TOWARDS WATER-SOLUBLE PEROXYL RADICAL CLOCKS

By

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ABSTRACT

Peroxyesters undergo homolytic O-O bond cleavage under thermal and photolytic conditions, forming an alkoxyl and acyloxyl radical, the latter of which rapidly decarboxylates to form a carbon centered radical. While peroxyesters are generally decomposed under anaerobic conditions to serve as sources of carbon-centered radicals to initiate radical reactions, it has been shown that β,γ-unsaturated peroxyesters can decompose to yield a delocalized carbon-centered radical which can serve as a precursor to a powerful peroxyl radical clock. This competitive kinetic approach is attractive as it allows the facile determination of rate constants for formal H-atom reactions between peroxyl radicals and reductants (e.g. phenolic and aromatic amine antioxidants) without the need for specialized equipment. Furthermore, this approach allows for the determination of this kinetic data in a wide variety of solvents.

Herein, we present our work towards the development of a water-soluble peroxyl radical clock, such that the kinetics of reactions of peroxyl radicals with water-soluble reductants can be determined. To do so, we have designed β,γ-unsaturated peroxyesters analogous to those we have previously developed, but that bear either carboxylate or poly(ethylene glycol) moieties to improve solubility in aqueous media. In addition, we have carried out a preliminary investigation of the use of alkyl and acyl xanthates as precursors to peroxyl radical clocks. These radical precursors offer the advantage that they have a pronounced ultraviolet absorption that tails into the visible, permitting efficient photochemical generation of the same alkyl radicals, but at longer wavelengths than that which are currently employed.

ACKNOWLEDGEMENTS

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The chemistry department has a vibrant bunch of people, and I have made a lot of friends along the way. I have my good friends in the Brown lab (Tamer, Pasha, Tony, Mark, Mark2, Alex), Veronique, Christina and Jitendra to thank, for keeping my spirits high and constantly providing encouragement.

Finally, I would like to thank my family for always being there for me. They have helped me tremendously and I am grateful for their support during my studies.

STATEMENT OF ORGINALITY

I hereby certify that all of the work described within this thesis is the original work of the author. Any published (or unpublished) ideas and/or techniques from the work of others are fully acknowledged in accordance with the standard referencing practices.

Jassimranjeet Dhanoa

May, 2010

TABLE OF CONTENTS

LIST OF SCHEMES:

LIST OF FIGURES

LIST OF TABLES

Chapter 1

Introduction

Free radical reactions play a prominent role in biology and medicine. In particular, reactive oxygen species (ROS), which comprise oxygen centred radicals and molecules derived there from, have been implicated in the onset and development of neurodegenerative diseases, heart disease and cancer.[1,2] ROS include superoxide ($O_2^{\bullet-}$) and its conjugate acid, hydroperoxyl radical ($HOO^{\bullet}$), hydroxyl ($HO^{\bullet}$) and peroxyl radicals ($ROO^{\bullet}$), as well as H_2O_2.[3] Among these species, $HO^{\bullet}$ has been studied the most extensively due to its high reactivity with most organic substrates and its probable involvement as an intermediate in various metal-dependent oxidant-generating systems.[3]

Radicals are produced under normal physiological processes or through the influence of exogenous species. Examples of exogenous species include compounds that occur naturally in the biosphere (e.g. ozone, NO_2, ethanol or tetradecanoyl phorbol acetate (TPA), industrial chemicals (e.g. carbon tetrachloride), or xenobiotics (e.g. benzo(α)pyrene).

Physiologically, superoxide is formed in all aerobically metabolizing cells, and appears to “leak” out of the mitochondrial electron transport causing the radicals to oxidize xenobiotics and/or initiate pathological changes.[3] Superoxide formation occurs through redox cycling, when electronegative compounds intercept electrons from normal cellular electron transport.

$$O_2 + e^{\ominus} \longrightarrow O_2^{\bullet-} \qquad (1)$$

Superoxide undergoes rapid dismutation to form hydrogen peroxide,

$$2O_2^{\bullet-} + 2H^+ \longrightarrow H_2O_2 + O_2 \qquad (2)$$

a reaction that is further accelerated by superoxide dismutases (SOD) in the body, and can also be protonated to form the hydroperoxyl radical:

$$O_2^{\bullet -} + H^+ \longrightarrow HOO^{\bullet} \quad (3)$$

The hydrogen peroxide formed via dismutation of superoxide, or from H-atom abstraction by hydroperoxyl reacts readily with low-valent metals, such as iron, to generate hydroxyl radicals by the Fenton reaction (4):

$$Fe^{2+} + H_2O_2 \longrightarrow Fe^{3+} + {}^{\ominus}OH + {}^{\bullet}OH \quad (4)$$

Peroxyl radicals are formed by the reaction of molecular oxygen with a carbon centred radical, which can be formed when hydroxyl (or hydroperoxyl) radicals abstract an H-atom from a molecule:

$$^{\bullet}OH + RH \longrightarrow R^{\bullet} + H_2O \quad (5)$$

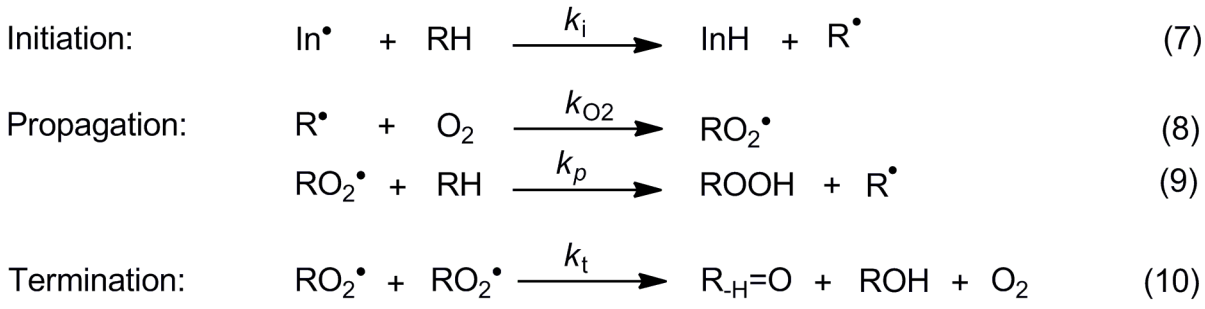
$$R^{\bullet} + O_2 \longrightarrow ROO^{\bullet} \quad (6)$$

The reactivity of peroxyl radicals in water – or in highly polar environments – is not well understood, even though peroxyl radicals are present in normal cells at high steady-state concentrations owing to the steady generation of superoxide, and therefore the reactions of hydroxyl (and hydroperoxyl) radicals with biomolecules in the oxygenated environment. Peroxyl radical formation can occur in the nuclear membrane, hence DNA associated with nuclear membrane-chromatin attachment sites are

presumed to be more susceptible to damage by reactive lipid radicals formed in adjacent membranes.[5] Biological processes, in which peroxyl radicals are generated, such as oxidizing lipids, are known to result in the oxidation of DNA bases.[5]

1.1 Lipid Peroxidation

Lipid peroxidation is a complex process, whereby molecular oxygen and lipid react through a free-radical chain mechanism, shown in **Scheme 1.1**. For most organic substrates in solution, the rate-determining step of the chain reaction is attack on the substrate by peroxyl radicals.[6] Under most conditions, the primary oxidation product derived from a substrate RH is the corresponding hydroperoxide, ROOH. The chain reaction can be represented by the series of steps shown below, more generally referred to as hydrocarbon autoxidation:



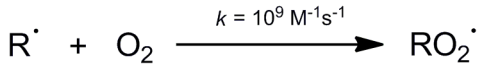
Scheme 1.1: The radical chain mechanism of hydrocarbon autoxidation

The initiation step basically involves any reaction which may yield a substrate derived alkyl radical (R•). Initiator radicals (In•), can be produced by photochemically or thermally using various sources, such as azo compounds and peroxides (i.e. H_2O_2, as in equation 4 and 5) for in vitro and in vivo reactions respectively. In the first propagation step, the substrate derived carbon-centred radical (R•) reacts with O_2 to form a peroxyl radicals (ROO•) at a diffusion controlled rate. In the second propagation step, the peroxyl radical abstracts a H-atom from a molecule of the substrate forming a hydroperoxide (ROOH) and a new carbon-centred radical (R•). In the final step, termination takes place when two substrate derived peroxyl radicals react to give non-radical products by the so-called Russell termination reaction,[7,8] believed to occur via a tetroxide intermediate.

Lipid peroxidation can have deleterious effects in biological systems. It has been implied that various pathological events including atherosclerosis, liver disease, tumorigenesis, and various neurological disorders, such as Alzheimer's disease are linked to lipid peroxidation.[9] Initiation and antioxidant defences are of key importance to the mechanism(s) of lipid peroxidation, and α-tocopherol (α-TOH) has attracted attention as a peroxidation chain breaker since its a potent lipophilic antioxidant and its readily available.[10]

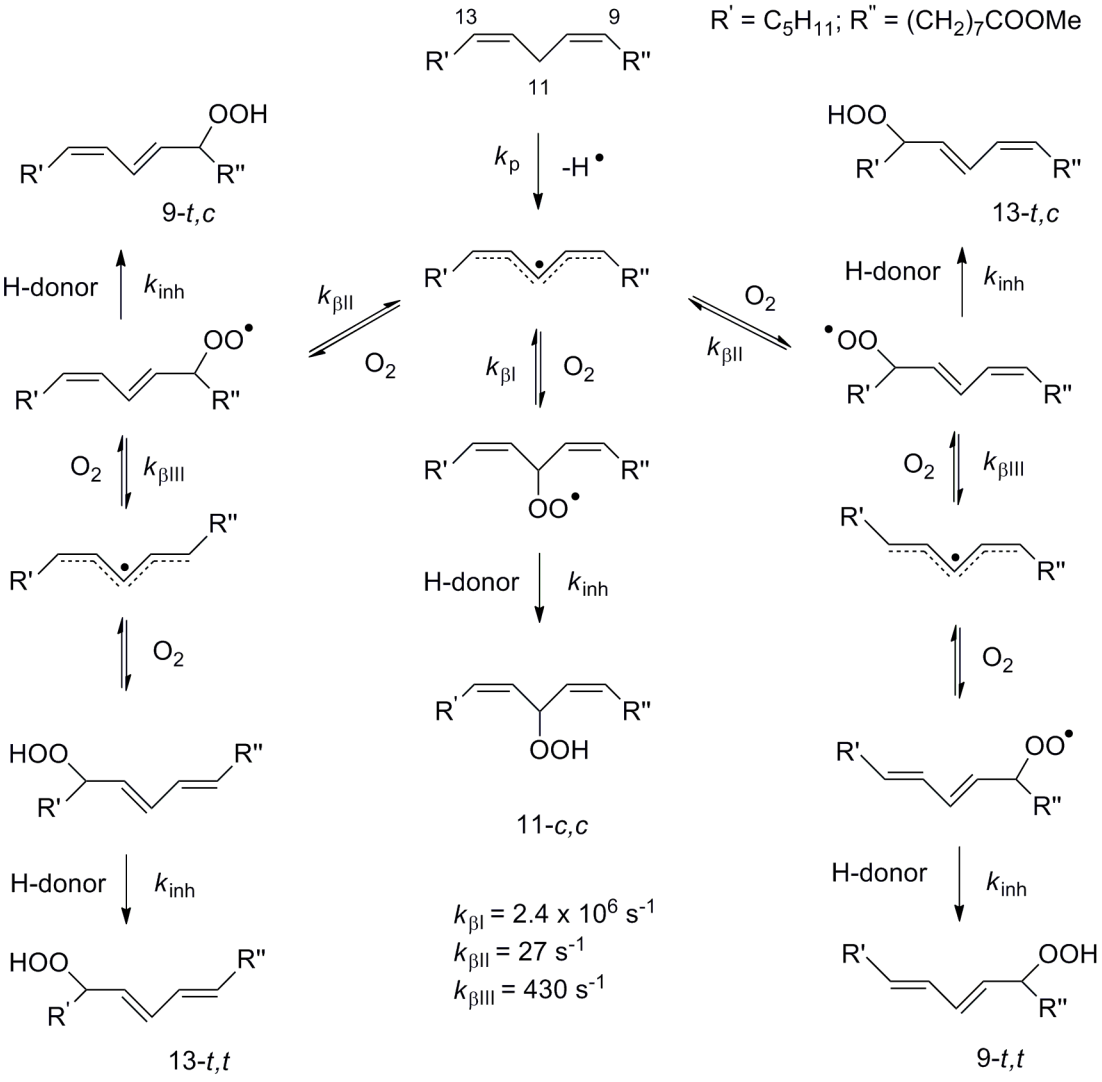
1.1.1 Reaction of Carbon-Centred Radicals with Oxygen

The reaction of oxygen with alkyl radicals to produce peroxyl radical occurs at or near the diffusion-controlled limit, with rate constants generally on the order of 10^9 $M^{-1}s^{-1}$; the formation of peroxyl radicals can occur within ≤10 μs.



(11)

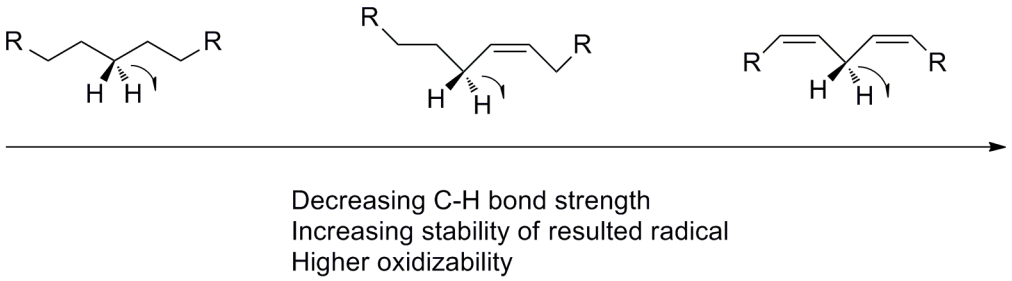
Radicals derived from the autoxidation of polyunsaturated lipids are highly stabilized and react reversibly, resulting in complex product mixtures, such as both trans, cis and trans, trans hydroperoxides as shown in the peroxidation of methyl linoleate (**Scheme 1.2**). Upon abstraction of a hydrogen atom from the bis-allylic position, reversible oxygen addition can occur at 3 different positions, C9, C11 and C13; the first, third and fifth carbon of the pentadienyl radical intermediate. In the case of methyl linoleate, a racemic mixture of up to five regioisomeric lipid hydroperoxides are formed: 9-*t,c*, 13-*t,c*, 13-*t,t*, 9-*t,t* and 11-*c,c*-hydroperoxyoctadienoates. The product distribution is dependent on the values of k_β of each of the intermediate peroxyl radicals, which reflects their relative propensities to fragment back to the alkyl radical and oxygen and either the k_p of the substrate or the k_{inh} of an antioxidant when it is present in the medium – either of which can donate a hydrogen atom to the peroxyl radical, trapping it as the hydroperoxide.



Scheme 1.2: Autoxidation of methyl linoleate, where k_p is the rate constant of H-atom abstraction, k_β is the rate constant of β fragmentation of intermediate peroxyl radicals and k_{inh} is the rate constant of peroxyl radical-trapping by antioxidants

1.1.2 H-Atom Abstractions by Peroxyl Radicals

The rate-limiting step in the propagation sequence of lipid peroxidation is H-atom abstraction from the lipid. Lipids are a good target for autoxidation due to their weak C-H bond. The bond dissociation enthalpy (BDE) is especially low at the allylic and bis-allylic positions of mono- and polyunsaturated lipids, respectively (see above in **Scheme 1.3**), since upon H-atom abstraction from these positions, resonance stabilized allyl and pentadienyl radicals are formed, respectively. The greater the stability of the resultant radical species, the lower the C-H BDE is and consequently, the higher the oxidizability of the lipid (**Scheme 1.3**).



Scheme 1.3: The different reactivities of saturated, monosaturated and polyunsaturated lipids

The rate constant of hydrogen atom abstraction by a free radical depends mostly on the activation energy of the reaction.[11] A reaction can only proceed rapidly if the activation energy is small; as a consequence, only exothermic hydrogen atom abstractions are likely to be fast. Oxidations would be rapid if the bond being formed (ROO-H) is as strong as the bond being broken (R-H) (**Eq. 8**). Peroxyl radicals are strongly resonance stabilized. The estimated bond dissociation energy for ROO-H is

approximately 88 kcal/mol, making it stronger than an allylic or benzylic C-H bond (~84 kcal/mol) or an aldehyde C-H bond (~86 kcal/mol).[6] The bond dissociation energy of ROO-H is comparable to a tertiary C-H bond in a saturated hydrocarbon (90-92 kcal/mol). The resonance weakened O-H, S-H, and N-H bonds of phenols, thiophenols, and aromatic amines provide hydrogen atoms which can be readily abstracted. Since peroxyl radicals can only abstract H atoms from weak C-H bonds, the abstraction is relatively slow and therefore the measurement of rate constants is effected by faster radical-radical decay processes.[6]

Since the peroxyl radical is unreactive in comparison to other oxygen derived radicals, it is very selective in its abstraction from hydrocarbons and it has a strong preference for the most weakly bound hydrogen atom. Abstraction from hydrocarbons, even when they contain an allylic or benzylic hydrogen, is generally a very slow process at 30 °C.[6] Although, the R-H bond dissociation energy is the most important factor in determining the rate constant for hydrogen atom abstraction from RH, steric and polar factors can also play a significant role.[6] Differences in rate constants, owing to sterics, have been observed both for changes in the structure of the peroxyl radical and for changes in the structure of the oxidizing substrate. For example, tertiary cumyl peroxyl radical is less reactive than the secondary tetralylperoxyl radicals toward cumene and tetralin. Generally, primary and secondary peroxyl radicals are about three to five times more reactive in hydrogen abstraction as tertiary peroxyl radicals.[6]

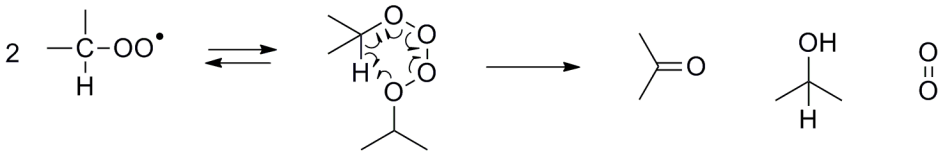
1.1.3 Peroxyl-Peroxyl Reactions

Rate measurements on the self-reactions of peroxyl radicals derived from secondary and tertiary alkyl radical have been carried out over a range of experimental conditions.[11] In terms of the self-reaction of primary alkylperoxyl radicals, they have only been carried out at room temperature and typically in aqueous solutions.[11] Primary alkyl peroxyl radicals can self-react very rapidly, near the collision rate, whereas secondary radicals react much more slowly, and tertiary radicals react very slowly. In order to study the kinetics of primary radicals, rapid kinetic techniques, such as pulse radiolysis or flash photolysis are required. Slower kinetic techniques such as interrupted photolysis can be used for secondary and tertiary radicals.[11] The slowness of the reaction between tertiary peroxyl radicals is due primarily due to the fact that the decomposition of a tetroxide intermediate to alkoxyl radicals and oxygen requires significant activation energy.[12]

Table 1.1: Chain termination Constants for Hydrocarbon Oxidation at 30 °C.[6]

Peroxyl Radical	**$2k_t$, $M^{-1}sec^{-1}$**	**Examples**
Primary, $RCH_2COO•$	2-4 x 10^8	p-Xylene, 1-octene
Secondary, RR'CHOO•	20-40 x 10^8 1-10 x 10^6	Ethylbenzene, styrene 3-Heptene
Tertiary, RR'R''COO•	0.1-60 x 10^4	Cumene
Hydroperoxyl, HOO•	1.2 x 10^9	1,4-cyclohexadiene

For primary and secondary peroxyls, the so-called Russell mechanism[7,8] is generally believed to be operative (**Eq. 12**), as proposed by Russell in 1957. The mechanism involves decomposition of a tetroxide through a cyclic transition state whereby one of the α-hydrogen atoms is transferred to the products ketone, alcohol and oxygen. Support for the Russell mechanism includes the observation that singlet oxygen is produced, as might be anticipated from this 6-membered pericyclic process.



(12)

The Russell termination mechanism is a plausible explanation for the observed low activation energy and rapid termination reactions of secondary peroxyl radicals compared with tertiary peroxyl radicals.

1.2 Antioxidants

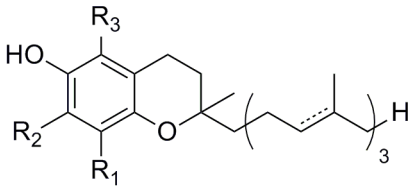
Although oxidation reactions are crucial for life, they can also be damaging; hence, plants and animals maintain complex systems of multiple types of antioxidants, such as glutathione, Vitamin C, Vitamin E as well as enzymes such as catalase, superoxide dismutase and various peroxidases. When there are low levels of antioxidants or inhibition of the antioxidant enzymes, oxidative stress can occur and may damage or kill cells.

Antioxidants are generally classified as preventive – i.e. they react with ROS or compounds that will give rise to ROS – such as the enzymes; and chain-breaking radical-trapping antioxidants – those that inhibit the chain reaction. Chain breaking

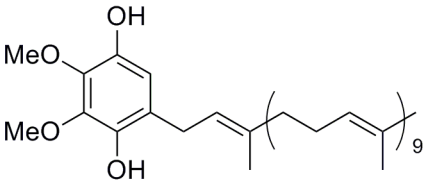
antioxidants are classified into two broad divisions, depending on whether they are soluble lipid soluble or water-soluble. Generally, lipid-soluble antioxidants protect cell membranes from lipid-peroxidation, whereas water soluble antioxidants react with oxidants in the cell cytosol and blood plasma. These antioxidants are generally made by plants, and therefore obtained from our diets. In body fluids and tissues, different antioxidants are present in a wide range of concentrations.

1.2.1 Lipid Soluble Antioxidants

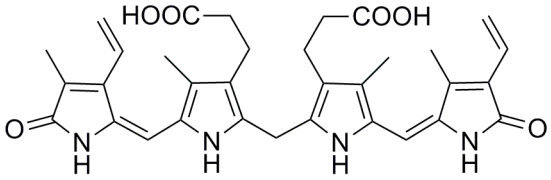
There are several lipophilic antioxidants which constitute membrane lipids and low density lipoprotein (LDL) particles; some examples include Ubiquinol (co-enzyme Q10), bilirubin, and Vitamin E. Vitamin E is a general name for the 4 congeners of each of the tocopherols and tocotrienols.[13] Of these congeners, α-tocopherol is the most abundant, and hence its phenolic radical trapping chain-breaking antioxidant ability has been thoroughly studied. α-Tocopherol is a potent, lipid-soluble phenolic antioxidant and it has been suggested that it is the only lipid-soluble chain breaking type of antioxidant present in human blood cells and rat liver.[13]



α-tocopherol: $R_1 = R_2 = R_3 = CH_3$
α-tocotrienol: $R_1 = R_2 = R_3 = CH_3$
β-tocopherol: $R_1 = R_3 = CH_3$, $R_2 = H$
β-tocotrienol: $R_1 = R_2 = R_3 = CH_3$
γ-tocopherol: $R_1 = R_2 = CH_3$, $R_3 = H$
γ-tocotrienol: $R_1 = R_2 = CH_3$, $R_3 = H$
δ-tocopherol: $R_1 = R_2 = R_3 = H$
δ-tocotrienol: $R_1 = R_2 = R_3 = H$



ubiquinol



bilirubin

Scheme 1.4: Several lipophilic antioxidants present in LDL particles in human serum.

Phenolic antioxidants inhibit lipid peroxidation by trapping chain-carrying peroxyl radicals by transfer of their phenolic H-atom to form a lipid hydroperoxide (**Eq. 13**).

$$ArOH + ROO^{\bullet} \underset{(\leftarrow)}{\overset{k_{inh}}{\longrightarrow}} ArO^{\bullet} + ROOH \qquad (13)$$

To break the radical chain reaction, the antioxidant has to intercept the radical in the propagation step and form a phenoxyl radical that is unreactive to either oxygen (**Eq.14**) or the substrate (**Eq.15**):

$$ArO^{\bullet} + O_2 \xrightarrow{\text{very slow}} Ar(=O)OO^{\bullet} \qquad (14)$$

$$ArO^{\bullet} + RH \xrightarrow{\text{very slow}} ArOH + R^{\bullet} \qquad (15)$$

Hence, the phenoxyl radical decays either through reacting with itself (**Eq. 16**) or more likely reacting with another chain-carrying radical (**Eq.17**).

$$ArO^{\bullet} + ArO^{\bullet} \xrightarrow{fast} \text{Products} \quad (16)$$

$$ArO^{\bullet} + ROO^{\bullet} \xrightarrow{fast} \text{Non-radical Products} \quad (17)$$

Assuming that chain termination occurs only via the two consecutive reactions **13** and **17** and taking quasi-stationary conditions, the rate of inhibition under a steady rate of initiation (R_i) and low [ROOH] is given by **Eq. 18**.

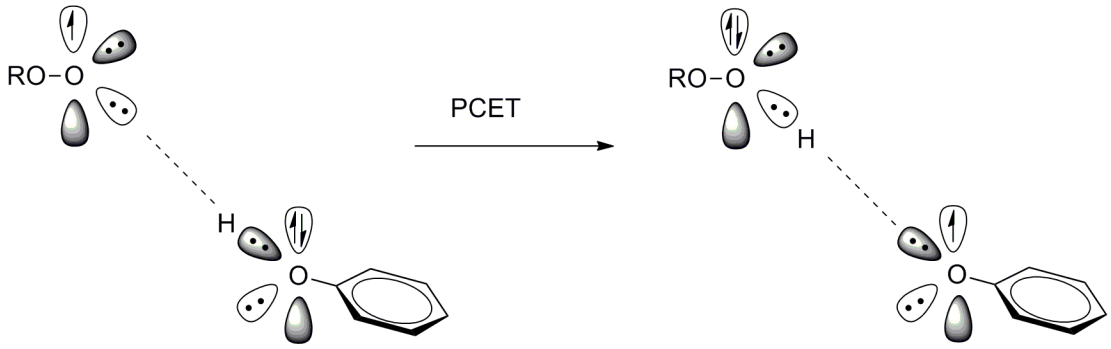
$$R_{O_2}^{inh} = \frac{k_p[RH]R_i}{nk_{inh}[ArOH]_0} \quad (18)$$

In this equation, k_p and k_{inh} represent the rate constants for the propagation and inhibition steps, respectively, and n represents the stoichiometric factor. It has been shown that in many cases the rate follows this law at least in the early stage (induction period) of inhibition of oxidation.[14] It also shows that for ArOH to be efficient k_{inh} must be several orders of magnitude greater than k_p. Consequently, antioxidant ability is not an absolute property of ArOH, but also depends on the substrate. Substrates that are difficult to oxidize can be protected by relatively slow antioxidants.

The antioxidant ability of phenols depends on the ability to give up a hydrogen atom as seen in **Eq. 13**. This ability is dependent on the bond dissociation enthalpy (BDE) of the phenolic O–H bond. An increase in the electron density of the aromatic ring leads to a lower BDE by stabilizing the electron deficient phenoxyl radical. Pedulli and co-workers has shown that having an electron donating (ED) group in the para position of α-Tocopherol lowered the O–H BDE by several kcal/mol; *p*-MeO phenol has an O–H BDE of (82.8 ± 0.8) kcal/mol versus (88.3 ± 0.2) kcal/mol for phenol.[15,16] The O-

H BDE in α-tocopherol is even lower (78.2 ± 0.8) kcal/mol[16] thus making it a good antioxidant.

The mechanism for **Eq.13** is historically believed to be an H-atom transfer (HAT) mechanism but has recently been revised to a proton-coupled electron transfer (PCET).[32] In this mechanism, complex formation between the phenol and peroxyl radical precedes the reaction, whereby the phenolic hydrogen is H-bonded to the peroxyl radical. This hydrogen is then moved as a proton from the phenol to the lone pair on the peroxyl radical along with the coupled movement of an electron from the 2p lone pair of the phenolic oxygen to the orbital containing the unpaired electron on the peroxyl radical, such that the electron travels between two formally non-bonding orbitals (**Scheme 1.5**).



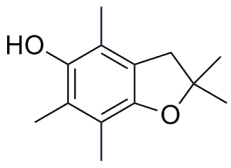
Scheme 1.5: PCET mechanism between a peroxyl and a phenol

The Hammett equation is a classic way of expressing substituent (Y) effects on rates and chemical equilibria. Applied to ArO–H BDEs for *para* substituted phenols, both theoretical and experimental values show an excellent linear correlation with σ+ (Y).[17]

Furthermore, it has been established that the effect of a substituent on the H atom abstraction from ArOH by radicals was due to a substituent induced change in ArO–H BDE and not to polar effects of the substituent on the transition state.[18] Hence, phenols with electron-donating substituents in the *ortho* and *para* positions would make faster antioxidants and, as exemplified the case of α-TOH, which possesses a *para* alkoxy moiety and *ortho* alkyl groups. Rate constants ($M^{-1}s^{-1}$) k_{13} (refer to **Eq. 13**) for H-atom abstraction from a few selected phenols are shown in **Table 1.2**.

