The Synthesis and Antioxidant Capacities of a range of Resveratrol and Related Phenolic Glucosides.

A thesis presented in fulfilment of the requirements for the degree of

Doctor of Philosophy

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Abstract

The resveratrol analogues have attracted great attention by scientists as these compounds exhibit numerous bioactive properties due to their outstanding antioxidant capacity. However, the role of the antioxidant activity of these molecules is still not quite clear. This thesis details the development and biological evaluation of a library of resveratrol analogues in order to provide a better understanding of their pharmaceutical value.

This thesis begins with an overview of an important hydroxylated stilbene (resveratrol) and its analogues present in natural plants, food and beverage. Consequently, these studies are summarised and aided in the selection of a new library of substrates to be synthesised herein and biologically evaluated.

Chapter two details the successful synthesis of resveratrol glycosides from resveratrol. Pleasingly, all chemical transformations carried out herein were performed in excellent yields. In-vitro anti-oxidant studies on these substrates revealed glycosylation of resveratrol leads to a decreased antioxidant capacity. In addition, these studies suggested the para hydroxyl group on resveratrol has a higher reactivity than the meta hydroxyl group.

Chapter three details the synthesis of a hydroxylated resveratrol (piceatannol) and many of its glycosides. Almost all of the targeted compounds were prepared by applying a modified strategy designed for resveratrol glycosides in high efficiency. The anti-oxidant assays suggested that piceatannol is a more powerful antioxidant than resveratrol. The assays also revealed that the antioxidant activity of piceatannol glycosides is quite dependent on the glycosylation position.

Chapter four then details the preparation of several common resveratrol dimers. The individual products were obtained via a one step oxidation of resveratrol followed by acetylation of the products, separation, and base hydrolysis. In addition, successful isomerisation of some of the trans-dimers into their cis forms was achieved in this study. With a simple protocol now in place to synthesise such resveratrol dimers, it paves the way for future work on the synthesis of glucosylated dimers of resveratrol. Such compounds would be expected to have a diverse range of antioxidant properties and other related bioactivities and are worthy of further exploration.
Finally, Chapter five contains the associated experimental procedures and characterisation data for all synthesised resveratrol and piceatannol analogues along with a range of oligomers.
List of publications


Declaration

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

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Q Mao

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## Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>ABS</td>
<td>Absorbance</td>
</tr>
<tr>
<td>br</td>
<td>Broad</td>
</tr>
<tr>
<td>COSY</td>
<td>Correlation spectroscopy</td>
</tr>
<tr>
<td>cm</td>
<td>Centimetres</td>
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<td>d</td>
<td>Doublet</td>
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<tr>
<td>DCM</td>
<td>Dichloromethane</td>
</tr>
<tr>
<td>DMSO</td>
<td>Dimethyl sulfoxide</td>
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<tr>
<td>dd</td>
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</tr>
<tr>
<td>ddd</td>
<td>Doublet of doublet of doublets</td>
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<tr>
<td>DPPH</td>
<td>2,2-Diphenyl-1-picrylhydrazyl-1,1-diphenyl-2-picrylhydrazyl</td>
</tr>
<tr>
<td>Et</td>
<td>Ethyl</td>
</tr>
<tr>
<td>Et₂O</td>
<td>Diethyl ether</td>
</tr>
<tr>
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<td>Ethyl acetate</td>
</tr>
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<td>Ethanol</td>
</tr>
<tr>
<td>FeCl₃</td>
<td>Ferric chloride</td>
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<tr>
<td>FRAP</td>
<td>Ferric cyanide reducing antioxidant power assay</td>
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<tr>
<td>g</td>
<td>Grams</td>
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<td>GC</td>
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<td>GCMS</td>
<td>Gas chromatography mass spectrometry</td>
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<td>Hours</td>
</tr>
<tr>
<td>HCl</td>
<td>Hydrochloric acid</td>
</tr>
<tr>
<td>HMBC</td>
<td>Heteronuclear multiple bond correlation</td>
</tr>
<tr>
<td>HMQC</td>
<td>Heteronuclear multiple quantum coherence</td>
</tr>
<tr>
<td>HPLC-DAD</td>
<td>High-Performance Liquid Chromatography-Diode-Array Detection</td>
</tr>
<tr>
<td>HRMS</td>
<td>High resolution mass spectrometry</td>
</tr>
<tr>
<td>Hz</td>
<td>Hertz</td>
</tr>
<tr>
<td>hν</td>
<td>Light/irradiation</td>
</tr>
<tr>
<td>J</td>
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</tr>
<tr>
<td>K₂CO₃</td>
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</tr>
<tr>
<td>L</td>
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<tr>
<td>LC-MS</td>
<td>Liquid chromatography-mass spectrometry</td>
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<tr>
<td>Lit.</td>
<td>Literature</td>
</tr>
<tr>
<td>Symbol</td>
<td>Term</td>
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<td>------</td>
</tr>
<tr>
<td>m</td>
<td>Multiplet</td>
</tr>
<tr>
<td>M</td>
<td>Molar (moles/litre)</td>
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<tr>
<td>min.</td>
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</tr>
<tr>
<td>m/z</td>
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</tr>
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</tr>
<tr>
<td>M.pt.</td>
<td>Melting point</td>
</tr>
<tr>
<td>MS</td>
<td>Mass spectrometry</td>
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</tr>
<tr>
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<td>Sodium</td>
</tr>
<tr>
<td>NaHCO₃</td>
<td>Sodium bicarbonate</td>
</tr>
<tr>
<td>NMR</td>
<td>Nuclear magnetic resonance</td>
</tr>
<tr>
<td>OAc</td>
<td>Acetate</td>
</tr>
<tr>
<td>Piv</td>
<td>Pivaloyl</td>
</tr>
<tr>
<td>ppm</td>
<td>Parts per million</td>
</tr>
<tr>
<td>psi</td>
<td>Pounds per square inch</td>
</tr>
<tr>
<td>q</td>
<td>Quartet</td>
</tr>
<tr>
<td>Rᶠ</td>
<td>Retension factor</td>
</tr>
<tr>
<td>rt</td>
<td>Room temperature</td>
</tr>
<tr>
<td>s</td>
<td>Singlet</td>
</tr>
<tr>
<td>t</td>
<td>Triplet</td>
</tr>
<tr>
<td>TBS</td>
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</tr>
<tr>
<td>THF</td>
<td>Tetrahydrofuran</td>
</tr>
<tr>
<td>TLC</td>
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</tr>
<tr>
<td>UV</td>
<td>Ultra-violet</td>
</tr>
<tr>
<td>d</td>
<td>Chemical shift</td>
</tr>
<tr>
<td>m</td>
<td>Micro</td>
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“God in his goodness sent the grapes, to cheer both great and small; little fools drink too much, and great fools not at all.”

-Anonymous

‘Wine is a biochemical challenge. It is a daunting task to probe the alchemy of this elixir and to determine what lies ‘at the heart of the matter’.’

-Goldberg
CHAPTER 1: INTRODUCTION.

1.1 Polyphenolics and Antioxidants.

Natural occurring polyphenolic compounds are of great interest to those in the health and nutritional fields due to the numerous reports over the decades that these compounds exhibit unique health benefits including anticancer, anticarcinogenic, antibacterial, antiviral, antioxidant and anti-inflammatory activities.\textsuperscript{1-5} These compounds originate from a large variety of dietary foods such as fruits, vegetables, nuts, grains and beverages such as wine, beer, tea and fruit juice.\textsuperscript{1-5} For example, berries such as grapes, blueberries, strawberries and cherries are fruits that are well known to contain high levels of a diverse range of polyphenolic compounds. \textit{Figure 1.1}. 

\textit{Figure 1.1}. Berries are highly rich in polyphenols.

Given the high interest in these compounds over the past decades, constant effort has been made by chemists to identify and quantify of these substances with the expectation to understand better how to create a healthier diet for humans. With the aid of modern detection and separation techniques such as HPLC, LC-MS and NMR, numerous polyphenolic compounds have been identified and separated from plants and foodstuffs thus far. These compounds may be classified into many sub-groups, including phenolic acids, stilbenes, chalcones, coumarins, flavonoids, and anthocyanins. \textit{Figure 1.2}.\textsuperscript{6-10} It has now been well established that there is a positive relationship between the pharmaceutical benefits of polyphenols and their antioxidant properties due to the finding that the latter plays a key role in quenching of harmful free radicals that cause many destructive diseases such as cardiovascular disease and cancer.\textsuperscript{11-13}
In order to examine the antioxidant capacity of these polyphenolic compounds, scientists have developed a variety of useful assays (DPPH, TEAC, FRAP, HOCl and deoxyribose assay) which are able to be conducted on large sample sizes or individually purified compounds with ease.\textsuperscript{14-17} The performance of each assay has also been examined. For example, Soobrattee et al. have measured the antioxidant capacity of some common polyphenols in fruits and vegetables based on a range of \textit{in-vitro} assays including TEAC, FRAP, HOCl and the deoxyribose assay, \textit{Table 1.1}.\textsuperscript{18} It was found that individual polyphenolic compounds may perform different antioxidant activities in different type of assays. For example, quercetin shows the highest antioxidant value in TEAC assay while cyanidin chloride is the most powerful antioxidant in FRAP assay. Thus, any single assay is not able to accurately evaluate the antioxidant activity of a polyphenolic compound.
Table 1.1. Antioxidant capacities of some common polyphenols measured by TEAC, FRAP, HOCl and the deoxyribose assay.18

<table>
<thead>
<tr>
<th>Compounds</th>
<th>Classification</th>
<th>TEAC (mmol trolox/L)</th>
<th>FRAP (µM Fe(II)/L)</th>
<th>HOCl scavenging assay (µM, IC50)</th>
<th>Deoxyribose assay (Rate constant M⁻¹s⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gallic acid</td>
<td>Simple phenolic compounds</td>
<td>3.62</td>
<td>5.25</td>
<td>281.0</td>
<td>------</td>
</tr>
<tr>
<td>Ellagic acid</td>
<td></td>
<td>1.05</td>
<td>4.39</td>
<td>499.49</td>
<td>3.57 x 10¹²</td>
</tr>
<tr>
<td>Ferulic acid</td>
<td>Hydroxycinnamic acids</td>
<td>0.98</td>
<td>1.33</td>
<td>22.65</td>
<td>1.55 x 10¹²</td>
</tr>
<tr>
<td>Rosmarinic acid</td>
<td></td>
<td>4.50</td>
<td>6.21</td>
<td>23.55</td>
<td>1.82 x 10¹²</td>
</tr>
<tr>
<td>3-Coumaric acid</td>
<td></td>
<td>0.17</td>
<td>------</td>
<td>1878.17</td>
<td>0.95 x 10¹²</td>
</tr>
<tr>
<td>Catechin</td>
<td>Flavanols</td>
<td>3.16</td>
<td>2.47</td>
<td>52.06</td>
<td>2.93 x 10¹²</td>
</tr>
<tr>
<td>Epicatechin</td>
<td></td>
<td>3.58</td>
<td>2.90</td>
<td>23.87</td>
<td>3.57 x 10¹²</td>
</tr>
<tr>
<td>Luteolin</td>
<td>Flavones</td>
<td>2.23</td>
<td>2.23</td>
<td>91.19</td>
<td>4.52 x 10¹²</td>
</tr>
<tr>
<td>Apigenin</td>
<td></td>
<td>2.01</td>
<td>2.01</td>
<td>330.46</td>
<td>3.44 x 10¹²</td>
</tr>
<tr>
<td>Quercetin</td>
<td>Flavonols</td>
<td>3.68</td>
<td>7.39</td>
<td>85.5</td>
<td>5.80 x 10¹²</td>
</tr>
<tr>
<td>Myricetin</td>
<td></td>
<td>3.07</td>
<td>4.58</td>
<td>105.07</td>
<td>5.10 x 10¹²</td>
</tr>
<tr>
<td>Rhamnetin</td>
<td></td>
<td>1.27</td>
<td>0.07</td>
<td>864.29</td>
<td>1.70 x 10¹²</td>
</tr>
<tr>
<td>Isoquercitrin</td>
<td></td>
<td>2.72</td>
<td>5.52</td>
<td>77.88</td>
<td>2.90 x 10¹²</td>
</tr>
<tr>
<td>Naringenin</td>
<td>Flavanones</td>
<td>0.13</td>
<td>0.44</td>
<td>204.34</td>
<td>1.40 x 10¹²</td>
</tr>
<tr>
<td>Cyanidin chloride</td>
<td>Anthocyanidins</td>
<td>0.96</td>
<td>12.28</td>
<td>55.79</td>
<td>1.65 x 10¹²</td>
</tr>
</tbody>
</table>

Due to the structural differences of the polyphenolic compounds, the mechanism of action with regards to the antioxidant activity may vary quite markedly. However, it is generally
accepted that the phenolic hydroxyl moieties on these substrates are crucial when it comes to the antioxidant performance of these compounds due to its significant radical quenching potential. For example, the radical scavenging mechanism for caffeic acid has been described by Li et al., *Scheme 1.1.* In this particular case caffeic acid quenches the DPPH radical by donating a hydrogen atom from the para-hydroxyl moiety to form the stable DPPHH substrate. Further oxidation of the resonance stabilised phenoxy radical (A) ultimately leads to the formation of the ortho-quinone (B). When considering the antioxidant capacity or strength of a substrate to behave as an antioxidant it is the ease of abstraction of the first hydrogen atom of the hydroxyl moiety which is considered the most important factor coupled with the stability of the newly formed phenoxy radical.

![Scheme 1.1](image)

*Scheme 1.1.* The radical scavenging mechanism of caffeic acid towards the DPPH radical.

In addition, Brown et al., has investigated the mechanism of the antioxidant action of catechin towards Cu$^{2+}$, which is a significant contributor to the oxidation of low density protein (LDL) in the human body. He has suggested that catechin is capable of capturing free Cu$^{2+}$ ions through chelation, thus can aid to diminish copper mediated LDL protein oxidation.

Due to the enormous number of polyphenolic compounds that exist in nature coupled with their structural complexity and yet to be determined antioxidant properties, this thesis will focus on the synthesis of a range of new polyphenolic compounds derived...
from the extremely well known antioxidant compound resveratrol and related derivatives coupled with a comprehensive evaluation of their antioxidant capacities.

1.2 Resveratrol.

Resveratrol (1) is a naturally occurring polyphenolic that was first isolated in 1963 from the roots of the polygonaceous plant, *Polygonum cuspidatum*, with the root extract being used as an anti-inflammatory drug in Asian countries for hundreds of years, *Figure 1.3*.21

![Figure 1.3. Structure of trans-resveratrol (1).](image)

A number of years later trans-resveratrol (1) was found in grapes (*Vitis vinifera*) for the first time in 1976 with further studies detecting its abundant presence in red wine.22 Over the last few decades, it has been found that this natural product is key to the origins of many bioactivities related to human health, including anti-tumour, antioxidant and anti-platelet aggregation properties.23 In particular, evidence has been found that it is very effective in the prevention of cardiovascular diseases.24-26 This finding has attracted great attention from scientists as many consider that the high level of resveratrol (1) in red wine together with its pronounced bioactivity may provide the answer for the phenomenon of the so called “French paradox”.27 Given that resveratrol (1) has received wide attention as a beneficial compound to human health, its market demand as a nutritional supplement has risen rapidly over the last decade. Consequently, a variety of resveratrol supplements such as grape skin extracts have been developed by many pharmaceutical companies and are now sold throughout the world. This international recognition of the health benefits of consuming resveratrol, coupled with its natural abundance in nature has many believing that this compound will undoubtedly play a greater role in the improvement of human health in the near future.

1.2.1 Natural Sources of Resveratrol.
Resveratrol (1) has been found in abundance in a variety of dietary food such as grapes, peanuts, mulberries, and strawberries. In addition, it is also present in many other plants such as *Eucalyptus*, *Gnetum montanum* and *Picea sp. Spruce*. It is suggested that the formation of *trans*-resveratrol (1) results from externally induced environmental stress factors such as adverse weather conditions, excessive heat, insect, animal or pathogenic attack. Figure 1.4 highlights many of plant species in which resveratrol has been identified.

![Figure 1.4](image)

**Figure 1.4.** Natural sources of resveratrol.  

### 1.2.2 Isomerisation of *trans*-Resveratrol and *cis*-Resveratrol.

Resveratrol may exist in either of two geometrical forms designated as *trans*-resveratrol (1a) and *cis*-resveratrol (1b), depending on the structural configuration about the double bond, Figure 1.5. Although the *trans* isomer (1a) is believed to be the naturally occurring product, the latter (1b) can always be detected together with its *trans* isomer in plants.
such as grape. This is not surprising as isomerisation between the two isomeric forms can readily occur. It has been documented that trans-resveratrol (1a) can be converted into the cis form (1b) under UV light irradiation whilst the reverse transformation can be achieved under thermal conditions, Figure 1.4. Moreover, it should be noted that cis-resveratrol (1b) is considerably less stable compared to trans-resveratrol (1a) and it is not easy to be separated from plants in a pure state due to the rapid cis to trans isomerisation at ambient temperature. Consequently, biological studies focusing only on the cis form have rarely been reported in the literature with only a few examples reported on the anticancer activity and anti-aggregation properties of the cis form.

Figure 1.5. Isomerisation between trans-resveratrol (1a) and cis-resveratrol (1b).

The rate of photo-isomerisation of trans-resveratrol (1a) into the cis-form (1b) over time has been examined by Bernard et al., Figure 1.6. In this study, the trans to cis isomerisation under irradiation of a solution of trans-resveratrol was monitored using HPLC, $^1$H NMR and UV absorbance over time. The results analysed by the three techniques were very consistent and indicated that 80% conversion of the trans-resveratrol into the cis form was achieved after 40 minutes of UV irradiation at a 350 nm wavelength.
Figure 1.6. Isomerisation of trans-resveratrol (1a) and cis-resveratrol (1b) by UV light irradiation.35

1.2.3 Levels of trans-Resveratrol in Foods and Beverages.

The levels of resveratrol in dietary foods and beverages have been investigated in many studies. The following table depicts the content of resveratrol (both trans and cis) in common foods and beverages, Table 1.2. It has been found that bilberries (up to 16 ng/g) and blueberries (up to 32 ng/g) bear relatively higher resveratrol levels compared with that found in grapes and other dietary foods. Red wine is obviously the richest source of resveratrol (up to 14.3 mg/L) when compared to other soft or alcoholic beverages.38

Table 1.2. Total resveratrol levels (trans and cis) in common foods and beverages.

<table>
<thead>
<tr>
<th>Dietary food</th>
<th>Approximate Concentration</th>
<th>Beverages</th>
<th>Approximate Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bilberries</td>
<td>16 ng/g</td>
<td>Ports and sherries</td>
<td>0.1 mg/L</td>
</tr>
<tr>
<td>Blueberries</td>
<td>32 ng/g</td>
<td>Red grape juice</td>
<td>0.50 mg/L</td>
</tr>
<tr>
<td>Boiled peanuts</td>
<td>5.1 µg/g</td>
<td>Red wines</td>
<td>14.3 mg/L</td>
</tr>
<tr>
<td>Roasted peanuts</td>
<td>0.055 µg/g</td>
<td>White grape juice</td>
<td>0.05 mg/L</td>
</tr>
<tr>
<td>Grapes</td>
<td>3.54 µg/g</td>
<td>White wines</td>
<td>2.1 mg/L</td>
</tr>
<tr>
<td>Peanut butter</td>
<td>1.4 µg/g</td>
<td>Cranberry raw juice</td>
<td>0.2 mg/L</td>
</tr>
<tr>
<td>Peanuts</td>
<td>1.92 µg/g</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pistachios</td>
<td>1.67 µg/g</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
1.2.4 Levels of Resveratrol in Wine.

Since wine is a rich source of resveratrol and is consumed by many, the determination of resveratrol levels in wines has become of great significance given the perceived associated health benefits (vide infra). Red wines possess much higher levels of resveratrol (both trans and cis) over white wines mainly as a result of the different vinification techniques. Typically longer grape skin contact time is involved during red wine making processes, which allows for greater extraction of resveratrol into the wine. White wines are pressed off the skins immediately to aid in avoiding oxidation, which limits the possibility of extraction of resveratrol into the wines medium. In addition, many other factors can influence the levels of resveratrol such as UV light exposure on grapes, enzyme additions during wine making, grape variety and even fining agents which result in a diminished level of resveratrol in a finished wine. Furthermore, some of the resveratrol may be present in the form of glucosides (such as piceid, vide infra) which may be hydrolysed to release the free parent resveratrol during wine making processes by the action of hydrolytic enzymes or even by the acidity of the wine itself. Quantification of resveratrol levels in wines is usually achieved by measuring the total level of resveratrol and its glucosides. Table 1.3 depicts the individual and total levels of trans and cis-resveratrol (1a and 1b) and the mono-glycoside (piceid) in some Spanish red and white wines respectively as determined by HPLC analysis.

As expected the total resveratrol levels including trans/cis-resveratrol (1a and 1b) and trans/cis-piceid in red wines was found to be much higher than that in the white wines. The individual levels of resveratrol in a particular red wine or white wine are quite dependent on the grape variety, appellation and vintage age. For example, among the red wine varieties, Pinot Noir and Merlot contain significantly higher levels of resveratrol than the otherreds. Notably, both the red wines and the white wines contain significant levels of trans/cis-piceid indicating that resveratrol glucosides are also important constitutes when evaluating the total resveratrol level of a wine or other beverage.
Table 1.3. Concentration of resveratrol and piceid in some Spanish red and white wines.\textsuperscript{37,40}

<table>
<thead>
<tr>
<th>Red wines</th>
<th>Pinot Noir</th>
<th>Merlot</th>
<th>Merlot</th>
<th>Cabernet Sauvignon</th>
<th>Cabernet Sauvignon</th>
<th>Tempranillo</th>
<th>Grenache</th>
</tr>
</thead>
<tbody>
<tr>
<td>Appellation</td>
<td>Penedès</td>
<td>Costers del Segre</td>
<td>Penedès</td>
<td>Navarra</td>
<td>Penedès</td>
<td>Priorat</td>
<td></td>
</tr>
</tbody>
</table>

| trans-resveratrol (mg/L) | 8.00 | 4.64 | 7.74 | 0.95 | 1.32 | 1.87 | 2.83 |
| cis-resveratrol (mg/L)  | 2.13 | 0.67 | 2.48 | 0.20 | 0.38 | 0.35 | 0.41 |
| trans-piceid (mg/L)     | 0.97 | 4.01 | 2.36 | 0.96 | 1.00 | 1.26 | 2.66 |
| cis-piceid (mg/L)       | 0.57 | 0.68 | 1.20 | 0.40 | 0.52 | 0.54 | 0.82 |
| Total resveratrol and piceid (mg/L) | 11.67 | 10.00 | 13.78 | 2.51 | 3.22 | 4.02 | 6.72 |

<table>
<thead>
<tr>
<th>White wines</th>
<th>Albariño</th>
<th>Chardonnay</th>
<th>Macabeo</th>
<th>Parellada</th>
<th>Riesling</th>
<th>Verdejo</th>
<th>Xarel.lo</th>
</tr>
</thead>
<tbody>
<tr>
<td>Appellation</td>
<td>Rias Baixas</td>
<td>Penedès</td>
<td>Penedès</td>
<td>Penedès</td>
<td>Valladolid</td>
<td>Alella</td>
<td></td>
</tr>
</tbody>
</table>

| trans-resveratrol (mg/L) | 0.080 | 0.047 | 0.095 | 0.174 | 0.041 | 0.080 | 0.353 |
| cis-resveratrol (mg/L)  | 0.021 | 0.020 | 0.005 | 0.030 | 0.013 | 0.059 | 0.062 |
| trans-piceid (mg/L)     | 0.080 | 0.036 | 0.252 | 0.425 | 0.143 | 0.209 | 0.422 |
| cis-piceid (mg/L)       | 0.071 | 0.035 | 0.042 | 0.196 | 0.100 | 0.224 | 0.217 |
| Total resveratrol and piceid (mg/L) | 0.252 | 0.138 | 0.394 | 0.825 | 0.297 | 0.572 | 1.054 |
1.3 Perceived Health Benefits of trans-Resveratrol (1a).

Numerous biological studies over the decades have revealed that resveratrol (1) is effective in the prevention and treatment of a wide variety of diseases including diabetes, cancer, inflammation and neurodegenerative diseases.\(^{24,25,32}\) In addition to this, positive outcomes from animal studies reveal a positive life span improvement.\(^{32}\) Among these useful pharmaceutical studies, the most promising findings which have attracted the interest of scientists are that resveratrol aids in the prevention of cardiovascular diseases and various cancers and that the antioxidant properties of resveratrol are beneficial to human health. These attributes of resveratrol are detailed further in the forthcoming sections.

1.3.1 Antioxidant Capacity of trans-Resveratrol (1a): The French Paradox.

Free radicals and reactive oxygen species (ROS) are continually generated in the human body and are considered to be a threat to long term human health, as an accumulation of these reactive species in the body enhances oxidative stress and may cause damage to crucial biomolecules including nucleic acids, lipids and proteins, which subsequently lead to a wide variety of diseases such as premature ageing, inflammation, heart disease and cancer. It is well known that natural antioxidants in fruits and vegetables such as vitamin C, vitamin E, fibre and folic acid play a significant role in reducing oxidative stress and are thus able to reduce the incidence of related diseases caused by ROS.\(^{26,41}\) Recent \textit{in vitro} studies have shown that a variety of novel polyphenolic compounds found in plants may exert more effective antioxidant activity against ROS compared with the classical antioxidants such as vitamins E and C.\(^{42}\) These novel polyphenolic compounds include stilbenes, flavonoids and anthocyanins.\(^{43-48}\) One of the most significant findings for the compound trans-resveratrol (1a) was that it is able to significantly prevent the oxidation of low-density lipoprotein (LDL) during peroxidative degradation through chelating and free radical scavenging effects.\(^{26,49}\) This property is thought to be associated with the “French Paradox” due to the evidence that the oxidation of LDL leads to an increased risk of cardiovascular diseases.\(^{50,51}\) Other useful antioxidant properties of resveratrol include its ability to inhibit chemiluminescence and the generation of $O_2^-$ and the production of ROS and thiobarbituric acid-reactive substances in blood platelets.\(^{24}\) Furthermore, it aids
in the prevention of oxidative stress-induced cellular damage and protects against the progression of atherosclerosis.\textsuperscript{52}

It is believed that the substantial antioxidant capacity of resveratrol is due to its structure which includes the presence of a central carbon-carbon double bond which is conjugated to the vital \textit{para}-hydroxyl moiety of the phenolic grouping.\textsuperscript{53-55} The two functional moieties enable the molecule to quench harmful radicals in the body by either hydrogen atom transfer or electron transfer processes while the resulting resveratrol radical is stabilised by delocalisation and then self-condenses into harmless resveratrol dimers as depicted in Scheme 1.2.\textsuperscript{56} Consequently, a variety of \textit{in-vitro} antioxidant tests have been developed for a thorough investigation of the antioxidant capacity of this novel compound and several of its analogues. Typical assays include the ferric cyanide reducing antioxidant power assay (FRAP), the hydrogen peroxide scavenging activity, the superoxide anion radical scavenging activity, the DPPH radical scavenging assay and the ABTS radical cation decolonisation assay.\textsuperscript{53,57-60}

\textbf{Scheme 1.2.} Mechanism of how resveratrol quenches free radicals.\textsuperscript{56}
The “French paradox” can be briefly described as people who lived in the south of France and have a daily diet rich in saturated fats and consume red wine regularly suffer much less coronary heart disease compared with other countries where people share the same dietary sources but do not consume red wine. According to statistics released in 1992, people in France suffered the lowest mortality rates caused by cardiovascular diseases compared with other western world countries such as Australia, USA, and Italy, Figure 1.7. Japan was excluded here as their diet is vastly different from the other countries.

![Figure 1.7](image)

**Figure 1.7.** The mortality rates caused by cardiovascular diseases in some developed counties around the world. It is now generally accepted that red wine which is rich in polyphenolic compounds such as resveratrol may be responsible for this so called “French Paradox.” These polyphenols appear to possess significant health benefits such as anti-cancer, anti-carcinogenic, anti-bacterial, anti-viral and anti-inflammatory activities and many other pharmaceutical benefits. Scientists have been debating whether resveratrol performs the key role towards the phenomenon of the “French paradox” over the years, however, it is clear that consumption of red wines which do contain high levels of resveratrol and its derivatives while white wines contain much less resveratrol is an important factor towards...
the observed health benefits.\textsuperscript{27,37,65} However, the concept of the “French Paradox” is still inconclusive as the mechanism involved is not fully understood and more \textit{in vitro} and \textit{in vivo} studies need to be carried out along with a thorough investigation of the anti-cardiovascular properties of all the other possible polyphenolic compounds that may be found in wines to ascertain which of these are the key antioxidants.

1.3.2 Resveratrol and Cancer.

Cancer is recognised as one of the most destructive human diseases and it was reported to be responsible for 12\% of all deaths around the world in 2000. Moreover, scientists anticipate that the total cases of cancer will further increase by 50\% by 2020 in both developed and developing countries.\textsuperscript{66} An important finding related to resveratrol is that this compound has chemopreventive and chemotherapeutic effects on a wide variety of cancer types, such as colon, pancreas, stomach, prostate, ovary, liver, lung and cervical cancers according to many \textit{in vitro} and \textit{in vivo} studies.\textsuperscript{32} It has been suggested that the chemopreventive activity of resveratrol is performed by blockage of carcinogen activation and increased detoxification via inhibition of phase I and induction of phase II enzymes, modulation of cell cycle and inducment of apoptosis and, anti-angiogenic effects and suppression of invasion and metastasis.\textsuperscript{32} For example, an \textit{in vitro} study reported by Delmas et al., has shown that the proliferation of rat hepatoma and human hepatoblastoma HepG2 cells can be significantly inhibited by adding resveratrol to the culture medium.\textsuperscript{67} It has also been shown by Atten et al., that resveratrol is capable of inhibiting the proliferation of nitrosamine-stimulated human gastric adenocarcinoma KATO-III and RF-1 cells.\textsuperscript{68} Similarly, evidence has been found that resveratrol is effective in the inhibition of prostate carcinogenesis by diverse cellular mechanisms related to tumor initiation, promotion and progression.\textsuperscript{32} Apart from the pronounced anticancer property of resveratrol itself, some studies have shown that a combination of resveratrol and other chemotherapeutic agents help to potentiate the effects in the treatment of cancer.\textsuperscript{54} This provides an opportunity for lower doses of chemotherapeutic agents and thus aids to reduce their negative side effects. However, it should be noted that the clinical data with regards to the benefits of this novel compound is still quite limited as only a few trials have been performed on humans thus far. In addition to this, the negative effects of resveratrol towards human health is still not quite clear due to the fact that limited research has been carried out on such possible side effects.
1.4 Quantification and Detection of Resveratrol by HPLC and LC-MS.

The quantification and detection of resveratrol in foods and beverages can be readily achieved by the use of modern analytical instruments such as HPLC and LC-MS. HPLC analysis using a reverse phase column coupled with a UV-Vis detector or fluorescence detector is the most common method for resveratrol analysis. For example, *trans*-resveratrol (1a) in grape extracts may be detected by rapid high-performance liquid chromatography coupled with a fluorescence detector. As depicted in Figure 1.8, a strong signal of resveratrol separated from other peaks can be observed at the specific emission wavelength of 403 nm. However, it should be noted that due to the low sensitivity and poor selectivity of these detectors, this method is not suitable for the analysis of samples bearing a complex mixture of constituents.

![Fluorescence excitation and emission spectra of trans-resveratrol (1a) (left) and detection of trans-resveratrol in grapes extracts at the emission wavelength of 403 nm (right).](image)

*Figure 1.8.* Fluorescence excitation and emission spectra of *trans*-resveratrol (1a) (left) and detection of *trans*-resveratrol in grapes extracts at the emission wavelength of 403 nm (right).69

Therefore, the use of a more specific and sensitive MS detector has become popular in recent years. For example, the quantitative analysis of *trans*- and *cis*-resveratrol (1a, b) in wine, grape, and cranberry products by HPLC coupled with a DAD and MS detector was developed in 2002.70 This study highlighted that the use of a MS detector provides excellent sensitivity for the measurement of resveratrol levels in food extracts with a detection limit of 0.31 pmol. The MS of resveratrol displays a parent ion at m/z 228.9 (M+1) together with the common identification fragments (m/z 182.9, 134.9, 118.9 and 106.9) as depicted in Figure 1.9. In addition to these HPLC methods, the use of GC-MS techniques has also been reported previously.71
Chapter 1 Introduction

1.5 Biosynthesis of Resveratrol in Plants.

The biochemical synthesis of resveratrol in plants has been elucidated and occurs via a series of enzymatically processes as highlighted in Scheme 1.3.²⁴ The synthesis begins with the amino acid phenylalanine (2) which is transformed into cinnamic acid (3) by deamination which is catalysed by the enzyme phenylalanine ammonia lyase. Enzymatic hydroxylation to p-coumaric acid (4) followed by conversion of the free acid into p-coumaroyl CoA (6) occurs with the aid of CoA ligase (5). The final step in the synthesis involves the condensation of the p-coumaroyl CoA (6) with malonyl CoA (7) in the presence of stilbene synthase to furnish trans-resveratrol (1a).

Scheme 1.3. Enzymatic synthesis of resveratrol in plants.²⁴
1.6 Previous Chemical Syntheses of *trans*-Resveratrol (1a).

Due to the high demand of *trans*-resveratrol in the nutraceutical supplement market, scientists have developed a number of short synthetic strategies to produce this compound. Usually, these synthetic strategies require the construction of the stilbene skeleton by the use of coupling reactions such as Wittig couplings, Suzuki cross-coupling, Heck reactions or Perkins condensations. For example, Guiso et al., has reported a five step synthesis employing the use of a Heck reaction, *Scheme 1.4*. The total synthesis began with the silylation of the simple phenolic aldehyde (8) to afford the corresponding OTBS protected aldehyde (9) in the presence of *N,N*-diisopropyl-ethylamine and DMF. Aldehyde (9) was then transformed into the styrene (10) by treatment with ylide followed by acetylation to furnish the acetate (11). This acetate was then coupled with 4-iodophenyl acetate (23) in the presence of palladium acetate to afford resveratrol acetate (13), which was deacetylated to afford *trans*-resveratrol (1a) in excellent overall yield (70%).

