Synthesis of α-carboline analogs utilizing Retrosynthesis and Organometalic Reactions and Examination of the Optimal Conditions for the Bischler-Napieralski Reaction

Tom-André Fredriksen

Master Thesis in Chemistry

Department of Chemistry, University of Bergen

Laboratory for Drug Discovery in Neuroscience (LDDN), Brigham and Woman's Hospital and Harvard Medical School

September 2020

Acknowledgment

I would like to thank my supervisor Hans-René Bjørsvik for his constant support and guidance throughout the project, as well as giving me the opportunity to join his research group and work in his laboratory.

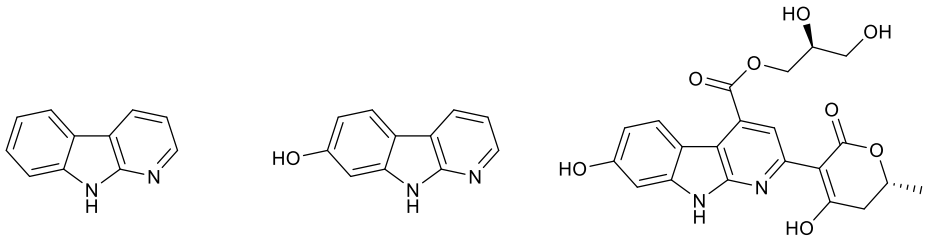
A special gratitude goes out to Davide Cirillo, Francesco Angelucci, Eirin Alme, Frida Johanne Lundevall and Sara Barranco Campos for always being available to help me improve the synthesis, based on their knowledge and experience. Additionally, I would like to thank Bjarte Holmelid, Cecilie Sævdal Dybsland, Marit Bøe Vaage and Nils Åge Frøystein for teaching me how to use the instruments imperative for either analysis or purification of the performed reactions.

I want to thank Dr. Kevin Hodgetts for letting me join his research team, and for always having useful solutions to any problem I faced. Thank you to all members of the LDDN research team for letting me feel welcome to the new lab, the weird lunch conversations, and the great atmosphere at the lab, which made my brief period there a joyful and educational experience.

Finally, I want to thank Stian L. Årvik for being a great co-worker, roommate, and friend. You always had interesting and inventive suggestions whenever we faced problems. Thank you for making tough days at the laboratory more enduring with quirky jokes and your positive mood.

Abstract

Pyrido[2,3]indoles (α-carbolines), have shown interesting properties regarding treatment of neurodegenerative disorders like Alzheimer's and Parkinson's disease, as well as antitumor and antiviral effects [1].This thesis attempts to discover a synthetic pathway for the natural product mescengricin (**3**), by applying retrosynthesis and known organometallic reactions, like Suzuki-Miyuara coupling and palladium catalysed borylation. In order to efficiently develop a synthetic route for mescengricin, modelling reactions applying less substituted compounds was performed, resulting in α-carboline (**1**) with high yield, and 9H-pyrido[2,3-b]indol-7-ol (**2**) with poor yield.



1 a-carboline

2 9H-pyrido[2,3-b]indol-7-ol

3 Mescengricine

The Bischler-Naperialski reaction, in a three-step synthesis of the natural product Edustomin H has been optimized, using a purified starting material in the design developed by M.Eriksen. By performing the reaction, with a pure compound the yield increased from 67% to 96%.

Index

1. Introduction

This project is centred around carboline structures, primarily focused on α-carbolines, and the development of a reliable synthetic route for carboline analogs. Secondly, will the improved method to synthesize Eudistom H, M. Eriksen reported [2] will be recreated, and the Bischler-Napieralski reaction be compared, using a telescope method to a pure starting material.

1.1 Carbolines

Carbolines are indole alkaloids, and their structures consist of a three-cyclic pyrido indole rings. Like most alkaloids, carbolines can be found in nature and especially in plants. Alkaloids often are of interest due to their diverse and important biological effects on humans and animals [3]. Carbolines possess properties that could be utilized in the war against neurodegenerative disorders and are shown to have interesting antitumor and antiviral effects that could be important in medical chemistry [1]. There are three different carbolines: α, β, and γ- carbolines, each with different properties. α-Carbolines are not as common in the nature as β- and γ- carbolines, however, since α derivatives have the potential of being the lead compound for the treatment of neurodegenerative disorders like Alzheimer's and Parkinson's disease [4] they are of high interest. In particular, mescengricin has received a lot of attention.

Mescengricin

Mescengricin is a pyrido[2,3]indole (α-carboline), and was isolated first from *Streptomyces griseflavus* by Seto *et al* [5]. *Streotomyces griseus* is a species of bacteria that grows in soil, but also has been reported to grow in deep sea sediments as well. *Streptomyces griseus* are most known for their ability to produce antibiotics. Seto *et al*, were able to extract only 3.3 mg of mescingricin, and this amount was used in NMR for structure determination that showed an α-carboline core substituted by a glycerol-ester and a hydroxydihydropyrone-ring. When testing mescingricin´s biological activity, it was discovered that mescingricin exhibits a strong protective activity of neural cells against L-glutamate toxicity [5]. It previously has been reported that overexposure to glutamate can result in neurodegeneration, which effectively leads to disorders that could cause permanent disability or death [6]. Not much else is known about mescingrincin's biological properties due to the low amount isolated. As a result, the discovery of a high yielding synthetic route is of high importance. Access to a synthetic sample will allow confirmation of the structure and additional biological testing.

1.2 Optimization of Bischler-Napieralski reaction

In 1986 T. Hino *et al* reported a three-step synthetic route to create Eudistomin H [7] (figure 1). Later, M. Eriksen successfully performed these experiments telescoped (without purification), and focused on optimizing the conditions in the Bischler-Napieralski reaction using chemometrics. In this project, the reactions performed by M. Eriksen will be recreated, and the product from the first reaction will be purified. By comparing the results using a pure compound to those using a telescoped method, important information will be gained. For example, whether the pollutants effect the reaction, or whether other conditions now are superior.

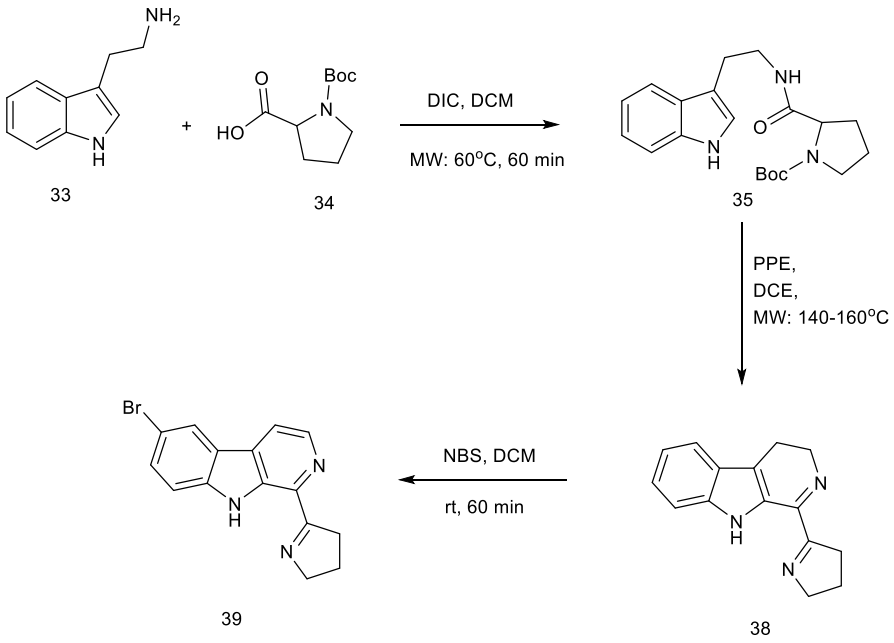


Figure 1 The synthetic route T. Hino et al developed [7].

1.3 Aim of study

The aim of the study is to create a solid and robust synthetic route for α-carboline (**1**) and 9H-pyrido[2,3-b]indol-7-ol (**2**), by utilizing retrosynthesis and organometallic reactions. The methods developed in the modelling of these compounds will be used to synthesize Mescengricin (**3**). If the target compound is not reached, a second objective will be to discover a synthetic route that could be continued in the future.

The optimization of the Bischler-Napieralski reactions performed by M. Eriksen also will been examined. The aim will be to discover if a significant difference in yield is achieved when the reaction is performed with a pure starting material, as compared to a telescoped method.

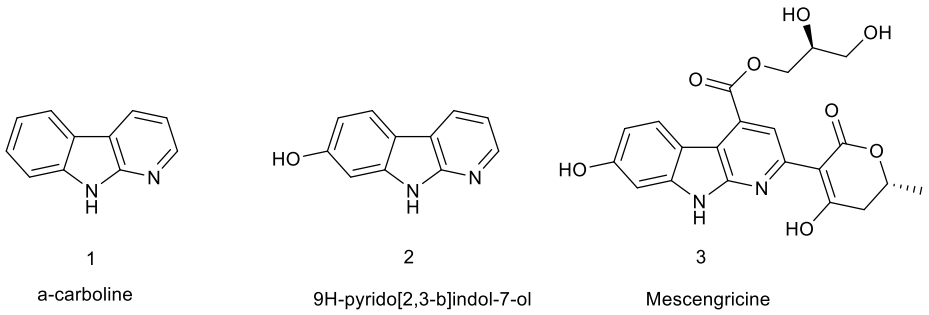


Figure 2 structures of the target compounds: α-carboline, 9H-pyrido[2,3-b]indol-7-ol and mescingricin

2. Theory and instrumentation

Microwave

Over the last decades, environmentally friendly processes have been developed, such as reduction of poisons solvents and hazardous reagents. For a long time, an alternative source for heating the reactions was neglected. This, however, was changed in 1986, when the first microwave irradiation was reported [8]. Utilizing microwave irradiation as a source of heat often reduces the reaction time and uses less energy as compared to conventional heating sources like oil or water baths.

When microwave irradiation is used as the heating source, microwaves are focused on the sample and heating the reaction mixture directly, unlike conventional heating methods, where the vessel is heated. In typical microwave ovens, the magnetron produces microwaves with a wavelength of 12.25 cm (2.45GHz) [8]. The figure below demonstrates that the microwaves are aimed at the reaction vessel and guided through a "tube" to the sample. In most of the modern microwave ovens, it is possible to monitor the temperature during the reaction. Due to a fiber-optic temperature probe and sealed reaction vessels, the temperature and pressure can be measured [8]. Additionally, a magnetic stirrer generally is installed in the instrument to ensure stable temperature in the reaction mixture.

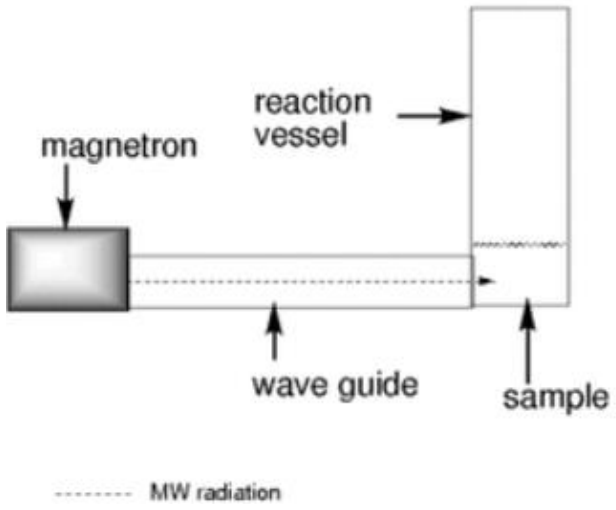


Figure 3 Schematic of a microwave oven designed for organic synthesis, obtained from Microwave-Assisted Reactions in Organic Synthesis [8].

Automatic flash column chromatography

Purifying the target compound often is a challenging and time consuming prosses most chemists face on a daily basis. Flash column chromatography uses a packed column with a stationary phase, generally silica, and an eluent system consisting of a mixture of organic solvent.

Generally, in flash column chromatography, the silica is mixed in a slurry of the impure product that is placed on top of the packed column and a solvent system elutes the compounds through the solids, and the eluate are collected in different fractions. Flash column chromatography exploits the difference in partitioning behaviour between the mobile and the stationary phases to separate the compounds [9]. The different compounds interact with the stationary phase based on relative solubility, the charge or absorption. Compounds that interact strongly to the solids will eluate later (higher retention time), while compounds that interact less strongly will have shorter retention time.

In recent time, this process has been automated with instruments that pumps the mobile phase through a prepacked column forcing the compounds to elute faster (see figure below). In modern instruments, the eluate often passes through a detector that measures the UV absorbent at a specified wavelength, and this signal is sent to a computer which makes a chromatograph of the process. The compounds are finally collected in fractions.

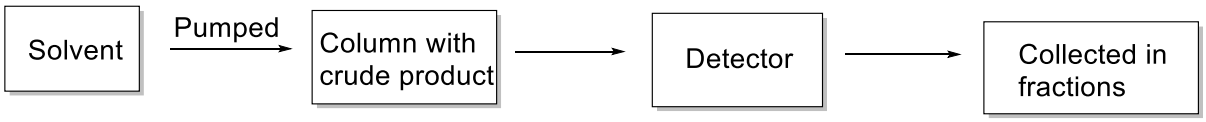


Figure 4 Schematic of automatic flash column chromatography

Gas Chromatography – Mass Spectrometry (GC-MS)

GC-MS is a commonly used analytical instrument in organic chemistry. The GC performs an efficient separation, using a carrier gas and a column, and the separated compounds are introduced to the MS, where they are identified. This efficient analytical tool is limited to small and volatile compounds.

In a GC instrument, a continues flow of a carrier gas, like helium, passes through the injection port, the column, and the detector. To ensure a satisfactory separation and reproducible retention times, the flow rate of the carrier gas is carefully controlled. The sample is injected into the heated injection port, where it is vaporized, and the carrier gas guides it through the column [10]. The column normally consists of a 15-30 meter long tube coated with a stationary phase like silica. The interaction between the stationary phase and the carrier gas, results in different elution times for the different compounds. From the column, the separated compounds pass through a detector before being introduced to the mass spectrometry.

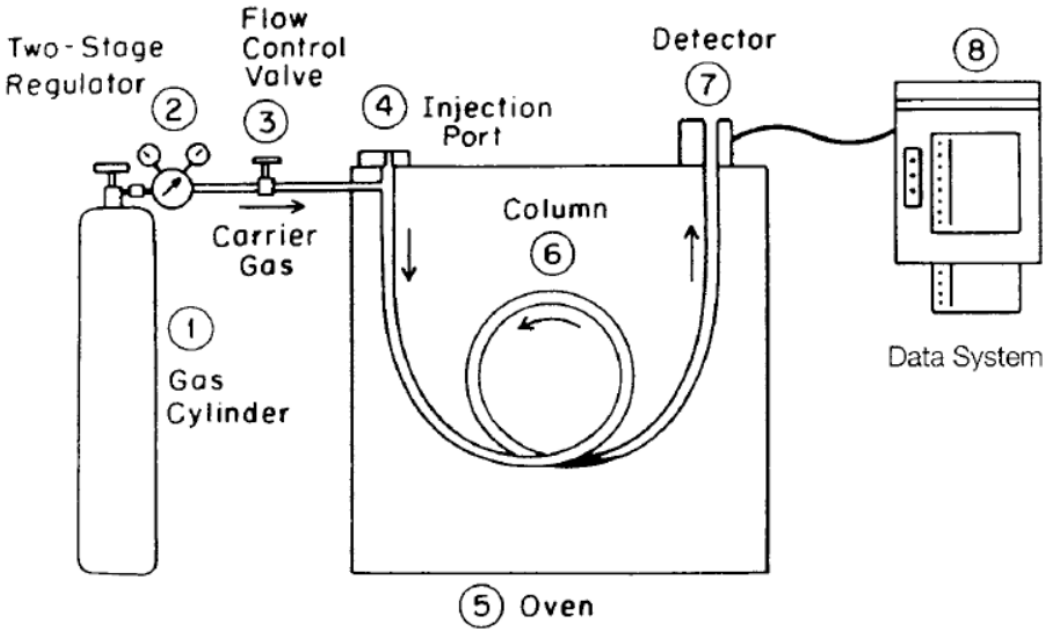


Figure 5 typical components in a gas chromatograph, obtained from Basic Gas Chromatography [10].

When the effluent elutes from the GC, they are introduced to the ion source in the mass spectrometer. In the ion source, ions are created using electron ionization (EI), ion trap, or time of flight [11], by blasting electrons at the compounds, resulting in fragmentation. The mass of these fragments is divided by charge giving the m/z ratio. Generally, the charge is +1, resulting in the m/z ratio to represents the molecular weight. The fragmented ions pass

through a quadrupole, which is programmed by a computer to only direct ions of a certain mass through to the detector.

Liquid Chromatography – Mass Spectrometry (LC-MS)

The LC-MS is an efficient and effective analytical tool in organic chemistry and other industries. Liquid chromatography performs a separation using a column consisting of a stationary phase (generally, silica) and a suited mobile phase, and the mass spectrometry then is able to sort and identify the separated compounds from the LC.

For the MS to perform an optimal analysis, a sufficient separation of the different compounds is needed to obtain this, a suited mobile phase is required. High pressure pumps deliver a constant flow of mobile phase through the system. The sample then is injected into this "river" of eluent and is guided into the column where the separation occurs [12]. The separation is caused by the affinity that the different molecules have towards the mobile and the stationary phases. Compounds with different polarities will pass through the column at a different rate. The column could consist of different solids, like alumina, but silica is the most common. When the compounds elute the analytical column, the sample is introduced to a detector, normally a UV detector, before being introduced to the MS.

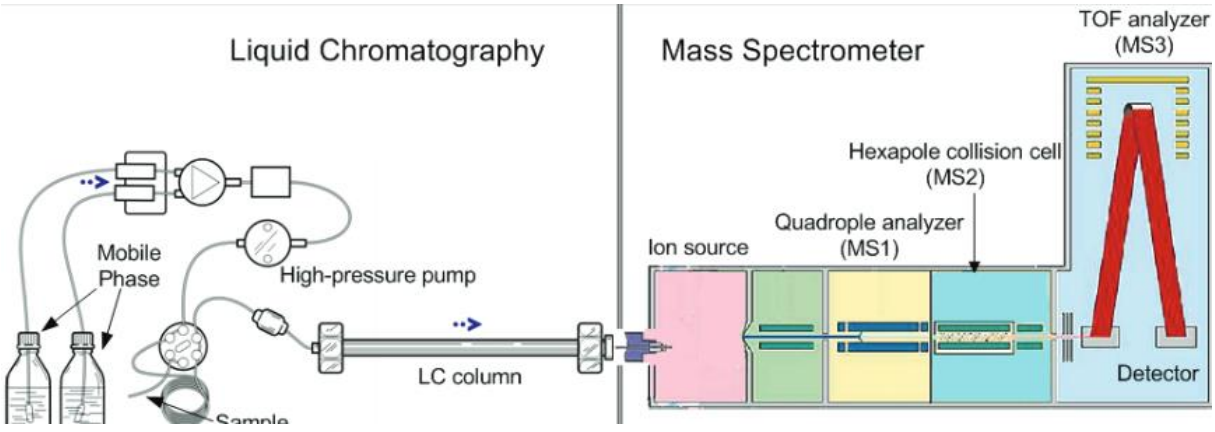


Figure 6 Schemtic diagram of LC-MS instrument, made by B. Zhou [13]

When the effluent elutes the LC, it is introduced to the ion source/ ion chamber, where it is ionized. When a mass spectrometer is coupled with a LC, the most common ionization technique is electronspray ionization (ESI), where the compounds are converted into gaseous ions [14]. Other techniques also can be used, such as atmospheric pressure chemical ionization

(APCI). The gaseous ions are introduced to a mass filter, generally a quadrupole, that is programmed only to let ions of a certain mass pass through to the detector.

Nuclear magnetic resonance (NMR)

Nuclear magnetic resonance is one of the most useful and robust tools for structure determination in organic chemistry. In organic synthesis, generally, proton (^{1}H) and carbon (^{13}C) are the most common nuclei to be analysed.

For the most useful nuclei, there are two quantum states that can be visualized as having the spin axis pointing "up" or "down" [15]. In a thermal equilibrium, these spin axes have the same energy, and it will be exactly a 1:1 ratio of the spin axis pointing up or down. In the presence of an external magnetic field that is aligned with the up state, the energy levels will be affected resulting in the "up" state having a lower energy than the "down" state. Due to this quantum anomaly, it is not possible for any states in between, and it will result in a small excess of the nuclei with a spin axis pointing up, due to the lower energy state. By irradiating the lower state with the right frequency, it is possible to reverse the spin orientation, resulting in a larger excess of the "up" state. A difference in population between the two states is imperative to receive an NMR signal, and a greater difference results in higher intensity.

Valuable information for structure determination is multiplicity, which is caused by the "split-split" occurrence. If two hydrogen (H_1 and H_2) are bonded to adjacent carbons, the magnetic nuclei of H_2 will be aligned with ("up") or against ("down") the magnetic field of the instrument. H_2 will slightly disturb how H_1 experience the external magnetic field. Because the resonant frequency is always proportional to the magnetic field, the resonant frequency for H_1 is changed [15]. Since approximately 50% of the H_2 nuclei are in the "up" state, and the remaining nuclei are in the "down" state, the H_1 resonance is split into a pair of resonance peaks with equal intensity (a doublet), and H_2 will experience equal effect from H_1.

Modern NMR instruments use a pulsed Fourier transform (FT) to record the spectrum. The individual magnetic fields of the nuclei are combined to give a rotating magnetic field that creates an electric voltage in a coil that is placed next to the sample [15]. Over a short amount of time, the individual nuclei get unsynchronised and the signal deteriorates; this is called free induction decay (FID). This signal gets sent to a computer that performs a Fast Fourier Transform, and convert the FID signal to a plot of intensity as a function frequency.

Melting point

Melting point analysis is a useful method to investigate the purity of a compound. The apparatus consists of a heating chamber and a glass “window” to inspect the sample. The sample vessel is placed in the oven, and, as the temperature raises, the compound will start to decompose and melt. The melting point interval is measured from where the first droplet is observed to the whole sample is observed as a liquid.

3. Method and Results

Retrosynthesis:

Building challenging and complex molecules is a problem that organic chemists have faced since the early days of chemistry. Retrosynthesis introduces a new way for chemists to approach the problem of finding a new synthetic route for the desired product using the disconnection approach [16]. Robert Robinson was the first to suggest this idea, when he published his famous tropinone synthesis in 1917 [17]. In the disconnection approach, the desired product is the starting point, and bonds are disconnected, which results in smaller molecules or "fragments of the product." To obtain acceptable starting materials, several disconnections are generally required. When performing a retrosynthesis, it is necessary to consider in which order to perform the disconnections. If the target molecule possesses a bulky functional group that could cause steric hindrance for the following reactions, that group generally should be disconnected first. The same applies if there is a group that deactivates the molecule [17].

Organometallic catalyst

Organometallic catalysis has received a lot of attention over last decades due to its wide range of reactions and tolerance for functional groups. In organometallic catalysts, generally a ligand coordinates to a transition metal (with some exceptions, such as the Grignard reagent). The metal normally is the catalytical centre at which, the reaction proceeds [18]. For the reactions to take place, the ligands play a significant role by changing the electronic properties of the metal and adjusting the steric hindrance around the metal [18]. In several reactions, the same metal can be used, but the ligand often changes depending on the reagents. The Suzuki Miyuara coupling is the main organometallic catalytical reaction used in this project, and it is described later.

3.1 α-Carboline

Retrosynthesis:

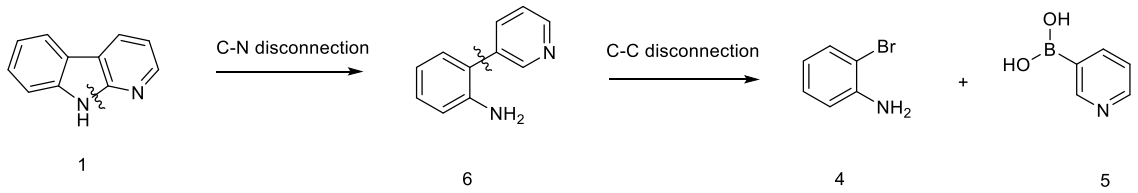


Figure 7 Retrosynthesis of a-carboline

The most evident start for α-carboline (**1**) is to disconnect the carbon-nitrogen bond (C-N) between the nitrogen and the pyridine, resulting in a biaryl pyridine(**6**). Generally, in retrosynthesis, it is a good strategy to disconnect large groups, if possible. Using this strategy, the next disconnection is the C-C bond coupling the biaryl pyridine. Considering that Suzuki coupling is a widely used reaction in organic chemistry to couple two arenes, the starting material should have a halide and a boronic acid present, giving compound **4** and **5** as the two starting materials.

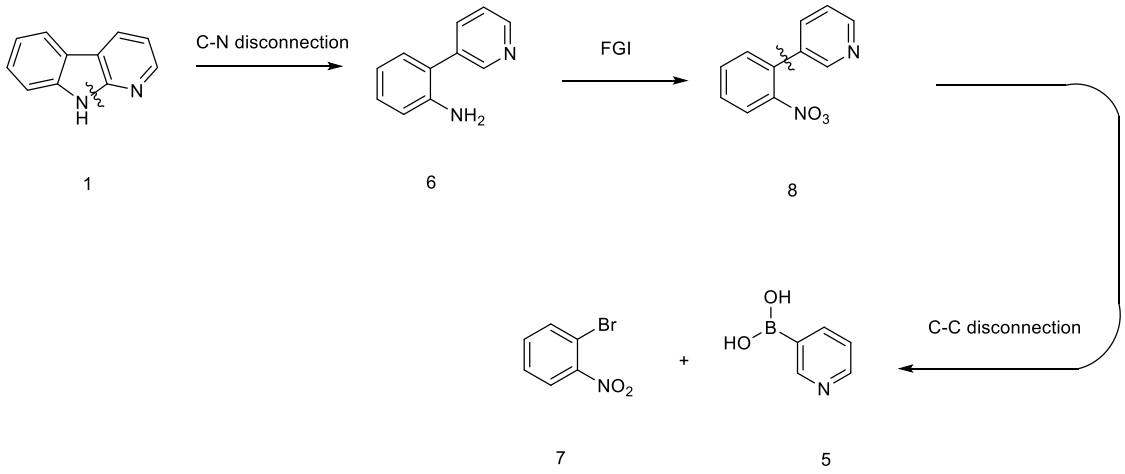
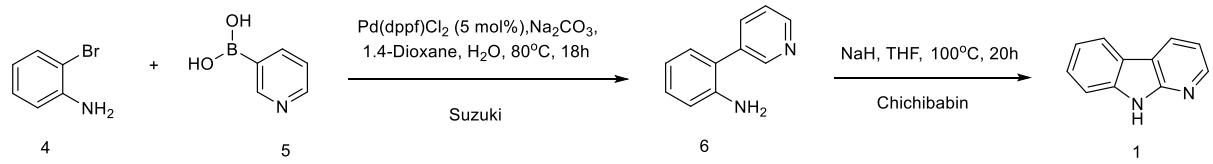


Figure 8 second retrosynthesis of a-carboline

A second retrosynthesis was performed to synthesize compound **1**. The same disconnections were performed; however, an additional step was introduced, where the amine (**6**) went through a functional group interconversion (FGI) resulting in compound **8**. Similar to the previously retrosynthesis, a Suzuki coupling was performed to couple compound **7** and **5**.

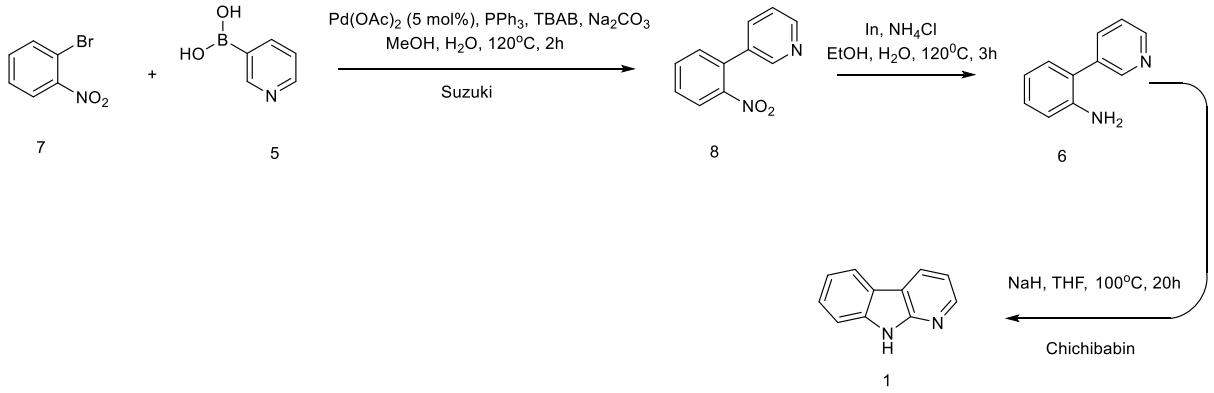
Total synthesis:

Approach 1



The synthetic route for α-carboline (**1**) first was a Suzuki coupling between the two starting materials **4** and **5,** to form the product (**6**). A C-N bond must be formed in the ortho position, relative to the nitrogen in the pyridine. Chichibabin reaction was performed to obtain the product in good yield. Different approaches in the final step, such as C-H activation, also was attempted with no success.

Approach 2



In the second approach to synthesize α-carboline (**1**), a Suzuki coupling was performed between 1-bromo-2-nitrobenzene (**7**) and pyridine-3-yl boronic acid (**5**) to obtain the product as 3-(2-nitrophenyl)pyridine (**8**). Reducing compound **8** resulted in 2-(pyridine-3-yl)aniline (**6**). Chichibabin reaction was performed once more to synthesize α-carboline (**1**).

Organometallic reactions:

The Suzuki Miyuara coupling was developed by the Japanese chemists, Akira Suzuki and his assistant, Norio Miyuara. Akira Suzuki was born September 12, 1930, in Mukawa-cho Japan. In 2010, he was awarded the Nobel Prize for his work using palladium catalysis for organic

synthesis; he shared this prize with two other chemists: Richard F. Heck and Negishi Ei-Ichi[19].

The Suzuki Miyuara coupling is an organometallic reaction, and one of the most widely used reactions to form a carbon-carbon bond. The Suzuki Miyuara reaction involves a coupling between an organoboron and an organic halide [20]. Palladium generally, is the metal used in Suzuki reactions with a ligand. Phosphine ligands are the most commonly used ligands; however, different ligands can be applied, such as dba. The use of Suzuki Miyuara coupling has increased tremendously over the last decades, and it has become one of the most efficient and robust ways to form biaryls and other aromatic compounds.

Compared to other cross coupling reactions, like Grignard and Stille cross coupling, the Suzuki Miyuara coupling have a higher tolerance towards functional groups, such as carbonyls and water.

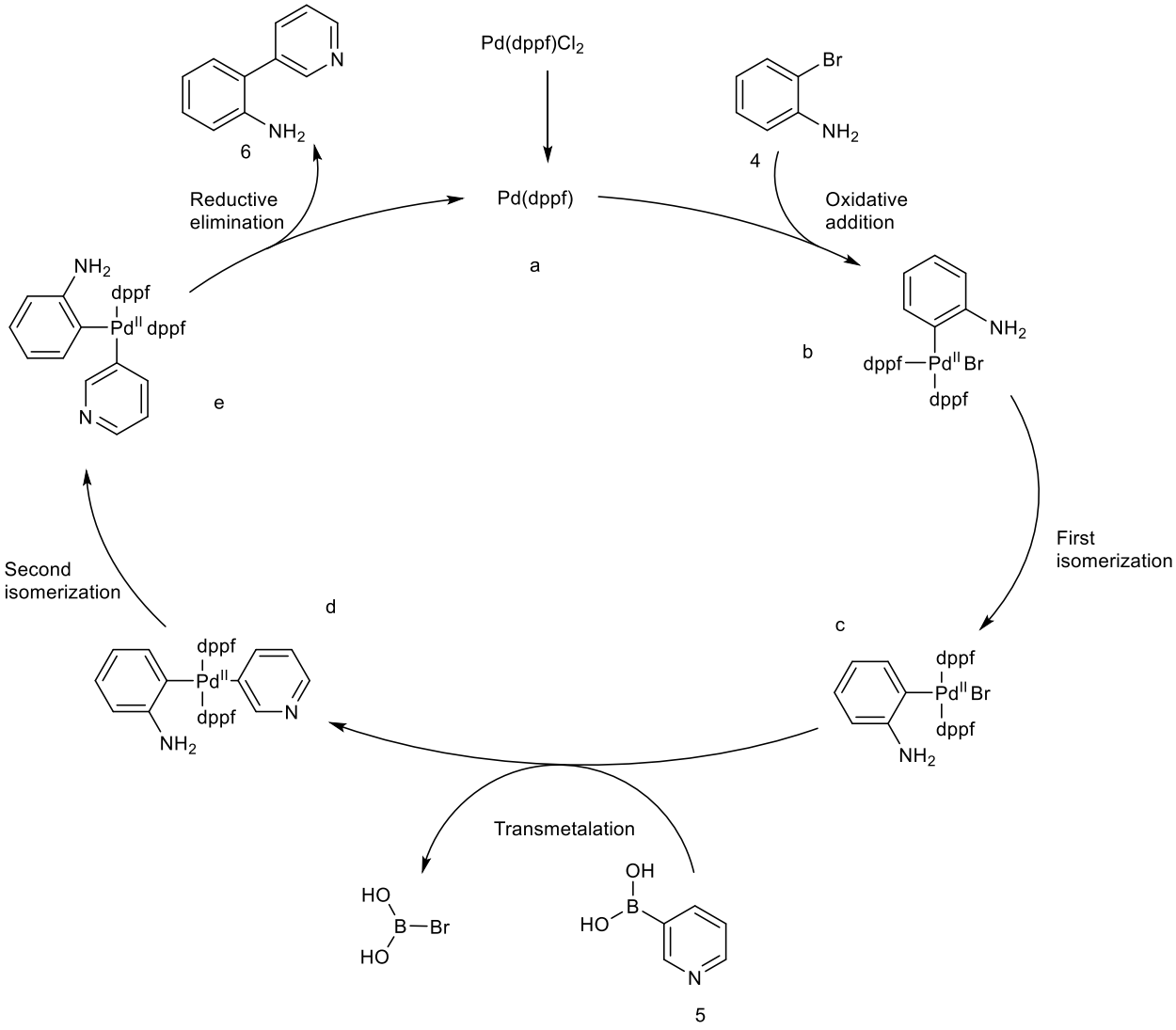


Figure 9 Proposed catalytical cycle based on Braga et al [20]. In the catalytical cycle there is only one dppf ligand, but it is forming two single bonds to palladium. For simplistic reasons two dppf ligands are shown in the figure.

In the reaction to synthesize compound (**6**), Pd(dppf)Cl_2 was chosen as the catalyst. Before the reaction can start, the precatalyst must be activated, forming Pd(dppf) and reducing palladiums oxidation state from (II) to (0). The first step of the catalytical cycle is an oxidative addition with an organohalide, then the bond between the halide and the aromatic compound breaks, and both form a bond to palladium resulting in a Pd (II) complex (**b**). The reaction rate depends on how activated the organohalide is (the reaction is faster if the organohalide is deactivated). An isomerization of the complex follows, changing positions of the molecules connected to the catalyst(**c**). Transmetalation is the next step, where the organoboron (**5**) is introduced. The boronic acid reacts with the halide and leaves the catalytical cycle as the pyridine forms a bond to the catalyst. A second isomerization follows,

rearranging the molecules connected to palladium, making the final step possible (**d**). In the final step of the catalytical cycle, the two organic compounds are coupled with each other and leave the cycle. The reductive elimination step removes two bonds connected to the catalysts, reducing palladium back to initial state (0), and, as a result recreating the catalyst.

A characteristic future of the Suzuki Miyuara coupling is that a base is required for the reaction to take place. The most common bases are K_2CO_3 and K_3PO_4 although other bases also could be applied and are important to promote the reaction. In the catalytical cycle, transmetalation is the delaying step. The boronic acid is more reactive in a basic environment; by adding a base, the reaction rate increases [21].