Table 1.2: Rate constants (k_{13}) for H-atom abstraction from a few selected phenols (ArOH) by ROO•

ArOH	**k_{13} ($M^{-1}s^{-1}$)**	**Reference**
Phenol	6.5×10^3	19
α-Tocopherol	3.2×10^6	20
I	5.7×10^6	21
II	1.1×10^7	22
III	2.9×10^7	22



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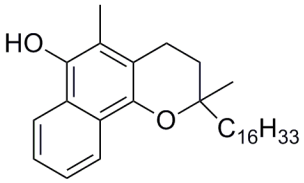

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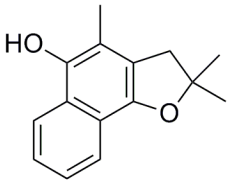
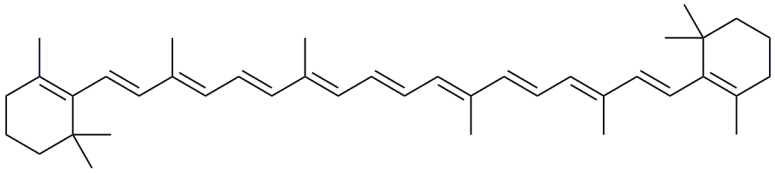


III

In addition to vitamin E, a few other lipid-soluble chain breaking antioxidants have been reported. It has been shown[23] that bilirubin, the end product of heme metabolism in mammals, can function as an antioxidant of either homogeneous solution or liposomes.[13,24] The antioxidant activity of bilirubin increases as the experimental concentration of oxygen is decreased.

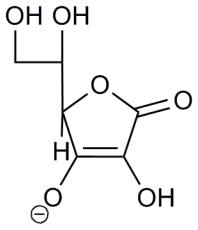
β-Carotene is known to be an effective quencher of singlet oxygen and can also function as a radical-trapping antioxidant. β-Carotene is highly unsaturated, thus making it very reactive towards peroxyl radicals. The peroxyl radical most likely adds to one of the unsaturated carbons, rather than hydrogen atom abstracting, to give a resonance-stabilized carbon-centered radical, which must react with oxygen reversibly.[13]



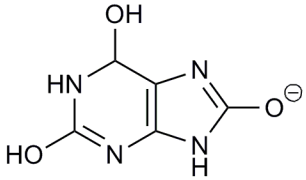
β-Carotene

1.2.2 Water Soluble Chain-Breaking Antioxidants

There are several chain-breaking water soluble antioxidants known in nature. For example, ascorbic acid, uric acid, cysteine and glutathione scavenge oxygen-centred radicals and suppress the oxidation of lipids.



Ascorbate (pK_a = 4.25)

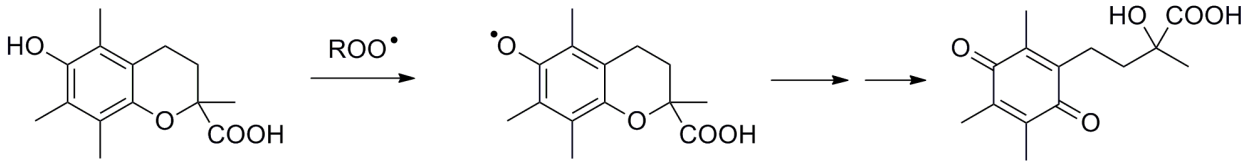


Urate (pK_a = 3.89)

It has been shown by Matsushita[25] in 1963, that uric acid is as effective an antioxidant as α-tocopherol in the oxidation of linoleic acid in water at 37°C, and since then other experiments have confirmed uric acid's antioxidant ability.[26,27] Matsushita also showed that adenine, guanosine, xanthine, hypoxanthine, uric acid, uricil, orotic acid, and ribonucleic acid were as effective antioxidants as α-tochopherol.[25]

Trolox, the water soluble antioxidant derivative of vitamin E, is often used in biological and biochemical applications to reduce oxidative stress or damage. Since its synthesis thirty-five years ago,[24] this synthetic phenolic antioxidant has been examined for its antioxidant action by a number of investigators in a variety of systems.

Trolox exhibits the structural requirements for effective antioxidant action; specifically, it is a hindered phenol with a *para* ether oxygen in a ring system allowing the structure to possess the requirement for stereoelectronic stabilization[21] of the resulting phenoxyl radical, **2**, on hydrogen atom transfer to an active oxygen radical, such as a peroxyl:

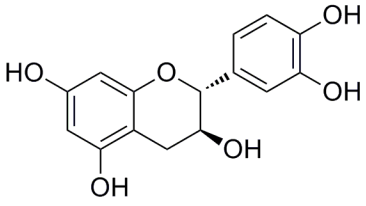


1 **2** **3**

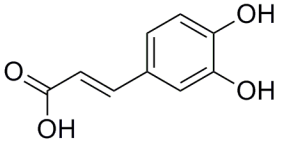
Trolox has advantages over other antioxidants (i.e. Vitamin E) because it has the same antioxidant activity, due to its chromanol structure, however it has a carboxy group which effects moderate water-solubility which makes it more versatile over lipid-soluble antioxidants. For example, Trolox does not have to be incorporated into the lipid membrane by solvent extraction and co-evaporation methods; it can be added directly

to the intact system.[24] This makes it convenient for studies on natural biological systems and for quantitative studies on model systems.

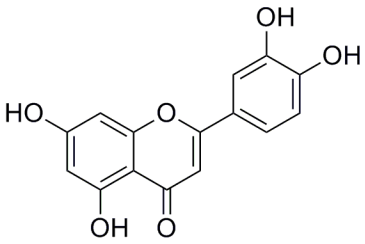
Polyphenols are a complex family of molecules, ubiquitous in plants[28] and include the flavonoids and phenolic acids. Little is known about their fate in the gastro-intestinal tract; however they have attracted a lot of research interest due to their complex structure and potent in vitro antioxidant activity. Commonly occurring flavonoids are quercetin derivatives (onion, apple), catechins (tea), anthocyanins (berries) and hesperitin derivatives (citrus fruits), while commonly occurring phenolic acids are caffeic acid, occurring in many fruit and vegetable and chlorogenic acid (coffee). The antioxidant activity of dietary nutrients is also susceptible to its environment, the presence of proteins and other antioxidants. Certain polyphenols may readily pass the gastrointestinal barrier (e.g. caffeic acid) while other are poorly absorbed (e.g. rutin). This makes it very difficult to correlate the antioxidant capacity of a compound in vitro with its effect in vivo.



Catechin



Caffeic Acid



Quercetin

1.3 Other Reactions of Peroxyls Radicals

The lifetime of peroxyl radicals can last for several seconds at 37°C, which is sufficient time for them to diffuse freely in solution, such as in biological systems. Peroxyl radicals can undergo a variety of reactions with suitable substrates. The most common reactions with molecules include hydrogen atom abstractions and addition to unsaturated systems as exemplified above, but also radical displacement, and oxygen atom transfer as shown below.

1.3.1 Radical Displacement

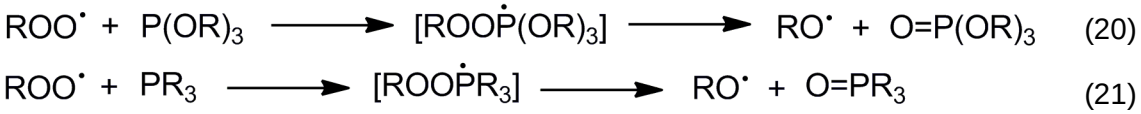
The autoxidation of organoboron compounds is a free-radical chain reaction which almost certainly involves a radical displacement on boron as one of the propagation steps.

$$ROO^{\bullet} + BR_3 \longrightarrow [ROO\dot{B}R_3] \longrightarrow ROOBR_2 + R^{\bullet} \qquad (19)$$

Other alkyl derivatives of group III metals also react readily with oxygen, presumably by the same sort of mechanism.

1.3.2 Oxygen Atom Transfer

The autoxidation of trialkyl phosphites and trialkyl phosphines occur by rapid chain reactions (**Eq. 19** and **Eq. 20**). Walling and Rabinowitz, were the first to propose that the propagation step involves the intermediate formation of a phosphoranyl radical with nine electrons on phosphorus.[6] The presence of unoccupied d orbitals in phosphorus allows for the valence shell expansion.



(20)

(21)

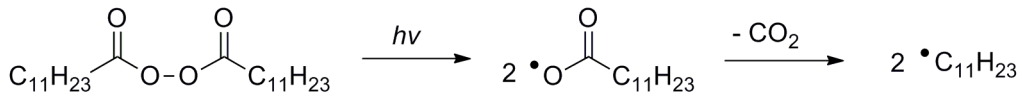
1.4 Production and Detection of Peroxyl Radicals

Peroxyl radicals are known to form through a variety of chemical processes, including the diffusion controlled reaction of carbon radicals with oxygen. Peroxyl radicals have been detected by electron spin resonance spectroscopy in hydrocarbon autoxidations (both in the solid and liquid phase) and in the radical and ceric ion oxidation of hydroperoxides.[6] The *g* values (line positions) of peroxyl radicals range from 2.014-2.019.[6,29] This value is unique in comparison to other radicals commonly present in oxidizing systems (e.g., R· or R_2NO·) which have *g* values near that of that of the free electron (2.0023) as a result of almost complete quenching of orbital angular momentum.[6] Peroxyl radicals exhibit optical absorption only in the UV, generally with λ_{max} near 250 nm and relatively low molar absorptivities ($\varepsilon = 10^3$ L mol^{-1} cm^{-1}).[11] However, both ethenyl and phenyl peroxyl radical absorb in the visible region in aqueous solution.

1.4.1 Photolytic and Thermolytic Production of Peroxyl Radicals

Diazenes (azo compounds), diacylperoxides, peroxyesters, peroxyoxalates and xanthates are a few examples of compounds which can be used as sources of alkyl radicals, which under aerobic conditions, give rise to peroxyl radicals. For example,

dilauroyl peroxide is a source of alkyl radicals via the following decomposition, but can react further with oxygen to form peroxyl radicals as shown in **Eq. 22**.



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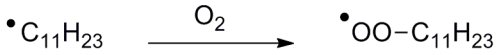
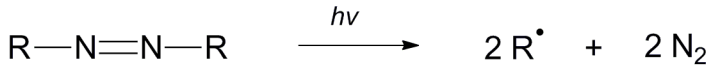


(22)

The production of the alkyl radical as precursors of peroxyl radicals has been studied thoroughly using photolytic methods. The most common examples are the photolysis of azo compounds, ketones, and less frequently, the photolysis of compounds containing metal carbon-bonds.[11] In the case of azo compounds, photodecomposition can result in the formation of two alkyl radicals, which may be identical if the compound is symmetrical.



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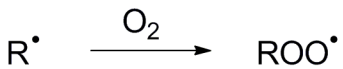
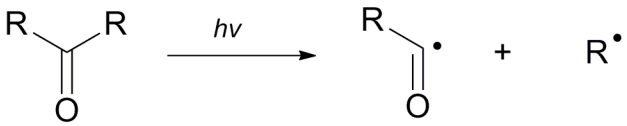


(24)

In the case of ketones, photolysis yields an alkyl and acyl radical, allowing both of them to react with oxygen to produce peroxyl radicals. The kinetic measurements may be for concurrent reactions with different rate constants, since there are two products being formed.



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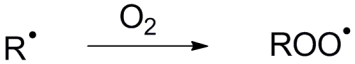


(26)

A common photolytic technique is to generate a reactive intermediate, which has the ability to abstract a hydrogen atom from a solute or the solvent to produce the desired alkyl radical.[11] It is common to implement photolysis to produce selective reactants, i.e. the photolysis of di-*tert*-butyl peroxide (DTBP) is used to generate *tert*-butoxyl radicals which subsequently react with the solvent.

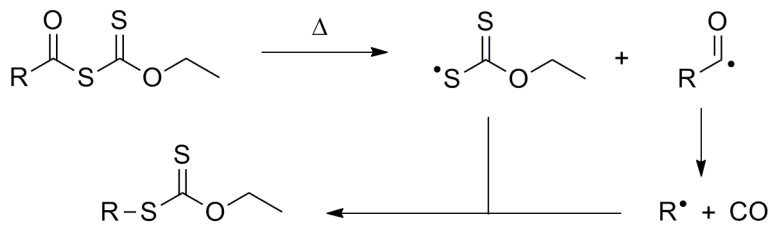
$$(\text{tert-BuO})_2 \xrightarrow{h\nu} 2\ \text{tert-BuO}^{\bullet} \quad (27)$$

$$\text{tert-BuO}^{\bullet} + \text{RH} \longrightarrow \text{tert-BuOH} + \text{R}^{\bullet} \quad (28)$$

$$\text{R}^{\bullet} \xrightarrow{O_2} \text{ROO}^{\bullet} \quad (29)$$

The *tert*-butoxyl radical is far more selective than the hydroxyl radical, thus allowing for greater control over which alkyl radical and subsequent peroxyl radical is formed. In terms of radical selectivity, tertiary radicals predominate over secondary, which in turn will predominate over primary.

Xanthates, which are classified under dithiocarbonates, exhibit very diverse chemistry and encompass mainly radical processes. Xanthates play important roles in the Chugaev elimination, but more importantly the Barton-McCombie radical deoxygenation[30] has had an important impact in organic synthesis, especially in carbohydrate chemistry, and as a convenient source of radicals from alcohols in general. Barton, George, and Tomoeda explored the photochemical behaviour of xanthates, and reasoned that since the xanthate residue has a pronounced ultraviolet absorption; it might serve as a probe for energy absorption, leading to bond fission.[31] The decomposition of an S-acyl xanthate is shown in **Scheme 1.6**, whereby an alkyl radical is formed.



Scheme 1.6: The decomposition of S-acyl xanthates in the absence of O_2. In the presence of O_2 the alkyl radical will be trapped to yield a peroxyl radical.

1.4.2 Production of Peroxyl Radicals by Pulse Radiolysis

Alkyl radicals can be generated by irradiating solutions with high energy ionizing radiation,[11] allowing for the reaction of primary radiolytic species with the solvent or solute. When exposed to radiation, water dissociates into the products shown in **Eq. 30**. In aqueous solutions e^-_{aq} or •OH radicals are responsible for radical formation by their reaction with various organic compounds. If the •OH radical is used to generate alkyl radicals, the solutions typically contain N_2O to convert the e^-_{aq} into •OH. As a result, the yield of the desired alkyl radical increases and the possibility of complications occurring are minimized since the other reactions of the electron are not favourable.

$$H_2O \longrightarrow e^-_{aq} + H^\bullet + HO^\bullet + H_2 + H_2O_2 \quad (30)$$

$$e^-_{aq} + N_2O \longrightarrow N_2 + {}^\bullet OH + OH^- \quad (31)$$

$$HO^\bullet + RH \longrightarrow H_2O + R^\bullet \quad (32)$$

$$R^\bullet + O_2 \longrightarrow ROO^\bullet \quad (33)$$

In contrast, if e^-_{aq} is used to generate the desired alkyl radical, the •OH radical cannot be converted to the hydrated electron, but instead it can be converted to a reducing radical capable of producing more of the alkyl radical. Otherwise, secondary products resulting from •OH must be taken into account in the kinetic analysis.

Alkyl radicals are commonly formed by hydrogen abstraction from the solvent or a solute, assisted by primary or secondary radiolytic products. This process can result in a mixture of alkyl radicals formed and will depend on the selectivity of the reactant radical. The hydroxyl radical is considerably non-selective and is mostly used to produce radicals from simple precursors, such as methanol, acetone, or acetonitrile, where they all yield the same alkyl radical. In the case of 2-propanol, which is a slightly more complicated, there is a preference towards alkyl radicals being secondary (86%) versus primary (13%). The hydroxyl radical can react with precursor which contain aromatic moieties, which leads can lead to more complications since the OH adduct radicals can form in addition to radicals formed by abstraction from the side chains.

1.5 Techniques for Measuring Peroxyl Radical Kinetics

1.5.1 Direct EPR (Electron Paramagnetic Resonance)

Peroxyl radicals can be detected by their optical absorption or EPR spectra and rate constants can be directly obtained using this method. Decay traces can be recorded by intercepting the UV-light used to generate the radicals with a mechanical shutter and monitoring the time-course of EPR signal intensity. For example, EPR spectroscopy has proven to be useful for studying acid-accelerated reactions of peroxyl radicals with phenols.[32] Although EPR is unsuitable for monitoring these reactions at room

temperature, Fukuzumi et al.[33] showed that the kinetics of reactions of cumylperoxyl radicals with substituted dimethylaniline derivatives and phenols can be studied in propionitrile at 193 K. By modifying the procedure slightly, Valgimigli et al. were able to measure the k_1 values directly for the reactions of cumylperoxyl radicals with various substituted phenols have been studied by monitoring the signal decay of the cumylperoxyl radical formed upon photolysis of a mixture of cumene (2M) and di-tert-butylperoxide (2M) in oxygenated propionitrile in the presence of the phenols between 193– 213 K.

1.5.2 Laser Flash Photolysis (LFP)

Peroxyl radicals are known to have weak absorptions in the UV, generally with a peak at or below 250 nm. To circumvent this problem the rates of reaction of peroxyl radicals with inorganic and organic compounds can sometimes be determined by monitoring the build-up of optical absorption of the product radical.[11] If the product radical exhibits a weak absorption at $\lambda > 250$ nm, as in the case of fatty acids, the rate constants could be determined using competition kinetics using compounds such as ABTS (2,2'-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid) or porphyrins as reference reactants.[11]

The main disadvantage of laser driven systems is that ultraviolet lasers have a fixed wavelength. Reactants in photochemical reactions can have a wide variety of absorption wavelengths, some of which may not be accessible to a given laser source. Therefore, several different types of lasers are often necessary to provide coverage of the UV range of common organic and inorganic reactants. For example, 355 nm is a

suitable wavelength for diazo compounds, but would not be ideal for ketones since they absorb around 180 nm.

Recently, it has been shown that it is possible to observe directly, in real time, the absorption of peroxyl radicals, with a suitable selection of “R groups” in a convenient spectral range (near UV/visible part of the spectrum). For example, the laser excitation of di-tert-butylperoxide in the presence of triethylamine (TEA) in an oxygen saturated solution, results in a new transient exhibiting a long lifetime (~4ms) with its absorption maximum at 380 nm.[34] Compared to the parent alkylperoxyl radicals previously observed, the TEA-$O_2^{\bullet}$ radical is characterized by the presence of a new and intense absorption band in the near UV/visible wavelength range. Although, there have been other red shifted radicals reported in literature; the ease of this approach can offer new opportunities.

1.5.3 Uninhibited Autoxidation

The oxidation of a hydrocarbon (R-H) in the liquid phase by atmospheric oxygen is shown in **Eq. 7-10**. This radical chain process includes three important steps of initiation, propagation and termination. It is essential that the rate of propagation be faster than the rate of termination[38] for this sequence to be considered a chain reaction.

Since H-atom abstraction from hydrocarbons by ground-state dioxygen ($^3O_2$) is usually very slow, autoxidations require a source of radicals for the initiation step to occur and be persistent.[14] There are various radical initiators which have been explored in kinetic studies, such as transition metals/ROOH or H_2O_2 and azo compounds.[14]

The rate of oxygen uptake in autoxidations is first order in the concentration of substrate, RH and one-half order in the rate of chain initiation, R_i. The kinetics of these reactions can also be measured by looking at the production of product as well as simply consumption of starting material, making **Eq. 34** valid.[40]

$$\frac{d[ROOH]}{dt} = \frac{-d[O_2]}{dt} = \frac{k_p}{2k_t^{1/2}[RH]} R_i^{1/2} \qquad (34)$$

For quantitative kinetic studies, the rate of initiation R_i must be controlled, for example by using a thermal azo initiator, R-N=N-R which decomposes at a known and controlled rate to form alkyl radicals known to react very rapidly with molecular oxygen to form propagating peroxyl radicals.[14] Another alternative is that R_i can be controlled by photochemical initiation. The "cage effect" on radicals generated in bilayers is known to be much greater than in homogenous solutions and the R_i must be measured, for example by the addition of a phenolic antioxidant, ArOH, known to trap two peroxyls, and by measuring time τ, during which oxygen uptake is suppressed.[14]

1.5.4 Inhibited Autoxidation

Inhibited autoxidations involve the use of a substance which can reduce the rate of autoxidation, thereby acting as antioxidants or retarders. Assuming quasi-stationary conditions, the rate of inhibition is represented by the **Eq. 35**. Typically, these experiments are monitored for the oxygen uptake for autoxidation of an organic hydrocarbon substrate in the presence of an antioxidant or a retarder over time. In kinetically controlled experiments, antioxidants can be easily recognized as simply

retarders or chain breaking antioxidants, based on the oxygen uptake profile. In these types of profiles, chain breaking antioxidants have a clear induction period, from which the stoichiometric factor can be determined (as in **Eq. 36**).

$$\frac{d[ROOH]}{dt} = \frac{-d[O_2]}{dt} = R_{O_2}^{inh} = \frac{k_p[RH]R_i}{nk_{inh}[ArOH]_0} \qquad (35)$$

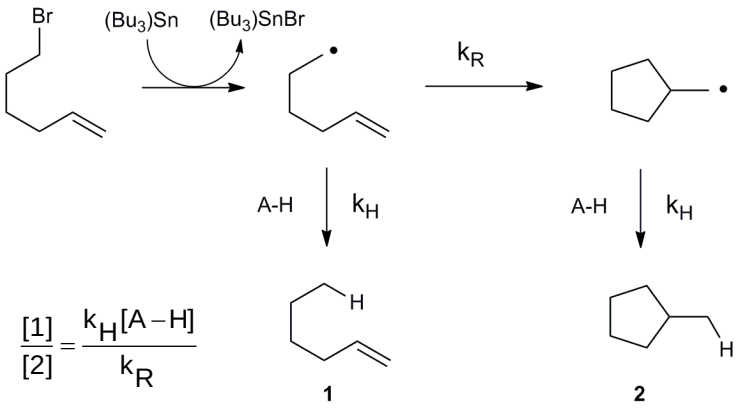
The stoichiometric factor of ArOH is calculated as follows:

$$n = \frac{R_i T}{[ArOH]_0} \qquad (36)$$

Where T and $[ArOH]_0$ are the induction period and the initial concentration of inhibitor, respectively. The product R_i x T gives the moles/litre of peroxyl radicals released by the initiator in the time, T. Therefore, the value of n represents the number of oxidation chains terminated by one molecule of inhibitor. Hence, n is generally found to be close to 1.0 or 2.0 for various phenols.[14]

1.5.5 Radical Clocks

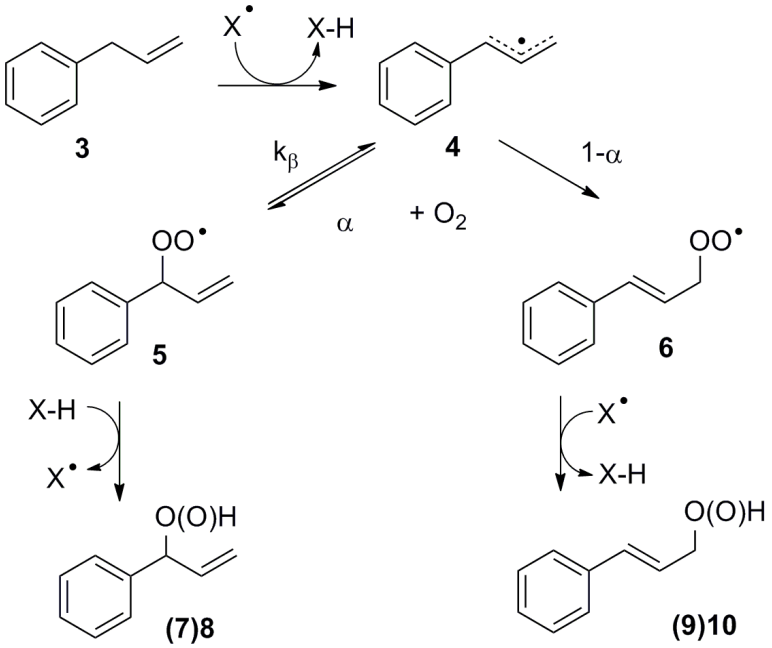
Radical clocks are an indirect method for determining radical-molecule reaction kinetics by competition between a unimolecular reaction having a known rate constant and a bimolecular reaction with an unknown rate constant. The 5-hexenyl radical cyclization is an example of a radical clock, which involves a competition between a 5-*exo-trig* cyclization (k_R) and abstraction of a hydrogen atom from a substrate A-H (k_H). From a kinetic analysis of the product ratio [1]/[2], is directly proportional to k_H/k_R.



Scheme 1.7: 5-Hexenyl Radical Cyclization Clock

The radical clock method[41] is ideally suited to applications involving carbon-centered radicals due to the vast literature documenting the kinetics of skeletal rearrangements of alkyl radicals. Dozens of clocks have been used to for determining reaction rate constants ranging from 10^{-1} to 10^{12} M^{-1} s^{-1}.[42] It is necessary that radical clocks must be "standardized" to an absolute rate constant. In the case of radical cyclization, a number of methods have been used to calibrate various clocks, including the rotating sector method, flash photolysis, pulse radiolysis and inhibited autoxidation.[41]

A peroxyl radical clock approach was first suggested by Porter and co-workers in 2006,[41] involving an indirect method for determining peroxyl radical-molecule reaction kinetics by competition between the β-fragmentation of a non-conjugated peroxyl radical and a bimolecular reaction to trap the peroxyl with an unknown rate constant (refer to **Scheme 1.8** below).



Scheme 1.8: The Kinetically Controlled Oxidation of Allylbenzene

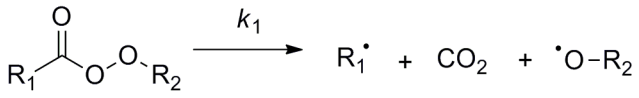
The mechanism for the autoxidation of allylbenzene (**Scheme 1.8**) proceeds by H-atom abstraction from the benzylic position to yield an α-vinylbenzyl radical (**4**), which is trapped by O_2 generating the nonconjugated and conjugated peroxyl radicals **5** and **6,** respectively. The fraction of α-vinylbenzyl radicals trapped at the benzylic position and leading to the non-conjugated peroxyl is defined as α, analogous to the partitioning of O_2 at the central position of the nonconjugated dienes.[41] Similarly, this nonconjugated peroxyl radical (**5**) undergoes *β*-fragmentation (k_β) in competition with H-atom transfer.

This approach has been demonstrated for determining rate constants for H-atom transfer (HAT) from simple phenols to peroxyl radicals in benzene,[41] however, it has one major limitation. It requires that X•, the radical derived from X–H, carry the oxidation chain reaction. This can be problematic for studying phenols that yield either persistent

(e.g. butylated hydroxy toluene, BHT) or highly stabilized (e.g. 6-amino-3-pyridinols)[10] radicals or other X–H that give rise to radicals that have difficulty carrying the chain (e.g. aromatic amines). Furthermore, because the chain-propagating reaction is so slow, a large concentration of substrate is required (e.g. 2.6 M of **3**) to ensure that sufficient oxidation of products occurs.[41] However, with such a large concentration of the substrate, it is difficult to study the kinetic solvent effect (KSE) on these types of reaction systems. This has severely limited studies of lipid peroxidation processes under kinetically-controlled conditions.[41] This limitation has resulted in an incomplete picture of the product distribution arising from the oxidation of polyunsaturated lipids and is likely of the reasons why the origin of many lipid derived toxins produced in vitro and in vivo are unknown. Furthermore, fundamental concerns such as what controls the rate of oxygen addition to alkyl radicals and fragmentation from peroxyl radicals have remained to not be fully understood.

Peroxyesters as Precursors for Radical Clocks

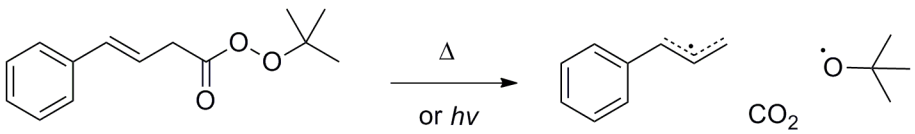
Peroxyesters $R_1C(O)OR_2$ are known for their weak O-O bond, making them ideal initiators of radical processes for chemical reactions in the laboratory.[43] The decomposition of peroxyesters is believed to occur through the concerted cleavage of two bonds (for stabilized R_1), leading to an alkyl and alkoxyl radical. The decomposition is characterized by the following parameters[43]:



(37)

1) The activation energy for the decomposition (30 ± 2 kcal/mol) is close to the O-O bond strength and depends slightly on the substituents R1 and R2.
2) The activation entropy is in the range of 36 - 54 J mol^{-1} K^{-1}.
3) The rate of decomposition (k_1) depends on the solvent viscosity. The cage effect will play an important role if the solvent viscosity is increased. This will result in a lower rate of decomposition (k_1).

Peroxyesters are generally decomposed under anaerobic conditions to serve as sources of carbon-centered radicals to initiate radical reactions; in the presence of O_2, it can serve as a precursor to a peroxyl radical clock.[44] Jha and Pratt have shown that the decomposition of peroxyesters, such as compound **9**, can generate a carbon centered radical which is analogous to the one shown in **Scheme 1.8**, by Porter and co-workers.



