Naphthopyranones - isolation, bioactivity, biosynthesis and synthesis

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The 1*H*-naphtho[2,3-*c*]pyran-1-one (naphthopyranone) moiety forms the structural framework of a group of secondary metabolites that have been isolated from a range of organisms including fungi, bacteria, lichen and plants. This review documents the known naturally occurring naphthopyranones – their isolation, biosynthesis and biological activity. A survey of methods reported for the synthesis of naphthopyranone natural products is presented.

1. Introduction

Natural products that include a naphthopyranone (1*H*-naphtho[2,3-*c*]pyran-1-one) core structure **A** are widely dispersed in Nature, having been isolated from a variety of sources including fungi, bacteria, lichen and plants. The bioactivity of this group, often the directing force for isolation of these compounds, encompasses antibiotic, cytotoxic, antioxidant, immunoregulatory, antimalarial and antifungal activities amongst the diverse range of identified bioactivities. Given that natural products and natural product-inspired compounds still represent a significant proportion of currently available pharmaceuticals,[1] investigation of this structure class for potential lead-compounds in the development of pharmaceuticals or for use as biochemical probes warrants their ongoing exploration.[2]

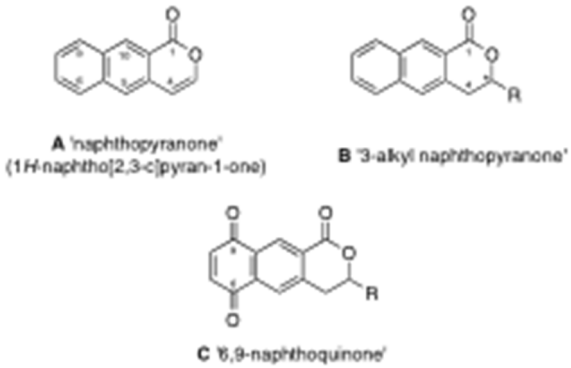

Figure 1: Typical core structural motifs **A**-**C** of naphthopyranone natural products (the numbering depicted in **A** is commonly adopted and is used for consistency throughout this discussion).

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The structural framework **A** defining this group is depicted in Figure 1 and can be seen to consist of a linear tricyclic system incorporating naphthalene and fused δ-lactone (α-pyrone) rings. The structure **A** is an extension of the bicyclic isocoumarin (1*H*-2-benzopyran-1-one) skeleton, the later of which often co-occur with naphthopyranone natural products.[3] It is worth noting that natural products based on the isomeric naphthopyran-4-one (γ-pyrone) structure also regularly occur in nature, a detailed discussion of which is available.[4] Oxygenation in **A** is typically present at the C-7, C-9 and C-10 positions as a consequence of their polyketide biosynthetic origin. More often they exist in the 3,4-dihydro form **B**, with the presence of an alkyl group at C-3 imparting asymmetry to the structures. Oxidation of the naphthalene to a naphthoquinone, as in the 6,9-quinone **C**, is a regularly encountered modification of the parent structure. Furthermore, both symmetrical and unsymmetrical dimerization of naphthopyranones at C-6, C-8 and/or through ether linkages provides further structural and stereochemical diversity within this family. A small number of ring-fused naphthopyranone derivatives, as exemplified by structures **D**-**G** (Figure 2), are known to occur naturally and have been included in the current discussion.

Figure 2: Ring-fused variations **D**-**G** of the naphthopyranone core structure.

Early investigations of naphthopyranones often focussed on the quinone-containing members of the family,[5] likely as a consequence of these pigments being responsible for the colouration displayed by the organism (e.g. fungi) from which they were sourced. Later, with bioactivity-guided isolation of natural products being more prevalent, newer and more diverse members of the naphthopyranone family of natural products have been identified. Not only do members of the family have potential in the area of medicinal chemistry, additionally, their widespread occurrence across a variety species has made them very useful biomarkers in the area of taxonomic identification.

This review provides an overview of known naturally occurring naphthopyranones, exemplifying the structural variety based on the core structures **A**-**G**, along with known bioactivity of members of this group. A discussion of the reported syntheses of members of this group is given with particular emphasis on the use of synthesis for the confirmation of structure, resolution of structural uncertainties such as stereochemistry and for the investigation of biosynthesis. For the purposes of the following discussion members of the naphthopyranone family of natural products have been arbitrarily classified as belonging to one of five sub-groups based on certain structural features that they possess. These are:

i) toralactone-based naphthopyranones ('**A**-type', Fig. 1);
ii) naphthopyranones asymmetric at C-3 ('**B**-type', Fig. 1);
iii) oxidized (quinonoid) naphthopyranones ('**C**-type', Fig. 1);
iv) naphthopyranones coupled to a non-naphthopyranone moiety; and
v) naphthopyranones having extended ring systems ('**D**-**G**-type', Fig. 2).

In addition, naphthopyranones isolated as a result of engineered polyketide synthesis are included as a separate group. Some structures, in particular the heterodimeric and quinonoid species, can be considered to belong to more than one sub-group and, as such, the separation into these groups is merely to assist in the discussion and ease of comparison of these natural products.

2. Isolation and bioactivity

2.1 Toralactone-based naphthopyranones

Toralactone **1** was first isolated in 1973 as a constituent of the seeds of *Cassia tora* L.,[6] then later isolated from *C. obtusifolia* L.[7] and is also found as a metabolite in a cultivated mycobiont of the lichen *Pyrenula* sp.[8] Toralactone **1** represents the quintessential structure of the naphthopyranone group of compounds and itself displays antimicrobial activity.[7] A series of glycosides of toralactone **1** are known, including the 9-*O*-glucoside **2** that has been obtained from *Eriocaulon buergerianum*,[9] *E. ligulatum*[10] and *Paepalanthus chiquitensis*[11] along with numerous other naphthopyranones, flavonoids and xanthones. *Eriocaulon buergerianum* ("Gujingcao") is used in traditional Chinese medicine for its ophthalmic, anti-inflammatory and antimicrobial effects. Toralactone glucoside **2** was shown to have α-glucosidase inhibitory activity (IC_{50} 106.7 μM).[12]

[[STRUCTURES 1-16 TO BE INSERTED HERE]]

From the seeds of *Cassia obtusifolia* L. have been isolated the gentiobioside cassiaside C **3**[13] and cassiaside C_2 **4**.[14] Cassiaside C_2 **4** inhibits histamine release from rat peritoneal exudate mast cells induced by an antigen-antibody reaction. Cassiaside C **3** and cassiaside C_2 **4** also occur in *C. tora* and were shown to possess negligible antibacterial effects against *Pseudomonas aeruginosa* and four strains of MRSA.[15] However, the aglycone toralactone **1** showed significant activity (MIC of 64 μg/mL) against the MRSA strains tested. Accompanying **3** and **4** in *Cassia tora* is a further member of this series of toralactone glycosides, triglucoside **5**, isolated as part of a bioassay-guided study searching for plant compounds

with estrogen-like activity.[16] In comparing a series of natural and synthetic phenolic compounds the estrogen-like activity of cassiaside C **3** was modest, whereas the aglycone toralactone **1** was shown to exhibit significant estrogenic activity. Additionally, synthetically-derived *nor*-toralactone (obtained by cleaving the C-7 methyl ether of toralactone **1**) showed further enhancement of estrogenic activity in an estrogen-dependent MCF-7 cell-proliferation assay and also anti-estrogenic activity in a concentration dependent manner.

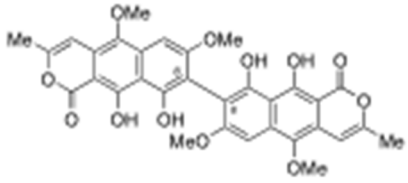
From the roots of *Cassia torosa* CAV., but not in the seeds and seedlings of this plant, were isolated 8-methyltoralactone **6** and the corresponding 10-*O*-methyl ether **7**, along with several phytosterols and anthraquinone pigments.[17] The location of the methyl group at C-8 was established from a series of NOE experiments and derivatization. The cassia plant has a long history as a traditional medicine in Asian countries such as Japan, China and Korea where it has been used as a diuretic, laxative, antiasthenic and for the treatment of conditions such as hepatitis and eye disease. Whilst toralactone **1** and its various glycosides contribute to the activity of cassia seeds and extracts, other components including anthraquinones and hydroanthracenes within the plant may also have important roles.[18]

Isotoralactone **8** was isolated from the seeds of *Cassia obtusifolia* L. and the structure established from spectral analysis and derivatization.[19] Thus, upon acetylation of **8** a diacetate was formed and shown to be identical with the diacetate of toralactone **1**, indicating migration of the double-bond takes place under these conditions. Isomerization of isotoralactone **8** to toralactone **1** could be effected by treatment with acid. Both isotoralactone **8** and toralactone **1** display antimicrobial activity against *Staphylococcus aureus* (MIC 2-3 μg/mL) and *Escherichia coli* (MIC 6-12 μg/mL)[7] and show only moderate inhibitory activity towards cAMP phosphodiesterase.[20]

The chemical constituents of the Eriocaulaceae plant family have been shown to be highly informative in taxonomic studies. In particular, the occurrence of naphthopyranone and flavonoid derivatives in *Paepalanthus* species has allowed these natural products to be used effectively as taxonomic markers for identification of genera in the Eriocaulaceae family.[21] Paepalantine **9** is the main constituent in the capitula of *Paepalanthus bromelioides*, an ornamental plant native to Brazil, and shows antimicrobial activity against various bacteria including *Bacillus cereus* (MIC 7.5 μg/mL), *Staphylococcus aureus* (MIC 7.5 μg/mL) and *S. epidermidis* (MIC 15 μg/mL).[22] Paepalantine **9** has also been isolated from *Paepalanthus vellozioides* and shown to be mutagenic in CHO and McCoy cell assays,[23] cytotoxic in both *in vitro* and *in vivo* mammalian systems[24] and protects mitochondria exposed to oxidative stress conditions by scavenging of superoxide radicals.[25] The capitula of *Paepalanthus bromelioides* also contains the glycosides **10** and **11**,[26] whilst the aerial parts of *P. microphyllus* contains **10** and **12**-**14**.[27] A number of the glycosides of paepalantine have also been assessed for cytotoxic[28] and mutagenic[29] activity.

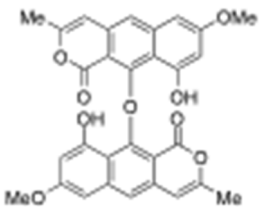
The flowers of *Paepalanthus geniculatus* contain naphthopyranone glycosides **15** and **16**, the 7-de-*O*-methyl analogues of **10** and **12**, along with paepalantine glycosides **11** and **12**.[30] The additional free hydroxy groups in **15** and **16** enhance the radical-scavenging capacity in comparison to **10** and **12**, as measured using the trolox equivalent antioxidant capacity (TEAC) assay.

The C-8/C-8' coupled dehydro dimer **17** of paepalantine has been isolated from *P. bromelioides* and has an IC_{50} of 55.9 μM on colorimetric assay for toxicity.[31] The paepalantine dimer **17** inhibits human kallikrein 5 and 7 serine proteases, the over-expression of which occurs in endocrine-related malignancies, with K_i values of 24.4 μM and 12.2 μM, respectively.[32]



17

Eriocauline **18**, a C-10/C-10' ether-linked dimer of toralactone **1**, has been isolated from the dichloromethane extracts from capitulae of *Eriocaulon ligulatum*,[33] an ornamental flower exported from Brazil. Whilst not possessing an asymmetric centre, circular dichroism (CD) and specific rotation data suggest that eriocauline **18** displays atropisomerism. The dichloromethane extract from *E. ligulatum*, in which there is a prevalence of eriocauline **18**, showed mutagenic activity (Ames test) in one of the four strains of *Salmonella typhimurium* tested.

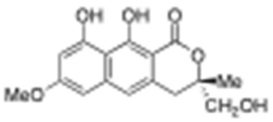


18 eriocauline

Naphthopyranones, particularly toralactone **1** and paepalantine **9** and their various glycosides, along with other co-metabolites such as flavonoids and xanthones have been used as reliable markers for the identification of *Eriocaulon* species using techniques such as high-performance liquid chromatography (HPLC) and mass spectrometry (MS).[34] The rapid throughput and ability to identify known compounds using the combined techniques of HPLC, HRMS, MS/MS and UV-absorption also serves as a valuable tool for dereplication during the search for new bioactive compounds from fungal sources.[35,36]

2.2 Naphthopyranones asymmetric at C-3

Cassialactone **19** was isolated, along with isotoralactone **8**, from the seeds of *Cassia obtusifolia*.[19] The originally proposed structure for cassialactone was revised to **19** on the basis of X-ray analysis of the triacetate derivative.[37] The (*S*)-configuration at C-3 in **19** was established by analysis of the CD spectrum of a *p*-methoxybenzoyl derivative. Cassialactone **19**, and a related dimeric species that incorporates C-8-methyl cassialactone, appear to be the only naturally occurring naphthopyranones that are disubstituted at C-3.



19 cassialactone

(*R*)-Semivioxanthin **20** was first identified as a metabolite of *Penicillium citreo-viride*[38] and the stereochemistry deduced by comparison of the CD spectrum to that of the isocoumarin (*R*)-mellein. (*R*)-Semivioxanthin **20** has also been found in *Aspergillus ochraceus* LCJ11-102 associated with the coral *Dichotella gemmacea*,[39] the endophytic fungus *Aspergillus* sp. HS-05 isolated from leaves of *Huperzia serrata*[40] and also in capitulae of the plant *Eriocaulon buergerianum*.[34] Notably, the enantiomeric species (*S*)-semivioxanthin **24** is also known to occur naturally having been isolated recently from mycelium of *Cryptosporiopsis abietina*,[41] the fungus responsible for stem canker of the Hinoki cypress in Japan. (*S*)-Semivioxanthin **24** showed abscisic activity against Hinoki cypress leaves and inhibited spore germination of the fungus *Cladosporium herbarum*. Semivioxanthin (of unspecified stereochemistry) isolated from an unidentified marine-derived fungus showed immunostimulatory effects resulting from promotion of tumor necrosis factor (TNF)-α production through nuclear factor (NF)-κB and mitogen-activated protein kinase (MAPK) signalling pathways.[42]

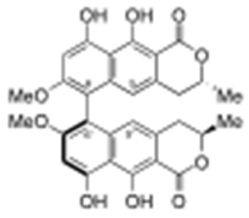
Further structural modification of semivioxanthin, such as glycosylation or biaryl coupling, has commonly taken place prior to isolation. For example, the glycosides **21**-**23** of (*R*)-semivioxanthin occur in leaves of the Brazilian plants *Paepalanthus vellozioides* and *P. latipes*[43] and also in the Chinese plant *Eriocaulon buergerianum*.[9] The (*R*)-stereochemistry at C-3 in **22** and **23** was established by comparison of the CD spectra of the corresponding aglycones with (*R*)-semivioxanthin **20**.