Chichibabin:

The Chichibabin reaction was first reported over a century ago by Aleksei Chichibabin. The reaction generally is used to introduce an amine group in the ortho position of a pyridine. In this reaction, however, an intramolecular reaction occurs, and the mechanism is similar to a S_nAr substitution.

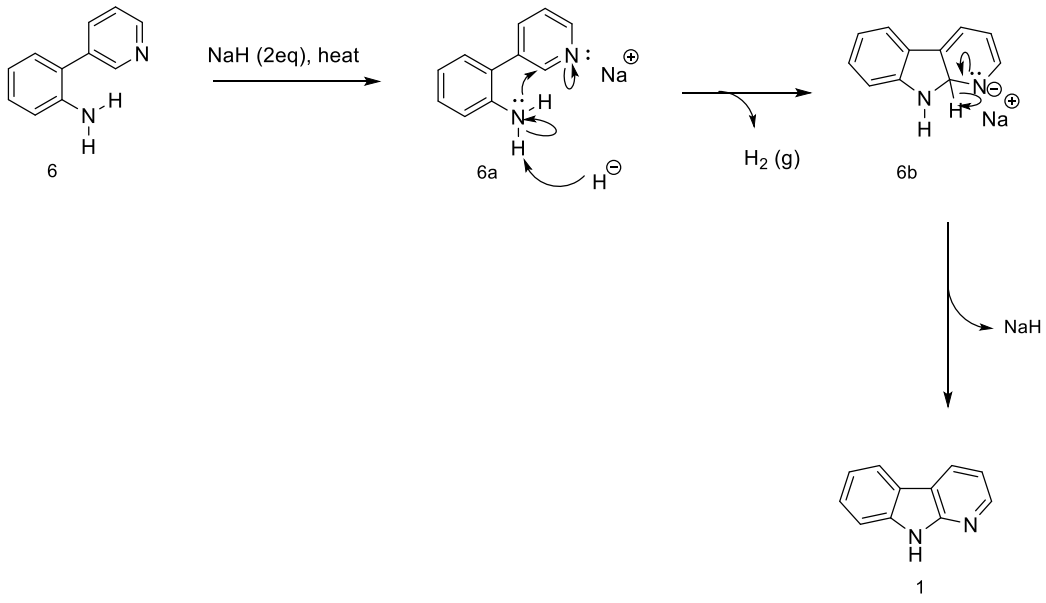


Figure 10 Chichibabin reaction mechanism proposal based on K. Breuker et al mechanism proposal [22]

Compound **6** is deprotonated by a strong base in NaH (**6a**), activating the amine group to perform a nucleophilic attack. The only position that can be attacked is the ortho position on the pyridine ring. As the amine performs the attack, the nitrogen in the pyridine withdraws an electron pair, resulting in a negative charge on the nitrogen (**6b**). This intermediate is non-

favourable and requires high energy for the reaction to take place. The sodium cation, however, stabilizes the negative charge. In the final step, aromaticity is restored when the available electron pair on the nitrogen forms a double bond with the adjacent carbon, resulting in NaH leaving the compound.

In the second approach, compound (**8**) also was synthesized by performing a Suzuki coupling with 1-bromo-2-nitrobenzene (**7**) and pyridine-3-yl boronic acid (**5**) with $Pd(PPh_3)_4$ as the catalyst, to obtain the product (**8**). The Suzuki coupling follows the same catalytical cycle as shown in figure 9. The reaction resulted in high yield (88%).

Indium reduction:

Reducing a nitro group into an amine can be performed in several ways. Using indium powder and ammonia chloride was the method of choice, and the reaction mechanism is provided below.

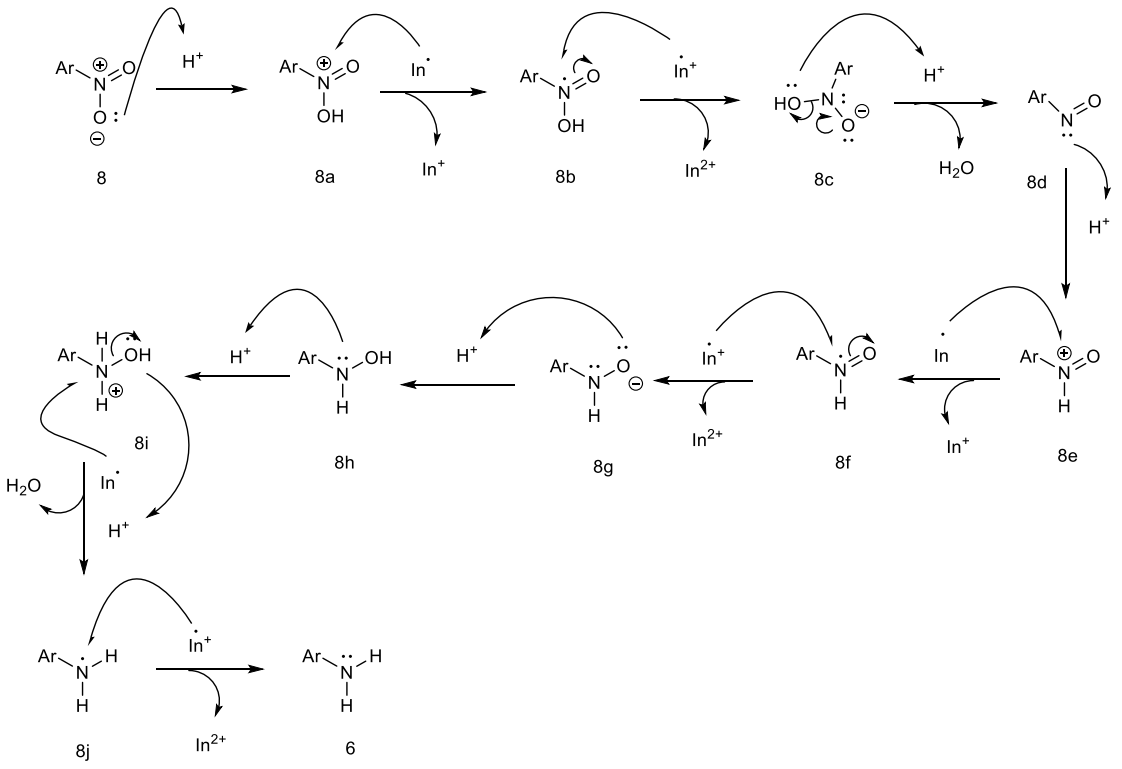


Figure 11 Mechanism proposal based on V. Elumalai el al proposal [23].

The nitro group is protonated by a weak acidic salt (NH_4Cl) to form **8a**. Indium donates one electron to the nitrogen giving In^+. In^{2+} is significantly more stable than In^+ and, therefore, wants to donate another electron to the nitrogen [24]. As nitrogen receives the second electron,

oxygen withdraws an electron pair from the π bond, giving it a negative charge (**8c**). The hydroxy group receives a proton from the weak acid and forms water, which functions as a leaving group, as the negatively charged oxygen restores the double bond with nitrogen forming **8d**. The available electron pair on nitrogen forms a bond with a proton that is received from the acid, before step 2 and 3 are repeated to form **8g**. The negatively charged oxygen receives another proton, forming **8h**. Nitrogen forms a new bond to a proton, giving nitrogen a positive charge. The hydroxy group is protonated and emigrates the compound (**8i-8j**). Steps 2 and 3 are repeated once more to give nitrogen the right amount of valence electrons resulting in the product (**6**) in decent yield (52%).

The Chichibabin reaction was again performed to reach the target compound (**1**) with high yield (84%)

Results

Table 1 The yield in % for each step of the synthetic route and the overall yield in % of the synthetic route

	Step 1	Step 2	Step 3	Overall
Approach 1 (yield %)	87	84	-	73
Approach 2 (yield %)	88	52	84	38

-: step not required

As mentioned earlier, a second cyclisation step was attempted by performing a C-H activation, using $Pd(OAc)_2$ IMES*HCl as the catalyst and H_2O_2 as the oxidizing agent. In all attempts, the reaction ran for 5 hours. The temperature and the amount of oxidizing agent were adjusted, as shown in table below. None of the reactions led to the desired product.

Table 2 Results of the C-H activation

Catalyst	H_2O_2 (mL)	Time (h)	Temperature (^oC)	Conversion (%)	Yield (%)
$Pd(OAc)_2$ IMES*HCl (5 %mol)	0.25	5	120	100	0
$Pd(OAc)_2$ IMES*HCl (5 %mol)	0.1	5	120	100	0
$Pd(OAc)_2$ IMES*HCl (5 %mol)	0.05	5	120	100	0
$Pd(OAc)_2$ IMES*HCl (5 %mol)	0.05	5	100	<10	0
$Pd(OAc)_2$ IMES*HCl (5 %mol)	0.05	5	80	<10	0

3.2 9H-pyrido[2,3-b]indol-7-ol

Retrosynthesis:

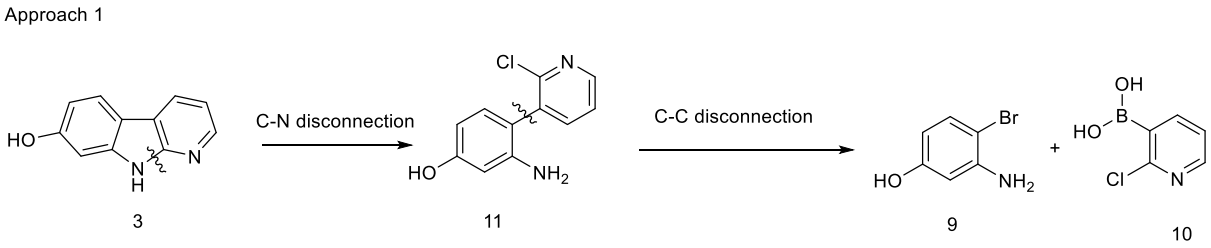


Figure 12 Retrosynthesis of 9H-pyrido[2,3-b]indol-7-ol

The retrosynthesis of 9H-pyrido[2,3-b]indol-7-ol was performed by disconnecting the same bonds as the retrosynthesis of α-carboline, starting with a C-N disconnection and giving the biaryl (**11**). A C-C disconnection results in compound **9** and **10** as the starting materials.

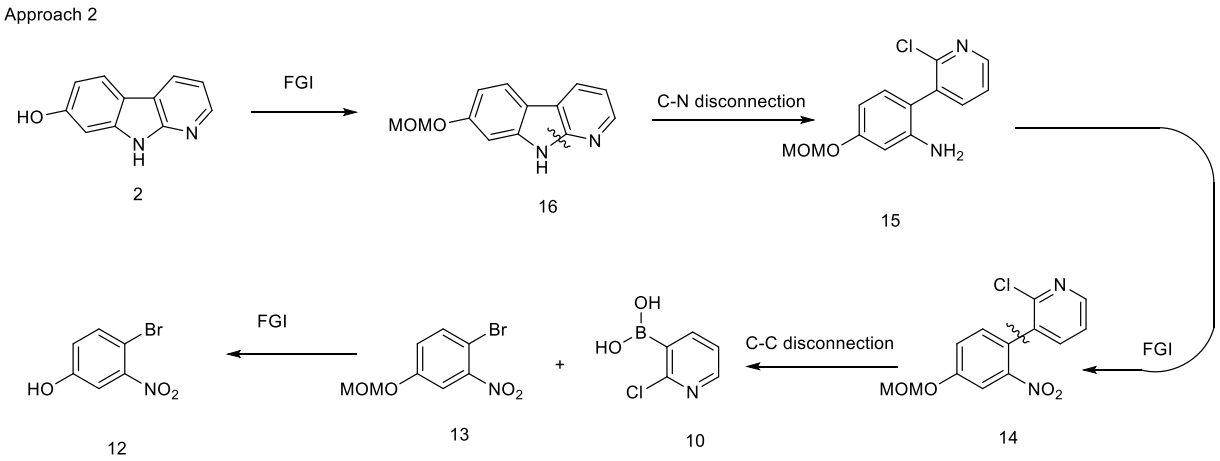


Figure 13 A second retrosynthesis of 9H-pyrido[2,3-b]indol-7-ol

A hydroxy group in the target compound (**2**) is fairly acidic. Protecting the alcohol appeared to be the first step of the retrosynthesis, giving compound **16.** Like the retrosynthesis of α-carboline, the first disconnection was the C-N bond, resulting in compound **15**. An FGI on the amine group followed, leading to **14**. Coupling two arenes normally is performed by a Suzuki coupling, resulting in an organoborane and an organic halide, compounds (**10**) and (**13**), respectively. The final step is to remove the protection of the alcohol, making compounds **12** and **10** as the two starting materials.

Total synthesis:

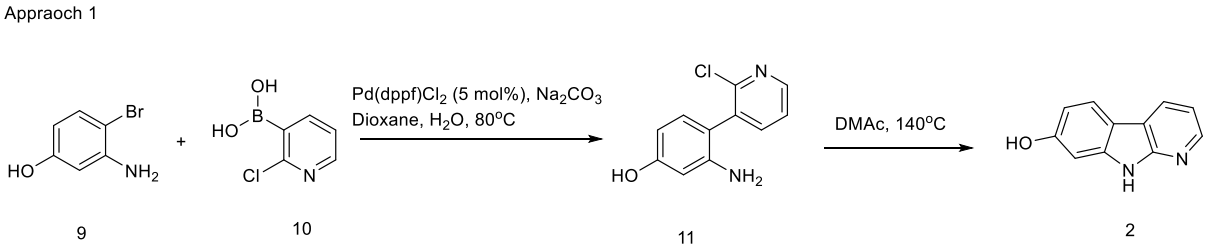


Figure 14 The total synthesis of 9H-pyrido[2,3-b]indol-7-ol using the the first retrosynthesis

The starting materials **9** and **10** was coupled in a Suzuki reaction with $Pd(dppf)Cl_2$ as the catalyst. It is believed the reaction follows the catalytical cycle presented in figure 9 in chapter 3.1. The coupling resulted in the target compound **11**, with 28% yield. The biaryl **11** was dissolved in DMAc and heated to 140°C for 4 hours. LC-MS suggested a conversion of ~30% to the target compound (**2**), but the compound was never isolated. It is assumed the reaction follows a similar S_nAr mechanism as shown in figure 18. However, no base is used to activate the amine; rather, the high temperature is enough for the amine to perform a

nucleophilic attack in the ortho position in the pyridine ring, resulting in a negative charge on nitrogen in the pyridine. When aromaticity is restored, HCl leaves the compound, giving **2** as the product.

The second approach:

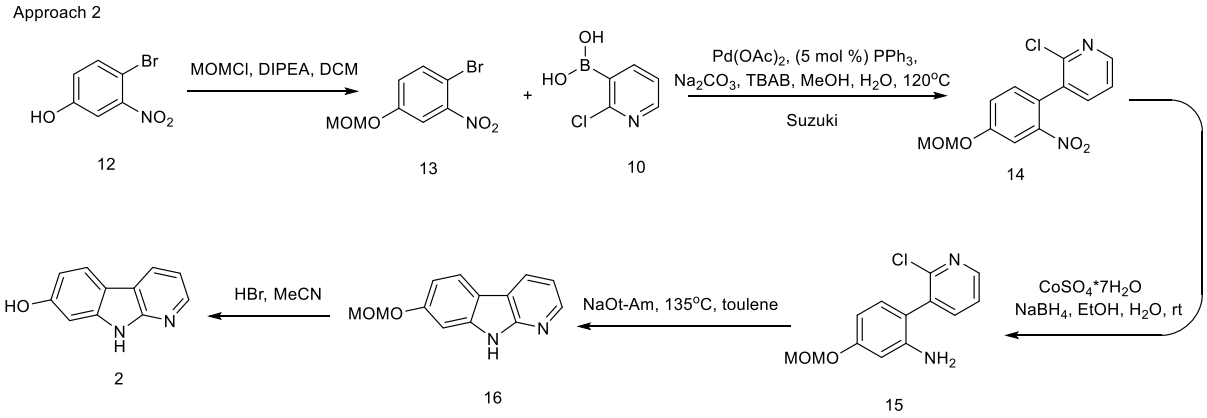


Figure 15 The total synthesis of 9H-pyrido[2,3-b]indol-7-ol using the second retrosynthesis

Protecting the alcohol in compound **12** with a methoxymethoxy ether was the first step of the synthesis. A Suzuki Miyuara coupling followed with compounds **10** and **13** as the reagents, resulting in the biaryl pyridine (**14**). Reducing the nitro group with cobalt sulphate and sodium borohydride followed to synthesize compound **15**. A strong, bulky base in sodium tert-pentoxide in toluene, combined with heat, was used to close the ring, giving compound **16**. Hydrobromic acid was applied to remove the protection group, leading to the target compound (**2**).

MOM protection of 4-bromo-3-nitrophenol (12).

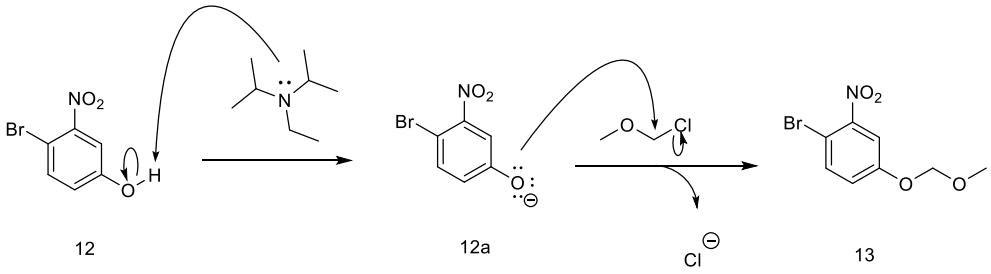


Figure 16 Protection of the alcohol, reaction mechanism proposal

Diisopropylethylamine deprotonates the acidic hydroxy proton, giving oxygen a negative charge to form **12a**. The oxygen performs a nucleophilic attack on the chloromethyl methyl

ether, in a S_n2 reaction. The chloride functions as the leaving group, giving compound **13** as the target molecule. This method also was attempted to protect 3-amino-4-bromophenol (**9**), but with no success.

Organometalic reactions:

The Suzuki-Miyuara coupling was performed to prepare the biaryl (**14**), the reaction is described, in 3.1 α-carboline and the catalytical cycle is shown figure 9. In contrast to figure 9, tetrakis $Pd(PPh_3)_4$ is the catalyst used in this reaction. The main difference between the two catalysts are the oxidation state, $Pd(dppf)Cl_2$ has a oxidation state of (II), while $Pd(PPh_3)_4$ has an oxidation state of (0). Results of the reaction with different conditions are provided in the table below. The reaction using a phase transfer catalyst ran for 2 hours, while the other reactions was running for 18 hours.

Table 3 Results of the different Suzuki reactions attempted.
Cat-catalyst, PTC-phase transfer catalyst

Cat	Mol (%)	Base	T(^{o}C)	Solvent	PTC	Yield (%)
$Pd(PPh_3)_4$	5	Na_2CO_3	120	MeOH, H_2O	TBAB	88
$Pd(PPh_3)_4$	3	Na_2CO_3	120	MeOH, H_2O	TBAB	88
$Pd(PPh_3)_4$	2	K_2CO_3	80	DMAc, H_2O	-	25
$Pd(PPh_3)_4$	5	K_2CO_3	100	DMAc, H_2O	-	38

Cobalt reduction

A number of reactions could perform the reduction of the nitro compound (**14**), like Pd/C which is probably the most common method. It was however attempted to utilize cobalt sulphate and sodium borohydride to perform the reduction. The proposed mechanism is provided below.

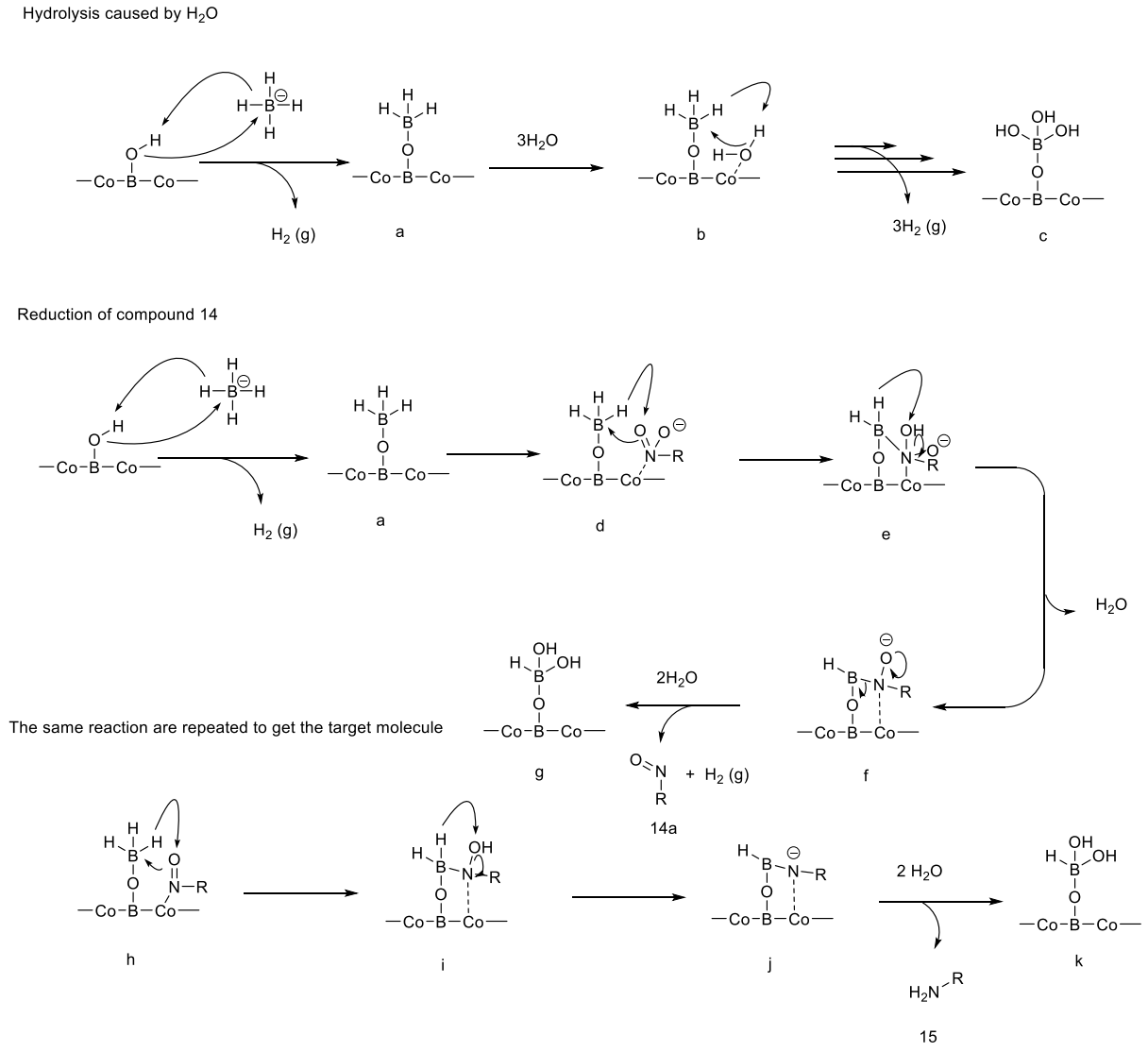


Figure 17 Proposed mechanism of the cobalt reduction. based on F. J. Lundevall reports [25]

The first step of the mechanism shows how water affect the reaction. Sodium borohydride reacts with the cobalt boron complex to form **a**. Water coordinates to cobalt, resulting in hydrogen leaving the complex as hydrogen gas (H_2) and OH forms bond to the boron complex **c**. When this reaction occurs with compound **14**, the nitro group coordinates to cobalt (**d**) and

one of the oxygens gets protonated, nitrogen uses the electron pair from the N=O bond to form a bond to boron. As a second protonation happens, nitrogen withdraws the electron pair from the bond to oxygen, and water leaves the compound **f**. It is believed that, when water enters the reaction, oxygen donates an electron pair to form a double bond to nitrogen, and the compound leaves as **14a**, forming **g**. **14a** enters a new cobalt boron complex, and similar steps are repeated, as the oxygen first gets protonated and nitrogen forms a bond to boron. The hydroxy group gets protonated again and leaves the compound as water. When water enters the complex, nitrogen gets protonated, resulting in the amine (**15**). This reaction was only performed once and resulted in 45% yield of the target compound (**15**).

S_nAr Intramolecular Substitution:

Nucleophilic aromatic substitution reaction (S_nAr) is a widely used reaction in organic chemistry. The S_nAr reaction normally takes place as either an addition-elimination or an elimination-addition mechanism. In both mechanisms, a non-favourable intermediate is created, which generally causes harsh reaction condition for the reaction to take place.

Generally, only aromatic compounds with electron-withdrawing substituents can stabilize the negative charge that occurs in the intermediate. The electron-withdrawing group also should be in ortho- or/and para- position relative to the leaving group, which limits the reaction.

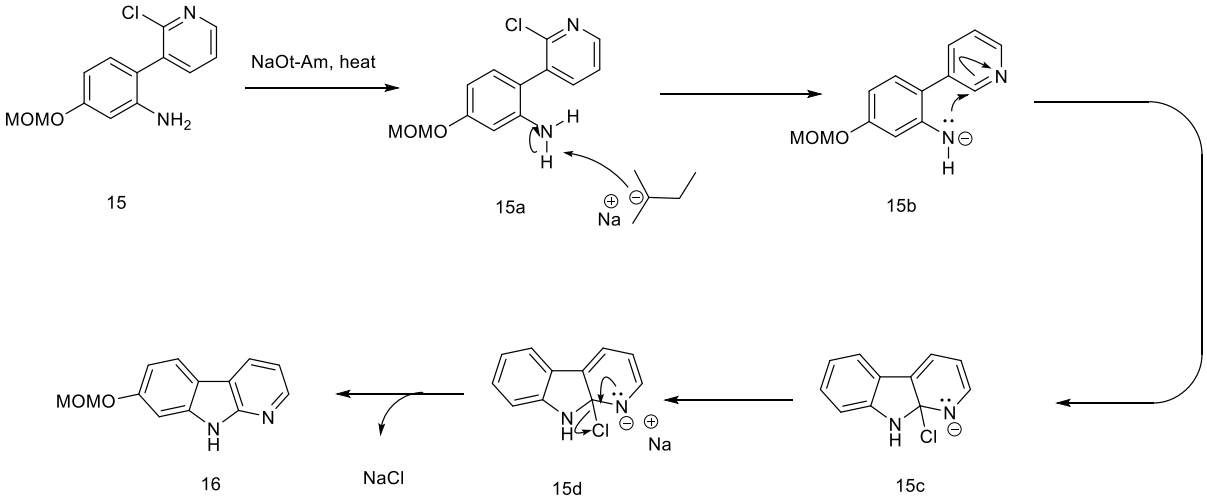


Figure 18 SnAR intramolecular mechanism proposal based on C. N. Neumann reports [26].

In the reaction mechanism proposal above, sodium tert-pentoxide is used as a base to activate the amine group to perform a nucleophilic attack in the ortho position of the pyridine ring (**15b**). Like the Chichibabin reaction, the nitrogen in the pyridine withdraws an electron pair from the C=N bond, giving the nitrogen a negative charge (**15c**). Sodium tert-pentoxide was chosen as the base because it is a strong, non-nucleophilic base, which means it will not cause a competing reaction. The negatively charged intermediate is stabilized by the sodium cation (**15d**). In the last step, chloride works as a leaving group, and aromaticity is restored.

Results

The results for the two synthetic routes are displayed in the table below. Both synthetic routes gave an unsatisfactory overall yield. The target compound was never isolated, resulting in a precise overall yield never was calculated.

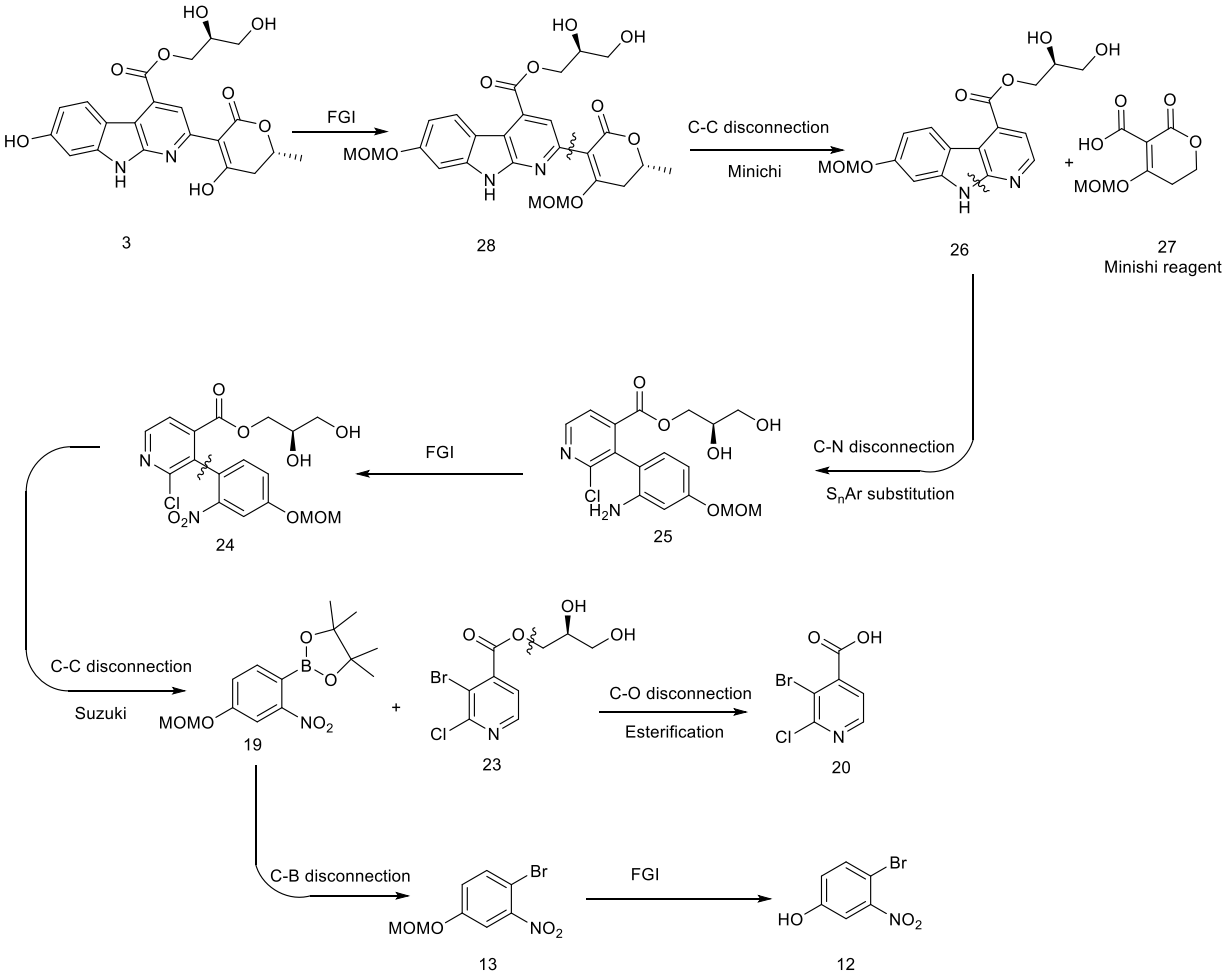
Table 4 The yield in % for each step of the 9H-pyrido[2,3-b]indol-7-ol synthetic route. The final product was not isolated, and the overall yield is only an estimate from the MS analysis.

Route #	Steps					
	1	2	3	4	5	Overall
1 yield (%)	28	unknown	-	-	-	<10%
2 yield (%)	98	88	55	25	Unknown	<5%

-: Step not required

3.3 Mescengricin

Retrosynthesis:



Retrosynthesis of the minishi reagent

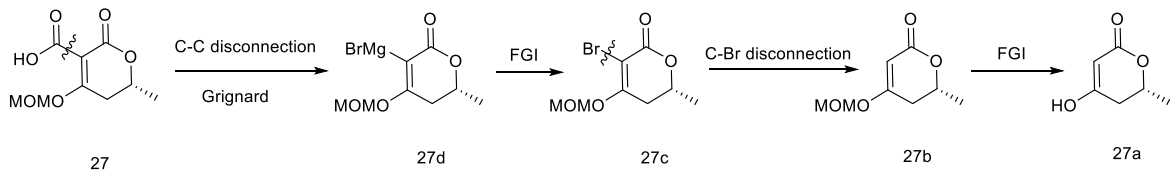


Figure 19 Retrosynthesis of mescengricin

The target compound (**3**) has four hydroxy groups, where at least two need to be protected, applying an FGI to form compound **28**. Performing a C-C disconnection between the α-carboline core and the pyran ring results in compounds **26** and **27**. Like the retrosynthesis for α-carboline and 9H-pyrido[2,3-b]indol-7-ol, a C-N disconnection opens up the conjugated system producing compound **25**. A second FGI follows to convert the amine into a nitro complex (**24**). Breaking the C-C bond by coupling the biaryl is the next disconnection; like

the previous synthetic routes, Suzuki coupling was the method of choice giving compounds **19** and **23**. Both reagents for the Suzuki coupling are not commercially available, additional C-O disconnection was performed on compound **23**, resulting in compound **20**. To the boronic ester (**19**), a C-B disconnection was performed, followed by an FGI to remove the protection group. The retrosynthesis gives 4-bromo-3-nitrophenol (**12**) and 3-bromo-2-chloro-isonicotinic acid (**20**) as the two starting materials.

For the Minishi reagent (**27**) a C-C disconnection was performed where the acid group is removed, resulting in the Grignard reagent (**27d**). An FGI followed changing the Grignard reagent into a bromo compound (**27c**). Breaking the C-Br would be the next step followed by an FGI, removing the protection group and giving **27a** as the starting material.

Total synthetic route for Mescengricin:

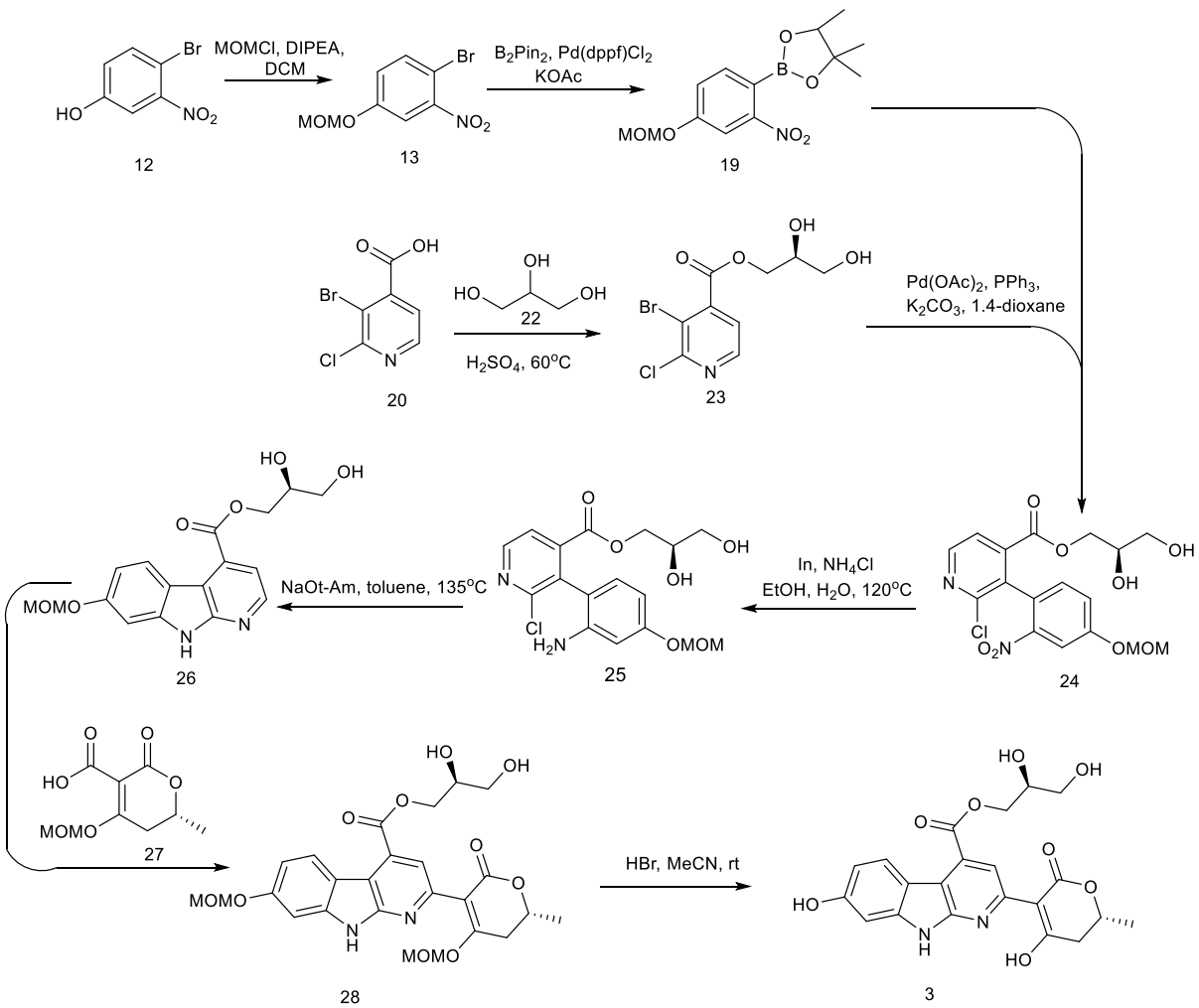


Figure 20 Planed synthetic route for mescengricin

Like the synthesis of 9H-pyrido[2,3-b]indol-7-ol, protecting the hydroxy group in compound **12**, was the first step. To perform a Suzuki reaction, an organoborane must be prepared. This was done by a palladium catalyst borylation of **13**, giving **19** as the product. Before the coupling of the two arenes can be done, **20** must undergo an esterification resulting in **23**. **19** and **23** were used as the two reagents in the Suzuki coupling, creating compound **24**. Performing a reduction using indium powder and NH_4Cl before a S_nAr reaction is used, gives **26** as the product. The pyran ring (**27**) should be coupled in a Minishi reaction. In the final step, the protection groups are removed, resulting in the target compound (**3**).

Synthetic route for the Minishi reagent.

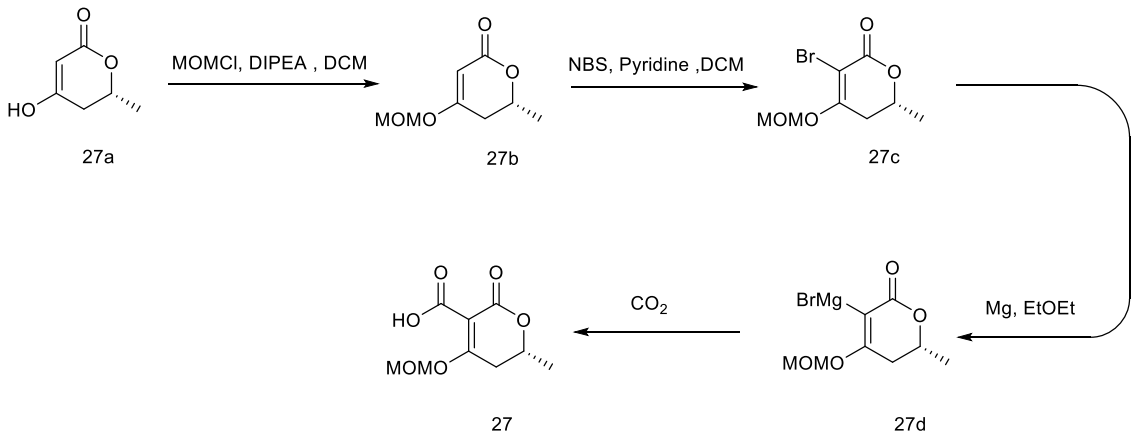


Figure 21 Planed synthetic route for the minishi reagent

In the synthetic route to create the Minishi reagent (**27**), compound **27a** gets protected before it goes through a brominating reaction, resulting in **27c**. Forming the Grignard reagent is next. Reacting the **27d** with dry ice (CO_2) will give the Minshi reagent (**27**).

MOM protection of 4-bromo-3-nitrophenol (12):

The protection of **12** is described in chapter 3.2 9H-pyrido[2,3-b]indol-7-ol, and the mechanism is provided in figure 16.

Fischer esterification:

A Fischer esterification was used on 3-bromo-2-chloroisonicotinic acid (**20**) to produce 2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (**23**) by using glycerol (**22**) and sulfuric acid. Resulting in 84% yield.

Organometalic reactions:

Miyaura borylation:

The palladium catalysed borylation is a one-step procedure to prepare arylboronic esters. Both boronic acids and boronic esters can be used in a Suzuki coupling. Generally, multiple steps are required to synthesize an arylboronic acid. While the boronic esters are easier prepared with simple workup [27], they are not as reactive as boronic acids.

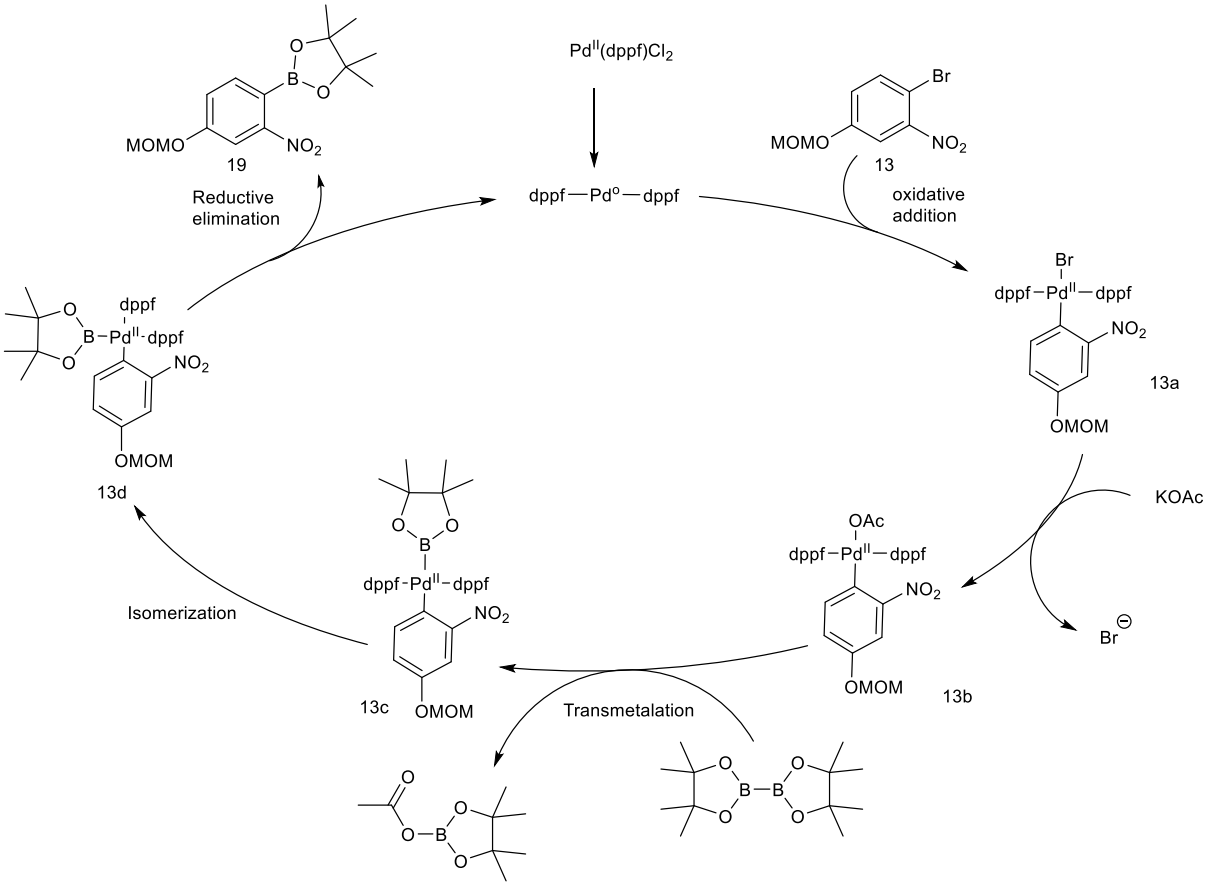


Figure 22 Mechanism proposal based on T. Ishiyama et al reports [27]. In the catalytical cycle there is only one dppf ligand, but it is forming two single bonds to palladium. For simplistic reasons two dppf ligands are shown in the figure

The Miyaura borylation follows a similar catalytical cycle as the Suzuki reaction. The catalyst must be reduced to Pd (0) before the oxidative addition takes place and the aryl halide is introduced. Palladium is oxidized back to (II) resulting in **13a**. In the following step, there is an exchange of ligands. The halide leaves the catalytical cycle as acetate coordinates to palladium (**13b**). In the transmetalation step, bis(pinacolate)diborane was introduced, removing the acetate, and forming a bond between the palladium and the boronic ester (**13c**). Isomerization of the complex followed, rearranging the compounds coupled to the catalytical

metal. Reductive elimination was the final step, resulting in **19**, as the catalyst was recreated. The results from the reaction are provided in the table below.

Table 5 Results of the Miyuara borylation with different conditions

Catalyst	Solvent	Base	Temperature	Yield (%)
$Pd(dppf)Cl_2$	DMF	KOAc	100	77
$Pd(dppf)Cl_2$	Dioxane	KOAc	80	77

A palladium catalysed borylation was also attempted for 4-bromo-3-nitrophenol (**12**), resulting in the boronic ester (**30**) with 30% yield. A Miyuara borylation also was performed on 3-amino-4-bromophenol (**9**), with no success.

Suzuki coupling:

The Suzuki coupling performed follows the same catalytical cycle as described in chapter 3.1 figure 9, with $Pd(OAc)_2$ as the catalyst and PPh_3 as the ligand. In some of the reactions, methyl-3-bromo-2-chloroisonicotinate (**21**) was used instead of 2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (**23**). The results are provided in the table below.

Table 6 The results of the Suzuki reaction

Catalyst	Solvent	Base	Temperature (^oC)	Yield (%)
$Pd(OAc)_2$, PPh_3 (2 mol%)	DMF, H_2O	K_2CO_3	100	0
$Pd(OAc)_2$, PPh_3 (4 mol%)	THF, H_2O	NaOH	80	0[a]
$Pd(OAc)_2$, PPh_3 (3 mol%)	DMAc	K_2CO_3	100	0[a]
$Pd(OAc)_2$, PPh_3 (2.5 mol%)	Dioxane	Cs_2CO_3	120	0

a = reaction was performed with compound **21** instead of compound **23**.

Indium reduction and S_nAr intramolecular substitution:

Indium reduction and S_nAr intramolecular substitution are the two-following reactions after the Suzuki coupling. Both reactions have been described in chapter 3.1 and 3.2, respectively. Unfortunately, the Suzuki coupling did not run successfully, and these reactions were never attempted for compounds **24** and **25**.

Results

Protecting compound **12** resulted in 98% yield. The borylation of compound **13** resulted in 77% yield, and the esterification of compound **20** gave 84% yield.

The synthetic route to create the Minishi reagent is provided in figure 21. Protecting the alcohol with a MOM group was performed with the same reaction as for compound (**12**), with 78% yield. Brominating the compound followed, which proved to be difficult, and no successful reactions were performed, see table below.

Table 7 Results of the attempted brominating reaction

Brominating agent	Solvent	Conditions	Yield (%)
DBH	Chloroform	Dark, rt	0
DBH	Chloroform (anhydrous)	Dark, rt	0
NBS	DCM, Pyridine	rt	0

3.4 Optimization of the Bischler-Napieralski reaction

Total synthesis:

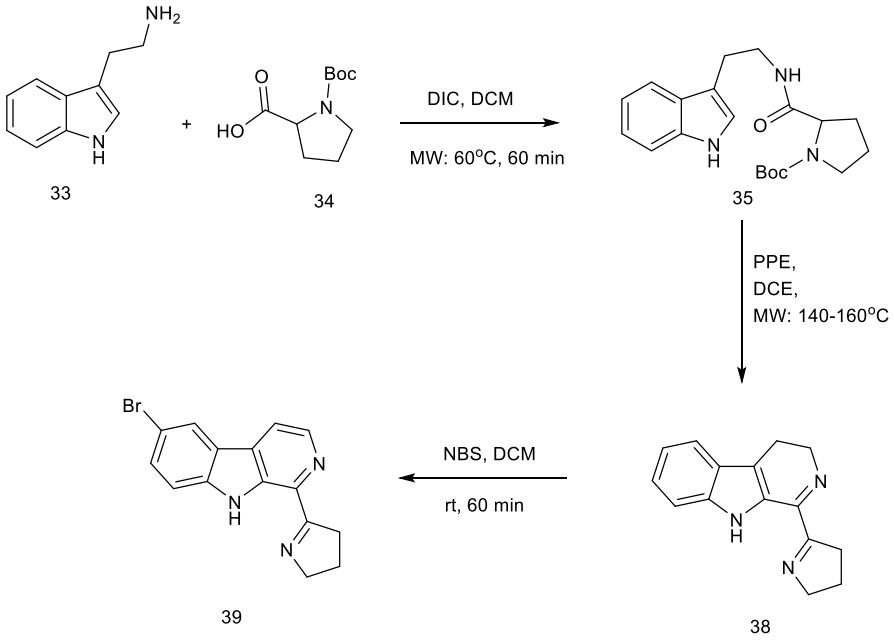


Figure 23 Synthetic route for Eudistomin H

N'-(N-Boc-prolyl)tryptamine:

Converting a carboxylic acid directly into an amide by using an amine has proven to be difficult. This is especially true given that amines generally are basic, which results in a stable, unreactive carboxylate. Introducing a coupling agent like DIC promotes the reaction.

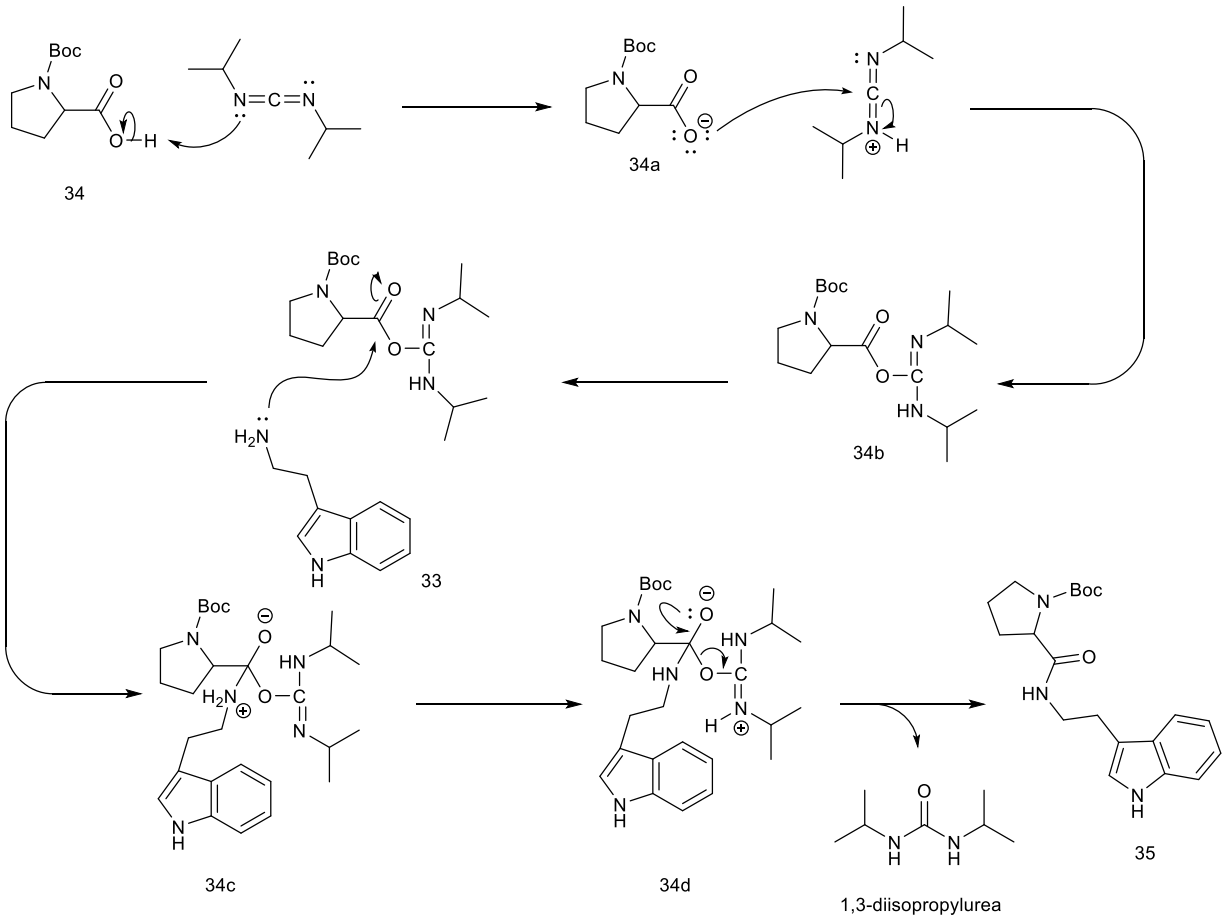


Figure 24 Reaction mechanism for converting carboxylic acids based on N. Fattahi et al proposed mechanism [28].

The carboxylic acid gets deprotonated by the coupling reagent, forming a carboxylate. The oxygen later performs an attack on the coupling reagent forming **34b**. The amine (**33**) then performs a nucleophilic attack on the carboxylic group, forcing oxygen to withdraw an electron pair from the double bond and giving it a negative charge. A proton exchange follows, resulting in a positive charge on the nitrogen in the coupling reagent (**34d**). The negatively charged oxygen donates an electron pair, restoring the double bond to carbon as the coupling reagent functions as a leaving group, forming 1,3-diisopropylurea and the target compound (**35**). The reaction resulted in 54% yield.

Bischler-Napieralski reaction:

The Bischler-Napieralski reaction is an important method for development of isoquinolines. August Bischler and Bernard Napieralski were the first to report this method back in 1893. The method is an intramolecular electrophilic aromatic substitution, where a condensing agent is used [29]. In the reaction performed in this project, polyphosphate ester (PPE) was chosen as the condensing agent.

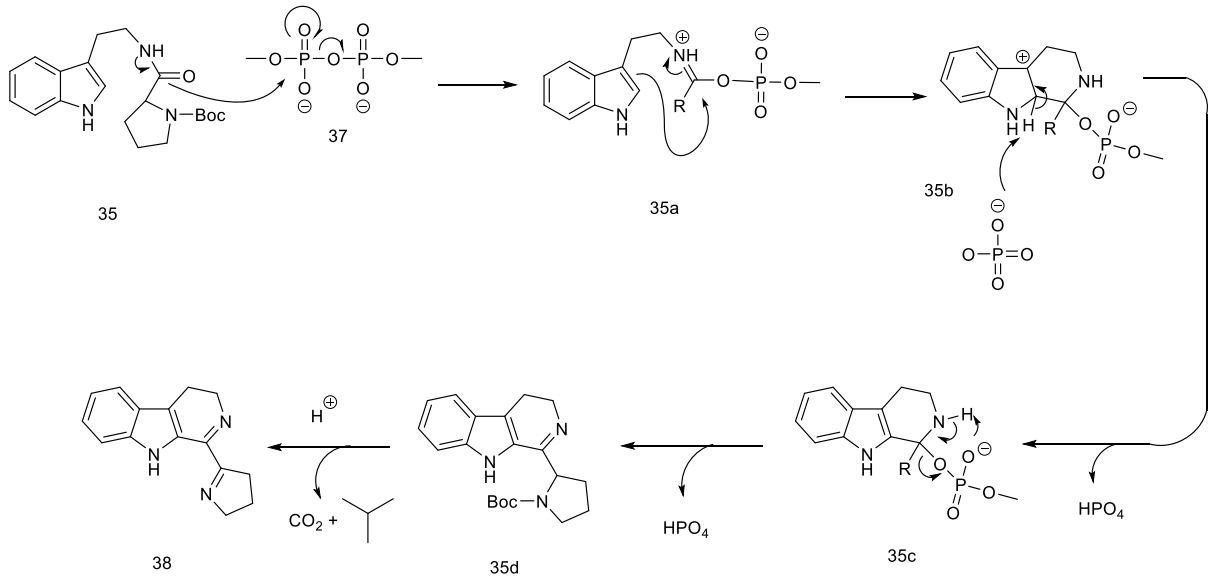


Figure 25 Proposed Bischler-Naprialski reaction mechanism based on reported mechanism by J. J. Li [30].

The nitrogen in the amide (**35**) uses the available electron pair to form a double bond to carbon, making the oxygen perform a nucleophilic attack on the condensing agent (PPE) (**37**) and forming **35a**. The π bond in the aromatic compound acts a Lewis base and forms a bond to the imide as the positively charged nitrogen withdraws an electron pair, resulting in **35b**. The left-over PPE deprotonates the compound restoring the double bond (**35c**). In the final step, nitrogen donates an electron pair to form a double bond to the adjacent carbon, as oxygen receives a proton, and the phosphate functions as a leaving group and forms isoquinoline (**35d**). The deprotection of the Boc group is not shown in the mechanism above, but it is believed that the acid produced in the reaction protonates the Boc group, resulting in isobutane functions as a leaving group. The nitrogen is left with an acid group which eventually will leave as carbon dioxide, resulting in the target compound **38**. The results from this reaction is provided in the table below.

Table 8 The results from the Bischler-Napieralski reaction

#	*Experimental variables*						Response
	Coded values			Real values			
	x_1	x_2	x_3	z_1 [g PPE]	z_2 [mL DCE]	z_3 [°C]	Yield (%)
1	0	0	0	3	2	150	58
2	-1	-1	-1	2	1	140	53[a]
$2_{(2)}$	-1	-1	-1	2	1	140	58[a]
3	+1	+1	+1	4	3	160	63[a]
4	+1	+1	-1	4	3	140	72
5	0	0	0	3	2	150	60
6	-1	+1	-1	2	3	140	81
$6_{(2)}$	-1	+1	-1	2	3	140	123
7	-1	-1	+1	2	1	160	74
8	-1	+1	+1	2	3	160	90
9	+1	-1	-1	4	1	140	87
10	+1	-1	+1	4	1	160	Explosion
11	+1	-1	0	4	1	150	Explosion
12	+1	+0.5	+0.5	4	2.5	155	96
13	+1	+0.5	+1	4	2.5	160	62
14	0	0	0	3	2	150	66[b]
15	0	0	0	3	2	150	75[b]
16	0	0	0	3	2	150	82[b]
17	0	0	0	3	2	150	73[b,a]

a: Reactions carried out after reparation of the microwave
b: Reaction time of 40 min.

Eudistomin H:

Synthesizing eudistomin H from isoquinoline (**38**) was performed in a brominating reaction using DBH. In the same reaction, a dehydrogenation occurs, resulting in a complete conjugated core (β-carboline). Halogenation of aromatic compounds is a type of electrophilic substitution, and is a useful reaction in organic synthesis. By brominating an aromatic compound, it is possible to add other substituents to the ring. Reaction mechanism proposal is provided below.

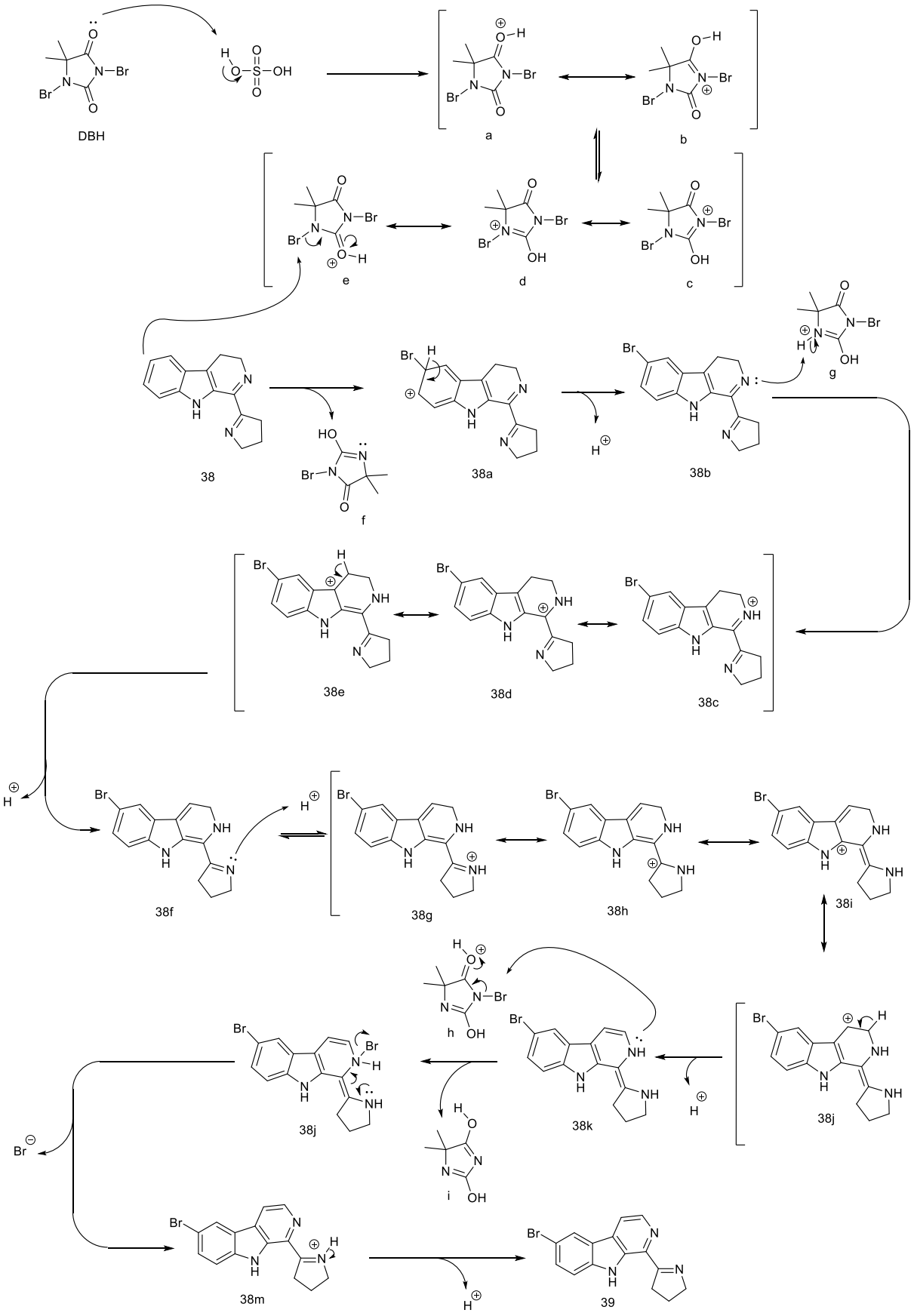


Figure 26 Mechanism proposal based of Sandtorv et al [31]

The mechanism proposal is based on the work of Sandtorv *et al* [26]. Dibromo hydantoin gets protonated by the sulfuric acid, creating a positive charge of the molecule. Compounds **a-e** in the scheme above show the resonance structures caused by the positive charge. The isoquinoline (**38**) performs a nucleophilic attack on compound **e**, resulting in **38a**. As the proton leaves, aromaticity is restored. The available electron pair on nitrogen in compound **38b** takes a proton from **g**, resulting in a positive charge. The resonance structures **38c-38e** show how the formal charge is shared between the compound. **38e** displays that another proton is kicked out as a π-bond is formed (**38f**). Nitrogen in the pyrrole gets protonated, and compounds **38g-38j** shows the resonance structures to stabilize the compound. When the formal charge is on a carbon that could form a double bond to another carbon, hydrogen leaves the compound, resulting in **38k**. The remaining bromo in **h** forms a bond to nitrogen giving **38j**, and both bromo atoms on DBH have left, resulting in **i**. As nitrogen donates an electron pair to form a double bond, a chain reaction occurs, wherein another double bond must change position, and a bond must be formed to nitrogen, as bromide leaves the molecule (**38m**). In the final step, proton functions as a leaving group, giving the target compound **39**.

The reaction to form eudistomine H was performed using both NBS and DBH as a bromo source. The results are shown in the table below.

Table 9 Results of the bromination, with different bromo reagents.

Reagent	Yield (%)
NBS	61
DBH	62

Results:

Table below shows the yield (%) for each reaction performed, and the overall yield for the synthetic route.

Table 10 the result in yield from each step of the synthetic route

Yield (%) of each step in the synthetic route			
Step 1	Step 2	Step 3	Overall
54	96	62	32

4. Discussion and Conclusion

4.1 α-Carboline

The first approach to synthesize α-carboline was done in two steps using a Suzuki-Miyuara coupling, to form a biaryl pyridine (**6**), followed by a chichibabin reaction to cyclize the molecule resulting in the target compound (**1**).

The Suzuki coupling in the first approach gave a consistent yield between 62-87%. The experiments with lower yields are probably caused by a loss of product in the work up. Considering that the solvent used in the reaction is soluble in water, this could cause problems in the extractions, if the solvent is not properly removed in advance. Emulsion was observed during some of the extractions, which likely caused some loss of the product to the water phase. Generally, the reaction and purification occurred without any problems. The NMR spectra in the appendix (figure 33 and 34) shows some traces of impurities in the aliphatic area.

The second approach also performed a Suzuki coupling, using a different starting material in 1-bromo-2nitrobenzene (**7**), a different ligand, and a phase transfer catalyst with the same base and boronic acid. The reaction was carried out in harsher conditions, but in reduced time. TLC was used to monitor the reaction, and the analyses suggested no side reactions occurred; however, full conversion did not happen. Considering no visible changes in the analysis were noted when the reaction ran for 1 hour to 2 hours, it was concluded that the catalyst had decomposed, and the reaction would never deliver 100% conversion. It is speculated that adding additional catalyst to the reaction mixture could have kickstarted the reaction, but this was never attempted. The NMR spectra in the appendix (figure 35 and 36) confirmed the product was made with a small amount of unidentified impurities in the aliphatic area.

The product from the Suzuki coupling in the second approach went through a reduction reaction using indium powder and ammonium chloride. The reaction was monitored using GC-MS. After 3 hours, the GC-MS suggested 100% conversion of the starting material, and only one product was formed (see figure 27). From the GC analyses, it was expected to have an excellent yield; however, the yield proved to be only 52%. Considering the workup consisted of washing the product through a pad of celite, some of the product could have been lost in the celite. However, a TLC analysis confirmed the product went through the celite, which suggested the loss of product did not happen in this part of the workup. A drying agent

was added to the filtrate before the solvent was removed under reduced pressure. It is possible that some of the product became trapped in the drying agent, but it seems unlikely that a large amount would be lost in this step. The NMR (figure 33 and 34 in the appendix) confirms high purity of the product with only traces of impurities. One of the reductions of compound **8** had significantly lower yield than the previous reactions using indium powder and ammonia chloride. It is speculated that the poor yield was caused by poor dissolution of indium and ammonia chloride. Previously, an ultrasonic bath was used to ensure dissolution. However, when this reaction was performed, the dissolution was performed under stirring. TLC analysis also confirmed several unidentified side reactions occured.

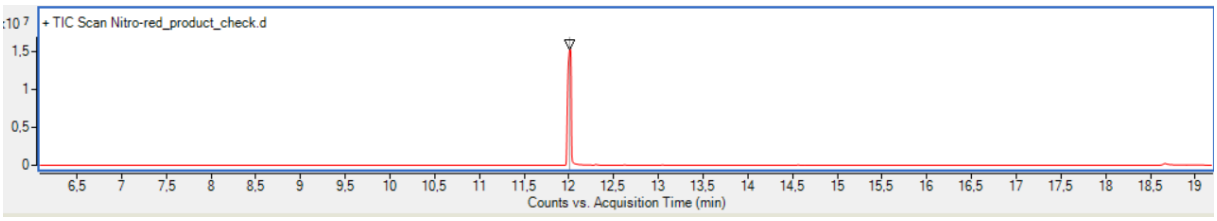


Figure 27 Indium reduction monitored by GC-MS

The Chichibabin reaction produced the target product with high yield. The reaction was monitored by TLC, which suggested that the reaction stopped before full conversion. Considering it two equivalents of the amine are normally used in the Chichibabin reaction [32], full conversion was not expected. Separating the starting material and the target compound was done without problems using column chromatography. The ^{1}H-NMR spectra (figure 37) shows a peak at ~3.25ppm, which is consistent with DOH [33] (deuterated water), which could suggest some moisture in the product. Besides traces of impurities in the aliphatic area, the NMR spectra confirms high purity of the target compound. It is reported that by adding an additive, like LiI, could increase the reaction rate and result in higher yield [32]. A reaction was performed where LiI was added, and this resulted in 0% conversion of the starting material.

C-H activation also was attempted to cyclize the biaryl (**6**), using hydrogen peroxide as an oxidation agent, with no success. The reaction was monitored using GC-MS, and the analyses suggested a N-oxide was formed. If sufficient amounts of oxidation are were available, ^{1}H-NMR confirmed the starting material was oxidized back to the nitro compound (**8**).

Conclusion

A robust synthetic route has been achieved to form α-carboline in a two-step synthesis. A second approach to synthesize α-carboline also was performed as a modelling route for more complex molecules with the α-carboline core, with success.

4.2 9H-pyrido[2,3-b]indol-7-ol

After successfully synthesizing α-carboline, a similar two-step synthesis was attempted to form 9H-pyrido[2,3-b]indol-7-ol. Considering that 9H-pyrido[2,3-b]indol-7-ol has an acidic proton in the hydroxy group, the Chichibabin reaction could not be used without a protecting group. By placing a good leaving group in the desired position of the pyridine, a cyclization should be possible.

A Suzuki coupling was performed using 3-amino-4-bromophenol (**9**) and 2-chloro-pyridin-3-yl boronic acid (**10**) as the starting material and Pd(dppf)Cl_2 as the catalyst. The yield was significantly lower than when compound **4** and **5** were used, and the reaction was difficult to recreate. Why the reaction was unstable and granted low yield is unclear, but it is speculated that the additional halide group could cause some steric hindrance, as well as a competing reaction, where the coupling occurs in the ortho position relative to the nitrogen in the pyridine. The hydrogen in the hydroxy group, is as mentioned before, acidic, and the alcohol could be deprotonated by the base used in the reaction. How a deprotonated hydroxy group would behave in a Suzuki reaction is uncertain; however, it is speculated that it could coordinate to the catalytical metal. This could complicate the reaction and, possibly, lead to side products, which are confirmed by the LC-MS analysis (figure 28). Additionally, the reaction resulted in poor yield, despite decent conversion. Performing the workup proved to be difficult; significant amounts of the product got trapped in the water phase. Neutralizing the solution was attempted, but with limited success. Due to an unexpected change of laboratory and corrupted data files, NMR spectra of compound **11** is not available.

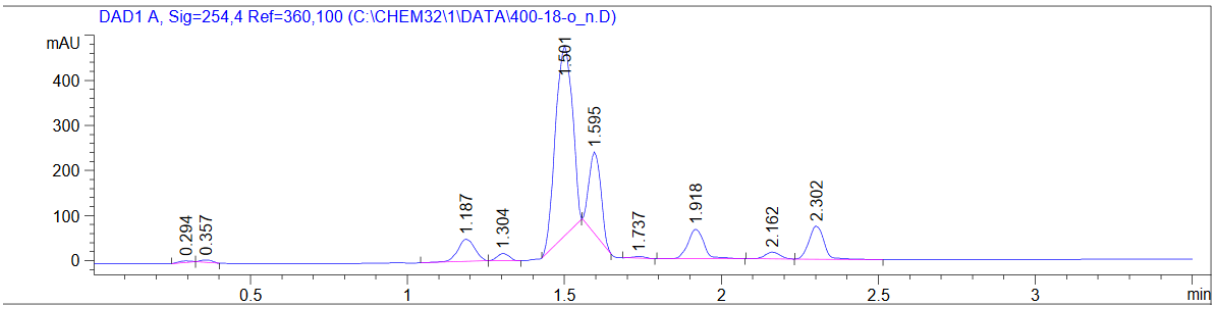


*Figure 28 LC-spectra of the reaction mixture in the synthesis for compound (**11**), where the peak at 1.501 is the product.*

Due to the unprotected hydroxy group, a different approach was made to cyclize compound (**11**), then the Chichibabin reaction used to produce α-Carboline. The cyclization was performed by heating the compound. The reaction was monitored by LC-MS, and the analysis suggested the reaction was working, but finished shortly with low conversion (see figure below). After the reaction was stopped, it was discovered that the reaction mixture was acidic. Considering chloride function as a leaving group and the amine loses a proton, it is likely that hydrochloric acid is produced, acidifying the reaction mixture. With an acidic reaction mixture, it is possible the amine group in the reagent gets protonated and precipitates, resulting in the reaction to stop. Considering particles were observed in the solution, this is a likely the explanation. Due to unexpected change of laboratory, the workup was never finalized, and the product was not isolated.

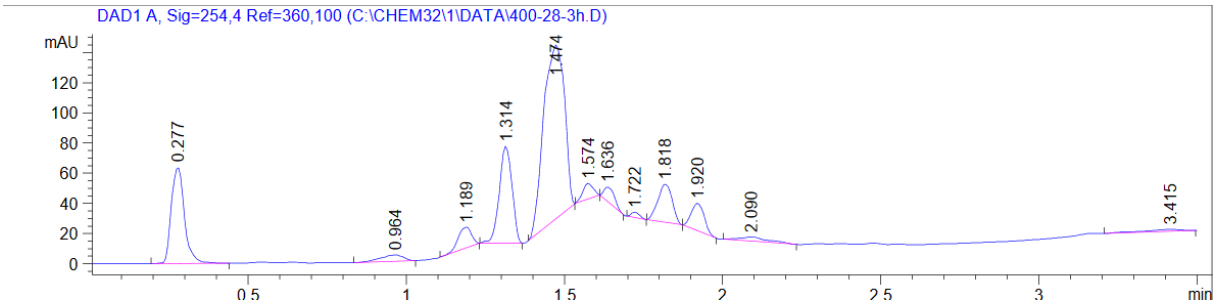


*Figure 29 The LC- analysis performed on the reaction mixture to form compound **2**. The peak observed at 1.314 min is suggested to be the product*

For the second approach, it was decided to protect the alcohol to improve the overall yield. Converting the alcohol into a methoxymethoxy group was attempted on 3-amino-4-bromophenol (**9**) and 4-bromo-3-nitrophenol (**12**). Protecting **12** was done easily with ~100% conversion and excellent yield (see figure 30 for GC analysis), and NMR (figure 39 and 40 in the appendix) confirmed the product (**13**) with traces of impurities. Compound **9**, however, proved to be difficult. The reaction was monitored using GC-MS, and the major product from the reaction had mass of 220.2 (figure 31). It is well-known that bromo consists of two

isotopes: 79 and 81 MW with approximately 1:1 ratio. The spectra below confirm bromo is not present in the major product, and likely functions as a leaving group. It is speculated that the base (DIPEA) used in the reaction has performed a nucleophilic attack on the ring, resulting in a S_nAr substitution kicking out bromo. As mentioned before S_nAr substitution reactions generally forms unfavourable intermediates that demands harsh reaction conditions. It is therefore unlikely that this occurs under ambient temperature, but it cannot be excluded. Considering this undesired reaction did not occur with **12** the main factor is probably the activation of the rings.

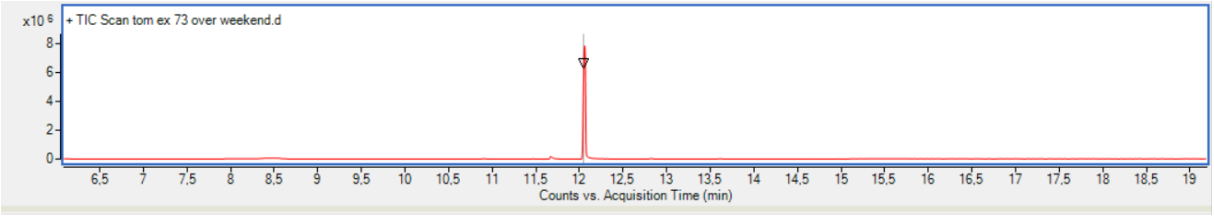


Figure 30 Protection of compound 12 monitored with GC-MS

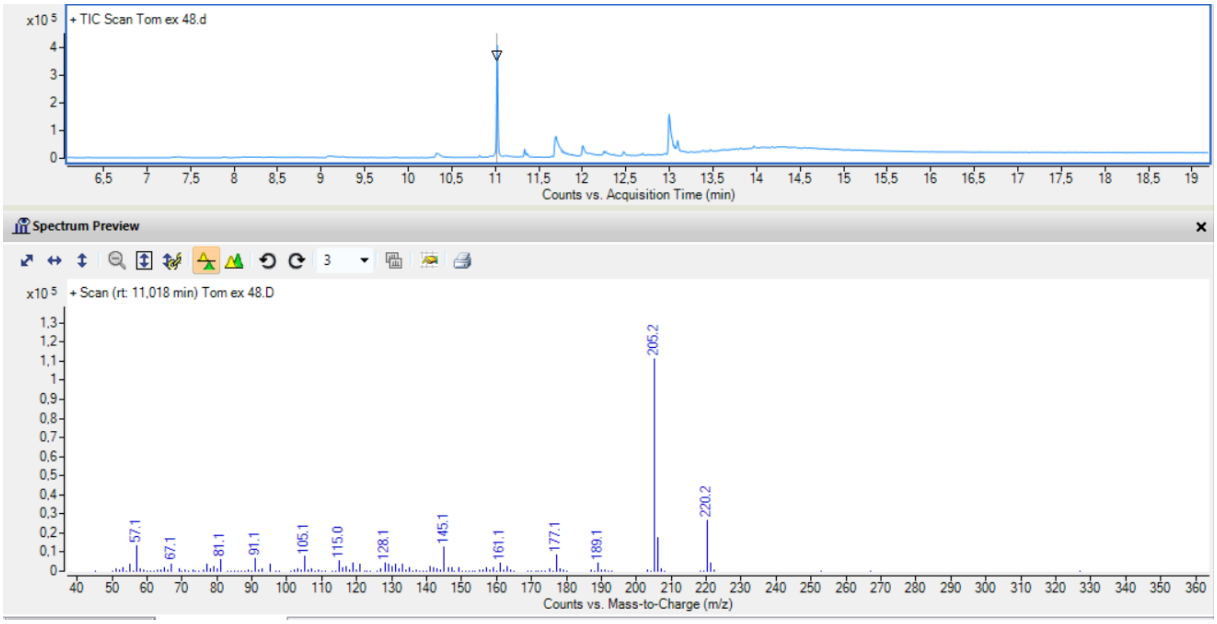


Figure 31 attempted protection of compound 9 GC-MS spectra

The Suzuki coupling was performed with 1-bromo-4-(methoxymethoxy)-2-nitrobenzene (**13**) and 2-chloropyridin-3-yl boronic acid (**10**) with tetrakis as the catalyst. The reaction was monitored with TLC, and, after 2 hours, it appeared the catalyst had decomposed, and the reaction stopped. TLC confirmed a small amount of starting material was left in the reaction mixture, and no side reactions had taken place. Adding additional catalyst might have resulted in 100% conversion; this, however, was never attempted. The reaction consistently delivered

yield >60%, and work up and purification was done easily. The NMR spectra in the appendix (figure 41 and 42) suggests the product (**14**) is of high purity with traces of solvent and other impurities. Coupling **13** and **10** also was performed without a phase transfer catalyst, a different base, solvent system, and lower temperature, but the reaction time was increased, which resulted in significantly lower yield. Why the reaction did not run as well is difficult to explain, but considering the two compounds possess some steric bulk, harsher conditions might be required.

Reducing the nitro biaryl (**14**) was done using cobalt sulphate and sodium borohydride. Like the indium reduction, a complete resolution seemed imperative for the reaction to run. The reaction was left stirring for 10 minutes at ambient temperature. A TLC analysis suggested a small amount of starting material had not reacted. It previously was observed that the reaction could form side reaction, if the reaction ran for too long. Therefore, the reaction was stopped after 10 minutes, without full conversion. From the TLC analysis, it was estimated a conversion of 70-90% due to the size difference of the two peaks observed on the silica coated plate. However, only 45% yield was isolated, which suggested a significant loss of product in the workup. The workup consists of a filtration, using a drying agent to eliminate water, and removes the solvent under reduced pressure. None of these steps are likely to lose a significant amount of product. However, a silica column was used to purify the compound, and it is possible some product got trapped in the column. Optimizing the workup from the reaction possibly could increase the overall yield in the synthetic routes significantly.

To cyclize the compound, a strong bulky base was used as well as a high temperature. Sodium tert-pentoxide dissolved in toluene was chosen as the strong base. This reaction also was attempted with potassium tert-butoxide as a solid, with no success. The reaction was monitored using TLC, and, after 12 hours, it appeared that approximately 50% of the starting material was converted. It was attempted to increase the temperature from 135^{o}C to 150^{o}C to see if the reaction rate would accelerate, but, after additional 2 hours, no visible changes in the TLC analysis, and the reaction was stopped. It proved to be difficult to separate the product from the starting material, and a decision was made to telescope the reaction and deprotect the compound. Separation was attempting once the target compound was made. The yield proved to be 50%, but, from the ^{1}H-NMR analysis (figure 45 and 46 in the appendix), it was approximately 1:1 ratio of the product and the starting material, which implies the yield of the product was ~25%. In the aromatic area of the spectra, it was expected to be observed a

singlet of doublets, two doublets, two doublets of doublets and a triplet, all with an integration value of 1H. By comparing the NMR spectra to the 2-(2-chloropyridin-3-yl)-5-(methoxymethoxy)aniline (**15**) analysis, those peaks could be excluded. After removing the peaks that belonged to the starting material (**15**), a singlet of doublets, one doublet, two doublets of doublets and a mulitiplet was left in the aromatic area. One doublet and a triplet are missing from the expected spectra; however, the integration value of the multiplet was 2H, which suggests an overlap between two signals. All the other signals had an integration value of 1H. By including a MS analysis, and the main peak showed a mass of 229.5 (calc M^+ = 229.3), it was concluded the product (**16**) was created.

Deprotecting 7-(methoxymethoxy)-9H-pyrido[2,3-b]indole (**16**) was done using HBr. Due to lack of time the product was not isolated but was analysed using LC-MS with direct infusion. The product appeared to have been created in a small amount, with an unidentified compound with a mass of 304 as the major product.

Conclusion

Two synthetic routes have been created to form 9H-pyrido[2,3-b]indole (**2**), but with poor yield. The second approach are probably the most robust synthetic route and small corrections of the conditions and the workup could increase the overall yield significantly.

4.3 Mesengricin

The first step of the synthetic route was protecting the hydroxy group on 4-bromo-3-nitrophenol (**12**) giving compound **13** as the product. This reaction have already been discussed in chapter 4.2 9H-pyrido[2,3-b]indol-7-ol. A Miyuara borylation reaction followed, producing the organoboronic ester (**19**). It was attempted to monitor the reaction with GC-MS, but it proved to be challenging identifying the product, since the calculated mass was not observed in the spectra. The analysis still provided information that the starting material was consumed (see figure below). A workup and purification were performed, and NMR confirmed the target compound (**19**) was created with traces of impurities and high yield (figure 47 and 48 in the appendix). The MS spectra below show some unexpected fragmentations, which has not been identified. It was expected that the pinacolate boron could be fragmented of the compound in the MS. However, a peak with m/z =294 was the highest mass registered, which means a mass of 16 is missing from the compound (**19**). If the product had an ionization of M^+ that suggest one methyl group was fragmented, which seems

unlikely. The other fragmentations also have not been identified; it is, however, obvious that bromo left the compound, considering the easily recognizable isotopes.

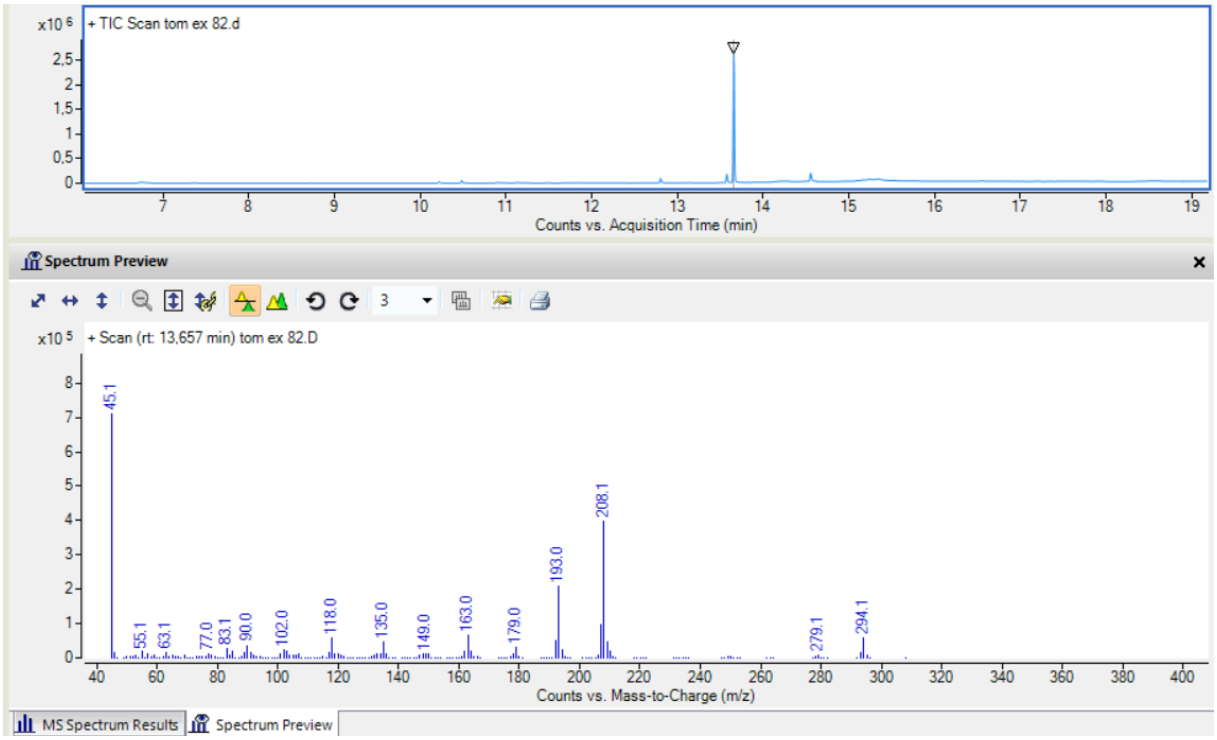


Figure 32 Monitoring borylation reaction with compound 13

Performing a Miyuara borlylation on 4-bromo-3-nitrophenol (**9**) without protecting the alcohol also was attempted. This proved to be difficult, however, a successful reaction was performed to create compound **30** with low yield. The NMR spectra in the appendix (figure 53 and 54) shows a pure spectrum of the compound. However, it proved to be difficult to recreate the reaction and with varying results. It is speculated that the reason why the reaction ran poorly is due to the hydroxy group. The hydrogen is acidic, and the weak base used in the borylation reaction could deprotonate it. This would complicate the reaction, and a competing reaction could occur. By protecting the alcohol, the reaction became more robust and the yield increased significantly. According to T. Ishiyama *et al*, [27] KOAc is the best base to achieve high yield in a Miyuara borylation, despite it does not accelerate the reaction by activating the transmetalation step like stronger bases. Stronger bases, such as K_2CO_3 and K_3PO_4, could push the reaction further, promoting biaryl as a biproduct. Considering biaryl could be a potential sideproduct of the reaction; harsher conditions where not explored.

3-bromo-2-chloroisonicotonic acid (**20**) was converted into methyl 3-bromo-2-chloroisonicotinate (**21**) and 2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (**23**) using

Fischer esterification. **21** was easily made with high purity and excellent yield (see figure 49 and 50 in the appendix). **23** however, proved to be more difficult. The reaction did run well, but the excess of glycerol made the extraction challenging and loss of product to the water phase seemed to be inevitable. Considering glycerol has three hydroxy groups, the reaction could result in two isomers of the product. NMR analysis confirmed two isomers was created and leftover unreacted glycerol was still left in the product (figure 51 and 52 in the appendix).

Performing Suzuki coupling where the boronic acid/ester is in ortho position relative to a nitro group is challenging and normally results in poor yield. Therefore a Miyuara borylation was attempted on 2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (**23**) to form 2,3-dihydroxypropyl 2-chloro-3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)isonicotinate (**31**). This was however unsuccessful, and from the NMR analysis it appeared that unreacted bis(pinacolate)diboron was the major product. Compound **23** possesses significant steric bulk, and it is speculated that the steric hindrance, is the main factor for the reaction to not take place, but with harsher conditions the reaction might proceed.

Methyl-3-bromo-2-chloroisonicotinate (**21**) was significantly easier to synthesize and purify than 2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (**23**) and compound **21** was therefore used in some of the Suzuki reactions to model the reaction. From the results in chapter 3.3 (table 7) it is shown that none of the Suzuki reactions attempted were successful. All the reactions were monitored using LC-MS with direct infusion. The reactions using a solvent system consisting of an organic solvent and water suggested the reaction had worked. MS analysis showed a large peak that was consistent with m/z = M-Cl which had been observed several times in earlier experiments. After the workup and purification however, NMR confirmed it was a compound similar to the starting material: 2-(4-(methoxymethoxy)-2-nitrophenyl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (**19**) but with an additional proton signal in the aromatic area, and the boronic ester signal lost. Protodeboronation is a normal side reaction that can occur in reactions where boronic acids and boronic esters are used with a proton source like water. The NMR spectra confirmed a protodeboronation has occurred. Considering undesirable side reaction is common when a proton source is available, it was decided the following reactions should be performed without water. These reactions resulted in unreacted starting material.

As mentioned earlier it is generally challenging to perform Suzuki reactions where the boronic acid or the boronic ester is in the ortho position relative to a nitro group. The two

arenes attempted to couple also possess steric bulk that possibly effect the reaction. It is still believed that the coupling is possible, it might however demand harsh conditions. The Suzuki coupling from previous experiments with less substituents using TBAB as a phase transfer catalyst was not attempted due to the risk of protodeboronation. In retrospect this reaction, still should have been attempted.

The remaining steps of the synthetic route consists of an indium reduction, a S_nAr intramolecular substitution, a Minishi reaction to couple the pyran ring, and deprotecting the hydroxy groups. It is believed that the indium reduction should run without any problems. It is unclear how selective the reaction is, and if the ester group possibly could be reduced as well. V. Elumalai *et al* [34] have however, performed this reaction on acid and ketone compounds, where only the nitro group has been reduced in high yield. This suggest a high selectivity towards nitro and can therefor assume the reduction would not cause any problems. The S_nAr intramolecular substitution to cyclize the compound, uses a strong base that potentially could deprotonate the two hydroxy groups in the glycerol ester and cause problems. An additional step could possibly be required to protect these groups. This could be performed by producing an acetal.

The Minishi reagent proved to be difficult to synthesize. Protecting the hydroxy group in the pyran ring was the first step followed by brominating the compound, adding magnesium to form a Grignard reagent and finally form a carboxylic acid where the Grignard reagent reacts with carbon dioxide. The protection did not cause any problems, using the same reaction as performed when protecting 4-bromo-3-nitrophenol (**12**). Brominating the compound was attempted with both NBS and DBH with different solvents and conditions with no success. All the reactions were monitored using GC-MS. In most of the reactions it appeared nothing happened, and the starting material never reacted. However, in one reaction where DBH was used as the bromo source in chloroform, it appeared an undesired reaction happened, where the bromide performed a nucleophilic attack on the protecting group recreating the unprotected pyran ring. Every brominating attempts were performed under ambient temperature, by increasing the temperature it is possible the reaction could have worked. Using a different bromo source like Br_2 could also been attempted. Considering the brominating of the isoquinoline (**34**) did run well, it could have been attempted to add sulfuric acid to activate the bromo source. Due to lack of time, the Minishi reagent was not prioritized, and few attempts were performed.

Conclusion

Two reactions were successfully performed forming the two compounds to couple in the Suzuki reaction. The Suzuki reaction however, never resulted in the target compound and the aim to synthesize mescengricin was not reached. Due to the modelling reactions performed with less substituted rings, it is believed the synthetic route could result in the target compound (**3**)

4.4 Optimization of the Bischler-Napieraplski reaction

N'-(N-Boc-prolyl)tryptamine:

The reaction performed to synthesize N'-(N-Boc-prolyl)tryptamine (**35**) was a quick reaction with a simple workup. It however, proved to be difficult to form a solid from the crude, and NMR confirmed, 1,3-diisopropylurea polluted the product. It was attempted to wash the product with additional aqueous citric acid (0.5M) with no result. Purify the crude using chromatography was the next approach, which resulted in a significant reduction of 1,3-diisopropylurea. Everything was unfortunately not removed, and figure 57 and 58 in the appendix confirms the product was made with diisopropyl urea as the main contaminate, the spectra suggest other small impurities as well.

PPE:

The synthesis of PPE (**37**) was performed by dissolving P_2O_5 (**36**) in diethyl ether and DCM and heating the reaction to 70^{o}C and let the mixture stir for three days. Followed by removing the solvent under reduced pressure. Considering PPE is a polymer, it is difficult to calculate the exact molecular weight, it was however, 60 g of the starting material (**36**) and therefor expected with full conversion to have approximately 60 g of the target compound (**37**). After removing the solvent, the target compound proved to be 110 g, which suggest almost 200% yield. It is believed that a significant amount of the solvent got trapped in the viscous liquid and that resulted in the inaccurate yield. ^{1}H-NMR did confirm, solvent was left in the product. Determine the quality of the product was attempted by performing a ^{31}P-NMR. Due to lack of experience with phosphorus NMR analysis, a good conclusion could not be made. However, when the product was used in the following reaction, and the reaction took place, which suggest PPE was successfully synthesized.

Bischler-Napieralski reaction:

Table 11 Yield of the Bischler-Napieralski reaction telescoped compared to purified starting material

#	Coded values			Yield (%)	
	X_1	X_2	X_3	Without purification	With purification
1	0	0	0	33	58
2	-1	-1	-1	34	56
3	+1	+1	+1	67	63[2]
4	+1	+1	-1	33	72
5	0	0	0	29	60
6	-1	+1	-1	34	81
7	-1	-1	+1	32	74
8	0	0	0	32	74[1]
9	-1	+1	+1	32	90
10	+1	-1	-1	35	87
11	+1	-1	+1	20	Explosion
12	+1	+0.5	+0.5	-	96
13	+1	+0.5	+1	-	62

1: Average of the four reaction with X_1 X_2 X_3 = 0 but the run time increased to 40 minutes.
-: Reactions not run.
2: Experiments carried out after reparation of the microwave

By comparing the telescoped method M. Eriksen performed, to the reactions carried out with a pure starting material, it became evident that purity have a significant impact on the outcome of the reaction. The average yield of the telescoped method was: 35%, while the average yield with a pure starting material was: 73% (excluding the experiment that exploded). With this in mind, it is clear that impurities from the telescoped reaction interfere with the reaction, and the yield will increase significantly with a pure reagent.

Considering 1,3-diisopropyluera was the main contaminate in the synthesis of N'-(N-Boc-prolyl)tryptamine (**35**), it is speculated that a competing reaction could occur in the Bischler-Napieralski reaction. The ^{1}H-NMR spectra in figure 59 in the appendix suggest that 1,3-diisopropylurea has been removed from the product, if that is caused by a reaction or the workup is unclear, but a competing reaction cannot be dismissed. Another aspect that should be considered with a polluted starting material, is that the amount of reagent becomes inaccurate, which results in an unreliable yield. The NMR spectra confirms the product has been made, with some impurities in the aliphatic area. The multiplet at ~ 2.95 ppm have an integration value of 4H, and it should have a value of 2H. Considering the other peaks in the spectra is consistent with what is expected, it was concluded the high integration value was caused by overlapping unidentified impurities.

Table 9 in chapter 3.4 shows that the reaction vessel exploded during two of the experiments. The reason why is not clear, but these experiments were running with high amount of condensing agent (PPE), high temperature and low amount of solvent. It is believed that the solution became to saturated, meaning the molecules are too close to each other, and with the high temperature, the reaction happens to fast. When the reaction mixture is heated to 160^{o}C the pressure would normally be ~7-8 bar. As explained in the reaction mechanism in chapter 3.4 it is believed that CO_2 are produced when the deprotection occurs. It is possible that if the reaction happens to quickly and CO_2 are rapidly produced, when the pressure is already ~8 bar, the pressure will continue to rise, which was observed. When the pressure reached 20 bar the reaction vessel exploded.

It is also important to note that four 0-experiments was performed when the time was increased from 30 minutes to 40 minutes. The results from table 9 in chapter 3.4 show that every experiment deliver higher yield than the reactions performed with 30 minutes. Increasing the time of the reaction is an important aspect, by increasing the time it might be

possible to change other variables like decreasing the temperature and still receive the same result.

Eudistomin H:

Brominating the isoquinoline (**38**) to form Euditomine H (**39**) was performed with two different sources of bromo. The source of bromo appeared to be of little significance, it did however prove to be slightly better to use DBH over NBS. It is a significant discovery that DBH function as well as NBS. By utilizing DBH instead of NBS the atom economy increases, and the reaction becomes “greener”.

Conclusion

The synthetic route proved to have an overall yield of 32%, which is lower than previously reported by M. Eriksen. In this project the focus was to optimize the Bishler-Napieralski reaction which was accomplished. The other reactions performed to create Euditomin H have been reported earlier, in higher yield than obtained in this project. M. Eriksen reported 94% yield in the first step [2]. By including this result, the overall yield would increase to 56%, compared to the 44% yield achieved in previous reports.

5. Summary and further work

5.1 Summary

The primary aim of the project was to discover a solid synthetic route for α-carboline and 9H-pyrido[2,3-b]indol-7-ol that could be used as a model for the synthesis of mescengricin. α-carboline was successfully synthesized in a two-step synthesis with high yield (73% overall), and a three-step synthesis with poor yield (38% overall). Two alternative routes was also discovered to create and 9H-pyrido[2,3-b]indol-7-ol, both however, resulted in poor yields <10% and <5% respectively. The two synthetic routes were based on the discoveries made in the synthesis of α-carboline. The first approach is similar to the two-step synthesis of α-carboline, but an additional halide is introduced to perform a S_nAr substitution to cyclize the compound. A significant decrease in yield was achieved in both reactions. Protecting the hydroxy group was performed in the second approach, which resulted in decent yield in the following reactions. It was however, challenging to perform a deprotection, and therefore the compound was never isolated, and an overall yield not calculated. A conclusion was made that protecting the alcohol, results in a more robust synthetic route. In order to synthesize mescengricin additional reactions was performed before the Suzuki reaction could take place. These reactions did run well, and a decent yield was achieved. The Suzuki reaction was never successfully performed and the project to synthesize mescenrgricin failed. If mescengricin could not be synthesized it would be attempted to develop a synthetic route that potentially could be continued in the future, and it is believed the reactions presented in this project could lead to the target compound (**3**).

A secondary objective was to examine the optimal conditions for the Bischler-Naperialski reaction, by using a pure starting material, compared to the telescoped method M. Eriksen performed. The results obtained from these reactions suggest a significant difference in the achieved yield, and which conditions that are optimal for the reaction. Based on these results, it was concluded that purifying the product from the first reaction, in the synthetic route to form Euditomin H, does make a significant difference and should be performed.

5.2 Further work

For future projects, the synthetic route for mescengricin could be completed. Although all attempts on the Suzuki reaction to couple compound **19** and **23** failed, it is still believed the reaction could work under different conditions. Once the coupling is performed the following reactions should run as expected, possibly protecting the two hydroxy groups in the glycerol ester would be necessary. Creating the Minishi reagent still require some work, brominating the compound was never successfully performed, but the reaction should run, if a different source of bromo is used, or the conditions are changed.

A number of synthesis performed in this project have the potential to optimized. Especially the reduction of nitro groups, which appeared to have ~100% conversion from the analysis but, resulted in ~50% yield. Deprotecting the alcohol also have significant potential for improvement, it is known reactions to remove the MOM protection group, but due to lack of time, only one method was attempted. The cyclisation to form α-carboline utilizing the Chichibabin reaction resulted in high yield, it is however reported that by adding an additive like NaI the reaction could result in higher yield, which could be attempted. Considering the similarities in the mechanism between the Chichibabin reaction and the S_nAr cyclisation, adding an additive could possibly achieve positive results in the S_nAr reaction as well.

In the synthetic route to form Eudistomin H, two of the reactions now deliver yield > 90%, which suggest further work are not required. Brominating the compound is the only step that could be improved significantly.

6. Experimental

6.1 General methods

Chemicals

All reagents and solvents used in this project were purchased commercially from different sources and were used as received.

Experimental description

TLC analysis were performed on alumina foil coated with silica gel (silica gel matrix, L x W 10 cm x 20 cm) with fluorescent indicator 254 nm, purchased from Sigma-Aldrich. The mobile phases used are specified in the text.

Instruments description

Automatic flash column chromatography was carried out using a CombiFlash Companion instrument, using Agela flash column silica prepacked columns, and on Interchim Puriflash XS 420 with Silica High Capacity Duo 20 µm columns delivered by Biotage with the eluent specified in the text.

NMR spectra were recorded on a Bruker Biospin AV500 (500MHz for ^{1}H) and (125MHz for ^{13}C). Chemical shift values are reported in ppm, and the multiplicity are reported as: singlet (s), singlet of doublets (sd), doublet (d), doublet of doublet (dd), doublet of doublet of doublet (ddd), doublet of quartet of doublet (dqd), doublet of triplet (dt), triplet (t), triplet of doublet (td), triplet of triplet (tt), triplet of triplet of doublet (ttd), quartet (q), pentet (p) and multiplet (m).

GC-MS analyses were performed with a capillary gas chromatograph equipped with a fused silica column using helium as carrier gas. The gas chromatograph was coupled with a mass spectrometer using electron ionization (EI) as ionization source.

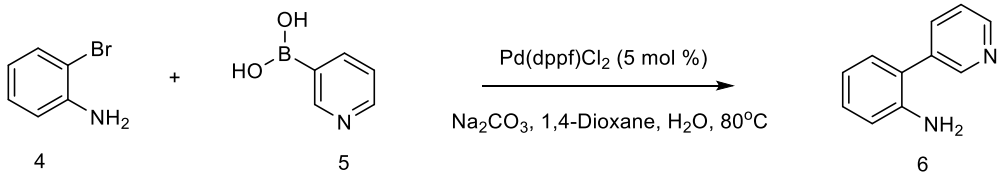
Electrospray ionization (ESI+) and low-resolution mass spectra (LRMS) were recorded on an Agilent 1260 infinity binary liquid chromatograph coupled to an Agilent 6120 quadrupole mass spectrometer.

Microwave: All microwave assisted reactions were carried out on Biotage initiator sixty instrument. Temperature and adsorption are specified in the text.

6.2 Experimental procedures

α-Carboline

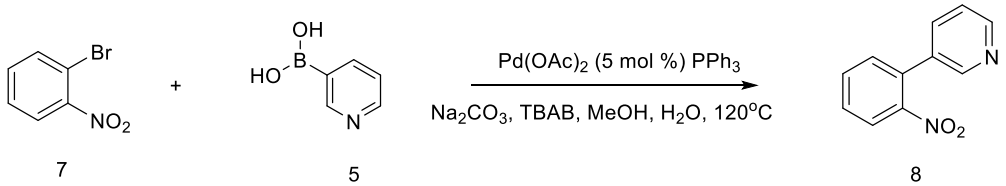
2-(3-pyridinyl)aniline (6)[35]



4 5 6

Pyridin-3-yl boronic acid (**5**) (0.092g, 0.749 mmol) Na_2CO_3 (0.245g, 2.312 mmol) and $Pd(dppf)Cl_2$ (0.020g, 0.027 mmol) were placed in a tube reactor and dissolved in 1,4-dioxane (3.5 mL) and H_2O (1,4 mL). 1-bromoaniline (**4**) (0.100g, 0.581 mmol) was added to the mixture. The reaction mixture was flushed with argon before the tube was sealed and heated to 80°C and left stirring overnight. After the solution was cooled to room temperature, the solvent was removed using a stream of nitrogen. The residue was diluted in 1.5 M NaOH (30 mL) and extracted with DCM (3x30 mL). The combined organic phases were dried over $MgSO_4$, and the solvent was removed under reduced pressure to obtain the crude product. Further purification was performed using silica column chromatography to obtain the product (**6**) as a yellow oil (0.0859g, 87% yield). R_f = 0.38 (Hex:EtOAc:MeOH 70:15:15). ^{1}H NMR (500 MHz, DMSO): δ 8.62 (d, 1H), 8.54 (dd, 1H), 7.85 (dt, 1H), 7.45 (ddd, 1H), 7.10 (td, 1H), 7.01 (dd, 1H), 6.80 (dd, 1H), 6.67 (td, 1H), 4.91 (s, 1H) ppm. ^{13}C NMR (125 MHz, DMSO): δ = 149.33, 147.71, 145.52, 136.12, 135.39, 130.25, 128.86, 123.63, 122.23, 116.82, 115.48 ppm. m/z (EI): m/z (%) 170.2 (100, M^+), 155.1 (1), 143.1 (33), 128.1 (1), 115.1 (24), 102.1 (1).

3-(2-nitrophenyl)pyridine (8)[23]

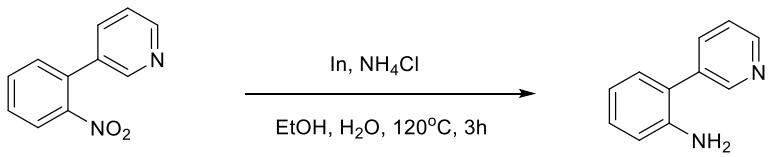


7 5 8

2-bromonitrobenzene (**7**) (0.145g, 0.718 mmol), 3-pyridinyl-boronaic acid (**5**) (0.149g, 1.212 mmol), Na_2CO_3 (0.102g, 0.962 mmol), TBAB (0.071 mmol), $Pd(OAc)_2$ (0.008g, 0.036 mmol) and PPh_3 (0.019g, 0.072 mmol) were placed in a tube reactor. A mixture of MeOH (4 mL) and H_2O (1.2 mL) was used to dissolve the reagents. The mixture was sparged with argon

before the tube was sealed; it then was heated to 120°C and left stirring for 2 hours. After the solution was cooled to room temperature, the solvent was removed under reduced pressure. The residue was dissolved in water (30 mL) and extracted with diethyl ether (2 x 30 mL); the organic layers were combined and dried over Na_2CO_3, and the solvent was removed under vacuo to obtain the crude product. Further purification was performed using silica column chromatography to obtain the product (**8**) as a yellow oil (0.126 g, 88% yield). R_f = 0.3. (Hex:EtOAc 4:6). ^{1}H NMR (500 MHz, CDCl): δ 8.67(dd, 1H), 8.60 (dd, 1H), 8.00 (dd, 1H), 7.76 (m, 2H), 7.58 (ddd, 1H), 7.44 (dd, 1H), 7.38 (ddd, 1H) ppm. ^{13}C NMR (125 MHz, CDCl): δ = 149.25, 148.39, 135.61, 133.74, 133.08, 132.87, 132.17, 132.14, 129.20, 124.67, 123.28 ppm.

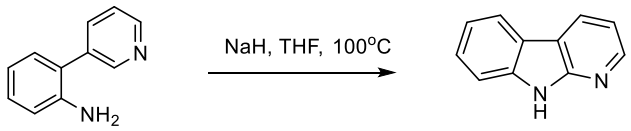
2-(3-Pyridinyl)aniline Indium reduction (6)[23]



8 6

NH_4Cl (0.055g, 1.03 mmol) and Indium powder (0.163g, 1.42 mmol) was dissolved in H_2O (1.2 mL) and transferred to a tube reactor. 3-(2-nitrophenyl)pyridine (**8**) (0.0967g, 0.483 mmol) was dissolved in ethanol (4 mL) and added to the solution. The tube was sealed, heated to 120°C, and left stirring. The reaction was monitored using LC-MS, and, after 3 hours, the starting material was consumed. After the solution had cooled to room temperature, the mixture was diluted with EtOAc (30 mL) and filtered through a pad of celite. Additional EtOAc (20 mL) was used to wash through the pad of celite. The filtrate was dried over Na_2SO_4, and solvent was removed under reduced pressure to obtain the product (**6**) observed as a yellow oil (0.043g, 52% yield). ^{1}H NMR (500 MHz, CDCl): δ 8.72 (dd, 1H), 8.66 (dd, 1H), 7.82 (dt, 1H), 7.38 (ddd, 1H), 7.20 (ddd, 1H), 7.11 (dd, 1H), 6.86 (td, 1H), 6.79 (dd, 1H), 3.75 (s, 2H) ppm. ^{13}C NMR (125 MHz, DMSO): δ = 149.33, 147.71, 145.52, 136.12, 135.39, 130.25, 128.86, 123.63, 122.23, 116.82, 115.48 ppm. m/z (EI): m/z (%) 170.2 (100, M^+), 155.1 (1), 143.1 (33), 128.1 (1), 115.1 (24), 102.1 (1).

α-Carboline (1) [32]

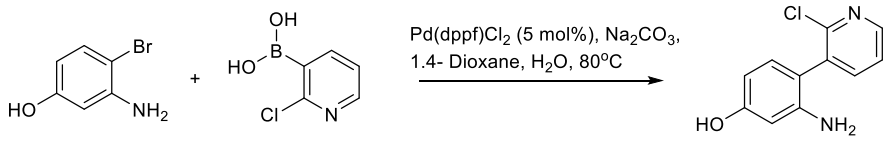


6 1

2-(3-pyridinyl)aniline (**6**) (0.064g, 0.376 mmol) was dissolved in THF (2 mL) in a tube reactor. NaH (60%) (0.045g, 1.876 mmol) was added carefully, as gas formation was observed upon addition. The tube was sealed when the gas ceased, and the mixture was heated to 100°C, the reaction was monitored using TLC. After 20 hours, the reaction appeared to have stopped, and the mixture was allowed to cool to room temperature before the solvent was removed under vacuo. The residue was quenched in ice cold water (15 mL), and the pH was neutralized using HCl (1M). The solution was extracted with DCM (3 x 30 mL), and the organic layers were combined and dried over $MgSO_4$. The solvent was removed under reduced pressure to obtain the crude product. Further purification was performed using silica column chromatography to obtain the product (**1**) as white crystals (0.053g, 84% yield). R_f = 0.5 (Hex:EtOAc 6:4), m.p = 210.2-214.6°C. ^{1}H NMR (500 MHz, DMSO): δ 8.43 (ddd, 1H), 8.35 (dd, 1H), 8.09 (dp, 1H), 7.44 (dt, 1H), 7.38 (ddd, 1H), 7.14 (m, 2H) ppm. ^{13}C NMR (125 MHz, DMSO) δ = 151.87, 146.02, 138.75, 128.33, 126.53, 121.10, 120.33, 119.33, 115.12, 114.90, 111.18 ppm. MS (ESI+): $(M+H)^+$ calcd for $C_{11}H_8N_2$ 169.2 found = 169.4

9H-pyrido[2,3-b]indol-7-ol

3-amino-4-(2-chloropyridin-3-yl)phenol (11)[35]

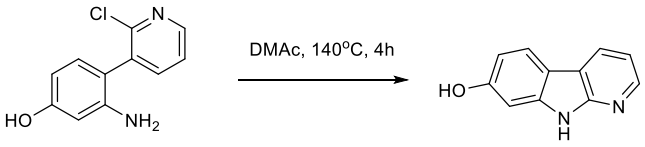


9 10 11

3-Amino-4-bromophenol (**9**) (0.186 g, 0.989 mmol), 2-chloropyridin-3-yl boronic acid (**10**) (0.205 g, 1.303 mmol), Na_2CO_3 (0.423 g, 3.991 mmol) and $Pd(dppf)Cl_2$ (0.036 g, 0.049 mmol) were placed in a 50 mL round bottom flask. 1.4-dioxane (7.0 mL) and H_2O (2.8 mL) was added as solvents. The round bottom flask was capped and flushed with argon before being heated to 80°C and left stirring overnight. After the reaction mixture was cooled to ambient temperature, the solvent was removed under reduced pressure. The residue was dissolved in water (30 mL) and extracted with DCM (3 x 30 mL). The combined organic

layers were dried with Na_2SO_4 before the solvent was removed. Further purification was performed using silica column chromatography to obtain the product (**11)** (0.066 g, 28% yield). R_f =0.063 (DCM:MeOH 95:5). NMR spectra not available due to corrupted file. MS (ESI+): $(M+H)^+$ calcd for $C_{11}H_7N_2OCl$ 221.66 found = 221.6.

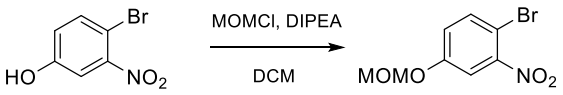
9H-pyrido[2,3-b]indole (2)



11 2

3-amino-4-(2-chloropyridin-3-yl)phenol (**11**) (0.186 g, 0.840 mmol) was placed in a 25 mL round bottom flask, and dissolved in DMAc (2 mL). The flask was capped before the reaction mixture was heated to 140°C and left stirring for 4 hours. Workup and purification were not performed, and NMR was not recorded. MS (ESI+): $(M+H)^+$ calcd for $C_{11}H_7N_2O$ 185.2 found =185.1

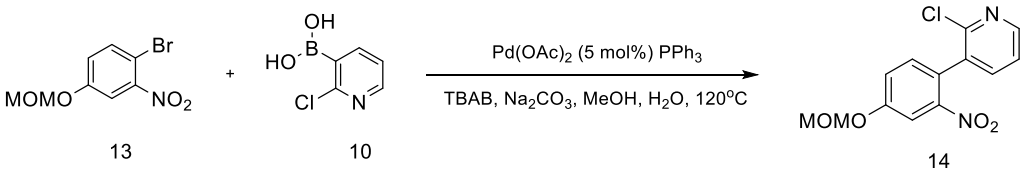
MOM protection of 4-bromo-3-nitrophenol (13) [36]



12 13

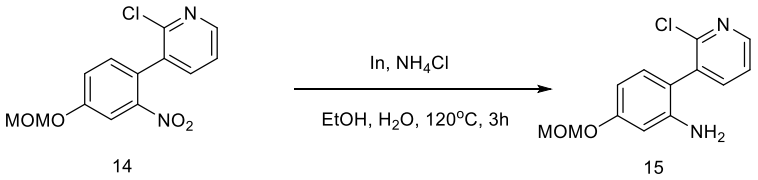
4-Bromo-3nitrophenol (**12**) (1.001 g, 4.591 mmol) was placed in a dry 100 mL round bottom flask and dissolved in anhydrous DCM (10 mL). The round bottom flask was capped and flushed with nitrogen before cooled to 0°C. Diisopropylethyl amine (1,6 mL, 9.182 mmol) and chloromethyl methyl ether (0.7 mL, 9.182 mmol) was added to the solution; the mixture was stirred for 30 minutes at 0°C and was left over night at room temperature. The mixture was diluted with 10% NaOH (30 mL) and extracted with DCM (3 x 30 mL). The combined organic layers were dried over $MgSO_4$, before being washed through a silica plug using additional DCM (200 mL). The solvent was removed under reduced pressure to obtain the product (**13**) as an orange oil. The product was cooled to 0°C to give an orange solid (1.183 g, 98% yield). ^{1}H NMR (500 MHz, CDCl): δ 7.53 (d, 1H), 7.47 (d, 1H), 7.05 (dd, 1H), 5.13 (s, 2H), 3.41 (s, 3H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 156.77, 150.23, 135.48, 121.52, 113.47, 105.77, 94.69, 56.41 ppm.

2-Chloro-3-(2-nitrophenyl)pyridine (14)[23]



A tube reactor was charged with 1-bromo-4-(methoxymethoxy)-2-nitrobenzene (**13**) (0.262 g, 1.000 mmol), (2-chloropyridin-3-yl)boronic acid (**10**) (0.276 g, 1.754 mmol), Na_2CO_3 (0.144 g, 1.359 mmol), TBAB (0.031g, 0.096 mmol), $Pd(OAc)_2$ (0.008g, 0.036 mmol), PPh_3 (0.019g, 0.072 mmol). A mixture of MeOH (4 mL) and H_2O (1 mL) was added to the tube. The reaction mixture was purged with argon before the tube was sealed, heated to 120°C, and left stirring for 2 hours. The solution was cooled to room temperature before the solvent was removed under reduced pressure. The residue was diluted with water (40 mL) and extracted with diethyl ether (2 x 30 mL). The organic layers were combined, dried over Na_2SO_4, and the solvent removed under vacuo. Further purification was performed using silica column chromatography to obtain the product (**14**) as a yellow oil (0.229g, 78% yield). R_f = 0.3 (Hex:EtOAc 7:3) ^{1}H NMR (500 MHz, CDCl): δ 8.47 (dd, 1H), 7.91 (dd,1H), 7.84 (d, 1H), 7.54 (m, 3H), 5.40 (s, 2H), 3.45 (s, 3H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 157.21, 149.02, 148.62, 148.23, 139.64, 133.48, 133.30, 124.76, 123.37, 121.76, 111.82, 94.22, 56.03 ppm.

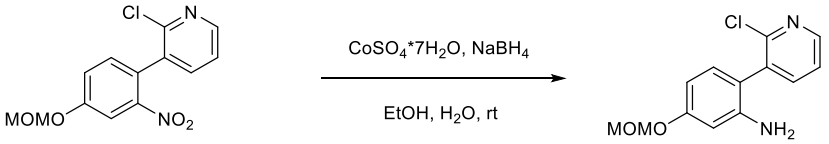
2-(2-chloropyridin(3-yl)-5-(methoxymethoxy)aniline indium reduction (15) [23]



NH_4Cl (0.064 g, 1.196 mmol) and indium powder (0.196, 1.707 mmol) was dissolved in water (1.2 mL) and transferred to a tube reactor. 2-chloro-3-(4-(methoxymethoxy)-2-nitrophenyl)pyridine (**14**) (0.174 g, 0.590 mmol) was dissolved in ethanol and added to the tube. The tube was sealed, heated to 120°C, and left stirring. The reaction was monitored using GC-MS, and, after 3 hours, the starting material was consumed. After the solution had cooled to room temperature, the mixture was diluted with EtOAc (30 mL) and filtered through a pad of celite. Additional EtOAc (20 mL) was used to wash through the celite. The filtrate was dried over Na_2SO_4, and the solvent was removed under reduced pressure to obtain the

product (**15**) observed as a yellow oil (0.078 g, 50% yield).) ^{1}H NMR (500 MHz, DMSO): δ 8.31 (dd, 1H), 7.66 (dd, 1H), 7.38 (dd, 1H), 6.74 (d, 1H), 6.35 (d, 1H), 6.22 (dd, 1H), 5.06 (s, 2H), 4.77, (s, 2H), 3.32, (s, 3H) ppm. ^{13}C NMR (125 MHz, DMSO) δ =157.82, 150.41, 148.38. 146.82, 141.39, 134.30, 130.93, 123.27, 115.64, 103.97, 101.93, 93.54, 55.45 ppm.

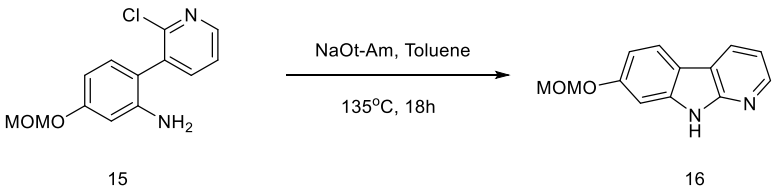
2-(2-chloropyridin(3-yl)-5-(methoxymethoxy)aniline Cobolt reduction (15) [25]



14 15

2-chloro-3-(4-(methoxymethoxy)-2-nitrophenyl)pyridine (**14**) (0.222g, 0.775 mmol) was dissolved in ethanol (4 mL) and added to a 50 mL round bottom flask. Co_2SO_4 * $7H_2O$ (0.212, 0.972 mmol) was dissolved in H_2O (1 mL) and added to the round bottom flask. $NaBH_4$ (0.114, 3.010 mmol) was added slowly to the pink mixture. Upon addition, dark precipitate was observed instantly. The solution was left stirring at room temperature for 10 minutes, before being quenched with water (15 mL) and filtrated using a Büchner funnel to remove the precipitate. The filtrate was diluted with EtOAc (50 mL) and water (50 mL). The aqueous phase was extracted with EtOAc (2 x 30 mL). The combined organic layers were dried over Na_2SO_4, and the solvent removed using reduced pressure. Further purification was performed applying silica column chromatography (Hexane:EtOAc 1:1) to obtain the product (**15**) as a yellow oil (0.089g, 45% yield). ^{1}H NMR (500 MHz, DMSO): δ 8.31 (dd, 1H), 7.66 (dd, 1H), 7.38 (dd, 1H), 6.74 (d, 1H), 6.35 (d, 1H), 6.22 (dd, 1H), 5.06 (s, 2H), 4.77, (s, 2H), 3.32, (s, 3H) ppm. ^{13}C NMR (125 MHz, DMSO) δ = 157.82, 150.41, 148.38. 146.82, 141.39, 134.30, 130.93, 123.27, 115.64, 103.97, 101.93, 93.54, 55.45 ppm.

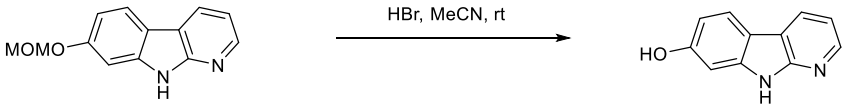
7-(methoxymethoxy)-9H-pyrido[2,3-b]indole (16)



15 16

2-(2-chloropyridin(3-yl)-5-(methoxymethoxy)aniline (**15**) (0.089g, 0.339 mmol) was dissolved in toluene (1 mL) and transferred to a dry tube reactor. Sodium tert-pentoxide (40% in toluene) (0.30 mL, 1.02 mmol) was added slowly to the tube. The mixture was purged with argon before the tube was sealed, heated to 135°C, and left stirring for 18 hours. The solution was cooled to room temperature before being quenched in ice cold water (15 mL). HCl (1M) was added to neutralize the solution before it was extracted with DCM (3 x 30 mL). The organic phases were combined and dried over $MgSO_4$, and the solvent was removed under vacuo to obtain the product (**16**) as yellow crystals (0.038g, 49% yield). ^{1}H NMR (500 MHz, DMSO): δ 8.45 (m, 2H), 8.10 (d, 1H), 7.22 (dd, 1H), 7.18 (sd, 1H), 6.97 (dd, 1H), 5.34 (s, 2H), 3.49 (s, 3H) ppm. ^{13}C NMR (125 MHz, DMSO) δ = 156.45, 148.38, 144.56, 141.40, 130.94, 127.41, 122.03, 114.97, 109.91, 101.97, 97.70, 94.31, 55.57 ppm. MS (ESI+): $(M+H)^+$ calcd for $C_{13}H_{13}N_2O_2$ 229.5 found = 229.5.

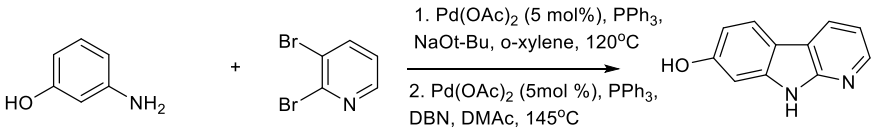
Deprotection of 7-(methoxymethoxy)-9H-pyrido[2,3-b]indole (16) [37]



16 2

7-(methoxymethoxy)-9H-pyrido[2,3-b]indole (**16**) was dissolved in MeCN (2 mL) and placed in a 50 mL round bottom flask. HBr (48%) (10 mL) was added to the flask, and the mixture was left stirring overnight. The solution was quenched with $NaHCO_3$ (s) and filtered. Further workup was not performed. Product (**2**) confirmed by LC-MS. MS (ESI+): $(M+H)^+$ calcd for $C_{11}H_9N_2O$ = 185.2 found = 185.1

9H-pyrido[2,3-b]indole (2) [38]

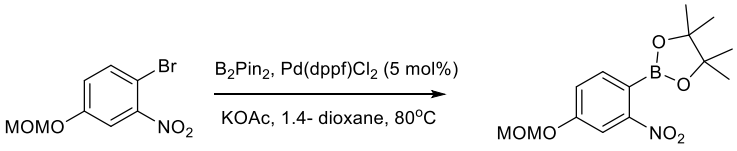


17 18 2

A 100 mL round bottom flask was charged with 3-aminophenol (**17**) (0.210 g, 1.110 mmol), 2,3-dibromopyridine (**18**) (0.237 g, 1.000 mmol), $Pd(OAc)_2$ (0.017 g, 0.052 mmol), PPh_3 (0.027 g, 0.100 mmol) and NaOt-Bu (0.114 g, 1.190 mmol). O-xylene 2.5 mL was added before the flask was capped and flushed with argon. The reaction mixture was heated to 120°C and left stirring for 3 hours. The solution was cooled to room temperature before additional $Pd(OAc)_2$ (0.017 g, 0.052 mmol), PPh_3 (0.027 g, 0.100 mmol), DBN (0.3 mL, 2.456 mmol) and DMAc (2.5 mL) were added. The flask was capped and flushed with argon once more before the mixture was heated to 145°C. The reaction was monitored with LC-MS, no product was observed in the analysis, and the reaction was stopped. Workup was not performed.

Mescengricin

2-(4-(methoxymethoxy)-2-nitrophenyl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (19) [39]

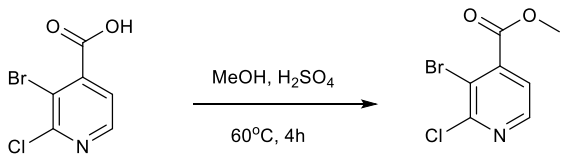


13 19

B_2Pin_2 (0.206 g, 0.811 mmol), KOAc (0.197 g, 2.007 mmol), $Pd(dppf)Cl_2$ (0.025 g, 0.034 mmol) were added to a dry tube reactor and dissolved in anhydrous 1.4-dioxane. 1-bromo-4-(methoxymethoxy)-2-nitrobenzene (**13**) (0.176 g, 0.672 mmol) was added to the stirred mixture. The tube was flushed with argon before it was sealed, heated to 80°C, and left stirring overnight. The reaction mixture was cooled to room temperature, and solvent was removed using a gentle stream of nitrogen. The residue was dissolved in EtOAc (30 mL) and washed with distilled water (2 x 30 mL). The organic layer was dried over Na_2SO_4, and the solvent was removed using reduced pressure. Further purification was performed using silica column chromatography to obtain the product (**19**) as a yellow solid (0.160 g, 77% yield). R_f = 0.84 (Hexane:EtOAc 7:3). ^{1}H NMR (500 MHz, CDCl): δ 7.73 (d, 1H), 7.39 (d, 1H), 7.24 (dd,

1H), 5.16 (s, 2H), 3.40 (s, 3H), 1.34 (s, 12H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 158.52, 152.39, 133.87, 121.78, 110.73, 94.43, 84.547 56.26, 24.71 ppm.

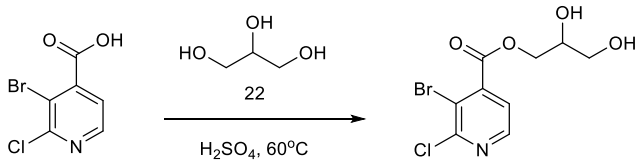
Methyl 3-bromo-2-chloroisonicotinate (21) [40]



20 21

3-bromo 2-chloroisonicotinic acid (**20**) (0.100 g, 0.419 mmol) was dissolved in methanol (2 mL) in a dry 25 mL round bottom flask. H_2SO_4 (98%) (0.03 mL, 0.559 mmol) were added before the mixture was heated to reflux and left stirring for 4 hours. The mixture was cooled to room temperature and concentrated under vacuo. The residue was dissolved in saturated aqueous $NaHCO_3$ and extracted with DCM (3 x 30 mL). The combined organic layers were dried over $MgSO_4$, and the solvent was removed under reduced pressure to obtain the product (**21**) observed as brown crystals (0.096 g, 91% yield). ^{1}H NMR (500 MHz, CDCl): δ 8.34 (d, 1H), 7.34 (d, 1H), 3.91 (s, 3H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 164.97, 153.00, 147.85, 143.80, 122.36, 118.39, 53.33 ppm.

2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (23) [40]

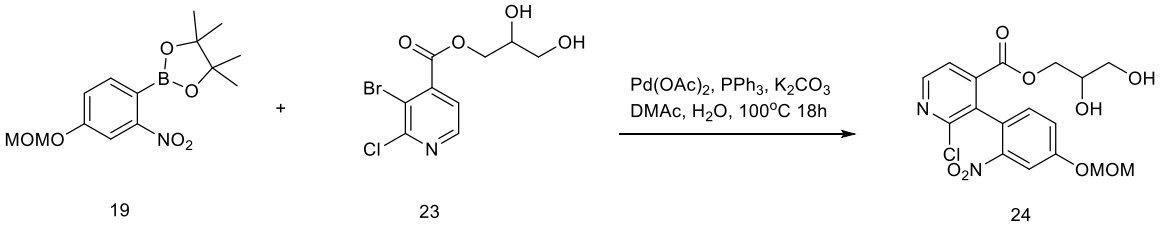


20 23

3-Bromo 2-chloroisonicotinic acid (**20**) (0.099 g, 0.419 mmol), anhydrous glycerol (**22**) (3 mL, 41.047 mmol), and H_2SO_4 (98%) (0.04 mL, 0.600 mmol) were placed in a dry 50 mL round bottom flask. The round bottom flask was capped before the mixture was heated to 60°C and left stirring overnight. The mixture was cooled to room temperature, quenched in saturated aqueous $NaHCO_3$ (20 mL), and extracted with DCM (3 x 30 mL). The combined organic layers were dried over $MgSO_4$ and the solvent was removed under reduced pressure to obtain the product (**23**) as a brown oil (0.109 g, 84% yield). ^{1}H NMR (500 MHz, DMSO): δ 8.57 (d, 1H), 7.72 (d, 1H), 5.09 (d, 1H), 4.76 (t, 1H), 4.46 (d, 2H), 4.24 (dd, 1H), 3.79 (ttd,

1H), 3.65 (m, 1H) ppm. ^{13}C NMR (125 MHz, DMSO) δ = 164.19, 151.14, 148.95, 122.93, 116.88, 72.46, 68.98, 67.96, 62.42 ppm.

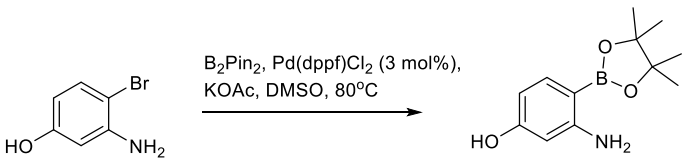
2,3-dihydroxypropyl 2-chloro-3-(4-(methoxymethoxy)-2-nitrophenyl)isonicotinate (24) [41]



19 23 24

A 50 mL round bottom flask was charged with 2-(4-(methoxymethoxy)-2-nitrophenyl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (**19**) (0.142 g, 0.459 mmol), 2,3-dihydroxypropyl 3-bromo-2-chloroisonicotinate (**23**) (0.139 g, 0.448 mmol), K_2CO_3 (0.144 g, 1.042 mmol), $Pd(OAc)_2$ (0.002 g, 0.009 mmol) and PPh_3 (0.006 g, 0.023 mmol). The compounds were dissolved in a mixture of DMAc and H_2O (4:1) (5 mL), before the flask was capped, and the mixture was purged with argon. The solution was heated to 100°C and left stirring for 18 hours. After the mixture had cooled to room temperature the solvent was removed using a gentle stream of nitrogen. EtOAc (30 mL) was used to dissolve the residue and the solution was washed with water (2 x 30 mL). Na_2SO_4 was added as a drying agent to the combined organic layers before the solvent was removed under vacuo. The product (**24**) was not observed in the NMR analysis.

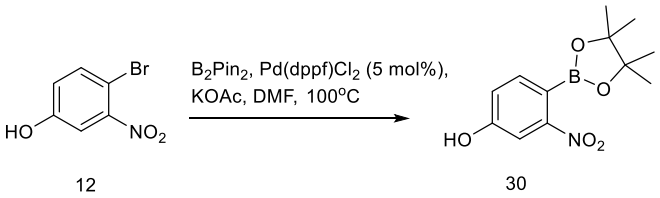
3-amino-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenol (27) [42]



9 29

A 50 mL round bottom flask was charged with B_2Pin_2 (0.372 g, 2.900 mmol), KOAc (0.779 g, 7.940 mmol) and $Pd(dppf)Cl_2$ (0.058 g, 0.0790 mmol). The flask was capped and flushed with argon. 3-amino-4-bromophenol (**9**) (0.501 g, 2.660 mmol) was dissolved in DMSO (15 mL) and injected into the flask. The mixture was heated to 80°C under stirring, and monitored by LC-MS. After 18 hours LC-MS suggested no product was formed and the reaction was stopped. Workup was not performed.

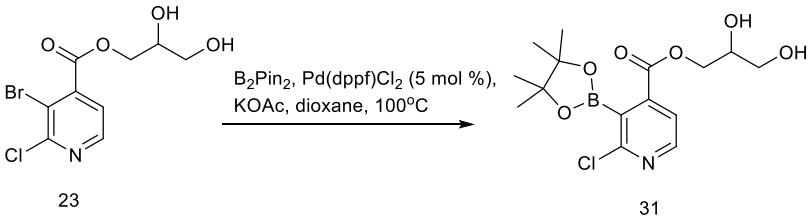
3-nitro-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenol (30) [39]



12 30

4-bromo-3-nitrophenol (**12**) (0.218g, 1.00 mmol), B_2Pin_2 (0.305g, 1.2 mmol), potassium acetate (0.294g, 3.00 mmol) and $Pd(dppf)Cl_2$ (0.036g, 0.05 mmol) was placed in a tube reactor and dissolved in DMF (3 mL). The tube was flushed with argon, capped and heated to 100°C and left stirring for 2 hours. The solvent was removed using a stream of nitrogen. Ethyl acetate was used to dissolve the residue, and the mixture was washed with distilled water (30 mL). The organic layer was dried with $MgSO_4$, and the solvent was removed under reduced pressure. Further purification was performed using a silica column with a gradient elution system consisting of dichloromethane and methanol to obtain the product (**30**) (0.078 g, 30% yield) as a yellow solid. ^{1}H NMR (500 MHz, DMSO): δ 10.67 (s, 1H), 7.67 (d, 1H), 7.36 (d, 1H), 7.02 (dd, 1H), 1.17 (s, 12H) ppm. ^{13}C NMR (125 MHz, DMSO) δ = 157.57, 150.09, 121.22, 112.13, 100.83, 82.81, 24.80 ppm.

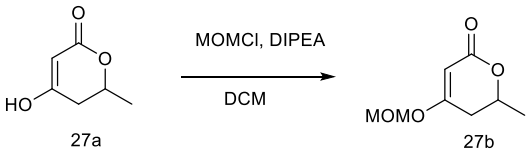
2,3-dihydroxypropyl 2-chloro-3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)isonicotinate (31) [43]



23 31

A tube reactor was charged with 2,3-dihydroxypropyl -3-bromo-2-chloroisonicotinate (**23**) (0.062 g, 0.199 mmol), B_2Pin_2 (0.077 g, 0.303 mmol), KOAc (0.060 g, 0.611 mmol), $Pd(dppf)Cl_2$ (0.007 g, 0.009 mmol). Anhydrous 1,4-dioxane (4 mL) was used to dissolve the compounds. The tube was purged with argon before being sealed and the mixture heated to 100°C and left stirring overnight. The solution was cooled to room temperature before the solvent was removed using a stream of nitrogen. The residue was dissolved in ethyl acetate (30 mL) and washed with water (2 x 30 mL). Na_2SO_4 was added to the organic layer as a drying agent and the solvent was removed under reduced pressure. The product was not found in the NMR analysis.

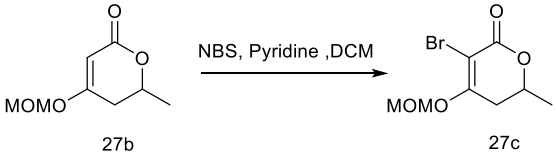
4-(methoxymethoxy)-6-methyl-5,6-dihydro-2H-pyran-2-one (27b) [36]



27a 27b

5,6-Dihydo-4-hydroxy-6-methyl-2H- pyran-2-one (**27a**) (0.128 g, 0.999 mmol) was placed in a dry 50 mL round bottom flask and dissolved in anhydrous DCM (3 mL). The solution was cooled to 0^{o}C before N,N-diisopropylethylamine (0.35 mL, 2.009 mmol) and chloromethyl methyl ether (0.15 mL, 1.975 mmol) were added under a nitrogen atmosphere. The reaction mixture was stirred at 0^{o}C for 30 minute and left stirring at ambient temperature overnight. The mixture was diluted with 10% aqueous NaOH (30 mL) before being extracted with DCM (3 x 30 mL). The combined organic layers were dried over $MgSO_4$ and the solvent was removed under reduced pressure. Further purification was applied using silica column chromatography (Hexane:EtOAc 1:1) to obtain the product (**27b**) as a clear oil (0.133 g, 78% yield). ^{1}H NMR (500 MHz, DMSO): δ 5.19 (d, 1H), 5.17 (s, 2H), 4.51 (dqd, 1H), 3.40 (s, 3H), 2.48 (m, 3H), 1.33 (d, 3H) ppm. ^{13}C NMR (125 MHz, DMSO) δ = 170.05, 166.14, 94.23, 92.13, 71.79, 56.61, 33.34, 20.10 ppm.

3-Bromo-4-(methoxymethoxy)-6-methyl-5,6-dihydro-2H-pyran-2-one (27c) [44]

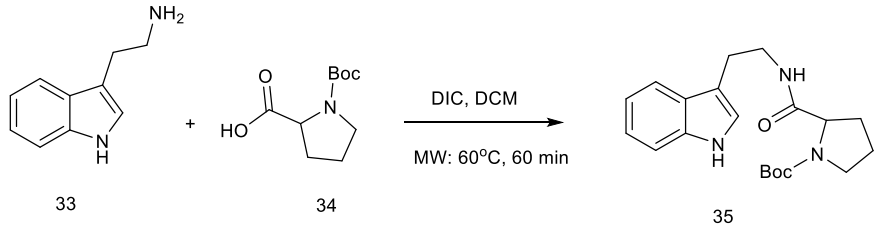


27b 27c

4-(methoxymethoxy)-6-methyl-5,6-dihydro-2H-pyran-2-one (**27b**) (0.105 g, 0.610 mmol), NBS (0.131 g, 0.736 mmol) and pyridine (0.3 mL, 3.724 mmol) were added to a 25 mL round bottom flask. DCM (3 mL) was added as a solvent before the mixture was left stirring at ambient temperature. The reaction was monitored using GC-MS, and after 5 hours, it was suggested the reaction did not start and workup was not performed.

Optimization of the Bischler-Napieralski reaction

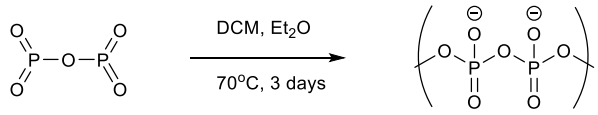
N'-(N-Boc-prolyl)tryptamine (35) [2]



33 34 35

A microwave reactor tube was charged with N-Boc-proline (**34**) (2.003 g, 9.314 mmol), tryptamine (**33**) (2.106 g, 13.134 mmol) and DIC (1.9 mL, 12.13 mmol). DCM (10 mL) was used to dissolve the compounds, before the tube was sealed and placed in the microwave. The mixture was heated to 60°C for 1 hour with adsorption settings on normal. The solution was diluted with 0.5 M aqueous citric acid (75 mL) and extracted with DCM (150 mL). The organic phase was first washed with saturated aqueous solution of $NaHCO_3$ (100 mL) and then brine (100 mL). $MgSO_4$ was added to the organic layer as a drying agent before the solvent was removed under reduced pressure to obtain the crude as a dark oil. Further purification was performed using silica column chromatography to obtain the product (**35**) as a light brown solid (1.802 g 54% yield). R_f = 0.11 (Hexane:EtOAc 1:1). ^{1}H NMR (500 MHz, CDCl): δ 8.26 (s, 1H), 7.53 (d, 1H), 7.29 (dt, 1H), 7.12 (t, 1H), 7.05 (t, 1H), 6.97 (s, 1H), 4.14 (d, 2H), 3.78 (p, 1H), 3.52 (d, 2H), 3.23 (m, 2H), 2.88 (q, 2H), 2.01 (d, 1H), 1.74 (m, 2H), 1.31 (m, 9H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 157.62, 136.44, 122.25, 122.13, 119.42, 118.77, 118.64, 113.20, 111.28, 42.19, 42.15, 40.83, 28.32, 28.38, 25.99, 25.35, 23.52, 23.40 ppm.

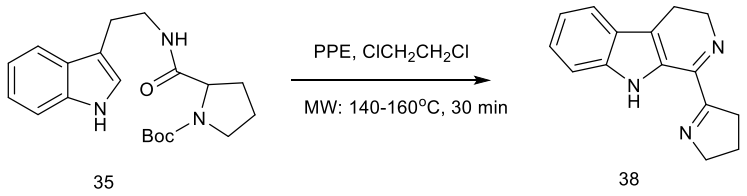
PPE (37) [2]



36 37

P_2O_5 (**36**) (60 g, 42.285 mmol) was added to a dry 500 mL round bottom flask. Anhydrous DCM (100 mL) and diethyl ether (200 mL) was added to the flask. The reaction mixture was heated to 70°C and left stirring for 3 days. After the solution was cooled to room temperature the solvent was removed under reduced pressure to obtain the product (**37**) as a clear brown viscous liquid (110 g).

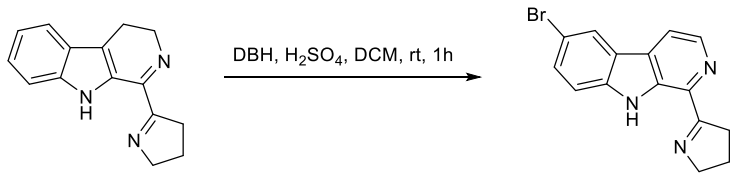
1-(3,4-dihydro-2H-pyrrol-5-yl)-4,9-dihydro-3H-pyrido[3,4-b]indole (38) [2]



35 38

A mixture of N'-(N-Boc-prolyl)tryptamine (**35**) (0.100 g, 0.279 mmol), PPE (**37**) (4 g) and 1,2-dichloroethane were added to a reactor tube. The tube was sealed and placed in the microwave. The reaction mixture was heated to 155°C for 30 minutes under very high absorption. After the solution had cooled to room temperature it was quenched with water (20 mL) and left stirring overnight. 3.5 M aqueous KOH (15 mL) was used to basify the solution, the reaction mixture turned orange upon addition of the base. The solution was extracted with toluene (2 x 20 mL) and the combined organic phases were dried over Na_2SO_4. The solvent was removed under vacuo to obtain the product (**38**) as a dark oil (0.066 g, 96% yield). R_f = 0.11 (Hexane:EtOAc 1:1). ^{1}H NMR (500 MHz, CDCl): δ 10.36 (s, 1H), 7.59 (d, 1H), 7.41 (dd, 1H), 7.25 (m, 2H), 7.12 (t, 1H), 4.17 (t, 2H), 4.10 (t, 2H), 2.95 (m, 4H), 1.98 (q, 2H), 1.14 (d, 1H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 175.86, 154.32, 136.59, 128.01, 124.97, 124.37, 119.84, 119.80, 116.26, 112.28, 62.39, 49.33, 34.25, 23.51, 21.60, 19.02 ppm.

Eudistomin H (39) [2]



38 39

1-(2-pyrrolinyl)-3,4-dihydro-β-carboline (**38**) (0.016 g, 0.046 mmol), DHB (0.015 g, 0.053 mmol) and sulfuric acid (98%) (0.8 mL, 14.927 mmol) were placed in a 25 mL round bottom flask and dissolved in DCM (1.2 mL). The flask was capped and flushed with nitrogen. The mixture was left stirring for 1 hour under room temperature and dark conditions. The solution was diluted with additional DCM (30 mL) and washed with saturated aqueous $NaHCO_3$ (30 mL) then with brine (30 mL). The organic layer was dried with Na_2SO_4 before the solvent was removed under vacuo. Further purification was performed with silica column chromatography to obtain the product (**39**) as a brown oil (0.012 g, 62% yield). ^{1}H NMR (500 MHz, CDCl): δ 8.52 (d, 1H), 8.27 (dd, 1H), 7.95 (d, 1H), 7.65 (dd, 1H), 7.47 (dd, 1H), 4.27

(m, 2H), 3.32 (ddt, 2H), 2.10 (dq, 3H) ppm. ^{13}C NMR (125 MHz, CDCl) δ = 139.33, 138.52, 131.33, 124.58, 116.14, 113.46, 112.84, 62.19, 34.90, 31.94, 29.67, 29.37, 22.70, 21.85, 14.13 ppm.

7. References

1. R. Ghahremarizadeh, S. Ahadi, A. Bazgir, *Tetrahedron Lett*. **2009**, *50*, 7379-7381.

2. M. Eriksen, Synthesis of the Marine Alkaloid Eudistomin H and Novel Derivatives. Master thesis, University of Bergen, **2013**.

3. W.A. Kakula-Koch, J. Widelski, Alkaloids. In *Pharmacognosy, Academic Press*: Lublin, **2016**, pp 163-198.

4. A. D. Wadsworth, B. J. Naysmith, M. A. Brimble, *Eur. J. Med. Chem.* **2015,** *97*, 816-829

5. K. Shin-ya, J.-S. Kim, K. Furihata, Y. Hayakawa, H. Seto, *J. Asian Nat. Prod. Res.* **2000**, *2*, 121-132.

6. D. W. Choi, *J. Neurosci.* **1990**, 10, 2493-2501.

7. Hino, T.; Ziping, L.; Seki, H.; Hara, R.; Kuramochi, T.; Nakagawa, *M. Chem. Pharm. Bull*. **1989**, *37*, 2596–2600

8. N. Kuhnert, *Angew. Chem. Int. Ed.* **2002**, *41*, 1863-1866

9. P. Ayare, V. Khanvilkar, N. Chalak, *PharmaTutor*, **2014**, *2* (5), 89-103.

10. H. M. Mcnair, J. M. Miller, *Basic Gas Chromatography*, Wiley, Hoboken, **2009**. pp 1-233.

11. J. Sneddon, S. Masuram, J. C. Richert, *Anal Lett*, **2007**, *40*, 1003-1012.

12. W. M. A. Niessen *Liquid Chromatography-Mass Spectrometry*, Taylor & Francis, Boca Raton, **2006**. pp 1-632.

13. B. Zhou, Computational Analysis of LC-MS/MS Data for Metabolite Identification, Master thesis, Virginia Polytechnic Institute and State University, **2011**

14. M. Wilm, *Mol. Cell. Proteomics*, **2011**, *10* M111.009407

15. N. E. Jacobsen, Fundamentals of NMR Spectroscopy in Liquids. In *NMR spectroscopy explained: Simplified Theory, Applications and Examples for Organic Chemistry and Structural Biology*; Wiley, Hoboken, **2007**. pp 1- 39.

16. R. O. de Souza, L. S. M. Miranda, U. T. Bornscheuer, *Chem. Eur. J.* **2017**, *23*, 12040-12063.

17. S. Warren, P. Wyatt, *Organic synthesis: The Disconnection Approach*, Wiley: Croydon, **2008**.

18. B. Zhao, Z. Han, K. Ding, *Angew. Chem. Int. Ed.* **2013**, *52*, 4744-4788.

19. J. E. Luebering, Suzuki Akira https://www.britannica.com/biography/Suzuki-Akira (accessed 17. March 2020)

20. A. A. C. Braga, G. Ujaque, F. Maseras, *Organometallics*, **2006**, *25*, 3647-3658.

21. R. Martin, S. L. Buchwald, *Acc. Chem. Res.* **2008**, *41*, 1461-1473.

22. K. Breuker, H. C. van der Plas, *J. Org. Chem.* **1979**, *44*, 4677-4680

23. V. Elumalai, C. Gambarotti, H.-R. Bjørsvik, *Eur. J. Org Chem.* **2018**, 1984-1992.

24. R.Piercy, N. A. Hampson, *J. Appl. Electrochem*. **1975**, 5, 1-15.

25. F. J. Lundevall, V. Elumalai, A. Drageset, C. Totland, H.-R. Bjørsvik, *Eur. J. Chem.* **2018**, 3416-3425.

26. C. N. Neumann, J. M. Hooker, T. Ritter, *Nature.* **2016,** *534*, 369-373.

27. T. Ishiyama, M. Murata, N. Miyaura, *J. Org. Chem*. **1995**, *60*, 7508-7510.

28. N. Fattahi, M. Ayubi, A. Ramazani, *Tetrahedron*. **2018**, *74*, 4351-4356.:

29. M. M. Heravi, S. Khaghaninejad, N. Nazari, Adv. Heterocycl. Chem. **2014**. *112*. 183-234.

30. J. J. Li, *Bischler-Napieralski reaction*. In: *Name Reactions*, Springer, Heidelberg, 2006, pp 57-58.

31. A. H. Sandtorv, H.-R. Bjørsvik, *Adv. Synth. Catal.* **2013**, *355*, 499-507

32. J. H. Pang, A. Kaga, S. Roediger, M.H Lin, S. Chiba, *Asian J. Org. Chem.* **2019**, *8*, 1058-1060.

33. G. R. Fulmer, A. J. Miller, N. H. Sherden, H. E. Gottlieb, A. Nudelman, B. M. Stoltz, J. E. Bercaw, K. I. Goldberg, *J. Org. Chem.* **1997**, *62*, 7512-7515.

34. V. Elumalai, H.-R. Bjørsvik, *Tetrahedron Lett.* **2016**. *57*. 1224-1226

35. A. L. Pumphrey, H. Dong, T. G. Driver, *Angew. Chem. Int. Ed.* **2012**, *51*, 5920-5923.

36. J. S. Wright, H. Shadnia, J. M. Anderson, T. Durst, M. Asim, M. El-Salfiti, C. Choueiri, M. A. C. Pratt, S. C. Ruddy, R. Lau, K. E. Carlson, J. A. Katzenellenbogen, P. J. O'Brien, L. Wan, *J. Med. Chem.* **2010,** *54*, 433-448.

37. D. L. Boger, K. C. Cassidy, S. Nakahara, *J. Am. Chem. Soc.* **1993**, *115*, 10733-10741.

38. J. K. Laha, P. Petrou, G. D. Cuny, *J. Org. Chem.* **2009**, *74*, 3152-3155.

39. N. Jana, F. Zhou, T. G. Driver, *J. Am. Chem. Soc.* **2015**, *137*, 6738-6741.

40. D. J. Kahl, K. M. Hutchings, E. M. Lisbeth, A. J. Haak, J. R. Leipprandt, T. Dexheimer, D. Khanna, P.- S. Tsou, P. L. Campbell, D. A. Fox, B. Wen, D. Sun, M. Bailie, R. R. Neubig, S. D. Larsen, *J. Med. Chem*. **2019**, *62*, 4350-4369.

41. M.-J. Lai, H.-Y. Lee, H.-Y. Chuang, L.-H. Chang, A.-C. Tsai, M.-C. Chen, H.-L. Huang, Y.-W. Wu, C.-M. Teng, S.-L. Pan, Y.-M. Liu, S. Mehndiratta, J.-P. Liou, *J. Med. Chem.* **2015**, *58*, 6549-6558.

42. T. Ishiyama, M. Murata, N. Miyaura, *J. Org. Chem.* **1995**, *60*, 7508-7510.

43. F. Vogt, M. Witschel, T. Seiser, C. Lopez, T. Seitz, G. Kraemer, T. Newton, W. Trevor, D. Schachtschabel, K. Kreuz, *Preparation of bipyridines and herbicidal compositions containing them forweed control.*(Carl-Bosch-Strasse), Patent PCT/EP2017/068636, **2018**