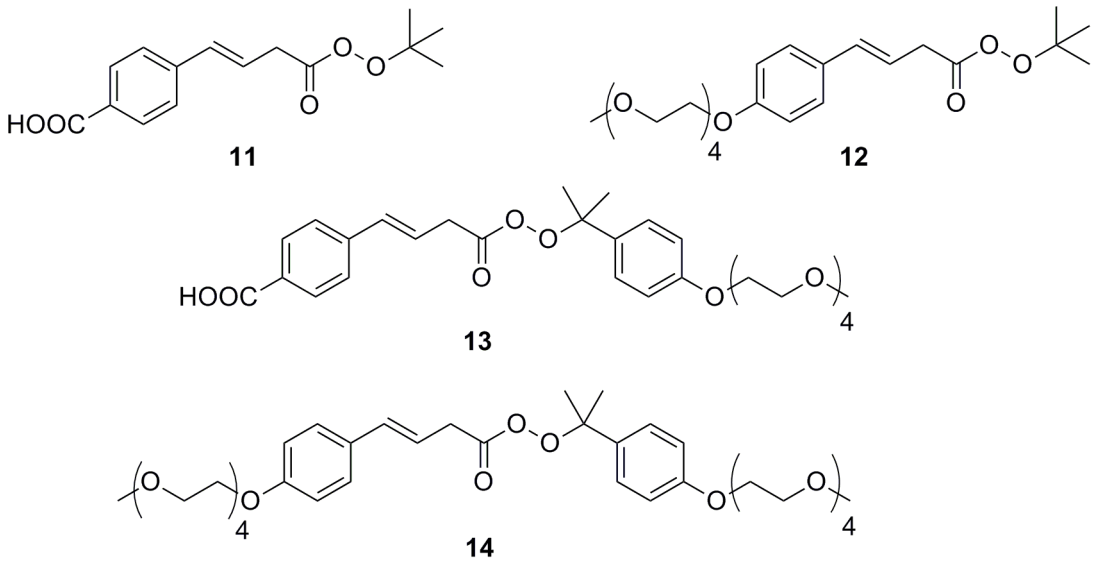
9

Under thermolytic or photolytic conditions, peroxyester **9** can decompose to give rise to the carbon centered radical **4**, which is oxygenated to produce the non-conjugated **(5)** and conjugated **(6)** peroxyl radicals in **Scheme 1.7**. This approach has been more feasible for determining the kinetic solvent effects on the β-fragmentation of peroxyl radicals, which in turn, allows the determination of KSEs on the reactions of radical trapping antioxidants with peroxyl radicals.[44]

The decomposition of perester **9** has been studied under thermolytic (37°C, 4 h) aerobic conditions in the presence of a good H-atom donor (e.g. α-tocopherol, α-TOH), yielding the same non-conjugated and conjugated hydroperoxides **(5 and 6)** as the method reported by Porter and co-workers in the kinetically controlled autoxidation of **3**.[44] The decomposition of **9** (analyzed by GC following PPh_3 reduction of **7** and **8**, respectively) was found to be dependent on the concentration of α-TOH. The rate constant for β-fragmentation of **5** was determined to be k_β = 1.7 (±0.1) x 10^5 s^{-1} at 37 °C using k_H = 3.5 x 10^6 $M^{-1}s^{-1}$ at 25 °C and α = 0.76 ± 0.01 , in good agreement with that determined in the autoxidation of allylbenzene at 37 °C (k_β = 2.6 (±0.3) x 10^5 and α = 0.74 ± 0.12 (at 37 °C).[44] Overall, this competitive kinetic approach is appealing as it allows the facile determination of rate constants for H-atom (PCET) reactions between peroxyl radicals and reductants (e.g. phenolic and aromatic amine antioxidants) without the need for specialized equipment. Furthermore, this technique allows us to probe medium effects since only a small amount of the peroxyester is necessary to generate sufficient products to obtain kinetics.

1.6 Objectives

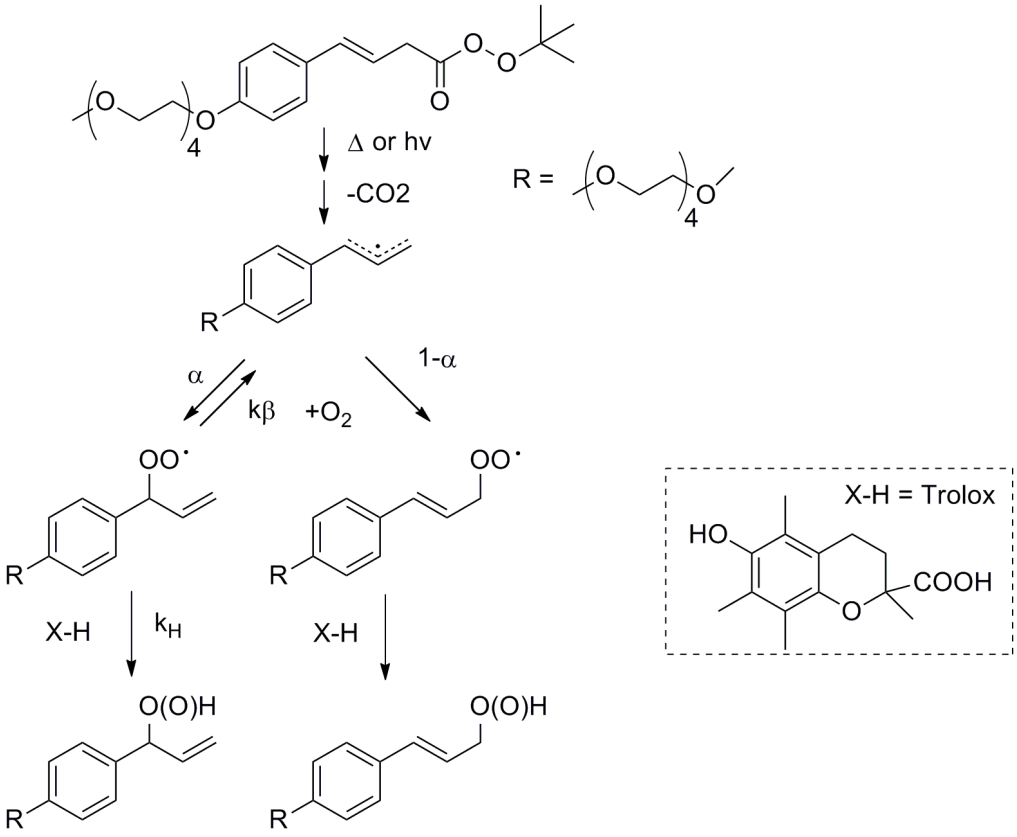
The primary objective of this project is to develop water-soluble peroxyl radical clocks, which extend the range of media in which the competitive kinetic approach is applicable to include aqueous media – by using either carboxylated peroxyester **11** and/or **13** or peroxyesters substituted with poly(ethylene glycol) oligomers, as in **12** and/or **14**. The use of peroxyesters as radical clocks can be applied to measure the rate of peroxyl radicals with water soluble H-atom donors.



Scheme 1.9: Target compounds initially proposed

Once synthesized, the water-soluble peroxyesters will be decomposed in aqueous media, and the intermediate peroxyl radicals trapped using the water-soluble analog of α-tocopherol, Trolox, for which a rate constant for reaction with peroxyl radicals can be estimated to be 1.4 x 10^6 $M^{-1}s^{-1}$. By examining the ratio of non-conjugated to conjugated hydroperoxides formed as a function of the concentration of

Trolox, we can determine the beta fragmentation rate of the non-conjugated peroxyl radical in water. With this in hand, we will be able to determine the kinetics of the reactions of other substrates with peroxyl radicals in water.

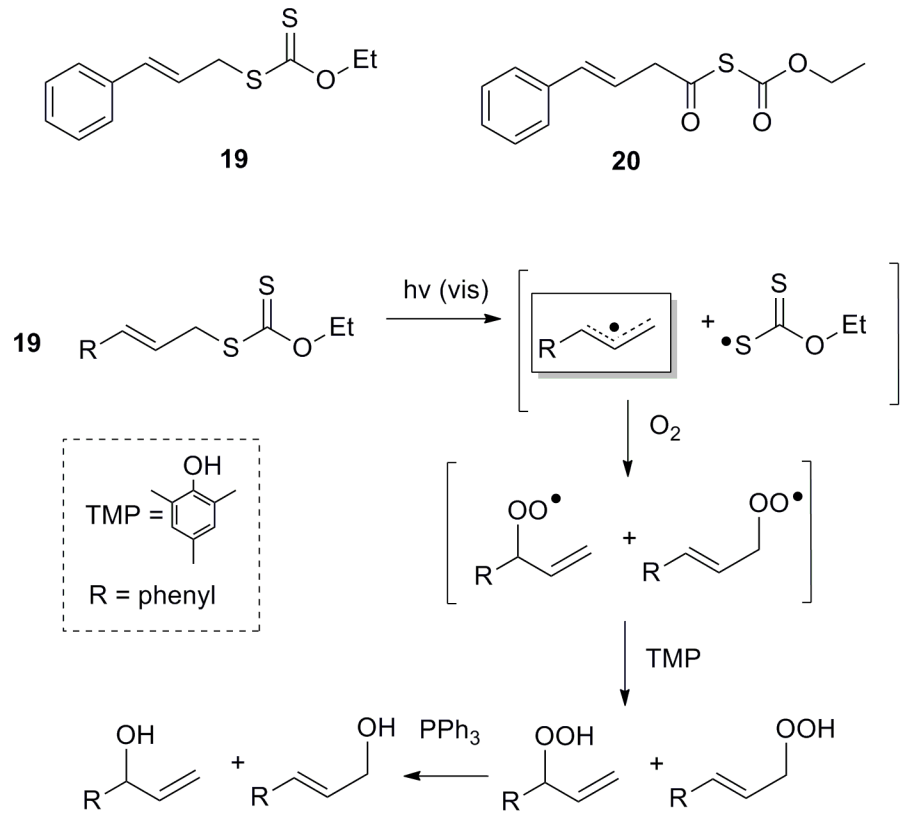


Scheme 1.10: The decomposition of a PEGylated water-soluble clock in water.

Xanthates as Radical Clocks

Taking into consideration the fact that xanthates decompose well under photolytic conditions, we were interested in using them as radical clocks to see if we could improve product yields in our decomposition experiments. We were interested in

studying the decomposition of an alkyl and acyl xanthate (compound **15** and **16**). In the case of the alkyl xanthate, the decomposition scheme is shown below.



Scheme 1.11: The decomposition pathway and quenching experiment of an alkyl xanthate.

Chapter 2

Results and Discussion

2.1. Efforts Towards a Carboxylated Peroxyl Radical Clock

Our first synthetic target was a peroxyester analogous to the 4-phenyl-perbut-3-enoate previously studied, but with a ring carboxylate (shown below, compound **11**) since a carboxy group appeared to be a simple way to improve water solubility. Four convenient routes were envisioned. The first involved a Stille coupling of a vinyl di-organotin precursor and methyl 4-bromobenzoate, the second also a Stille coupling, but with a vinyl organotin precursor and methyl 4-bromobenzoate, the third a Sonogashira reaction of methyl 4-bromobenzoate with 3-butynol, and finally a Wittig reaction with 4-formyl methylbenzoate and (2-carboxyethyl)tri-phenylphosphonium bromide

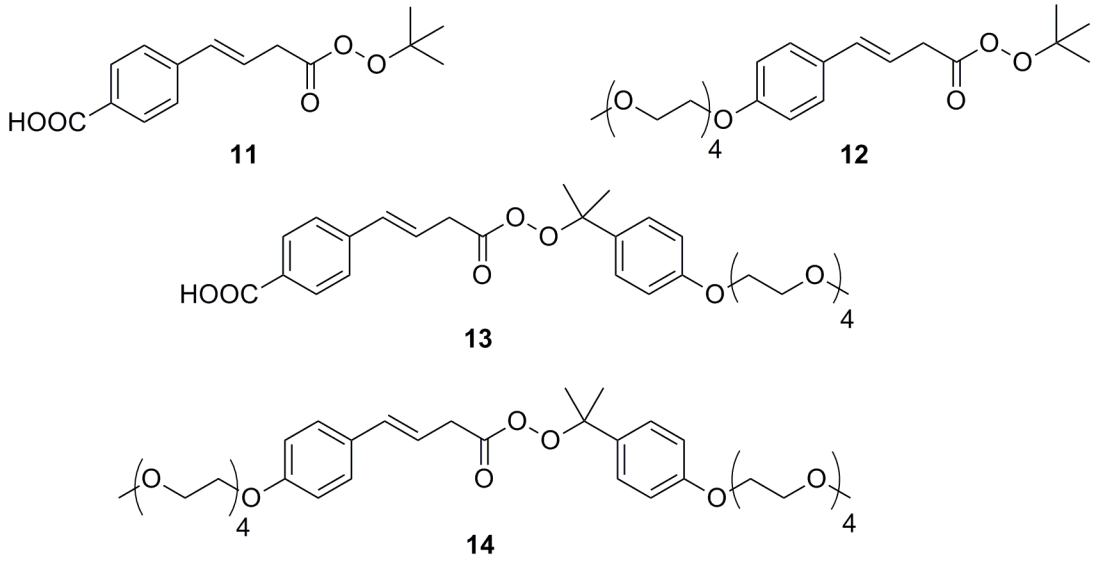


Figure 2.1: Proposed Target Compounds

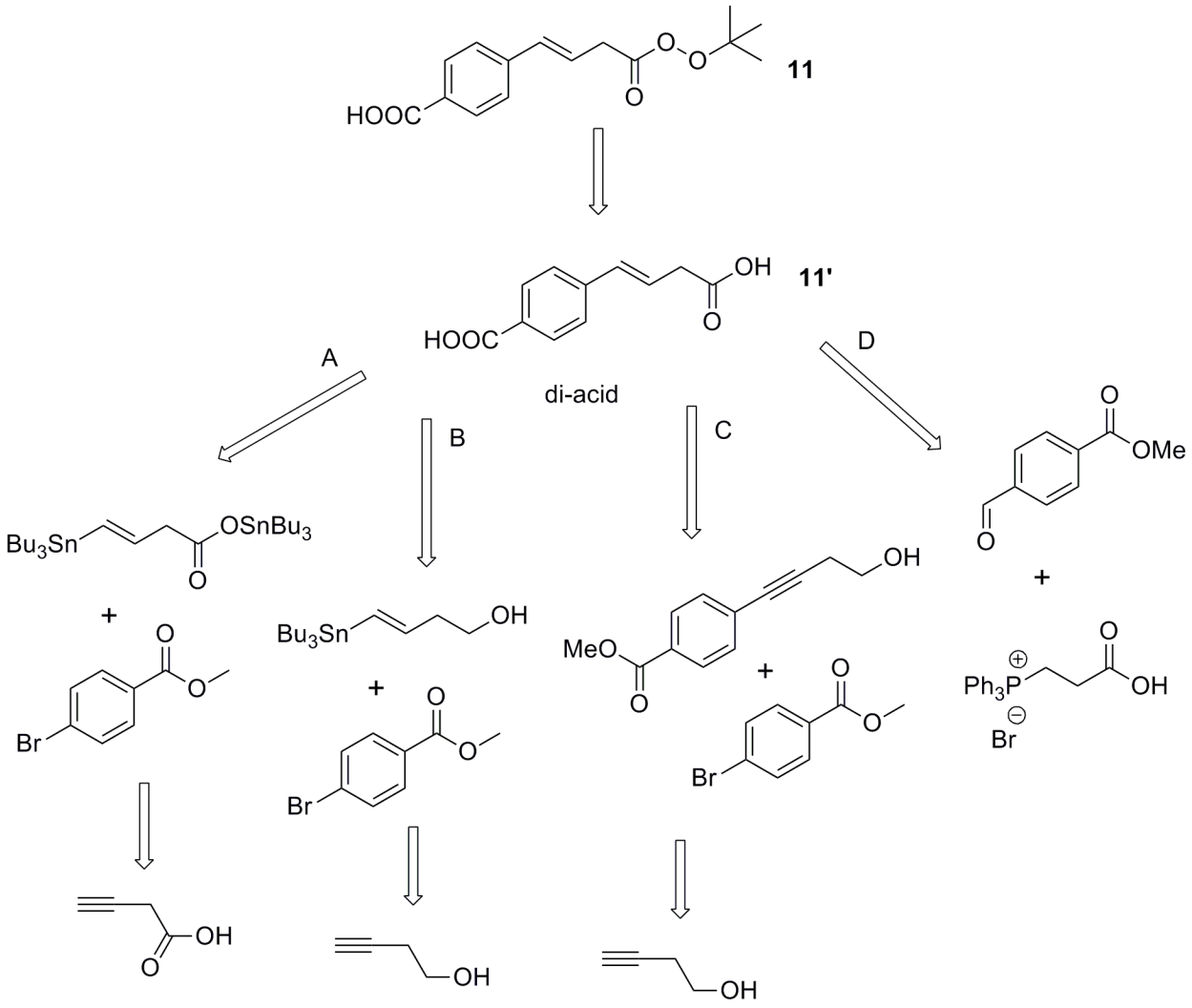


Figure 2.2: The retrosynthetic pathway towards the diacid **11'**

2.1.1. Stille Route A to the Diacid **11'**

The first approach that we took towards synthesizing the di-acid was a route involving the Stille reaction as shown in **Figure 2.2**. The reaction sequence involves a Jones oxidation, hydrostannation, Stille reaction, and lastly, hydrolysis

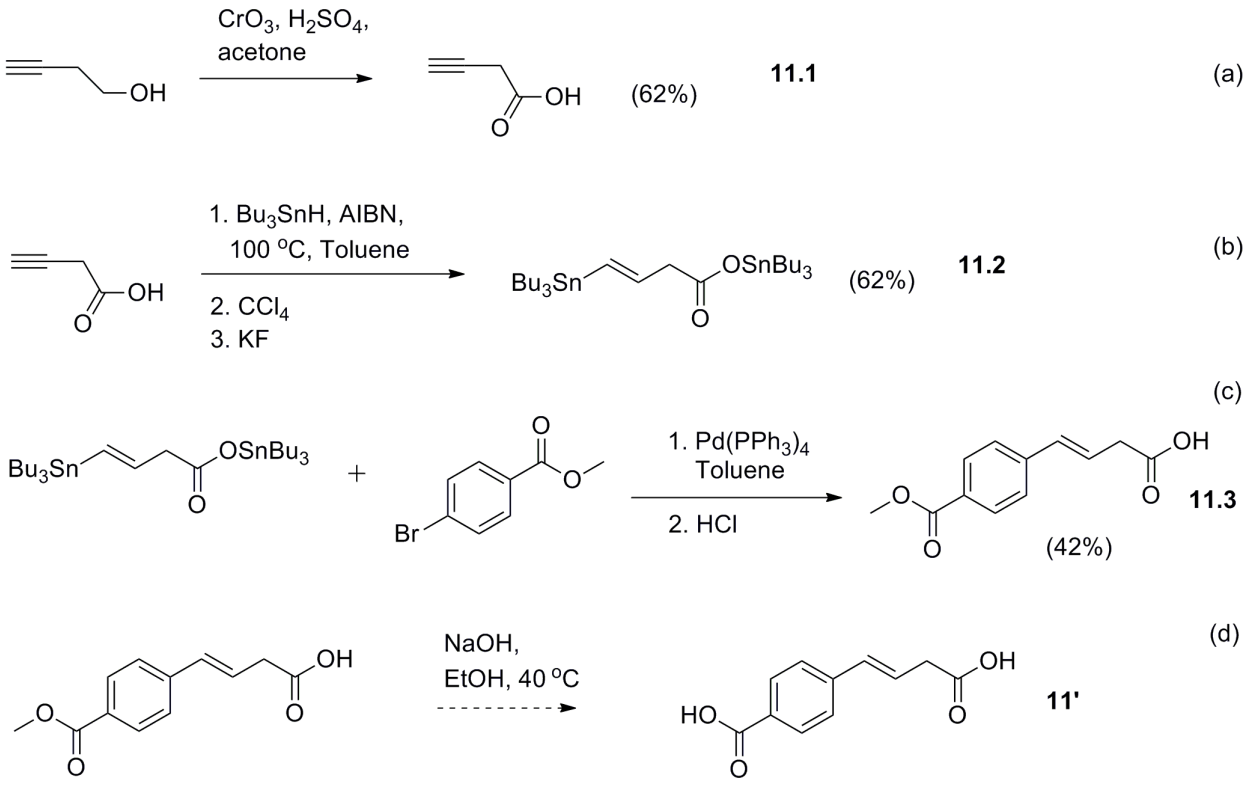


Figure 2.3: Stille Route A to the diacid **11'**

The Jones oxidation is useful for converting primary and secondary alcohols with chromic acid to carboxylic acids and ketones, respectively.[45] The reaction was discovered in 1946 with E.R.H. Jones and his colleagues, when they converted alkynyl carbinols with chromic acid (CrO_3 and dilute sulphuric acid) to the corresponding ketones without oxidizing the triple bond. Jones oxidations are typically carried out in

acetone, since it dissolves most substrates and it reacts with any excess oxidant so it protects the product from being oxidized further. In addition, excess of the oxidant is not recommended since it can oxidize other functional groups. The Jones oxidation does not affect double or triple bonds, but α, β-unsaturated double bonds can undergo isomerization.

The Jones oxidation on 3-butynol proceeded fairly well according to a procedure reported in literature,[46] and afforded a yield of 62%. The reaction was complete within 2 hours (by TLC) and purified by an acid/base wash. This method of purification is effective for isolating the carboxylic acid, since the alcohol starting material is easily removed.

The second step in the synthesis is preparation of the vinyl di-organotin reagent, obtained from the radical hydrostannation of 3-butynoic acid (shown in **Figure 2.3 (b)**). Hydrostannation reactions of alkenes and alkynes which involve stannyl radicals have been explored thoroughly, and are often tolerant of a wide variety of functional groups and can be used for preparing functionally-substituted organotin compounds. Hydrostannation of alkenes and alkynes are typically carried out at 80-110°C with azobis(isobutyronitrile) (AIBN), a radical initiator.

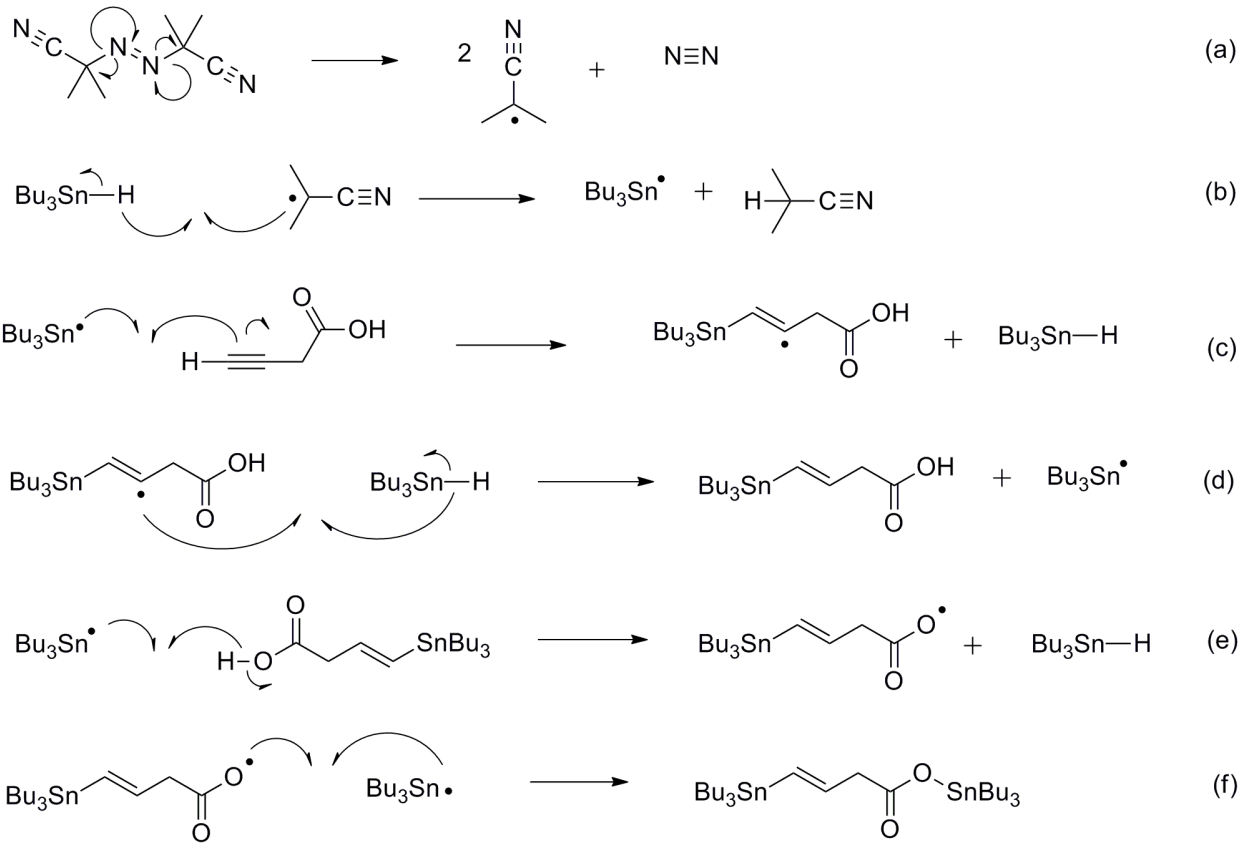


Figure 2.4: The hydrostannation of 3-butynoic acid

Note that in this mechanism (**Figure 2.4**), AIBN is the initiator and decomposes readily to form two 2-cyanoprop-2-yl radicals which can then initiate Bu_3SnH to react with the substrate. The stereochemistry of step (c) of **Figure 2.4** is undefined, however under the reaction conditions there was a preference for the Z isomer (Z:E, 85:15). Since there is an excess of Bu_3SnH used in the reaction (2.4 eq.), the second equivalent repeats the cycle, generating another Bu_3Sn•, which reacts with the acyloxy radical in step (f), the radical termination step which generates the desired product.

The hydrostannation reaction went to completion within 3 hours using the procedure described by Duchêne et. al.[47] Excess Bu_3SnH was quenched by reacting with carbon tetrachloride, followed by KF to convert the unreacted Bu_3SnH to Bu_3SnF, which is filtered off. After purification by column chromatography, the product was isolated with a yield of 62%.

A Stille cross coupling reaction was carried out with the vinyl di-organotin precursor (**Figure 2.3 (c)**). The reaction involves a Pd(0)-catalyzed coupling reaction between an organostannane and an organic electrophile to form a new C-C σ bond.[45] The organotin precursor has many advantages; they are tolerant to many functional groups and not sensitive to moisture or oxygen unlike most organometallic compounds.[48] However, the biggest disadvantages are that they are highly toxic and difficult to remove traces of the by-products of the reaction mixture.[48]

Originally, the reaction was attempted with p-bromobenzoic acid as the coupling partner with the vinyl di-organotin. The reaction did not seem to proceed to an appreciable extent, and hence a new coupling partner, methyl 4-bromobenzoate was used. This substrate worked better, and the reaction was complete within 12 hours at 100°C.

The Stille cross coupling reaction operates through a catalytic cycle and has three steps: 1) oxidative addition; 2) transmetallation; and 3) reductive elimination. The active catalyst is believed to be a 14-electron Pd(0) catalyst. The cross-coupling is achieved through the reductive elimination step, and upon workup the final product is obtained by hydrolysing the stannyl ester with 1N HCl. Although the crude NMR (after

workup) indicated that the reaction was successful, there was some difficulty in removing alkyl based tin by-products by column chromatography.

The final step towards making the di-acid is hydrolysis of the methyl ester as shown in **Figure 2.3 (d)**. A crude sample of the methyl protected di-acid was used for the hydrolysis, since pure material could not be obtained. The hydrolysis reaction did not go to completion, and it still remained a challenge to remove the tin-derived by-products from the small amount of product that was formed.

2.1.2 Stille Route B to Diacid **11'**

We were interested in improving the Stille reaction by changing the vinyltin precursor. In order to do this the following synthetic route was explored:

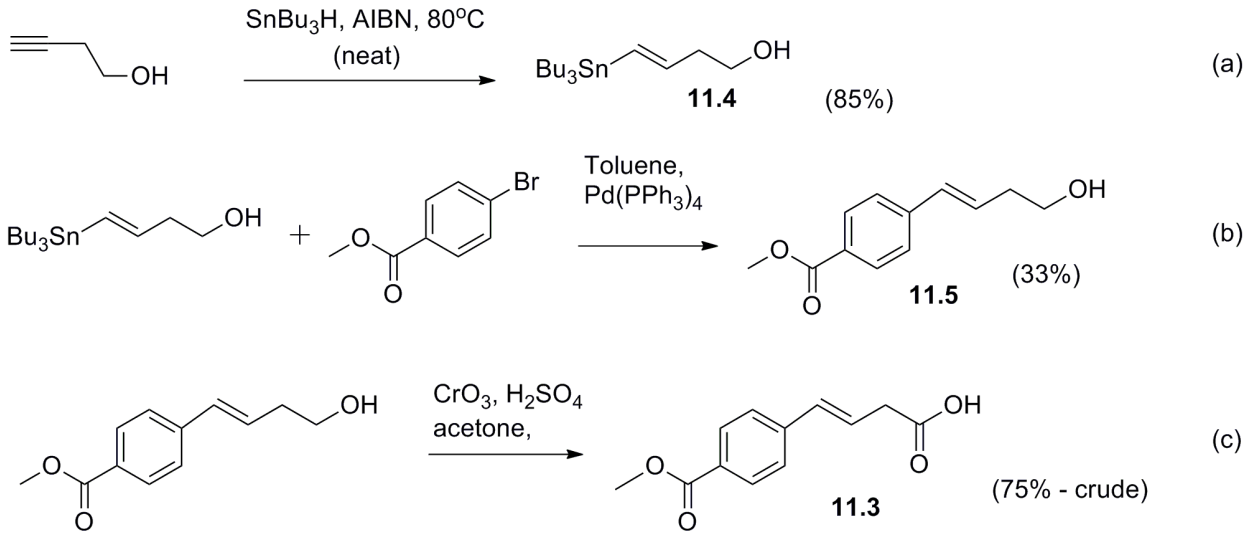


Figure 2.5: Stille Route B to the Diacid **11'**

This second Stille route involved the formation of the mono-vinyl tin compound through a hydrostannation, which has been reported in literature.[49] The reaction went to completion within 8 hours and was purified by column chromatography, affording a yield of 85%. The second step was the Stille cross-coupling, which was carried out with methyl 4-bromobenzoate. This reaction worked better in comparison to the analogous Stille reaction which was carried out on with the vinyl di-organotin reagent, discussed in **Section 2.1.1**.

