Chapter 4

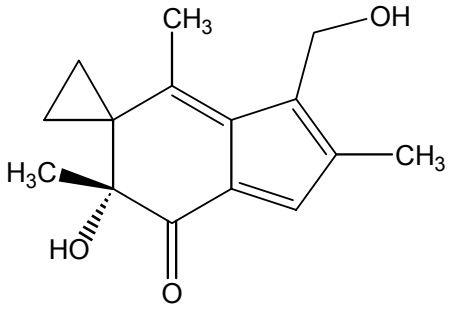
Total Synthesis of Hydroxymethylacylfulvene

Abstract

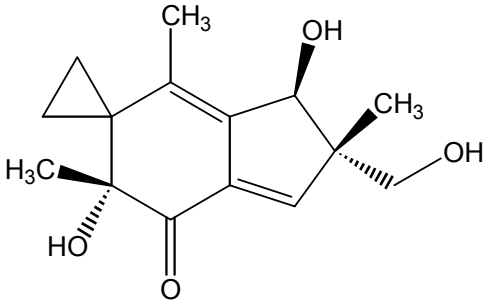
Hydroxymethylacylfulvene (**1**) (HMAF, also MGI 114) is a promising antitumor compound derived from the sesquiterpene illudin S (**2**). It is less cytotoxic than illudin S to normal cells and exhibits much greater selectivity in toxicity to malignant cells. A simple synthetic method is demonstrated to rapidly construct the skeleton of HMAF and its analogs starting from 2-(3-methoxyphenyl)ethanol. In particular this synthesis opens the door to enantioselective aromatic oxidation chemistry.

Introduction

Despite massive efforts put forward during the 20th century, human cancers continue to devastate the lives of millions of people worldwide. It has been estimated that over 100,000 Americans are diagnosed with cancer per year. The development of an antitumor drug that is highly efficient and less toxic to normal cells is needed. Hydroxymethylacylfulvene **1** is derived from the sesquiterpene illudin S by treatment with dilute sulfuric acid and excess paraformaldehyde. Illudin S was first isolated from the basidiomycete *Omphalotus illudens*. During the past 10 years, extensive preclinical studies on HMAF and related fulvenes have been conducted, leading to phase I human clinical trials which began in December 1995. Phase II clinical trials targeting several solid tumor types are now in progress under the sponsorship of MGI Pharma and the National Cancer Institute.[1,2]



HMAF
1

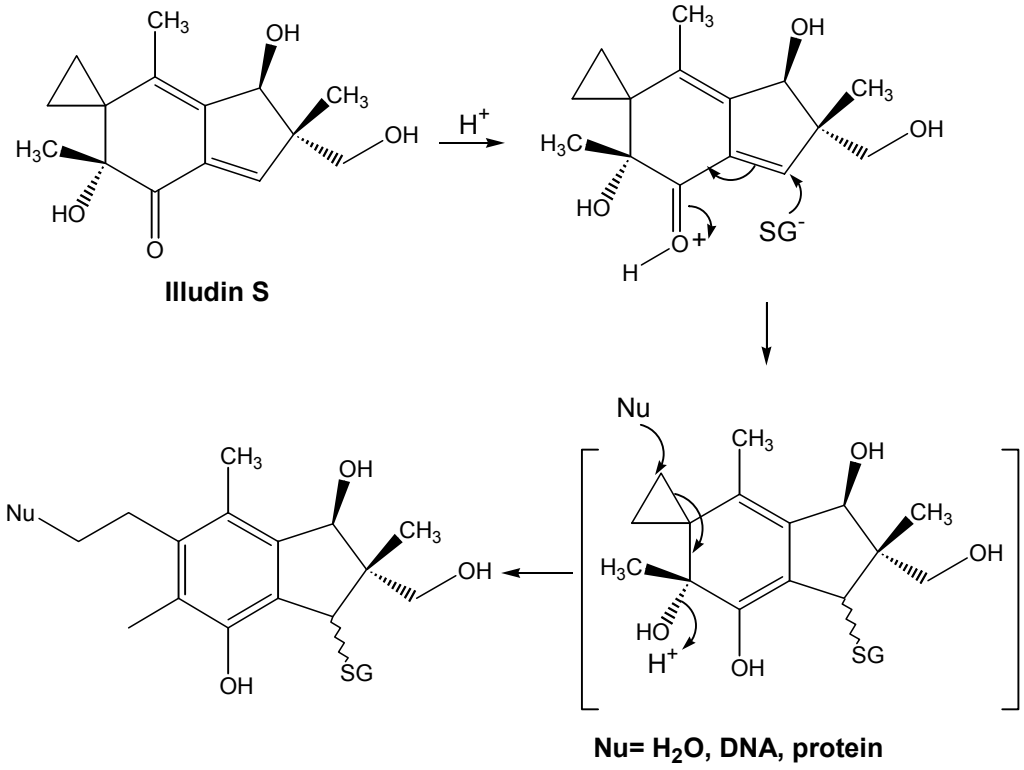


Illudin S
2

It is well known that many antitumor natural products behave as alkylating agents (Scheme 1). Among them are illudin compounds which react preferentially with thiols, and their cytotoxicity is attributed to the ability to react with vital thiol enzymes. Thiols