![Chemical reaction diagram](image)

*Scheme 1.4. Synthesis of resveratrol via Heck reaction.*

Andrus et al., also reported that the synthesis of *trans*-resveratrol (1a) can be achieved in four steps by the use of a Heck coupling reaction, *Scheme 1.5*. The starting acid (14)
was transformed to the acyl chloride (15) and then condensed with the styrene (16) in the presence of the amino salt (17) and palladium acetate to afford the triacetate (13). Hydrolysis under basic conditions then afforded trans-resveratrol (1a) in a good yield of 73%.

Scheme 1.5. Synthesis of resveratrol via Heck coupling.

In addition, Chen et al., has reported a one-step synthesis of resveratrol by hydrolysis of the known glycoside of resveratrol known as picead (18) with the aid of beta-glycosidase. Scheme 1.6. The transformation was successfully achieved at 60°C after 4 hours in 56% yield.

Scheme 1.6. Synthesis of resveratrol by enzymatic hydrolysis of picead (18).
1.7 Other Known Resveratrol Derivatives in Nature and Their Bioactivities.

In the past few decades, the number of resveratrol derivatives that are closely related structurally to the parent compound and which have been isolated from plants has increased rapidly. It has been found that many of these compounds are simply substituted resveratrol derivatives. As summarised in Figure 1.10, they can be sub-classified into several groups including resveratrol, that has been methoxylated, glycosylated, hydroxylated or those that contain additional carbon substituents.

![Diagram of resveratrol derivatives]

**Figure 1.10.** Other important resveratrol derivatives isolated from plants.
It is believed that many of these substituted resveratrol derivatives exhibit promising bioactivities such as chemo-preventive and anti-oxidant activities like the parent compound (1a). For example, a previous study has reported six methylated resveratrol derivatives, namely pterostilbene (19), desoxyrhapontigenin (24), pinostilbene (25), 3-hydroxyl-5,4’-dimethoxystilbene (26), resveratrol-trimethyl-ether (27), and 3,4,5,4’-tetrahmethoxy-trans-stilbene (28). Among these analogues, pterostilbene (19), which is also present in grapes and in heartwood extracts of *Pterocarpus marsupium*, shows a similar antioxidant capacity and anticarcinogenic activity to resveratrol itself according to animal studies. Again the para-hydroxyl moiety has been left unsubstituted and is considered the key reactive point in terms of antioxidant capacity. Interestingly, the tetra-methoxylated resveratrol derivative (28), exhibits even better bioactivity against cancer cells than resveratrol itself.

![Methylated Resveratrol Derivatives](image.png)

*Figure 1.11.* Some methylated resveratrol derivatives identified from plants. In addition, scientists have recently identified in plants many polyhydroxylated resveratrol derivatives which may display important chemopreventive and therapeutic bioactivities towards certain diseases, *Figure 1.12.* It has been reported that these compounds exhibit high antioxidant capacities and possess significant pharmaceutical potential. For example, a promising finding is that 3,5,3’,4’,5’-pentahydroxy-trans-stilbene (30) can efficiently inhibit human colon cancer cell proliferation via oxidative...
stress mediated apoptotic cell death and it may be able to be used as an potential colorectal cancer chemotherapeutic agent.\textsuperscript{77,78}

\textbf{Figure 1.12}. Some important polyhydroxylated stilbenes found in plants.\textsuperscript{32,77,78}

There are also a large number of resveratrol derivatives of higher structural complexity compared to the simple substituted resveratrol analogues described above. The most common compounds found in nature are a variety of resveratrol dimers, trimers and tetramers, some of which are depicted in \textbf{Figure 1.13}.\textsuperscript{79}

\textbf{Figure 1.13}. Some important resveratrol oligomers found in plants.\textsuperscript{79-82}
Many biological studies have revealed that these resveratrol oligomers are also of potential pharmacological usage. For example, the resveratrol dimer trans-ε-viniferin (33) extracted from *Vitis vinifera* displays high levels of antioxidant activity towards superoxide radicals in a DMSO medium. Another dimer named pallidol (35), which has been isolated from red wine exhibits a strong singlet oxygen quenching capability.

Moreover, the cyclic trimeric resveratrol derivative named α-viniferin (36), isolated from *Caragana chamlagu* is used as a traditional medicine in Korea and possesses anti-inflammatory potential in adjuvant-induced arthritis or carrageenin-induced paw edema in animal studies. Finally, vaticanol C (37), was found to have a potent suppressive activity of cell growth in colon cancer cell lines in a recent study.

1.8 Examples of Syntheses of Other Important Related Resveratrol Derivatives.

The development of extraction techniques for the isolation of natural products over the decades, many resveratrol analogues have been isolated from plant extracts to meet the demand in pharmaceutical research. However, due to the fact that these compounds are usually present in plants in limited quantities, extraction of large quantities is often associated with high cost and low efficiency. Thus, a variety of synthetic strategies for the preparation of related resveratrol analogues have been developed in recent times. A typical example previously reported is the total synthesis of Combretastatin A-4 (45), which has gained rapid recognition in the health arena due to its promising anti-cancer activity. The soluble form (Combretastatin A-4 phosphate) has been under clinic trials for treatment of acute myelogenous leukemia and relapsed ovarian cancer. The total synthesis of Combretastatin A-4 by Gaukroger in 2001 is depicted in Scheme 1. The precursor, 3-hydroxy-4-methoxybenzaldehyde (39) was silylated and then transformed into the dibromostyrene (40) using the Corey-Fuchs Wittig-type bromination. Desilylation of the dibromide (38) with tetrabutylammonium fluoride followed by a stereoselective reduction of (41) using tributyltin hydride and tetrakis (triphenylphosphine)palladium(0) afforded the (Z)-vinyl bromide (42). Treatment of (42) with the boronic acid (43), in the presence of tetrakis (triphenylphosphine)palladium(0) and sodium carbonate in DME, furnished the Combretastatin A-4 (45) in good yield.
In 2009, Moran et al., has synthesised two bioactive compounds (46) and (47) which have the same carbon core as resveratrol itself, however, differ in that the usual hydroxyl substituents had been replaced by alternative substituents, Scheme 1.8. The synthesis of (46) involved a Heck coupling whilst preparation of (47) utilised a Wittig reaction. The anti-proliferative properties of these two compounds were then evaluated in several cancer lines with the results suggesting that the two compounds are more effective anti cancer agents compared to resveratrol itself.
Scheme 1.8. Synthesis of related resveratrol analogues (46) and (47).

In 2008, Huang et al., synthesised a range of modified resveratrol analogues based on the parent compound (1a) and examined their cytotoxic activity against epidermoid tumor Cell Line KB and their xanthine oxidase inhibitory activity, Scheme 1.9.

Scheme 1.9. Synthesis of a range of resveratrol analogues.
Treatment of resveratrol (1a) with POCl₃ in DMF and MeCN, afforded the two formylated products (48) and (49) in 72% and 9% yields, respectively. Methylation of (48) with MeI and K₂CO₃ in DMF gave three individual methoxylated derivatives (50a-c). By replacing MeI with two other reagents BrCH₂CO₂Et or Br(CH₂)₃Br under the same conditions, afforded products (51a-c) and (52a,b), respectively. Base hydrolysis of the esters (51a-c) gave the corresponding carboxylic acids (53a-c). The biological assays showed that formylation of resveratrol (1a) remarkably improves the antitumor activity whilst alkylation generally lead to a decrease in antitumor activity. Moreover, derivative (48) showed the strongest xanthine oxidase inhibitory activity when compared to the others although only limited derivatives were screened, Table 1.4.

**Table 1.4.** Cytotoxic activity against human epidermoid tumor Cell Line KB and xanthine oxidase inhibitory activity of the synthesised resveratrol analogues.

<table>
<thead>
<tr>
<th>Compound</th>
<th>Cytotoxic Activity against Epidermoid Tumor Cell Line KB, IC₅₀ [μM]</th>
<th>Xanthine Oxidase Inhibitory Activity, IC₅₀ [μM]</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>&gt; 30</td>
<td>13.9 ± 0.25</td>
</tr>
<tr>
<td>48</td>
<td>4.9 ± 0.15</td>
<td>2.5 ± 0.12</td>
</tr>
<tr>
<td>49</td>
<td>2.8 ± 0.11</td>
<td>---</td>
</tr>
<tr>
<td>50a</td>
<td>6.0 ± 0.18</td>
<td>26.6 ± 0.55</td>
</tr>
<tr>
<td>50b</td>
<td>12.9 ± 0.26</td>
<td>16.9 ± 0.33</td>
</tr>
<tr>
<td>50c</td>
<td>9.3 ± 0.14</td>
<td>---</td>
</tr>
<tr>
<td>51a</td>
<td>7.4 ± 0.21</td>
<td>---</td>
</tr>
<tr>
<td>51b</td>
<td>7.1 ± 0.15</td>
<td>---</td>
</tr>
<tr>
<td>51c</td>
<td>7.3 ± 0.23</td>
<td>---</td>
</tr>
<tr>
<td>52a</td>
<td>4.1 ± 0.12</td>
<td>---</td>
</tr>
<tr>
<td>52b</td>
<td>8.9 ± 0.33</td>
<td>---</td>
</tr>
<tr>
<td>53a</td>
<td>9.5 ± 0.24</td>
<td>---</td>
</tr>
<tr>
<td>53b</td>
<td>11.6 ± 0.38</td>
<td>---</td>
</tr>
<tr>
<td>53c</td>
<td>&gt; 30</td>
<td>---</td>
</tr>
</tbody>
</table>

1.9 Resveratrol Glucosides.

Resveratrol glycosides have been found in a variety of plant families. It has been reported that these analogues are formed via enzymatic induced glycosylation of resveratrol and...
which is believed to aid to protect the parent compound from enzymatic oxidation. Thus, glycosylation is able to extend the half-life of cis and trans-resveratrol (1a,b) in the plant cells and therefore helps maintain its biological properties. In addition, other roles of resveratrol glycosides in plants such as improved solubility have also been suggested. In plants, the most common resveratrol glycoside found is piceid (18). Figure 1.14. This mono-glycosidated resveratrol is abundant in grapes and also present in many other plants such as Rheum undulatum and polygonum cuspidatum (PC) which are utilise for traditional medicine in Asian countries. Other reported glycosylated resveratrol derivatives include the two di-glucosidated resveratrols (54 and 55), which have been identified from the roots of Carex distachya Desf. (Cyperaceae) and Morus alba cell cultures, respectively. A recent study has shown resveratroloside (56) and the resveratrol tri-glycoside (57) are present in grape cell cultures.

Figure 1.14. Resveratrol glycosides identified in plants.

1.9.1 Bioactivity of Resveratrol Glycosides.

Compared with the in-depth biological and clinic studies on trans-resveratrol (1a), limited work has been carried out on the resveratrol glycosides. This is partly due to the fact that these compounds are usually present in plants in very low levels. Hence, it is difficult to extract these compounds from plants and carry out a full-scale investigations of their potential pharmaceutical properties and usage. However, the pharmaceutical potential of these analogues for disease prevention and treatment has been reported in recent years.
Several *in-vitro* biological studies have shown that some resveratrol glycosides bear similar pharmaceutical bioactivities to that of resveratrol itself; such as excellent antioxidant activity and anti-platelet aggregation.\textsuperscript{101} For example, it has been reported that picead (18) has the potential to prevent LDL-oxidation and reduce cardiovascular diseases.\textsuperscript{102} Resveratroloside (56), another mono-glycosylated resveratrol, exhibits antioxidant and prolyl endopetidase inhibitory activities.\textsuperscript{103} It should be noted that the consumption of resveratrol glycosides by humans may in fact enhance the overall health benefit of free resveratrol as these analogues may be hydrolysed back to the parent compound in the stomach or intestine and thus lead to an increase in the level of free resveratrol in the tissues/organs within the body.\textsuperscript{104} Therefore, the *in-vivo* pharmaceutical performance of these compounds is expected to be more complex than that reported in the *in-vitro* studies.

### 1.9.2 Resveratrol Glycosides in Wine.

Previous wine analyses have shown that picead (18) is the most common resveratrol glycoside found in wine. Both the *trans* and *cis* isomers have been detected in a variety of wines including red, rosé and white wines. Several studies have found that in some wines, picead (18) is more concentrated than the parent resveratrol itself. For example, the levels of picead (18) and resveratrol (1) in 120 commercial Portuguese and French wines were measured.\textsuperscript{105} The results showed that the levels of picead (18) (up to 50.8 mg/L for *trans*-picead (18a) and up to 17.9 mg/L for *cis*-picead (18b) were significantly higher than that of resveratrol (1) (up to 6.8 mg/L for *trans*-resveratrol (1a) and up to 9.5 mg/L for *cis*-resveratrol (1b)) in these red wines. Compared with red wine, rosé wine bears relatively lower level of picead (18) but higher than in white wine. For example, the measurement of both *trans* and *cis*-picead in Spanish white and rosé wines in 1996 showed that up to 3.52 mg/L of picead (18) was detected in the rosé wine in comparison with 1.80 mg/L in the white wine.\textsuperscript{37} Apart from picead (18), other resveratrol glycosides still remain unknown in wines as little effort has been made with regards to the identification of these compounds in previous wine analyses. However, it should be noted that some other analogues such as resveratroloside (56) and tri-glycosided resveratrol (57) are very likely to be present, as they have been detected in grape cell cultures.
1.9.3 Previous Syntheses of Resveratrol Glycosides.

A number of ways to prepare resveratrol glycosides have been reported in the literature. These strategies usually require the use of glucosylation reagents such as \( \alpha \)-bromotetra-\( O \)-acetyl-\( D \)-glucose (58) or tri-fluoroacetimidate to achieve the glycosylation step.\textsuperscript{101,103} For example, Scheme 1.10 depicts a one-step synthesis of the mono-glucoside piceid (18) via a direct coupling reaction of \textit{trans}-resveratrol (1a) with \( \alpha \)-bromotetra-\( O \)-acetyl-\( D \)-glucose (58) in the present of methanolic sodium solution. By carefully controlling the amount of \( \alpha \)-bromotetra-\( O \)-acetyl-\( D \)-glucose (58) added, the reaction affords the desired mono-glucoside (18) in 25% yield.\textsuperscript{101}

\[
\text{Scheme 1.10. One step synthesis of picead (18) from \textit{trans}-resveratrol (1a).}^{101}
\]

It has been reported that resveratrol glycosides can be also synthesised via glucosylation of simple aromatics followed by a coupling reaction between the glucosylated intermediate with another arene to build up the skeleton of resveratrol using Heck type coupling reactions or Wittig condensations.\textsuperscript{106-109} For example, Scheme 1.11 depicts the total synthesis of piceid (18) and resveratroloside (56). The synthesis of piceid (18) began with the glucosylation of the idophenol (59) with \( \alpha \)-bromotetra-\( O \)-acetyl-\( D \)-glucose (58) to furnish acetate (60) in low yield. The acetate (60) was then coupled with the styrene (16) in the presence of Pd(\( \text{OAc} \)\textsubscript{2}) catalyst to afford the desired protected resveratrol (61). Deprotection of the acetyl moieties on (61) furnished the targeted piceid (18) in high yield for this last step. A similar strategy was also applied on the synthesis of resveratroloside (56) as shown in \textit{Scheme 1.11}.
Scheme 1.11. The total synthesis of picead (18) and resveratroloside (56).¹⁰⁶
In 2006, Zhang et al., reported a convenient method to synthesise the resveratrol mono-
glucosides (18 and 56) and di-glucosides (73 and 54) by regioselective protection of 
trans-resveratrol (1a) with OTBS groupings followed by glucosylation of these silylated 
resveratrols with 2,3,4,6-tetra-O-benzoyl-D-glucopyranosyl trifluoroacetimidate.\textsuperscript{103} As 
depicted in Scheme 1.12 silylation of resveratrol afforded four OTBS protected 
resveratrol derivatives (65-68). After separation by silica column chromatography, these 
compounds were coupled with the trifluoroacetimidate to afford the four glycosylated 
resveratrol derivatives (69-72) in satisfactory yields. Deprotection of these derivatives 
with NaOMe afforded the desired glycosides (18, 54, 56 and 73) in excellent yields.

\textbf{Scheme 1.12.} The synthesis of resveratrol glycosides (18, 54, 56 and 73).\textsuperscript{103}

In addition to these chemical approaches to resveratrol glycosides, enzymatic 
transformation of trans-resveratrol (1a) to piceid (18) has also been developed recently, 
\textit{Scheme 1.13}.\textsuperscript{22} It should be noted that these methods have been shown to be more
environmentally friendly and less costly compared to conventional chemical strategies. For example, treatment of resveratrol with whole-cell suspensions of *Streptococcus mutans* under very mild conditions afforded piceid (18) in a low 18% yield.

![Scheme 1.13. Enzymatic transformation of trans-resveratrol (1a) to piceid (18).](image)

1.10 Thesis Aims.

As highlighted above previous literature has shown wine is a rich source of many important resveratrol derivatives, including several resveratrol glycosides, hydroxylated resveratrols, methylated resveratrols and also a range of resveratrol oligomers. However, due to the complex matrix of wine, only a few have been thus far identified while many are still unknown in wine but have been found in other plant material. Furthermore, many of these resveratrol derivatives have been shown to exhibit useful biological and pharmaceutical properties. Thus, a thorough investigation of whether many of these other resveratrol derivatives are found in wine and their bioactivities is important for a better understanding of the potential health benefits of wine. Therefore the aim of this research study is to synthesise a range of both known and unknown resveratrol derivatives and to examine the antioxidant capacity of these analogues. The targeted compounds are to include all ten *cis* and *trans*-resveratrol glycosides, a range of resveratrol dimers such as the viniferins and pallidol, and also to include all eleven possible glucosides of *trans*-piceatannol, Figure 1.15. In short, the synthesis of these stilbene glycosides was to be achieved by glycosylation of the stilbene aglycones whereas the synthesis of resveratrol dimers was to be accomplished by the dimerisation of the appropriate resveratrol analogues.
Figure 1.15. Proposed library of resveratrol, piceatannol glucosides and associated dimers for determination of their antioxidant capacities.

In addition, the antioxidant capacity of the targeted resveratrol and piceatannol glycosides will be measured individually utilising *in-vitro* FRAP and DPPH assays in order to gain an understanding of the effect that glycosylation has on the antioxidant performance of the parent compounds. Such investigations will also allow us to further fine tune our current understanding of the mechanistic basis behind the antioxidant capacities of this group of important natural stilbenes and highlight further why consumption of these derivatives in the form of foodstuffs and beverages may have beneficial effects on human health.
CHAPTER 2: SYNTHESIS OF TRANS/CIS-RESVERATROL GLUCOSIDES AND THEIR ANTIOXIDANT CAPACITIES.

As described within Chapter 1 there were 10 mono-, di- and tri-glucosides of cis and trans-resveratrol to be prepared. This Chapter details the successful synthesis of all of these proposed analogues along with their full chemical characterisation. Furthermore, the antioxidant capacities of these derivatives were then determined utilising both the FRAP and DPPH assays in order to gain an understanding of the effect that glycosylation has on the antioxidant performance of the parent compounds. A mechanistic picture of the origins of this antioxidant capacity is then discussed. Whilst the proposed antioxidant tests only require small quantities of pure chemical material for evaluation it was important to consider that if any of the target compounds displayed excellent antioxidant activity then the chemical processes for their synthesis should also be able to be scaled up to ensure ample quantities for additional future biological evaluations. Consequently, with this in mind, all chemical transformations carried out here for the proposed synthesis of the targets were almost exclusively performed within the hundreds of milligrams to multi-gram scale.

2.1 Synthetic Strategy for Rapid Access to the Resveratrol Glucosides.

The synthesis of all the trans-resveratrol glucosides was foreseen to be achieved in only four steps, and was to begin with the non exhaustive silylation of trans-resveratrol (1a) utilising TBDMSCI in a similar synthetic strategy to that recently developed by Zhang.\textsuperscript{103} Whilst we knew this strategy would lead to a mixture of all possible mono-, di- and tri-trans-(1a)-OTBDMS derivatives we were confident that separation of the individual derivatives would be possible and thus allow for rapid access to all necessary analogues, Scheme 2.1. Unlike the method of Zhang who employed the 2,3,4,6-tetra-O-benzoyl-d-glucopyranosyl trifluoroacetimidate as the glycosylating reagent, Scheme 1.12,\textsuperscript{103} we decided to utilise the orthoester (74) reported by Kunz,\textsuperscript{110} which has previously been successfully employed for the glycosylation of numerous alcohols and phenolics under mild reaction conditions.\textsuperscript{111} The final steps in the synthesis of the desired trans-resveratrol glucosides would involve desilylation followed by depivaloylation, Scheme 2.1. Moreover, with the pure individual trans-resveratrol glucosides in hand, it was foreseen that each
could be individually isomerised into its cis analogue through exposure to UV light, thus completing the first synthesis of all mono-, di- and tri-cis-resveratrol glucosides (1b).

**Scheme 2.1.** Synthetic strategy for rapid access to the resveratrol glucosides exploiting the Kunz reagent (74).
Chapter 2 cis/trans-Resveratrol Glucosides and Their Antioxidant Capacities

2.2 Preparation of Kunz Reagent (74).

Before the preparation of the requisite glucosides of resveratrol could begin it was first necessary to synthesise the Kunz reagent (74) from D-glucose (75), Scheme 2.2. Thus treatment of D-glucose (75) in the presence of pivaloyl chloride, triethylamine and DMAP in dichloromethane afforded the penta-pivaloylated glucose (76) in 84% yield as a white solid.\textsuperscript{110} Treating this latter compound (76) with HBr (33% in acetic acid) in dichloromethane afforded 2,3,4,6-tetra-O-pivaloyl-\(\alpha\)-D-glucopyranosyl bromide (77) in a very good 89\% yield again as a white solid. This bromide (77) was then treated with S-collidine, acetophenone oxime and silver triflate in dichloromethane to furnish the Kunz reagent (74) in 76\% yield. The \(^1\)H NMR data and the melting points of compounds (76), (77) and (74) were all in accordance with those previously reported in the literature.\textsuperscript{110,112-114}

\[\text{Scheme 2.2. Synthesis of the Kunz reagent (74).}\]
2.3 Non-Exhaustive Silylation of trans-Resveratrol (1a) with TBDMSCl.

Non exhaustive silylation of trans-resveratrol (1a) with TBDMSCl (1.8 equivalents) afforded a mixture of five requisite silyl ethers (65-68, 78) which were easily separable by column chromatography as had been previously found by Zhang et al.\textsuperscript{103} This one pot procedure afforded the requisite mono- and di-OTBDMS protected analogues (65) (25.3 %), (66) (10.7 %), (67) (18.4 %), (68) (11.3 %) and (78) (17.3%) and could be conducted on multi gram scale. The \textsuperscript{1}H NMR spectra of these silylated compounds were all in agreement with those reported by Zhang et al.\textsuperscript{103} The key feature within the \textsuperscript{1}H NMR spectra of these compounds was the presence of the trans C=C bond clearly identifiable by the typical coupling constant of $J = 16.2$ Hz between the two protons signals at ca. 6.95 and 6.82 ppm $\delta$ for all derivatives (65-68, 78).

\begin{equation}
\begin{array}{c}
\text{TBDMSO} \\
\text{OH} \\
\text{(65) 25.3%}
\end{array}
\end{equation}

\begin{equation}
\begin{array}{c}
\text{TBDMSO} \\
\text{OH} \\
\text{(66) 10.7%}
\end{array}
\end{equation}

\begin{equation}
\begin{array}{c}
\text{TBDMSO} \\
\text{OH} \\
\text{(67) 18.4%}
\end{array}
\end{equation}

\begin{equation}
\begin{array}{c}
\text{TBDMSO} \\
\text{OH} \\
\text{(68) 11.3%}
\end{array}
\end{equation}

\begin{equation}
\begin{array}{c}
\text{TBDMSO} \\
\text{OH} \\
\text{(78) 17.3%}
\end{array}
\end{equation}

\textbf{Scheme 2.3.} Non-exhaustive silylation of trans-resveratrol (1a) with TBDMSCl.
2.4 Synthesis of trans-Resveratrol Glycosides Utilising the Kunz Reagent (74).

With the four mono- and di-OTBDMS protected trans-resveratrol precursors (65-68) in hand, we next attached the protected glucoside units to the free hydroxyls utilising the Kunz reagent in the presence of boron trifluoride diethyl etherate to furnish the mono- and diglucoside derivatives (79-82) in very good yields (75-85%), Scheme 2.4.

Scheme 2.4. Synthesis of the protected resveratrol glycosides (83-86) utilising the Kunz reagent.
The key features within the $^1$H NMR spectra of these compounds was the presence of the trans C=C bond clearly identifiable by the typical coupling constant of $J = 15.6$ or $16.2$ Hz between the two protons at ca. 6.94 and 6.80 ppm, along with the presence of the anomeric proton resonance at ca. 5.1 ppm for the glucose unit(s) for all derivatives (79-89). Furthermore, the anomeric $^{13}$C signal appeared at ca. 99 ppm indicating the attachment of the protected glucose unit(s). Desilylation of these glycosides (79-89) with TBAF furnished the required protected glycosides (83-86) in excellent yields (82-90%). The fact that these substrates contained either one or two protected glucose units were clearly evident from HRMS analysis with the individual molecular ions [M+H]$^+$ determined to be (727.3688, 727.3688, 1225.6517, 1225.6517) for compounds (83-86) respectively.

Scheme 2.5. Basic hydrolysis of (83-86) to furnish the four mono- and di-glucosidated
trans-resveratrol derivatives (18, 54, 56 and 73).

Global deprotection of all pivaloyl groupings within (83-86) utilising NaOMe at room temperature afforded the desired four mono- and di-OTBDMS protected trans-resveratrol precursors (18, 54, 56 and 73) in excellent yields, Scheme 2.5. The $^1$H and $^{13}$C NMR data of these four mono- and di-glucosidated trans-resveratrol derivatives (18, 54, 56 and 73) were all in accordance with those reported previously although subtle differences were noted due to the fact that many of these other groups measured the NMR spectra in pyridine-$d_5$ + 5% D$_2$O, $d_6$-DMSO, $d_6$-acetone or MeOD whilst we measured ours in $d_4$-MeOH.\textsuperscript{39, 100, 103, 106, 108, 115, 116} The standout key features in the $^1$H NMR spectra of these derivatives was the clear presence of the trans C=C at approximately 7.0 and 6.8 ppm displaying a coupling constant around 16 Hz and the presence of a doublet at ca. $\delta$ 4.9 ppm ($J = 7$ Hz) for the proton on the anomeric carbon atom of the sugar moiety(ies) whilst the $^{13}$C NMR resonance indicated the presence of the either one or two anomeric carbon atoms at approximately 102 ppm for the sugar unit(s).

In order to gain access to the tri-glucoside of trans-resveratrol (1a), the bis-protected glucoside (86) was first treated with the Kunz reagent to furnish (87) in a good yield of 70% followed by deprotection with NaOMe to afford (88) in an excellent yield of 94% as depicted in Scheme 2.6.

\begin{align*}
\text{(86)} & \xrightarrow{\text{a}} \text{(87) 70\%} \\
\text{(88) 94\%}
\end{align*}

(a) Kunz reagent, BF$_3$OEt$_2$, CH$_2$Cl$_2$, 0°C; 
(b) NaOMe (20 equiv.), MeOH, rt.
Scheme 2.6. Synthesis of the tri-glycosided trans-resveratrol (87).

During the writing of this thesis Pandey et al. reported the enzynatic biosynthesis of the tri-glucoside (88). Unfortunately they did not quote any physical or spectroscopic data (\(^1\)H or \(^{13}\)C NMR) except to provide the mass spectrum in ESI positive mode. They reported a HRMS + Na\(^+\) of 737.2266 which correlated well with our low resolution ESI \(m/z\) 736.9 (\(M^+ + Na^+\)). Inspection of the \(^1\)H NMR revealed the presence of the expected aromatic signals along with the clear presence of two distinctly different glucose units. Once again the protons resonance about the central C=C bond appeared at 6.96 and 7.09 ppm and displayed a coupling of 16.8 Hz indicating the presence of a trans double bond.

Figure 2.1. \(^{13}\)C NMR spectrum of the trans-tri-glycoside (88).

Confirmation of the structure of the tri-glucoside (88) was much clearer from inspection of the \(^{13}\)C NMR, Figure 2.1. A total of ten signals were clearly visible between 105.7 and 160.6 ppm confirming the presence of the backbone of the resveratrol unit. The two ipso OGlu about the aromatic rings appeared downfield at 160.6 and 159.3 ppm whilst the
signals for the $\text{C}=$C appeared at 130.5 and 128.3 ppm. As expected, tri-glucoside (88) incorporates two different glucose units and due to symmetry all $^{13}\text{C}$ signals for the sugar units should appear as a close set of two signals. This was indeed true for all carbon signals except one (75.4 ppm), which appeared as a singlet. Finally, two anomeric carbon signals at 102.7 and 102.6 ppm confirmed the presence of the two different glucose units and supports the expected $\beta$-D-stereochemistry, Figure 2.1.

2.4.1 HPLC and UV-Vis Analysis of the trans-Resveratrol Analogues.

Samples of all trans-resveratrol analogues (1a, 18, 54, 56, 73 and 88) were analysed individually by HPLC coupled to a DAD detector. The analyses were performed on a C18 reverse phase column (250 x 4.5 mm) employing a two-solvent system (Solvent A : 1 M acetic acid; Solvent B: 80% acetonitrile with 20% A), following modified conditions reported by Riberiro de Lima et al. Each individual chromatogram was monitored simultaneously at 306 nm which corresponds to the $\lambda_{\text{max}}$ for trans-resveratrol (1a).

![Figure 2.2](image_url)

*Figure 2.2.* Separation of the trans-resveratrol analogues (1a, 18, 54, 56, 73 and 88) by reverse phase HPLC.

After recording the individual retention times of the trans-resveratrol glucosides, a mixture of all compounds (1a, 18, 54, 56, 73 and 88) was prepared and injected into the HPLC to acquire a chromatogram displaying all six trans-resveratrol analogues, *Figure*
2.2. It can clearly be concluded that \textit{trans}-resveratrol (1a) and the five glucosides were completely separated utilising this procedure and that this method of analysis may be applicable to the detection of these compounds in wine. Furthermore, and as expected, the order of elution follows their expected polarities i.e. the more polar tri-glucoside (88) elutes first followed by the di-glucosides (54 and 73), then the mono-glucosides (18 and 56) and finally followed by \textit{trans}-resveratrol (1a) itself being the least polar of all compounds injected.

The UV-Vis absorption spectra (190 to 600 nm) of the individual \textit{trans}-resveratrol analogues (1a, 18, 54, 56, 73 and 88) were acquired from the HPLC runs, Figure 2.3.
Comparison of the UV-Vis spectra of the mono-, di- and trans-triglucosides with the parent trans-resveratrol (1a) indicated that all spectra were essentially identical. The λ max absorbance peak for all derivatives occurred between 304-306 nm. This is not surprising, as one would not expect glycosidation of the phenolic groupings within (1a) to alter the conjugation throughout these analogues.

2.5 Preparation of the cis-Resveratrol Glucosides.

With the five mono-, di- and trans-triglucosides in hand, we next prepared the corresponding cis analogues (1b, 89-93) utilising irradiation with light of wavelength (365 nm) utilising a modified version of the procedure previously reported for the isomerisation of trans-resveratrol (1a) into cis-resveratrol (1b) by Allen et al. 34

Scheme 2.7. Isomerisation of the trans-resveratrol analogues (1a, 18, 54, 56, 73 and 88) into the cis-resveratrol analogues (1b, 89-93).

Specifically 10 mg of each of the trans-resveratrols (1a, 18, 54, 56, 73 and 88) was individually dissolved in d4-MeOH (5 mL) and the solutions irradiated with UV light (365 nm) for 40 min. The irradiated samples were then immediately subjected to HPLC-DAD
analysis employing the same conditions for the analysis of trans-resveratrol glycosides. The individual chromatographic spectrums were acquired, an example of which is shown in Figure 2.4 for the isomerisation of the trans-resveratrol diglucoside (54) into its cis counterpart (92). Irradiation for times > 40 minutes failed to increase the yield of the cis isomers indicating that these reactions had reached a steady state phase. It should be noted that the isomerisation of trans-resveratrol (1a) into its cis-counterpart (1b) is highly dependent on the sample matrix, although ratios of 10 : 90 (trans : cis) are often observed when the isomerisation is conducted in methanol. Finally, the solutions were directly analysed by $^1$H NMR immediately after irradiation in order to determine the ratio of the cis isomers relative to the trans analogues. These analyses indicated that the cis-glucosides were formed in yields ranging from 90 to 95%, Scheme 2.7. It should be further noted that the samples of the cis-resveratrol analogues (1b, 89-93) would undergo slow isomerisation back into their more stable trans counterparts over a number of hours, however, this isomerisation was slow if the samples were kept dilute and in the cold. Thus, for the antioxidant capacity studies below the samples were used immediately after their synthesis.

![Figure 2.4](image)

**Figure 2.4.** HPLC analysis of the cis/trans-isomerisation of the resveratrol diglucoside trans-(54) into cis-(92).

Confirmation of the structures of the cis-resveratrol analogues (1b, 89, 91-93) was readily made by comparison with the $^1$H and $^{13}$C NMR data already reported in the literature. The diglucoside (90) is yet to have been reported. Key features in the $^1$H
NMR resonance of these compounds indicated the presence of a cis C=C at ca. 6.5 and 6.4 ppm displaying a coupling constant of ca. 12 Hz. Furthermore, the anomeric proton(s) of the sugar unit(s) appeared as a doublet between 4.61-4.93 ppm $\delta$ and displayed a coupling constant between 6.6-7.8 Hz. In addition, analysis of the $^{13}$C NMR spectra of these compounds indicated the presence of the appropriate number of anomeric carbon signals at ca. 102 ppm $\delta$.