[[STRUCTURES 20-26 TO BE INSERTED HERE]]

From a culture of *Penicillium janthinellum* was isolated 7-de-*O*-methylsemivioxanthin **25**, the (*R*)-stereochemistry being determined by comparison of the CD spectrum with that of (*R*)-semivioxanthin **20**.[44] The polyketide biosynthetic origin of **25** was demonstrated by supplementation of the growing culture with ^{13}C-labelled acetate.

Vioxanthin **26** was first isolated in 1966 from *Trichophyton violaceum*[45] and has since been isolated from *Penicillium citreo-viride*,[38] *P. aurantiogriseum* obtained from cereal samples,[46] *Aspergillus ochraceus*[47] and the thermophilic fungus *Malbranchea pulchella* var. *sulfurea* isolated from compost.[48] Vioxanthin **26** isolated from the later source was labelled 'Tf-26Vx' and shown to be highly active against Gram-positive bacteria (MIC range of 0.045-1.56 μg/mL) and strictly anaerobic Gram-negative bacteria. Initially the (*P*,*R*,*R*)-configuration[49] of vioxanthin **26** obtained from *Penicillium citreo-viride* was proposed by Zeeck and co-workers based on interpretation of the CD spectrum and co-occurrence of (*R*)-semivioxanthin **20**.[38] The CD spectrum of **26** shows a strong positive Cotton effect to longer wavelength and a strong negative one to shorter wavelength that corresponds to 'positive exciton chirality',[50] which in the case of **26** corresponds to a *P*-configuration at the stereogenic axis according to the Prelog-Helmchen rules. This conclusion has been rigorously confirmed by a combination of total synthesis and feeding experiments using ^{13}C-labelled 7-de-*O*-methylsemivioxanthin.[51] Application of the exciton chirality method, or comparison of CD spectra to that of vioxanthin **26**, has routinely been used to deduce the axial chirality of dimeric naphthopyranones.

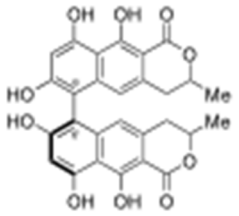
Pigmentosin A **27**, a 6,6'-binaphthopyranone, was obtained from the lichen *Hypotrachyna immaculata* collected in Australia and shown to be the regioisomer of vioxanthin **26**.[52] In this regard, HMBC and HMQC correlations and a significant upfield shift of the H-5 proton signal in the ^{1}H NMR spectrum of **27** due to shielding by the adjacent aromatic ring established the site of connectivity. The recent stereoselective synthesis of pigmentosin A **27** has established the absolute stereochemistry depicted.[53] The synthetic material was shown to inhibit the growth of *Bacillus subtilis* with an MIC of 20 μM. The regioisomers pigmentosin A **27** and vioxanthin **26** have been isolated as co-metabolites in the lichen *Hypotrachyna toiana* found in Malaysia.[54]



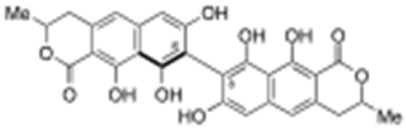
27 pigmentosin A

Another pair of regioisomeric binaphthopyranones found as co-metabolites are aschernaphthopyrones A **28** and B **29**.[55] Isolated from the scale insect pathogenic fungus *Aschersonia paraphysata*, **28** and **29** are the C-7 de-*O*-methyl analogues of pigmentosin A **27** and vioxanthin **26**, respectively. Whilst the axial stereochemistry of aschernaphthopyrones A **28** and B **29** could be determined by interpretation of their CD spectra, the absolute stereochemistry at C-3/C-3' in **28** and **29** is yet to be established. Aschernaphthopyrone A **28** shows moderate

antimalarial activity against *Plasmodium falciparum* K1 with an IC_{50} of 7.3 μM in addition to cytotoxicity against NCI-H187 cells (human small-cell lung cancer) and KB cells (oral human epidermoid carcinoma) with IC_{50} values of 4.5 μM and 17 μM, respectively. Aschernaphthopyrone B **29** showed much weaker activity in similar assays.



28 aschernaphthopyrone A

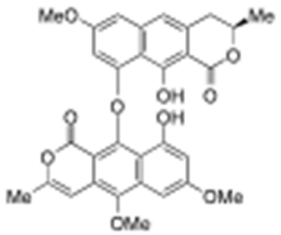


29 aschernaphthopyrone B

The antitumor antibiotics PF1158A **30**, PF1158B **31** and PF1158C **32**, produced in the culture fluids of *Geosmithia argillacea* PF1158, are inhibitors of P388 mouse leukemia cells.[56] The antibiotic compounds **30-32** are novel trimers composed of three semivioxanthin moieties.

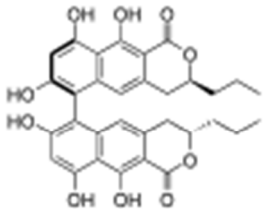
[[STRUCTURES 30-32 TO BE INSERTED HERE]]

The ether bridged naphthopyranone dimer planifolin **33** is found in the capitula of *Paepalanthus planifolius*.[57] The two components of planifolin **33** correspond to semivioxanthin **20** and paepalantine **9**. The stereochemistry at C-3 in the semivioxanthin moiety of planifolin **33** was assumed to be (*R*) due to the presence of various glycosides of (*R*)-semivioxanthin **20** in several other *Paepalanthus* species. Planifolin **33** shows mutagenic activity, as measured using the Ames test, and cytotoxicity to cells of the McCoy mouse fibroblast cell line.[58] The cytotoxicity index (CI_{50}) for planifolin of 12.8 μg/mL compares favourably to cisplatin (CI_{50} 41.9 μg/mL).

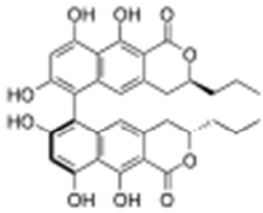


33 planifolin

The atropisomeric talaroderxines A **34** and B **35** were isolated from the ascomycetous fungus *Talaromyces derxii* cultivated on rice and were shown to have strong antibacterial activity against *Bacillus subtilus*.[59] The existence of the talaroderxines as a pair of atropisomers was inferred from the ^{13}C NMR spectrum, which showed discrete doubling of some resonances. The isomers **34** and **35** were separated in the form of their hexaacetyl derivatives and subsequent methanolysis regenerated the individual natural products. The position of the biaryl link in **34** and **35** was established by difference NOE experiments involving the respective hexamethyl ethers. The axial chirality in the talaroderxines A **34** and B **35** was established from their circular dichroism spectra whilst the (*S*)-configuration at C-3 and C-3' was determined by chemical correlation with (*S*)-(+)-3-hydroxyhexanoic acid. Using a combination of in silico screening and bioassay testing, talaroderxines A **34** and B **35** were shown to be inhibitors of botulinum neurotoxin serotype A (BoNT/A) having IC_{50} values of 3.7 μM and 10.1 μM, respectively, in a BoNT/A protease assay.[60] The talaroderxines used in this study were obtained from liquid cultures of a coprophilous *Delitschia* sp., isolated from a sample of kangaroo dung.



34 talarodenxine A

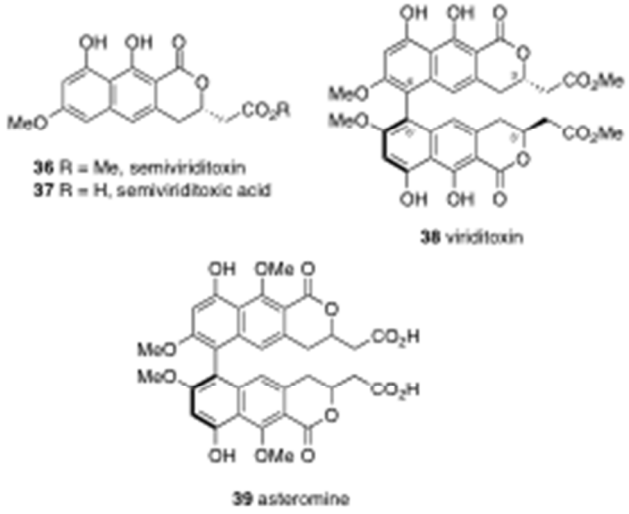


35 talarodenxine B

The fungus *Paecilomyces variotii*, isolated from the larvae of the mountain pine beetle *Dendroctonus ponderosae*, produces the metabolites semiviriditoxin **36** and semiviriditoxic acid **37**,[61] the structures of which were established by using chemical and spectroscopic methods. The (*S*)-stereochemistry was deduced in both cases by comparison of their CD spectra with that of (*R*)-semivioxanthin **20**.

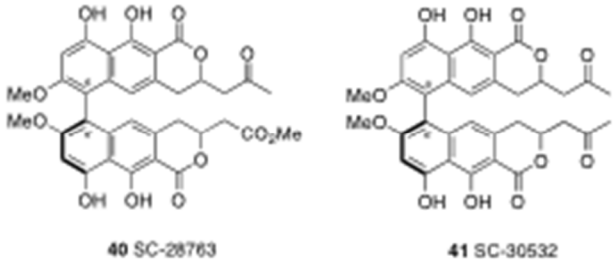
Viriditoxin **38**, the C-6/C-6' dehydro dimer of semiviriditoxin **36**, occurs in several fungi including *Aspergillus viridinutans*,[62] *A. brevipes*,[63] *A. fumigatus*[64] and *Spicaria divaricata*.[65] It was erroneously assigned a C-8/C-8' coupled structure that was subsequently revised on the basis of difference NOE experiments involving viriditoxin tetramethyl ether.[66] These showed correlation between H-8 and both the C-7 and C-9 methoxy groups. The chirality of the C-6/C-6' axis in **38** was established from the CD spectrum. Recently, the absolute configuration at the stereogenic centres C-3 and C-3' in **38** has been determined by total synthesis.[67] Viriditoxin **38** has an LD_{50} of 2.8 mg/kg in mice,[62] and is reported to block the polymerization of FtsZ, a tubulin-like GTPase that has an essential role in bacterial cell division, with an IC_{50} of 8.2 μg/mL.[68] However, a later study has shown that the conditions required to observe inhibition of FtsZ polymerization by

viriditoxin **38** are very specific and that the toxicity of this compound may preclude its further development in drug discovery.[69] Viriditoxin **38** shows activity against a range of bacterial pathogens including methicillin-resistant and methicillin-sensitive strains of *Staphylococcus aureus* (MIC 4-8 μg/mL) and vancomycin-resistant and vancomycin-sensitive *Enterococcus faecalis* and *Enterococcus faecium* (MIC 2-16 μg/mL).[68]



Asteromine **39** was found in a strain of the fungus *Mycosphaerella asteroma*. It is phytotoxic to *Cucumis sativus* (cucumber), weakly antibacterial and antifungal and is lethal to *Artemia salina* shrimp at 0.1 mM.[70] The structure and axial chirality of asteromine **39** was established by spectroscopic analysis of the dimethyl ester, however, the stereochemistry at C-3 and C-3' is not known.

SC-28763 **40** and SC-30532 **41** were isolated from *Spicaria divaricata*,[65,71] along with the related metabolite SC-28762 that is structurally identical to viriditoxin **38**. Originally proposed as having a C-8/C-8' biaryl axis, structural revision to the depicted C-6/C-6' coupled systems **40** and **41** has been proposed in line with the revision of the structure of viriditoxin **38**. Both SC-28763 **40** and SC-30532 **41** inhibited the anaerobic organisms *Clostridium perfringens* (MIC 1-16 μg/mL) and *Trichomonas vaginalis* (MIC 1 μg/mL), SC-28763 **40** also showed modest activity against two strains of *Candida albicans* (MIC 50-75 μg/mL), the plant pathogen *Verticillium albo-atrum* (MIC 50 μg/mL) and the bacterium *Staphylococcus aureus* (MIC 50 μg/mL). Viriditoxin (SC-28762) **38** showed little or no activity against these microorganisms.



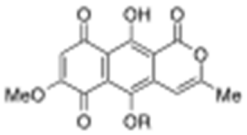
Monapinones A-E **42**-**46** and their co-occurring homo- and heterodimers the dinapinones **47**-**56** were identified as part of a program screening microbial metabolites for inhibitors of triacylglycerol (TG) synthesis using Chinese hamster ovary-K1 (CHO-K1) cells.[72] Isolated from the culture broth of *Talaromyces pinophilus* (originally named *Penicillium pinophilum*) FKI-3864, the monapinones **42**-**46** were only produced when the production medium was supplemented with seawater. The monapinones exhibited weak TG inhibition, whereas dinapinones A2 **48** and AB2 **50** showed the most potent activities having IC_{50} values of 0.65 μM and 1.17 μM, respectively. Interestingly, whilst the IC_{50} values of the individual atropisomers dinapinone A1 **47** and A2 **48** were >12 μM and 0.65 μM, respectively, a 1:1 mixture of **47** and **48** had an IC_{50} value of 0.054 μM. The relative stereochemistry of the hydroxylated C-3 side-chains in **42**-**56** was established from detailed analysis of ^{1}H NMR coupling constants and ROESY experiments whilst the axial configuration was assigned based on the observed Cotton effects in the CD spectra. The 3*S* configurations of **42** and **43** were deduced by comparison of their respective CD spectra with (*R*)-semivioxanthin **20**. The *in vitro* dimerization of **42** using the lysate prepared from *P. pinophilum* FKI-3864 gave **47** and **48**, establishing their absolute stereochemistries.

[[STRUCTURES 42-56 TO BE INSERTED HERE]]

2.3 Quinonoid naphthopyranones

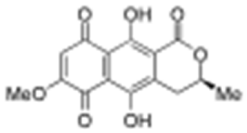
Anhydrofusarubin lactone **57** occurs in several fungi including *Nectria haematococca*[73] and *Fusarium solani*[74] along with several other naphthoquinones. 5-Methoxy-3,4-dehydrosemixanthomegnin **58**, the 5-*O*-methyl ether of anhydrofusarubin lactone **57**, has been isolated from the capitula of *Paepalanthus latipes*.[75] The naphthoquinone **58** shows *in vitro* cytotoxicity against McCoy cells with a CI_{50} value of 35.8 μg/mL, which can be further enhanced by ascorbate-driven redox cycling.[76] Naphthoquinone **58** also exhibits cytotoxicity against murine mammary tumor cells (LM2) and lung adenocarcinoma cells (LPO7) with CI_{50} values of 74.6 and 6.2 μM, respectively.[77] Furthermore, **58** shows anti-inflammatory activity as indicated by suppression of nitric oxide (NO) generation on lipopolysaccharide-stimulated

macrophages, the suppression not being due to direct scavenging of NO.



