The Completed Total Synthesis of Louisianin C and Studies Toward the Total Synthesis of Azacridone A

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Scholar of the College Thesis
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Table of Contents

Preface

Total Synthesis is a field of Organic Chemistry that focuses on the construction of various compounds. These compounds can be known, such is the case in the synthesis of natural products like Lactonamycin, or they can be creations of the imagination, like the Molecular Motor. In both cases, this particular concentration of science involves a keen use of intellect as well as a substantial amount of creativity.

Many wonder whether or not synthesis is a worthwhile investment of time and money. The most often used answer to this question is that synthesis produces helpful compounds like aspirin or antibiotics such as erythromycin. Other good target molecules include scarce natural products for research purposes. A good synthesis makes them inexpensive and easy to create industrially resulting in less expensive drugs and vitamins in the market for the consumer. But in many, if not most cases the synthetic target proves to not be useful to the human body, generally due to toxicity or its presence as a carcinogen.

Aside from these compounds that end up being harmful, many compounds that seem to have little value to humanity to begin with are synthesized anyway and are also heavily funded. It is these compounds that receive a lot of criticism from both scientists and non-scientists, and it is these compounds that are often times the most important to total synthesis.

It is true that total synthesis when studied on its own has little value to humanity. There is little gained in synthesizing a natural product using chemistry that has been known for years. The goal of total synthesis should not be to simply create the natural

product. It should also be to develop new methodology along the way. It is through the development of new reactions that Chemistry progresses.

This is not to say that methodology should be approached first, and then a multi-step reaction sequence involving the new chemistry should be attempted. This is not progress. This is simply a proof of what has already been shown to occur. This approach to Chemistry holds its head only slightly above those Chemists that consitently synthesize molecules without having discovered anything new. There is a time and a place for a 'methodology then synthesis' approach. That place is Catalysis, a different speciality than total synthesis, one that deals with the improvement of synthetic method, not the discovery.

Through careful design and synthesis of molecules, we as scientists learn the mechanics of chemistry. We can learn how molecules interact with one another, and how different types of bonding are going to affect the chemistry of different reactions. Using the results of published reactions and syntheses scientists can use the previously performed chemistry in application to their own molecules. With the recent rise in publications being placed on the Internet it has become easier to stay up-to-date with current chemistry. This in turn fosters an accelerated growth in the field and allows us to move forward into more complex reactions and target molecules at a faster pace.

With this progress chemists and biochemists hope to research and produce molecules that will have biological activity and will be applicable to the human body through shorter and more selective synthetic routes. This growth in the field also provides chemists with the knowledge to go back to older syntheses and make them shorter and more cost efficient. With the current political and public interest in

pharmaceuticals and healthcare costs, making syntheses shorter means lower costs to the consumers. Thus establishing a reasoning behind the high investment in total synthesis.

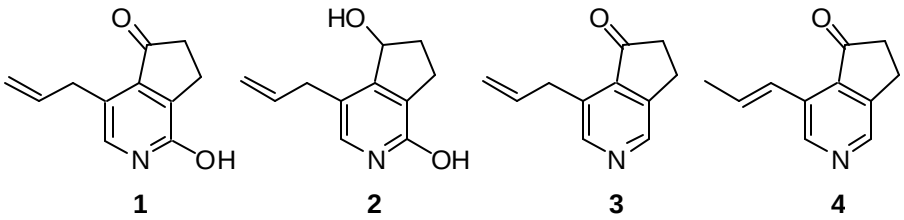
Behind the advanced science of total synthesis lies a certain amount of creativity and knowledge that when applied correctly benefits everyone both in the science and in the community.

It is with this in mind that I have opted to perform research in the field of natural product synthesis, and I propose this research for designation of Scholar of the College under the direction of Vanderslice Professor of Chemistry T. Ross Kelly.

Louisianin C: Introduction and Total Synthesis

The louisianins are a family of four alkaloids isolated from a cultured broth of *Streptomyces* sp. WK-4028, which was isolated from a soil sample collected in Louisiana, U.S.A.[1] These molecules show anti-androgenic activity, primarily through the inhibition of the growth of testosterone-responsive SC 115 cells. (SC 115 cells are testosterone-responsive mouse mammary carcinomas.)[2] Androgens aid the growth of the prostate gland. This can be normal or abnormal cell growth. In the case of abnormal androgenic cell production, the daughter cells that are produced are those that result in prostate cancer. The louisianins have shown promise for the inhibition of androgenic cell production, and consequently could inhibit prostate cancer cell production.

The structures of louisianins A-D (**1**-**4**) as determined by Omura et al. (**Figure 1**) all have a common, centralized pyridine ring with two major constituents attached: a fused five-membered ring and an allylic side chain. Louisianins A (**1**) and B (**2**) have a hydroxyl group present on the pyridine ring while C (**3**) and D (**4**) do not.[2]



1 **2** **3** **4**

Figure 1. Louisianins A, B, C, and D.

[1] Another alkaloid, with the same carbon skeleton as the louisianins, has been isolated from a source from Uluday Turkey: Henne P.; Grabley S.; Thiericke R.; Zeeck A. *Ann*. **1997**, *5*, 937.

[2] Takamatsu, S.; Kim, Y.-P.; Hayashi, M.; Furuhata, K.; Takayanagi, H.; Komiyama, K.; Woodruff, H. B; Omura, S. *J. Antibiot.* **1995**, *48*, 1090.

Omura et al. have shown conversion of **1** to **4**, and in the process have isolated **3**. At first **1** was originally found to be the most potent inhibitor of the SC 115 proliferation, while **3** and **4** showed only slight inhibition. It was later found that **3** and **4** showed anti-angiogenic activity and potently suppressed the tube formation of vascular endothelical cells *in vitro*. While **1** was found to be produced in large amounts by a cultured fermentation broth of *Streptomyces* sp. WK-4028, **3** and **4** were found in only very small quantities.[3]

The discovery of the anti-angiogenic activity of louisianin C (**3**) and its scarcity in nature made the small alkaloid a good target for synthesis by the Kelly Lab. Aside from this, conversion of **3** to **4** was shown by Omura et al. to be possible in relatively high yield making **3** the more synthetically important target between the two choices.

Research on the synthesis of louisianin C began 4 years ago with little success. With a newly developed retrosynthesis, the compound was completed within one year (2002) and the first completed total synthesis was recently accepted for publication (**Scheme 1,** page 8).

The strategy behind the new synthetic pathway lies in the symmetry of the compounds in the first two steps and the key method for cyclization of the five-membered ring. The first step of the synthesis involved the protection of the reactive carbon (C-4) on the 3,5-dribromopyridine (**5**). The carbon in the 4 position on the ring was lithiated to give **6,** according to the method of Gu.[4] This was subsequently quenched with trimethylsilylchloride. The trimethylsilyl group (TMS), now in place on the C-4 position, acts not only as a bulky protector of the carbon *para* to the nitrogen, but it also

[3] Sunazuka, T.; Zhi-Ming, T.; Harigaya, Y.; Takamatsu, S.; Hayashi, M.; Komiyama, K.; Omura, S. *J. Antibiot.* **1997**, *50*, 274.
[4] Gu, Y. G.; Bayburt, E. K. *Tetrahedron Lett.* **1996**, *37*, 2565.

acts as a leaving group later in the synthesis. With this site on the ring protected and the future leaving group in place, synthesis proceeded with a Stille coupling between two allylic groups and the C-3 and C-5 positions of the pyridine nucleus to yield compound **8** in 96 % yield.[5]

The next step involved the selective hydroxylation of only one of the allylic groups, thus destroying the symmetry of the compound. This regioselective anti-Markonikov hydroxylation was performed using 9-borabicyclo[3.3.1]nonane (9-BBN) and hydrogen peroxide. Boron trifluoride diethyl etherate was used to protect the nitrogen atom in the pyridine ring, preventing the ring nitrogen from interacting with the one equivalent of 9-BBN adduct that followed.[6] (Only one equivalent of 9-BBN was used to avoid the possibility of dihydroxylation.) *N*,*N*,*N'*,*N'*-tetramethylethylenediamine (TMEDA) was then added to quench the boron trifluoride diethyl etherate, removing it from the pyridine ring. A slight white precipitate formed in the solution as evidence of this reaction. This left only the 9-BBN to react with the hydrogen peroxide which yielded alcohol **9** in 43% yield with 45% recovery of starting material. The yield for this step based on unrecovered starting material was 77%.