### 2.5.1 UV-Vis Analysis of the cis-Resveratrol Analogues.

We next examined the individual UV-Vis absorption spectra of the cis-resveratrols prepared above, Figure 2.5. It was found that the maximum absorbance wavelength of these cis-analogues decreases to around 285 nm and represents a blue-shift when compared to the trans-analogues, which is in good agreement with that reported for the cis-resveratrol glycosides.\textsuperscript{31,34} Once again it can also be see that there is no significant difference between the absorbance maxima for these cis-resveratrol glucosides.
2.6 Antioxidant Capacities of the cis/trans-Resveratrol Glycosides.

With all cis/trans-resveratrol glycosides now prepared, we next determined the antioxidant capacities of all these cis/trans-resveratrol analogues employing both the in-vitro FRAP and DPPH assays. These assays are commonly utilised to determine the antioxidant activities of polyphenolics and would allow us to compare our results with those already reported in the literature for some of the resveratrol analogues. Furthermore, any subtle differences when employing these two different methods of determination of antioxidant capacity would be revealed.

2.6.1 Utilisation of the FRAP Assay.

The FRAP (Ferric Ion Reducing Antioxidant Power Assay) is a novel method for assessing the “antioxidant power” of phenolics and was developed by Benzie et al. in 1996.

During the FRAP assay, the ferric salt $\text{Fe}^{3+} \text{(TPTZ)}_2\text{Cl}_3$ is used as an oxidant and is reduced by the presence of an antioxidant to the corresponding ferrous salt $\text{Fe}^{2+} \text{(TPTZ)}_2\text{Cl}_3$ through a redox electron-transfer reaction, *Scheme 2.8*. This process is accompanied with an increase in the absorbance at 593 nm, which is proportional to the concentration of the phenolic antioxidant with the absorbance change is readily measured employing a UV-Vis spectrometer.

\[
\begin{align*}
\text{FRAP reagent} & \quad \text{antioxidant} \\
\text{Fe}^{3+} + \text{TPTZ} & \quad \text{(phenolics)} \\
\text{colourless} & \quad \text{blue}
\end{align*}
\]

\(\text{TPTZ} = 2,4,6\text{-tripyridyl-s-triazine}\)

*Scheme 2.8*. The reduction pathway of the FRAP complex with a phenolic antioxidant.

In this study, the FRAP reagent was prepared according to a previous reported method and is described herein within the experimental section. In addition, and as
suggested by Hengst et al., a calibration curve for the FRAP assay was constructed for trolox (vitamin E) using solutions in concentration ranges of 7.5-15 µM. A linear plot of absorbance at 593 nm vs concentration was constructed following the FRAP assay detailed below. Equation established: \( y (\text{Trolox}) = 0.045x + 0.021, \ r^2 = 1.000, \ Figure\ 2.6.\)

**Figure 2.6.** The calibration curve (Change of absorbance vs concentration of trolox) measured by the FRAP assay.
Figure 2.7. Antioxidant capacities of the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) employing the FRAP assay.

The stock solutions of cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) were prepared at a concentration of 400 µM/L in MeOH. The FRAP reagent (3 mL) was then mixed with 0.3 mL of the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) individually and topped up to 4 mL with MeOH. The samples were incubated at 37 °C for 45 minutes before analysis and the absorbance recorded at 593 nm, respectively. The results were repeated in triplicate and expressed by trolox equivalence and are displayed in Figure 2.7.

2.6.2 Utilisation of the DPPH Assay.

The DPPH assay utilises the stable 2,2-diphenyl-1-picrylhydrazyl radical (DPPH radical) in a methanolic solution, which displays a purple colour in solution and a characteristic UV-Vis absorption maximum at 515 nm. The addition of a phenolic antioxidant quenches the DPPH radical and results in a loss of colour or decrease in absorbance, which is proportional to the concentration of the phenolic compound added, Scheme 2.9. 53

\[
\begin{align*}
\text{(DPPH radical)} & \quad \text{(Purple colour)} \\
\text{(phenolic antioxidant)} & \quad \text{(colourless)}
\end{align*}
\]

Scheme 2.9. The reaction mechanism for the DPPH assay in the presence of a phenolic antioxidant.

The DPPH reagent was prepared according to the method previously reported. 62,119 Trolox (Vitamin E) was again utilised as a standard to construct a linear calibration curve (Change of absorbance vs concentration of trolox) as shown in Figure 2.8.
Figure 2.8. The calibration curve (change of absorbance vs concentration of trolox) measured by DPPH assay.

Figure 2.9. Antioxidant capacities of the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) employing the DPPH assay.

The same stock solution of the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) prepared for the FRAP assay above were used for the DPPH test. The individual samples were prepared by mixing 3 mL of the DPPH solution (60 µmol) with 0.3 mL of
the resveratrol analogues and toped up to 4 mL with MeOH. The mixed solutions were kept in dark for 30 minutes before analysis. The absorbance for the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) was recorded and the results expressed as trolox equivalence according to the equation \( y = -0.024 + 0.456 \) established above. Figure 2.8.

2.7 Discussion on the Antioxidant Capacities of the cis/trans-Resveratrol Glycosides.

The antioxidant performance of the parent cis and trans-resveratrols (1a/b) and the corresponding ten mono-, di- and cis/trans-triglucosides (18, 54, 56, 73, 88, 89-93) employing the FRAP and DPPH assays are depicted in Figures 2.7 and 2.9. Globally, the antioxidant capacity trends found between utilising the two types of assays were very similar although some subtle differences were found for the cis-glucosidated analogues. In terms of antioxidant capacity strength, the parent cis and trans-resveratrols (1a/b) displayed the highest antioxidant strength and there was no difference between these geometrical isomers. Previously it has been reported that the cis-resveratrol (1b) has a slightly higher antioxidant capacity when compared to the trans derivative (1a) although the IC\(_{50}\) values are very close.\(^{121,122}\) The cis and trans-tri-glycosides (88 and 93) displayed no antioxidant capacity as expected as there are no free phenolic hydroxyls to participate in hydrogen atom abstraction.

It has been well established experimentally\(^{56,123-125}\) and theoretically\(^{126}\) that hydrogen atom abstraction from the para 4'-OH within the A ring of resveratrol is more favourable than hydrogen abstraction from the meta 3'-OH or 5'-OH groupings within the B ring of the cis and trans-resveratrols (1a/b). Given that theoretical calculations\(^{126}\) suggest little difference between the bond strengths of the 4'-OH and those of the 3'-OH or 5'-OH groupings within resveratrol and its glycosidated analogues, suggests that the main driving force for hydrogen abstraction from the 4'-OH within the A ring is a result of a lower activation barrier to abstraction due to the formation of eight stabilised resonance structures whilst abstraction from the 3'-OH or 5'-OH within the B ring only results in the possible formation of four resonance structures, Scheme 2.10. Given that hydrogen atom abstraction from cis-resveratrol (1b) leads to the same number of resonance contributors and the fact that these resonance structures are interchangeable between the two manifolds (e.g. delocalisation of (102) may lead to (98) after bond rotation) suggests that little difference between the antioxidant capacities of the cis and trans analogues should
be seen as observed herein.

\[ \text{Scheme 2.10.} \text{ H-atom abstraction from the } para \ 4'-\text{OH vs the } meta \ 3'\text{-OH or } 5'\text{-OH within resveratrol and the associated resonance structures.} \]
Glycosidation of the 4’-OH grouping within the A ring of resveratrol would naturally block this key para 4’-OH grouping and should lead to a dramatic decrease in antioxidant capacity. Indeed, as can be seen in Figures 2.7 and 2.9, glycosidation of the 4’-OH grouping within the A ring of both cis- and trans-resveratrol (compounds 90 and 56 respectively) resulted in a dramatic reduction in antioxidant capacity. Reductions of approximately 80% and 60% were seen for the FRAP and DPPH assays respectively, Figures 2.7 and 2.9. Consequently, taking into account that the para 4’-OH is completely blocked and cannot contribute to the overall antioxidant capacity and that the B ring has two exposed meta 3’-OH and 5’-OH groupings allows us to suggest for the first time that these meta hydroxyl groupings within resveratrol contribute somewhere in the order of 10-20% individually toward the overall antioxidant capacity of resveratrol itself. A recent study by Sato et al. in 2014 also found that the trans-glucoside (56) displayed very low antioxidant capacity when compared to trans-resveratrol (1a) itself and correlates well with our findings.115 Again both the cis- and trans-glucosidated analogues (90 and 56) displayed the same levels on antioxidant capacities, again suggesting that the configuration about the central stilbenic double bond is of little consequence to overall antioxidant activity for these glucosidated derivatives.

Given that each of the free meta 3’-OH and 5’-OH groupings within the B ring of resveratrol appear to contribute in the order of 10-20% of the antioxidant capacity of the cis and trans-resveratrols, then we expected that glycosidation of one of the 3’-OH or 5’-OH groupings within the B ring of resveratrol should result in a decrease in antioxidant capacity by some 10-20%. Indeed, for trans-(18) a decrease of some 20% in antioxidant capacity was found in both assays whilst a 20% reduction for cis-(89) was found in the DPPH assay although no decrease in activity was found when employing the FRAP assay, and highlights that more than a single antioxidant assay should be employed when analysing a series of closely related phenolics in order to avoid aberrant conclusions based on subtle differences in the oxidants employed in these antioxidant capacity tests. It should be noted that a recent study by Sato et al. in 2014 also found that the trans-glucoside (18) displayed a slightly poorer antioxidant capacity when compared to trans-resveratrol (1a) itself, which correlates well with our findings.115
Glycosidation of both the meta 3’-OH of the B ring and the para 4’-OH of the A ring was expected to result in the cis and trans-resveratrol di-glucosides (91 and 73) which display very poor antioxidant capacities in the range of 10-20% given that the meta 5’-OH still remains free. However, the trans-resveratrol di-glucoside (73) displayed even poorer antioxidant activity levels of only 2-5% when compared to trans-resveratrol (1a) itself, perhaps suggesting that diglycosidation of the trans analogue results in substantial twisting of the stilbenic moiety, which in turn results in a strengthening of the meta 5’-OH bond due to a decrease in the energetic favourability of the four potential resonance structures (103-106), Scheme 2.10. Conversely, the fact that the sugar units are inductively withdrawing may also aid in the strengthening of the meta 5’-OH thus resulting in the observed lowering of antioxidant strength. The cis-resveratrol di-glucoside (91) displayed an antioxidant capacity level of some 20% when compared to cis-resveratrol itself within the FRAP assay as expected, however, displayed an enhanced antioxidant capacity in the order of 50% when compared to cis-resveratrol itself (1b) within the DPPH assay. Again differences in observed antioxidant capacity strength appear to be due to the choice of antioxidant employed vs subtle structural and electronic differences in the ground state structures of the cis and trans-resveratrol di-glucosides (91 and 73). Overall, however it can be concluded that these di-glucosides display much poorer antioxidant activities when compared to their parent counterparts (1a/b) as would be expected.

Finally, glucosidation of both meta 3’-OH and 5’-OH groupings within the B ring of the cis and trans-resveratrols (92 and 54) whilst leaving the para 4’-OH grouping free within the A ring resulted in observed antioxidant capacity levels of around 50% for the cis and trans-resveratrols (92 and 54) when compared to the parent analogues themselves (1a/b) for both assays although a slightly elevated level was found for the cis-diglucoside (92) in the DPPH assay. This trend is again consistent with the fact that the para 4’-OH grouping within the A ring dominates the antioxidant capacities of these resveratrol derivatives but also again highlights that there is still a significant combined contribution from both meta 3’-OH and 5’-OH groupings within the B ring.

The results above on the observed antioxidant capacities of the parent cis and trans-resveratrols (1a/b) and the corresponding ten mono-, di- and triglucosides (18, 54, 56, 73, 88, 89-93) employing the FRAP and DPPH assays highlight several key findings.
Globally it can be concluded that the \textit{para} $4'$-OH grouping within the A ring dominates the antioxidant capacities of these resveratrol derivatives with the \textit{meta} $3'$-OH and $5'$-OH groupings within the B ring contributing around some 10-20\% individually toward the overall antioxidant capacity of resveratrol itself. Furthermore, there is little difference seen between the observed antioxidant capacity strength and the stereochemistry about the central stilbenic C=C double bond. This latter finding bodes well with the fact that the \textit{cis} series of resveratrol analogues were only of ca. 95\% purity in terms of \textit{cis} orientation (i.e. contained ca. 5\% \textit{trans} after synthesis), a fact that clearly would make no real difference to the overall observed antioxidant capacities as the individual antioxidant capacities are essentially the same for both the \textit{cis} and \textit{trans} series of compounds.

\textbf{2.8 Conclusions for Chapter Two.}

In summary, this chapter details the successful synthesis of all ten mono-, di- and triglucosides of \textit{cis/trans}-resveratrol (18, 54, 56, 73, 88, 89-93). The synthesis began with a one-pot synthesis of the TBDMSO-protected resveratrols, which were readily separated by column chromatography, \textit{Scheme 1.3}. The Kunz reagent was then employed to attach the requisite protected glucose units, \textit{Scheme 1.4}. Simple deprotection allowed for the ready access to these resveratrol glucosides and the reactions could be carried out on large scale. The \textit{trans}-analogues were then transformed into the corresponding \textit{cis}-isomers neatly by irradiation with UV light (365 nm) resulting in conversions of 92-95\%, \textit{Scheme 2.7}. Examination the individual UV-Vis absorption spectra of the \textit{cis} and \textit{trans}-resveratrols prepared above indicated that the maximum absorbance wavelength of these \textit{cis}-analogues decreases to around 285 nm and represents a blue-shift when compared to the \textit{trans}-analogues ($\lambda_{\text{max}}$ 304-306 nm), \textit{Figures 2.3 and 2.5}. There was no significant difference between the absorbance maxima for the \textit{cis}-resveratrol glucosides or their \textit{trans} counterparts, which was not surprising, as one would not expect glycosidation of the phenolic groupings within (1a/b) to alter the conjugation throughout these analogues. All the \textit{cis} and \textit{trans}-glucosides seperated nicely by HPLC and were fully characterised by NMR.

Antioxidant capacities of the parent \textit{cis} and \textit{trans}-resveratrols (1a/b) and the corresponding ten mono-, di- and triglucosides (18, 54, 56, 73, 88, 89-93) employing the
FRAP and DPPH assays were then measured, the results of which are displayed in Figures 2.7 and 2.9. Globally it can be concluded that the para 4'-OH grouping within the A ring dominates the antioxidant capacities of these resveratrol derivatives with the meta 3'-OH and 5'-OH groupings within the B ring contributing around some 10-20% individually toward the overall antioxidant capacity of resveratrol itself. Furthermore, there is little difference seen between the observed antioxidant capacity strength and the stereochemistry about the central stilbenic C=C double bond.

There has been some debate as to whether the levels of trans-resveratrol consumed in a human diet, by the injection of wine are sufficient to explain the perceived health benefits of the French paradox.\textsuperscript{127} This is essentially based on the fact that the observed antioxidant activity of trans-resveratrol is low when compared to other polyphenolics found in wine, coupled with its relatively low levels found in wine is not sufficient to explain the enhance health benefits observed. Given that it is known that glycosylation of bioactive substrates such as resveratrol can enhance their water solubility, intestinal absorption and physiochemical stability,\textsuperscript{104,115,128} coupled with the fact that many of these other potentially potent resveratrol glucosides have not had their biological effects measured previously, it may be possible to propose the following.

"The consumption of a larger pool of resveratrol glycosides by humans, whether it be from specific foodstuffs or from wine, may in fact enhance the overall health benefit of free resveratrol as the glucosylated analogues may be hydrolysed back to the parent free resveratrol in the stomach/intestine/tissues and thus lead to an increase in the overall level of free resveratrol in the tissues/organs within the body."

For example, if we take the extreme case of the trans-triglucoside (88) which displays no antioxidant activity but has been consumed in a human diet. If it undergoes hydrolysis within human tissues then it can hydrolyse into either of the di-glucosides (54 or 73), which do display some antioxidant activity and thus could provide some additional health benefits. Further downstream hydrolysis of either of these di-glucosides will lead to the formation of either mono-glycosides (18 or 56), which again have significant antioxidant activity. Finally, hydrolysis of either of these mono-glucosides will result in the formation of the parent trans-resveratrol (1a), which displays the highest antioxidant activity overall. Moreover, the same is true if any of the cis series of potential resveratrol glucosides were
consumed in a human diet as this could also add to the overall health benefits in the same way. Consequently, we suggest that in order to get a much clearer picture on how the perceived benefits of the French paradox are linked to resveratrol consumption, then one needs to first determine whether all these other potential resveratrol glucosides are present in wine and what their levels are, and carry out full biological studies employing this pool of resveratrol glucosides. Finally, such considerations may need to be taken into account for all biological studies where the perceived health benefits for humans are being correlated with the foodstuff being consumed on a daily basis.
CHAPTER 3: SYNTHESIS OF PICEATANNOL GLUCOSIDES AND THEIR ANTIOXIDANT CAPACITIES

3.1 Piceatannol and its Bioactivity.

Piceatannol (20) was isolated for the first time from Euphorbia lagascae in 1984 and since then has been found in many plant species such as sugar cane, peanuts, *Rheum undulatum, rhubarb*, vaccinium berries and grapes. The synthesis and accumulation of this compound in plants is reported to be associated with environmental stresses such as fungal attack, ultraviolet exposure, and microbial infection. The biological activity of piceatannol in living organism is still not clear due to limited *in-vivo* studies. However, recent *in-vitro* assays have shown that this compound displays many biological properties such as anticancer, antioxidant and anti-inflammatory activities. Several studies have suggested that piceatannol (20) may be used as a potential anti-cancer agent due to its anti-tumour properties. This is supported by studies showing that the anti-proliferative activity of piceatannol is associated with the inhibition of the progression of Caco-2 cells through the S phase of the cell cycle. In addition, Ashikawa has reported that piceatannol is able to suppress the activation of the nuclear transcription factor by inhibiting IkBα kinase and p65 phosphorylation. Furthermore, piceatannol displays immunosuppressive and antitumorigenic activities in several cell lines as a protein-tyrosine kinase inhibitor. This property is suggested to be important for the prevention of graft rejection.

Apart from these important bioactivity studies, it has been reported that piceatannol displays promising antioxidant capacity according to several antioxidant capacity studies. For example, Pierre et al., have reported that piceatannol exhibits much better antioxidant activity in DPPH and LDL assays than resveratrol. The mechanism of its antioxidant activity was investigated by Shang et al., and is depicted in Scheme 3.1. In short, a radical (107) is generated from the parent compound (20) via either hydrogen atom transfer (HAT) or a sequential proton loss electron transfer (SPLET) mechanism followed by the loss of another hydrogen atom to afford the key diradical (108). The diradical (108) subsequently rearranges to the o-quinone (111) which then condenses with another piceatannol molecule via a Diels-Alder reaction to yield the
piceatannol dimer (112).

**Scheme 3.1.** A proposed mechanism of the antioxidant activity of piceatannol.

3.1.1 Piceatannol Glycosides in Plants.

In plants, enzymatic glycosylation of piceatannol can afford several piceatannol glycosides. *Figure 3.1.* For example, a mono piceatannol glycoside named astringin (32) has been identified as the major constitutive antifungal compound in Sitka spruce (*Picea sitchensis*). A recent finding has indicated that this compound is also present in grapes together with its parent compound. In addition, another mono piceatannol glycoside
(113) has been found in several plants, including rhubarb, which is widely used as a traditional Chinese medicinal herb and a Korean plant termed rhubarb R. undulatum. Apart from these mono glycosides, the identification of a diglycosidated piceatannol (114) has also been reported previously.

\[ \text{Figure 3.1. The structure of piceatannol glycosides (32), (113) and (114).} \]

3.1.2 Piceatannol and its Glycoside (Astringin) in Wine.

Piceatannol (20) and its mono-glycoside (astringin) (32) have been found in a variety of wines. A previous wine analysis study has shown that the two compounds are present in both red and white wines in a significant level in a survey of 120 red and white wines. Another wine study in 2007 quantified trans-astringin levels in 19 red and 3 white Italian wines. It was reported that the concentration in red wine varied between 9-35 mg/L compared with 9-15.6 mg/L in the white wine. In recent years, the use of new techniques such as ultra-high-pressure liquid chromatography (UPLC) and stir bar sorptive extraction with gas chromatography–mass spectrometry (SBSE-TD-GC–MS) has greatly eased the analysis of wine components from the complex matrix. This has facilitated the identification of important stilbenes such as resveratrol (1), oxyresveratrol (21) and piceatannol (20) and their glycosides. For example, many stilbenes including piceatannol and astringin have been identified in some Burgundy red wines. A recent wine study in 2013 has also shown for the first time that both trans and cis isomers of piceatannol, astringin and oxyresveratrol are present in 15 red, rose and white wine samples.

3.1.3 Strategies for the Synthesis of Piceatannol.

As piceatannol (20) has been recognised as another important molecule in a similar fashion to resveratrol due to its significant pharmaceutical potential towards human health,
coupled with the limited supply from nature, a number of synthetic strategies have been
developed for the large scale preparation of this compound. The syntheses begin from
cheap aromatic compounds such as 3,5-dihydroxybenzoic acid (14) or vanillin (122).146
For example, a seven step synthesis of piceatannol employing 3,5-dihydroxybenzoic acid
(14) as the precursor has been reported by Piao et al., Scheme 3.2.133,146 The total
synthesis began with the simple esterification and methylation of the precursor (14) to
afford (116). Reduction and bromination of (116) furnished the bromide (118). Conversion to the phosphonate (119) followed condensation with the benzaldehyde (120)
lead to the formation of the methylated piceatannol (121). Demethylation of (121) yielded
piceatannol (20) as the final product, however, the yields were not reported in this study.

Scheme 3.2. Synthesis of piceatannol via Wittig-Horner reaction.

In 2002, Han et al. reported an alternative route for the preparation of piceatannol (20),
Scheme 3.3.147 Vanillin (122) was benzylated with benzyl bromide in the presence of
potassium carbonate in acetone and then reacted with trimethylsilyl diazomethane and n-buthyllithium to furnish the ethynylbenzene (124) via Colvin rearrangement. The condensation of (124) with (125) catalysed by PdCl₂(PPh₃)₂ and CuI in the presence of triethylamine yielded the alkyne (126) in very high yield. Bromination of (126) followed by zinc reduction furnished the protected piceatannol (128) in 80% yield. This compound was then deprotected utilising BBr₃ to furnish targeted (20) in 50% yield.

Scheme 3.3. Synthesis of piceatannol via Pd catalysed coupling.

Importantly, a direct transformation of resveratrol (1a) into piceatannol (20) via IBX oxidation has been reported recently, Scheme 3.4. The transformation can be simply achieved by the oxidation of resveratrol (1a) to the quinone (111) in the presence of IBX (129) at -78°C in MeOH followed by reduction of the intermediate quinone (111) with the reduction agent Na₂S₂O₄.
Chapter 3 Synthesis and Antioxidant Capacity Studies of Piceatannol Glucosides

Scheme 3.4. Synthesis of piceatannol (20) by IBX oxidation.

3.2 Synthesis of Piceatannol and Piceatannol Glycosides.

Encouraged by the successful synthesis of all possible resveratrol glycosides in the previous chapter, we decided that we would attempt to prepare piceatannol (20) and all possible forms of its glycosides (32, 113, 114, 130-137) as the extension of this research. Considering the potential time constraints for this project, we focused on only the synthesis of trans-piceatannol analogues in this study, Figure 3.2.

Figure 3.2. The targeted piceatannol glycosides to be synthesised.
3.2.1 Synthesis of IBX (129).

Considering that the synthesis of piceatannol (20) may be easily accomplished in a one pot reaction by IBX oxidation of resveratrol (1) followed by Na₂S₂O₄ reduction, we decided to repeat this reaction by applying the same procedure reported by Lucia et al. The oxidising reagent, IBX (129), was first prepared according to the procedure developed by Giuffredi et al., Scheme 3.5. 2-Benzoi acid (138) was treated with oxone (139) in water at 70 °C to afford IBX (129) in 70% yield. The ¹H and ¹³C NMR data of the product (129) were all in accordance with that reported by Giuffredi et al.

\[
\begin{align*}
\text{Compound} & \quad \text{Reagent} & \quad \text{Condition} & \quad \text{Yield} \\
(138) & \quad 2\text{KHSO}_3.\text{KHSO}_4.\text{K}_2\text{SO}_4 & \quad \text{water, 70 °C, 3h} & \quad 70\% \\
(139) & & & \\
(129) & & & \\
\end{align*}
\]

Scheme 3.5. Synthesis of IBX (129).

3.2.2 Synthesis of Piceatannol (20).

Treatment of trans-resveratrol (1a) in MeOH with 1.5 equivalents of IBX at -20 °C for 1 h followed by the addition of aqueous Na₂S₂O₄ solution afforded piceatannol (20) in 30% yield, together with an unknown byproduct in 20% yield which was not further characterised, Scheme 3.6. The structure of (20) was confirmed by the loss of the aromatic proton signal (H-3’) in the ¹H NMR spectrum of the starting material and further confirmed by the molecular ion [M+H]⁺ (245.0812) calculated for C₁₄H₁₃O₄ in the HRMS spectrum. All proton and carbon signals of this compound (20) in the ¹H NMR and ¹³C NMR spectra were in accordance with that reported by Han et al., Figure 3.3. The HMBC spectrum of (20) allows each individual proton and carbon signal to be assigned, Figure 3.3.
Scheme 3.6. A one-pot synthesis of piceatannol (20) from resveratrol (1a).

Figure 3.3. The proton-carbon NMR correlations of piceatannol (20).
NMR solvent: $d_6$-acetone

3.2.3 Proposed Strategy for the Synthesis of Piceatannol Glycosides.

Encouraged by the successful transformation of resveratrol (1a) into piceatannol (20) via IBX oxidation and Na$_2$S$_2$O$_4$ reduction, we attempted to apply the same strategy to furnish all the targeted piceatannol glycosides (32, 113, 114, 130-137). The proposed synthetic routes are depicted in Scheme 3.7 and Scheme 3.8, in which the pivaloylated resveratrol's (83 and 86) and the silylated resveratrol (66) were to be used as the starting material to synthesise all the targeted compounds. By modification of the resveratrol analogue (83)
via IBX oxidation-Na$_2$S$_2$O$_4$ reduction, the glycosylated piceatannol (140) was envisaged to be prepared. Glycosylation of (140), purification, followed by depivaloylation should therefore afford the targeted compounds (32, 114, 131 and 132), Scheme 3.7.

**Scheme 3.7.** Proposed strategy for the synthesis of piceatannol glycosides (32, 114, 131 and 132).

Similarly, the silylated piceatannol (144) was to be prepared by the use of IBX oxidation and Na$_2$S$_2$O$_4$ reduction, Scheme 3.8. Non-selective glycosylation of (144) was then to be carried out to give the protected glycosides (146) and (147), which are to be deprotected to furnish the piceatannol glycosides (113) and (130). Glycosylation of (144) with excessive Kunz reagent would allow for the synthesis of compound (145). Depivaloylation of this product (145) was to be carried out to afford desired glucoside (133) while further glycosylation of (142) followed by depivaloylation was to be completed to furnish two more piceatannol glycosides (135) and (137). In addition, IBX oxidation of (86) followed by Na$_2$S$_2$O$_4$ reduction, should allow for the pivaloylated piceatannol diglucoside (143) to be produced. This compound (143) was then to be
depivaloylated to afford the desired glycoside (131). Finally, a further glycosylation of (143) following by deprotection was to be carried out to prepare the final two glycosides (134) and (136), Scheme 3.8.

**Scheme 3.8.** Proposed strategies for the synthesis of the piceatannol glycosides (113, 130, 131 and 133-137).

### 3.2.4 Synthesis of the Piceatannol Glycosides (32), (114) and (132).

Treatment of glucoside (83) with 1.5 equivalents of IBX (129) followed by Na₂S₂O₄ reduction afforded the pivaloylated astringin (140) in 74% yield, Scheme 3.9. Formation of (140) was confirmed by the disappearance of the aromatic proton H-3' in the ^1^H NMR spectrum and further confirmed from its mass spectrum, where a molecular ion [M+H]^+ (743.3630) calculated for C₄₀H₅₅O₁₃ was observed. The pivaloylated glucoside (140) was then hydrolysed in a sodium methoxide solution to furnish the piceatannol mono-
glycoside (32) in 35% yield, Scheme 3.9. The $^1$HNMR and $^{13}$CNMR data of (32) were all accordance with that reported by Teguo et al.\textsuperscript{151}


With pivaloylated glucoside (140) in hand, we next attempted to prepare the three piceatannol diglucosides (131), (132) and (114) by glycosylation of (140) with Kunz reagent followed by base hydrolysis, Scheme 3.10. By treatment of (140) with one equivalents of Kunz reagent over 30 minutes afforded three separable pivaloylated diglycosides (141), (142) and (143) in 1.8%, 6.8% and 7.8%, respectively. It was also noted that the reaction also lead to the formation of a small amount of pivaloylated piceatannol triglycosides although they were formed in less than 1% yield.

The three pivaloylated piceatannol diglycosides (141-143) were confirmed by the individual molecular mass m/z [MH$^+$] (1241.6437, 1241.6514 and 1241.6476) in their HRMS spectra, all in accordance with the calculated molecular mass for C$_{66}$H$_{97}$O$_{22}$. In order to further investigate the structural difference of the three isomers (141-143), the NMR spectra including $^1$H, $^{13}$C, COSY, HSQC and HMBC for each product were obtained on a 500 MHz NMR instrument.
Scheme 3.10. Synthesis of the piceatannol diglycosides (132) and (114).

With regards to the compound (141), the structure was deduced from its analysis of $^1$H NMR spectra, where the chemical shift of H-2 was exactly the same as that of H-6 at $\delta = 6.98$ ppm as expected for a symmetrically substituted diglucoside. This suggests that the two positions (2 and 6) are structurally identical and thus indicates both position 3 and 5 have been glycosylated as depicted in Figure 3.4. This is further supported by the identical chemical shifts for C-2 and C-6 at $\delta = 109.0$ ppm in the $^{13}$C NMR spectrum. Considering that this compound (141) was produced in a very small yield and we wished access to larger quantities it was decided not to hydrolyse (141) into the glucoside (131) at this stage but explore an alternative route as detailed in Scheme 3.8.
In reference to the other two pivaloylated diglycosides (142) and (143), the individual $^{13}$C-NMR spectra indicated that the carbon signals (H-2 and H-6) were separated from one another (Compound 142: C-2 and C-6 at $\delta = 108.7, 106.2$ ppm; Compound 143: C-2 and C-6 at $\delta = 108.9, 106.4$ ppm). This suggests that the positions 2 and 6 on both compounds are not structurally identical. Therefore, it could be deduced that the glycosylation occurred either on positions 3’ or 4’. The structure of the two compounds (142) and (143) were differentiated from their individual HMBC NMR spectra in which a correlation signal of the anomeric proton (H$_{glu}$-1”) with C-3’ (for compound (142)) and C-4’ (for compound (143)) were observed, respectively, Figure 3.5.

By comparing the $^1$HNMR spectra of the two isomers (142 and 143), we noticed that the chemical shift of H-2’ ($\delta = 7.35$ ppm) for compound (142) was much more downfield than that of H-2’ ($\delta = 7.14$ ppm) for compound (143), as shown in Figures 3.6 and 3.7.
This may be due to that the attachment of the sugar moiety on position 3’ which can induce a significant down field shift of the adjacent proton (H-2’). It should be noted that this observation would be quite useful in identifying the regioselectivity of the pairs of glycosylated piceatannol isomers to be prepared below such as (146 and 147).

![Figure 3.6. The aromatic proton signals of (142). NMR solvent: d6-acetone](image)

![Figure 3.7. The aromatic proton signals of (143). NMR solvent: d6-acetone](image)
The purified compounds (142 and 143) were then hydrolysed individually in base to furnish the diglycosides (132) and (114) in excellent yields. The individual products were deduced by the absence of the pivaloyl groupings in the $^1$HNMR spectra (Figures 3.8 and 3.9.) and further confirmed by the same molecular ion $[M+H]^+$ (569).

**Figure 3.8.** The $^1$HNMR spectrum of (132). NMR solvent: $d_6$-MeOH
3.2.5 Synthesis of the Piceatannol Glycoside (130).

Encouraged by the successful synthesis of the piceatannol diglycosides (114 and 132), we next investigated the preparation of a range of glycosides namely (113, 130, 133, 135 and 137) employing compound (66) as the key precursor as shown in Scheme 3.11. By treatment of the silylated resveratrol (66) with 1.5 equiv. of IBX in MeOH followed by Na$_2$S$_2$O$_4$ reduction, the silylated piceatannol (144) was obtained in 73% yield. The compound was deduced by the loss of the proton signal H-3’ in its proton NMR spectrum and further confirmed by HRMS m/z [MH$^+$] (473.2538) calculated for C$_{26}$H$_{41}$O$_4$Si. The glycosylation was carried out with the use of 1.5 equivalents of Kunz reagent (74) with the expectation of preparing the desired three pivaloylated products (146), (147) and (148) in the one reaction. Interestingly, TLC analysis suggested that the glycosylation only afforded a mixture of the pivaloylated monoglycosides (146) and (147) in 1:1 ratio, with no pivaloylated diglycoside (148).

A possible reason for this is that when a hydroxyl group on the catechol (144) (either the meta 3’-OH or the para 4’-OH) is attached with a pivaloylated sugar moiety, it may cause strong steric hindrance to its adjacent hydroxyl group thus prevent the latter to be further glycosylated. In order to confirm this possibility, we repeated this reaction with the use of excessive Kunz reagent (4 equiv.). TLC analysis once again confirmed the presence of the mixture of (146) and (147) but failed to indicate the presence of any compound (148). Due to the failure of preparing the indeterminate (148), the synthesis of the targeted compounds (133, 135 and 137) was not able to be achieved by the above synthetic strategy.