57 R = H, anhydrofusarubin lactone
58 R = Me, 5-methoxy-3,4-dehydro-semixanthomegnin

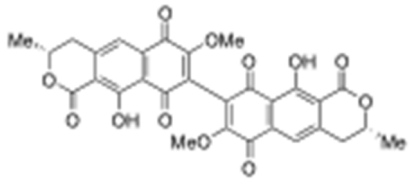
Haemoventosin **59** is a red pigment isolated in 1971 from the lichen *Ophioparma ventosa*.[78] The initially proposed furanonaphthoquinone structure for haemoventosin was subsequently revised, twice, most recently to the quinonoid δ-lactone formulation **59**.[79] The structure **59** evolved from extensive NMR experiments with the monoacetyl derivative of haemoventosin. The (*S*)-stereochemistry at C-3 in **59** was proposed after comparison of the circular dichroism spectrum of 10-*O*-acetylhaemoventosin with that of (*R*)-10-*O*-acetylsemixanthomegnin. The spectra were close to being mirror images of one another and therefore haemoventosin **59** should possess the (*S*)-stereochemistry at C-3.



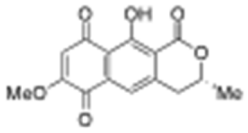
59 haemoventosin

The dimeric pyranonaphthoquinone xanthomegnin **60**, first isolated from the mould *Trichophyton megnini*[80] and subsequently from *T. rubrum*[81] and *T. violaceum*,[82] occurs in many fungi including *Nannizzia cajetani*,[83] *Microsporium cookei*,[84] and various *Penicillium*[38,85-87] and *Aspergillus*[86,88] species. The linear structure of xanthomegnin **60** superseded an earlier angular formulation in 1978 on the basis of ^{13}C NMR experiments[89] and was later confirmed by synthesis from natural semivioxanthin **20**, a co-metabolite of **60** in *Penicillium citreo-viride*.[38] The CD spectrum of xanthomegnin **60** isolated from *P. citreo-viride*, along with the ^{1}H NMR spectrum that shows splitting of the hydroxyl signals, indicates that **60** exists as a mixture of atropisomers. Xanthomegnin is a potent inhibitor of oxidative phosphorylation,[84,90] it inhibits the growth of *Bacillus subtilus* [83] and is a genotoxic carcinogen.[91]

Whilst the occurrence of xanthomegnin **60** is widespread across a range of organisms, the corresponding monomer semixanthomegnin **61** has only recently been found to occur naturally as a metabolite in *Eriocaulon buergerianum*, which also contains glycosides of semivioxanthin **20**.[9] Semixanthomegnin **61** had previously been prepared by total synthesis[92] and by semi-synthesis from semivioxanthin **20** obtained from *Penicillium citreo-viride*.[38] Semixanthomegnin **61** showed an MIC value of 64 μg/mL against a strain of *Staphylococcus aureus*.

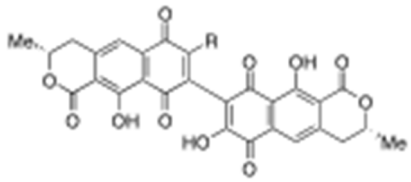


60 xanthomegnin



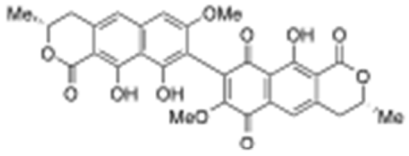
61 semixanthomegnin

Luteosporin **62** and plastatin **63** are phospholipase A_2 inhibitors isolated from *Penicillium chermesinum*, plastatin **63** (IC_{50} 1.6 μg/mL) being the more active of the two.[93] Luteosporin **62** had previously been isolated from *Microsporum cookei* [90,94] and was synthesized as an intermediate *en route* to its 7,7'-di-*O*-methyl ether xanthomegnin **60** from semivioxanthin **20**.[38] Plastatin **63** has been prepared, albeit in very low yield, by ammonolysis of natural xanthomegnin **60** followed by relactonization.[93]

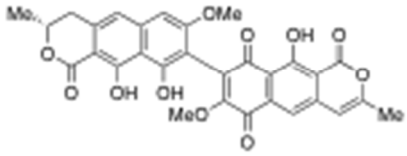


62 R = OH, luteosporin
63 R = NH_2, plastatin

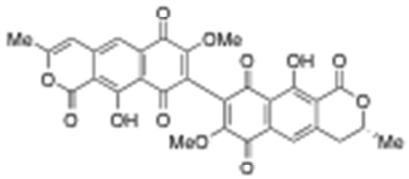
Viomellein **64** was first isolated from *Aspergillus sulphureus* and the structure was established by spectroscopic analysis and comparison with xanthomegnin **60**. Oxidative degradation of **64** and comparison of the CD spectrum of the resulting isocoumarin with that of (*R*)-mellein established the (*R*) absolute configuration for **64**.[88] Viomellein **64** has since been found in *A. ochraceus*,[86] *A. melleus*,[88] *Penicillium viridicatum*,[85] *P. cyclopium*[86] and also in *Nannizzia cajetani*, where it co-occurs with 3',4'-dehydroviomellein **65**, xanthomegnin **60**, 3,4-dehydroxanthomegnin **66** and 3,4,3',4'-bisdehydroxanthomegnin **67**.[83]



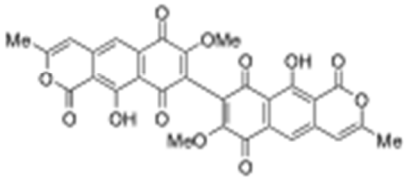
64 viomellein



65 3',4'-dehydroviomellein

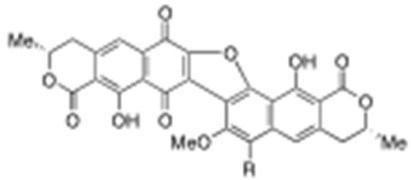


66 3,4-dehydroxanthomegnin



67 3,4,3',4'-bisdehydroxanthomegnin

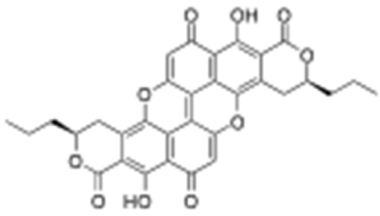
Rubrosulphin **68** and viopurpurin **69** are co-metabolites in *Aspergillus sulphureus*, *A. melleus*[88] and *Penicillium viridicatum*,[85] with viopurpurin **69** also being obtained from *Trichophyton violaceum*[45] The structure of rubrosulphin **68** was established by spectroscopic analysis and confirmed by conversion of viomellein **64** to rubrosulphin **68** on heating in acetone in the presence of potassium carbonate.[88]



68 R = H, rubrosulphin
69 R = OH, viopurpurin

The mycotoxins xanthomegnin **60**, viomellein **64**, vioxanthin **26**, and a number of other related metabolites are produced in characteristic combinations by several fungal species, including *Penicillium aurantiogriseum*, the most common cereal-borne *Penicillium* species of worldwide occurrence.[95] The presence of these compounds and their ease of detection has allowed their use as taxonomic markers in the identification and classification of these fungal species.[96] Due to the economic value of food crops that are susceptible to contamination by mycotoxin-producing organisms,[97] analytical protocols have been developed to assess for the presence and levels of mycotoxins including naphthopyranones, aflatoxins, isocoumarins such as ochratoxin A and numerous other structure classes in a variety of food products and culture extracts.[98]

The green colour of wood infected with the fungus *Chlorociboria aeruginosa* is due to the presence of the extended quinone xylindein **70**. A process for artificially colouring wood with *C. aeruginosa* was patented and oak stained in this way was used for making "Tunbridge ware", a special kind of marquetry. The commercial application of wood stained by *Chlorociboria* sp. is still an area of active interest.[99] Over one hundred years ago Liebermann obtained the pigment in crystalline form for the first time.[100] Structural studies of xylindein **70** were severely hampered by its extremely low solubility in all common organic solvents. In the 1920's Kogl[101] made the first advances in the structure elucidation of xylindein and the complete structure was elucidated, independently, by Todd and co-workers[102] and by Edwards and Kale.[103] The methods relied on derivatization and degradative studies. Recently, X-ray crystallographic analysis of a bis *para*-bromobenzyl derivative of the natural product established that xylindein **70** itself has (3*S*,3'*S*) chirality.[104]



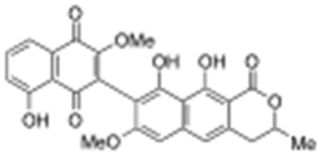
70 xylindein

2.4 Naphthopyranones coupled to non-naphthopyranones

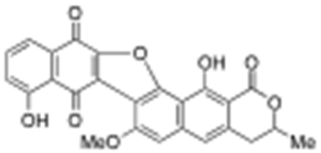
As can been seen from the examples discussed above, both homodimeric and heterodimeric naphthopyranones are commonly encountered. In contrast, only a few naphthopyranones that are coupled to non-naphthopyranone aromatic systems have been isolated.

The naphthoquinone-coupled naphthopyranones xanthoviridicatin D **71** and G **72** were isolated, along with the previously known metabolites xanthomegnin **60**, viomellein **64**, rubrosulphin **68**, and viopurpurin **69**, from the mould *Penicillium viridicatum*.[105] Xanthoviridicatin D **71** differs from viomellein **64** by the absence of a lactone ring in the naphthoquinone moiety. Similarly, xanthoviridicatin G **72**, that contains an additional ether linkage, differs from the co-metabolite rubrosulphin **68** by the absence of one of the lactone rings. Conversion of xanthoviridicatin D to xanthoviridicatin G occurred upon heating in the presence of base in a manner similar to the formation of rubrosulphin **68** from viomellein **64** under these conditions. Later, the isomeric xanthoviridicatins E **73** and F **74** were isolated, through a bioassay-directed isolation protocol, from the fermentation broth of an endophytic strain of *Penicillium chrysogenum*.[106] Xanthoviridicatins E **73** and F **74**

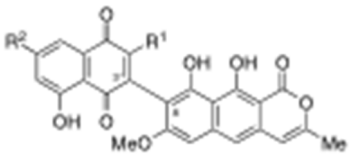
inhibited the cleavage reaction of HIV-1 integrase with IC_{50} values of 6 μM and 5 μM, respectively. Xanthoviridicatins E **73** and F **74** differ from xanthoviridicatin D **71** by having further unsaturation of the lactone ring (i.e. incorporate a toralactone **1** moiety) and additional substitution on the naphthoquinone.



71 xanthoviridicatin D

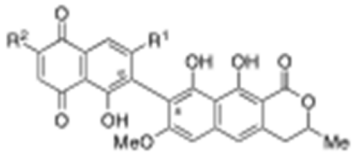


72 xanthoviridicatin G

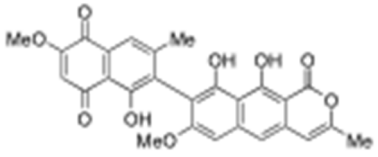


73 R^1 = OMe, R^2 = Me, xanthoviridicatin E
74 R^1 = Me, R^2 = OMe, xanthoviridicatin F

From the culture broth of *Penicillium radicum* strain FKI-3765-2 were isolated the biaryl compounds xanthoradones A-C **75-77**.[107] Interestingly, **75** and **76** possess the dihydronaphthopyranone moiety exhibited by xanthoviridicatin D **71** coupled to the trisubstituted naphthoquinone moiety contained in xanthoviridicatins E **73** and F **74**. However, the biaryl bond in xanthoradones A and B is between C-8 of the dihydronaphthopyranone and C-6' of the naphthoquinone, whereas in the xanthoviridicatins E and F the naphthoquinone moiety connects at C-3'. Xanthoradone C **77** is isomeric to xanthoviridicatin E **73**, differing only by the location of the biaryl bond at C-6' on the naphthoquinone. The nature of the Cotton effect in the CD spectra of xanthoradones A-C is similar to (*M*)-vioxanthin **26**, suggesting **75-77** also have biaryl axes with (*M*)-configuration. Xanthoradones A and B exhibit moderate anti-MRSA activities, having an MIC of 4.0 μg/mL and 2.0 μg/mL, respectively. In combination with the β-lactam antibiotic imipenem, xanthoradones A-C **75-77** reduce the MIC of imipenem from 16 to 0.060, 0.030 and 0.50 μg/mL, respectively, suggesting unsaturation in the lactone is unfavourable for potentiating the activity of imipenem against the MRSA K-24 strain. Xanthoradones A **75** and B **76** show cytotoxicity on Jurkat cells with IC_{50} values of 23.2 μg/mL and 2.6 μg/mL, respectively.

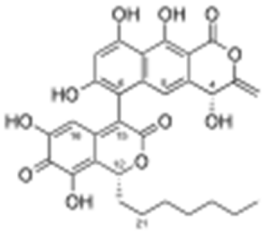


75 R^1 = Me, R^2 = OMe, xanthoradone A
76 R^1 = OMe, R^2 = Me, xanthoradone B



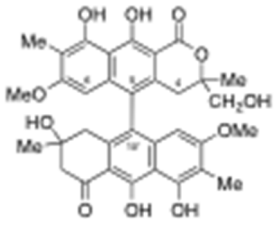
77 xanthoradone C

Cytosporacin **78** was isolated from the fermentation broth of the fungus *Cytospora rhizophorae* ATCC38475 and shown to contain a naphthopyranone linked to an isochromandione moiety.[108] The structure **78**, including the presence of a C-6/C-15 linkage, was established by extensive NMR analysis that also indicated the presence of two rotamers. Upon separation the individual components rapidly re-equilibrated to the original 6:5 mixture of rotamers. With the two aryl systems in **78** lying perpendicular to each other, the relative stereochemistry of the two distant asymmetric centres could be established by the observation of an NOE between H-16 and both H-4 and H-5 for one rotamer, and an NOE between H-5 and both H-16 and H-21 for the other rotamer. A ^{13}C-labelling experiment established the polyketide biosynthetic origin of **78**. Cytosporacin **78** showed modest *in vitro* activity against Gram-positive bacteria.