44. K. C. Nicolaou, H. Ding, J.-A. Richard, D. Y.-K. Chen *J. Am. Chem. Soc*. **2010**, *132*, 3815–3818

8. Appendix

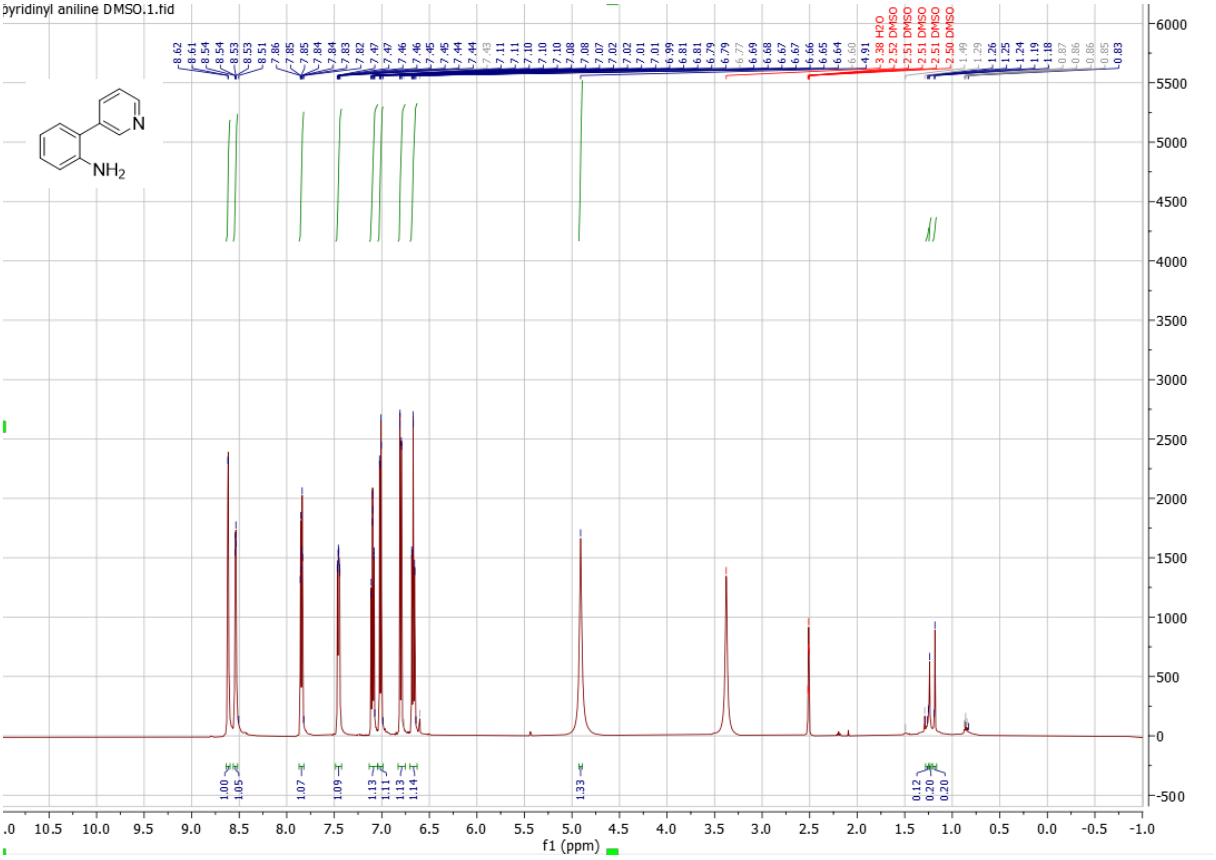


Figure 33 ^{1}H-NMR of compound 6

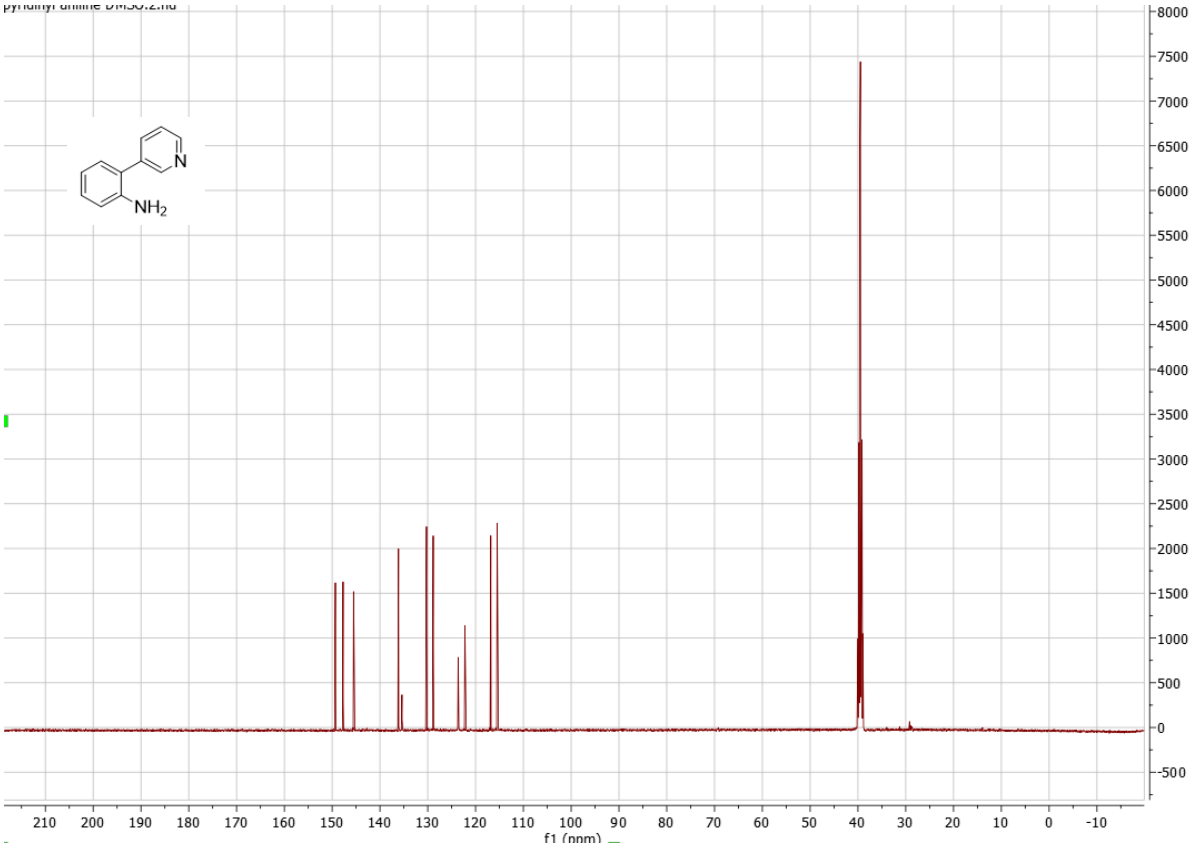


Figure 34 ^{13}C-NMR spectra of compound 6

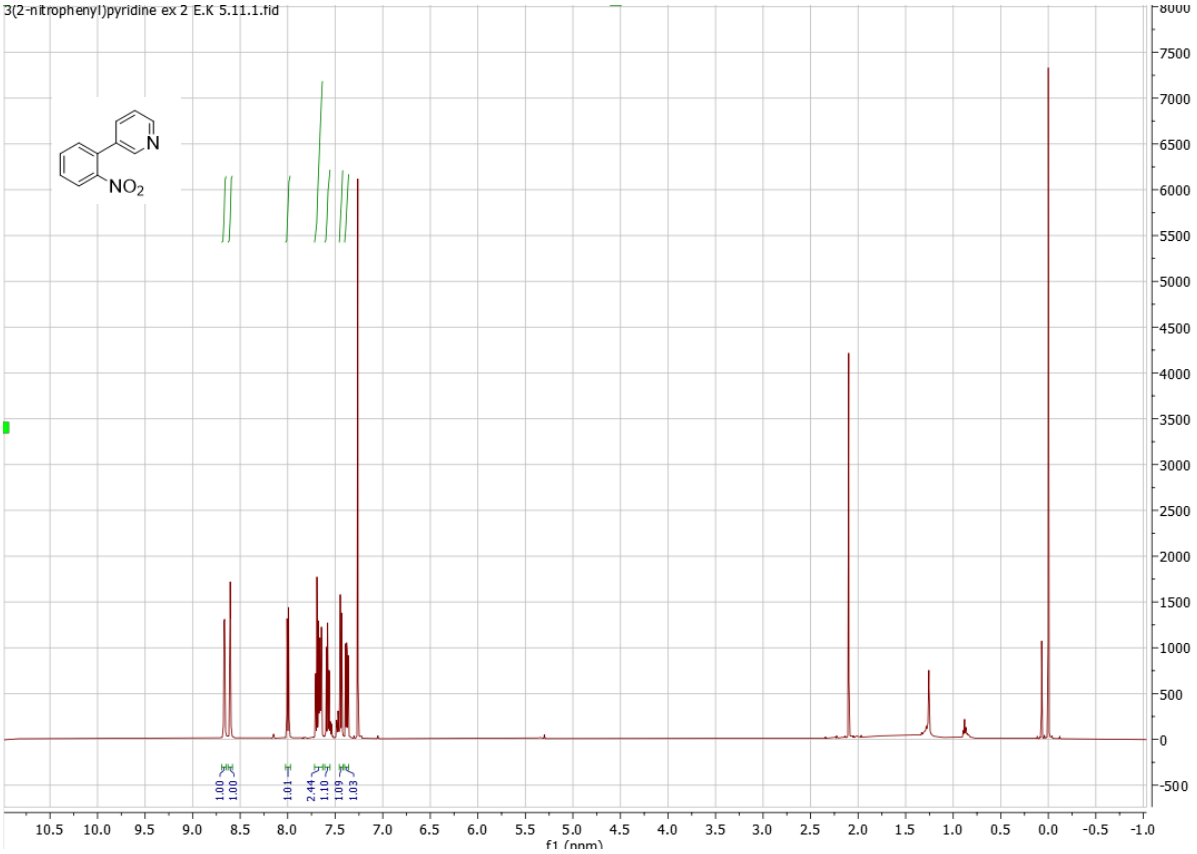


Figure 35 ^{1}H-NMR spectra of compound 8

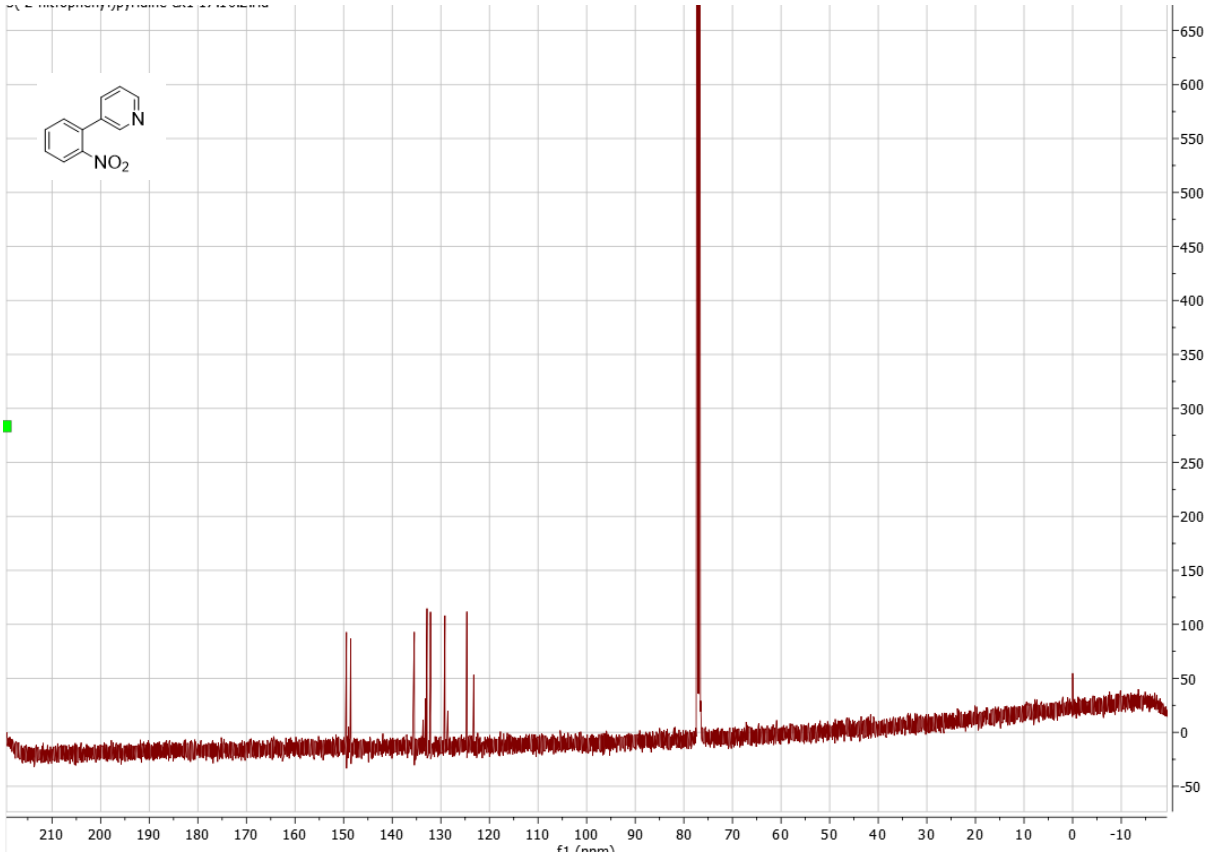


Figure 36 ^{13}C-NMR spectra of compound 8

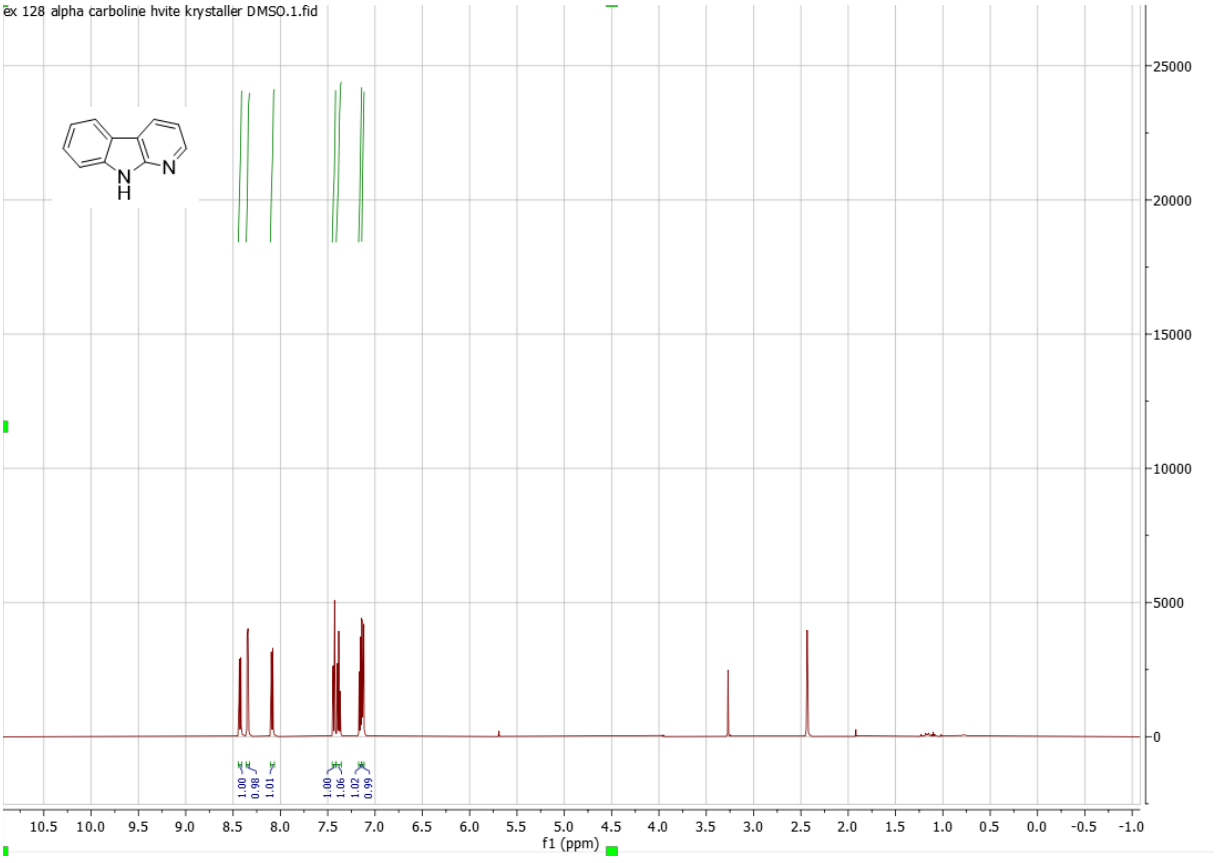


Figure 37 ^{1}H-NMR spectra of compound 1

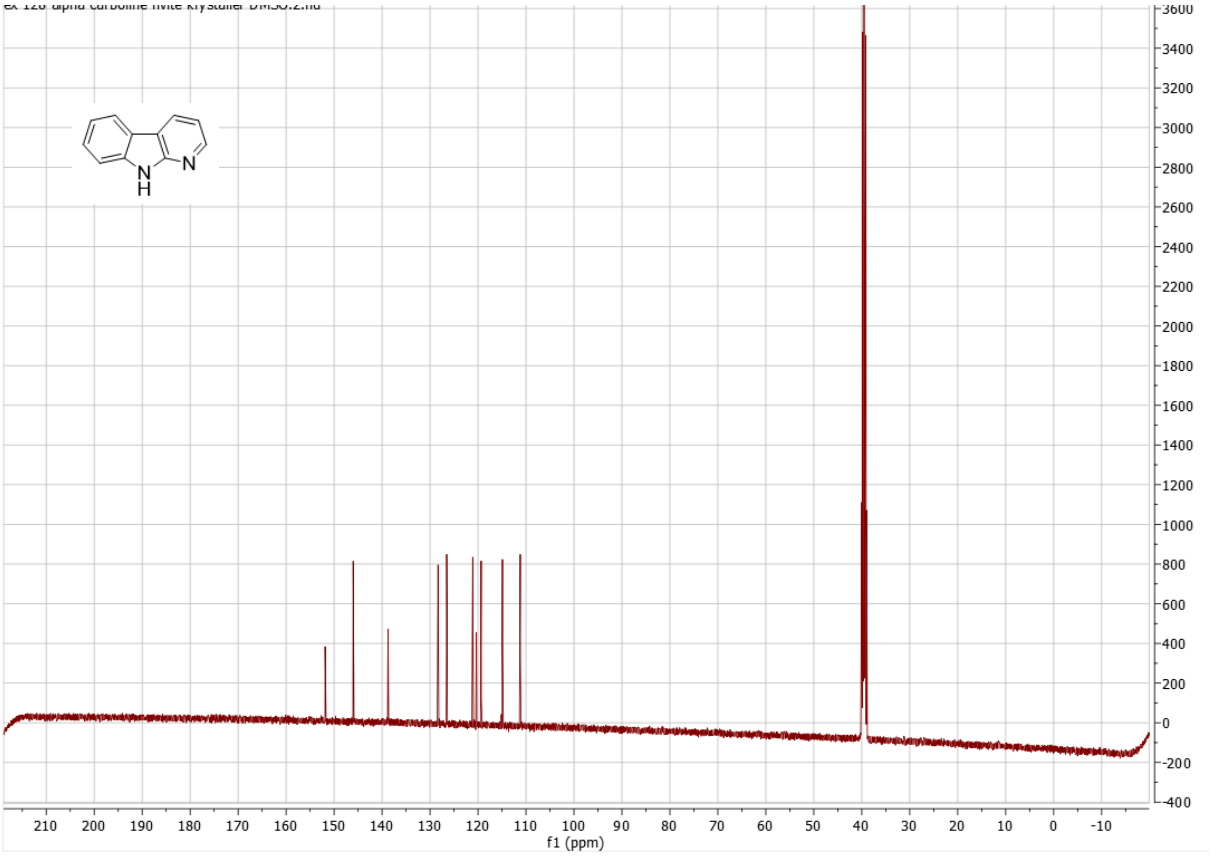


Figure 38 ^{13}C-NMR spectra of compound 1

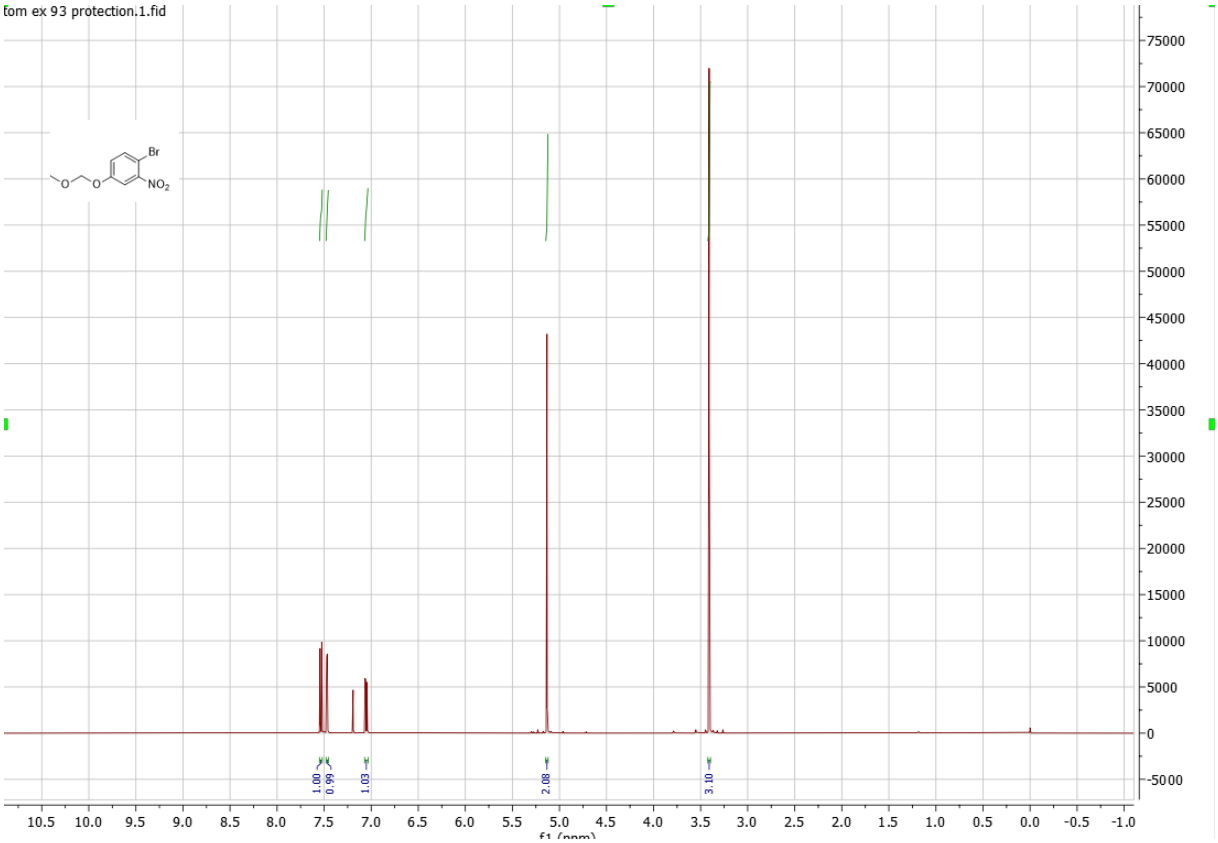


Figure 39 ^{1}H-NMR spectra of compound 13

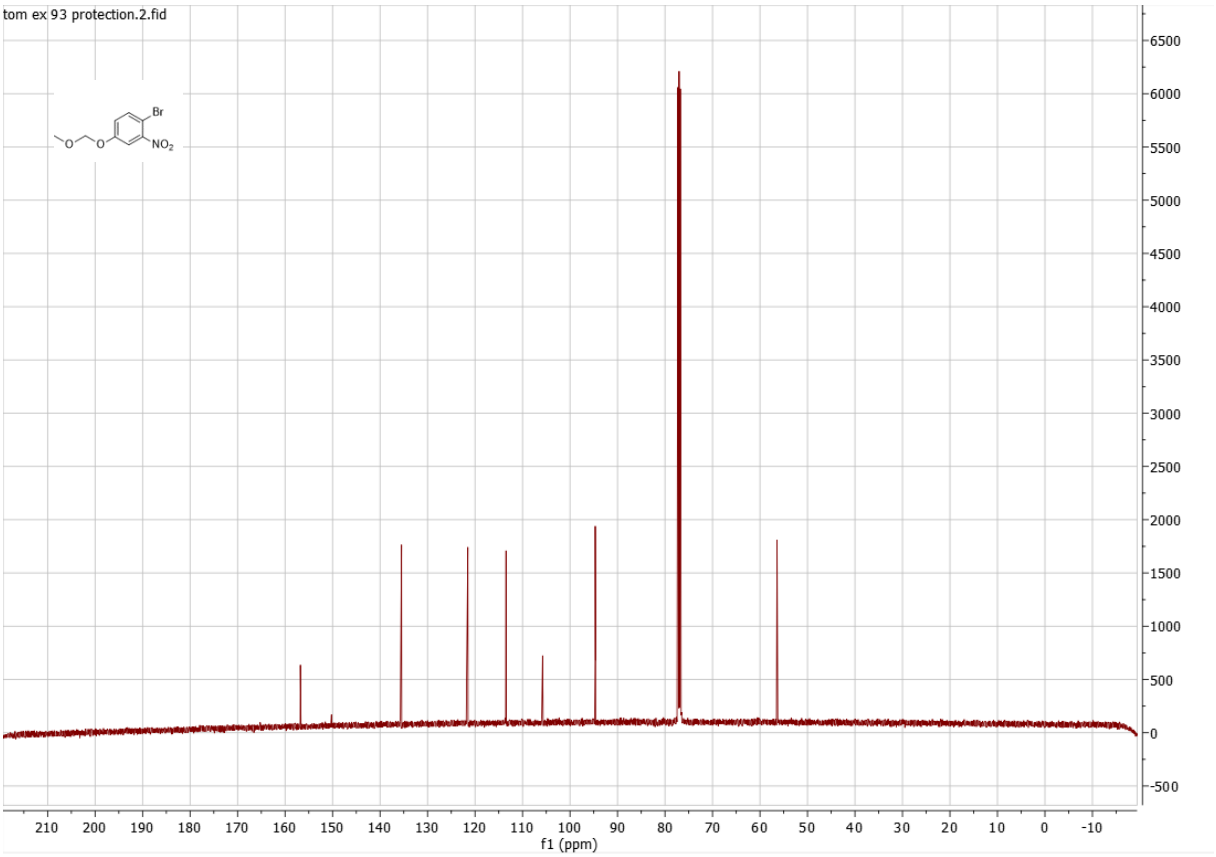


Figure 40 ^{13}C-NMR spectra of compound 13

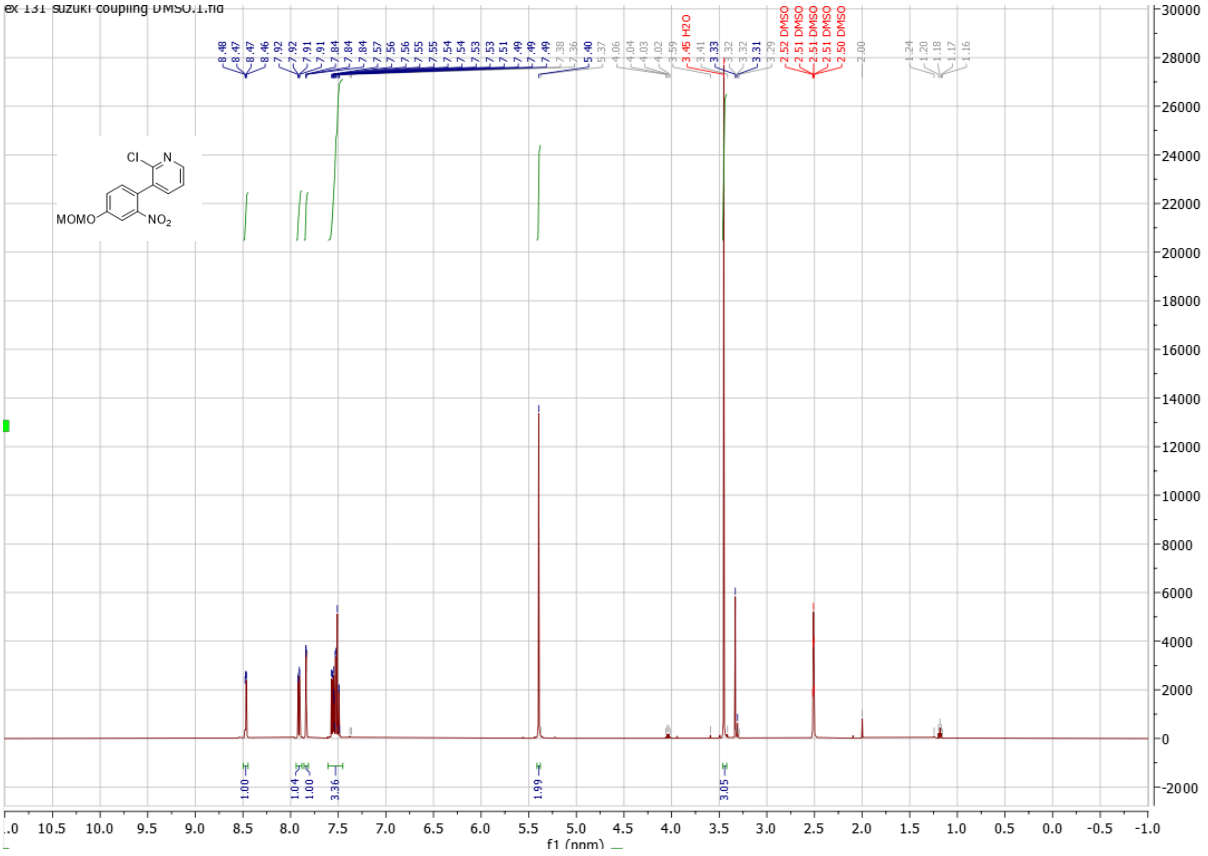


Figure 41 [1]H-NMR spectra of compound 14

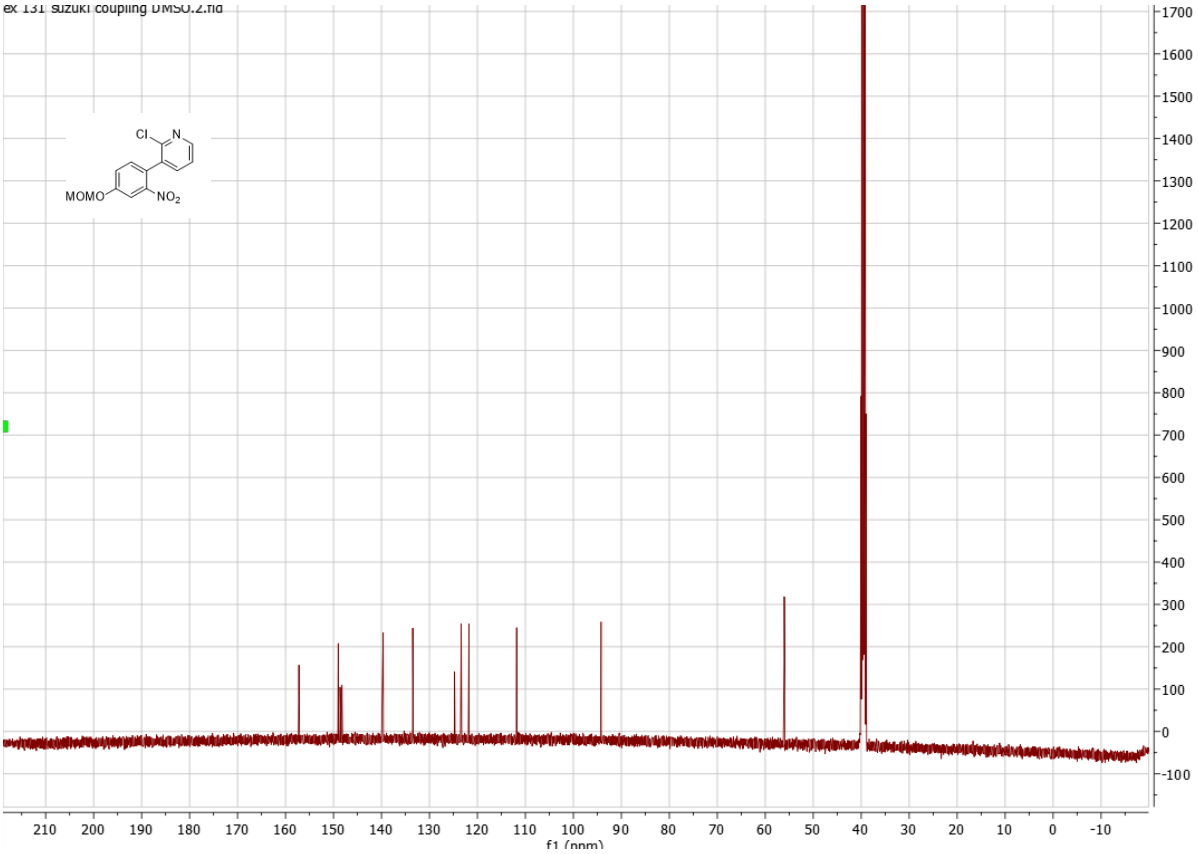


Figure 42 [1]H-NMR spectra of compound 14

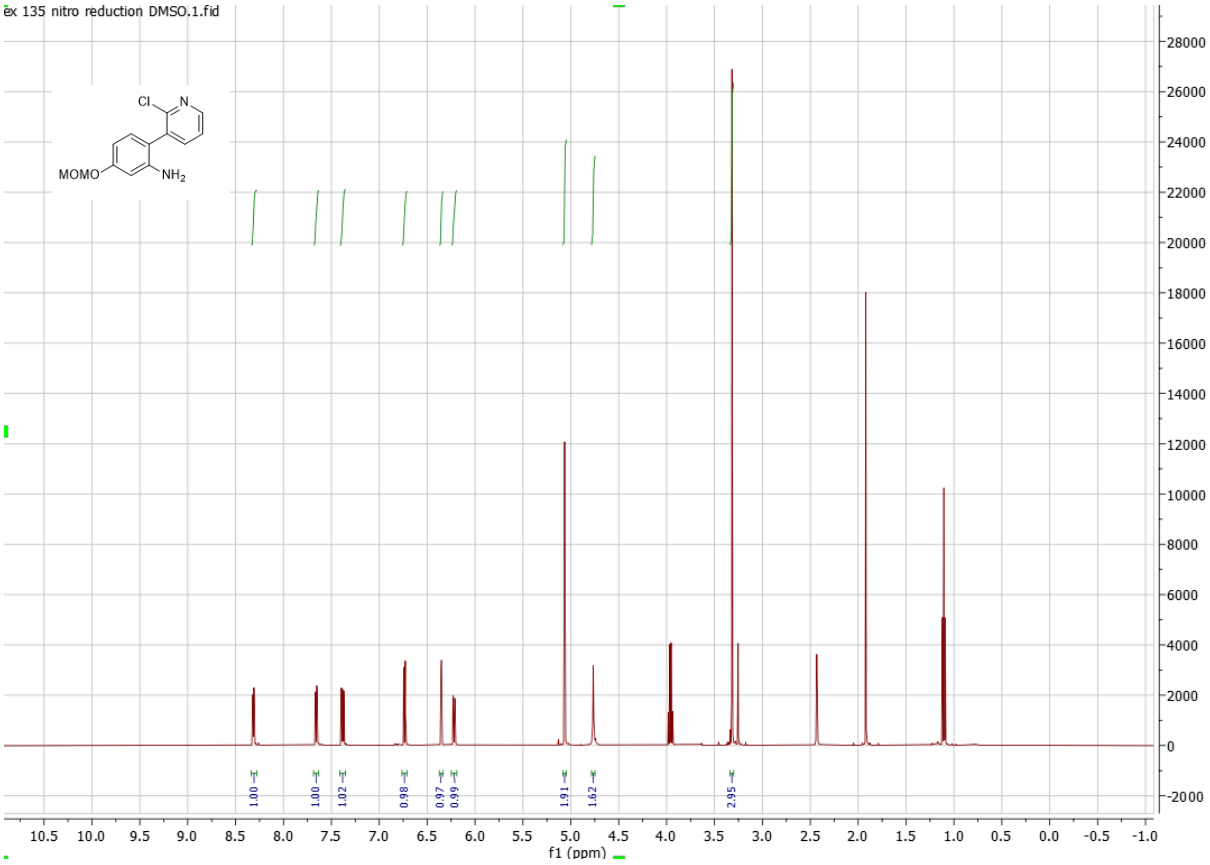


Figure 43 ^{1}H-NMR spectra of compound 15

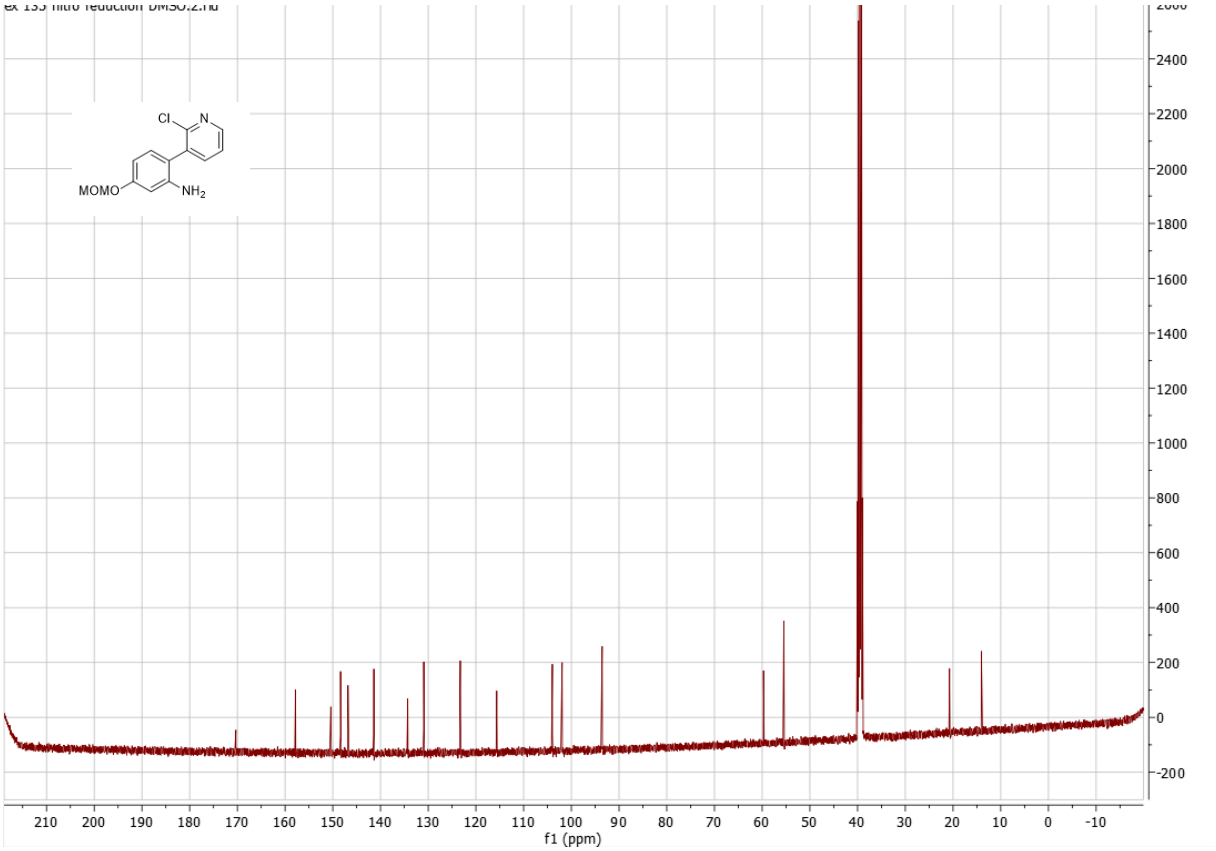


Figure 44 ^{13}C-NMR spectra of compound 15

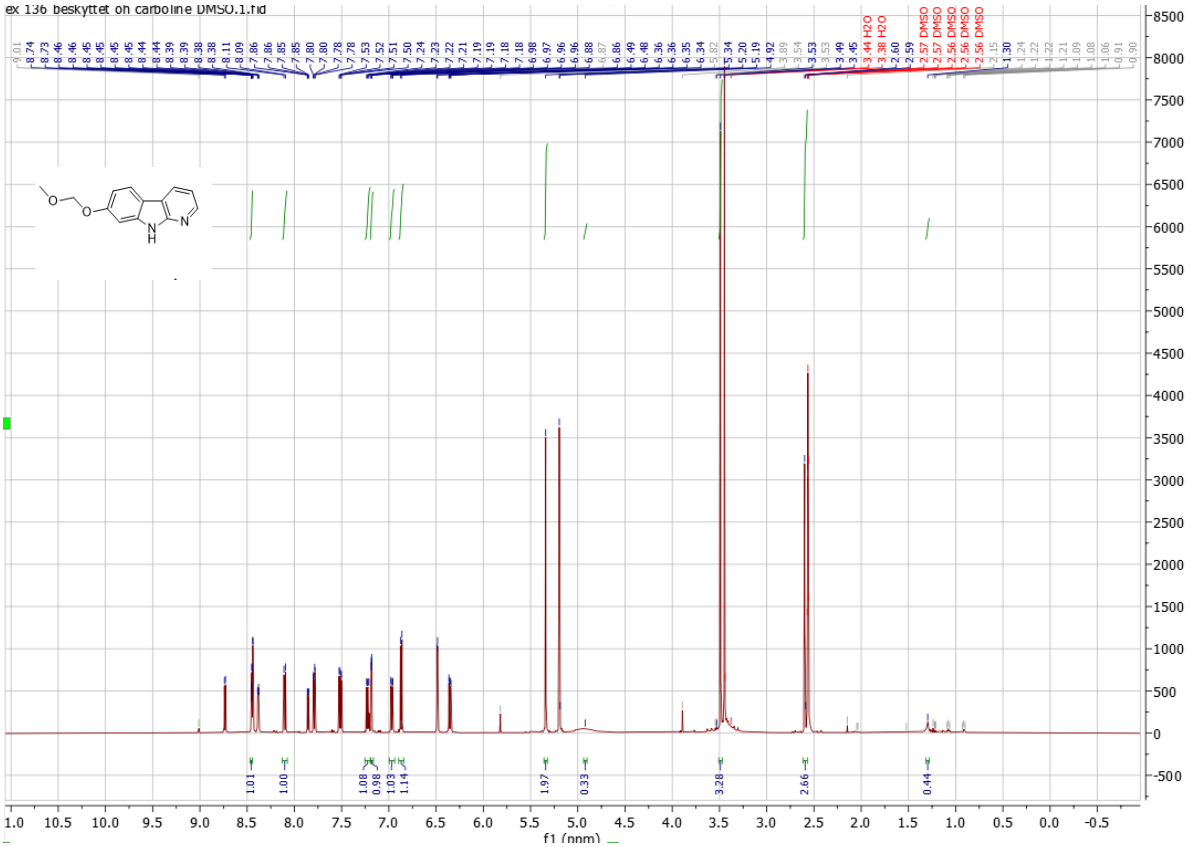


Figure 45 ^{1}H-NMR spectra of compound 16

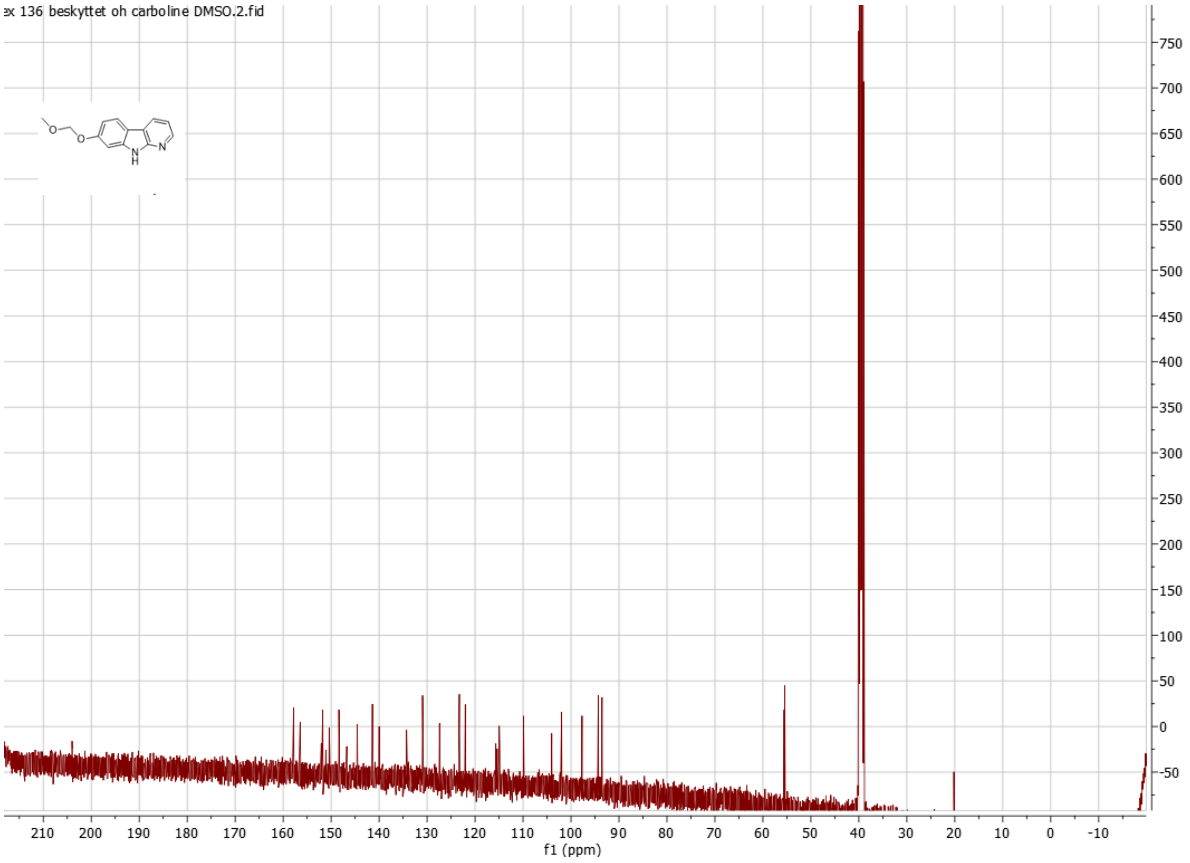


Figure 46 ^{13}C-NMR spectra of compound 16

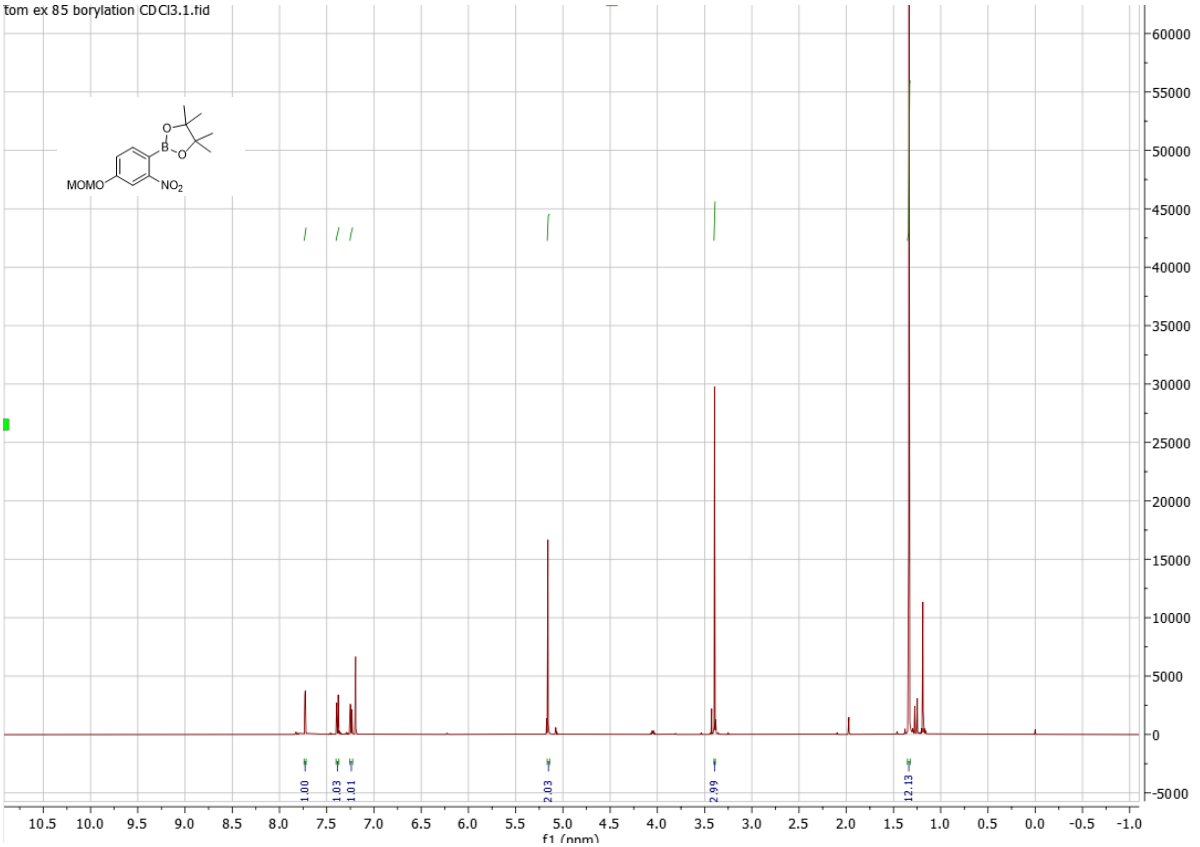