Upon desilylation of the mixture of (146) and (147) followed by separation using flash column chromatography, the pivaloylated monoglycosides (149) and (150) were afforded in low yields. Since the structure of the two compounds can be simply differentiated by comparing the chemical shift of the individual proton signal (H-2’) in the $^1$HNMR spectra as mentioned above, we deduced that a more downfield shift of the proton signal (H-2’) at $\delta = 7.36$ ppm suggested compound (149) was glycosylated on the 3’-OH while the other regioisomer with H-2’ at $\delta = 7.11$ ppm indicated formation of the other isomer (150). The
structure of the two products was further confirmed from their individual HMBC NMR spectra, where the correlation of the anomeric proton (H_{\text{glu-1}}) with C-3’ on (149) and C-4’ on (150) could be observed, respectively. The two substrates (149 and 150) were then hydrolysed individually in a sodium methoxide solution as accomplished for the pivaloylated glycosides (114) and (132). Compound (130) was recovered in only 9% yield whereas (113) was not recovered at all. The low yield of the glycoside (130) and the failure of the recovery of (113) suggest the two piceatannol glycosides (149) and (150) are not stable under strong basic conditions and thus the depivaloylation strategy for these two compounds (149) and (150) would need to be modified.

Scheme 3.11. Synthesis of the piceatannol mono-glycoside (130).
3.2.6 Synthesis of the Piceatannol Glycosides (131), (134) and (136).

Employing a similar synthetic route as designed for (130), the piceatannol glycosides (131), (134) and (136) were prepared as shown in Scheme 3.12.

Scheme 3.12. Synthesis of the piceatannol glycosides (131), (134) and (136).

Treatment of the pivaloylated resveratrol diglycoside (86) with 1 equivalents of IBX followed by Na$_2$S$_2$O$_4$ reduction furnished the pivaloylated piceatannol diglycoside (141)
in 79% yield. The $^1$H NMR of this product was completely consistent with that of (141) obtained previously, Scheme 3.10. Upon depivaloylation of (143) under basic conditions, the glycoside (131) was recovered in 49% yield. The product was deduced by the disappearance of the pivaloyl group in the $^1$H NMR spectrum.

Glycosylation of (141) with Kunz reagent (1 equiv.) afforded a mixture of (151) and (152) in 1:1 ratio. By repeating the reaction using excessive Kunz reagent (3 equiv.) afforded none of the compound (153). This was consistent with the conclusion made earlier that the positions 3’ and 4’ could not be both glycosylated by the use of Kunz reagent due to steric hindrance.

Upon separation by flush column chromatography, two fractions containing mixtures of (151) and (152) in an 7:3 and 2:8 ratio individually were collected, Scheme 3.12. In order to separate the pure compounds (151) and (152), the two mixtures were acetylated individually to give two acetylated mixtures ((154) and (155) in an 7:3 and 2:8 ratio, respectively). By dissolving the two mixtures individually in hot MeOH (50°C) followed by cooling down the solutions to 0°C, the pure compounds (154) and (155) were recrystallised from the two mixtures. The two products (154) and (155) were confirmed by the molecular mass m/z [MH$^+$] (1781.9830, 1781.9836) calculated for C$_{94}$H$_{141}$O$_{32}$ in the HRMS spectra. The individual structures of the two isomers (154) and (155) were determined from their HMBC NMR spectra, in which the correlation signals of the anomeric proton (H$_{\text{gluc}-1}$) with C-3’ (for compound (154)) and C-4’ (for compound (155)) were observed, respectively. Upon hydrolysing the acetates (154) and (155) under basic conditions, the targeted products (134) and (136) were furnished in excellent yields.

3.3 Antioxidant Capacity Studies of the Piceatannol Glucoside Analogues.

With the majority (8 out of 12) of the desired piceatannol glucoside analogues now synthesised, we next investigated the antioxidant capacity of these compounds by in-vitro FRAP and DPPH assays, as described for the resveratrol analogues in Chapter 2. All synthesised compounds were tested with a final concentration of 15 µmmol in both assays. Trans-resveratrol (1a) was used as the reference compound and the measured absorbance from the FRAP and DPPH assays was expressed as resveratrol equivalence.
3.3.1 FRAP and DPPH Assays.

The antioxidant performance of the piceatannol analogues (20, 32, 114, 130-132, 134 and 136) in FRAP and DPPH assays is summarised in Figures 3.10 and 3.11, respectively.

**FRAP Assay**

![FRAP Assay Graph](image1)

**Figure 3.10.** The antioxidant performance of the piceatannol analogues in the FRAP assay.

**DPPH Assay**

![DPPH Assay Graph](image2)
Figure 3.11. The antioxidant performance of piceatannol analogues in the DPPH assay.

Globally, the antioxidant capacity trends exhibited by the *trans* piceatannol analogues (20, 32, 114, 130-132, 134 and 136) from the FRAP and DPPH assays were very similar. It can be seen that piceatannol (20) displays the highest antioxidant activity, some 1.62 or 1.37 fold higher than that of the reference compound *trans* resveratrol (1a). This is not supervising as piceatannol (1a) possesses one more meta hydroxyl moiety than resveratrol, enabling an increased capability of generating phenoxy free radicals.

By comparing with the measured antioxidant values of (20), (32) and (131) with the *para* glycosylated analogues (130), (114) and (136) for both assays, it is again clear that the blockage of the *para* hydroxyl group significantly decreases the antioxidant activity (50%-90%) in a similar fashion to what we observed during the antioxidant capacity tests for resveratrol glycosides in Chapter 2. Thus, we can again conclude that this position plays a dominant role in determining the antioxidant capacity of the parent compounds as well as that for resveratrol itself. The antioxidant performance of (130) is roughly 40% and 50% of that of piceatannol (20) in FRAP and DPPH assays, respectively. This suggests that the contribution of the three *meta* hydroxyl groups on piceatannol (20) towards the total antioxidant capacity is approximately equal to that contributed by the 4’-OH. Due to the fact that the piceatannol glycosides (113) and (133) were not prepared in our study, we were not able to differentiate the antioxidant capability of the *meta* 3-OH or the 5-OH on the B ring with the *meta* 3’-OH on the A ring. However, it should be noted that the *meta* 3’-OH significantly enhance the antioxidant activity of the *para* 4’-OH as we have noticed that the antioxidant performance of astringin (32) is 30% and 10% higher than that of resveratrol (1a) in both the FRAP and DPPH assays, respectively, although the two compounds both bear one *para* hydroxyl group and two *meta* hydroxyl groups.

It has been suggested by Shang et al., that because the *meta* 3’-OH and the *para* 4-OH’ are a catechol grouping they have an enhanced co-effect towards the antioxidant activity. A proposed mechanism for this is described in Scheme 3.13, in which astringin (32) can be transformed to the quinone (160) in the presence of the FRAP reagent via the sequential proton loss electron transfer (SPLET) mechanism and then condensed with its individual parent molecules via Diels-Alder reaction to yield the dimers (162). This
hypothesis is further supported by our findings that all the piceatannol analogues (20, 32 and 131) bearing the catechol group exhibited outstanding antioxidant activity in both the FRAP and DPPH assays.

Scheme 3.13. A proposed mechanism for the ferric reducing capability of compound (32).

3.4 Conclusions for Chapter Three.
Chapter 3 Synthesis and Antioxidant Capacity Studies of Piceatannol Glucosides

In summary, this chapter details the successful synthesis of the hydroxylated resveratrol (piceatannol) and seven piceatannol glycosides (32, 130-132, 134 and 136). The synthesis of piceatannol (20) was achieved by the IBX oxidation of trans resveratrol (1a) followed by Na₂S₂O₄ reduction in a one-pot reaction. The synthesis of the piceatannol glucosides began with the modification of the protected resveratrol analogues (83), (66) and (86) to afford the corresponding piceatannol analogues (140), (141) and (144) via the IBX oxidation/Na₂S₂O₄ reduction system. By selective glycosylation of these compounds with the use of Kunz reagent (74) followed by deprotection of the OTBDMS and pivaloyl groups, seven piceatannol glycosides (32, 130-132, 134 and 136) were obtained. Furthermore, it was noted that sterics inhibited the successful glycosylation of the catechol moiety within compounds (133, 135 and 137). The FRAP and DPPH assays of the piceatannol analogues revealed that the presence of the para 4'-OH again plays a dominate role towards the total antioxidant capacity and contributes to approximately half of the total antioxidant activity of piceatannol. Moreover, it was found that the catechol grouping which is made up of the meta 3'-OH and para 4'-OH moieties can significantly enhance the antioxidant capacity, highlighting that piceatannol (20) possesses a stronger radical scavenging activity than resveratrol (1a) itself. Overall it may be concluded that piceatannol (20) is of more competitive pharmaceutical value than resveratrol (1a) although the former has been much less studied in the past.
CHAPTER 4: RESVERATROL OLIGOMERS

4.1 Resveratrol Dimers in Grapes and Wine.

Resveratrol oligomers are an important group of resveratrol analogues found in grapes and wines. It has been suggested that these compounds are produced from enzymatic oxidation of resveratrol caused by environmental stresses such as fungal infections.\(^{152}\) The most common resveratrol oligomers found in wine are simple resveratrol dimers such as epsilon-viniferin (33), delta-viniferin (163) and pallidol (35), Figure 4.1.\(^{153-156}\) These compounds are derived from the oxidation of resveratrol (1) catalysed by 4-hydroxystilbene peroxidases and can be usually detected together with the parent compound resveratrol (1) in wine analysis.

![Resveratrol oligomers](image)

**Figure 4.1.** Several common resveratrol dimers found in wine.

For example, the levels of epsilon-viniferin (33) and pallidol (35) have been analysed in a study of 21 commercial wines from the south of France, Table 4.1. Epsilon-viniferin (33) was detected in dry red and sweet white wines at levels between 0.1 and 1.63 mg/L. In contrast, pallidol (35) was not found in the sweet white wines but was detected in a few dry reds, rose and white wines, at levels between 0.38 and 2.22 mg/L.\(^{157}\)
Table 4.1. The levels of epsilon-viniferin (33) and pallidol (35) in some commercial wines from the south of France.

<table>
<thead>
<tr>
<th>Wine</th>
<th>type</th>
<th>Vintage</th>
<th>Color</th>
<th>Epsilon-viniferin (mg/L)</th>
<th>Pallidol (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cabernet Sauvignon</td>
<td>Red dry</td>
<td>1998</td>
<td>Red</td>
<td>1.63</td>
<td>--</td>
</tr>
<tr>
<td>Cabernet Sauvignon</td>
<td>Red dry</td>
<td>1999</td>
<td>Red</td>
<td>0.1</td>
<td>--</td>
</tr>
<tr>
<td>Cabernet Sauvignon</td>
<td>Red dry</td>
<td>1999</td>
<td>Red</td>
<td>1.35</td>
<td>2.22</td>
</tr>
<tr>
<td>Merlot</td>
<td>Red dry</td>
<td>1998</td>
<td>Red</td>
<td>0.53</td>
<td>--</td>
</tr>
<tr>
<td>Tempranillo</td>
<td>Red dry</td>
<td>1998</td>
<td>Red</td>
<td>0.13</td>
<td>--</td>
</tr>
<tr>
<td>Egiodola</td>
<td>Red dry</td>
<td>1999</td>
<td>Red</td>
<td>1.12</td>
<td>1.33</td>
</tr>
<tr>
<td>rose from Syrah</td>
<td>rose dry</td>
<td>1999</td>
<td>rose</td>
<td>--</td>
<td>0.38</td>
</tr>
<tr>
<td>Chardonnay (red)</td>
<td>White dry</td>
<td>1999</td>
<td>White</td>
<td>--</td>
<td>0.30</td>
</tr>
<tr>
<td>Chardonnay</td>
<td>White dry</td>
<td>1999</td>
<td>White</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Chardonnay</td>
<td>White dry</td>
<td>1999</td>
<td>White</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Sauvignon</td>
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<td>White</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Sauvignon</td>
<td>White dry</td>
<td>2000</td>
<td>White</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Viognier</td>
<td>White dry</td>
<td>1999</td>
<td>White</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Semillion</td>
<td>Botrytised sweet white</td>
<td>1986</td>
<td>White</td>
<td>0.11</td>
<td>--</td>
</tr>
<tr>
<td>Semillion</td>
<td>Botrytised sweet white</td>
<td>1991</td>
<td>White</td>
<td>0.10</td>
<td>--</td>
</tr>
<tr>
<td>Semillion</td>
<td>Botrytised sweet white</td>
<td>1993</td>
<td>White</td>
<td>0.14</td>
<td>--</td>
</tr>
<tr>
<td>Semillion</td>
<td>Botrytised sweet white</td>
<td>1994</td>
<td>White</td>
<td>0.17</td>
<td>--</td>
</tr>
<tr>
<td>Semillion</td>
<td>Botrytised sweet white</td>
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<td>0.08</td>
<td>--</td>
</tr>
<tr>
<td>Semillion</td>
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<td>1997</td>
<td>White</td>
<td>0.13</td>
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<tr>
<td>Jhu I,jnm Semillion</td>
<td>Botrytised sweet white</td>
<td>1998</td>
<td>White</td>
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<tr>
<td>Semillion</td>
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<td>1999</td>
<td>White</td>
<td>0.12</td>
<td></td>
</tr>
</tbody>
</table>

Apart from viniferins (33 and 163) and pallidol (35), many other resveratrol dimers such as viniferaether (164), malibatol A (165), ampelopsin A (166), ampelopsin B (167) and ampelopsin D (168) have been identified in grapes and wines, Figure 4.2.\textsuperscript{158, 159}
Figure 4.2. Other common resveratrol dimers found in wine.

Formation of (+)-ampelopsin B:

Scheme 4.1. Formation of ampelopsin B (167) and ampelopsin D (168) from epsilon-viniferin (33).
Takaya et al., has revealed that several of the dimers such as ampelopsin B (167) and ampelopsin D (168) in plants are produced from the precursor epsilon-viniferin (33) via a protonation/cyclisation process and further suggested that product dimer outcome is dependent on the position of protonation of epsilon-viniferin (33). For example, ampelopsin B (167) may be formed from epsilon-viniferin (33) via the protonation of the stilbenic double bond followed by intramolecular-cyclisation whereas ampelopsin D (168) may be formed from (33) via the protonation of the oxygen atom on the dihydrofuran ring, as depicted in Scheme 4.1.

In addition to resveratrol dimers, some complicated resveratrol oligomers such as resveratrol tetramers have also been found in grapes. For example, a resveratrol tetramer, hopeaphenol (171), was identified for the first time in wines from North Africa together with other common resveratrol analogues including trans-piceid (2a), pallidol (35) and trans-epsilon-viniferin (33), Figure 4.3.

![Figure 4.3. The chemical structure of hopeaphenol (171).](image)

More recently, two new resveratrol tetramers cis-vitisin A (172) and vitisin B (173) have also been separated and characterised from a common grapevine (Vitis vinifera) native to the Mediterranean region, central Europe, and southwestern Asia, Figure 4.4.
4.2 Synthesis of Resveratrol Oligomers.

Due to the fact that many resveratrol oligomers have been shown to have important bioactivities, coupled with the difficulty in extracting large quantities of these compounds from plants for pharmaceutical research, efforts have been made on the synthesis of these oligomers in recent years. Generally, the synthesis of some typical resveratrol oligomers such as epsilon-viniferin (33), delta-viniferin (163) and pallidol (35) are easy to accomplish as these compounds can be obtained from the oxidation of resveratrol (1) directly by employing a variety of reagents such as oxidating reagents, peroxidases or radical initiators.\textsuperscript{154} For example, Takaya et al., investigated the oxidation products of resveratrol by using a variety of oxidising agents.\textsuperscript{154} In their study, three dimers (33), (163) and (35) were afforded in varying quantities as shown in Scheme 4.2 and Table 4.2.

Scheme 4.2. The oxidised products formed from resveratrol by the use of oxidising agents.
The outcome of the oxidation of resveratrol when employing Tl(NO$_3$)$_3$, K$_3$[Fe(CN)$_6$], Ce(SO$_4$)$_2$, FeCl$_3$ and MnO$_2$ as oxidants is depicted in Table 4.2. Treatment of resveratrol (1a) with the oxidising reagent (Tl(NO$_3$)$_3$) over 5 minutes furnished the dimer (33) in 30.1% yield. In contrast, the use of K$_3$[Fe(CN)$_6$] afforded three different dimers (33, 163 and 35) in 21.9%, 22.5% and 15.7% yields respectively. Treatment of resveratrol with FeCl$_3$ or MnO$_2$ furnished the major resveratrol dehydrodimer (33) in very high yield, 97% and 91% respectively whereas the use of Ce(SO$_4$)$_2$ at low temperature gave compounds (33 and 164) in very poor yields.

Table 4.2. Treatment of resveratrol with various oxidising agents.

<table>
<thead>
<tr>
<th>Reagent</th>
<th>Solvent</th>
<th>Temperature (°C)</th>
<th>Time</th>
<th>33 (%)</th>
<th>163 (%)</th>
<th>35 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tl(NO$_3$)$_3$</td>
<td>MeOH</td>
<td>-50</td>
<td>5 min</td>
<td>30.1</td>
<td>---</td>
<td>---</td>
</tr>
<tr>
<td>K$_3$[Fe(CN)$_6$]</td>
<td>MeOH</td>
<td>25</td>
<td>10 min</td>
<td>21.9</td>
<td>22.5</td>
<td>15.7</td>
</tr>
<tr>
<td>Ce(SO$_4$)$_2$</td>
<td>MeOH</td>
<td>-50</td>
<td>26 h</td>
<td>3.7</td>
<td>8.4</td>
<td>---</td>
</tr>
<tr>
<td>FeCl$_3$</td>
<td>Acetone</td>
<td>25</td>
<td>20 h</td>
<td>0.9</td>
<td>97.0</td>
<td>1.5</td>
</tr>
<tr>
<td>MnO$_2$</td>
<td>CH$_2$Cl$_2$</td>
<td>25</td>
<td>24 h</td>
<td>---</td>
<td>91.0</td>
<td>9.0</td>
</tr>
</tbody>
</table>

The dimerisation of resveratrol has been previously investigated and it is generally accepted that the formation of resveratrol dimers proceeds in a radical manner. For example, Shang$^{56}$ reported the mechanism of the reaction of resveratrol with the galvinoxyl radical by analysing the resveratrol dimer formed, Scheme 4.3.$^{56}$ In this study, a resveratrol radical (94) is generated via either hydrogen atom transfer (HAT) or sequential proton loss electron transfer (SPLET) at the 4’-OH position of resveratrol and stabilised by delocalisation via the formation of a number of resonance structures (98) and (99) as depicted in Figure 2.7. The self-condensation of radical (98) affords the product (35) whereas its coupling reaction with either of the radicals (99 or 94) furnishes the dimers (33) and (163) respectively.
Scheme 4.3. The proposed mechanism for the formation of resveratrol dimers (33), (163) and (35).
Compared with the viniferins (33 and 163) and pallidol (35), the synthesis of other resveratrol oligomers usually requires multiple steps. For example, a total synthesis of the resveratrol oligomers ampelopsin D (168) and isoampelopsin (181) has been reported by Snyder et al., in 2007, *Scheme 4.4.* \(^{161}\) In this study, the starting material (175) was coupled with the aldehyde (176) in the presence of \(n\)-butyllithium to form the alcohol (177). Treatment of (177) with acid (TsOH) generates the intermediate (177) which undergoes a cascade reaction featuring cation generation, cyclisation and thiol addition to afford compound (180), which was then transformed to the product (168) via several steps. Isomerisation of (168) in the presence of hydrochloric acid at 80°C gave the other desired product (181).

*Scheme 4.4.* Total synthesis of the resveratrol oligomers ampelopsin D (168) and isoampelopsin (181).
4.3 Preparation of Some Typical Resveratrol Dimers.

In this study, some typical resveratrol dimers including epsilon-viniferin (33), delta-viniferin (163) and pallidol (35) were prepared for wine analysis. The idea being with these authentic compounds in hand we could later quantify their concentrations in a variety of wine samples. The synthesis of these compounds was achieved by employing a direct oxidation of resveratrol with K₃Fe(CN)₆, according to the modified procedure reported by Takaya et al. As we wished not to have to purify by preparative HPLC as described by Takaya et al., coupled with the fact that we wished to synthesise large quantities of these dimers, we devised a simple purification procedure that relied upon acetylation prior to purification.

4.3.1 Preparation of trans-Viniferins Acetates (182 and 183) and Pallidol Acetate (184).

Treatment of trans-resveratrol (1a) with K₃Fe(CN)₆ and K₂CO₃ in MeOH at ambient temperature afforded a mixture of three resveratrol dimers, delta-viniferin (163), epsilon-viniferin (33) and pallidol (35) respectively. Scheme 4.5. Acetylation of these dimers by treatment of Ac₂O and Et₃N in CH₂Cl₂ and DMSO furnished three acetates (182), (183) and (184), which were then subjected to flash column chromatography to afford a pure fraction of pallidol hexaacetate (184) in 19.4% yield and a mixed fraction of trans-delta-viniferin pentaacetate (183) and trans-epsilon-viniferin pentaacetate (182) in 16.5% yield.

The proton and carbon NMR spectra for (184) were both in good agreement with those reported by Khan et al in 1986. An observed correlation between H-8 and H-8’ in the ROESY spectrum suggested that these two protons were on the same face suggesting a cis-configuration about the central fused ring system for (184).

In order to confirm the cis-configuration, crystals of (184) were grown by recrystallising from EtOAc. Examination of the X-ray structure confirmed the cis orientation about the central fused ring system as depicted in Figure 4.5.
**Scheme 4.5.** Synthesis of the dimer acetates (182), (183) and (184).

**Figure 4.5.** The crystal structure of pallidol hexaacetate (184).
4.3.2 Separation of the trans-Viniferins Acetates (182 and 183).

Due to the difficulty in separating the two viniferin acetates (182) and (183) by flash column chromatography, we attempted to apply recrystallisation as an alternative approach to accomplish the task. Scheme 4.6. By carefully recrystallising the mixture from MeOH, at ambient temperature furnished pure (183) (6.9%). The mother liquors were then placed at –20 °C resulting in the precipitation of a mixture of (182) and (183). The mother liquors were then evaporated to afford pure (182) (2.8%).

The structures of trans-epsilon-viniferin pentaacetate (182) and trans-delta-viniferin pentaacetate (183) were deduced from their individual proton NMR spectrums with the main differences being about the aromatic region, Figures 4.6 and 4.7.

Figure 4.6. The $^1$HNMR spectrum of (182). NMR solvent: CDCl$_3$
The structure of the two isomers (182) and (183) were further confirmed by the long-range correlation signals of C-1b with the nearby protons in their HMBC NMR spectra. As shown in Figures 4.8 and 4.9, the correlation signal of C-1b with the proton signals (H-8a and H-8b) and (H-5b and H-8b) can be observed for (182) and (183), respectively. Furthermore, the proton and carbon NMR spectra of the dimer acetate (182) were both identical to those reported by Khan et al. and Ito et al., previously.156,162
Figure 4.9. Some important HMBC signals of (182).

Figure 4.10. Some important HMBC signals of (183).
4.3.3 Deacetylation to Afford Dimers (33), (35) and (163).

Deacetylation of the dimer acetates (182), (183) and (184) was achieved via base hydrolysis in aqueous MeOH in the presence of K$_2$CO$_3$, Scheme 4.7. Column chromatography furnished the target dimers (33), (163) and (35) in excellent yields. The proton and carbon NMR spectra of the three dimers were all in accordance with those reported in the literature, pallidol (35),$^{156}$ trans-epsilon-viniferin (33),$^{162-164}$ trans-delta-viniferin (163).$^{165}$

Scheme 4.7. Hydrolysis of dimer acetates (182), (183) and (184).

4.3.4 Preparation of cis-Viniferins (185) and (186).

With the trans-isomers (33) and (163) in hand, we next attempted to prepare the corresponding cis isomers (185) and (186) by UV irradiation of these compounds as we did for the trans-resveratrol analogues mentioned in Chapter 2. The isomerisation was
achieved by employing a modified procedure to that reported by Yao et al.\textsuperscript{163} Scheme 4.8.

\textbf{Scheme 4.8.} Isomerisation of the resveratrol dimers (33) and (163) by UV light irradiation.

UV irradiation of a solution of (33) and (163) in \textit{d}_4-MeOH for half an hour, the trans-isomers (33) and (163) were transformed into the cis-isomers (185) and (186) in 50\% and 75\% yield, respectively. The cis-adducts (185) and (186) were individually determined by the coupling constant of the olefinic proton signals (H-7 and H-8, \( J = 12 \text{ Hz} \)), which is typical for such cis-structures. The proton and carbon NMR spectra of the cis isomers were also in accordance with those reported in the literature, cis-epsilon-viniferin (185)\textsuperscript{163} cis-delta-viniferin (186).\textsuperscript{166} The yields for both cis-isomers (185) and (186) were calculated by integrating of trans/cis-signals in the \textsuperscript{1}HNMR spectra. Notably, the cis isomers isomerised back to the trans isomers at ambient temperature over time.

\textbf{4.4 Summary for Chapter Four.}

This chapter details the synthesis of three resveratrol dimers (33), (163) and (35) formed via the dimerisation of resveratrol using K\textsubscript{3}Fe(CN)\textsubscript{6} as the oxidising agent in a one-pot reaction. The separation of the three products (33), (163) and (35) were successfully achieved by acetylation of the mixture of (33), (163) and (35) into the corresponding
acetates (182), (183) and (184) and then purified by the use of flash column chromatography and recrystallisation followed by base hydrolysis. The crystal structure of (184) was successfully determined by X-ray diffraction spectroscopy for the first time. In addition, the cis-viniferins (185) and (186) were prepared by UV light irradiation of the corresponding trans-viniferins (33) and (163), respectively. With a simple protocol now in place to synthesise such resveratrol dimers, it paves the way for future work on the synthesis of glucosylated dimers of resveratrol. For example, if we were to take the glucosylated resveratrol analogues synthesised in Chapter 2 and subjected them to oxidative dimerisation then we would expect to form a broad range of glucosylated dimers of resveratrol related to those prepared within this chapter. Such compounds would be expected to have a diverse range of antioxidant properties and other related bioactivities and are worthy of further exploration. Moreover, some of these glucosylated dimers of resveratrol may exist in wines and other foodstuffs and their synthesis would aid in finding such natural products in nature. Indeed as a side project to this PhD thesis we sent a number of our authentic dimers of resveratrol to Prof. Cedric Saucier at UBC, Canada where he utilised them to confirm the presence of such dimers in Canadian wines.

CHAPTER 5: EXPERIMENTAL.

5.1 General Experimental.

Solvents and reagents
Dry organic solvents were purchased and dispensed using a Puresolv™ solvent purification system, except tetrahydrofuran (THF), which was dried over sodium wire with benzophenone as indicator and freshly distilled prior to use. Pyridine was dried by storage on 4Å molecular sieves. General organic solvents were obtained and distilled where needed. Reagents other than those synthesised were purchased from Sigma-Aldrich Chemical Company Ltd., and used without further purification.

Naming of synthesised compounds
Compounds are labeled using common nomenclature as they would appear in the literature, followed by the IUPAC name as generated using ACD/Labs 12.0 software.

Chromatography
Column chromatography was performed using Davisil 40-63 μm silica gel. Thin layer chromatography was performed using Merck silica gel 60 F254 alumina sheets (20 x 20 cm) and visualised under 254 nm light or developed in either vanillin or potassium permanganate dip.

Melting points
Melting points were obtained using a Büchi B-540 melting point apparatus and are uncorrected.

NMR
The $^1$H and $^{13}$C spectra of synthesised compounds were recorded in either CDCl$_3$, $d_4$-MeOH, $d_6$-Acetone or $d_6$-DMSO and acquired on either a Varian Gemini 300, Bruker 500, or Varian Unity Inova 600 Fourier transform spectrometer as indicated. $^1$H spectra were referenced to the TMS peak at 0.00 ppm and the $^{13}$C Spectra were referenced to residual CDCl$_3$ at 77.00 ppm, $d_4$-MeOH at 49.5 ppm, $d_6$-acetone at 29.9 ppm or $d_6$-DMSO at 39.5 ppm. Chemical shifts (δ) are reported in ppm and coupling constants (J) in Hz. $^1$H NMR
multiplicities are described by the following abbreviations: singlet (s), doublet (d), triplet (t), quartet (q), pentet (p), sextet (sext), multiplet (m) and broad (br) for broadened signals.

**X-ray crystallography**
X-ray crystallography was performed by Prof Edward R. T. Tiekink at the Department of Chemistry, University of Malaya, Malaysia.

**Infrared spectra**
Infrared spectra were recorded on a Lambda Scientific FTIR 7600 series spectrophotometer, either neat or as a nujol mull, as indicated.

**Accurate Mass determinations**
Accurate mass determination was performed by the Central Science Laboratories, University of Tasmania.

**Specific rotations**
Specific rotations were measured with a PolAAr 21 polarimeter and referenced to the D sodium line (589 nm) at 20°C in a cell with 1 dm path length. The concentration (c) is specified in g/100 mL and the solvent used as reported.

**Computational Chemistry**
Theoretical calculations were performed using the Spartan ’08 software package, with final geometries and/or energies highlighted where needed.

5.2 Experimental for Chapter 2.

1,2,3,4,6-Penta-\(\text{O}-\)Pivaloyl-\(\beta\)-\(\text{D}\)-glucopyranoside (76).
To a solution of pivaloyl chloride (100 mL, 0.813 mol), triethylamine (113 mL, 0.815 mol), DMAP (900 mg, 7.37 mmol) in dichloromethane (200 mL) at 0°C was added \((R)-\text{D-glucopyranoside (75)}\) (18.3 g, 0.102 mol). The mixture was stirred at ambient temperature for a further 48 h and then the reaction mixture quenched with 2M H\(_2\)SO\(_4\) (300 mL). The mixture was then extracted with dichloromethane (3 x 100 mL) and the combined organics washed with 2M H\(_2\)SO\(_4\) (200 mL), 1M NaHCO\(_3\) (200 mL), and finally water (200 mL). The organics were then dried over anhydrous MgSO\(_4\) and concentrated in vacuo to afford crude (76) which was recrystallised from EtOH to furnish known (76) (51 g, 84%) as a white solid. M.p. 156.3-158.4 °C, Lit.\(^{114}\) M.p. 156-158 °C. \(^1\)H NMR (300 MHz, CDCl\(_3\)): \(\delta 5.69 (1\text{H}, d, J = 8.1 \text{ Hz}), 5.37 (1\text{H}, dd, J = 9.3, 9.3 \text{ Hz}), 5.23 (1\text{H}, dd, J = 9.3, 9.3 \text{ Hz}), 5.17 (1\text{H}, dd, J = 9.6, 9.6 \text{ Hz}), 4.20-3.82 (3\text{H}, m), 1.23, 1.18, 1.16, 1.13, 1.12 (45\text{H}, 5s). All physical and spectroscopic data were in accord with those previously reported.\(^{112}\)

2,3,4,6-tetra-O-Pivaloyl-\(\text{D-glucopyranosyl bromide (77).}\)

To a stirred solution of (76) (49 g, 81.2 mmol) in dichloromethane (250 mL) at 0°C, was added dropwise HBr (41 mL, 33% in acetic acid, 234 mmol) and the reaction mixture kept at ambient temperature for 24 h. The mixture was then quenched with water (200 mL) and the organics extracted into dichloromethane (3 x 100 mL). The combined organics were washed with saturated NaHCO\(_3\) (200 mL), water (200 mL), and dried over anhydrous MgSO\(_4\) followed by concentration in vacuo. Recrystallisation of the crude solid from EtOH afforded (77) (42 g, 89%) as a white solid. M.p. 141.5-143.4 °C, Lit.\(^{114}\) M.p. 142-143 °C. \(^1\)H NMR (300 MHz, CDCl\(_3\)): \(\delta 6.62 (1\text{H}, d, J = 4.2 \text{ Hz}), 5.63 (1\text{H}, dd, J = 9.9, 9.9 \text{ Hz}), 5.21 (1\text{H}, dd, J = 9.9, 9.9 \text{ Hz}), 4.80 (1\text{H}, dd, J = 9.9, 4.2 \text{ Hz}), 4.33 (1\text{H}, ddd, J = 9.9, 4.2, 2.1 \text{ Hz}), 4.16-4.18 (2\text{H}, m), 1.22, 1.18, 1.17, 1.15 (36\text{H}, 4 x s). All physical and spectroscopic data were in accord with those previously reported.\(^{113}\)
1,2-\textit{O}-(1-N-1-Phenylethylideneaminoxy)-2,2-dimethylpropyldene-3,4,5-tri-\textit{O}-pivaloyl-\textit{D}-glucopyranoside (74) (Kunz reagent).