78 cytosporacin

Torosaol II **79** represents an unusual heterodimeric system that consists of a naphthopyranone half that closely resembles cassialactone **19** (with additional methylation at C-8) and the other component being a dihydroanthracenone (7-methyltorosachrysone).[109] The anisotropic effect resulting from the biaryl bond being located between C-5 and C-10' in **79** results in an upfield shift of signals in the ^1H NMR spectrum of **79** in comparison to the monomer cassialactone **19**. The effect is particularly evident in signals from the C-4 methylene and C-6 aromatic protons, as is observed in related biaryl systems.[110] Isolated from the roots of *Cassia torosa* along with a number of other dihydroanthracenones, torosaol II **79** is reported to have an ED_{50} of 5.5 μg/mL against an *in vitro* KB cell line.

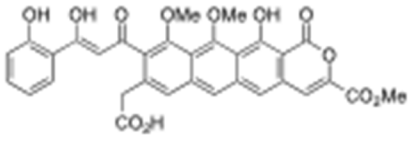


79 torosaol II

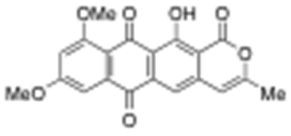
2.5 Naphthopyranones with extended ring systems

Natural products that possess the tricyclic naphthopyranone core structure are the focus of the present discussion, however, it is worth noting that the naphthopyranone moiety can also be incorporated within an extended ring system. In such natural products, ring fusion may be present on the lactone ring or as an extension of the aromatic system, as can be seen by the examples that follow.

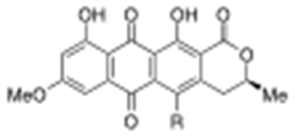
In contrast to the relatively widespread occurrence of natural naphthopyranones, anthrapyranones are rare natural products. Thermorubin **80**, produced by the bacterium *Thermoactinomyces antibioticus*,[111] possesses strong antibiotic properties resulting from blocking the initiation phase of bacterial protein synthesis.[112] The structure **80** was firmly established by single crystal X-ray analysis,[113] and numerous derivatives of thermorubin have been prepared[114] and used for structure activity studies.[115] Thermorubin **80** shows activity against several bacteria including *Staphylococcus aureus* (MIC 0.006 μg/mL) and *Streptococcus pneumoniae* (MIC 0.05 μg/mL).[115] A recently obtained co-crystal structure of thermorubin complexed with the 70S ribosome from *Thermus thermophilus* shows conformational changes in the ribosome that are likely to be responsible for the activity.[116] Thermorubin **80** inhibits aldose reductase in a dose-dependent manner with IC_{50} values of 1.3 nM and 54 nM on rat lens and human aldose reductases, respectively, whilst not inhibiting other reductases.[117] The use of thermorubin **80** and its derivatives as aldose reductase inhibitors for the prevention and treatment of complications resulting from diabetes has been patented.[118]



80 thermorubin



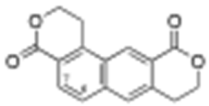
81 anhydroleprolutein



82 R = H, dermolactone
83 R = OH, 5-hydroxydermolactone

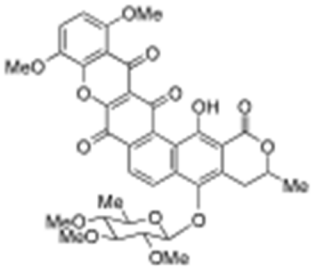
Several toadstools from the subgenus *Leprocybe* of *Cortinarius* contain anhydroleprolutein **81** as a trace constituent,[110] whilst from the Australian toadstool *Dermocybe sanguinea* (sensu Cleland) have been isolated the quinones dermolactone **82** and 5-hydroxydermolactone **83**.[119] The stereochemistry of dermolactone **82** was established by synthesis of the (*S*)-enantiomer **82** and direct comparison of the chiroptical and spectroscopic properties with the natural material. Interestingly, this comparison established that the natural material is an anisochiral mixture in which the (*S*)-enantiomer **82** predominates to the extent of 64 to 36%.[120]

The plant *Swertia mileensis*, used in traditional Chinese medicine to treat viral hepatitis, has been the subject of investigations to identify the active anti-hepatitis B virus (HBV) compounds. As a result of these studies swerilactone E **84** was isolated and shown to possess significant activitiy, based on an anti-HBV assay on the Hep G 2.2.15 cell line *in vitro*, inhibiting the secretion of hepatitis B virus surface antigen (HBsAg) (IC_{50} 0.22 mM) and hepatitis B virus e antigen (HBeAg) (IC_{50} 0.52 mM).[121] Accompanying swerilactone E **84** in *S. mileensis* was the 6,7-dihydro derivative that showed lower activity. The lack of oxygenation in **84** and the additional 8,9-fused lactone ring suggests a novel biosynthetic origin. The biogenesis of more complex members of the swerilactone family has been proposed to involve secoiridoid intermediates.[122]



84 swerilactone E

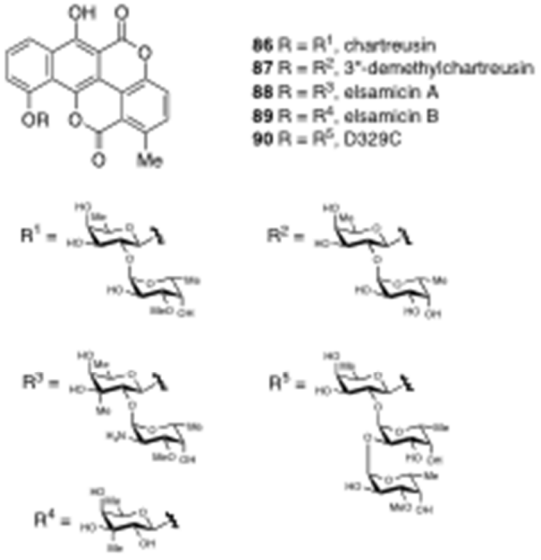
Polycyclic xanthone IB-00208 **85** was isolated from the culture broth of a marine-derived species of *Actinomadura*.[123] The relative stereochemistry of the sugar moiety, but not C-3 of the pyrone ring, could be established by spectroscopic analysis. Along with good activity against Gram-positive organisms (MIC values of 0.09-1.4 nM), IB-00208 **85** exhibits potent cytotoxic activity (MIC 1 nM) against both human (A-549, HT-29 and SK-MEL-28) and murine (P388D1) tumor cell lines.



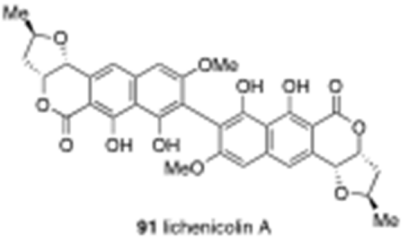
85 IB-00208

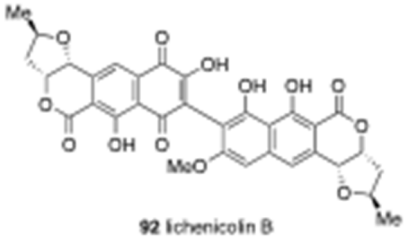
Chartreusin **86** was first isolated in 1953 from the culture broth of soil-derived *Streptomyces chartreusis* and shown to have antibiotic activity,[124] the complete structure being

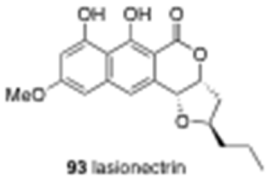
elucidated in 1960.[125] Chartreusin **86** was shown to exhibit excellent antitumor activity against murine P388, L1210 leukemia and B16 melanoma cell lines, however its rapid elimination through the bile and slow gastrointestinal absorption limits its clinical potential.[126] In the search for related compounds with greater therapeutic potential, an unidentified actinomycete strain J907-21 isolated from a soil sample of El Salvador was shown to produce elsamicins A **88** and B **89**.[127] Elsamicin A **88** exhibited greater antitumour activity than chartreusin **86**, attributed to the greater solubility resulting from the presence of an amino sugar, whereas elsamicin B **89** was nearly inactive. Further related compounds have been isolated, including 3''-demethylchartreusin **87** from *Streptomyces chartreusis* that shows similar antitumor activity to chartreusin **86**,[128] D329C **90** from a mutant of *S. chartreusis*,[129] and from marine sediment Streptomycete isolates the 2''- and 4''-monoacetates of chartreusin **86**.[130] Using the combined techniques of biosynthetic engineering and mutasynthesis has given rise to further analogues of potential therapeutic value that are not available readily through synthetic protocols alone.[131] The significant antitumor effects of chartreusin **86** and related structures principally involves binding to DNA[132] but can also involve DNA-cleavage through the production of free radicals.[133] A wealth of pharmacological data has been obtained on chartreusin **86**, elsamicins A **88** and B **89** and related compounds[134] allowing a number of members of this family of anti-cancer agents to proceed to phase II clinical trials.[135]



Lichenicolins A **91** and B **92** were isolated from an unidentified lichenicolous fungus, strain *LL*-RB0668.[136] The structures were determined by NMR analysis with the ^{13}C NMR shift data for lichenicolin A **91** being almost identical to semivioxanthin **20** in the aromatic region except for the C-8 signal, indicating a C-8/C-8' linkage between the equivalent halves. The location of the biaryl linkage was further established from the ROESY spectrum that was also used to establish the *cis* ring junction between the lactone (α-pyrone) and furan rings and the relative orientation of the furan methyl substituent. Lichenicolin B **92**, obtained as a minor isolation product, has undergone oxidation of one of the naphthalene moieties to give the unsymmetrical dimer. Interestingly, lichenicolin A showed moderate to good antimicrobial activity against a range of Gram-positive and Gram-negative bacteria, whereas lichenicolin B was found to be inactive.

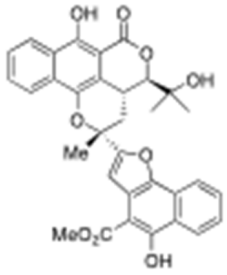






More recently the monomeric furan-fused naphthopyranone lasionectrin **93** has been isolated from a species of *Lasionectria* obtained from forest fern leaf litter in Equatorial Guinea.[137] Bioassay-guided fractionation led to the isolation of **93** that exhibits an IC_{50} value of 11.0 μM against *Plasmodium falciparum Pf*3D7. The relative stereochemistry depicted in **93** was determined by NMR analysis.

From the roots of *Rubia cordifolia* was isolated rubicordifolin **94** that was shown to possess significant cytotoxic activity *in vitro* against Chinese hamster lung (V-79) cells (IC_{50} 4.7 μg/mL), human nasopharynx carcinoma (KB) cells (IC_{50} 1.2 μg/mL) and P388 lymphocytic leukemia cells (IC_{50} 2.9 μg/mL), and *in vivo* antitumor activity against Sarcoma 180 ascites in mice.[138] The relative stereochemistry depicted in **94** was established by NOE analysis of a sample of rubicordifolin prepared during later synthetic studies that also established a likely biosynthetic origin of rubicordifolin **94**, which is found naturally as a racemate.[139]

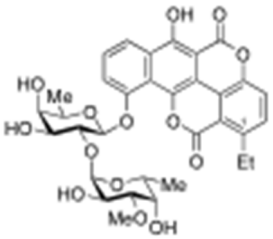


94 rubicordifolin

2.6 Engineered naphthopyranones

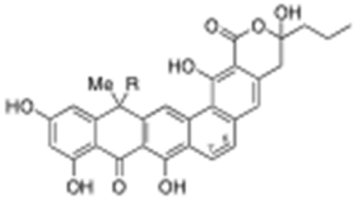
Whilst a number of the naphthopyranones discussed thus far have been isolated after modification of the growing media conditions in which the source organism is cultivated, the emerging area of metabolic engineering has opened up new avenues for the isolation of novel secondary metabolites. In this regard the ability to manipulate polyketide synthase (PKS) gene clusters allows mutants to be produced in a deliberate and controlled manner.[140] By inactivating genes responsible for a specific step in the biosynthesis of polyketide metabolites, intermediates (or shunt products) can be isolated that both give information about biosynthetic pathways and may also have their own beneficial properties. A number of novel naphthopyranones have been isolated using such processes.

The C-1 methyl substituent of chartreusin **86** is derived biosynthetically from an acetyl starter unit being incorporated into the chartreusin PKS. In order to produce the chartreusin analogue homochartreusin **95**, Hertweck constructed a *cha* PKS mutant complimented with genes coding for aclacinomycin biosynthesis that produce a polyketide primed with propionate. As a result, an ethyl substituent is incorporated into homochartreusin **95** at C-1.[131] Homochartreusin **95** and chartreusin **86** showed similar antitumoral and cytostatic activity in K-562 and HeLa tumour cell lines, whereas synthetic norchartreusin that lacks an alkyl substituent at C-1 showed significantly lower activity.



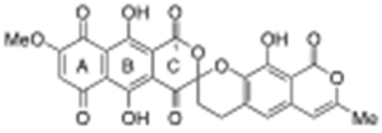
95 homochartreusin

By cloning the gene cluster responsible for the biosynthesis of the benastatins and subsequent deletion of the *benQ* gene, a new group of novel metabolites were formed.[141] Amongst these were benastatins F **96** and H **97** that were accompanied by their respective 6,7-dihydro analogues. Benastatin H **97** showed antiproliferative effects against the L-929 mouse fibroblast cell line (GI_{50} 22.4 μg/mL) and human leukemia cell line K-562 (GI_{50} 24.7 μg/mL) and cytotoxicity against HeLa cells (GI_{50} 22.8 μg/mL). The *benQ* gene determines starter unit selection during biosynthesis of the benastatins and, as such, it was established that the products **96** and **97** likely arise by priming with a butyrate rather than the usual hexanoate primer that is used in production of benastatins from a non-mutated *Streptomyces* sp.



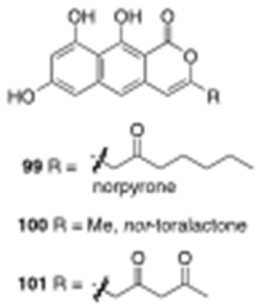
96 R = Me, benastatin F
97 R = OH, benastatin H

In order to gain further insight into how the crucial 5,6-spiroacetal moiety contained in the rubromycin family of compounds is formed biosynthetically, a series of gene-deletion variants of the griseorhodin A gene cluster were constructed. As a result, the biosynthetic intermediate lenticulone **98** was isolated and shown to possess a naphthopyranone moiety in which the C-ring forms part of a 6,6-spiroketal.[142] The key modification in transforming lenticulone **98** to griseorhodin A is excision of the C-1 carbonyl group in **98**, possibly involving a decarboxylative process, to form a 5,6-spiroketal system. Lenticulone **98** showed similar bioactivity to griseorhodin A in antiproliferative assays using K-562 cells (GI_{50} 0.035 μmol/mL), cytotoxicity against HeLa cells (CC_{50} 0.090 μmol/mL) and antibacterial activity against *Staphylococcus carnosus* and *Bacillus subtilis*.