The next step of the reaction involved the use of *o*-iodoxybenzoic acid (IBX). IBX is the precursor to the Dess-Martin periodinane reagent which oxidizes alcohols to aldehydes and ketones. It was recently found that IBX performs the same function with similarly mild reaction conditions, and is much easier to make than Dess-Martin reagent. It is worth noting that, in many cases, an *N*-oxide can be formed when using IBX in the

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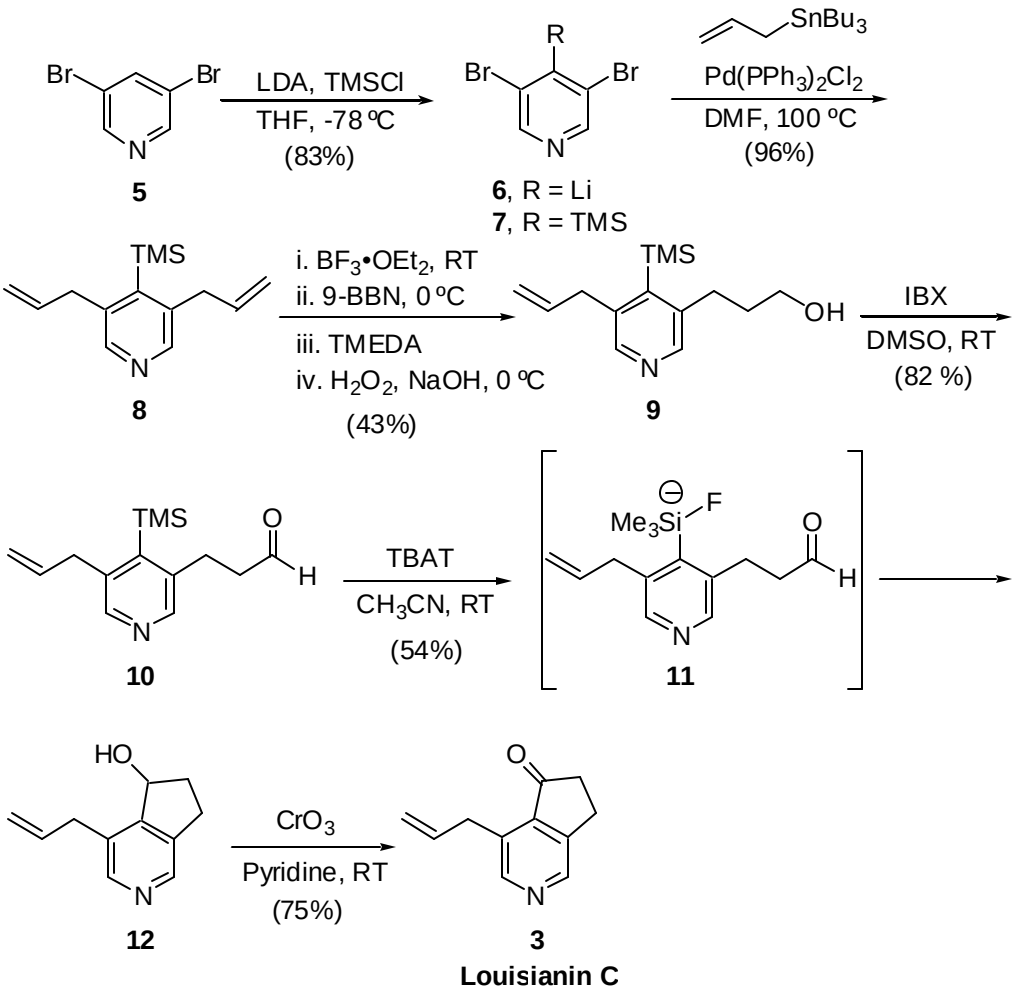
[5] The same coupling conditions were used as for the preparation of compound **14** in: Tilley, J. W.; Clader, J. W.; Zawoiski, S.; Wirkus, M.; LeMahieu, R. A.; O'Donnell, M.; Crowley, H.; Welton, A. F. *J. Med. Chem.* **1989**, *32*, 1814.

[6] Brown, H. C.; Vara-Prasad, J. V. N.; Zee, H.-S. *J. Org. Chem.* **1986**, *51*, 439.

presence of a nucleuphilic nitrogen atom. Finney and More have used an Fmoc protecting group to prevent such from happening.[7] It was decided that with a primary alcohol as the target, the competing oxidation of the aromatic, pyridine nitrogen would not be a significant enough problem to add another step for protection. In the end, the result was an 82% yield of **10** from **9** with no evidence of a side reaction TLC.[8]



Scheme 1. The synthesis of louisianin C (**3**).

[7] More, J.D.; Finney, N.S. *Org. Lett.* **2002**, *4*, 3001.
[8] Frigerio, M.; Santagostino, M. *Tetrahedron Lett.* **1994**, *35*, 8019.

Aldehyde **10** is the precursor to the key cyclization of the fused five-membered ring. Tetrabutylammonium fluoride (TBAF) was the first reagent used for the attempted removal of the TMS group, however it was largely unsuccessful. Tetrabutylammonium triphenylsilyldifluoride (TBAT) was then tried as a fluoride source.[9] The benefits of using TBAT instead TBAF were that it was anhydrous and did not contain a naked (and basic) fluoride ion.[10] The reaction is presumed to have passed through a species similar to **11**.[11] It was here that the TMS protecting group acted as a leaving group, that lead to the cyclization of the aldehyde to yield the five-membered ring in compound **12** in 54% yield.

The final step of the synthesis was from the literature of Omura et al. IBX was originally attempted, but required heating and the use of multiple equivalents. This may have been a cause of the ability of the pyridine nitrogen to compete for oxidation with an inhibited secondary alcohol such as the one present in **12**.[8]

In any case, the results were inferior to that of the chromium trioxide oxidation published by Omura et al.[3] Louisianin C (**3**) was isolated in an overall yield of 11% in 6 steps. The spectrometric data for our compound matches that published by Omura et al. This is the first synthesis of any of the louisianins and it confirms the structure proposed by Omura et al of louisianin C (**3**).[2]

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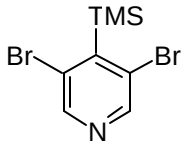
[9] Pilcher, A. S.; DeShong, P. *J. Org. Chem.* **1996**, *61*, 6901.
[10] Pilcher, A. S.; Ammon, H. L.; DeShong, P. *J. Am. Chem. Soc.* **1995**, *117*, 5166.
[11] Chuit, C; Corriu, R. J. P; Reye, C.; Young, J. C. *Chem. Rev*. **1993**, *93*, 1371.