To a stirred solution of (77) (28.7 g, 48.7 mmol) in dichloromethane (400 mL) at -40 °C was added S-collidine (10 mL, 76.8 mmol), acetophenone oxime (8.1 g, 59.9 mmol) and silver triflate (12.8 g, 49.8 mmol). The reaction was stirred for a further 18 h at room temperature. The mixture was then filtered and the volatiles removed \textit{in vacuo}. The residue was then separated by column chromatography (increasing polarity from 0% to 15% ethyl acetate in petroleum spirit as eluant) to afford (74, 76%) as a white solid. M.p. 111.3-113.5 °C. \textit{H NMR} (300 MHz, CDCl3): δ 7.70-7.67 (2H, ArH, m), 7.40-7.34 (3H, ArH, m), 6.20 (1H, d, $J = 6.0$ Hz), 5.24 (1H, dd, $J = 6.0$, 3.3 Hz), 5.01 (1H, dd, $J = 9.6$, 6.0 Hz), 4.68 (1H, dd, $J = 6.0$, 2.7 Hz), 4.25-4.11 (3H, m), 2.24 (3H, s), 1.19, 1.18, 1.16, 1.13 (36H, 4 x s). All physical and spectroscopic data were in accord with those previously reported.\(^{110}\)

\textbf{Synthesis of (TBDMS)-oxy analogues of \textit{trans}-resveratrol;}

3-\{\textit{[\textit{tert}-butyl(dimethyl)silyl]oxy}\}-5-\{\textit{[\textit{tert}-butyl(dimethyl)silyl]oxy}\}phenyl ethenyl]phenol (65), 4-\{\textit{[\textit{E}-2-(3,5-bis\{\textit{[\textit{tert}-butyl(dimethyl)silyl]oxy}\}phenyl) ethenyl]phenol (66), 3-\{\textit{[\textit{tert}-butyl(dimethyl)silyl]oxy}\}-5-\{\textit{[\textit{E}-2-(4-hydroxyphenyl) ethenyl]phenol (67), 5-\{\textit{[\textit{E}-2-(4-\{\textit{[\textit{tert}-butyl(dimethyl)silyl]oxy\}phenyl)ethenyl]benzene-1,3-diol (68), and \{4-\{\textit{[\textit{E}-2-(3,5-bis\{\textit{[\textit{tert}-butyl(dimethyl)silyl]oxy\}phenyl) ethenyl]phenoxy\}\textit{[\textit{tert}-butyl]dimethylsilane} (78). To a solution of \textit{trans}-resveratrol (1a) (8.3 g, 36.4 mmol) in anhydrous acetonitrile (150 mL) and dimethyl sulfoxide (15 mL) was added \textit{tert}-butyldimethylsilyl chloride (9.87 g, 65.5 mmol), anhydrous imidazole (5.0 g, 73.4 mmol) and 4-dimethylaminopyridine (0.15 g, 1.2 mmol) at 0°C. The reaction was
stirred for 12 h, allowing the temperature to warm to ambient temperature. The reaction mixture was then quenched with saturated sodium bicarbonate (100 mL) and the organic layer washed with water (3 x 50 mL). The combined aqueous layers were extracted with ethyl acetate (3 x 50 mL) and the combined organics dried over anhydrous magnesium sulphate, filtered and concentrated in vacuo. The residue was then separated by column chromatography (increasing polarity from 2% to 25% ethyl acetate in petroleum spirit as eluent) to furnish the five titled compounds (65) (4.2 g, 25.3 %), (66) (1.8 g, 10.7 %), (67) (2.3 g, 18.4 %), (68) (1.4 g, 11.3 %) and (78) (3.6 g, 17.3%). All spectral data for all derivatives (65-68, 78) were consistent with that previously reported by Zhang.  

Di-OTBDMS (65): \(^1\)H NMR (200 MHz, CDCl\(_3\)): \(\delta\) 7.37 (2H, d, \(J = 8.6\) Hz), 6.95 (1H, d, \(J = 16.2\) Hz), 6.83 (2H, d, \(J = 8.6\) Hz), 6.82 (1H, d, \(J = 16.2\) Hz), 6.58 (1H, d, \(J = 2.0\) Hz), 6.54 (1H, d, \(J = 2.0\) Hz), 6.24 (1H, dd, \(J = 2.0, 2.0\) Hz), 0.99, 0.97 (18H, 2s), 0.22, 0.21 (12H, 2s).  

Di-OTBDMS (66): \(^1\)H NMR (200 MHz, CDCl\(_3\)): \(\delta\) 7.38 (2H, d, \(J = 8.6\) Hz), 6.95 (1H, d, \(J = 16.2\) Hz), 6.83 (1H, d, \(J = 16.2\) Hz), 6.82 (2H, d, \(J = 8.6\) Hz), 6.59 (2H, d, \(J = 2.0\) Hz), 6.24 (1H, dd, \(J = 2.0, 2.0\) Hz), 0.99, (18H, s), 0.21 (12H, s).  

Mono-OTBDMS (67) \(^1\)H NMR (200 MHz, CDCl\(_3\)): \(\delta\) 7.37 (2H, d, \(J = 8.6\) Hz), 6.95 (1H, d, \(J = 16.2\) Hz), 6.83 (2H, d, \(J = 8.6\) Hz), 6.82 (1H, d, \(J = 16.2\) Hz), 6.57 (1H, dd, \(J = 2.0, 2.0\) Hz), 6.54 (1H, dd, \(J = 2.0, 2.0\) Hz), 6.24 (1H, dd, \(J = 2.0, 2.0\) Hz), 0.98, (9H, s), 0.21 (6H, s).  

Mono-OTBDMS (68) \(^1\)H NMR (200 MHz, CDCl\(_3\)): \(\delta\) 7.30 (2H, d, \(J = 8.6\) Hz), 6.90 (1H, d, \(J = 16.2\) Hz), 6.77 (2H, d, \(J = 8.6\) Hz), 6.76 (1H, d, \(J = 16.2\) Hz), 6.52 (2H, d, \(J = 2.0\) Hz), 6.25 (1H, dd, \(J = 2.0, 2.0\) Hz), 0.97, (9H, s), 0.19 (6H, s).  

Tri-OTBDMS (78): \(^1\)H NMR (200 MHz, CDCl\(_3\)): \(\delta\) 7.37 (2H, d, \(J = 8.6\) Hz), 6.96 (1H, d, \(J = 16.2\) Hz), 6.83 (1H, d, \(J = 16.2\) Hz), 6.82 (2H, d, \(J = 8.6\) Hz), 6.59 (2H, d, \(J = 2.0\) Hz), 6.24 (1H, dd, \(J = 2.0, 2.0\) Hz), 0.99, 0.98 (27H, 2s), 0.21, 0.20 (18H, 2s).

To a mixture of (65) (256 mg, 0.56 mmol), boron trifluoromethane (20 mL) at 0°C was added the Kunz reagent (390 mg, 0.616 mmol). The reaction was kept at 0°C for 30 min, before allowing it to warm to ambient temperature. The reaction mixture was quenched with saturated sodium bicarbonate (15 mL) and extracted with dichloromethane (3 x 20 mL). The combined organic extracts were dried over magnesium sulphate, filtered and the volatiles removed in vacuo. The residue was subjected to column chromatography (increasing polarity from 2% to 20% ethyl acetate in petroleum spirit as eluant) yielded the title compound (79) as a white solid (438 mg, 82%). M.p. 146-148°C. [α]D25 = -8.7° (c = 2.16, ethanol). IR cm⁻¹: 2970, 1744, 1599, 1512.

1H NMR (600 MHz, CDCl₃): δ 7.37 (2H, H₂-2' and H-6', d, J = 7.8 Hz), 6.94 (1H, H-8, d, J = 15.6 Hz), 6.82 (2H, H-3' and H-5', d, J = 7.8 Hz), 6.80 (1H, H-7, d, J = 15.6 Hz), 6.68 and 6.67 (2H, H-2 and H-6, 2s), 6.34 (1H, H-4, s), 5.41 (1H, H_glu-3, t, J = 9.6 Hz), 5.28 (1H, H_glu-2, dd, J = 9.6, 8.4 Hz), 5.22 (1H, H_glu-4, t, J = 9.6 Hz), 5.10 (1H, H_glu-1, d, J = 8.4 Hz), 4.23 (1H, H_glu-6a, d, J = 11.4 Hz), 4.14 (1H, H_glu-6b, dd, J = 11.4, 5.4 Hz), 3.91 (1H, H_glu-5, dd, J = 9.6, 5.4 Hz), 1.18, 1.17, 1.16 and 1.14 (36H, (C=O)C(CH₃)₃, 4s), 0.99 and 0.98 (18H, SiC(CH₃)₃, 2s), 0.21 and 0.20 (12H, Si(CH₃)₂, 2s).

13C NMR (75 MHz, CDCl₃): δ 178.3, 177.4 and 176.7 (C=O), one (C=O) masked, 158.3, 157.1 and 155.9 (C-3, C-5 and C-4'), 140.2 (C-1), 130.6 (C-1’), 129.5 (C-8), 128.1 (C-2’ and C-6’), 126.5 (C-7), 120.6 (C-3’ and C-5’), 113.2 and 108.0 (C-2 or C-6), 108.5 (C-4), 99.7 (C_glu-1), 72.9, (C_glu-5), 72.3 (C_glu-3), 71.2 (C_glu-2), 68.0 (C_glu-4), 62.2 (C_glu-6), 39.1 (C(CH₃)₃), three (C(CH₃)₃) carbons masked, 27.43, 27.40, 27.36, 27.33, (C(CH₃)₃),
25.9 (SiC(CH₃)₃), one (SiC(CH₃)₃) carbon masked, 18.5 (SiC(CH₃)₃), one (SiC(CH₃)₃) carbon masked, -4.06 and -4.11 (Si(CH₃)₂), one set masked. HRMS (ESI⁺) calcd. for (M⁺ + H) C₅₂H₈₅O₁₂Si₂: 955.5423; Found: 955.5416.

4-[(E)-2-(3,5-Bis[(terti-butyl(dimethyl)silyl)oxy]phenyl)ethenyl]phenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (80).

The title compound (80) was prepared from (66) (150 mg, 0.328 mmol), boron trifluoroetherate (0.180 mL, 1.44 mmol) in dry dichloromethane (20 mL) and the Kunz reagent (229 mg, 0.361 mmol) using the method described above for the synthesis of (79). Column chromatography (increasing polarity from 2% to 20% ethyl acetate in petroleum spirit as eluant) afforded the title compound (80) as a white solid (235 mg, 75%). M.p. 161-163 °C. [α]D²⁵ = -6.9⁰ (c = 0.72, ethanol). IR cm⁻¹ 2973, 1735, 1594, 1482. ¹H NMR (600 MHz, CDCl₃): δ 7.39 (2H, H-2’ and H-6’, d, J = 7.8 Hz), 6.96 (2H, H-3’ and H-5’, d, J = 7.8 Hz), 6.93 (1H, H-8, d, J = 15.6 Hz), 6.80 (1H, H-7, d, J = 15.6 Hz), 6.59 (2H, H-2 and H-6, s), 6.24 (1H, H-4, s), 5.42 (1H, H₃glu-3’’, t, J = 9.6 Hz), 5.30 (1H, H₃glu-2’’, dd, J = 9.6, 8.4 Hz), 5.17 (1H, H₃glu-4’’, t, J = 9.6 Hz), 5.08 (1H, H₃glu-1’’, d, J = 8.4 Hz), 4.28 (1H, H₃glu-6a’’, d, J = 12.0 Hz), 4.07 (1H, H₃glu-6b’’, dd, J = 12.0, 6.0 Hz), 3.92 (1H, H₃glu-5’’, dd, J = 9.6, 6.0 Hz), 1.24, 1.18, 1.15 and 1.14 (36H, (C=O)C(CH₃)₃, 4s), 0.99 (18H, SiC(CH₃)₃, s), 0.22 (12H, Si(CH₃)₂, s). ¹³C NMR (75 MHz, CDCl₃): δ 178.3, 177.4 and 176.8 (C=O), one (C=O) masked, 157.0 (C-3, C-5 and C-4’), one C-O masked, 139.4 (C-1), 132.8 (C-1’), 128.1 and 128.0 (C-7 and C-8), 127.9 (C-2’ and C-6’), 117.2 (C-3’ and C-5’), 111.9 (C-2 and C-6), 111.8 (C-4), 99.8 (C₃glu-1’’), 73.0 (C₃glu-5’’), 72.3 (C₃glu-3’’), 71.1 (C₃glu-2’’), 68.2 (C₃glu-4’’), 62.5 (C₃glu-6’’), 39.1 (C(CH₃)₃), three (C(CH₃)₃) carbons masked, 27.43, 27.42, 27.37 and 27.33 (C(CH₃)₃), 26.0 (SiC(CH₃)₃), 18.5 (SiC(CH₃)₃), -4.07 (Si(CH₃)₂). HRMS (ESI⁺) calcd. for (M⁺ + H) C₅₂H₈₅O₁₂Si₂: 955.5423; Found: 955.5407.
3-\{\text{[tert-Butyl(dimethyl)silyl]oxy}\}-5-\{\text{[(E\text{-})-2-\{4-\{\text{[2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)]-\beta-D-glucopyranosyl]oxy}\}phenyl]ethenyl}phenyl-2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)]-\beta-D-glucopyranoside\} (81).

The title compound was prepared from (67) (171 mg, 0.499 mmol), boron trifluoroetherate (0.552 mL, 4.393 mmol), dichloromethane (20 mL) and the Kunz reagent (696 mg, 1.098 mmol) using the method described above for the synthesis of (79). Column chromatography (increasing polarity from 2\% to 20\% ethyl acetate in petroleum spirit as eluant) yielded the title compound (67) as a white solid (568 mg, 85\%). M.p. 231-233 °C. [\alpha]_D^{25} = -14.6° (c = 0.27, dichloromethane). IR cm\(^{-1}\) 2973, 1736, 1593, 1482. 1\text{H} NMR (600 MHz, CDCl\(_3\)): \(\delta\) 7.38 (2H, H-2' and H-6', d, J = 8.4 Hz), 6.97 (2H, H-3' and H-5', d, J = 8.4 Hz), 6.94 (1H, H-8, d, J = 16.2 Hz), 6.83 (1H, H-7, d, J = 16.2 Hz), 6.70 and 6.68 (2H, H-2 and H-6, 2s), 6.36 (1H, H-4, s), 5.42 (1H, H\text{glu}-3 or H\text{glu}-3'', t, J = 9.6 Hz), 5.41 (1H, H\text{glu}-3 or H\text{glu}-3'', t, J = 9.6 Hz), 5.30 (1H, H\text{glu}-2 or H\text{glu}-2'', dd, J = 9.6, 8.4 Hz), 5.29 (1H, H\text{glu}-2 or H\text{glu}-2'', dd, J = 9.6, 8.4 Hz), 5.22 (1H, H\text{glu}-4 or H\text{glu}-4'', t, J = 9.6 Hz), 5.18 (1H, H\text{glu}-4 or H\text{glu}-4'', t, J = 9.6 Hz), 5.11 (1H, H\text{glu}-1 or H\text{glu}-1'', d, J = 8.4 Hz), 5.09 (1H, H\text{glu}-1 or H\text{glu}-1'', d, J = 8.4 Hz), 4.28 (1H, H\text{glu}6a or H\text{glu}6a'', dd, 12.3, 1.8 Hz), 4.22 (1H, H\text{glu}6a or H\text{glu}6a'', dd, 12.3, 1.8 Hz), 4.14 (1H, H\text{glu}6b or H\text{glu}6b'', dd, J = 12.3, 5.4 Hz), 4.09 (1H, H\text{glu}6b or H\text{glu}6b'', dd, J = 12.3, 6.0 Hz), 3.90 (2H, H\text{glu}5 and H\text{glu}5'', m), 1.24, 1.19, 1.18, 1.17, 1.16, 1.15, 1.14, 1.13 (72H, (C=O)(CH\(_3\)), 8s), 0.99 (9H, Si(CH\(_3\)), s), 0.22 (6H, Si(CH\(_3\)), s). 13\text{C} NMR (75 MHz, CDCl\(_3\)): \(\delta\) 178.06, 178.01, 177.22, 177.18, 176.53, 176.37 (C=O), two (C=O) masked, 158.1, 156.86 and 156.79 (C-3, C-5 and C-4'), 139.5 (C-1), 132.2 (C-1''), 128.6 (C-8), 127.8 (C-2' and C-6'), 127.3 (C-7), 116.9 (C-3' and C-5'), 113.1 and 107.9 (C-2 or C-6), 108.4 (C-4),
99.49 and 99.46 (C_{glu}-1 or C_{glu}-1’’), 72.7 and 72.5 (C_{glu}-5 or C_{glu}-5’’), 72.0, (C_{glu}-3 and C_{glu}-3’’), 70.9 and 70.8 (C_{glu}-2 or C_{glu}-2’’), 67.9 and 67.7 (C_{glu}-4 or C_{glu}-4’’), 62.2 and 62.0 (C_{glu}-6 or C_{glu}-6’’), 38.8 and 38.7 (C(CH_{3})_{3}), 27.15, 27.13, 27.11, 27.10, 27.08 and 27.04 (C(CH_{3})_{3}), two (C(CH_{3})_{3}) masked, 25.6 (SiC(CH_{3})_{3}), 18.2 (SiC(CH_{3})_{3}), -4.35 and -4.37 (Si(CH_{3})_{2}). HRMS (ESI') calcd. for (M^+ + H) C_{72}H_{111}O_{21}Si: 1339.7387; Found 1339.7382. Anal. calcd. for C_{72}H_{111}O_{21}Si: C, 64.55; H, 8.28; Found C, 64.61; H, 8.02.


The title compound was prepared from (82) (200 mg, 0.584 mmol), boron trifluoroetherate (0.645 mL, 5.139 mmol), dichloromethane (20 mL) and the Kunz reagent (814 mg, 1.285 mmol) using the method described above for the synthesis of (79). Column chromatography (increasing polarity from 2% to 20% ethyl acetate in petroleum spirit as eluant) yielded the title compound (82) as a white solid (657 mg, 84%). M.p. 130-133 °C. \([\alpha]_D^{25} = -15.7^\circ\) (c = 0.95, ethanol). IR cm\(^{-1}\) 2975, 1746, 1512, 1482. \(^1\)H NMR (600 MHz, CDCl\(_3\)): \(\delta\) 7.35 (2H, H-2’ and H-6’), d, \(J = 9.0\) Hz), 6.94 (1H, H-8, d, \(J = 15.6\) Hz), 6.82 (2H, H-3’ and H-5’), d, \(J = 7.8\) Hz), 6.80 (1H, H-7, d, \(J = 15.6\) Hz), 6.77 (2H, H-2 and H-6, s), 6.44 (1H, H-4, s), 5.41 (2H, H_{glu}-3 and H_{glu}-3’, t, \(9.6\) Hz), 5.26 (2H, H_{glu}-2 and H_{glu}-2’, dd, \(J = 9.6, 8.4\) Hz), 5.20 (2H, H_{glu}-4 and H_{glu}-4’, dd, t, \(J = 9.6\) Hz), 5.11 (2H, H_{glu}-1 and H_{glu}-1’), d, \(J = 8.4\) Hz), 4.23 (2H, H_{glu}-6a and H_{glu}-6a’, dd, \(J = 12.0, 1.8\) Hz), 4.12 (2H, H_{glu}-6b and H_{glu}-6b’), dd, \(J = 12.0, 5.4\) Hz), 3.91 (2H, H_{glu}-5 and H_{glu}-5’, ddd, \(J = 9.6, 5.4, 1.8\ Hz), 1.17, 1.16, 1.15 and 1.14 (72H, (C=O)C(CH_{3})_{3}, 4s), 0.97 (9H, SiC(CH_{3})_{3}, s), 0.22 (6H, Si(CH_{3})_{2}, s). \(^{13}\)C NMR (75 MHz, CDCl\(_3\)): \(\delta\) 178.3, 177.5, 176.8
and 176.7 (C=O), 158.3 and 156.1 (C-3, C-5 or C-4’), 140.7 (C-1), 130.3 (C-1’), 128.7 (C-8), 128.1 (C-2’ and C-6’), 126.1 (C-7), 120.7 (C-3’ and C-5’), 109.2 (C-2 and C-6), 105.4 (C-4), 99.3 (C_{glu}-1 and C_{glu}-1’), 72.9 (C_{glu}-5 and C_{glu}-5’), 72.3 (C_{glu}-3 and C_{glu}-3’), 71.2 (C_{glu}-2 and C_{glu}-2’), 68.0 (C_{glu}-4 and C_{glu}-4’), 62.2 (C_{glu}-6 and C_{glu}-6’), 39.1 and 39.0 (C(CH₃)₃), 27.42, 27.38, 27.36 and 27.32, (C(CH₃)₃), 18.5 (SiC(CH₃)₃), -4.1 (Si(CH₃)₂). HRMS (ESI⁺) calcd. for (M⁺+H) C₇₂H₁₁₁O₂₁Si: 1339.7387; Found: 1339.7383.

3-Hydroxy-5-[(E)-2-(4-hydroxyphenyl)ethenyl]phenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (83).

To a solution of (79) (145 mg, 0.152 mmol) in dry tetrahydrofuran (20 mL) was added tetra-n-butylammonium fluoride (0.456 mL, 0.456 mmol) at 0°C. The reaction was kept at 0°C for 20 min, and then allowed to attain ambient temperature. The reaction mixture was then quenched with saturated sodium bicarbonate (15 ml) and extracted with ethyl acetate (3 × 20 ml). The combined organic extracts were dried over magnesium sulphate, filtered and the volatiles removed in vacuo. Column chromatography (increasing polarity from 5% to 35% ethyl acetate in petroleum spirit as eluant) yielded the title compound (83) as a white solid (155 mg, 85%). M.p. 123-127, (dec.). [α]D²⁵ = -19.8° (c = 0.96, ethanol). IR cm⁻¹: 2978, 1741, 1597, 1517, 1482. ¹H NMR (600 MHz, CDCl₃): δ 7.35 (2H, H-2’ and H-6’, d, J = 8.4 Hz), 6.93 (1H, H-8, d, J = 15.6 Hz), 6.83 (2H, H-3’ and H-5’, d, J = 8.4 Hz), 6.77 (1H, H-7, d, J = 15.6 Hz), 6.66 and 6.62 (2H, H-2 or H-6, 2s), 6.40 (1H, H-4, s), 5.40 (1H, H_{glu}-3, t, J = 9.6 Hz), 5.29 (1H, H_{glu}-2, dd, J = 9.6, 7.8 Hz), 5.21 (1H, H_{glu}-4, t, J = 9.6 Hz), 5.04 (1H, H_{glu}-1, d, J = 7.8 Hz), 4.27 (1H, H_{glu}-6a, d, J = 12.0 Hz), 4.10 (1H, H_{glu}-6b, dd, J = 12.0, 6.0 Hz), 3.86 (1H, H_{glu}-5, dd, J = 9.6, 6.0 Hz), 1.19, 1.17, 1.15, and
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1.14 (36H, (C=O)(CH₃)₃, 4s). ¹³C NMR (75 MHz, CDCl₃): δ 178.4, 176.8, 176.7 and 176.2 (C=O), 158.0, 156.8 and 155.5 (C-3, C-5 or C-4’), 140.0 (C-1), 129.4 (C-1’), 129.0 (C-8), 127.8 (C-2’ and C-6’), 125.5 (C-7), 115.5 (C-3’ and C-5’), 107.7 and 107.1 (C-2 or C-6), 103.1 (C-4), 99.0 (C glu-1), 72.3, (C glu-5), 71.9 (C glu-3), 71.2 (C glu-2), 68.0 (C glu-4), 62.3 (C glu-6), 39.1 (C(CH₃)₃), 27.43, 27.40, 27.36 and 27.33 (C(CH₃)₃). HRMS (ESI⁺) calcd. for (M⁺+H) C₄₀H₅₅O₁₂: 727.3694; Found: 727.3688.

4-[(E)-2-(3,5-Dihydroxyphenyl)ethenyl]phenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (84).

The title compound was prepared from (80) (145 mg, 0.152 mmol), tetrahydrofuran (20 mL) and tetra-n-butylammonium fluoride (0.456 mL, 0.456 mmol) using the method described above for the synthesis of (83). The residue was then subjected to column chromatography (increasing polarity from 5% to 35% ethyl acetate in petroleum spirit as eluant) to furnish the title compound (84) as a white solid (91 mg, 82%). M.p. 125-128 °C, (dec.). [α]D²⁵ = -11.0° (c = 0.63, ethanol). IR cm⁻¹ 2976, 1740, 1598, 1482. ¹H NMR (600 MHz, CDCl₃): δ 7.29 (2H, H-2’ and H-6’, d, J = 9.0 Hz), 6.92 (1H, H-8, d, J = 15.6 Hz), 6.91 (2H, H-3’ and H-5’, d, J = 9.0 Hz), 6.76 (1H, H-7, d, J = 15.6 Hz), 6.56 (2H, H-2 and H-6, s), 6.32 (1H, H-4, s), 5.39 (1H, H glu-3’’, t, J = 9.6 Hz), 5.29 (1H, H glu-2’’, dd, J = 9.6, 7.8 Hz), 5.15 (1H, H glu-4’’, t, J = 9.6 Hz), 4.96 (1H, H glu-1’’, d, J = 7.8 Hz), 4.28 (1H, H glu-6a’’, dd, J = 12.6 1.2 Hz), 4.03 (1H, H glu-6b’’, dd, J = 12.0, 7.2 Hz), 3.81 (1H, H glu-5’’, ddd, J = 9.6, 7.2, 1.2 Hz), 1.23, 1.19, 1.15 and 1.14 (36H, (C=O)(CH₃)₃, 4s). ¹³C NMR (75 MHz, CDCl₃): δ 178.9, 178.0 and 177.0 (C=O), one (C=O) masked, 157.0 and 156.7 (C-3, C-5 or C-4’), 140.1 (C-1), 132.5 (C-1’), 128.6 (C-8), 127.9 (C-2’ and C-6’), 127.5 (C-7), 117.1 (C-3’ and C-5’), 106.2 (C-2 and C-6), 103.4 (C-4), 99.5 (C glu-1’’), 72.6, (C glu-5’’), 72.2 (C glu-3’’), 71.1 (C glu-2’’), 68.2 (C glu-4’’), 62.5 (C glu-6’’), 39.2
(C(CH₃)₃), 27.39, 27.33, 27.32 and 27.30, (C(CH₃)₃). HRMS (ESI⁺) calcd. for (M⁺+H) C₄₀H₅₅O₁₂: 727.3694; Found: 727.3688.


The title compound was prepared from (81) (300 mg, 0.224 mmol), tetrahydrofuran (20 mL) and tetra-n-butyrammonium fluoride (0.448 mL, 0.448 mmol) using the method described above for the synthesis of (83). Column chromatography (increasing polarity from 5% to 30% ethyl acetate in petroleum spirit as eluant) yielded the title compound (85) as a white solid (247 mg, 90%). M.p. 142-145 °C, decompose. [α]D²⁵ = -24.8° (c = 1.01, ethanol). IR cm⁻¹ 2979, 1740, 1600, 1482. ¹H NMR (600 MHz, CDCl₃): δ 7.37 (2H, H-2' and H-6', d, J = 7.8 Hz), 6.97 (2H, H-3' and H-5', d, J = 7.8 Hz), 6.95 (1H, H-8, d, J = 16.2 Hz), 6.82 (1H, H-7, d, J = 16.2 Hz), 6.68 and 6.63 (2H, H-2 or H-6, 2s), 6.43 (1H, H-4, s), 5.42 (1H, H_glu-3 or H_glu-3''', t, J = 9.6 Hz), 5.41 (1H, H_glu-3 or H_glu-3''', t, J = 9.6 Hz), 5.31 (1H, H_glu-2 or H_glu-2''', dd, J = 9.6, 8.4 Hz), 5.29 (1H, H_glu-2 or H_glu-2''', dd, J = 9.6, 8.4 Hz), 5.20 (1H, H_glu-4 or H_glu-4''', t, J = 9.6 Hz), 5.18 (1H, H_glu-4 or H_glu-4''', t, J = 9.6 Hz), 5.11 (1H, H_glu-1 or H_glu-1''', d, J = 8.4 Hz), 5.08 (1H, H_glu-1 or H_glu-1''', d, J = 8.4 Hz), 4.28 (1H, H_glu-6a or H_glu-6a''', d, J = 12.0 Hz), 4.26 (1H, H_glu-6a or H_glu-6a''', d, J = 12.0 Hz), 4.11 (1H, H_glu-6b or H_glu-6b''', dd, J = 11.4, 6.0 Hz), 4.08 (1H, H_glu-6b or H_glu-6b''', dd, J = 11.4, 6.0 Hz), 3.90 (2H, H_glu-5 and H_glu-5'''), 1.24, 1.21, 1.19, 1.18, 1.17, 1.16, 1.15, 1.14 (72H, (C=O)C(CH₃)₃, 8s). ¹³C NMR (75 MHz, CDCl₃): δ 178.6, 178.4,
177.2, 176.9, 176.8 (C=O), three (C=O) masked, 158.5, 157.5 and 157.1 (C-3, C-5 or C-4’), 140.1 (C-1), 132.4 (C-1’), 129.1 (C-8), 128.1 (C-2’ and C-6’), 127.4 (C-7), 117.2 (C-3’ and C-5’), 108.3 and 107.7 (C-2 or C-6), 103.8 (C-4), 99.7 and 99.5 (C_{glu}-1 or C_{glu}-1’), 73.0 and 72.9 (C_{glu}-5 or C_{glu}-5’), 72.3 (C_{glu}-3 and C_{glu}-3’), 68.2 (C_{glu}-4 and C_{glu}-4’), 62.5 (C_{glu}-6 and C_{glu}-6’), 39.1, 39.0 (C\(_{(CH_3)_3}\)), 27.41, 27.35 and 27.32 (C\(_{(CH_3)_3}\)), five (C\(_{(CH_3)_3}\)), masked. HRMS (ESI\(^+\)) calcd. for (M\(^+\)+H) C\(_{66}H_{97}O_{21}\): 1225.6522; Found: 1225.6517.

3-[(E)-2-(4-Hydroxyphenyl)ethenyl]-5-[[2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-\(\beta\)-d-glucopyranosyl]oxy]phenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-\(\beta\)-d-glucopyranoside (86).

The title compound was prepared from (82) (350 mg, 0.261 mmol), tetrahydrofuran (20 mL) and tetra-n-butyrammonium fluoride (0.522 mL, 0.522 mmol) using the method described above for the synthesis of (83). Column chromatography (increasing polarity from 5% to 30% ethyl acetate in petroleum spirit as eluant) yielded the title compound (86) as a white solid (281 mg, 88%). M.p. 123-127 °C. [\(\alpha\)]\(_D\)\(^{25}\) = -11.2° (c = 0.53, ethanol). IR \(cm^{-1}\) 2976, 1740, 1605, 1518, 1482. \(^1\)H NMR (600 MHz, CDCl\(_3\)): \(\delta\) 7.37 (2H, H-2’ and H-6’, d, \(J = 8.4 \text{ Hz}\)), 6.94 (1H, H-8, d, \(J = 15.6 \text{ Hz}\)), 6.82 (2H, H-3’ and H-5’, d, \(J = 8.4 \text{ Hz}\)), 6.80 (1H, H-7, d, \(J = 15.6 \text{ Hz}\)), 6.78 (2H, H-2 and H-6, s), 6.44 (1H, H-4, s), 5.41 (2H, H_{glu}-3 and H_{glu}-3’, t, \(J = 9.6 \text{ Hz}\)), 5.27 (2H, H_{glu}-2 and H_{glu}-2’, dd, \(J = 9.6, 8.4 \text{ Hz}\)), 5.20 (2H, H_{glu}-4 and H_{glu}-4’, t, \(J = 9.6 \text{ Hz}\)), 5.14 (2H, H_{glu}-1 and H_{glu}-1’, d, \(J = 8.4 \text{ Hz}\)), 4.23 (2H, H_{glu}-6a and H_{glu}-6a’, d, \(J = 12.0 \text{ Hz}\)), 4.11 (2H, H_{glu}-6b and H_{glu}-6b’, dd, J = 12.0, 6.0 Hz), 3.92 (2H, H_{glu}-5 and H_{glu}-5’, dd, \(J = 9.6, 6.0 \text{ Hz}\)), 1.17, 1.16, 1.15 and 1.14...
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(72H, (C=O)C(CH₃)₃, 4s). ¹³C NMR (75 MHz, CDCl₃): δ 178.5, 177.6, 176.9 and 176.8 (C=O), 158.2 and 156.7 (C-3, and C-5 or C-4’), 140.7 (C-1), 130.2 (C-1’), 129.3 (C-8), 128.4 (C-2’ and C-6’), 125.5 (C-7), 116.0 (C-3’ and C-5’), 109.2 (C-2 and C-6), 105.2 (C-4), 99.2 (C gluc-1 and C gluc-1’), 72.8 (C gluc-5 and C gluc-5’), 72.0 (C gluc-3 and C gluc-3’), 71.9 (C gluc-2 and C gluc-2’), 68.0 (C gluc-4 and C gluc-4’), 62.2 (C gluc-6 and C gluc-6’), 39.1 and 39.0 (C(CH₃)₃), 27.46, 27.40, 27.36 and 27.30 (C(C(CH₃)₃)). HRMS (ESI⁺) calcd. for (M⁺+H) C₆₆H₉₇O₂₁: 1225.6522; Found: 1225.6517.