98 lenticulone

By deconstruction and reassembly of the iterative polyketide synthase (IPKS) responsible for biosynthesis of aflatoxin B_1, Townsend showed that production of the usual anthraquinone biosynthetic intermediate could be attenuated in favour of the new shunt product norpyrone **99**.[143] The production of norpyrone **99** occurred in constructed enzymes in which the thioesterase/Claisen cyclase (TE/CLC) domain was not present, favouring spontaneous product release via pyrone formation rather than intramolecular Claisen reaction to form an anthrone product.

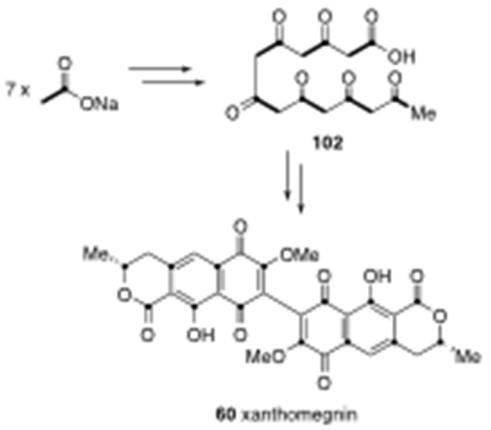


Polyketide synthase CTB1 from *Cercospora nicotianae* is responsible for the first assembly steps towards the biosynthesis of the perylenequinone cercosporin. A reconstructed CTB1 set of domains was shown to produce *nor*-toralactone **100** as the major product.[144] The thioesterase (TE) domain of CTB1 was shown to accelerate pyrone **100** formation possibly by promoting keto/enol tautomerization in the active site leading to release of **100** through enol-lactonization. The starter unit acyltransferase (SAT) domain of CTB1 initiates polyketide extension by utilizing acetyl-CoA selectively, leading to generation of *nor*-toralactone **100** via heptaketide formation. By using 'domain-swapped' CTB1 that incorporates a hexanoyl starter unit an analogue of *nor*-toralactone **100** was produced and proposed to be the naphthopyranone **101**.[145] As evidenced by the examples outlined above, the ability to engineer the enzymes responsible for polyketide biosynthesis provides exciting opportunities to custom design new polyketide-derived products, including novel naphthopyranones.

3. Biosynthesis

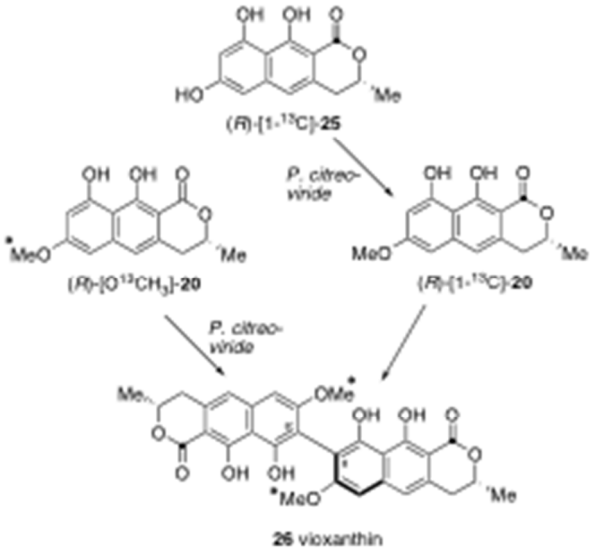
The methods that have been used to investigate the biosynthesis of naphthopyranones somewhat reflect the techniques available at the time of their isolation, developing in their complexity in parallel with advances in the field of polyketide biosynthesis.[146] Early investigations principally relied on isotopic labelling experiments, whilst more recent studies have utilized engineered enzymes to probe biosynthetic pathways, including the type II PKS genes and enzymes responsible for the synthesis of aromatic polyketides.[147]

The biosynthesis of xanthomegnin **60** has been studied by feeding sodium [1-^{13}C]- and [1,2-$^{13}C_2$]-acetate to *Aspergillus melleus*.[148] The ^{13}C-enriched xanthomegnin **60** so obtained (Scheme 1) showed enhancement of seven of the fourteen carbon signals in the ^{13}C NMR spectrum consistent with the homodimeric structure, the enhanced signals corresponding to alternate carbons as would be expected for a regular heptaketide **102** assembly. Labelled viomellein **64** was also isolated and showed it to be derived from fourteen intact acetate units.



Scheme 1: Incorporation of sodium [1-^{13}C]-acetate into xanthomegnin **60**.

The process by which formation of dimeric naphthopyranones takes place is itself an interesting aspect of the biosynthesis of these natural products. To investigate this Müller prepared the ^{13}C-labelled naphthopyranones (*R*)-[1-^{13}C]-**25** and (*R*)-[O^{13}CH$_3$]-**20** (Scheme 2).[51] The ^{13}C-labelled substrates **25** and **20** were then used in feeding experiments with *Penicillium citreo-viride* resulting in the isolation of double-labelled vioxanthin **26** in both cases. This observation established that C-7 *O*-methylation of **25** to produce semivioxanthin **20** takes place before regio- and atroposelective phenolic coupling to form the C-8/C-8' dimer, vioxanthin **26**.



Scheme 2: Incorporation of ^{13}C-labelled substrates **20** and **25** into vioxanthin **26**.

It would be anticipated that other naphthopyranones originate from a similar folding pattern during their biosynthesis to that elucidated for xanthomegnin **60** and vioxanthin **26**. Direct evidence to confirm this proposition has been obtained for 7-de-*O*-methylsemivioxanthin **25** (Figure 3).[44] In the case of cytosporacin **78**, incorporation of [1,2-$^{13}C_2$]-acetate confirmed that the naphthopyranone moiety is formed

from a heptaketide in the usual fashion and the isochromandione is derived from an octaketide.[108] It is pertinent to note that for the monapinones **42**-**46**, and their corresponding homo- and heterodimers, the C-3 alkyl substituent consists of a C_7 or C_9 chain length, consistent with incorporation of an additional acetate group for some members. A similar observation can be made for the lichenicolins A **91** and B **92** that have a C_3 unit at C-3, whereas the monomeric lasionectrin **93** that bears a close structural resemblance but is isolated from a different organism has a C_5 unit at C-3.

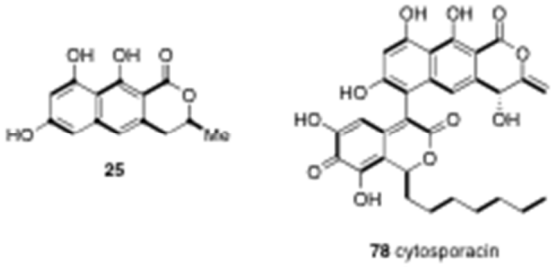
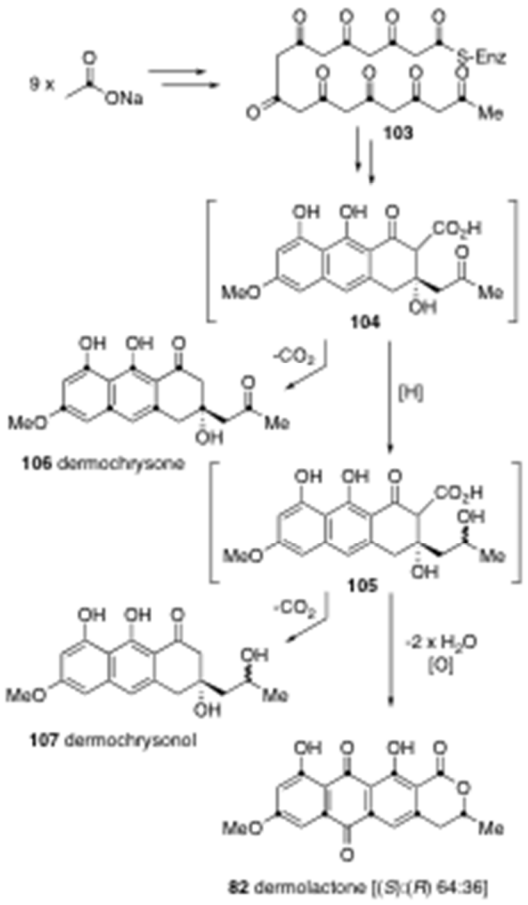


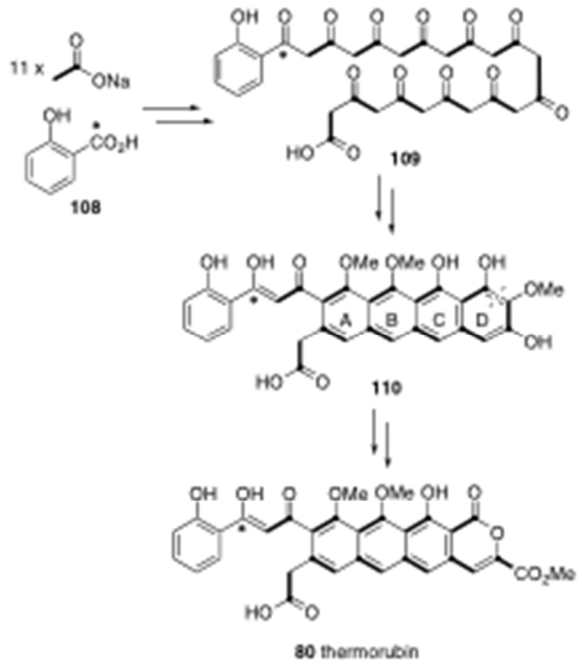
Figure 3: Incorporation of labelled acetate ([1-^{13}C] or [1,2-$^{13}C_2$]) into naphthopyranones.

Dermolactone **82** was shown to originate from a nonaketide precursor **103** (Scheme 3) by incorporation of [1-^{13}C]-acetate into the product **82**.[119] Whilst dermochrysone **106**, a co-metabolite of dermolactone **82** in *Dermocybe sanguinea* (sensu Cleland), has (*S*) stereochemistry, dermochrysonol **107** occurs as a mixture of diastereoisomers epimeric at the secondary alcohol stereocentre. This is consistent with the isolation of dermolactone **82** as an unequal mixture of enantiomers, suggesting that reduction of **104** to **105** occurs non-stereoselectively.



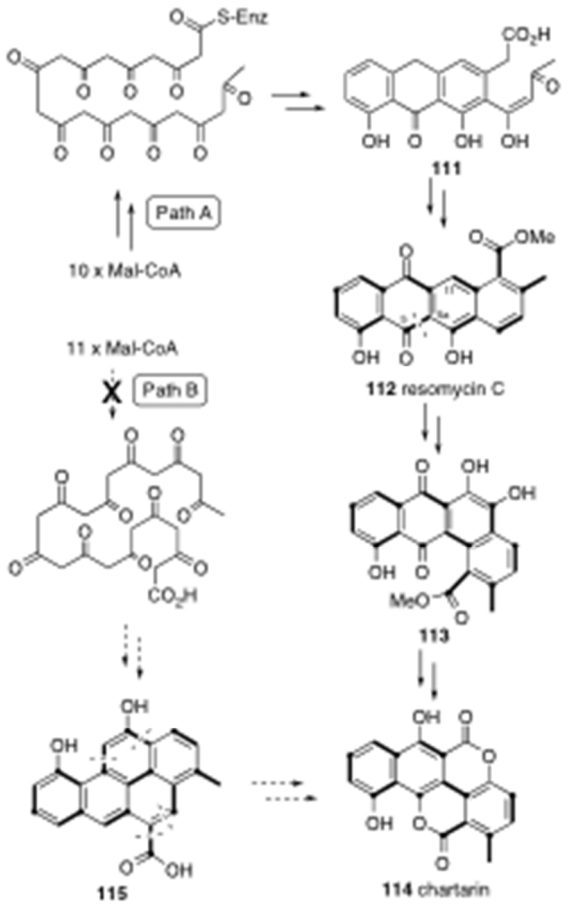
Scheme 3: Incorporation of [1-^{13}C]-acetate and proposed biosynthesis of dermolactone **82**.

A number of naphthopyranones require some modification to the regular linear folding pattern and condensation process seen in the examples above. The biosynthesis of thermorubin **80** was postulated, amongst other possibilities, to be based on the combination of an undecaketide and salicylic acid.[113] Direct evidence for the biosynthetic origin of thermorubin **80** was obtained by Aragozzini and co-workers by feeding *Thermoactinomyces vulgaris* with [1-^{13}C]-, [2-^{13}C]- and [1,2-$^{13}C_2$]-acetate.[149] Incorporation of the labelled acetate (Scheme 4) showed that the A-D rings in **80** originate from an undecaketide chain **109**, the lactone being formed by a Woodward oxidative fission of ring D in the proposed intermediate **110**. Labelled salicylic acid **108** was also incorporated into **80** (73% isotopic enrichment of C-3'), accounting for the origin of the additional phenyl ring, salicylic acid being available naturally via the shikimate pathway.



Scheme 4: Incorporation of [1-^{13}C]-, [1,2-$^{13}C_2$]-acetate and ^{13}C-labelled salicylic acid **108** into thermorubin **80**.

Early investigations into the biosynthesis of chartreusin **86** showed that the aglycone chartarin **114** is derived from acetate, however, clearly the end product **114** (Scheme 5) is not formed directly from folding of a polyketide intermediate and has instead undergone rearrangement. Feeding experiments using [1-^{13}C]-, [2-^{13}C]- and [1,2-$^{13}C_2$]-acetate identified the origin of each carbon in the skeleton of **114** and led to the initial proposal that chartreusin **86** is formed via an angularly folded undecaketide intermediate **115** (Path B).[150] A revised biosynthesis of chartreusin **86** involving the more conventional linearly fused decaketide **111** (Path A) has been proposed after a detailed analysis, by pathway dissection, of the genes involved.[151] An engineered mutant lacking the *chaZ* gene produces the known anthracyclic polyketide resomycin C **112** in lieu of chartreusin **86**. Resomycin C **112** may undergo C-5/C-5a bond cleavage followed by bond formation between C-5/C-11 to give angular intermediate **113**. Loss of 'C-6', possibly by oxidation, and lactone formation would give chartarin **114**.



Scheme 5: Biosynthesis of chartarin **114**, the aglycone of chartreusin **86**.