Louisianin C: Experimental Data[12]

Preparation of 3,5-Dibromo-4-(trimethylsilyl)pyridine (**7**)**:**



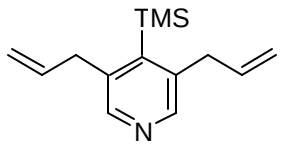
7

A solution of 3,5-dibromopyridine (**5**) (2.00 g, 8.44 mmol) in dry tetrahydrofuran (67 mL) was added by cannula over 30 min to a stirring solution of lithium diisopropylamide [prepared by adding at –78 ºC 2.5 M *n*-butyllithium in hexanes (3.4 mL, 8.6 mmol, 1.0 equiv) to diisopropylamine (1.21 mL, 8.61 mmol, 1.02 equiv) and stirring for 5 min] in THF (50 mL) under nitrogen at –78 °C. After 5 min, trimethylsilyl chloride (1.50 mL, 11.8 mmol, 1.40 equiv) was added in one portion. The reaction was monitored by TLC (silica, 49:1, hexanes:ethyl acetate) and, after a period of 25 min, the solution was quenched with a satd aqueous solution of ammonium chloride (10 mL) and was allowed to reach room temperature.[4] The resulting mixture was extracted twice with diethyl ether. The combined organic extracts were washed once with water and once with brine. The organic phase was dried over anhyd magnesium sulfate, filtered, and concentrated in vacuo to afford the crude product as yellow crystals (2.40 g). Flash column chromatography on silica gel (5 x 18 cm) using 49:1 hexanes:ethyl acetate as eluent gave the purified product (2.17 g, 83% yield): white flakes, mp 55-56 ºC; IR (KBr) 2985, 2949, 2897, 1476, 1245, 1164 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.56 (s, 2H), 0.57 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 150.3, 149.6, 128.1, 2.9; HRMS (ESI) calcd

[12] For general procedures see: Kelly, T. R.; Lebedev, R. L. *J. Org. Chem.* **2002**, *67*, 2197.

$C_8H_{12}Br_2NSi$ [M+H]: 307.9106; found 307.9092; Anal. calcd for $C_8H_{11}Br_2NSi$: C, 31.09; H, 3.59; N, 4.53; found C, 30.97; H, 3.31; N, 4.36.

Preparation of 3,5-Diallyl-4-(trimethylsilyl)pyridine (8):

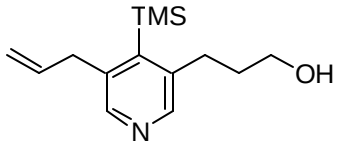


8

A solution of 3,5-dibromo-4-(trimethylsilyl)pyridine (**7**) (8.22 g, 26.6 mmol) and allyltributyltin (17.6 mL, 79.8 mmol, 3.00 equiv) in dimethylformamide (132 mL) was degassed by bubbling nitrogen through it for 30 min. Dichlorobis(triphenylphosphine)palladium(II) (1.87 g, 2.66 mmol, 10 mol %) was then added, and the reaction flask was heated at 100 ºC.[5] When the reaction was complete [ca. 20 h, TLC monitoring (silica, 20:1, hexanes:ethyl acetate)] the vessel was cooled to room temperature and the mixture was filtered through a pad (4 cm diameter, 4 cm height) of activated basic alumina (Brockmann I) on a fine porosity sintered glass funnel to remove the palladium and tin residues. The basic alumina was washed with 400 mL of hexanes. The combined filtrate and washes were extracted three times with 1 M ammonium hydroxide to further remove tin residues, and the organic layer was washed 3 times with water, once with brine, dried over anhyd $MgSO_4$ and concentrated in vacuo. The pale yellow oil was purified by flash column chromatography on silica gel (6 x 22 cm) using a 9:1 hexanes:ethyl acetate as eluent to yield a slightly yellow oil (6.15 g, 96% yield): pale yellow oil, IR (neat, NaCl) 3081, 3006, 2979, 2954, 2901, 1638, 1409, 1253 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 8.21 (s, 2H), 6.04-5.90 (m, 2H), 5.11 (dd, 2H, J = 10.2, 1.8

Hz), 4.86 (dd, 2H, J = 17.1, 1.8 Hz), 3.54-3.51 (m, 4H), 0.40 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.8, 146.4, 139.2, 137.6, 116.2, 37.5, 3.0; HRMS (ESI) calcd $C_{14}H_{22}NSi$ [M+H]: 232.1522; found 232.1524; Anal. calcd for $C_{14}H_{21}NSi$: C, 72.66; H, 9.15; N, 6.05; found C, 72.76; H, 8.85; N, 5.89.

Preparation of 3-Allyl-5-(3-hydroxypropyl)-4-(trimethylsilyl)pyridine (9):

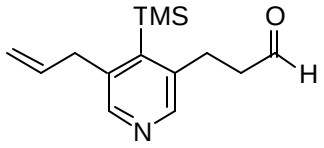


9

Boron trifluoride diethyl etherate (1.42 mL, 11.2 mmol, 1.00 equiv) was added dropwise over 5 min to a stirred solution of 3,5-diallyl-4-(trimethylsilyl)pyridine (**8**) (2.60 g, 11.2 mmol) in THF (112 mL) under nitrogen at 0 ºC.[6] A 0.50 M solution of 9-borabicyclo[3.3.1]nonane in THF (27 mL, 14 mmol, 1.2 equiv) was added dropwise, over 15 min, at room temperature. The mixture was left to stir overnight (15 h) to ensure a completed reaction. The colorless solution was treated with *N*,*N*,*N'*,*N'*-tetramethylethylenediamine (0.85 mL, 5.6 mmol, 0.5 equiv), and after stirring for ~1 min a white precipitate formed. The reaction vessel was cooled to 0 ºC, and a 2.5 M sodium hydroxide:35% hydrogen peroxide (1:1, v/v, 60.5 mL) was added in one portion. The excess peroxide was quenched by addition of aliquots of a satd aqueous solution of sodium sulfite (60 mL) until peroxide indicator strips (EM Quant, Fisher cat no: 10011-1) were negative. The aqueous mixture was then extracted three times with 20 mL portions of dichloromethane, and the combined organic extracts were washed with water and brine, dried over anhyd magnesium sulfate, filtered, and concentrated in vacuo to yield a

yellow oil (2.66 g). The oil was purified by flash column chromatography on silica gel (4 x 18 cm) using 7:3 ethyl acetate:hexanes as eluent. Two purified compounds were isolated: the starting material 3,5-diallyl-4-(trimethylsilyl)pyridine (**8**) (1.16 g, 45% yield) and the desired product 3-allyl-5-(3-hydroxypropyl)-4-(trimethylsilyl)pyridine (**9**) (1.20 g, 43%, 77% based on unrecovered **8**): clear colorless oil, IR (neat, NaCl) 3342, 2979, 2949, 1680, 1413, 1254 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 8.25 (s, 1H), 8.18 (s, 1H), 6.02-5.91 (m, 1H), 5.10 (dd, 1H, *J* = 10.2, 1.7 Hz), 4.86 (dd, 1H, *J* = 17.1, 1.7 Hz), 3.72 (t, 2H, *J* = 6.3 Hz), 3.52-3.49 (m, 2H), 2.86-2.80 (m, 2H), 1.86-1.78 (m, 2H), 1.82 (bs, 1H), 0.42 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 148.2, 147.9, 146.1, 142.4, 139.6, 137.5, 116.4, 61.9, 37.6, 36.2, 30.1, 3.1; HRMS (ESI) calcd for $C_{14}H_{23}NOSi$ [M+H]: 250.1627; found 250.1623.