Sodium metal (55 mg, 2.39 mmol) was dissolved in methanol (10 mL) to form sodium methoxide and then compound (83) (100 mg, 0.138 mmol) was added. The reaction mixture was stirred at ambient temperature for 48 h, diluted with water (10 mL), neutralized with Dowex 50-X (H⁺) resin, filtered and concentrated. Column chromatography (8% methanol in ethyl acetate as eluant) yielded the title compound (18) as a white solid (51 mg, 95%). M.p. 225-228 °C, (dec.), [α]D²⁵ = -52.2° (c = 0.44, methanol), (Lit., M.p. 226-229 °C,¹⁰⁶ Lit., [α]D²⁵ = -64° (c = 0.32, methanol)).¹⁰⁶ ¹H NMR (600 MHz, CD₃OD): δ 7.36 (2H, H-2’ and H-6’, d, J = 7.8 Hz), 7.01 (1H, H-8, d, J = 16.2 Hz), 6.84 (1H, H-7, d, J = 16.2 Hz), 6.77 (2H, H-3’ and H-5’, d, J = 7.8 Hz), 6.79 and 6.62 (2H, H-2 or H-6, 2s), 6.46 (1H, H-4, s), 4.90 (1H, H gluc-1, d, J = 7.2 Hz), 3.93 (1H, H gluc-6a, d, J = 12.0 Hz), 3.72 (1H, H gluc-6b, dd, J = 12.0, 6.0 Hz), 3.50-3.44 (3H, H gluc-2, H gluc-3 and H gluc-5, m), 3.40 (1H, H gluc-4, t, J = 9.0 Hz). ¹³C NMR (150 MHz, CD₃OD): δ 160.9, 160.0 and 158.9 (C-3, C-5 or C-4’), 141.9 (C-1), 130.8 (C-1’), 130.5 (C-8), 129.4 (C-2’ and C-6’), 127.2 (C-7), 117.0 (C-3’ and C-5’), 108.9, 107.5 (C-2 and
C-6)), 104.6 (C-4), 102.9 (C$_{glu}$-1), 78.7, (C$_{glu}$-5), 78.5 (C$_{glu}$-3), 75.4 (C$_{glu}$-2), 72.0 (C$_{glu}$-4), 63.1 (C$_{glu}$-6). ESI-MS m/z: 391 [M+H]$^+$. All other physical and spectroscopic details were in accordance with those previously reported.\textsuperscript{39, 103, 106, 108, 115, 116}


The title compound was prepared from compound (84) (80 mg, 0.110 mmol), methanol (10 mL) and sodium metal (60 mg, 2.60 mmol) using the method described above for the synthesis of (18). Column chromatography (8% methanol in ethyl acetate as eluant) yielded the title compound (56) as a white solid (41 mg, 95%). M.p. 207-212 °C. [\(\alpha\)\textsubscript{D}\]\textsuperscript{25} = -42.3° (c = 0.42, methanol), (Lit., M.p. 210-215 °C,\textsuperscript{106} Lit., [\(\alpha\)\textsubscript{D}\]\textsuperscript{25} = -73.9° (c = 0.39, methanol).\textsuperscript{106} \textsuperscript{1}H NMR (600 MHz, CD$_3$OD): \(\delta\) 7.45 (2H, H-2’ and H-6’, d, \(J = 7.8\) Hz), 7.06 (2H, H-3’ and H-5’, d, \(J = 7.8\) Hz), 6.99 (1H, H-8, d, \(J = 16.2\) Hz), 6.87 (1H, H-7, d, \(J = 16.2\) Hz), 6.46 (2H, H-2 and H-6, s), 6.18 (1H, H-4, s), 4.92 (1H, H$_{glu}$-1’’, d, \(J = 6.6\) Hz), 3.90 (1H, H$_{glu}$-6a’’, d, \(J = 12.0\) Hz), 3.71 (1H, H$_{glu}$-6b’’, dd, \(J = 12.0, 6.0\) Hz), 3.48-3.42 (3H, H$_{glu}$-2’’, H$_{glu}$-3’’ and H$_{glu}$-5’’, m), 3.42 (1H, H$_{glu}$-4’’, t, \(J = 9.0\) Hz). \textsuperscript{13}C NMR (150 MHz, CD$_3$OD): \(\delta\) 160.2 and 159.2 (C-3, C-5 or C-4’), 141.5 (C-1), 133.7 (C-1’), 129.4 (C-8), 129.07 (C-2’ and C-6’), 127.05 (C-7), 118.4 (C-3’ and C-5’), 106.4 (C-2 and C-6), 103.5 (C-4), 102.8 (C$_{glu}$-1’’), 78.7, (C$_{glu}$-5’’), 78.5 (C$_{glu}$-3’’), 75.4 (C$_{glu}$-2’’), 71.9 (C$_{glu}$-4’’), 63.0 (C$_{glu}$-6’’). ESI-MS m/z: 391 [M+H]$^+$. All other physical and spectroscopic details were in accordance with those previously reported.\textsuperscript{103, 106, 115, 116}
4-{(E)-2-[3-(β-D-Glucopyranosyloxy)-5-hydroxyphenyl]ethenyl}phenyl-β-D-glucopyranoside (73).

The title compound was prepared from (85) (128 mg, 0.104 mmol), methanol (10 mL) and sodium metal (85 mg, 3.70 mmol) using the method described above for the synthesis of compound (18). Column chromatography (15% methanol in ethyl acetate as eluant) yielded the title compound (73) as a white solid (53 mg, 97%). M.p. 200-204 °C, (dec.). [α]_{D}^{25} = -54.4° (c =1.01, methanol), Lit., [α]_{D}^{25} = -72.5° (c = 1.03, methanol).

1H NMR (600 MHz, CD_{3}OD): δ 7.45 (2H, H-2′ and H-6′, d, J = 9.0 Hz), 7.07 (2H, H-3′ and H-5′, d, J = 9.0 Hz), 7.05 (1H, H-8, d, J = 16.8 Hz), 6.91 (1H, H-7, d, J = 16.8 Hz), 6.80 and 6.65 (2H, H-2 or H-6, 2s), 6.48 (1H, H-4, s), 4.93 (1H, H_{glu}-1 or H_{glu}-1′′, d, J = 7.8 Hz), 4.91 (1H, H_{glu}-1 or H_{glu}-1′′, d, J = 7.2 Hz), 3.92 (1H, H_{glu}-6a or H_{glu}-6a′′, d, J = 12.0 Hz), 3.90 (1H, H_{glu}-6a or H_{glu}-6a′′, d, J = 12.0 Hz), 3.71 (2H, H_{glu}-6b and H_{glu}-6b′′, m), 3.51-3.40 (8H, H_{glu}-2, H_{glu}-3, H_{glu}-4, H_{glu}-5, H_{glu}-2′′, H_{glu}-3′′, H_{glu}-4′′ and H_{glu}-5′′, m).

13C NMR (150 MHz, CD_{3}OD): δ 160.9, 160.0 and 159.1 (C-3, C-5 or C-4′), 141.6 (C-1), 133.6 (C-1′), 130.0 (C-8), 129.2 (C-2′ and C-6′), 128.6 (C-7), 118.4 (C-3′ and C-5′), 109.0, 107.7 (C-2 and C-6), 104.9 (C-4), 102.8 and 102.6 (C_{glu}-1 or C_{glu}-1′), 78.6 and 78.5 (C_{glu}-5 or C_{glu}-5′), 78.4 and 78.3 (C_{glu}-3 or C_{glu}-3′), 75.4 and 75.3 (C_{glu}-2 or C_{glu}-2′), 71.9 and 71.8 (C_{glu}-4 or C_{glu}-4′), 63.0 and 62.9 (C_{glu}-6 or C_{glu}-6′). ESI-MS m/z: 553 [M+H]^+. All other physical and spectroscopic details were in accordance with those previously reported.
3-([2,3,4,6-tetrakis-O-(2,2-Dimethylpropanoyl)]-β-D-glucopyranosyl)oxy]-5-[(E)-2-(4-hydroxyphenyl)ethenyl]phenyl-β-D-glucopyranoside (54).

The title compound was prepared from (86) (150 mg, 0.122 mmol), methanol (10 mL) and sodium metal (90 mg, 3.91 mmol) using the method described above for the synthesis of compound (18). Column chromatography (15% methanol in ethyl acetate as eluant) yielded the title compound (54) as a white solid (63 mg, 99%). M.p. 165-168 °C, (dec.). [α]_D^{25} = -69.6° (c = 0.16, methanol), Lit., [α]_D^{25} = -71.5° (c = 1.01, methanol).

1H NMR (600 MHz, CD_3OD): δ 7.37 (2H, H-2’ and H-6’, d, J = 8.4 Hz), 7.06 (1H, H-8, d, J = 15.6 Hz), 6.94 (2H, H-2 and H-6, s), 6.88 (1H, H-7, d, J = 15.6 Hz), 6.76 (2H, H-3’ and H-5’, d, J = 8.4 Hz), 6.75 (1H, H-4, s), 4.94 (2H, H_glu-1 and H_glu-1’, d, J = 6.6 Hz), 3.93 (2H, H_glu-6a and H_glu-6a’, d, J = 11.4 Hz), 3.69 (2H, H_glu-6b and H_glu-6b’, d, J = 6.6 Hz), 3.53 (2H, H_glu-5 and H_glu-5’, d, J = 9.0, 6.6 Hz), 3.40-3.45 (4H, H_glu-2, H_glu-2’, H_glu-3 and H_glu-3’, m), 3.36 (2H, H_glu-4 and H_glu-4’, t, J = 9.0 Hz). 13C NMR (150 MHz, CD_3OD): δ 160.6 and 159.0 (C-3, C-5 or C-4’), 141.9 (C-1), 131.0 (C-1’), 130.7 (C-8), 129.5 (C-2’ and C-6’), 126.7 (C-7), 117.0 (C-3’ and C-5’), 110.2 (C-2 and C-6), 105.3 (C-4), 102.7 (C_glu-1), 78.7, (C_glu-5), 78.5 (C_glu-3), 75.4 (C_glu-2), 72.0 (C_glu-4), 63.2 (C_glu-6).

ESI-MS m/z: 553 [M+H]^+. All other physical and spectroscopic details were in accordance with those previously reported.

3-([2,3,4,6-tetrakis-O-(2,2-Dimethylpropanoyl)]-β-D-glucopyranosyl)oxy]-5-[(E)-2-(4-([2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)]-β-D-glucopyranosyl)oxy]phenyl)ethenyl]phenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (87).
The title compound was prepared from compound (86) (200 mg, 0.163 mmol), boron trifluoride etherate (0.09 mL, 0.716 mmol), dichloromethane (20 mL) and the Kunz reagent (113 mg, 0.178 mmol) using the method described above for the synthesis of (79). Recrystallisation from methanol yielded the title compound (87) as a white solid (197 mg, 70%). M.p. 215-217 °C. [α]D^25 = -38.8° (c = 0.59, ethanol). IR cm⁻¹ 2975, 1735, 1598, 1482. ^1H NMR (600 MHz, CDCl₃): δ 7.37 (2H, H-2’ and H-6’, d, J = 8.4 Hz), 6.97 (2H, H-3’ and H-5’, d, J = 8.4 Hz), 6.95 (1H, H-8, d, J = 16.2 Hz), 6.83 (1H, H-7, d, J = 16.2 Hz), 6.78 (2H, H-2 and H-6, s), 6.46 (1H, H-4, s), 5.43 (1H, H-glu-3’’, t, J = 9.6 Hz), 5.41 (2H, H-glu-3 and H-glu-3’, t, J = 9.6 Hz), 5.31 (1H, H-glu-2’’, dd, J = 9.6, 7.8 Hz), 5.26 (2H, H-glu-2 and H-glu-2’, dd, J = 9.6, 7.8 Hz), 5.20 (2H, H-glu-4 and H-glu-4’, t, J = 9.6 Hz), 5.18 (1H, H-glu-4’’, t, J = 9.6 Hz), 5.14 (2H, H-glu-1 and H-glu-1’, d, J = 7.8 Hz), 5.09 (1H, H-glu-1’’, d, J = 7.8 Hz), 4.29 (1H, H-glu-6a’’, d, J = 12.0 Hz), 4.23 (2H, H-glu-6a and H-glu-6a’, d, J = 12.0 Hz), 4.10 (2H, H-glu-6b and H-glu-6b’, dd, J = 12.0, 5.4 Hz), 4.08 (1H, H-glu-6b’’, dd, J = 12.0, 5.4 Hz), 3.92 (3H, H-glu-5, H-glu-5’ and H-glu-5’’, m), 1.18, 1.17, 1.16, 1.15, 1.14 (108H, (C=O)(CH₃)₃, 5s, 3s masked). ^13C NMR (75 MHz, CDCl₃): δ 178.3, 177.5, 176.8, 176.7 (C=O), three (C=O) masked), 158.3 and 157.2 (C-3, C-5 or C-4’), 140.2 (C-1), 132.2 (C-1’), 129.7 (C-8), 128.1 (C-2’ and C-6’), 127.1 (C-7), 117.2 (C-3’ and C-5’), 109.4 (C-2 and C-6), 105.6 (C-4), 99.7 and 99.2 (C-glu-1, C-glu-1’ or C-glu-1’’), 73.0 and 72.9 (C-glu-5, C-glu-5’ or C-glu-5’’), 72.3 (C-glu-3, C-glu-3’ and C-glu-3’’), 71.2 and 71.1 (C-glu-2, C-glu-2’ or C-glu-2’’), 68.2 and 68.0 (C-glu-4, C-glu-4’ or C-glu-4’’), 62.5 and 62.2 (C-glu-6, C-glu-6’ or C-glu-6’’), 39.1, 39.0, (C(CH₃)₃), 27.41, 27.37, 27.32 (C(CH₃)₃, five (C(CH₃)₃ masked). HRMS (ESI⁺) calcd. for [M+H]^+ C₆₆H₇₇O₂₁: 1723.9351; Found: 1723.9346.
4-{(E)-2-[3,5-bis(β-D-Glucopyranosyloxy)phenyl]ethenyl}phenyl-β-D-glucopyranoside (88).

The title compound was prepared from (87) (135 mg, 0.078 mmol), methanol (10 mL) and sodium metal (100 mg, 4.35 mmol) using the method described above for the synthesis of compound (18). Column chromatography (20% methanol in ethyl acetate as eluant) yielded the title compound (87) as a white solid (52 mg, 94%). M.p. 215-220°C, (dec.). [α]D25 = -79.4° (c = 0.84, methanol). 1H NMR (600 MHz, CD3OD): δ 7.46 (2H, H-2' and H-6', d, J = 9.0 Hz), 7.09 (1H, H-8, d, J = 16.8 Hz), 7.07 (2H, H-3' and H-5', d, J = 9.0 Hz), 6.96 (1H, H-7, d, J = 16.8 Hz), 6.96 (2H, H-2 and H-6, s), 6.76 (1H, H-4, s), 4.94 (2H, Hglu-1 and Hglu-1', d, J = 7.8 Hz), 4.92 (1H, Hglu-1'', d, J = 7.8 Hz), 3.93 (2H, Hglu-6a or Hglu-6a’, d, J = 11.4 Hz), 3.88 (1H, Hglu-6a’’, d, J = 12.0 Hz), 3.68 (3H, Hglu-6b, Hglu-6b’ and Hglu-6b’’, m), 3.56-3.44 (9H, Hglu-2, Hglu-3, Hglu-5, Hglu-2’, Hglu-3’, Hglu-5’, Hglu-2’’, Hglu-3’’ and Hglu-5’’, m), 3.41 (1H, Hglu-4’’, d, J = 9.0 Hz), 3.36 (2H, Hglu-4 and Hglu-4’, d, J = 9.0 Hz). 13C NMR (150 MHz, CD3OD): δ 160.6 and 159.3 (C-3, C-5 and C-4’), 141.6 (C-1), 133.4 (C-1’), 130.5 (C-8), 129.3 (C-2’ and C-6’), 128.2 (C-7), 118.4 (C-3’ and C-5’), 110.4 (C-2 and C-6), 105.7 (C-4), 102.7 and 102.6 (Cglu-1, Cglu-1’ or Cglu-1’’), 78.7 and 78.6 (Cglu-5, Cglu-5’ or Cglu-5’’), 78.41 and 78.38 (Cglu-3, Cglu-3’ or Cglu-3’’), 75.4 (Cglu-2, Cglu-2’ and Cglu-2’’), 72.1 and 71.8 (Cglu-4, Cglu-4’ or Cglu-4’’), 63.1 and 63.0 (Cglu-6, Cglu-6’ or Cglu-6’’). ESI m/z 736.9 [M + Na+].
**HPLC Analysis of the cis/trans-Resveratrol Glucosides.**

HPLC analyses were performed using a Hewlett-Packard (HP) 1100 gradient liquid chromatograph with a diode array detector coupled to Chem Station HP 79995A software following modified conditions reported by Maria et al.\textsuperscript{105} A SS Wacosil C18 reverse phase column (250 x 4.5 mm, 5 pm particle size) with a guard column was used at 30°C. Individual samples or mixtures of glucosides (25 µL) were injected and the flow rate was controlled at 0.5 mL/min. Two solvents were used for the separation: Solution A: 1M acetic acid in Milli-Q-water; Solution B: 20% of Solution A and 80% acetonitrile HPLC grade. The eluent was monitored at 306 nm and the elution profile was as follows: 0 min, 100% A, 0% B; 10 min, 100% A, 0% B; 20 min, 90% A, 10% B; 30 min, 80% A, 20% B; 40 min, 60% A, 40% B; 45 min, 0% A, 100% B; 50 min, 90% A, 10% B; 52 min, 100% A, 0% B; 56 min, 100% A, 0% B.

**(Z)-5-(4-hydroxy styryl)benzene-1,3-diol (1b).** **Common name: cis-Resveratrol.**

\[
\begin{align*}
\text{HO} & \quad \text{5} \\
\text{4} & \quad \text{1} \\
\text{3} & \quad \text{2} \\
\text{2'} & \quad \text{1'} \\
\text{3'} & \quad \text{4'} \\
\text{6} & \quad \text{8} \\
\text{7} & \quad \text{6'} \\
\text{5} & \quad \text{OH}
\end{align*}
\]

(1b)

A solution of (1a) (10 mg) in \(d_4\)-MeOH (0.5 mL) was irradiated under UV light (365 nm) for 40 min. The cis/trans-isomerisation was then analysed by \(^1\)H NMR spectroscopy (600 MHz) indicating that the cis-isomer (1b) was afforded in 90% yield. \(^1\)H NMR (300 MHz, \(d_4\)-MeOH): \(\delta\) 7.12 (2H, H-2' and H-6'), d, \(J = 8.4\) Hz), 6.70 (2H, H-3' and H-5', d, \(J = 8.4\) Hz), 6.42 (1H, H-8, d, \(J = 12.0\) Hz), 6.33 (1H, H-7, d, \(J = 12.0\) Hz), 6.27 (2H, H-2 and H-6, dd, \(J = 2.1, 2.1\) Hz), 6.20 (1H, H-4, dd, \(J = 2.1, 2.1\) Hz). All other physical and spectroscopic details were in accordance with those previously reported.\textsuperscript{39}
3-Hydroxy-5-[(Z)-2-(4-hydroxyphenyl)ethenyl]phenyl-β-D-glucopyranoside \((89)\).
Common name: *cis*-piceid.

![Structure of 3-Hydroxy-5-[(Z)-2-(4-hydroxyphenyl)ethenyl]phenyl-β-D-glucopyranoside](image)

The title compound \((89)\) was prepared in the same manner as \((1b)\) above from \((18)\). The *trans/cis* isomerisation was then analysed by \(^1\)H NMR spectroscopy (600 MHz) indicating that the *cis*-isomer \((89)\) was afforded in 92\% yield. \(^1\)H NMR (600 MHz, \(d_4\)-MeOH): \(\delta\)
7.08 (2H, H-2’ and H-6’, d, \(J = 9.0\) Hz), 6.65 (2H, H-3’ and H-5’, d, \(J = 9.0\) Hz), 6.51 and 6.39 (2H, H-2 or H-6, 2s), 6.46 (1H, H-8, d, \(J = 12.0\) Hz), 6.38 (1H, H-4, s), 6.34 (1H, H-7, d, \(J = 12.0\) Hz), 4.69 (1H, H\(_{\text{glu}}\)-1, d, \(J = 7.8\) Hz), 3.76 (1H, H\(_{\text{glu}}\)-6a, dd, \(J = 12.0, 1.8\) Hz), 3.70 (1H, H\(_{\text{glu}}\)-6b, dd, \(J = 12.0, 5.4\) Hz), 3.38-3.34 (4H, H\(_{\text{glu}}\)-2, H\(_{\text{glu}}\)-3, H\(_{\text{glu}}\)-4 and H\(_{\text{glu}}\)-5, m). \(^{13}\)C NMR (150 MHz, \(d_4\)-MeOH): 160.6, 159.8 and 158.2 (C-3, C-5 or C-4’), 141.6 (C-1), 131.9 (C-2’ and C-6’), 131.8 (C-1’), 130.4 (C-8), 129.6 (C-7), 116.5 (C-3’ and C-5’), 111.3 and 109.8 (C-2 or C-6), 104.6 (C-4), 102.8 (C\(_{\text{glu}}\)-1), 78.4, (C\(_{\text{glu}}\)-5), 78.3 (C\(_{\text{glu}}\)-3), 75.3 (C\(_{\text{glu}}\)-2), 71.5 (C\(_{\text{glu}}\)-4), 62.7 (C\(_{\text{glu}}\)-6). ESI-MS m/z: 391 [M+H]\(^+\). All other physical and spectroscopic details were in accordance with those previously reported.\(^{39}\)


![Structure of 4-[(Z)-2-(3,5-Dihydroxyphenyl)ethenyl]phenyl-β-D-glucopyranoside](image)
The title compound (90) was prepared in the same manner as (1b) above from (56). The trans/cis isomerisation was then analysed by \(^1\)H NMR spectroscopy (600 MHz) indicating that the cis-isomer (90) was afforded in 95% yield. \(^1\)H NMR (600 MHz, \(d_4\)-MeOH): \(\delta\) 7.13 (2H, H-2’ and H-6’, d, \(J = 8.4\) Hz), 6.94 (2H, H-3’ and H-5’, d, \(J = 8.4\) Hz), 6.44 (1H, H-8, d, \(J = 12.6\) Hz), 6.39 (1H, H-7, d, \(J = 12.6\) Hz), 6.18 (2H, H-2 and H-6, dd, \(J = 1.8\) Hz), 6.11 (1H, H-4, dd, \(J = 1.8, 1.8\) Hz), 4.86 (1H, H\(_{\text{glu}}\)-1’’, d, \(J = 6.6\) Hz), 3.87 (1H, H\(_{\text{glu}}\)-6a’’, dd, \(J = 12.0, 1.8\) Hz), 3.68 (1H, H\(_{\text{glu}}\)-6b’’, dd, \(J = 12.0, 6.0\) Hz), 3.48-3.34 (4H, H\(_{\text{glu}}\)-2’’, H\(_{\text{glu}}\)-3’’, H\(_{\text{glu}}\)-4’’ and H\(_{\text{glu}}\)-5’’), m. \(^13\)C NMR (150 MHz, \(d_4\)-MeOH): 160.6 and 158.6 (C-3, C-5 or C-4’’), 141.4 (C-1), 133.2 (C-1’’), 131.7 (C-2’’ and C-6’’), 130.98 and 130.93 (C-8 or C-7), 117.8 (C-3’’ and C-5’’), 108.7 (C-2 and C-6), 103.0 (C-4), 102.7 (C\(_{\text{glu}}\)-1’’), 78.6, (C\(_{\text{glu}}\)-5’’), 78.5 (C\(_{\text{glu}}\)-3’’), 75.4 (C\(_{\text{glu}}\)-2’’), 71.9 (C\(_{\text{glu}}\)-4’’), 63.0 (C\(_{\text{glu}}\)-6’’). ESI-MS m/z: 391 [M+H]\(^+\).


The title compound (91) was prepared in the same manner as (1b) above from (73). The trans/cis isomerisation was then analysed by \(^1\)H NMR spectroscopy (600 MHz) indicating that the cis-isomer (91) was afforded in 94% yield. \(^1\)H NMR (600 MHz, \(d_4\)-MeOH): \(\delta\) 7.17 (2H, H-2’ and H-6’, d, \(J = 9.0\) Hz), 6.96 (2H, H-3’ and H-5’, d, \(J = 9.0\) Hz), 6.50 (1H, H-8, d, \(J = 12.0\) Hz), 6.46 and 6.37 (2H, H-2 or H-6, 2s), 6.42 (1H, H-7, d, \(J = 12.0\) Hz), 6.39 (1H, H-4, dd, \(J = 1.8, 1.8\) Hz), 4.92 (1H, H\(_{\text{glu}}\)-1 or H\(_{\text{glu}}\)-1’’, d, \(J = 7.2\) Hz), 4.61 (1H, H\(_{\text{glu}}\)-1 or H\(_{\text{glu}}\)-1’’), d, \(J = 7.2\) Hz), 3.88 (1H, H\(_{\text{glu}}\)-6a’’, dd, \(J = 11.4, 2.4\) Hz), 3.74-3.67 (3H, H\(_{\text{glu}}\)-6a, H\(_{\text{glu}}\)-6b and H\(_{\text{glu}}\)-6b’’, m), 3.48-3.16 (8H, H-2, H-3, H-4, H-5, H-2’’, H-3’’, H-4’’...
and H-5”, m). $^{13}$C NMR (150 MHz, $d_4$-MeOH): 160.5, 159.8 and 158.6 (C-3, C-5 or C-4’), 141.1 (C-1), 133.3 (C-1’), 131.8 (C-2’ and C-6’), 131.4 (C-8), 130.8 (C-7), 118.1 (C-3’ and C-5’), 111.7 and 109.5 (C-2 or C-6), 104.6 (C-4), 102.8 and 102.6 (C$_{\text{glu}-1}$ or C$_{\text{glu}-1”}$), 78.5 and 78.1 (C$_{\text{glu}-5}$ and C$_{\text{glu}-5”}$ or C$_{\text{glu}-3}$ and C$_{\text{glu}-3”}$), 75.3 and 75.1 (C$_{\text{glu}-2}$ and C$_{\text{glu}-2”}$), 71.8 and 71.5 (C$_{\text{glu}-4}$ and C$_{\text{glu}-4”}$), 62.9, and 62.6 (C$_{\text{glu}-6}$ and C$_{\text{glu}-6”}$). ESI-MS m/z: 553 [M+H]$^+$. All other physical and spectroscopic details were in accordance with those previously reported.$^{117}$

3-($\beta$-d-Glucopyranosyloxy)-5-[(Z)-2-(4-hydroxyphenyl)ethenyl]phenyl-$\beta$-d-glucopyranoside (92).

The title compound (92) was prepared in the same manner as (1b) above from (54). The trans/cis isomerisation was then analysed by $^1$H NMR spectroscopy (600 MHz) indicating that the cis-isomer (92) was afforded in 92% yield. $^1$H NMR (600 MHz, $d_4$-MeOH): $\delta$ 7.07 (2H, H-2’ and H-6’, d, $J = 8.4$ Hz), 6.68-6.64 (5H, H-2, H-6, H-3’, H-5’ and H-4, m), 6.50 (1H, H-8, d, $J = 11.4$ Hz), 6.37 (1H, H-7, d, $J = 11.4$ Hz), 4.76 (2H, H$_{\text{glu}-1}$ and H$_{\text{glu}-1”}$, d, $J = 7.8$ Hz), 3.80 (2H, H$_{\text{glu}-6a}$ and H$_{\text{glu}-6a”}$, dd, $J = 11.4$, 1.8 Hz), 3.66 (2H, H$_{\text{glu}-6b}$ and H$_{\text{glu}-6b”}$, dd, $J = 11.4$, 6.6 Hz), 3.46-3.32 (H-2, H-3, H-4, H-5, H-2’, H-3’, H-4’ and H-5’, m). $^{13}$C NMR (150 MHz, $d_4$-MeOH): 160.6 and 160.2 (C-3, C-5 or C-4’), 141.4 (C-1), 132.3 (C-8), 131.9 (C-2’ and C-6’), 131.8 (C-1’), 129.3 (C-7), 116.7 (C-3’ and C-5’), 112.7 (C-2 and C-6), 105.4 (C-4), 102.6 (C$_{\text{glu}-1}$), 78.4, (C$_{\text{glu}-5}$), 78.3 (C$_{\text{glu}-3}$), 75.2 (C$_{\text{glu}-2}$), 71.7 (C$_{\text{glu}-4}$), 62.8 (C$_{\text{glu}-6}$). ESI-MS m/z: 553 [M+H]$^+$. All other physical and spectroscopic details were in accordance with those previously reported.$^{100}$

The title compound (93) was prepared in the same manner as (1b) above from (88). The trans/cis isomerisation was then analysed by $^1$H NMR spectroscopy (600 MHz) indicating that the cis-isomer (93) was afforded in 92% yield. $^1$H NMR (600 MHz, $d_4$-MeOH): $\delta$ 7.17 (2H, H-2' and H-6', d, $J = 8.4$ Hz), 6.98 (2H, H-3' and H-5', d, $J = 8.4$ Hz), 6.69 (1H, H-4, s), 6.65 (2H, H-2 and H-6, s), 6.56 (1H, H-8, d, $J = 12.6$ Hz), 6.47 (1H, H-7, d, $J = 12.6$ Hz), 4.93 (1H, $\text{H}_{\text{glu}}$-1'', d, $J = 7.8$ Hz), 4.73 (2H, $\text{H}_{\text{glu}}$-1 and $\text{H}_{\text{glu}}$-1', d, $J = 7.2$ Hz), 3.88 (1H, $\text{H}_{\text{glu}}$-6a'', dd, $J = 11.4$, 2.4 Hz), 3.77-3.67 (5H, $\text{H}_{\text{glu}}$-6a, $\text{H}_{\text{glu}}$-6a', $\text{H}_{\text{glu}}$-6b, $\text{H}_{\text{glu}}$-6b' and $\text{H}_{\text{glu}}$-6b'', m), 3.50-3.31 (12H, H-2, H-3, H-4, H-5, H-2', H-3', H-4', H-5', H-2'', H-3'', H-4'' and H-5'', m). $^{13}$C NMR (150 MHz, $d_4$-MeOH): 160.3 and 158.7 (C-3, C-5 or C-4'), 141.0 (C-1), 133.2 (C-1'), 131.9 (C-8), 131.8 (C-2' and C-6'), 130.5 (C-7), 117.9 (C-3' and C-5'), 112.7 (C-2 and C-6), 105.7 (C-4), 102.64 and 102.58 ($\text{C}_{\text{glu}}$-1, $\text{C}_{\text{glu}}$-1' or $\text{C}_{\text{glu}}$-1''), 78.65, 78.48, 78.38 and 78.28 ($\text{C}_{\text{glu}}$-5 and $\text{C}_{\text{glu}}$-5' or $\text{C}_{\text{glu}}$-5'' and $\text{C}_{\text{glu}}$-3 and $\text{C}_{\text{glu}}$-3' or $\text{C}_{\text{glu}}$-3''), 75.36 and 75.18 ($\text{C}_{\text{glu}}$-2 and $\text{C}_{\text{glu}}$-2' or $\text{C}_{\text{glu}}$-2''), 71.80 and 71.70 ($\text{C}_{\text{glu}}$-4 and $\text{C}_{\text{glu}}$-4' or $\text{C}_{\text{glu}}$-4''), 62.88 and 62.79 ($\text{C}_{\text{glu}}$-6 and $\text{C}_{\text{glu}}$-6' or $\text{C}_{\text{glu}}$-6''). ESI-MS m/z: 713 [M-H]$^+$. All other physical and spectroscopic details were in accordance with those previously reported.
Determination of the antioxidant capacities of cis/trans-resveratrol (1a/b) and their glucosides (18, 54, 56, 73, 88, 89-93) employing the FRAP method.

Sample Preparation: Standard solutions of trolox, cis/trans-resveratrol and all cis/trans-resveratrol glucosides were prepared by dissolving the substrate in methanol before the FRAP assay was conducted. Trolox (20 mg), cis/trans-resveratrol (20 mg) and all cis/trans-resveratrol glucosides (20 mg), were each separately dissolved in 25 mL of methanol and further diluted to afford a final concentration of 200 µM as standard stock solutions. All solutions were protected from light (wrapped in aluminum foil) at 4°C.

Calibration plots: A calibration curve for the FRAP assay was constructed for trolox using solutions in concentration ranges of 7.5-15 µM. A linear plot of absorbance at 593 nm vs concentration was constructed following the FRAP assay detailed below. Equation established: \( y = 0.045x + 0.021, r^2 = 1.000 \).

FRAP assay: FRAP assays were performed by the method developed by Benzie and Strain with slight modifications. The following FRAP reagents were prepared: 250 mL of 0.3 M acetate buffer (pH = 3.6), 50 mL of TPTZ (0.02 M) solution prepared by dissolution of TPTZ (156.25 mg) in 2.0 mL HCl (1M) and diluted with water and 10 mL of aqueous ferric chloride solution (0.02 M). Then 1.0 mL of each of the latter two mixtures were combined and further diluted with 34 mL of the acetate buffer and pre-warmed at 37°C. A volume of 3.0 mL of this reagent was mixed with a 0.3 mL of the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) in methanol, and made up to a final volume of 4 mL at a final concentration of 15 µM. The solution was shaken and incubated at 37°C in the dark for 45 minutes and the absorbance was then measured at 593 nm at ambient temperature, using a Cintra 40 UV-Vis spectrophotometer (Cintra, Australia). Reference samples were prepared by mixing 3.0 mL of the FRAP reagent with 1.0 mL of methanol and were used to zero the instrument before replacing with the actual sample. Single analyses from triplicate samples were performed at the indicated time points and expressed as trolox equivalents extracted from the calibration curves above.
Determination of the antioxidant capacities of cis/trans-resveratrol (1a/b) and their glucosides (18, 54, 56, 73, 88, 89-93) employing the DPPH method.

**Sample Preparation:** Standard solutions of trolox, cis/trans-resveratrol and all cis/trans-resveratrol glucosides were prepared by dissolving the substrate in methanol before the DPPH assay was conducted. Trolox (20 mg), cis/trans-resveratrol (20 mg) and all cis/trans-resveratrol glucosides (20 mg), were each separately dissolved in 25 mL of methanol and further diluted to afford a final concentration of 400 μM as standard stock solutions. All solutions were protected from light (wrapped in aluminum foil) at 4°C.

**Calibration plots:** A calibration curve for the DPPH assay was constructed for trolox using solutions in concentration ranges of 0-15 μM. A linear plot of absorbance at 515 nm vs concentration was constructed following the DPPH assay detailed below. Equation established: \( y \) (Trolox) = -0.024x + 0.456, \( r^2 = 1.000 \).

**DPPH assay:** DPPH assays were performed following previously described methods\(^{62,119}\) with slight modification. Fresh DPPH solutions (250 mL, 60 μM in methanol) were prepared and stored in the dark at 4°C. A volume of 3.0 mL of this reagent was mixed with 0.3 mL of the cis/trans-resveratrol analogues (1a/b, 18, 54, 56, 73, 88, 89-93) in methanol, and made up to a final volume of 4 mL at a final concentration of 15 μM. The solutions were kept at ambient temperature in the dark for 30 minutes and the absorbance was measured at 515 nm. Reference samples were pure methanol and used to zero the instrument before replacing with the actual sample. Single analyses from triplicate samples were performed at all indicated time points and expressed as trolox equivalents extracted from the calibration curve above.

5.3 Experimental for Chapter 3.

2-Iodoxybenzoic acid (129).
To a stirred solution of 2-iodobenzoic acid (138) (5 g, 0.020 mol) in water (80 mL) was added oxone (139) (20 g, 0.033 mol). The reaction mixture was kept at 70 °C for 3 h, cooled down to 0°C and left for another 1.5 h. The precipitated product in the solution was collected by filtration, washed with ice water and acetone and then dried under vacuum to afford pure iodobenzoic acid (129) (3.9 g) in 70% yield as white solid.

\[ \text{1H NMR (500 MHz, } d_6\text{-DMSO): } \delta 8.16 (1\text{H, d}, J = 7.5 \text{ Hz}), 8.02 (1\text{H, d}, J = 7.5, 7.5 \text{ Hz}), 7.97 (1\text{H, dd}, J = 7.5, 7.5 \text{ Hz}), 7.84 (1\text{H, dd}, J = 7.5, 7.5 \text{ Hz}). \]

\[ \text{13C NMR (125 MHz, } d_6\text{-DMSO): } \delta 168.5 \text{ (C = O), 147.4, 133.8 (ArC), 132.4, 131.3, 131.1, 125.9 (ArCH).} \]

4-[(E)-2-(3,5-Dihydroxyphenyl)ethenyl]benzene-1,2-diol (20).