react readily at room temperature, adding to the α,β-unsaturated carbonyl and giving a cyclohexadiene intermediate which rapidly undergoes opening of the cyclopropane and loss of the tertiary hydroxyl. Reaction with thiols, e.g., methylthioglycolate, cysteine and glutathione is pH-dependent, with the optimum pH being 5.6-6.1. Not surprisingly, toxicity can be modulated by varying glutathione levels in cells. A third generation analog hydroxymethylacylfulvene (HMAF) caused complete tumor regression in all animals at the maximum tolerated dose of 10 mg/kg (iv) three times per week for 3 weeks. This resulted in increased life span of more than 150%. HMAF has also been found to exhibit outstanding activity against breast, colon, and skin cancer cell lines derived from human tumors.[1,3-9]

Scheme 1. Studies on the mechanism of action of illudins.



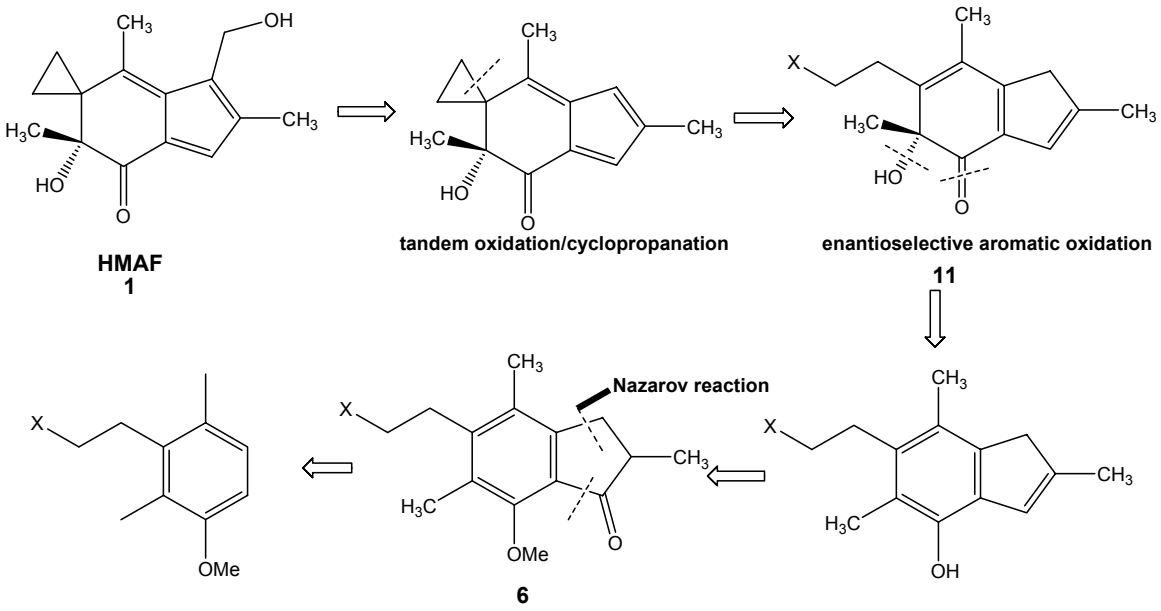
Biologically active natural products will have a profound effect on the field of medicine in the 21st century. However, these molecules are commonly isolated in small quantities and, therefore, cannot be subjected to advanced biological testing whereby the mechanism by which they act can be understood. Chemical synthesis can produce these compounds in the mass quantities necessary for intensive research and therapy.

Retrosynthesis

Several syntheses of structurally related compounds have appeared in the chemical literatures,[10-13] but these syntheses are not suitable for large scale production. A retrosynthetic analysis of the total synthesis of Hydroxymethylacylfulvene **1** (HMAF, also MGI 114) is illustrated in Scheme **2**. Starting from 2-(3-methoxyphenyl)ethanol, our strategy should lead to an efficient and versatile synthesis of HMAF and provide access to several derivatives of these products in sufficient amounts for further biological testing.