Preparation of 3-[5-Allyl-4-(trimethylsilyl)pyridin-3-yl]propanal (10):



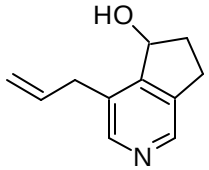
10

CAUTION! IBX is explosive under impact or upon heating to >200 ºC.[13] *o*-Iodoxybenzoic acid[14] (IBX, 1.16 g, 4.15 mmol, 1.2 equiv) was added to a stirred solution of 3-allyl-5-(3-hydroxypropyl)-4-(trimethylsilyl)pyridine (**9**) (861 mg, 3.46 mmol) in dimethylsulfoxide (8.3 mL) at room temperature, giving a homogenous solution. Reaction progress was monitored by TLC (silica, ethyl acetate); after 1 h the starting material was consumed. Water (10 mL) was then added to the solution and a white

[13] Plumb, J. B.; Harper, D. J. *Chem. Eng. News*. **1990**, *July 16*, 3.
[14] The IBX was prepared according to the following reference: Frigerio, M.; Santagostino, M.; Sputore, S. *J. Org. Chem.* **1999**, *64*, 4537.

precipitate formed which was removed by filtration through a pad (2 cm diameter, 1 cm height) of Celite® on a fine porosity sintered glass funnel. The Celite® was rinsed with dichloromethane to ensure complete recovery of product. The resulting combined biphasic filtrate and rinse was separated and the aqueous phase was extracted with two 15 mL portions of additional dichloromethane. The combined organic layers were washed twice with 10 mL portions of water (to remove any excess DMSO) and brine, and dried over anhyd magnesium sulfate. After concentration in vacuo, the oil was purified by flash column chromatography on a silica gel (2 x 17 cm) using 95:5 dichloromethane:methanol as eluent to afford 3-[5-allyl-4-(trimethylsilyl)pyridin-3-yl]propanal (**10**) (700 mg, 82% yield) as a clear colorless oil. The product was stored under nitrogen at –24 ºC: clear colorless oil, IR (neat, NaCl) 2978, 2953, 2899, 1724, 1412, 1255 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 9.84 (s, 1H), 8.24 (s, 1H), 8.22 (s, 1H), 6.02-5.92 (m, 1H), 5.11 (dd, 1H, *J* = 10.2, 1.7 Hz), 4.86 (dd, 1H, *J* = 17.1, 1.7 Hz), 3.51 (dd, 2H, *J* = 3.9, 1.7 Hz), 3.11-3.05 (m, 2H), 2.76 (t, 2H, *J* = 8.4 Hz), 0.41 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 199.9, 148.8, 147.5, 146.2, 140.5, 139.6, 137.4, 116.5, 46.6, 37.5, 25.9, 3.0; HRMS (ESI) calcd for $C_{14}H_{22}NOSi$ [M+H]: 248.1471 found 248.1472.

Preparation of 6,7-Dihydro-4-(2-propenyl)-5*H*-cyclopenta[*c*]pyridin-5-ol (12):

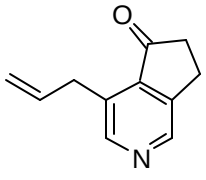


12

A solution of 3-[5-allyl-4-(trimethylsilyl)pyridin-3-yl]propanal (**10**) (172 mg, 0.696 mmol) in dry acetonitrile (11 mL) was transferred over 10 min by cannula under

nitrogen to a flame-dried flask containing tetrabutylammonium triphenylsilyldifluoride (TBAT, 756 mg, 1.39 mmol, 2.0 equiv) at room temperature.[10] The reaction was refluxed at 85 ºC, and was monitored by TLC (silica, ethyl acetate). After 1 h the starting material was consumed. The reaction was cooled, and ten mL of 1 M sulfuric acid was added, and the solution was washed two times with dichloromethane to remove impurities. The aqueous layer was then made basic (pH ≈ 10) by addition of 2.5 M sodium hydroxide. The milky aqueous layer was extracted three times with 10 mL portions of dichloromethane, the combined organic extracts were washed once each with water and brine, dried over anhyd magnesium sulfate, and concentrated in vacuo to give a yellow oil (97 mg). The oil was purified by preparative thin layer chromatography (silica, 95:5 dichloromethane:methanol) to afford **12** as a clear colorless oil (66 mg, 54% yield). The spectra are in agreement with those reported for **12**. The HCl salt of **12** was prepared by bubbling HCl gas (generated by adding conc. sulfuric acid to ammonium chloride) into a solution of **12** in ether. The ether was decanted, leaving behind a clear colorless oil, that was placed under high vacuum, which caused the HCl salt to crystallize: white needles, mp 134-135 ºC; IR (neat, NaCl) [M+H] 3300, 3039, 2972, 2912, 2692 cm^{-1}; ^{1}H NMR [M+H] (300 MHz, $CDCl_3$) δ 8.36 (s, 1H), 8.28 (s, 1H), 6.02-5.83 (m, 1H), 5.52 (dd, 1H, *J* = 6.6, 5.5 Hz), 5.29-5.23 (m, 1H), 5.17 (s, 1H), 3.78 (dd, 2H, *J* = 7.2, 5.5 Hz), 3.41-3.25 (m, 1H), 3.11-2.95 (m, 1H), 2.71-2.58 (m, 1H), 2.36-2.29 (m, 1H), 2.34 (bs, 1H); ^{13}C NMR [M+H] (100 MHz, $CDCl_3$) δ 163.1, 143.1, 138.3, 137.7, 134.4, 133.0, 119.1, 74.0, 34.8, 33.6, 28.3; HRMS (ESI) [M+H] calcd for $C_{11}H_{13}NO$: 176.1075, found: 176.1072.

Preparation of 6,7-Dihydro-4-(2-propenyl)-5*H*-cyclopenta[c]pyridin-5-one, louisianin C (3):



3

Louisianin C

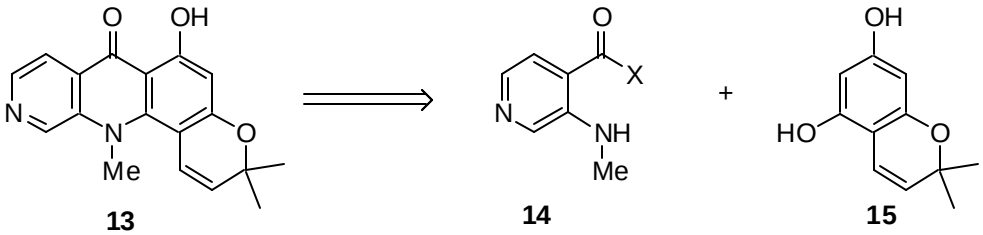
Following the procedure in reference 3, chromium trioxide (100 mg, 1.00 mmol, 3.0 equiv) was added in one portion to a stirred solution of 6,7-dihydro-4-(2-propenyl)-5*H*-cyclopenta[*c*]pyridin-5-ol (**12**) (59.1 mg, 0.337 mmol) in pyridine (3.2 mL) at room temperature. The reaction was monitored by TLC (silica, ethyl acetate). After consumption of the starting material (2 h) the reaction mixture was diluted with water (10 mL). The mixture was extracted twice with 10 mL portions of dichloromethane and the combined organic extracts were washed with water and brine and dried over anhyd magnesium sulfate. The solvent was evaporated in vacuo to give a clear colorless oil. The oil was purified using preparative thin layer chromatography (silica, ethyl acetate) to afford **3** (42 mg, 72% yield) as a clear colorless oil: IR (neat, NaCl) 2976, 2925, 1718 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.76 (s, 1H), 8.47 (s, 1H), 6.02-5.92 (m, 1H), 5.09 (dd, 1H, *J* = 6.8, 1.6 Hz), 5.06 (m, 1H), 3.80 (d, 2H, *J* = 6.8 Hz), 3.16 (apparent t, 2H, *J* = 6.0 Hz), 2.72 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 206.9, 148.9, 147.9, 147.6, 139.5, 135.2, 132.8, 116.7, 36.6, 32.7, 23.3; HRMS (ESI) calcd for $C_{11}H_{12}NO$ [M+H]: 174.0919; found 174.0913. The spectra are in agreement with those reported for **3**.[2]

Azacridone A: Introduction and Progress toward Total Synthesis

Azacridone A (**13**) is an example of an acridone type alkaloid that was isolated from the roots of Marsh grapefruit (*C. paradisi*). Acridone molecules have been known to be biologically active, most importantly to humans as antitumor and antiviral agents. These molecules are most often found in Rutaceous (*Citrus*) plants.

The molecule was isolated through extraction with refluxing acetone. The extracts were separated by chromatograpy and then run on a preparative TLC to isolate azacridone A (**13**).

