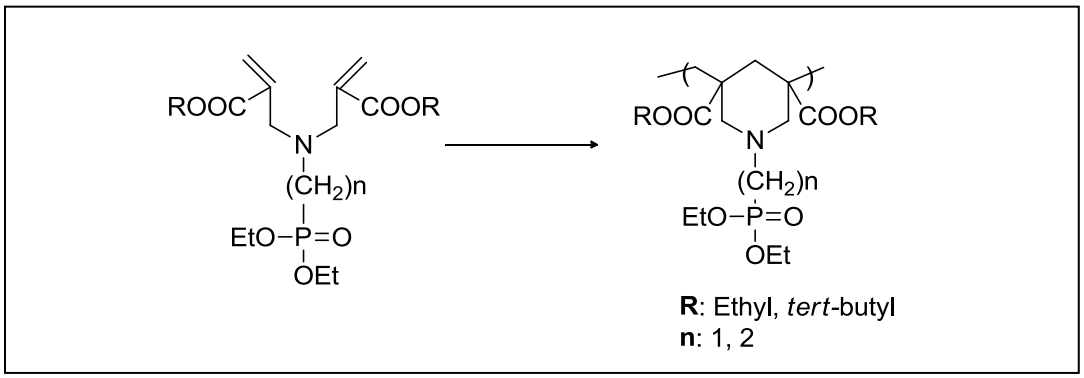


Figure 5.17. Polymerization of **C1-C4**.

The monomers **C1-C4** were bulk and solution polymerized thermally at 60-80 °C using AIBN as the radical initiator and standard freeze-evacuate-thaw procedures (Table 5.2). Monomers were very reactive and bulk polymerized in a very short time. Monomer **C1** was the most reactive monomer among the amine-linked bismethacrylates

and gave both soluble and crosslinked polymers depending on the polymerization conditions. Monomer **C1** even polymerized in the refrigerator without any initiator to give a completely soluble polymer with a M_n value of 53.000. Bulk polymerization of **C3** gave only soluble polymers with M_n value of 72.000. This indicates that increasing number of methylene groups increase cyclization efficiency (Figure 5.17 and Figure 5.18). Bulk polymerizations of **C2** and **C4** were slower than **C1** and **C3** and gave soluble polymers with very low molecular weights around 2.000 and 7.900. Increasing bulkiness of the ester substituent from ethyl to *tert*-butyl decreased the rate of polymerization, consistent with the lower molecular weights of poly-**C2** and poly-**C4**.

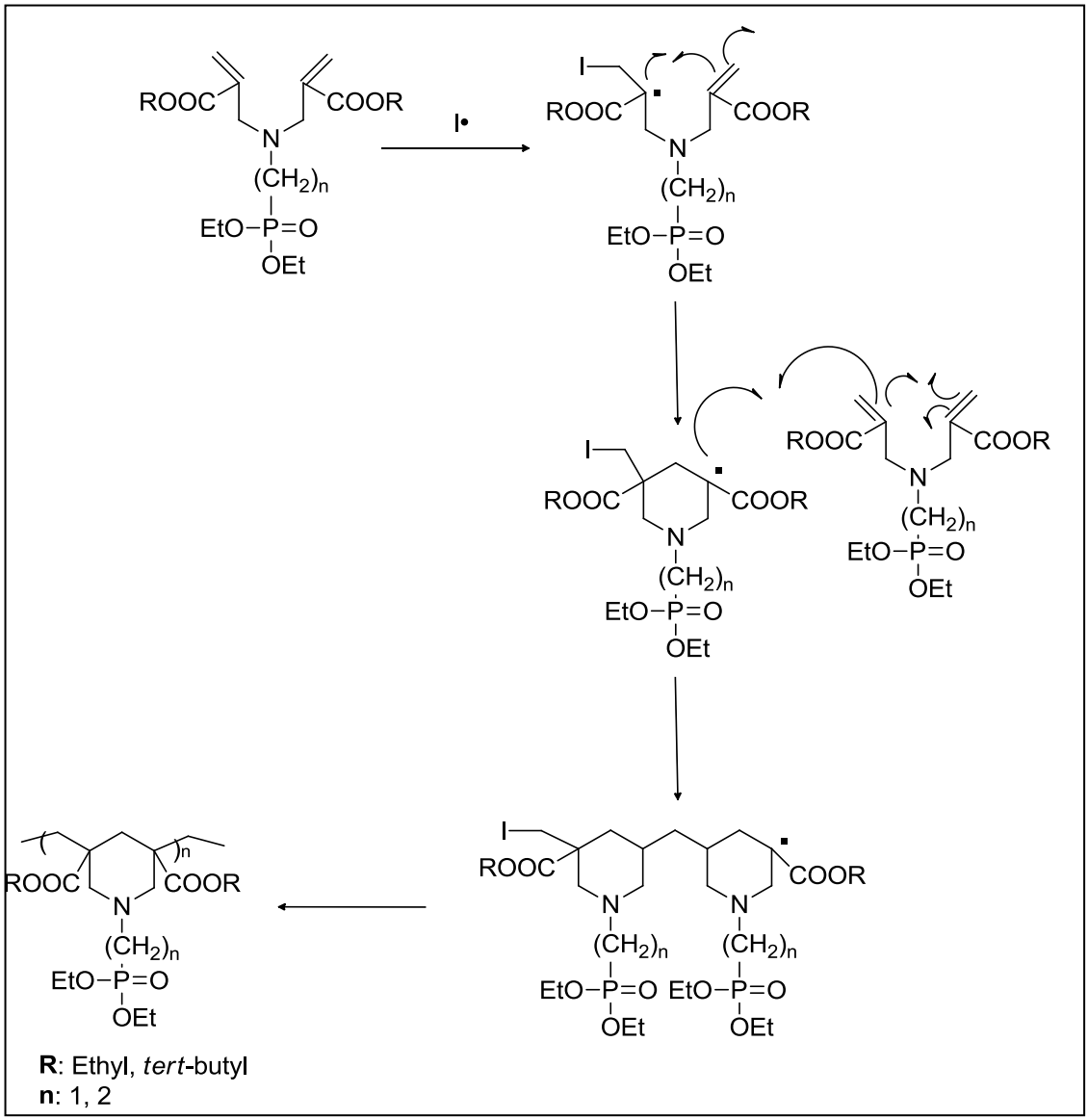


Figure 5.18. Cyclopolymerization mechanism of **C1-C4**.

Table 5.2. Thermal polymerization conditions, polymer characterization results and differences in chemical shift values (Δδ) of monomers.

Polymer	Temp (°C)	Solvent	[M]	Time (min)	Conversion (%)	M_n	Δδ (ppm)
C1	04	-	-	-	-	53.000	11.17
C1[a]	70	Bulk	-	40	crosslinked	-	-
C1[b]	70	Bulk	-	43	22	51.300	-
C1[c]	60	Acetone	3	56	crosslinked	-	-
C2[a]	70	Bulk	-	241	20	1.800	13.41
C2[c]	60	Acetone	3	48[d]	-	-	-
C2[b]	80	Bulk	-	240	20	2.900	-
C3[a]	70	Bulk	-	44	12	72.000	11.98
C3[c]	60	Acetone	3	65	15	5.800	-
C4[a]	70	Bulk	-	205	19	7.900	14.36
C4[c]	60	Acetone	3	30[d]	-	-	-
C5[a]	60	Bulk	-	25	crosslinked	-	9.77
C5[c]	60	Acetone	3	30	crosslinked	-	-
C6[a]	60	Bulk	-	35	crosslinked	-	9.81
C6[c]	60	Acetone	3	31	crosslinked	-	-

[a] [AIBN]: 1 mol %.

[b] [AIBN]: 0.5 mol %.

[c] [AIBN]: 0.024 M.

[d] Time (h).

The effect of vinyl substituents on polymerizability has been related to chemical shift differences of vinyl carbons of typical monomers. The stronger the electron-donating power of the substituents and/or the more bulky the substituents, the larger the difference and the lower the radical polymerizability [185, 186]. The chemical shift differences of the derivatives synthesized are shown in Table 5.2. The smaller chemical shift difference of **C1** and **C3** proves the reactivity of these monomers giving high molecular weight polymers due to reduced chain transfer of hydrogens on the connecting CH_2 groups compared to **C2** and **C4**. The slightly higher chemical shift difference of **C3** from **C1** or **C4** from **C2** indicates that the larger steric effect of diethyl 2-aminoethylphosphonate compared to diethyl aminomethylphosphonate.

The polymers were soluble in a variety of organic solvents such as acetone, THF, ether, methylene chloride and methanol but insoluble in water and hexane. Because monomers **C1** and **C3** have the same solubility with their polymers, purification of the polymers was only possible after precipitation into mixture of methanol:water (1:1) twice. However, polymer **C2** and **C4** were easily purified by precipitation into hexane where their monomers were soluble. ^{1}H NMR spectrum of the soluble polymers showed no residual double bonds, indicating very high cyclization efficiencies (Figure 5.3, Figure 5.5 and Figure 5.10).

We also investigated photopolymerization of the synthesized monomers with photodifferential scanning calorimetry using Irgacure 651, to determine their relative rate of polymerizations and conversions. All the polymerizations were performed under identical conditions of temperature, initiator concentration and light intensity. Figure 5.19 and Table 5.3 show the time dependences of the polymerization rate and conversions for monomers **C1**-**C4**. Some important observations are: (i) Photopolymerization behaviors of the synthesized monomers are similar to each other. For example, monomers **C1**-**C4** show maximum rates of 0.022, 0.017, 0.023 and 0.019 s^{-1} after 22.8, 19.2, 17.4 and 19.2 s. This behavior is typical of monofunctional (meth)acrylates. (ii) Monomers with *tert*-butyl groups showed lower polymerization rates and conversions compared to those of ethyl groups. (iii) Degree of conversion of the synthesized monomers was very high (72-98 %). The degree of conversion depends on the polymerization temperature and the T_g of the system which indicates its mobility. DSC analysis of the polymers obtained from thermal

polymerizations showed glass transition temperatures of 95 °C for poly-**C2**, 100 °C for poly-**C4** and 50 °C for poly-**C3**. The T_g value of poly-**C1** was not obvious. These results indicated the effect of bulky *tert*-butyl groups increasing T_g of polymers and explains low conversion of *tert*-butyl ester monomers. It was also indicated that changing the number of methylene units between amine and phosphonate groups did not affect flexibility of the system and thus T_g values of the polymers.

Table 5.3. Rates and conversions of synthesized monomers and AHM in photopolymerization.