A more detailed knowledge of the mechanisms involved in the biosynthesis of polyketides, including those leading to a selection of naphthopyranones, is beginning to emerge through the use of gene manipulation. As a representative example, *nor*-toralactone **100** is formed through the involvement of a non-reducing polyketide synthase (NR-PKS). Such systems are typically characterised by a set of six catalytic domains (Scheme 6); the starter unit acyltransferase (SAT), ketoacyl synthase (KS), malonyl acyltransferase (MAT), product template (PT), acyl-carrier protein (ACP) and thioesterase (TE) domains. By constructing the CTB1 domain responsible for the first assembly steps of cercosporin, *nor*-toralactone **100** could be isolated.[144] The biosynthesis proceeds along a typical pathway of starter unit selection (controlled by SAT), chain extension, regioselective aldol cyclization controlled by the PT domain and product release from TE. Product release from the enzyme may occur by hydrolysis to form carboxylic acid **116** followed by hemiketal formation and dehydration to give pyrone **100**. However, selective labelling of the polyketide starter unit by use of [^{18}O]acetyl-CoA led to formation of ^{18}O-labelled *nor*-toralactone **100** indicating that product release occurs by TE-catalysed pyrone formation (enol-lactonization).

[[SCHEME 6 TO BE INSERTED HERE]]

Scheme 6: Biosynthesis of *nor*-toralactone **100** by the PKS domain of CTB1.

In the case of *nor*-toralactone **100** the TE domain is crucial to catalyse product release and resultant pyrone formation.[144] In other systems, such as the constructed NR-PKS responsible for the synthesis of aflatoxin, the absence of the thioesterase/Claisen cyclase (TE/CLC) domain can lead to pyrone formation as a shunt product in lieu of the usual formation of an anthrone product that results from product release via a Claisen reaction.[143] Similarly, the biosynthetic mechanisms involved in the formation of naphthopyranones such benastatins F **96** and H **97**,[141] lenticulone **98**[142] and chartreusin **86**[131] are understood in intimate detail, from how PKS primer units are chosen, individual roles of genes for control of polyketide chain length and folding pattern, and subsequent modifications such as alkylation, oxygenation and glycosyl transfer. This developing field in which polyketide synthesis is manipulated at the gene level has enormous potential and could conceivably lead to the ability to undertake de novo synthesis of polyketide products using designed enzymes.

4. Synthesis

4.1 Semisynthesis

Despite the common occurrence of naphthopyranones as natural products the number of reported syntheses of members of this group is limited. A selection of naphthopyranones have been obtained by semisynthesis whereby one naphthopyranone, isolated from a natural source, has been converted to a closely related natural product. Examples of this are prevalent in the early investigations of quinonoid-containing naphthopyranones. In particular, studies by Zeeck and co-workers in 1979 were crucial in verifying the structure of xanthomegnin **60** by its synthesis from a co-metabolite.[38] Thus, oxidation using Fremy's salt of (*R*)-semivioxanthin **20** (Scheme 7), obtained from *Penicillium citreo-viride*, gave semixanthomegnin **61** that was not itself identified as a natural product until 2008. Demethylation of **61** followed by dimerization using persulphate gave luteosporin **62** that upon methylation delivered xanthomegnin **60** that was shown to be identical to natural xanthomegnin. Xanthomegnin **60** has itself been converted, in low yield, to plastatin **63** by treatment with ammonium hydroxide and relactonization.[93] Finally, viomellein **64** obtained from *Aspergillus sulphureus* was shown to undergo conversion to rubrosulphin **68** upon heating with potassium carbonate, thus supporting the assigned structures of these co-metabolites.[88] In combination, these studies have proved particularly useful in providing confirmation of the structural relationships between this group of natural products.

[[SCHEME 7 TO BE INSERTED HERE]]

Scheme 7: Interconversion of quinonoid naphthopyranones through semisynthesis.

The majority of total syntheses of naphthopyranones that have appeared employ either a biomimetic strategy, involve an annulation strategy based on a tandem Michael addition-Dieckmann condensation reaction or use a Diels-Alder cycloaddition approach. The use of these methods in the total synthesis of naphthopyranone natural products will be discussed in the following sections.

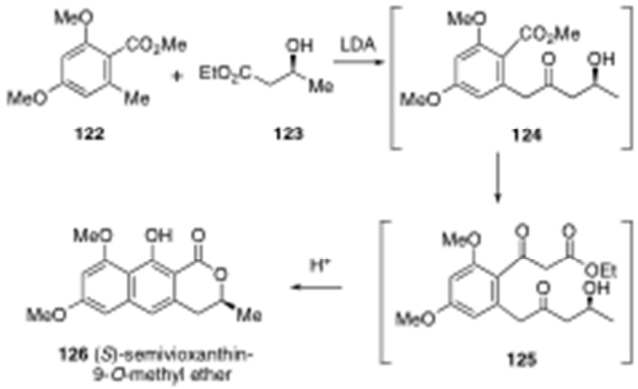
4.2 Biomimetic approaches

The first synthesis of (±)-semivioxanthin **20** involved the intramolecular condensation of a synthetic polyketide.[152] It began from ketal-protected diethyl β-oxoglutarate **117** that was reacted with two equivalents of the dianion from *tert*-butyl acetoacetate to give the polyketide **118** (Scheme 8). With calcium diacetate intermediate **118** suffered a series of intramolecular condensations to give **119**. A series of protection and deprotection steps lead to the diester **120**, which was subjected to a selective Claisen reaction with the enolate from *tert*-butyl acetate to give the keto-ester **121**. Removal of the *tert*-butyl ester in **121**, decarboxylation and final reduction gave (±)-semivioxanthin **20** in a yield of 4% over the ten steps.

[[SCHEME 8 TO BE INSERTED HERE]]

Scheme 8

A concise synthesis of (*S*)-semivioxanthin-9-*O*-methyl ether **126** has been developed in which ethyl (*S*)-3-hydroxybutanoate **123** is the source of chirality.[153] Thus, treatment of methyl orsellinate dimethyl ether **122** with LDA generates the benzylic anion, with subsequent exposure to ester **123** giving (*S*)-semivioxanthin-9-*O*-methyl ether **126** in 35% yield (Scheme 9). The formation of **126** requires reaction between **122** and two molecules of ethyl (*S*)-3-hydroxybutanoate **123**, firstly forming **124**. The likely partially condensed polyketide intermediate **125** may be formed by reaction of **124** with the anion of ethyl acetate, generated by retro aldol reaction of **123**, or by direct reaction between **123** and **124** followed by loss of acetaldehyde.



Scheme 9

During synthetic studies of *Rubiaceae* natural products the facile preparation of rubicordifolin **94** was achieved by treating the postulated biosynthetic intermediate vinyl naphthoquinone **127** with phenylboronic acid (Scheme 10).[139] Under these conditions naphthoquinone **127** undergoes reversible cyclization to form *ortho*-quinone methide **128**, calculations showing this to be a viable process. Naphthofuran **129**, also formed from **127** via cyclization and dehydration, then undergoes hetero-Diels-Alder cycloaddition in combination with **128** to lead ultimately to rubicordifolin **94**. Favourable π-stacking in the transition state during cycloaddition and the steric bulk of the hydroxyisopropyl group in **128** appear to control the diastereoselectivity of the reaction. Prenylated naphthoquinones related to **127** are found in *Rubiaceae* sp. and, as such, this pathway may account for the occurrence of rubicordifolin **94** naturally as a racemate.

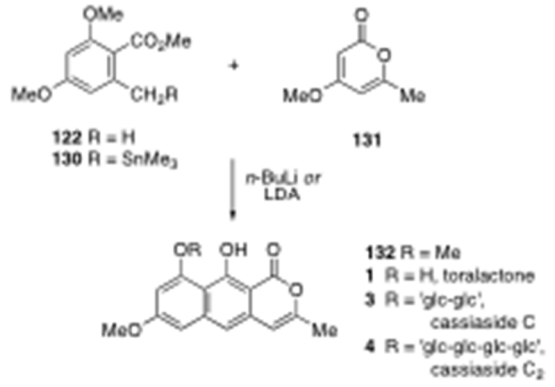
[[SCHEME 10 TO BE INSERTED HERE]]

Scheme 10

4.3 Annulation approaches

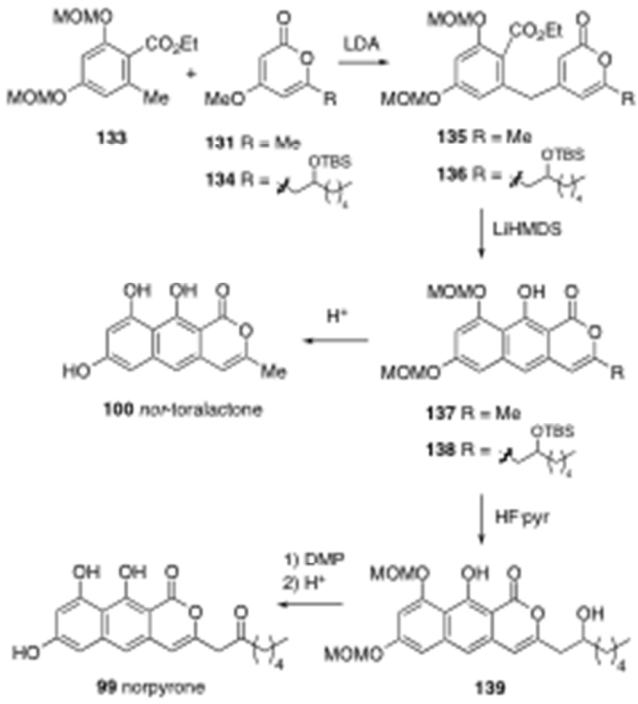
The Staunton-Weinreb annulation procedure is ideally suited to the rapid construction of naphthopyranones and has thus become a standard method for the preparation of this class of compounds, both in the synthesis of naturally occurring naphthopyranones and for the preparation of versatile naphthopyranone intermediates *en route* to other structure classes.[154] Examples of the use of this approach for the synthesis of naturally occurring naphthopyranones will be discussed in this section.

Developed by Staunton for the synthesis of toralactone **1**, the Staunton-Weinreb annulation involves the generation of an *ortho*-toluate anion that is able to undergo a tandem Michael addition-Dieckmann condensation with α,β-unsaturated lactones.[154] In the case of toralactone **1** the stannylated orsellinate **130** and the lactone **131** were used (Scheme 11).[155] Firstly, deprotonation of the orsellinate **122** with LDA and trapping of the resultant anion gave the trimethylstannyl derivative **130**. Subsequent treatment of **130** with *n*-butyllithium generated the anion of **122** that underwent a tandem Michael-Dieckmann reaction with the α,β-unsaturated lactone **131** to give toralactone-9-*O*-methyl ether **132**. Direct reaction of the lactone **131** with the anion of **122** generated from LDA gave the same product **132** but in lower yield. Subsequently, the ether **132** has been converted to toralactone **1** by selective ether cleavage using boron tribromide.[156] More recently, toralactone **1** prepared in this way was then converted to cassiaside C **3** and cassiaside C_2 **4** by selective glycosylation of the C-9 hydroxy group.[157]



Scheme 11

The syntheses of *nor*-toralactone **100** and norpyrone **99** have been undertaken using a similar approach to that used in the synthesis of toralactone **1**. The anion formed from orsellinate **133** (Scheme 12) underwent addition to lactone **131**, quenching the reaction at low temperature leading to isolation of the addition-elimination product **135**. Further base treatment completed formation of the naphthopyranone **137** and subsequent exposure to acid gave *nor*-toralactone **100**.[144] In an analogous fashion reaction between orsellinate **133** and lactone **134** gave **136** that was not isolated but instead immediately converted to naphthopyranone **138**. After silyl group removal to form **139**, oxidation and protecting group removal took place efficiently to give norpyrone **99**.[143] The availability of synthetic *nor*-toralactone **100** and norpyrone **99** was important for their use as standards for comparison and identification of products isolated from engineered PKS-containing organisms.



Scheme 12

A concise synthesis of (±)-semivioxanthin **20**, reported by Deshpande, involves a tandem Michael-Dieckmann reaction between orsellinate **140** and lactone **141** (Scheme 13).[158] Removal of the protecting group gave semivioxanthin **20** that, upon oxidation, delivered semixanthomegnin **61**. More recently this approach was adapted to a stereoselective synthesis of both (*R*)- and (*S*)-semivioxanthin.[159] For the synthesis of (*R*)-semivioxanthin **20** (Scheme 14), enzymatic reduction of *tert*-butyl 3,5-dioxohexanoate afforded *tert*-butyl (*R*)-5-hydroxy-3-oxohexanoate **142** in 99.4% e.e.. The ester **142** was subsequently lactonized and then converted to the enol methyl ether (*R*)-**141**. Tandem Michael-Dieckmann reaction between the orsellinate **143** and the lactone (*R*)-**141** followed by protecting group removal then gave (*R*)-semivioxanthin **20**. (*S*)-Semivioxanthin **24** (97% e.e.) was prepared similarly beginning from *tert*-butyl (*S*)-5-hydroxy-3-oxohexanoate *ent*-**142**, which was processed in a similar manner to the (*R*)-enantiomer. The ^{13}C-labelled naphthopyranones (*R*)-[1-^{13}C]-**25** and (*R*)-[O^{13}CH$_3$]-**20** (Scheme 2), used to investigate the biosynthesis of vioxanthin **26** in *Penicillium citreo-viride*, were prepared using a similar strategy to that depicted in Scheme 14.[51]

[[SCHEME 13 TO BE INSERTED HERE]]

Scheme 13

[[SCHEME 14 TO BE INSERTED HERE]]

Scheme 14

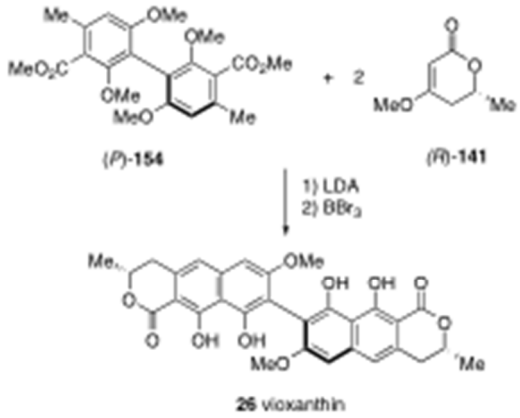
Since a variety of readily available substituted orsellinates have been shown to react with α,β-unsaturated lactone electrophiles,[154,160] the ability to access the chiral lactone electrophilic component efficiently is an important consideration in applying the above strategy to a range of naphthopyranone targets. To this end, it has been shown that lactones such as (*R*)-**141**, **147** and **148** are readily available in two steps from terminal epoxides **144**-**146** (Scheme 15). The simplest of these lactones, methyl substituted lactone (*R*)-**141**, underwent Staunton-Weinreb annulation with orsellinate **149** to form **150** with final deprotection delivering (*R*)-semivioxanthin **20** in 17% yield over four steps from (*R*)-propylene oxide **144**.[161] Beginning with racemic propyl oxirane **145** the C-3 propyl-substituted naphthopyranone **152** was also available in four steps (17% yield).[162] Naphthopyranone **152** constitutes one half of the symmetrical dimers talaroderxines A **34** and B **35** and also a potential precursor to xylindein **70**. For the first total synthesis of (*S*)-semiviriditoxin **36**, chiral epoxide **146** was used to prepare lactone **148** that underwent Staunton-Weinreb annulation to generate naphthopyranone **153**. Straightforward conversion to (*S*)-semiviriditoxin **36** was then effected. Spectroscopic comparison of synthetic (*R*)-semivioxanthin **20**, synthetic (*S*)-semiviriditoxin **36** and the reported data for naturally occurring **36** confirmed the (*S*)-stereochemistry of semiviriditoxin **36** from the fungus *Paecilomyces variotii*.[161]

[[SCHEME 15 TO BE INSERTED HERE]]

Scheme 15

With numerous naphthopyranone natural products occurring as dimers, synthetic methods for accessing such structures have been developed. Two general methods can be envisaged for the preparation of dimeric systems using the Staunton-Weinreb annulation, firstly, oxidative biaryl coupling of a monomeric naphthopyranone precursor, and secondly, a double annulation of a bis-orsellinate precursor. Both of these methods have been applied to the synthesis of homodimeric naphthopyranononones.