What sets azacridone A (**13**) apart from other acridone derivatives is the presence of the second nitrogen in the leftmost ring. This is particularly interesting because at the time of isolation the only other known acridone compound with a second nitrogen was a synthetic compound. In other words, azacridone A (**13**) was the first naturally occurring aza-type acridone ever to be isolated.[15]



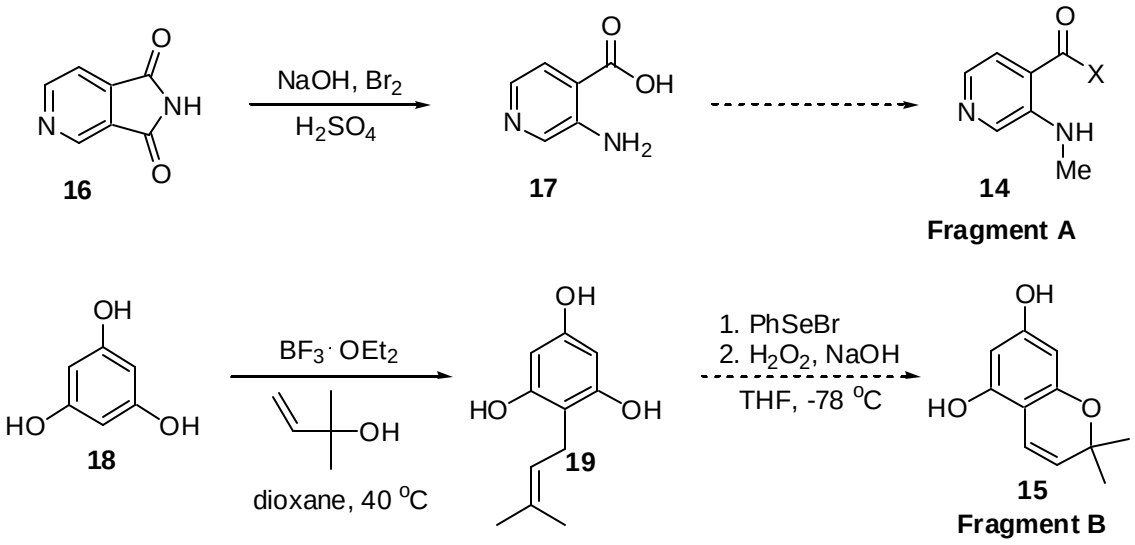
Scheme 2. Retrosynthesis of azacridone A (**13**).

[15] Takemura, Y.; Isono, Y.; Ju-Ichi, M.; Omura, M.; Ito, C.; Furukawa, H. *Chem. Pharm. Bull.* **1993**, *41*, 789.

This compound was chosen as a target for synthesis based on the possibility of biological activity and its somewhat complex four-ring structure that could easily be approached through the retrosynthesis as shown above (**Scheme 2**).

Fragment B (**15**) is a known benzopyran moiety, and there is strong literature precedence to support the formation of fragment A (**14**). The proposed synthesis of these two fragments is shown below (**Scheme 3**).

Synthesis of fragment A (**14**) began with commercially available compound 3,4-pyridinedicarboximide (**16**), which when treated with bromine, causes the imide ring to open and subsequently go through a Hoffman rearrangement. The resulting compound was quenched with sulfuric acid, which precipitated an orange solid (**17**).[16]



Scheme 3. The proposed synthesis of fragments A (**14**) and B (**15**).

The remainder of the synthesis of **14** has yet to be attempted. The proposed synthesis involves the conversion to an ester. This will be performed not only to make

[16] Chen, Q.; Deady, L. W. *Aust. J. Chem.* **1993**, *46*, 987.

the carbonyl less reactive, but also because of literature precedent that supports a ring closure between the ester and the aromatic portion of **15**.[17] With this in mind, the methylation of the amino group will most likely follow the esterification, which in turn will be followed by the reaction of the nitrogen with fragment B (**15**). These last two steps could most likely be performed in any order.

Synthesis of fragment B (**19**) began with starting material **18,** the commercially available compound phloroglucinol. This compound is known go through keto-enol tautomerization affording both the trihydroxyl form shown (**Scheme 3**), and its 1,3,5-triketylcyclohexane tautomer. Proton NMR shows that the predominant form of **18** is the aromatic tautomer. Compound **19** was made by prenylating the ring at a position *para* to one of the hydroxyl groups. This was performed by the removal of the alcohol from 2-methyl-3-buten-2-ol by boron trifluoride diethyl etherate.[18] This presumably forms a delocalized positive charge on the primary carbon of the butene. The aromatic ring then attacks to the primary carbocation to yield the prenylated compound **19**, which like **18** shows preference for the aromatic tautomer.

It is believed that the benzopyran moiety **15** will have preference for the enol tautomer as well. There is literature precedent for making **15** using a selenium bromide resin. Other literature supports the possibility of forming the same compound using phenylselenium bromide, which is a more cost effective and time efficient process than the making and using of a resin.[19]

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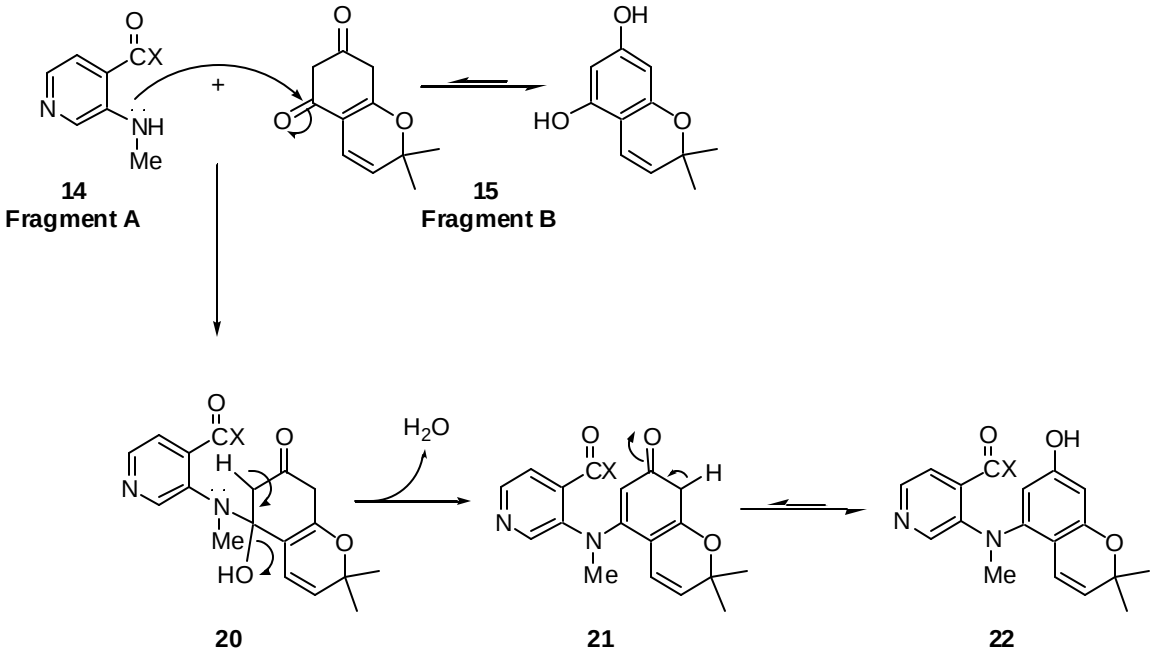
[17] Wilthopf, P.; Lackner, H. *Tetrahedron*. **1987**, *43*, 4549.
[18] Lee, H.-H. *J.Chem. Soc,. Perkin Trans. I.* **1981**, 3205.
[19] Nicolaou, K. C.; Pfefferkorn, J. A.; Roecker, A. J.; Cao, G.-Q.; Barluenga, S.; Mitchell, H. J. *J. Am. Chem. Soc.* **2000**, *122*, 9939.

The mechanism of the proposed addition of **14** to **15** is disputable due to the fact that the nitrogen can undergo a Michael addition to the aromatic ring or it can add to the tautomeric diketyl compound of **15.** This is dependent on the predominant tautomer of **15**. As was previously discussed, it is presumed that the enol tautomer is the preferred form, however, if one draws the attempted Michael addition, one can see through electron pushing that there is no place to stabilize the anion produced by amino addition. The only way the reaction could occur would be through an S_N2 type addition, which is highly unlikely on an aromatic ring. For this reason the addition of fragment A (**14**) to fragment B (**15**) will initially be attempted with the ketyl approach in mind (**Scheme 4**).



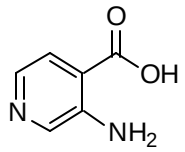
Scheme 4. The proposed addition mechanism of **14** to **15**.

The next step will be the ring closure of fragments A (**14**) and B (**15**), which will be accomplished by the deprotonation of the carbon positioned *ortho* to the hydroxyl

group on the aromatic ring (**22**).[17] The deprotonation will be performed using an organolithium reagent to be followed by acylation of the ester. The resulting cyclization will be the completed alkaloid azacridone A (**13**).

Azacridone A: Experimental Data[12]

Preparation of 3-aminoisonicotonic acid (17):

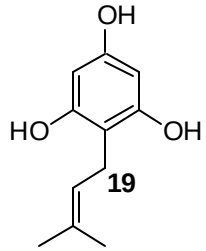


17

3,4-Pyridinedicarboximide (**16**, 1.00 g, 6.75 mmol) was added in portions to an ice-cold solution of sodium hypobromite, prepared by adding Br_2 (393 µL, 7.63 mmol, 1.13 equiv) dropwise to stirring of 10% NaOH (20 mL). A further 12 mL of 10 % NaOH were added to the resulting pale brown solution, and the reaction vessel was subsequently heated to 85 ºC over 30 min. The reaction vessel was then held at 85 ºC for 10 minutes before being cooled to room temperature. The resulting bright yellow solution was treated with 50% sulfuric acid solution at 0 ºC until a pH ~3 was achieved. With the addition of the acid, a bright orange precipitate formed. The solid was filtered and washed with water (3 x 5 mL). The resulting filtrate was neutralized, and the solid was dried under vacuum overnight to yield **18** (730 mg, 79% yield): orange solid, mp 300-302 ºC; IR (KBr) 3379, 3272, 2490, 2144, 1625, 810 cm^{-1}; ^1H NMR (400 MHz, DMSO) δ 8.19 (s, 1H), 7.72 (d, 1H, *J* = 5.2 Hz), 7.45 (d, 1H, *J* = 4.8 Hz); ^{13}C NMR (100 Hz, DMSO) d 168.4, 145.7, 140.5, 135.1, 122.9; LRMS (ESI) calcd for $C_6H_7N_2O_2$ [M+H]: 139.05; found 139.07. The spectra are in agreement with those reported for **17**.[16,20]

[20] For m.p. reference: Hurd, C. D.; Bethune, V. G. *J. Org. Chem.* **1970**, *35*, 1471.

Preparation of 2-(3-methylbut-2-enyl)phloroglucinol (19):



19

Boron trifluoride diethyl etherate (1.5 mL, 12 mmol, 0.30 equiv) was added dropwise (5 min) to a dry stirring solution of phloroglucinol (**18**, 5.00 g, 39.7 mmol) in dioxane (25 mL) at 40 ºC under N_2. After 15 min, 2-methyl-3-buten-2-ol (2.1 mL, 20 mmol, 0.50 equiv) was added dropwise (5 min) to the stirring solution at 40 ºC. After 1 h, the reaction was cooled to room temp, and the solution was extracted with diethyl ether (2 x 20 mL) and water (25 mL). The organic layers were combined, washed with water (4 x 15 mL) and brine (15 mL), dried over $MgSO_4$, and concentrated in vacuo. The crude mixture was purified using column chromatography (silica gel, 4 x 17 cm) with ethyl acetate:benzene (gradient 1:20-1:5) as eluent. The white needles isolated were then recrystallized ($CHCl_3$) to yield **19** (770 mg, 10 %)[18]: white needles, mp 98-100 ºC; IR (KBr) 3235, 1615, 1043 cm^{-1}; ^1H NMR (400 MHz, DMSO) d 8.79 (s, 2H), 8.69 (s, 1H), 5.74 (s, 2H), 5.10 (bt, 1H, J = 6.4 Hz), 3.02 (d, 2H, J = 6.8 Hz), 1.65 (s, 1H), 1.58 (s, 1H); ^{13}C NMR (100 MHz, DMSO) d 156.1, 155.6, 128.3, 124.6, 105.2, 94.1, 25.7, 21.8, 17.9; LRMS calcd 195.10; found 195.14 (trace amount); LRMS calcd $C_{11}H_{14}O_3Na$ [M + Na]: 217.08; found 217.12.[21,22]

[21] Proton NMR reference was performed in acetone. This was confirmed and then the product was run in DMSO for better peak resolution of the alcohols: Fukai, T.; Fujimoto, T.; Hano, Y.; Nomura, T.; Uzawa, J. *Heterocycles.* **1984**, *22*, 2805.

[22] For IR and m.p. reference see: Finnegan, R. A.; Morris, M. P.; Djerassi, C. *J. Org. Chem.* **1961**, *26*, 1180.

NMR Spectra for

Louisianin C and Azacridone A Compounds

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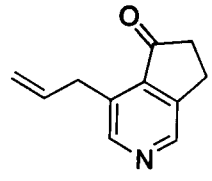

3

Louisianin C

'3C OBSERVE

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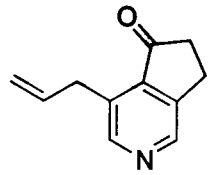

3

Louisianin C

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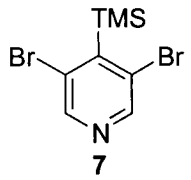

7

13C OBSERVE

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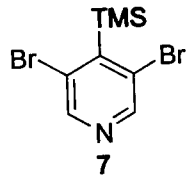
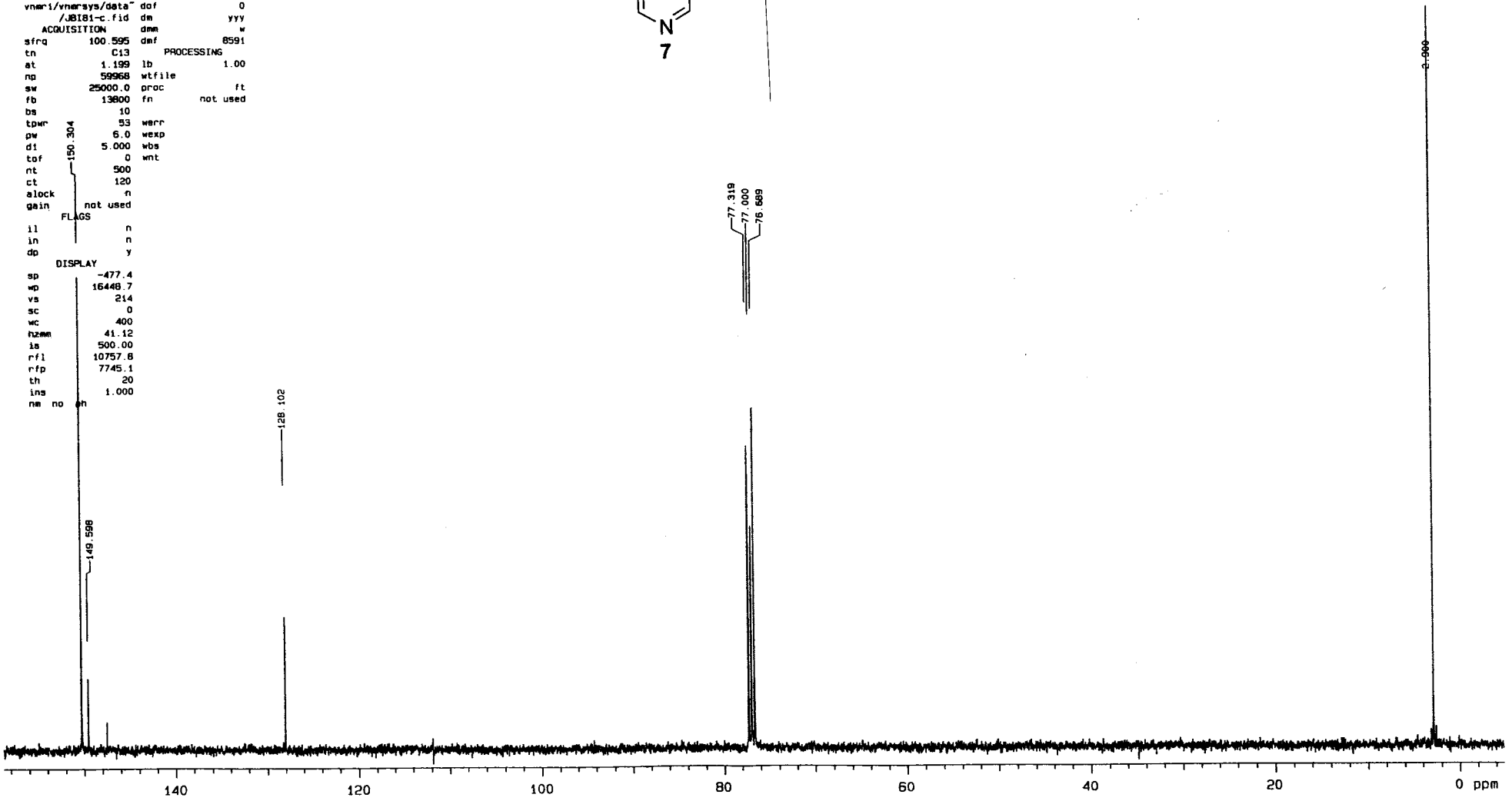