This route seemed to be very attractive since it involved a mono vinyl tin precursor, and we hoped that this would mean that less of the tin-derived by-products would contaminate the product, which was the case since a pure yield of 33% was obtained. The next step in the synthesis was the Jones oxidation to form the methyl protected di-acid (**Figure 2.5 (c)**). The reaction was left to stir for 4 hours, but did not entirely go to completion. When the reaction was repeated again, it was left to stir overnight, however the amount of product formed did not change. An attempt to purify the product by column chromatography was made, however, the R_f for the starting material and product were too similar, making the separation impossible under various conditions.

2.1.3 Sonogashira Route to **11'**

We also explored a route which involved a Sonogashira cross-coupling reaction to build the framework, followed by a reduction and an oxidation to make the di-acid. Since the Stille cross-coupling reactions previously described proved to be difficult in purifying, we decided to abandon the tin route and focus on a "cleaner" approach. Furthermore, this

synthetic route appeared to be more attractive since it was one step less compared to the Stille pathways. **Figure 2.6** shows the Sonogashira synthetic pathway towards the di-acid.

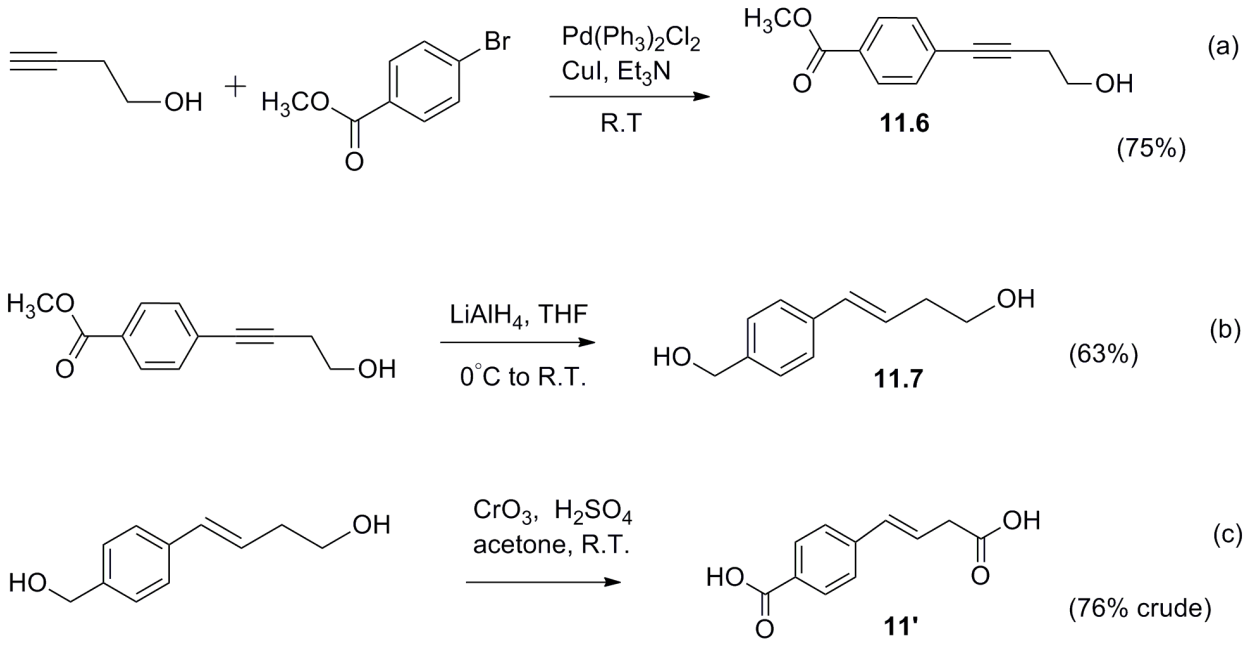


Figure 2.6: Sonogashira to the diacid **11'**

The Sonogashira reaction involves the palladium catalyzed coupling of terminal alkynes with aryl and vinyl halides.[45] Some general features of the reaction are that they can be carried out at room temperature and catalytic amounts (0.5-5 mol%) of copper (I) usually applied to activate the alkyne. For the reaction to work best, $Pd(PPh_3)_2Cl_2$ or $Pd(PPh_3)_4$ should be used and thorough deoxygenation is necessary to maintain the activity of the Pd catalyst.

The Sonogashira reaction worked very well under the conditions[50] shown in **Figure 2.6 (a)**. The reaction went to completion within 24 hours and there was no need

to add additional catalyst. A yield of 75% was obtained, which was encouraging compared to the poor yields obtained from the Stille cross coupling reactions.

The second step involved reduction of the Sonogashira product to a diol (**Figure 2.6 (b)**). This was accomplished by using lithium aluminum hydride, where the first 2 equivalents were used for reducing the ester and another 1.5 equivalents was used for reducing the alkyne to the trans-alkene. The reduction of propargylic alcohols with lithium aluminum hydride has been reported in literature.[51] It has generally been accepted that the reduction proceeds through a hydride transfer from the aluminum bound to oxygen to the β-carbon of the triple bond, leading to the trans olefin.[51] After workup, and column chromatography, the pure diol afforded a yield of 63%.

The final step in this synthetic route was conversion of the diol to the di-acid. This was initially attempted by a Jones oxidation, however it appeared the reaction did not go to completion with excess oxidant. Other attempts at this oxidation were made, such as using Na_2CrO_7 (3 mol %), HNO_3 (40 mol%), and $NaIO_4$ (4.4 eq) at 0°C for 8 hours. In both cases, the product was too difficult to isolate as a result of various aldehyde impurities (from incomplete oxidation).

2.1.4 Wittig Route to Diacid **11'**

The Wittig reaction appeared to be a useful approach since it was a single step from commercially available starting materials. The Wittig reaction has several advantages over other olefination methods; in particular, it occurs with total positional selectivity (i.e., a carbonyl group is converted into an alkene). Furthermore, the factors which effect E and Z stereoselectivity are well understood and can be controlled by using the

appropriate phosphorus reagent and reaction conditions.[52] There are a wide variety of phosphorus reagents that can be used for Wittig reactions, and the nature of these reagents makes it convenient for the Wittig reaction to be divided into three main classes: the “classic” Wittig reaction of phosphonium ylides, the Horner-Wadsworth-Emmons reaction of phosphonate anions, and the Horner-Wittig reaction of phosphine oxide anions.

The nature of the ylide and carbonyl compound can have a profound effect on the products observed in a Wittig reaction. Stabilized ylides are those that possess an R group that is anion stabilizing/electron-withdrawing. These types of ylides tend to be less reactive compared to other ylides and usually only react with aldehydes to give the E-alkene; a result of thermodynamic control. Consequently, the less crowded and hence favoured trans-oxaphosphatane gives rise to the observed E-alkene. Other factors that favour the E-alkene are aprotic reactions conditions, and addition of a catalytic amount of benzoic acid. Some factors that disfavour E-alkene geometry are use of α-oxygenated aldehydes or methanol as a solvent as well as using lithium and magnesium salts in DMF as solvent.[52]

In contrast, non-stabilized ylides possess an R group that is anion destabilizing/electron donating (e.g. alkyl groups). Reactions of these ylides with an aldehyde or ketone are generally under kinetic control and favour the Z-alkene as a product. Some factors that favour the Z-alkene are bulky and/or aliphatic aldehydes, bulky phosphorus ligands (phenyl ligands), lithium free conditions/low temperatures and hindered ylides. Factors that disfavour the Z-alkene include: small phosphorus ligands,

cyclohexyl ligands, cyclic phosphorus ligands and aromatic or α, β-unsaturated aldehydes.

The nature of the reactant carbonyl group in the substrate can influence the outcome of the Wittig reaction; for example, primary aliphatic aldehydes favour Z-alkenes, while aromatic or α,β-unsaturated aldehydes tent to reduce the Z-selectivity, especially in polar aprotic solvents. A higher Z-selectivity is also observed with aryl alkyl ketones compared to unsymmetrical dialkyl ketones.

Phosphonium salts are usually prepared from triphenylphosphine, and the phosphorus ylides are generated before the reaction or in situ.[52] Strong bases (such as BuLi) under inert conditions are required to make non-stabilized ylides, whereas weaker bases (i.e. alkali metal hydroxides in aqueous solution) can be used to make stabilized ylides. Furthermore, the ylides are water and oxygen sensitive. In terms of chemoselectivity, the phosphorus ylides react fast with aldehydes, slower with ketones and other carbonyl compounds (such as esters and amides) remain unchanged.[52]

Originally, the Wittig reaction was attempted based on a literature procedure by Maryanoff and co-workers[53]. They were interested in studying the stereochemistry of Wittig reactions and their effect of nucleophilic groups in the phosphonium ylide. They documented a Wittig reaction which was carried out with (2-carboxyethyl)tri-phenylphosphonium bromide, benzaldehyde and LiHMDS, however, no isolated yield was reported. Nonetheless, we attempted a few Wittig reactions (summarized in **Table 2.1**), replacing the benzaldehyde with 4-formyl benzoic acid, and later p-methoxy benzaldehyde (entry 5, **Table 2.1**).

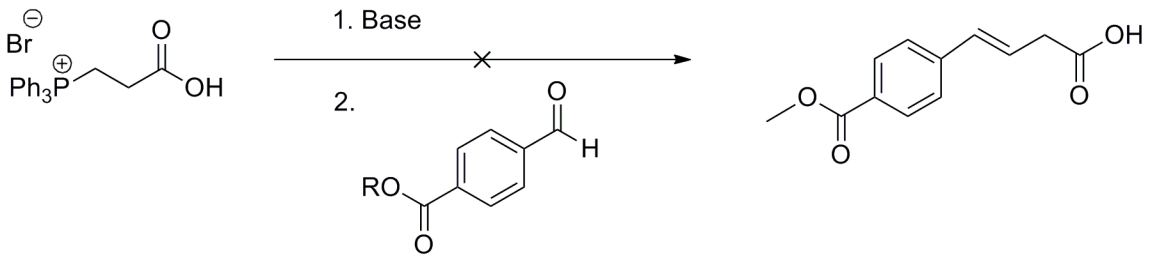


Figure 2.7: General scheme for the Wittig reaction towards making the methyl protected di-acid. R = H

The conditions in the Maryanoff paper were reproducible for benzaldehyde, however when the conditions were applied to 4-formyl benzoic acid, no product was formed. We attempted using different bases however, none of the reaction conditions were suitable to make the di-acid directly. The reaction appeared to work when p-methoxy benzoic acid was used, however some of the cis olefin was also observed (0.82:0.18 E:Z).

Table 2.2: Conditions for the Wittig Reaction towards the preparation of the di-acid

Entry	A (eq)	R	PS (eq)	Base (eq)	Solvent(s)	Temp (°C)	Comments
1	1	H	1.5	LiHMDS (3.5)	THF	0°C (ylide)→ rm temp 12 h	S.M. was recovered
2	1	H	1.5	K^{t}OBu (3.5)	THF	0°C (ylide)→ rm temp 12 h	S.M. was recovered
3	1	H	1	NaH (3.5)	DMSO	80°C (base) →10 °C (PS + aldehyde→rm temp 21 h.	S.M. was recovered
4	1	H	1.1*	NaH (2.2)	THF and N-methyl-pyrrolidone	RT	S.M. was recovered
5	0.8	~CH_3	1*	LiHMDS (2.2)	THF	RT	Trace amounts of product formed.

A = Aldehyde, PS = phosphonium salt, * = recrystallized material was used ,~ = 4-formyl methylbenzoate was used

In an effort to prevent the phosphonium salt from being deprotonated at the carboxyl terminus, we decided to synthesize an ethyl protected phosphonium salt. The phosphonium salt was made by the refluxing ethyl 3-bromopropanoate with triphenylphosphine in acetonitrile. However, when the new phosphonium salt was substituted into the reaction, the reaction did not work and unreacted starting material (aldehyde), the hydrate and hydrolyzed phosphonium salt was recovered.

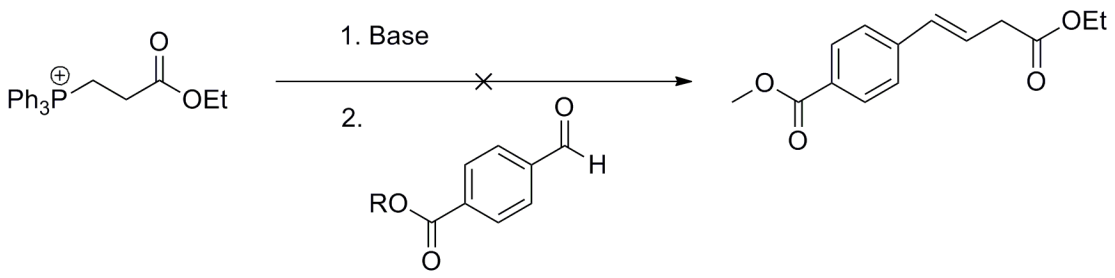
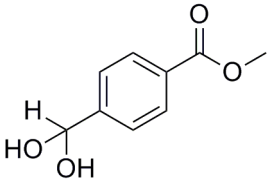


Figure 2.8: General scheme for the Wittig reaction conditions towards the protected di-acid

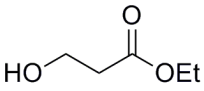
Table 2.3: Modified Wittig reaction conditions

Entry	Aldehyde (Eq)	(PS)	Base (Eq)	Solvent(s)	Temp (°C)	Comments
1	0.8	1.0	LiHMDS (2.2)	THF	RT 12 h	S.M., hydrate and hydrolyzed PS were recovered
2	1.0	1.0	K^{t}OBu (2.0)	THF	RT 12 h	Same as above
3	1.0	1.0	K^{t}OBu (2.0)	Benzene/ THF	RT 12 h	Same as above

Hydrate:



Hydrolyzed Phosphonium Salt:



An alternative procedure was reported by Minami and co-workers [54], whereby they synthesized a variety of (*E*)-4-(Substituted Phenyl)-3-butenoic acids. The main difference in their experimental procedure was that they specifically used KO^tBu as a base and changed the order of addition. The procedure involved stirring the aldehyde and phosphonium salt in THF at 0°C and adding KO^tBu dropwise to the reaction.

The di-acid was directly attempted using 4-formyl benzoic acid, however no product was isolated. Although a variety of different substituted benzaldehydes were tested, this compound was not reported by Minami and co-workers. We then attempted the reaction with 4-formyl methyl benzoate, which gave a reported yield of 23%.[54] We discovered that this prep worked very well on this substrate, with triphenylphosphine oxide being the major contaminant. The product obtained had a yield of 41%, considerably better than that reported in literature. The purification proved to be challenging, as triphenylphosphine oxide co-eluted with some of the product after column chromatography. Hence, it was difficult to obtain a higher yield for this reaction.

In the final step, the di-acid was hydrolyzed using sodium hydroxide in ethanol at 40°C. Although the reaction did not go to completion after several hours, it was worked up and the product was isolated by column chromatography, affording a yield of 56%. Hence, the Wittig reaction route appeared to be the most successful and shortest for obtaining the di-acid.

Since these synthetic routes proved to be very challenging, and there was insufficient time and material available to optimize the peroxyester synthesis using the di-acid, this approach was abandoned.

2.2 Efforts Towards a PEGylated Peroxyl Radical Clock

2.2.1 Synthesis of PEGylated Peroxyester **12**

The introduction of poly(ethylene glycol) (PEG) substituents on substrates is a popular strategy for increasing their water solubility and is commonly used for biotechnical and biomedical applications. PEG with molecular weights less than 1000 g/mol are viscous colourless liquids.[55] PEG is usually made by an anionic initiation process with few chain-transfer and termination steps, thus the molecular weight distributions are generally narrow. However, the commonly used monomethyl ethers of PEG have a broad molecular weight distribution because of the presence of high molecular weight PEG (i.e. the polymer with two hydroxyl terminal groups). This PEG is made from trace hydroxide acting as an initiator, and since this difunctional polymer grows at both ends, it has a higher molecular weight than the monomethyl ether, which grows at only one end.[55] In terms of selecting a PEG compound suitable for our synthesis, we decided to use tetraethylgylcol(monomethylether) since it was commercially sold at an affordable price in high purity (98% GC); most importantly, it would give the same water solubility as other PEG compounds.

PEGs exhibit a wide variety of interesting solubility properties as they are soluble in water and in many organic solvents, including toluene, methylene chloride, ethanol, and acetone; hence PEG is often considered to be amphiphilic. PEG is insoluble in hexane and similar aliphatic hydrocarbons and in ethyl ether, ethylene glycol, molecules which closely resemble PEG. Most importantly, PEG has the ability to solubilize other molecules when covalently attached to a compound.

In order to incorporate a PEG group into the target compound proposed, we decided to take the following synthetic route:

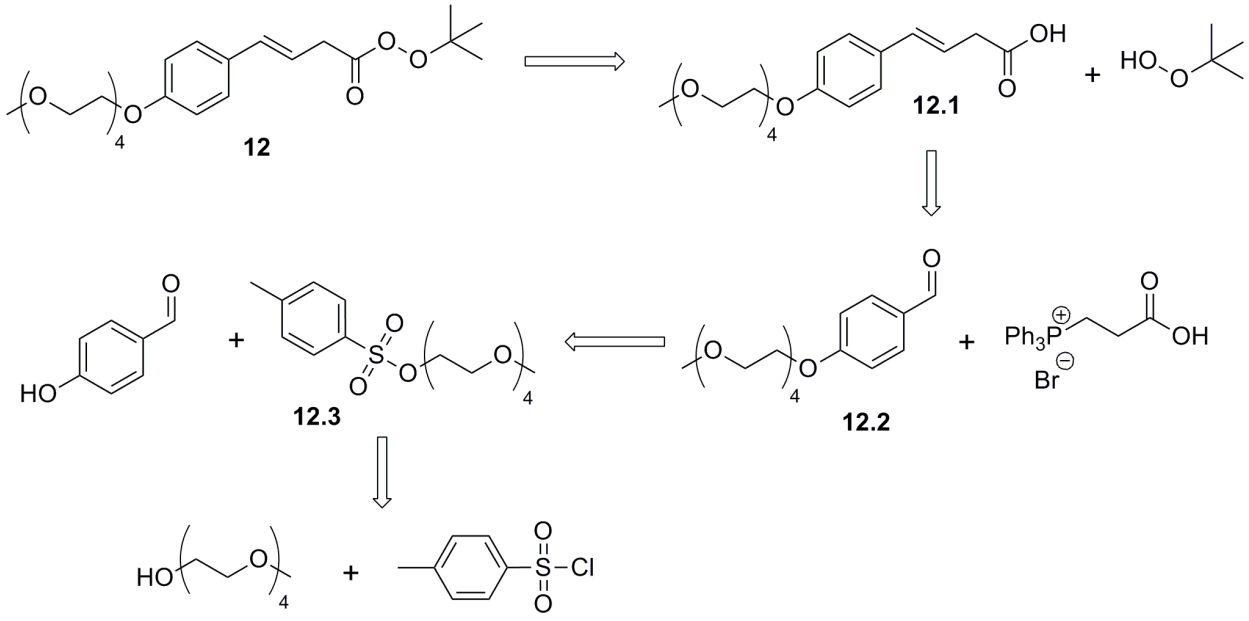


Figure 2.9: Proposed synthetic route for the PEGylated Peroxyester **12**

In order to make the PEGylated acid, the substituted aldehyde had to be prepared starting from the activation of the tetraethyleneglycol(monomethylether) at its alcohol terminus with p-toluene sulfonyl chloride according to a literature procedure.[56] The mechanism for this reaction is shown below:

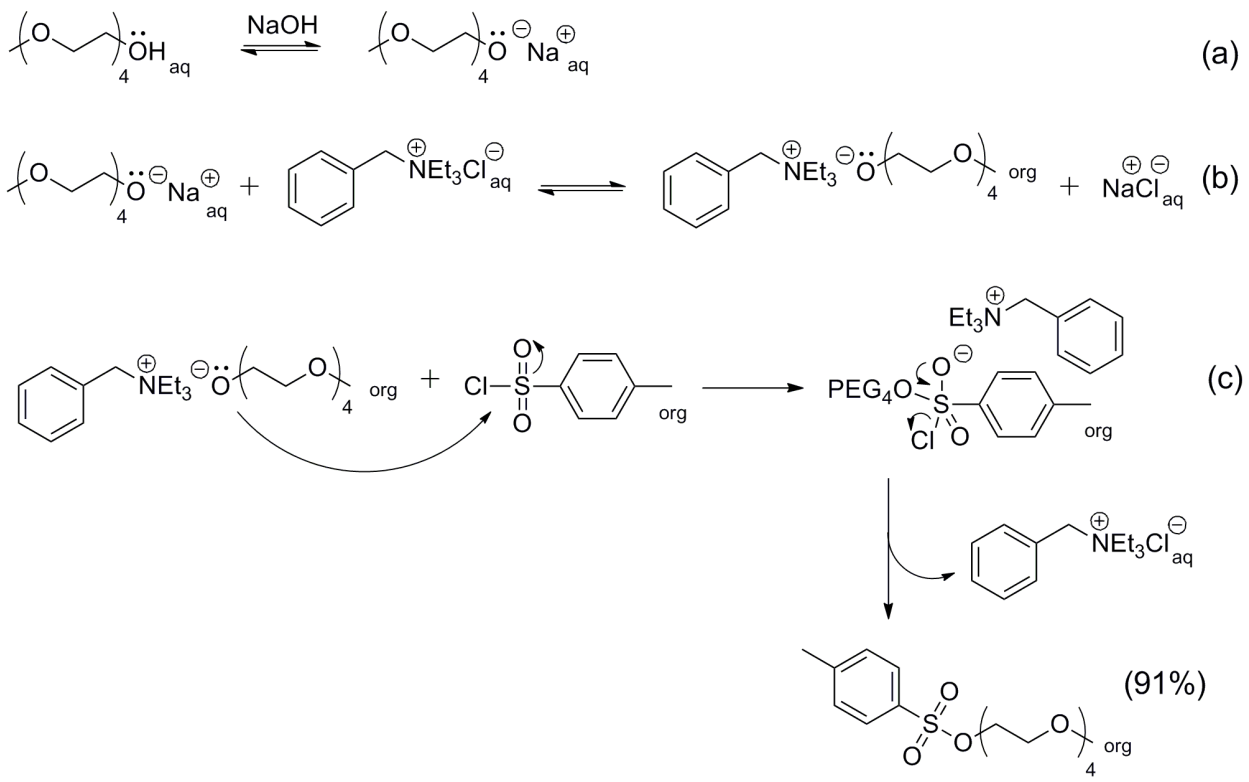


Figure 2.10: Tosylation of tetraethyleneglycol(monomethylether) with p-toluene sulfonyl chloride.

The tosylation reaction proceeds with a slight excess of p-toluene sulfonyl chloride and a catalytic amount of benzyltriethylammonium chloride (BTACl). In this reaction BTACl is a phase transfer catalyst and is initially in the aqueous phase with tetraethyleneglycol(monomethylether), whereas the organic phase contains the organic reactant p-toluene sulfonyl chloride in dichloromethane. Since the phases are mutually immiscible the reaction does not proceed unless the catalyst, BTACl is present. The catalyst transfers continuously reacting anions into the organic phase in the form of lipophilic ion-pairs produced according to the ion-exchange equilibrium (1b), where they react further, with p-toluene sulfonyl chloride affording nucleophilic substitution (1c). The

tosylation reaction proceeded well within 18 hours with a yield of 91% after purification by column chromatography.

The second step in the synthesis towards the target compound is the PEGylation of 4-hydroxybenzaldehyde through etherification. The reaction is carried out under basic conditions with K_2CO_3 (3 fold excess) in N,N-dimethylformamide. The reaction proceeds by deprotonation of the hydroxyl group of 4-hydroxybenzaldehyde with K_2CO_3, followed by nucleophilic substitution on the carbon next to the sulfonyl group of the tosylated-PEG. The reaction conditions[12] called for excess base, equimolar amounts of 4-hydroxybenzaldehyde and tosylPEG, heating under reflux for 48 hours in DMF. The reaction proceeded well within 48 hours with a yield of 88% after purification by column chromatography.

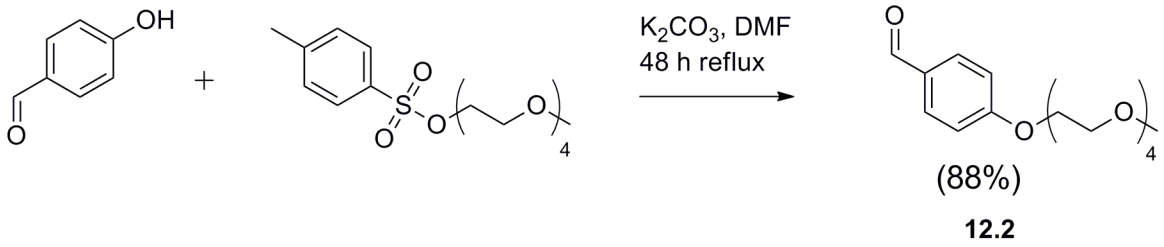


Figure 2.11: Etherification of 4-hydroxybenzaldehyde with tosylated PEG.

The third step in the synthesis towards the PEGylated peroxyester is the “classic” Wittig reaction. Since its discovery, the Wittig reaction has become one of the most important and most effective methods for the synthesis of alkenes. We decided that the Wittig reaction would be a good approach to synthesizing the framework for the compound, by reacting PEGylated aldehyde and 2-carboxyethyl)triphenylphosphonium bromide.

It is essential to have a high purity alkyl triphenylphosphonium halide for Wittig reactions, especially for the generation of unstabilized ylides. Although the phosphonium salt, ((2-carboxyethyl)triphenylphosphonium bromide) was purchased from ACROS in 97% purity, it appeared to contain some triphenylphosphine and water as impurities when checked by ^{1}H NMR and had to be purified. The impure phosphonium salt was purified by recrystallizing with dichloromethane and tetrahydrofuran to afford material that was pure by ^{1}H NMR.

The Wittig reaction was carried out with recrystallized (2-carboxyethyl)triphenyl-phosphonium bromide (1 eq), PEGylated aldehyde (0.95 eq) and KO^{t}Bu (2.1 eq). The reaction was first performed by stirring the phosphonium salt and aldehyde in THF (concentration of 0.84 M), cooling to 0°C and adding KO^{t}Bu in THF (concentration of 2.5 M). The solution turned bright orange as base was added, most likely indicating the formation of the ylide. An excess of base is required since there are two sites that can be deprotonated; the proton on the carboxyl group of the phosphonium salt or the proton on the carbon adjacent to the phosphine (which gives rise to the ylide).

The reaction was left to stir at room temperature and appeared to be complete by TLC after 12 hours. For the work up, the reaction was quenched with 10% H_2SO_4 (pH = 2) and then extracted with ethyl acetate. The crude product was a viscous oil with triphenylphosphine oxide as the major impurity. The crude product was originally purified by column chromatography, with silica gel (mobile phase 96/4 dichloro-methane/methanol). However, it was discovered (after ^{1}H NMR) that the fractions containing the desired product appeared to have an impurity that co-eluted with it. By NMR it appeared as though the impurity was aromatic and contained a PEG moiety as

well. After careful examination, it appeared as if the impurities were a result of a Cannizzaro reaction (of the aldehyde starting material). The reaction was repeated and a longer column and slightly modified TLC conditions were used to try to separate the impurity but there was no success with this approach.

We decided that optimizing the Wittig reaction was the best approach to minimizing the amount of unwanted side-reactions from occurring. We opted to use 4-methoxybenzaldehyde instead of the PEGylated aldehyde, as a model substrate to optimize the reaction since it is inexpensive. Furthermore, using the substrate 4-methoxybenzaldehyde proved to be more useful at confirming the Cannizzaro reaction impurities.

