Synthesis of analogs of aflatoxin B2 and glycinoeclepin A

Beth Ellen Johnston
Iowa State University

Follow this and additional works at: https://lib.dr.iastate.edu/rtd

Part of the Organic Chemistry Commons

Recommended Citation
INFORMATION TO USERS

This manuscript has been reproduced from the microfilm master. UMI films the text directly from the original or copy submitted. Thus, some thesis and dissertation copies are in typewriter face, while others may be from any type of computer printer.

The quality of this reproduction is dependent upon the quality of the copy submitted. Broken or indistinct print, colored or poor quality illustrations and photographs, print bleedthrough, substandard margins, and improper alignment can adversely affect reproduction.

In the unlikely event that the author did not send UMI a complete manuscript and there are missing pages, these will be noted. Also, if unauthorized copyright material had to be removed, a note will indicate the deletion.

Oversize materials (e.g., maps, drawings, charts) are reproduced by sectioning the original, beginning at the upper left-hand corner and continuing from left to right in equal sections with small overlaps. Each original is also photographed in one exposure and is included in reduced form at the back of the book.

Photographs included in the original manuscript have been reproduced xerographically in this copy. Higher quality 6" x 9" black and white photographic prints are available for any photographs or illustrations appearing in this copy for an additional charge. Contact UMI directly to order.
Synthesis of analogs of aflatoxin B₂ and glycinoeclepin A

Johnston, Beth Ellen, Ph.D.
Iowa State University, 1992
Synthesis of analogs of aflatoxin B₂ and glycinoeclepin A

by

Beth Ellen Johnston

A Dissertation Submitted to the
Graduate Faculty in Partial Fulfillment of the
Requirements for the Degree of
DOCTOR OF PHILOSOPHY

Department: Chemistry
Major: Organic Chemistry

Approved:

Signature was redacted for privacy.

In Charge of Major Work

Signature was redacted for privacy.

For the Major Department

Signature was redacted for privacy.

For the Graduate College

Iowa State University
Ames, Iowa

1992
DEDICATION

This dissertation is dedicated to my family, whose love, support, and faith in me made it possible.
# TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>GENERAL INTRODUCTION</td>
<td>1</td>
</tr>
<tr>
<td>PAPER I: TOTAL SYNTHESIS OF RACEMIC DEMETHOXYAFLATOXIN B₂</td>
<td>3</td>
</tr>
<tr>
<td>INTRODUCTION</td>
<td>4</td>
</tr>
<tr>
<td>HISTORICAL</td>
<td>5</td>
</tr>
<tr>
<td>RESULTS AND DISCUSSION</td>
<td>27</td>
</tr>
<tr>
<td>CONCLUSION</td>
<td>36</td>
</tr>
<tr>
<td>EXPERIMENTAL</td>
<td>37</td>
</tr>
<tr>
<td>REFERENCES</td>
<td>44</td>
</tr>
<tr>
<td>PAPER II: SYNTHETIC APPROACHES TOWARD ANALOGS OF GLYCINOECLEPIN A</td>
<td>49</td>
</tr>
<tr>
<td>INTRODUCTION</td>
<td>50</td>
</tr>
<tr>
<td>HISTORICAL</td>
<td>51</td>
</tr>
<tr>
<td>RESULTS AND DISCUSSION</td>
<td>64</td>
</tr>
<tr>
<td>CONCLUSION</td>
<td>115</td>
</tr>
<tr>
<td>EXPERIMENTAL</td>
<td>116</td>
</tr>
<tr>
<td>REFERENCES</td>
<td>132</td>
</tr>
<tr>
<td>APPENDIX: BIOLOGICAL TESTING</td>
<td>137</td>
</tr>
<tr>
<td>GENERAL SUMMARY</td>
<td>138</td>
</tr>
<tr>
<td>ACKNOWLEDGMENTS</td>
<td>139</td>
</tr>
</tbody>
</table>
ABBREVIATIONS

The following abbreviations are used in this dissertation:

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ar</td>
<td>aryl</td>
</tr>
<tr>
<td>9-BBN</td>
<td>9-borabicyclo[3.3.1]nonane</td>
</tr>
<tr>
<td>Bn</td>
<td>benzyl</td>
</tr>
<tr>
<td>BPS</td>
<td>tert-butyldiphenylsilyl</td>
</tr>
<tr>
<td>Bu</td>
<td>butyl</td>
</tr>
<tr>
<td>t-Bu</td>
<td>tert-butyl</td>
</tr>
<tr>
<td>Bz</td>
<td>benzoyl</td>
</tr>
<tr>
<td>DBU</td>
<td>1,8-diazabicyclo[5.4.0]undec-7-ene</td>
</tr>
<tr>
<td>DDQ</td>
<td>2,3-dichloro-5,6-dicyano-1,4-benzoquinone</td>
</tr>
<tr>
<td>DIBAL-H</td>
<td>diisobutylaluminum hydride</td>
</tr>
<tr>
<td>DME</td>
<td>dimethoxyethane</td>
</tr>
<tr>
<td>DMF</td>
<td>dimethylformamide</td>
</tr>
<tr>
<td>DMSO</td>
<td>dimethylsulfoxide</td>
</tr>
<tr>
<td>Et</td>
<td>ethyl</td>
</tr>
<tr>
<td>g</td>
<td>gram</td>
</tr>
<tr>
<td>h</td>
<td>hour(s)</td>
</tr>
<tr>
<td>HMDS</td>
<td>hexamethyldisilazane</td>
</tr>
<tr>
<td>HMPA</td>
<td>hexamethylphosphoric triamide</td>
</tr>
<tr>
<td>LDA</td>
<td>lithium diisopropylamide</td>
</tr>
<tr>
<td>LTA</td>
<td>lead tetraacetate</td>
</tr>
<tr>
<td>m-CPBA</td>
<td>m-chloroperoxybenzoic acid</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Definition</td>
</tr>
<tr>
<td>--------------</td>
<td>------------</td>
</tr>
<tr>
<td>Me</td>
<td>methyl</td>
</tr>
<tr>
<td>MEM</td>
<td>(2-methoxyethoxy)methyl</td>
</tr>
<tr>
<td>min</td>
<td>minute(s)</td>
</tr>
<tr>
<td>mmol</td>
<td>millimole(s)</td>
</tr>
<tr>
<td>MMC</td>
<td>methyl methoxymagnesium carbonate</td>
</tr>
<tr>
<td>MMPP</td>
<td>monoperoxyphthalic acid, magnesium salt</td>
</tr>
<tr>
<td>MOM</td>
<td>methoxymethyl</td>
</tr>
<tr>
<td>Ms</td>
<td>methanesulfonyl (mesyl)</td>
</tr>
<tr>
<td>MKV</td>
<td>methyl vinyl ketone</td>
</tr>
<tr>
<td>NBS</td>
<td>N-bromosuccinimide</td>
</tr>
<tr>
<td>PCC</td>
<td>pyridium chlorochromate</td>
</tr>
<tr>
<td>Ph</td>
<td>phenyl</td>
</tr>
<tr>
<td>PM</td>
<td>Phenylmenthyl</td>
</tr>
<tr>
<td>PPTS</td>
<td>pyridium p-toluenesulfonate</td>
</tr>
<tr>
<td>i-Pr</td>
<td>isopropyl</td>
</tr>
<tr>
<td>PTSA</td>
<td>p-toluenesulfonic acid</td>
</tr>
<tr>
<td>Rf</td>
<td>retention factor</td>
</tr>
<tr>
<td>TBDMS</td>
<td>tert-butyldimethylsilyl</td>
</tr>
<tr>
<td>Tf</td>
<td>trifluoromethanesulfonyl</td>
</tr>
<tr>
<td>THF</td>
<td>tetrahydrofuran</td>
</tr>
<tr>
<td>THP</td>
<td>tetrahydropyran</td>
</tr>
<tr>
<td>TLC</td>
<td>thin layer chromatography</td>
</tr>
<tr>
<td>TMS</td>
<td>trimethylsilyl</td>
</tr>
<tr>
<td>Tr</td>
<td>triphenylmethyl (trityl)</td>
</tr>
<tr>
<td>Ts</td>
<td>p-toluenesulfonyl (tosyl)</td>
</tr>
</tbody>
</table>
GENERAL INTRODUCTION

For the synthetic organic chemist, a natural product represents challenge: the synthesis itself, as well as development and application of methodology toward that end. The synthetic chemist is also interested in the synthesis of simpler analogs of such a compound in the hope they will display improved, similar, or at least acceptable biological activity. Such analogs are useful for structure-activity studies and may be more suitable for commercial purposes than compounds obtained from a natural source. Intermediates formed in attempts at total syntheses are often suitable analogs.

The goal of the first part of this research was to explore the use of silver salts to promote radical cyclizations of appropriate substrates to form variously substituted furofuran systems and to apply this methodology to the synthesis of demethoxy aflatoxin B₂, a analog of aflatoxin B₂, a potent naturally occurring toxin. While such reactions have been reported previously, the conditions employed here are much milder and more convenient. In the second part of this research, various analogs of glycinoeclepin A, the naturally occurring hatching stimulus of the soybean cyst nematode, were prepared and tested for hatch-stimulating ability. While few of the compounds tested were active, this information shed light on which portions of the molecule were necessary for activity.
Explanation of the Dissertation Format

This dissertation is divided into two separate papers, each in publishable format. The structures and references for each are therefore numbered independently. Each paper is preceded by an introduction. The first paper covers the synthesis of racemic demethoxy aflatoxin B₂. The second paper deals with the synthesis of analogs of glycinoeclepin A. A general summary of both papers will follow the second paper.
PAPER I: TOTAL SYNTHESIS OF RACEMIC DEMETHOXYAFLATOXIN B₂
INTRODUCTION

This manuscript will describe the total synthesis of racemic demethoxy aflatoxin B₂. The key step in the synthesis is a silver-mediated oxidative radical cyclization to form the aflatoxin B₂ furo[2,3-b]furan system. The conditions developed for this step differ from those reported for many similar methods in that they are mildly basic, rather than acidic. A second unique feature of this synthesis is that, unlike many earlier reported syntheses of aflatoxins, a von Pechmann cyclization was not employed to append the D ring.
HISTORICAL

The aflatoxins are a class of mycotoxins, or toxic fungal metabolites, produced by strains of *Aspergillus flavus*, *A. parasiticus*, and *A. nomius*. Although there are numerous aflatoxins, the four major toxins are B₁, B₂, G₁, and G₂. Aflatoxins M₁ and M₂, metabolites of B₁

![Chemical structures](image)

and B₂, are also biologically significant. While the aflatoxins are often considered together for toxicological purposes, this dissertation will deal primarily with aflatoxins B₁ and B₂ due to their close relationship to each other and to the target molecule.
Demethoxyaflatoxin B₂

Aflatoxins pose significant health hazards to both humans and livestock animals. They are potent liver toxins, and in differing exposure levels in animals they can cause death, liver damage or cancer, a reduction in milk and egg production, and immune suppression. Aflatoxins have also been associated with hepatic, renal, colonic, nasal, and pulmonary neoplasms in various species. Commercially, cattle, poultry, and swine are affected, and chronic, as well as acute, exposure results in serious health problems.

In humans, acute aflatoxicosis has been reported in Taiwan, Uganda, and India with numerous fatalities. A disease responsible for the deaths of several children in Thailand has been linked to aflatoxin. Chronic low-level exposure to aflatoxins is also a concern, as aflatoxins have been implicated as a possible cause of human liver cell carcinoma (LCC). Many studies, particularly in parts of Africa and Asia, have shown a link between the two. Unfortunately, other risk factors of the test populations (for example, exposure to the hepatitis B virus) and conflicting evidence from other studies have left the matter unsettled.
The International Agency for Research on Cancer did, however, place aflatoxin B\textsubscript{1} on its list of human carcinogens in 1988.

Aflatoxins are often found in foods and feeds in the United States. Contamination can occur on the crop in the field or during storage or transportation. Aflatoxins have been found in peanuts, corn, wheat, rice, cottonseed, copra, nuts, various foods, milk, eggs, and cheese. Although strict control of foods in the United States makes acute or chronic aflatoxicosis an insignificant problem here, long-term exposure to low levels is a concern because of the possible link to LCC. Contaminated feeds, which are less tightly regulated, can cause severe livestock health problems.

The above factors translate into economic losses. Detection of toxins, decontamination where applicable, livestock veterinary care and losses, and disposal of contaminated commodities are all costly. Improved methods of detection and analysis are needed, as are further toxicological studies and development of safe means of detoxification and decontamination.\textsuperscript{1} Analogs of the aflatoxins could be highly useful for these applications. Toward that end, a synthetic plan for an analog of aflatoxin B\textsubscript{2} was formulated.

Many total and partial syntheses of the aflatoxins have been reported since Buchi and co-workers\textsuperscript{2} determined the structures of aflatoxins B\textsubscript{1}, B\textsubscript{2}, G\textsubscript{1}, and G\textsubscript{2} in the 1960's. A review by Schuda\textsuperscript{3} summarizes the early work of the Buchi group in this area, as well as that of others.
Buchi and co-workers reported the first total synthesis of aflatoxin B$_1$. Starting from phloroacetophenone-4-methyl ether (8) or dihydroxy-4-methylcoumarin (9), the key intermediate, coumarin 10, was prepared. Oxidation of the allylic methyl to an aldehyde and reduction of the coumarin, which also caused hydrolysis, formed an intermediate which spontaneously cyclized to lactone 11. The phenol from deprotected 11 underwent von Pechmann condensation to yield lactone 12. Hydrolysis of the esters of 12 re-formed the $\gamma$-lactone. Acid chloride formation, Lewis acid catalyzed cyclization, and reduction
of the γ-lactone provided 13 (aflatoxin B$_{2a}$), whose acetate underwent pyrolysis to afford aflatoxin B$_1$.

In 1971, Buchi and Weinreb$^5$ published an improved route to aflatoxin B$_1$ which used a milder variation of the von Pechmann condensation. Intermediate 11 was converted to diacetate 14,

which underwent pyrolysis and deprotection to yield a phenol which in turn afforded aflatoxin B$_1$ upon von Pechmann condensation with a cyclic β-bromoenone with zinc carbonate and sodium bicarbonate.

Roberts published the first synthesis of an aflatoxin B$_2$ ABC ring system.$^6$ Starting from 9, coumarin 15 was prepared. Catalytic hydrogenation, acetate formation, and hydride reduction of the lactone
produced the ABC ring system 16. Compound 16 condensed with 2-carboethoxycyclopentanone (17) to yield tetrahydrodeoxoaflatoxin B1 (18), an analog of aflatoxin B2. A total synthesis of aflatoxin B2 was realized when 16 underwent von Pechmann condensation with diethyl β-keto adipate to yield an ester whose acid chloride cyclized under Lewis acid conditions to afford 2.
In 1986, Castellino and Rapoport\textsuperscript{8} reported a new route to 16 and thus a formal total synthesis of aflatoxin B\textsubscript{2}. Amine 21 reacted with an excess of aldehyde 20 to produce oxime 22 in 83% yield with an E/Z ratio of 18:1. Using conditions developed to avoid Beckmann rearrangement, 22 was subjected to excess anhydrous hydrochloric acid in THF at 65-70 °C in a sealed tube and 3,4-oxaza Cope product 23 resulted in 87% yield. Selective hydrolysis of 23 to a bis-mesylate was not possible; a mixture of mono-mesylates was always produced. The hydrolysis product mixture was cyclized to form the furofuran system.
Benzylation of the phenol allowed separation of the isomers. Hydrolysis, methylation, and transfer hydrolysis then afforded the target molecule 16 in 47% overall yield.