Figure 47 ^{1}H-NMR spectra of compound 19

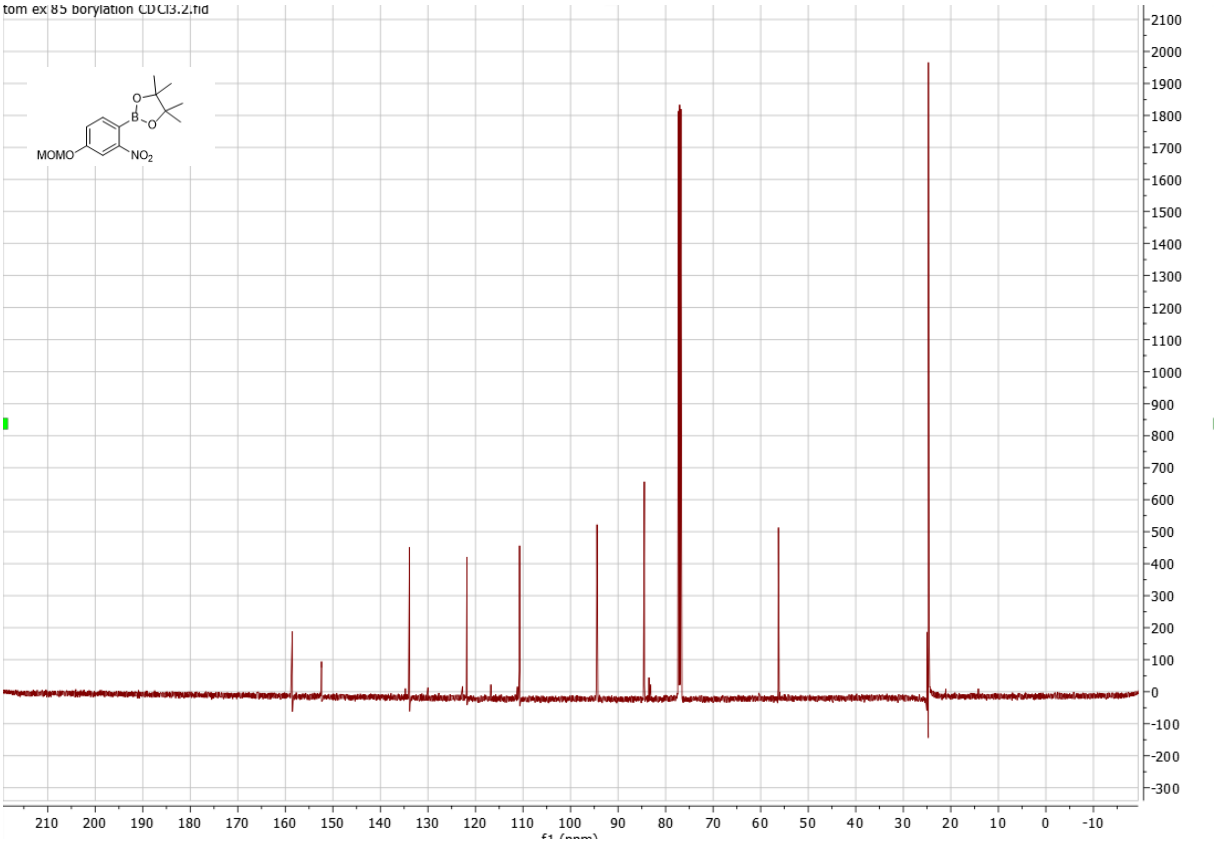


Figure 48 C-NMR spectra of compound 19

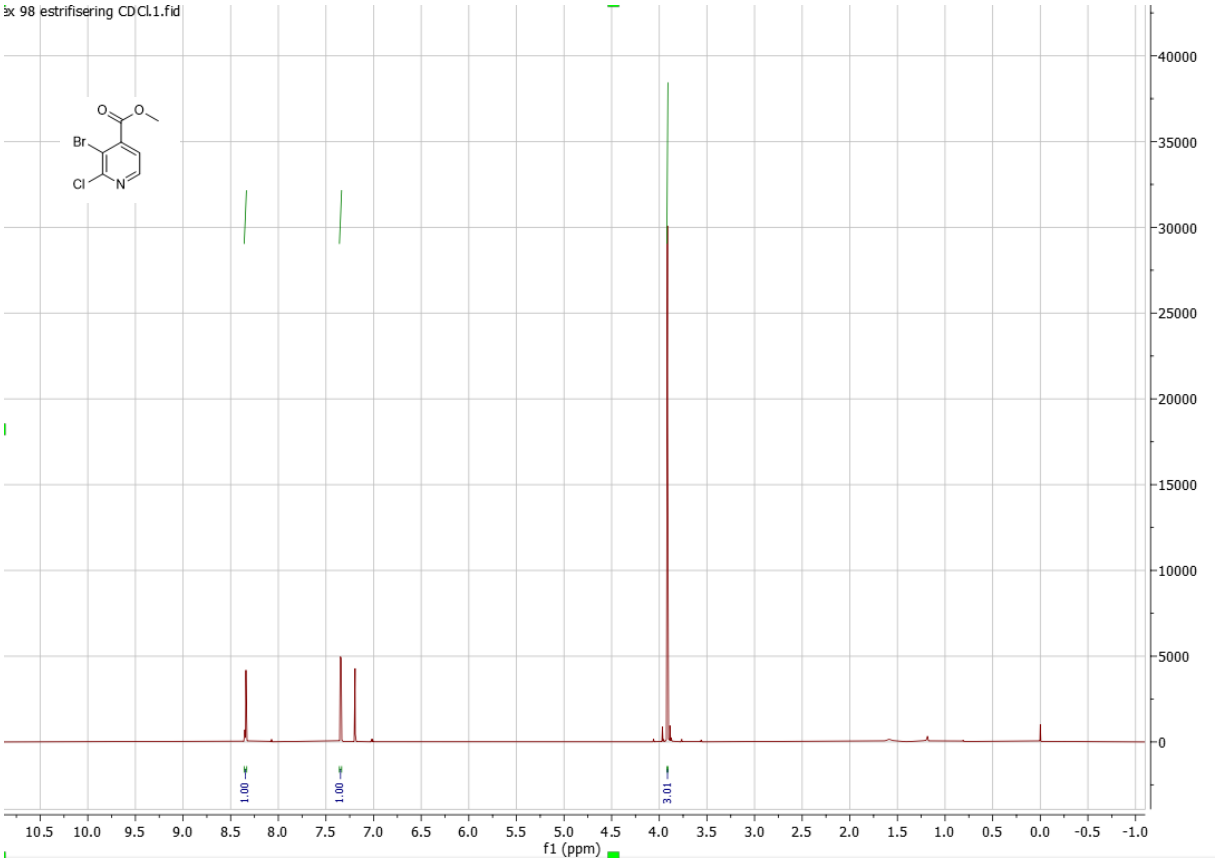


Figure 49 ^{1}H-NMR spectra of compound 21

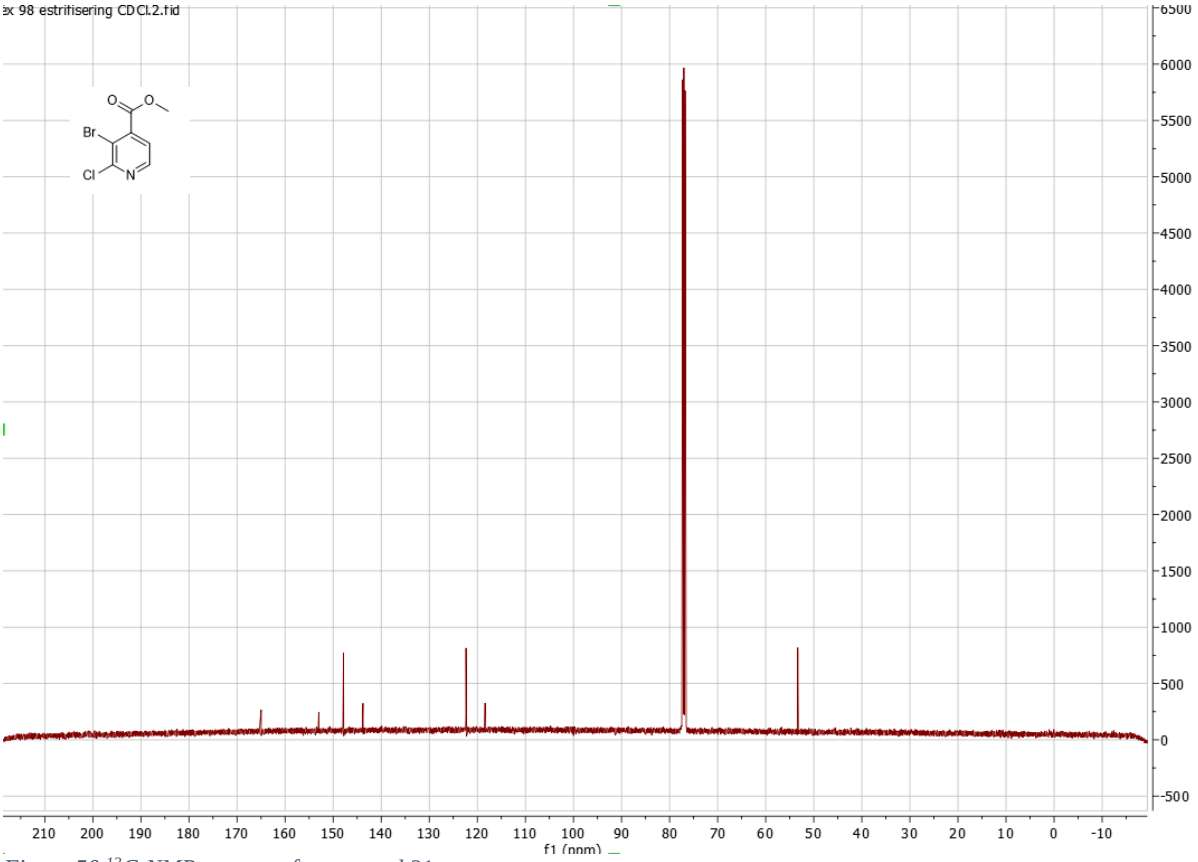


Figure 50 ^{13}C-NMR spectra of compound 21

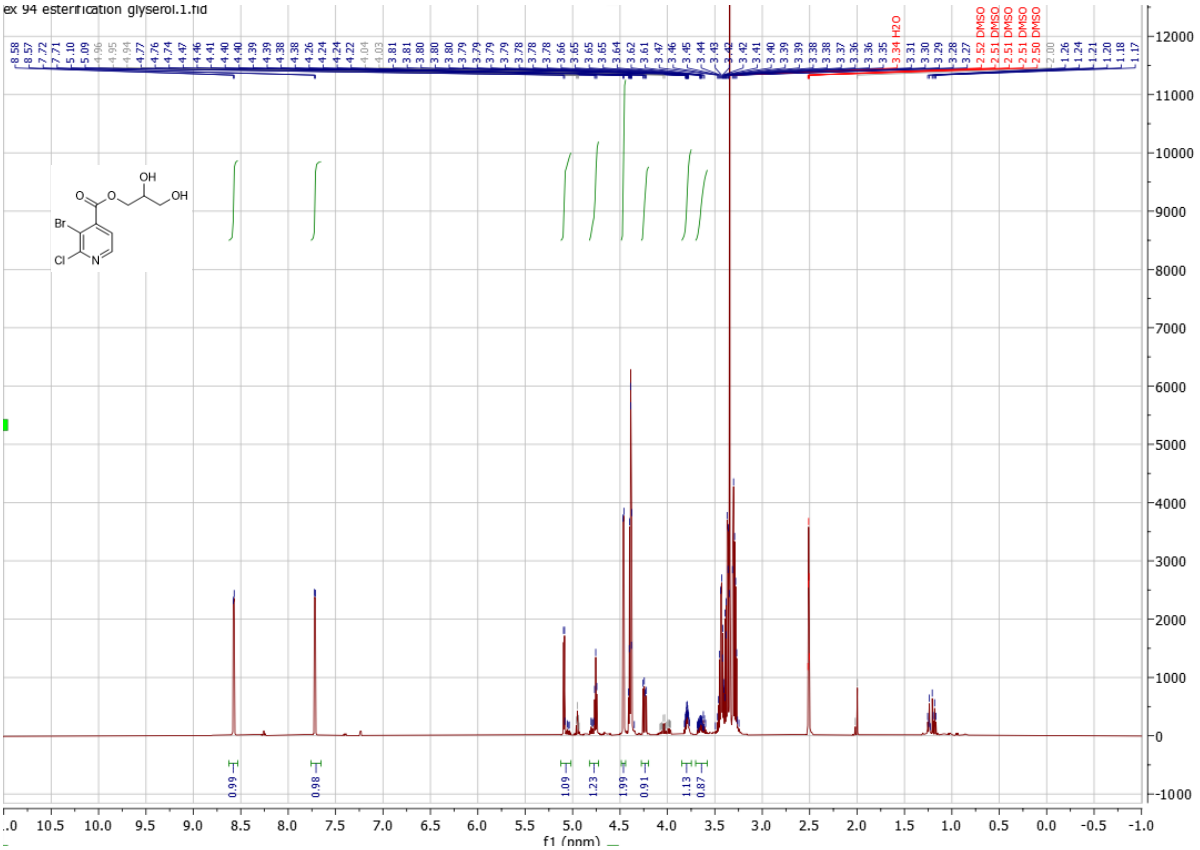


Figure 51 [1]H-NMR spectra of compound 23

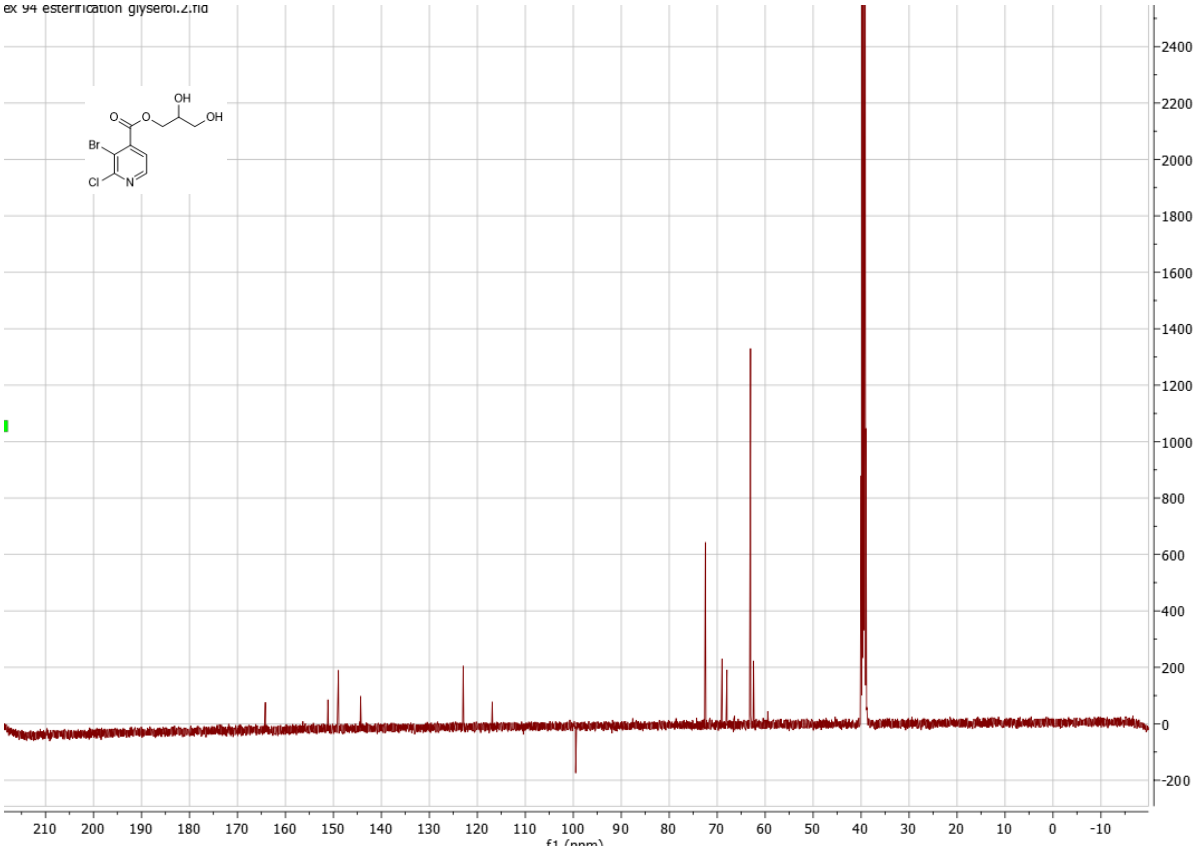


Figure 52 ^{13}C-NMR spectra of compound 23

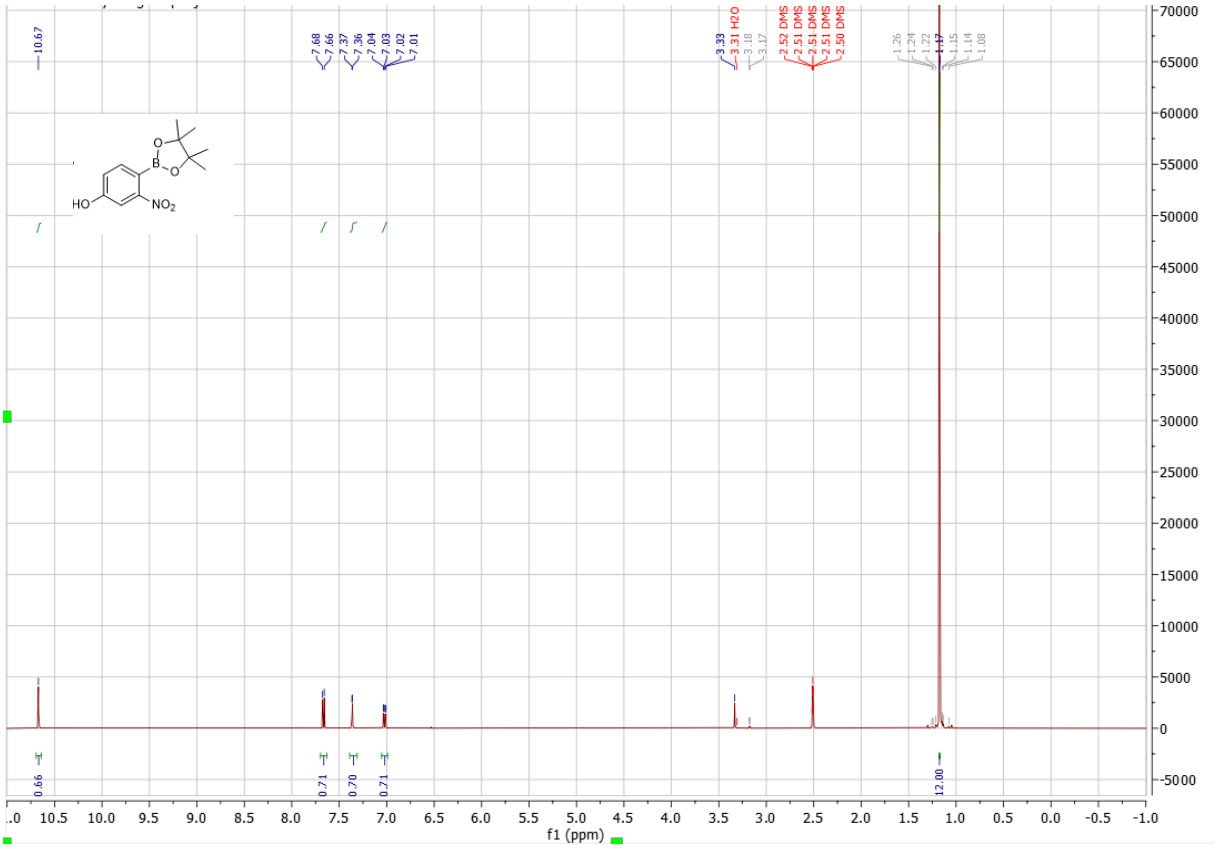


Figure 53 [1]H-NMR spectra of compound 30

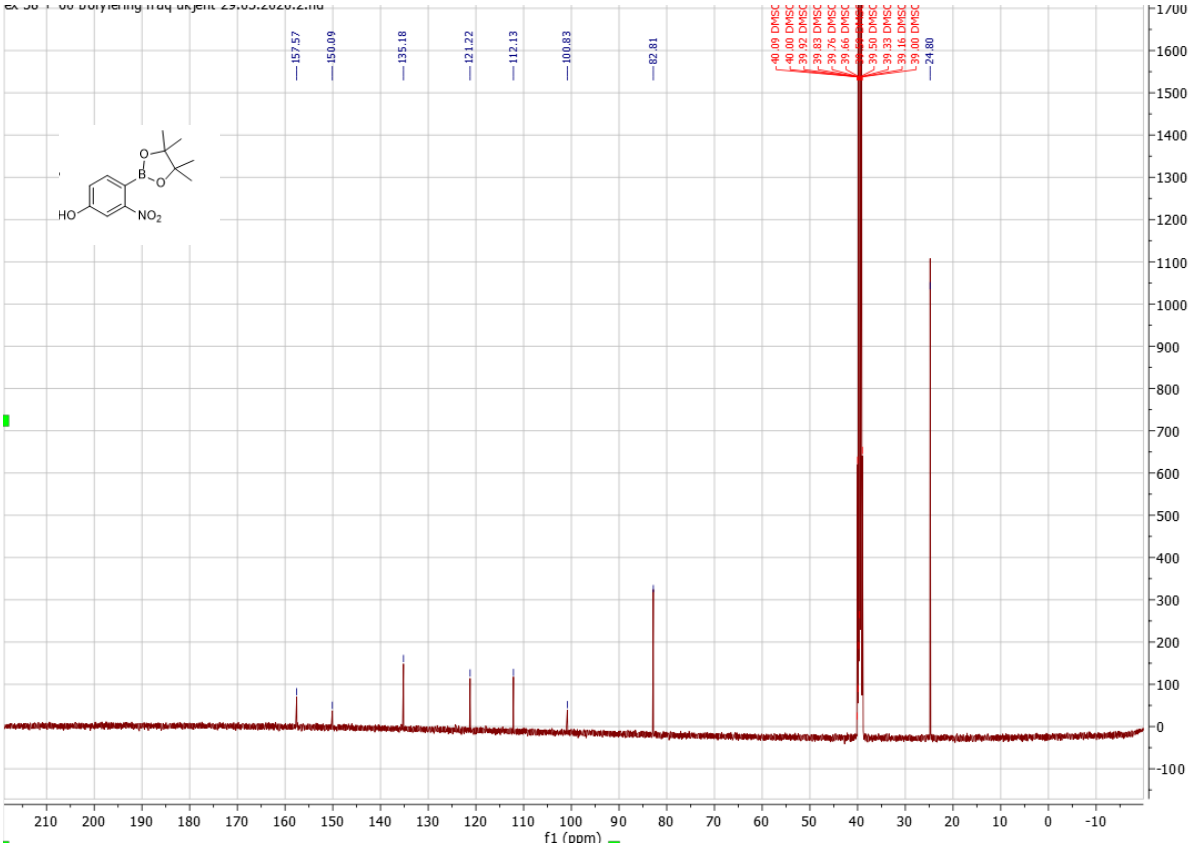


Figure 54 ^{13}C-NMR spectra of compound 30

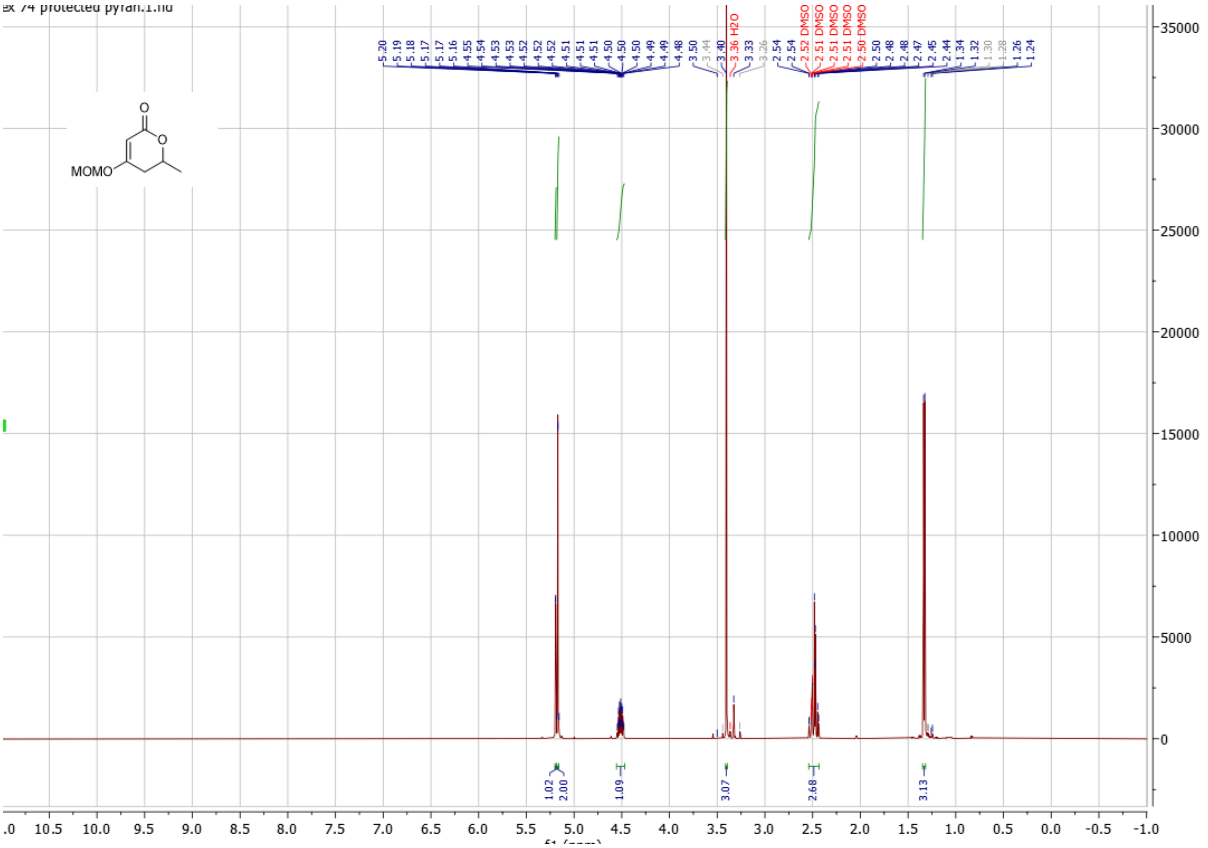


Figure 55 ^{1}H-NMR spectra of compound 27a

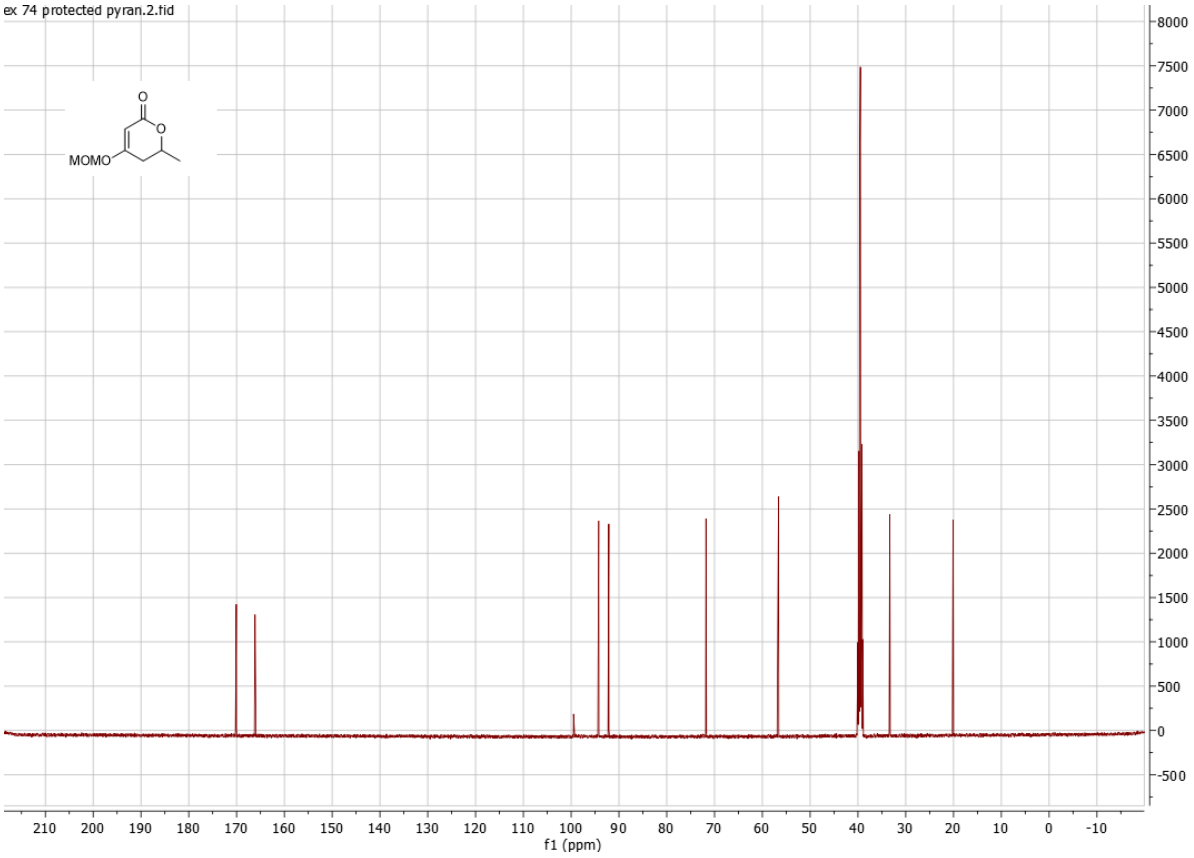


Figure 56 ^{13}C-NMR spectra of compound 27a

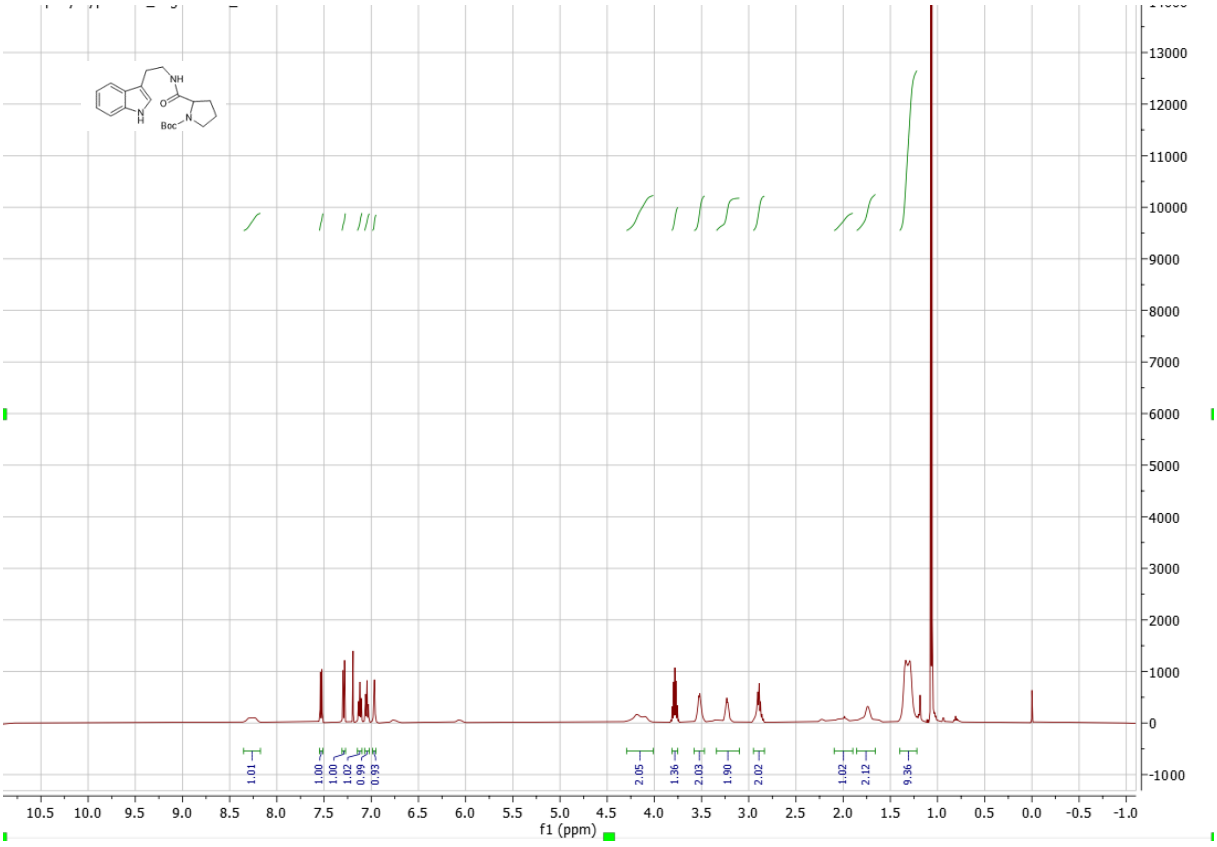


Figure 57 ^{1}H-NMR spectra of compound 35

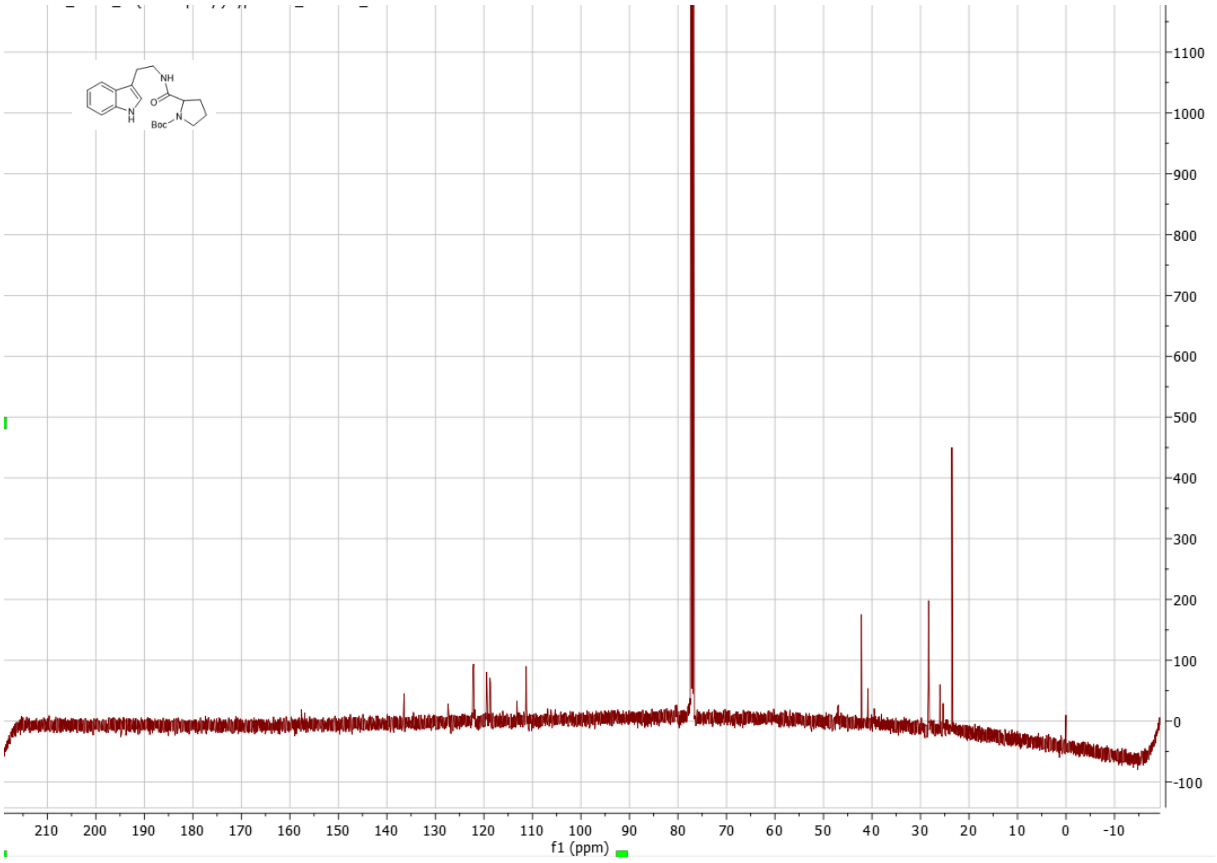


Figure 58 ^{13}C-NMR spectra of compound 35

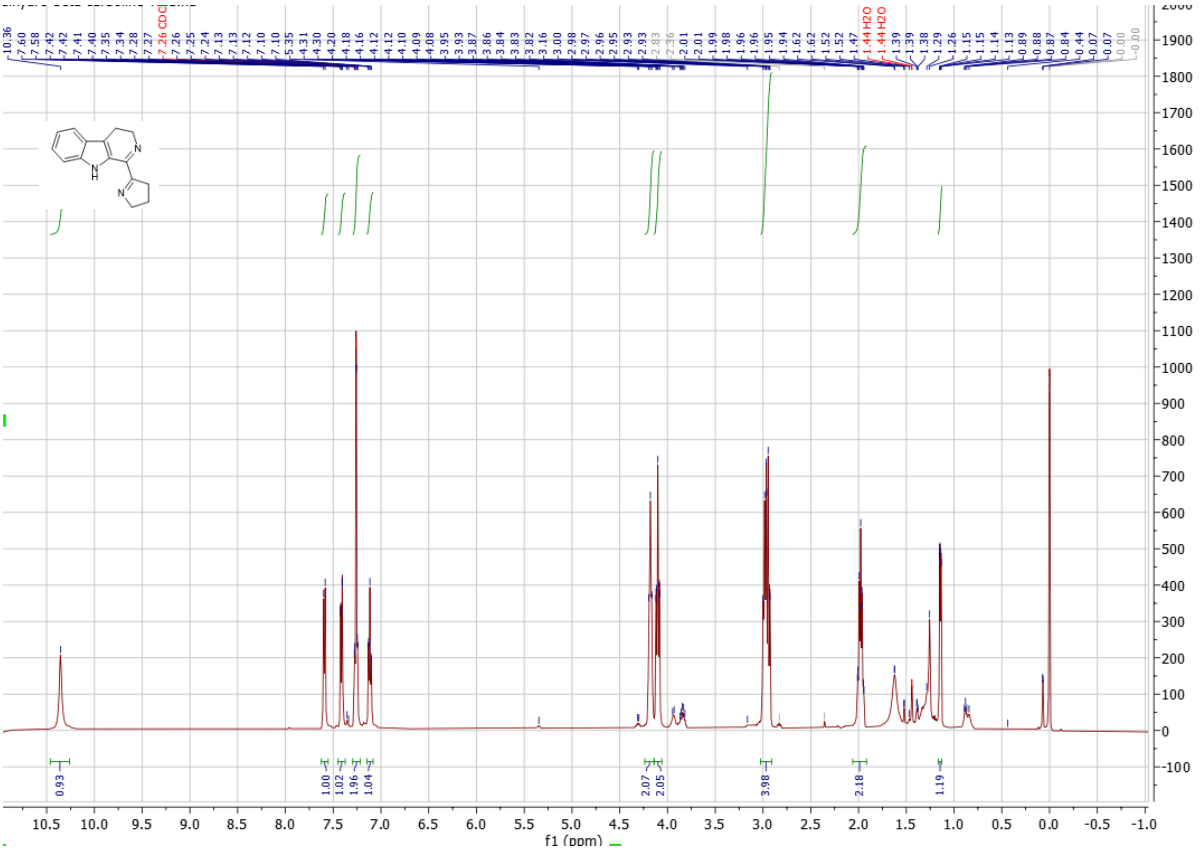


Figure 59 [1]H-NMR spectra of compound 38

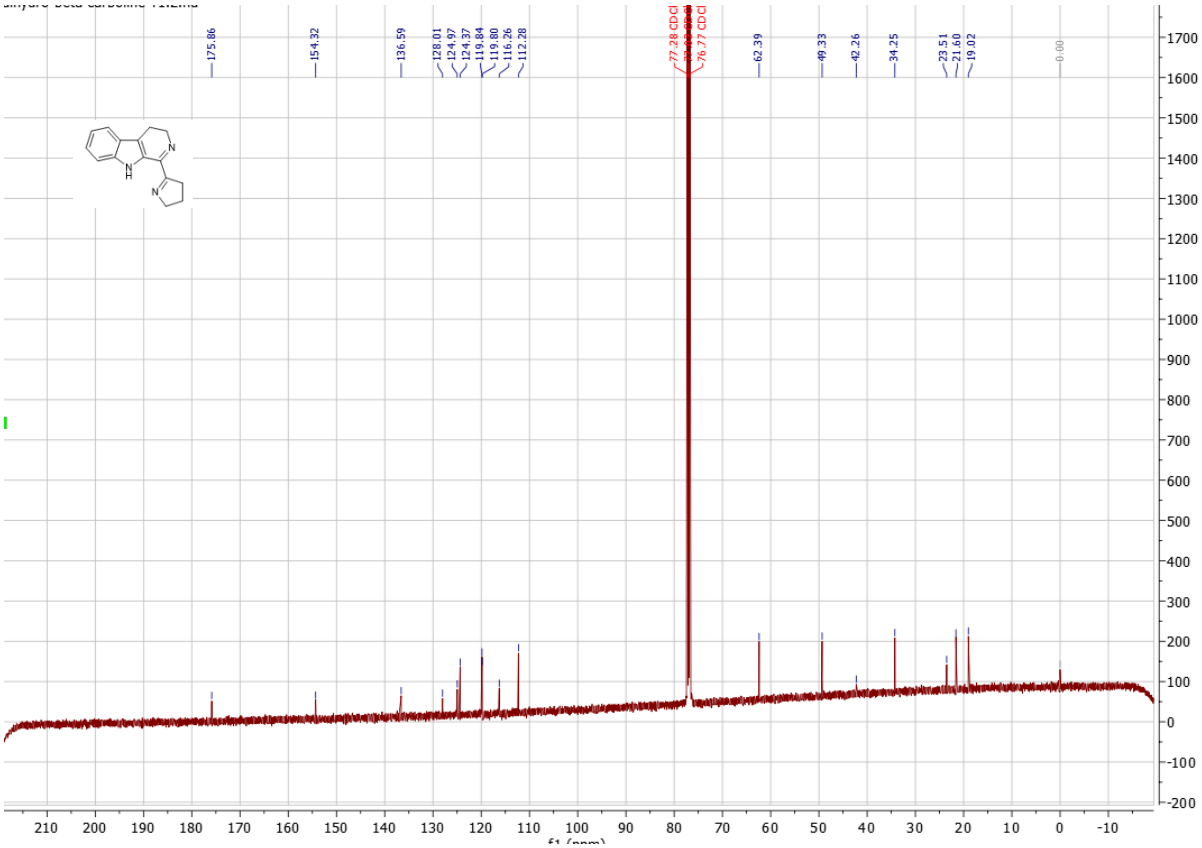


Figure 60 ^{13}C-NMR spectra of compound 38

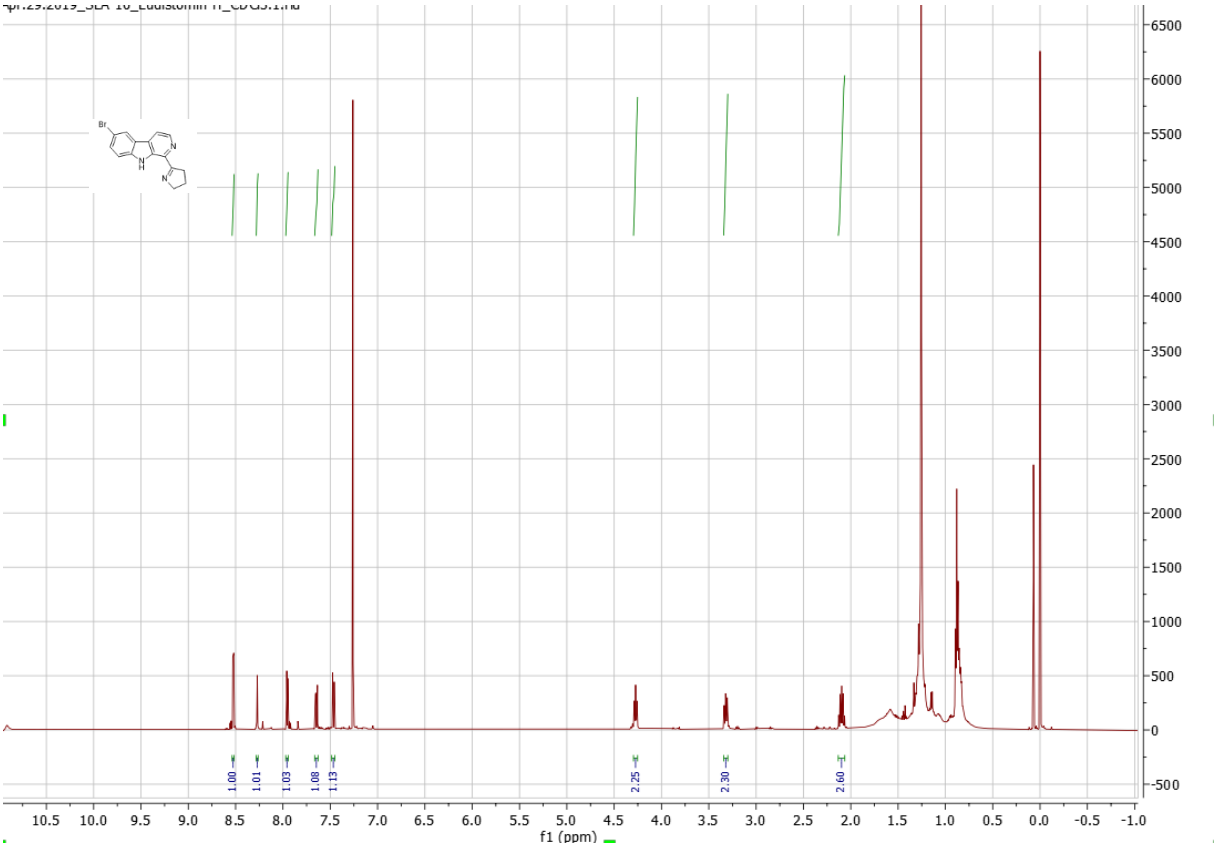


Figure 61 ^{1}H-NMR spectra of compound 39

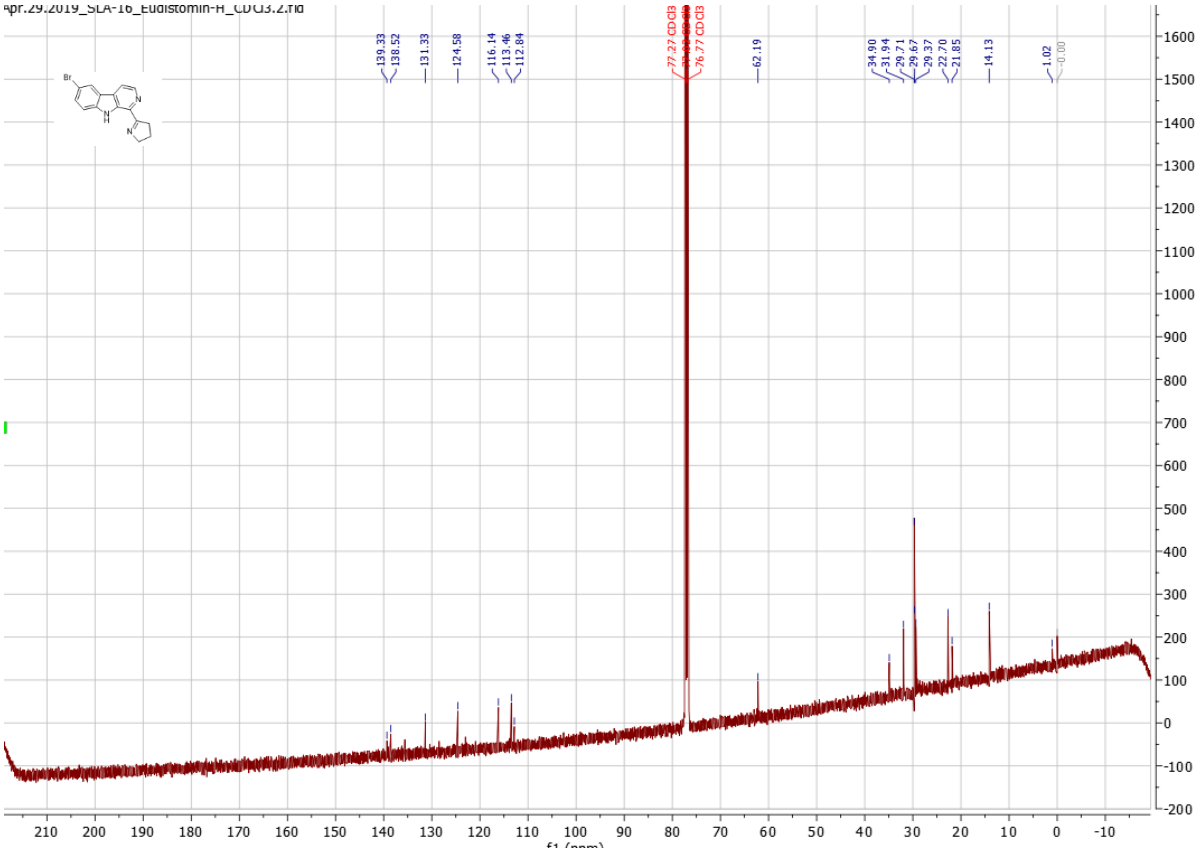


Figure 62 ^{13}C-NMR spectra of compound 39