7



cs2-153

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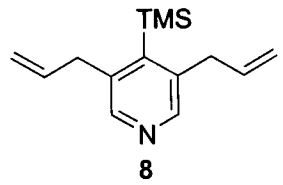

8

13C OBSERVE

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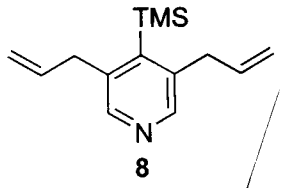

8

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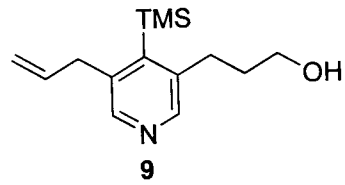

9

13C OBSERVE

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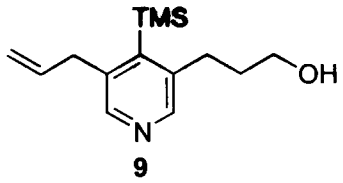
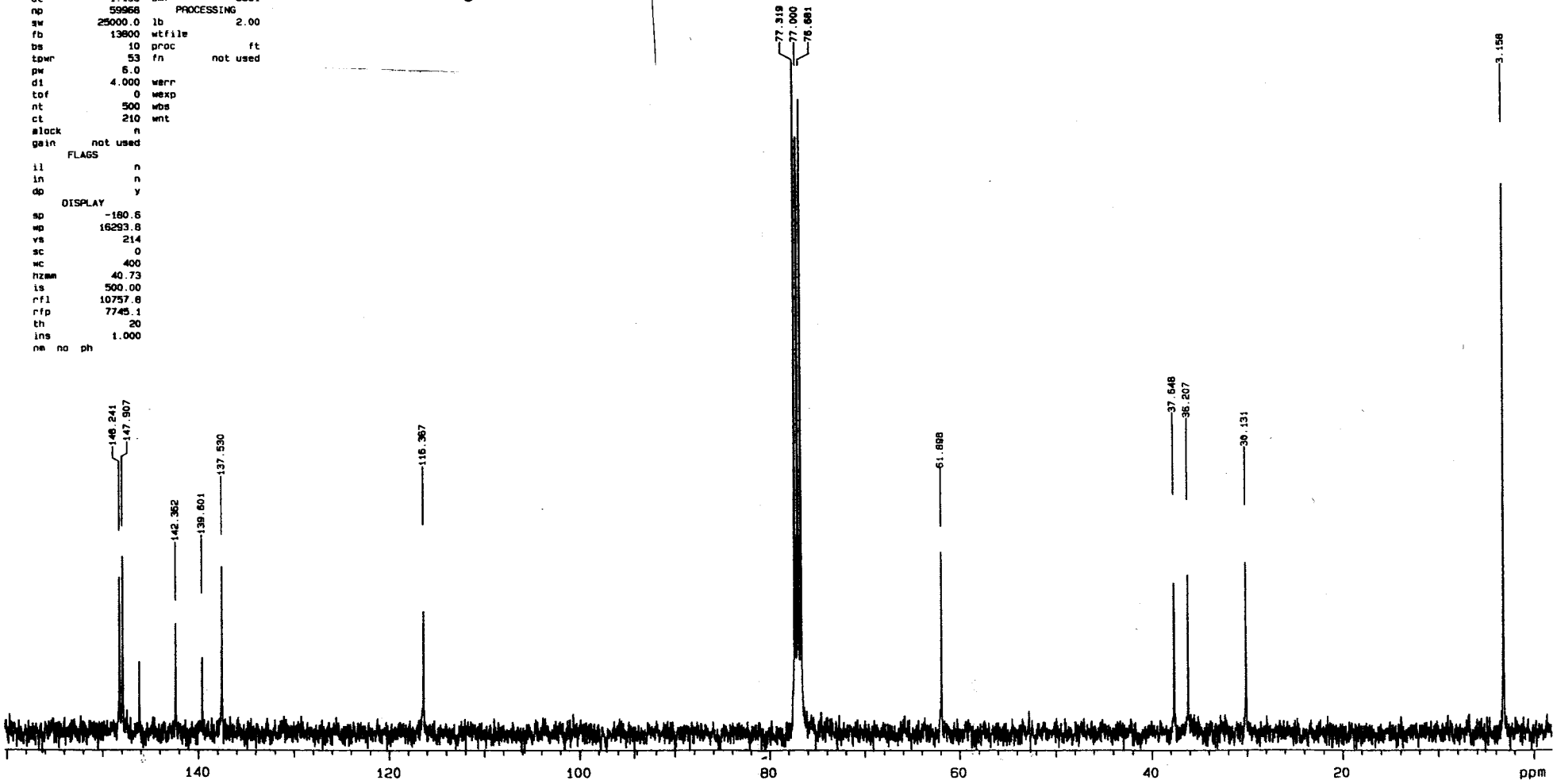


9



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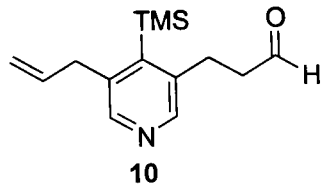
rfp	2177.4		
th	20		
ins	1.000		
ai	ph		

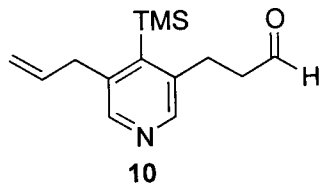


10

13C OBSERVE

exp1 std13c

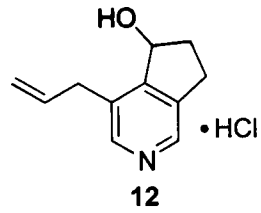
SAMPLE		DEC. & VT	
date	Nov 19 2002	dfrq	400.022
solvent	CDCl3	dn	H1
file	exp	dpwr	47
ACQUISITION		dof	0
sfrq	100.595	dm	yyy
tn	C13	dmm	w
at	1.199	dmf	9925
np	59968	PROCESSING	
sw	25000.0	lb	2.00
fb	13800	wtfile	
bs	10	proc	ft
tpwr	57	fn	not used
pw	6.0		
d1	4.000	werr	
tof	0	wexp	
nt	500	wbs	
ct	140	wnt	
alock	n		
gain	not used		
FLAGS			
il	n		
in	n		
dp	y		
DISPLAY			
sp	-3018.1		
wp	25000.0		
vs	214		
sc	0		
wc	400		
hzmm	0.98		
is	500.00		
rfl	10763.1		
rfp	7745.1		
th	20		
ins	1.000		
nm no ph			