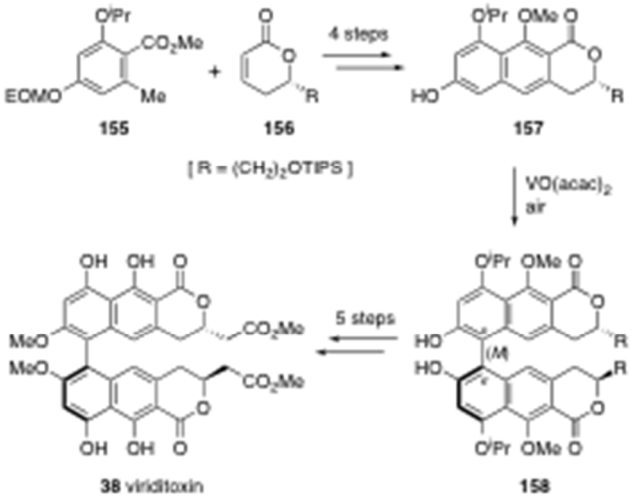
The enantioselective synthesis of both (*M*,*R*,*R*)- and (*P*,*R*,*R*)-vioxanthin **26** was undertaken by Müller as part of an investigation of the biosynthesis of biarylic secondary metabolites.[51] For the synthesis of (*P*,*R*,*R*)-vioxanthin **26** (Scheme 16), atropisomerically pure bis-orsellinate (*P*)-**154** was used. A double Staunton-Weinreb annulation with lactone (*R*)-**141** followed by selective deprotection gave the dimeric naphthopyranone (*P*,*R*,*R*)-vioxanthin **26**. Similarly, the atropisomer (*M*,*R*,*R*)-vioxanthin could be prepared from bis-orsellinate (*M*)-**154**. Access to both (*M*,*R*,*R*)- and (*P*,*R*,*R*)-vioxanthin **26** by synthesis was crucial for confidently assigning the absolute stereochemistry of natural vioxanthin **26**.[51]



Scheme 16

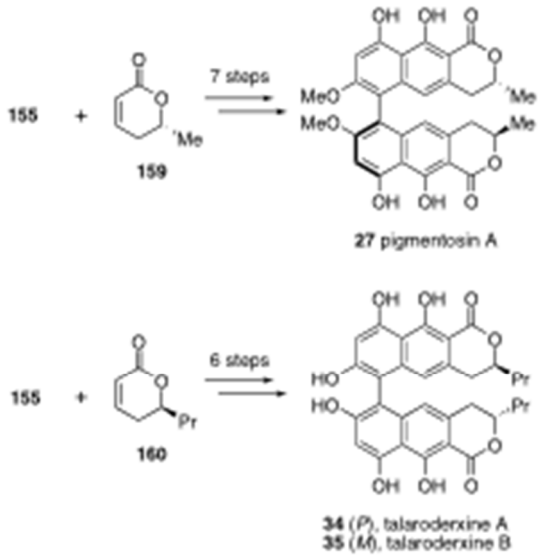
For the synthesis of viriditoxin **38**, Shaw used a vanadium-catalyzed naphthopyranone dimerization procedure as the key step in forming the C-6/C-6' homodimeric system (Scheme 17).[67] In this approach the required naphthopyranone intermediate **157** was prepared by annulation between orsellinate **155** and lactone **156** followed by protecting group manipulation. The β-naphthol **157** then underwent oxidative coupling using VO(acac)$_2$ to give the naphthopyranone dimer **158**, with the remote stereocentres exerting diastereocontrol to produce the desired atropisomer (*M*)-**158** (dr 76:24). Improved

atroposelectivity (dr 89:11) was obtained using a chiral BINOL-derived vanadium catalyst. A further five steps from **158** completed the first synthesis of viriditoxin **38** and established the absolute configuration of the natural product.



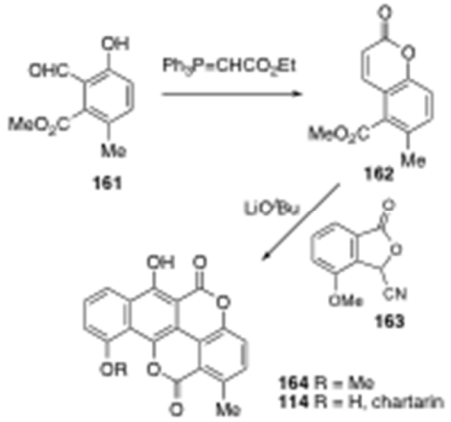
Scheme 17

This methodology has been further adapted to the synthesis of the dimeric naphthopyranones pigmentosin A **27** and talaroderxines A **34** and B **35** (Scheme 18).[53] Thus, annulation between orsellinate **155** and the methyl-substituted lactone **159** leads, ultimately, to pigmentosin A **27**. The corresponding propyl-substituted lactone **160** was reacted similarly giving rise to each of the talaroderxines A **34** and B **35**. In the later case the choice of chiral vanadium catalyst used for the phenolic coupling enabled effective control of the axial stereochemistry to deliver the desired atropisomeric product.

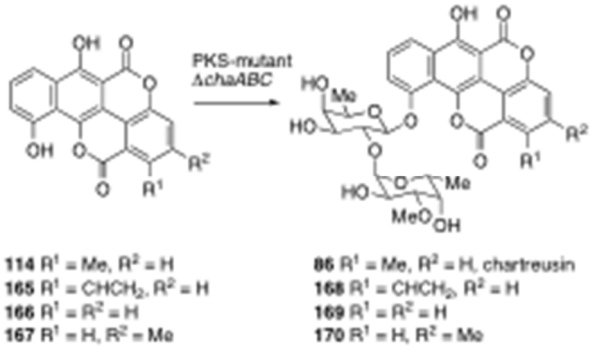


Scheme 18

The synthesis of chartarin **114**, the aglycone of chartreusin **86**, has been undertaken by a number of groups.[163] An efficient synthesis of chartarin **114** from benzaldehyde **161** has been achieved by Mal and co-workers[164] in which a Hauser-Kraus annulation between coumarin **162** and cyanophthalide **163** (Scheme 19) was used to rapidly assemble the benzonaphthopyranone core **164**, final demethylation providing chartarin **114**. A similar strategy was used by Hertweck to prepare chartarin **114** and the analogues **165-167** (Scheme 20).[131] When synthetic chartarin **114** and **165-167** were added to the heterologous host *Streptomyces albus*, a chartreusin (*cha*) PKS null mutant not capable of producing chartarin **114**, chartreusin **86** and analogues **168-170** were isolated successfully. Vinylchartreusin **168** showed slightly lower activity against K-562 and HeLa tumour cells when compared to chartreusin **86**, however upon photoactivation a 12-fold increase in the activity of **168** was shown (GI_{50} 0.6 μM) in a colon adenocarcinoma cell line (HT-29), resulting from [2 + 2] photo adduct formation between vinylchartreusin **168** and DNA. Notably, norchartreusin **169** showed a significant loss in cytotoxic activity, whereas chartreusin regioisomer **170** retained activity. As demonstrated in the preparation of chartreusin **86**, the modern approach of linking chemical synthesis with biosynthesis (mutasynthesis) has vast potential for accessing analogues not readily obtainable either through total synthesis or derivatization of natural products.



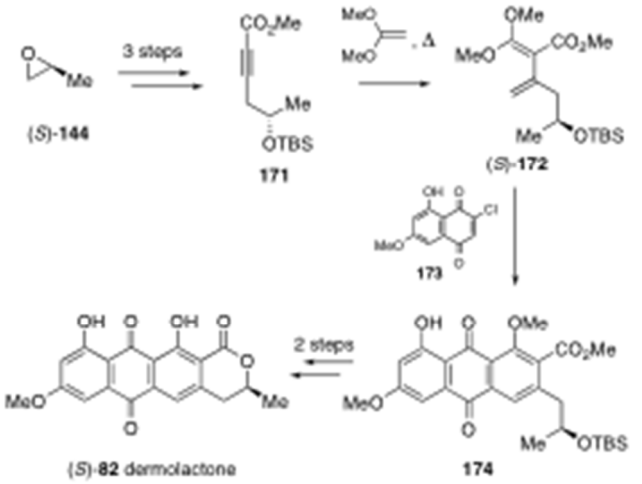
Scheme 19



Scheme 20

4.4 Diels-Alder approaches

Whilst quinonoid-containing naphthopyranones have been prepared by oxidation of the corresponding naphthol, quinonoid compounds are also suited to synthesis directly using a Diels-Alder approach. To establish the stereochemistry of dermolactone **82**, its stereoselective total synthesis was undertaken. The reaction sequence that leads to (*S*)-dermolactone **82** is summarized in Scheme 21.[120] Thus, reaction between (*S*)-propylene oxide **144** and lithium acetylide gave (*S*)-pent-4-yn-2-ol that was silylated and carbomethoxylated to afford the hexynoate ester **171**. Reaction of the ester **171** with ketene dimethyl acetal then gave the highly functionalized chiral butadiene (*S*)-**172**. When diene (*S*)-**172** was heated with 2-chloro-1,4-naphthoquinone **173**, anthraquinone **174** was obtained regioselectively. Removal of the silyl protecting group under acidic conditions, lactonization and finally selective demethylation of the sterically most encumbered 12-*O*-methyl ether group afforded (*S*)-dermolactone **82** in high enantiomeric purity. The availability of enantiomerically pure (*S*)-dermolactone **82**, obtained by synthesis, was crucial in identifying that natural dermolactone exists as an unequal mixture of enantiomers.



Scheme 21

The potential versatility of the highly functionalized diene **172** to be used in the synthesis of other lactone-fused aromatic systems was realized by its reaction with various dienophiles. Thus, the (*R*)-enantiomer of **172**, prepared from (*R*)-propylene oxide in the same manner as described above for (*S*)-**172**, when heated with 2,5-dichlorobenzoquinone **175** formed naphthoquinone **176** (Scheme 22). Substitution using methoxide and lactone formation completed the first total synthesis of (*R*)-semixanthomegnin **61**.[92] The flexibility of this Diels-Alder approach was further demonstrated by the synthesis of ochratoxin-α **178**. In this case diene (*R*)-**172** reacted effectively with the acetylenic dienophile methyl propiolate to give benzoate **177** that was further transformed to ochratoxin-α **178**, completing a formal synthesis of ochratoxin A **179**.[165] Ochratoxin A **179** co-occurs with xanthomegnin **60** in several *Penicillium* species, including those contaminating agricultural products,[97] and is a nephrotoxin, immunosuppressant, teratogen and carcinogen that is implicated in several human diseases.[166] A similar Diels-Alder strategy to that shown in Schemes 21 and 22 has been explored for the synthesis of xylindein **70**.[167]

[[SCHEME 22 TO BE INSERTED HERE]]

Scheme 22

5. Conclusions

Scientific interest in the chemistry of naphthopyranone compounds has a long history extending back to the 19th century when the blue-green pigment responsible for the natural staining of wood used for decorative purposes was being investigated, later the responsible agent being identified as xylindein **70**. Over 130 years on from these early studies, state-of-the-art techniques have now demonstrated the possibility of constructing engineered polyketide synthases designed to produce novel naphthopyranones, such as norpyrone **99**. Along the way naphthopyranones have been shown to occur in a range of organisms including fungi, bacteria, lichen and plants, with most examples showing some level of antibiotic, cytotoxic, antioxidant, immunoregulatory, antimalarial or antifungal activity, to name but a few of the diversity of bioactivities identified. Some members have shown significant potential for further development, such as analogues of chartreusin **86** that have proceeded to phase II clinical trials for the treatment of cancer. The preparation of naphthopyranones, either by semisynthesis or total synthesis, has played an important role in confirming structural details such as absolute stereochemistry in a number of naturally occurring naphthopyranones. The desire to develop new methods to enable efficient synthesis of these entities has expanded the broader art of organic synthesis, with recent developments in the area of mutasynthesis beginning to have a beneficial impact in the preparation of naphthopyranones and novel analogues otherwise not readily available through total synthesis. After a long history of investigation, new opportunities are now opening up for exploitation of naphthopyranones, particularly for their pharmaceutical potential.

