Synthesis and Evaluation of Functionalized Benzoboroxoles as anti-Tuberculosis Agents

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Abstract

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Several aminobenzoboroxole derivatives have been prepared starting from o-boronobenzaldehyde. Nitration of the benzoboroxole followed by Pd-C mediated hydrogenation provided 6-aminobenzoboroxole. Using this amine as a common intermediate, numerous structurally interesting benzoboroxoles have been prepared employing aromatic bromination, N-alkylation, reductive amination, and N-amidation. 3-Substituted functionalized benzoboroxoles have been synthesized via Baylis-Hillman reaction followed by nitration and reduction which provided highly functionalized amino benzoboroxoles.

These derivatives have been evaluated for their anti-tubercular activity on mycobacterium tuberculosis H37Rv using 7H9 and GAST protocols. Based on these studies, a potent benzoboroxole analog has also been identified for further development.

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LIST OF ABBREVIATIONS

TB	Tuberculosis
HIV	Human immunodeficiency virus
MDR-TB	Multidrug resistant TB
XDR-TB	Drug resistant strains of TB
H37Rv	*Mycobacterium tuberculosis* strains
MIC	Minimum inhibitory concentration
EDC	1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide
DMAP	4-Dimethylaminopyridine
HCl	Hydrochloric acid
NMP	*N*-methylpyrrolidine
BSA	bis-(trimethylsilyl)acetamide
BCG	*Mycobacterium bovis*
NaH	Sodium hydride
THF	Tetrahydrofuran
HPLC	High performance liquid chromatography
$Pd(PPh_3)_4$	Tetrakis(triphenylphosphine)palladium(0)
mPTPB	Mycobacterium tuberculosis protein tyrosine phosphatase
HOBT	Hydroxybenzotriazole
DMF	*N*,*N*-dimethylformamide
PCl_5	Phosphorus pentachloride
$POCl_3$	Phosphorus (V) oxychloride
NH_4OH	Ammonium hydroxide
KHMDS	Potassium bis(trimethylsilyl)amide
DMSO	Dimethyl sulfoxide
DABCO	1,4-diazabicyclo[2.2.2]octane
MCF-7	Michigan Cancer Foundation-7
MBC	Minimum bactericidal concentration
MAC	Minimum anaerobicidal concentration

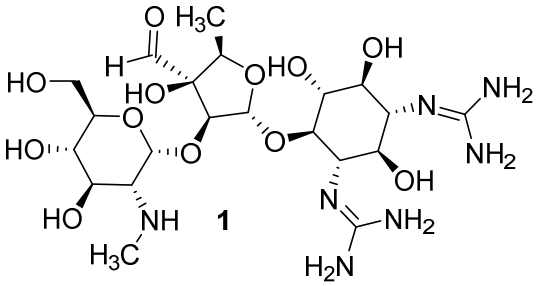
INH	Isoniazid
CFU	Colony forming units (growth of bacteria)
7H9	Liquid growth medium for *M. Smeg.*
GAST	Glycerol-alanine-salts-tween
TLC	Thin layer chromatography
^{1}H NMR	Proton (^{1}H) nuclear magnetic resonance spectrum
^{13}C NMR	Carbon (^{13}C) nuclear magnetic resonance spectrum
Hz	Hertz
MHz	Mega Hertz
J	Coupling constant
Pd-C	Palladium on carbon
$NaHCO_3$	Sodium bicarbonate
$MgSO_4$	Magnesium sulfate anhydrous
$NaBH_4$	Sodium borohydride
CO_2	Carbondioxide
IMEM	Improved minimum essential medium
DPBS	Dulbecco's phosphate buffered saline
SRB	Sulforhodamine-B
NaOH	Sodium hydroxide
NaCl	Sodium chloride
OADC	Oleic Albumin Dextrose Catalase
OD	Optical density
RPMI	Roswell Park Memorial Institute growth medium
FBS	Fetal bovine serum
PBS	Phosphate buffered saline
EDTA	Ethylenediaminetetraacetic acid
SDS	Sodium dodecyl sulfate

Chapter 1: Introduction

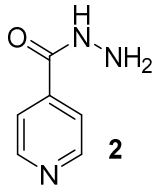
Tuberculosis (TB) is a highly contagious chronic bacterial infection that affects nearly one third of world's population, which results in latent TB.[1] About 10% of this affected population develop active disease over their lifetime. TB is the most common disease in Asia and Africa with India and China constituting about 40% of world's TB cases. This infection is spread through the air and causes millions of deaths every year making it the second greatest killer next to HIV. The disease is rapidly progressed in HIV affected patients of which 25% death is due to TB.

In 1944, Streptomycin **1** was the first effective anti-tubercular drug, which was considered to be the "cure" for tuberculosis but most of the cured patients had a relapse and the standard four-drug regimen of isoniazid **2**, rifampin **3**, ethambutol **4**, and pyrazinamide **5** came into existence during 1952-1961. However, the prolonged treatment with these first line drugs over period of six to nine months resulted in resistant strains of TB.

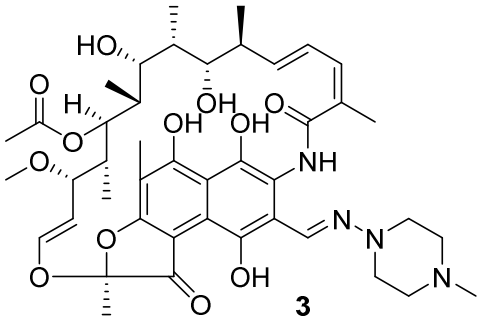
TB is caused by *Mycobacterium tuberculosis*. The emergence of multi-drug resistant TB (MDR-TB) and extremely drug resistant strains of TB (XDR-TB) is a cause of great concern. According to WHO, 3.7% of the world's new cases and 20% of previously treated cases were estimated as MDR-TB. Hence, novel, highly effective therapeutic strategies are urgently required for the treatment of this epidemic. Some of the new generation compounds that are in various stages of clinical development for the treatment of TB are listed below.[2]



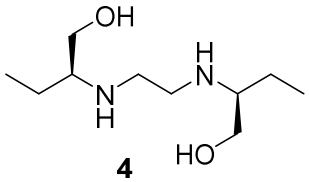
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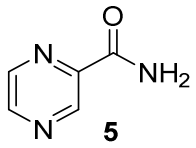
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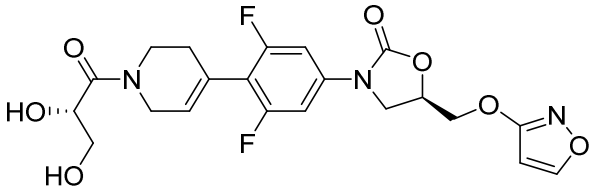
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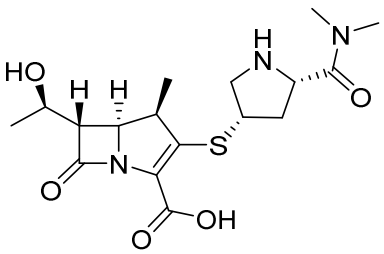
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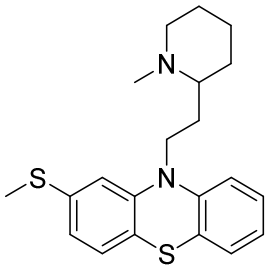
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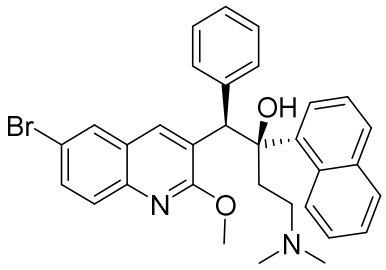
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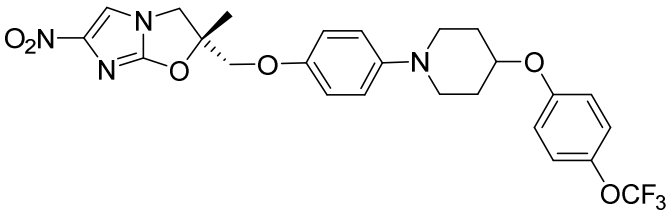
Meropenem