Rodrigo et al.\textsuperscript{9} also published a new route to 16 which involved a lead tetraacetate-mediated cyclization of an aromatic iodide as a key step. Iodination of 3,5-dimethoxyphenol (25) and formation of the ether provided 26 as a substrate for LTA-iodine intramolecular conjugate addition to yield 27 after deiodination. Reduction and iodination of 27 provided 28. Compound 28 cyclized with LTA-I\textsubscript{2} to afford a tricyclic product which underwent deiodination and transfer hydrogenolysis to yield 16 in ca. 4% overall yield, completing the formal total synthesis of aflatoxin B\textsubscript{2}.

Rodrigo et al.\textsuperscript{10} later used an iodide as a key intermediate in their total synthesis of aflatoxin B\textsubscript{2}. Tosylation of 28, cyclization with LTA-I\textsubscript{2},
and deprotection afforded diiodide 29, which could also be produced by diiodination of 16. Deiodination with one equivalent of sodium

\[
\begin{align*}
28 & \quad \text{→} \quad 29 \\
& \quad \text{MeO} \\
29 & \quad \text{→} \quad 30 \\
& \quad \text{MeO} \\
& \quad \text{I}
\end{align*}
\]

hydride followed by one equivalent of n-BuLi at -100 °C and benzylation of the phenol provided iodide 30. Michael addition of the mixed cuprate from 30 to ethyl cyclopentenone-2-carboxylate afforded 31. Cyclization of 31 with trifluoroacetic acid and dehydrogenation with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) provided aflatoxin B₂ in 2.3% overall yield from 3,5-dimethoxyphenol.

In 1991, Messeguer et al. synthesized an aflatoxin B₁ model, 3a,8a-dihydro-4,6-dimethoxyfuro[2,3-b]benzofuran (34). The synthetic approach was based upon Buchi's work, and started with 3,5-dimethoxyphenol (25). Condensation of 25 with methylacetoacetate with methanesulfonic acid catalysis yielded coumarin 32, which
provided an aldehyde upon allylic oxidation. When treated with zinc in acetic acid, the lactone opened and recylcized to tricyclic lactone 33. Reduction of 33 and pyrolysis of its corresponding carbonate led to the target, 34. Epoxidation of 34 with dimethyl dioxirane provided a compound for comparative biological testing.

Recently, Pirrung et al. used the cycloaddition of cyclic rhodium carbenoids to aromatic heterocycles\(^{12}\) to generate heterocyclic systems with potential as aflatoxin intermediates. Reaction of 35 with dihydrofuran or furan with rhodium acetate produced 36 and 37 respectively.
While not covered in this dissertation, syntheses of various other aflatoxins have also been reported. Buchi et al. reported a synthesis of aflatoxin M₁ in 1981. Kraus et al. recently published an approach to aflatoxin M₂ using Type II photocyclization reactions.

The idea of applying metal-mediated oxidative radical cyclizations to the synthesis of rings was attractive, and had an extensive history. In 1968, Bush and Finkbeiner and Heiba et al. almost simultaneously published reports that olefins and acetic acid reacted in the presence of manganese(III) acetate to produce γ-lactones. Bush and Finkbeiner reported low to modest unoptimized yields of lactones using mono- and disubstituted olefins, Mn(OAc)₃•2H₂O, and acetic anhydride in acetic acid at reflux temperature. They felt the •CH₂CO₂H radical was not involved, but a manganese-acid complex of some sort was. Heiba et al. also used mono- and disubstituted olefins, but
reported moderate to good yields of lactones. They employed \( \text{Mn(OAc)}_3 \cdot \text{2H}_2\text{O} \) and sodium acetate in boiling acetic acid. Propionic acid afforded lactones with an \( \alpha \)-methyl substituent. They felt that \( \cdot \text{CH}_2\text{COOH} \) radicals were produced by thermolysis of the manganic complex, and that they added to the olefins faster than they abstracted allylic protons or were oxidized.

After the publication of this pioneering work, numerous extensions, enhancements, and explanations followed. In 1973, Nikishin \textit{et al.}\(^\text{17}\) reported that in the presence of manganese(III) or cobalt(III) and copper(II) acetate, diethyl malonate and diethyl alkylmalonates reacted with olefins to produce diethyl alk-2-enylmalonates. These, in turn, produce \( \gamma \)-lactones upon acidic hydrolysis.

\[
\text{HCR'(CO}_2\text{Et})_2 + \text{R}_2\text{CH}_2\text{C(R}_3\text{)=CH}_2 \xrightarrow{\text{Mn(OAc)}_3 \text{ or Co(OAc)}_3, \text{Cu(OAc)}_2} \xrightarrow{\text{H}_2\text{O, H}^+} \text{R}_2\text{R}_1\text{R}_3\text{O} \]

Heiba and Dessau extended their earlier work in 1974 to include the reaction of \( \beta \)-diketones and \( \beta \)-ketoesters\(^\text{18}\) and then carboxylic acids\(^\text{19}\) with olefins in the presence of manganic acetate. A wide range of yields, mostly satisfactory, were reported for both processes. In the more extensive study on the acids, it was found that various acids, including propionic and cyanoacetic, could be used, as well as high-valent metals other than manganese. Conditions were optimized
(elevated temperatures, addition of carboxylate salt, and addition of acetic anhydride to scavenge large amounts of water if the Mn(OAc)₃ was prepared \textit{in situ}). A complete mechanism was also proposed:
A study by Okano\textsuperscript{20} employing acetic acid/acetic anhydride or propionic acid/propionic anhydride as a solvent system proposed that the radicals involved in the reactions were from the solvent, rather than from the manganese complex itself. The yields of lactones in this study were generally low.

Kurosawa\textit{ et al.}\textsuperscript{21} found that olefins reacted with malonic acid in acetic acid in the presence of manganese(III) acetate to produce substituted 2,7-dioxaspiro[4.4]nonane-1,6-diones in variable yields.

\[
\text{Ph} = \begin{array}{c}
\text{Ph} \\
\text{H} \\
\text{Ph}
\end{array} + \text{HOAc} \xrightarrow{\text{Mn(OAc)}_3, \text{HOAc}} \text{Ph} \xrightarrow{\text{84\%}}\text{Ph} \xrightarrow{\text{42}}
\]

In 1983, Nikishin\textit{ et al.}\textsuperscript{21} found that saturated acids and amides could be cyclized to $\gamma$-lactones in moderate yields, along with $\delta$-lactone by-products, using Na$_2$S$_2$O$_8$-CuCl$_2$ in aqueous media. Regioselectivity for $\gamma$- vs. $\delta$-lactones was found to be greater for the acids. The proposed mechanism is shown:
Corey and co-workers published several reports on the use of manganese(III) acetate to form heterocycles. In 1984, Corey and Kang used intramolecular cyclization to produce tricyclic lactones with possible applications toward natural products such as the gingkolides.

These reactions occurred at 23-40 °C in reasonable yields.

With reactive acids, such as cyanoacetic acid, maximum lactone yields were achieved with short reaction times. Longer times allowed further reaction to form 48. Ethyl hydrogen malonate and ethyl
hydrogen chloromalonate\textsuperscript{24} also gave good yields of $\alpha$-substituted $\gamma$-lactones.

Corey and Ghosh\textsuperscript{25} published a synthesis of furans using manganese(III)-promoted cyclizations of enol ethers with $\beta$-dicarbonyl compounds. Cyclic and acyclic enol ethers were used, and a variety of

[$\text{\begin{align*} &\text{\(CH_2\)n} OMe + COOEt &\xrightarrow{\text{Mn(III)}} \text{\(CH_2\)n} OMe + \text{\(C\)O} OEt \end{align*}}\]  

$\beta$-dicarbonyl compounds were also found to be successful. Mechanistically, Corey felt that coordination of the carbonyl with manganese formed a "radicaloid" which then attacked the olefin.

Fristad and coworkers were also active in this area of research. Fristad and Peterson\textsuperscript{26} reported that acetic anhydride appeared to enhance reaction rates, but did so because it was oxidized more rapidly than acetic acid. Addition of the anhydride altered the product mixture; unsaturated acids were produced along with reduced amounts of lactones. Optimized reaction conditions and regio- and stereochemical outcomes were discussed. A mechanism involving a radical as part of a manganese complex was also proposed. Ernst and Fristad\textsuperscript{27} used manganese(III) acetate to effect intramolecular cyclizations of unsaturated activated carboxylic acids to bicyclo[3.3.0]- and -[4.3.0]-lactones. Formation of 2,7-dioxaspiro[4,4]nonane-1,6-diones from
malonic acid and olefins was reported by Fristad and Hershberger,\textsuperscript{28} with due note of essentially the same results published earlier by Kurosawa.\textsuperscript{21} Rapid exchange with ligands in the manganese complex was proposed. Synthesis of several $\alpha$-substituted-$\gamma$-lactones using substituted acetic acids was reported.\textsuperscript{29} Although similar reactions

\[
R^1R^2C=CR^3R^4 + [\text{Mn}_3\text{O}] + \text{HO}_2\text{CCH}_2X \xrightarrow{\Delta \text{HOAc}} \begin{array}{c}
\text{O} \\
\text{X} \\
\hline
\text{R}_1 \\
\text{R}_2 \\
\text{R}_3 \\
\text{R}_4
\end{array}
\]

\[
X = \text{Cl, CH}_2\text{Cl, CN, COOMe}
\]

had been published, this report investigated more substituents ($X=\text{S(O)Ph, SO}_2\text{Ph, NO}_2, \text{P(O)(OE)}_2, \text{NMe}_3^+, \text{and Br}$ were also used and found unsatisfactory) and details of stereochemistry and optimal conditions were described. Use of a manganese(III) acetate mediated cyclization in a route to substituted tetralones was also reported.\textsuperscript{30}
In a departure from the widely used manganese(III) acetate in acetic acid, Citterio et al.\textsuperscript{31} found that oxidative deprotonation of carbonyl compounds occurred with iron(III) salts in acetonitrile or acetic anhydride. Moderate yields of \( \gamma \)-lactones, along with other oxidative addition products, were achieved when \( \beta \)-diesters reacted with olefins, inter- or intramolecularly, in the presence of iron(III) perchlorate nonahydrate in acetonitrile.

Paquette et al.\textsuperscript{32} used manganese (III) mediated cyclization to produce a key intermediate in a synthesis of (\( \pm \))-14-epiupial.
Interestingly, the epimer of 58 with the MOM group cis to the methyl group refused to cyclize. Stereogenic factors are believed to be involved, as well as possible heteroatom effects.

Surzur et al. investigated the reaction of allylic esters of acetoacetic acid and malonic acid with manganese(III) acetate, copper(II) acetate, and potassium acetate in acetic acid.33 Substituted γ-lactones, bis γ-lactones, or cyclopropyl γ-lactones were produced, depending on the substitution pattern of the starting material.
In a short review of lactone synthesis by electron transfer and radical chemistry, Surzur and Bertrand described intramolecular cyclizations of acrylic acids in aqueous acetonitrile with Na2S2O8 and catalytic silver nitrate and copper(II). The reaction of allylic esters of
acetoacetic and malonic acids with manganese(III) and copper(II) to produce various lactones and bis-lactones was further explored. Propargylic esters were also found to react under the same conditions to produce \( \gamma \)-lactones.

![Chemical structure](image)

Vanadium(V) compounds can be used for one-electron oxidations. Hirao et al. found that ethyl acetoacetate reacted with \( \alpha \)-methylstyrene and \( \text{VO(OEt)Cl}_2 \) in ethanol to afford lactone 72 in 52% yield.35

![Chemical structure](image)

Snider and co-workers have been very active in the area of manganese(III)-mediated oxidative radical cyclizations.36 As this work deals primarily with the formation of carbocycles, rather than heterocycles, it will not be detailed here. However, a recent publication detailing new, milder reaction conditions is relevant to the work to be presented. Until this work, most metal-mediated oxidative radical cyclizations were performed under rather harsh conditions: carboxylic
acid solvents, and often elevated temperatures. Ethanol\textsuperscript{37} was recently employed as a solvent, and the results compared to cyclizations performed in acetic acid. It was found that a greater percentage of 5-exo cyclization products was produced in cyclizations in ethanol than in acetic acid. Ethanol also acted as a reducing agent for primary and vinyl radicals. If copper(II) acetate is used to oxidize primary cyclopentanemethyl radicals, alkenes are mainly produced in ethanol, while alcohols and lactones are produced preferentially in acetic acid. It should be noted, however, that products from this radical are minor overall products in the reaction in acetic acid. Products from a cyclohexyl radical predominate. Of course, the main advantage of ethanol as a solvent is that acid-sensitive substrates, such as many enol ethers, may be used in these metal-mediated radical cyclizations.

In view of the work published at the time our work on aflatoxin analogs was begun, we were drawn to a report by Saegusa \textit{et al.} which used the relatively mild conditions of silver(I) oxide in DMSO to dimerize \( \beta \)-ketoesters.\textsuperscript{38}

\[
\begin{align*}
R\text{CH}_2\text{X} & \xrightarrow{\text{Ag}_2\text{O}} \xrightarrow{\text{X}X} \xrightarrow{R\text{CH}C\text{C}R} \xrightarrow{YY} X, Y = \text{COR, CO}_2\text{R, etc.} \\
\text{Y} & \xrightarrow{\Delta} \text{DMSO} \\
\end{align*}
\]

Hoping to employ these mild conditions to perform cyclizations like those described above, work was begun.
RESULTS AND DISCUSSION

The strategy initially envisioned was to use the conditions of Saegusa to form a substituted aflatoxin $\text{B}_2[2,3-b]$furofuran system and then continue with construction of the remaining three rings.

\[
\text{Aflatoxin B}_2
\]

Cyclizations were initially run with silver(I) oxide in DMSO. However, it was discovered that acetonitrile, with a dielectric constant similar to DMSO, could also be used successfully and more conveniently. Easily prepared silver(I) carbonate was then found to work well as an oxidant. Being mildly basic and employing a readily removable solvent, silver(I) carbonate in near-boiling acetonitrile provides an attractive alternative to the widely used conditions using carboxylic acids as solvents for oxidative radical cyclizations.

Ethyl acetoacetate reacted with an excess of 2,3-dihydrofuran and silver carbonate in hot acetonitrile to afford furofuran 75 in 50% yield after chromatographic purification. It was hoped that 75 could be converted to diene 76, which could undergo a Diels-Alder reaction to append the A ring to the system. When 75 was treated with LDA and
TMSCI, diene 76 did not result. It appeared that C- rather than O-silylation had occurred. Unfavorable steric interactions in an initial O-silylated product may have encouraged the transfer of the silicon to the carbon. When 75 was treated with the more reactive TMSOTf and diisopropylethylamine, it decomposed, likely due to reaction with the acetal.

An intramolecular cyclization of an appropriately substituted furofuran was next eyed to build the A ring. Compound 75 reacted with LDA and acetaldehyde to produce an alcohol, which afforded ketone 77 on oxidation with pyridinium chlorochromate (PCC). Unfortunately, attempts to cyclize 77 with sodium methoxide in
methanol at temperatures from -15 °C to 120 °C (sealed tube) failed to produce the desired benzofuran 78. A theory for this failure is that there may have been an insufficient concentration of the necessary ketone enolate under the thermodynamic conditions employed. Compound 81 was then synthesized, with the hope its greater tendency toward enolization would facilitate cyclization. Treatment of 75 with LDA and aldehyde 79 and subsequent oxidation of the alcohol produced with PCC provided ketoacetal 80. The ketal proved more difficult to hydrolyze than anticipated; it resisted PTSA in acetone and silica gel and aqueous oxalic acid in dichloromethane. Treatment with 5% HCl in THF yielded diketone 81. Numerous attempts were made to cyclize 81: KOH in methanol or water, NaOMe/MeOH, LiH, t-BuOLi, and t-BuOMg, all at temperatures from 0 °C to 86 °C. The desired
benzofurofuran was not produced. Heating at 210 °C in an open tube also failed to yield 82. These reactions generally returned starting material and/or decomposition products.