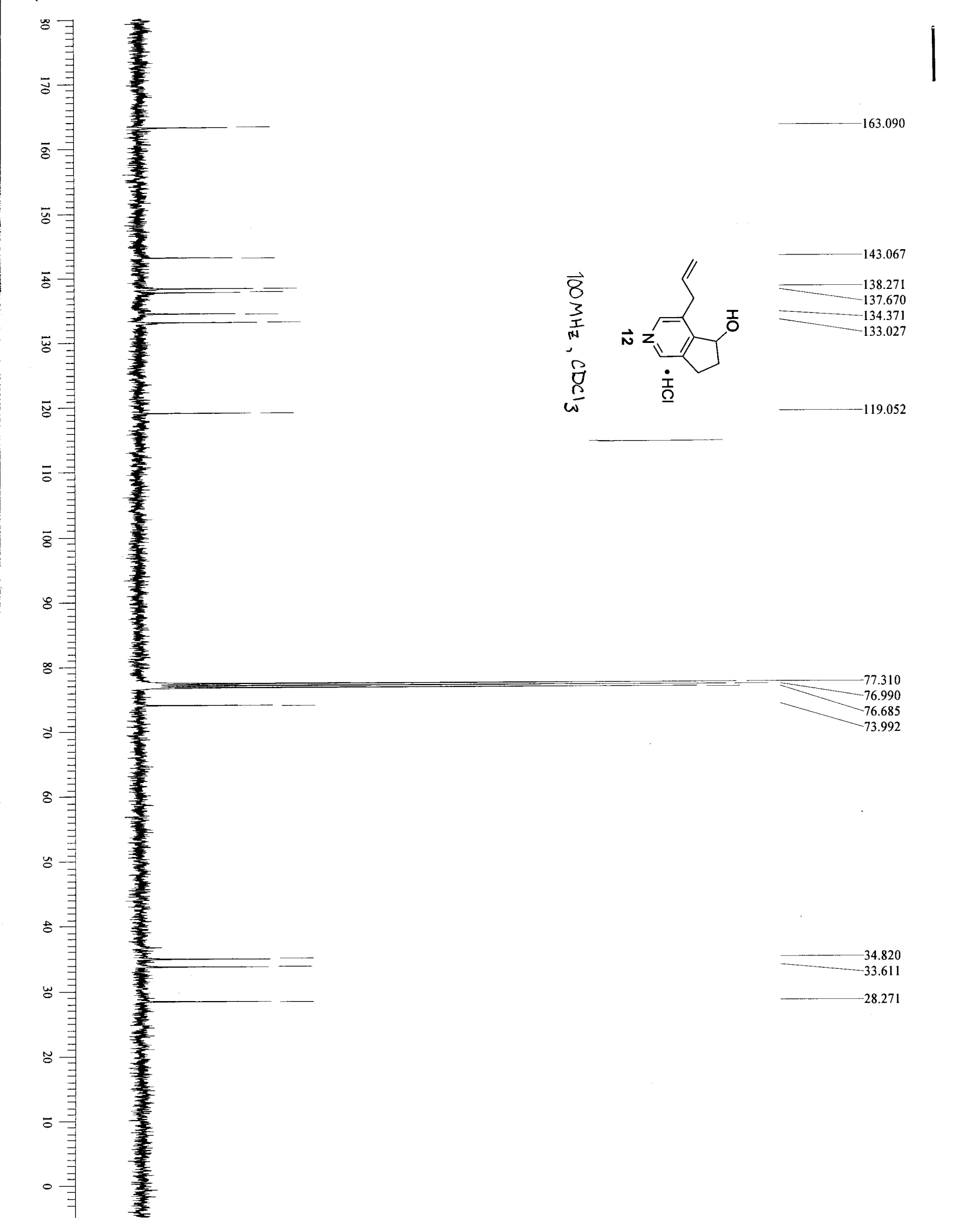
10

exp1 pulse sequence: std1h

SAMPLE		DEC. & VT	
date	Jan 9 03	dfrq	299.922
solvent	CDCl3	dn	H1
file	exp	dpwr	30
ACQUISITION		dof	0
sfrq	299.923	dm	nnn
tn	H1	dmm	c
at	3.543	dmf	200
np	32000	dseq	undefined
sw	4516.2	dres	undefined
fb	2500	homo	n
bs	4	PROCESSING	
pw	7.2	wtfile	
d1	1.000	proc	ft
tof	871.4	fn	32768
nt	120	math	f
ct	28		
alock	n	werr	
gain	not used	wexp	
FLAGS		wbs	
il	n	wnt	
in	n		
dp	y		
hs	nn		
DISPLAY			
sp	11.3		
wp	2986.3		
vs	86		
sc	0		
wc	400		
hzmm	7.47		
is	2060.72		
rfl	2481.8		
rfp	2171.4		
th	20		
ins	1.000		
ai	ph		



12

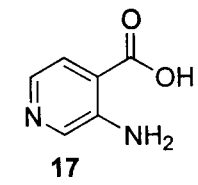

163.090
143.067
138.271
137.670
134.371
133.027
119.052
77.310
76.990
76.685
73.992
34.820
33.611
28.271
100 MHz, CDCl3
12
HO
N
•HCl

8.189
7.718
7.705
7.449
7.436
1.0
1.0
1.0
400MHz, DMSO
17
N
NH2
O
OH

13C OBSERVE

exp1 std13c

SAMPLE		DEC. S VT	
date	Mar 31 2003	dfrq	400.024
solvent	DMSO	dn	H1
file	exp	dpwr	55
ACQUISITION		dof	0.0
sfrq	100.595	dm	yyy
tn	C13	dmm	w
at	1.199	dmf	10000
np	59968	PROCESSING	
sw	25000.0	lb	1.00
fb	13800	wtfile	
bs	4	proc	ft
tpwr	57	fn	not used
pw	10.0		
d1	1.000	werr	
tof	0	wexp	
nt	500	wbs	wft
ct	148	wnt	
alock	n		
gain	not used		
FLAGS			
il	n		
in	n		
dp	y		
DISPLAY			
sp	-3046.6		
wp	25000.0		
vs	214		
sc	0		
wc	400		
hzmm	1.29		
is	500.00		
rfl	7039.9		
rfp	3993.3		
th	20		
ins	1.000		
nm no ph			



17

400 MHz, DMSO

8.841
8.796
8.700
5.789
5.746
5.136
5.120
5.102
3.333
3.037
3.020
2.502
1.702
1.660
1.629
1.585
-0.000
1.9
1.8
2.0
1.0
0.9
2.3
2.3
3.0
400 MHz, DMSO
HO
OH
OH
19

13C OBSERVE

exp1 std13c

SAMPLE		DEC. & VT	
date	Mar 31 2003	dfrq	400.024
solvent	DMSO	dn	H1
file	exp	dpwr	55
ACQUISITION		dof	0.0
sfrq	100.595	dm	yyy
tn	C13	dmm	w
at	1.199	dmf	10000
np	59968	PROCESSING	
sw	25000.0	lb	1.00
fb	13800	wtfile	
bs	10	proc	ft
tpwr	57	fn	not used
pw	10.0		
d1	1.000	werr	
tof	0	wexp	
nt	500	wbs	wft
ct	230	wnt	
alock	n		
gain	not used		
FLAGS			
il	n		
in	n		
dp	y		
DISPLAY			
sp	-1475.7		
wp	18444.6		
vs	214		
sc	0		
wc	400		
hzmm	46.11		
is	500.00		
rfl	7039.9		
rfp	3993.3		
th	28		
ins	1.0000		
nm no ph