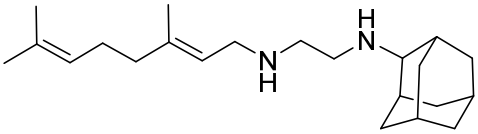
Thioridazine



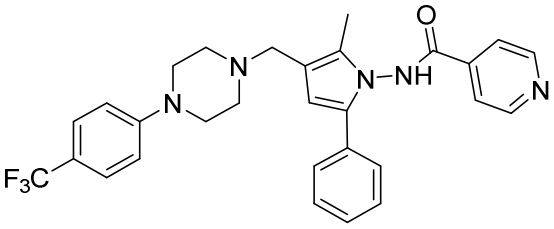
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OPC-67683



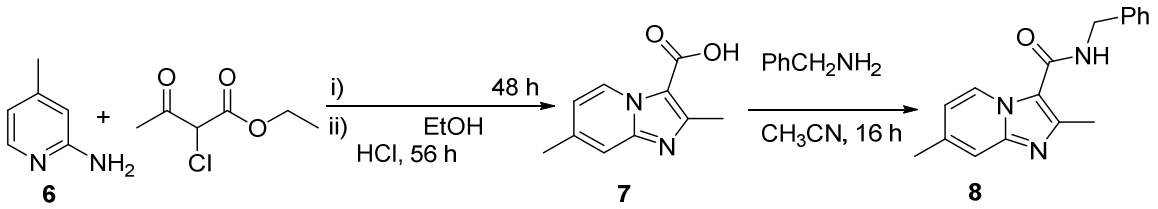
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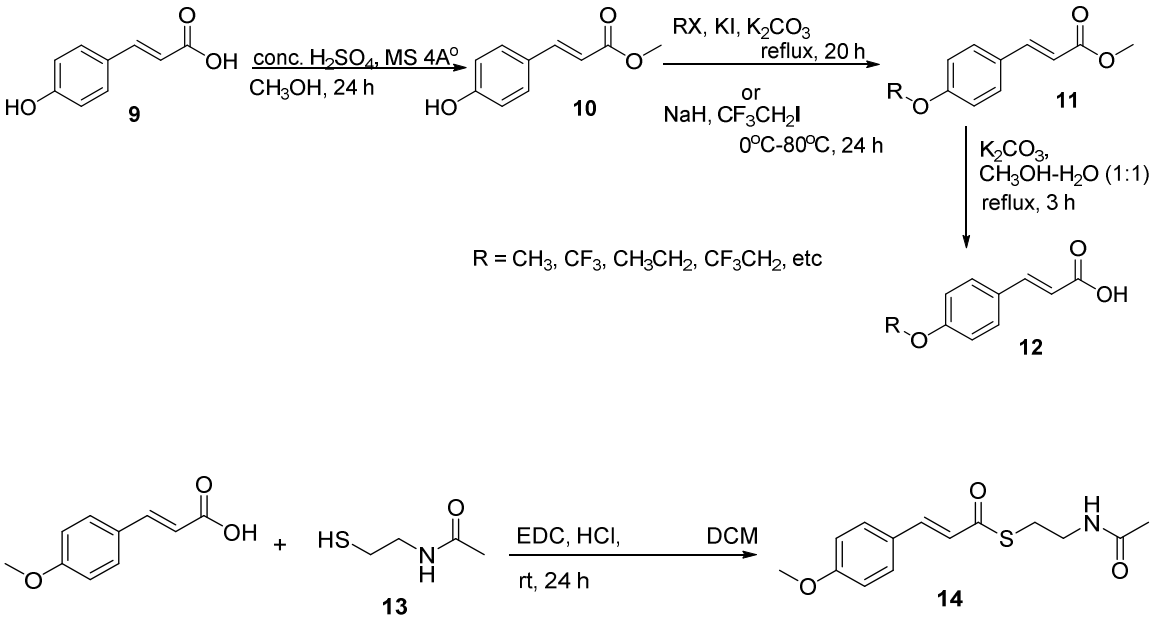
Owing to the importance of the treatment of MDR-TB, numerous papers have been published. Some of the molecules that are being developed as *anti*-tubercular agents have been delineated below.

Miller and coworkers reported novel imidazo[1,2-a]pyridine-3-carboxamides as potent *anti*-tubercular agents against multi drug resistant and extended drug resistant strains. The synthesis of these compounds was achieved by condensing 2-amino-4-picoline **6** and ethyl-2-chloroacetoacetate in 1,2-dimethoxyethane in the presence of lithium hydroxide in ethanol to obtain ethyl 2,7-dimethylimidazo[1,2-a]pyridine-3-carboxylate **7**. This compound **7** was coupled with benzyl amine in acetonitrile using EDC and DMAP, to yield *N*-benzyl-2,7-dimethylimidazo[1,2-a]pyridine-3-carboxamide **8** (**Scheme 1.1**).[3] The synthesized compounds were evaluated for their *in vitro* anti-tuberculosis activity versus replicating, non-replicating, multi- and extensive drug resistant Mtb strains. The MIC values were found to be in the following range. H37Rv (MIC TB = 0.4 – 1.9 µM), MDR-TB (MIC = 0.07-2.2 µM), XDR-TB (MIC = 0.07 – 0.14 µM), VERO toxicity (IC_{50} > 100 µM).



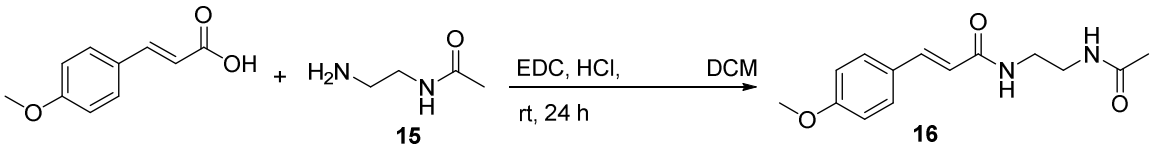
Scheme 1.1: Synthesis of imidazo[1,2-a]pyridine-3-carboxamides as potent *anti*-tubercular agents

Baltas *et. al.* reported a series of thioester, amide, hydrazide, and triazolophthalazine derivatives of 4-alkoxy cinnamic acid as potential *anti*-tubercular agents. *(E)*-4-hydroxycinnamic acid **9** was treated with sulfuric acid in methanol and stirred for 24 h to obtain methyl ester **10**. This ester **10** was reacted with alkyl halide in presence of potassium iodide, potassium carbonate and refluxed in acetone to yield *(E)*-alkyl-3-(4-alkoxyphenyl)prop-2-enoate **11**. The ester **11** was hydrolyzed with potassium carbonate in methanol-water to obtain *(E)*-4-alkoxycinnamic acid **12**. This acid **12** and N-acetylcysteamine **13** were coupled using DMAP and EDC in dry dichloromethane at room temperature to obtain *(E)*-S-2-acetamidoethyl 3-(4-methoxyphenyl)prop-2-enethioate **14** (**Scheme 1.2**).[4] The MIC value of **14** on H37Rv was found to be > 1900 μM.