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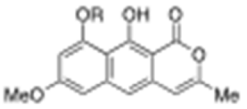
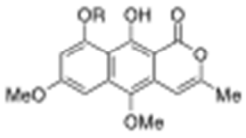
THE FOLLOWING GRAPHICS/SCHEMES NEED TO BE INSERTED INTO THE TEXT AT THE INDICATED POSITIONS

[[STRUCTURES 1-16]]:



1 R = H, toralactone
2 R = R^1
3 R = R^2, cassiaside C
4 R = R^3, cassiaside C_2
5 R = R^4



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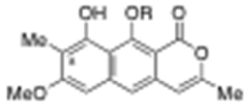
10 R = R^1
11 R = R^5
12 R = R^2
13 R = R^6
14 R = R^7



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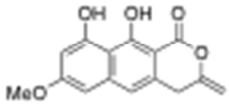
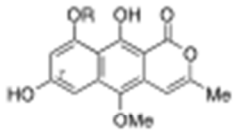
7 R = Me



8 isotoralactone



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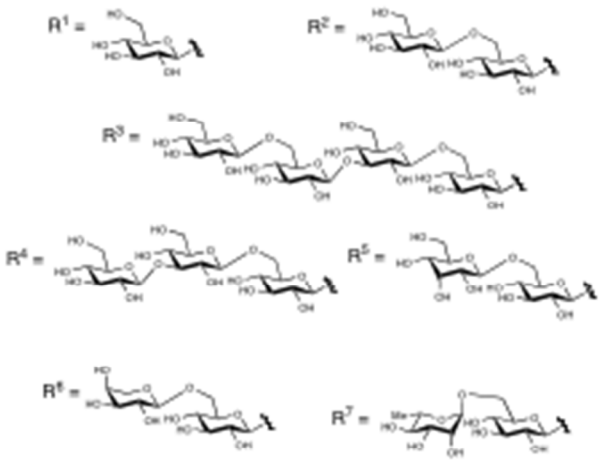
16 R = R^2



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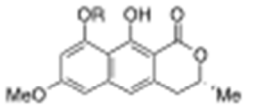
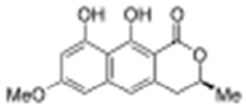
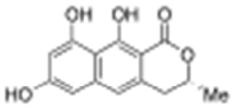


20 R = H, (*R*)-semivioxanthin
21 R = R^1
22 R = R^2
23 R = R^3



24 (*S*)-semivioxanthin



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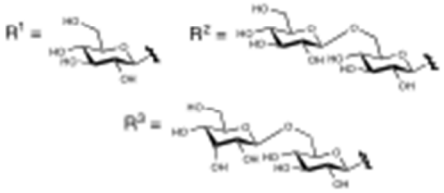
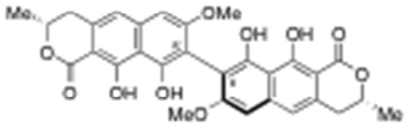
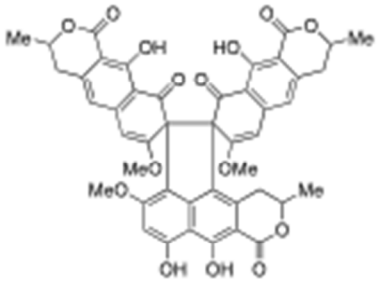


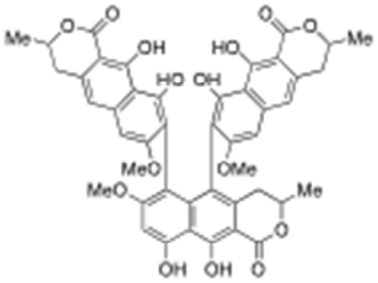


26 vioxanthin

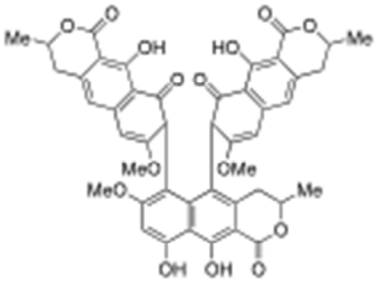
[[STRUCTURES 30-32]]:



30 PF1158A

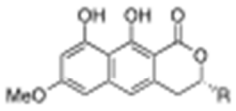


31 PF1158B

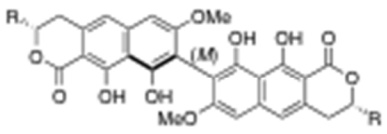


32 PF1158C

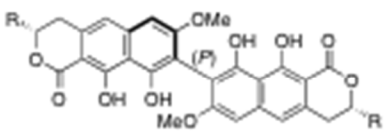
[[STRUCTURES 42-56]]:



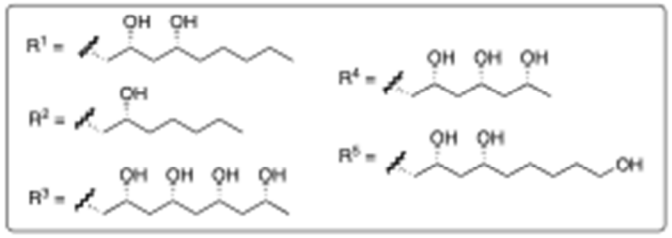
42 R = R^1, monapinone A
43 R = R^2, monapinone B
44 R = R^3, monapinone C
45 R = R^4, monapinone D
46 R = R^5, monapinone E

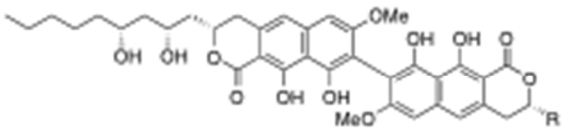


47 R = R^1, dinapinone A1



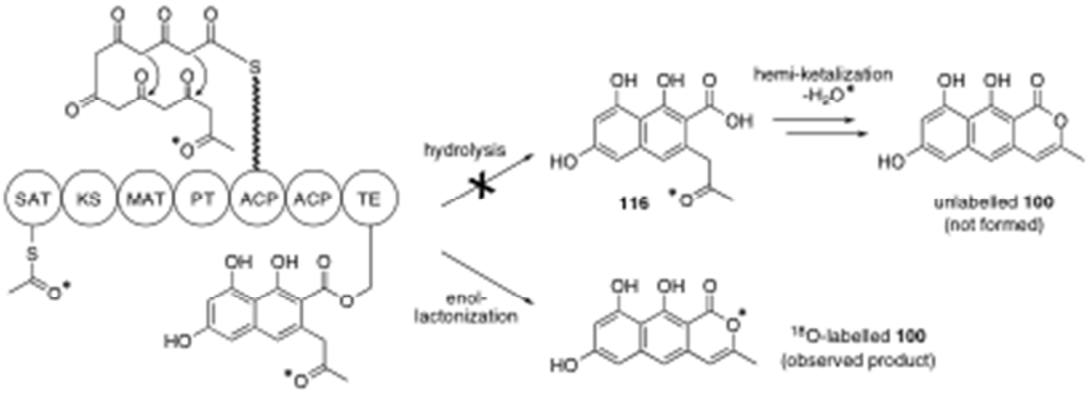
48 R = R^1, dinapinone A2





axis-(*M*)	axis-(*P*)
49 R = R^2, dinapinone AB1	**50** R = R^2, dinapinone AB2
51 R = R^3, dinapinone AC1	**52** R = R^3, dinapinone AC2
53 R = R^4, dinapinone AD1	**54** R = R^4, dinapinone AD2
55 R = R^5, dinapinone AE1	**56** R = R^5, dinapinone AE2

[[SCHEME 6]]:



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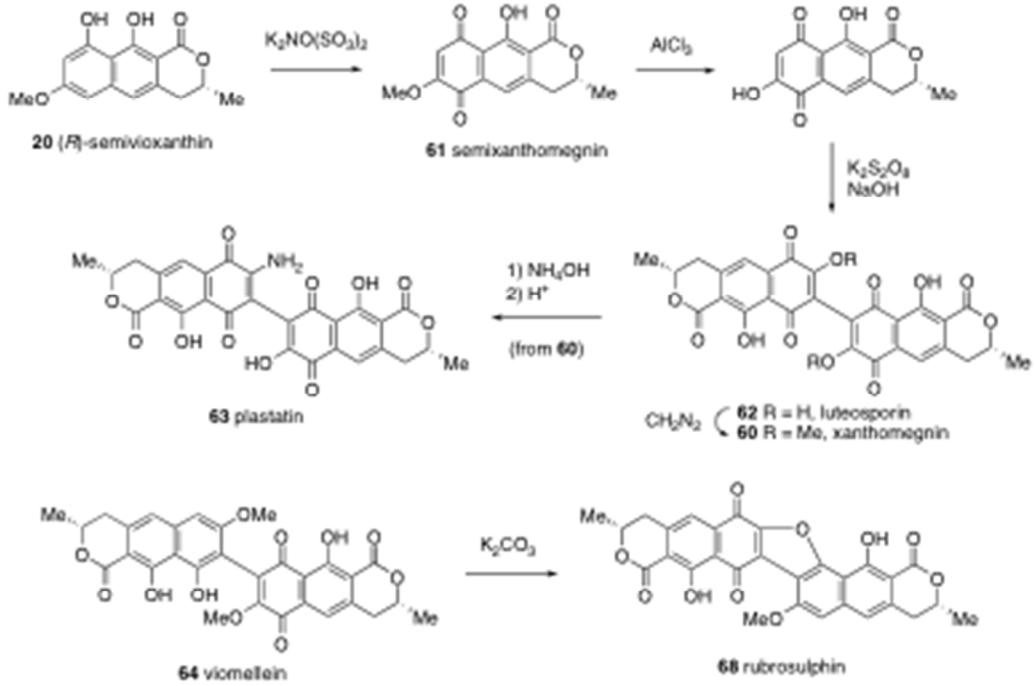

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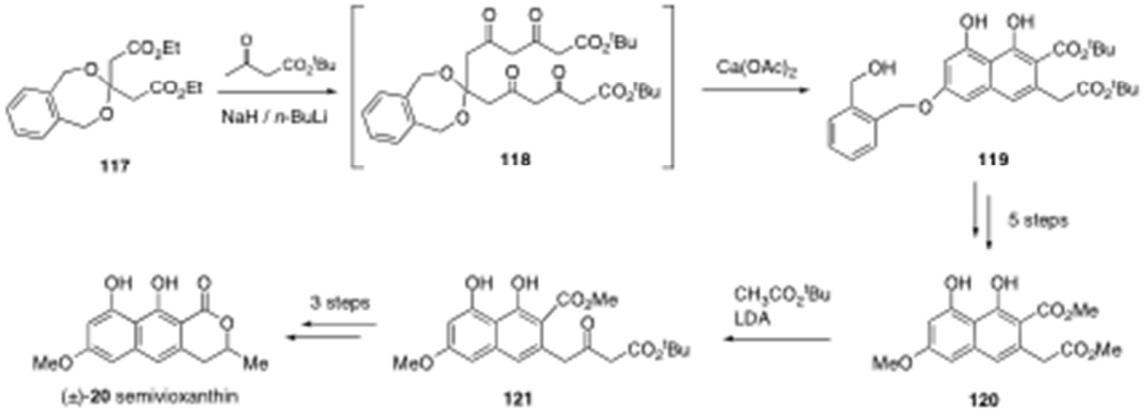
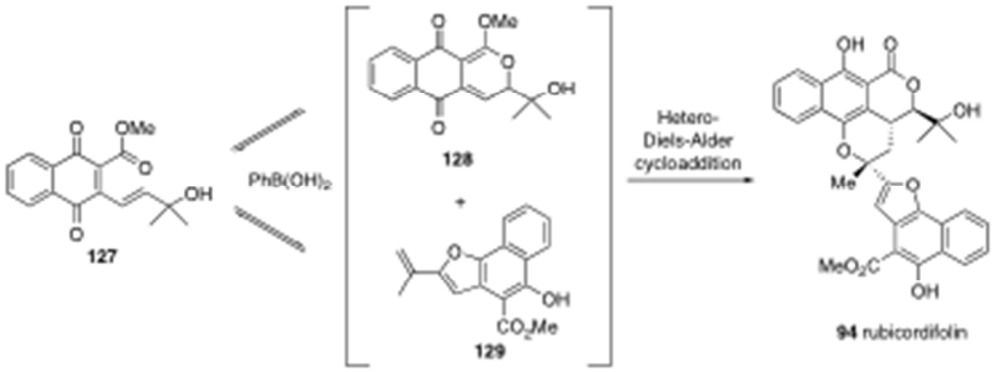
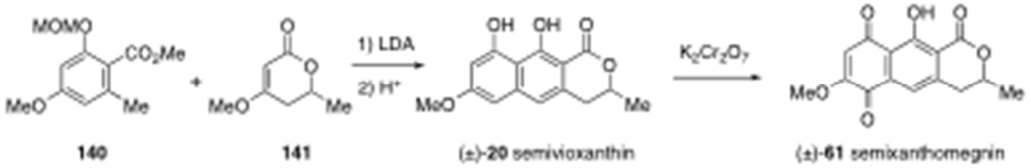


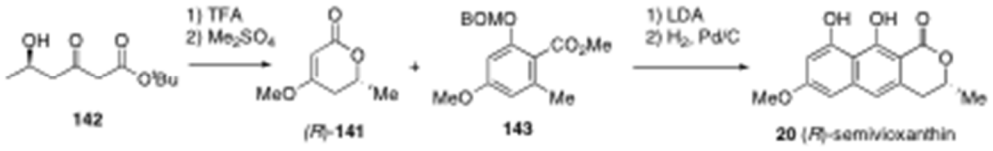
[[SCHEME 10]]:



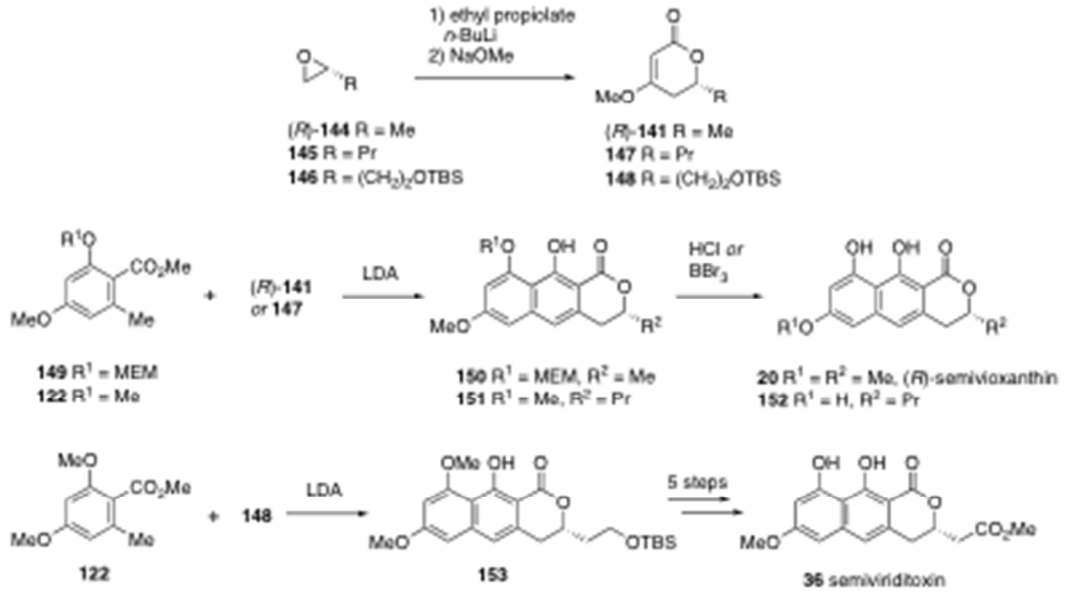
[[SCHEME 13]]:



[[SCHEME 14]]:



[[SCHEME 15]]:



[[SCHEME 22]]: