CHAPTER ONE

A Biological and Chemical Introduction to the Zoanthamine Alkaloids

1.1 Isolation of Zoanthamine Alkaloids

1.1.1 *The Order Zoantharia*

The order zoantharia comprises an intriguing group of marine polyps. This order has been morphologically classified into at least a dozen genera, and recent analysis of their respective mitochondrial DNA has elucidated the relationships between them.[1] Species in this order are widely dispersed throughout the temperate and tropical littoral regions of the Indian, Pacific, and Atlantic Oceans. These soft corals are generally aggressive colonizers of reef environments. In the wild, they are known to reproduce both sexually[2] and by asexual budding of new polyps.

Figure 1.1 Representative Zoanthids

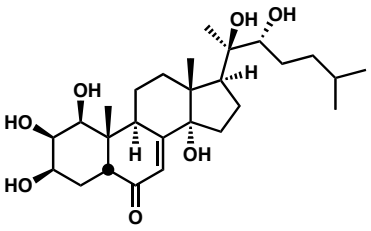
The polyps have a tube-shaped body and are radially symmetric. Atop the body are tentacles that guide food to the central orifice for digestion. Additionally, zoanthids frequently contain symbiotic microalgae, which provide energy via photosynthesis. These dinoflagellate algae also play an important role in the biosynthesis of secondary metabolites originally isolated from the zoanthids.[3] When alarmed, the polyps contract their tentacles inward, and some species also expel from their bodies a stream of water laden with powerful toxins as a means of defense from predators. As a result of their vibrant pigmentation, zoanthids are prized as specimens for marine aquariums and are commonly sold as "sea mats."

1.1.2 *Natural Products Isolated from the Order Zoantharia*

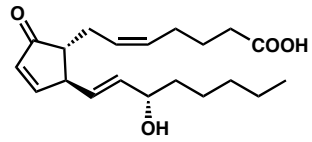
A rich diversity of natural product archetypes has been isolated from species in the order zoantharia (Figure 1.2). Zoanthusterone (**1**) is a representative ecdysteroid isolated from a *Zoanthus sp*.[4] Prostaglandins like PGA_2 (**2**), isolated from *Palythoa kochii*, have been shown to stabilize microtubules in a manner similar to paclitaxel.[5] A family of more than a dozen natural products based on the highly fluorescent zoanthoxanthin (**3**) skeleton has been isolated from *Parazoanthus axinellae*.[6] A related demethylated structure, parazoanthoxanthin A, has shown significant anticholinesterase activity.[7] Both red[8] and yellow[9] fluorescent proteins containing novel chromophores have been isolated and crystallographically studied. Perhaps the best known compound isolated from these marine organisms is palytoxin (**4**). Palytoxin was isolated from *Palythoa sp*. in the Hawaiian islands and remains one of the most toxic compounds

known, with an LD_{59} of 15 μg/kg in mice.[10] The structure of palytoxin was determined and later synthesized by Kishi.[11]

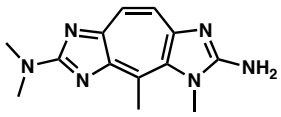
Figure 1.2 Representative Natural Products from Zoanthids



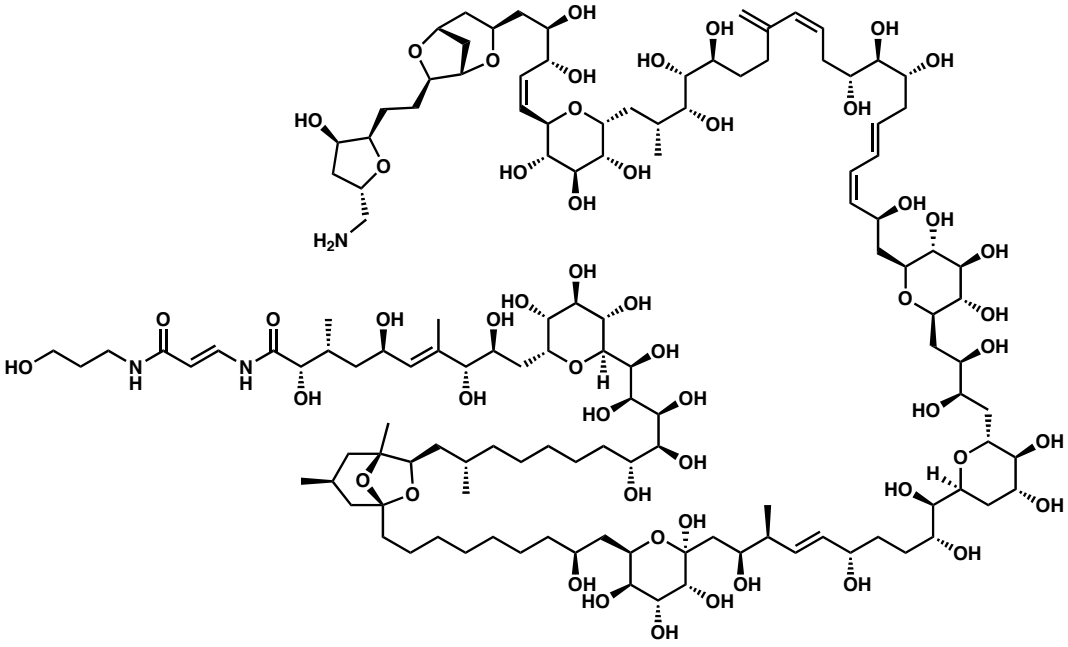
Zoanthusterone ***(1)***



PGA_2 ***(2)***



Zoanthoxanthin ***(3)***



Palytoxin ***(4)***

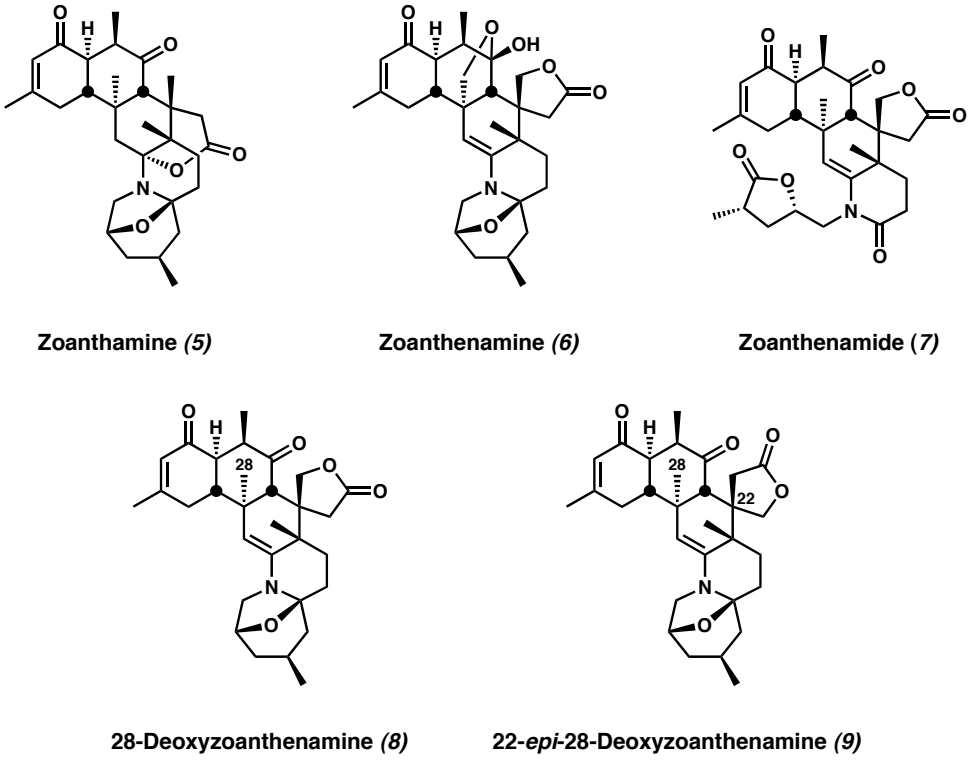
1.2 The Zoanthamine Natural Products

1.2.1 *Isolation and Structural Characterization*

In 1984, the isolation of the natural product zoanthamine (**5**) from a species of the genus Zoanthus off the Visakhapatnam coast of India was disclosed. The connectivity

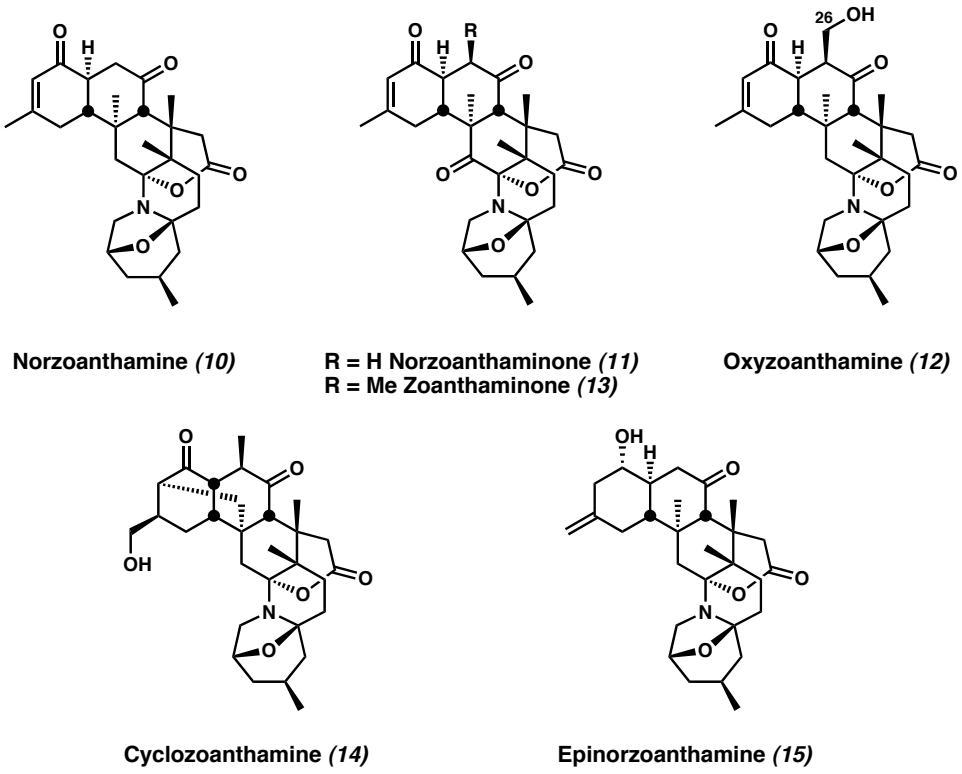
and relative stereochemistry of the previously unknown alkaloid skeleton was unambiguously determined by single crystal X-ray diffraction.[12] This isolation effort also afforded the related natural products zoanthenamine (**6**) and zoanthenamide (**7**).[13] The same group later described 28-deoxyzoanthenamine (**8**) and 22-*epi*-28-deoxyzoanthenamine (**9**) from a Zoanthus species isolated in the Bay of Bengal.[14] The latter four structures were deduced by comparison with zoanthamine's spectroscopic data. Originally, this isolation effort was undertaken in search of a known eye irritant produced by the Zoanthus species. However, all five of the isolated alkaloids showed some inhibition of phorbol myristate acetate (PMA) inflammation in mouse ears. The function of the zoanthamine alkaloids in the producing organisms is unknown.

Figure 1.3 Zoanthamine Natural Products Isolated by Rao



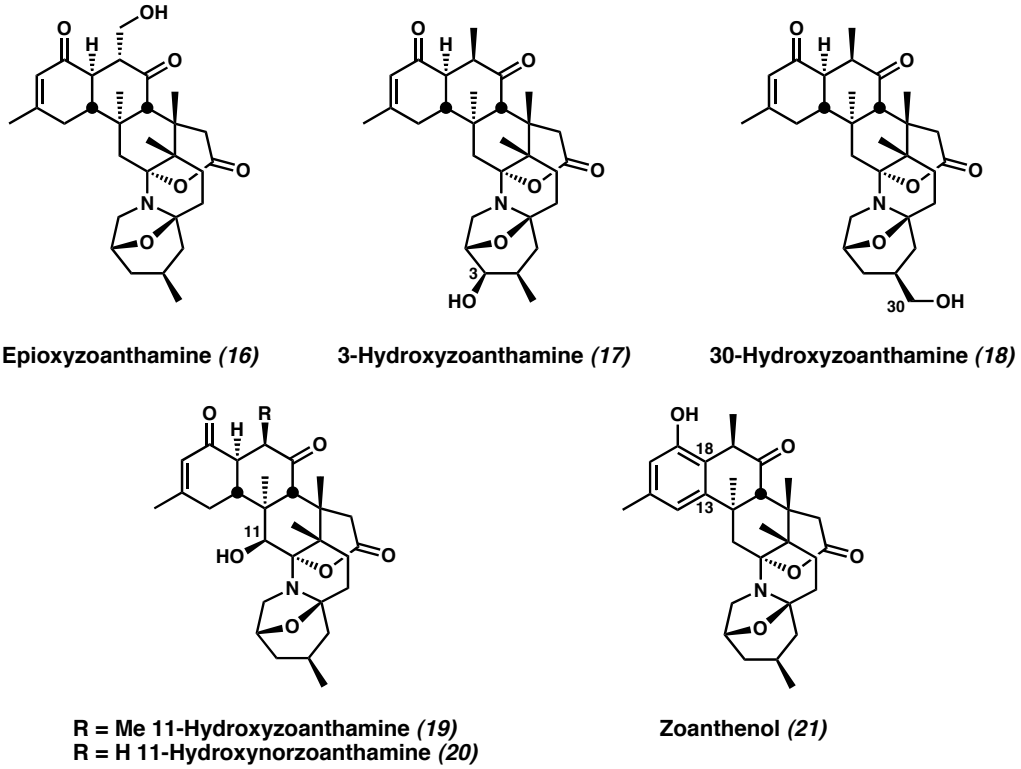
In 1995, Uemura and coworkers identified five new zoanthamine natural products isolated from a Zoanthus species collected off the Ayamaru coast of the Amami Islands south of Japan.[15] Among these, norzoanthamine (**10**) and norzoanthaminone (**11**) lack the C(26) methyl group, while in oxyzoanthamine (**12**) the C(26) methyl group has been oxidized. The relative configuration of norzoanthamine was confirmed by X-ray diffraction.[15] The absolute configuration of norzoanthamine, later determined by NMR analysis of MTPA derivatives, is shown in Figure 1.4.[16] The C_{30} alkaloid related to norzoanthaminone (**11**), zoanthaminone (**13**), was amenable to X-ray crystal structure determination as disclosed by Clardy.[17] Cyclozoanthamine (**14**) and epinorzoanthamine (**15**) represent intriguing modifications to the enone functionality typical of zoanthamines. Both structures were assigned by extensive NOE experiments.[15]

Figure 1.4 Zoanthamine Natural Products Isolated by Uemura and Clardy



Zoanthids isolated from the Canary Islands by Norte and coworkers in 1996 provide zoanthamine natural products with several interesting oxidation patterns (Figure 1.5). Epioxyzoanthamine (**16)** is unique in its C(19) stereochemistry, which was determined by comparison with NMR data for oxyzoanthamine.[18] 3-Hydroxyzoanthamine (**17)** and 30-hydroxyzoanthamine (**18)** show novel sites of oxidation, while 11-hydroxyzoanthamine (**19)** and 11-hydroxynorzoanthamine (**20)** are presumably related to zoanthaminone and norzoanthamineone. Finally, zoanthenol (**21**) has a unique oxidized aromatic A ring, which removes the C(13) and C(18) stereocenters. As a result of the structural change, extensive HMBC and ROESY correlation experiments were performed to confirm its structure and relative stereochemistry.[19]

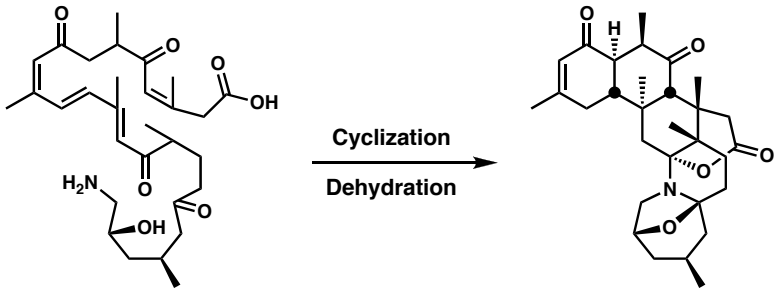
Figure 1.5 Zoanthamine Natural Products Isolated by Norte



1.2.2 *Biosynthesis of the Zoanthamine Natural Products*

The C_{30} skeleton of the zoanthamine natural products led to initial biosynthetic speculation focused on triterpene precursors.[12] However, no normal head-to-tail isoprene connectivity could account for the zoanthamine skeleton. More recently, Uemura has proposed that the zoanthamine natural products may arise via a polyketide construction (Scheme 1.1).[16,20] The proposed intermediate **22** accounts for most of the oxygenation found in the zoanthamines. While no definitive statement can be made about biosynthesis, there appears to be support for the polyketide hypothesis.[18]

Scheme 1.1 Hypothetical Polyketide Precursor

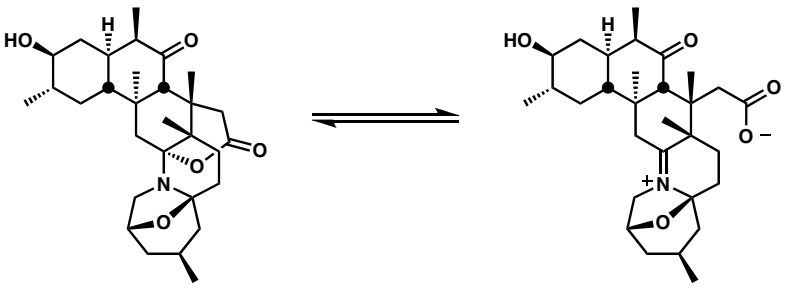


Hypothetical Polyketide Precursor *(22)* **Zoanthamine *(5)***

Another factor complicating the understanding of zoanthamine alkaloid biosynthesis is the role of the symbiotic dinoflagellate algae that are commonly contained in zoanthids. Although such symbiotic strains are difficult to culture in isolation, Nakamura and coworkers have been able to produce quantities of *Symbiodinium sp*. free of zoanthids.[21] Algae of the genus *Symbiodinium* have been isolated from zoanthids of the genus Zoanthus.[22] While the algae produced different distributions of metabolites depending on the media used, Nakamura's group was able to isolate a new C_{30} alkaloid

they named zooxanthellamine (**23)** after significant experimentation with culture conditions. Zooxanthellamine exists as a mixture of imine and lactone forms (Figure 1.6). Its structure and relative configuration were delineated from extensive NMR studies. Zooxanthellamine was shown to have the same sense of absolute configuration as the zoanthamine alkaloids by NMR comparison of its MTPA esters.[21] The remarkable similarity between zooxanthellamine and zoanthamine has called into question the role of the zoanthids in producing the zoanthamine natural products.[23] It may be that the zoanthids play only a small role in adjusting the oxidation state of the completed zoanthamine skeleton. The subtle variations in the alkaloids' structures could be determined by factors in the marine environment or by the host zoanthid species. Alternatively, different algae species may be involved in the production of different zoanthamines.

Figure 1.6 Structure of Zooxanthellamine

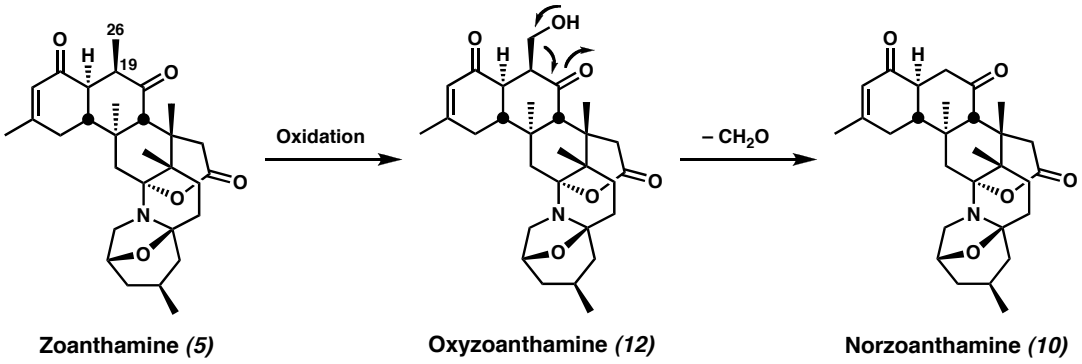


Zooxanthellamine *(23)*

Another intriguing biosynthetic question involving the zoanthamine natural products is the origin of the norzoanthamine-type alkaloids, which lack the C(26) methyl group. The isolation of oxyzoanthamine prompted Uemura and coworkers to propose a

mechanism for the demethylation of zoanthamines (Scheme 1.2). They propose oxidation of the zoanthamine to the intermediate oxyzoanthamine, which then undergoes a retro aldol, formally releasing formaldehyde and the noralkaloid.[15]

Scheme 1.2 Proposed Biosynthesis of the Norzoanthamines

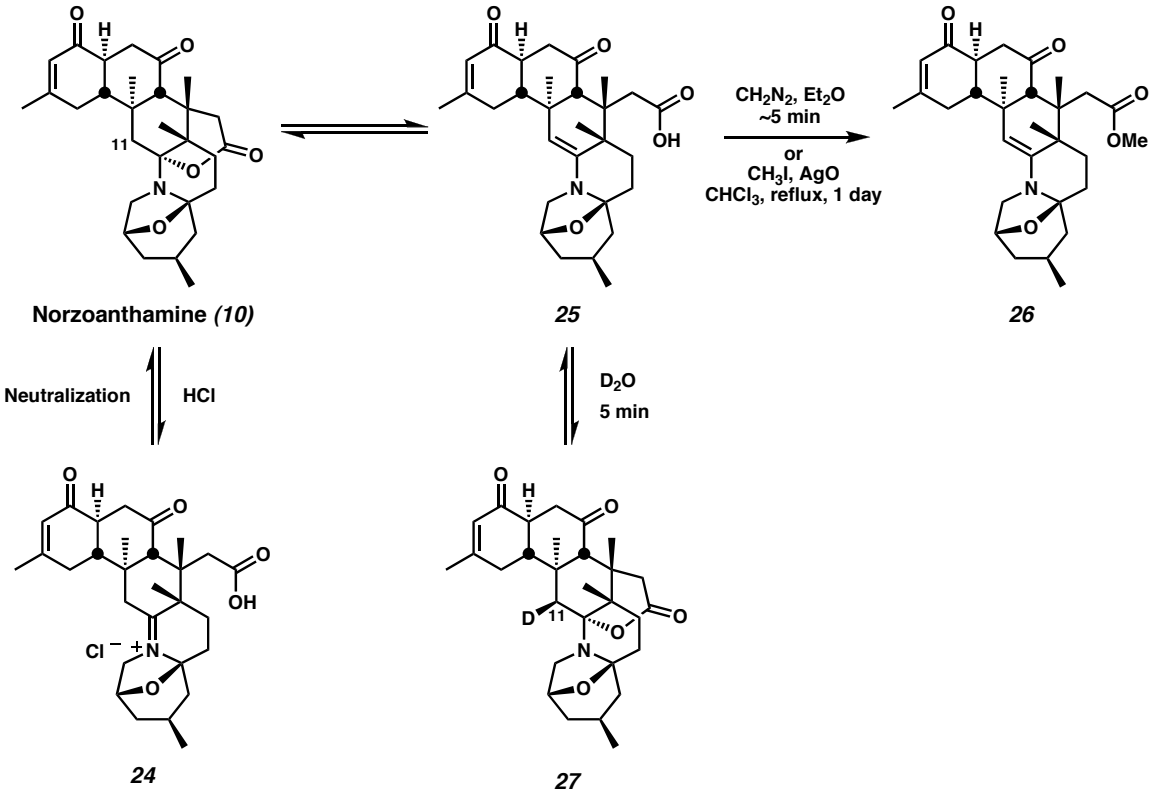


1.2.3 *Chemical Reactivity of the Zoanthamine Natural Products*

Fluxion between lactone and imine isomers, similar to the equilibration observed with zooxanthellamine, has been demonstrated in several zoanthamine natural products (Scheme 1.3). Norzoanthamine forms iminium **24** under acidic conditions and reverts upon neutralization.[20] Under neutral to basic conditions, elimination forms enamine **25**. The equilibrium between norzoanthamine and enamine **25** was demonstrated by the conversion of norzoanthamine to methyl ester **26** in minutes upon exposure to diazomethane.[20] Furthermore, NMR spectra of norzoanthamine in D_2O show specific and complete deuterium incorporation at the 11β position to give deuteride **27** in minutes.[18,19] Similar rates of deuterium incorporation were observed with zoanthenol, 3-hydroxynorzoanthamine, and 30-hydroxnorzoanthamine. In contrast, the 11β-hydroxyzoanthamines did not show significant deuterium incorporation, suggesting that

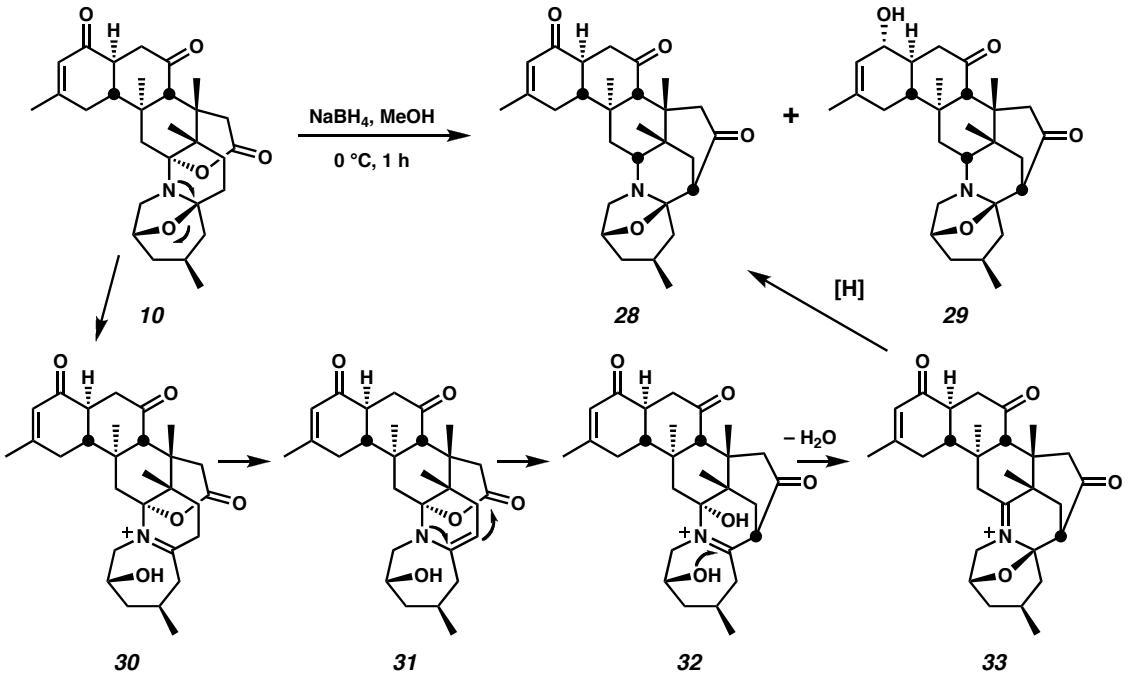
the elimination to an enamine is inaccessible.[19] This fluxional behavior in aqueous media at physiologically relevant pHs may play an important role in determining the bioactivities of these molecules.

Scheme 1.3 Equilibria Between Lactone and Enamine Isomers of Norzoanthamine



The hemiaminal region of the zoanthamine alkaloids also shows intriguing reactivity under reductive conditions. Treatment of norzoanthamine with sodium borohydride generates two anomalous products, enone **28** and allylic alcohol **29** (Scheme 1.4).[20,16] Under the reaction conditions, enamine **31** is believed to attack the lactone in an intramolecular fashion to afford keto-iminium **32**. Dehydration generates iminium **33**, which undergoes reduction to give enone **28**. Further reduction affords allylic alcohol **30**.

Scheme 1.4 Anomalous Reduction of Norzoanthamine



1.3 Biological Activities of Zoanthamine Alkaloids

1.3.1 *Antiosteoporotic Activity*

Perhaps the best studied and most well-known biological activity of the zoanthamine alkaloids is the antiosteoporotic effect first reported by Uemura in 1996.[24] Norzoanthamine and its hydrochloride salt have been shown in vivo to prevent the symptoms of osteoporosis in ovariectomized mice, a pharmaceutical model for postmenopausal osteoporosis. Osteoporosis is a loss of bone mineral density that often results when osteoclasts reabsorb bone tissue at a rate faster than it is regenerated.[25] In the model, ovariectomized mice, lacking in estrogen, quickly lose bone mass and strength. At doses of 0.4 and 2.0 mg/kg/d (five days a week for 4 weeks, *p.o.*) of norzoanthamine hydrochloride, the ovariectomized mice retained femur weight at statistically higher rates than untreated ovariectomized mice.[26] Additionally, with doses

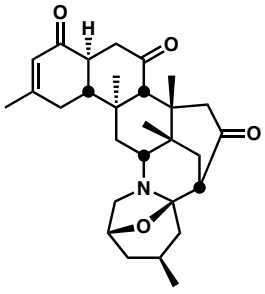
from 0.016-0.4 mg/kg/d of norzoanthamine hydrochloride the femurs of ovariectomized mice maintained strength, as measured by failure load, nearly comparable to unovariectomized control mice.[27] Finally, mice treated with norzoanthamine hydrochloride were observed to possess cortical bone significantly thicker than that found in the control animals.[28]

In analogy to estrogen replacement therapy in postmenopausal women, ovariectomized mice can be rescued from the effects of osteoporosis by treatment with 17β-estradiol. However, treatment with norzoanthamine hydrochloride shows interesting differences from estrogen therapy. Uterine atrophy, a side effect of estrogen deficiency after ovariectomy, is not affected by treatment with norzoanthamine hydrochloride.[28] This ability to restore bone strength without eliciting other side effects associated with steroid hormone receptors is a highly desirable property.

The origin of norzoanthamine's antiosteoporotic effect may lie in its ability to suppress the production of interleukin 6 (IL-6). Norzoanthamine and its hydrochloride salt **24** have been shown to suppress the excretion of IL-6 from preosteoblastic cells at concentrations of 13 and 4.6 μg/ml, respectively, in vitro.[29] IL-6 is in turn involved in stimulating the generation of osteoclasts, which reabsorb bone tissue. Estrogen derives its antiosteoporotic properties from the inhibition of IL-6 production.[30] However, in vitro studies on osteoblasts with norzoanthamine hydrochloride showed no effect on osteoclast formation. Also, the suppression of IL-6 secretion has not yet been demonstrated in vivo.[28] These last two points, along with the lack of uterine weight gain in ovariectomized mice, suggest that the zoanthamine alkaloids may act by a mechanism distinct from estrogen's.[31]

The need to find non-estrogen therapies to prevent the onset of osteoporosis has spurred considerable effort to define a structure activity relationship (SAR) for norzoanthamine's antiosteoporotic effects, despite its unclear molecular target. Via semi-synthesis, Uemura and coworkers produced and tested a number of norzoanthamine derivatives.[20,29] It should be noted that all of the derivatives assayed were significantly less efficacious (higher IC_{50} values) in limiting IL-6 production than norzoanthamine (Figure 1.7).[32] The removal of the olefin (ketone **35** and diol **36**) caused some loss in activity. However, disruption of the lactone/hemiaminal functionality (carboxylic acid **37** and ester **27**) caused the greatest drops in activity.[20]

Figure 1.7 SAR Study of Norzoanthamine's Ability to Inhibit IL-6 Production



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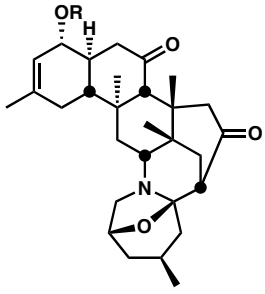
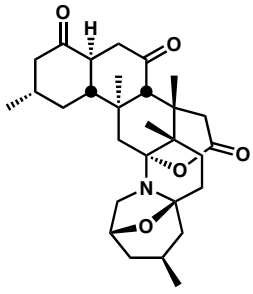


29, R = H
34, R = OAc



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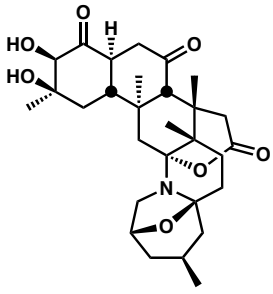

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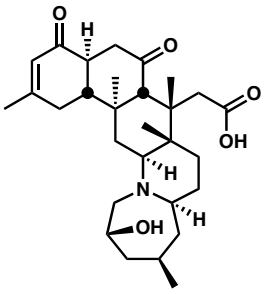

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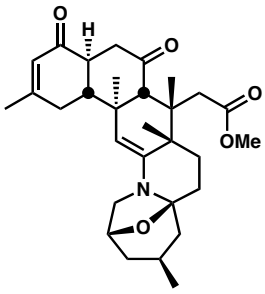

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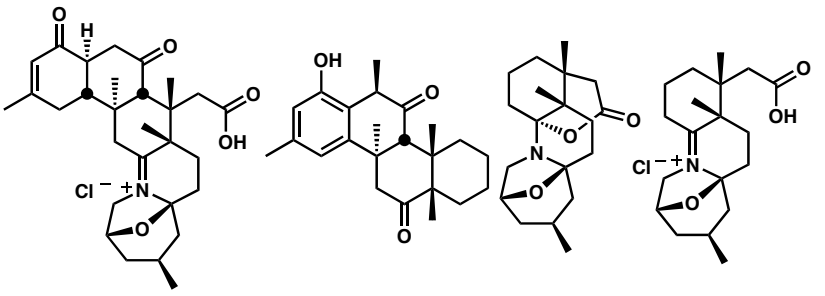
Compound	IC_{50} (μg/mL) for Suppression of IL-6 Production
Norzoanthamine *(10)*	13
28	25
29	30
34	23
35	45
36	35
37	42
26	>100

More recently, Hirama and coworkers have studied the SAR of zoanthenol-related molecules to inhibit the growth of IL-6 dependent MH-60 cells (Figure 1.8). In their assays, the zoanthamine hydrochloride salts **24** and **38** showed the strongest inhibition of IL-6 production with IC_{50} values of 13 and 26 μM, respectively. A truncated mimic of zoanthenol's "northern" carbocyclic region **39** and a mimic of the zoanthamine "southern" heterocyclic region **40** both showed very poor activity. However, iminium **41**, the hydrochloride salt of heterocycle **40**, demonstrated a significant amount of the activity found in the zoanthamine hydrochlorides.[33] This result provides further support for two trends: (a) the hydrochloride salt form of a zoanthamine molecule is a more active inhibitor of IL-6 production than the natural product, and (b) the heterocyclic portion of the molecule contains a large part of the pharmacophore for IL-6 inhibition.

Figure 1.8 SAR of Zoanthenol Analogues' Inhibition of IL-6 Dependent Cell Growth



Compound	IC_{50} (μM) for IL-6 Dependent Cell Growth
24	13
38	26
39	>100
40	>100
41	~70

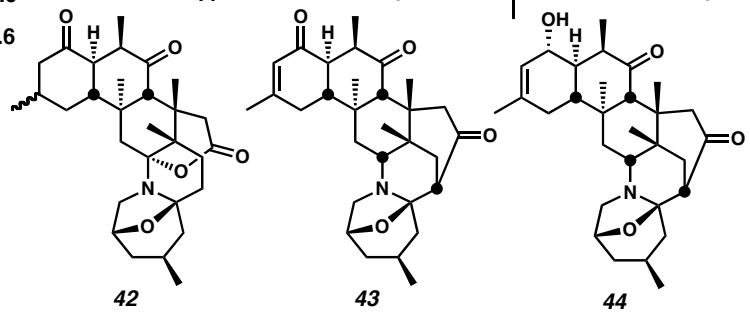
1.3.2 *Miscellaneous Biological Activities*

A multitude of other biological activities have been discovered for molecules in the zoanthamine family. As previously mentioned, zoanthamine, zoanthenamine, and zoanthamide were found to be inhibitors of PMA-induced inflammation in mouse ear.[12,13]

In addition to the isolation of norzoanthamine, norzoanthamineone, oxyzoanthamine, cyclozoanthamine, and epinorzoanthamine, Uemura and coworkers reported that these structures had significant cytotoxicity against P388 murine leukemia cells (Table 1.1).[15] The most potent cytotoxicity was displayed by norzoanthaminone. The antibacterial properties of zoanthamine and several of its reduced derivatives have been investigated.[34] In disk susceptibility experiments, the zoanthamine alkaloids showed broad spectrum activity against both Gram negative and Gram positive bacteria (Table 1.1). More recently the effect of zoanthamine alkaloids on human platelet aggregation has been investigated.[35] These experiments showed that at concentrations of 0.5 mM 11-hydroxyzoanthamine and methyl ester **26** prevent the majority of platelet aggregation caused by collagen, arachidonic acid, and thrombin. In contrast, oxyzoanthamine and zoanthenol were highly effective inhibitors of aggregation in the presence of collagen at 0.5 mM, but showed almost no activity in the presence of arachidonic acid or thrombin.

Table 1.1 Summary of Miscellaneous Biological Activities

Cytotoxicity		**Antibacterial Activity**				
			Inhibition Zone (dia. in mm)			
Compound	**IC_{50} (μg/ml) for Inhibition of P388 Murine Leukemia Cells**	**Compound**	**Gram Negative**		**Gram Positive**	
			S. typhimurium	***E. coli***	***B. sphaericus***	***S. aureus***
Norzoanthamine *(10)*	**24.0**	**Zoanthamine *(5)***	**6**	**6**	**8**	**7**
Norzoanthaminone *(11)*	**1.0**	***42***	**12**	**6**	**8**	**10**
Oxyzoanthamine *(12)*	**7.0**	***43***	**7**	**6**	**8**	**10**
Cyclozoanthamine *(14)*	**24.0**	***44***	**6**	**7**	**7**	**9**
Epinorzoanthamine *(15)*	**2.6**					



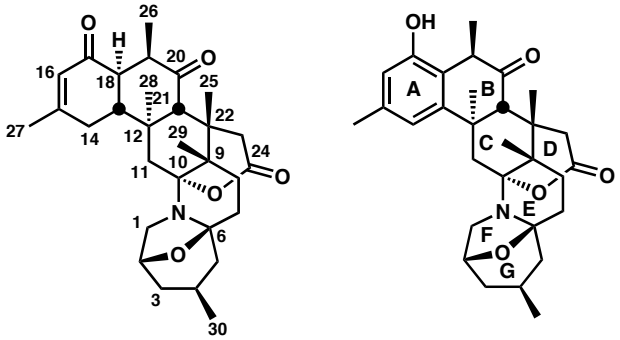
42 ***43*** ***44***

1.4 Approaches Toward the Synthesis of Zoanthamine Natural Products

1.4.1 *General Remarks*

The intriguing diversity of biological activities and the challenging structures of the zoanthamine alkaloids have inspired no fewer than seven synthetic chemistry groups to publish strategies toward the total synthesis of these molecules. Carbon numbering and ring naming will refer to the zoanthamine numbering scheme throughout this thesis (Figure 1.9). Though the focus of the work in our laboratories has been toward the synthesis of zoanthenol, a survey of the synthetic work toward any of the zoanthamines is instructive due to the similar challenges posed by all the molecules in this family. Many researchers have focused their efforts on the synthesis of the ABC rings, which poses a significant synthetic challenge due to its stereochemical density. For example, the C ring contains three quaternary stereocenters in vicinal and non-vicinal relationships. In addition to the difficulty of synthesizing quaternary centers, their steric bulk also renders even routine transformations on nearby functionally recalcitrant. Other researchers have focused on the synthesis of the heterocyclic DEFG rings. DEFG ring synthesis presents the challenge of forming the heterocycles with the correct hemiaminal connectivity.

Figure 1.9 Carbon Numbering and Ring Naming Scheme for the Zoanthamine Alkaloids



Zoanthamine *(5)* **Zoanthenol *(21)***

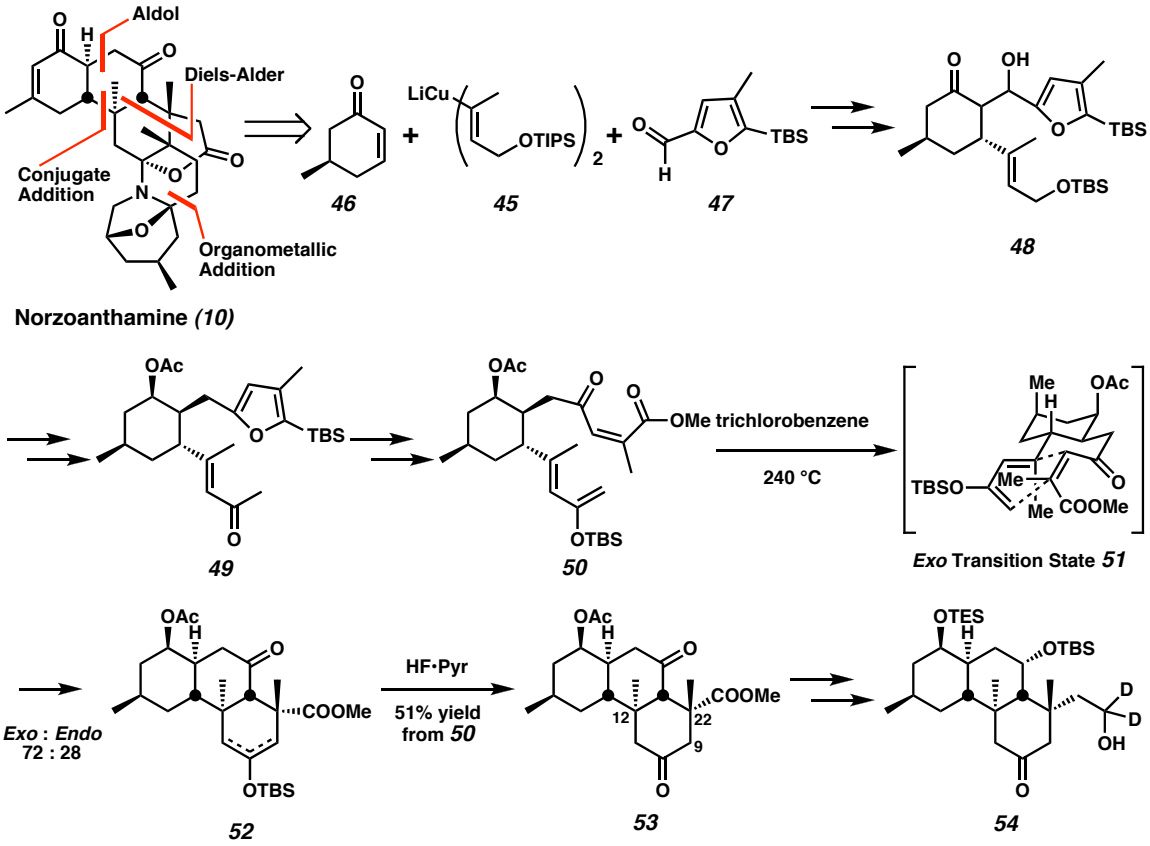
1.4.2 *Miyashita's Synthesis of Norzoanthamine*

Twenty years after the isolation of the first zoanthamine alkaloids, Miyashita and coworkers reported the only completed total synthesis to date of a zoanthamine alkaloid with their synthesis of norzoanthamine.[36] This impressive 41-step effort included several creative solutions to problems that likely arose during the execution of the synthesis. Their Diels-Alder strategy for the construction of the ABC ring system of norzoanthamine was disclosed in 2002 (Scheme 1.5).[37] Addition of cuprate **45** to enantiopure enone **46** followed by aldol reaction with aldehyde **47** provided ketone **48**. This efficient three component coupling sequence established all the carbons of the ABC ring system. After further manipulation, furan **49** was oxidized and its enone silylated to give Diels-Alder substrate **50**. Upon heating to 240 °C, the Diels-Alder reaction proceeded predominantly through the desired *exo* transition state **51** (72:28). After silyl cleavage, diastereomerically pure ketone **53** was isolated in 51% yield. This impressive Diels-Alder reaction sets both the C(12) and C(22) quaternary centers of norzoanthamine with the correct relative stereochemistry. However, this strategy provided little functionality to aid in the construction of the C(9) quaternary stereocenter. After significant functional group manipulation, ketone **53** was converted to ketoalcohol **54**.

Ketoalcohol **54** contains suitable functionality to attempt the formation of the C(9) quaternary center (Scheme 1.6). To that end, acylation of ketoalcohol **54** with dimethyl carbonate and lithium *tert*-butoxide proceeded regioselectively, presumably due to lactone formation, and was followed by quenching with methyl iodide to give *O*-alkylated β-ketolactone **55**. Upon treatment with lithium *tert*-butoxide and methyl iodide in DMPU, the *O*-alkylated β-ketolactone **55** underwent C-alkylation to give quaternized

δ-lactone **56**. Impressively, this difficult transformation was accomplished in 83% yield as a single diastereomer.

Scheme 1.5 Miyashita's Diels-Alder Construction of the ABC Rings of Norzoanthamine

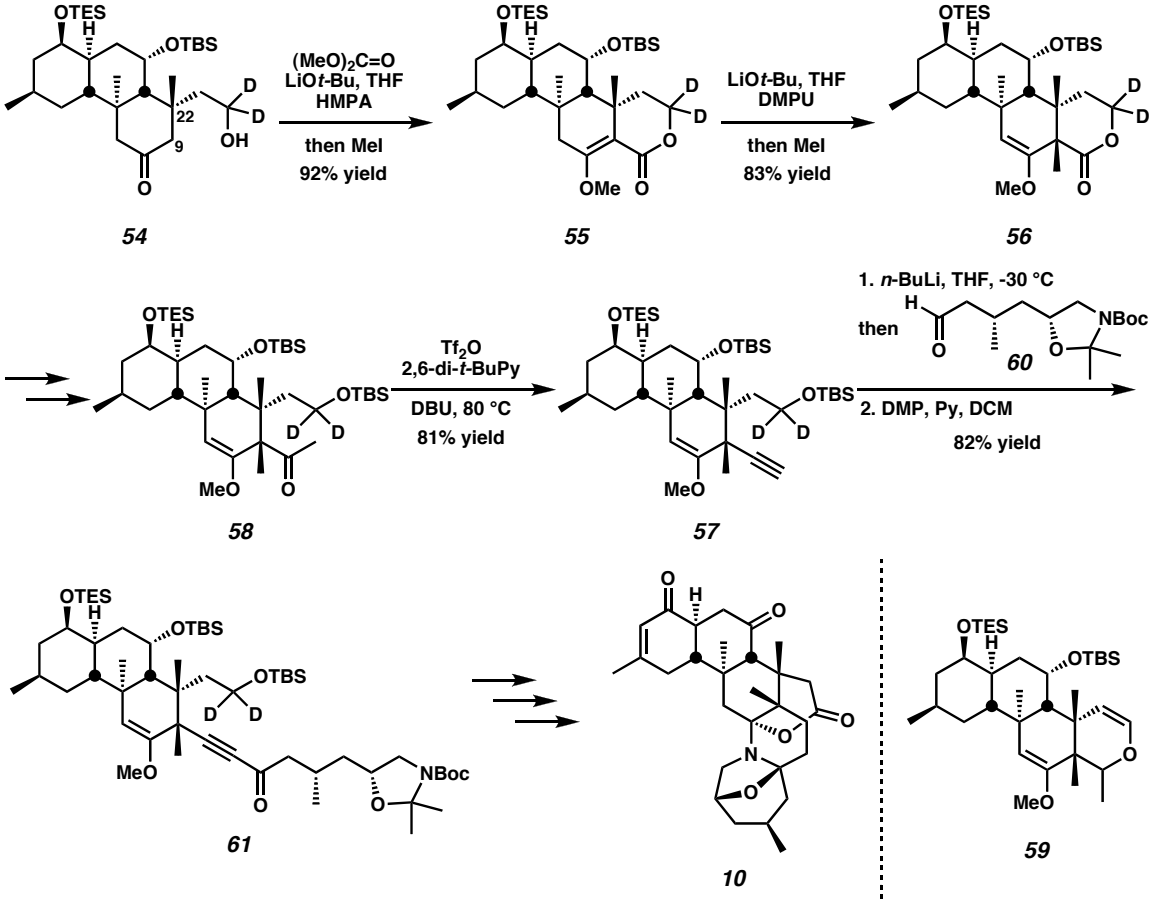


The next challenge lay in converting δ-lactone **56** to acetylene **57** (Scheme 1.6). The addition of methyl lithium with subsequent silylation converted δ-lactone **56** to ketone **58**. While the dehydration of ketone **58** appeared routine, it was at this point that Miyashita's clever incorporation of deuterium proved necessary. When the hydrido equivalent of ketone **58** was treated with triflic anhydride, the intermediate carbocation was predominantly intercepted by intramolecular hydride addition leading to

dihydropyran **59**. However, deuteride transfer proved much slower, and the deutero ketone **58** could be dehydrated to alkyne **57** with only minor amounts of the dihydropyran formed.

The carbon skeleton of norzoanthamine was completed by the addition of aldehyde **60** to the lithium salt of alkyne **57** and oxidation of the resulting alcohol to ynone **61**. Completion of the target required a further twelve steps of deprotection, oxidation state adjustment, and dehydration. In addition to the impressive synthetic accomplishment, this synthesis also served to unambiguously confirm the absolute stereochemistry of norzoanthamine that had been deduced from NMR experiments.

Scheme 1.6 The Completion of Norzoanthamine

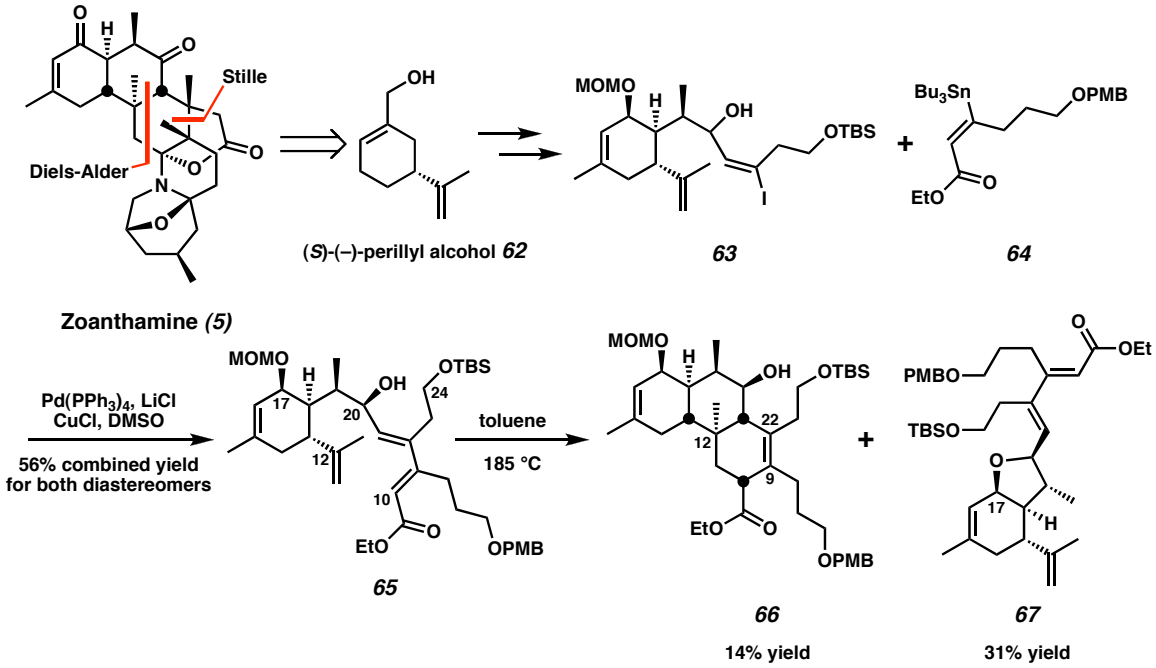


1.4.3 *Tanner's Diels-Alder Approach to the Zoanthamine ABC Ring System*

Tanner and coworkers also chose to assemble the ABC rings via a Diels-Alder approach (Scheme 1.7). Their synthesis began with (*S*)-(–)-perillyl alcohol **62** as a chiral synthon for the A ring.[38] After numerous steps, (*S*)-(–)-perillyl alcohol was transformed into iodide **63**.[39] Stille coupling with stannane **64** proved difficult and required conditions reported by Corey to afford reasonable yields of the Diels-Alder substrate **65**.[40,41] Though not the originally conceived or most direct substrate, diene **65** evolved through significant experimentation.[42] The alcohol oxidation state at C(20) was found to be necessary to prevent elimination of the primary TBS group at C(24), which occurred from the corresponding ketone. However, without the C(20) carbonyl, the diene was no longer electron deficient enough to support the desired inverse demand Diels-Alder reaction. Thus the stannane fragment was redesigned to stannane **64**, incorporating an ester moiety at the C(10) position that would need to be excised later in the synthesis. Of the two C(20) alcohol diastereomers isolated, only diene **65** underwent thermal Diels-Alder reaction. The reaction proceeded with high diastereoselectivity to afford β,γ unsaturated ester **66**, albeit in modest yield. The major product isolated involved displacement of C(17) methoxymethyl ether to give tetrahydrofuran **67**. This problem can likely be circumvented with a more robust protecting group at C(17) or protection of the C(20) alcohol. While this Diels-Alder strategy nicely establishes the quaternary center at C(12), it requires the formation of the difficult vicinal C(9) and C(22) quaternary centers at a late stage. Tanner proposes to carryout this transformation by conjugate addition into the latent C(10) enone, which is to be revealed by oxidative cleavage of the ethyl ester in

Diels-Alder adduct **66**, followed by methyl iodide trapping. No model studies were presented to demonstrate this strategy's ability to form vicinal quaternary stereocenters.[43]

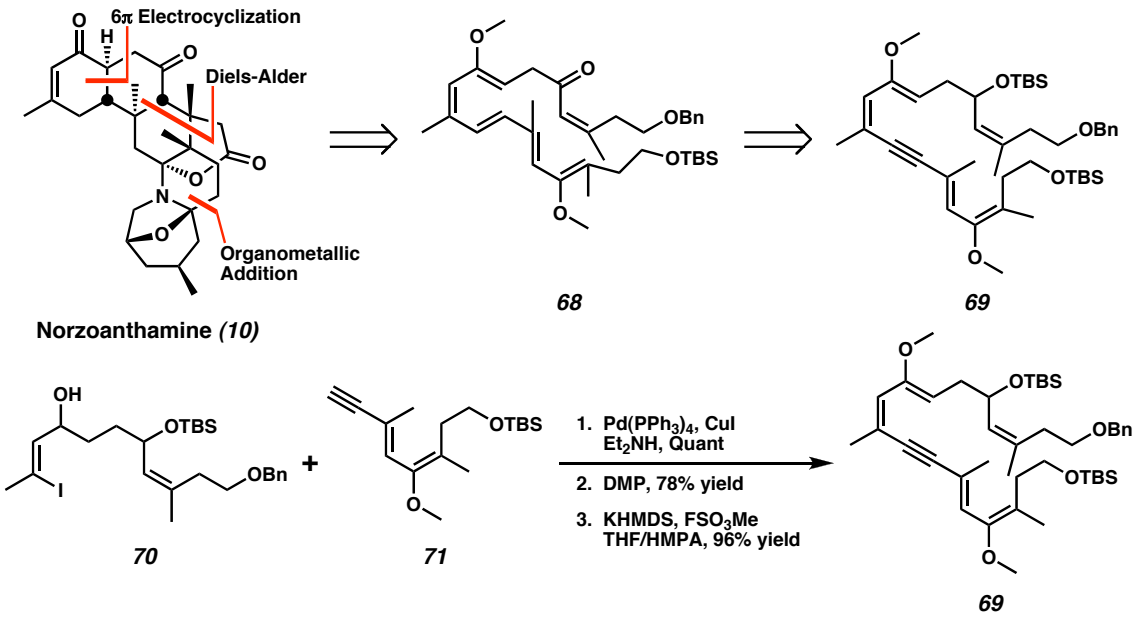
Scheme 1.7 Tanner's Approach to the ABC Ring System



1.4.4 *Uemura's Biomimetic Approach to the Norzoanthamine ABC Ring System*

Recently, Uemura and coworkers have proposed a synthetic strategy based on their biosynthetic hypothesis, which purports that the zoanthamine alkaloids arise from a linear polyketide skeleton, which then undergoes numerous cyclizations.[44] To support this hypothesis, they endeavored to synthesize and cyclize polyene **68**, which would in turn arise from enyne **69** (Scheme 1.8). Vinyl iodide **70** and alkyne **71** were efficiently assembled by Sonogashira coupling[45] and conversion to enyne **69** was completed by oxidation and methylation. No report has appeared on the selective reduction of enyne **69** to the linear polyene **68** or on attempts to cyclize polyene **68**.

Scheme 1.8 Uemura's Biomimetic Cyclization Approach to Norzoanthamine



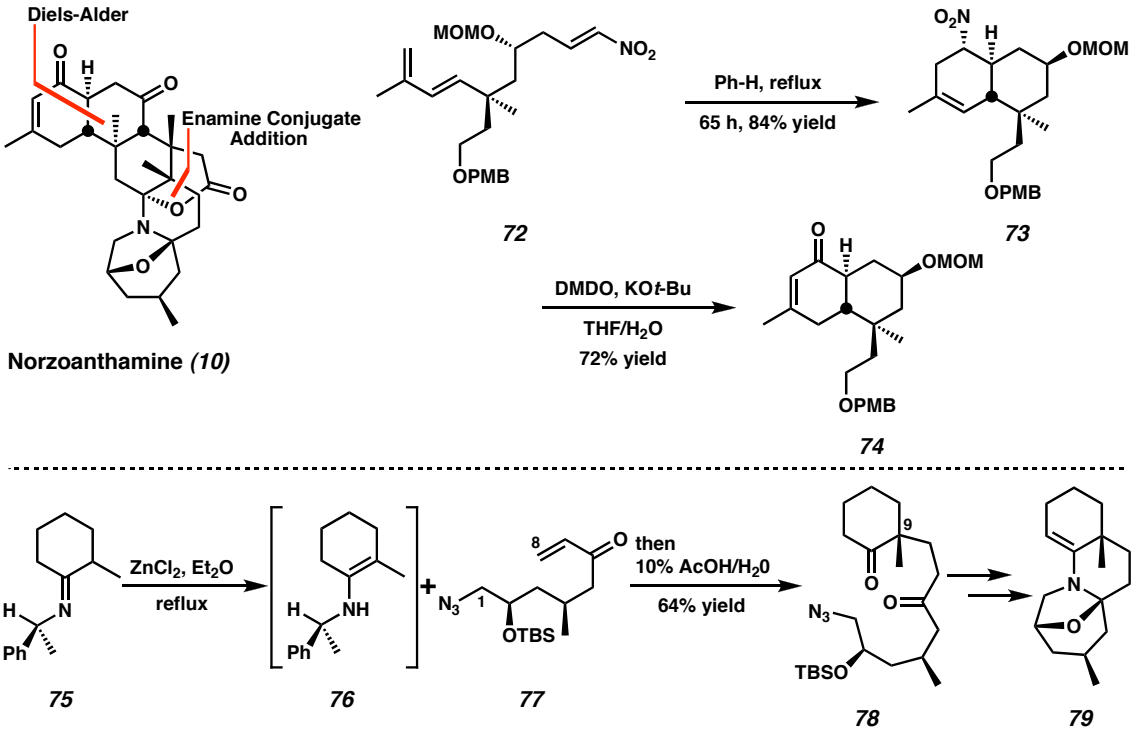
1.4.5 *Williams's Approach to the Norzoanthamine AB and EFG Ring Systems*

Williams and coworkers have contributed approaches to both the synthesis of the carbocyclic AB rings and the heterocyclic EFG rings. Their Diels-Alder strategy constructs the AB rings and subsequently appends the C ring (Scheme 1.9).[46] In the event, nitro alkene **72** underwent reaction in refluxing benzene via an *endo* transition state to afford decalin **73** in good yield and 10:1 dr. A Nef reaction[47] converted the nitro moiety to the desired ketone and facilitated olefin migration. The product enone **74** has the necessary stereochemistry and functionality to begin C ring annulation.

In addition, the Williams group demonstrated an efficient strategy to append the C(1)-C(8) fragment to the ABC ring system and stereospecifically establish the C(9) quaternary center.[48] When heated with zinc (II) chloride, chiral imine **75** generates a significant amount of enamine **76** at equilibrium. The enamine **76** undergoes conjugate addition from the β-face (over the smaller methyl group) of the energy minimized

conformation depicted. Enone **77** was prepared in enantioenriched form using the Evans chiral oxazolidinone.[49] Hydrolysis of the intermediate imine affords diketone **78** with excellent diastereoselectivity (22:1). Upon Staudinger reduction and silyl deprotection of azide **78**, the desired imine formation, ketalization, and dehydration occur without further intervention to give the EFG model enamine **79**.

Scheme 1.9 William's Efforts Toward Norzoanthamine

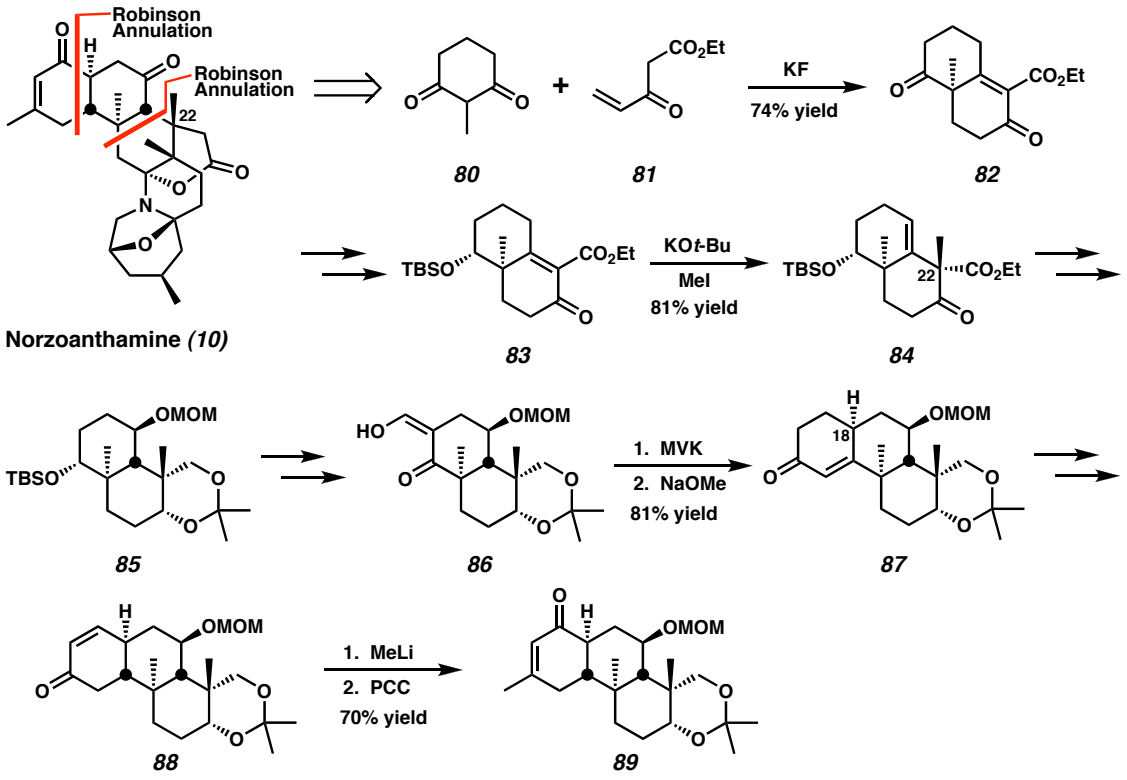


1.4.6 *Theodorakis's Annulation Approach to the Norzoanthamine ABC Ring System*

The annulation strategy proposed by Theodorakis and coworkers is unique in that it starts with the B ring intact and sequentially appends the C and A rings.[50] Condensation of diketone **80** and ketoester **81** with potassium fluoride conditions previously described by their group afforded enone **82** (Scheme 1.10).[51] The formation of

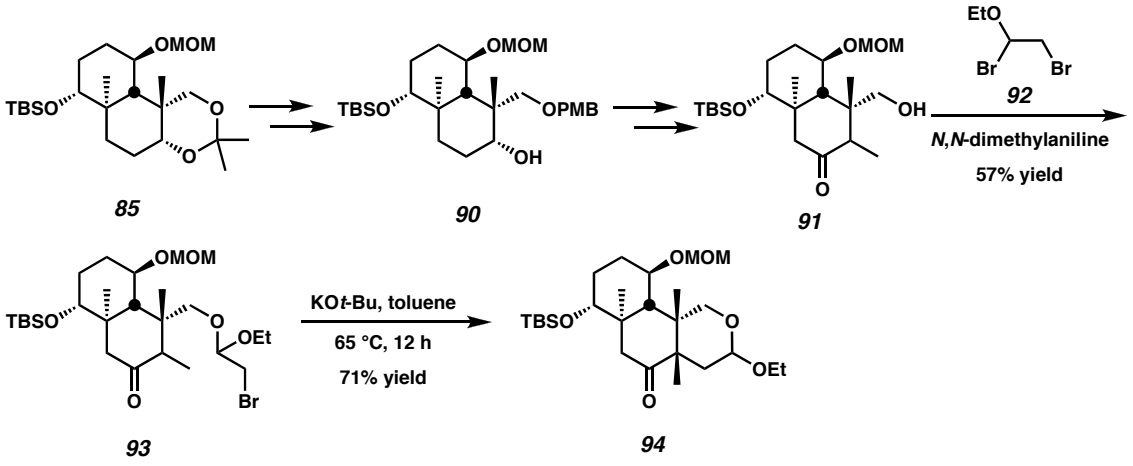
the quaternary stereocenter at C(22) was the next challenge. Treatment of α,β unsaturated ketoester **83** with potassium *tert*-butoxide and methyl iodide produced the quaternized ketoester **84** with complete diastereomeric control. After significant functional group modification, hydroxymethylene-ketone **86** was ready for A ring annulation. A two-step Robinson annulation[52] protocol gave enone **87** as a single isomer at the newly formed C(18) center. After straightforward transformation to enone **88**, methyl lithium addition and PCC oxidation afforded the transposed enone **89**, which contained all the functionality and stereochemistry in the AB rings.

Scheme 1.10 Theodorakis's Annulation Approach to the ABC Ring System



In a related study, Theodorakis demonstrated that the installation of the difficult C(9) quaternary center was possible from intermediate acetonide **85** (Scheme 1.11).[53] Acetonide **85** was advanced by selective protection to give alcohol **90**, which could in turn be converted to methyl ketone **91**. Acetal formation between Stork's dibromo-acetal reagent **92**[54] and the alcohol moiety of methyl ketone **91** produced bromide **93**. Exposure of bromide **93** to base gave a single intramolecular alkylation product **94** in 71% yield. The efficiency of this protocol is impressive given the difficulty of establishing vicinal quaternary centers. Additionally, the alkylation gave complete selectivity for the desired C(9) diastereomer of acetal **94**, as affirmed by X-ray structure determination. Taken in conjunction with Theodorakis's other work, this strategy solved the difficult problem of generating all three of the C ring quaternary centers and produced a norzoanthamine ABC ring system well accoutered for the completion of the total synthesis.

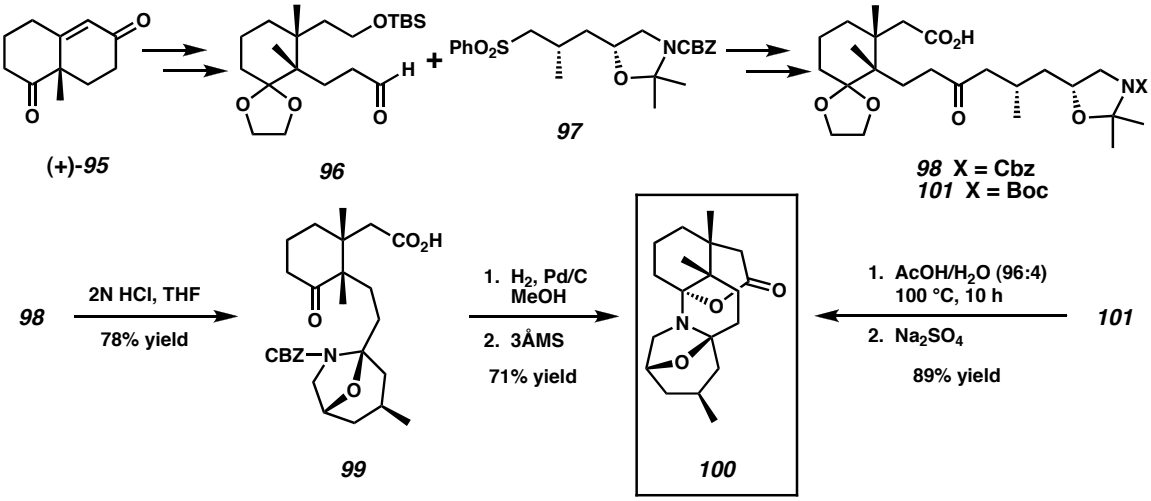
Scheme 1.11 Theodorakis's Installation of the C(9) Quaternary Center



1.4.7 *Kobayashi's Synthesis of the Heterocyclic CDEFG Zoanthamine Ring System*

In 1998 Kobayashi and coworkers disclosed an enantioselective route to the complete CDEFG ring system.[55] The Wieland-Miescher ketone **95**[56] served as the starting material to produce aldehyde **96** (Scheme 1.12). The coupling of sulfone **97**'s lithium salt to aldehyde **96** and oxidation state adjustment completed the Cbz protected cyclization substrate **98**. Treatment with hydrochloric acid removed the acetonide and formed the FG rings. Tricyclic intermediate **99** underwent hydrogenolysis and dehydration to complete the pentacyclic hemiaminal **100**. A one-pot, two-step protocol for cyclization, using Boc protected substrate **101** and acidic conditions, was subsequently investigated and gave an excellent yield of the hemiaminal **100**.[57]

Scheme 1.12 Kobayashi's Sulfone Approach to the Heterocyclic DCEFG Ring System

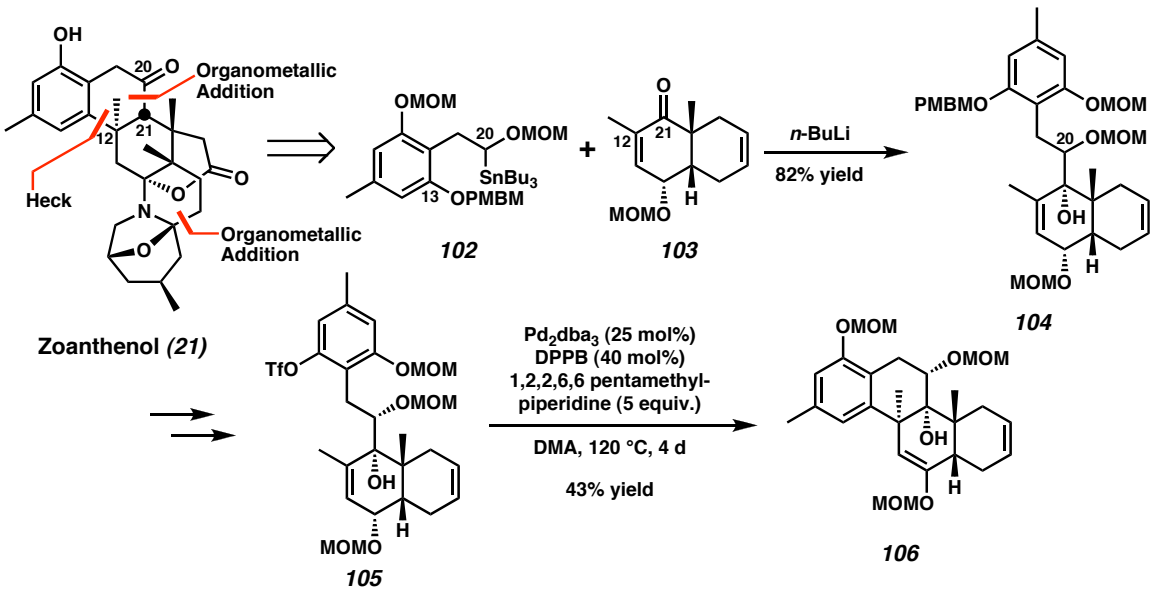


1.4.8 *Hirama's Strategy for the Zoanthenol ABC Ring System*

The strategy proposed by Hirama and coworkers is uniquely geared toward the synthesis of zoanthenol's ABC ring system. The key Heck[58] disconnection of the C(12)-

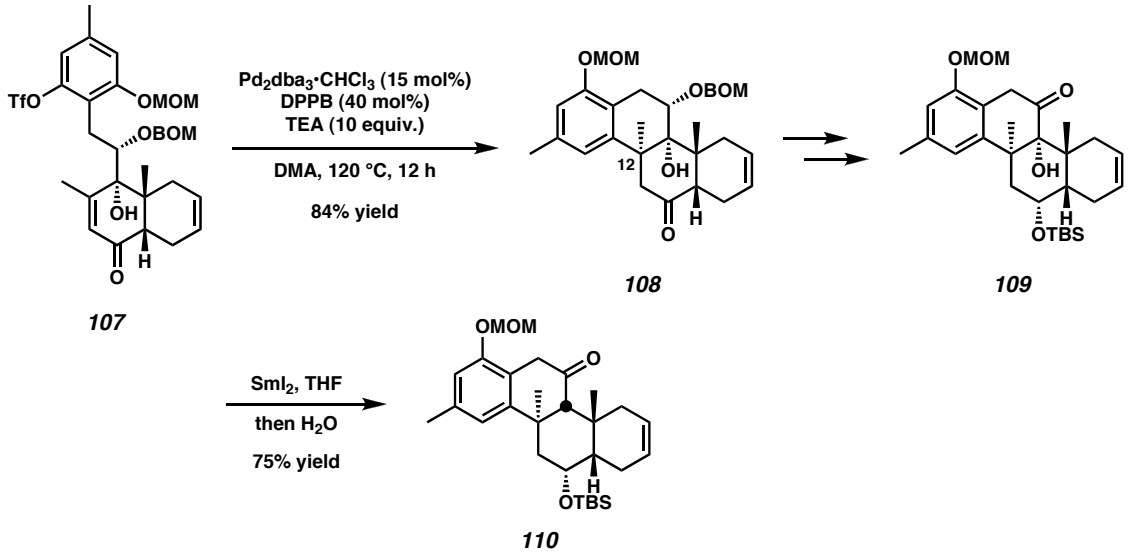
C(13) bond relies on the aromatic A ring unique to zoanthenol (Scheme 1.13). Further disconnection of the B ring at the C(20)-C(21) bond reveals stannane **102** and enone **103** as appropriate fragments. The C ring synthon, enone **103**, was envisioned to arise in enantioenriched form from an asymmetric quinone Diels-Alder reaction also developed in the Hirama group.[59] In practice, transmetallation of stannane **102** and addition into enone **103** afforded tertiary alcohol **104** as a mixture of diastereomers at C(20). Straightforward conversion of tertiary alcohol **104** to triflate **105** allowed for the investigation of the key intramolecular Heck reaction. After significant optimization, conditions were discovered to produce the desired enol ether **106** in modest yield.[60] Though the reaction did proceed with excellent diastereoselectivity, it had several drawbacks, including reduction of the substrate, high palladium loading, and long reaction times.

Scheme 1.13 Hirama's Heck Strategy for the Zoanthenol ABC Ring System



In light of these problems, Heck substrate **105** was altered to increase the electrophilicity of the accepting olefin. As shown in Scheme 1.14, exposure of enone **107** to reductive Heck conditions produced ketone **108** in excellent yield. With the difficult C(12) stereocenter established, their next goal was the reduction of the tertiary alcohol moiety of ketone **109**. Samarium (II) iodide gave the reduced ketone **110** as a single diastereomer and in good yield.[61]

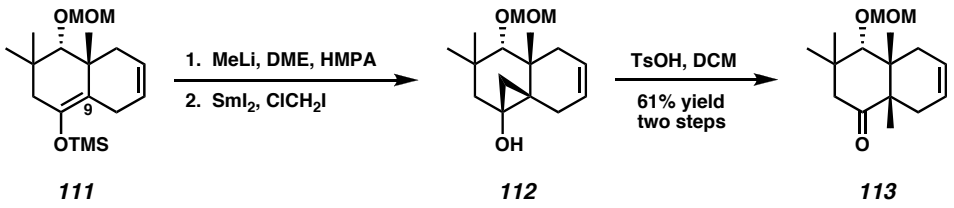
Scheme 1.14 Hirama's Alternate Heck Strategy



107 *108* *109* *110*

The largest challenge that remains in Hirama's synthesis is the establishment of the C(9) quaternary stereocenter. However, his group has already demonstrated a highly diastereoselective methylation of silyl enol ether **111** as a model of methylation at C(9) (Scheme 1.15).[62] The methylation was achieved by samarium (II) iodide promoted cyclopropanation and acid-mediated ring opening to give methyl ketone **113** and its C(9) epimer with a favorable 3:1 dr.

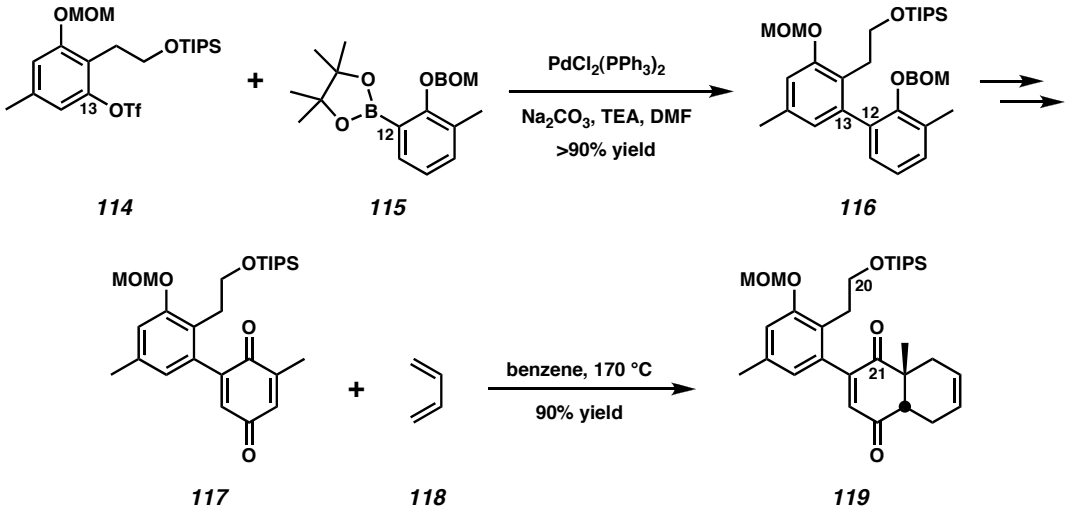
Scheme 1.15 Hirama's Installation of the C(9) Methyl



111 *112* *113*

Recently, Hirama and coworkers have disclosed an alternate strategy for the assembly of zoanthenol's ABC ring system. This strategy reverses the order in which the B ring bonds are formed.[63] As depicted in Scheme 1.16, Suzuki coupling of aryl triflate **114** and borane **115** unite the A and C ring synthons via the C(12)-C(13) bond to yield biaryl **116**. Upon the elaboration of Diels-Alder adduct **119**, the final B ring bond, C(20)-C(21), will likely be constructed by an organometallic addition analogous to the synthesis of tertiary alcohol **104**.

Scheme 1.15 Hirama's Alternate Heck Strategy



114 *115* *116*

117 *118* *119*

1.5 Conclusion

The zoanthamine alkaloids are a structurally unique family of natural products. Though they are isolated from soft coral of the order zoantharia, it may be that symbiotic algae play a large role in the biosynthesis of these secondary metabolites. Their biosynthesis is believed to be polyketide in origin, but no specifics of the pathway are known. The benefit of these complicated natural products to the producing organisms is unknown, but the isolation of various zoanthamine alkaloids in the Indian, Pacific, and Atlantic Oceans suggests that these widespread metabolites may have an important function. Species in the order zoantharia have produced several classes of biologically active natural products. The zoanthamines are no exception. Antiosteoporotic, antibiotic, anti-inflammatory, and cytotoxic biological activities have been discovered in various zoanthamines. As a result, these molecules have garnered increasing attention from synthetic chemists.

As synthetic targets, the zoanthamine alkaloids are a challenge to current synthetic methods and an inspiration for the creation of new reactions. In the contemporary era, it is common for newly isolated natural products of interesting structure or biological significance to succumb to total synthesis in one to two years' time. By comparison, twenty years passed between the isolation of zoanthamine and Miyashita's total synthesis of norzoanthamine in 2004. Any successful synthesis of these alkaloids requires expertise in both carbocyclic and heterocyclic chemistry. Construction of the carbocyclic ABC rings is hindered by the stereochemical density of this tricycle. In particular, the three quaternary centers of the C ring represent a major challenge. This challenge has inspired a number of creative annulation strategies utilizing Diels-Alder,

Heck, and Robinson annulation reactions. The heterocyclic DEFG rings are topographically complex and contain a number of sensitive functional groups. Pioneering syntheses of the heterocyclic region of these molecules determined the feasibility of different cyclization strategies.

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(27) The dose range given in the text (0.016-0.4 mg/kg/d) for $p < 0.05$ is from ref. 20. However, in ref. 27 only dosing at 0.4 mg/kg/day gave a significant ($p < 0.05$) increase in failure load. No explanation for this disparity is given.

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