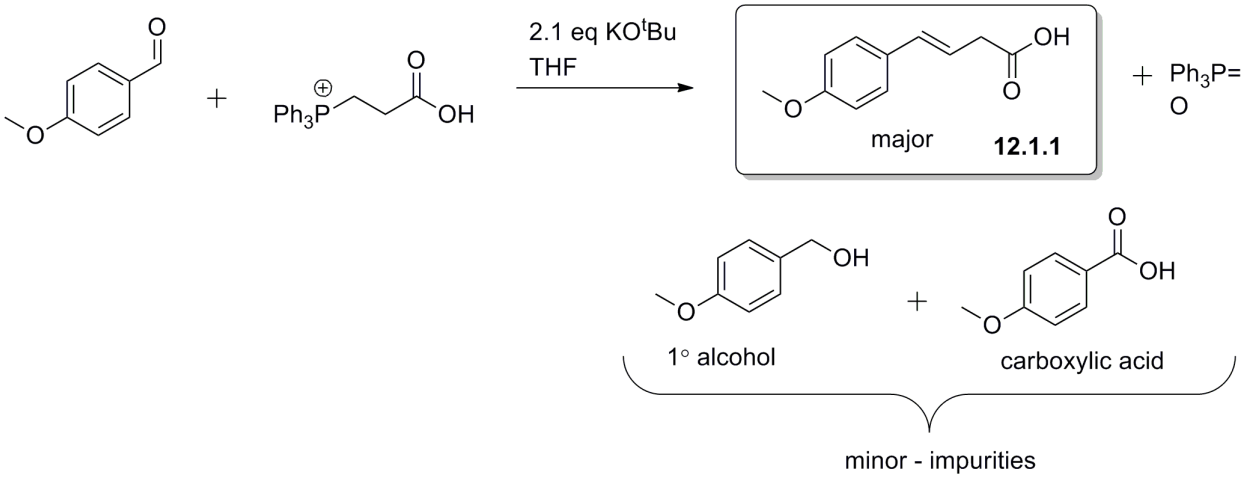


Figure 2.12: Model reaction with p-methoxy benzaldehye showing the desired products and proposed impurities

Generally, the Cannizzaro reaction occurs with aliphatic or aromatic aldehydes with no α-hydrogen. It involves an intermolecular hydride transfer reaction (**Figure 2.13**), when reacted with concentrated NaOH (50 wt%) or other strong bases (e.g. alkoxides).[45] This disproportionation reaction proceeds by one molecule of aldehyde oxidizing the other to give a carboxylic acid and is reduced to the corresponding primary alcohol with a maximum yield of 50%.

In the case of the Wittig reaction on either the PEGylated substrate or the model substrate, the Cannizzaro reaction takes place to some extent with trace amounts of water. In the presence of water, potassium tert-butoxide will react to form potassium hydroxide, which allows the Cannizzaro reaction to proceed. It is believed that the reaction occurs through the following mechanism:

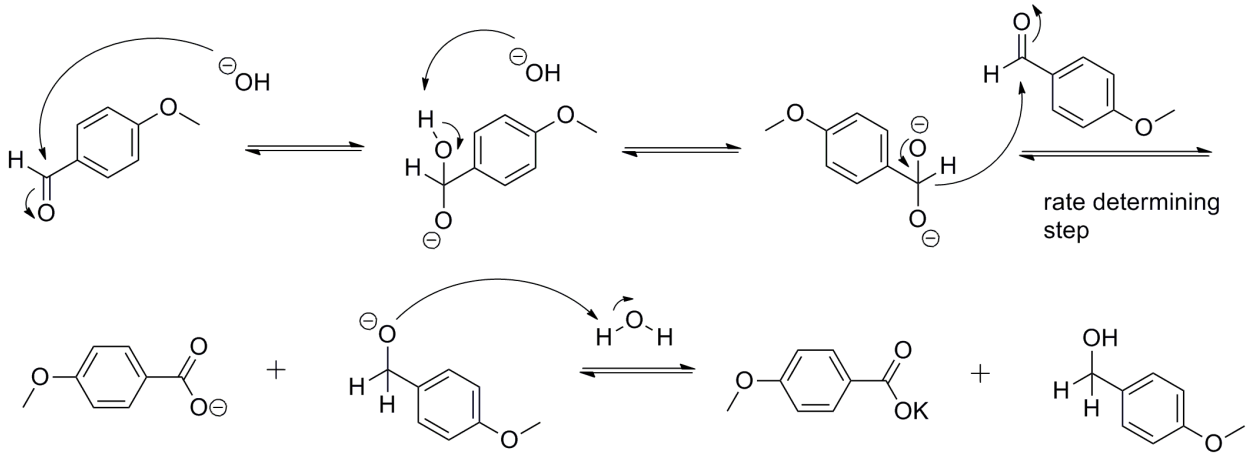


Figure 2.13: Mechanism for the Cannizzaro reaction

Optimization reactions were carried out on our model substrate under various conditions. We examined several factors, such as the concentration of the reactants

and carried out the reactions with freshly distilled THF. It appeared that varying the concentrations of the reactants had a direct effect on the ratio of the Cannizzaro products observed (the benzyl alcohol and carboxylic acid). The following table shows the ratios of the various products formed in a crude reaction to form the model compound:

Table 2.4: Product ratios in the Wittig reaction to form the model compound

Conditions	**Wittig : Alcohol**	**Wittig : Carboxylic Acid**
[0.84 M] Aldehyde, [2.5 M] KO^tBu;	1 : 0.10	1 : 0.22
[0.42 M] Aldehyde, [1.25 M] KO^tBu	1 : 0.07	1 : 0.18
[1.1 M] Aldehyde, [3.3 M] KO^tBu	1 : 0.05	1: 0.15

Once the reactions were complete (checked by TLC), they were concentrated and characterized by ^1H NMR. Although the reactions were crude, the impurity peaks were well resolved and the ratios could be compared easily, thus making it easy to extract information. To obtain the Wittig to alcohol ratio, an olefin proton (1 H, 6.51 ppm) from the Wittig product was compared with the benzylic proton (4.61 ppm). Similarly, for the Wittig to carboxylic acid ratio, the olefinic proton was compared to the aromatic protons at 8.07 ppm.

Based on the optimization reactions, shown in **Table 2.3**, the concentrated reaction conditions ([1.1 M] Aldehyde, [3.3 M] KO^tBu) worked the best at minimizing the amount of side products, which is displayed through the highest Wittig product to

Cannizzaro ratio. The concentrated reaction conditions worked the best since the least amount of water was present, due to the fact that less solvent was used. Although the THF was freshly distilled, it confirms how water sensitive this reaction is.

Using the concentrated reaction conditions, the Wittig reaction on the model compound was carried out on a slightly larger scale. An aqueous workup was implemented and the product was purified by column chromatography to remove triphenylphosphine oxide (60/40 Pentane/Ethyl acetate) affording a yield of 36%. Unsatisfied with this yield, the reaction was repeated under the exact conditions, however after workup the product was recrystallized with ethanol/water to afford a yield of 49%. This yield is satisfactory compared to the literature yield[54] reported (45%).

According to the mechanism shown in **Figure 2.13**, the carboxylic acid salt is formed in the Cannizzaro reaction, however upon workup the respective acid is formed. The workup requires the reaction to be quenched with acid (pH = 2), and as a result, the carboxylic acid is observed in the crude ^{1}H NMR (after extraction with ethyl acetate and drying) with about 4% of the sample containing the acid impurity. The carboxylic acid impurity was also confirmed by spiking the sample with p-methoxybenzoic acid, causing the impurity peaks at 8.05 ppm and 6.95 ppm to grow in significantly (**Figure 2.14 (b)**). Although the carboxylic acid impurity could not be completely removed after purification, the benzyl alcohol could, through the use of acid-base chemistry.

Since Wittig reactions are very sensitive to moisture, the quality of the base can affect the outcome. It is often recommended that KO^{t}Bu should be sublimed when first received from the manufacturer. Furthermore, due to the hygroscopic nature of KO^{t}Bu, it

is recommended to be stored in the absence of air. Exposure to air can result in the bottle containing high percentages of carbonate and hydroxide.[57]

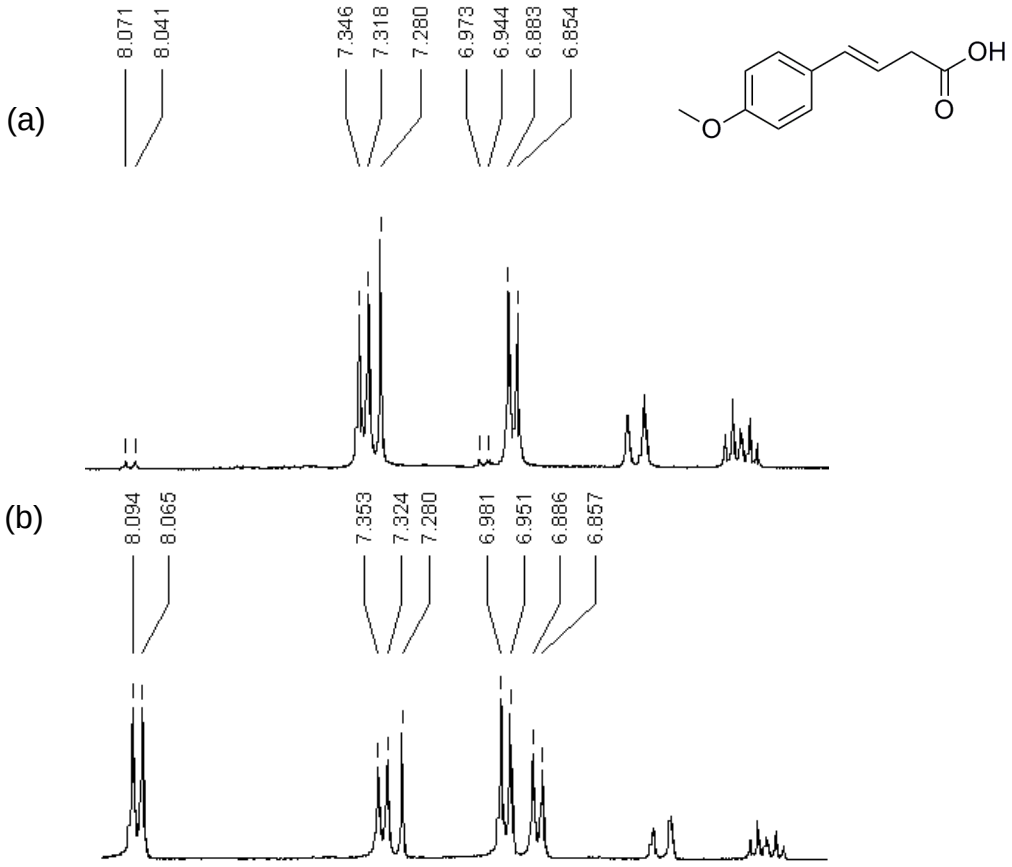
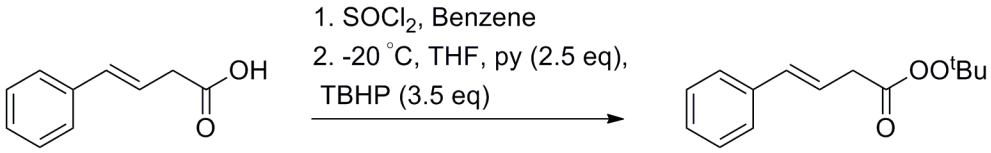


Figure 2.14: (a) ^{1}H NMR of the purified model compound 12.1.1 (containing ~4% carboxylic impurity (b) Model compound spiked with p-methoxy benzoic acid.

A problem often associated with the Wittig reaction is the removal of triphenylphospine oxide from the desired products, which was the case for both the model compound and the PEGylated acid. The phosphine oxide has peculiar properties, in that attempts to remove it from the crude reaction mixture by column chromatography

can be complicated and resulting in co-elution with the desired product. Many experimental procedures precipitate out the triphenylphosphine oxide with ether during the workup and then triturate out the product with additional ether to leave the triphenylphosphine oxide as a crystalline solid. Usually this procedure works best when mixtures of ether and hexane are used, employing as much hexane as the product solubility will allow.

To ensure that the final step of the PEGylated peroxyester went smoothly, we decided to optimize the reaction using the model compound (carboxylic acid). The first attempt at making the model perester, was based on a literature procedure,[44] where a phenyl perester was synthesized:



The model perester was made in two parts; the first part of being the formation of the acid chloride with thionyl chloride in benzene. This reaction was monitored by TLC, by quenching a small portion of the reaction mixture with methanol to form the analogous methyl ester thus allowing the reaction to be followed more easily.

Once the acid chloride was formed, the second part of the reaction involved concentrating the reaction by removing benzene and excess thionyl chloride. The acid chloride was then dissolved in THF and cooled down to -20°C, before pyridine (2.5 eq) and tert-butyl hydroperoxide (3.5 eq. 5.5 M solution in decane, dried over 4 Å molecular

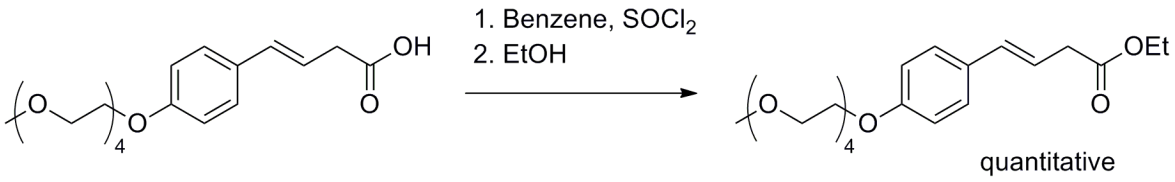
sieves). The reaction appeared to be complete with the precipitation of the pyridinium salt, however the reaction did not appear to go to completion when checked by TLC. Although the product spot could be visualized, there seemed to be some starting material reformed (carboxylic acid). The reaction was worked up by quenching with ice-water, washing with dilute acid and then base. The product was purified quite easily with silica gel and a short column, to afford a white solid with a yield of 49% (Compound **12.1.2**).

Satisfied with this result, we attempted the reaction with PEGylated acid under the same conditions. To our surprise, the reaction did not work as well with the PEGylated acid. Based on the NMR result, we were able to conclude a few things regarding this experiment. It is likely that rotovapping the acid chloride to remove the excess benzene and thionyl chloride may have been a poor choice. The conditions for making an acid chloride from thionyl chloride results in the formation of HCl and it is likely that the PEG chain is susceptible to cleavage due to the fact that HCl is being concentrated in the flask. Although the chemistry worked well on the model compound, having a PEG chain introduces more complications because it is more sensitive to acidic conditions. An attempt to purify the crude product (PEGylated peroxyester) with a short silica column was made, however the fractions which contained the desired product contained an unknown PEGylated impurity which could not be removed.

Known methods for the synthesis of peroxyesters are based on the acylation of hydroperoxides with appropriate agents, such as acid chlorides[12] and anhydrides[58]; and in some cases, carboxylic acids[59] themselves. Among the acylating agents mentioned, the most accessible are carboxylic acids. They are capable of acylating hydroperoxides

in the presence of strong mineral acids. The procedure requires no preliminary transformation of carboxylic acid into some acylating agent. However, this method requires a large concentration of mineral acid to be used, which can impose synthetic restrictions, especially in the case of the PEGylated acid substrate. Furthermore, the reaction is reversible; therefore dehydrating agents (such as Na_2SO_4, $MgSO_4$, or B_2O_3) are usually added to increase the conversion. Satisfactory yields have been reported in literature for the acylation of tert-butyl and tert-amyl hydroperoxides with lower fatty acids.[58] Acylation of hydroperoxides with carboxylic acids can also be affected in the presence of imidazole.[58] When imidazoles are used, the yields of the peroxyesters can reach 60-80%, but the reaction is accompanied by side reactions making the product difficult to purify.[58]

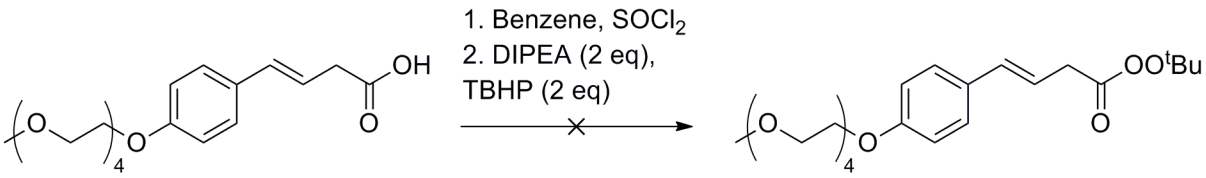
Our next approach was to find new conditions to make the PEGylated perester which didn't involve a highly acidic environment. We were interested in a one pot, direct approach if possible. Preferably, we wanted to make the acid chloride and directly quench the reaction with pyridine and tert-butyl hydroperoxide. This idea was prompted by the following result observed:



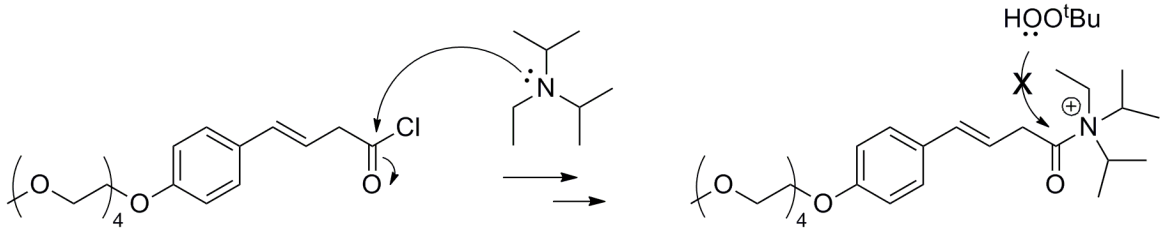
Directly quenching the acid chloride with excess ethanol allows for the formation of the ester shown above, which was confirmed by ^1H NMR. The reaction goes to completion almost instantly and there is no need to cool the reaction down extensively.

Keeping this in mind, we wanted to design an experiment that would work as straightforward as this one.

Initially we designed an experiment that involved making the acid chloride, followed by quenching the reaction with *N*,*N*-Diisopropylethylamine (DIPEA or Hünig's base) and tert-butyl hydroperoxide. The conditions are shown below:



Since there is excess thionyl chloride present and HCl is generated in the formation of the acid chloride, adding base before the addition of TBHP was necessary to facilitate the reaction. DIPEA seemed to be a suitable base since it is sterically hindered and correspondingly non nucleophilic.



One problem with this reaction was that it was carried out at room temperature, and hence a lot of heat/gas evolved from the reaction. Peroxyesters are generally made at low temperatures, such as -20°C or lower.[12] By TLC and ^{1}H NMR it was confirmed that the reaction was unsuccessful. Since Hünig's base is sterically hindered, it is likely that once the acyl pyridinium salt is formed, it is too crowded for TBHP to attack the

carbonyl and hence the desired perester is not formed. The following diagram displays the crowded reaction centre.

Based on the previous experiment, we decided that cooling the reaction would be important for minimizing the amount of heat evolved in synthesizing the peroxyester. However, since benzene was used as the solvent, the lowest temperature that we could cool the reaction down to, without freezing, was about 5°C. Initially the reaction was performed by cooling the acid chloride in benzene to 5°C and adding pyridine (2 eq.) and TBHP (2 eq.) dropwise. Precipitation of the pyridinium salt took place and when checked by TLC, the peroxyester was being formed and some starting material was present (PEGylated carboxylic acid) as well. The product was purified using a short silica column, however the PEG chain integrated too high by ^{1}H NMR and other impurities were evident. Furthermore, when another batch of the product was made, it could not be purified using neutral alumina either. Hence, it appeared that this peroxyester may be unstable to chromatographic techniques.

The reaction was repeated with the same conditions, but an aqueous workup was used to purify the product from the pyridinium chloride salt. Although the reaction worked better under these conditions, we varied the amount of TBHP added to the reaction to see if we could minimize having an excess of unreacted TBHP. After carrying out the reaction with 1.5 eq., 1.2 eq., and 1 eq. of TBHP, we discovered that adding 1 eq. was sufficient enough to form the peroxyester, as expected.

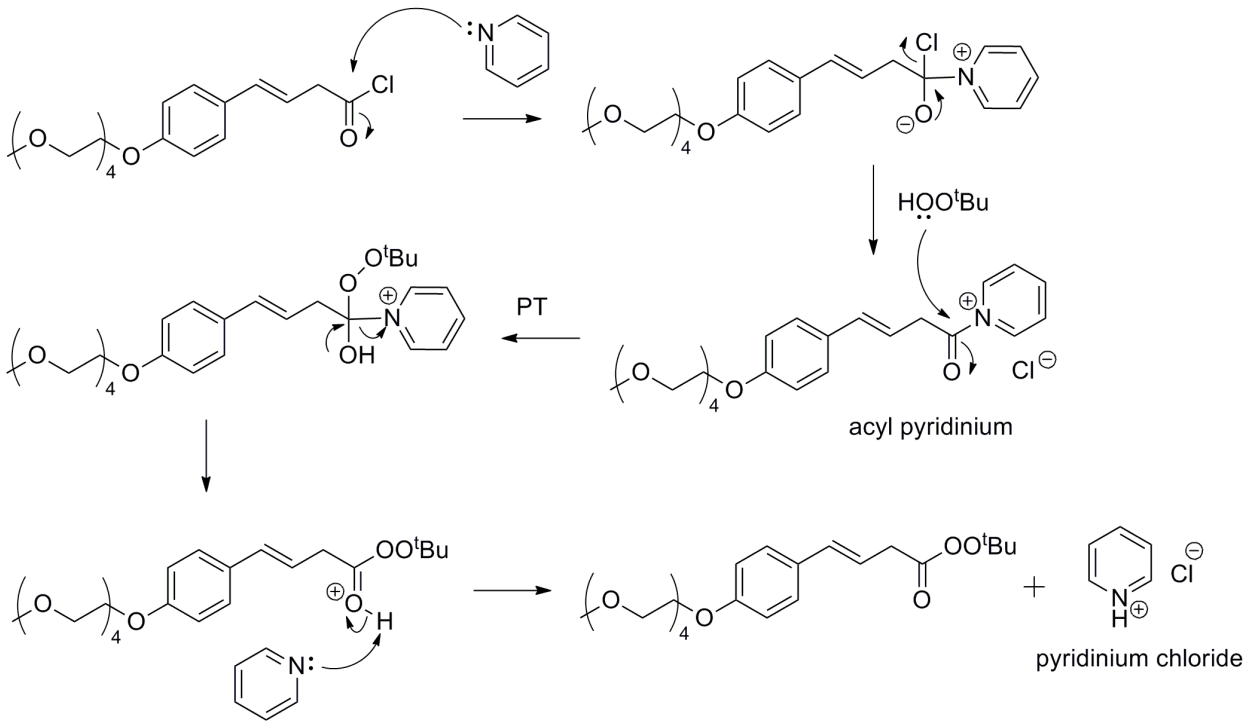


Figure 2.15: Mechanism for the synthesis of PEGylated peroxyester (PT = proton transfer)

One problem with the peroxyester synthesis appeared to be the hydrolysis of the product. In all cases of synthesizing the acid chloride, there appeared to be complete conversion. However, in the subsequent step of forming the peroxyester it was observed by TLC that the starting material (carboxylic acid) was being reformed. This is likely due to the fact that even the slightest amount of water present in the pyridine may reverse the reaction to some degree.

Although the starting material (PEGylated acid) could not be removed from the product, there appeared to be an appreciable amount of product being formed in the reaction, which was encouraging. Hence, several small scale batches of the product

were made for determining the right separation conditions on the HPLC (explained in further detail in **Section 2.2.2**).

We found that the synthesis proved to be problematic on later attempts to make more PEGylated peroxyester and the results could not be reproduced, as shown in **Figure 2.16**. The problem appeared to arise in the conversion of the acid chloride to the peroxyester. Extra care was taken such as distilling fresh benzene and pyridine to avoid water from interfering with the reaction. In every attempt, after pyridine was added to the reaction an insoluble red tar-like substance adhered to the flask, something which was not initially observed. Thus, the subsequent reaction with TBHP did not occur to a significant extent and upon aqueous workup the NMR appeared to mostly contain starting material (PEGylated acid) in addition to hydrolyzed PEG.

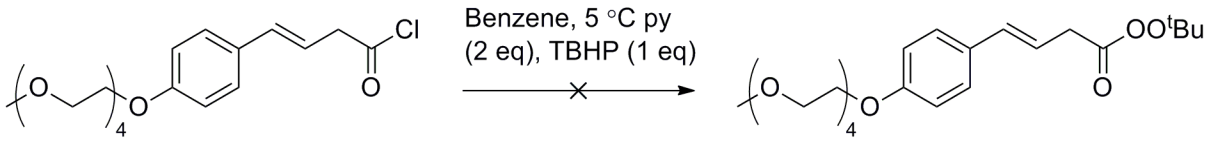


Figure 2.16: Reaction conditions for the PEGylated peroxyester synthesis

Additional attempts were made by substituting benzene for diethyl ether, so that the reaction could be cooled down to -20 to -25 °C, which is the temperature that several peroxyesters have been reported to made at.[12, 60]. After the addition of pyridine, an insoluble orange tar-like substance was formed, making it difficult to react with the TBHP, similar to the case mentioned above.

There is literature precedence of insoluble tar-like complexes formed upon addition of pyridine to acid chlorides, in the case of acetyl, cinnamoyl and 4-phenyl-3-butenoyl chlorides. In these cases, the addition the order of addition was reversed with TBHP added first, followed by pyridine. This method of reversing the reagents was attempted, but there was no success at making the peroxyester using this method. There was enough material available to carry out preliminary decomposition experiments; however the synthesis of the PEGylated peroxyester (**12**) needs to be revisited in order to determine why the original reaction conditions are not reproducible.

2.2.2 Preliminary Studies of PEGylated Peroxyester Decomposition

In order to carry out decomposition experiments, it was necessary to synthesize the products being formed in such experiments. The following figure shows the products that are formed in a typical decomposition reaction:

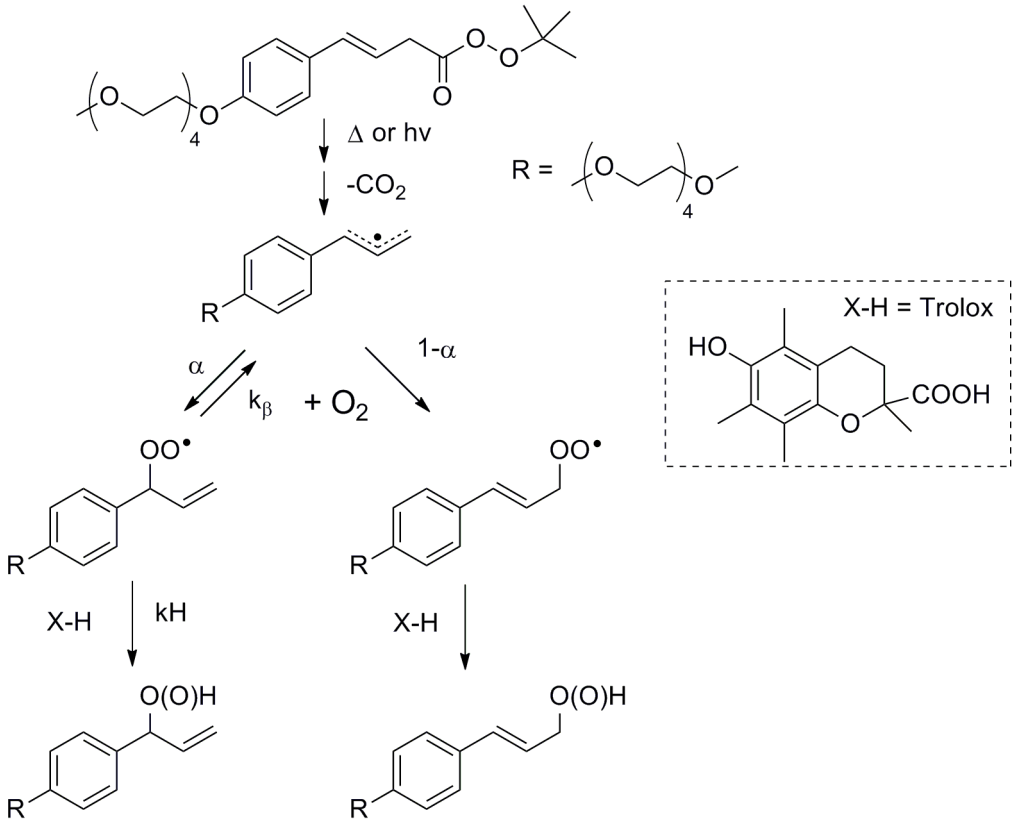
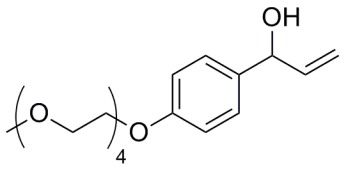
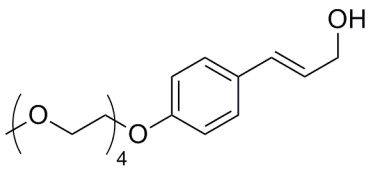


Figure 2.17: Expected decomposition pathway of the PEGylated peroxyester **12**

In the decomposition experiment shown above, the peroxyester can be heated in the presence of an H-atom donor, such as Trolox to give the two hydroperoxide products shown above. To obtain the alcohol products, TCEP (tris(2-carboxyethyl)phosphine) was used to reduce the hydroperoxides. Therefore, it is these alcohol products that we were interested in synthesizing, since they are the products of this decomposition experiment.



12.2.1



12.2.3

Figure 2.18: Non- conjugated (**12.2.1**) and conjugated (**12.2.3**) standards synthesized for decomposition studies.

Both the non-conjugated and conjugated standards were synthesized from the PEGylated aldehyde. The conjugated alcohol was made by a Horner-Wadsworth Emmons olefination followed by hydrolysis, whereas the non-conjugated alcohol was made by a Grignard reaction.

Conjugated Standard

Similar to the Wittig reaction, the Horner-Wadsworth Emmons (HWE) reaction is an effective way for synthesizing olefins. Aldehydes and ketones can be used to prepare alkenes by reacting them with carbanions of alkyl diphenyl phosphine oxides. The HWE olefination has several advantages over the “classic” Wittig reaction. The phosphorus carbanions are more nucleophilic than the corresponding phosphorus ylides, making them very reactive towards all aldehydes and ketones under milder reaction conditions. Furthermore, the phosphorus byproducts in the HWE olefination are water-soluble and hence readily separated from the desired product. Phosphonates that lack a α-substituent at R^1 (for example, COO^-, COOR, CN, SO_2R, vinyl, aryl $P(O)(OR_2)$,

OR or NR_2) generally result in a low yield of the product alkene.[52] The stereochemical outcome of the HWE reaction is mostly dependent on the nature of the phosphonate used. In general, bulky substituents at both the phosphorus and the carbon adjacent to the carbanion favor formation of the E-alkene.

The reaction conditions for the HWE olefination are shown in **Figure 2.19** to make the PEGylated ester. It is generally accepted that for base sensitive substrates, the use of a metal salt (LiCl and NaI) and a weak amine base (e.g. DBU) has shown to be effective at avoiding epimerization. The use of lithium salts allow for milder reaction conditions in general; lithium likes to form a tight complex with the carbanion derived from phosphonate **1** as shown in **2**, enhancing the acidity of **1**, shown in **Figure 2.19 (b).**[61] Adding a lithium salt to the reaction allows for easy deprotonation with an amine (i.e. DBU) under very mild conditions.

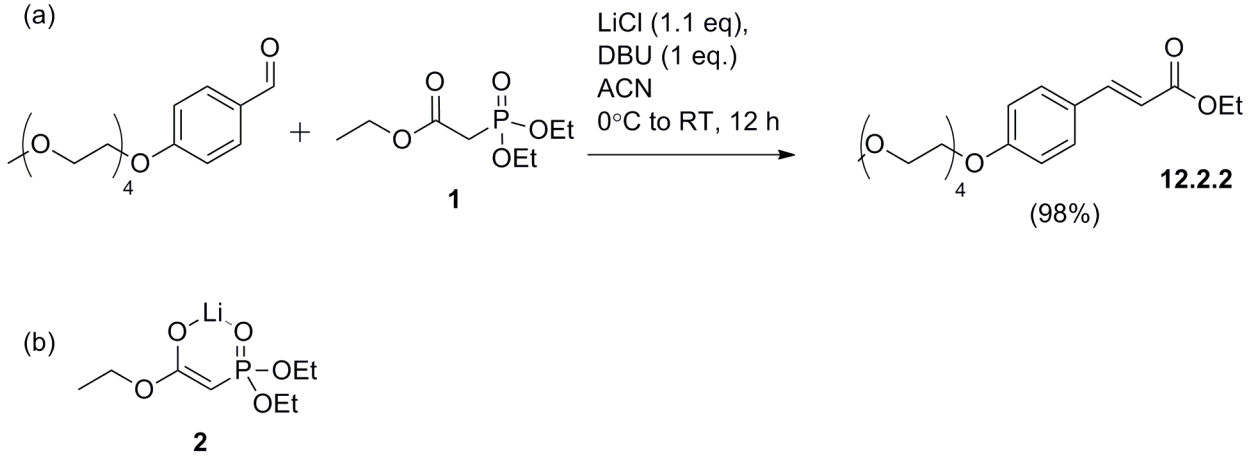
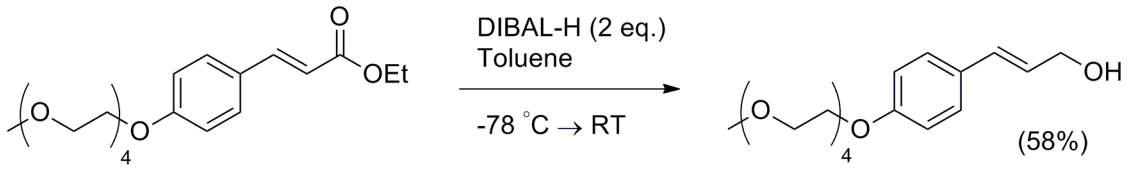


Figure 2.19: (a) Reaction conditions for the Horner-Wadsworth-Emmons olefination on the PEGylated aldehyde **(b)** Li^+ tight ion complex coordinated to the phosphonate.

The HWE olefination was initially carried out at 0°C, then allowed to warm up to room temperature to stir for 12 hours. The olefination reaction proceeded very well by TLC and after work-up a colourless oil was obtained in 98% yield.

The last step in the conversion of the HWE olefination product to the conjugated alcohol is reduction of the ester. This was accomplished by reducing the ester with DIBAL (Diisobutylaluminium hydride) by the conditions shown in below.



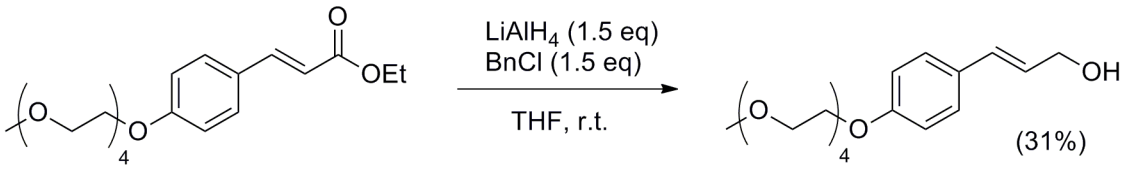
12.2.3

The reaction conditions used were based on a literature procedure [62] whereby reduction of a para substituted ethyl cinnamate to the respective cinnamyl alcohol took place. The best yield obtained using this prep was (58%), since the reaction did not go to completion and starting material had to be removed from the product by column chromatography.

Reduction of conjugated carbonyl compounds with reducing agents such as lithium aluminum hydride and sodium borohydride are generally complicated by the competing 1,4 and 1,2 processes.[63] Hence, reagents such as AlH_3 (generated from $AlCl_3$) and DIBAL-H are the most popularly used reductants for the conversion of α, β-unsaturated esters into unsaturated alcohols. Although these approaches have been shown to be very useful, the methodologies for the reduction are associated with

several disadvantages such as expensive reagents, harsh reaction conditions, low temperature, long reaction time and much excessive use of reducing agents.

The reduction of the PEGylated α, β-unsaturated ester was also attempted with lithium aluminum hydride and benzyl chloride to see if the yield could be improved. This method was attempted because it was milder and the reagents were readily available. It has been shown that the reaction of $LiAlH_4$ and an alkyl halide (such as benzyl chloride) leads to the formation of AlH_3 as a by-product and hence acts as the reducing agent in the presence of the ester.[64] Several α, β-unsaturated esters have been reduced to alcohols using the conditions shown below, and yields between 80-90% have been reported[65] with a reaction time of 1 hour. However, when this reaction was performed on the PEGylated α, β-unsaturated ester the reaction did not proceed to completion after several hours. Hence, this method was not effective for preparing the conjugated alcohol and the DIBAL-H method was the best approach at making the compound.



12.2.3

Non-Conjugated Standard

The Grignard reaction has become one of the most versatile methods for making C-C bonds. The Grignard reaction, discovered in 1900 by V. Grignard, is very useful for converting aldehydes and ketones to secondary and tertiary alcohols respectively with

an alkyl halide and magnesium metal (Mg).[45] Grignard reagents are usually prepared by reacting alkyl, aryl, and vinyl halides with magnesium metal in aprotic nucleophilic solvents (e.g. ethers, tertiary amines). Generally, Grignards are thermodynamically stable but air and moisture sensitive and incompatible with acid functionalities (alcohols, thiols, phenols, carboxylic acids, 1° and 2° amines, terminal alkynes)[45] Grignard reagents contain a highly polarized C-Mg bond, with the partial negative charge on the carbon atom, thus making them good carbon nucleophiles. Furthermore, in most carbon heteroatom multiple bonds the carbon atom is partially positively charged so the formation of C-C bonds with the nucleophilic Grignard occurs readily. Generally, one equivalent of a Grignard reagent, followed by an aqueous workup, is sufficient to convert aldehydes to secondary alcohols (formaldehyde to primary alcohols), nitriles to ketones, and carbon dioxide to acids.

Grignard reactions are often accompanied by some side products. It is common for Grignard reagents formed from an alkyl halide to give the undesired Wurtz coupling products.[45] In addition, the presence of oxygen from air, and moisture can interfere with the reaction by consuming some of the reagent to give alkoxides and alkanes. Furthermore, the Grignard can act as a base if there is a proton in the α-position and enolize the substrate. Whereas if the reagent has a β-hydrogen and the substrate is hindered, reduction of the carbonyl group may occur by an intramolecular hydride transfer.

The non-conjugated alcohol was made by reacting vinyl magnesium bromide with the PEGylated aldehyde. Synthesizing this compound was not very trivial and various reaction conditions were explored, as shown in **Table 2.4** on the next page.

Table 2.4: Reaction conditions for the synthesis of the non-conjugated standard

Entry	Conditions	TLC	Workup	NMR
1	Aldehyde: 1.0 eq [0.4 M], Vinyl-MgBr: 1.1 eq Et_2O, 0°C → rm temp (2h)	Starting material consumed	Acidify with 10% HCl, extract with E_2O	Conjugated alcohol formed
2	Aldehyde: 1.0 eq [0.73 M], Vinyl-MgBr: 1.1 eq THF, 0°C (2h)	Reaction did not go to completion - (5%) aldehyde present	Acidify with 10% HCl, extract with E_2O	Mixture of aldehyde (5%) and conjugated alcohol
3	Aldehyde: 1.0 eq [0.73 M], Vinyl-MgBr: 1.2 eq THF, 0°C (2h)	Starting material consumed	Quench with sat. NH_4Cl and extract with DCM. [66]	Non-conjugated product formed (86% yield)

The synthesis of the PEGylated non-conjugated standard was attempted under three conditions. Vinyl magnesium bromide was commercially available and all solvents and glassware were free of water. The conditions described in the first entry of this table show that the starting material was consumed; however upon workup it appeared that the conjugated alcohol was isolated. The same result was observed in the second case as well, even though the reaction was performed at a higher concentration and lower temperature. Quenching the reaction with dilute HCl promotes the isomerization of the non-conjugated product to the more stable, conjugated alcohol.

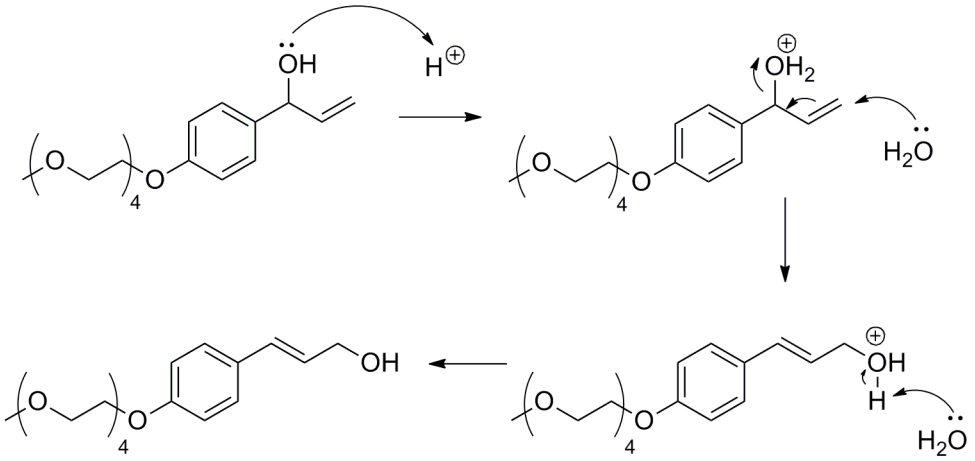


Figure 2.20: The isomerization reaction of the non-conjugated alcohol to the conjugated alcohol.

One interesting result is that when the reaction is quenched with NH_4Cl, followed by extraction with DCM, the desired, non-conjugated product is observed in high yield. The explanation for this result is that since there is no acid used in the workup, the acid catalyzed isomerization reaction does not take place, hence the conjugated product is not observed. Although the first two "failed" reactions in **Table 2.4** did not give the desired product, this synthesis would have been more useful if it had been done before the chemistry for the non-conjugated alcohol was developed (Horner-Wadsworth Emmons olefination, followed by hydrolysis).

2.2.3 HPLC decomposition experiments

In order to carry out analyses of the decomposition experiments of the PEGylated peroxyester by HPLC, it was necessary to determine at which wavelength the decomposition products absorb. This was carried out using the Varian Cary 50 Bio UV-Visible Spectrometer. The UV spectra for each standard with their λ_{max} shown in **Figure 2.21.**

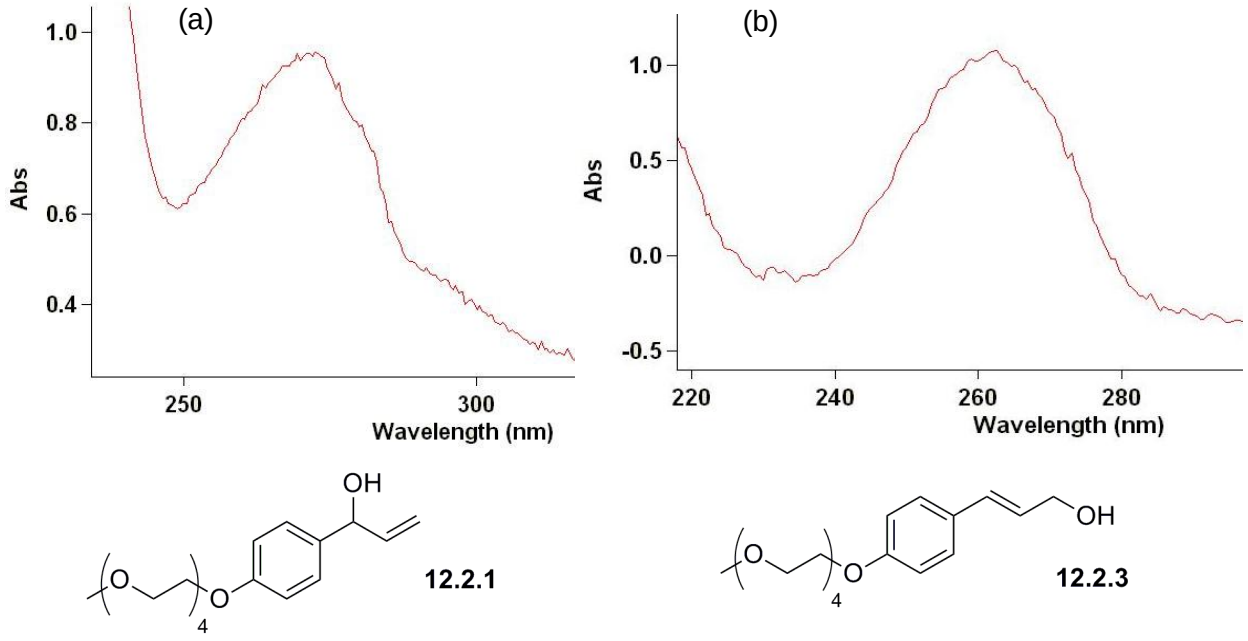


Figure 2.21: The UV spectra of (a) non-conjugated, λ_{max} = 274 nm (b) conjugated alcohol λ_{max} = 262 nm taken in methanol, 1 cm UV quartz cell.

The absorbance spectra of the non-conjugated and conjugated alcohol gave a peculiar result, which is not intuitive. The λ_{max} for the non-conjugated alcohol was measured to be higher than the conjugated alcohol. One would expect the conjugated alcohol to have a higher λ_{max} since it has a styrene chromophore, and generally, having

a conjugated compound extends the pi network of electrons, causing a shift in the λ_{max}. The λ_{max} for each compound was measured again, to rule out the possibility of error, but the result was the same. Furthermore, the PEGylated peroxyester was found to have a λ_{max} of 263 nm as well, consistent with that observed with the conjugated standard.

The alcohol standards were run separately on the HPLC (XBridge Prep (reverse phase) C8 column (10 x 150 mm)) using a mobile phase of 60/40 MeOH/ddH_2O at 1 mL/min separately at λ_{max} = 262 nm and 274 nm for the conjugated and non-conjugated alcohols respectively. The retention time for the conjugated and non-conjugated alcohol are R_t = 11.65 min and 12.63 min for the conjugated and non-conjugated alcohols respectively. Note that there was some contamination of the conjugated alcohol shown in **Figure 2.23** from carry over.

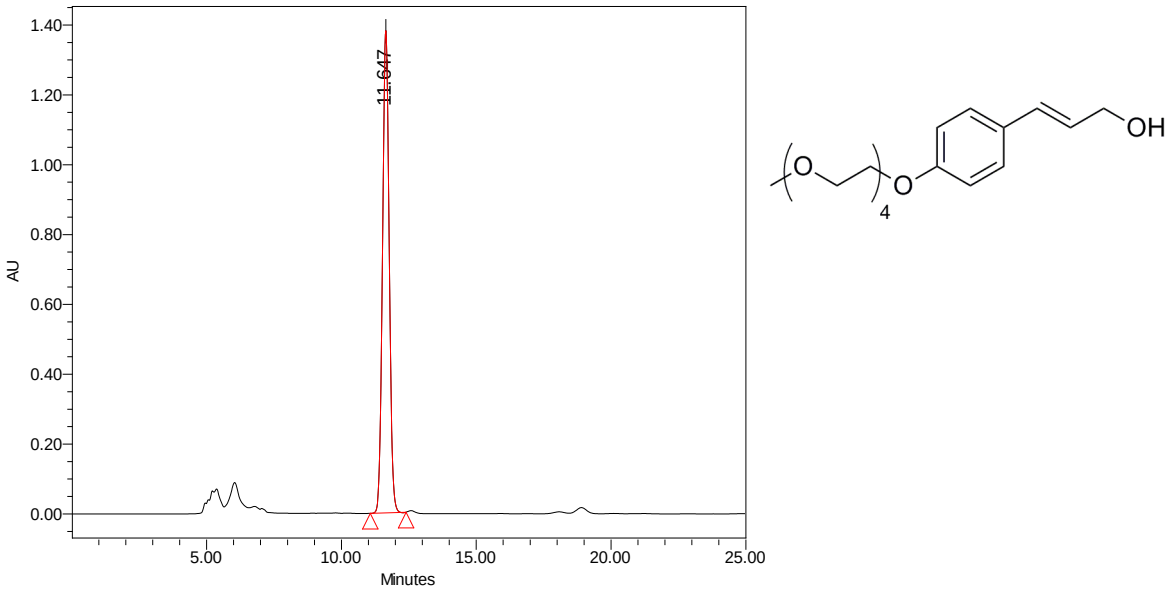


Figure 2.22: (a) Conjugated Standard 12.2.3 @ 262 nm. HPLC conditions: 1 mL/min, 60/40 (Methanol/ddH_2O). R_T = 11.65 min

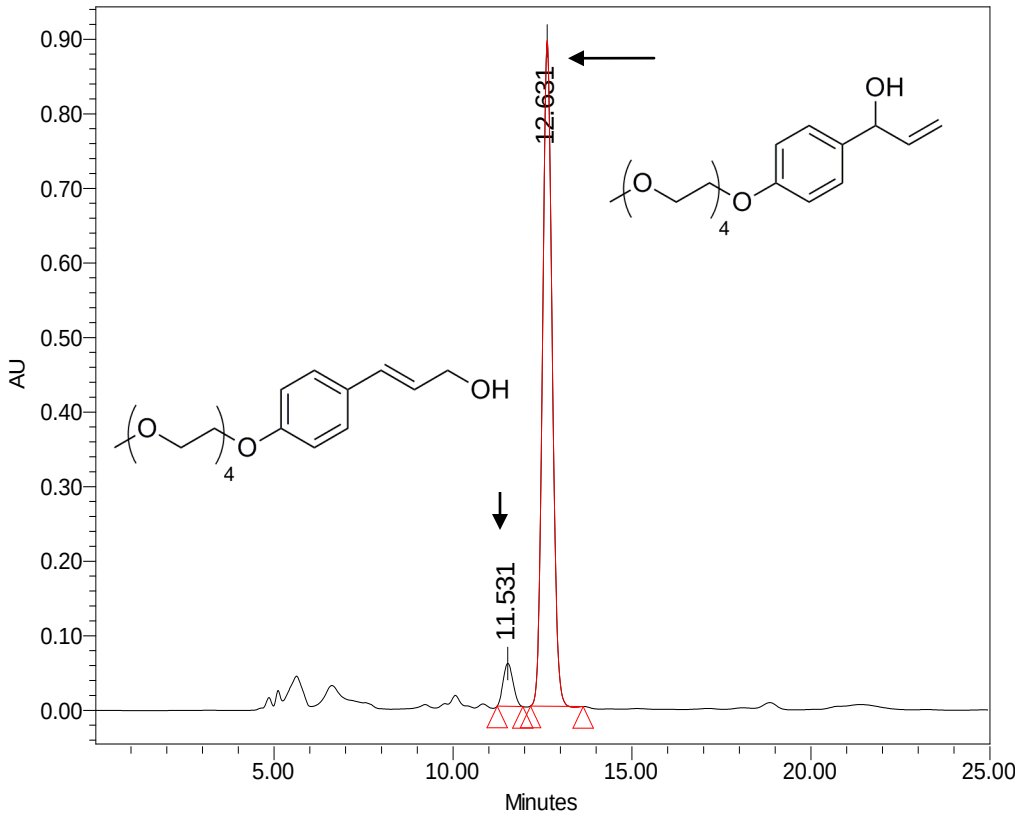


Figure 2.23: Non-conjugated Standard **12.2.1** @ 274 nm. HPLC conditions: 1 mL/min, 60/40 (Methanol/ddH_2O). R_T = 12.63 min

A decomposition experiment was carried out on the PEGylated peroxyester in the presence of Trolox. The blank peroxyester sample (without Trolox) is shown in **Figure 2.24,** before Trolox was added. In the presence of Trolox, at 37°C, the peroxyester decomposes within 1 hour, by the disappearance of the peak at 39.9 minutes in **Figure 2.25** and the formation of the conjugated alcohol product and small amounts of the non-conjugated product. This preliminary experiment shows that the peroxyester has a relatively fast decomposition rate, proving how unstable this compound is.

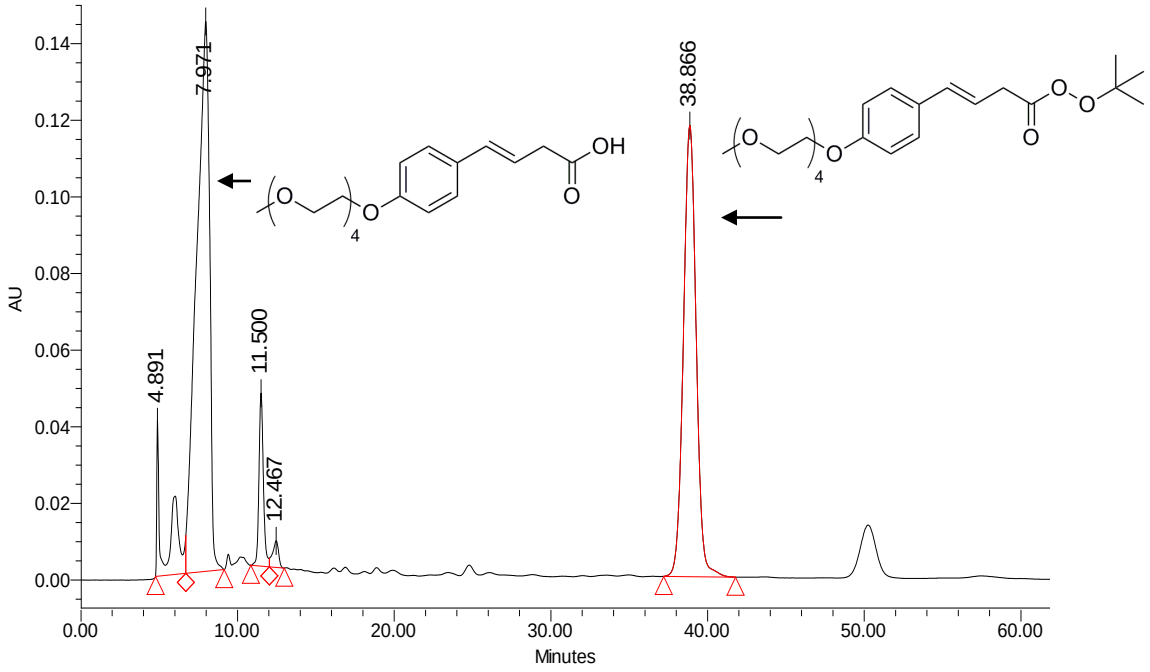


Figure 2.24: PEGylated Peroxyester before Trolox is added (R_T = 39.9 min for peroxyester, R_T = 7.97 min for PEGylated carboxylic acid) @ 262 nm. Flow: 1 mL/min, Mobile phase: 60/40 (Methanol/ddH_2O).

To make these results more conclusive, a co-injection of the conjugated standard should have been made to verify if the peak at 11.50 min (**Figure 2.25**) is indeed the conjugated alcohol standard. Nonetheless, it is apparent that the PEGylated peroxyester decomposes in the presence of Trolox and future work will have to be carried out to determine how the product ratios change in the presence Trolox.

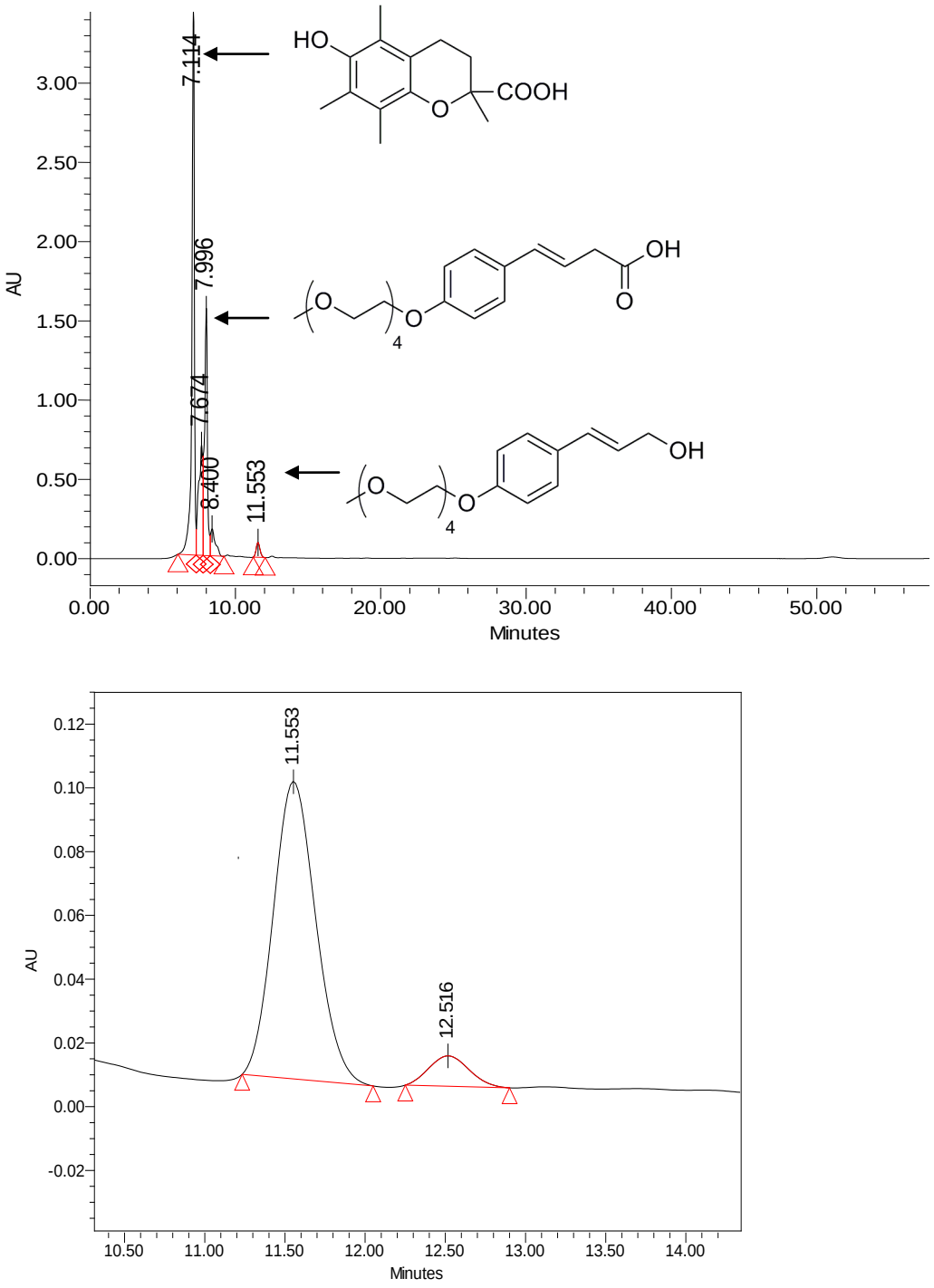


Figure 2.25: (a) PEGylated Peroxyester 12 after incubating for 1 hour in Trolox **(b)** Cross-section of chromatograph (a)

2.3 Towards Di-PEGylated Peroxyesters For Further Increased Water Solubility

Aside from the PEGylated peroxyester, we were interested in making an analog, the Di-PEGylated Peroxyester. This analog would surely be water soluble but most we were hoping it would increase the product yield in decomposition experiments. The Di-PEGylated Peroxyester is shown below:

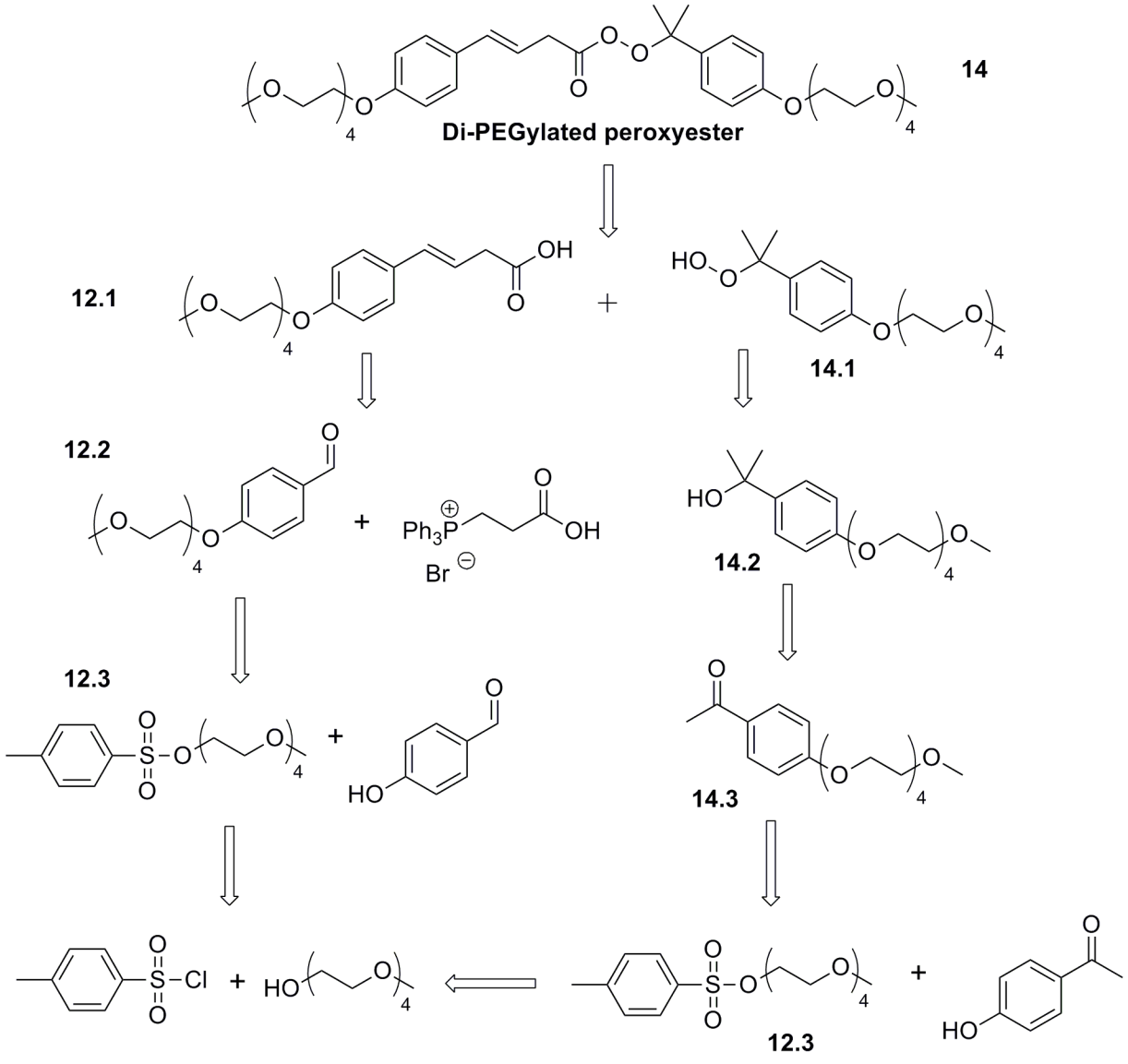


Figure 2.26: Proposed synthetic route to the Di-PEGylated peroxyester **14**

The actual synthesis of the Di-PEGylated peroxyester was not attempted, since the synthesis of the PEGylated hydroperoxide proved to be very difficult. The route towards the PEGylated hydroperoxide is shown in **Figure 2.27.** The first step is conversion of tosyl PEG with 4'hydroxyacetophenone to the PEGylated ketone, with a yield of 90% after column chromatography. The reaction mechanism is analogous to the synthesis of the PEGylated aldehyde, **12.2** since the conditions were very similar.

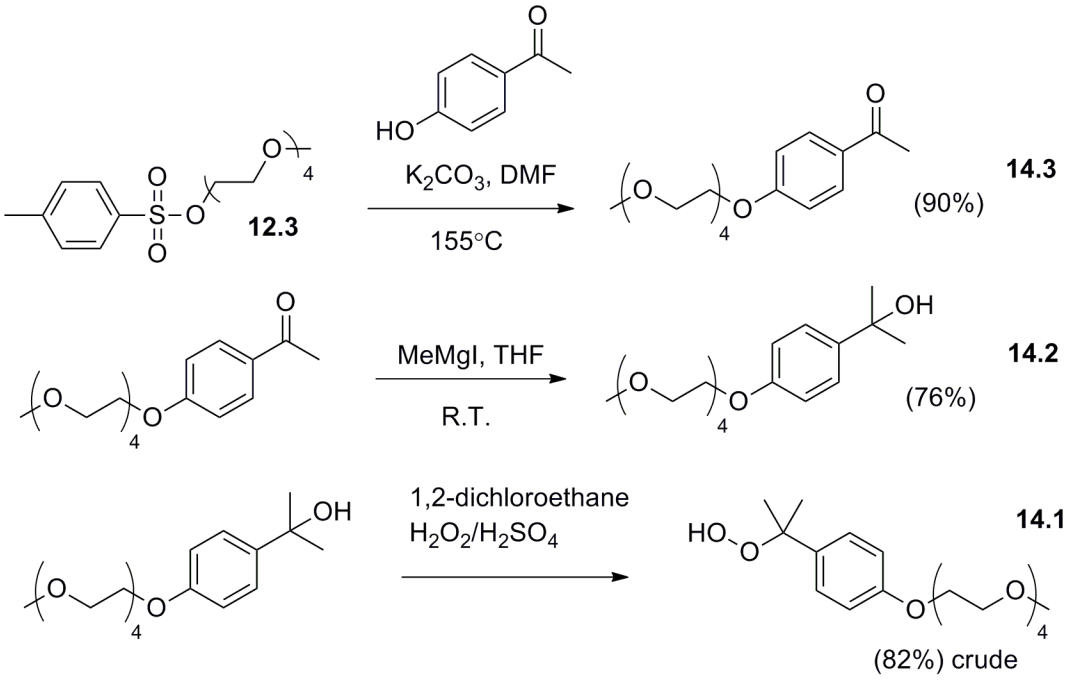


Figure 2.27: The steps towards making the PEGylated hydroperoxide

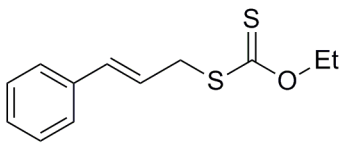
The second step of the synthesis was the Grignard reaction with the PEGylated ketone. The Grignard reagent, MeMgI (1.36 eq) was commercially available as a 3 M solution, and the reaction was carried out in THF at room temperature and went to completion within 4 hours, affording a yield of (76%).

The final step towards the synthesis of the PEGylated hydroperoxide was oxidation of the alcohol product using H_2O_2/H_2SO_4 based on a procedure reported by Mazurkiewicz and co-workers.[67] The reaction appeared to proceed well, as there was evidence of a more polar spot on the TLC plate (confirmed with hydroperoxide stain), corresponding to the hydroperoxide and the starting material appeared to be consumed. Based on the ^1H NMR it was evident that starting material was converted to the hydroperoxide product. However under these acidic conditions, it appeared that the PEG chain was being cleaved since the ^1H NMR integrations of the CH_2 protons corresponding to the PEG was too high. Furthermore, the CH_2 protons of the PEG chain had small impurity peaks adjacent to them, most likely a result of free PEG present. Unfortunately, there was not enough time to optimize the reaction conditions, but a crude yield of 82% was obtained.

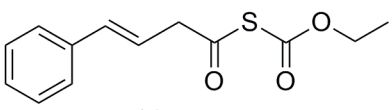
2.4 Xanthates as Precursors to Peroxyl Radical Clocks

2.4.1 Synthesis of Xanthates

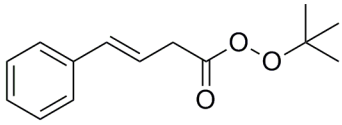
We were interested in synthesizing xanthates as precursors to the unsymmetrical allyl radical that drives the clock approach since they decompose readily under photolytic conditions. Furthermore, we were hoping that these xanthates (**19**, **20**) would increase the yield of decomposition products and could be used as an improvement to the first generation radical peroxyester radical clock (**21**).



19



20



21

Irradiation of xanthates with visible light generates radicals that can be trapped with alkenes or can provide other radicals through elimination reactions. Various radicals can be generated with the appropriate type of xanthate precursor. For example, acyl radicals can be made from S-acyl xanthates, such as O-ethyl derivative **22**. The sensitivity of xanthates towards visible light (leading to acyl radicals) was established by Barton, George, and Tomoeda,[30] invoking a radical recombination mechanism, instead of a chain process. The preparation of S-acyl xanthates is quite simple in theory, as they require an acid chloride reacting with a xanthate salt, such as potassium ethyl xanthate.

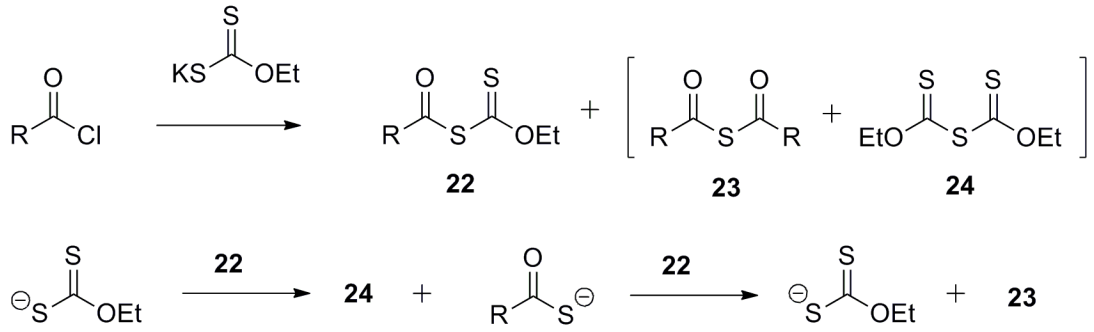
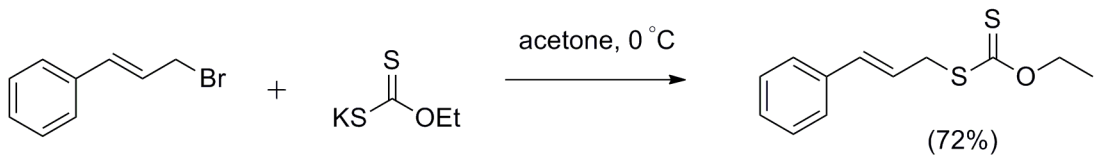
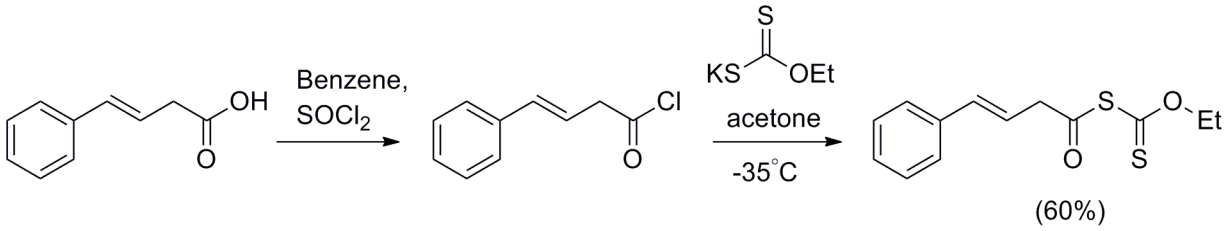


Figure 2.28: Formation of S-acyl xanthate and its decomposition by an ionic chain mechanism

Compound **19**, an S-alkyl xanthate was made in one step by reacting cinnamyl bromide with potassium O-ethyl xanthate in acetone at 0 °C. This S_N2 reaction proceeds very well within 1 hour, affording a yield of 72%.



Compound **20**, an S-acyl xanthate was synthesized in two steps. The first step was conversion of trans-styryl acetic acid to the acid chloride, followed by the formation of the S-acyl xanthate:



The synthesis of the S-acyl xanthate was not trivial. The reaction was carried out at 35°C and the xanthate salt was added dropwise in acetone to the acid chloride. The reaction was left to stir for 2 hours, but after this period there appeared to be several spots on the TLC plate. There appeared to be some decomposition of the product as well, and initially very low yields were obtained.

Several chemists have found that the preparation of acyl xanthates is often not very reproducible.[31] This is due to the fact that variable amounts of the thioanhydride **23** and O,O-diethyl xanthic anhydride **24** are formed in addition to the desired S-acyl xanthate **22**.[31] The reasoning behind this is that the S-acyl xanthate can decompose completely by an ionic chain mechanism in the presence of a small quantity of the xanthate salt.[31] The modified experimental procedure published by Zard avoids an excess of the xanthate salt, whereby the salt is added to a slight excess of the acid chloride in the dark. This modified procedure was used and the yield of the reaction to make the acyl xanthate improved to 60%.

2.4.2 UV and Decomposition Studies

We were interested in determining the molar extinction coefficient of the acyl and alkyl xanthate. Hence the absorbance was monitored over a wide concentration range to obtain the molar extinction coefficient (ε) by using UV spectroscopy. The extinction coefficients were determined by Beer-Lambert Law:

$$A = \varepsilon cl$$

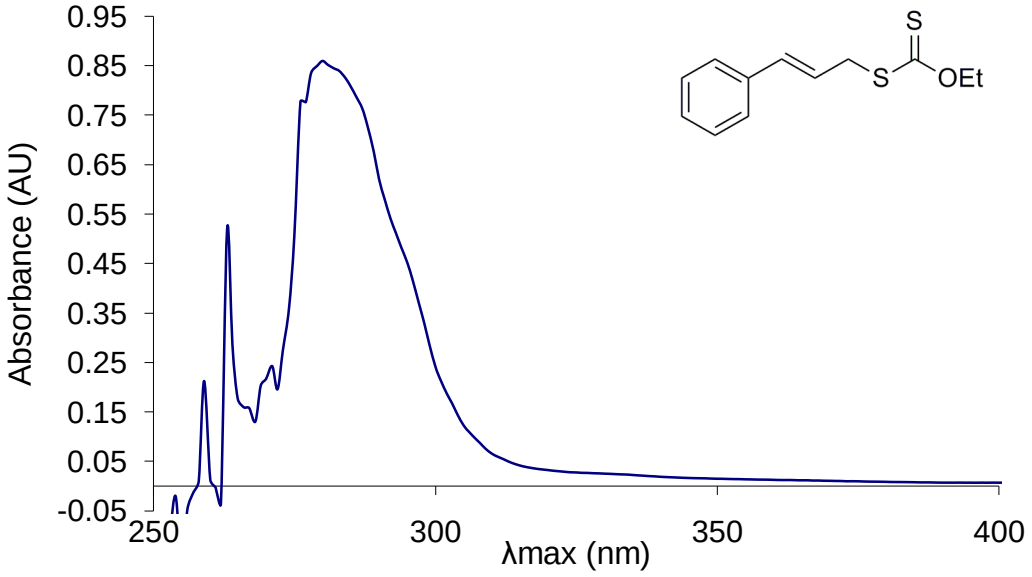


Figure 2.29: The absorbance spectrum of alkyl xanthate **19**. Concentration: 0.22 mM xanthate in benzene, 1 cm UV quartz cell, λ_{max} (nm) = 279 nm.

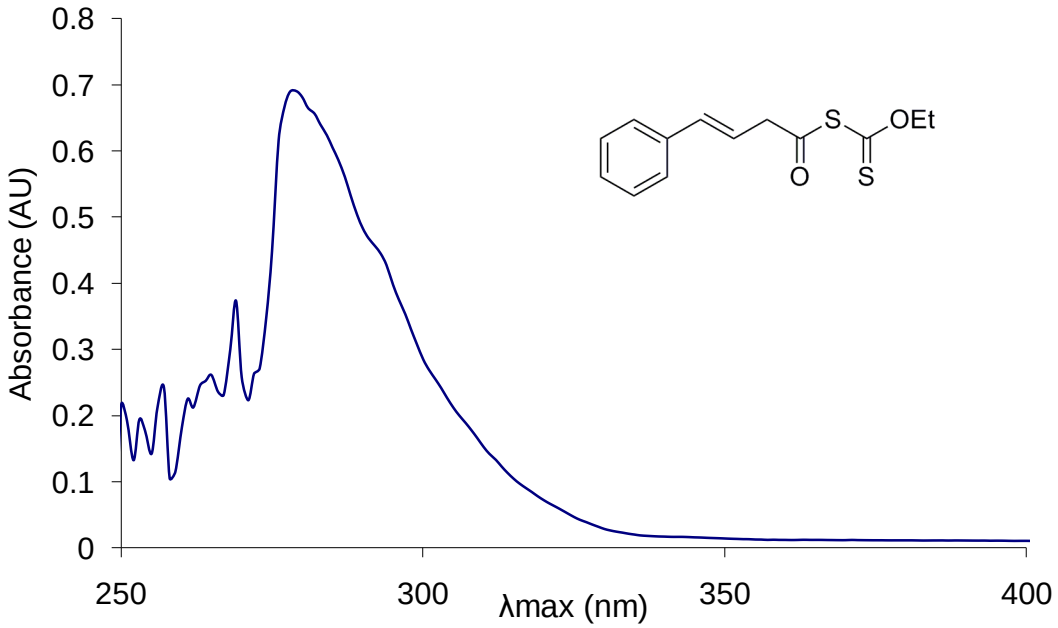
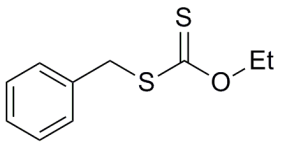
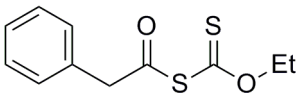


Figure 2.30: The absorbance spectrum of S-acyl xanthate **20**. Concentration: 0.22 mM xanthate in benzene, 1 cm UV quartz cell, λ_{max} (nm) = 279 nm.

For the UV-vis experiments, the samples were prepared in benzene. Although a background subtraction scan was taken, there is still a great deal of noise since benzene absorbs around 260 nm. Nonetheless, the xanthates both had absorptions which were shifted to higher wavelengths (279 nm) and the molar extinction coefficients were obtained from the slope of standard curves, since the path length (l) is 1 cm.



19'



20'

The molar extinction coefficients are compared with other similar acyl and alkyl xanthates shown in **Table 2.5** reported in literature by Barton, et. al (19' and 20') [30]. The values obtained were determined from the slope of standard curves, since the path length (l) is 1 cm.

Table 2.5: The molar extinction coefficients for acyl and alkyl xanthates at various λ_{max}

λ_{max} (nm)	ε ($M^{-1}cm^{-1}$) **19**	ε ($M^{-1}cm^{-1}$) **19'**	ε ($M^{-1}cm^{-1}$) **20**	ε ($M^{-1}cm^{-1}$) **20'**
279	28, 030	10, 900	22,599	
300	-	-	-	8,600
353	-	63	-	-
366	79	-	-	-
395	-	-	55	54

The molar absorpivity is a measure of how strongly a chemical species absorbs light at a given wavelength. From the experimental data obtained for compound **19** and **20**, we can see that the molar absorpivity at 353-395 nm is approximately the same compared to compounds **19'** and **20'** due to the n to π* transition in the xanthate moiety. Hence this experiment confirmed that with high molar absorptivities these compounds should be effective at absorbing light (of the appropriate wavelength) and would be good candidates for our decomposition experiments.

The decomposition of both xanthates was studied and characterized using the GC. The standards for the decomposition experiments were commercially available: α-vinylbenzyl alcohol and cinnamyl alcohol. The retention time of the alkyl and acyl xanthates was found to be 12.2 min and 11.90 minutes, respectively, using the chromatographic method mentioned in **Section 3.10**. The retention times of the alcohol standards, α-vinyl alcohol and cinnamyl alcohol, were 10.18 min and 15.00 min, respectively.

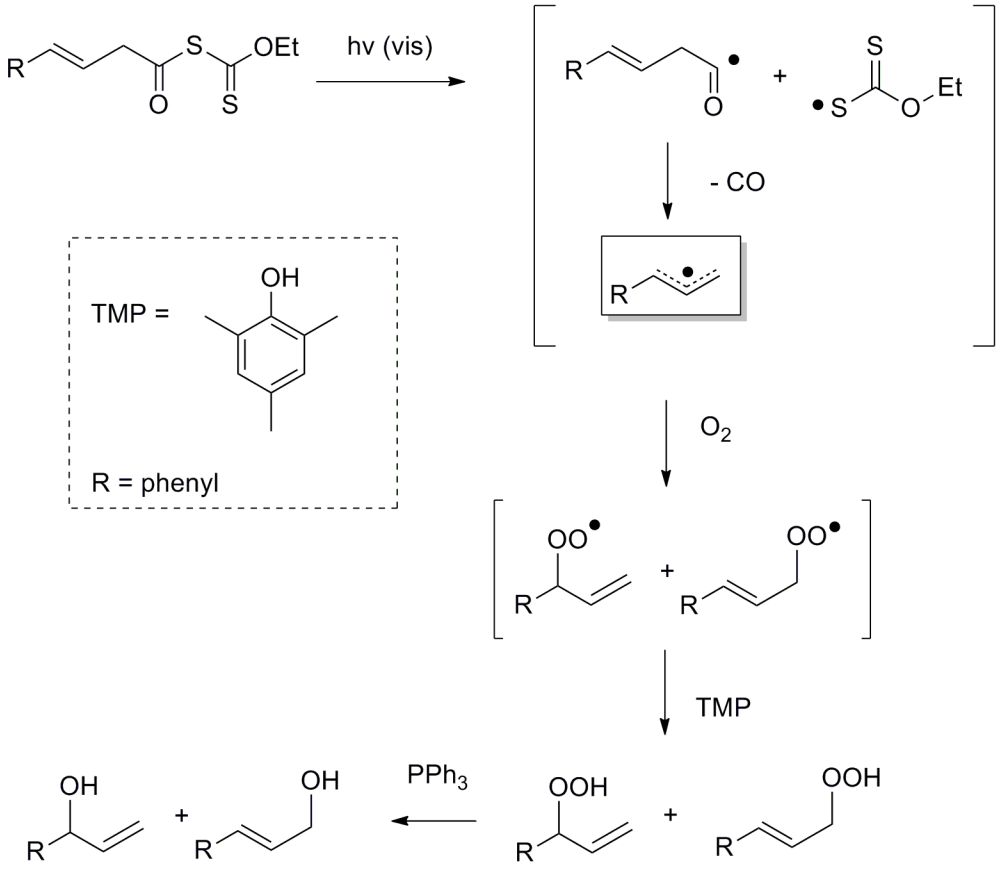


Figure 2.31: The decomposition of the S-acyl xanthate **20**

Solutions of the xanthates were prepared in benzene in the presence of 2,4,6-trimethylphenol (TMP) and the samples were irradiated for 1 hour using a sodium lamp (power in 430 W) . The reactions were quenched with PPh_3 and analyzed on the GC. However, the lack of product being formed (cinnamyl alcohol and α-vinylbenzyl alcohol) with both of the xanthates suggested that the experiments were not worth carrying on Although the xanthates appeared to be decomposing, (checked by TLC before they were analyzed by GC), it was difficult to extract the peak areas since the chromatography after 15 minutes was poor. We attempted to repeat the experiment by irradiating the samples in the photoreactor, but that did not improve the product formation of the conjugated and non-conjugated alcohols significantly.

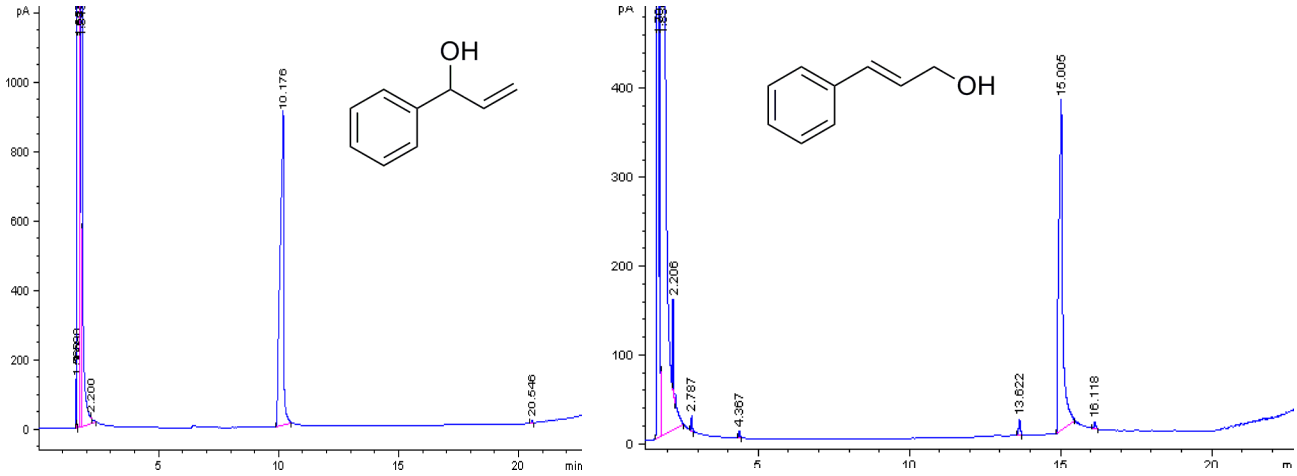


Figure 2.32: The GC chromatographs obtained for α-vinyl benzyl alcohol (left) and cinnamyl alcohol (right).

2.5 Conclusions and Outlook

The synthesis towards the carboxylated water-soluble target **11,** proved to be very challenging. Several routes towards compound **11'** were explored; however, the Wittig route worked the best. The synthesis of compound **11**, in addition to its decomposition products (alcohol standards) will be necessary for running decomposition experiments in the future.

The PEGylated route compound **12**, was synthesized successfully. Preliminary decomposition experiments have been performed and have shown that **12** decomposes in approximately 1 hour in the presence of Trolox. Furthermore, growth of the non-conjugated and conjugated products was evident in the HPLC chromatographs. It would be beneficial to repeat the experiments and determine the rate of beta fragmentation by varying the concentration of Trolox and seeing what effect it has on the formation of the non-conjugated and conjugated products.

The alkyl and acyl xanthates, **19** and **20**, were synthesized in fairly high yields. The decomposition experiments, carried out with TMP in benzene did not show a significant amount of product being formed by GC, but this was likely due to the fact that the chromatography was poor since the hydroperoxide products were visible by TLC (with a hydroperoxide stain). The decomposition of **19** and **20** will be studied further to optimize the conditions for the separation of the products. Furthermore, by varying the concentration of TMP, we can monitor the ratios of the decomposition products to obtain a rate constant.

CHAPTER 3

EXPERIMENTAL DETAILS

3.1 General

All chemicals were purchased from Sigma Aldrich or TCI unless otherwise indicated. THF and benzene were freshly distilled from a solution of blue ketyl formed by the reaction of sodium with a small amount of benzophenone. ^{1}H NMR spectra were recorded at 300 or 400 MHz. GC analysis was carried out with an Agilent Technologies 6890 equipped with a DB-5 column (30 m x 0.32 mm x 0.25 mm). The non-conjugated and conjugated alcohols from the xanthate decomposition were separated using the following temperature program: 95 °C for 2 min; 95-100 °C @ 0.5°C/min; 100-150°C @ 5°C/min; 150-280°C @ 40°C/min; 280 °C for 2.8 min. HPLC analysis was carried out on Waters Delta 600 equipped with a Waters 2487 Dual λ Absorbance Detector, using a XBridge Prep (reverse phase) C8 column (10 x 150 mm). The decomposition products were separated on the HPLC with a mobile phase of 60:40 methanol: ddH_2O (doubly distilled water) at 1 mL/min

3.2 Stille Route A to the Diacid 11'

But-3-ynoic acid, **11.1**

A solution of chromium trioxide (5.75 g, 57.5 mmol) and H_2SO_4 (5 M, 66 mL) was cooled to 0°C. A solution of 3-butyn-1-ol (1.85 g, 26.4 mmol) in 27 mL of acetone was added dropwise to the chromium trioxide solution. The reaction mixture was allowed to warm up to room temperature and was left to stir for 2 hours. The contents of the flask were poured onto an ethyl acetate/water mixture. The aqueous layer was extracted with ethyl acetate (3 x 15 mL). The organic extracts were combined, washed with brine and dried

with $MgSO_4$. The solvent was removed and the product was dried under vacuum. Yield: 1.4 g (62%) yellow solid. ^{1}H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ 3.40 (d, 2H), 2.27 (t, 1H).[46]

Tributylstannyl-4-(tributylstannyl)but-3-enoate, **11.2**

Under N_2 atmosphere, a toluene solution (5 mL) of but-3-ynoic acid (0.052 g, 0.62 mmol), tributyltin hydride (0.415 g, 1.43 mmol) and azobis(isobutyronitrile) (1 mg) was stirred at 100°C. After 3 hours, toluene was eliminated under vacuum and then carbon tetrachloride (0.74 mL) was added to react with the excess tributyltin hydride. After the solution was stirring for 1 hour, potassium fluoride solution (0.5 M, 0.8 mL) with acetone was added. The solution was filtered and then extracted with diethyl ether and then dried over magnesium sulphate. Yield: 0.253 g (62%) colourless oil. ^{1}H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ E isomer: 6.15 (dt, J = 18.9 Hz, 5.0 Hz, 1H), 6.09 (d, J = 18.9 Hz, 1H), 3.19 (d, J = 5.1 Hz, 2H), 0.87-1.63 (m, 71H). Z isomer: 6.69 (dt, J = 12.6 Hz, 7.1 Hz, 1H), 6.00 (dt, J = 12.6, 1.3 Hz, 1H), 3.08 (dd, J = 7.1 Hz, 1.3 Hz, 2H).[47]

Methyl 4-bromobenzoate

A flask containing 4-bromo-benzoic acid (10.0 g, 50 mmol), methanol (50 mL) and 0.3 g of sulphuric acid was refluxed for 24 hours. After cooling to room temperature, methanol was removed under reduced pressure and the resulting residue was extracted with methylene chloride. The organic extract was washed with water, dried over $MgSO_4$, filtered and concentrated under reduced pressure. Yield: 9.3 g (87%) white solid. ^{1}H

NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ 7.89/7.58 (^3J = 8.0 Hz, 4H, C_6H_4), 3.91(s, 3 H).[68]

(E)-methyl 4-(4-hydroxybut-1-en-1-yl)benzoate **11.3**

In a 10 mL flask were introduced toluene (0.84 mL), Tributylstannyl-4-(tributylstannyl)but-3-enoate (0.200 g, 0.30 mmol), methyl 4-bromobenzoate (0.065 g, 0.30 mmol) and tetrakis(triphenylphospine)palladium (3 mol%). The mixture was degassed under vacuum and stirred overnight at 100 °C. After cooling, the stannyl ester is hydrolyzed with 10 mL of 1 N HCl. After extraction with diethyl ether, the organic layer was treated with 1N NaOH solution. The aqueous layer was washed with ether and then acidified with 1N HCl solution and then extracted with ether. After removal of the solvents under reduced pressure, the product was purified by column chromatography on SiO_2 with EtOAc/MeOH (95/5). Yield: 0.026 g (42%) colourless oil. 7.33/6.87 (d, ^3J = 8.7 Hz, 4H, C_6H_4), 6.47 (d, ^3J = 16 Hz, 1H) 6.17 (m, ^3J = 16, 7 Hz, 1H) 3.82 (s, 1H, OCH_3), 3.30 (d, 2H, CH_2) ppm.