Monomers	**Max. Rate (s^{-1})**	**Conversion (%)**
C1	0.022	89
C2	0.017	82
C3	0.023	98
C4	0.019	72
C5	0.075	90
C6	0.056	80
AHM	0.060	71

Thermal bulk and solution polymerization of monomers **C5** and **C6** at 60 °C using AIBN gave crosslinked polymers (Table 5.2). The photopolymerization behaviors of these monomers were investigated using photo-DSC and compared with that of AHM (Figure 5.19b). It was clearly seen that: (i) the shape of the rate curves is similar to that of AHM, typical to multifunctional monomers where gel effect is observed at low conversions, (ii) monomers **C5** and **C6** polymerize at similar or higher rates than AHM, even though AHM has two double bonds. The maximum rates of polymerizations reached were 0.075 and 0.056 for monomers **C5** and **C6** (Table 5.3). The smaller chemical shift difference of **C5** compared to **C6** also confirms its reactivity. The conversions (90 and 80% for **C5** and **C6**) were higher than AHM (71 %). The reactivity of monomers **C5** and **C6** is conjectured to be due to the extra hydrogen abstraction and/or hydrogen bonding due to the NH group they have, in addition to the OH group they have in common with AHM. To check this conjecture, photopolymerization of these monomers was tried in the presence of

benzophenone (BP), which is known as hydrogen abstraction photoinitiator (Figure 5.19b). The significantly higher photopolymerization rate of monomers **C5** and **C6** with respect to AHM supports our conjecture. Additionally, the polymers obtained from thermal polymerizations of these monomers were insoluble indicating the importance of chain transfer.

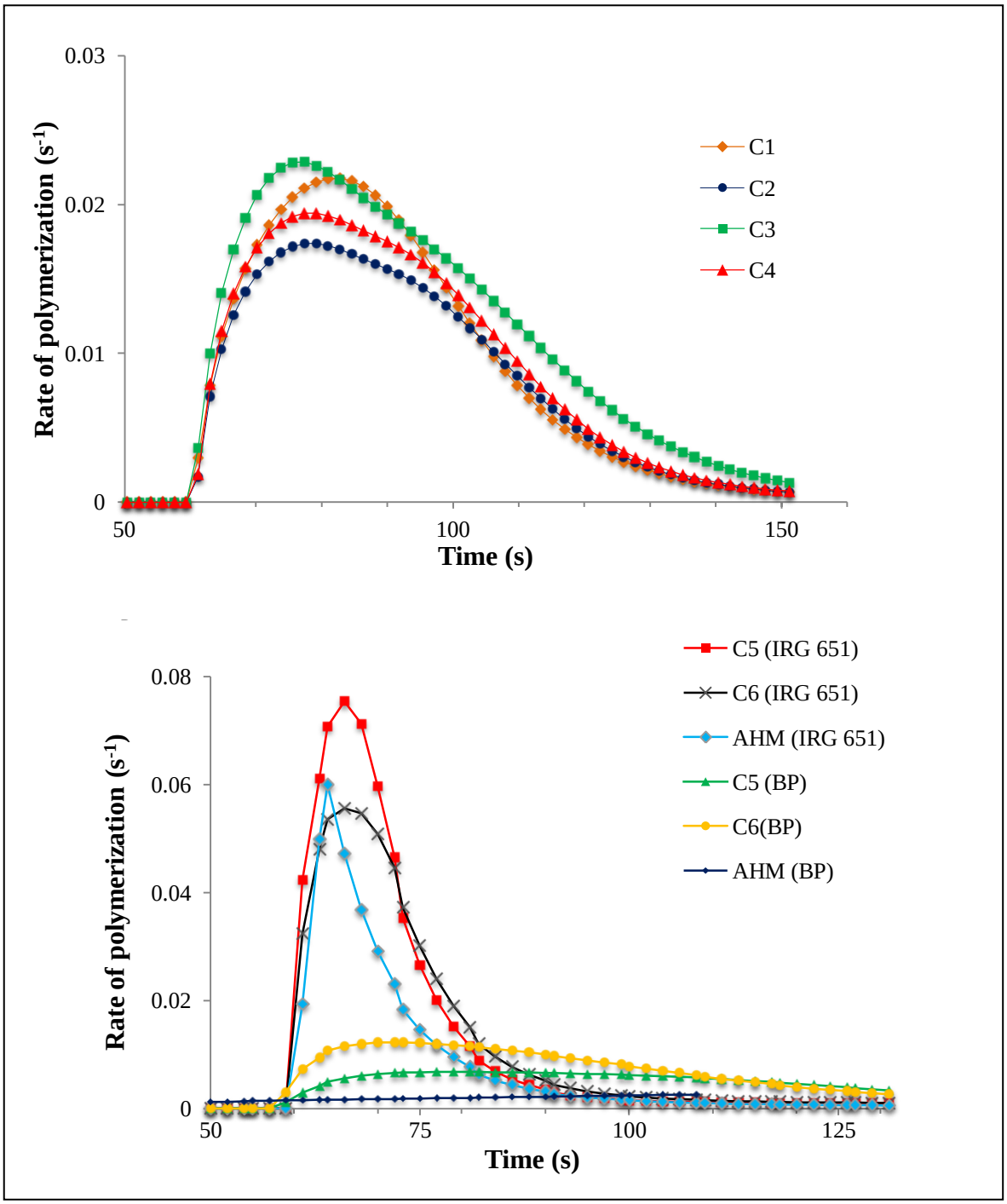


Figure 5.19. Photopolymerizations of monomers a) **C1-C4**; b) **C5, C6** and AHM.

Jansen *et al*. [187] investigated the rate of polymerization of different acrylates as a function of hydrogen bonding capability for systems containing amide, urethane and urea groups, and found that the monomers capable of forming hydrogen bonds show 3-6 times higher polymerization rates compared to their non-hydrogen bonding analogues. The high reactivities were suggested to be due to pre-organization via hydrogen bonding to bring the double bonds close to each other, enhancing propagation, although reduction in termination rate may also be involved or be the cause. It is known that hydrogen bonding decreases with increasing temperature, which tends to decrease the polymerization rate, whereas chain transfer increases with increasing temperature which tends to increase the polymerization rate. In order to investigate the effect of temperature change on polymerization rate, monomer **C5** was polymerized at 30, 40 and 70 °C. We found that rate of polymerizations remains almost constant with temperature. This can be explained by competing effects of activation which occurs by increasing temperature and deactivation which is due to decreased hydrogen bonding.

The thermal stabilities of the homopolymers were investigated by TGA under nitrogen at 10 °C/minute. Polymers obtained from AHM derivatives, **C5** and **C6,** showed the same thermal degradation behavior which takes place in a broad temperature range. The char yields of these polymers were found to be 26 % at 580 °C. The polymers containing *tert*-butyl group, poly-**C2** and poly-**C4**, also showed the TGA plots of the same shape. Both polymers started to lose weight around 200 °C due to decomposition of the *tert*-butyl ester group and then degraded gradually to give char yields of 31 % and 35 % for polymer **C2** and **C4**. The high char yields of these polymers are probably due to formation of carboxylic acids in addition to phosphonic acids which may contribute to crosslinking in the early stages of degradation. The polymers **C1** and **C3** with the ethyl ester group showed the highest thermal stability. The first step of degradation of polymer **C3** starts at significantly higher temperature (300 °C) compared to other polymers. Initial weight loss of polymer **C1** is probably due to residual solvent or monomer. The char yields of these polymers (20 % and 17 %) were lower compared to the other monomers (Figure 5.20).

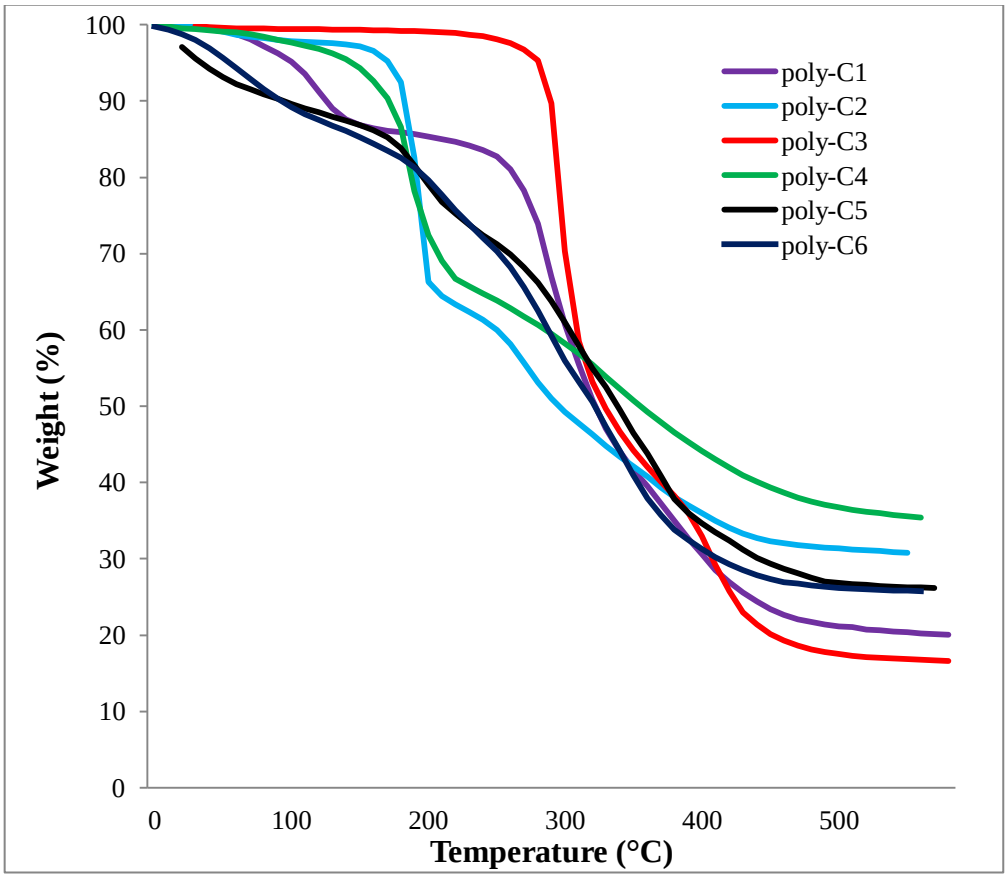


Figure 5.20. TGA curves of polymers.

5.4. Conclusion

In the first part of this work, four novel aminophosphonate-containing 1,6-heptadienes were synthesized by the reaction of diethyl aminomethylphosphonate or diethyl 2-aminoethylphosphonate and EBBr or TBBr. These monomers displayed high polymerization tendencies to give crosslinked or soluble polymers depending on monomer structures and polymerization conditions. The *tert*-butyl ester monomers showed lower polymerization rates compared to ethyl ester monomers. The correlation between the chemical shift differences of the double bond carbons and the reactivities of the monomers seems to hold. The studies on the hydrolysis of phosphonate and/or *tert*-butyl groups of these monomers to give new monomers with zwitterionic structure in aqueous solution within a pH range are continuing. Polymers that will be obtained from these monomers will have potential applications in water treatment, coatings, remediation and pharmaceutics.

In the second part of this work, two novel aminophosphonate-containing methacrylate monomers were synthesized via convenient Michael addition of diethyl aminomethylphosphonate or diethyl 2-aminoethylphosphonate to AHM. The high rates of polymerization of these monomers was attributed to both hydrogen bonding interactions due to additional NH groups as well as chain transfer reactions.

All of the homopolymers obtained produced char on burning indicating their potential as flame-retardant materials.

6. SYNTHESIS AND COPOLYMERIZATIONS OF ZWITTERIONIC MONOMERS

6.1. Introduction

Ion containing water soluble polymers have great importance based on their various applications [122]. These charged polymers can be classified as polyelectrolytes and polyampholytes. Polyelectrolytes contain either anionic or cationic functional groups [118]. Among cationic polyelectrolytes poly(diallyldimethylammonium chloride) (poly-DADMAC) is the most widely produced polymer [188-190]. It has been widely used in many application areas such as flocculent in water treatment, antimicrobial biocide, biocatalyst agents in medical, biological and food applications, antistatic agent and electro conductive coating [118, 191].

Polyelectrolytes which have both cationic and anionic repeat groups are called polyampholytes [118, 121, 192]. Zwitterionic polymers are also known as polymeric betaines have both charges on different atoms in a single repeat unit. Zwitterionic polymers have biomimetic character and thus used for designing non-biofouling materials and surfaces [193-195]. They are generally synthesized from zwitterionic monomers having a quarternary ammonium as the cationic moiety and a sulfonate (sulfobetaine) [119, 120], a carboxylate (carbo- or carboxybetaines) [119, 122, 123], a phosphonate / phosphinate (phosphobetaines) [119, 124, 196] or dicyanoethenolates [197] as anionic groups. These polymers have intra- and interchain ionic contacts resulting in an ionically cross-linked network structure which generally makes them insoluble in pure water. Therefore, in comparison to polyampholytes, polybetaines are hardly water soluble. However, the solubility is increased by addition of low molecular weight electrolytes (e.g. NaCl) which screen the net attractive electrostatic interaction between the polymer chains, allowing the zwitterionic polymers to dissolve [198, 199].

The alpha-aminomethylene phosphonate betaines are useful in a variety of applications including chelating agents for scale inhibition, soil anti-redeposition agents in detergents, as crystal modifiers [126, 127]. Both monomeric and polymeric

aminomethylphosphonic acids exhibit good metal complexation ability and used for treatment of wastewater, groundwater and seawater [112, 113, 126, 172-174, 200]. Besides their chelating abilities to metals, aminophosphonic acids which are structural analogues of amino acids exhibits biological activities such as inhibition of enzymes, antibacterial activity, neuroative compounds, anticancer drugs or pesticides [175].

In the previous part of the thesis, new monomers containing aminophosphonate groups were synthesized. In this part, hydrolysis of phosphonate and/or *tert*-butyl groups of these monomers to give new monomers with zwitterionic structure in aqueous solution was studied. A series of phosphobetaines, carboxybetaines or both type with one or two methylene units between the cationic and anionic moiety were synthesized. To best of our knowledge, the monomers having both carboxylic and phosphonic acid groups together with the nitrogen will represent the first examples of this class of ionic monomers and polymers. The effect of pH on the structure of the monomers and their copolymerization behavior with DADMAC were investigated.

6.2. Experimental

6.2.1. Materials and Apparatus

6.2.1.1. Materials. Ethyl α-bromomethacryate (EBBr), *tert*-butyl α-bromomethacryate (TBBr), diethyl aminomethylphosphonate and diethyl 2-aminoethylphosphonate were synthesized according to literature procedures [153, 154, 177, 178]. Trimethylsilyl bromide (TMSBr; Aldrich, Taufkirchen, Germany) was distilled before use. TFA, DADMAC (65 wt % aq), 2,2'-azobis(N,N-amidinopropane) dihydrochloride (V-50), solvents and starting materials were obtained from Aldrich and used as received.

6.2.1.2. Apparatus. The monomer characterization involved ^{1}H, ^{13}C and ^{31}P NMR spectroscopy (Varian Gemini 400 MHz) and Fourier transform infrared (FTIR) spectroscopy (T 380). The solution viscosities were meausered using a Cannon-Ubbelohde viscometer at 30 °C.

6.2.2. Synthesis of Monomers

6.2.2.1. (Bis(2-(ethoxycarbonyl)allyl)amino)methylphosphonic acid (**D1**). TMSBr (0.459 g, 3.0 mmol) was added dropwise to a solution of **C1** (0.395 g, 1.0 mmol) in 1 mL of dry CH_2Cl_2 in an ice bath under nitrogen. After refluxing the volatile components were removed under vacuum. Methanol (5 mL) was added and the mixture was stirred at room temperature overnight. After removal of the solvent, the crude product was washed with ether to give yellow solid 39 % yield.

^{1}H NMR (MeOD), δ = 1.42 (t, 6H, CH_3), 3.44 (m, 2H, CH_2), 4.04 (s, 4H, CH_2), 4.19 (m, 4H, O-CH_2), 6.31 (2H, CH_2=), 6.58 ppm (2H, CH_2=).

FTIR: 3500-2500 (OH), 2912 (CH), 1696 (C=O), 1630 (C=C), 1148 (P=O), 1031 cm^{-1} (P-O).

6.2.2.2.((Bis(2-(*tert*-butoxycarbonyl)allyl)amino)methyl)phosphonic acid (**D2**). **D2** was synthesized using the same procedure as for **D1** using **C2** instead of **C1** and product was obtained as yellow solid in 35% yield.

^{1}H NMR (MeOD), δ = 1.44 (s, 18H, CH_3), 3.40, 3.44 (d, 2H, CH_2-P), 4.19 (s, 4H, CH_2-N), 6.18 (2H, CH_2=), 6.49 ppm (2H, CH_2=).

^{13}C NMR (MeOD), δ = 28.31 (*C*H_3), 37.2, 37.65(CH_2-P), 58.65 (CH_2-N), 84.69 ((CH_3)$_3$*C*), 132.34 (CH_2=), 136.80 (C=), 165.17 ppm (C=O).

FTIR: 3500-2500 (OH), 2972 (CH), 1691 (C=O), 1630 (C=C), 1150 (P=O), 978 cm^{-1} (P-O).

6.2.2.3. (2-(Bis(2-(ethoxycarbonyl)allyl)amino)ethyl)phosphonic acid (**D3**). **D3** was synthesized using the same procedure as for **D1** using **C3** instead of **C1** and product was obtained as yellow solid in 35 % yield.

^{1}H NMR (MeOD), δ = 1.29 (t, 6H, CH_3), 2.07 (m, 2H, CH_2-P), 3.35 (m, 2H, CH_2-N), 4.04 (s, 4H, CH_2), 4.25 (m, 4H, O-CH_2), 6.27 (2H, CH_2=), 6.63 ppm (2H, CH_2=).

^{13}C NMR (MeOD), δ = 14.16 (*C*H_3CH_2O), 22.61, 23.97 (CH_2-P), 51.42 (CH_2-N), 55.28 (*C*H_2C=), 62.88 (CH_3*C*$H_2$0), 131.78 (CH_2=), 136.09 (C=), 166.40 ppm (C=O).

^{31}P NMR (MeOD), δ = 17.99 ppm.

FTIR: 3500-2500 (OH), 2983 (CH), 1717 (C=O), 1636 (C=C), 1222 (P=O), 1020 (P-O) cm^{-1}.

6.2.2.4. (2-(Bis(2-(*tert*-butoxycarbonyl)allyl)amino)ethyl)phosphonic acid (**D4**). Monomer **D4** was synthesized using the same procedure as for **D1** using **C4** instead of **C1** and product was obtained as yellow-brown solid in 30 % yield.

^{1}H NMR (MeOD), δ = 1.55 (s, 18H, CH_3), 2.33 (m, 2H, CH_2-P), 3.47 (m, 2H, CH_2-N), 4.09 (s, 4H, CH_2), 6.26 (2H, CH_2=), 6.59 ppm (2H, CH_2=).

6.2.2.5. 2,2'-((((diethoxyphosphoryl)methyl)azanediyl)bis(methylene))diacrylic acid (**D5**). CF_3COOH (0.44 g, 3.91 mmol) was added to **C2** (0.25 g, 0.56 mmol) dropwise in an ice-bath under nitrogen and the solution was stirred at room temperature overnight. After evaporation of CF_3COOH, the product was washed with diethyl ether and dried under vacuum to give the pure product as white solid in 43 % yield.

^{1}H NMR (MeOD), δ = 1.39 (t, 6H, CH_3), 3.84 (m, 2H, CH_2), 4.29 (s, 4H, CH_2), 4.30 (m, 4H, O-CH_2), 6.34 (2H, CH_2=), 6.76 ppm (2H, CH_2=).

^{13}C NMR (MeOD), δ = 16.77 (*C*H_3CH_2O), 46.15, 47.67 (CH_2-P), 59.91 (*C*H_2C=), 66.81 (CH_3*C*H_2O), 130.99 (CH_2=), 138.46 (C=), 169.95 ppm (C=O).

FTIR: 3500-2500 (OH), 2984 (CH), 1688, 1668(C=O), 1630 (C=C), 1252 (P=O), 1014 and 967 cm^{-1} (P-OEt).

6.2.2.6. 2,2'-(((2-(diethoxyphosphoryl)ethyl)azanediyl)bis(methylene))diacrylic acid (**D6**). **D6** was synthesized using the same procedure as for **D5** using **C2** instead of **C1** and the pure product was obtained as white crystals in 33 % yield (mp. 117-120 °C).

^{1}H NMR (MeOD), δ = 1.05 (t, 6H, CH_3), 2.20 (m, 2H, CH_2), 3.15 (m, 2H, CH_2), 3.84 (s, 4H, CH_2-C=), 3.87 (q, 4H, O-CH_2), 6.02 (2H, CH_2=), 6.40 ppm (2H, CH_2=).

^{13}C NMR (MeOD), δ = 16.83 (*C*H_3CH_2O), 21.01, 22.42 (CH_2-P), 30.90 (CH_2-N), 56.66 (*C*H_2C=), 64.73 (CH_3*C*H_2O), 132.51 (CH_2=), 137.96 (C=), 169.04 ppm (C=O).

FTIR: 3500-2500 (OH), 2988 (CH), 1688, 1668 (C=O), 1630 (C=C), 1251 (P=O), 1015 and 968 cm^{-1} (P-O-Et).

6.2.2.7. 2,2'-(((phosphonomethyl)azanediyl)bis(methylene))diacrylic acid (**D7**). CF_3COOH (0.35 g, 3.1 mmol) was added to **D2** (0.17 g, 0.4 mmol) dropwise in an ice-bath under nitrogen and stirred at room temperature overnight. Then unreacted CF_3COOH was evaporated and the product obtained as yellow-brown crystals in 25 % yield.

^{1}H NMR (MeOD), δ = 3.34, 3.38 (d, 2H, CH_2), 4.24 (s, 4H, CH_2-C=), 6.30 (2H, CH_2=), 6.70 ppm (2H, CH_2=).

FTIR: 3500-2500 (OH), 2960 (CH), 1689(C=O), 1629 (C=C), 1256 (P=O), 1014 cm^{-1} (P-O).

6.2.2.8. 2,2'-(((2-phosphonoethyl)azanediyl)bis(methylene))diacrylic acid (**D8**). Monomer **D8** was synthesized using the same procedure as for **D7** using **D4** instead of **D2**. The product obtained as yellow solid in around 20 % yield.

^{1}H NMR (MeOD), δ = 1.99 (m, 2H, CH_2-P), 3.12 (m, 2H, CH_2), 3.74 (s, 4H, CH_2-C=), 5.96 (2H, CH_2=), 6.34ppm (2H, CH_2=).