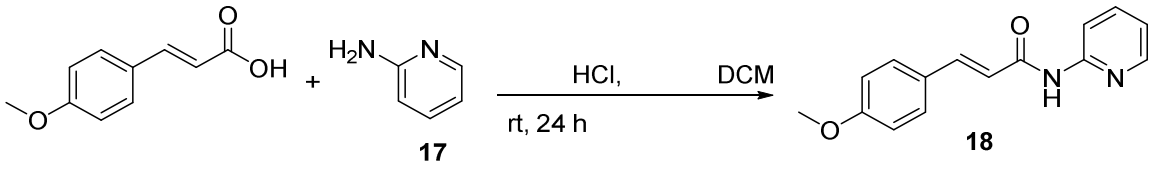
Scheme 1.2: Synthesis of *(E)*-S-2-acetamidoethyl 3-(4-methoxyphenyl)prop-2-enethioate

Similarly, **12** and *N*-acetylethylenediamine **15** were coupled using DMAP and EDC.HCl in dry dichloromethane at room temperature to obtain *(E)-N*-(2-acetamidoethyl)-3-(4-methoxyphenyl)prop-2-enamide **16** (**Scheme 1.3**).[4] The MIC value of **16** on H37Rv was found to be 225 μM.



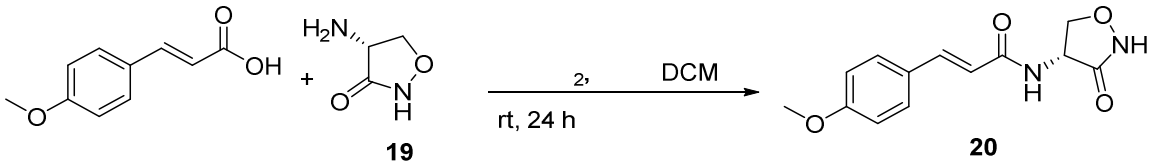
Scheme 1.3: Synthesis of *(E)-N*-(2-acetamidoethyl)-3-(4-methoxyphenyl)prop-2-enamide

12 and 2-amino pyridine **17** were coupled using DMAP and EDC in dry dichloromethane at room temperature to obtain *(E)-N*-(Pyridin-2-yl))-3-(4-mehoxyphenyl)prop-2-enamide **18** (**Scheme 1.4**).[4] The MIC value of **18** on H37Rv was found to be 248 μM.



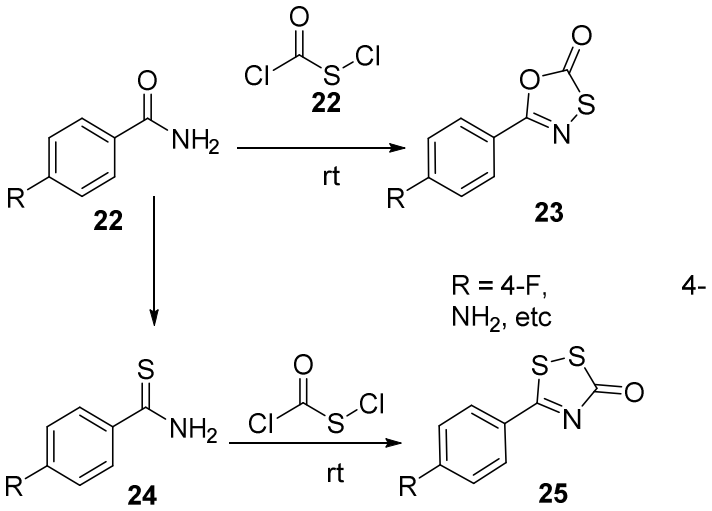
Scheme 1.4: Synthesis of *E)-N*-(Pyridin-2-yl))-3-(4-mehoxyphenyl)prop-2-enamide

12 and oxalyl chloride were stirred at room temperature for 2 h, and this reaction mixture was added to another round-bottom flask containing D-cycloserine **19** in dry dichloromethane, *N*-methylpyrrolidine (NMP) and bis-(trimethylsilyl)acetamide (BSA) and were stirred under N_2 at room temperature for 1 h, followed by the treatment with pyridine and acyl chloride and stirred at room temperature for 12 h to obtain *(E)-N*-(3-oxo-1,2-oxazolidin-4-yl)-3-(4-methoxyphenyl)prop-2-enamide **20** (**Scheme 1.5**).[4] The MIC value of **20** on H37Rv was found to be 950 μM.



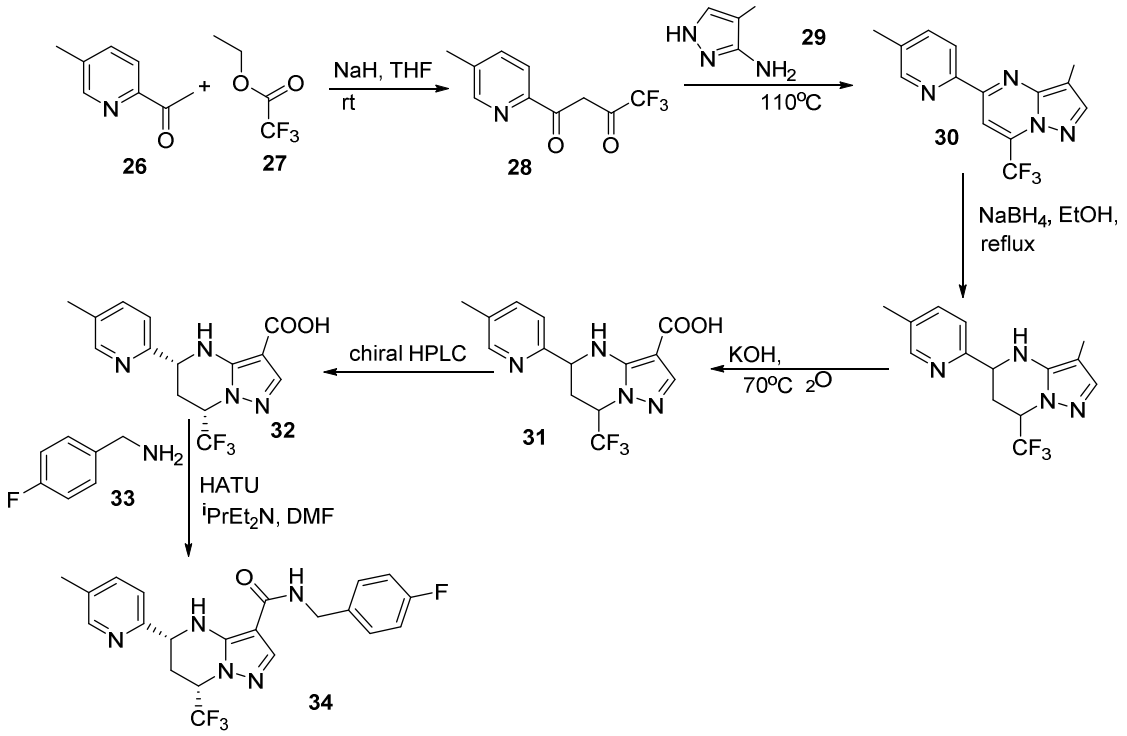
Scheme 1.5: Synthesis of *(E)-N*-(3-oxo-1,2-oxazolidin-4-yl)-3-(4-methoxyphenyl) prop-2-enamide

Wang, *et. al*. recently reported small molecules containing oxathiazol-2-one moiety as potent inhibitors of *Mycobacterium bovis* (BCG). Chlorocarbonylsulfenyl chloride **21** was added to the amide **22** in THF at room temperature to furnish 1,3,4-oxathiazol-2-one **23**. The amide **22** in THF was refluxed with Lawesson's reagent to obtain thioamide **24**. This **24** was treated with chlorocarbonylsulfenyl chloride at room temperature to get the corresponding 1,2,4-dithiazol-3-ones **25, 26** (**Scheme 1.6**).[5] The synthesized compounds were assessed for *anti*-tubercular activities on the virulent H37Rv strain and their MIC values found to be in the 8-64 µg/mL range.



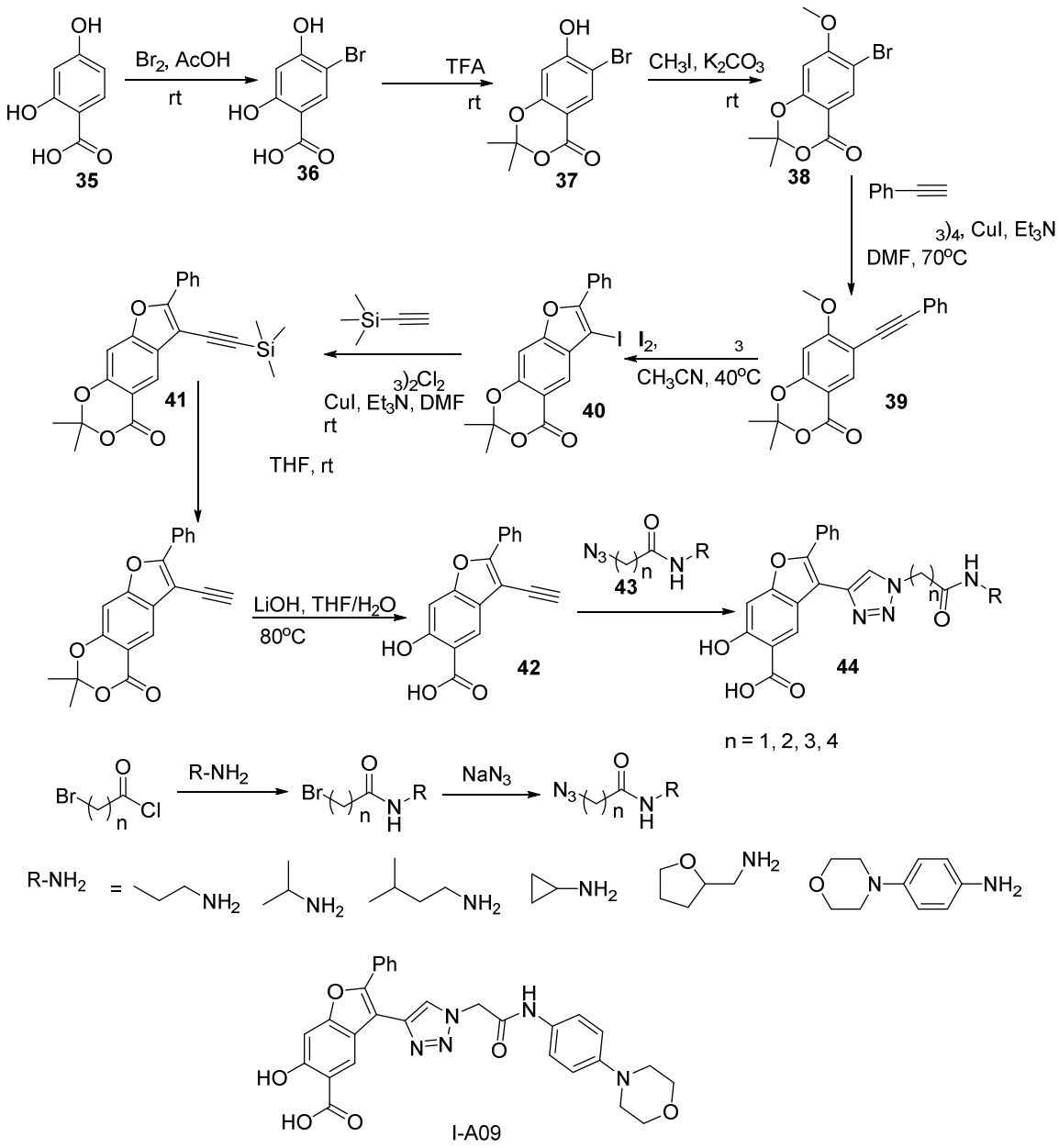
Scheme 1.6: Synthesis of molecules containing oxathiazol-2-one moiety

Yokokawa *et. al.* recently reported tetrahydropyrazolopyrimidine carboxamide derivatives as potent *anti*-tubercular agents. 5-methyl-2-acetylpyridine **26** was treated with NaH in THF and ethyl 2,2,2-trifluoroacetate **27** was added to afford 4,4,4-trifluoro-1-(5-methylpyridin-2-yl)butane-1,3-dione **28**, which was reacted with 3-amino-1H-pyrazole-4-carboxylic acid ethyl ester **29** to yield ethyl 5-(5-methylpyridin-2-yl)-7-(trifluoromethyl)pyrazolo[1,5-a]pyrimidine-3-carboxylate **30**. The ester **30** was converted to acid **31** via reduction with sodium borohydride followed by alkaline hydrolysis. The racemic acid was subjected to preparative chiral HPLC in order to effect the separation of enantiomers. The chiral acid **32** thus obtained was coupled with (4-fluorophenyl) methanamine **33** to afford (5R,7S)-N-(4-fluorobenzyl)-5-(5-methylpyridin-2-yl)-7-(trifluoromethyl)-4,5,6,7-tetrahydropyrazolo[1,5-a]pyrimidine -3-carboxamide **34** (**Scheme 1.7**).[6] Its MIC value in Mtb H37Rv strain was found to be 0.44 μM.



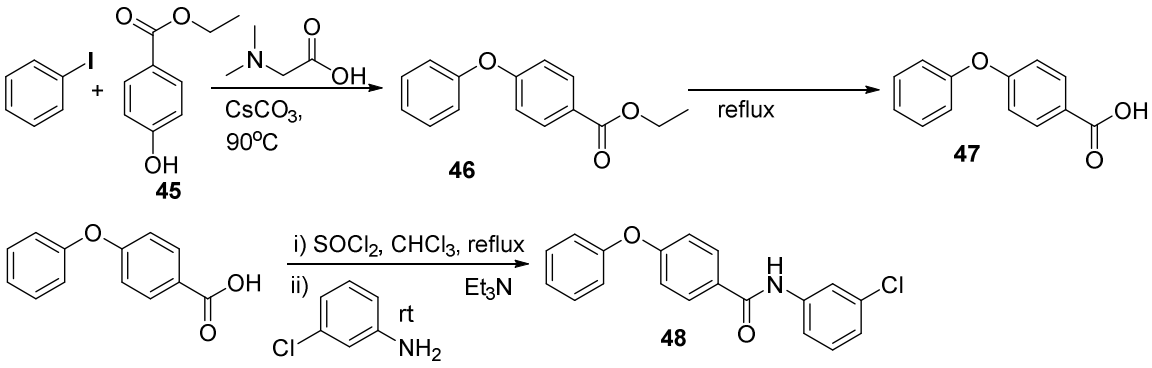
Scheme 1.7: Synthesis of tetrahydropyrazolopyrimidine carboxamides

Zhou *et. al.* have reported triazole derivatives as anti-tuberculosis agents that target mycobacterium protein tyrosine phosphatase B. 4-hydroxysalicylic acid **35**, upon regioselective bromination, afforded 5-bromo-4-hydroxysalicylic acid **36**. This acid **36** was protected with acetone in presence of trifluoroacetic anhydride/trifluoroacetic acid to obtain dioxanone **37**, which was alkylated using methyl iodide to yield the aryl methyl ether **38**. This was further coupled with phenylacetylene in the presence of $Pd(PPh_3)_4$ to get **39**, which was then treated with molecular iodine to induce cyclization. The iodo-olefin product **40** was then coupled with ethynyltrimethylsilane to furnish the compound **41**. Desilylation and deacetylation of **41** provided the core alkyne **42**, which was then coupled with an azide **43** using click chemistry to obtain the product **44** (**Scheme 1.8**).[7] The IC_{50} of *anti*-tubercular activity in compound I-A09 on mPTPB strain was found to be 1.26 μM.



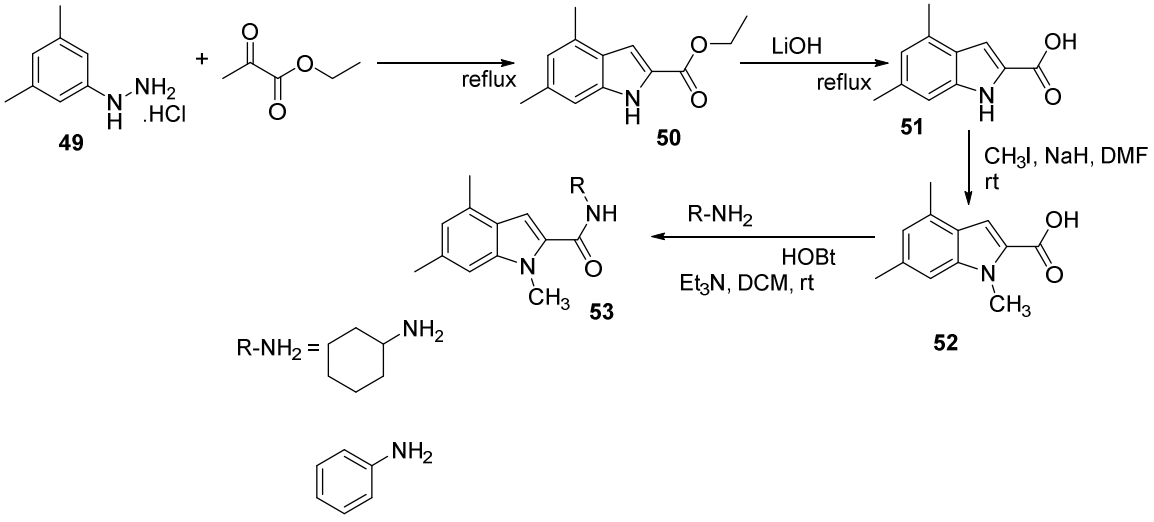
Scheme 1.8: Synthesis of triazole derivatives as anti-tuberculosis agents

Luo *et. al.* synthezied a series of compounds with a diphenyl ethers utilizing *N*,*N*-dimethylglycine-promoted Ullmann coupling reaction. Ethyl 4-phenoxybenzoate **46** was synthesized by condensing iodobenzene with ethyl 4-hydroxybenzoate **45**, which was then hydrolyzed to obtain the acid **47** by refluxing with sodium hydroxide in ethanol-water. The acid **47** was then treated with thionyl chloride to obtain the acyl chloride, which was then coupled with 3-chloroaniline to furnish the corresponding amide **48** (**Scheme 1.9**).[8] The *anti*-tubercular activity of compound **48** was evaluated against Mycobacterium tuberculosis H37Rv and its MIC was found to be 32 µg/mL.



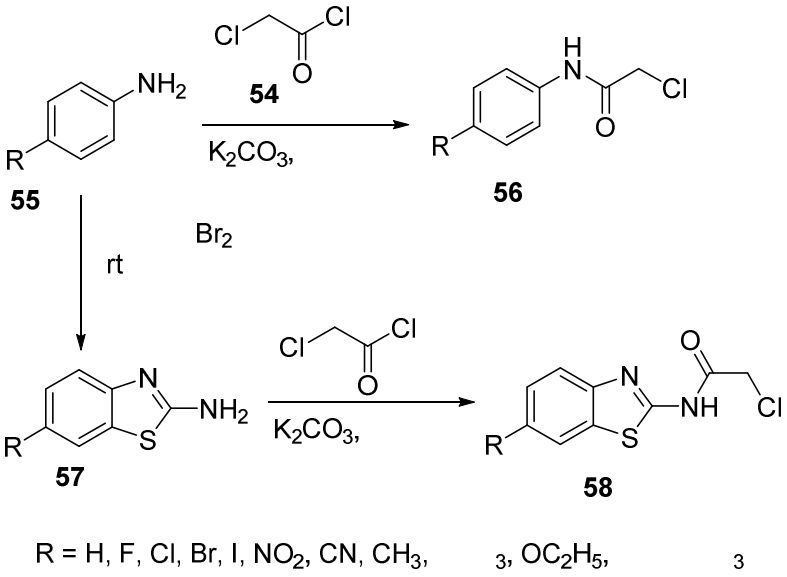
Scheme 1.9: Synthesis of diphenylether derivatives

Kozikowski *et. al.* reported the synthesis of indolecarboxamide derivatives as novel *anti*-tubercular agents against drug susceptible and drug resistant mycobacterium tuberculosis strains. The synthesis involved the condensation of 3,5-dimethylphenylhydrazine hydrochloride **49** and ethyl pyruvate under acidic conditions using Fischer indole synthesis to afford the disubstituted indole-2-carboxylate **50**, which upon hydrolysis in lithium hydroxide in ethanol afforded the corresponding acid **51**. *N*-Methylation of the acid **51** followed by hydrolysis yielded carboxylic acid **52**, which was subsequently coupled with the corresponding amine in the presence of EDC, HOBT, and triethylamine to obtain the amide **53** (**Scheme 1.10**).[9] The *anti*-tubercular activity of these compounds were evaluated on XDR-TB strains and its MIC values were found to be in the range of 0.012 – 500 µM.



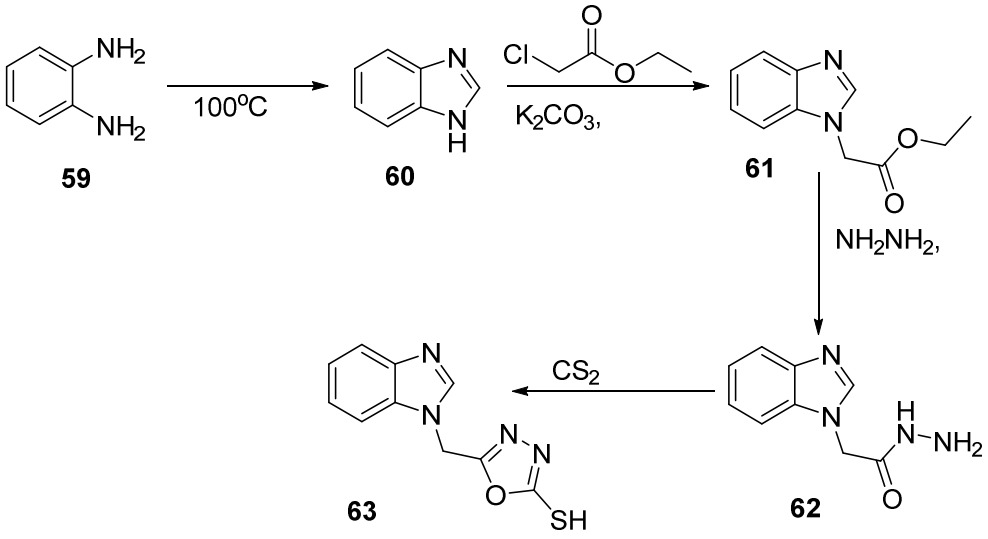
Scheme 1.10: Synthesis of indolecarboxamide derivatives

Patel *et. al.* reported benzimidazolyl-1,3,4-oxadiazol-2ylthio-N-phenyl (benzothiazolyl) acetamides as *anti*-bacterial, *anti*-fungal and *anti*-tuberculosis agents. The synthesis involved the coupling of chloroacetyl chloride **54** and amine **55** in presence of potassium carbonate and acetone to obtain corresponding amide **56**. Benzothiazole **57** was prepared by treating amine **55** with potassium thiocyanate in bromine-acetic acid mixture at room temperature. This thiazole **57** was then coupled with chloroacetyl chloride in the presence of potassium carbonate in benzene to furnish the corresponding amide **58** (**Scheme 1.11**).[10]



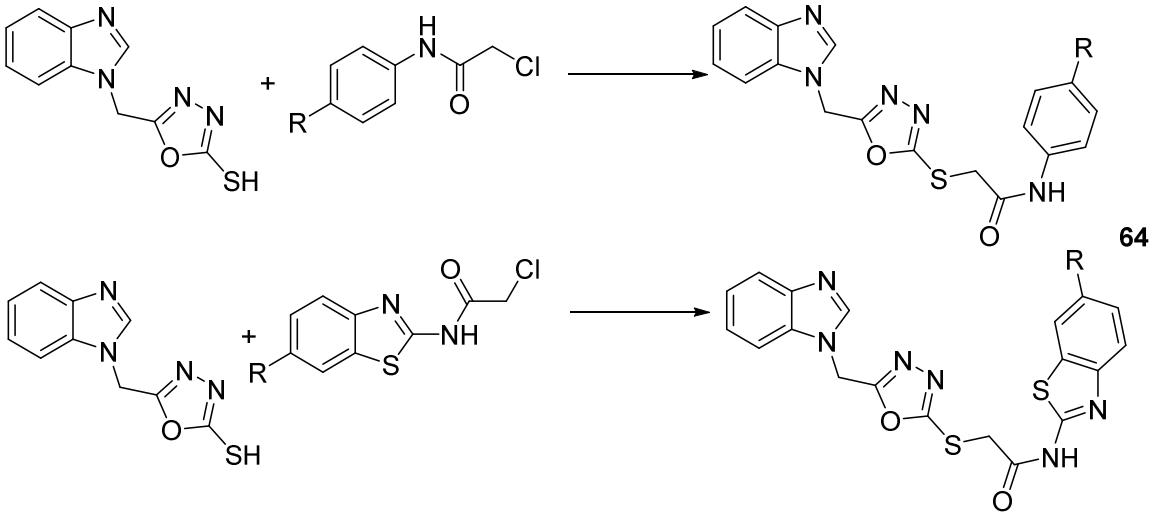
Scheme 1.11: Synthesis of chloroacetamides

o-Phenylene diamine **59** upon heating with formic acid at 100^{o}C yielded 1H-benzimidazole **60**. This was refluxed in ethyl chloroacetate in the presence of potassium carbonate and methanol to obtain ethyl 1H-benzimidazol-1-ylacetate **61**, which upon treatment with hydrazine in ethanol furnished 2-(1H-benzo[d]imidazol-1-yl)acetohydrazide **62**. This hydrazide **62** was reacted with carbon disulfide in ethanol to yield 5-((1H-benzo[d]imidazol-1-yl)methyl)-1,3,4-oxadiazole-2-thiol **63** (**Scheme 1.12**).[10]



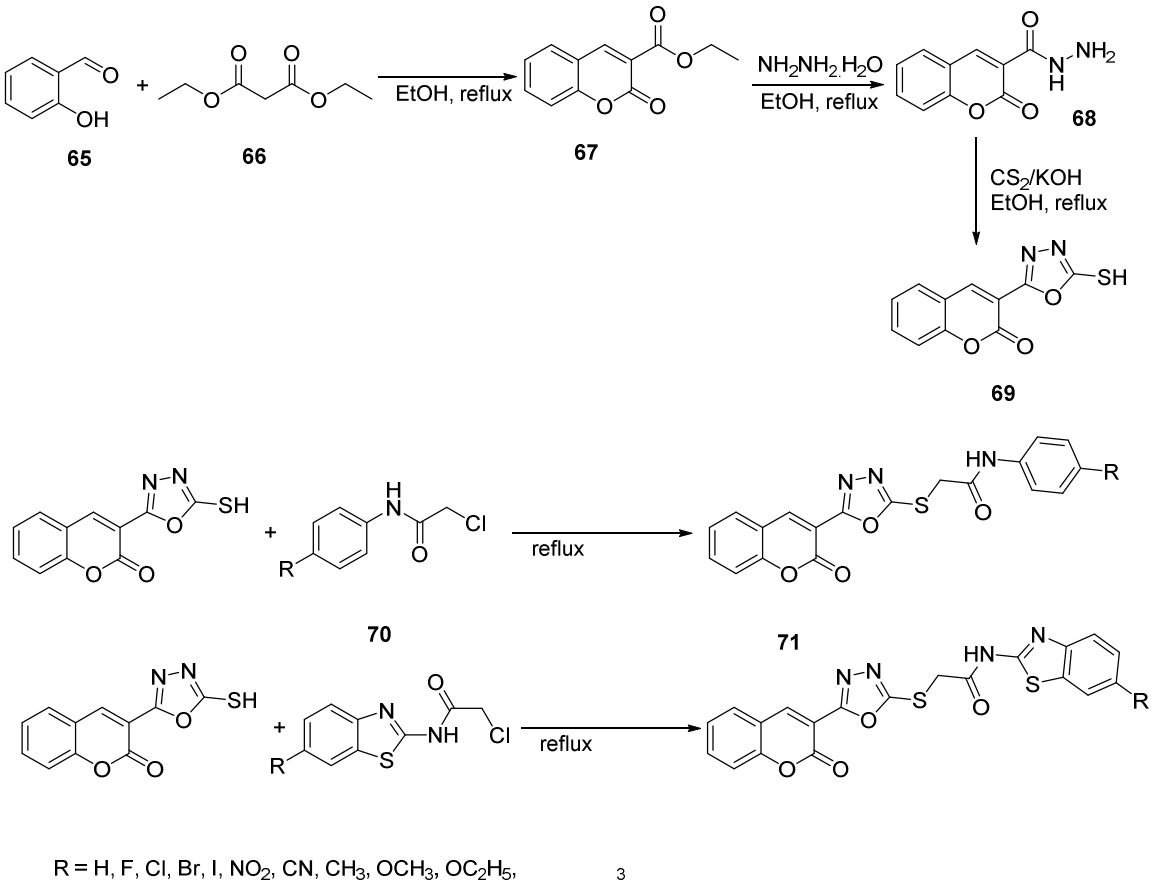
Scheme 1.12: Synthesis of 5-((1H-benzo[d]imidazol-1-yl)methyl)-1,3,4-oxadiazole-2-thiol

The thiol **63** was further condensed with the amide **56** in presence of potassium carbonate in acetone to furnish the corresponding benzimidazolyl-1,3,4-oxadiazol-2ylthio-N-phenyl-acetamides **64** (**Scheme 1.13**).[10] These compounds were evaluated for *anti*-tubercular activity on MTB H37Rv and their MIC values were found to be in the range of 25 – 500 µg/mL.



Scheme 1.13: Synthesis of benzimidazolyl-1,3,4-oxadiazol-2ylthio-N-phenyl-acetamides

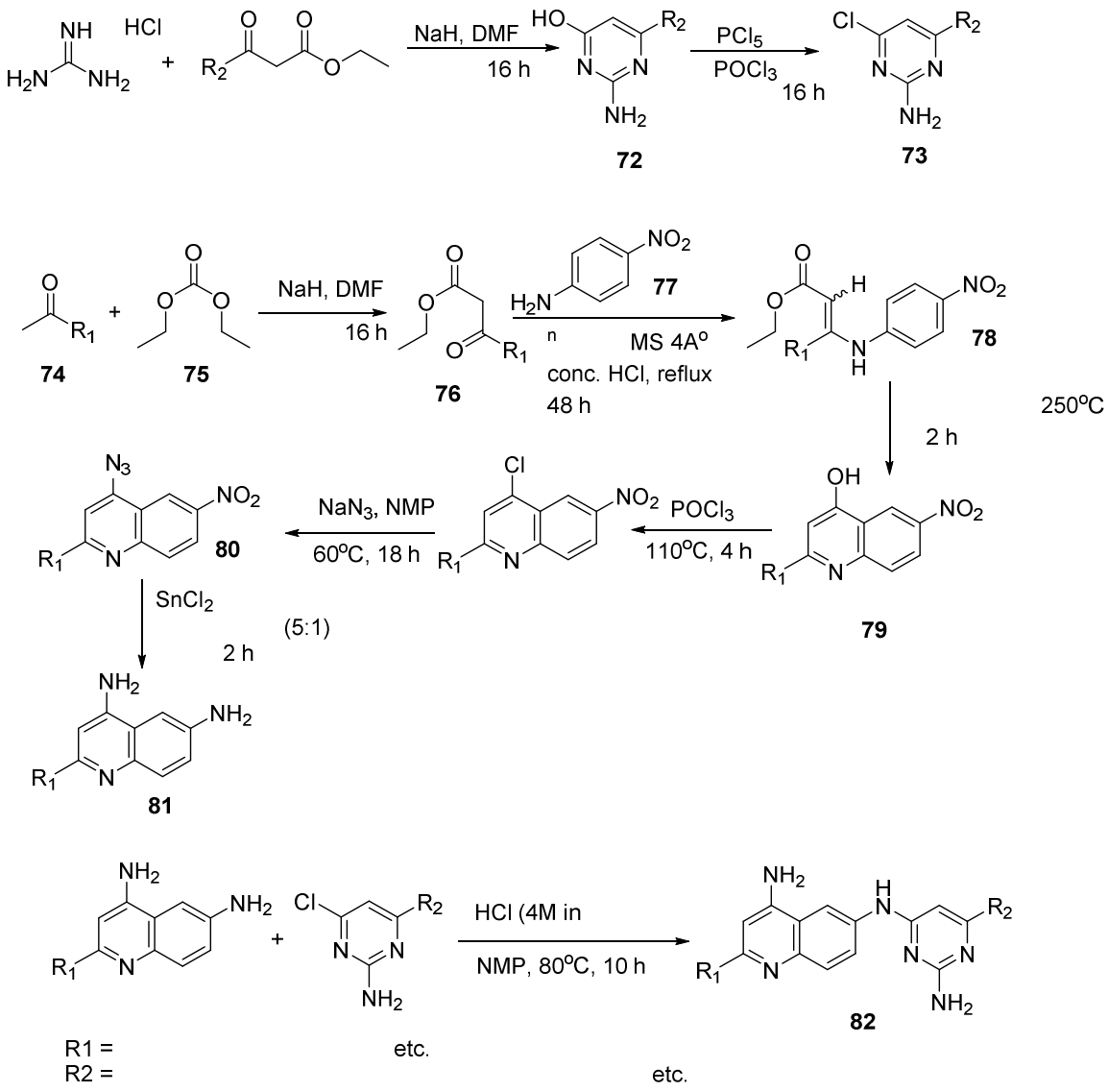
Patel *et. al.* recently reported coumarin-based 1,3,4-oxadiazol-2ylthio-N-phenyl/benzothiazolyl acetamides as *anti*-microbial and *anti*-tuberculosis agents. The synthesis involved the condensation of salicylaldehyde **65** and diethylmalonate **66** in presence of piperidine in ethanol to obtain ethyl 2-oxo-2H-chromene-3-carboxylate **67**, which was further reacted with hydrazine in ethanol to yield 2-oxo-2H-chromene-3-carbohydrazide **68**. This hydrazide **68**, upon treating with carbon disulfide in potassium hydroxide and ethanol yielded 3-(5-mercapto-1,3,4-oxadiazol-2-yl)-2Hchromen-2-one **69**. This was condensed with the amide **70** in presence of potassium carbonate in acetone to furnish the corresponding 2-(5-2-oxo-2Hchromen-3-yl)-1,3,4-oxadiazol-2-ylthio)-N-phenyl acetamides **71** (**Scheme 1.14**).[11] The *anti*-tubercular activity (MIC) of these compounds on MTB H37Rv was found to be in the range of 3.12–25 µg/mL.



Scheme 1.14: Synthesis of coumarin-based 1,3,4-oxadiazol-2ylthio-N-phenyl/benzothiazolyl acetamides

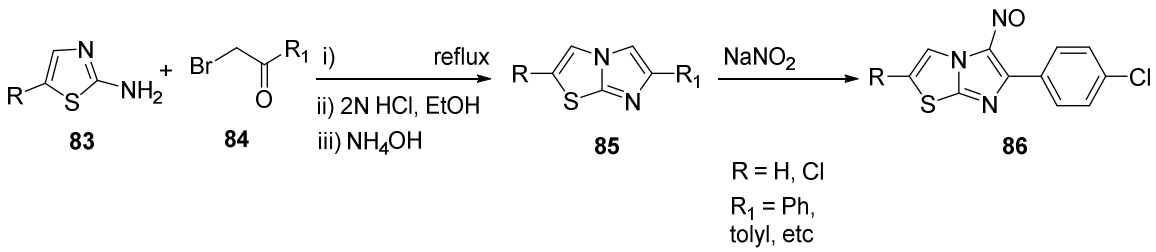
Shirude, *et. al.* reported quinolinyl pyrimidines as a novel class of *anti*-tubercular agents. The synthesis was achieved by treating guanidine hydrochloride and ethyl 3-(2-methoxyphenyl)-3-oxopropanoate with sodium hydride in DMF to obtain 2-amino-6-(2-methoxyphenyl)pyrimidin-4-ol **72**, which was refluxed in PCl_5 and $POCl_3$ to obtain 4-chloro-6-methylpyrimidin-2-amine **73**.

Substituted ketones **74** were condensed with diethyl carbonate **75** to obtain oxopropanoate **76**. This **76** was treated with 4-nitroaniline **77** to yield uncyclized product **78**, which was cyclized in Dowtherm A to get the corresponding 6-nitroquinolin-4-ol derivative **79**. This **79** was converted to 4-chloro and 4-azido derivatives **80** by reacting with phosphorous oxychloride and sodium azide respectively. The azido derivative **80** was further reduced to quinoline-4,6-diamine **81** in presence of tin chloride and ethanol-ethylacetate mixture. The diamine **81** upon treatment with **73** in presence of HCl in dioxane and NMP furnished pyrimidin-4-yl-quinoline-diamine **82** (**Scheme 1.15**).[12] The compounds were evaluated for their *anti*-tubercular activity on Mtb strain and its MIC values range from 0.81 – 3 μM.