The key features of this synthesis are: (1)Nazarov reaction to form bicyclic system (**6**);[14] (2) Enantioselective aromatic oxidation chemistry to install the free hydroxy at the chiral center of intermediates (**11**); (3) Tandem oxidation/cyclopropanation at the final stage to afford HMAF efficiently.

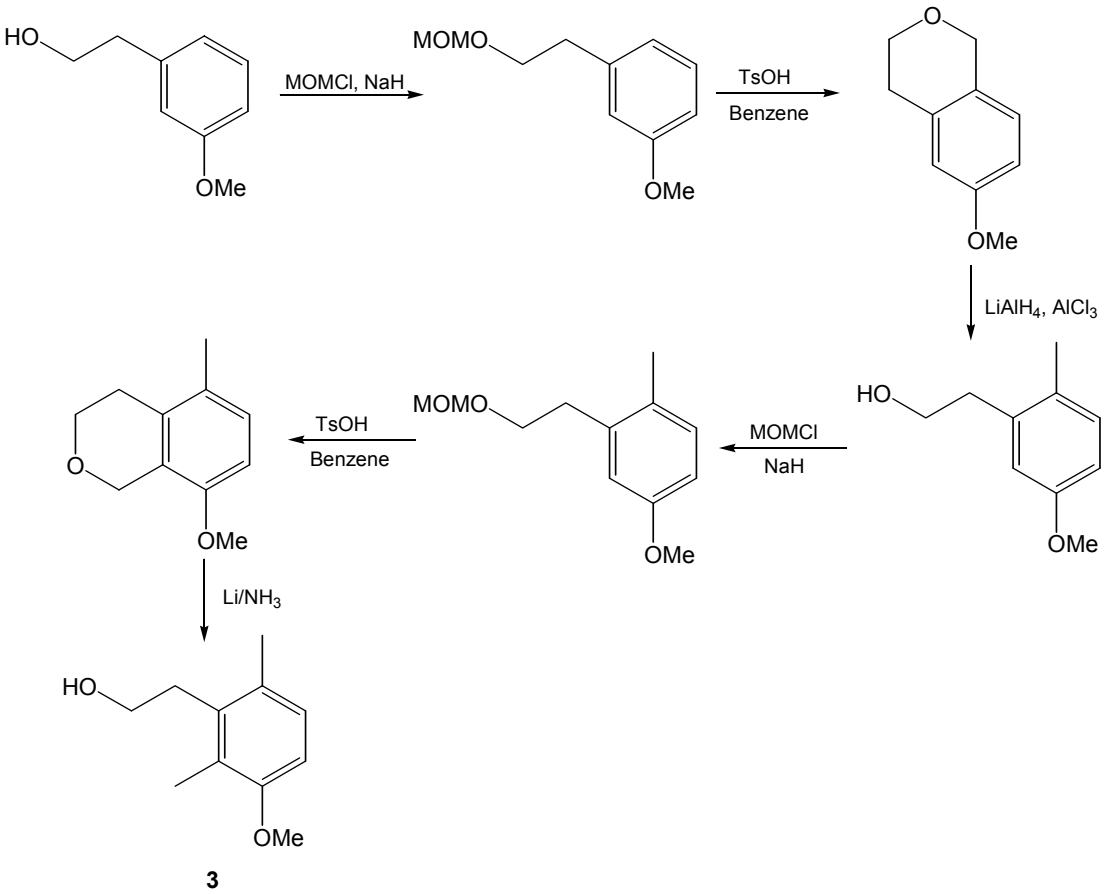
Scheme 2. Retrosynthesis of Hydroxymethylacylfulvene



Results and Discussions

Starting from (3-methoxy)-2-phenylethanol, compound (**3**) was synthesized as described in the literature.[15] The two methyl groups were introduced to the aromatic ring by Friedel-Crafts alkylation, followed by LAH or Li/NH_3 reduction (Scheme 3).