At this point, a new route to the benzofurofuran was sought. Direct formation via the developed silver(II) carbonate methodology seemed appropriate. Cyclohexane-1,3-dione reacted with 2,3-dihydrofuran and silver(II) carbonate to produce the furofuran 83 in good yields (55-70%).

With 83 in hand, attachment of the A-ring methoxy group and "handles" upon which to build the D and E rings were necessary. Treatment of 83 with cupric chloride dihydrate and lithium chloride in
DMF\textsuperscript{44} afforded phenol 84 in 22% yield. Unfortunately, attempts to produce a substituted phenol failed. Reaction of 83 with sulfuryl chloride\textsuperscript{45} did not produce the $\alpha,\alpha$-dichloride. Treatment with LDA and MoOPH\textsuperscript{46} gave a promising result, but was not pursued due to the hazards involved. While triethylamine and TMSOTf failed to produce the enol silyl ether of 83, LDA and TMSCl was successful.
Oxidations of 87 with palladium (II) acetate, MMPP (monoperoxyphthalic acid, magnesium salt), or m-CPBA all failed to yield the phenol 84. Efforts to add PhSCl or bromine to 87 were also fruitless.

As LDA had successfully generated the enolate of 83 in the formation of its enol silyl ether, use of the enolate was further explored. As addition to acetaldehyde was also promising, Michael addition to an appropriate Michael acceptor seemed attractive.

Treatment of 83 with LDA and ketoester 89 produced Michael adduct 90, which in turn led to enone 91 after formation of the selenide and oxidation. Reaction of DBU or PTSA with 91 failed to produce a cyclized product. Heating at ~200 °C in toluene in a sealed tube produced some cyclized product as well as the aromatic target molecule 7. Treatment of 92 with DDQ produced 7 in 50% yield. A selenide of 91 could also be prepared and eliminated to an aromatic product, but this route was not pursued.
While this route showed promise and did produce small amounts of demethoxy aflatoxin B$_2$, the thermal cyclization was unreliable and yields were quite low, so improvements were sought. Compound 83 reacted with LDA and MeSSO$_2$Me$^{51}$ to produce sulfide 93 in 84% yield. Oxidation of 93$^{52}$ produced sulfoxide 94. Attempts to react the lithium enolate of 94 with 89 produced what appeared to be an exo-eliminated
product. Michael addition using 93 was successful (78-84%). Conversion of 96 to 97 was accomplished in excellent yields (83-98%).

Formation of a phenyl selenide was unsuccessful. Oxidation of 97 produced bis-sulfoxide 98 plus starting material and a 16% yield of
demethoxyaflatoxin B₂. Heating the unpurified oxidation reaction mixture from 97 at 150 °C in toluene in a sealed tube for seven hours afforded 27% of 7 (from 97) after purification.

This improved route to demethoxyaflatoxin B₂ requires only six steps and proceeds in approximately 11% overall yield.
The total synthesis of racemic demethoxyaflatoxin B$_2$ is described. The key step in the synthesis is a silver-mediated oxidative radical cyclization to form the aflatoxin B$_2$ benzofuro[2,3-b]furan system. The conditions developed for this step differ from, and are complementary to, those reported for many similar methods in that they are mildly basic, rather than acidic. A sequence of Michael addition to an E-ring precursor by the ABC ring system, phenyl sulfenylation, oxidation, and cyclization of the D ring completed the synthesis in a concise manner.
EXPERIMENTAL

Unless otherwise noted, materials were obtained from commercial suppliers and were used without purification. Diethyl ether and tetrahydrofuran (THF) were distilled from sodium benzophenone ketyl. Benzene was distilled from lithium aluminum hydride. Methylene chloride and acetonitrile were purified by distillation from calcium hydride. Apparatus for experiments requiring anhydrous conditions was flame-dried under a stream of nitrogen or dried in a 150 °C oven for 12 h and cooled under a stream of nitrogen or in a desiccator. Flash chromatography was performed on EM Science Kieselgel 60 (mesh 230-400). Thin layer chromatography was performed using EM Science Kieselgel F254 prepared plates with a thickness of 0.25 mm. The solvent systems were suitable mixtures of hexanes (H) and ethyl acetate (EA) unless otherwise noted. The abbreviation sg represents silica gel. Infrared spectra were obtained on a Perkin-Elmer 1320 spectrophotometer and are reported in cm\(^{-1}\). Proton nuclear magnetic resonance spectra (300 MHz) were obtained using a Nicolet Magnetics Corporation NT-300 spectrometer. All chemical shifts are reported in \(\delta\) relative to tetramethylsilane as an internal standard. Splitting patterns are designated as s (singlet), d (doublet), t (triplet), q (quartet), dd (doublet of doublets), dt (doublet of triplets), ABq (AB quartet), and m (multiplet); a br prefix indicates a broadened pattern. Carbon-13 NMR spectra (75.47 MHz) were obtained on a Varian Associates VXR-300 spectrometer and are reported in \(\delta\) relative to CDCl\(_3\) (77.00 ppm). High
resolution mass spectra were obtained on a Kratos model MS-50 spectrometer. Low resolution mass spectra were obtained on a Finnegan 4023 mass spectrometer. Melting points were determined on a Fisher-Johns melting point apparatus and are uncorrected. Elemental analyses were performed by Galbraith Laboratories. The purity of all title compounds was determined to be >90% by 300 MHz proton NMR and/or elemental analysis.

**General Procedure for the Radical Cyclization:** A suspension of dicarbonyl compound (1 equiv), vinyl ether (10 equiv) and freshly prepared silver(I) carbonate (2 equiv) in dry MeCN (3 mL/mmol of dicarbonyl compound) was heated at reflux under nitrogen until TLC analysis indicated that no dicarbonyl compound remained. The mixture was cooled, filtered through Celite and concentrated in vacuo. The residue was purified by sg chromatography using H:EA.

**75:** $^1$H NMR (CDCl$_3$) $\delta$ 1.29 (t, $J = 6.9$ Hz, 3H), 2.00-2.11 (m, 2H), 2.23 (d, $J = 1.8$ Hz, 3H), 3.62-3.79 (m, 2H), 4.00-4.08 (m, 1H), 4.10-4.28 (m, 2H), 6.07 (d, $J = 6.3$ Hz, 1H); IR (NaCl, neat) 2980, 1690, 1640 cm$^{-1}$; HRMS: $m/e$ for C$_{10}$H$_{14}$O$_4$, calcd. 198.08921, measured 198.08866; CMR (CDCl$_3$) $\delta$ 13.72, 14.09, 31.31, 46.72, 59.13, 66.55, 103.13, 109.25, 164.92, 168.09; m.p.: 41.5 °C-45.5 °C; TLC (4:1 H:EA) R$_F$ = 0.35.

**83:** light yellow oil. $^1$H NMR (CDCl$_3$) $\delta$ 2.00-2.18 (m, 4H), 2.30-2.40 (t, $J = 6.6$ Hz, 2H), 2.40-2.55 (m, 2H), 3.59-3.68 (m, 1H), 3.68-3.80 (m, 1H), 4.09 (td, $J = 8.1$, 0.6 Hz, 1H), 6.24 (d, $J = 5.7$ Hz, 1H); IR (CDCl$_3$) 2980, 1765 (w), 1720 (w), 1630 (br) cm$^{-1}$; HRMS: $m/e$ for C$_{10}$H$_{12}$O$_3$

**Ethyl 2-(2,4-dioxopenty1)-furo[2,3-b]furan-3-carboxylate (81):** To a solution of LDA (1.2 mmol, prepared from diisopropylamine and n-BuLi at 0°C) in 0.5 mL of THF at -78 °C was added ester 75 (0.198 g, 1.00 mmol) in 1.5 mL of THF over 2 min. The solution was stirred at -78 °C for 30 min. Aldehyde 79 (0.200 g, 1.5 mmol) in 1.5 mL of THF was added dropwise and the solution was allowed to warm to 0 °C. The solution was cooled to -78°C, quenched with 0.13 mL of acetic acid and diluted with water. The aqueous layer was extracted with CH₂Cl₂. The combined organic layers were dried and concentrated in vacuo. The residue (RF (1:1 H:EA) = 0.25) was immediately oxidized.

To a suspension of PCC (0.289 g, 1.3 mmol) and Florisil (0.57 g) in CH₂Cl₂ (3 mL) was added the above alcohol (0.220 g, 0.67 mmol). After TLC showed that no alcohol remained, the suspension was poured into 40 mL of ether and filtered through Celite. The residue was dissolved in 1.1 mL of THF and 1.1 mL of 5% HCl and the solution was stirred for 22 h. After the usual workup, the residue was purified by sg chromatography using 2:1 H:EA to afford 0.18 g (100% yield) of 9. Compound 77 was prepared from ester 75 and acetaldehyde by the first two steps of this procedure.

**77:** ¹H NMR (CDCl₃) δ 1.28 (t, J = 7.1 Hz, 3H), 2.00-2.19 (m, 2H), 2.23 (s, 3H), 3.76-3.83 (m, 2H), 3.80 (dd, J = 17.0, 42.2 Hz, 2H), 4.02-4.12
(m, 1H), 4.12-4.25 (m, 2H), 6.45 (d, J = 6 Hz, 1H); IR (NaCl, neat) 2985, 2885, 1735, 1690, 1640 cm\(^{-1}\); TLC (1:1 H:EA) RF = 0.43.

81: \(^1\)H NMR (CDCl\(_3\)) \(\delta\) 1.28 (td, J = 7.1, 1.2 Hz, 3H), 2.04 (s, 3H), 2.05-2.17 (m, 2H), 2.25 (s, 1H), 3.65-3.85 (m, 4H), 4.02-4.12 (m, 1H), 4.12-4.25 (m, 2H), 5.56 (s, 1H), 6.15 (d, J = 6 Hz, 1H); IR (NaCl, neat) 2980, 2880, 1695, 1640, 1610 cm\(^{-1}\); HRMS: \textit{m/e} for C\(_{14}\)H\(_{18}\)O\(_6\) calcd. 282.11034, measured 282.11069; TLC (2:1 H:EA) RF = 0.35.

2,3,3a,4,5,6,7,8a-Octahydro-5-methylthio-[2,3-b]-benzofuran-4-one (93): To a stirred solution of LDA [4.27 mmol, prepared from distilled diisopropylamine (4.66 mmol, 0.65 mL) and n-BuLi (4.27 mmol)] in 2 mL dry THF at -78 °C under nitrogen was added dropwise ketone 83 (0.70 g, 3.88 mmol) in 7.8 mL dry THP. After 30 min at -78 °C, MeSSO\(_2\)Me (4.66 mmol, 0.48 mL) was added dropwise and the reaction mixture stirred while warming to to 0 °C. When the reaction appeared to be done by TLC, the mixture was recooled to -78 °C, quenched with 30 mL of a pH 7 buffer, the cooling bath was removed, and the solution pH adjusted to 6. The aqueous layer was extracted with CH\(_2\)Cl\(_2\) and the combined organics washed with brine, dried over Na\(_2\)SO\(_4\), and concentrated. The crude sulfide (1.01 g) was purified by flash chromatography on sg (1:4 H:EA) to yield the keto-sulfide 93 (0.74 g, 3.27 mmol) in 84% yield. Compound 93 was a light yellow oil: \(^1\)H NMR (CDCl\(_3\)) \(\delta\) 2.04-2.08 (m, 2H), 2.09-2.14 (m, 1H), 2.17 and 2.20 (s, 3H), 2.31-2.49 (m, 2H), 2.60-2.82 (m, 1H), 3.26 (t, J = 4.1 Hz, 1H), 3.60-3.78 (m, 2H), 6.23 and 6.27 (d, J = 5.7 Hz, 1H); IR (film) 2980, 1733, 1635, 1405 cm\(^{-1}\); TLC (1:4 H:EA) RF = 0.53.
2,3,3a,4,5,6,7,8a-Octahydro-5-methylthio-5-(3-oxo-2-(1-oxo-2-oxabutyl)-cyclopentyl)furo-[2,3-b]benzofuran-4-one (96): To a solution of LDA (0.484 mmol, prepared from n-BuLi (0.484 mmol) and diisopropylamine (0.52 mmol) in 0.2 mL of THF at -78 °C) was added a solution of ketone 93 (0.100 g, 0.44 mmol) in 1 mL of THF over 2 min. The solution was stirred at -78 °C for 30 min. Ketoester 89 (0.0746 g, 0.48 mmol) in 1 mL of THF was added dropwise and the solution was stirred at -78°C for 30 min. Acetic acid was added to quench the reaction; water was added and the aqueous layer was extracted with CH₂Cl₂. The combined organic layers were dried and concentrated. The residue was purified by chromatography using 1:1 H:EA to afford 0.14 g (84% yield) of 96. Compound 96 was a light yellow oil: ¹H NMR (CDCl₃) δ 1.25-1.32 (m, 3 H), 1.55-1.86 (m, 1 H), 1.90 and 1.98 (s, 3 H), 2.01-2.08 (m, 2 H), 2.10-2.14 (m, 1 H), 2.19 (d, J = 5.7 Hz, 1 H), 2.20-2.40 (m, 1 H), 2.41-2.48 (m, 2 H), 2.70-2.95 (m, 1 H), 3.15-3.29 (m, 1 H), 3.46-3.50 (m, 1 H), 3.55-3.80 (m, 3 H), 4.06-4.14 (m, 1 H), 4.14-4.25 (m, 2 H), 6.24 and 6.28 (br d, J = 6 Hz, 1 H); IR (film) 2980, 1750, 1720, 1635 cm⁻¹; TLC (1:4 H:EA) Rf = 0.55.

Demethoxy aflatoxin B₂ (7): To a stirred suspension of NaH (0.0098g, 0.41 mmol) (washed three times with hexanes and dried with N₂) in 2 mL of THF at 0 °C under N₂ was added the diketoester 96 in 1 mL of THF. After 30 min at 0 °C, PhSCl was added (0.0684 g, 0.47 mmol), the solution was stirred for 5 min, the ice bath was removed, and the mixture was stirred at room temperature. TLC after 80 min and 140 min showed starting material, and after stirring overnight
showed little change. The reaction mixture was added dropwise to 10 mL each of ether, pentane, and saturated aqueous sodium bicarbonate, plus ice. The aqueous layer was extracted with 10 mL of 1:1 ether: pentane. The combined organic layers were washed with brine, dried over Na₂SO₄, and concentrated. The material was purified by flash sg column to remove the PhSSPh (1:1 H:EA) to yield 0.13 g crude material.

The bis-sulfide (0.13 g) was taken up in acetonitrile, cooled in an ice/salt bath, and 0.5 M aqueous NaIO₄ (1.06 mmol, 2.13 mL) was added dropwise. After stirring at ice/salt temperature for 1 h, the mixture was stored in a refrigerator overnight. The flask was then removed from the refrigerator and the mixture was stirred at room temperature for 24 h, during which time a white precipitate formed. The mixture was filtered, extracted with CH₂Cl₂, and the combined organic layers was washed with brine, dried over Na₂SO₄, and concentrated to yield 0.10 g of the crude bis-sulfoxide.

The crude bis-sulfoxide was dissolved in 12 mL of dry toluene. The solution was degassed with argon for 10 min and then heated at 150 °C in a sealed tube for 7 h. The mixture was diluted with water. The aqueous layer was extracted with CH₂Cl₂, and the combined organic layers was dried over Na₂SO₄ and concentrated. Purification by flash chromatography on sg (1:4 H:EA) yielded 0.0207 g (18 % yield from 93) of the aflatoxin analog 7. Compound 7 was a beige solid: ¹H NMR (CDCl₃) δ 2.27-2.37 (m, 2H), 2.70-2.77 (m, 2H), 3.16-3.22 (m, 2H), 3.62 (q, J_AB = 9, 17Hz, 1H), 4.12-4.26 (m, 2H), 6.50 (d, J = 6 Hz, 1H), 6.87 (d, J = 9 Hz, 1H), 7.58 (d, J = 9 Hz, 1H); IR (CH₂Cl₂) 3045, 1765, 1697, 1610
cm\(^{-1}\); MS: \(m/e\) 284; HRMS: calcd. 284.06847, measured 284.06826; CMR (CDCl\(_3\)) \(\delta\) 24.50, 31.50, 34.90, 44.18, 67.80, 107.65, 112.38, 113.46, 114.82, 118.84, 127.35, 153.05, 155.11, 166.41, 176.54, 200.67; TLC (1:4 H:EA) RF = 0.16; mp (decomposed).
REFERENCES


PAPER II: SYNTHETIC APPROACHES TOWARD ANALOGS OF GLYCINOECLEPIN A
This manuscript will describe the syntheses of various analogs of glycinoeclepin A, the natural hatching stimulus for the soybean cyst nematode. While a total synthesis of the molecule was not achieved, analogs including modified C and D rings and modified A, B, and C rings have been synthesized and tested for hatch-stimulating activity.

For the sake of simplicity, many of the compounds described here have been shown without stereochemistry. Unless stereochemistry is explicitly detailed, these compounds should be considered to be mixtures of isomers.
Nematodes of various types have troubled agriculturalists for centuries. Because nematodes generally have a very narrow host plant range, crop rotations of host and non-host plants generally provide effective pest management. Chemical nematicides have also been used, but most effective ones have been deemed environmentally unsafe. Resistant plant varieties can also be used, but are often lower yielding than susceptible cultivars. Over time, nematodes can also adapt to resistant varieties.

In recent years, the soybean cyst nematode (SCN) has become an increasing problem for soybean growers in the United States. Infestation is particularly widespread in Missouri and other southern bean-growing states, but is spreading rapidly to the north. Many regions in the area of infestation, or in its path, depend heavily on soybeans, and the multiyear crop rotations necessary to reduce SCN populations to a non-damaging level are not economically feasible. With chemical control environmentally unsound and resistant plant cultivars often not satisfactory for various reasons, an effective, safe, economical means of SCN control is clearly needed.

SCN overwinters in eggs encased in protective cysts in the soil. The eggs can remain viable for years within the cysts, hatching when conditions become favorable. Among these conditions are pH, moisture, temperature, chemicals in the soil, root diffusates, and chemical
hatching factors. We believed that if conditions could be modified to stimulate SCN egg hatching at a time when no host plants were available, the juveniles would die, leaving fields safe for planting at the appropriate time. Most of the conditions required for hatching are beyond the farmer's control. However, the hatching stimulus for SCN, glycinoeclepin A, is known, and if it (or a sufficiently active analog) could be prepared in adequate quantities, we believe it could be used to trigger non-seasonal hatching of the encysted eggs. Glycinoeclepin A stimulates hatching at extremely low concentrations and is released during the normal growth cycle of soybeans. Thus, its use should be both economically and environmentally sound.

![Glycinoeclepin A](image)

Glycinoeclepin A (1) was first isolated\(^1\) in microgram quantities from 113 kg dried 'Hon-kintoke' kidney bean roots and was found to be active at \(10^{-11}\) to \(\sim 10^{-12}\) g/ml. Its structure was determined\(^2\) in 1985 using various spectral means and X-ray crystallography, all on the derived bis(p-bromophenacyl) ester.
As only 1.25 mg of the diester of glycinoeclepin A could be isolated from approximately a ton of bean root, it was obvious that an alternate source of glycinoeclepin A was needed. Since 1986, several researchers have reported syntheses of glycinoeclepin A or analogs of it. Analogs are an attractive option to glycinoeclepin A, for even if they are only partially as active as the highly active parent, they could still possess sufficient activity to be commercially useful.

Sakakibara applied for a Japanese patent in 1986 for several highly substituted multicyclic carbonyl compounds, which he intended as intermediates for glycinoeclepin A synthesis. In 1987, he followed this work with an approach to glycinoeclepin A's side chain which involved a Claisen rearrangement. Although the acid moiety was never placed on the side chain, the terminal double bond was seen as a ready "handle" for doing so. Also in 1987, Sakakibara published a synthesis of two active glycinoeclepin A analogs. Having noticed that four of the
The molecule's five oxygen-containing functional groups appeared to form a space appropriate for a "guest," structures containing the appropriate partial structures were constructed. A key step in the synthesis was
the halocyclization to form the precursor to the AB ring system. Compound 11a was accompanied by a lactone which could be converted back to 11a. Both 11a and 12 exhibited approximately 50% hatching stimulation at $10^{-5}$ g/ml.

Sasai et al. synthesized an analog of the CD ring system of glycinoeclepin A. The diene 14 underwent a Diels-Alder reaction

![diagram](attachment:image.png)

with dimethyl acetylene dicarboxylate to form a bicyclic adduct. Further transformations yielded the diacid 15, which showed hatching activity at $10^{-9}$ g/ml.

In 1988, Murai et al. published the first total synthesis of glycinoeclepin A. A convergent route was followed, in which the AB ring system was constructed and joined to the C ring, followed by a final cyclization to yield the D ring. Functionalization and opening of a 6-membered ring was used to set the stereochemistry of the D-ring side chain during construction of the C ring.

A Baker's yeast reduction of 2,2-dimethylcyclohexane-1,3-dione yielded an optically active keto alcohol. Formation of the exo methylene group allowed for halocyclization to form the AB ring system, as in
Sakakibara’s work.

Construction of the C and D rings began with R-(-)-carvone. The molecule was manipulated to produce a functionalized bicyclic system, one ring of which was oxidized to a lactone and then opened with base to reveal a handle for the formation of the D ring, with its side chain already in place.
The AB and C ring moieties were first joined with a lactone linkage. The desired C-C bond between the AB and C rings was established using KF/18-crown-6. Opening of the lactone ring and functional group manipulation set the stage for cyclization to form the D ring. Further functional group manipulation led to glycinoeclepin A.
Mori and Watanabe\textsuperscript{8} reported a total synthesis of glycinoeclepin A in 1989. They employed many strategies similar to those of previous workers, but used a unique method of forming the C ring. The synthesis began with a Baker's yeast reduction of 2,2-dimethyl-1,3-cyclohexanedione, followed by functionalizations and a halocyclization to form the AB ring system with a "handle." To form the D ring and its
side chain, the bicyclic dione 27 was prepared from 3-methyl-2-cyclopentenone and reduced with Baker's yeast. Functionalization and ring expansion yielded compound 28. Compound 28 was condensed with 26 in the presence of methyl lithium and zinc chloride.

Functionalization of the tetracyclic system led to compound 30, which underwent reductive cyclization upon treatment with lithium dimethylcuprate. Simultaneous opening of the lactone ring freed the D-ring side chain. Final functional group manipulation led to
glycinoeclepin A. A second total synthesis of glycinoeclepin A published by Mori and Watanabe\(^9\) in 1991 was nearly identical, but used much more efficient means of final ester cleavage and hydroxyl deprotection than the previous work.

Fukuzawa \textit{et al.}\(^{10}\) used biomimetic methods to prepare glycinoeclepin A analog 32.

9,10-Seco-cycloartanes were prepared by Sakamaki \textit{et al.} as intermediates for glycinoeclepins.\(^{11}\)

\(X = \text{lower alkyl, alkylidene, haloalkyl, hydroxyalkyl, CO}_2\text{H, alkoxy carbonyl}
\)

\(R = \text{H, pyranyl, methoxymethyl, ethoxyethyl}\)
In 1990 Corey and Houplis\textsuperscript{12} reported a total synthesis of glycinoeclepin A. This synthesis was quite different than those previously reported in that it began at the D ring and built the other rings on in turn, and it used intramolecular oxymercuration rather than halocyclization to form the AB ring system. Reaction of the potassium enolate of cyclopentanone 35 with ester 36 (prepared from the acid and (-)\textit{8}-phenylmenthol) yielded a product with 95:5 enantioselectivity and 5:1 diastereoselectivity for C-17 to C-20. The corresponding methyl ester gave a racemic product. Further transformations of 37 led to diene 38, which underwent a Diels-Alder reaction with 3-(p-toluenesulfonyl)propionic acid and further functionalization to afford compound 39. Coupling of 39 with triflate 40 (prepared
enantioselectively from 2,2-dimethylcyclohexane-1,3-dione via Baker's yeast reduction and appropriate functionalization) yielded compound

\[
\begin{align*}
39 \quad + \quad 40 \quad \rightarrow \quad 41
\end{align*}
\]

Once the ketone in 41 was reduced and chloroacetylated and the silyl protecting group was removed, intramolecular oxymercuration/demercuration formed the ether linkage of the AB ring system.

\[
\begin{align*}
41 \quad \text{\rightarrow} \quad 42
\end{align*}
\]

Compound 42 underwent rearrangement with FeCl₃ in acetic anhydride to give compound 43, which was converted to glycinoeclepin A in five steps. The p-bromophenacyl ester was identical to that of an authentic sample by HPLC, MS, IR, 500 MHz proton NMR, and optical rotation. This total synthesis is the most simple and brief of those reported to date.
42 \[1.1 \text{ equiv FeCl}_3 \]
\[10:1 \text{ Ac}_2\text{O} \]
\[\text{CH}_2\text{Cl}_2 \]

\[\xrightarrow{\text{1}} \]

43

1
RESULTS AND DISCUSSION

Upon brief inspection, glycinoeclepin A appears to be a relatively straightforward synthetic target, its four contiguous stereogenic centers providing the greatest challenge. Synthesis of the two bicyclic units and connection of them seemed a reasonable strategy. Unfortunately, synthesis of the AB system in particular proved much more difficult than anticipated. In light of our primary goal of synthesizing field-useable compounds with SCN hatching activity, simple analogs appeared more attractive than glycinoeclepin A itself. In order to determine which functionalities were necessary for biological activity, many analogs were prepared and tested. Variations included the presence or absence of various functional groups, the number and size of rings, and the length of the side chain. We postulated initially that the acid moieties were important, the hydroxyl group less important or unimportant, and the methyl groups unimportant. Based on these ideas, syntheses were begun.
Compound 44\textsuperscript{13} looked like a good precursor to an analog of glycineclepin A's CD ring system. Although it was a hexahydro-naphthalene, rather than a hexahydroindane, it had appropriate double bonds, a carboxyl functionality in the correct C-ring position, a hydroxyl for functionalization toward the AB system, and a ketal which could be opened to approximate the side chain. Unfortunately, the literature reports were not reproducible, and 44 itself was not synthesized.

\[
\begin{align*}
\text{HO} & \\
\text{COOEt} \\
44
\end{align*}
\]

2-Methyl-1,3-cyclohexanedione reacted with ethyl acrylate (methyl acrylate was also used successfully later) in the presence of triethylamine in boiling ethyl acetate to produce the ester 45\textsuperscript{14} in approximately 75% purified yield. The next step was to be addition of lithioacetylenylethyl ether to the dione with subsequent dehydration

\[
\begin{align*}
\text{O} & \\
\text{O} \\
\text{+} & \\
\text{Et}_3\text{N} \\
\text{\rightarrow} & \\
\text{O} & \\
\text{OEt} & \\
45
\end{align*}
\]
and ester formation using sulfuric acid in ethanol.\textsuperscript{12} Unfortunately, the anion addition did not work well, and only traces of the desired product were ever formed. The corresponding addition to benzaldehyde was

\[ \text{Li-OEt} \quad 45 \xrightarrow{\text{Et}_2\text{O}, -78^\circ \text{C}} \quad \text{46} \]

successful, so the failure to produce 47 was attributed to peculiarities of the molecule and a new method was selected. The Horner-Emmons\textsuperscript{15} modification of the Wittig reaction seemed a logical choice. Treatment of triethylphosphonoacetate with sodium hydride and 45 gave a 66-

\[ 45 + (\text{EtO})_2\text{P(0)}\text{CH}_2\text{CO}_2\text{Et} + \text{NaH} \xrightarrow{\text{THF}} \quad 48 \]

70\% yield of 48 after flash column chromatography on silica gel. Before attempting the intramolecular cyclization of the diester, transformation of the ketone moiety was undertaken. Reaction of 48 with ethylene
glycol and p-toluenesulfonic acid in boiling benzene\textsuperscript{13} gave acceptable yields of ketal 49.

Initial attempts to cyclize 49 were not as successful as those reported in the literature.\textsuperscript{13,16} Direct trapping of the enolate failed.

to give satisfactory results. If no trapping agent was added, the enol could be isolated,\textsuperscript{13} but it rearranged within 24 h to the enone. The methoxymethyl protected enol could be made from the freshly prepared enol and chloromethylmethyl ether with diisopropylethylamine in moderate to good yields.
In order to free the acid moiety and produce an analog suitable for testing, attempts were made to hydrolyze the ester. Treatment of 50 with lithium hydroxide in THF/methanol/water simply returned starting material. BuSLi in HMPA\textsuperscript{17} also failed to cleave the ester. These difficulties, coupled with the discovery that 50 decomposed slowly upon storage, led to alterations in the target molecule.
As 49 was stable and readily accessible, it was used as a convenient starting point. In order to approximate the D-ring side chain, the acetal of 49 was opened using triethylsilane and boron trifluoride etherate\textsuperscript{18} to afford the alcohol in 61-80\% yield. Triethylsilane with trifluoroacetic acid failed to open the acetal. Cyclization of the diester with \textit{in situ} trapping of the enolate was next attempted. Treatment of 53 with potassium tert-butoxide in benzene
at room temperature, followed by addition of chlorotrimethylsilane, led to an unsatisfactory mixture of products. Cyclization of 53 with potassium tert-butoxide, followed by quenching of the enolate with H₂O, gave a promising result by proton NMR, albeit in relatively low yield.

Unfortunately, an attempt to form the methoxymethyl protected enol resulted in a large number of product spots by TLC and low mass return.

Concerned that the free alcohol group in 53 might be one of the
problems in the above work, it was protected as a tert-butyl-
dimethylsilyl ether. Cyclization of 55 with potassium tert-butoxide
showed promise, but an attempt to protect the enol as a methoxy-
methyl ether did not proceed cleanly. Cyclization with \textit{in situ} trapping
of the enolate with chloromethylmethyl ether failed. Cyclization and \textit{in situ} trapping using THF as the solvent at room temperature gave a very
similar TLC profile, and the product proved difficult to purify by

\[
55 + \text{t-BuOK} \xrightarrow{\text{R.T.}} 56
\]

chromatography. Similar attempts to cyclize 55 (THF, 0 °C) and trap the
enolate with benzoyl chloride or methyl iodide proved unsatisfactory.

When 55 was treated with potassium tert-butoxide in THF at 0 °C,
the cyclized enolate could be trapped with chlorotrimethylsilane to
yield enol silyl ether 57 in 75-85% yield. Treatment of 57 with
potassium fluoride and benzyl bromide failed to produce the desired
benzyl compound 58.
Based on the initial positive result, other trapping experiments were examined. Treatment of 55 with potassium tert-butoxide followed by methanesulfonyl chloride gave a small amount of the

55 + t-BuOK + MeSO₂Cl \xrightarrow{\text{THF, 0°C}} 59

(minor)
desired mesylate, but the reaction mixture was a multitude of spots by TLC analysis. An attempt to generate the mesylate and allow it to react \textit{in situ} with tetrahydrofurfuryl alcohol also gave an unclear result. A later attempt to trap the enolate with benzyl bromide also gave several products.

Treatment of \(55\) with potassium tert-butoxide followed by careful aqueous workup gave enol \(56\) which converted to enone \(61\) with time or with silica gel chromatography. Treatment of \(61\) with benzyl alcohol and p-toluenesulfonic acid failed to give the desired unsaturated benzyl
ether, which would have very roughly approximated the AB system of glycinoeclepin A. Unfortunately, 61 proved unstable, decomposing within days at 0 °C. Therefore, this route was dismissed as a feasible path to analogs.

At this point, interest in compound 53 was renewed. A second attempt was made to cyclize 53 with potassium tert-butoxide using THF at 0 °C and \textit{in situ} trapping of the enolate with chlorotrimethylsilane. Treatment of the presumed bicyclic enol silyl ether with potassium fluoride and benzyl bromide in dimethylformamide yielded what

\[
\begin{array}{c}
\text{OH} \\
\text{O} \\
\text{Et} \\
\text{t-BuOK} \\
\text{KF} \\
\text{(COCl)}_2 \\
\text{BnBr} \\
\text{DMSO} \\
\text{THF, 0°C} \\
\text{DMSO} \\
\end{array}
\]

appeared to be a benzyl ester/benzylvinyl ether. While Swern oxidation of this product did produce an aldehyde, this path was abandoned as more promising paths arose.

Treatment of alcohol 53 with Jones' reagent\textsuperscript{20} produced an acid, but in low yields. A two step oxidation of this alcohol involving a Swern oxidation\textsuperscript{21} to the aldehyde followed by a silver oxide oxidation\textsuperscript{22} to the
acid proved much more effective, with yields up to 86%. Initially, the product of both oxidations was believed to be the monoacid 62. Integration of the ester region in the proton NMR spectrum did not support this structure. Treatment of the oxidation product with diazomethane\textsuperscript{23} yielded a product with two new methyl ester peaks. The proton NMR and infrared spectrum supported triester 63.

In an earlier attempt to cyclize 55 with sodium methoxide in methanol, transesterification resulted. This transesterification was also successful on unprotected alcohol/diester 53. In an effort to simplify
spectra, bis-methyl ester 64 was prepared in > 95% yield. Compound 64 underwent Swern oxidation in quantitative yield, but upon further oxidation with silver oxide in aqueous ethanol product 66 resulted and resisted a third transesterification attempt. Based on these results, oxidation of aldehyde 67 (from Swern oxidation of 64) was performed using aqueous methanol and diacid 68 resulted. It was ultimately discovered that both the bis-methyl and bis-ethyl ester gave the same
oxidation product when methanol was used as the solvent. Thus, prior transesterification was unnecessary.

As compound 68 contained approximations of glycinoeclepin A's D-ring side chain, B-ring "acid" (masked as an ester), and further oxygen-containing functionality, it was submitted as an analog for biological testing. Results to date are inconclusive. Lithium hydroxide hydrolysis to the triacid 69 afforded a second analog for testing. Compound 69 has not yet been tested.
Attempts were then made to produce closer glycinoeclepin A analogs from 68. Treatment of 68 with diazomethane in dichloromethane yielded triester 70. When subjected to potassium tert-butoxide and chlorotrimethylsilane in THF at 0 °C, compound 70 failed to produce the desired bicyclic enol silyl ether 71.

It was hoped that treatment of 68 with strong base would produce an anion which quench α-to the ester, allowing the introduction of an A ring approximation. 2-Furaldehyde was chosen as the electrophile because of its ring size and ring oxygen. It was hoped that the alcohol produced could be eliminated to form the unsaturated furan. Unfortunately, treatment of 68 with various bases failed to yield the addition product 73. Conditions used included three, four, or five equivalents of lithium diisopropylamide, three equivalents of potassium hydride, and five equivalents of potassium tert-butoxide in THF, DMF, or DMSO.
Bis-Wittig products from a precursor of 68 were also seen as potential analogs, but their synthesis proved elusive. Treatment of 45 with two equivalents of the Wittig reagent 75 returned starting material, while treatment with two equivalents of sodium hydride and two equivalents of triethylphosphonoacetate also failed to yield the desired product. Resubmission of the mono-Wittig product did not produce the bis-product.
Pursuit of analogs along this line was halted at this point and new pathways explored. The method of Mukaiyama\textsuperscript{24} for coupling enol silyl ethers and $\alpha,\beta$-unsaturated carbonyl compounds seemed a likely starting point.

It was hoped that 77 or 78 would couple with an appropriate enol ether under Mukaiyama's conditions and that the resulting adduct could be cyclized to give a CD ring system analog with a masked acid moiety and a handle upon which to build the D-ring side chain.

Treatment of ethyl or benzyl acetate with LDA followed by addition of acrolein produced the unsaturated alcohols 75 and 76 in greater than 71\% yield. Jones oxidation\textsuperscript{20} afforded the dicarbonyl compounds. The coupling was attempted first on a model system, and gave promising
results by proton NMR. Based on this result, a cyclic ketone for the desired analog was prepared.25

Treatment of 2-methyl-2-cyclopenten-1-one with titanium(IV) chloride and allyltrimethylsilane afforded moderately good yields of 83 after purification. Formation of the enol silyl ether using HMDS, Et₃N, and TMSI at 0 °C invariably led to a product mixture containing some of the kinetic product, as well as the desired thermodynamic product. It
was later discovered that this problem could be overcome by starting the reaction at a lower temperature,\textsuperscript{26}

Reaction of a mixture of 84 and 85 with 77 in the presence of titanium (IV) chloride afforded an initially promising crude mixture.

Silica gel chromatography failed to yield the desired product definitively and attempted cyclization of the tentatively identified product gave unclear results. Allowing the coupling reaction to warm to
room temperature did not appear to improve the results. Use of the ethyl ester, rather than the benzyl ester, also failed to produce the desired product cleanly.

Attempts to couple 84 with acrolein in the presence of titanium(IV) chloride failed to produce appreciable amounts of the coupled aldehyde. Methylvinyl ketone (MVK) also failed to produce...
satisfactory results. A combination of titanium(IV) chloride and titanium(IV) isopropoxide served to couple 84 and MVK, albeit in low yield. Extension to the unsaturated β-ketoester was not promising.

The dicarbonyl compound 90 could be cyclized to the desired bicyclic compound 91 using potassium hydroxide in boiling ethanol.

However, it was discovered that 91 could be produced much more efficiently (approximately 50% yield) by a two step process in which 84 reacted with MVK in the presence of catalytic potassium hydroxide.
in ether. When the unpurified reaction mixture was heated with excess potassium hydroxide, 91 was formed.

As 91 contained the bicyclic system of glycineclepin A, a precursor for its side chain, and an enone capable of undergoing further functionalization, it was used as a precursor for several analogs. Treatment of 91 with sodium hydride, followed by bubbling carbon dioxide gas through the reaction mixture,\textsuperscript{29} afforded carboxylic acid 92 in 30% yield. Addition of a catalytic amount of potassium hydride increased the yield to 43%. A sample of 92 was submitted for biological testing and was found to be inactive.

Further functionalization of the side chain was the next goal. Toward this end, a selective ozonolysis of 92 using Sudan Red 7B as an indicator\textsuperscript{30} was performed, and an aldehyde resulted. The aldehyde could be further oxidized using either Jones' reagent or silver oxide in aqueous ethanol, with the latter affording greater yields. Unfortunately, the ozonolysis proved somewhat unreliable, failing in some cases to go
to completion. In other cases, the ozonolysis intermediate failed to decompose completely to the aldehyde. Model systems, including carvone and heptene, were employed to test reaction conditions. Solvents used were methanol, dichloromethane, and 2:1 dichloromethane/ethanol. Sudan Red 7B, Solvent Red 23, and Sudan III were used in different cases, and dimethylsulfide (or triphenylphosphine) was used to decompose the intermediates. No suitable conditions were found, so the route was abandoned.

As functionalization of the side chain in 91 was not readily achieved by ozonolysis, it was hoped that it could be performed before cyclization. Ozonolysis of the unpurified product mixture from the reaction of 83, MVK, and potassium hydroxide in aqueous ethanol was only marginally successful. Oxidation with silver oxide produced an acid, as evidenced by the infrared spectrum. Cyclization of the unpurified mixture with potassium hydroxide in boiling ethanol was tentatively promising by proton NMR, but the ester produced by reaction with diazomethane did not appear promising by proton NMR.
Use of the unpurified reaction mixture 95 made determination of results difficult, so further attempts at oxidation were made on 83. Treatment of 83 with potassium permanganate, acetic acid, and Adogen 464\textsuperscript{32} afforded acid 97 in 71-78% yield. Reaction of 97 with MVK and

\[ \text{83} + \text{Kmno}_4 + \text{CH}_3\text{CO}_2\text{H} + \text{Adogen 464} \xrightarrow{\text{H}_2\text{O}} \text{97} \]
potassium hydroxide in EtOH/Et₂O and heating the unpurified reaction mixture with potassium hydroxide in ethanol appeared to yield the enone 98; however, after reaction of 98 with diazomethane the results were unclear.

Treatment of 83 with ruthenium(III) chloride trihydrate and sodium periodate³³ afforded 97, accompanied by a trace of the corresponding aldehyde.

\[
83 + \text{RuCl}_3 \cdot 3\text{H}_2\text{O} + \text{NaIO}_4 \xrightarrow{\text{CCl}_4/\text{CH}_2\text{CN}/\text{H}_2\text{O}} 97 + \text{aldehyde}
\]

It was hoped that a tert-butyl ester of 97 would cyclize more readily than 97, and be readily cleaved to the desired acid once cyclization was achieved. Toward that end, 97 was treated with isobutylene and sulfuric acid in ether. Unfortunately, the reaction primarily returned starting material.

\[
97 + \xrightarrow{\text{Et}_2\text{O}} + \text{H}_2\text{SO}_4 \xrightarrow{\text{S. M.}}
\]
An attempt was also made to react 2-methylcyclopent-2-ene-1-one with the tert-butyldimethylsilyl enol ether of ethyl acetate in the presence of mercury(II) iodide and triethylamine.\textsuperscript{34} While the starting material was consumed, the product was not clean and defied purification by chromatography. An attempt to couple 100 with MVK using titanium(IV) chloride and titanium(IV) isopropoxide\textsuperscript{27} gave an unclear result. Attempted cyclization of the unpurified reaction mixture did not afford the desired bicyclic compound.

At this point, a return to the bicyclic intermediate seemed prudent. Treatment of 92 with ruthenium(III) chloride and sodium periodate afforded a mixture of starting material, aldehyde, and acid, as determined by infrared spectra and proton NMR.
A sidechain epoxide appeared to be a desirable intermediate. With the hope that epoxidation would occur preferentially at the more electron rich double bond, 91 was treated with m-chloroperoxybenzoic acid to yield epoxide 103 in 28-30%. Addition of butylated hydroxytoluene (BHT, 104) to the reaction mixture as a radical inhibitor and raising the temperature increased yields to 38%. Use of the sulfide inhibitor 105 and higher temperatures increased yields to 42%.
An attempt to epoxidize acid 92 was not satisfactory.

Various methods were employed in an effort to open the epoxide ring. Treatment of 103 with boron trifluoride etherate did not give a clear result. Aluminum trichloride in dioxane did effect opening to

\[
\text{103} + \text{BF}_3\cdot\text{Et}_2\text{O} \xrightarrow{\text{Et}_2\text{O}} \xrightarrow{\text{R.T.}} \text{106}
\]

\[ \text{103} + \text{AlCl}_3 \\
\text{or Et}_2\text{AlCl} \\
\xrightarrow{1. \text{dioxane}} \\
\xrightarrow{2. \text{HCl}} \text{106 (minor)} \\
\xrightarrow{\text{R.T. or ~12 °C}} \\
\]

\[ \text{103} + \text{Et}_2\text{AlCl} \\
\xrightarrow{1. \text{CH}_2\text{Cl}_2} \\
\xrightarrow{2. \text{NaHCO}_3} \\
\xrightarrow{-78 \text{ °C} \rightarrow 0 \text{ °C}} \\
\xrightarrow{0 \text{ °C} \rightarrow \text{R.T.}} \text{106} \\
\]

\[ \text{103} + \text{AlCl}_3 \\
\xrightarrow{1. \text{CH}_2\text{Cl}_2} \\
\xrightarrow{2. \text{HCl}} \text{106 (trace)} \\
\]

the aldehyde, as did diethylaluminum chloride in dioxane. The latter afforded better yields. Diethylaluminum chloride in dichloromethane with an aqueous sodium bicarbonate quench failed to produce the aldehyde at temperatures from -78 °C to room temperature. Aluminum
trichloride in dichloromethane with an acidic aqueous workup yielded only a trace of the desired aldehyde.

Epoxide 103 was next treated with sodium hydride, catalytic potassium hydride, and carbon dioxide in the hopes that the epoxide would open, either during the reaction or during the basic workup.

While the epoxide did not open to form 107, acid 108 was produced in 66% yield. Treatment of 108 with periodic acid\(^{39}\) afforded an aldehyde.
The product of silver oxide oxidation of 109 was difficult to identify by proton NMR, but treatment of the unpurified reaction mixture with diazomethane showed it to be several products with none dominant.

Treatment of 103 with periodic acid\textsuperscript{39} afforded aldehyde 102. An attempt to purify 102 by chromatography failed and the material began to decompose when concentrated in vacuo. Treatment of 102 with silver oxide initially gave unclear results, but acid 98 was produced when the reaction was repeated on a larger scale. A sample of 98 was sent for biological testing to determine whether its short side chain had an effect on biological activity. The test results were inconclusive.

Further modifications of the side chain were sought. Attempts to add carbon tetrabromide to olefin 91 using light or benzoyl peroxide and heat\textsuperscript{40} failed. Treatment of 91 with ethyl diazoacetate\textsuperscript{41}
produced only small amounts of a possible ester along with returned starting material and several other products. Compound 91 reacted

$$91 + N_2CHCO_2Et + \text{Rh(II) acetate} \rightarrow 113$$

with N-bromosuccinimide and water\textsuperscript{42} to produce bromohydrin 114, which in turn produced epoxide 103 in 40% yield over the two steps.
An attempt to use this method to produce the hydroxy nitrile failed, affording instead the bromohydrin and the epoxide.

Osmylation\textsuperscript{43} of 91 afforded diol 116 in 76-85\% yield. Oxidation of the diol with the Jones’ reagent produced the $\alpha$-ketoacid 117.

Selective hydroborations were next examined. Treatment with dicyclohexylborane (prepared \textit{in situ})\textsuperscript{44} afforded the desired alcohol 118 in 63-79\% yield. 9-Borabicyclo[3.3.1]nonane (9-BBN)\textsuperscript{45} failed to produce 118 from 91.
An attempt to produce $\alpha,\beta$-unsaturated acid 119 from 118 failed, as did an attempt to produce bromide 120.
Treatment of 118 or its precursor 91 with methyl methoxy-magnesium carbonate did not yield the desired acid 121.

\[
\begin{align*}
\text{91: } R &= \text{C} = \text{C} \\
\text{118: } R &= \text{CH}_2\text{CH}_2\text{OH} \\
\end{align*}
\]

Oxidation of 118 with Jones' reagent produced acid 122 in excellent yield. A sample of 122 was sent for biological testing, with inconclusive results. An attempt to produce 122 from the olefin via a boronic acid was not successful.
As with 118, the α,β-unsaturated acid was not formed when 122 was treated with base and CO₂. An attempt to reduce the enone to the alcohol with diisobutylaluminum hydride (DIBAL-H)⁴⁹ also failed.
To help confirm the identity of 122, its methyl ester 125 was prepared using diazomethane. Due to its superior solubility
properties and greater ease of handling compared to the acid, it was seen as a valuable intermediate for further analog synthesis. The ester could be hydrolyzed after the desired compounds were made.

Enol ethers of 125 were seen as potential analogs. Toward that end both 122 and 125 were treated with cerium trichloride heptahydrate and sodium borohydrate in methanol. While

\[ 125 + \text{CeCl}_3 \cdot 7\text{H}_2\text{O} + \text{NaBH}_4 \xrightarrow{\text{MeOH}} 126 \]

the acid produced an unsatisfactory product mixture, the ester afforded good yields of the alcohol after purification.

Using an oxygen, rather than a methylene, to link the analog AB and C rings did not seem significant, and a tetrahydropyranyl ether seemed a good starting approximation of the glycinoeclepin A A ring.

\[ 126 + \text{THPO} + \text{PPTS} \xrightarrow{\text{CH}_2\text{Cl}_2, \text{R.T.}} 127 \]
Treatment of 126 with dihydropyran and pyridinium-p-toluenesulfonate (PPTS)\(^1\) unfortunately did not produce the desired ether 127.

Hoping to introduce further oxygen functionality for an analog, 126 was treated with diisopropylethylamine and chloromethyl methyl ether to afford ether 128 in good yields. The methoxy ethyl ether

\[
\text{126} + \text{i-Pr}_2\text{NEt} + \text{ROCH}_2\text{Cl} \xrightarrow{\text{CH}_2\text{Cl}_2, 0^\circ\text{C} \rightarrow \text{R.T.}} \text{128} \quad \text{R} = \text{CH}_3 \\
\text{129} \quad \text{R} = \text{CH}_2\text{CH}_2\text{OCH}_3
\]

was produced similarly in excellent yield, and contained an additional oxygen to help create a "host" area for an analog.

Hydrolysis of 129 with lithium hydroxide afforded acid 130 on a small scale.
Treatment of 126 with sodium hydroxide and chloroacetic acid\textsuperscript{52} in water was initially promising, with possible hydrolysis of the ester, but this avenue was not further pursued, as more promising routes were found.

\[
126 + \text{ClCH}_2\text{CO}_2\text{H} + \text{NaOH} \rightarrow \text{H}_2\text{O} \rightarrow 131
\]

\[
\text{R} = \text{H, Me}
\]

At this point, attention was once again focused on the enone. Treatment of 125 with ethylene glycol and PTSA produced ketal 132 in 47\% yield. While reaction of 132 with triethylsilane and boron trifluoride etherate failed to open the ketal, hydrolysis of the ester with
lithium hydroxide afforded acid 134, which was sent for biological testing. The test results were inconclusive.

\[
132 + \text{Et}_3\text{SiH} + \text{BF}_3 \cdot \text{Et}_3\text{O} \xrightarrow{\text{CH}_2\text{Cl}_2 \atop 0^\circ \text{C} \rightarrow \text{R.T.}} \quad \text{133}
\]

\[
132 + \text{LiOH} \cdot \text{H}_2\text{O} \xrightarrow{\text{THF/MeOH/H}_2\text{O} \atop 3.6 : 3.6 : 1} \quad \text{134}
\]

Lithio tert-butylacetate reacted with 125 to produce alcohol 135, which underwent elimination to yield unsaturated ester 136 plus isomers with acid catalysis. Unfortunately, cleavage of the tert-butyl ester proved difficult. Both trifluoroacetic acid\textsuperscript{53} and iodotrimethylsilane\textsuperscript{54} failed to produce the desired acid 137 cleanly.
AcO-Bu-t + LDA + 125 \rightarrow \text{THF} \rightarrow -78^\circ C \rightarrow \text{t-BuO}\text{OMe} \rightarrow 135

135 \xrightarrow{\text{PTSA}} \text{THF} \rightarrow \text{R.T.} \rightarrow \text{t-BuO}\text{OMe} \rightarrow 136 + \text{isomers}

136 + \text{CF}_3\text{CO}_2\text{H} \xrightarrow{\text{CH}_2\text{Cl}_2} \text{R.T.} \rightarrow \text{HO}\text{OMe} \rightarrow 137

136 + \text{TMSI} \xrightarrow{\text{CCl}_4} \text{R.T.} \rightarrow 137
Lithio ethylacetate also reacted with 125 to produce an alcohol, which afforded the unsaturated ester and its isomers with PPTS or PTSA or

\[
\text{OEt} + \text{LDA} + 125 \rightarrow \begin{array}{c}
\text{HO} \\
\text{EtO} \\
\text{Me}
\end{array}
\]

\[\text{THF} \quad -78^\circ C\]

\[138\]

\[
138 \xrightarrow{\text{PPTS or PTSA} \ \text{THF} \ \text{R.T.}} \begin{array}{c}
\text{HO} \\
\text{EtO} \\
\text{Me}
\end{array}
\]

\[+ \text{isomers}\]

\[139\]

PPTS. The initial attempt to hydrolyze 139 with lithium hydroxide succeeded only in cleaving the ethyl ester. At 50 °C, the desired diacid was produced.
The hydroxy diacid was desired as an analog. Treatment of 138 with lithium hydroxide over several days served to cleave the esters, but some elimination occurred during the acidic workup. In order to avoid this problem, the reaction was repeated, the reaction mixture
neutralized, and the solvents removed. The salt 144 was sent for biological testing, with inconclusive results.

\[ \text{138} \xrightarrow{\text{LiOH}} \text{144} \]

A Horner-Emmons reaction between 125 and triethylphosphonoacetate led to 139, but in lower yield than the route described above. Trimethylphosphonoacetate showed little promise.

\[ \text{125} + (\text{EtO})_2\text{POCH}_2\text{CO}_2\text{Et} + \text{NaH} \xrightarrow{\text{THF}} \text{139} \]

An effort was then made to produce analogs with better representations of the AB ring system. Dilithio-N-methylbenzamide was known to react with some carbonyl compounds. Phthalides could be produced from these initial adducts.\(^{55}\) Armed with this knowledge,
a model system was studied. Reaction of cyclohexenone with dilithio-N-methylbenzamide (prepared \textit{in situ}) followed by treatment of the unpurified adduct with potassium hydroxide in boiling aqueous ethanol produced phthalide 146 in moderate yield.

With the phthalide in hand, numerous attempts were made to open it to the corresponding acid. Treatment of 146 with PTSA, boron trifluoride etherate, trifluoroacetic acid, lithium hydroxide, or activated zinc in aqueous formic acid\textsuperscript{56} returned starting material. Treatment with iodontrimethylsilane\textsuperscript{54} was not promising. Reaction with activated zinc, catalytic copper(II) sulfate, and pyridine in boiling aqueous potassium hydroxide\textsuperscript{56} did produce an acid of undetermined purity.
Treatment of the analog precursor 125 with dilitio-N-methylbenzamide as in the model system was tentatively promising, although the methyl ester appeared to be cleaved during the KOH step. Re-

\[ \text{125} \quad \text{145} \quad \text{148} \]

formation of the ester with diazomethane allowed for purification. This delivered what appeared to be the desired phthalide, but in low yield. At this point, this route was abandoned to seek more promising ones.

Treatment of N-methylbenzamide with two equivalents of n-butyllithium and 2-furaldehyde afforded alcohol 150. It was hoped that 150 could be cyclized to the phthalide and later opened to yield
the desired acid 152. Unfortunately, reaction of 150 with potassium hydroxide in boiling aqueous ethanol failed to yield the phthalide 151.

\[
\begin{align*}
\text{150} & \quad \xrightarrow{\text{1. THF/Et}_2\text{O}} \quad \xrightarrow{\text{hexanes}} \quad 150 \\
\end{align*}
\]

An attempt was then made to remove the hydroxyl functionality to produce an amide which could be converted to an acid. Treatment of

\[
\begin{align*}
\text{150} & \quad \xrightarrow{\text{EtOH/H}_2\text{O}} \quad \text{reflux} \quad 151 \\
\end{align*}
\]

\[
\begin{align*}
\text{150} & \quad \xrightarrow{\text{NaBH}_4 + BF_3\cdot Et_2O} \quad \text{glyme or diglyme} \quad \text{0°C} \quad 153 \\
\end{align*}
\]
150 with sodium borohydride and boron trifluoride etherate in glyme or diglyme\textsuperscript{57} failed to completely remove the hydroxyl functionality. Flash silica gel chromatography yielded a small amount of the desired product. A different route to 152 was then sought.

Treatment of methyl-o-toluate (prepared from o-toluic acid and methanol) with bromine and sunlamp irradiation\textsuperscript{58} afforded bromide 154 in good yield. Although some starting material remained, the product was pure enough to carry on. Treatment of 154 with difuryl-cuprate\textsuperscript{58} yielded furan 155 in 28-49\% yield after chromatography. Compound 155 underwent a Diels-Alder\textsuperscript{59} reaction with maleic

\[
\text{O} \quad \text{O} \\
\text{Br} \\
\text{Me} \\
\text{OMe} \\
\text{Et}_2\text{O} \\
\text{THF}
\]

\[
\text{O} \quad \text{O} \\
\text{Me} \\
\text{OMe} \\
\text{Et}_2\text{O} \\
\text{THF}
\]
anhydride to afford adduct 156 in moderate yield. Hydrogenation of
the adduct produced anhydride 157. Treatment of 157 with water did
not appear to produce the desired diacid. Reaction with sodium
hydroxide decomposed the starting material. As a more promising
route was found, this one was abandoned.

A [4+3] cycloaddition to furan 155 was next explored.
Bromination of 3-methyl-2-butanone affords bromide 159 in good
yield. In a model system, the enol silyl ether of 159 reacted with furan
and zinc chloride to produce the cyclized compound 161. After this
positive result, 155 and 160 were combined in the presence of zinc chloride. Chromatography of the reaction mixture yielded starting material, the desired cycloadduct 162, and an uncyclized addition product 163. Boron trifluoride etherate did not effect cyclization when used in place of zinc chloride.

Hydrolysis of 162 with lithium hydroxide required several days at room temperature and did not reach completion. However, the acid was produced in 81-84% yield, and was submitted for biological testing. Acid 164 has not yet been tested.
TMSO\(_\text{Br} \) + 155 \rightarrow \text{CH}_2\text{Cl}_2 \rightarrow -78 \, ^\circ\text{C} \rightarrow \text{R.T.} \rightarrow 162

13-20%

\begin{align*}
+ &\text{S. M.} \\
40-45% &\rightarrow \end{align*}

\begin{align*}
+ &\text{S. M.} \\
15-16% &\rightarrow \end{align*}

162 + LiOH\cdot\text{H}_2\text{O} \rightarrow \text{THF}/\text{MeOH}/\text{H}_2\text{O} \rightarrow 162 + \text{O}

3.6:3.6:1

\begin{align*}
162 &\rightarrow \text{164} \\
\text{162} &\rightarrow \end{align*}
CONCLUSION

Several analogs of glycinoeclepin A were synthesized and submitted for testing of their hatch-stimulating ability. The analogs possessed a variety of the functional groups found in glycinoeclepin A and were constructed using a variety of synthetic methods. While only one of the analogs tested to date (compound 68) has shown any activity, this information is valuable in the determination of which functionalities are critical for stimulation of soybean cyst nematode hatching.
EXPERIMENTAL

Unless otherwise noted, materials were obtained from commercial suppliers and were used without purification. Diethyl ether and tetrahydrofuran (THF) were distilled from sodium benzophenone ketyl. Benzene was distilled from lithium aluminum hydride. Methylene chloride and acetonitrile were purified by distillation from calcium hydride. Apparatus for experiments requiring anhydrous conditions was flame-dried under a stream of nitrogen or dried in a 150°C oven for 12 h and cooled under a stream of nitrogen or in a desiccator. Flash chromatography was performed on EM Science Kieselgel 60 (mesh 230-400). Thin layer chromatography was performed using EM Science Kieselgel F254 prepared plates with a thickness of 0.25 mm. The solvent systems were suitable mixtures of hexanes (H) and ethyl acetate (EA) unless otherwise noted. The abbreviation sg represents silica gel. Infrared spectra were obtained on a Perkin-Elmer 1320 spectrophotometer and are reported in cm⁻¹. Proton nuclear magnetic resonance spectra (300 MHz) were obtained using a Nicolet Magnetics Corporation NT-300 spectrometer. All chemical shifts are reported in δ relative to tetramethylsilane as an internal standard. Splitting patterns are designated as s (singlet), d (doublet), t (triplet), q (quartet), dd (doublet of doublets), dt (doublet of triplets), ABq (AB quartet), and m (multiplet); a br prefix indicates a broadened pattern. Carbon-13 NMR spectra were obtained on a Varian Associates VXR-300 spectrometer and are reported in δ relative to CDCl₃ (77.00 ppm). High resolution
mass spectra were obtained on a Kratos model MS-50 spectrometer. Low resolution mass spectra were obtained on a Finnegan 4023 mass spectrometer. Melting points were obtained on a Fisher-Johns melting point apparatus and are uncorrected. Elemental analyses were performed by Galbraith Laboratories. The purity of all title compounds was determined to be >90% by 300 MHz proton NMR and/or elemental analysis.

**Ethyl 2-[(ethoxycarbonyl)methylene]-1-methyl-6-oxocyclohexane propionate (48):** To a stirred suspension of NaH (1.27 g of a 60% mull in mineral oil, 31.8 mmol, washed three times with hexanes and dried with N₂) in 64 mL dry THF at 0 °C under N₂ was added dropwise triethylphosphonoacetate (7.13 g, 31.8 mmol) in 25 mL THF. After 45 min at 0 °C, dione 45₁₄ (6.00 g, 26.5 mmol) in 27 mL THF was added dropwise. The mixture was allowed to warm to room temperature with stirring, then heated to 50 °C in an oil bath. The reaction was monitored by TLC. When starting material was still present after two days, the mixture was heated to reflux and stirred for three h. The mixture was cooled to room temperature, 200 mL water was added, and the aqueous layer was extracted with methylene chloride. The combined organic layers were washed with brine, dried over Na₂SO₄, and concentrated in vacuo. The residue was purified by sgc chromatography using 3:1 H:EA to yield 48 (5.93 g, 76%) as a pale oil. (Large quantities of 48 were purified by high vacuum distillation, collecting the material distilling from 137 °C to 150 °C at 30-33 μm Hg.)
48: $^1$H NMR (CDCl$_3$) $\delta$ 1.25 (d, J = 0.6 Hz, 3H), 1.22-1.32 (m, 6H), 1.61-1.79 (m, 1H), 1.85-2.09 (m, 2H), 2.13-2.35 (m, 3H), 2.39-2.71 (m, 3H), 3.65 (dt, J = 13.8, 4.5 Hz, 1H), 4.07-4.20 (m, 4H), 5.75 (s, 1H); IR (Neat) 2975, 2935, 2900, 2870, 1720 (Broad), 1630 cm$^{-1}$; MS (Cl-NH$_3$) $m/e$ 297 (M+H), 314 (M+NH$_4$); TLC (3:1 H:EA) $R_F$ = 0.38.

General Procedure for Ketalization: A mixture of the ketone (1 equiv), ethylene glycol (2 equiv), and PTSA (0.12 equiv) in dry benzene (0.14 M in the ketone) was heated to reflux in dry benzene under nitrogen in a flask fitted with a Dean-Stark trap and a reflux condenser. Reflux was maintained until TLC indicated the reaction to be complete. If necessary, activated molecular sieves and/or additional PTSA was added to the reaction flask to drive the reaction to completion. The reaction mixture was cooled to room temperature, washed with saturated aqueous NaHCO$_3$, water, and brine, dried over Na$_2$SO$_4$, and concentrated in vacuo. The crude product was purified by sg chromatography using H:EA.

Ethyl 2-[(ethoxycarbonyl)methylene]-6,6-(ethylenedioxy)-1-methylcyclohexane propionate (49): $^1$H NMR (CDCl$_3$) $\delta$ 1.03 (s, 3H), 1.24 (t, J = 7.2 Hz, 3H), 1.28 (t, J = 7.2 Hz, 3H), 1.49 (dt, J = 13.2, 4.2 Hz, 1H), 1.60-1.72 (m, 2H), 1.72-1.85 (m, 1H), 1.86-2.05 (m, 4H), 2.11-2.22 (m, 2H), 3.83-4.00 (m, 4H), 4.05-4.17 (m, 4H), 5.66 (s, 1H); IR (Neat) 2975, 2950, 2880, 1730, 1715, 1635 cm$^{-1}$; MS (Cl-NH$_3$) $m/e$ 295, 317, 334, 341 (M+H), 358 (M+NH$_4$); TLC (3:1 H:EA) $R_F$ = 0.41.
1',2',3',6',7',7'a-Hexahydro-1-[3-(hydroxycarbonyl)ethyl]-7'a-methyl-spiro[1,3-dioxolane-2,5'-[5H]indene] (132):  
$^1$H NMR (CDCl$_3$) δ 0.91 and 1.12 (s, 3 H), 1.28-2.03 (m, 9 H), 2.21-2.40 (m, 4 H), 3.67 (s, 3 H), 3.88-3.98 (m, 4 H), 5.26 and 5.31 (br s, 1 H); IR (Neat) 2950, 2880, 1735; TLC (5:1 H:EA) $R_F$ = 0.26.

Ethyl 2-[(ethoxycarbonyl)methylene]-6-(2-hydroxyethoxy)-1-methylcyclohexane propionate (53): To a solution of 49 (9.57 g, 28.1 mmol) in 100 mL dry methylene chloride at 0 °C under nitrogen was added triethylsilane (4.90 g, 42.2 mmol) dropwise. BF$_3$·Et$_2$O was then added dropwise and the solution allowed to stir while warming to room temperature overnight. TLC indicated that starting material remained; thus 50% more triethylsilane was added, the mixture cooled to 0 °C, 50% more BF$_3$·Et$_2$O was added, and the mixture was stirred at room temperature and monitored by TLC. After four h, the mixture was heated to reflux and again monitored by TLC. After five h at reflux, heating was ceased and the mixture stirred while cooling to room temperature overnight. TLC then indicated the reaction to be essentially complete. The mixture was washed with 70 mL saturated aqueous NaHCO$_3$, 500 mL ether was added, the organics washed with 100 mL saturated aqueous NaHCO$_3$ and 100 mL brine, dried over Na$_2$SO$_4$, and concentrated in vacuo. Purification of the crude alcohol by flash sg chromatography (3:1 H:EA) afforded 53 (7.45 g, 77%) as a pale oil: $^1$H NMR (CDCl$_3$) δ 1.17 (s, 3 H), 1.25 (t, J = 7.1 Hz, 3 H), 1.29 (t, J = 7.1 Hz, 3H), 1.59-2.26 (m, 9H), 3.01 (dd, J = 4.4, 10.7 Hz, 1H), 3.38-
120

3.43 (m, 1H), 3.57-3.73 (m, 4H), 4.07-4.18 (m, 4H), 5.69 (s, 1H); IR (Neat) 3480 (broad), 2980, 2935, 2865, 1730, 1715, 1630 cm⁻¹; MS (Cl-NH₃) m/e 360 (M+NH₄); TLC (1:1 H:EA) Rₚ = 0.36.

Ethyl 2-[(ethoxycarbonylmethylene)-6-(2-hydroxyethoxy)-1-methylcyclohexane propionate (67b): To a solution of DMSO (0.66g, 8.41 mmol) in 21 mL dry methylene chloride at -78 °C under N₂ was added dropwise oxalyl chloride (0.80 g, 6.31 mmol). After 10 min at -78 °C, 53 (1.44 g, 4.21 mmole) in 4.2 mL dry methylene chloride was added dropwise along with a 1 mL rinse. The mixture was stirred at -78 °C for one h, during which a white precipitate formed. Triethylamine was then added dropwise, the mixture was stirred while warming to 0 °C, and stirred at 0 °C for 10 min. The mixture was then poured into 20 mL saturated aqueous NH₄Cl and 120 mL ether, the organics washed with water and brine, dried over Na₂SO₄, and concentrated in vacuo. The crude aldehyde was obtained as a yellow oil in nearly quantitative yield (1.43 g, > 99% crude): ¹H NMR (CDCl₃) δ 1.22 (s, 3H), 1.22-1.31 (m, 6H), 1.61-2.32 (m, 9H), 3.05 (dd, J = 4.2, 10.8 Hz, 1H), 3.64-3.74 (m, 1H), 3.93-4.18 (m, 6H), 5.71 (s 1H), 9.73 (s, 1H); IR (Neat) 3445 (broad), 2975, 2955, 2865, 1733 (broad), 1710 (broad), 1630 cm⁻¹; TLC (1:1 H:EA) Rₚ = 0.40.

The bis-methyl ester was prepared analogously:

Methyl 2-[(methoxycarbonylmethylene)-6-(2-hydroxyethoxy)-1-methylcyclohexane propionate (67a): ¹H NMR (CDCl₃) δ 1.21 (s, 3H), 1.63-2.25 (m, 10H), 3.00-3.12 (m, 1H), 3.65 (s, 3H), 3.70 (s, 3H), 3.94-4.15 (m(ABq?), 2H), 5.72 (s, 1H), 9.73 and 9.73
General Procedure for Silver Oxide Oxidation: To a 0.2 M solution of the aldehyde (1 equiv) in absolute ethanol or methanol was added dropwise a 4.67 M aqueous solution of silver nitrate (2.33 equiv). A 1.07 M aqueous solution of potassium hydroxide (5.35 equiv) was then added dropwise (a black ppt formed), and the mixture stirred at room temperature for 2.5-4.5 h. When TLC indicated the reaction to be complete, the silver salts were filtered on a fritted funnel and washed with water. The aqueous layer was washed with ether, acidified to pH = 3, and extracted with methylene chloride and ether. The organic layers from the second extraction were dried over Na₂SO₄ and concentrated in vacuo.

2-[(Methoxycarbonyl)methylene]-6-(3-hydroxy-1-oxa-3-oxopropyl)-1-methylcyclohexane propionic acid (68): Viscous yellow oil which became a pale yellow foam when placed under vacuum for several hours; ¹H NMR (CDCl₃) δ 1.21 (s, 3H), 1.65-2.35 (m, 9H), 3.11-3.16 (m, 1H), 3.55-3.65 (m, 1H), 3.70 (s, 3H), 4.04-4.21 (m(ABq?), 2H), 5.72 and 5.73 and 5.76 (br s, 1H); IR (CDCl₃) 3400-3000 (OH), 2970, 2940, 2865, 1720, 1710 (broad), 1630 cm⁻¹; MS (CI-NH₃) m/e 244, 258, 274, 288, 304, 318, 332 (M+NH₄), 346 (M+NH₄ for residual ethyl ester).

1,2,3,6,7,7a-Hexahydro-7a-methyl-5-oxo-1-[5H]indene acetic acid (98): Yellow oil; ¹H NMR (CDCl₃) δ 1.06 (s, 3H), 1.20-1.45 (m, 1H), 1.72-1.93 (m, 1H), 1.93-2.21 (m, 1H), 2.21-2.75 (m, 8H), 5.80
and 5.86 (br s, 1H); IR (Neat) 3600-3080 (-OH), 2940, 2920, 1725, 1705, 1660, 1645 cm\(^{-1}\); MS (CI-NH\(_3\)) \(m/e\) 226 (M+NH\(_4\)), 243 (M+NH\(_4\)+NH\(_3\)), 434 (2M+NH\(_4\)).

General Procedure for Lithium Hydroxide Hydrolysis: To a 0.13 M solution of the ester (1 equiv) in 3.6:3.6:1 THF:MeOH:H\(_2\)O at room temperature under N\(_2\) was added LiOH\(\cdot\)H\(_2\)O in one portion. The reaction flask was briefly flushed with N\(_2\), and the mixture stirred at room temperature or 50°C until TLC showed the reaction to be complete. The mixture was then cooled to room temperature, concentrated to remove THF and MeOH, the residue taken up in water, the aqueous extracted with ether and methylene chloride, acidified to pH = 3, extracted with ether, the organic layers from the second extraction washed with a small portion of brine, dried over Na\(_2\)S\(_2\)O\(_4\), and concentrated in vacuo.

2-([Hydroxycarbonyl)methylene]-1-methyl-6-(3-hydroxy-1-oxa-3-oxopropyl) cyclohexane propionic acid (69): white foam; \(^1\)H NMR (Acetone D-6) \(\delta\) 1.26 (s, 3H), 1.65-2.30 (m, 9H), 3.22 (dd, J = 4.4, 10.7 Hz, 1H), 3.62-3.75 (m, 1H), 4.10-4.23 (m(ABq?), 2H), 5.76 (s, 1H), 10.61 (br s); IR (Film) 3500-3000 (-OH), 2940, 2871, 2679, 2575, 1740, 1701, 1686, 1630 cm\(^{-1}\); MS (CI-NH\(_3\)) \(m/e\) 242, 318 (M+NH\(_4\)); TLC (6:3:1 CHCl\(_3\):MeOH:AcOH) \(R_F\) = 0.67; mp = 171-176 °C.

1',2',3',6',7',7'a-Hexahydro-1-[3-(methoxycarbonyl)-ethyl]-7'a-methyl-spiro[1,3-dioxolane-2,5'-[5H]indene] (134): white solid; \(^1\)H NMR (CDCl\(_3\)) \(\delta\) 0.92 and 1.13 (s, 3H), 1.32-2.03 (m, 9H), 2.33-2.42 (m, 4H), 3.89-3.98 (m, 4H), 5.26 and 5.31 (br s, 1H); IR (CDCl\(_3\))
123

3450-3100 (-OH), 2956, 2934, 2892, 1707 cm⁻¹; MS (CI-NH₃) m/e 267 (M+H), 284 (M+NH₄), 301 (M+NH₄+NH₃); TLC (6:3:1 CHCl₃:MeOH:AcOH) Rₑ = 0.80; mp = 102-105 °C.

Lithium 5-[2-lithiumoxycarbonyl)ethyl]-1,2,3,6,7,7a-hexahydro-5-hydroxy-7a-methyl-1-[5H]indene-3-propionate (144): The lithium carboxylate was prepared by the above method without acidic workup. The reaction mixture was neutralized and concentrated to afford a tan powder: ¹H NMR (D₂O) δ 0.73 and 0.93 (s, 3H), 1.07-1.35 (m, 5H), 1.42-1.82 (m, 6H), 1.87-2.10 (m, 2H), 3.18 (s, 2H), 5.01 and 5.81 (br s, 1H).

1-[2-(Hydroxycarbonyl)benzyl]-4,4-dimethyl-8-oxa-3-oxobicyclo[3.2.1]octene (164): light orange powder; ¹H NMR (CDCl₃) δ 0.93 (s, 3H), 1.23 (s, 3H), 2.40 (d, J = 16.2 Hz, 1H), 2.71 (d, J = 15.9 Hz, 1H), 3.52-3.63 (m (ABq?), 2H), 4.43 (s, 1H), 6.10-6.16 (m 2H), 7.30-7.52 (m, 3H), 7.98 (d, J = 7.8 Hz, 1H); IR (Film) 3130 (broad), 3074, 2967, 2928, 1709, 1696 cm⁻¹; MS (CI-NH₃) m/e 287 (M+H), 304 (M+NH₄); HRMS m/e for C₁₇H₁₈O₄, calcd 286.12051, measured 286.12010; mp = 131-142 °C.

2-Methyl-3-(2-propenyl)-cyclopentan-1-one (83): To a solution of 2-methyl-2-cyclopenten-1-one (9.23 g, 96 mmol) in 128 mL of dry methylene chloride at -78 °C under N₂ was added TiCl₄ (36.42 g, 192 mmole) dropwise over 12 minutes. After 20 minutes' stirring at -78°C, allyl trimethylsilane (27.42 g, 240 mmol) was added dropwise over 20 min. The dark purple mixture was stirred at -78 °C for two h, the cold bath exchanged for a normal ice bath, and the mixture stirred
an additional two h. It was then re-cooled to -78 °C, quenched with 160 mL water, the aqueous extracted with methylene chloride, and the organics washed with water and brine, dried over Na$_2$SO$_4$, and concentrated. Distillation of the crude yellow oil at 5 mm Hg and collecting the material distilling from 67-100 °C yielded 83 as a colorless liquid (9.11g, 69%): $^1$H NMR (CDCl$_3$) $\delta$ 1.01 and 1.08 (d, $J = 7.2$ Hz, d, $J = 6.6$ Hz, 3H), 1.33-1.51 (m, 1H), 1.67-2.01, (m, 2H), 2.01-2.15 (m, 2H), 2.15-2.26 (m, 1H), 2.26-2.49 (m, 2H), 5.00-5.13 (m, 2H), 5.69-5.91 (m, 1H); IR (Neat) 3070, 2960, 2925, 2870, 1735, 1635 cm$^{-1}$; TLC (9:1 H:EA) $R_F = 0.32$.

1,2,3,6,7,7a-Hexahydro-7a-methyl-5-oxo-1-(2-propenyl)-[5H]indene (91): To 43 mL dry ether at room temperature under N$_2$ was added 4.29 mL 2.5 M ethanolic potassium hydroxide. The mixture was cooled in an ice/salt bath and 83 (8.32 g, 60.2 mmol) was added dropwise (neat + 2 - 1.0 mL ether rinses). Methyl vinyl ketone (4.22 g, 60.2 mmol) in 10.72 mL dry ether was then added slowly dropwise with the bath temperature maintained at -6° to -7 °C and the mixture stirred at -7 °C for 45 min. The ice bath was removed and the mixture stirred an additional 45 minutes at room temperature, after which it was poured into 43 mL 10% aqueous HCl. The aqueous layer was saturated with NaCl and extracted with ether, and the organics dried over Na$_2$SO$_4$ and concentrated in vacuo. The crude amber oil was checked by $^1$H NMR then taken up in 43 mL 10% ethanolic potassium hydroxide and heated to reflux, where the mixture was stirred for 30 min. The cooled mixture was neutralized with glacial
acetic acid to pH = 6, concentrated, and the residue taken up in 200 mL water. The aqueous layer was extracted with ether, the organics washed with brine, dried over Na$_2$SO$_4$, and concentrated to yield an amber oil. Purification by sg chromatography provided 91 as a light yellow oil (6.68 g, 58 % over two steps): $^1$H NMR (CDCl$_3$) $\delta$ 1.04 and 1.25 (s, 3H), 1.44-1.79 (m, 3H), 1.89-2.15 (m, 3H), 2.17-2.73 (m, 5H), 4.93-5.10 (m, 2H), 5.64-5.88 (m, 2H); IR (Neat) 3065, 2960, 2940, 2920, 2865, 1665, 1635 cm$^{-1}$; MS (CI-NH$_3$) m/e 191 (M+H), 208 (M+NH$_4$), 381 (2M+H); TLC (4:1 H:EA) $R_F = 0.34$.

1,2,3,6,7,7a-Hezhydro-4-hydroxycarbonyl-7a-methyl5-oxo-1-(2-propenyl)-[5H]indene (92): To a suspension of NaH (0.30 g of a 60% mull in mineral oil, 7.39 mmol) and KH (two small drops of a 35% dispersion in mineral oil) in 14.7 mL dry dimethoxyethane (DME) at room temperature was added rapidly enone 91 (1.58 g, 8.30 mmole) in 2.1 mL DME. The mixture was stirred at room temperature overnight. The dark mixture was then cooled to 0 °C and CO$_2$ (passed through a CaCl$_2$ drying tube) was bubbled through it for 75 min. 14.8 mL 10% aqueous NaOH was added and the mixture stirred at 0 °C for 15 min. The aqueous layer was extracted with ether, acidified to pH = 2-3, stirred for 30 min, and extracted with ether. The combined organics from the second extraction were dried over Na$_2$SO$_4$ and concentrated to afford a yellow oil which began to crystallize upon cooling: $^1$H NMR (CDCl$_3$) $\delta$ 1.13 and 1.34 (s, 3H), 1.52-1.95 (m, 3H), 1.99-2.33 (m, 4H), 2.54-2.93 (m, 2H) 3.27 (dd, J = 5.3, 9.5 Hz, 2H), 4.98-5.14 (m, 2H), 5.61-5.87 (m, 1H); IR (Neat) 3500-3100 (-OH), 3065, 2965,
126

2925, 2760, 1740, 1620, 1593 cm⁻¹; MS (CI-NH₃) m/e 235 (M+H), 252 (M+NH₄), 486 (2M+NH₄); TLC (3:1 H:EA) Rₚ = 0.06 (streak).

1,2,3,6,7,7a-Hexahydro-7a-methyl-1-(oxiranylmethyl)-5-oxo-[5H]indene (103): To a solution of 91 (0.57 g, 3.00 mmol) in 15 mL dry dichloroethane at room temperature under N₂ was added 3- tert-butyl-4-hydroxy-5-methylphenyl sulfide (5.7 mg, 0.016 mmol) then m-chloroperoxybenzoic acid (0.93 g of 67%, 3.59 mmol). The mixture was heated to 90 °C and stirred at that temperature for one h. TLC showed starting material was still present and starch iodide paper tested negative for peroxides. 4.2 mg inhibitor and 0.47 g m-CPBA were added and heating resumed for one h. TLC indicated the reaction was complete. The mixture was diluted with methylene chloride, washed with aqueous NaHSO₃ and aqueous NaHCO₃, and the aqueous extracted with methylene chloride. The organics were washed with brine, dried over Na₂SO₄, and concentrated in vacuo. Purification of the residue by sg chromatography provided 103 as a pale yellow oil: ¹H NMR (CDCl₃) δ 1.03 and 1.26 (s, 3H), 1.60-1.87 (m, 4H), 1.96-2.20 (m, 3H), 2.30-2.84 (m, 6H), 2.87-2.97 (m, 1H), 5.78 and 5.83 (br s, 1H); IR (Neat) 3040, 2980, 2945, 2925, 2865, 1700, 1665 cm⁻¹; MS (CI-NH₃) m/e 207 (M+H), 224 (M+NH₄), 241 (2M+NH₄+NH₃), 413 (2M+H), 430 (2M+NH₄); TLC (1:1 H:EA) Rₚ = 0.30.

1,2,3,6,7,7a-Hexahydro-7a-methyl-5-oxo-1-(2-oxoethyl)-[5H]indene (102): To a suspension of periodic acid (0.28 g, 1.23 mmol) in 17.5 mL dry ether at room temperature under N₂ was added rapidly dropwise epoxide 103 (0.25 g, 1.23 mmol) in 4.9 mL dry ether.
A precipitate began to form immediately, and the mixture was stirred vigorously at room temperature for three h. TLC showed starting material to be present, so 0.08 g periodic acid (27%) was added. The mixture was stirred until TLC showed it to be complete (2.5 h). The mixture was filtered through Celite, washed with saturated aqueous NaHCO₃ until no more orange color appeared in the aqueous layer, washed with brine, dried over Na₂SO₄, and concentrated in vacuo. An attempt to purify the aldehyde from another experiment had led to partial decomposition; thus the product, a yellow oil (0.14 g, 59%), was carried on unpurified; \(^1\)H NMR (CDCl₃) \(\delta\) 1.05 and 1.29 (s, 3H), 1.68-1.85 (m, 1H), 1.91-2.03 (m, 1H), 2.03-2.22 (m, 1H), 2.30-2.78 (m, 8H), 5.78 and 5.80 and 5.84 (br s, 1H), 9.79-9.83 (m, 1H); IR (Neat) 2960, 2920, 2860, 1720, 1663 (broad) cm⁻¹; TLC (20:10:1 ether: hexane: ethanol) \(R_F\) = 0.20.

\(1,2,3,6,7,7a\)-Hexahydro-1-(3-hydroxypropyl)-7a-methyl-5-oxo-[5H]indene (118): To a 1.0 M solution of borane in THF (17.34 mL, 17.34 mmol) at 0 °C under N₂ was added dropwise cyclohexene (2.85 g, 34.68 mmol) in dry THF. The ice bath was removed and the mixture stirred for 30 min at room temperature, during which time a white precipitate formed. The mixture was recooled to 0 °C, and allyl compound 91 (3.00 g, 15.8 mmol) in 7.9 mL dry THF was added dropwise. The mixture was allowed to stir with no further addition of ice to the cooling bath. TLC showed the reaction to be complete after 2.5 h. 10.4 mL ethanol, 3.48 mL 6 N NaOH, and 6.95 mL 30% H₂O₂ were added sequentially; this mixture was heated at 50 °C
for one h, and cooled to room temperature. The aqueous material was absorbed by Na₂SO₄, the Na₂SO₄ rinsed well with ether, the organics washed with brine, the brine back-extracted with ether, and the combined organics dried over Na₂SO₄ and concentrated. Compound 118 was obtained as a pale oil (2.60 g, 79%) after sg chromatography: ¹H NMR (CDCl₃) δ 1.02 and 1.24 (s, 3H), 1.24-1.37 (m, 1H), 1.41-1.79 (m, 6H), 1.95-2.13 (m, 2H), 2.29-2.80 (m, 5H), 3.64-3.70 (m, 2H), 5.76 and 5.81 (br s, 1H); IR (Neat) 3420 (broad), 2930, 2855, 1650 (broad); TLC (1:4 H:EA) Rₛ = 0.27.

1,2,3,6,7,7a-Hexahydro-7a-methyl-5-oxo-1-[5H]indene-
3-propionic acid (122): Alcohol 118 was taken up in reagent grade acetone and cooled to 0 °C. Jones reagent was added dropwise until a red color persisted. The mixture was stirred at 0 °C for three h, when TLC indicated the reaction to be complete. 2-Propanol was added until a green color persisted and the quenched mixture stirred at 0 °C for 20 min. Enough brine was added to dissolve the sticky blue-green residue and the aqueous layer was saturated with NaCl and extracted with ether and methylene chloride. The combined organic layers were washed with brine, the brine back-extracted with ether, and the organics dried over Na₂SO₄ and concentrated in vacuo. Base extraction with cold 1 N NaOH and ether afforded 0.74 g (87%) of 122 as a yellow oil: ¹H NMR (CDCl₃) δ 1.04 and 1.24 (s, 3H), 1.42-1.63 (m, 2H), 1.63-1.92 (m, 2H), 1.92-2.12 (m, 2H), 2.28-2.77 (m, 7H), 5.77 and 5.83 (br s, 1H); IR (Neat) 3500-3000 (-OH), 2940, 2860, 1730, 1705, 1660, 1635 cm⁻¹; MS (Cl-NH₃) m/z 223 (M+H), 240 (M+NH₄) TLC (1:4 H:EA) Rₛ = 0.11 (streak).
Methyl 1,2,3,6,7,7a-Hexahydro-7a-methyl-5-oxo-1-[5H]indene-3-propionate (125): A solution of diazomethane in ether was added via disposable pipet without stirring to a solution of acid 122 (0.56 g, 2.52 mmol) in methylene chloride in an Erlenmeyer flask until a bright yellow color persisted. The mixture was allowed to stand at room temperature for one h, N2 was bubbled through the solution for 45 min (the yellow color dispersed), and the solvent was removed in vacuo. Sg chromatography (2:1 H:EA) yielded 0.52 g (87%) of 125 as a pale yellow oil: 1H NMR (CDCl3) δ 1.04 and 1.24 (s, 3H), 1.48-1.56 (m, 1 H), 1.65-1.89 (m, 2H), 1.93-2.10 (m, 2H), 2.24-2.75 (m, 8H), 3.69 (s, 3H), 5.76 and 5.82 (br s, 1H); IR (Neat) 2949, 2868, 1734, 1669 (broad) cm⁻¹; MS (Cl-NH3) m/e 237 (M+H), 254 (M+NH₄), 473 (2M+H); HRMS calcd. for C₁₄H₂₀O₃ 236.14124, measured 236.14123; TLC (2:1 H:EA) Rf = 0.24.

Methyl 5-[2-(ethoxycarbonyl)ethyl]-1,2,3,6,7,7a-hexahydro-5-hydroxy-7a-methyl-1-[5H]indene-3-propionate (138): To a 1 M solution of LDA (prepared at 0 °C from diisopropylamine (0.15 g, 1.52 mmol) in 0.50 mL dry THF and n-BuLi (0.685 mL in hexane, 1.40 mmol)) at -78 °C under N2 was added ethyl acetate (0.12 g, 1.40 mmol) in 1.26 mL dry THF. The mixture was stirred at -78 °C for 90 min, and enone 125 (0.30 g, 1.27 mmol) in 1.27 mL dry THF was added dropwise. TLC showed the reaction to be essentially complete after 3.5 h. The mixture was quenched with 10% acetic acid in methylene chloride, stirred at -78 °C for 10 minutes, water was added, and the cooling bath removed. The mixture was stirred while warming
to room temperature, the pH adjusted to 7, the aqueous layer extracted with methylene chloride, and the organics washed with water and brine, dried over Na$_2$SO$_4$, and concentrated in vacuo. Purification by sg chromatography (2:1 H:EA) afforded 0.18 g (45%) of 138 as a pale yellow oil: $^1$H NMR (CDCl$_3$) $\delta$ 0.91 and 1.11 (s, 3H), 1.29 (t, $J$ = 7.2 Hz, 3H), 1.31-1.53 (m, 1H), 1.68-2.02 (m, 5H), 2.02-2.48 (m, 3H), 2.52 (s, 2H), 3.68 (s, 3H), 4.18 (q, $J$ = 7.2 Hz, 2H), 5.21 and 5.30 (br s, 1H); IR (Neat) 3510, 2950, 2865, 1747, 1728 cm$^{-1}$; TLC (2:1 H:EA) R$_F$ = 0.37.

Methyl 2-(2-furylmethyl)benzoate (155): To a solution of furan (4.82 g, 70.8 mmol) in 29 mL dry ether at 0 °C under N$_2$ was added n-BuLi (2.10 M in hexane, 32.48 mL, 68.2 mmol) dropwise. The ice bath was removed and the mixture stirred at room temperature protected from light for 70 min. 19.7 mL dry THF was added to dissolve the precipitate which formed, and the amber solution transferred over 20 min via canula to a suspension of copper(I) bromide•dimethyl sulfide complex (7.11 g, 34.6 mmol) in 44 mL dry THF at -45 °C under N$_2$. 5 mL additional THF was required to dissolve precipitate which fell back out of solution. After 45 min at -45 °C, bromoester 154$^{58}$ in 29 mL dry ether was added dropwise via canula, and the mixture allowed to stir while warming to room temperature, protected from light, in the ice bath overnight. The mixture was poured into saturated aqueous NH$_4$Cl, the aqueous extracted with ether, the organics washed with brine, dried over Na$_2$SO$_4$, filtered through Celite on a fritted funnel, and concentrated. Purification of the brown oil by sg chromatography provided 155 as a yellow oil (1.67g, 39%): $^1$H NMR
131

(CDC\textsubscript{3}) \textdelta 3.86 (s, 3H), 4.39 (s, 2H), 5.95-5.96 (m, 1H), 6.27 (dd, J = 1.8,3.0 Hz, 1H), 7.23-7.32 (m, 3H), 7.40 (td, J = 7.5, 1.2 Hz, 1H), 7.91 (dd, J = 1.2, 7.8 Hz, 1 H); IR (Neat) 3067, 2950, 1719, 1601 cm\textsuperscript{-1}; MS (CI-NH\textsubscript{3}) m/e 217 (M+H), 234 (M+NH\textsubscript{4}); HRMS m/e for C\textsubscript{13}H\textsubscript{12}O\textsubscript{3} calcd. 216.07864, measured 216.07857; TLC (30:1 H:EA) R\textsubscript{F} = 0.26.

1-(2-Methoxycarbonylbenzyl)-4,4-diethyl-8-oxa-3-oxo-bicyclo[3.2.1]octene (162): Zinc chloride (0.47 g, 3.47 mmol) was fused under vacuum in the reaction flask. Furan 155 (0.50 g, 2.31 mmol) in 2.31 mL dry methylene chloride was added and the mixture cooled to -78 °C. Enol silyl ether 160\textsuperscript{*1} in dry methylene chloride was added dropwise and the mixture stirred at -78 °C for one h. TLC showed no product being formed, so the mixture was warmed to -3 °C over 75 min. TLC showed no change, so the ice bath was removed. TLC after 30 min showed product, with no change after 30 more min. The mixture was poured into water, the organics washed with water, the aqueous extracted with methylene chloride, and the organics washed with brine, dried over Na\textsubscript{2}SO\textsubscript{4}, and concentrated. 162 was obtained as a yellow oil (0.14 g, 20%) after sg chromatography of the crude brown oil: \textsuperscript{1}H NMR (CDC\textsubscript{3}) \textdelta 0.92 (s, 3H), 1.24 (s, 3H), 2.64 (d, J = 15.9 Hz, 1H), 3.33 (d, J = 15.9 Hz, 1H), 3.55 (s, 2H), 3.89 (s, 3H), 4.39 (d, J = 1.5 Hz, 1H), 6.03-6.13 (m, 2H), 7.26-7.31 (m, 1H), 7.37-7.47 (m, 2H), 7.86 (dd, J = 1.2, 7.8 Hz, 1H); IR (Neat) 3072, 2952, 2927, 2869, 1717, 1702 cm\textsuperscript{-1}; MS (CI-NH\textsubscript{3}) 301 (M+H), 318 (M+NH\textsubscript{4}); HRMS m/e for C\textsubscript{18}H\textsubscript{20}O\textsubscript{4} calcd. 300.13616, measured 300.13643; TLC (10:1 H:EA) R\textsubscript{F} = 0.15.
133

Koho JP 02 83,385 [90 83,385] (Cl. C07D493108), 23 March
113, 59621h.

96, 219.
15. Wadsworth, Jr., W. S.; Emmons, W. D. J. Am. Chem. Soc. 1961,
83, 1733-8.
18. Kraus, G. A.; Frazier, K. A.; Roth, B. D.; Taschner, M. J.;
(b) Von Pechmann, H. Chem Ber. 1895, 28, 855.


APPENDIX: BIOLOGICAL TESTING

For the purpose of biological testing, zinc sulfate is the standard for SCN hatching activity to which synthetic compounds are compared. If a compound's activity is significantly less than that of ZnSO₄, it is considered inactive. If its activity is greater than or equal to that of ZnSO₄, it is considered active. To date, compounds 68, 92, 98, 122, 134, and 144 have been tested. 68 was active on its first test, negative on its second, and positive on its third. Unfortunately, the standard exhibited aberrant behavior on the third trial, and more testing has been ordered. Compound 92 tested negative. Compounds 98, 122, 134, and 144 all gave inconclusive test results due to unusual behavior of the standard. Compounds 69 and 164 have not yet been tested.
This dissertation has shown the value of the application of synthetic organic chemistry to other disciplines. Analogs of compounds which have potential as toxins or as agricultural aids have been synthesized. Demethoxyaflatoxin B\textsubscript{2} could be useful in structure/activity studies of the aflatoxins, as well as in studies on their mode of toxic action and means of detoxifying them. The synthetic methodology developed for the synthesis of the aflatoxin furofuran ring system complements the existing methods of radical oxidative cyclization in that mildly basic, rather than acidic conditions are used.

Analogs of glycinoeclepin A hold great potential as a safe and economical means of control of the soybean cyst nematode. Although only one of the analogs submitted for testing has shown hatch-stimulating activity to date, the negative tests shed light on the portions of the molecule necessary for biological activity.
First of all, I must thank God, through whose grace all things truly are possible.

Next, I must thank the many others who have contributed to my graduate career: Dr. Kraus, for his guidance and enthusiasm; members of the Kraus group, both past and present, for their help and support; and the various members of the services and support systems, who kept things running smoothly all these years.

I am grateful to my family for their continuing support.

Special thanks to those who made this dissertation possible through the generous loans of their computers, expertise, and time.

Finally, I give my heartfelt thanks to my husband, Benny, without whose love, support, typing, and Chemdrawing this dissertation would not have been possible. His patience, sacrifice, and unselfish assistance should qualify him for sainthood.

Too many others have helped me to be listed here by name, but not one has been forgotten.