To a solution of trans-resveratrol (1a) (101 mg, 0.443 mmol) in methanol (20 mL) at -20 °C was slowly added o-iodoxybenzoic acid (IBX) (129) (101 mg, 1.11 mmol) with stirring. The reaction was allowed to warm up to room temperature over 1 h. The reaction was then quenched with a solution of Na$_2$S$_2$O$_4$ (85% grade) (300 mg, 1.46 mmol) in water (40 mL) and then acidified to pH 4 with 1M hydrochloric acid. The organic were extracted with ethyl acetate (3 x 30 mL) and the combined extracts were dried over anhydrous sodium sulfate. The volatiles were removed in vacuo and the crude product purified by flash chromatography (increasing polarity from 10% to 40% ethyl acetate in petroleum spirit as eluant) to furnish the title compound (20) (35 mg, 32%) as a yellow solid.

\[ \text{1H NMR (500 MHz, } d_6\text{-acetone): } \delta 7.06 (1\text{H, H-2', s}), 6.93 (1\text{H, H-8, d}, J = 16.5 \text{ Hz}), 6.87 (1\text{H, H-6'}, d, J = 8.0 \text{ Hz}), 6.79 (1\text{H, H-5'}, d, J = 8.0 \text{ Hz}), 6.78 (1\text{H, H-7, d}, J = 16.5 \text{ Hz}), 6.51 (2\text{H, H-2 and H-6, s}), 6.24 (1\text{H, H-4, s}). \]

\[ \text{13C NMR (125 MHz, } d_6\text{-acetone): } \delta 159.6, 159.62 (\text{C-3 and C-5}), 146.22 (\text{C-3' and C-4'}), 140.7 (\text{C-1}), 130.5 (\text{C-1'}), 129.3 (\text{C-8}), 126.8 (\text{C-7}), 119.8 (\text{C-6'}), 116.2 (\text{C-5'}), 113.7 (\text{C-2'}), 105.5 (\text{C-2 and C-6}), 102.7 (\text{C-4}). \]

ESI-HRMS m/z: 245.0808 [M+H]$^+$, calcd. for C$_{28}$H$_{21}$O$_6$, 245.0812.
3-[(E)-2-(3,4-Dihydroxyphenyl)ethenyl]-5-hydroxyphenyl-2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (140).

To a solution of the resveratrol glycoside (83) (500 mg, 0.690 mmol) in methanol (50 mL) at -20 °C was slowly added α-iodoxybenzoic acid (IBX) (129) (291 mg, 1.04 mmol) with stirring. The mixture was allowed to warm up to room temperature over 1 h. The reaction was quenched with a solution of Na₂S₂O₄ (85% grade) (283 mg, 1.38 mmol) in water (150 mL) and then acidified to pH 4 with 1M hydrochloric acid. The organics were extracted with ethyl acetate (3 x 50 mL) and the combined ethyl acetate layers dried over anhydrous sodium sulfate. The volatiles were removed in vacuo and the crude product purified by flash chromatography (increasing polarity from 5% to 35% ethyl acetate in petroleum spirit as eluant) to afford the title product (140) (380 mg, 74%) as a yellow solid.

¹H NMR (500 MHz, d6-acetone): δ 7.08 (1H, H-2’, s), 7.01 (1H, H-8, d, J = 16.0 Hz), 6.91 (1H, H-6’, d, J = 9.0 Hz), 6.86 (1H, H-7, d, J = 16.0 Hz), 6.82 (1H, H-5’, d, J = 9.0 Hz), 6.75 (2H, H-2 and H-6, s), 6.46 (1H, H-4, s), 5.60 (1H, H_glu-1, d, J = 8.0 Hz), 5.49 (1H, H_glu-3, dd, J = 9.5, 9.5 Hz), 5.26-5.20 (2H, H_glu-2 and H_glu-4, m), 4.34-4.20 (3H, H_glu-5 and H_glu-6), 1.19, 1.17, 1.15, 1.14 (36H, 12 x CH₃, 4s). ¹³C NMR (125 MHz, d6-acetone): δ 178.12, 177.41, 177.03, 176.95 (C=O), 159.61, 159.57 (C-3 and C-5), 146.35, 146.22 (C-3’ and C-4’), 141.2 (C-1), 130.5, 130.0 (C-8 and C-1’), 126.3 (C-7), 120.1 (C-6’), 116.3 (C-5’), 114.0 (C-2’), 108.6, 106.3 (C-2 and C-6), 103.8 (C-4), 99.5 (C_glu-1), 73.11, 72.96 (C_glu-3 and C_glu-5), 72.0 (C_glu-2), 68.8 (C_glu-4), 62.7 (C_glu-6), 39.43, 39.40, 39.39, 39.31 (4 x C(CH₃)₃), 27.53, 27.49, 27.47, 27.44 (4 x C(CH₃)₃). ESI-HRMS m/z: 743.3637 [M+H]⁺, calcd. for C₂₈H₂₁O₆, 743.3633.
3-[(E)-2-(3,4-Dihydroxyphenyl)ethenyl]-5-hydroxyphenyl-β-D-glucopyranoside (32).

Sodium metal (40 mg, 1.74 mmol) was dissolved in methanol (10 mL) to form sodium methoxide and then (140) (26 mg, 0.035 mmol) was added. The reaction mixture was stirred at rt for 48 h under N\textsubscript{2} protection, diluted with water (10 mL), neutralised with Dowex 50-X (H\textsuperscript{+}) resin (300 mg), filtered, and concentrated. Column chromatography (10% methanol in ethyl acetate as eluant) yielded the title compound (32) as a white solid (5 mg, 35%).

\textsuperscript{1}H NMR (500 MHz, \textit{d}\textsubscript{4}-methanol): δ 7.02 (1H, H-2’, s), 6.97 (1H, H-8, d, \textit{J} = 16.5 Hz), 6.88 (1H, H-6’, d, \textit{J} = 8.0 Hz), 6.82 (1H, H-7, d, \textit{J} = 16.5 Hz), 6.80 (1H, H-6, s), 6.81 (1H, H-5’, d, \textit{J} = 8.0 Hz), 6.63 (1H, H-2’, s), 6.48 (1H, H-4, s), 4.92 (1H, H\textsubscript{glu}-1, d, \textit{J} = 7.0 Hz), 3.93 (1H, H\textsubscript{glu}-6a, d, \textit{J} = 11.0 Hz), 3.68 (1H, H\textsubscript{glu}-6b, dd, 11.0, \textit{J} = 5.0 Hz), 3.54-3.43 (4H, H\textsubscript{glu}-2, H\textsubscript{glu}-3, H\textsubscript{glu}-4, and H\textsubscript{glu}-5). \textsuperscript{13}C NMR (125 MHz, \textit{d}\textsubscript{4}-methanol): δ 160.5, 159.6 (C-3 and C-5), 146.6, 146.5 (C-3’ and C-4’), 141.4 (C-1), 131.0 (C-1’), 130.3 (C-8), 126.7 (C-7), 120.3 (C-6’), 116.4 (C-5’), 114.1 (C-2’), 108.3, 107.8 (C-2 and C-6), 104.1 (C-4), 102.4 (C\textsubscript{glu}-1), 78.2, 78.1 (C\textsubscript{glu}-3 and C\textsubscript{glu}-5), 74.9 (C\textsubscript{glu}-2), 71.5 (C\textsubscript{glu}-4), 62.6 (C\textsubscript{glu}-6 and C\textsubscript{glu}-6’). ESI-MS m/z: 407.08 [M+H]\textsuperscript{+}.

To a mixture of compound (140) (310 mg, 0.417 mmol) and boron trifluoride etherate (0.22 ml, 1.75 mmol) in dry dichloromethane (40 mL) at 0°C was added the orthoester (74) (278 mg, 0.438 mmol). The reaction was kept for 30 min. and allowed to warm up to room temperature and then quenched with saturated sodium bicarbonate (40 mL). The lower dichloromethane layer was collected, dried over anhydrous magnesium sulphate, filtered, and concentrated in vacuum. The residue was then separated by column chromatography (increasing polarity from 3% to 20% ethyl acetate in petroleum spirit as eluant) to afford (144) (35 mg, 6.8%), (143) (40 mg, 7.8%) and (141) (8 mg, 1.8%), respectively.
Compound (141): For characterisation data see alternative synthesis below.

Compound (142): $^1$H NMR (500 MHz, $d_6$-acetone): $\delta$ 7.35 (1H, H-2', s), 7.14 (1H, H-6', d, $J = 8.0$ Hz), 7.04 (1H, H-8, d, $J = 16.5$ Hz), 6.95 (1H, H-7, d, $J = 16.5$ Hz), 6.97 (1H, H-5', d, $J = 8.0$ Hz), 6.75 (2H, H-2 and H-6, s), 6.46 (1H, H-4, s), 5.57-5.47 (4H, $H_{\text{glu-1/2'/2''/3'}}$ and $H_{\text{glu-3/4'}}$, m), 5.26-5.19 (4H, $H_{\text{glu-2/2''/2''/3'}}$ and $H_{\text{glu-4/4''/5'5''}}$, m), 4.30-4.20 (6H, $H_{\text{glu-5/5'5''}}$ and $H_{\text{glu-6/6''}}$, m), 1.21, 1.18, 1.16, 1.14, 1.13 (72H, 24 x CH$_3$, s, 3s masked). $^{13}$C NMR (125 MHz, $d_6$-acetone): $\delta$ 178.26, 178.23, 177.57, 177.51, 177.13, 177.03, 177.00 (C=O, one C=O masked), 159.8, 159.4 (C-3 and C-5), 148.5, 145.6 (C-3' and C-4''), 140.9 (C-1), 130.3 (C-1''), 129.6 (C-8), 127.1 (C-7), 123.5 (C-6''), 117.4 (C-5'''), 116.1 (C-2''), 108.7, 106.2 (C-2 and C-6), 103.8 (C-4), 100.4, 99.4 (C$_{\text{glu-1 and C-glu-1''}}$), 73.18, 73.08, 72.90, 72.82 (C$_{\text{glu-3/3'' and C-glu-5/5''}}$), 72.3, 72.0 (C$_{\text{glu-2 and C-glu-2''}}$), 68.66, 68.61 (C$_{\text{glu-4 and C-glu-4''}}$), 62.60, 62.56 (C$_{\text{glu-6 and C-glu-6''}}$), 39.42, 39.39, 39.35, 39.34, 39.27 (C(C$_{\text{CH}_3}$)$_3$, three (C(C$_{\text{CH}_3}$)$_3$ masked), 27.48, 27.47, 27.44, 27.41, 27.39, 27.32 (C(C$_{\text{CH}_3}$)$_3$, two C(C$_{\text{CH}_3}$)$_3$ masked). ESI-HRMS m/z: 1241.6466 [M+H]$^+$, calcd. for C$_{28}$H$_{21}$O$_6$, 1241.6514.

Compound (143): $^1$H NMR (500 MHz, $d_6$-acetone): $\delta$ 7.14 (1H, H-5', d, $J = 8.0$ Hz), 7.12 (1H, H-2', s), 7.04 (1H, H-8, d, $J = 16.5$ Hz), 6.97 (1H, H-6', d, $J = 8.0$ Hz), 6.95 (1H, H-7, d, $J = 16.5$ Hz), 6.75 (2H, H-2 and H-6, s), 6.46 (1H, H-4, s), 5.50-5.39 (4H, $H_{\text{glu-1/2'/2''/3'}}$ and $H_{\text{glu-3/4'}}$, m), 5.26-5.19 (4H, $H_{\text{glu-2/2''/2''/3'}}$ and $H_{\text{glu-4/4''/5'5''}}$, m), 4.32-4.16 (6H, $H_{\text{glu-5/5'5''}}$ and $H_{\text{glu-6/6''}}$, m), 1.21, 1.19, 1.17, 1.16, 1.15 (72H, 24 x CH$_3$, 5s). $^{13}$C NMR (125 MHz, $d_6$-acetone): $\delta$ 178.3, 177.6, 177.50, 177.48, 177.10, 177.09, 177.01 (C=O), 159.8, 159.5 (C-3 and C-5), 148.5 (C-4''), 145.4 (C-3''), 140.7 (C-1), 134.2 (C-1''), 129.5 (C-8), 128.3 (C-7), 119.4 (C-6''), 117.8 (C-5''), 114.7 (C-2'), 108.9, 106.4 (C-2 and C-6), 104.1 (C-4), 100.5, 99.4 (C$_{\text{glu-1 and C-glu-1''}}$), 73.11, 73.06, 72.98, 72.87 (C$_{\text{glu-3/3'' and C-glu-5/5''}}$), 72.3, 72.0 (C$_{\text{glu-2 and C-glu-2''}}$), 68.8, 68.7 (C$_{\text{glu-4 and C-glu-4''}}$), 62.6 (C$_{\text{glu-6 and C-glu-6''}}$), 39.41, 39.39, 39.37, 39.34, 39.27 (C(C$_{\text{CH}_3}$)$_3$, three C(C$_{\text{CH}_3}$)$_3$ masked), 27.47, 27.43, 27.38, 27.30 (C(C$_{\text{CH}_3}$)$_3$, four C(C$_{\text{CH}_3}$)$_3$ masked). ESI-HRMS m/z: 1241.6466 [M+H]$^+$, calcd. for C$_{28}$H$_{21}$O$_6$, 1241.6437.
The title compound was prepared from (142) (16 mg, 0.0129 mmol), methanol (10 mL), and sodium metal (40 mg, 1.74 mmol) using the method described for the synthesis of compound (32). Column chromatography (20% methanol in ethyl acetate as eluant) yielded the title compound (132) as a white solid (7 mg, 95%).

\[ \text{ESI-MS } m/z: 569.09 \ [\text{M+H}]^+. \]

The title compound was prepared from (143) (15 mg, 0.0121 mmol), methanol (10 mL), and sodium metal (45 mg, 1.96 mmol) using the method described for the synthesis of compound (32). Column chromatography (20% methanol in ethyl acetate as eluant) yielded the title compound (114) as a white solid (6.8 mg, 98.8%).

$^1$H NMR (500 MHz, $d_4$-methanol): $\delta$ 7.11 (1H, H-5’, d, $J = 8.5$ Hz), 7.00 (1H, H-2’, s), 6.94 (1H, H-8, d, $J = 16.5$ Hz), 6.91 (1H, H-6’, d, $J = 8.5$ Hz), 6.84 (1H, H-7, d, $J = 16.5$ Hz), 6.75, 6.57 (2H, H-2 and H-6, 2s), 6.42 (1H, H-4, s), 4.84 (1H, H$_{\text{glu}}$-1 or H$_{\text{glu}}$-1’’, d, $J = 7.0$ Hz), 4.73 (1H, H$_{\text{glu}}$-1 or H$_{\text{glu}}$-1’’, d, $J = 7.5$ Hz), 3.88 (1H, H$_{\text{glu}}$-6a or H$_{\text{glu}}$-6a’’, d, $J = 11.0$ Hz), 3.85 (1H, H$_{\text{glu}}$-6a or H$_{\text{glu}}$-6a’’, d, $J = 12.0$ Hz), 3.69-3.64 (2H, H$_{\text{glu}}$-6b and H$_{\text{glu}}$-6b’’, m), 3.45-3.33 (8H, H$_{\text{glu}}$-2, H$_{\text{glu}}$-3, H$_{\text{glu}}$-4, H$_{\text{glu}}$-5, H$_{\text{glu}}$-2’’, H$_{\text{glu}}$-3’’, H$_{\text{glu}}$-4’’, and H$_{\text{glu}}$-5’’). $^{13}$C NMR (125 MHz, $d_4$-methanol): $\delta$ 160.8, 159.9 (C-3 and C-5), 148.8 (C-4’), 146.9 (C-3’), 141.3 (C-1), 134.9 (C-1’), 129.9 (C-8), 128.9 (C-7), 120.3 (C-6’), 119.0 (C-5’), 114.9 (C-2’), 108.9, 107.5 (C-6 and C-2), 104.8 (C-4), 104.5, 102.7 (C$_{\text{glu}}$-1 and C$_{\text{glu}}$-1’’), 78.7, 78.6, 78.4, 77.9 (C$_{\text{glu}}$-3/C$_{\text{glu}}$-3’’ and C$_{\text{glu}}$-5/C$_{\text{glu}}$-5’’), 75.3, 75.2 (C$_{\text{glu}}$-2 and C$_{\text{glu}}$-2’’), 71.8, 71.6 (C$_{\text{glu}}$-4 and C$_{\text{glu}}$-4’’), 62.9, 62.7 (C$_{\text{glu}}$-6 and C$_{\text{glu}}$-6’’). ESI-MS m/z: 569.00 [M+H]$^+$. 

![Diagram of compound 114]
4-[(E)-2(3,5-bis[{tert-Butyl(dimethyl)silyl]oxy}phenyl)ethenyl]benzene-1,2-diol (144).

To a mixture of the silylated resveratrol (66) (600 mg, 1.31 mmol) in methanol (40 mL) at -20 °C was slowly added o-iodoxybenzoic acid (IBX) (129) (552 mg, 1.97 mmol) with stirring. The reaction mixture was allowed to warm up to room temperature over 1 h and then quenched with a solution of Na$_2$S$_2$O$_4$ (85% grade) (537 mg, 2.62 mmol) in water (120 mL) followed by acidification to pH 4 with 1M hydrochloric acid. The organics were extracted with ethyl acetate (3 x 50 mL) and the combined ethyl acetate layers dried over anhydrous sodium sulfate and concentrated under vacuum. The crude product was purified by flash chromatography (increasing polarity from 2% to 20% ethyl acetate in petroleum spirit as eluant) to afford the title product (144) (450 mg, 73%) as a yellow solid.

$^1$H NMR (500 MHz, $d_6$-acetone): $\delta$ 7.10 (1H, H-2', s), 7.03 (1H, H-8, d, $J = 16.5$ Hz), 6.93 (1H, H-6', d, $J = 8.0$ Hz), 6.90 (1H, H-7, d, $J = 16.5$ Hz), 6.81 (1H, H-5', d, $J = 8.0$ Hz), 6.71 (2H, H-2 and H-6, s), 6.27 (1H, H-4, s), 1.01 (9H, SiC(CH$_3$)$_3$, s), 0.24 (6H, Si(CH$_3$)$_2$, s). $^{13}$C NMR (125 MHz, $d_6$-acetone): $\delta$ 159.73 (C-3 and C-5), 145.29, 145.22 (C-3’and C-4’), 140.1 (C-1), 129.7 (C-1’), 129.1 (C-8), 125.5 (C-7), 119.2 (C-6’), 115.3 (C-5’), 113.0 (C-2’), 111.4 (C-2 and C-6), 110.8 (C-4), 17.9 (SiC(CH$_3$)$_3$), -5.1 (Si(CH$_3$)$_2$).

ESI-HRMS m/z: 473.2538 [M+H]$^+$, calcd. for C$_{28}$H$_{21}$O$_6$, 473.2538.

5-[(E)-2-(3,5-Dihydroxyphenyl)ethenyl]-2-hydroxyphenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (149) and 4-[(E)-2-(3,5-Dihydroxyphenyl)ethenyl]-2-hydroxyphenyl 2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-β-D-glucopyranoside (150).
To a mixture of (141) (410 mg, 0.867 mmol), boron trifluoride etherate (0.52 mL, 4.16 mmol) in dry dichloromethane (50 mL) at 0°C was added the orthoester (74) (659 mg, 1.04 mmol). The reaction was allowed to warm up to room temperature over 30 min. and then the reaction mixture was quenched with saturated sodium bicarbonate (40 mL). The lower dichloromethane layer was collected, dried over anhydrous magnesium sulphate, filtered, and concentrated under vacuum. The residue was then purified by column chromatography (increasing polarity from 0% to 20% ethyl acetate in petroleum spirit as eluant) to give an inseparable mixture of the compounds (146) and (147) (500 mg) in 1:1 ratio. The mixture was dissolved in dry tetrahydrofuran (30 mL) at 0°C and TBAF (0.754 mL, 0.754 mmol) was added. The reaction was allowed to warm up to room temperature over 30 min. and then quenched with saturated sodium bicarbonate (30 mL) and extracted with ethyl acetate (3 x 20 mL). The combined organic extracts were dried over anhydrous magnesium sulphate, filtered and concentrated under vacuum. The residue was then purified by column chromatography (increasing polarity from 5% to 40% ethyl acetate in petroleum spirit as eluant) to afford (149) (129 mg, 20%) and (150) (155 mg, 24%), respectively.
Compound (149): \(^1\)H NMR (500 MHz, \(d_6\)-acetone): \(\delta 7.36\) (1H, H-2', s), 7.18 (1H, H-6', d, \(J = 9.0\) Hz), 6.99 (1H, H-8, d, \(J = 16.0\) Hz), 6.89 (1H, H-7, d, \(J = 16.0\) Hz), 6.88 (1H, H-5', d, \(J = 9.0\) Hz), 6.53 (2H, H-2 and H-6, s), 6.28 (1H, H-4, s), 5.60 (1H, H\textsubscript{glu}-1'', d, \(J = 8.0\) Hz), 5.51 (1H, H\textsubscript{glu}-3'', dd, \(J = 9.0, 9.0\) Hz), 5.28-5.23 (2H, H\textsubscript{glu}-2'', and H\textsubscript{glu}-4'', m), 4.32-4.19 (3H, H\textsubscript{glu}-5'' and H\textsubscript{glu}-6'', m), 1.20, 1.17, 1.16, 1.14 (36H, 12CH\textsubscript{3}, 4s). \(^{13}\)C NMR (125 MHz, \(d_6\)-acetone): \(\delta 178.10, 177.44, 177.41, 176.97\) (C=O), 159.6 (C-3 and C-5), 148.1 (C-3'), 145.2 (C-4'), 140.7 (C-1), 130.8 (C-1'), 128.8 (C-8), 127.9 (C-7), 123.5 (C-6'), 117.2 (C-5'), 115.7 (C-2'), 105.7 (C-2 and C-6), 102.8 (C-4), 100.6 (C\textsubscript{glu}-1'''), 73.2, 73.0 (C\textsubscript{glu}-3''' and C\textsubscript{glu}-5'''), 72.4 (C\textsubscript{glu}-2'''), 68.8 (C\textsubscript{glu}-4'''), 62.6 (C\textsubscript{glu}-6'''), 39.46, 39.42, 39.32 (C(CH\textsubscript{3})\textsubscript{3}), 27.53, 27.45, 27.37, 27.36 (C(CH\textsubscript{3})\textsubscript{3}). ESI-HRMS m/z: 743.3637 [M+H]\(^+\), calcd. for C\textsubscript{28}H\textsubscript{21}O\textsubscript{6}, 743.3630.

Compound (150): \(^1\)H NMR (500 MHz, \(d_6\)-acetone): \(\delta 7.13\) (1H, H-5', d, \(J = 8.5\) Hz), 7.11 (1H, H-2', s), 6.97 (1H, H-8, d, \(J = 16.5\) Hz), 6.96 (1H, H-6', d, \(J = 8.5\) Hz), 6.88 (1H, H-7, d, \(J = 16.5\) Hz), 6.52 (2H, H-2 and H-6, s), 6.26 (1H, H-4, s), 5.49-5.47 (2H, H\textsubscript{glu}-1''', and H\textsubscript{glu}-3'''', m), 5.27-5.21 (2H, H\textsubscript{glu}-2''', and H\textsubscript{glu}-4'''', m), 4.25-4.15 (3H, H\textsubscript{glu}-5'''', and H\textsubscript{glu}-6'''', m), 1.19, 1.18, 1.16, 1.14 (36H, 12CH\textsubscript{3}, 4s). \(^{13}\)C NMR (125 MHz, \(d_6\)-acetone): \(\delta 178.19, 178.61, 177.47, 177.08\) (C=O), 159.72 (C-3 and C-5), 148.5 (C-4'), 145.2 (C-3'), 140.2 (C-1), 133.0 (C-1'), 129.0 (C-7), 128.5 (C-8), 119.3 (C-6'), 117.9 (C-5'), 114.5 (C-2'), 105.8 (C-2 and C-6), 103.1 (C-4), 100.5 (C\textsubscript{glu}-1'''''), 73.1, 72.9 (C\textsubscript{glu}-3'''', and C\textsubscript{glu}-5'''''), 72.3 (C\textsubscript{glu}-2'''''), 68.8 (C\textsubscript{glu}-4'''''), 62.6 (C\textsubscript{glu}-6'''''), 39.40, 39.38, 39.34, 39.27 (C(CH\textsubscript{3})\textsubscript{3}), 27.46, 27.43, 27.37, 27.30 (C(CH\textsubscript{3})\textsubscript{3}). ESI-HRMS m/z: 743.3637 [M+H]\(^+\), calcd. for C\textsubscript{28}H\textsubscript{21}O\textsubscript{6}, 743.3630.

4-[(E)-2-(3,5-Dihydroxyphenyl)ethenyl]-2-hydroxyphenyl \(\beta\)-D-glucopyranoside (130).
The title compound was prepared from (150) (20 mg, 0.027 mmol), methanol (10 mL), and sodium metal (36 mg, 1.57 mmol) using the method described for the synthesis of compound (32). Column chromatography (10% methanol in ethyl acetate as eluant) yielded compound (130) as a white solid (1 mg, 9%).

\(^1\)H NMR (500 MHz, \(d_4\)-methanol): \(\delta\) 7.16 (1H, H-5', d, \(J = 8.0\) Hz), 7.03 (1H, H-2', s), 6.94 (1H, H-6', d, \(J = 8.0\) Hz), 6.93 (1H, H-8, d, \(J = 16.5\) Hz), 6.83 (1H, H-7, d, \(J = 16.5\) Hz), 6.45 (2H, H-2 and H-6, s), 6.18 (1H, H-4, s), 4.78 (1H, H\(\text{glu-1}'''\), d, \(J = 7.0\) Hz), 3.90 (1H, H\(\text{glu-6a d,} J = 11.5\) Hz), 3.74 (1H, H\(\text{glu-6b}'''', \dd, J = 11.5, 5.0\) Hz), 3.50-3.38 (4H, H\(\text{glu-2}''', H\(\text{glu-3}''', H\(\text{glu-4}'''', H\(\text{glu-5}''''). 13^C NMR (125 MHz, \(d_4\)-methanol) In part due to insufficient compound: \(\delta\) 160.0 (C-3 and C-5), 129.3 (C-8), 120.1 (C-6'), 119.1 (C-5'), 114.8 (C-2'), 106.2 (C-6 and C-2), 78.7, 77.9 (C\(\text{glu-3}'''', C\(\text{glu-5}''')), 75.1 (C\(\text{glu-2}''''), 71.6 (C\(\text{glu-4}'''), 62.7 (C\(\text{glu-6}'''). ESI-MS m/z: 407.05 [M+H].

3-[(E)-2-(3,4-Dihydroxyphenyl)ethenyl]-5-[[2,3,4,6-tetra-O-(2,2-dimethylpropanoyl)-\(\beta\)-D-glucopyranosyl]oxy]phenyl-2,3,4,6-tetra-O-(2,2-dimethylpropanoyl)-\(\beta\)-D-glucopyranoside (141).

![Diagram of compound 141](image)

To a mixture of resveratrol diglycoside (86) (400 mg, 0.326 mmol) in methanol (40 mL) at -20 °C was slowly added \(\alpha\)-iodoxybenzoic acid (IBX) (129) (137 mg, 0.489 mmol) with stirring. The reaction mixture was allowed to warm up to room temperature over 1 h. The reaction mixture was quenched with a solution of Na\(_2\)S\(_2\)O\(_4\) (85% grade) (133 mg, 0.652 mmol) in water (120 mL) and then acidified to pH 4 with 1M hydrochloric acid. The organic were extracted with ethyl acetate (3 x 50 mL) and the combined ethyl acetate
layers dried over anhydrous sodium sulfate and concentrated under vacuum. The crude product was purified by flash chromatography (increasing polarity from 5% to 30% ethyl acetate in petroleum spirit as eluant) to afford the title product (141) (320 mg, 79%) as a yellow solid.

$^1$H NMR (500 MHz, $d_6$-acetone): $\delta$ 7.09 (1H, H-8, d, $J = 16.5$ Hz), 7.07 (1H, H-2’, s), 6.98 (2H, H-2 and H-6, s), 6.91 (1H, H-7, d, $J = 16.5$ Hz), 6.88 (1H, H-6’, d, $J = 8.0$ Hz), 6.81 (1H, H-5’, d, $J = 8.0$ Hz), 6.57 (1H, H-4, s), 5.60 (2H, H$_{\text{glu}}$-1 and H$_{\text{glu}}$-1’, d, $J = 8.0$ Hz), 5.47 (2H, H$_{\text{glu}}$-3 and H$_{\text{glu}}$-3’, dd, $J = 9.5$ Hz), 5.24-5.19 (4H, H$_{\text{glu}}$-2/2’ and H$_{\text{glu}}$-4/4’, m), 4.32 (2H, H$_{\text{glu}}$-5 and H$_{\text{glu}}$-5’, m), 4.25-4.18 (4H, H$_{\text{glu}}$-6 and H$_{\text{glu}}$-6’, m), 1.19, 1.16 (72H, 24 x CH$_3$, 2s). $^{13}$C NMR (125 MHz, $d_6$-acetone): $\delta$ 178.22, 177.50, 177.07, 177.00 (C=O), 159.1 (C-3 and C-5), 146.71, 146.36 (C-3‘ and C-4’), 141.7 (C-1), 131.4 (C-8), 130.0 (C-1’), 125.6 (C-7), 120.2 (C-6’), 116.2 (C-5’), 114.0 (C-2’), 109.0 (C-2 and C-6), 104.8 (C-4), 99.0 (C$_{\text{glu}}$-1 and C$_{\text{glu}}$-1’), 73.01, 72.94 (C$_{\text{glu}}$-3/3’ and C$_{\text{glu}}$-5/5’), 72.0 (C$_{\text{glu}}$-2 and C$_{\text{glu}}$-2’), 68.6 (C$_{\text{glu}}$-4 and C$_{\text{glu}}$-4’), 62.6 (C$_{\text{glu}}$-6 and C$_{\text{glu}}$-6’), 39.3, 39.2 (C(CH$_3$)$_3$, two C(CH$_3$)$_3$ masked), 27.46, 27.41, 27.39 (C(CH$_3$)$_3$, one C(CH$_3$)$_3$ masked). ESI-HRMS m/z: 1241.6466 [M+H]$^+$, calcd. for C$_{28}$H$_{21}$O$_6$, 1241.6476.

3-[(E)-2-(3,4-Dihydroxyphenyl)ethenyl]-5-((β-D-glucopyranosyloxy)phenyl-β-D-glucopyranoside (131).

![Diagram of 3-[(E)-2-(3,4-Dihydroxyphenyl)ethenyl]-5-((β-D-glucopyranosyloxy)phenyl-β-D-glucopyranoside (131).](image)

The title compound was prepared from (141) (15 mg, 0.016 mmol), methanol (10 mL), and sodium metal (45 mg, 1.96 mmol) using the method described for the synthesis of compound (32). Column chromatography (20% methanol in ethyl acetate as eluant) yielded the title compound (131) as a white solid (4.5 mg, 49%).
$^1$H NMR (500 MHz, $d_4$-methanol): $\delta$ 7.01 (1H, H-8, d, $J = 16.0$ Hz), 6.99 (1H, H-2', s), 6.92 (2H, H-2 and H-6, s), 6.86 (1H, H-6', d, $J = 8.0$ Hz), 6.83 (1H, H-7, d, $J = 16.0$ Hz), 6.74 (1H, H-4, s), 6.73 (1H, H-5', d, $J = 8.0$ Hz), 4.93 (2H, H$_{\text{glu}}$-1 and H$_{\text{glu}}$-1', d, $J = 7.0$ Hz), 3.93 (1H, H$_{\text{glu}}$-6a or H$_{\text{glu}}$-6a', d, $J = 11.0$ Hz), 3.87 (1H, H$_{\text{glu}}$-6a or H$_{\text{glu}}$-6a', d, $J = 11.0$ Hz), 3.70-3.67 (2H, H$_{\text{glu}}$-6b and H$_{\text{glu}}$-6b'), 3.54-3.43 (8H, H$_{\text{glu}}$-2, H$_{\text{glu}}$-3, H$_{\text{glu}}$-4, H$_{\text{glu}}$-5, H$_{\text{glu}}$-2', H$_{\text{glu}}$-3', H$_{\text{glu}}$-4' and H$_{\text{glu}}$-5'). $^{13}$C NMR (125 MHz, $d_4$-methanol): $\delta$ 160.4 (C-3 and C-5), 141.6 (C-1), 131.1 (C-1'), 130.8 (C-8), 120.6 (C-6'), 114.2 (C-2'), 109.9 (C-2 and C-6), 103.4 (C-4), 102.4 (C$_{\text{glu}}$-1 and C$_{\text{glu}}$-1'), 78.5, 78.4, 78.2 (C$_{\text{glu}}$-3/ C$_{\text{glu}}$-3' and C$_{\text{glu}}$-5/ C$_{\text{glu}}$-5'), 75.1 (C$_{\text{glu}}$-2 and C$_{\text{glu}}$-2'), 71.8, 71.5 (C$_{\text{glu}}$-4 and C$_{\text{glu}}$-4'), 62.94, 62.87 (C$_{\text{glu}}$-6 and C$_{\text{glu}}$-6'). ESI-MS m/z: 569.05 [M+H]$^+$. 

5-[(E)-2-(3,5-bis{[2,3,4,6-tetrakis-O-(2,2-Dimethylpropanoyl)-$\beta$-D-glucopyranosyl]oxy}phenyl)ethenyl]-2-hydroxyphenyl-2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-$\beta$-D-glucopyranoside (151) and 3-[(E)-2-(3-hydroxy-4-{[2,3,4,6-tetrakis-O-(2,2-Dimethylpropanoyl)-$\beta$-D-glucopyranosyl]oxy}phenyl)ethenyl]-5-{[2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-$\beta$-D-glucopyranosyl]oxy}phenyl-2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-$\beta$-D-glucopyranoside (153).
To a solution of compound (141) (330 mg, 0.266 mmol), boron trifluoride etherate (0.13 mL, 1.06 mmol) in dry dichloromethane (60 mL) at 0°C was added the orthoester (74) (169 mg, 0.266 mmol). The reaction was allowed to warm up to room temperature over 30 min. and then quenched with saturated sodium bicarbonate (40 mL). The lower dichloromethane layer was collected, dried over anhydrous magnesium sulphate, filtered, and concentrated in vacuum. The residue was then purified by column chromatography (increasing polarity from 0% to 20% ethyl acetate in petroleum spirit as eluant) to afford two fractions (fraction A: a mixture of (151) and (152) in 7:3 ratio (120 mg); fraction B: a mixture of (151) and (152) in 2:8 ratio (230 mg)).

Compound (151): $^1$H NMR (500 MHz, $d_6$-acetone) (in part): $\delta$ 7.38 (1H, H-2', s), 7.00 (2H, H-2 and H-6, s), 6.92 (1H, H-5', d, $J$ = 8.5 Hz), 6.62 (1H, H-4, s). ESI-MS m/z: 1739.9 [M+H]$^+$.  

Compound (152): $^1$H NMR (500 MHz, $d_6$-acetone) (in part): $\delta$ 7.03 (2H, H-2 and H-6, s), 6.62 (1H, H-4, s). ESI-MS m/z: 1739.9 [M+H]$^+$.  


The mixture (fraction A, 120 mg) was dissolved in dichloromethane (20 mL) and treated with acetic anhydride (0.033 mL, 0.348 mmol) and triethylamine (0.049 mL, 0.348 mmol). The reactions was kept for 2 h at room temperature and then quenched with
saturated sodium bicarbonate (30 mL). The organic substrates were extracted into ethyl acetate (3 x 30 mL) and the combined organics dried over anhydrous magnesium sulphate, filtered and concentrated in vacuum. Upon flash column chromatography (increasing polarity from 0% to 20% ethyl acetate in petroleum spirit as eluant), a mixture of the acetates (154) and (155) were obtained in 7:3 ratio (115 mg in total). Compound (154) (15 mg) was recrystallised from the mixture from MeOH as a white solid.

M.p. 126.5-129.0 °C. $^1$H NMR (500 MHz, $d_6$-acetone): $\delta$ 7.44 (1H, H-2’, s), 7.27 (1H, H-6’, d, $J = 8.5$ Hz), 7.20 (1H, H-8, d, $J = 16.5$ Hz), 7.14 (1H, H-7, d, $J = 16.5$ Hz), 7.10 (1H, H-5’, d, $J = 8.5$ Hz), 7.02 (2H, H-2 and H-6, s), 6.65 (1H, H-4, s), 5.62 (1H, H$_{\text{glu}}$-1”’, d, $J = 8.0$ Hz), 5.59 (2H, H$_{\text{glu}}$-1 and H$_{\text{glu}}$-1’, d, $J = 8.0$ Hz), 5.50-5.45 (3H, H$_{\text{glu}}$-3, H$_{\text{glu}}$-3’’, and H$_{\text{glu}}$-3’’, m), 4.41-4.21 (9H, H$_{\text{glu}}$-5/ H$_{\text{glu}}$-5'/ H$_{\text{glu}}$-5'' and H$_{\text{glu}}$-6/ H$_{\text{glu}}$-6'/ H$_{\text{glu}}$-6'', m), 2.25 (3H, COCH$_3$), 1.16, 1.15, 1.13, 1.12 (96H, 32 x CH$_3$, 4s, 4s masked).

$^{13}$C NMR (125 MHz, $d_6$-acetone): $\delta$ 177.07, 176.98, 176.94 (CO(CH$_3$)$_3$, five CO(CH$_3$)$_3$ masked), 169.2 (COCH$_3$), 159.10 (C-3 and C-5), 149.5 (C-3’ and C-4’), 140.9 (C-1), 130.2 (C-8), 129.5 (C-7), 124.6 (C-6’), 121.6 (C-5’), 113.9 (C-2’), 109.8 (C-2 and C-6), 105.3 (C-4), 99.1, 98.6 (C$_{\text{glu}}$-1, C$_{\text{glu}}$-1’ and C$_{\text{glu}}$-1’’), 73.03, 73.01, 72.92, 72.84 (C$_{\text{glu}}$-3/ C$_{\text{glu}}$-3’/ C$_{\text{glu}}$-3’’ and C$_{\text{glu}}$-5/ C$_{\text{glu}}$-5’/ C$_{\text{glu}}$-5’’), 72.0, 71.8 (C$_{\text{glu}}$-2, C$_{\text{glu}}$-2’ and C$_{\text{glu}}$-2’’), 68.52, 68.50 (C$_{\text{glu}}$-4, C$_{\text{glu}}$-4’ and C$_{\text{glu}}$-4’’), 62.67, 62.52 (C$_{\text{glu}}$-6, C$_{\text{glu}}$-6’ and C$_{\text{glu}}$-6’’), 39.42, 39.39, 39.38, 39.36, 39.35, 39.34, 39.29 (C(CH$_3$)$_3$, one (C(CH$_3$)$_3$ masked), 27.47, 27.37, 27.33, 27.32, 27.26 (C(CH$_3$)$_3$, three (C(CH$_3$)$_3$ masked), 20.56 (COCH$_3$). ESI-HRMS m/z: 1781.9400 [M+H]$^+$, calcd. for C$_{28}$H$_{21}$O$_6$, 1781.9830.

3-[(E)-2-(3-acetoxy-4-[[2,3,4,6-tetrakis-O-(2,2-Dimethylpropanoyl)-$\beta$-D-glucopyranosyl]oxy]phenyl]ethenyl]-5-[[2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-$\beta$-D-glucopyranosyl]oxy]phenyl-2,3,4,6-tetrakis-O-(2,2-dimethylpropanoyl)-$\beta$-D-glucopyranoside (155).
The mixture (fraction B, 230 mg) was dissolved in dichloromethane (30 mL) and treated with acetic anhydride (0.066 mL, 0.696 mmol) and triethylamine (0.098 mL, 0.696 mmol). The reactions were kept for 2 h at room temperature and then quenched with saturated sodium bicarbonate (30 mL). The organic substrates were extracted into ethyl acetate (3 x 30 mL) and the combined organic extracts dried over anhydrous magnesium sulphate, filtered and concentrated in vacuum. Upon flash column chromatography (increasing polarity from 0% to 20% ethyl acetate in petroleum spirit as eluant), a mixture of the acetates (154) and (155) were obtained in 2:8 ratio (200 mg in total). Compound (155) (35 mg) was recrystallised from the mixture from MeOH as a white solid.

M.p. 162.5-164.7 °C. ¹H NMR (500 MHz, d₆-acetone): δ 7.36 (1H, H-6', d, J = 8.5 Hz), 7.34 (1H, H-2', s), 7.28 (1H, H-5', d, J = 8.5 Hz), 7.21 (1H, H-8, d, J = 16.5 Hz), 7.07 (1H, H-7, d, J = 16.5 Hz), 7.03 (2H, H-2 and H-6, s), 6.61 (1H, H-4, s), 5.60 (2H, H_glu-1 and H_glu-1', d, J = 8.0 Hz), 5.58 (1H, H_glu-1'', d, J = 8.0 Hz), 5.50-5.45 (3H, H_glu-3, H_glu-3', and H_glu-3'', m), 5.28-5.20 (6H, H_glu-2/ H_glu-2'/ H_glu-2'' and H_glu-4/ H_glu-4'/ H_glu-4''', m), 4.37-4.23 (9H, H_glu-5/ H_glu-5'/ H_glu-5''' and H_glu-6/ H_glu-6'/H_glu-6''', m), 2.29 (3H, COCH₃), 1.24, 1.18, 1.17, 1.16, 1.14 (96H, 32 x CH₃, 5s). ¹³C NMR (125 MHz, d₆-acetone): δ 177.14, 177.06, 177.01 (CO(CH₃)₃, five C=O masked), 169.2 (COCH₃), 159.1 (C-3 and C-5), 149.1 (C-3' and C-4'), 141.12, 141.03 (C-1 and C-4'), 133.0 (C-1'), 129.6 (C-8), 128.4 (C-7), 126.4 (C-6'), 121.8 (C-2'), 115.9 (C-5'), 109.5 (C-2 and C-6), 105.3 (C-4), 99.0, 98.6 (C_glu-1 and C_glu-1'''), 72.98, 72.91, (C_glu-3/3''' and C_glu-5/5'''), 71.91, 71.75 (C_glu-2 and C_glu-2'''), 68.7, 68.6 (C_glu-4 and C_glu-4'''), 62.7, 62.6 (C_glu-6 and C_glu-
The title compound was prepared from (154) (15 mg, 0.0085 mmol), methanol (10 mL), and sodium metal (25 mg, 1.09 mmol) using the method described for the synthesis of compound (32). Column chromatography (30% methanol in ethyl acetate as eluant) yielded the title compound (134) as a white solid (5.5 mg, 89%).

$^1$H NMR (500 MHz, $d_4$-methanol): $\delta$ 7.50 (1H, H-2’, s), 7.08 (1H, H-6’, d, $J = 8.0$ Hz), 7.06 (1H, H-8, d, $J = 16.5$ Hz), 6.96 (2H, H-2 and H-6, s), 6.94 (1H, H-7, d, $J = 16.5$ Hz), 6.81 (1H, H-5’, d, $J = 8.0$ Hz), 6.74 (1H, H-4, s), 4.91 (2H, H$_{\text{glu}}$-1 and H$_{\text{glu}}$-1’, d, $J = 7.0$ Hz), 4.80 (1H, H$_{\text{glu}}$-1”’, d, $J = 7.0$ Hz), 3.98 (1H, H$_{\text{glu}}$-6a”’, d, $J = 11.5$ Hz), 3.94 (2H, H$_{\text{glu}}$-6a and H$_{\text{glu}}$-6a’, d, $J = 11.0$ Hz), 3.72-3.76 (3H, H$_{\text{glu}}$-6b, H$_{\text{glu}}$-6b’, H$_{\text{glu}}$-6b”’, m), 3.52-3.54 (12H, H$_{\text{glu}}$-2, H$_{\text{glu}}$-3, H$_{\text{glu}}$-4, H$_{\text{glu}}$-5, H$_{\text{glu}}$-2’, H$_{\text{glu}}$-3’, H$_{\text{glu}}$-4’, H$_{\text{glu}}$-5’, H$_{\text{glu}}$-2”’, H$_{\text{glu}}$-3”’, H$_{\text{glu}}$-4”’ and H$_{\text{glu}}$-5”’). $^{13}$C NMR (125 MHz, $d_4$-methanol): $\delta$ 160.4 (C-3 and C-5), 148.6 (C-3’), 147.4 (C-4’), 141.6 (C-1), 131.4 (C-1’), 130.6 (C-8), 127.4 (C-7), 124.1 (C-6’), 117.4 (C-5’), 116.6 (C-2’), 110.0 (C-2 and C-6), 105.4 (C-4), 104.7, 102.6 (C$_{\text{glu}}$-1, C$_{\text{glu}}$-1’ and C$_{\text{glu}}$-1”’), 78.9, 78.6, 78.3, 78.0 (C$_{\text{glu}}$-3/ C$_{\text{glu}}$-3’/C$_{\text{glu}}$-3”’ and C$_{\text{glu}}$-5/ C$_{\text{glu}}$-5’/C$_{\text{glu}}$-5”’), 75.3, 75.2 (C$_{\text{glu}}$-2, C$_{\text{glu}}$-2’ and C$_{\text{glu}}$-2”’), 71.99, 71.96 (C$_{\text{glu}}$-4, C$_{\text{glu}}$-4’ and C$_{\text{glu}}$-4”’), 63.03, 62.99 (C$_{\text{glu}}$-6, C$_{\text{glu}}$-6’ and C$_{\text{glu}}$-6”’). ESI-MS m/z: 731.00 [M+H]$^+$. 

5-{(E)-2-[3,5-bis(β-D-Glucopyranosyloxy)phenyl]ethenyl}-2-hydroxyphenyl-β-D-glucopyranoside (134).
The title compound was prepared from (155) (20 mg, 0.0113 mmol), methanol (10 mL), and sodium metal (32 mg, 1.39 mmol) using the method described for the synthesis of compound (32). Column chromatography (30% methanol in ethyl acetate as eluant) yielded the title compound (136) as a white solid (8 mg, 97%).

$^1$H NMR (500 MHz, $d_4$-methanol): $\delta$ 7.16 (1H, H-5’, d, $J = 8.0$ Hz), 7.06 (1H, H-2’, s), 7.04 (1H, H-8, d, $J = 16.5$ Hz), 6.96 (1H, H-6’, d, $J = 8.0$ Hz), 6.95 (2H, H-2 and H-6, s), 6.93 (1H, H-7, d, $J = 16.5$ Hz), 6.77 (1H, H-4, s), 4.94 (2H, H$_{\text{glu-1}}$ and H$_{\text{glu-1’}}$, d, $J = 6.5$ Hz), 4.78 (1H, H$_{\text{glu-1’’’}}$, d, $J = 7.0$ Hz), 3.93 (2H, H$_{\text{glu-6a}}$ and H$_{\text{glu-6a’}}$, d, $J = 12.0$ Hz), 3.90 (1H, H$_{\text{glu-6a’’’}}$, d, $J = 12.0$ Hz), 3.73-3.67 (3H, H$_{\text{glu-6b}}$, H$_{\text{glu-6b’}}$, H$_{\text{glu-6b’’}}$, m), 3.55-3.41 (12H, H$_{\text{glu-2}}$, H$_{\text{glu-3}}$, H$_{\text{glu-4}}$, H$_{\text{glu-5}}$, H$_{\text{glu-2’}}$, H$_{\text{glu-3’}}$, H$_{\text{glu-4’}}$, H$_{\text{glu-5’}}$, H$_{\text{glu-2’’’}}$, H$_{\text{glu-3’’’}}$, H$_{\text{glu-4’’’}}$ and H$_{\text{glu-5’’’}}$). $^{13}$C NMR (125 MHz, $d_4$-methanol): $\delta$ 160.5 (C-3 and C-5), 148.8 (C-4’), 147.0 (C-3’), 141.3 (C-1), 134.7 (C-1’), 130.5 (C-8), 128.4 (C-7), 120.3 (C-6’), 119.0 (C-5’), 115.0 (C-2’), 110.2 (C-2 and C-6), 105.5 (C-4), 104.4, 102.5 (C$_{\text{glu-1}}$, C$_{\text{glu-1’}}$ and C$_{\text{glu-1’’’}}$), 78.6, 78.5, 78.3, 78.7 (C$_{\text{glu-3}}$/C$_{\text{glu-3’}}$/C$_{\text{glu-3’’’}}$ and C$_{\text{glu-5’}}$/C$_{\text{glu-5’}}$/C$_{\text{glu-5’’’}}$), 75.22, 75.16 (C$_{\text{glu-2}}$, C$_{\text{glu-2’}}$ and C$_{\text{glu-2’’’}}$), 71.9, 71.6 (C$_{\text{glu-4}}$, C$_{\text{glu-4’}}$ and C$_{\text{glu-4’’’}}$), 63.0, 62.7 (C$_{\text{glu-6}}$, C$_{\text{glu-6’}}$ and C$_{\text{glu-6’’’}}$). ESI-MS m/z: 731.12 [M+H]$^+$.
5.4 Experimental for Chapter 4.

Preparation of a mixture of trans-epsilon-viniferin pentaacetate (182), trans-delta-viniferin pentaacetate (183) and Pallidol hexaacetate (184).

To a solution of trans-resveratrol (1a) (500 mg, 2.19 mmol) and K$_2$CO$_3$ (245 mg, 1.77 mmol) in MeOH (120 mL) at room temperature was slowly added an aqueous solution (10 mL) of K$_3$Fe(CN)$_6$ (575 mg, 1.75 mmol) over 5 minutes and the mixture stirred for a further 30 minutes. The mixture was then concentrated in vacuo and loaded directly onto a flash chromatography column and the organics eluted with ethyl acetate. The fraction containing the crude dimers was then concentrated in vacuo and then dissolved in CH$_2$Cl$_2$ (60 mL) followed by the addition of DMSO (10 mL), Ac$_2$O (0.83 mL, 8.8 mmol) and Et$_3$N (1.23 mL, 8.9 mmol) and the reaction mixture kept at ambient temperature for a further 24 hours. The reaction was then quenched with NaHCO$_3$ (30 ml) and the organics extracted with EtOAc (3 x 30 mL). The combined organics were washed with water (20 mL), dried over anhydrous MgSO$_4$ and the volatiles removed in vacuo. The acetates (182), (183) and (184) were then separated by flush column chromatography (increasing polarity from 20% to 50% ethyl acetate in petroleum spirit) to afford pallidol hexaacetate (150 mg, 19.4%) and a mixture of (182) and (183) (120 mg, ratio 1:2, 16.5%). Recrystallisation of
pallidol hexaacetate (184) from EtOAc gave the product as colourless cubes. The two viniferin acetates (182) and (183) were further separated by simple recrystallisation (MeOH) at ambient temperature to furnish pure (183) (50 mg, 6.9%). The mother liquors were then placed at –20 °C resulting in the precipitation of a mixture of (182) and (183) (45 mg). The mother liquors were then evaporated to afford pure (182) (20 mg, 2.8%).

**trans-epsilon-Viniferin pentaacetate; 3-(Acetyloxy)-5-[6-(acetyloxy)-2-[4-(acetyloxy)phenyl]-4-{(E)-2-[4-(acetyloxy)phenyl]ethenyl}-2,3-dihydro-1-benzofuran-3-yl]phenyl acetate (182):** 

$^1$H NMR (600 MHz, CDCl$_3$): δ 7.33 (2H, H-2a and H-6a, d, $J = 9.0$ Hz), 7.18 (2H, H-10b and H-14b, d, $J = 9.0$ Hz), 7.09 (2H, H-3a and H-5a, d, $J = 9.0$ Hz), 6.98 (2H, H-11b and H-13b, d, $J = 9.0$ Hz), 6.94 (1H, H-6b, d, $J = 1.8$ Hz), 6.90 (1H, H-12a, dd, $J = 1.8, 1.8$ Hz), 6.87 (1H, H-8b, d, $J = 16.8$ Hz), 6.85 (2H, H-10a and H-14a, dd, $J = 1.8, 1.8$ Hz), 6.65 (1H, H-4b, dd, $J = 1.8, 1.8$ Hz), 6.53 (1H, H-7b, d, $J = 16.8$ Hz), 5.60 (1H, H-7a, d, $J = 7.2$ Hz), 4.59 (1H, H-8a, d, $J = 7.2$ Hz), 2.34, 2.30, 2.27, 2.26 (15H, 5 x COCH$_3$, 4s). $^{13}$C NMR (150 MHz, CDCl$_3$): δ 169.41, 169.37, 169.34, 168.69 (5 x COCH$_3$), 160.9 (C-3b), 152.1 (C-5b), 151.8 (C-11a and C-13a), 150.6 (C-4a), 150.3 (C-12b), 144.4 (C-9a), 138.0 (C-1a), 135.3 (C-1b), 134.4 (C-9b), 130.3 (C-8b), 127.7 (C-10b and C-14b), 126.7 (C-2a and C-6a), 124.0 (C-7b), 123.8 (C-2b), 121.9 (C-3a and C-5a), 121.7 (C-11b and C-13b), 118.5 (C-10a and C-14a), 114.8 (C-12a), 110.7 (C-6b), 102.8 (C-4b), 92.7 (d, C-7a), 56.6 (d, C-8a), 21.12, 21.09 (5 x COCH$_3$).

**trans-delta-Viniferin pentaacetate; 3-(Acetyloxy)-5-{(E)-2-[2-[4-(acetyloxy)phenyl]-3-[3,5-bis(acetyloxy)phenyl]-2,3-dihydro-1-benzofuran-5-yl]ethenyl}phenyl acetate (183):** 

$^1$H NMR (600 MHz, CDCl$_3$): δ 7.37 (1H, H-6b, d, $J = 7.8$ Hz), 7.34 (2H, H-2a and H-6a, d, $J = 8.4$ Hz), 7.20 (1H, H-2b, s), 7.10 (2H, H-3a and H-5a, d, $J = 8.4$ Hz), 7.07 (2H, H-10b and H-14b, d, $J = 1.8$ Hz), 7.01 (1H, H-7b, d, $J = 16.8$ Hz), 6.95 (1H, H-5b, d, $J = 7.8$ Hz), 6.93 (1H, H-12a, dd, $J = 1.8, 1.8$ Hz), 6.85 (1H, H-8b, d, $J = 16.8$ Hz), 6.82 (2H, H-10a and H-14a, dd, $J = 1.8, 1.8$ Hz), 6.78 (1H, H-12b, dd, $J = 1.8, 1.8$ Hz), 5.55 (1H, H-7a, d, $J = 8.4$ Hz), 4.54 (1H, H-8a, d, $J = 8.4$ Hz), 2.30, 2.29, 2.26 (15H, 5 x COCH$_3$, 3s). $^{13}$C NMR (150 MHz, CDCl$_3$): δ 169.33, 168.96, 168.81 (5 x COCH$_3$), 159.92 (C-4b), 151.41 (C-11a and C-13a), 151.15 (C-11b and C-13b), 150.66 (C-4a), 143.70 (C-9a), 139.93 (C-9b), 137.70 (C-1a), 130.71 (C-1b), 130.14 (C-7b), 129.74 (C-3b), 128.50 (C-6b), 126.92 (C-2a and C-6a), 124.94 (C-8b), 123.40 (C-2b), 121.91 (C-3a
and C-5a), 118.76 (C-10a and C-14a), 116.61 (C-10b and C-14b), 114.73 (C-12a), 113.90 (C-12b), 110.00 (C-5b), 93.38 (C-7a), 57.29 (C-8a), 21.12, 21.09 (5 x COCH₃).

**Pallidol hexaacetate;** (4bS,9bS)-5,10-bis[4-(Acetyloxy)phenyl]-4b,5,9b,10-tetrahydroindeno[2,1-a]indene-1,3,6,8-tetrayl tetraacetate (184): M.p. 213.0–215.4 °C. ¹H NMR (600 MHz, CDCl₃): δ 7.15 (4H, H-2/2’ and H-6/6’, d, J = 8.4 Hz), 7.05 (4H, H-3/3’ and H-5/5’, d, J = 8.4 Hz), 6.88 (2H, H-12 and H-12’, d, J = 1.8 Hz), 6.76 (2H, H-14 and H-14’, d, J = 1.8 Hz), 4.45 (2H, H-7 and H-7’, t, J = 1.8 Hz), 4.17 (2H, H-8 and H-8’, t, J = 1.8 Hz), 2.30, 2.28, 1.69 (18H, 6 x COCH₃, 3s). ¹³C NMR (150 MHz, CDCl₃): δ 169.46, 169.01, 167.82 (6 x COCH₃), 150.9 (C-13 and C13’), 149.5 (C-4 and C-4’), 147.9 (C-9 and C9’), 147.4 (C-11 and C-11’), 140.6 (C-1 and C1’), 133.6 (C-10 and C-10’), 128.7 (C-2/2’ and C-6/6’), 121.8 (C-3/3’ and C-5/5’), 115.4 (C-14/14’), 115.1 (C-12/12’), 61.2 (C-8 and C-8’), 55.7 (C-7 and C-7’), 21.11, 21.09, 19.95 (6 x COCH₃).

**trans-epsilon-viniferin;** 5-{6-Hydroxy-2-(4-hydroxyphenyl)-4-[(E)-2-(4-hydroxyphenyl)ethenyl]-2,3-dihydro-1-benzofuran-3-yl]benzene-1,3-diol (33).

To a mixture of (182) (16 mg, 0.024 mmol) in MeOH (10 mL) and water (20 mL) was added K₂CO₃ (30 mg). The reaction mixture was kept at room temperature for 24 h and then neutralised with diluted HCl. The product was extracted with EtOAc (3 x 20 mL) and the combined extracts washed with water (20 mL), dried over anhydrous MgSO₄ and the volatiles removed *in vacuo*. The crude product was then subjected to chromatography employing ethyl acetate in petroleum spirit (7:1) as eluent to furnish the title compound (33) (10 mg, 92%).
\(^1\)H NMR (600 MHz, \(d_6\)-acetone): \(\delta 7.19\) (2H, H-2a and H-6a, d, \(J = 8.4\) Hz), \(7.17\) (2H, H-10b and H-14b, d, \(J = 9.0\) Hz), \(6.91\) (1H, H-8b, d, \(J = 16.5\) Hz), \(6.84\) (2H, H-3a and H-5a, d, \(J = 8.4\) Hz), \(6.74\) (2H, H-11b and H-13b, d, \(J = 9.0\) Hz), \(6.73\) (1H, H-6b, brs), \(6.70\) (1H, H-7b, d, \(J = 16.5\) Hz), \(6.33\) (1H, H-4b, d, \(J = 2.4\) Hz), \(5.43\) (1H, H-7a, d, \(J = 5.4\) Hz), \(4.45\) (1H, H-8a, d, \(J = 5.4\) Hz).

\(^13\)C NMR (150 MHz, \(d_6\)-acetone): \(\delta 161.9\) (C-3b), \(159.3\) (C-11a and C-13a), \(159.2\) (C-5b), \(160.0\) (C-4a and C-12b), \(144.4\) (C-9a), \(135.9\) (C-1b), \(133.3\) (C-1a), \(129.6\) (C-8b), \(129.3\) (C-9b), \(128.3\) (C-10b and C-14b), \(127.4\) (C-2a and C-6a), \(122.9\) (C-7b), \(119.3\) (C-2b), \(116.0\) (C-11b and C-13b), \(115.8\) (C-3a and C-5a), \(106.5\) (C-10a and C-14a), \(103.8\) (C-6b), \(101.7\) (C-12a), \(96.4\) (C-4b), \(93.4\) (d, C-7a), \(56.8\) (C-8a). ESI-HRMS m/z: 453.1344 [M-H], calcd. for C\(_{28}\)H\(_{21}\)O\(_6\), 453.1376.

trans-delta-Viniferin; 5-[(E)-2-(3,5-Dihydroxyphenyl)ethenyl]-2-(4-hydroxyphenyl)-2,3-dihydro-1-benzofuran-3-yl]benzene-1,3-diol (163).

![Chemical Structure](image)

To a mixture of (184) (30 mg, 0.045 mmol) in MeOH (10 mL) and water (20 mL) was added K\(_2\)CO\(_3\) (40 mg). The reaction mixture was kept at room temperature for 24 h and then neutralised with diluted HCl. The product was extracted with EtOAc (3 x 20 mL) and the combined extracts washed with water (20 mL), dried over anhydrous MgSO\(_4\) and the volatiles removed \textit{in vacuo}. The crude product was then subjected to chromatography employing ethyl acetate in petroleum spirit (7:1) as eluent to furnish the title compound (163) (19 mg, 93%).

\(^1\)H NMR (600 MHz, \(d_4\)-MeOH): \(\delta 7.35\) (1H, H-6b, d, \(J = 7.8\) Hz), \(7.17\) (2H, H-2a and H-6a, d, \(J = 8.4\) Hz), \(7.17\) (1H, H-2b, s), \(6.97\) (1H, H-7b, d, \(J = 16.8\) Hz), \(6.84\) (1H, H-5b, d, \(J = 7.8\) Hz), \(6.77\) (2H, H-3a and H-5a, d, \(J = 8.4\) Hz), \(6.77\) (1H, H-8b, d, \(J = 16.8\) Hz), \(6.42\) (2H, H-10b and H-14b, d, \(J = 1.8\) Hz), \(6.19\) (1H, H-12a, dd, \(J = 1.8, 1.8\) Hz), \(6.14\)
(1H, H-12b, dd, J = 1.8, 1.8 Hz), 6.11 (2H, H-10a and H-14a, dd, J = 1.8, 1.8 Hz), 5.36 (1H, H-7a, d, J = 8.4 Hz), 4.39 (1H, H-8a, d, J = 8.4 Hz). \(^{13}\)C NMR (150 MHz, \(d_4\)-MeOH): \(\delta\) 161.4 (C-4b), 160.4 (C-11a and C-13a), 160.2 (C-11b and C-13b), 159.2 (C-4a), 145.9 (C-9a), 141.6 (C-9b), 133.3 (C-1b), 132.88 (C-3b), 132.83 (C-1a), 129.8 (C-7), 129.1 (C-6b), 129.1 (C-2a and C-6a), 127.9 (C-8b), 124.6 (C-2b), 116.9 (C-3a and C-5a), 110.8 (C-5b), 108.2 (C-10a and C-14a), 106.3 (C-10b, 14b), 103.2 (C-14b), 102.9 (C-12a), 94.9 (C-7a), 59.2 (C-8a). ESI-HRMS m/z: 453.1344 [M-H], calcd. for C\(_{28}\)H\(_{21}\)O\(_6\), 453.1356.

**Pallidol; 5,10-bis(4-Hydroxyphenyl)-4b,5,9b,10-tetrahydroindeno[2,1-a]indene-1,3,6,8-tetrol (35).**

![Chemical Structure](image)

To a mixture of (185) (30 mg, 0.042 mmol) in MeOH (10 mL) and water (20 mL) was added K\(_2\)CO\(_3\) (35 mg). The reaction mixture was kept at room temperature for 24 h and then neutralised with diluted HCl. The product was extracted with EtOAc (3 x 20 mL) and the combined extracts washed with water (20 mL), dried over anhydrous MgSO\(_4\) and the volatiles removed *in vacuo*. The crude product was then subjected to chromatography employing ethyl acetate in petroleum spirit (7:1) as eluent to furnish the title compound (35) (15 mg, 79%).

\(^1\)H NMR (300 MHz, \(d_6\)-DMSO): 3.74 (2H, brs, H-8 and H-8’), 4.43 (2H, brs, H-7 and H-7’), 6.13 (2H, d, J=1.8 Hz, H-14 and H-14’), 6.46 (2H, d, J = 1.8 Hz, H-12 and H-12’), 6.63 (4H, d, J = 9.0 Hz, H-3, H-5, H-3’ and H-5’), 6.90 (4H, d, J = 9.0 Hz, H-2, H-6, H-2’ and H-6’). \(^{13}\)C NMR (75 MHz, \(d_6\)-DMSO) \(\delta\) 158.2 (C-11 and C-11’), 155.4 (C-4 and C-4’), 154.4 (C-13 and C-13’), 148.9 (C-9 and C-9’), 136.5 (C-1 and C-1’), 128.1 (C-2, C-
2’, C-6’ and C-6’), 122.1 (C-10 and C-10’), 115.2 (C-3, C-3’, C-5 and C-5’), 102.2 (C-14 and C-14’), 101.7 (C-12 and C-12’), 59.2 (C-8 and C-8’), 52.7 (C-7 and C-7’). ESI-HRMS m/z: 453.1344 [M-H], calcd. for C_{28}H_{21}O_{6}, 453.1347.

cis-epsilon-Viniferin;
5-{6-Hydroxy-2-(4-hydroxyphenyl)-4-[(Z)-2-(4-hydroxyphenyl)ethenyl]-2,3-dihydro-1-benzofuran-3-yl]benzene-1,3-diol (185).

A solution of (33) (10 mg) in d_6-acetone (0.5 mL) was irradiated under UV light (365 nm) for 1 h. The trans/cis isomerisation was then immediately analysed by ^1^H NMR which indicated the cis-isomer (185) was afforded in 50% yield.

^1^H NMR (600 MHz, d_6-acetone): δ 7.04 (2H, H-2a and H-6a, d, J = 8.4 Hz), 7.00 (2H, H-10b and H-14b, d, J = 9.0 Hz), 6.80 (2H, H-3a and H-5a, d, J = 8.4 Hz), 6.64 (2H, H-11b and H-13b, d, J = 9.0 Hz), 6.30 (2H, H-4b and H-6b), 6.24 (1H, H-8b, d, J = 12.0 Hz), 6.20 (1H, H-12a, dd, J = 1.8 Hz), 6.03 (2H, H-10a and H-14a, dd, J = 1.8 Hz), 6.02 (1H, H-7b, d, J = 12.0 Hz), 5.28 (1H, H-7a, d, J = 5.4 Hz), 3.98 (1H, H-8a, d, J = 5.4 Hz).

cis-delta-Viniferin; 5-{5-[Z]-2-(3,5-Dihydroxyphenyl)ethenyl]-2-(4-hydroxyphenyl)-2,3-dihydro-1-benzofuran-3-yl]benzene-1,3-diol (186).
A solution of *trans*-epsilon viniferin (163) (10 mg) in *d*₄-MeOH (0.5 mL) was irradiated under UV light (365 nm) for 1 h. The *trans/cis* isomerisation was then immediately analysed by ¹H NMR which indicated the *cis*-isomer (186) was afforded in 75% yield.

¹H NMR (600 MHz, *d*₄-MeOH): δ 7.14 (2H, H-2a and H-6a, d, J = 8.4 Hz), 7.14 (1H, H-6b), 6.88 (1H, H-2b, s), 6.77 (2H, H-3a and H-5a, d, J = 8.4 Hz), 6.70 (1H, H-5b, d, J = 7.8 Hz), 6.43 (1H, H-7b, d, J = 12.0 Hz), 6.33 (1H, H-8b, d, J = 12.0 Hz), 6.22 (2H, H-10b and H-14b, d, J = 1.8 Hz), 6.15 (1H, H-12a, dd, J = 1.8, 1.8 Hz), 6.11 (2H, H-12b, dd, J = 1.8, 1.8 Hz), 6.04 (2H, H-10a and H-14a, dd, J = 1.8, 1.8 Hz), 5.30 (1H, H-7a, d, J = 9.0 Hz), 4.31 (1H, H-8a, d, J = 9.0 Hz).
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