^{13}C NMR (MeOD), δ = 22.56, 23.93 (CH_2-P), 49.72 (CH_2-N), 56.26 (CH_2C=), 131.94 (CH_2=), 136.55 (C=), 168.55 ppm (C=O).

FTIR: 3500-2500 (OH), 2926 (CH), 1692(C=O), 1630 (C=C), 1144(P=O), 976 cm^{-1} (P-O).

6.2.3. Viscosity Studies

Viscosity measurements were performed with Cannon-Ubbelohde Viscometer at 30 °C. All solutions were made at around 0.2 -0.3 g/dL in deionized water or in 0.1 N NaCl solutions and diluted incrementally.

6.2.4. Thermal Polymerizations

All thermal polymerizations were run in septum-sealed glass tubes that were subjected to freeze-thaw procedures and placed in 70-80 °C oil baths. The copolymerizations of monomers **(D2, D3, D5, D8)** with DADMAC in two different ratios were carried out (monomer: DADMAC, 5:95 and 20:80 mol%) in water ([M]= 2.7 M) in the presence of V-50 ($5x10^{-2}$ $molL^{-1}$). The copolymers were precipitated into acetone and dried under vacuum. Purification of the copolymers was performed by dissolution in water and dialysis against water with tubing with a molecular cutoff of 6000–8000. The polymer was recovered by freeze drying.

6.3. Results and Discussion

6.3.1. Monomer Synthesis and Characterization

The hydrolysis of the phosphonate and /or *tert*-butyl groups of monomers **C1-C4** was carried out to give new monomers with zwitterionic structure, **D1-D8** (Figure 6.1).

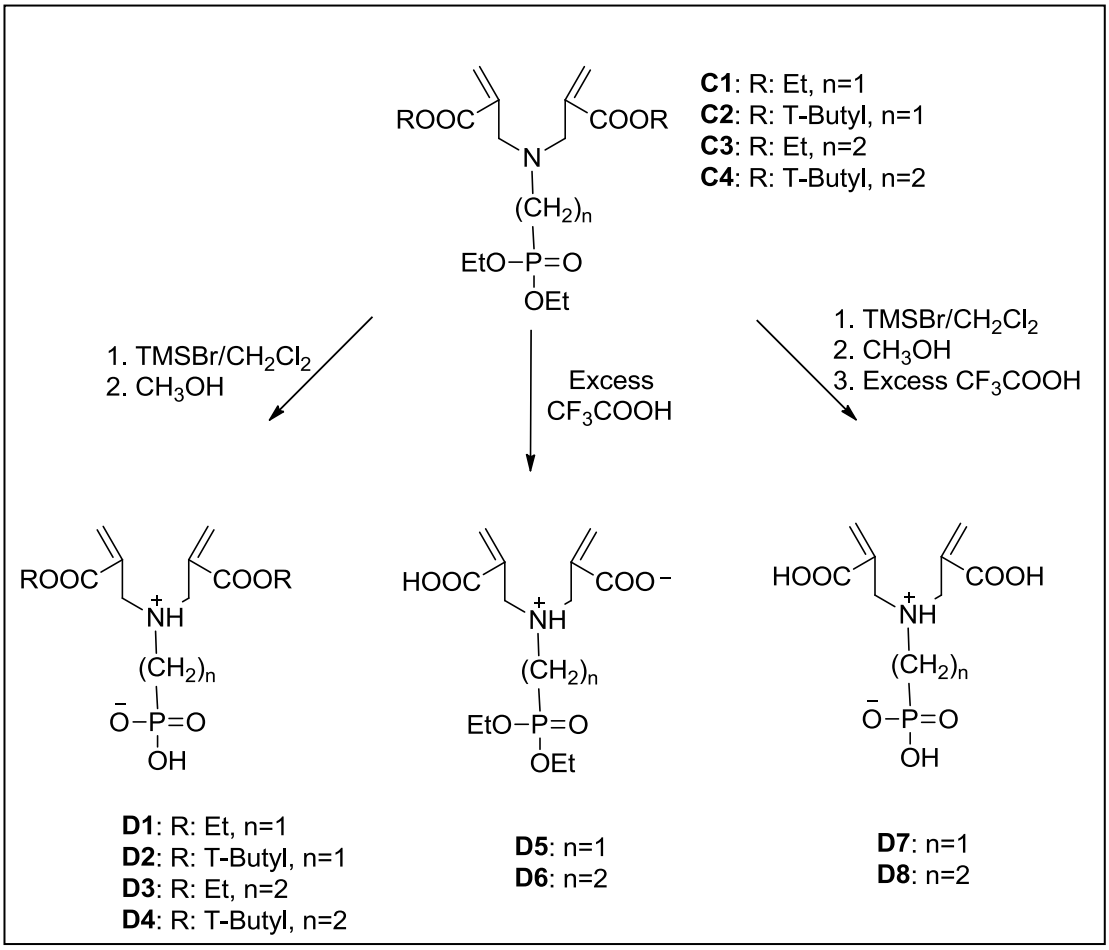


Figure 6.1. Synthesis of acidic monomers (**D1-D8)**.

The synthesized monomers can be divided into three groups. The monomers in the first group have phosphobetaine structures (**D1-D4**) and were synthesized by silylation of monomers **C1-C4** by TMSBr followed by methanolysis of the silyl ester derivatives. These water soluble monomers were obtained in 30-39 % yield as yellow solids after several washings with diethyl ether. The structures of the monomers were confirmed by ^{1}H, ^{13}C, ^{31}P NMR and FTIR spectroscopy. In the ^{13}C NMR spectrum of **D2**, the complete hydrolysis is supported by the absence of ethyl peaks of the phosphonate ester in **C2** around 16.32 and 61.68 ppm (Figure 6.2). Other characteristic peaks of this monomer were *tert*-butyl carbons at 28.31 and 84.69 ppm, and double bond carbons at 136.80 and 165.17 ppm. In the ^{1}H NMR spectra of **D3** and **D4**, methylene protons adjacent to phosphorus gave a multiplet at 2.07-2.33 ppm. The single peak of the phosphorus atom at 17.99 ppm in the ^{31}P NMR spectrum indicates purity of this monomer (Figure 6.3).

In the FTIR spectra, the monomers showed a broad absorption band between 2500 and 3500 cm^{-1} which corresponds to O-H and N-H stretching of acid and quaternary

ammonium groups (Figure 6.4). Other peaks observed at 1710, 1635 and 1250 cm^{-1} are due to C=O, C=C and P=O bonds.

The monomers in the second group (**D5** and **D6**) have carboxybetaine structure and were synthesized by the hydrolysis of the *tert*-butyl groups of monomers **C2** and **C4** with excess TFA. Monomers were solids and purified by several washings with diethylether. In the ^{13}C NMR spectrum of **D5**, the hydrolysis was confirmed by the absence of *tert*-butyl peaks of **C2** at 27.90 and 80.53 ppm. The doublet seen at 35.27 and 35.65 ppm is due to methylene carbon attached to phosphorus. In the ^{1}H NMR of **D6**, the protons attached to phosphorus and nitrogen show multiplets at 2.20 and 3.84 ppm, respectively. In the FTIR spectra of these monomers, the carbonyl peak showed splitting which is due to deprotonation of one of the carboxylic acid resulted in COO^-. For example, monomer **D3** showed different two peaks related to C=O stretchings of COOH and COO^- at 1687 and 1668 cm^{-1}, respectively (Figure 6.4). This situation proves the carboxybetaine structure of the monomers.

The monomers in the third group (**D7** and **D8**) were synthesized by further hydrolysis of monomers **D2** and **D4** with TFA. The hydrolysis was proved by the absence of *tert*-butyl peaks at 1.44 ppm in their ^{1}H NMR (Figure 6.5). In the ^{13}C NMR spectrum of **D8**, the methylene carbon adjacent to phosphorus showed a doublet at 22.56 and 23.93 ppm. Although these monomers (**D7** and **D8**) contained -COOH groups similar to **D5** and **D6**, different from the latter's they showed single carbonyl peak which is an evidence for deprotonation of phosphonic acid so the phosphobetaine structure was proved.

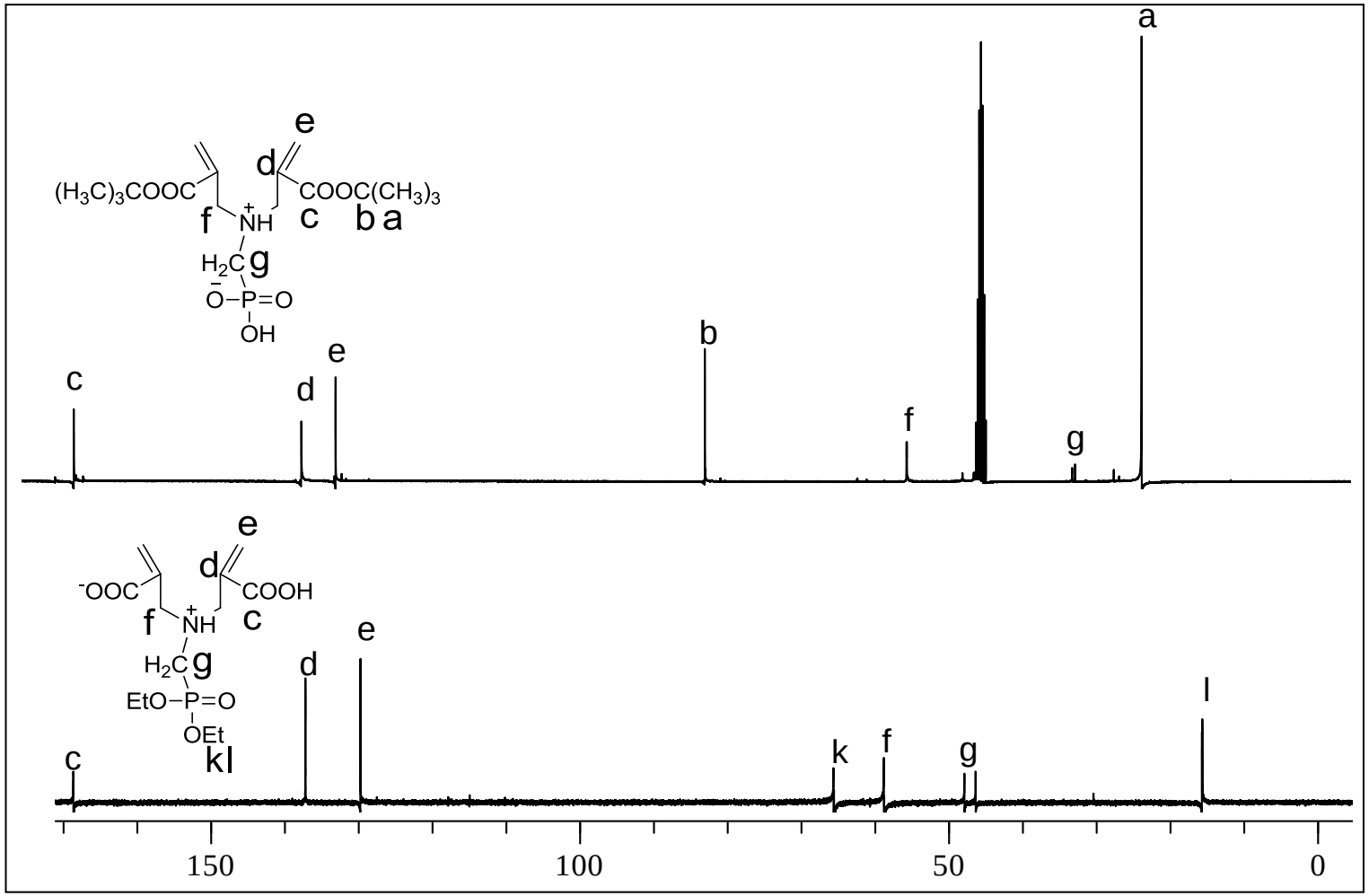


Figure 6.2. ^{13}C NMR spectra of **D2** and **D5**.

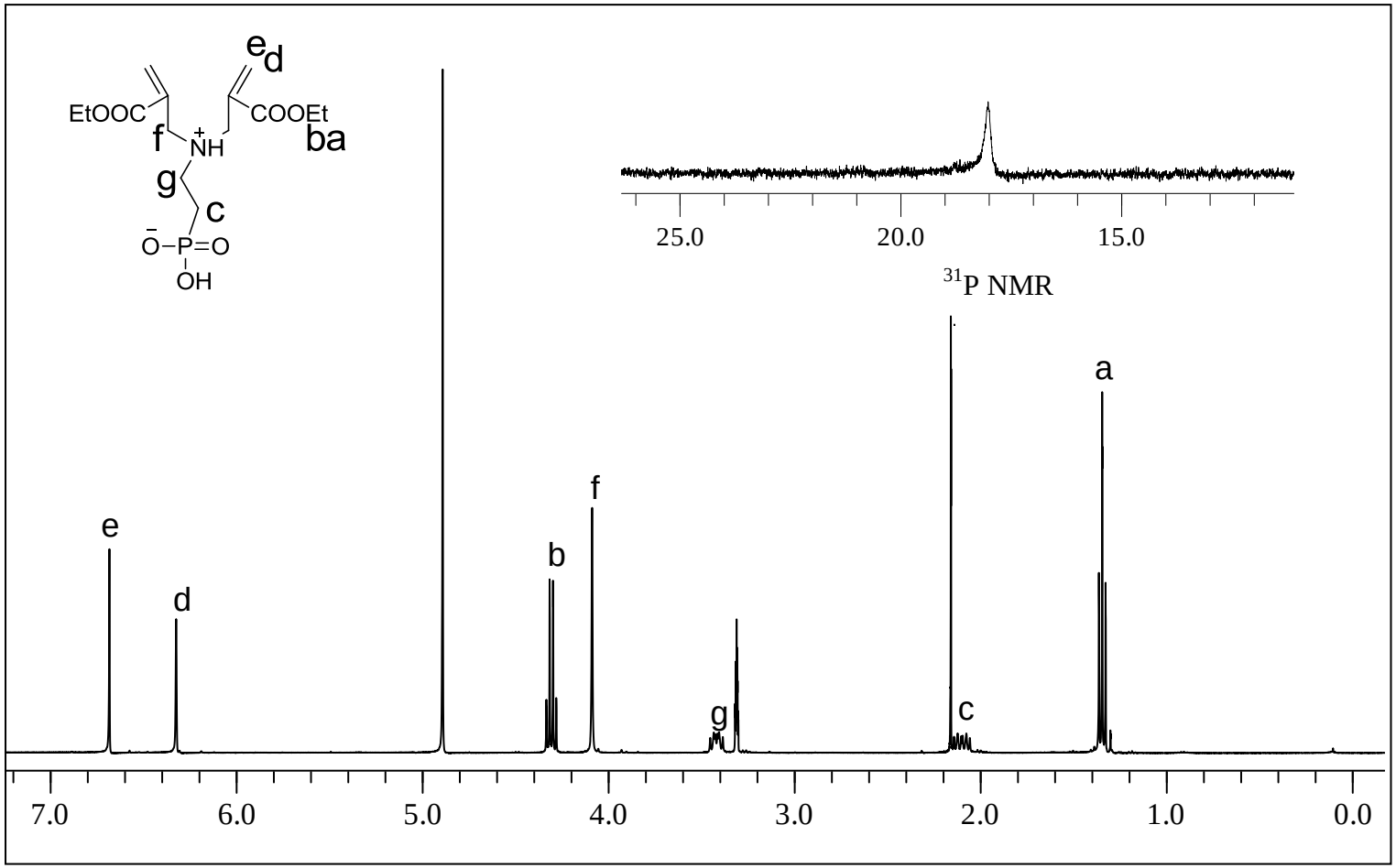


Figure 6.3. ^{1}H NMR and ^{31}P NMR spectraof **D3**.

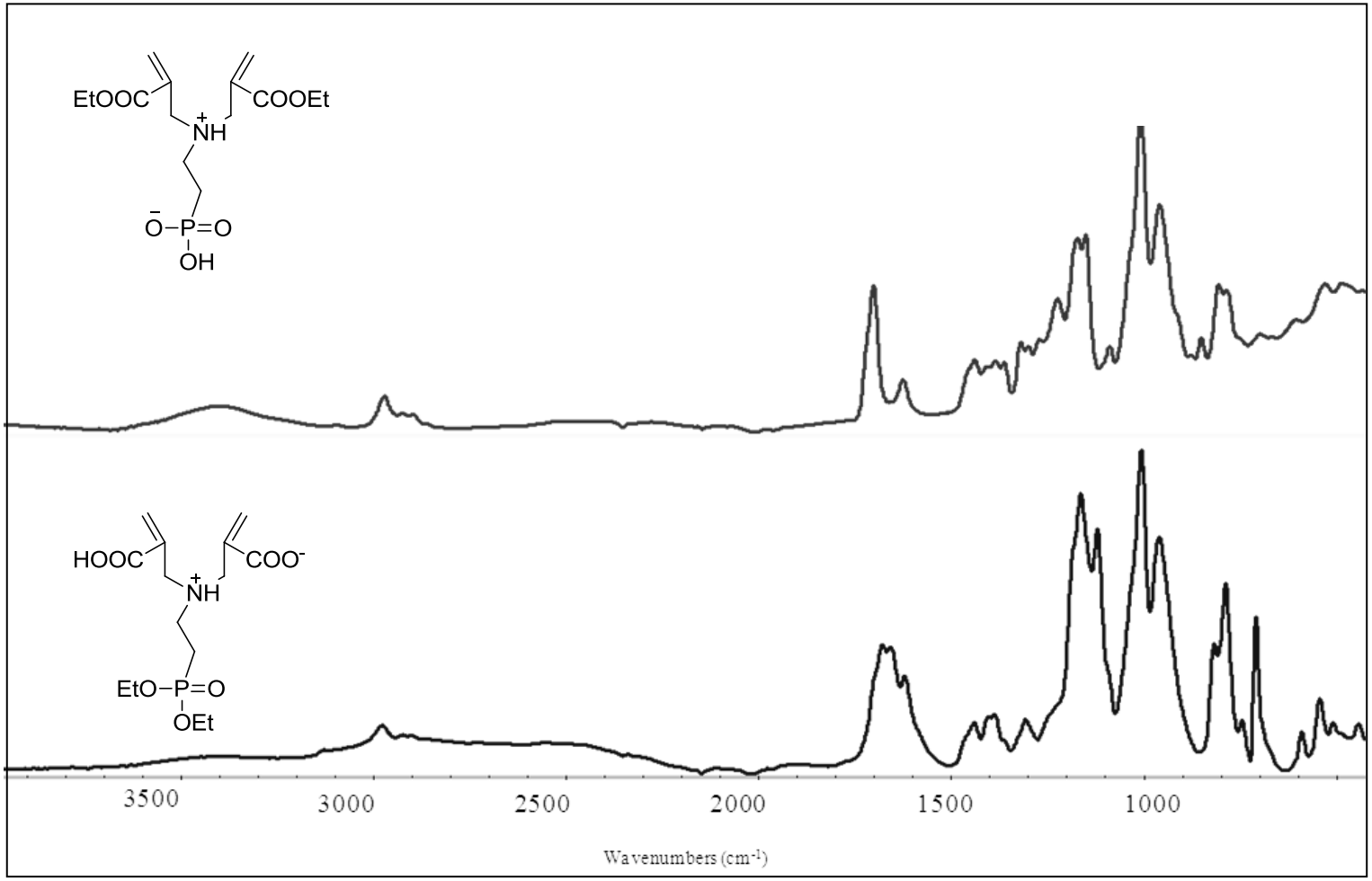


Figure 6.4. FTIR spectra of **D3** and **D6**.

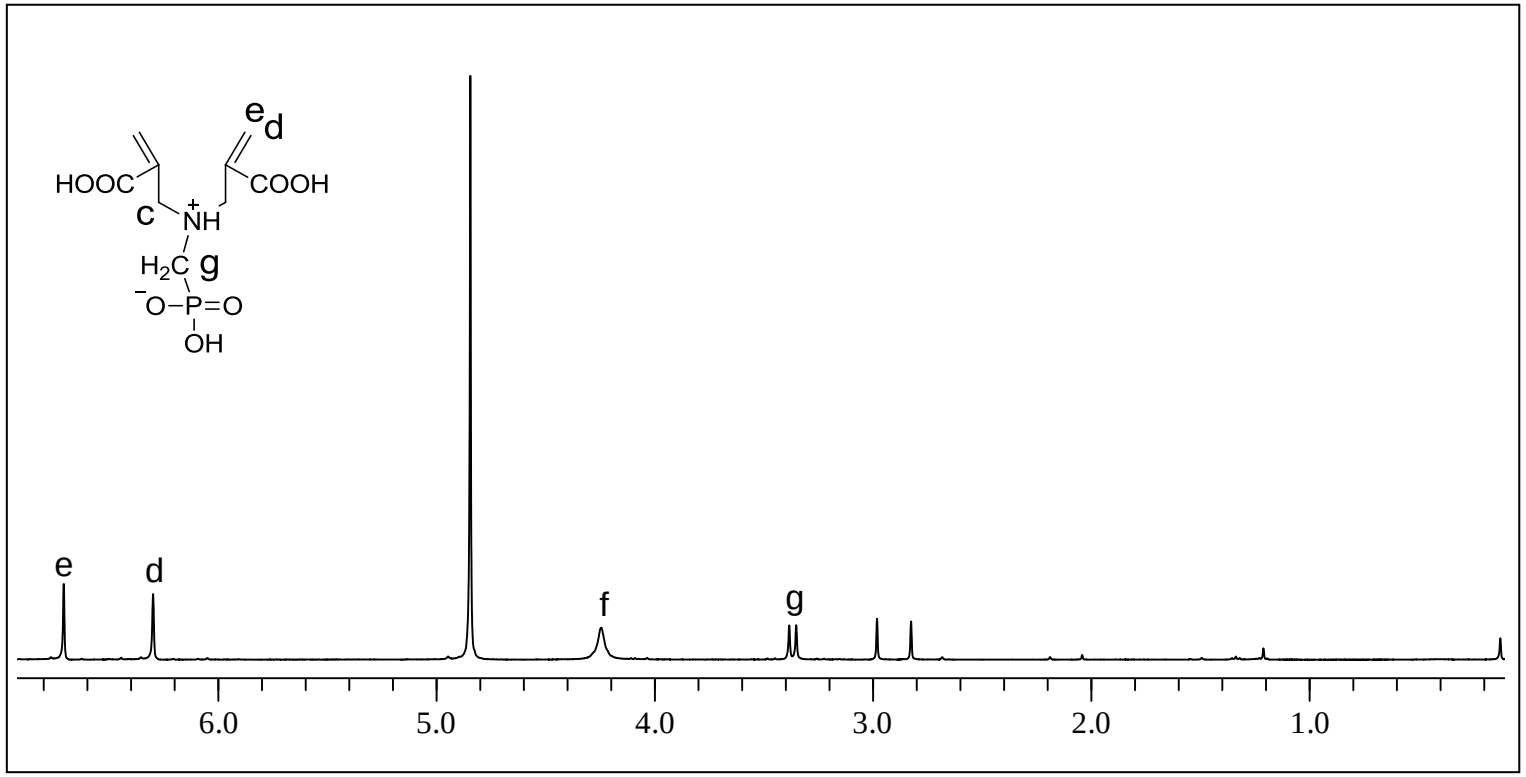


Figure 6.5. ^{1}H NMR spectrum of **D7**.

6.3.2. Viscosity Studies

Riedelsberg and Jaeger showed that monomers containing amino phosphonic acid-groups in their structure have phosphobetaine structure, and they are expected to dissociate in two steps: (i) deprotonation of second P-OH; and then (ii) deprotonation of NH^+ [112]. Because our monomers also have similar structures, carboxybetaine and phosphobetaine, we investigated their behavior upon pH change. Among three groups of monomers, one representative monomer was chosen from each, only phosphonic acid-containing monomer (**D3**); only carboxylic acid-containing monomer (**D5**) and both phosphonic and carboxylic acid-containing monomer (**D7**). The potentiometric titration of these monomers was conducted with standard NaOH (0.05129 M) and the results were shown in "pH" vs "α" where α is the nominal degree of neutralization in terms of equivalents of base added per phosphonic acid groups [201]. It was found that the monomers have zwitterionic structure in water at a pH around 2.4, 2.7 and 2.4 for **D3**, **D5** and **D7** (Figure 6.6).

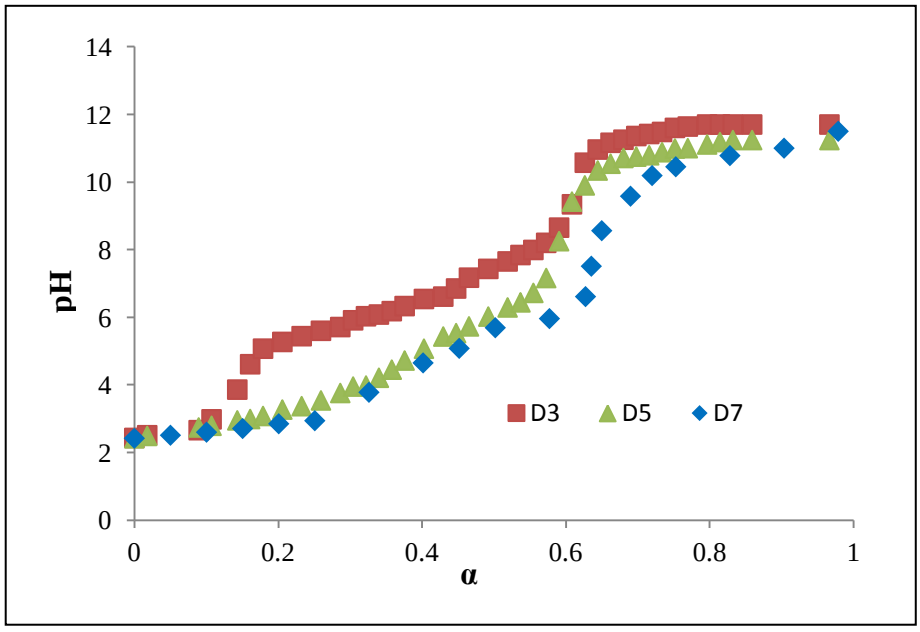


Figure 6.6. pH change for monomers **D3**, **D5** and **D7** upon titration with NaOH.

Monomer **D3** was expected to have the following dissociation mechanism as shown in Figure 6.7 [112].

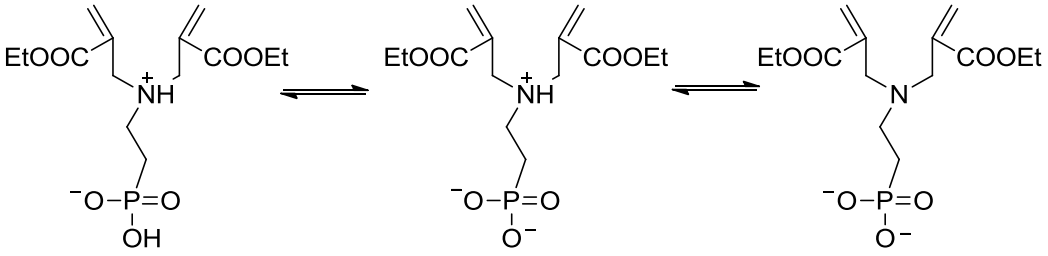


Figure 6.7. Proposed dissociation of **D3** with increasing pH.

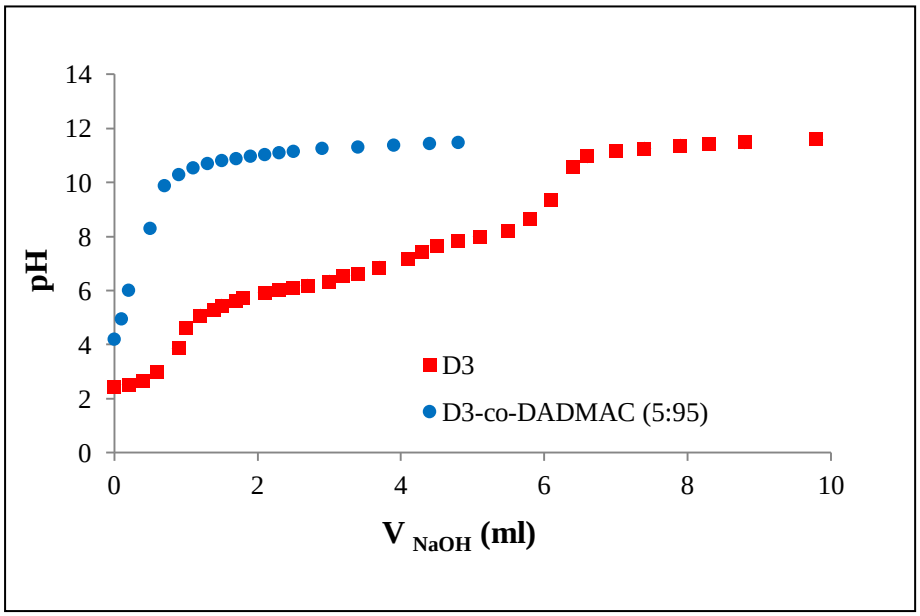


Figure 6.8. pH change for **D3** and **D3**-*co*-DADMAC upon titration with NaOH.

To investigate the pH-responsiveness of monomer **D3**, ^{1}H NMR spectrum of **D3** was also taken at different pH values (Figure 6.9). The pH value of the aqueous solution of this monomer was 2.4 before NaOH addition (Figure 6.9a). After the addition of NaOH (pH=12), we observed disappearance of the ethyl peaks at 1.29 and 4.25 ppm (Figure 6.9b). Also, the peaks corresponding to hydrogens attached to nitrogen at 4.04 ppm shifted upfield (3.22 ppm) due to deprotonation of the monomer. The addition of HCl converted this monomer to **D8** and resulted to peak shifts to downfield again due to protonation of nitrogen (Figure 6.9c).

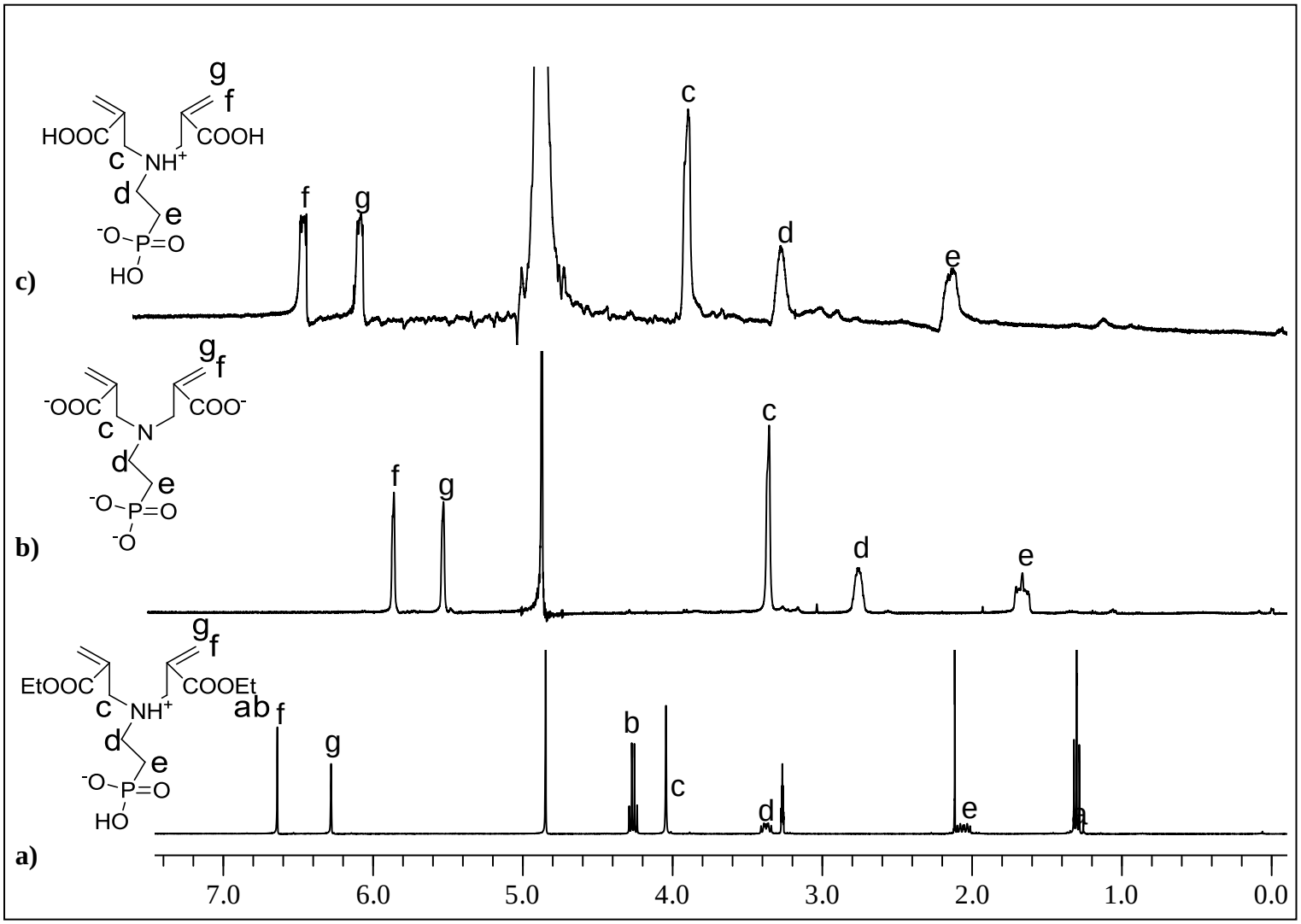


Figure 6.9. [1]HNMR spectra **D3 a)** at around pH= 2; **b)** at the end of titration (pH=12); **c)** after acidification again (pH=2).

6.3.3. Thermal Polymerizations

The monomers (**D2**, **D3**, **D5** and **D8**) were copolymerized with DADMAC in water at 70 °C using V-50. The total monomer and initiator concentrations were 2.7 $molL^{-1}$ and 0.5 x 10^{-2} $molL^{-1}$. DADMAC amounts in feed were 80 and 95 mol%. Table 6.1 shows the copolymerization conditions. The copolymers were isolated by precipitation into acetone and purified by dialysis against water. The copolymers were soluble in water and methanol but insoluble in in ether and acetone. The copolymerization yields were 60-80 % but decreased to about 10 % after dialysis, indicating highest fraction of short chains.

The incorporation of monomers into copolymers was confirmed by ^{1}H NMR and FTIR spectrum. In the FTIR spectra of copolymers, there are new peaks around 1710 and 1220 cm^{-1} corresponding to C=O and P=O, while there are none of these peaks in poly-DADMAC spectrum (Figure 6.10). ^{1}H NMR of pure poly-DADMAC shows peaks: 0.8-1.8 ppm due to 4H in the polymer backbone; 2,2-2.8 ppm due to cyclic protons attached to backbone (2H); 2.9-3.3 ppm and 3.6-3.9 ppm due to cyclic hydrogens and two methyl groups attached to nitrogen. The copolymer compositions were calculated by integration of peak at 3.6- 3.9 ppm with respect to a new peak around 4-4.2 ppm due to methylene groups of incorporated monomer (Figure 6.11).

Table 6.1. Polymerization conditions.

Monomers[a]	**DADMAC in the feed**	**DADMAC in the copolymer**[b]	**Yield**[c] **(%)**	**Intrinsic viscosity**[d] **(dL/g)**
D2-DADMAC	95	96	15	-[e]
D2-DADMAC	80	-[e]	6	-[e]
D3-DADMAC	95	97	10	2.0
D3-DADMAC	80	85	9	-[e]
D5-DADMAC	80	90	8	-[e]
D8-DADMAC	80	92	8	-[e]
DADMAC	100	100	12	1.6

[a] Polymerization time= 4h, [M]=2.7 $molL^{-1}$, $[V_{50}]= 0.5 \times 10^{-2}$ $molL^{-1}$.

[b] % Incorporation of monomers in the copolymer as determined by ^{1}HNMR.

[c] Calculated after dialysis.

[d] Intrinsic viscosities were performed in 0.1 N NaCl solution.

[e] This value was not calculated.

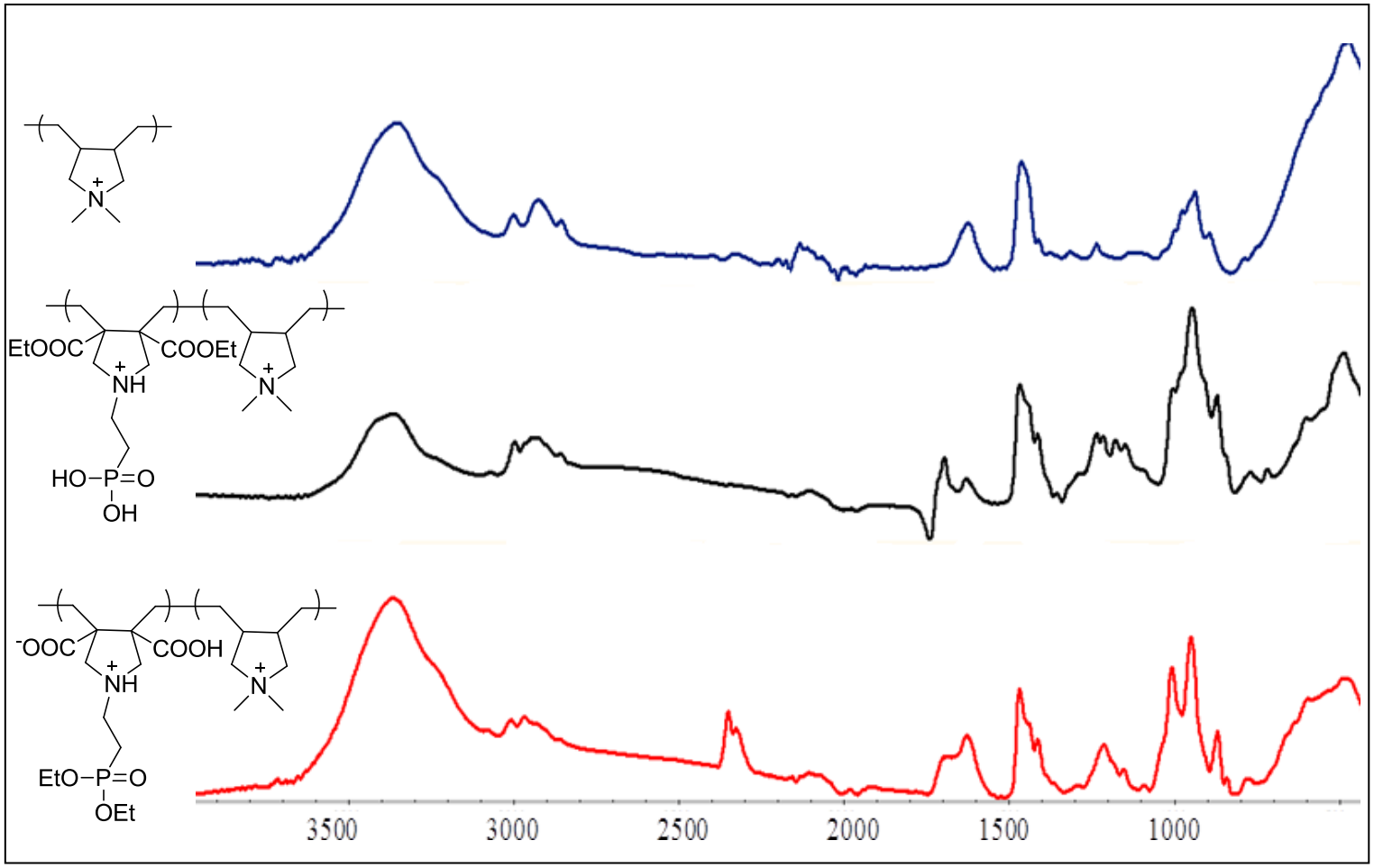


Figure 6.10. FTIR Spectrum of poly-DADMAC, **3**-*co*-DADMAC (5:95), **6**-*co*-DADMAC (5:95).

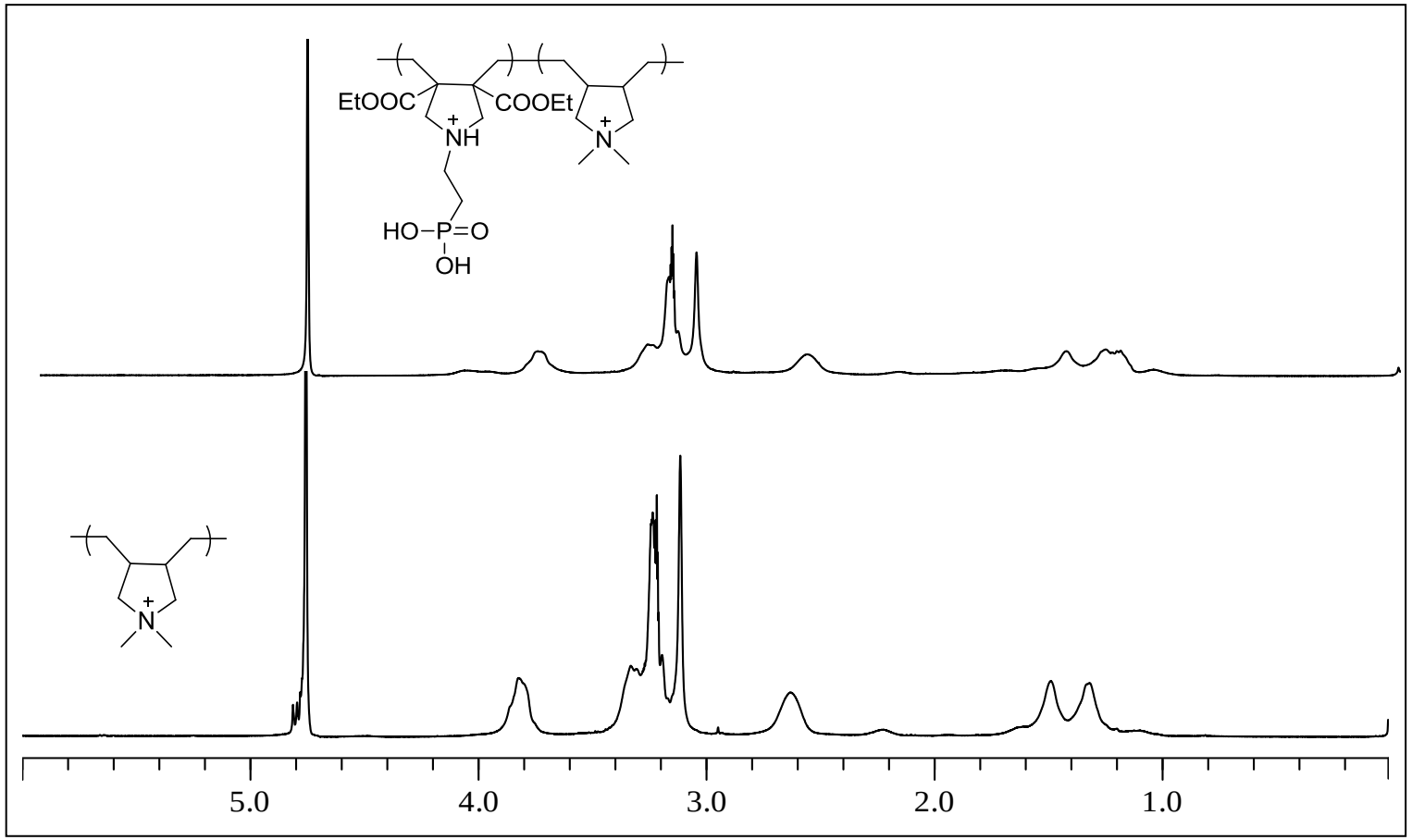


Figure 6.11. ^{1}H NMR spectra of poly-DADMAC and **D3**-co-DADMAC (20:80).

Viscosity curves of the **D3-*co*-DADMAC** copolymer and poly-DADMAC in water are presented in Figure 6.12. The synthesized copolymer showed a typical polyelectrolyte behavior, a strong increase in reduced viscosity at decreasing polymer concentrations. The viscosity of the copolymer was higher than that of poly-DADMAC.

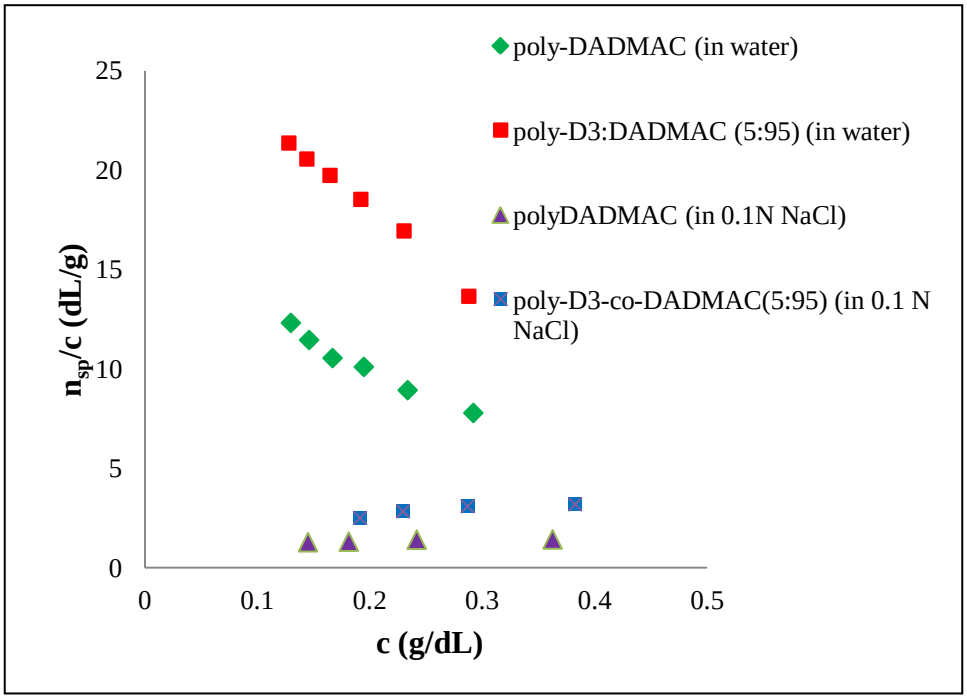


Figure 6.12. Viscosity-polymer concentration graph.

Also, one of the homopolymers (poly-**C1)** (M_n=53000) were hydrolyzed selectively with TMSBr and a totally water soluble polymer was obtained after purification by dialysis (Figure 6.13).

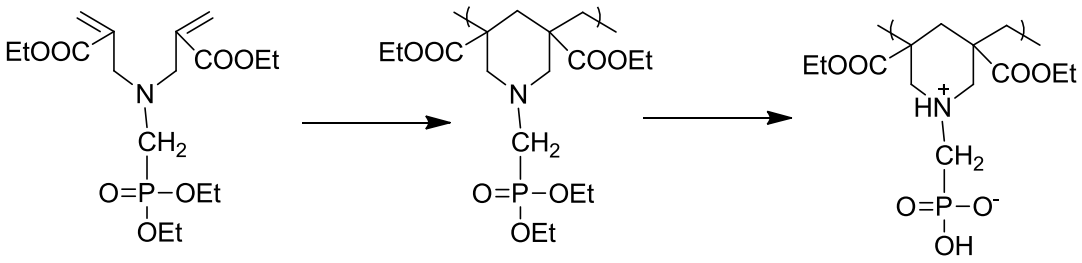


Figure 6.13. Synthesis of quaternary ammonium polymer.

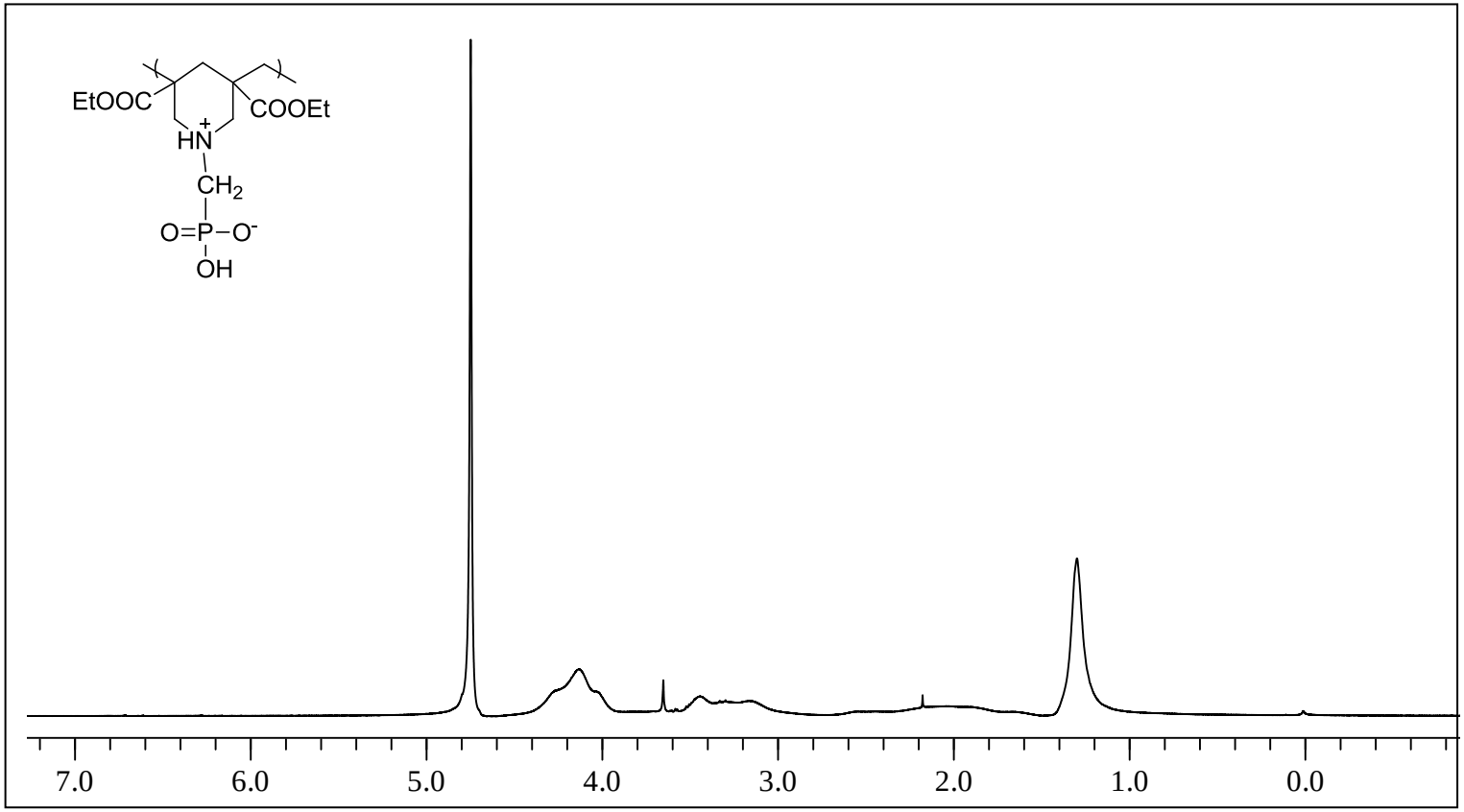


Figure 6.14.[1]H NMR spectrum of hydrolyzed polymer.

6.4. Conclusion

In this study, eight novel zwitterionic monomers with phosphobetaine and carboxybetaine structures were synthesized. pH responsiveness of these monomers were confirmed by ^{1}H NMR analysis. Copolymerization with DADMAC gave new polyelectrolytes. These monomers constitute a new class of monomers which may find biological and environmental applications.

7. CONCLUSION

In the first part of this study, two novel bisphosphonate-containing RHMA monomers were synthesized from reaction of ethyl α-bromomethacrylate (EBBr) and *tert*-butyl α-bromomethacrylate (TBBr) with tetraethyl 4-hydroxybutane-1,1-diyldiphosphonate. Their thermal bulk polymerizations, photopolymerizations and copolymerizations with poly(ethylene glycol) methyl ether methacrylate (PEGMA) were investigated. The homopolymerizations resulted in polymers with M_n values of 25000-56000; the copolymerizations yielded soluble polymers with 22-34 % incorporation of the new monomers; photopolymerizations gave some structure-reactivity correlation; and one of the homopolymers, upon hydrolysis of its bisphosphonate groups, can bond to hydroxyapatite (HAP) which makes these polymers good candidates to be used as biomaterials, in particular, for bone targeting applications.

In the second part of this study, five novel methacrylates, containing either bisphosphonate, or bisphosphonic acid**,** or carboxylic acid, or the last two together, were synthesized and evaluated. The bisphosphonate monomers were synthesized by the reactions of tetraethyl 1-hydroxyethane-1,1-diyldiphosphonate with EBBr and TBBr; same procedure failed with tetraethyl hydroxy(phenyl)methylenediphosphonate. Then, the bisphosphonate monomers were converted to bisphosphonic or carboxylic acid monomers by hydrolysis. Hydrolytic stability, the properties of copolymerizations with commercial dental monomers and HAP interactions make these monomers promising candidates for dental adhesives.

In another part, two different groups of novel aminophosphonate-containing methacrylates were synthesized. The route to the first group involves reactions of EBBr and TBBr with diethyl aminomethylphosphonate and diethyl 2-aminoethylphosphonate. Bulk and solution polymerizations at 60-80 °C with 2,2'-azobis(isobutyronitrile) (AIBN) gave crosslinked or soluble polymers depending on monomer structure and polymerization conditions. Increasing bulkiness from ethyl to *tert*-butyl decreases the polymerization rate, which also correlated well with the chemical shift differences of double bond carbons and consistent with the lower molecular weights of *tert*-butyl ester polymers (M_n= 1.800-7.900

vs. 50.000-72.000). The route to the second group involves the Michael addition reaction between diethyl aminomethylphosphonate and diethyl 2-aminoethylphosphonate with 3-(acryloyloxy)-2-hydroxypropyl methacrylate (AHM) to give secondary amines. The photopolymerization using differential scanning calorimeter showed that these monomers have similar or higher reactivities than AHM, even though AHM has two double bonds. The high rates of polymerization of these monomers was attributed to both hydrogen bonding interactions due to additional NH groups as well as chain transfer reactions.

The last part of the study involved sysnthesis of new zwitterionic monomers with carboxybetaine and phosphobetaine structures. They were synthesized by hydrolysis of aminophosphonate-containing methacrylates of the previous part and cyclopolymerized with diallyldimethyl ammonium chloride to give new water soluble polymers which may find application in metal binding.

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