Scheme 3. Preparation of the Aromatic Subunit of HMAF.[7]

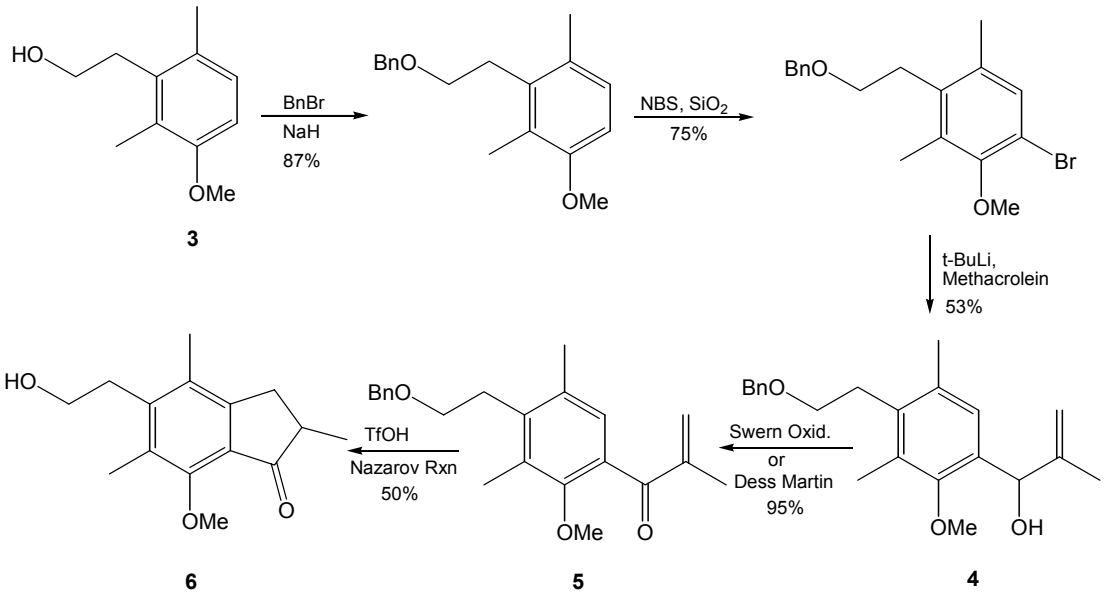


3

After benzyl protection of the hydroxyl group in 3-methoxy-2,6-dimethyl phenylethanol (**3**), selective α-bromization was achieved using NBS and silica gel

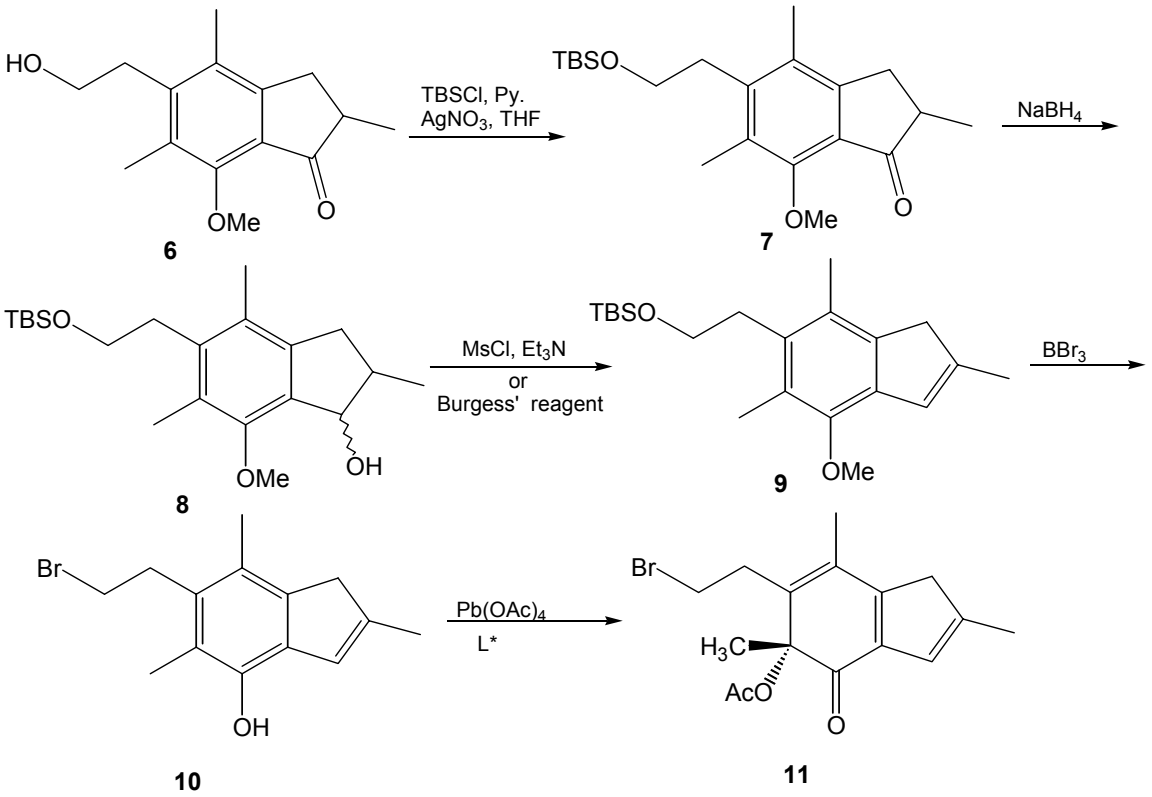
condition. Allylic alcohol (**4**) was formed by adding *t*-BuLi and methacrolein. Swern or Dess Martin oxidation gave α,β-unsaturated ketone (**5**) which underwent Nazarov reaction to form bicyclic system (**6**)(Scheme 4).

Scheme 4. Advancing the Aromatic Subunit---Nazarov Reaction



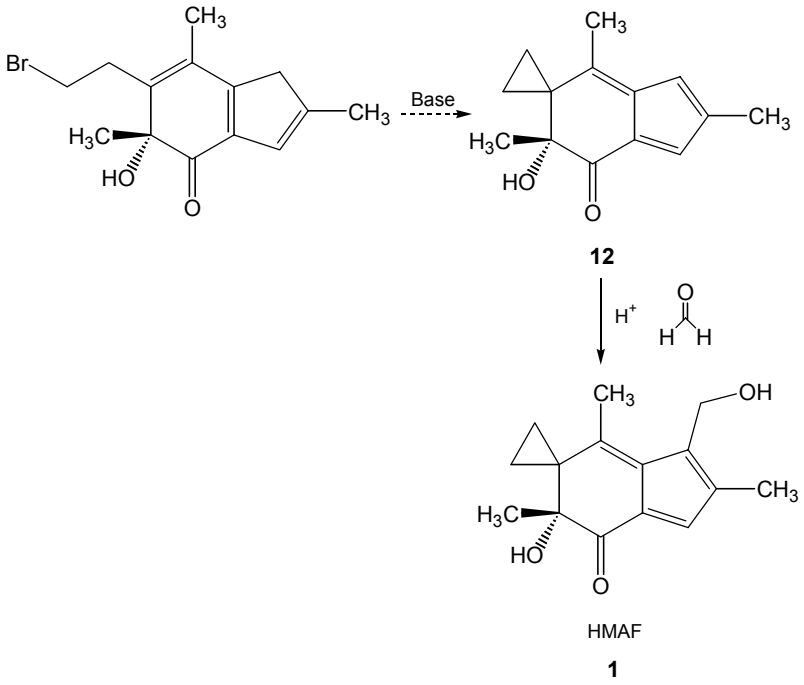
The free hydroxyl group of the compound (**6**) was protected by a silyl group. Ketone-alcohol-alkene transformation was achieved by reduction using $NaBH_4$ followed by treatment with Burgess' reagent, or MsCl and Et_3N. Phenol (**10**) was obtained by demethylation using BBr_3. Free hydroxy at the chiral center of intermediate (**11**) was designed to be installed by enantioselective aromatic oxidation chemistry, using $Pb(OAc)_4$ and chiral ligand followed by deacetylation. Instead of screening chiral ligand, enzymatic deacetylation will be a good way to achieve enantioselectivity (Scheme 5).

Scheme 5. Advancing the aromatic Subunit---Enantioselective aromatic oxidation



Final stage manipulation involves screening different base to achieve tandem oxidation/cyclopropanation to give known intermediate acylfulvene (**12**) which undergoes an ene reaction with formaldehyde to afford HMAF (**1**) (Scheme 6).

Scheme 6. Tandem oxidation/cyclopropanation



Conclusions

This research demonstrated a facile, efficient, and inexpensive synthesis for HMAF. In addition, a variety of acylfulvene analogues and their precursors will be synthesized and subsequently tested for biological activity. Although the HMAF and related molecules are known to have biological activity, the mechanisms by which they act remain to be further understood. The development of an efficient total synthesis of HMAF and its analogues would allow for the production of large quantities of each and thus render advanced biological testing of these molecules possible. The results of these experiments could establish precisely how these molecules function. Once the key molecular interactions are known, it should be possible to design molecules that have improved biological activity, bringing the world closer to uncovering the cure for cancer.

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PUBLICATIONS:

1. **Xin Qi,** Richard W. Roberts, "Synthesis and binding activity of λ N peptide-acridine conjugates directed against *BoxB* RNA" *Biochemistry*, *manuscript in preparation*.
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6. **Xin Qi**, John Montgomery, "New Three Component Synthesis of 1,3-Dienes Employing Nickel Catalysis" *J. Org. Chem.* **1999**, *64*, 9310-9313.