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Synthesis of natural product analogs as small molecule inhibitors

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Synthesis of natural product analogs as small molecule inhibitors

by

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To my parents

who love me unconditionally.

To my family

who always has faith in me.

To my friends

who are valuable in my life.

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NOMENCLATURE

Ac	acetyl
ACN	acetonitrile
Ar	aryl
Bn	benzyl
°C	degrees centigrade
calcd.	calculated
d	doublet
DBU	1,8-diazabicyclo[5.4.0]undec-7-ene
DCC	*N,N*′-dicyclohexylcarbodiimide
DCM	dichloromethane
dd	doublet of doublets
ddd	doublet of doublet of doublets
DIBAL	diisobutylaluminium hydride
DMAP	4-dimethylaminopyridine
DMF	*N,N*-dimethylformamide
equiv.	equivalent
Et	ethyl
Et_3N	trimethylamine
G	gram
h	hour
LRMS	low-resolution mass spectrometry

IC_{50}	50% inhibitory concentration
J	coupling constant
KO^tBu	potassium *tert*-butoxide
LDA	lithium diisopropylamide
LiO^tBu	Lithium *tert*-butoxide
LiTMP	lithium tetramethylpiperidine
M	molarity
m	multiplet
m-CPBA	meta-chloroperoxybenzoic acid
Me	methyl
mg	milligram
MHz	megahertz
min	minute
mL	milliter
MLV	modified live virus
mmol	millimole
mol%	mole percent
MOM	methoxymethyl
m.p.	melting point
n-Bu	normal butyl
NMR	nuclear magnestic resonance
P	para
PCC	pyridinium chlorochromate

PTSA	*p*-toluenesulfonic acid
PRRS	porcine reproductive and respiratory syndrome
PVN	percent virus neutralization
q	quartet
R_f	retention factor
s	singlet
sat.	saturated
t	triplet
TBAB	tetrabutylammonium bromide
TBAF	tetrabutylammonium fluoride
TBS	tert-butyldimethylsilyl
THF	tetrahydrofuran
TMAO	trimethylamine N-oxide
TMS	trimethylsilyl
UV	ultraviolet

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ABSTRACT

Natural products with interesting unique structures and special biological activities possess great potential in the development of novel pharmaceuticals. However, the evaluation of biological activity is always limited due to the low yield and difficulty of separation from plants and microorganisms. Synthetic organic chemistry provides an alternative tool to create such natural products from accessible materials. A huge mission for organic chemists is to develop novel strategies to produce natural products and their analogs in lab, as well as in quantitative scales. With this in mind, we have designed small molecule inhibitors towards Porcine reproductive and respiratory syndrome virus (PRRSV) and developed the synthetic routes to natural products psoracorylifols and furomollugin. In addition, a novel palladium nanoparticle catalyst was evaluated with the Suzuki-Miyaura reaction of aryl chloride.

Chapter one describes the design and synthesis of small molecule inhibitors of PRRSV. Natural 1-(E)- Atractylodinol was successfully produced in a seven-step direct route. More analogs with antiviral activity were synthesized based on this natural product. The goal to design a cost effective antiviral drug towards PRRSV was achieved.

The design of an approach to the unique 6,8-dioxabicyclo[3.2.1]octane skeleton in psoracorylifols B and C is discussed in chapter two. This special core structure was constructed via ketal formation in five steps.

The novel direct synthetic route to furomollugin is introduced in chapter 3. Furomollugin was synthesized by utilizing the Hauser-Kraus annulation as a key step.

The last chapter focuses on the evaluation and application of a novel palladium catalyst developed by Datye and co-workers. The utility of Datye catalyst was demonstrated in the Suzuki-Miyaura reaction of aryl chlorides and boronic acids. This catalyst showed great catalytic activity when aryl chlorides and aryl bromides were used for the Suzuki coupling in water media.

CHAPTER 1.

SMALL MOLECULE INHIBITORS TOWARDS PRRSV

1.1. Introduction

Porcine reproductive and respiratory syndrome (PRRS) is caused by Porcine reproductive and respiratory syndrome virus (PRRSV). Porcine reproductive and respiratory syndrome (PRRS), also named blue ear disease, is a widespread disease affecting domestic pigs. The clinical symptoms of PRRS are complicated but can be characterized by severe reproductive losses including abortions, mummified fetuses, weak born and stillborn young, post-weaning pneumonia, increased mortality, and growth retardation of young pigs. PRRSV, a small enveloped RNS virus, belongs to the Arteriviridae family which includes lactate dehydrogenase-elevating virus (LDV) of mice, equine arteritis virus (EAV), and simian hemorrhagic fever virus (SHFV) also.[1]

PRRS was first recognized in the United States in 1987, and the causative virus (PRRSV) was identified in Netherlands in 1991.[2] Currently, the PRRSV isolates are divided into two distinct strains: North American strain, VR-2332, and the European strain, the Lelystad virus (LV).[3, 4] Besides Europe and North America, it was also identified in China in 1995 and is present in Japan, Vietnam, the Philippines, Malaysia, and Korea among other agricultural countries in Asia. A few countries (Australia, New Zealand, several European countries, parts of Africa and India) did not find any evidence of this disease.[5]

After almost three decades, PRRS is still considered one of the most economically significant diseases in the global swine industry.[6] In 2005, the annual costs of PRRS for the American swine industry were approximately 560 million dollars, and recently, the estimated costs have increased to about 664 million dollars per year in the United States, with an additional 360 million dollars in annual veterinary costs. [7, 8] Additionally, in June 2006, an outbreak of a clinical syndrome called ‘‘high fever’’ emerged in pig farms and spread throughout more than 11 provinces in China, affecting over two million pigs within a few months, with high mortality rates, with PRRSV subsequently identified as the etiological agent.[9]

Control of PRRS virus has proven to be a challenge for swine practitioners throughout the world due to the complexity of PRRS disease. To control PRRS, current management strategies focus on the biosecurity and vaccination to prevent spreading of PRRSV. The Biosecurity protocols are targeted at reducing transmission risks and typically fall into one of three categories: Hygiene/Sanitation, Separation and Exclusion.[10] However, the biosecurity protocols can only be effective to the herds without disease, it is hard to eliminate the PRRS virus for the contaminated animals. Moreover, the biosecurity requires more personnel input and monetary investments to create a clean, virus free environment for uninfected pigs to stop infection.

Farmers combine biosecurity and vaccination to prevent the further economic loss caused by PRRS virus to increase the effectiveness of PRRS control. Vaccination is an effective tool to control PRRS if used correctly. The purpose of vaccination is to produce an immune response that will protect against clinical disease, but it does not prevent infection. Since PRRSV strains differ in virulence in infected pigs, it is important to

develop specific vaccination programs at the right time and place, based on individual farm diagnostic data, rather than promote standardized protocols. To control PRRS, modified live virus (MLV) vaccines appear to be more effective than killed preparations. Comparing with killed vaccines, MLV vaccines can give a strong, long-lasting immune response in fewer doses, have less chance to cause allergic reactions and behave more like the disease-causing virus. However, modified live vaccines should not be used in PRRS virus-negative herds, pregnant females, or breeding age boars.

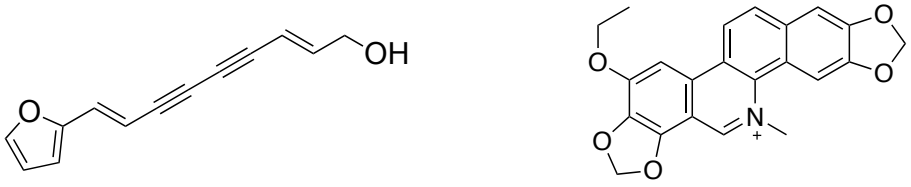
Although vaccines are used to treat PRRSV nowadays, there are still some significant drawbacks for the use of vaccines. PRRSV is genetically heterogeneous. Extensive sequence variation was found between the European and the U.S. isolates.[11] Biological and antigenic variations among PRRSV have been well reported too.[12] Moreover, Pathogenesis and pathogenic variation have been well documented although the mechanism of PRRSV pathogenesis is poorly understand. All these variations of PRRSV and immune evasion would reduce the effectiveness of vaccines and contribute to PRRSV persistence within an animal or herd. The new approaches in vaccine design may help in the long term to control and eliminate PRRSV. However, there is a need for alternate strategies and tools to reduce the economic burden of PRRS in the short term.

Our alternative strategy to control PRRS is the development and use of potent antiviral drugs. Vaccines may provide protection against a virus after days. Unlike vaccines, antiviral drugs can provide almost immediate treatment for animals, which would be beneficial in the event of a new outbreak or emergence of new virus variants. The antiviral drugs can be added to food to feed animals, which makes it possible to treat large numbers of animals at one time. The vaccine treatment can only be supplied to a

small amount of pigs due to the single injection of vaccines. The antiviral drugs can also be used in periods of increased susceptibility, such as post-weaning and transport. The final goal of our project is to develop potent and cost-effective small molecule inhibitors of PRRSV based on the natural products.

1.2. Results and Discussion

This is a collaboration project between Kraus lab and Carpenter lab in the Department of Animal Science at Iowa State University. We are responsible for all the organic synthesis of the compounds in this project and Carpenter lab conducts the biological tests of synthetic compounds. Ethoxysanguinarine and atracylodinol (Figure 1) were reported useful as anti-PRRSV drugs by Li group in 2013,[13] with IC_{50} values of 7.9 and 39.4 μmol/L, respectively. We chose natural product compound **1** atractylodinol as our target molecule for synthesis.



1
1-(E)-Atractylodinol

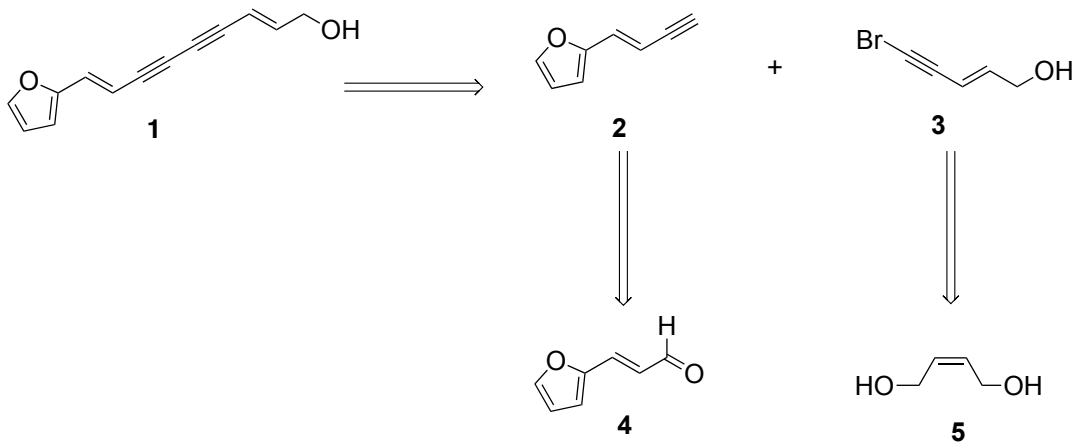
Ethoxysanguinarine

Figure 1. Structures of 1-(E)-Atractylodinol and Ethoxysanguinarin.

Natural 1-(E)- Atractylodinol was isolated from rhizomes of *Atractylodes lancea. A. lancea is* widely used in the traditional Chinese and Japanese medicines against

rheumatic diseases, digestive disorders, night blindness, and influenza.[14] The aqueous extract of *A. lancea* also helps the delay in gastric emptying[15] while the n-hexane extract of the rhizome exhibits good inhibitory activities in both 5-lipooxygenase (5-LOX) and cyclooxygenase-1 (COX-1) enzymatic in vitro assays.[16]

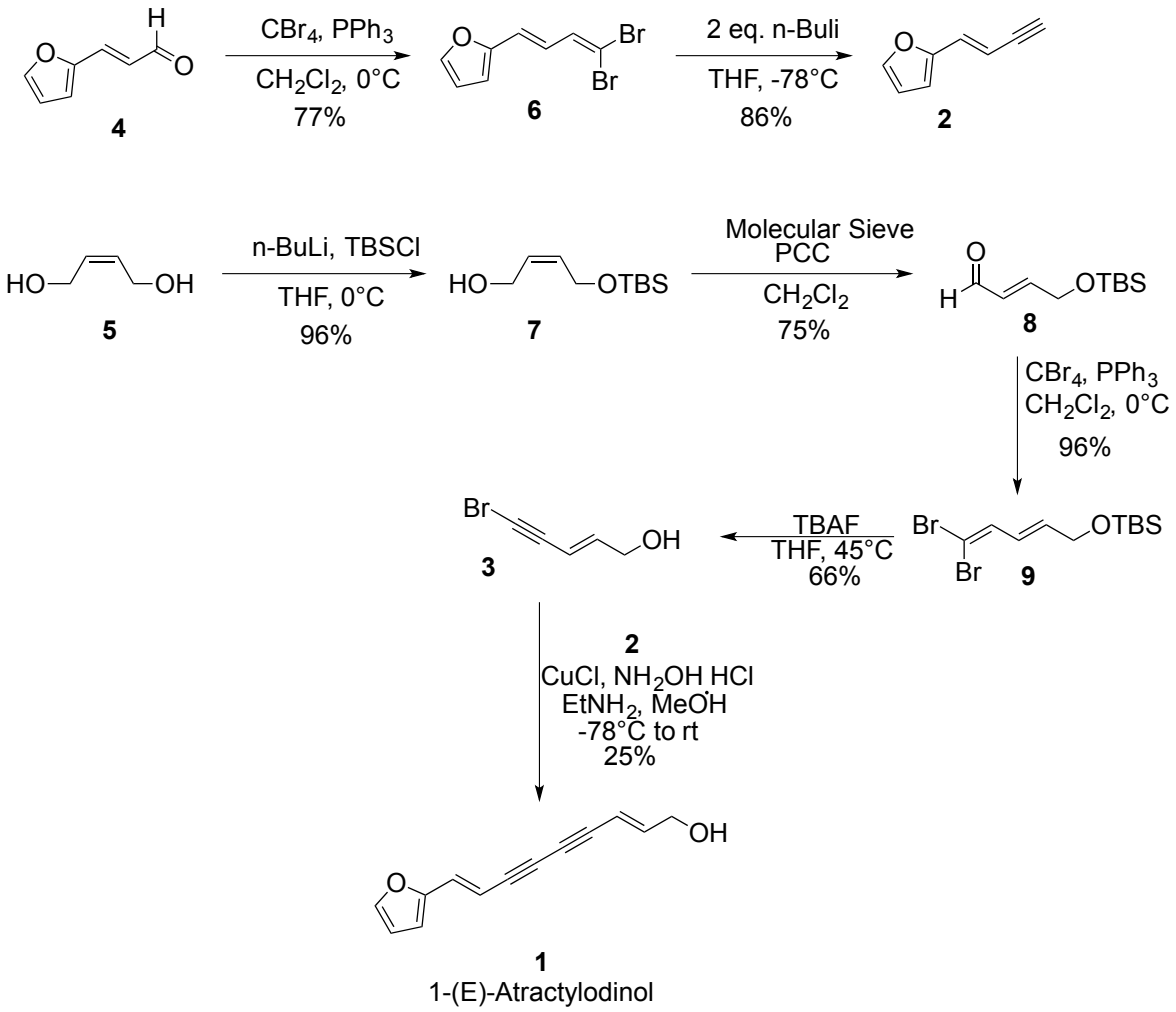
The reported isolation procedure gives 9.6 mg with about 70% of 1-(E)-atracylodiol from 570 g of dried and powered *A. lancea* rhizomes.[14] The low isolation yield and high cost cannot supply enough material for future biological activity tests in animals. Also to make the industrial quantitative production of the compound **1** is feasible, we decided to design a route to synthesize compound **1** in gram scale in the lab. The retrosynthetic route is shown in Scheme 1.



Scheme 1. Retrosynthetic route of 1-(E)-atractylodinol.

In our synthetic plan, coumpond **1** could be achieved by coupling reaction of furyl enyne **2** and bromo alcohol **3**. Furyl enyne **2** could be converted by well known Corey-Fuchs reaction from commercially available material 3-(2-furyl)acrolein **4**. From diol **5**, compound **3** is accessible through several steps including protection of one hydroxyl

group with TBSCl, PCC oxidation of the remaining alcohol followed by dibromonation, and deprotection of TBS ether to give free hydroxy group.



Scheme 2. Total synthesis of 1-(E)-atractylodinol.

We started the synthesis from 3-(2-furyl)acrolein **4**, as shown in Scheme 2. We chose to utilize Corey-Fuchs reaction to obtain the terminal alkyne from the aldehyde in two steps. Treatment of **4** with carbon tetrabromide and triphenylphosphine gave us dibromo compound **6**, which was converted to furyl enyne **2**.[17] The two-step conversion

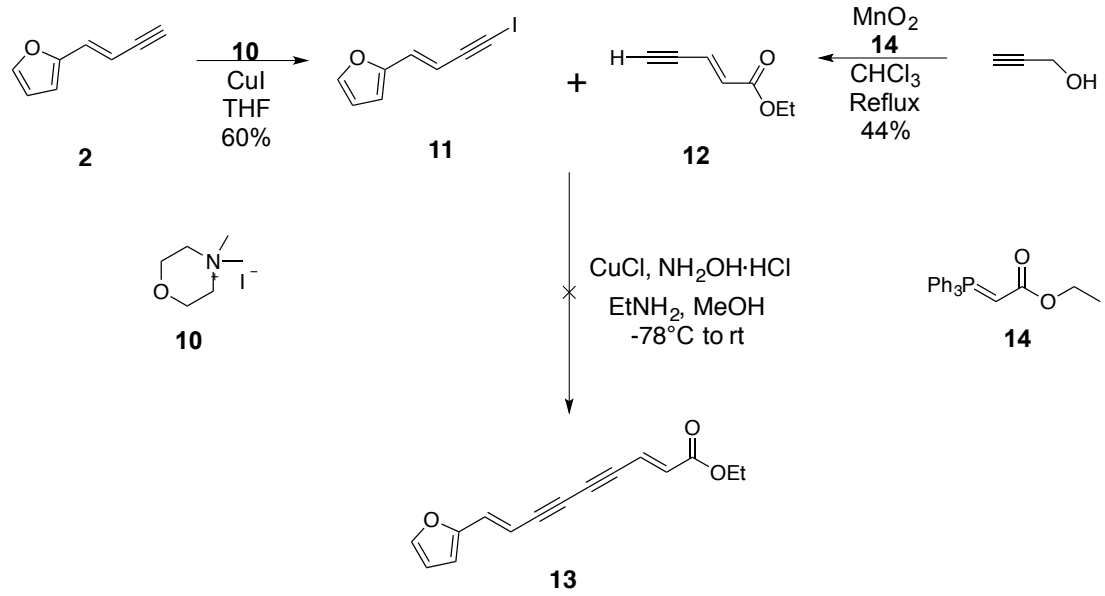
helped us to obtain intermediate **2** in a reasonable yield. The intermediate **2** played an important role in the whole project.

After successfully making enyne **2**, we focused our attention toward the other part of the target molecule. Starting from cis-2-butene-1,4-diol **5**, protection one hydoxyl group with with 1 equivalent of tert-butyldimethylsilyl chloride at 0 °C gave TBS ether **7**.[18] The terminal allylic alcohol was converted to α,β-unsuturated aldehyde **8** with oxidant pyridinium chlorochromate at room temperature in 75% yield. The condition of converting aldehyde **8** to dibromo TBS ether compound **9** was the same as the first step of Corey-Fuchs reaction, which was CBr_4 and PPh_3 at 0 °C. Treatment of compound **9** with TBAF in THF at 45 °C resulted in deprotection of the TBS group with F^-, as well as the elimiation of bromide with F^- working as the base to make bromo enyne alcohol **3** in 66% yield.[19]

After we had two coupling components in hand, we focused on finding the most suitable coupling condition. By utilizing a copper(I) catalyst with the apperence of ethyl amine and hydroxyamine hydrochloride in methanol, the Cadiot–Chodkiewicz coupling reaction of compounds **2** and **3** give us the desired natrual product **1** in 25% yield.[19] Although the yield was low for the final step, it was the only effective condition we found for this two- component coupling. To the best of our knowledge, no total synthesis route of 1-(E)-atractylodinol had been reported in literatures. Our synthetic route contains seven steps from two commercial available stating materials 3-(2-furyl)acrolein **4** and cis-2-butene-1,4-diol **5**, the total yield is 11%.

The in vitro test conducted by Alyssa Evans from the Carpenter group showed the percentage virus netralization (PVN) of compound **1** was 100%, which means that our

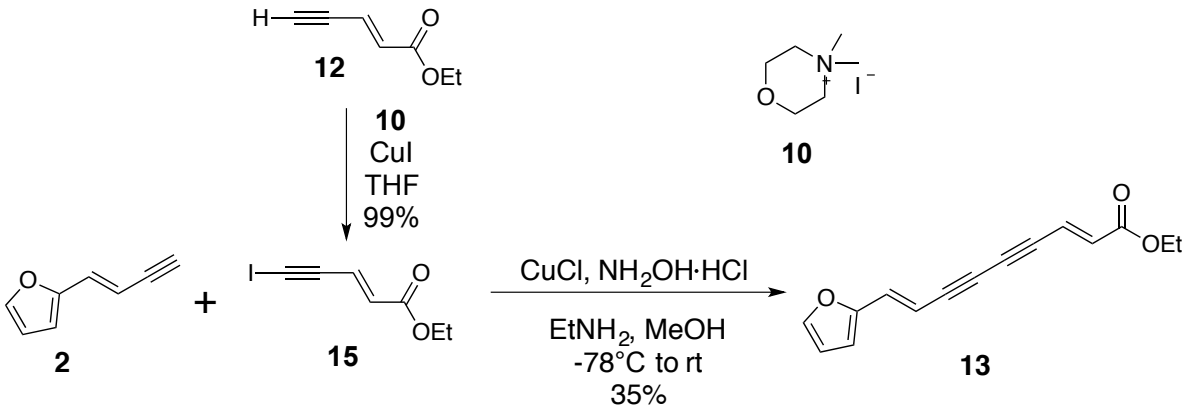
synthetic compound **1** is effective towards PRRSV as it was reported. However, the difficuty of operation and low yield of the coupling reaction at the final step made the synthetic route impractical for large scale production. To achieve our project goal, there were two options to choose. One was to develop a better synthetic route for desired natrual product **1**. The other one was to make natrual product analogs which have the same anti-PPRSV property as compound **1**. We decided to continue the project following two directions simultaneously.



Scheme 3. Original synthetic route of **13**.

To improve the synthetic route for natrual product **1**, as well as to make analog **13**, we designed a new route as shown in Scheme 3. From the prepared enyne **4**, the iodo enyne compound **11** was obtained with copper(I) catalyst and known morpholoine hydroiodide salt in 60% yield.[20, 21] Acetylene **13** was prepared from propargyl alcohol. Oxidation with manganese dioxide followed with Wittig reaction with

(carbethoxymethylene)-triphenylphosphorane **14** converted propargyl alcohol to compound **13** in one pot, the E isomer was isolated in 30% yield after flash column chromatography purification. Copper catalyzed Cadiot–Chodkiewicz coupling reaction was used to build C-C bond between iodoalkyne **11** and acetylene **12**. Unfortunately, the reaction did not go through as expected, ester analog **13** was not harvested after reaction and only the starting materials were recycled after the workup.



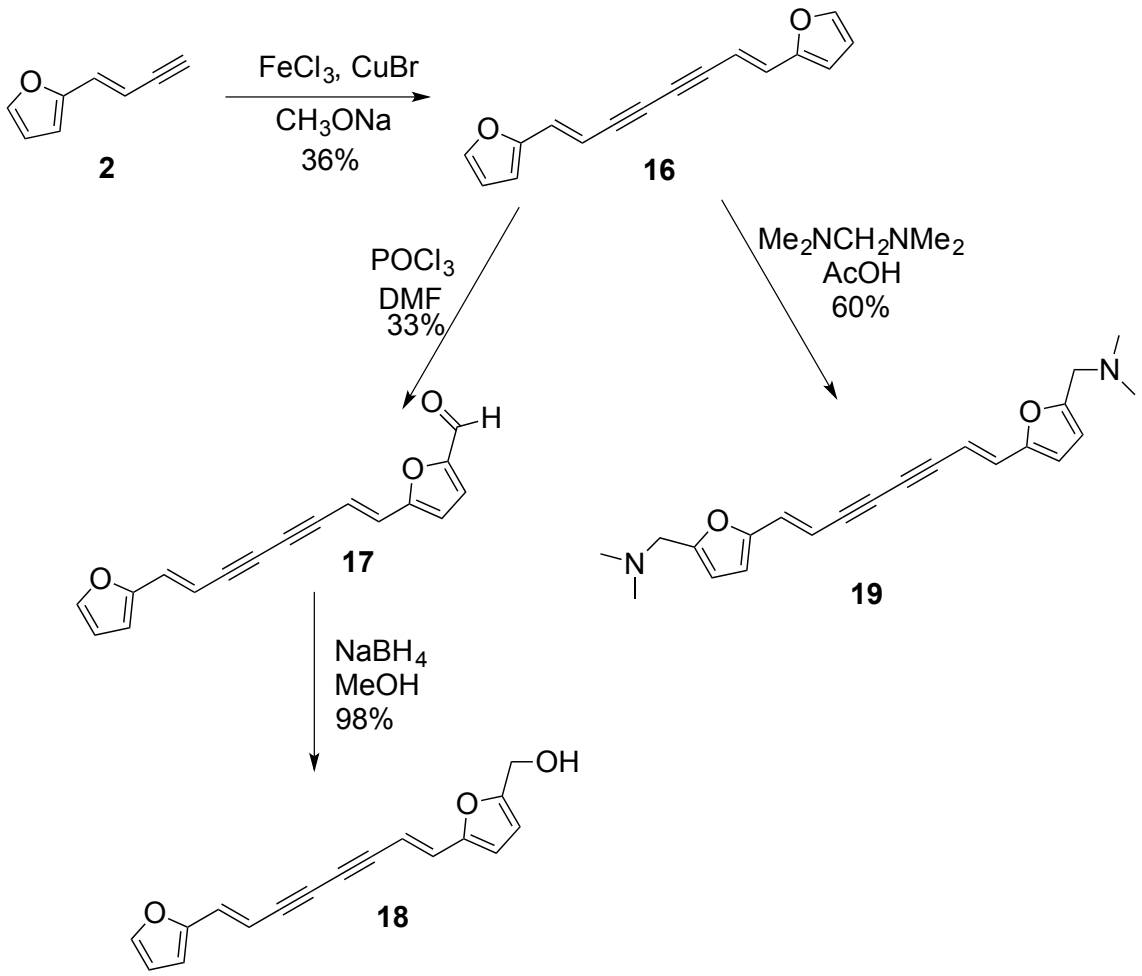
Scheme 4. Alternate synthetic route of **13**.

Due to the faliture of the original plan at the final stage, a new route was required to continue our project. We did not want to start from the beginning, so enyne **2** was kept in the alternate plan. Instead of a coupling reaction between furyl iodoalkyne **11** and ester **12**, iodoalkyne **15** and enyne **2** were carried on to the coupling reaction. Compound was made from terminal alkyne **12** with morpholine hydroiodide salt **10**, following the same procedure discribed before. The conversion yield was 99%, suprisingly high, compared to 66% yield for compound **11**. The same copper catalyzed coupling condition between **2** and **15** gave us the target molecule ester **13** in 35% yield.

It only took five steps to get compound **13** from commercially available materials. Compound **13** could be converted to natrual product **1** by one extra reduction step, which would give us a six-step synthetic route towards **1**. The shorter plan was appealing; however, the low yield of the coupling and difficulty of operation would still be a problem to produce large amounts of the desired product for animal testing. Meanwhile, the antiviral activity of compound **13** was also tested, it neutralized 99.3% of the PRRSV in vitro, which was not effective enough towards PRRSV due to the quick reproduction of virus. The research of synthetic route improvement stoppted at this point. We changed our focus to the synthesis of analogs with antiviral activity.

Starting from the previously prepared enyne **2**, homo-coupling reaction of termial alkyne **2** with iron(III) chloride and copper(I) bromide as co-catalyst gave us dimer **16** in 36% yield in Scheme 5.[22] To improve the yield of this reaction, we tried other possible conditions such as dimethyl formamide with copper(I) iodide as catalyst and tetrahydrofuran with nickel(II) chloride and copper(I) iodide as co-catalyst. However, all other conditions failed to deliver the dimer **16**. So we dicided to adopt this method to make dimer **16** despite the yield being as low as 36% percent. The in vitro test showed that compound **16** neutralized 99.3% of PRRS virus, a significant improvement from ester **13**, which was 95.6%. The result also showed that it was a valid plan to synthesize the anti-PRRSV natrual product analogs.

Although compound **16** neutrilized 95.6% of virus during in vitro test, it was still not an ideal antiviral drug because of the replication of virus. Afterwards, we put our focus mainly on the modification of compound **16** and development of new analogs. The plan of all the potential analogs are shown in schemes **5**, **6**, **7** and **8**.



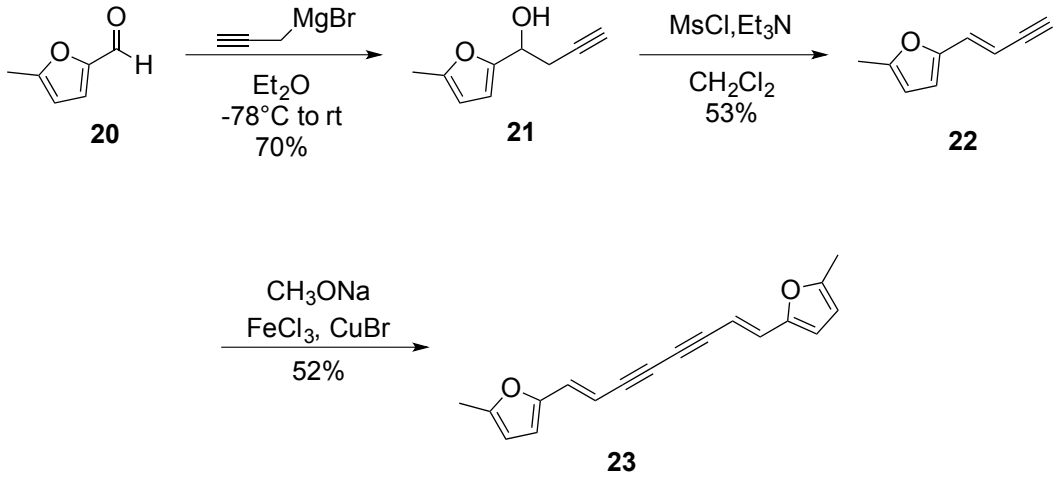
Scheme 5. Synthesis of dimers **16**, **17**, **18** and **19**.

Since dimer **16** showed good antiviral activity, our thought was to change the solubility of future analogs based on the structure of **16**. The in vitro test was performed in serum, in which the major component was water solution, we hoped that the better solubility in polar sovents would increase the antiviral property of our compounds. To improve the solubitity of analogs in water, we focused on installing polar functional groups, such as carbonyl groups, hydroxy groups and amino groups, on to the dimer **16**, a symmetric conjugated molecule.

In the beginning, our attention was foucused on installing an aldehyde on the furan ring. The well known Vilsmeier–Haack reaction was used for the transformation from dimer **16** to aldehyde **17** as seen in Scheme 5. The reaction of the dimethyl formamide with phosphorus oxychloride produces an electrophilic iminium cation, then the subsequent electrophilic aromatic substitution on dimer **16** produces an iminium ion intermediate, which is hydrolyzed to give the desired furyl aldehyde in 33% yield.[23, 24] To make the molecule more polar, the reduction reaction of aldehyde with $NaBH_4$ was applied to convert **17** to alcohol **18** in THF and methanol in 98% yield.[25] The in vitro test of aldehyde **17** and alcohol **18** showed that both compounds neutralized PRRSV effectively, the PVN is 96.6% and 98.6%, respectively. Although **17** and **18** would be useful against PRRSV potentially, the low yield of coupling step and formylation step limited the large scale production and application of these two drugs, besides the non-100% neutralization activity towards PRRSV.

Giving up the plan of installing aldehyde and hydroxyl groups was frustrating, but a novel plan attracted our attention. As shown in Scheme 5, instead of putting an aldehyde group, we attempted to add dimethyl amino group on the structure. Utiliziing bis(dimethylamino)methane in acetic acid gave us aminomethylation product **19** in 60% yield,[26] which neutralized 100% of virus in in vitro test as the natural product **1**. The synthetic route we developed for analog **19** contains only four steps from commerially available material 3-(2-furyl)acrolein **4**. Even though the yield for coupling step is only 36%, the simple operation with mild unpreotected condition makes large scale production possible. Moreover, we could easily convert compound **19** into hydrochloride salt or formic acid salt by treating with the corresponding acid. The salts would be hard to

decompose and much more stable to store at room temperature. The salt form also has better solubility compared to orgnic compound **19** when added into food as drug to feed animals. So far, we have successfully designed a scalable route to synthesize an effecive analog as anti-PRRSV drug, which makes animal tests possible.



Scheme 6. Synthesis of **23**.

With the success of antiviral drug **19**, we continued to design more analogs. Another idea was to put two formyl groups on the molecule, the attempt is shown in Scheme 6. Firstly, we tried to get dimer **23** from 5-methyl furfural **20**. Addition of freshly made propargylmagnesium bromide at -78 °C converted **20** into seconary alcohol **21** in 70% yield. Protection of alcohol with MsCl, followed by elimiation with triethylamine yielded enyne **22** in 53% yield as the mixture of cis and trans isomers. The previously used coupling condition was applied again to produce dimer **23** in 52% yield. The coupling yield was higher than previous applications, maybe due to the electron donating effect of the attached methyl groups on furan rings. Unfortunately, utilizing SeO_2 to

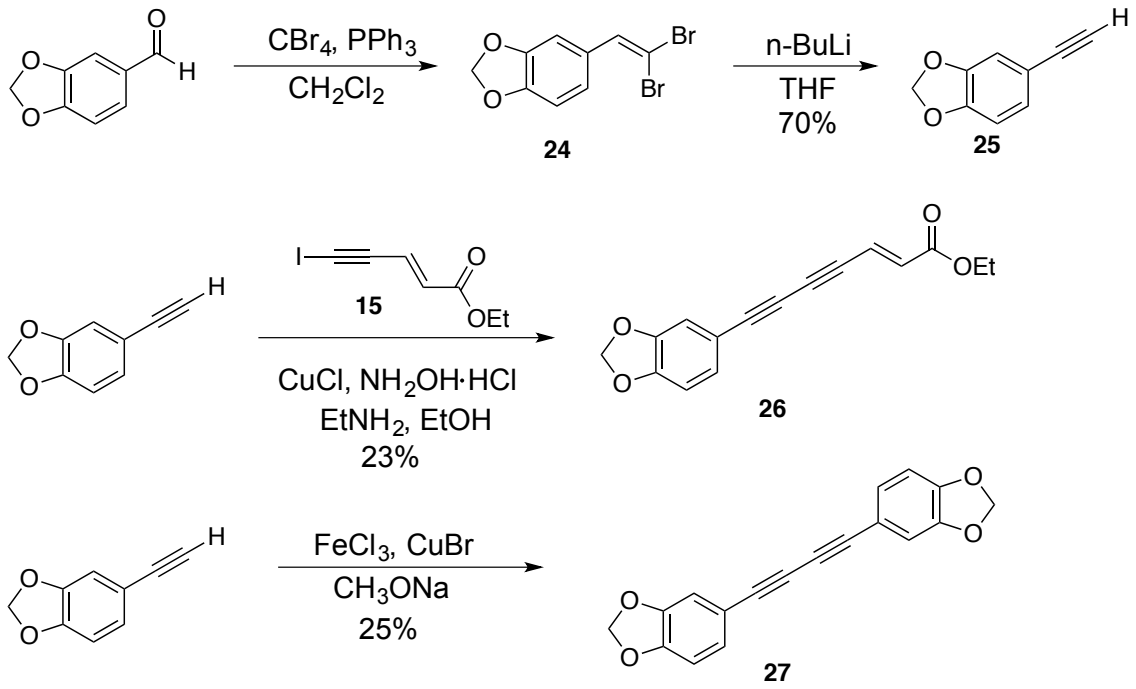
convert the terminal methyl groups into aldehyde groups falied. We could not make the dialdehyde compound as expected. However, the PVN value of compound **23** is 99%, which makes it as a good potential canditate for effective antiviral inhibitors.

A B

Scheme 7. Ring structure modification.

For further investigation, we intended to check whether the structrual stabilty of molecules would increase the antiviral property or not. As seen in Scheme 7, there are two double bonds in the furan ring and one double bond outside the ring structure, structure A on the left. The furan ring is not stable compared with a benzene ring, so we decided to put three double bonds together in one benzene ring to increase the structural stability. Moreover, the oxygen atom in furan ring was also kept in the structure B as the ether oxygen in the 5 member ring. Basically, we converted the furan ring structure A to a more stable benzene ring structure B keeping the same amount of double bonds and the oxygen atom. The synthetic route of target molecules ester **26** and dimer **27** are shown in Scheme 8. Both molecules started from the commercially available material piperonal. Corey-Fuchs reaction was used again to convert piperonal to termianl alkyne **25** in two steps, 70% yield. For **26,** the same conditon towards ester **13** was applied to give desired product. The iron/copper catalyzed terminal alkyne coupling condition was used to yield dimer **27** in 25% yield. The follwing in vitro test result showed that PVN value of

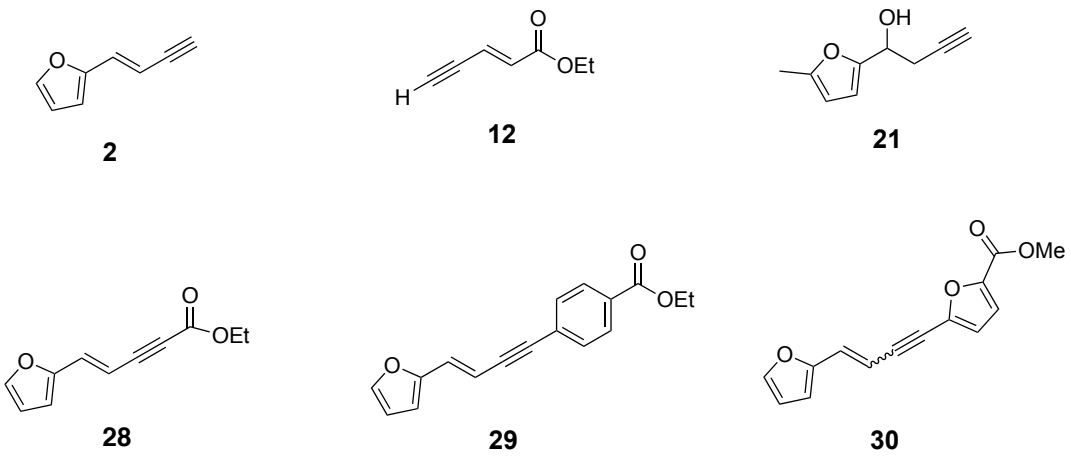
compound **26** and **27** were 56% and 11%, respectively, which indicated that neither of these two compounds would be effective to eliminate PRRSV. Moreover, the low PVN values also revealed that the stable benzene ring would not increase the effectiveness of analogs and the furan rings were one of the most crucial components for the antiviral propertity of our designed small molecule inhibitors.



Scheme 8. Synthesis of **26** and **27**.

Among all the privious synthesized molecules, we also found out that small simple molecules **2** and **12** showed anti-PRRSV activity. The PVN value of compound **2** was 93.4% and 95.6% for compound **13**. So we changed our focus on same small simple molecules which could be synthesized in less than four steps, and hoped that we could find a much more cost-efficient molecule inhibitor for PRRSV. The compounds we made

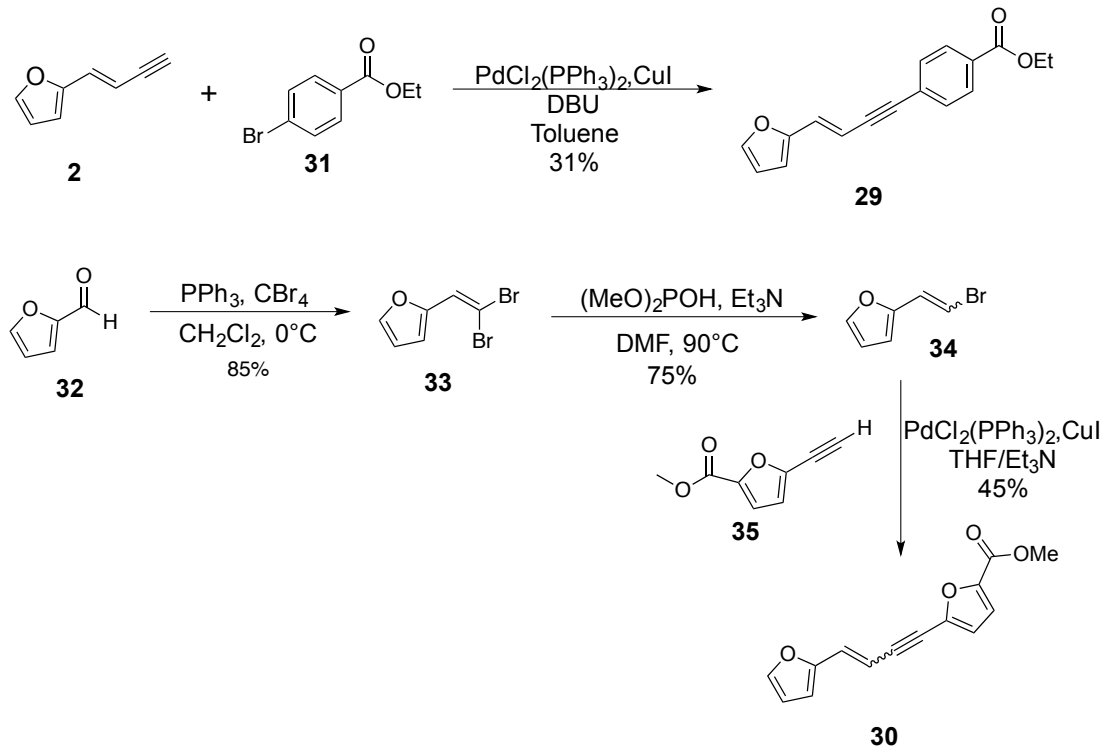
are ester **28**, **29** and **30**, beisdes secondary alcohol **21** which was an intermediate for compond **23**, as seen in Scheme 9.



Scheme 9. Other small molesules for biology test.

The synthetic methods towards compound **29** and **30** are exhibted in Scheme 10. The Pd and Cu catalyzed Sonogashira coupling reaction was performed between terminal alkyne **2** and aryl bromide **31** to build a carbon-carbon bond to yield ester **29** in 31% yield. For compound **30**, we started from furfural **32**, followed by the treatment of CBr_4 and PPh_3. Then the dibromo compound **33** was converted to bromo compound **34**, which was a mixture of cis and trans isomers, with dimethyl phosphite $(MeO)_2POH$ and triethyl amine (Et_3N) in DMF.[27] However, unlike compounds **2** and **12**, the new synthesized products **21**, **29** and **30** all had poor antiviral activity with PVN value below 65%. In addition, the most interesting result was that the PVN value of compound **28** was -24%, the only negative value among all the compounds. It means that compound **28** stimulated the replication of PRRSV, instead of killing the virus. At this point, none of our attempts to develop a more cost-effective antiviral drug compared to compound **19** was achieved.

For future animal test on mice and pigs, compound **19** was still the best designed analog for mass production.



Scheme 10. Synthesis of **29** and **30.**

1.3. Conclusions

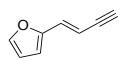
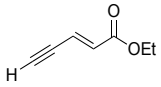
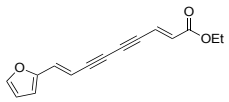
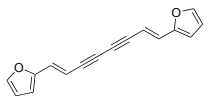
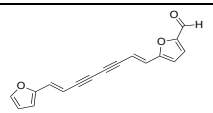
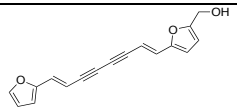
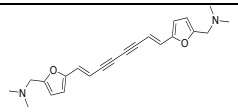
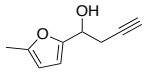
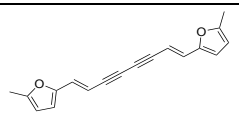
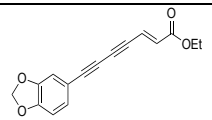
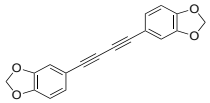
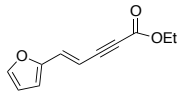
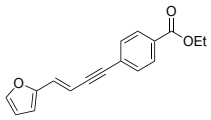
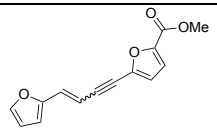
Natural 1-(E)-Atractylodinol **1** was successfully synthesized in seven steps and all the characterization data values (NMR and MS) of final product were identical to the literature values. One more efficient six-step synthetic route through ethyl ester analog **13** of compound **1** was also proposed. Even though the final reduction step was not carried out, there is evidence to support the feasibility of the reduction step. To the best of our knowledge, neither of these two efficient synthetic routes had been reported before.

Moreover, a series of analogs were synthesized based on the natural product **1**. We synthesized more than 15 molecules which were tested in vitro against PRRSV. The selective data of PVN is shown in Table 1. Among all the synthesized analogs, nine molecules showed more than 90% PVN value. Moreover, 10 μg of selected effective compounds could inactivate 10^3 to 10^5 infectious units of PRRSV and the EC_{50} values of effective compounds were 1-5 μg, respectively.

Compound **19** is the most cost efficent product, only four steps from comercially available starting material **4** in high yied. The development of compound **19** make the animal test feasible in the near future. We will focus on the large production of antiviral drug **19**, as well as the characterization method development for drug concentration level in serum.

In summary, we have sucessfully achieved our goal in development of the samll molecule inhibitors towards PRRSV. More tests including the animal test would be carried out in the future.

Table 1. Percent virus neutralization with 10 µg compound/200 FFU Virus

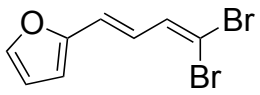
Number	Stucture	PVN (%)	Number	Stucture	PVN (%)	Number	Stucture	PVN (%)
1		100	2		93.4	12		95.6
13		99.3	16		95.6	17		96.6
18		98.6	19		100	21		42
23		99.0	26		56	27		11
28		-24	29		65.7	30		53

1.4. Experimental

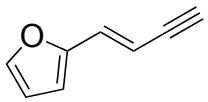
General Procedures

All starting materials were purchased from Sigma-Aldrich; THF was freshly distilled with LAH, other solvents were purchased from Fisher Scientific and used without further purification. All reactions were carried out in flame-dried glassware under argon with dry solvents under anhydrous conditions, unless specified. All yields refer to chromato-graphically isolated products. Reactions were monitored by thin- layer chromatography (TLC) carried out on 0.20 mm silica gel plates using UV light as a visualizing agent and potassium permanganate with heat as developing agents. Silica gel 60Å, particle size 0.032 – 0.063 mm, was used for flash column chromatography. ^{1}H and ^{13}C NMR spectra were acquired on a Varian VXR-300 or Bruker DRX-500 spectrometer. ^{1}H and ^{13}C chemical shifts (δ) are given in ppm relative to the residual protonated solvent peak ($CDCl_3$: $_{\delta}H$ = 7.26 ppm, $_{\delta}C$ = 77.0 ppm; CD_3OD: $_{\delta}H$ = 3.31 ppm, $_{\delta}C$ =49.0 ppm; $(CD_3)_2SO$: $_{\delta}H$ = 2.50 ppm, $_{\delta}C$ = 39.52 ppm; $(CD_3)_2CO$: $_{\delta}H$ = 2.05 ppm, $_{\delta}C$ = 29.84 ppm) as an internal reference. Low resolution mass spectra (LRMS) were recorded on an Agilent 6540 QTOF (quadrupole time of flight) mass spectrometer using ESI (electrospray ionization) or APCI (atmospheric-pressure chemical ionization), or EI (electron ionization) on an Agilent 6890 GC/MS.

Selected Experimental, Physical, and Spectral Data

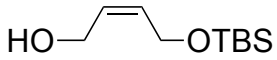


(*E*)-2-(4,4-Dibromobuta-1,3-dien-1-yl)furan (6) : To a solution of CBr_4 (2.6532 g, 8 mmol) in CH_2Cl_2 (50 mL) at 0 °C, PPh_3 (4.1966 g, 16 mmol) was added in portions over 3 minutes. The color of solution changed to dark brown. After adding, the mixture was stirred for 10 minutes under argon gas protection. Acrolein **4** (0.4885 g, 4 mmol) was added to the resulting solution over 5 minutes portionwise. After another 30 minutes stirring at 0 °C, TLC was checked to make sure the reaction was done. Evaporation of CH_2Cl_2, followed by a quick flash column chromatography (silica gel, EtOAc:hexanes 1:1) to remove most of salt gave a pale grey crude solid product. The purification of crude product by flash column chromatography (silica gel, EtOAc:hexanes 1:4) afforded **6** as a light yellow solid in 77% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.47–7.40 (m, 1H), 7.02 (dd, *J* = 10.4, 0.5 Hz, 1H), 6.68 (dd, *J* = 15.5, 10.4 Hz, 1H), 6.54–6.36 (m, 3H).

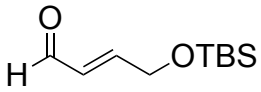


(*E*)-2-(But-1-en-3-yn-1-yl)furan (2): To a solution of dibromo compound **6** (0.5601 g, 2.02 mmol) in 15 mL THF, n-BuLi (2.5 M in Hexane solution, 1.62 mL, 4.03 mmol) was added dropwise over 10 minutes at -78 °C under argon protection. The resulting mixture stirred at -78 °C for 1 h, the the temperature was increased to room temparature. After 1 h stirring at room temperature, saturated NH_4Cl aqueous solution was added slowly to quench the reaction. The reaction mixture was extracted with diethyl ether (3 x 30 mL). The organic layers were collected, dried with anhydrous $MgSO_4$, filtered and

concentrated *in vacuo*. The residue was purified by flash column chromatography (silica gel, pentane) to afforded **2** as colorless oil in 86% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.38 (d, *J* = 1.8 Hz, 1H), 6.78 (d, *J* = 16.1 Hz, 1H), 6.45 – 6.31 (m, 2H), 6.03 (d, *J* = 18.4 Hz, 1H), 3.09 (d, *J* = 2.4 Hz, 1H). ^{13}C NMR (75 MHz, $CDCl_3$) δ = 151.98, 143.43, 130.34, 112.07, 110.68, 105.25, 83.04, 80.26.

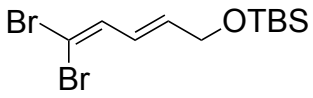


(*Z*)-4-((*Tert*-butyldimethylsilyl)oxy)but-2-en-1-ol (7): The diol **5** (0.4404 g, 5 mmol) was dissolved in anhydrous THF (20 mL), followed by addition of n-BuLi (2.5 M in Hexane solution, 2 mL, 5 mmol) at 0 °C over 5 minutes. After 1 h stirring at 0 °C, 5 mL THF solution of TBSCl (0.7536 g, 5 mmol) was added dropwise over 3 minutes. The resulting mixture was allowed to stir at room temperature for overnight. The reaction ws quenched by adding saturated NH_4Cl aqueous solution, followed by extraction with diethyl ether (2 x 20 mL). The organic layers was combined, dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give crude pruduct **7** in 96% yield. The crude product was used for next step without future purification; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 5.78 – 5.61 (m, 2H), 4.31–4.16 (m, 4H), 0.91 (d, *J* = 1.4 Hz, 9H), 0.09 (d, *J* = 2.8 Hz, 3H).

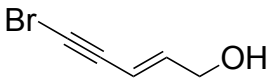


(*E*)-4-((*Tert*-butyldimethylsilyl)oxy)but-2-enal (8): Alcohol **7** (0.8201 g, 4.08 mmol) was added to a flame-dried flask with CH_2Cl_2 (15 mL) and molecular sieve (1 g). PCC

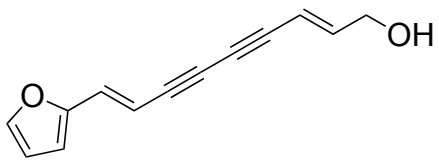
(0.9062 g, 4.21 mmol) was added in one portion at 0 °C, the solution color changed to orange. After overnight stirring at room temperature, the solvent was evaporated *in vacuo*, diluted with diethyl ether. The resulting suspension was filtered through a short silica gel column to remove solid. The residue was purified by flash column chromatography (silica gel, EtOAc:hexanes 1:19) to afforded **8** in 75% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 9.60 (d, *J* = 8.1 Hz, 1H), 6.89 (dt, *J* = 15.4, 3.3 Hz, 1H), 6.48–6.33 (m, 1H), 4.45 (dd, *J* = 3.3, 2.2 Hz, 2H), 0.92 (s, 9H), 0.09 (d, *J* = 1.6 Hz, 3H).



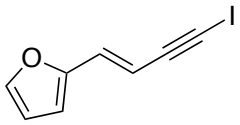
(*E*)-*Tert*-butyl((5,5-dibromopenta-2,4-dien-1-yl)oxy)dimethylsilane (9): Compound **9** was prepared by using previous described procedure for compound **6** in 96% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.96 (d, *J* = 10.3 Hz, 1H), 6.42–6.26 (m, 1H), 6.14–6.00 (m, 1H), 4.00 (d, *J* = 7.8 Hz, 2H), 0.91 (s, 9H), 0.08 (d, *J* = 7.5 Hz, 3H).



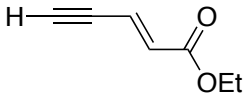
(*E*)-5-Bromopent-2-en-4-yn-1-ol (3): To a solution of the dibromoalkene obtained above **9** (0.3832 g, 1.08 mmol) in THF was added TBAF (1.0 M solution in THF, 0.4 mL, 0.4 mmol) at room temperature, then the mixture was stirred at 45 °C for 18 h and diluted with Et_2O. The mixture was washed with saturated aqueous NH_4Cl, H_2O, and brine and then dried over $MgSO_4$. Concentration gave the mixture of the corresponding TBS- deprotected bromoacetylene **3**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.38–6.23 (m, 1H), 5.72 (dt, *J* = 15.9, 1.9 Hz, 1H), 4.20 (dd, *J* = 5.0, 1.9 Hz, 2H).



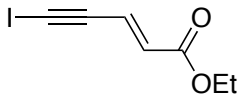
1-(E)-Atractylodinol (1): To a solution of $EtNH_2$ (70% aqueous solution, 2.2 mL) in MeOH (3 mL) was added CuCl (13.0 mg, 131.4 μmol) at room temperature that resulted in the formation of a blue solution. To the resulting mixture was added $NH_2OH{\cdot}HCl$ (54.8 mg, 0.788 mmol) at room temperature to discharge the blue color. The resulting colorless solution indicated the presence of Cu(I) salt. To the resulting mixture was added **2** (108.6 mg, 0.9196 mmol) in MeOH (2 mL) at room temperature, and the mixture was stirred at the same temperature for 10 min that resulted in the formation of a yellow suspension. To the resulting mixture was added bromoacetylene **3** (42.3 mg, 0.2627 mmol) in MeOH (2 mL) at −78 °C, and mixture was stirred at the same temperature for 30 min. The mixture was allowed to warm to room temperature for 3 h. The mixture was diluted with Et_2O, washed with H_2O and brine, and then dried over $MgSO_4$. Concentration and flash column chromatography (silica gel, EtOAc:hexanes 1:4) gave the corresponding product **1**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.39 (s, 1H), 6.81 (d, *J* = 16.0 Hz, 1H), 6.41 (dt, *J* = 11.1, 4.5 Hz, 3H), 6.11 (d, *J* = 15.9 Hz, 1H), 5.88 (d, *J* = 15.9 Hz, 1H), 4.26 (d, *J* = 3.3 Hz, 2H); ^{13}C NMR (75 MHz, $CDCl_3$) δ = 152.03, 145.44, 143.85, 131.30, 112.36, 111.55, 109.28, 104.83, 81.34, 81.09, 77.10, 75.03, 63.00.



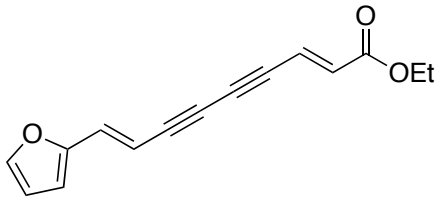
(*E*)-2-(4-Iodobut-1-en-3-yn-1-yl)furan (11): To 10 mL THF was added enyne **2** (0.1386 g, 1.17 mmol), morpholine hydroiodide salt (0.4787 g, 1.41 mmol) and CuI (0.0112 g, 0.06 mmol) at room temperature. The reaction was monitored with TLC until no starting material left. Saturated aqueous NH_4Cl solution was added to quench reaction, followed by the extraction with diethyl ether (3 x 15 mL). The combined organic layer was washed with saturated aqueous $Na_2S_2O_3$, dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo*; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.37 (d, *J* = 1.8 Hz, 1H), 6.72 (d, *J* = 16.0 Hz, 1H), 6.45 – 6.31 (m, 2H), 6.17 (d, *J* = 16.0 Hz, 1H).



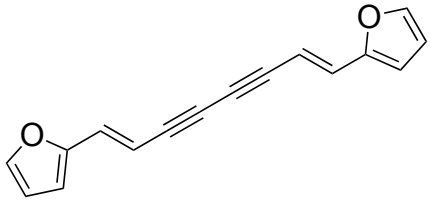
Ethyl (*E*)-pent-2-en-4-ynoate (12): To a flask with MnO_2 (4.3470 g, 50 mmol), **14** (2.0902 g, 6 mmol) in chloroform was added propargyl alcohol (0.2803 g, 5 mmol). The resulting mixture was refluxed for 18 h, and monitored with TLC. After reaction was finished, the mixture was filtered through celite, washed with CH_2Cl_2 and concentrated *in vacuo*. Diluted with pentane, filtered after 30 minutes stirring gave crude product. Flash column chromatography (silica gel, EtOAc:hexanes 1: 99) purification gave **12** in 45% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.25 – 6.06 (m, 2H), 4.24 (q, *J* = 7.2 Hz, 2H), 3.60 (dd, *J* = 2.5, 0.8 Hz, 1H), 1.31 (d, *J* = 14.3 Hz, 3H).



Ethyl (*E*)-5-iodopent-2-en-4-ynoate (15): The same procedure as compound **11** gave **15** in 99% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.25 – 6.06 (m, 2H), 4.24 (q, *J* = 7.2 Hz, 2H), 3.60 (dd, *J* = 2.5, 0.8 Hz, 1H), 1.31 (d, *J* = 14.3 Hz, 3H).

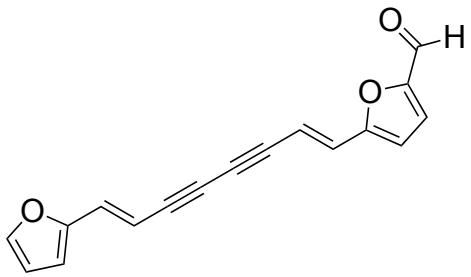


Ester (13): Compound **13** was prepared by using previous described procedure for compound **1** in 35% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.41 (s, 1H), 6.93–6.77 (m, 2H), 6.44 (d, *J* = 1.7 Hz, 2H), 6.32 (dd, *J* = 15.8, 2.7 Hz, 1H), 6.13 (d, *J* = 16.0 Hz, 1H), 4.22 (q, *J* = 7.1 Hz, 2H), 1.30 (t, *J* = 7.2 Hz, 3H).

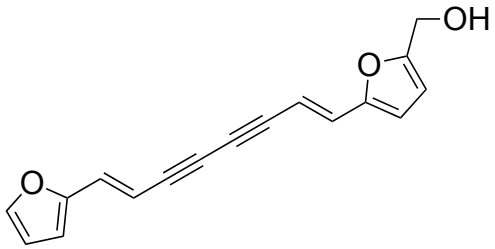


(1*E*,7*E*)-1,8-Di(furan-2-yl)octa-1,7-dien-3,5-diyne (16): Enyne **2** (0.1226 g, 1.038 mmol), $FeCl_3$ (16.8 mg, 0.1038mmol), CuBr (7.5 mg, 0.052 mmol) and CH_3ONa (162 mg, 3 mmol) were added to 5 mL of THF under ambient temperature in the open air. The reaction mixture was stirred under room temperature for 20 h. The progress of the reaction was monitored by TLC. After completion of the reaction, the solvent was

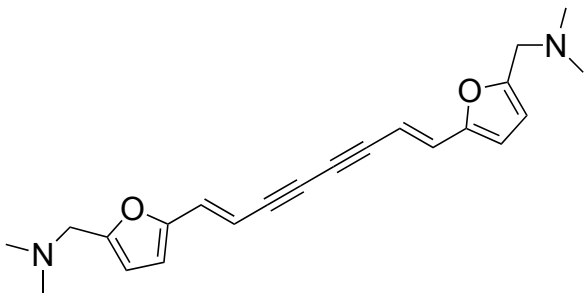
removed under reduced pressure and the crude product obtained was purified by column chromatography over silica gel to afford **16** in 36% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.40 (d, *J* = 1.8 Hz, 2H), 6.82 (d, *J* = 15.8 Hz, 2H), 6.47–6.35 (m, 4H), 6.15 (d, *J* = 15.8 Hz, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ = 151.92, 143.65, 130.93, 112.18, 111.32, 104.80, 82.54, 77.31.



5-((1*E*,7*E*)-8-(Furan-2-yl)octa-1,7-dien-3,5-diyn-1-yl)furan-2-carbaldehyde (17): To a DMF (2 mL) solution of Dimer **16** was added $POCl_3$ slowly at 0 °C. The resulting mixture was allowed to stir at 0 °C for 1 h, room temperature for 1 h, then 80 °C for 10 minute. When the reaction cooled down, aqueous NaOAc solution was added, followed by the extraction with CH_2Cl_2 (3 x 15 mL). Organic layer was washed with water, brine, dried over anhydrous $MgSO_4$. Concentration and flash column chromatography (silica gel, EtOAc:hexanes 4:1) gave compound **17** in 33% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 9.61 (s, 1H), 7.41 (s, 1H), 7.23 (d, *J* = 3.7 Hz, 1H), 6.84 (dd, *J* = 15.9, 3.1 Hz, 2H), 6.59–6.50 (m, 2H), 6.50–6.39 (m, 2H), 6.15 (d, *J* = 15.9 Hz, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ = 177.37, 156.51, 152.41, 151.78, 143.89, 131.58, 129.33, 122.86, 112.63, 112.28, 111.80, 111.35, 104.35, 84.34, 81.32, 80.44, 76.93.

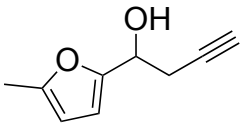


(5-((1*E*,7*E*)-8-(Furan-2-yl)octa-1,7-dien-3,5-diyn-1-yl)furan-2-yl)methanol (18): To a flask with aldehyde **17** (10 mg, 0.038 mmol) and THF:Methnol (1:1) solution was added $NaBH_4$ portionwise. After stirring at room temperature for 2 h, the solvent was evaporated under reduced pressure. The residue was extracted with H_2O (5 mL) and EtOAc (3 x 10 mL). Organic layer was dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give crude pruduct. **18** was obtained by flash column chromatography purification over silica gel (EtOAc:hexanes 1:4-1:2); ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.40 (s, 1H), 6.88 – 6.72 (m, 2H), 6.47 – 6.28 (m, 4H), 6.15 (d, J = 15.9 Hz, 2H), 4.61 (s, 2H).



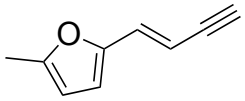
1,1'-(((1*E*,7*E*)-Octa-1,7-dien-3,5-diyne-1,8-diyl)bis(furan-5,2-diyl))bis(*N*,*N*-dimethylmethanamine) (19).: To a solution of **13** (0.1287 g, 0.549 mmol) in glacial AcOH (2 mL) cooled to below 10 °C as added portionwise, bis(dimethlyamino)methane (0.1684 g, 0.549 mmol), while the temperature was maintained at about 12 °C. The

solution was then allowed to rise to room temperature and left to stand overnight. The orange solution was poured onto crushed ice and stirred while K_2CO_3 (5 g) was added in portions, followed by extraction with CH_2Cl_2, (3 x 15 mL). Extracts were combined and back-extracted into 2% aqueous HCl (3 x 20 mL). Reneutralization to pH 10.5 with K_2CO_3, followed by extraction with CH_2Cl_2, drying $MgSO_4$, and concentration gave **19** in 60% yield; ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.74 (d, *J* = 15.9 Hz, 2H), 6.30 (d, *J* = 3.3 Hz, 2H), 6.22 (d, *J* = 3.3 Hz, 2H), 6.11 (d, *J* = 15.9 Hz, 2H), 3.44 (s, 4H), 2.25 (s, 12H). ^{13}C NMR (126 MHz, $CDCl_3$) δ = 154.07, 151.61, 130.81, 112.02, 111.16, 104.39, 82.69, 77.31, 55.92, 45.09.

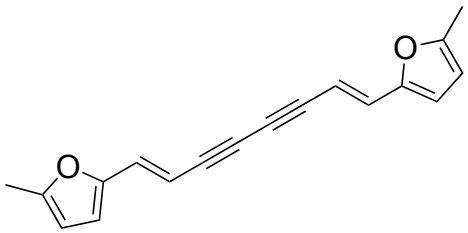


1-(5-Methylfuran-2-yl)but-3-yn-1-ol (21): To a three-neck flask with 10 mL of anhydrous diethyl ether and Mg turning (0.0576 g, 2.4 mmol) was added propargyl bromide (0.2 mL, 2.4 mmol) dropwise. The reflux was kept by slow addition of propargyl bromide. After addition, the reaction was allowed to cool down to room temperature slowly in 2 h. Freshly prepared propargylmagnesium bromide was transfer to another flask with 5-methylfurfural (0.2 mL, 2 mmol) in diethyl ether solution (5 mL) by cannel at -78 °C. The reaction was quenched by adding saturated NH_4Cl aqueous solution after 2 h, followed by extraction with diethyl ether (3 x 20 mL). The organic layers was combined, dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give crude pruduct **21** in 70% yield, which was used without further purification; 1H NMR (300

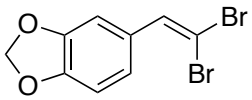
MHz, $CDCl_3$) δ = 6.21 (d, *J* = 3.1 Hz, 1H), 5.96–5.82 (m, 1H), 4.82 (t, *J* = 6.3 Hz, 1H), 2.81–2.71 (m, 2H), 2.37–2.25 (m, 3H), 2.07 (t, *J* = 2.7 Hz, 1H).



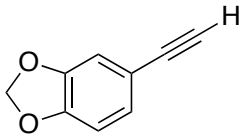
(*E*)-2-(But-1-en-3-yn-1-yl)-5-methylfuran (22): To a solution of alcohol **21** (0.1521g, 1.0 mmol) and Et3N (0.42 mL, 3.0 mmol) in CH_2Cl_2 (10 mL) was added MsCl (0.10 mL, 1.3 mmol) at 0 °C. The reaction was monitored by TLC after 2 h. Saturated NH_4Cl aqueous solution was added once the reaction completion, followed by extraction with diethyl ether (3 x 20 mL). The organic layers was combined, dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give crude pruduct **22** in 55% yield, which was mixture of cis and trans isomers; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.00 (d, *J* = 3.3 Hz, 0.37H), 6.71 (d, *J* = 16.0 Hz, 0.62H), 6.60 (d, *J* = 11.9 Hz, 0.38H), 6.24 (d, *J* = 3.2 Hz, 0.60H), 6.09–5.71 (m, 1.64H), 5.40 (dd, *J* = 11.9, 2.6 Hz, 0.37H), 5.30 (s, 0.35H), 3.44 (s, 0.34H), 3.06 (d, *J* = 2.5 Hz, 0.57H), 2.32 (d, *J* = 3.1 Hz, 3H)



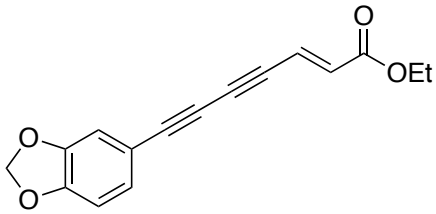
(1*E*,7*E*)-1,8-Bis(5-methylfuran-2-yl)octa-1,7-dien-3,5-diyne (23): The compound **23** was obtained according previous coupling procedure for compound **16**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.98 (d, *J* = 3.4 Hz, 1H), 6.75 (dd, *J* = 15.8, 10.2 Hz, 1H), 6.64 (dd, *J* = 11.8, 7.4 Hz, 1H), 6.28 (t, *J* = 4.0 Hz, 1H), 6.15–6.00 (m, 3H), 5.52 (dd, *J* = 11.8, 9.4 Hz, 1H), 2.38–2.29 (m, 6H).



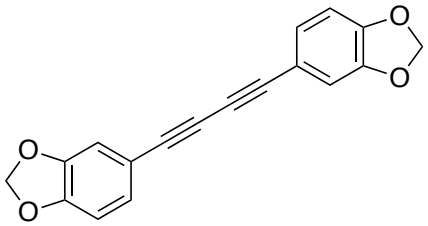
5-(2,2-Dibromovinyl)benzo[1,3]dioxole (24): The compound **24** was obtained according previous described procedure of compound **6**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.37 (d, *J* = 0.7 Hz, 1H), 7.19 (d, *J* = 1.8 Hz, 1H), 7.00–6.90 (m, 1H), 6.80 (d, *J* = 8.1 Hz, 1H), 5.99 (s, 2H).



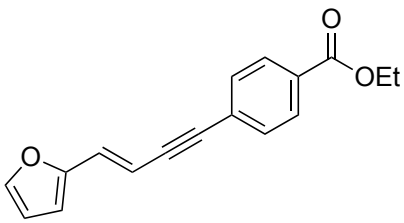
5-Ethynylbenzo[1,3]dioxole (25): The compound **25** was obtained according previous described procedure of compound **2**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.07 – 6.98 (m, 1H), 6.93 (d, *J* = 1.6 Hz, 1H), 6.75 (dd, *J* = 8.0, 0.4 Hz, 1H), 5.98 (s, 2H), 2.97 (s, 1H).



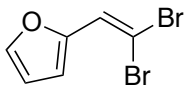
Ethyl (*E*)-7-(benzo[*d*][1,3]dioxol-5-yl)hepta-2-en-4,6-diynoate (26): The compound **26** was obtained according previous described procedure of compound **1**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.07 (dd, *J* = 8.2, 1.6 Hz, 1H), 6.93 (d, *J* = 1.6 Hz, 1H), 6.89–6.72 (m, 2H), 6.33 (d, *J* = 15.8 Hz, 1H), 6.00 (s, 2H), 4.23 (q, *J* = 7.2 Hz, 2H), 1.30 (t, *J* = 7.1 Hz, 3H).



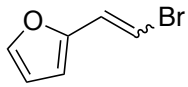
1,4-Bis(benzo[*d*][1,3]dioxol-5-yl)buta-1,3-diyne (27): The compound **27** was obtained according previous described procedure of compound **16**; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.06 (dd, *J* = 8.1, 1.6 Hz, 2H), 6.94 (d, *J* = 1.6 Hz, 2H), 6.76 (d, *J* = 8.0 Hz, 2H), 5.99 (s, 4H).



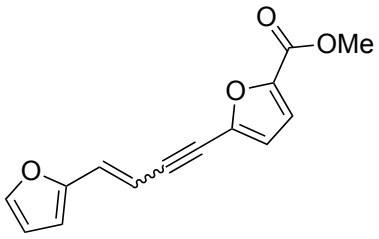
Ethyl (*E*)-4-(4-(furan-2-yl)but-3-en-1-yn-1-yl)benzoate (29): To a mixture of enyne **2** (0.0591 g, 0.5 mmol), ester **31** (0.0764 g, 0.33 mmol), $PdCl_2(PPh_3)_2$ (12.0 mg, 16.5 μmol) and CuI (3.3 mg, 16.5 μmol) in toluene was added DBU (0.1 mL). The solution color changed from yellow to dark brown after addition. The resulting mixture was allowed to stir at room temperature, under argon, for 24 h, then filtered and concentrated *in vacuo*. **29** was obtained by flash column chromatography purification over silica gel (EtOAc:hexanes 1:9); ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.04–7.95 (m, 2H), 7.55–7.45 (m, 2H), 7.41 (d, *J* = 1.8 Hz, 1H), 6.83 (d, *J* = 16.0 Hz, 1H), 6.48–6.35 (m, 2H), 6.28 (d, *J* = 16.0 Hz, 1H), 4.38 (q, *J* = 7.1 Hz, 2H), 1.40 (t, *J* = 7.1 Hz, 3H).



2-(2,2-Dibromovinyl)furan (33): The compound **33** was obtained according previous described procedure of compound **6**; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.47 – 7.37 (m, 2H), 6.95 (d, *J* = 3.5 Hz, 1H), 6.46 (dd, *J* = 3.5, 1.8 Hz, 1H).



2-(2-Bromovinyl)furan (34): To a solution of dibromo **33** (1.8675 g, 7.4 mmol) in DMF was added $(MeO)_2POH$ (3.2634 g, 29.7 mmol) and Et_3N (3.3757 g, 33.4 mmol). The resulting solution was heated to reflux for 20 h. After reaction completion, the mixture was quenched with a saturated aqueous solution of NH_4Cl and extracted with ethyl acetate (3 x 10 mL). The organic extracts were dried over $MgSO_4$, and concentrated *in vacuo*. The residue was purified by flash column chromatography giving compound **34**.



Methyl 5-(4-(furan-2-yl)but-3-en-1-yn-1-yl)furan-2-carboxylate (35): A solution of alkyne **35** (0.0364 g, 0.242mmol) in THF (2 mL) was added at room temperature, under argon, to a stirred mixture of **34** (0.0381 g, 0.221 mmol), $PdCl_2(PPh_3)_2$ (4.3 mg, 6 μmol), CuI (2.3 mg, 12.1 μmol), and Et_3N (50μL, 36.3 μmol) in THF (2 mL). After reaction completion (1 h), the mixture was quenched with a saturated aqueous solution of NH_4Cl

and extracted with ethyl acetate (3 x 10 mL). The organic extracts were washed with a saturated aqueous solution of NaCl, dried over $MgSO_4$, and concentrated *in vacuo*. The residue was purified by flash column chromatography leading to compound **35** in 45% yield.:

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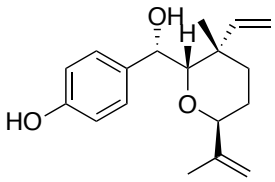
CHAPTER 2.

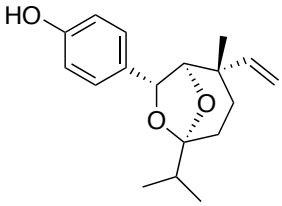
SYNTHETIC APPROACH TO THE PSORACORYLIFOLS*

2.1. Introduction

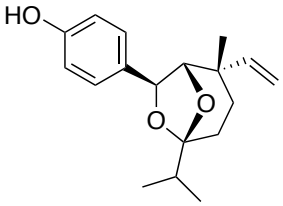
Psoracorylifols A–C (**1**–**3** in Figure 1) were isolated from the seeds of *Psoralea corylifolia* L. (Fabaceae), a well-known traditional Chinese medicine, which has been applied to cure gynecological bleeding, vitiligo and psoriasis.[1] In 2006, Yue and co-workers identified and characterized these molecules.[2] They reported that psoracorylifols A–C showed potent inhibitory activity against two strains of *Helicobacter pylori* (SS1 and ATCC 43504) with MICs of 25.0, 12.5 and 12.4 μg/mL, respectively.[2] Compounds **2** and **3** were especially effective against *H. pylori*-ATCC 43504, a drug resistant strain. These compounds exhibited activities ten times stronger than metronidazole, which is a critical ingredient for combination therapies of *H. pylori* infections.[3]



Psoracorylifol A
1



Psoracorylifol B
2



Psoracorylifol C
3

Figure 1. Structures of psoracorylifols A (**1**), B (**2**), and C (**3**).

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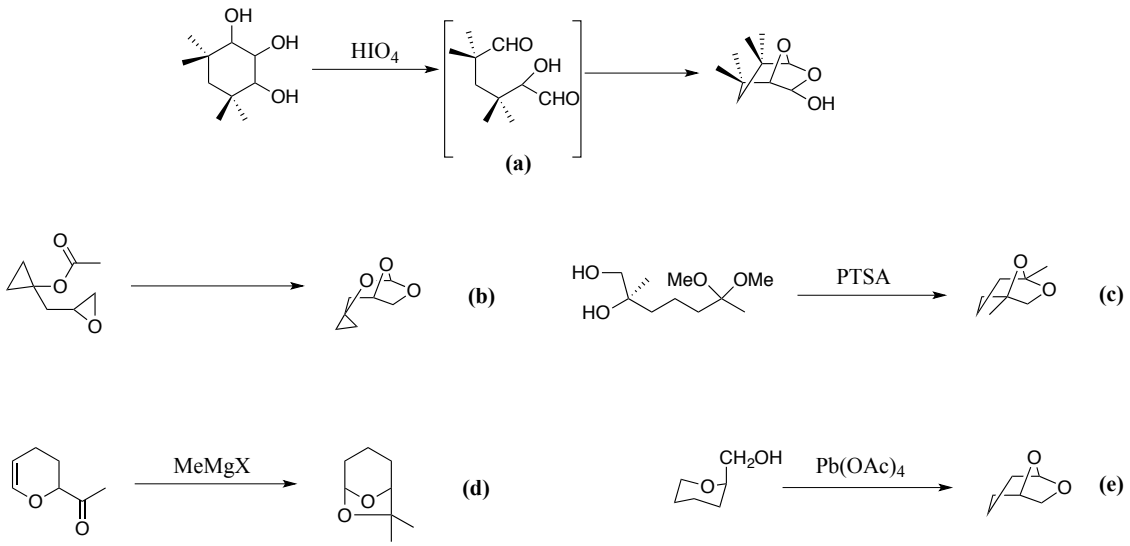
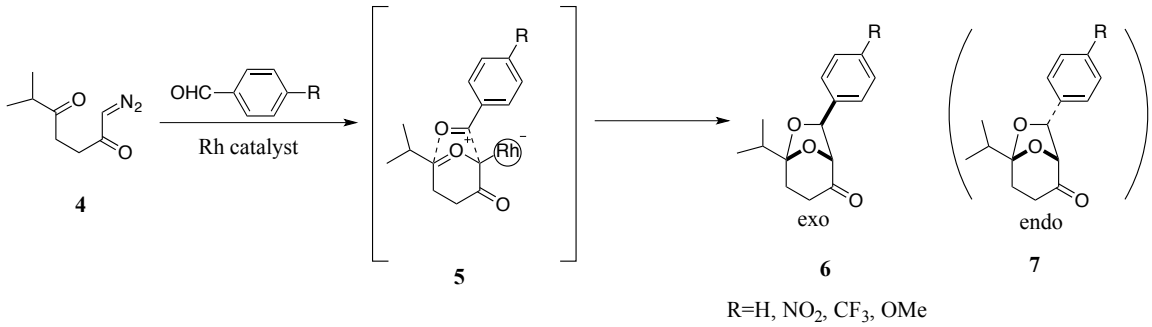
The 6,8-dioxabicyclo[3.2.1]octane skeleton is a common structural subunit which can be found in many biologically active natural products.[4] The selected synthetic routes to build such bicyclic ketals are shown in Scheme 1. Periodic acid cleavage of the cyclohexane triol gives the bicyclic ketal through a dialdehyde intermediate, as shown in Scheme 1(a).[5] The structure can also be derived from an epoxide in high yield, which Wasserman and co-workers discovered as part of a study on the rearrangement of carbonyl-epoxides, as shown in Scheme 1(b).[6] Moreover, this unique bicyclic skeleton can be prepared from a diol and a ketone or a diol and an aldehyde, which is made by deprotection of the carbonyl group or the hydroxyl groups in situ, as illustrated in Scheme 1(c).[7] In Scheme 1(d), an interesting method is shown to build a similar unit. Grignard and organolithium reagents attack the ketone and the desired structure forms after cyclization.[8] Cyclization of a pyranyl carbinol with lead tetraacetate via a radical mechanism has been reported to form the 6,8-dioxabicyclo[3.2.1]octane skeleton in low yields, as seen in Scheme 1(e).[9]



Scheme 1. Some synthetic routes to bicyclic ketals.

Recently, Hashimoto and coworkers reported a synthesis of the core skeleton for the psoracorylifols, as shown in Scheme 2.[10]They developed a novel catalytic asymmetric method to construct the exo-7-aryl-6,8-dioxabicyclo[3.2.1]octane framework of psoracorylifols B (**2**) and C (**3**). The exo- and enantioselective cycloadditions of a six-membered carbonyl ylide derived from 1-diazo-6-methyl-2,5-heptanedione (**4**) with aromatic aldehydes under the catalysis of chiral dirhodium(II) catalyst, have been achieved with high levels of asymmetric induction (up to 87% ee) by the use of electron-rich aromatic aldehydes. Although this method was able to generate the desired framework unit, it comes with some practical challenges. For example, the complete removal of transition-metal catalysts is important since the presence of metals could affect the bioactivity evaluation process. Furthermore, the preparation and strict operational conditions of the catalyst limit the industrial application.

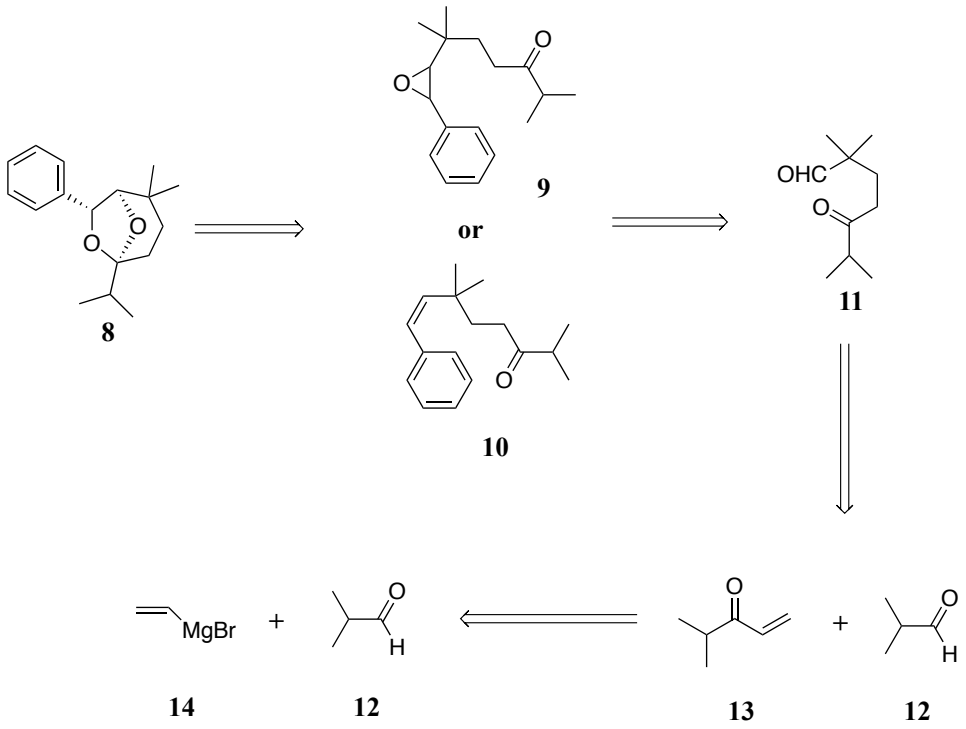


Scheme 2. Core structure construction of psoracorylifols B and C.

2.2. Results and Discussion

Our goal was to develop a more direct and strategically different synthetic route to psoracorylifol analogs that could be readily extendable to single enantiomer synthesis.

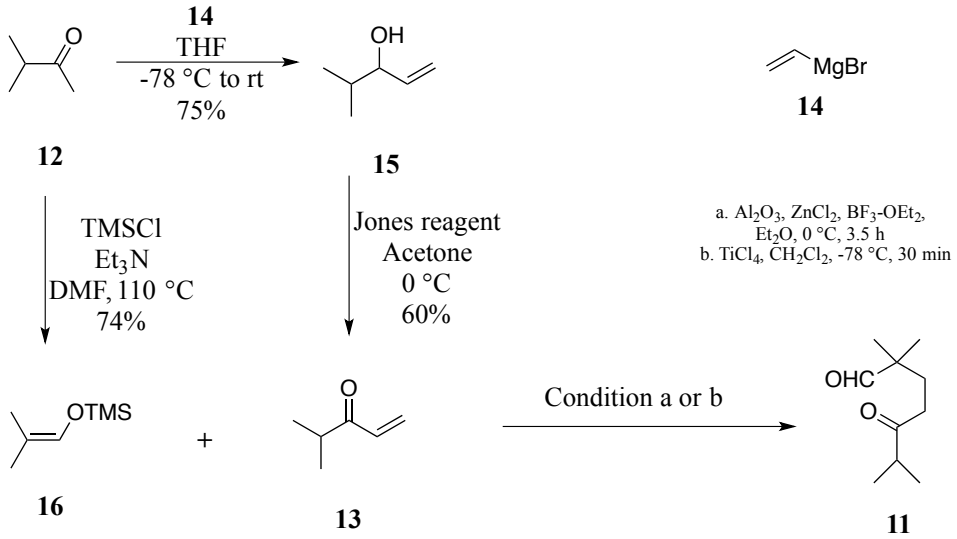
In order to identify an efficient synthetic pathway, we initially explored the strategy, as shown in Scheme 3. We chose to use ketal formation as a key reaction to build such a skeleton. Compound **8** was chosen as the targeted model system based on the framework of psoracorylifols. The 6,8-dioxabicyclo[3.2.1]octane skeleton could be constructed by cyclization after epoxide ring opening in compound **9** or dihydroxylation of the double bond in compound **10**. Both **9** and **10** would be converted from a keto aldehyde, which is prepared using the Michael addition of the enol silyl ether of isobutyraldehyde **12** with isopropyl vinyl ketone **13**.



Scheme 3. Retrosynthesis of **8**.

Starting with isobutyraldehyde **12**, addition of vinylmagnesium bromide **14** at -78 °C gave the secondary allylic alcohol **15** in 75% yield, as seen in Scheme 4. A Jones

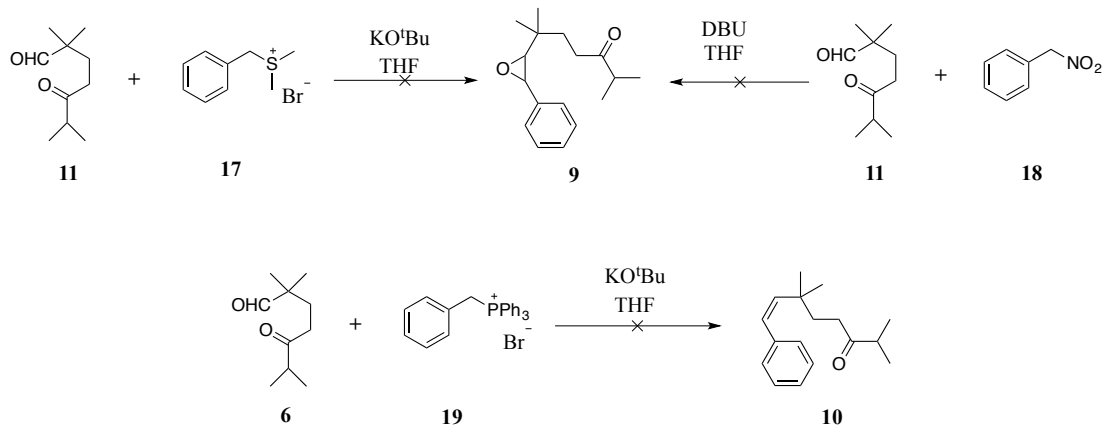
oxidation converted the secondary allylic alcohol **15** to a α,β-unsuturated ketone **13** in 60% yield at 0 °C.[11] After the preparation of compound **13**, enol silyl ether **16** was obtained from ketone **12** with the treatment of TMSCl and triethylamine at 110 °C in 74% yield.[7] With both compounds **16** and **13** in hand, we attempted to connect the two molecules by utilizing a Micheal addition. Keto aldehyde **11** was formed in two methods: one condition was with the Lewis acids aluminum oxide and boron trifluoride etherate at 0 °C, the second condition was with titanium (IV) chloride at -78 °C.[12, 13] Both conditions yielded keto aldehyde **11** based on the analysis of their respective NMR spectra , however neither of them gave us a pure product **11** due to the difficulty of separation. At this point, we used this material **11** for the next step without futhur purification.



Scheme 4. Synthesis of keto aldehyde **11**.

Our strategy for the next step was to prepare epoxide **9** or alkene **10,** as shown in Scheme 5. For epoxide **9**, commercially available sulfonium salt **17** was used according

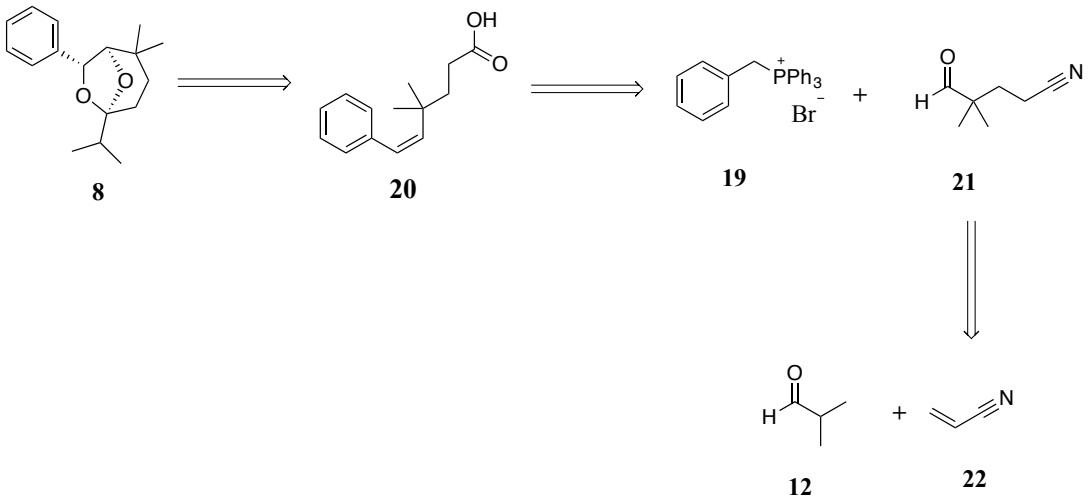
to the previously reported work done by our lab.[14] Potassium tert-butoxide was used as a strong base to form the anion at the benzylic position of sulfonium salt **17** by deprotonation. This molecule was then used to attack the aldehyde in compound **11** to form the epoxide by kicking off the dimethyl sulfide. However, epoxide **9** was not formed as we proposed. Furthermore, treatment with (nitromethyl)benzene **18** was not successful in producing the desired epoxide. Attemps to produce alkene **10** with the Wittig reagent triphenylphosphine benzyl bromide **19** did not work either. Analysis of the NMR spectra of the unpurified reaction suggested that intermolecular aldol-derived side reactions had intervened. Unfortunatelly, our original proposed synthetic route was abandoned at this stage due to the failure of synthesizing epoxide **9** and alkene **10**.



Scheme 5. Synthesis of **9** and **10**.

After the failure of our first synthetic plan, we came up with an alternative route, shown in Scheme 6. The desired product **8** would be converted from acid **20** after nucleophilic addition with isopropylmagnesium chloride and dihydroxylation with osmium tetroxide. Acid **20** could be synthesized from a Wittig reaction between

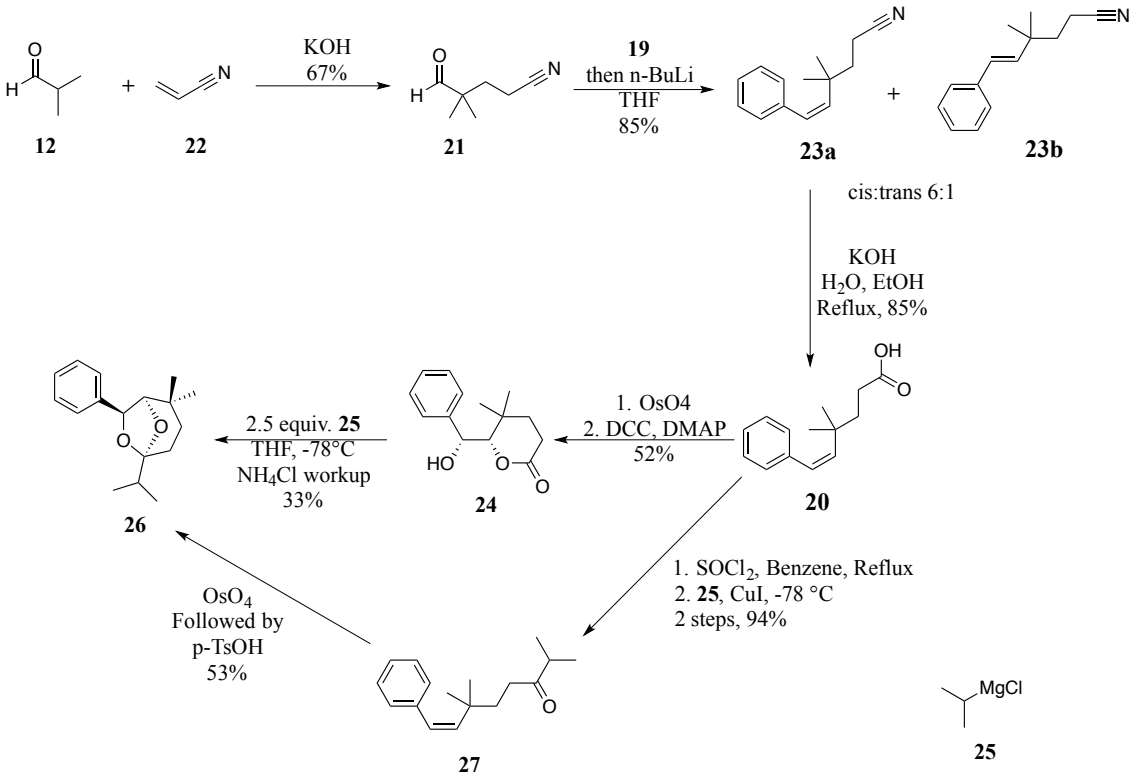
triphenyl phosphonium salt **19** and nitrile aldehyde **21** followed by the hydrolysis of a nitrile group. Isobutyraldehyde **12** and acrylonitrile would be used to make nitrile aldehyde **21**. Compared with the first plan, this route installed the isopropyl group on the molecule at the end of synthesis with the addition of isopropylmagneisium chloride, instead of making the isopropyl vinyl ketone at the beginning of synthesis. Both plans started from same starting material isobutyraldehyde **12.**



Scheme 6. Alternative route.

To test our proposed plan, we began with the synthesis of nitrile aldehyde **21** from aldehyde **12** and acrylonitrile **22**, as seen in Scheme 7. According to the method of Bruson[15], acrylonitrile was slowly added into the solution of aldehyde **12** and potassium hydroxide. Since is an exothermic reaction, the temperature of reaction must be monitored and kept below 55 °C during and after addition. The nitrile aldehyde **21** was purified with the Kugelrohr short path distillation to give a colorless liquid. This compound had the advantage that it could be made in 67% yield in multigram quantities.

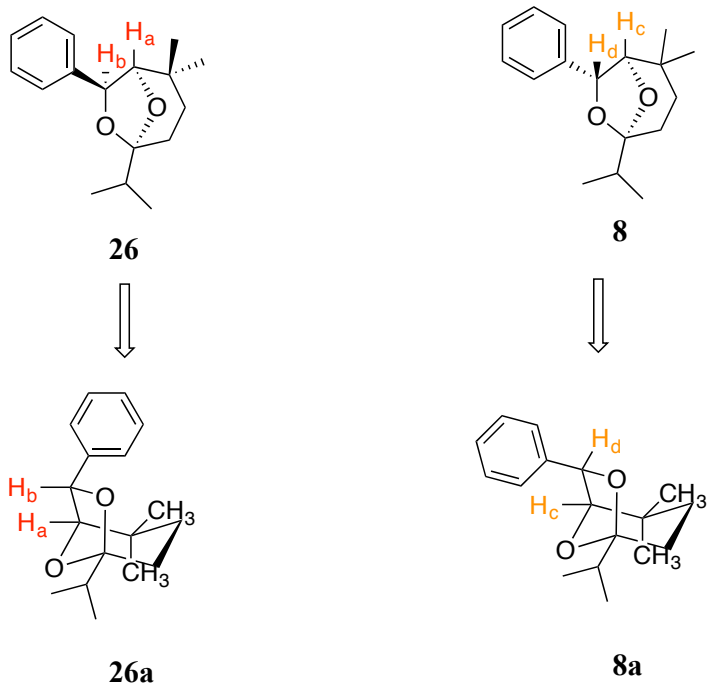
Fortunately, the reaction of triphenyl phosphonium salt **19** and n-butyllithium with aldehyde **21** provided nitrile cis isomer **23a** in 65% yield after silica gel chromatography, as shown in Scheme 6. (85% yield for **23a** and **23b**, **23a**:**23b**=6:1). Hydrolysis of **23a** with KOH in a co-solvent of ethanol and water provided carboxylic acid **20** in 85% yield.[16]



Scheme 7. Synthesis of compound **26**.

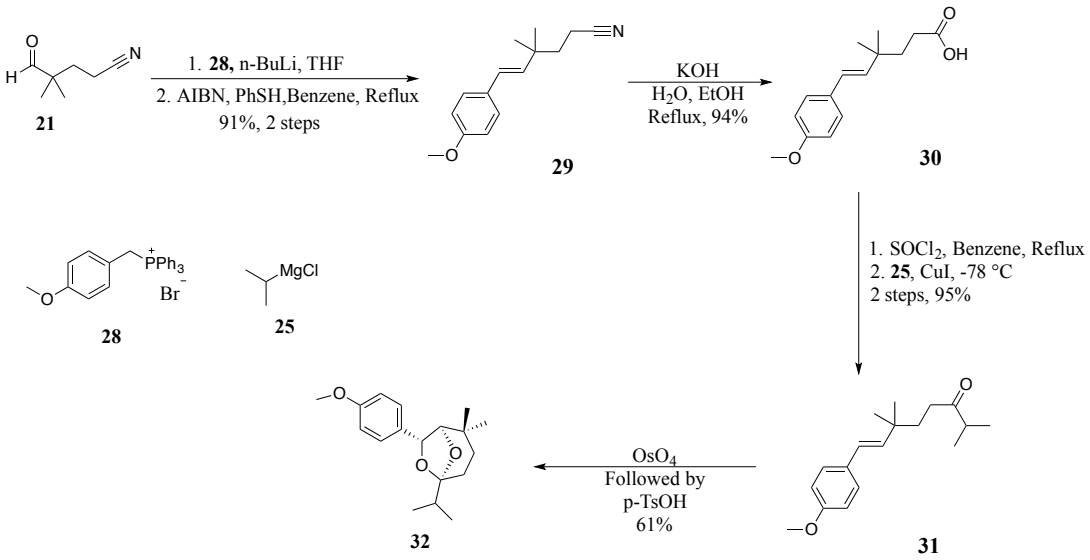
Although the resulting olefinic acid **20** could not be epoxidized with mCPBA, it did react with catalytic osmium tetraoxide and trimethylamine N-oxide dihydrate in acetone and water. Following this was a coupling reaction with DCC and DMAP to give lactone **24** in 52% yield for two steps, as shown in Scheme 7.[17] Lactone **24** reacted

readily with isopropylmagnesium chloride at -78 °C to afford ketal **26** in 33% yield, after a workup with an aqueous ammonium chloride solution. We tried to utilize isopropylmagnesium chloride to convert the nitrile group to ketone **27** in one step, which should be a easy step according to the literature. However, all our attempts failed to form the desired ketone **27** with this method. After some experimentation, we found that acid **20** reacted with thionyl chloride followed by an addition of isopropylmagnesium chloride at -78 °C in THF with the appearance of copper (I) iodide to form the ketone **27** in 94% yield. The reaction of ketone **27** with catalytic osmium tetraoxide and trimethylamine N-oxide dihydrate followed by treatment with PTSA afforded **26** in 53% yield. The second route to produce compound **26** through alkene **27** gave a higher overall yield compared to the route via lactone **24**.



Scheme 8. Three-dimensional structure of **8** and **26**.

After compound **26** was obtained, we tried to confirm the structure by comparing the NMR spectrum data of **26** with data in the literature.[2] In our compound **26**, the coupling constant for the methines protons H_a and H_b was 4.6 Hz, which indicated that there was coupling between the two protons. However, according to the literature, the coupling constant for the adjacent methine protons H_c and H_d in compound **8** should be less than 1 Hz, since it had the same stereochemistry in the dioxabicyclo[3.2.1]octane skeleton as psoracorylifol B (**2**). Based on this data, we concluded that the stereo-structure of compound **26** should be **26a** in Scheme 8. Comparing structure **26a** with **8a**, the interacton between H_a and H_b in **26a** caused the splitting of these two protons resulting in the coupling constant with a value of 4.6 Hz. The low coupling constant between H_c and H_d can be explained with the dihedral angle between H_c and H_d in structure **8a**, which was close to 90°.



Scheme 9. Synthesis of analog **32**.

Even though the stereochemistry of compound **26** was different with our target model molecule **8,** the dioxabicyclo[3.2.1]octane core structure could be formed by this direct method. For further investigation, we hypothesized a new pathway, in which the stereochemistry of the core structure would be constructed with the trans olefinic acid instead of cis isomer **20**. To check our hypothesis, as well as to synthesize a closer analog **32**, we reacted nitrile aldehyde **21** with 4-methoxybenzyl(triphenyl)phosphorene **28** to give trans and cis isomers, as shown in Scheme 9. All of the cis isomer in the crude mixture was converted to the trans isomer by radical rearrangement initialized with azobisisobutyronitrile and thiophenol in refluxing benzene.[18] This two-step procedure produced the trans isomer as the major product of the Wittig reaction and increased the yield to 91%, compared to 12% for **23b** in Scheme 6. Hydrolysis of the nitrile group converted it to a carboxylic acid with potassium hydroxide in aqueous ethanol at reflux in 94% yield. Treatment of acid **30** with thionyl chloride at high temperature followed by the addition of isopropylmagnesium chloride with copper (I) iodide at -78 °C formed ketone **31** in 95% yield for 2 steps. Final product **32** was derieved from **31** in 61% yield with the same procedure used for **26**, catalytic osmium tetraoxide and trimethylamine N-oxide dihydrate followed by treatment with PTSA. The coupling constant for the adjacent methine protons in compound **32** was less than 1 Hz, which indicated that ketal **32** had the same structure as psoracorylifol B (**2**).

2.3. Conclusions

Yue and coworkers first isolated psoracorylifols, a well-known traditional Chinese medicine, in 2006 from the seeds of *Psoralea corylifolia* L.[2] Since then psoracorylifols B (**2**) and C (**3**) attracted our attention due to their unique 6,8-dioxabicyclo[3.2.1]octane skeleton and valuable biological activities. In our work, we utilized ketal formation as the key step to form this unique bicyclic structure. The synthetic route we developed to synthesize ketal **32** only contains five steps from isobutyraldehyde **12** in 40% yield. The bicyclic ketal structure was confirmed by comparing NMR spectrum data with literature. This route will enable the analysis of stereoisomeric analogs of **2** in order to better understand the mechanism of action of this novel family of natural products.

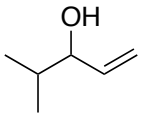
2.4. Experimental

General Procedures

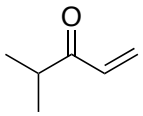
All starting materials were purchased from Sigma-Aldrich; THF was freshly distilled with LAH, other solvents were purchased from Fisher Scientific and used without further purification. All reactions were carried out in flame-dried glassware under argon with dry solvents under anhydrous conditions, unless specified. All yields refer to chromato-graphically isolated products. Reactions were monitored by thin layer chromatography (TLC) carried out on 0.20 mm silica gel plates using UV light as a visualizing agent and potassium permanganate with heat as developing agents. Silica gel

60Å, particle size 0.032 – 0.063 mm, was used for flash column chromatography. ^{1}H and ^{13}C NMR spectra were acquired on a Varian VXR-300 spectrometer. ^{1}H and ^{13}C chemical shifts (δ) are given in ppm relative to the residual protonated solvent peak ($CDCl_3$: $_{\delta}H$ = 7.26 ppm, $_{\delta}C$ = 77.0 ppm; CD_3OD: $_{\delta}H$ = 3.31 ppm, $_{\delta}C$ =49.0 ppm; $(CD_3)_2SO$: $_{\delta}H$ = 2.50 ppm, $_{\delta}C$ = 39.52 ppm; $(CD_3)_2CO$: $_{\delta}H$ = 2.05 ppm, $_{\delta}C$ = 29.84 ppm) as an internal reference. High resolution mass spectra (HRMS) were recorded on an Agilent 6540 QTOF (quadrupole time of flight) mass spectrometer using ESI (electrospray ionization) or APCI (atmospheric-pressure chemical ionization), or EI (electron ionization) on an Agilent 6890 GC/MS.

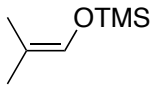
Selected Experimental, Physical, and Spectral Data



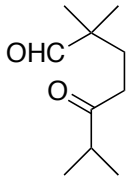
4-Methylpent-1-en-3-ol (15): To a solution of isobutyraldehyde (0.365 mL, 4 mmol) in THF (5 mL) was added vinylmagnesium bromide (1M in THF solution, 4.8 mL, 4.8 mmol) dropwise at 0 °C. After 30 minutes at 0 °C, the solution was allowed to warm to room temperature and sit for another 2 h. Saturated NH_4Cl aqueous solution was added slowly to quench the reaction followed by extraction with diethyl ether (3 x 30 mL). The organic layers were collected, dried with anhydrous $MgSO_4$, filtered and concentrated *in vacuo*. The residue was purified by flash column chromatography (silica gel, Et_2O:pentane 1:9) to afforded **15** as light yellow oil in 75% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 5.85 (ddd, *J* = 17.1, 10.4, 6.5 Hz, 1H), 5.29 (s, 1H), 5.27 – 5.08 (m, 2H), 3.90 – 3.79 (m, 1H), 1.79 – 1.65 (m, 1H), 1.00 – 0.78 (m, 6H).



4-Methylpent-1-en-3-one (13): To a solution of secondary alcohol **15** (0.2313 g, 2.3 mmol) in acetone (5 mL) at 0 °C was added a diluted Jones reagent (1 mL 8 N Jones reagent in 9 mL acetone) dropwise over 20 minutes until the orange color persisted. 2-Isopropanol was added dropwise until the color turned to dark green. The resulting mixture was concentrated and extracted with diethyl ether (3 x 30 mL). The organic layer was washed with water, sodium bicarbonate solution and brine, dried with anhydrous $MgSO_4$, filtered and concentrated *in vacuo*. The residue was purified by flash column chromatography (silica gel, Et_2O:pentane 1:9) to afforded **13** as light yellow oil in 60% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.52 – 6.36 (m, 1H), 6.26 (dd, *J* = 17.5, 1.7 Hz, 1H), 5.77 (dd, *J* = 10.4, 1.6 Hz, 1H), 2.96 – 2.75 (m, 1H), 1.13 (d, *J* = 6.9 Hz, 6H).

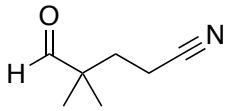


Enol silyl ether (16): To a flame-dried flask with isobutyraldehyde (0.183 mL, 2 mmol) and triethylamine (0.418 mL, 3 mmol) in DMF (2 mL) was added TMSCl (0.381 mL, 2 mmol). The mixture was heated at 110 °C for 3 h. After the reaction was cooled down, pentane (3x 20 mL) was added into flask. The clear pentane layer was collected, combined and concentrated *in vacuo* to give **16** in 74% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.00 (dd, *J* = 2.8, 1.4 Hz, 1H), 1.56 (d, *J* = 14.5 Hz, 6H), 0.16 (s, 9H).



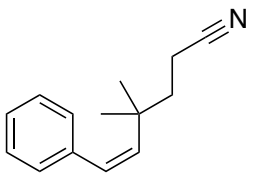
2,2,6-Trimethyl-5-oxoheptanal (11): Method A: The mixture of enone **13** (0.1350 g, 1.38 mmol), enol silyl ether **16** (0.1168g, 0.81 mmol), Al_2O_3 (0.5780 g) and $ZnCl_2$ (0.1560 g) was stirred at 0 °C for 30 minutes. After adding BF_3 etherate (0.2142 g, 0.17 mmol), the resulting mixture was stired at 0 °C for another 4 hours. Saturated NH_4Cl aqueous solution was added slowly to quench the reaction followed by extraction with diethyl ether (3 x 30 mL). The organic layers were collected, dried with anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give crude product **11**.

Method B: To a flame-dried flask with $TiCl_4$ (1 M solution in CH_2Cl_2, 0.54 mL, 0.54 mmol) was added enone **13** (0.053 g, 0.54 mmol) in CH_2Cl_2 at -78 °C. After stirring for 10 minutes, enol silyl ether **16** (0.0792 g, 0.54 mmol) was added at -78 °C dropwise. The reaction mixture was allowed to stir at – 78 °C for another 5 minutes, then quenched with saturated sodium bicarbonate aquesous solution. Diethyl ether was added for extraction (3 x 20 ml). The extracte was dried with anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give crude keto aldehyde **11**.

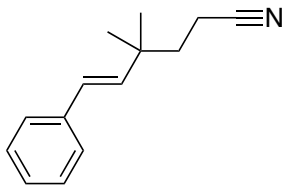


4,4-Dimethyl-5-oxopentanenitrile (21): To a solution of isobutyraldehyde (4 g, 5.06 mmol) and 50% aqueous KOH (0.2 g) was added acrylonitrile slowly at 0 °C. The

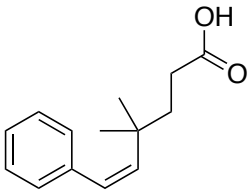
temperature was allowed to go back to room temperature after addition. After stirring at room temperature for 1 h and 55 °C for one hour, 4 M HCl aqueous solution was added until the pH value was 5. The mixture was washed with water (2 x 50 mL), followed by drying with anhydrous $MgSO_4$, filtration and concentration *in vacuo*. The product **21** was purified by Kugelrohr distillation in 67% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 9.36 (s, 1H), 2.31 – 2.19 (m, 2H), 1.83 - 1.79 (m, 2H), 1.05 (s, 6H).



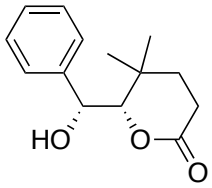
(*Z*)-4,4-dimethyl-6-phenyl-5-hexenenitrile (23a): To a solution of benzyltriphenylphophonium bromide **19** (4.97 g, 11.46 mmol) in 60 mL of THF at 0°C, *n*-BuLi (2.5 M in hexane solution, 4.6 mL, 11.46 mmol) was added dropwise. After stirring at 0°C for 1 h, nitrile aldehyde **21** (1.20 g, 9.55 mmol) was added to the solution. The resulting solution was stirred overnight at room temperature. Saturated NH_4Cl solution was added to quench the reaction, followed by extraction with ethyl acetate. The desired nitrile product was isolated by flash column chromatography (silica gel, EtOAc:hexane 1:10), in 73% combined yield, and a *cis*:*trans* ratio of 6:1; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 7.37 – 7.21 (m, 3 H), 7.19 – 7.09 (m, 2 H), 6.58 (d, *J* = 12.7 Hz, 1 H), 5.43 (d, *J* = 12.7 Hz, 1 H), 2.31 – 2.19 (m, 2 H), 1.71 – 1.59 (m, 2 H), 0.98 (s, 6 H).



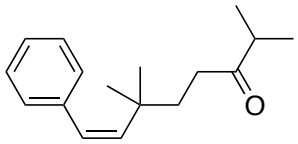
(E)-4,4-dimethyl-6-phenyl-5-hexenenitrile (23b): Compound **23b** was collected as a minor product of procedure of **23a**. ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.41 – 7.22 (m, 5H), 6.33 (d, *J* = 16.3 Hz, 1H), 6.07 (d, *J* = 16.3 Hz, 1H), 2.33 – 2.21 (m, 2H), 1.81 (m, 2H), 1.15 (s, 6H).



Z-4,4-Dimethyl-6-phenyl-5-hexenoic acid (20): To a solution of nitrile **23a** (0.40 g, 2 mmol) in 10 mL of 4:1 ethanol and water, KOH (0.34 g, 6 mmol) was added in one portion. After the reaction mixture had been heated to reflux for 1 h, another portion of KOH (0.17, 3 mmol) was added. The reaction was kept boiling for another 24 h. A 1M HCl solution was added to the cooled reaction mixture until the pH=3. Extraction with ethyl acetate gave 0.42 g (85%) of acid; ^{1}H NMR (300 MHz, $CDCl_3$): δ= 7.34 – 7.11 (m, 5H), 6.53 (d, J = 12.7 Hz, 1H), 5.46 (d, *J* = 12.7 Hz, 1H), 2.40 – 2.27 (m, 2H), 1.70 – 1.57 (m, 2H), 0.94 (s, 6H).

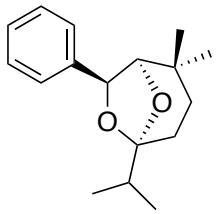


Lactone (24): To a 2:1 acetone and H_2O solution (20 mL) with acid **20** (0.22 g, 1 mmol) and of TMAO (0.45 g, 4 mmol), 0.63 mL of OsO_4 in t-BuOH solution (2.5% wt. %) was added dropwise. The reaction was monitored by TLC until the starting material was gone. Saturated sodium sulfite solution was added, and the mixture was stirred for 30 min followed by extraction with dichloromethane. The resulting product was dissolved in dichloromethane, followed by 1 equivalent of DCC and 10 mol % of DMAP. The reaction mixture was stirred overnight, then washed with saturated NH_4Cl solution, and brine. Flash column chromatography (silica gel, EtOAc:hexane 2:1) gave the desired lactone product in 52% yield; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 7.47 – 7.24 (m, 5H), 4.81 (t, *J* = 5.3 Hz, 1H), 4.30 (d, *J* = 5.7 Hz, 1H), 2.67 (d, *J* = 5.0 Hz, 1H), 2.44 (ddd, *J* = 11.8, 8.1, 6.4 Hz, 2H), 1.65 (dt, *J* = 13.2, 8.1 Hz, 1H), 1.57 – 1.43 (m, 1H), 1.12 (s, 3H), 0.92 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$): δ = 171.4, 141.1, 128.8, 128.6 127.7, 89.4, 74.6, 35.6, 32.4, 27.6, 27.5, 20.9.



(*Z*)-2,6,6-Trimethyl-8-phenyl-7-octen-3-one (27): The reaction mixture of carboxylic acid **20** (0.0867 g, 0.40 mmol) and thionyl chloride (0.058 mL, 0.8 mmol) in benzene solution was heated to reflux for 4 hr. When the temperature cooled down, the mixture was concentrated *in vacuo.* At – 78 °C, to the resulting mixture with CuI (0.1171 g, 0.62

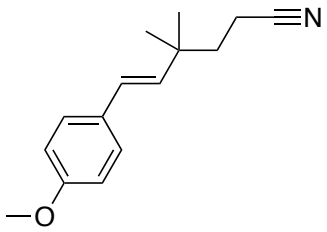
mmol) in THF was added isopropylmagnesium chloride (2 M in THF solution, 0.226 ml, 0.452mmol) dropwise. The reaction mixture was allowed to warm to room temperature and stir overnight. 1 M HCl aqueous solution was added to quench the reaction follow by extraction with EtOAc(3 x 30 mL), drying with anhydrous $MgSO_4$, filtration and concentration *in vacuo* to give crude product 27 in 94% yield without further purificaton; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.33 – 7.07 (m, 5H), 6.50 (d, *J* = 12.7 Hz, 1H), 5.46 (d, *J* = 12.7 Hz, 1H), 2.62 – 2.49 (m, 1H), 2.53 – 2.27 (m, 2H), 1.70 – 1.50 (m, 2H), 1.07 (dd, *J* = 7.3, 2.4 Hz, 6H), 0.94 (d, *J* = 2.8 Hz, 6H).



5-Isopropyl-2,2-dimethyl-7-phenyl-6,8-dioxabicyclooctane (26): Route A (from lactone **24**): To a solution of 62 mg (0.26 mmol) of lactone in THF solution at -78°C, 0.33 mL (0.66 mmol) of isopropyl magnesium chloride solution (2 M in THF) was added dropwise. The reaction was stirred at -78°C for 5 h. Then saturated NH_4Cl solution was added to quench the reaction, followed by extraction with ethyl acetate. Prep-TLC gave 23 mg (33%) of product.

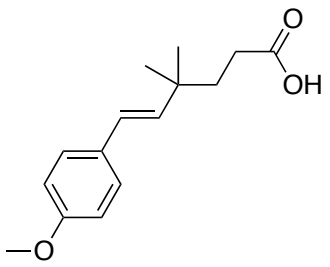
Route B (from alkene **27**): To a 2:1 acetone and H_2O solution (5 mL) with ketone **27** (0.091 g, 0.37 mmol) and of TMAO (0.083 g, 0.74 mmol), 0.23 mL of OsO_4 in t-BuOH solution (2.5% wt. %) was added dropwise. The reaction was monitored by TLC until the starting material was gone. Saturated sodium sulfite solution was added, and the mixture

was stirred for 30 min followed by extraction with dichloromethane. The resulting product was dissolved in dichloromethane, and was allowed to stir with p-TsOH (5 mg) at room temperature for overnight. The resulting mixture was filtered and concentrated *in vacuo*. The product **26** was purified with flash column chromatography (silica gel, EtOAc:hexane 1:4) in 53% yield; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 7.46 (d, *J* = 7.6 Hz, 2H), 7.38 – 7.18 (m, 3H), 5.25 (d, *J* = 4.6 Hz, 1H), 4.03 (dd, *J* = 4.6, 1.9 Hz, 1H), 2.11 – 1.66 (m, 4H), 1.21 – 1.13 (m, 1H), 1.11 – 1.00 (m, 9H), 0.17 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$): *δ* 137.8, 128.3, 127.2, 126.5, 111.3, 86.0, 81.5, 35.4, 34.6, 30.8, 28.5, 28.0, 26.1, 17.1, 17.1.

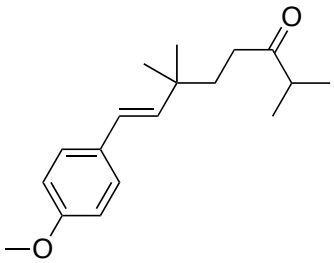


(E)-4,4-Dimethyl-6-(4-methoxyphenyl)-5-hexenenitrile (29): To a solution of (4-methoxybenzyl)triphenylphophonium bromide **28** (4.33 g, 10.3 mmol) in 60 mL of THF at 0°C, *n*-BuLi (2.5 M in hexane solution, 4.12 mL, 10.3 mmol) was added dropwise. After stirring at 0°C for 1 h, nitrile aldehyde **21** (1.1766 g, 9.4 mmol) was added to the solution. The resulting solution was stirred overnight at room temperature. Saturated NH_4Cl solution was added to quench the reaction, followed by extraction with ethyl acetate. To the crude product (1.9649 g, 8.57 mmol) in 50 mL benzene solution was added AIBN (0.1407g, 0.857 mmol) and thiophenol (0.44 mL, 4.285 mmol). The resulting mixture was heated to reflux for 9 h. After evaporation of solvent, the mixture was purified with flash column chromatography (silica gel, EtOAc:hexane 1:10) to give

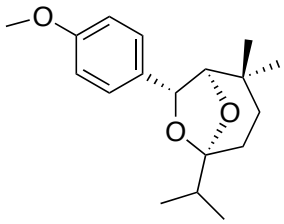
trans isomer **29** in 91% for 2 steps; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.29 (d, *J* = 8.4 Hz, 2H), 6.85 (d, *J* = 8.7 Hz, 2H), 6.26 (d, *J* = 16.2 Hz, 1H), 5.92 (d, *J* = 16.2 Hz, 1H), 3.81 (s, 3H), 2.37 – 2.20 (m, 2H), 1.85 – 1.72 (m, 2H), 1.13 (s, 6H).



E-4,4-Dimethyl-6-(4-methoxyphenyl)-5-hexenoic acid (30): Acid **30** was prepared according to previous hydrolysis procedure of acid **20** in 94% yield.



(E)-2,6,6-Trimethyl-8-(4-methoxyphenyl)-7-octen-3-one (31): Compound **31** was prepared according to previous procedure of ketone **27** in 95% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.28 (d, *J* = 8.7 Hz, 2H), 6.84 (d, *J* = 8.7 Hz, 2H), 6.22 (d, *J* = 16.2 Hz, 1H), 5.97 (d, *J* = 16.2 Hz, 1H), 3.80 (s, 3H), 2.57 (p, *J* = 7.0 Hz, 1H), 2.45 – 2.33 (m, 2H), 1.65 (m, 2H), 1.15 – 1.00 (m, 12H).



5-Isopropyl-7-(4-methoxyphenyl)-2,2-dimethyl-6,8-dioxabicyclooctane (32): Analog **32** was obtained according previous procedure for **26** (route B) in 61% yield; ^{1}H NMR (300 MHz, $CDCl_3$): δ 7.26 – 7.16 (m, 2H), 6.82 – 6.75 (m, 2H), 4.98 (br s, 1H), 3.72 (br s, 4H), 2.00 (p, *J* = 7.0 Hz, 1H), 1.68 (dd, *J* = 8.7, 2.0 Hz, 2H), 1.58 – 1.50 (m, 1H), 1.35 – 1.28 (m, 1H), 1.05 – 0.94 (m, 9H), 0.91 (s, 3H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 159.2, 135.5, 127.8, 113.9, 112.2, 90.1, 78.0, 55.5, 35.7, 32.8, 30.3, 26.4, 26.2, 24.6, 18.4, 17.8; HRMS (ESI-TOF) calcd for $C_{18}H_{26}O_3$ $[M + H]^+$ 291.1955, found 290.1882.

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CHAPTER 3.

TOTAL SYNTHESIS OF FUROMOLLUGIN*

3.1. Introduction

The roots of *Rubia cordifolia* L. (Rubiaceae) have been used in traditional Chinese medicine for centuries to treat joint inflammation, dysmenorrhea, hematorrhea, hemostasis, and psoriasis.[1,2] They are officially listed in the Chinese Pharmacopoeia, which is an official compendium of drugs, covering traditional Chinese and western medicines. The methanol extract of the dried roots of *R. cordifolia* can be fractionated by partitioning first between chloroform and water, and then between n-butanol and water. Anthraquinone, naphtho-hydroquinone, naphthohydroquinone are some of the compounds that have been isolated using this method since the 1970s.[3] Most of the isolated compounds contain the furonaphthalene subunit. Specific natural products are shown in Figure 1 and include furomollugin (**1**), mollugin (**2**), and rubilactone (**3**).[4-6]

The biological activities of these isolated compounds have been investigated after the antitumour activity of bicyclic hexapeptides, which were isolated from the extract of *Rubia cordfolia*, was reported by Itokawa and co-workers two decades ago.[7] In 1996, the inhibition activity of furomollugin (**1**) and mollugin (**2**) against hepatitis B virus (HBV), which commonly results in chronic and acute hepatitis, was reported by Ho and coworkers.[8] Compound **1** and **2** strongly suppressed the secretion of the hepatitis B

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surface antigen, both with IC_{50} values of 2.0 μg/mL, in human hepatoma Hep3B cells while having little effect on the viability of the cells. Compound **2** is thought to be a Janus kinase 2 (JAK2) inhibitor and inhibits lipopolysaccharide (LPS)-induced inflammatory responses by blocking the activation of the Janus kinase/ signal transducers and activators of the transcription (JAK-STAT) pathway.[9] According to the work conducted by Woo and co-workers, compound **1** showed weak cytotoxicity to a human colon carcinoma cell line (HT-29), possibility mediated through topoisomerase inhibition.[10]

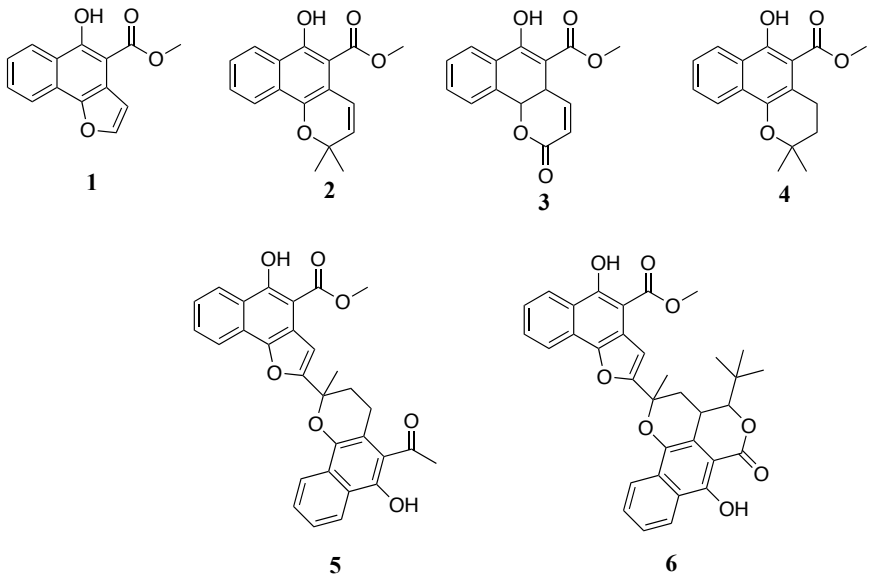
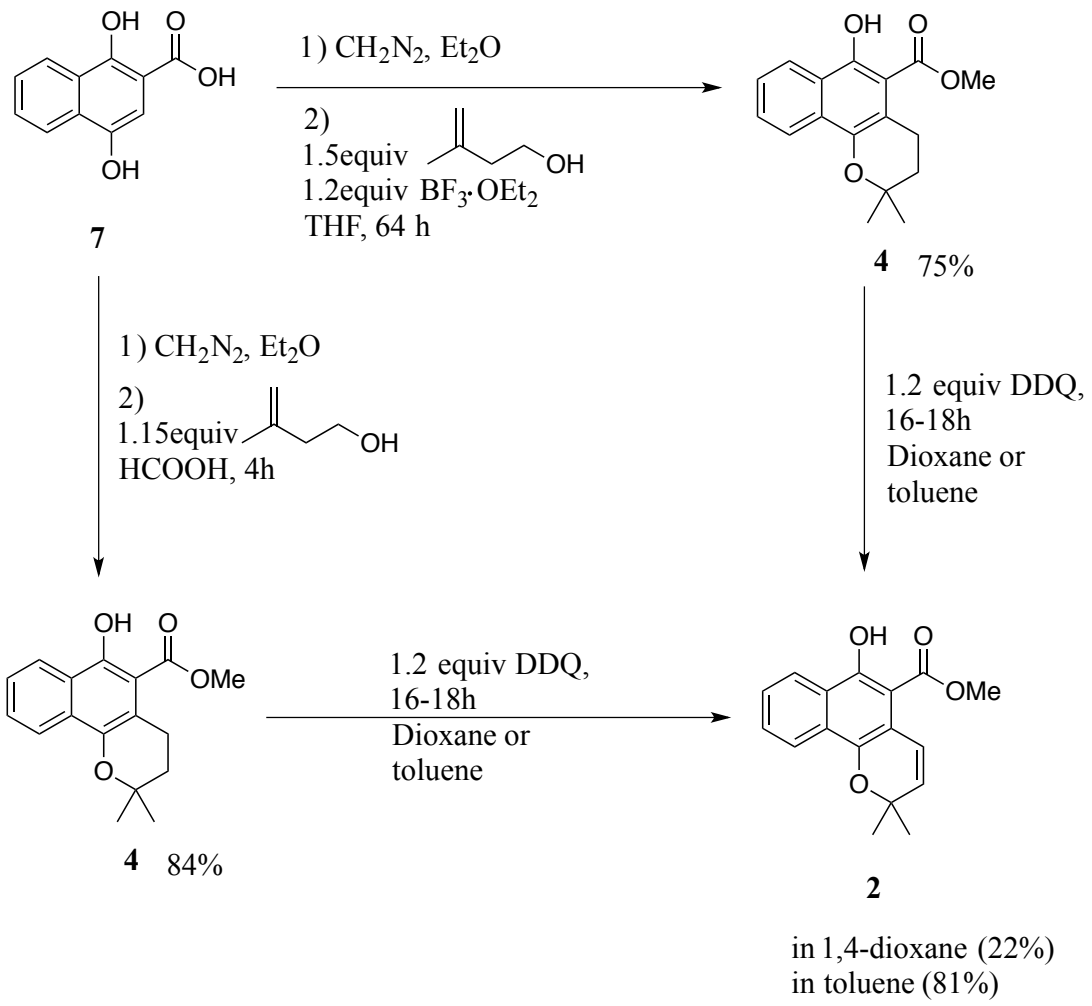


Figure 1. Some isolated compounds.

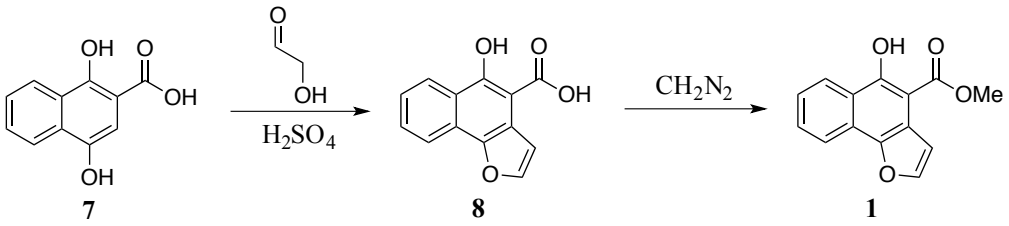
Since the isolation of these compounds from *R. cordifolia*, scientists have been working to find effective synthetic routes to produce the natural compounds with unique structures and interesting biological activities. Among all these analogs, mollugin (**2**) is well investigated and several synthetic routes have been reported since it was first

isolated. Most of the reported methods utilized acid-catalyzed cyclization of methyl 1, 4-dihydroxy-2-naphthoate and different reagents, including a very direct synthesis by De Kimpe, as shown in Scheme 1.[11] In the route developed by De Kimpe, methyl 1,4-dihydroxy-naphthalene-2-carboxylate was first prepared by esterification of the carboxylic acid **6** with diazomethane. Then the methyl ester was heated under reflux with 3-methyl-3-buten-1-ol in tetrahydrofuran (THF) and boron trifluoride diethyl etherate (BF_3-OEt_2) to yield 3,4-dihydromollugin **4** via BF_3-activation of the alcohol moiety by borate formation followed by subsequent *ortho*-alkylation of the aromatic alcohol and cyclization by attack of the phenol oxygen across the proton-activated carbon–carbon double bond of the isoprenyl group. Alternatively, the yield of the isolated 3,4-dihydromollugin **4** can be increased to 84% by using formic acid instead of BF_3-OEt_2. In the last step, 3,4-dihydromollugin **4** was converted to the desired mollugin **2** by oxidation with dichlorodicyano-*p*-benzoquinone (DDQ) in dioxane in 22% yield. This yield can be increased to 81% by using a DDQ oxidation of dihydromollugin **4** in toluene.

For furomollugin (**1**), there are also some reported methods. The first synthetic route was reported by Schildknecht and Straub in 1976, as seen in Scheme 2.[12] They synthesized furomollugin (**1)** from 1,4-dihydroxy-2-naphthoic acid **7**. The sulfuric acid catalyzed cyclization of compound **7** and glycolaldehyde formed acid **8**. The selective esterification with diazomethane gave desired compound **1**. This is the first reported synthesis of furmollugin **1**. However, the overall yield of this method is really low, only 2% from acid **7** to ester **1**.



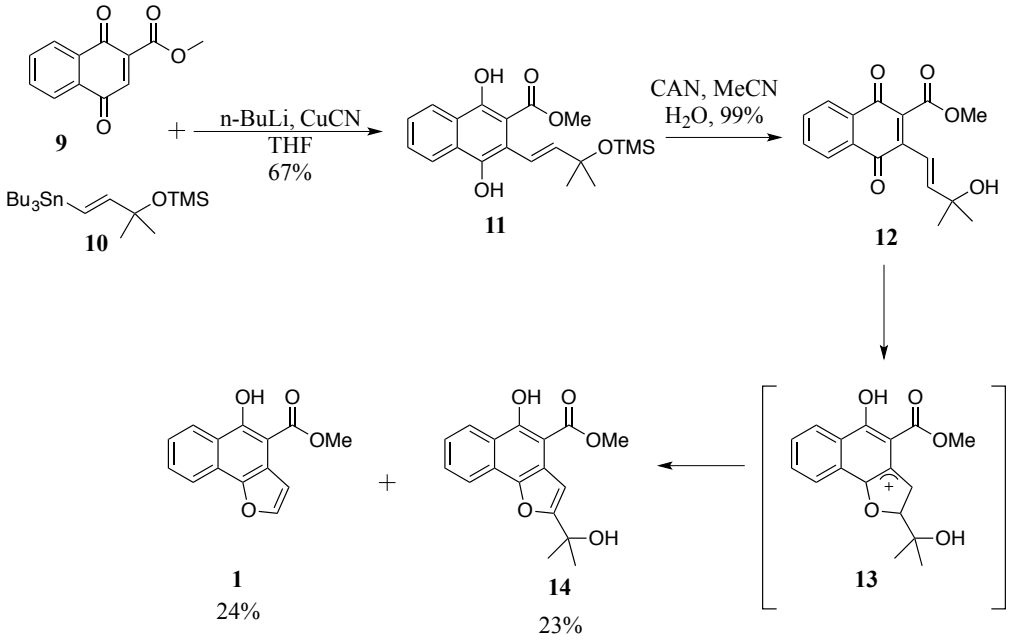
Scheme 1. Direct synthesis of **2** by De Kimpe.



Scheme 2. First synthesis of furomollugin (**1**) by Schildknecht and Straub.

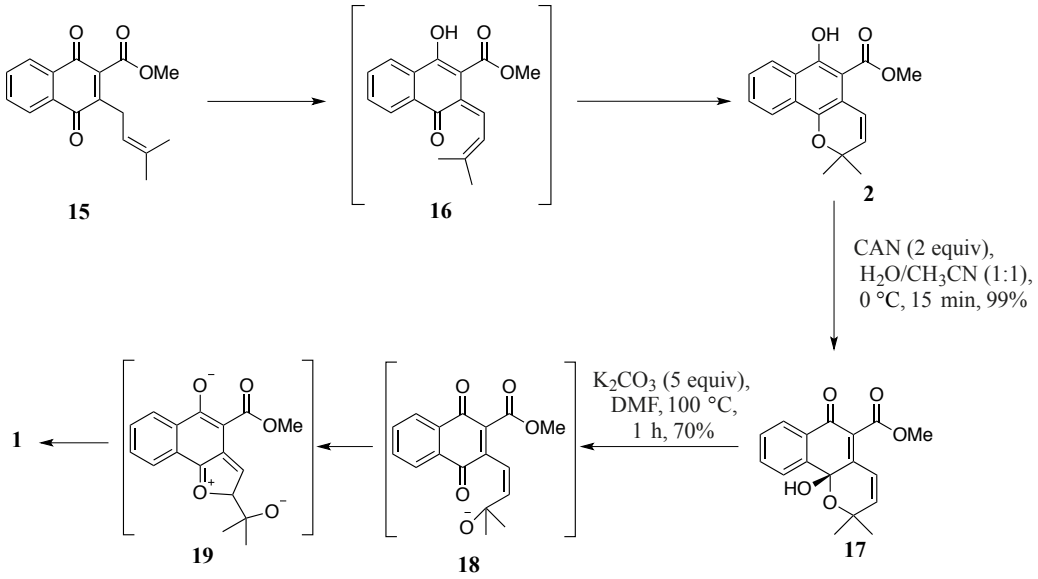
In 2005, another synthetic route to furomollugin **1** was reported by Trauner and co-workers in Scheme 3.[13] They started from 2-carbomethoxy- naphthoquinone **9**, which

was prepared by Fischer esterification of 1,4-dihydroxynaphthoic acid **7**, followed by oxidative demethylation, as exhibited in Scheme 3. Vinyl naphthohydroquinone **11** was prepared by conjugate addition of a vinyl cuprate derived from vinyl stannane **10** to 2-carbomethoxynaphthoquinone **9**, followed by tautomerization. Oxidation of **11** with aqueous cerium ammonium nitrate not only restored the quinone but also resulted in cleavage of the silyl ether to afford **12**. Vinyl quinone **12** was not stable and underwent conversion into naphthofuran **14** in 23% yield and furomollugin (**1**) in 24% yield. This reaction presumably proceeds through cation **13**, which undergoes either deprotonation to afford **14** or *retro*-Friedel-Crafts hydroxyalkylation to yield **1**. In their synthesis towards 5-hydroxybenzo[2,3-b]furan natural product rubicordifolin, furomollugin (**1**) could be produced as a minor byproduct in 36% yield.



Scheme 3. Synthesis of **1** by Trauner.

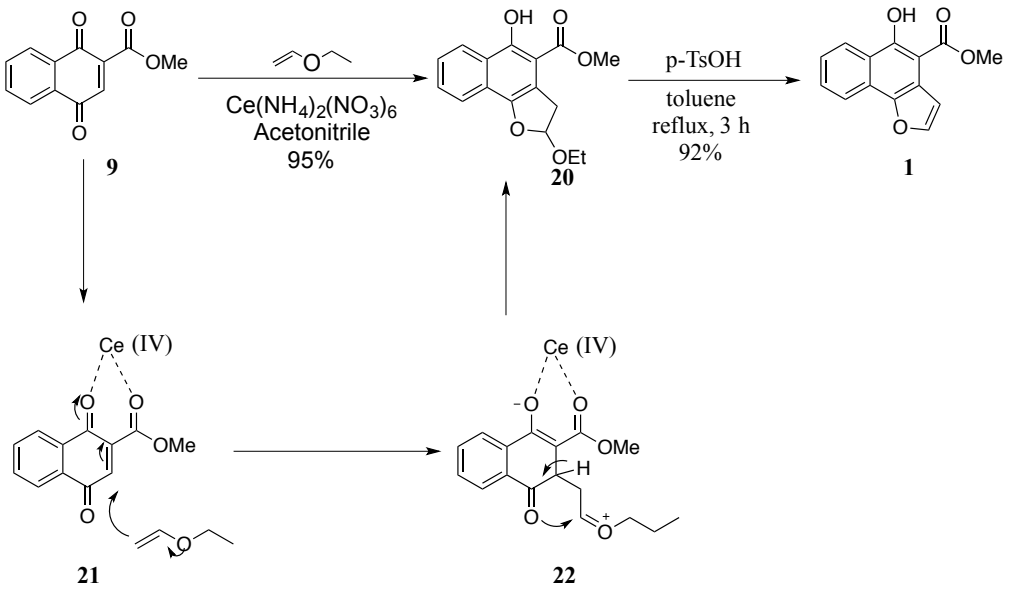
In 2008, Trauner and co-workers improved their synthesis method to furomollugin (**1**) in a total synthesis route of rubicordifolin, as seen in Scheme 4.[14] They made mollugin (**2**) from naphthoquinone **15**, which was also prepared from naphthoic acid **7**. Oxidation of **2** with cerium ammonium nitrate afforded **17**, which was converted to compound **1** under basic conditions. This conversion presumably undergoes ring opening to form **18**, then a five-member ring cyclization to provide **19**, followed by expulsion of acetone to afford **1**.



Scheme 4. Improved synthesis of **1** by Trauner.

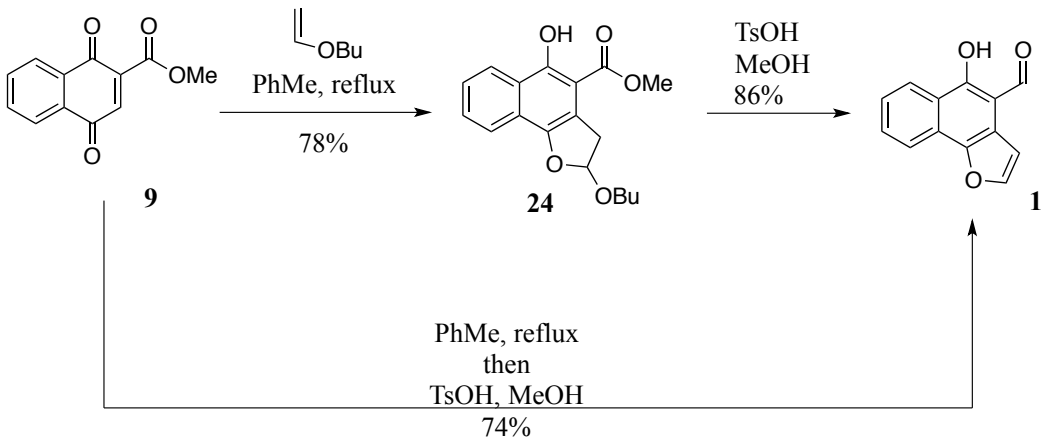
A direct synthesis of furomollugin (**1**) has been reported by Xia and Lee in 2013 as exhibited in Scheme 5. They chose previously described 2-carbomethoxy-naphthoquinone **9** as the starting material. The cyclization reaction of **9** with ethyl vinyl ether utilizing ceric ammonium nitrate as a Lewis acid catalyst. This reaction proceeds through complex **21**, followed by attack of vinyl ethyl ether to give another intermediate

22. Isomerization of **22** followed by intramolecular cyclization formed acetal **20.** Treatment of **20** with acid eliminated ethanol to provide the natural product in excellent yield over two steps.



Scheme 5. Synthetic route of **1** by Xia and Lee.

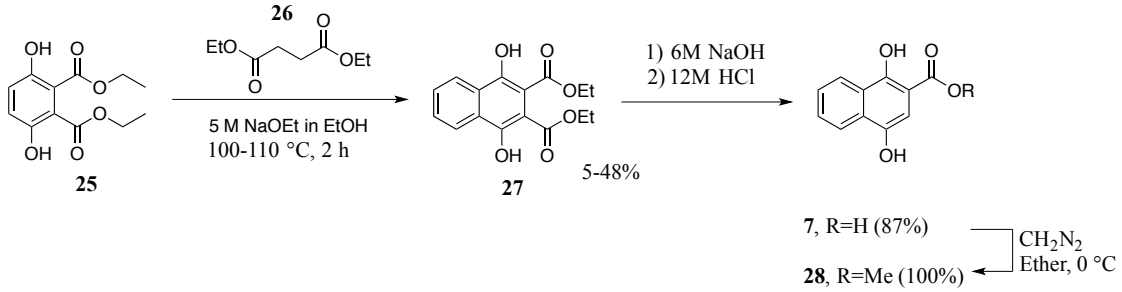
Moreover, a similar procedure was reported by Piggott in 2014, as illustrated in Scheme 6.[16] The reaction of **9** with butyl vinyl ether gave acetal **24** in good yield. Then acid-catalyzed aromatization afforded furomollugin (**1**). The one-pot procedure beginning with toluene and then switching the solvent to MeOH before acidification gave compound **1** in a slightly better yield than the two-pot process. Additionally, De Kimpe and coworkers reported an innovative ring contraction sequence that converted the mollugin skeleton to an isopropenyl-substituted furomollugin,[17] which was not covered here.



9 **24** **1**

Scheme 6. Synthetic route of **1** by Pigrott.

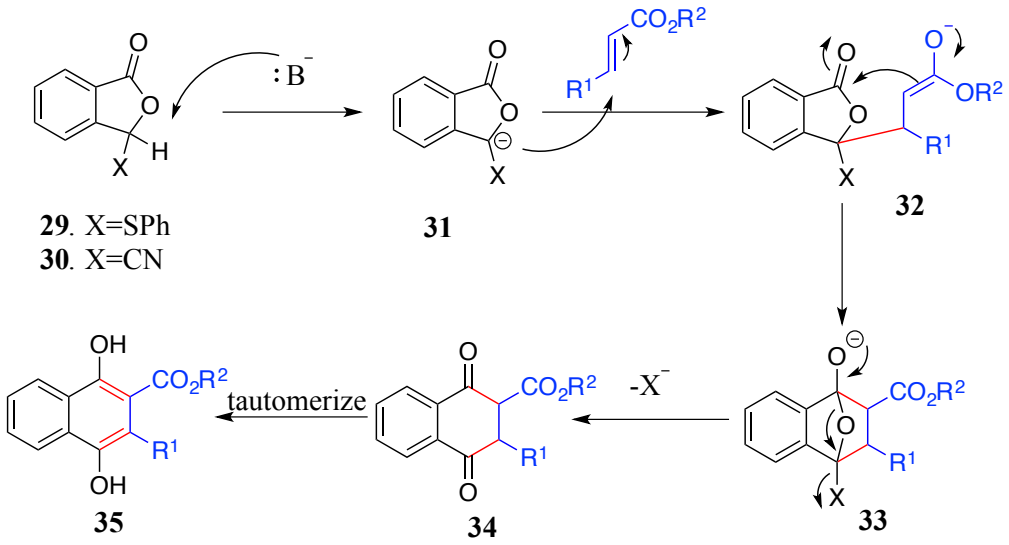
Most of the reported synthetic routes started from 2-carbomethoxy-naphthoquinone **9**, which was derived from 1,4-dihydroxy-2-naphthoic acid **7**. The preparation of **7** is shown in Scheme 7. Condensation of diethyl 3,6-dihydroxyphthalate **25** and diethyl succinate **26** followed by acid-catalysed decarboxylation afforded 1,4-dihydroxy-naphthalene-2-carboxylic acid **7**. This three-step synthesis suffers from the disadvantage of a low overall yield. The variable yields of the double Claisen condensation are often as low as 5% and mounting to 48%, pointing to a tricky reaction.[18] Due to this drawback, we wanted to design a novel synthetic route toward **1**.



25 **27**

Scheme 7. Preparation of naphthoic acid **7.**

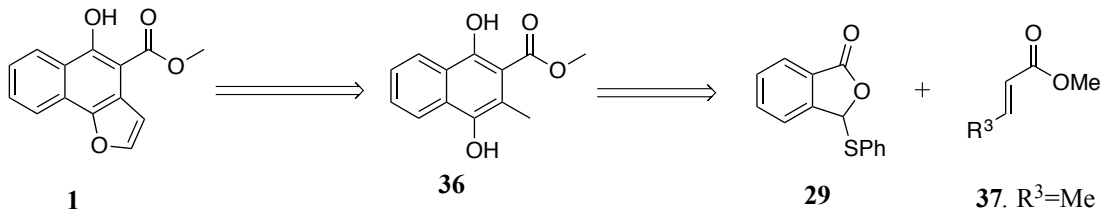
The Hauser-Kraus annulation is a good way to generate naphthalene hydroquinones from phthalide anions and α, β–unsaturated carbonyl compounds.[19, 20] The mechanism of the Hauser-Kraus annulation is shown in Scheme 8. Deprotection of the functionalized phthalide produced anion **31** followed by a Michael addition gives intermediate **32**. Then **32** undergoes a bicyclic ring formation followed by expulsion of the ketone to form naphthoquinone **34**. The desired product **35** is obtained after tautomerization. The feature of the Hauser-Kraus reactions is that four carbons of the newly formed cyclohexenone ring are supplied by the Michael acceptor.[21] A number of natural products have been synthesized via the efficient addition of phthalides **29** and **30** using a range of Michael acceptors.[22]



Scheme 8. Mechanism of the Hauser-Kraus annulation.

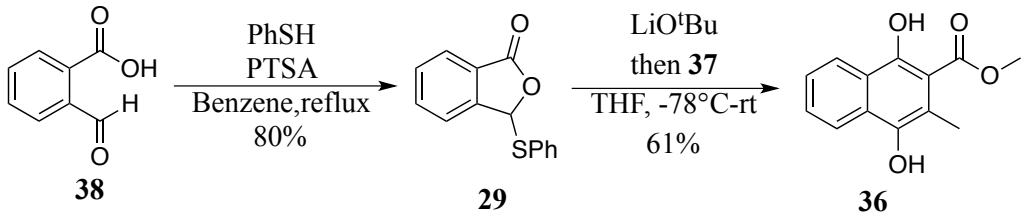
3.2. Results and Discussion

Our strategy for the synthesis of **1** involved the Hauser-Kraus annulation. The retrosynthetic analysis is depicted below in Scheme 9. The furan ring could be installed by an anion formation of the benzylic position of **36** follow by cyclization. Compound **36** would be formed from phthalide **29** and methyl crotonate **37**. And the functionalized phthalide **29** can be prepared based on the previous reported procedure.[22] In principle, it offers a pathway to compounds **1** – **3** depending on the structure of R^3 in ester **37**.



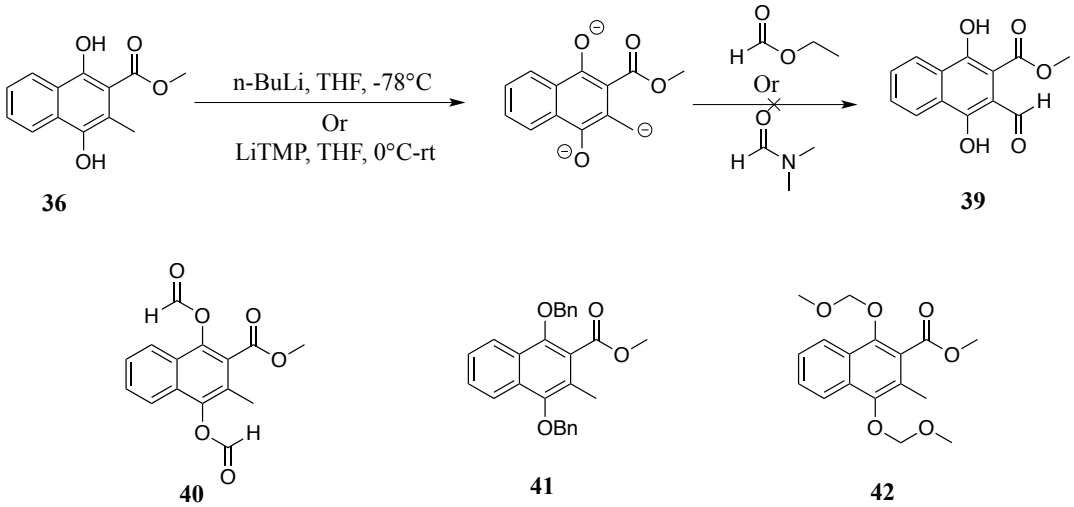
Scheme 9. Retrosynthesis of **1**.

To test our proposed route, we started to make the intermediate **36**. Treatment of 2-carboxybenzaldehyde **38** with thiophenol and para-toluenesulfonic acid (PTSA) afforded Phthalide **29** in 80% yield. Compound **36** was formed through the Hauser-Kraus annulation of **29** with methyl crotonate in 68% yield. During our work, we found that lithium tert-butoxide produced a cleaner reaction compared to potassium tert-butoxide. Additionally, we prepared some hydroxyl group protected naphthalene hydroquinones from **36**.

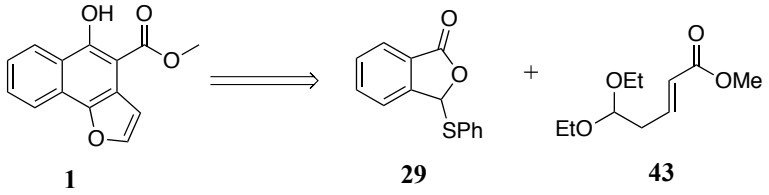


Scheme 10. Synthesis of **36**.

With compound **36** in hand, we tried to deprotonate the benzylic position of **36**, as shown in Scheme 11. First, a tri-anion was made with a strong base from **36**, then treated with ethyl formate or dimethylformamide (DMF). However, the deprotonation of **36** with 3 equivalents of lithium diisopropylamide (LDA) or lithium tetramethylpiperidide (LiTMP) returned recovered starting material. Next we tried to protect two hydroxyl groups with formyl groups to give compound **40**. We hypothesized that the formed anion on benzylic position would attack the closest carbonyl group to transfer the formyl group to the benzylic position. Unfortunately, the reaction didn't happen as we expected, only the starting material was recovered. At last, we changed the protection group to benzyl group and methoxymethyl group to get compound **41** and **42**. Again, the deprotonation of dibenzyl ether **41** or bis-MOM ether **42** with excess base (LDA, LiTMP) returned recovered starting material when treated with ethyl formate or DMF. While the rationale for our inability to acylate **36** or its ethers is not clear, we changed our synthetic plan as illustrated in Scheme 12. Instead of installing formyl group with the anion chemistry at the end, we used a Michael acceptor **43** with an acetal group which could be converted to aldehyde.

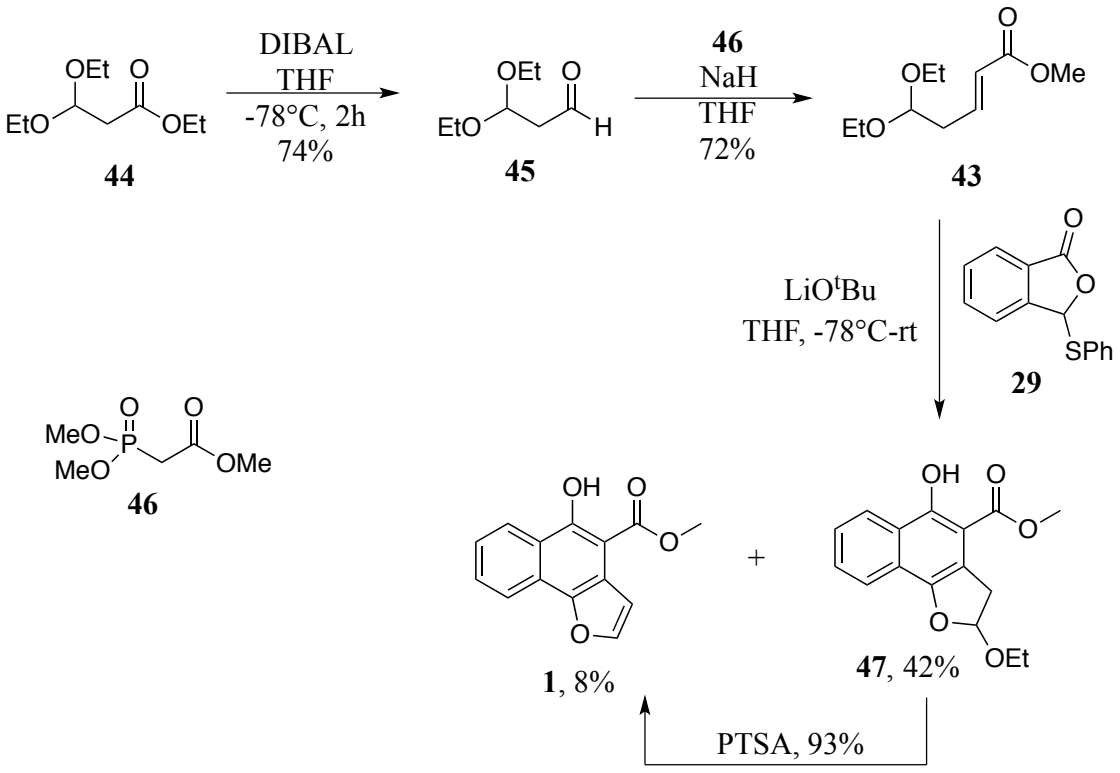


Scheme 11. Deprotonation of benzylic position.



Scheme 12. Alternate synthetic route to **1**.

To test this new route, ethyl 3,3-diethoxypropionate **44** was reduced to aldehyde **45** with diisobutylaluminium hydride (DIBAL) at -78 °C in 74% yield.[23] The resulting aldehyde was converted to the α,β-unsuturated acetal ester **43** by the Horner-Wadsworth-Emmons reaction (HWE) with trimethyl phosphonoacetate **46** in 72% yield. The Hauser-Kraus annulation with phthalide **29** and acetal ester **43** affored and furomollugin (**1**) in 8% yield and compound **47** in 42% yield which could be converted into **1** using para-toluenesulfonic acid.



Scheme 13. Synthesis of **1**.

3.3. Conclusions

Furomollugin (**1**) was isolated from the roots of *Rubia cordifolia* L. by Schildknecht and Straub in the 1970s. Since then, several synthetic routes towards this natural product have been reported. 1,4-Dihydroxynaphthoic acid **7** is a common starting material for these methods. A novel direct synthesis of **1** was achieved from commercially available materials. The key step of our synthetic route is the Hauser-Kraus annulation of functionalized phthalide **29** with a known acetal eter **43**. This efficient route will enable the broad evaluation of the antiviral activity of this interesting natural product (**1**). Moreover, this novel synthetic route offers a new pathway to other compounds in *R.*

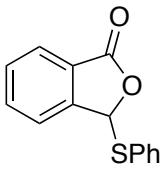
cordifolia family, such as mollugin (**2**), and rubilactone (**3**), depending on the structure of Michael acceptors.

3.4. Experimental

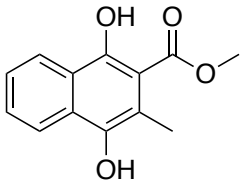
General Procedures

All starting materials were purchased from Sigma-Aldrich; THF was freshly distilled with LAH, other solvents were purchased from Fisher Scientific and used without further purification. All reactions were carried out in flame-dried glassware under argon with dry solvents under anhydrous conditions, unless specified. All yields refer to chromatographically isolated products. Reactions were monitored by thin layer chromatography (TLC) carried out on 0.20 mm silica gel plates using UV light as a visualizing agent and potassium permanganate with heat as developing agents. Silica gel 60Å, particle size 0.032 – 0.063 mm, was used for flash column chromatography. ^{1}H and ^{13}C NMR spectra were acquired on a Varian VXR-300 or Bruker DRX-500 spectrometer. ^{1}H and ^{13}C chemical shifts (δ) are given in ppm relative to the residual protonated solvent peak ($CDCl_3$: $_{\delta}H$ = 7.26 ppm, $_{\delta}C$ = 77.0 ppm; CD_3OD: $_{\delta}H$ = 3.31 ppm, $_{\delta}C$ =49.0 ppm; $(CD_3)_2SO$: $_{\delta}H$ = 2.50 ppm, $_{\delta}C$ = 39.52 ppm; $(CD_3)_2CO$: $_{\delta}H$ = 2.05 ppm, $_{\delta}C$ = 29.84 ppm) as an internal reference. High resolution mass spectra (HRMS) were recorded on an Agilent 6540 QTOF (quadrupole time of flight) mass spectrometer using ESI (electrospray ionization) or APCI (atmospheric-pressure chemical ionization), or EI (electron ionization) on an Agilent 6890 GC/MS.

Selected Experimental, Physical, and Spectral Data

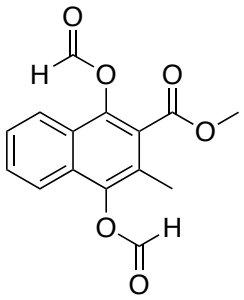


Phthalide (29): To a flame-dried flask with 2-carboxybenzaldehyde **38** (1.6569 g, 11.04 mmol) and PTSA (82.8 mg) in 50 ml benzene solution was added thiophenol (1.35 mL, 13.24 mmol). The resulting mixture was heated up to reflux with the Dean-Stark apparatus setup for 3 h. After the solution was cooled down, the mixture was concentrated *in vacuo*. The recrystallization of the crude product with hexane and ethyl acetate gave phthalide **29** in 80% yield as a white solid; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 7.79 (d, *J* = 7.6 Hz, 1H), 7.77 – 7.61 (m, 3H), 7.53 – 7.48 (m, 3H), 7.33 – 7.20 (m, 2H), 6.73 (s, 1H).

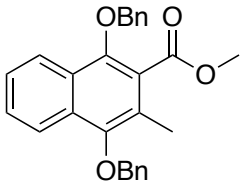


Methyl 1,4-dihydroxy-3-methyl-2-naphthoate (36): To a stirred solution of tert-butanol (1.1389 g, 15.37 mmol) in THF (50 ml) at 0 °C under argon atmosphere was added n-butyllithium (6.15 ml,15.37 mmol) and the mixture was allowed to stir at 0 °C for 30 min to get a lithium tert-butoxide solution. At -78°C, 10 ml of a THF solution of phthalide **29** (1.2009 g, 4.96 mmol) was added slowly into the prepared lithium tert-butoxide solution. The mixture was stirred at -78 °C for another 30 minutes, after which methyl crotonate **37** (0.7444 g, 7.43 mmol) was injected into the reaction mixture at -78 °C. The resulting mixture was stirred at -78 °C for 1 hr and then allowed to warm to room temperature to

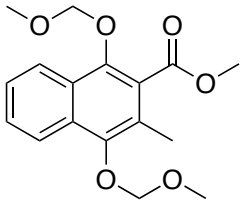
sit overnight. The reaction was quenched with 4 M HCl solution followed by the extraction with ethyl acetate (3 x 60 mL). The organic layer was dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give the crude product. The recrystallization with hexane and diethyl ether afforded **36** in 61% yield as a white solid; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 12.06 (s, 1H), 8.39 (d, *J* = 8.2 Hz, 1H), 8.05 (d, *J* = 8.3 Hz, 1H), 7.70 – 7.58 (m, 1H), 7.56 – 7.44 (m, 1H), 4.71 (s, 1H), 4.01 (s, 3H), 2.54 (s, 3H).



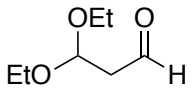
Methyl 1,4-bis(formyloxy)-3-methyl-2-naphthoate (40): To a solution of **36** (0.1131 g, 0.49 mmol) in THF was added DIPEA (0.19 mL, 1.07 mmol) at 0 °C. The acetic formic anhydride was added after 5 minutes. The reaction was monitored by TLC. After no starting material was left, saturated NH_4Cl solution was added to quench the reaction followed by extraction with ethyl acetate. The combined organic layer was dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give the crude product. The residue was purified with flash column chromatography (silica gel, EtOAc:hexane 1:5) to give **40** in 85% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 8.49 (s, 1H), 8.40 (s, 1H), 7.92 – 7.79 (m, 2H), 7.69 – 7.51 (m, 2H), 3.95 (s, 3H), 2.35 (s, 3H).



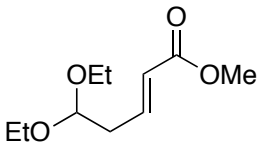
Methyl 1,4-bis(benzyloxy)-3-methyl-2-naphthoate (41): To an acetone solution of **36** (0.3836 mg, 1.65 mmol) and potassium carbonate (1.1414 g, 8.26 mmol) was added benzyl bromide (0.98 mL, 8.26 mmol). The resulting mixture was heated to 60 °C for reaction. After 48 h, the solution was cooled down followed by filtration and concentration *in vacuo*. The residue was extracted with water and ethyl acetate (3 X 30 mL). The combined organic layer was dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo* to give the crude product, which was purified with flash column chromatography (silica gel, EtOAc:hexane 1:5) to give **41** in 65% yield; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 8.17 – 8.08 (m, 2H), 7.62 – 7.31 (m, 12H), 5.14 (s, 2H), 4.98 (s, 2H), 3.91 (s, 3H), 2.41 (s, 3H).



Methyl 1,4-bis(methoxymethoxy)-3-methyl-2-naphthoate (42): Compound **42** was prepared according to the previous procedure of compound **40**.; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 8.10 (dd, J = 11.1, 8.4 Hz, 2H), 7.53 (t, J = 8.6 Hz, 2H), 5.13 (d, J = 14.3 Hz, 4H), 3.97 (s, 3H), 3.67 (s, 3H), 3.62 (s, 3H), 2.40 (s, 3H).

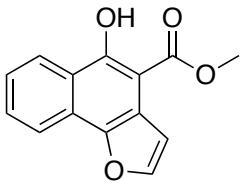


3,3-Diethoxypropanal (44): To a solution of methyl 3,3-diethoxypropanoate **44** (1.9113 g, 10.05 mmol) in anhydrous dichloromethane (5 mL) was added a solution of DIBAL (78.8 mL of a 1.0 M solution in toluene, 15.75 mmol) over 20 minutes at -78 °C. After addition, the reaction was stirred at -78 °C for an additional 45 minutes after which methanol (0.3 mL) was added. The reaction was then warmed to room temperature and stirred for 30 minutes. To the reaction was sequentially added water (~5 mL), stirred 10 minutes, and then solid Na_2SO_4. Upon stirring for 10 minutes the reaction mixture was filtered and concentrated *in vacuo*. The residue was purified with flash column chromatography (silica gel, Et_2O:pentane 1:5) to give **44** in 74% yield as a colorless oil; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 9.75 (t, J = 2.3 Hz, 1H), 4.96 (t, J = 5.5 Hz, 1H), 3.61 (ddq, J = 40.7, 9.3, 7.1 Hz, 4H), 2.73 (dd, J = 5.5, 2.3 Hz, 2H), 1.21 (t, J = 7.0 Hz, 6H).



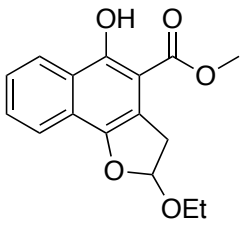
Methyl (*E*)-5,5-diethoxypent-2-enoate (43): Into a flame dried flask with a stir bar was placed sodium hydride (0.2175 g, 5.43 mmol) and 20 ml dry THF and the slurry is cooled to 0 °C. Trimethyl phosphonoacetate (1.0598 g, 5.82 mmol) is then added dropwise and the mixture warmed to room temperature and stirred for 30 min. Compound **44** (0.6975 g, 4.77 mmol) was then added as a solution in 5 ml THF. After stirring for 30 minutes, the reaction was quenched with saturated aqueous NH_4Cl solution and extracted with EtOAc. (3 x 50 mL) The combined organic layer was dried over Na_2SO_4, filtered and

concentrated *in vacuo*. The residue was purified with flash column chromatography (silica gel, EtOAc:hexane 1:20) to give **43** in 72% yield as a light yellow oil; ^{1}H NMR (300 MHz, $CDCl_3$) δ = 6.93 (dt, *J* = 15.1, 7.2 Hz, 1H), 5.98 – 5.85 (m, 1H), 4.58 (t, *J* = 5.6 Hz, 1H), 3.73 (s, 3H), 3.71 – 3.56 (m, 2H), 3.56 – 3.45 (m, 2H), 2.53 (ddd, *J* = 7.2, 5.5, 1.6 Hz, 2H), 1.21 (t, *J* = 7.0 Hz, 6H).



Furomollugin (1): To a stirred solution of tert-butanol (0.30 g, 4.1 mmol) in THF (5 ml) at 0 °C under argon atmosphere was added n-butyllithium (1.64 ml, 4.1 mmol) and the mixture was allowed to stir at 0 °C for 30 min to get a lithium tert-butoxide solution. At -78°C, 2 ml of a THF solution of phthalide **29** (0.33 g, 1.36 mmol) was added slowly into the prepared lithium tert-butoxide solution. The mixture was stirred for another 30 min at -78 °C, after which the acetal ester **43** (0.41 g, 2.0 mmol) was injected into the reaction mixture at -78 °C. The resulting mixture was stirred at -78 °C for 1 hr and then allowed to warm to room temperature to sit overnight. The reaction was quenched with 4 M HCl solution followed by the extraction with ethyl acetate. The organic layer was dried over $MgSO_4$ and evaporated to give crude product, which was purified by flash column chromatography on silica gel to give **47** in 42% yield and **1** in 6% yield. The 8 was converted to 1 by refluxing in toluene with PTSA in 93% yield; ^{1}H NMR (400 MHz, $CDCl_3$) δ = 12.27 (1H, s), 8.47 (1H, d, *J* = 8.4 Hz), 8.20 (1H, d, *J* = 8.3 Hz), 7.82 – 7.67 (2H, m), 7.60 – 7.48 (1H, m), 7.19 (1H, d, *J* = 2.2 Hz), 4.08 (3H, s); ^{13}C NMR (101 MHz,

$CDCl_3$) δ 172.0, 159.0, 144.4, 144.3, 130.1, 125.1, 124.9, 124.9, 122.9, 119.8, 119.7, 109.3, 99.2, 52.3.; HRMS (ESI-TOF) calcd for $C_{14}H_{11}O_4$ $[M + H]^+$ 243.0652, found 243.0658.



Methyl 2-ethoxy-5-hydroxy-2,3-dihydronaphtho[1,2-*b*]furan-4-carboxylate (47): 1H NMR (300 MHz, $CDCl_3$) δ = 11.81 (s, 1H), 8.37 (d, J = 8.5 Hz, 1H), 7.92 (d, J = 8.3 Hz, 1H), 7.61 (t, J = 7.5 Hz, 1H), 7.49 (t, J = 7.7 Hz, 1H), 5.87 (dd, J = 6.7, 2.5 Hz, 1H), 4.09 – 3.97 (m, 1H), 3.96 (s, 3H), 3.77 – 3.57 (m, 2H), 3.51 – 3.38 (m, 1H), 1.26 (t, J = 7.0 Hz, 3H).

3.5. References

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CHAPTER 4.

SUZUKI-MIYAURA REACTIONS OF ARYL HALIDES USING SUPPORTED PALLADIUM NANOPARTICLES

4.1. Introduction

Transition-metal compounds are extremely useful as catalysts for organic transformations which involve the construction of complex organic molecules such as pharmaceutical derivatives, natural products, and molecular materials.[1] Among many transition metals used for organic synthesis, palladium catalysts and reagents are particularly useful and versatile.[2] There are several features regarding palladium catalysts and reagents listed below. Most importantly, palladium catalysts offer an abundance of possibilities for carbon–carbon bond formation. No other transition metals can offer such versatile methods of the carbon–carbon bond formations as palladium. In 2010, the Noble prize was awarded to Heck, Negishi, and Suzuki for the development of palladium-catalyzed cross-coupling reactions in organic synthesis.[3] The Heck reaction,[4] the Suzuki-Miyaura reaction,[5] the Negishi coupling reaction,[6] and the Sonogashira reaction[7] are now among the most commonly used organometallic reactions in both industry and academia. Organopalladium reactions have been employed in a number of complex natural product syntheses as well as pharmaceutical syntheses.[8] In addition, valuable organopalladium reactions developed by Trost,[9] Tsuji,[10] Stille,[11] Kumada,[12] and Larock[13] occupy a significant place in the "palladium toolbox", as shown in Figure 1.

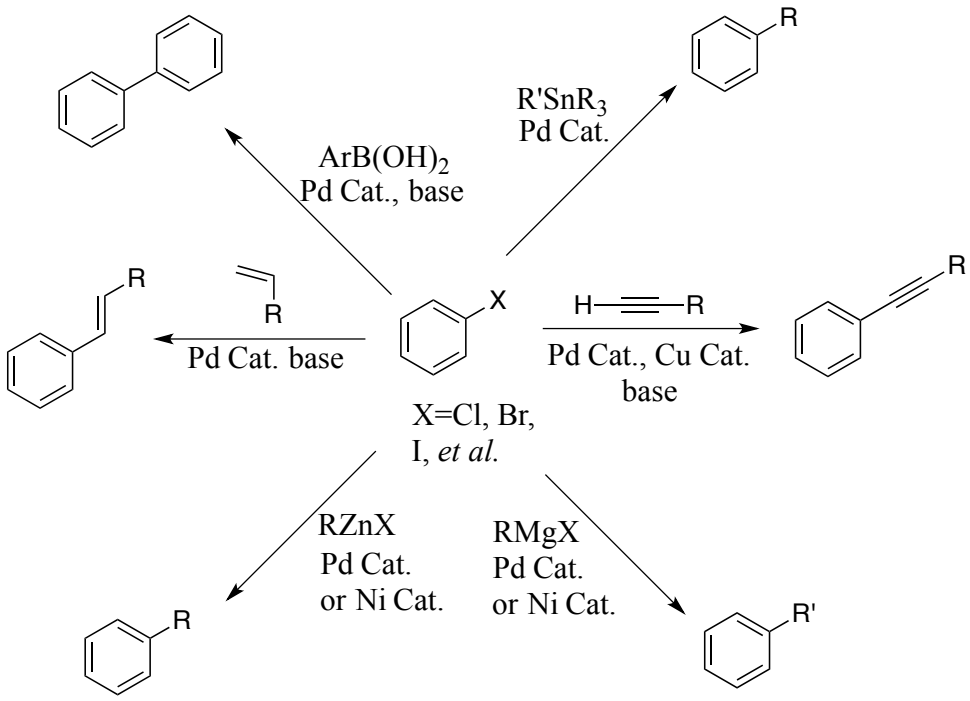


Figure 1. Palladium toolbox.

The second important feature of palladium catalysts and reagents is the tolerance with a variety of functional groups such as carbonyl and hydroxyl groups. Palladium-catalyzed reactions can be carried out with these functional groups smoothly. Although reactions involving palladium should be carried out carefully, palladium catalysts are not very sensitive to oxygen and moisture. Moreover, the toxicity of palladium has caused no serious problem so far. A number of industrial processes, particularly for the production of fine chemicals based on palladium-catalyzed reactions, have been developed and are currently being utilized. This fact reflects the advantages of using palladium catalysts commercially.[2]

Although homogeneous catalysts are suitable in small scale applications, the evolution to heterogeneous palladium catalysts has become a rapidly emerging area of

research.[14] Heterogeneous palladium catalysts are easy to recycle and separate from the product mixture. Additionally, there are opportunities to adjust the catalyst through variation in types of supports. Significantly, this approach avoids the need for expensive ligands. A number of solid materials, such as graphene,[15] titania,[16] polymers,[17] metal organic frameworks (MOFs),[18] mesoporous carbon,[19] and magnetic nanoparticles[20] have been employed as supports for palladium. Bimetallic catalysts have been devised, which in certain cases facilitate the separation of the catalyst.[21]

The heterogeneous palladium catalyst developed by Datye and coworkers is convenient to prepare from the reaction of palladium acetate with methanol at ambient temperature.[22] Additionally, the method of preparation is very flexible with regard to catalyst support, since the support is simply added as the reaction proceeds. Moreover, the particle size is 1.5 nm and no additional functionalization is required.

The Datye catalyst was initially evaluated in the Suzuki-Miyaura reaction. The Suzuki-Miyaura reaction is one of the most important and powerful synthetic methods for the construction of carbon-carbon bonds, in particular for the formation of unsymmetrical biaryls.[23] This reaction involves the palladium-mediated coupling of aryl boronic acids or their corresponding esters **1** with aryl halides **2**. The mechanism of this reaction is shown in Figure 2.[24] The first step is the oxidative addition of palladium catalyst **4** to the halide **2** to form the organopalladium species **5**. The treatment with bases gives intermediate **6**, which is converted to intermediate **7** after transmetalation with boron-ate complex **8**. Reductive elimination of the desired product **3** restores the original palladium catalyst **4** which completes the catalytic cycle.

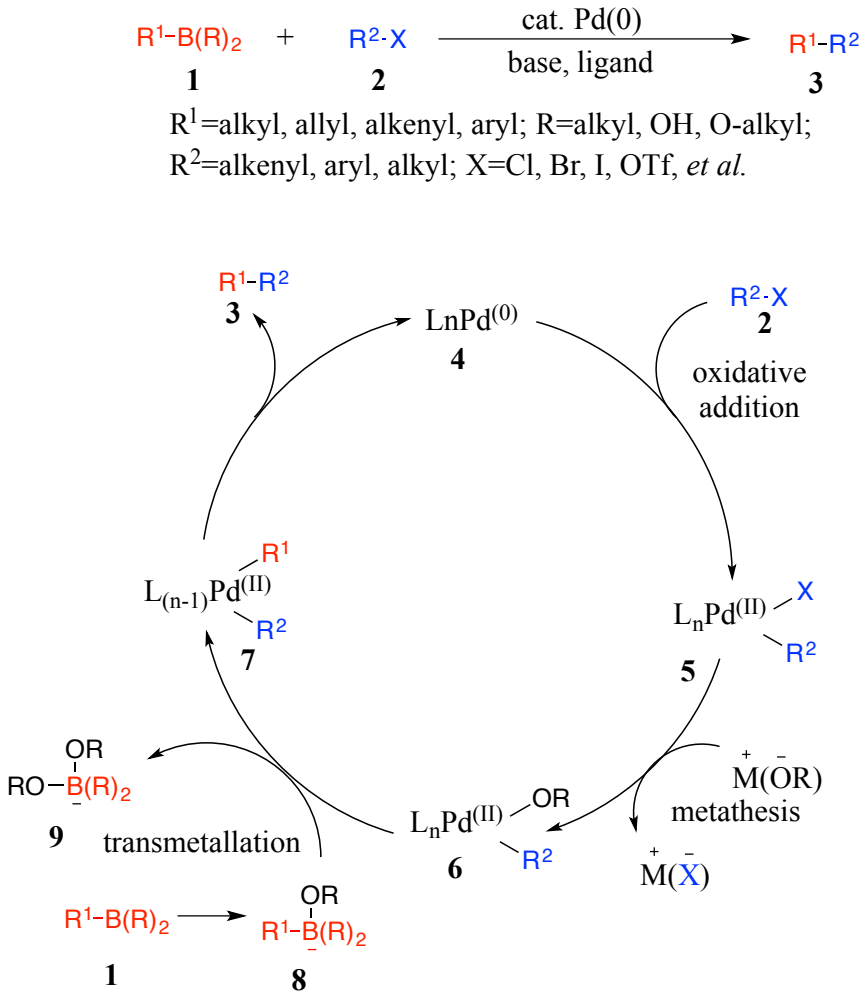
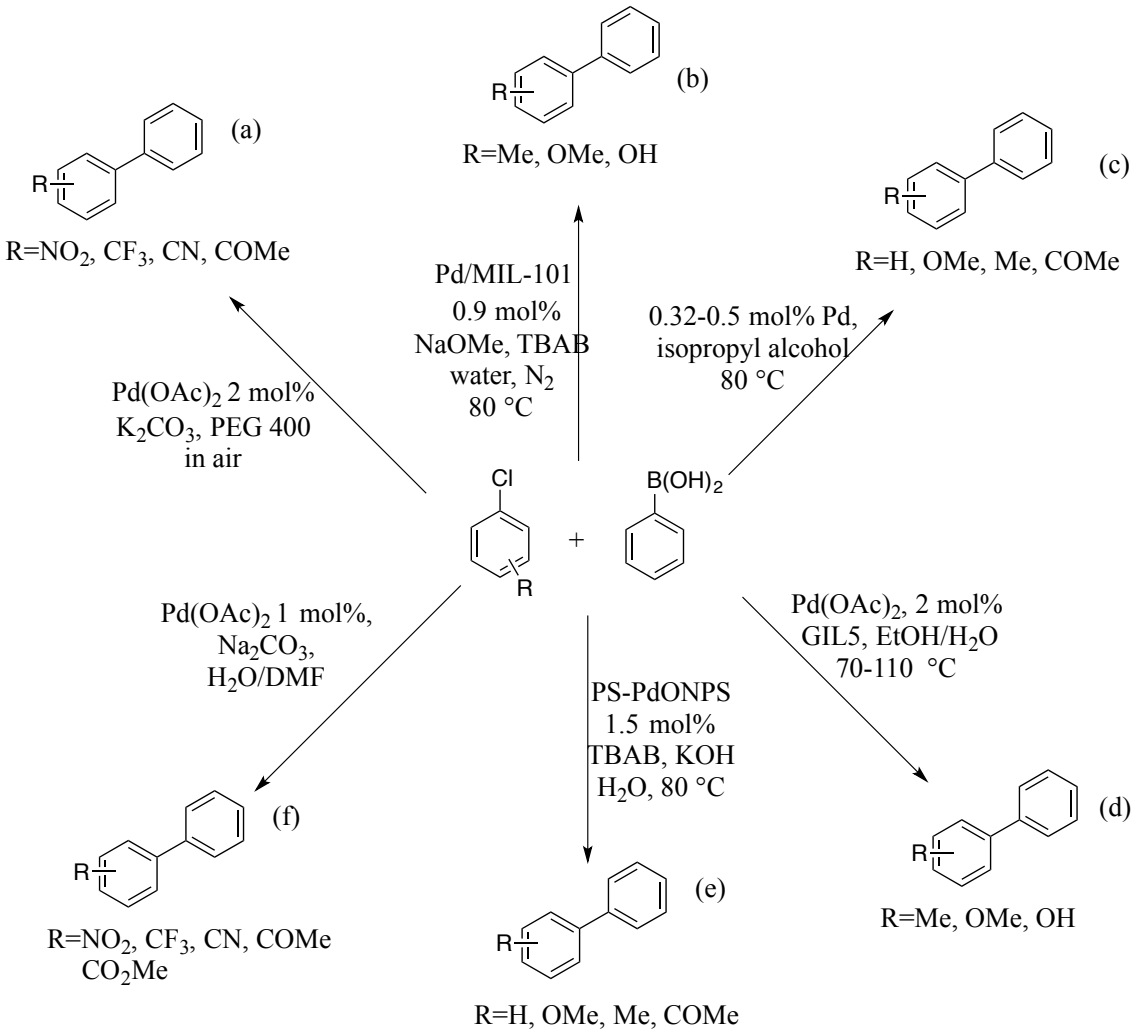


Figure 2. Mechanism of the Suzuki-Miyaura reaction.

Almost every reported palladium catalyst affords high yields of coupling products with aryl iodides or aryl bromides, but the yields with aryl chlorides are frequently much lower. However, the use of aryl chlorides is desirable because they are inexpensive and readily available from a practical point of view. Furthermore, most of the reported coupling reactions were carried out in organic solvents. From a sustainable chemistry viewpoint, water is cheap, nontoxic, and readily available green reaction medium for organic synthesis. Therefore, the development of heterogeneous catalysts that can

activate aryl chlorides and facilitate the coupling reactions of aryl chlorides in aqueous media is highly desirable.

A number of researchers have reported nanoparticle-mediated reactions of boronic acids with aryl halides. Liu and coworkers generated palladium nanoparticles in situ in PEG400 under aerobic conditions. Using 2 mole percent of palladium at 45 °C in PEG400, they achieved high yields of Suzuki coupling products with aryl chlorides and phenylboronic acid, as shown in Scheme 1(a).[25] Jiang and coworkers stabilized palladium nanoparticles on a metal-organic framework (MIL-101). Reaction of this catalyst at 80 °C in water with aryl chlorides provided coupling products in high yields, as seen in Scheme 1(b).[26] Yang and coworkers synthesized silica nanoparticles functionalized with N-heterocyclic carbenes. Using 0.3 mole percent palladium at 80 °C, they achieved very good yields of products from aryl chlorides, as shown in Scheme 1(c).[27] Li and coworkers prepared palladium-containing micelles stabilized by guanidinium ionic liquids (GIL5). Using 2 mole percent palladium at 100 °C afforded good to excellent yields (41% to 99%) of products from aryl chlorides, as exhibited in Scheme 1(d).[28] Ohtaka and coworkers recently reported linear polystyrene supported palladium catalysts that functioned as homogeneous catalysts in solution. Using this "catch-release" strategy they obtained very good yields of products from aryl chlorides and bromides, as shown in Scheme 1(e).[29] In Scheme 1(f), Liu and coworkers used an aqueous DMF solution and palladium catalysts at 100 °C and reported good to very good yields of Suzuki products with aryl chlorides.[30]



Scheme 1. Reported nanoparticle-mediated reactions.

Although utilizing most of these reported catalysts afforded high yields for the Suzuki-Miyaura reaction of boronic acids and aryl chlorides, none of the nanoparticle preparation protocols compared to the Datye catalyst in terms of operational convenience. Moreover, only a few of the catalyst systems described above achieved the goal of green chemistry to use water as the solvent for reactions. We utilized the Datye catalyst to find the best aqueous catalyst system for the Suzuki-Miyaura coupling of boronic acids and aryl chlorides.

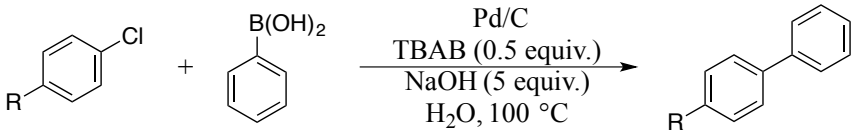
4.2. Results and Discussion

To test the possibility of using the Datye catalyst for the Suzuki-Miyaura reaction, we first compared the catalytic ability of this supported palladium on carbon nanoparticle catalyst (5 wt%) with commercially available palladium on carbon catalyst, which was purchased from Kawaken Fine Chemicals (10 wt%, 53.5% water content). Using 0.2 mole percent of the Datye catalyst with tetrabutylammonium bromide and sodium hydroxide at 100 °C in water produced the desired coupling product in 38% yield from 4'-chloro-acetophenone and phenylboronic acid, as shown in Table 1, Entry 2.[31] This yield was higher than the yield of the coupling reaction catalyzed with the commercially available palladium on carbon catalyst (Table 1, Entry 1). Furthermore, reaction of 4-chloroanisole at 100 °C in water with 2 mole percent of Datye catalyst loading afforded the desired coupling product in 42% yield, comparing to 34% yield with the catalyst purchased from Kawaken (Table 1, Entry 3-4).

The comparison test in Table 1 shows that the Datye catalyst is a better palladium catalyst for the Suzuki-Miyaura reaction in aqueous solution than the commercial catalyst. Moreover, the Datye catalyst requires a lower catalyst loading for the reaction compared to other reported catalysts. To further demonstrate the utility of this palladium catalyst for the Suzuki reaction, a number of aryl chlorides were reacted with representative commercially available boronic acids, which is shown in Table 2. We found that increasing the reaction time from two hours to 12 hours increased the reaction yield of 4'-chloro-acetophenone and phenylboronic acid to 72% from 38%, as seen in Table 2, Entry 1. Some other aryl chlorides were used for the Suzuki reaction with

phenylboronic acid and the Datye catalyst, as exhibited in Table 2, Entry 2-4. Using electron deficient aryl chlorides, such as methyl 2-chlorobenzate and methyl 4-chlorobenzoate, gave the desired coupling products **11** in 20% yield and **12** in 22% yield. Increasing the reaction time of electron efficient aryl chloride 4-chloroanisole did not increase the reaction yield, which was 42%.

Table 1. Comparison of catalytic activity.

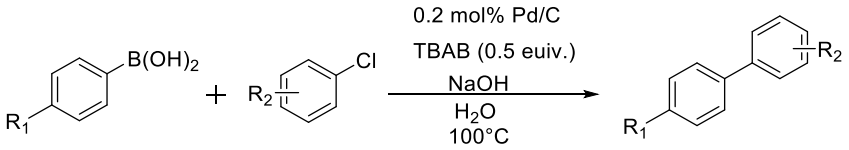


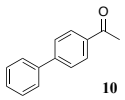
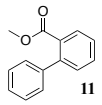
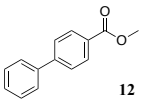
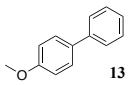
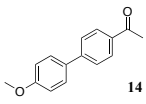
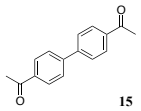
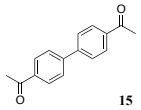
Entry	R	Pd/C source	Pd/C (%)	Time (h)	Yield (%)
1[a]	Ac	Kawaken	0.2	2	27
2[a]	Ac	Datye	0.2	2	38
3	OMe	Kawaken	0.2	6	34
4	OMe	Datye	0.2	6	42

[a.] 2.5 equiv. NaOH was used.

Since 4'-chloroacetophenone gave us the best yield among these aryl chlorides described above, we used this compound for the Datye catalyst catalyzed Suzuki reaction with a variety of boronic acids, as seen in Table 2, Entry 5-7. Using 4-methoxy-phenylboronic acid produced the product in a 45% yield at 100 °C in water, as seen in Table 2, Entry 5. The Suzuki reaction of 4-acetylphenylboronic acid with 4'-chloroacetophenone catalyzed with the Datye catalyst only gave 9% yield with 0.2 mole percent catalyst loading (Table 2, Entry 6). The yield was increased to 25% (based on NMR spectra) with 2 mole percent catalyst loading (Table 2, Entry 7).

Table 2. Suzuki-Miyaura reaction of aryl chlorides catalyzed with the Datye catalyst.



Entry	R_1	R_2	Pd (%)	Time (h)	Boronic acid (equiv)	NaOH (equiv)	Product	Yield (%)
1	H	4-Ac	0.2	12	1.1	2.5	 **10**	72
2	H	2-CO_2Me	0.2	12	1.1	2.5[a]	 **11**	20
3	H	4- CO_2Me	0.2	12	1.1	2.5[a]	 **12**	22
4	H	4-OMe	2	12	1.5	5	 **13**	42
5	4-OMe	4-Ac	0.2	2	1.1	2.5	 **14**	45
6	4-Ac	4-Ac	0.2	2	1.1	2.5	 **15**	9
7	4-Ac	4-Ac	2	2	1.1	2.5	 **15**	25

[a] The KF was used as base.

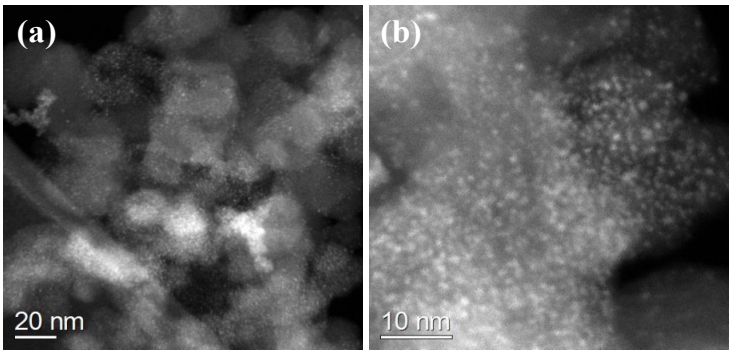


Figure 3. HAADF-STEM images at low (a) and high (b) magnifications of the Datye catalyst.

The Datye catalyst prepared by the Datye group consists of small, highly dispersed 1-3 nm palladium particles, as depicted in Figure 3. The size and shape of the palladium nanoparticle catalyst was determined by Datye group by using a high angle annular dark field (HAADF) scanning transmission electron microscopy (STEM) after the catalytic experiments, as illustrated in Figure 4. Although the utility of this palladium nanoparticle catalyst for the Suzuki coupling in water media was proven, the sintering of palladium particles was observed after reaction with aryl chlorides. In Figure 4(a) and 4(b), the spent catalyst sample from reaction of 4'-chloroacetophenone and 4-methoxyphenylboronic acid in dimethylformamide (DMF) shows significant sintering of palladium particles (over 20 nm) when the reaction was performed using dimethylformamide as the solvent. In contrast, the spent sample in Figure 4(c) shows less sintering of palladium particles when the reaction was performed using water as the solvent. Figures 4(c) and 4(d) appear to show a bimodal distribution, with palladium particles over 20 nm (d) and palladium particles between 1-3 nm (c).

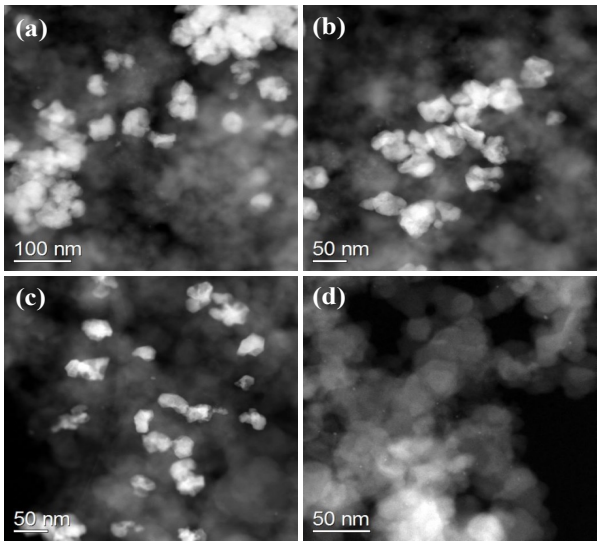
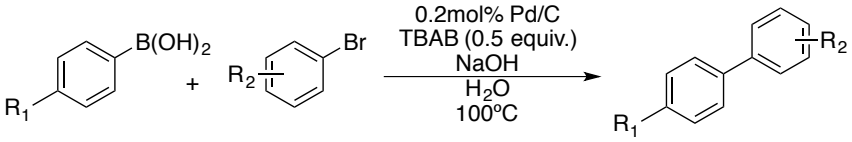
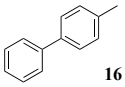
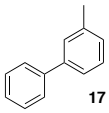
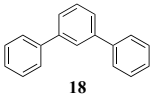


Figure 4. HAADF-STEM images of the sample (a,b) after reaction in DMF, and the sample (c,d) after reaction in water.

To further test the activity of catalyst, coupling reactions of aryl bromides with boronic acids were carried out, as shown in Table 2. The reaction of 4-bromotoluene and phenylboronic acid with 0.2 mole percent of the Datye catalyst, tetrabutylammonium bromide and sodium hydroxide at 100 °C in water produced the desired product **16** in 90% yield after column chromatography separation, as seen in Table 3, Entry 1. This result indicated that the Datye catalyst exhibits excellent catalytic activity for the Suzuki reaction of aryl bromides. Using 3-bromotoluene in the same condition afforded compound **17** in 89% yield, as shown in Table 3, Entry 2. In Table 3, Entry 3, utilizing 1,3-dibromobenzene with 2.2 equivalent of phenylboronic acid gave m-terphenyl **18** in 49% yield.

Table 3. Suzuki-Miyaura reaction of aryl bromides catalyzed with the Datye catalyst.



Entry	R_1	R_2	Pd (%)	Time (h)	Boronic acid (equiv)	NaOH (equiv)	Prodct	Yield (%)
1	H	4-Me	0.2	12	1.1	2.5	 16	90
2	H	3-Me	0.2	12	1.1	2.5	 17	89
3	H	3-Br	0.2	12	2.2	2.5	 18	49

After the catalytic experiment of aryl bromides, the catalyst was collected to check the size and shape by HAADF-STEM. The images are shown in Figure 5. All the spent samples from the Suzuki reaction of aryl bromides show a significant reduction in sintering of palladium, compared to the samples from reaction of aryl chloride. The size of a recycled pallidum nanoparticle catalyst is around 5 nm after reaction according

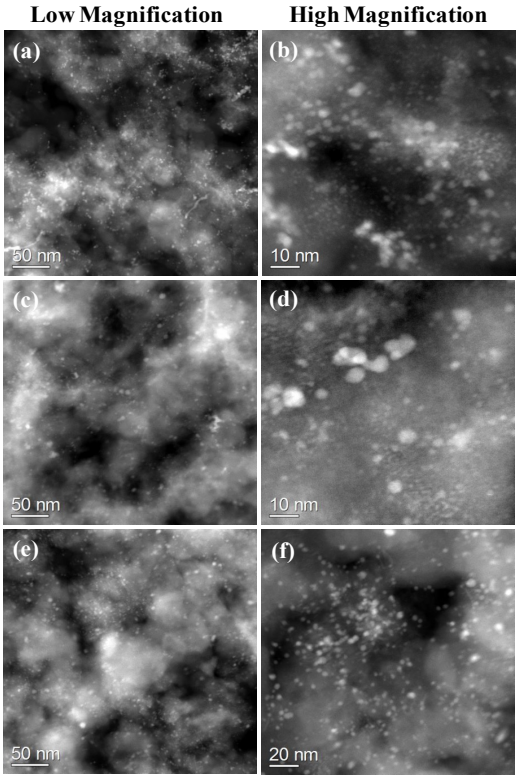


Figure 5. HAADF-STEM images at low (left) and high (right) magnifications of the samples from Table 3: Entry 1 (a, b); Entry 2 (e,f); Entry 3 (c, d).

to the HAADF-STEM images. This is not a significant change when compared to the size of the as-prepared catalyst, which is 1.5 nm.

The low yield of the catalyzed Suzuki-Miyaura reaction of aryl chlorides may be explained by the size change of particles during reactions. The sintering of palladium nanoparticles, when the reactions were performed with aryl chlorides, decreased the catalytic activity of the Datye catalyst. In contrast, a change in size of particles was not observed when reactions were performed with aryl bromide. The Datye catalyst can give a high yield of desired coupling product from aryl bromides.

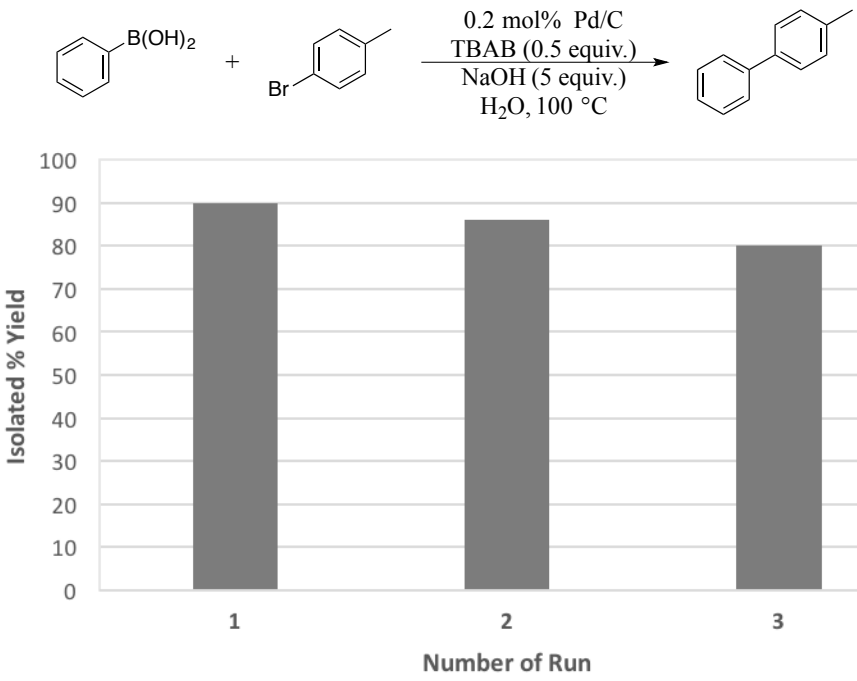


Figure 6. Recyclability test of the Datye catalyst.
The condition for each run: phenylboronic acid (0.1341 g, 1.1 mmol), 4-bromo toluene (0.1710 g, 1.0 mmol), tertbutylammonium bromide (0.1612 g, 0.5 mmol), sodium hydroxide (0.1 g, 2.5 mmol), catalyst (5 wt% Pd/C, 0.0043 g, 2.0 μmmol, recycled), water (5 mL), temperature: 100 °C, time: overnight.

Because the Datye catalyst was recovered intact in the reaction with aryl bromides, the recyclability of this catalyst can be studied. The reaction of 4-bromotoluene and phenylboronic acid with 0.2 mole percent palladium catalyst loading was carried out for the test, as shown in Figure 6. After three recycles, the catalyst was still active and the product yield was 80%, compared with 90% for first run. As the catalyst can be recycled for three consecutive runs in water, the Dayte catalyst represents a sustainable variant of palladium catalyzed coupling chemistry. Inductively Coupled Plasma Mass Spectrometry (ICP-MS) analysis revealed that only 0.3% of palladium species leached into the solution, which indicated that the Datye catalyst is a heterogeneous catalyst as expected.

4.3. Conclusions

The Datye catalyst can be used as a heterogeneous catalyst for the Suzuki-Miyaura reaction of aryl chlorides and boronic acids in water. The low reaction yields of aryl chlorides was presumably caused by the sintering of palladium particles. This catalyst also showed an excellent activity for the coupling reactions of aryl bromides. Even after several recycles, the activity was not decreased. There was no sintering observed when the reactions were performed with aryl bromides. In addition, the preparation of this palladium nanoparticle catalyst is really simple compared with other reported catalysts. Excellent catalytic activity, simple preparation and environmentally friendly conditions, are all features that make the Datye catalyst a good candidate for palladium catalysts in industry.

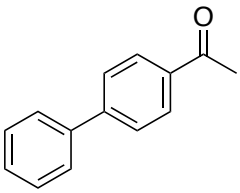
4.4. Experimental

General Procedures

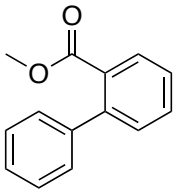
All starting materials were purchased from Sigma-Aldrich. All yields refer to chromatographically isolated products. Reactions were monitored by thin layer chromatography (TLC) carried out on 0.20 mm silica gel plates using UV light as a visualizing agent and potassium permanganate with heat as developing agents. Silica gel 60Å, particle size 0.032 – 0.063 mm, was used for flash column chromatography. ^{1}H spectra were acquired on a Varian 400 MR or Bruker DRX-500 spectrometer. ^{1}H chemical shifts (δ) are given in ppm relative to the residual protonated solvent peak ($CDCl_3$: $_{\delta}H$ = 7.26 ppm) as an internal reference. Recycled samples were dispersed in ethanol and mounted on holey carbon grids for examination in a JEOL 2010F 200 kV transmission electron microscope. Images were recorded in high angle annular dark field (HAADF) mode.

Selected Experimental, Physical, and Spectral Data

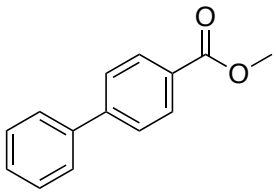
The preparation of the Datye catalyst: The catalyst was prepared by the Datye group. A solution of Pd acetate in methanol was ultrasonicated for 40 min to completely dissolve the Pd acetate, and the solution was added to Vulcan carbon XC-72 to obtain a Pd loading of 5 wt%. A Buchi rotary evaporator was used to gently remove methanol from the sample at 40 °C , and the dried product was collected.



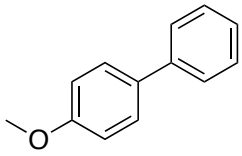
4-Acetylbiphenyl (10): The preparation of **10** is representative. To a sealed tube, phenylboronic acid (0.1341 g, 1.1 mmol) and 4-chloroacetophenone (0.1456 g, 1.0 mmol) was added into a 5 ml water solution with tertbutylammonium bromide (0.1619 g, 0.5 mmol) and sodium hydroxide (0.1000 g, 2.5 mmol). After the Datye catalyst (0.0043 g, 0.2 mol%) was added, the reaction mixture was heated to reflux for 12 hours. After the reaction was cooled down to room temperature, the solution was transferred to a centrifuge tube. 10 mL of methylene chloride was then added followed by 5 min centrifuge. Then the clear organic layers were transferred to a separatory funnel, followed by washing with water (2 x 5 mL), drying with anhydrous $MgSO_4$, filtration and concentration *in vacuo*. The desired product **1** was purified by flash column chromatography (silica gel, EtOAc:hexane 1:10), in 72% yield as a white solid; ^{1}H NMR (400 MHz, $CDCl_3$) δ = 8.07 – 8.00 (m, 2H), 7.73 – 7.59 (m, 4H), 7.52 – 7.38 (m, 3H), 2.64 (s, 3H).



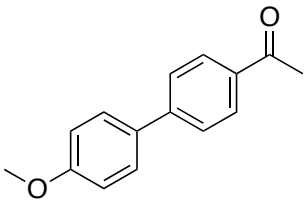
Methyl biphenyl-2-carboxylate (11): KF was used as a base instead of NaOH. The yield is based on NMR. ^{1}H NMR data was identical to corresponding literature NMR spectra.[32]



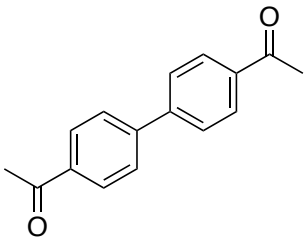
Methyl biphenyl-4-carboxylate (12): KF was used as a base instead of NaOH. ^{1}H NMR (400 MHz, $CDCl_3$): δ = 8.15 – 8.07 (m, 2H), 7.71 – 7.59 (m, 4H), 7.49 – 7.44 (m, 2H), 7.42 – 7.36 (m, 1H), 3.95 (s, 3H).



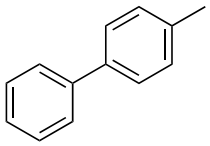
4-Methoxybiphenyl (13): ^{1}H NMR (400 MHz, $CDCl_3$): δ = 7.54 – 7.44 (m, 4H), 7.42 – 7.35 (m, 2H), 7.28 – 7.23 (m, 1H), 6.98 (d, *J* = 8.3 Hz, 2H), 3.86 (s, 3H).



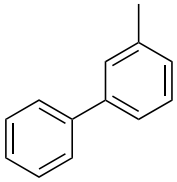
1-(4'-Methoxybiphenyl-4-yl)ethanone (14): ^{1}H NMR (400 MHz, $CDCl_3$) δ = 8.02 - 7.98 (m, 2H), 7.66 - 7.61 (m, 2H), 7.59 - 7.54 (m, 2H), 7.02 - 6.97 (m, 2H), 3.85 (s, 3H), 2.62 (s, 3H).



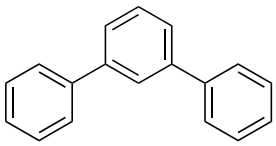
4,4'-Diacetylbiphenyl (15): ^{1}H NMR (500 MHz, $CDCl_3$) δ = 8.06 – 8.07 (m, 4H); 7.72 – 7.73 (m, 4H), 2.66 (s, 6H).



4-Methylbiphenyl (16): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.67 – 7.56 (m, 2H), 7.54 – 7.40 (m, 4H), 7.40 – 7.28 (m, 1H), 7.29 – 7.22 (m, 2H), 2.41 (s, 3H).



3-Methylbiphenyl (17): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.64 – 7.54 (m, 2H), 7.48 – 7.37 (m, 4H), 7.32 (td, *J* = 7.5, 2.1 Hz, 2H), 7.15 (d, *J* = 7.5 Hz, 1H), 2.41 (s, 3H).



m-Terphenyl (18): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.85 (d, *J* = 2.0 Hz, 1H), 7.69 (d, *J* = 7.6 Hz, 4H), 7.63 – 7.60 (m, 2H), 7.57 – 7.46 (m, 5H), 7.41 (t, *J* = 7.4 Hz, 2H).

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CHAPTER 5.

GENERAL CONCLUSIONS

In the first chapter, several small molecule inhibitors towards Porcine reproductive and respiratory syndrome virus (PRRSV) were developed. Natural 1-(E)-Atractylodinol was successfully synthesized in seven steps, which showed 100% inactivation of PRRSV in in vitro test. This is the first synthetic route towards 1-(E)-Atractylodinol. Based on the structure of 1-(E)- Atractylodinol, more than fifteen analogs were designed and synthesized. Among all the synthesized analogs, nine of them showed more than 90 percent virus netralization. Specifically, one dimethyl amino analog is the most cost efficient product to synthesize in order to make animal testing feasible in the near future.

Chapter 2 focused on developing a synthetic approach to the psoracorylifols. The unique 6,8-dioxabicyclo[3.2.1]octane skeleton can be formed concisely by utilizing ketal formation as a key step. The overall yield is 40% in 5 steps. This route will enable the analysis of psoracorylifols in order to better understand the mechanism of action of this novel family of natural products.

Chapter 3 describes a novel direct synthetic route towards furomollugin. The method constructs furomollugin from commercially available materials via the Hauser-Kraus annulation of a functionalized phthalide and a Michael acceptor. This synthetic route will enable a broad evaluation of the biological activities of furomollugin and offer a new pathway towards other compounds in this family.

The last chapter discusses the utility of the Datye catalyst for the Suzuki-Miyaura reaction of aryl chlorides in aqueous solution. As a novel heterogeneous catalyst, the Datye catalyst has several advanced features compared to other palladium catalysts, such as excellent catalytic activity, simple preparation and environmentally friendly conditions. All these features will make the Datye catalyst a great substantial palladium catalyst in industry.