3.3. Stille Route B to Compound 11'

Tri-n-butyl(4-hydroxybut-3-enyl)stannane, **11.4**

3-Butynol (0.5 g, 7.14 mmol), tri-n-butyltin hydride (2.4 mL, 9.05 mmol) and AIBN (100 mg, 0.60 mmol) were heated at 80°C for 7 h under argon. The reaction mixture was allowed to cool to room temperature. Purification by silica gel chromatography (petroleum ether 40/60: ethyl acetate, 4:1 as eluent) gave a colourless oil. Yield: 2.2 g, 85%; ^1H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ

0.90 (t, 15H, J = 7.2 Hz, 3xSnCH2), 1.29-1.46 (m, 15 H, 3 x $CH_3CH_2CH_2CH_2Sn$), 1.48 (m, 6H J = 7.2 Hz, 3 x CH_2CH_2Sn), 1.50 (s, 1H, OH, disappears upon addition of D_2O), 2.43 (m, 2H, CCH2), 3.67 (t, 2 H, J = 6.2 Hz, CH_2OH), 5.93 (dt, 1H, J = 6.2, 19 Hz, C(H)CH2), 6.12 (dt, J = 19 Hz, 1H, C(H)Sn).[49]

(E)-methyl 4-(4-hydroxybut-1-en-1-yl)benzoate, **11.5**

In a 25 mL flask were introduced toluene (1.7 mL), Tri-n-butyl(4-hydroxybut-3-enyl)stannane (0.200 g, 0.55 mmol), methyl 4-bromobenzoate (0.112 g, 0.52 mmol) and tetrakis(triphenylphospine)palladium (3 mol%). The mixture was degassed under vacuum and stirred overnight at 100 °C. After cooling, the stannyl ester is hydrolyzed with 10 mL of 1 N HCl. After extraction with diethyl ether, the organic layer was treated with 1N NaOH solution. The aqueous layer was washed with ether and then acidified with 1N HCl solution and then extracted with ether. After removal of the solvents under reduced pressure, the product was purified by column chromatography on SiO_2 with 50/50 Pentane/EtOAc. Yield: 0.090 g (80%) colourless oil. 7.33/6.87 (d, ^3J = 8.7 Hz, 4H, C_6H_4), 6.47 (d, ^3J = 16 Hz, 1H) 6.17 (m, ^3J = 16, 7 Hz, 1H) 3.82 (s, 1H, OCH_3), 3.30 (d, 2H, CH_2) ppm.

3.4. Sonogashira Route to Compound 11'

Methyl 4-(4-hydroxybut-1-yn-1-yl)benzoate **11.6**

To a flame dried flask, methyl 4-bromobenzoate (5.40 g, 25.1 mmol) and triethylamine (60mL) were added under a nitrogen atomosphere, $Pd(Ph_3)_2Cl_2$ (0.352 g, 2 mol%), CuI (0.050 g, 1 mol%) followed by 1-butynol (1.81 g, 25.11 mmol) were added and the reaction was left to stir for 24 hours. The reaction was reduced to dryness and SiO_2 along with ethyl acetate were added to the crude product and the solvent was removed. The SiO_2 was then packed onto a fritted funnel and washed with 50/50 Hexanes/EtOAc and then hexanes. The filtrate was then reduced to dryness to give the desired product. Yield: 3.84 g (75%), yellow solid. ^{1}H NMR ($CDCl_3$): δ 8.00/7.49 (d, ^{3}J = 8.7 Hz, 4H, C_6H_4), 3.94 (s, 3H), 3.86 (q, 2H), 2.75 (t, 2H) ppm.

(E)-4-(4-(hydroxymethyl)phenyl)but-3-en-1-ol, **11.7**

A solution of lithium aluminum hydride (0.195 g, 5.02 mmol) in 2.0 mL of dry THF was N_2 purged for 5 minutes. Alkyne (2.4.1) (0.500 g, 2.45 mmol) in 2.0 mL of THF was slowly added to the lithium aluminum hydride solution at 0°C. The reaction was left to stir for 3 hours and hydrolysis of the ester was apparent by TLC. Two additional equivalents of lithium aluminum hydride (0.195 g, 5.02 mmol) was added and the reaction was heated 40 and left to stir for 12 hours. (A crude NMR was taken to ensure that the reduction of the alkyne had occurred). The reaction was cooled to room temperature and 0.4 mL of water was slowly added, followed by 0.4 mL of 15% NaOH and 1.2 mL H_2O. The mixture was stirred for 30 minutes followed by the addition of two scoops of $MgSO_4$ and additional stirring for 15 minutes. The solids were filtered off and

the solvent was removed under reduced pressure. Yield: 0.275 g (63%), yellow solid. ^{1}H NMR ($CDCl_3$): δ 7.36/7.30 (d, ^{3}J = 8.2 Hz, 4H, C_6H_4), 6.47 (d, ^{3}J = 16 Hz, 1H), 6.32 (m, ^{3}J = 15 Hz, 7 Hz, 1H) 3.62 (s, 2H), 3.68 (t, 2H), 2.43 (t, 2H) ppm.

3.5. Wittig Route To Compound 11'

Methyl 4-Formyl Benzoate

To a suspension of 4-formylbenzoic acid (2.00 g, 13.3 mmol) in dry acetonitrile (40 mL), 1,8-Diazabicycloundec-7-ene (DBU) (2.2 mL, 14.7 mmol) and methyl iodide (1.1 mL, 17.3 mmol) were added. The mixture was stirred at room temperature for 2 hours and then concentrated under vacuum. The residue was dissolved in ethyl acetate (50 mL) and this solution was washed with H_2O, (3 x 20 mL), aqueous HCl (0.4 N) (3 x 10 mL) and then dried ($MgSO_4$). Yield: 1.94 g (89%) white solid. ^{1}H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ 10.1 (s, 1H, aldehyde) 8.22/7.98 (^{3}J = 8.2 Hz, 4H, C_6H_4), 3.99 (s, 3H, OCH_3) ppm.[68]

(E)-methyl 4-(4-hydroxybut-1-en-1-yl)benzoate, **11.3**

A solution of potassium tert-butoxide (0.108 g, 0.958 mmol) in dry tetrahydrofuran (0.38 mL) was added portion wise to a mixture of methyl 4-formylbenzoate (0.062 g, 0.456 mmol) and (2-carboxyethyl)triphenylphosphonium bromide (0.200 g, 0.48 mmol) in tetrahydrofuran (0.53 mL) under cooling with ice-water for over 20 minutes. The reaction mixture was stirred at the same temperature for additional 30 minutes and then brought up to room temperature. After stirring for 10 hours, the reaction mixture was poured onto ice water and 10% $NaHCO_3$ was added. The mixture washed with Et_2O and the

aqueous layer was adjusted to pH 2 with 10% HCl and then extracted with ethyl acetate several times. The organic layer was dried over $MgSO_4$ and then evaporated under reduced pressure. The impure product was purified by column chromatography (60/40 – hexanes/ethyl acetate). Yield: 0.05 g, 41% off-white solid. ^{1}H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ 7.33/6.87 (d, ^{3}J = 8.7 Hz, 4H, C_6H_4), 6.47 (d, ^{3}J = 16 Hz, 1H) 6.17 (m, ^{3}J = 16, 7 Hz, 1H) 3.82 (s, 1H, OCH_3), 3.30 (d, 2H, CH_2) ppm.[54]

(E)-4-(3-carboxyprop-1-en-1-yl)benzoic acid, **11'**

Ester **11.3** (0.100, g, 0.45 mmol) was dissolved in 7.8 mL of ethanol. A solution of 1M NaOH (1 mL) was added dropwise to the reaction and was left to stir overnight at 40°C. The reaction was quenched with 6M HCl to pH 2 and was then extracted with dichloromethane 10 times. The combined organic phases were dried over Na_2SO_4 and evaporated. Crude Yield: 50 mg (56%), yellow solid. ^{1}H NMR (acetone-d_6): δ 7.93/6.87 (d, ^{3}J = 8.4 Hz, 4H, C_6H_4), 6.66-6.52 (m, 2H), 3.30 (d, 2H, CH_2) ppm.

3.6. PEGylated Perester Synthesis and Standards

3,6,9,12-Tetraoxatridecyl-4-toluenesulfonate, **12.3**

Tetraethyleneglycol monomethyl-ether (5.00 g, 24.0 mmol) was dissolved in 24 mL of dichloromethane. Benzyltriethylammonium chloride (0.217 g, 0.96 mmol) and 20 mL of 30% aqueous NaOH solution were added dropwise. Then a solution of p-toluenesulfonyl chloride (4.88 g, 25.6 mmol) in 23 mL of dichloromethane was added dropwise and the reaction mixture was left to stir for 20 hours at room temperature. The

white precipitate, which was formed in the organic layer was dissolved by adding 30 mL of water. The organic phase was separated, washed with 3 x 30 mL of water, and dried with $MgSO_4$. The solvent was removed, and the crude yellow residue was purified by column chromatography over SiO_2 with CH_2Cl_2/MeOH (50:1). The first yellow band was discarded before the colourless product was eluted. Yield: 7.9 g (91%) colourless oil. ^1H ^1H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ 7.80/7.34 (^3J = 8.0 Hz, 4H, C_6H_4), 4.16/3.69 (4H, $SO_3CH_2CH_2$), 3.72-3.46 (m, 12H, OCH_2), 3.38 (s, 3H, OCH_3), 2.45 (s, 3H, aryl CH_3) ppm.[56]

4-(1,4,7,10,13-Pentaoxatetradecyl) benzaldehyde, **12.2**

4-Hydroxybenzaldehyde (1.53 g, 12.5 mmol) K_2CO_3 (5.18 g, 37.5 mmol) and **12.1** (5.00 g, 13.8 mmol) of were dissolved in 42 mL of dry DMF under N_2 atmosphere and heated under reflux. After 48 hours, the conversion was quantitative when monitored by TLC. The reaction was cooled down to room temperature and filtered, followed by evaporation of the filtrate. The residue was dissolved in CH_2Cl_2 and filtered again to remove any soluble components. The filtrate was evaporated and purified by column chromatography over SiO_2 with CH_2Cl_2/MeOH (100:1). Yield: 3.92 g (88%), nearly colourless viscous oil. ^1H NMR was in agreement with chemical shifts reported in literature NMR ($CDCl_3$): δ 9.89 (s, 1H, CHO) 7.83/7.04 (^3J = 8.9 Hz, 4H, C_6H_4), 4.22/3.90 (4H, aryl-OCH_2CH_2), 3.76-3.51 (m, 12H, OCH_2), 3.38 (s, 3H, OCH_3) ppm.[56]

4-(1,4,7,10,13-Pentaoxatetradecyl)-(E)-(3-carboxyprop-1-en-1-yl) benzene, **12.1**

A solution of potassium tert-butoxide (1.74 g, 15.5 mmol) in dry tetrahydrofuran (4.7 mL) was added portionwise to a mixture of **12.2** (2.19 g, 7.03 mmol) and (2-carboxyethyl)triphenylphosphonium bromide (3.07 g, 7.40 mmol) in tetrahydrofuran (6.4 mL) under cooling with ice-water for over 20 minutes. The reaction mixture was stirred at the same temperature for additional 30 minutes and then brought up to room temperature. After stirring for 10 hours, the reaction mixture was poured onto ice water and 10% $NaHCO_3$ was added. The mixture washed with Et_2O and the aqueous layer was adjusted to pH 2 with 10% HCl and then extracted with ethyl acetate several times. The organic layer was dried over $MgSO_4$ and then evaporated under reduced pressure. The semi-solid was recrystallized from cold ether. Yield: 2.02 g (78%) off-white semi solid. ^{1}H NMR ($CDCl_3$): δ 7.31/6.87 (^{3}J = 8.9 Hz, 4H, C_6H_4), 6.47 (d, 1H – olefin, ^{2}J = 15.8 Hz), 6.15 (m, 1H – olefin, ^{3}J = 15.5, 7 Hz) 4.14/3.87 (4H, aryl-OCH_2CH_2), 3.76-3.55 (m, 12H, OCH_2), 3.55 (s, 3H, OCH_3), 3.29 (d, 2H, CH_2) ppm.

(E)-tert-butyl 4-(4-(2-methoxyethoxy)phenyl)but-3-eneperoxoate, **12**

A solution of **12.1** (0.050 g, 0.136 mmol) in anhydrous benzene (0.45 mL) was treated with $SOCl_2$ (20 uL, 0.272 mmol) and the mixture was left to stir for 4 hours at room temperature. The conversion to the acid chloride was quantitative by TLC. The reaction was cooled down to 5°C, pyridine (22 uL, 0.272 mmol), followed by tert-butyl hydroperoxide (25 uL, 0.136 mmol) was added dropwise. The mixture was left to stir in the dark for an additional 30 minutes. The reaction mixture was quenched with ice-water and extracted with dichloromethane 10 times. Yield: 56 mg, (93% crude). ^{1}H NMR

($CDCl_3$): δ 7.30/6.88 (^3J = 8.7 Hz, 6H, C_6H_4), 6.50 (d, 1H – olefin, ^2J = 15.7 Hz), 6.14 (m, 1H – olefin, ^3J = 15.6, 7.2 Hz) 4.15/3.87 (7.5H, aryl-OCH_2CH_2), 3.75-3.55 (m, 24H, OCH_2), 3.39 (s, 5H, OCH_3), 3.29 (d, 2H, CH_2), 1.35 (s, 9H, t-butyl) ppm.

1-(4-(2-methoxyethoxy)phenyl)prop-2-en-1-ol, **12.2.1**

In an oven dried flask, 1.0 M vinyl magnesium bromide in THF was added to a solution of **12.2** (0.100 g, 0.320 mmol) in 0.45 mL of THF at 0 °C. After 15 minutes, the reaction was allowed to warm up to room temperature and stirred for an additional hour. The reaction was quenched by addition of saturated aqueous NH_4Cl and extracted with diethyl ether. The organic phase was washed with brine, dried over $MgSO_4$ and the solvent was subsequently removed to give the product. No purification was necessary. TLC conditions: CH_2Cl_2/MeOH (98%/2%). Yield: 94 mg (86%), colourless oil. ^1H NMR ($CDCl_3$): δ 7.26/6.88 (^3J = 8.6 Hz, 4H, C_6H_4), 6.02 (^3J = 16.2, 10.3, 5.9 Hz 1H, olefin), 5.30 (^3J = 17.1 Hz, 1H, carbon with OH), 5.14 (^3J = 15.5, 10.3 Hz, 2H, terminal H on olefin), 4.11/3.83 (4H, aryl-OCH_2CH_2), 3.71-3.51 (m, 12H, OCH_2), 3.35 (s, 3H, OCH_3) ppm.

(E)-ethyl 3-(4-(2-methoxyethoxy)phenyl)acrylate, **12.2.2**

A flask containing PEGylated aldehyde (0.400 g, 1.28 mmol) and lithium chloride (0.060 g, 1.41 mmol) in 4.3 mL of acetonitrile was cooled down to 0 °C. Triethylphosphonoacetate (0.26 mL, 1.28 mmol) was subsequently added. After 15 minutes, 1,8-Diazabicycloundec-7-ene (0.19 mL, 1.28 mmol) was added dropwise. The reaction was left to stir for 12 hours at room temperature. The reaction was then diluted

with diethyl ether washed with a saturated solution of ammonium chloride, brine and dried over $MgSO_4$, filtered over celite and the filtrate was concentrated to dryness. The crude product was purified by column chromatography over SiO_2 with CH_2Cl_2/MeOH (97%:3%). Yield: 0.477 g (98%), nearly colourless viscous oil. ^{1}H NMR ($CDCl_3$): δ 7.65/6.32 (d, ^{3}J = 15.9 Hz, 2H, olefin H) 7.45/6.93 (^{3}J = 7.5 Hz, 4H, C_6H_4), 4.26 (2H, CH_2 ethyl) 4.16/3.89 (4H, aryl-OCH_2CH_2), 3.74-3.52 (m, 12H, OCH_2), 3.39 (s, 3H, OCH_3) ppm.

*(E)-3-(4-(2-methoxyethoxy)phenyl)prop-2-en-1-ol,***12.2.3**

To an oven dried flask, diisobutylaluminium hydride (1.13 mL, 1.13 mmol) was added at -78 °C to **12.2.2** (0.174 g, 0.454 mmol) in 3.0 mL of toluene. The reaction was stirred at -78 °C for 1 hour and was left to heat up gradually to room temperature overnight. The reaction was diluted with CH_2Cl_2 and a saturated solution of sodium sulphate was added to precipitate out the aluminum salts. $MgSO_4$ was added, filtered off and then the filtrate was reduced to dryness. The crude product was purified by column chromatography over SiO_2 with CH_2Cl_2/MeOH (97%:3%). Yield: 90 mg (58%), nearly colourless viscous oil. ^{1}H NMR ($CDCl_3$): δ 7.31/6.87 (d, ^{3}J = 8.7 Hz, 4H, C_6H_4), 6.55 (d, ^{3}J = 15.9 Hz, 1H, olefin), 6.24 (m, ^{3}J = 15.8, 12, 5.9 Hz, 1H, olefin) 4.30 (d, ^{3}J = 5.9 Hz, CH_2) 4.14/3.86 (4H, aryl-OCH_2CH_2), 3.75-3.54 (m, 12H, OCH_2), 3.38 (s, 3H, OCH_3) ppm.[54]

3.7 Test Reactions – Model Compounds

(E)-4-(4-methoxyphenyl)but-3-enoic acid, **12.1.1**

A solution of potassium tert-butoxide (2.30 g, 20.2 mmol) in dry tetrahydrofuran (6.1 mL) was added portionwise to a mixture of p-methoxy benzaldehyde (1.5 g, 9.14 mmol) and (2-carboxyethyl)triphenylphosphonium bromide (3.99 g, 9.61 mmol) in tetrahydrofuran (8.1 mL) under cooling with ice-water for over 20 minutes. The reaction mixture was stirred at 0 ºC for additional 30 minutes and then brought up to room temperature. After stirring for 10 hours, the reaction mixture was poured onto ice water and 10% $NaHCO_3$ was added. The mixture washed with Et_2O and the aqueous layer was adjusted to pH 2 with 10% HCl and then extracted with ethyl acetate several times. The organic layer was dried over $MgSO_4$ and then evaporated under reduced pressure. The solid was recrystallized from cold ether. Yield: 0.86 g (49%), white solid. ^{1}H NMR was in agreement with chemical shifts reported in literature ($CDCl_3$): δ 7.33/6.87 (^3J = 8.7 Hz, 4H, C_6H_4), 6.49 (^3J = 15.9 Hz, 1H, olefin), 6.16 (^3J = 15.7, 7.1 Hz, m, 1H, olefin), 3.83 (s, 3H, CH_3), 3.30 (d, 2H, CH_2) ppm.[54]

(E)-tert-butyl 4-(4-methoxyphenyl)but-3-eneperoxoate, **12.1.2**

A solution of the p-methoxy acid (2.7.1) (0.100 g, 0.521 mmol) in anhydrous benzene (1.7 mL) was treated with $SOCl_2$ (76 uL, 1.04 mmol) and the mixture was left to stir for 4 hours at room temperature. The conversion to the acid chloride was quantitative by TLC. Benzene and excess $SOCl_2$ were removed reduced pressure. The residue obtained was dissolved in dry THF (3.5 mL), cooled to -20°C and pyridine (0.15 mL, 1.82 mmol) dissolved in 0.82 mL of THF was added slowly to the reaction, followed

by the slow addition of *tert*-butyl hydroperoxide (0.24 mL, 1.30 mmol). The mixture was left to stir in the dark for an additional 30 minutes followed by evaporation of most of the solvent. The concentrated crude reaction was loaded onto a SiO_2 column and purified with a mobile phase of Hexanes/EtOAc (80%/20%). Yield: 32 mg, (23%). ^{1}H NMR ($CDCl_3$): δ 7.32/6.87 (^{3}J = 8.7 Hz, 4H, C_6H_4), 6.51 (^{3}J = 16 Hz, 1H, olefin), 6.14 (^{3}J = 14.3, 7.0 Hz, 1H, m, olefin), 3.83 (s, 3H, CH_3), 3.26 (d, 2H, CH_2), 1.35 (s, 9H, t-butyl) ppm.

3.8 PEGylated Hydroperoxide

1-(4-(2-methoxyethoxy)phenyl)ethanone, **14.3**

4'-hydroxyacetophenone (0.854 g, 6.27 mmol), K_2CO_3 (2.60g, 18.8 mmol), **12.1** (2.50 g, 6.90 mmol) were dissolved in 20 mL of dry DMF under N_2 atmosphere and heated under reflux. After 48 hours, the conversion was quantitative when monitored by TLC. The reaction was cooled down and filtered, and the filtrate was evaporated. The residue was dissolved in CH_2Cl_2 and filtered again to remove any soluble components. The filtrate was evaporated and purified by column chromatography over SiO_2 with CH_2Cl_2/MeOH (98/2%). Yield: 1.84 g (90%), nearly colourless viscous oil. ^{1}H NMR ($CDCl_3$): δ 7.93/6.95 (^{3}J = 8.9 Hz, 4H, C_6H_4), 4.20/3.89 (4H, aryl-OCH_2CH_2), 3.76-3.54 (m, 12H, OCH_2), 3.38 (s, 3H, OCH_3) ppm.

2-(4-(2-methoxyethoxy)phenyl)propan-2-ol, **14.2**

A solution of PEGylated ketone (0.457 g, 1.40 mmol) in 0.42 mL of THF was added dropwise to a flask containing 3 M methyl magnesium iodide (0.63 mL, 1.9 mmol) in 0.42 mL of THF. The reaction was left to stir for 4 hours at room temperature. The reaction was quenched with the slow addition of 0.28 mL of H_2O followed by the extraction with diethyl ether (5 x 10 mL). The organic layer was dried with magnesium sulphate and the solvent was removed under reduced pressure. Yield: 0.364 g (76%), nearly colourless oil. ^{1}H NMR ($CDCl_3$): δ 7.94/6.97 (^{3}J = 8.9 Hz, 4H, C_6H_4), 4.21/3.90 (4H, aryl-OCH_2CH_2), 3.71-3.55 (m, 12H, OCH_2), 3.39 (s, 3H, OCH_3), 2.58 (s, 3H, CH_3), 1.62 (s, 3H, CH_3) ppm.

1-(2-hydroperoxypropan-2-yl)-4-(2-methoxyethoxy)benzene, **14.1**

To an intensively stirred mixture of alcohol (0.070 g, 0.206 mmol) in 1,2-dichloroethane (0.4 mL) was added at 50°C a solution of H_2O_2/H_2SO_4 (0.188 mL/1.85 uL). The reaction was left to stir for 3 hours and formation of the product was observed by TLC (visible with NaI/Acetic acid). The reaction was cooled to room temperature and 5 mL of 5% $NaHCO_3$ was added. The aqueous layer was washed with 1,2-dichloroethane (5 x 10 mL) and the organic layers were combined, then washed with $(NH_4)_2SO_4$, water, and dried with magnesium sulphate. The solvent was removed under reduced pressure to give a colourless oil. Crude Yield: 82%. ^{1}H NMR ($CDCl_3$): δ 7.81/7.78 (^{3}J = 8.5Hz, 4H, C_6H_4), 4.09/3.84 (4H, aryl-OCH_2CH_2), 3.71-3.57 (m, 12H, OCH_2), 3.40 (s, 3H, OCH_3), 2.58 (s, 3H, CH_3), 1.60 (s, 3H, CH_3) ppm.

3.9. Xanthate Synthesis

S-cinnamyl O-ethyl carbonodithioate, **19**

Cinnamyl bromide (0.500 g, 2.54 mol) and potassium ethyl xanthate (0.407 g, 2.54 mmol) were stirred at 0-10 °C in 5 mL of acetone for about 1 h. The insoluble inorganic halide was filtered, and the acetone solution was evaporated under vacuum to give a yellow solid. Yield: 0.432 g (72%) ^{1}H NMR ($CDCl_3$): δ 7.38-7.25 (^{3}J = 8.6 Hz, 5H, C_6H_5), 6.56 (^{3}J = 15.7 Hz, d, 1H, olefin H), 6.22 (^{3}J = 14.5, 7.2 Hz, m, 1H, olefin H), 4.69 (q, 2H, CH_2 ethyl), 3.49 (d, 2H, CH_2), 1.48 (t, 3H, CH_3 ethyl) ppm.^{13}C NMR ($CDCl_3$): δ 214.3, 136.8, 134.3, 129.0, 128.2, 126.8, 123.8, 123.4, 70.7, 39.0, 14.2 ppm.

(O-ethyl carbonothioic) (E)-4-phenylbut-3-enoic thioanhydride, **20**

Trans-styryl acetic acid (0.201 g, 1.24 mmol) was dissolved in 4 mL of dry benzene, followed by the addition of thionyl chloride (184 uL, 2.48 mmol). The reaction was left to stir for 4 hours and excess thionyl chloride and benzene were removed. The acid chloride was then dissolved in 1 mL of acetone and cooled to -30 °C. Potassium O-ethyl xanthate (0.199 g, 1.24 mmol) was added portionwise (with a dropping funnel) to the reaction mixture over 30 minutes. The reaction was stirred at -35 °C for 1 hour. The reaction was then allowed to warm up to room temperature. The solvent was removed in vacuuo, water (0.3 mL) was added to the residue and the product was extracted into CH_2Cl_2 (2 x 10 mL). The extract was washed with 10% $NaHCO_3$, then with water, and dried over $MgSO_4$. Removal of the solvent in vaccuo gave the desired product. Yield: 0.20 g (60%) yellow solid ^{1}H NMR ($CDCl_3$): δ 7.37-7.24 (^{3}J = 7.3 Hz, 5H, C_6H_5), 6.63 (^{3}J = 15.7 Hz, d, 1H, olefin H), 6.25 (^{3}J = 15.7, 7.5 Hz, m, 1H, olefin H), 4.68 (q, 2H, CH_2

ethyl), 3.97 (d, 2H, CH_2), 1.44 (t, 3H, CH_3 ethyl) ppm. ^{13}C NMR ($CDCl_3$): δ 203.5, 190.3, 136.3, 128.7, 128.1, 126.5, 119.2, 71.1, 47.8, 13.6 ppm.

3.10 Xanthate Decomposition Procedure (photolytic)

Stock solutions of TMP (trimethylphenol) (1.0 M and 2.0 M) and the desired xanthate (**19** or **20**, 0.1 M) were freshly prepared in benzene (without additional aeration) before use. TMP (0.1 M) and xanthate (**19** or **20**, 0.01 M, 10 µL of 0.1 M) were mixed and diluted up to 100 µL in benzene in 2 mL vials. The vials were capped were irradiated using UVB lamps (centred at 300 nm) in a Luzchem photoreactor (LZC-ORG) for 1 hour at 25 °C. After the irradiation, reaction mixtures were quenched with PPh_3 (50 uL in 1.0 M in benzene) and diluted up to 1.8 mL with hexanes. Decomposition products were determined by GC.

3.11 PEGylated Peroxyester Decomposition Procedure (thermolytic)

Stock solutions of peroxyester 12 (0.1 M) and trolox (1.0 M) were freshly prepared using MeOH/water. Perester 12 (0.01 M, 50 µL of 0.1 M) and Trolox (0.1 M, 50 µL of 1.0 M) were mixed and diluted up to 500 µL in 60/40 MeOH/water. 50 µl aliquots were withdrawn from the sample vial every 30 minutes and quenched with TCEP (tris(2-carboxyethyl)phosphine) (0.05 M, 50 µL of 0.50 M). 10 µL of the quenched reaction mixture was injected onto the HPLC every 30 minutes to analyze the decomposition products.

3.12 References

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