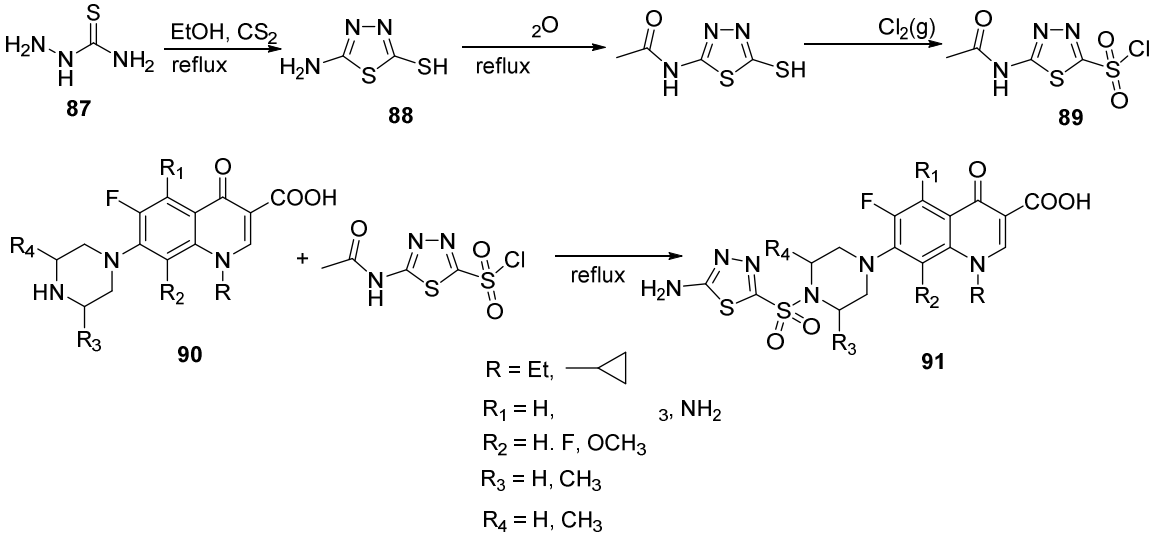
Scheme 1.15: Synthesis of quinolinyl pyrimidines

Andreani *et. al.* reported imidazo[2,1-*b*]thiazole derivatives as *anti*-tubercular agents. Briefly, thiazole **83** was treated with the 2-bromo-1-arylethanone **84** in acetone and refluxed in HCl and quenched with NH_4OH to obtain imidazothiazole **85**. This **85** was further treated with sodium nitrite in acetic acid to furnish nitrosoimidazothiazole **86** (**Scheme 1.16**).[13] The MIC values of these compounds **86** on Mtb strain were found to be in the range of 0.015 to 0.250 µg/mL.



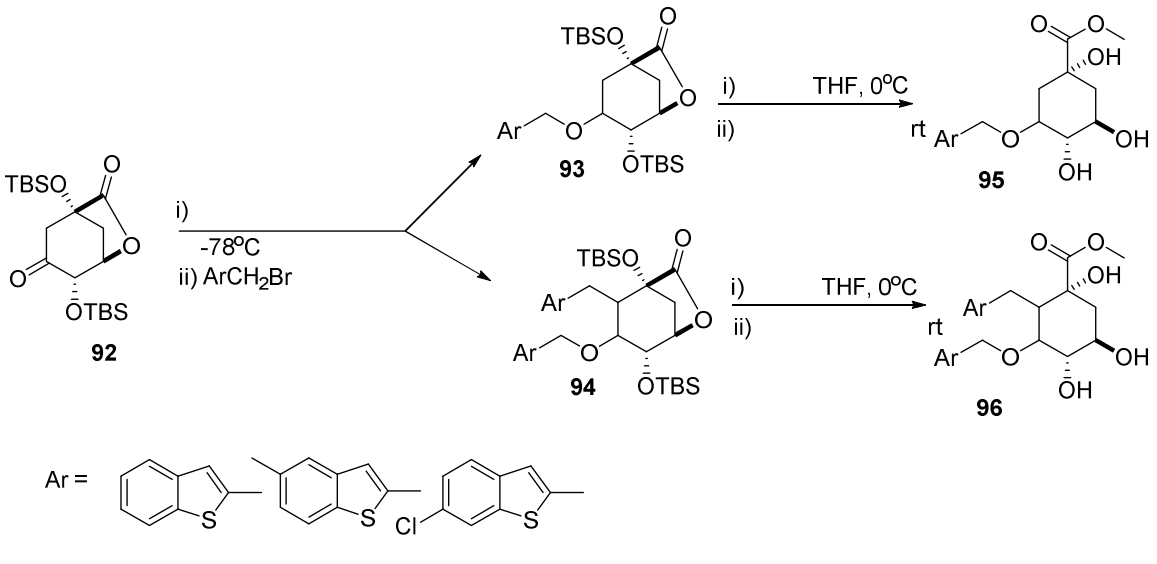
Scheme 1.16: Synthesis of imidazo[2,1-*b*]thiazole derivatives

Gadad *et. al.* reported 7-[4-(5-amino-[1,3,4]thiadiazole-2-sulfonyl)-piperazin-1-yl] fluoroquinolonic derivatives as *anti*-bacterial and *anti*-tubercular agents. The synthesis involved the formation of 2-amino-5-mercapto-1,3,4-thiadiazole **88** using the treatment of thiosemicarbazide **87** with carbon disulfide and anhydrous sodium carbonate. This thiazole **88** was acetylated and further treated with acetic acid and gaseous chlorine to obtain sulfonyl chloride **89**. This **89** was treated with 7-(1-piperazinyl) fluoroquinoline **90** in dried acetone, to furnish 7-[4-(5-acetylamino-[1,3,4]thiadiazole-2-sulfonyl)-piperazin-1-yl] fluoroquinoline **91** (**Scheme 1.17**).[14] The compounds were evaluated for *anti*-tubercular activity on H37Rv strain and thier MIC was found to be in the range of ~10 µg/mL.



Scheme 1.17: Synthesis of 7-[4-(5-amino-[1,3,4]thiadiazole-2-sulfonyl)-piperazin-1-yl] fluoroquinolonic derivatives

Gonzalez-Bello *et. al.* reported potent mycobacterium tuberculosis type II dehydroquinase inhibitors. The synthesis involved the treatment of ketone **92** with KHMDS at -78^{o}C followed by reaction with the corresponding bromide afforded a mixture of O-alkylated and dialkyl products **93** and **94** respectively, which were deprotected using TBAF in THF, followed by esterification to afford corresponding lactones **95** and **96** respectively (**Scheme 1.18**).[15] These lactones were evaluated for *anti*-tubercular acitvity on H37Rv, and the MIC values were found to be >160 µg/mL.



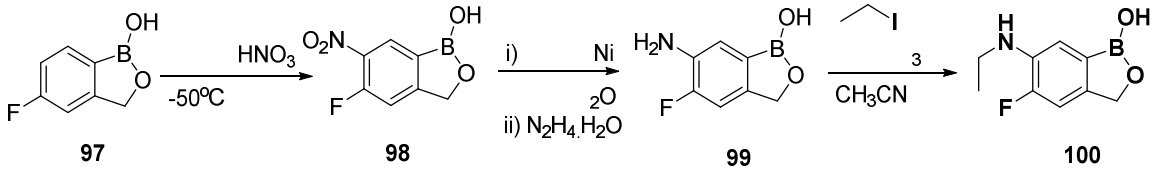
Scheme 1.18: Synthesis of dehydroquinase inhibitors

Our ongoing projects on the development of novel methodologies for the synthesis of functionalized benzoboroxoles have prompted us to evaluate benzoboroxole template towards the treatment of tuberculosis. The following schemes **1.19 – 1.20** delineate some of the medical applications of benzoboroxole small molecules.

Cusack *et. al.* reported the synthesis of *anti*-fungal benzoxaboroles. The synthesis involved the nitration of 5-fluorobenzoxaborole **97** with fuming nitric acid at -50^{o}C to obtain 6-nitro-5-fluoro-benzoxaborole **98**, which was reduced in presence of Raney Nickel in methanol-water and hydrazine monohydrate to yield 6-amino-5-fluoro-benzoxaborole **99**. Compound **99** was alkylated using ethyl iodide in presence of sodium bicarbonate in acetonitrile to furnish 6-ethylamino-5-fluoro-benzoxaborole **100** (AN3018) (**Scheme 1.19**).[16] The antifungal activity of **100** on different fungal species was found to be as follows.

C. albicans IC_{50} = 38 µM, MIC = 2 µg/mL

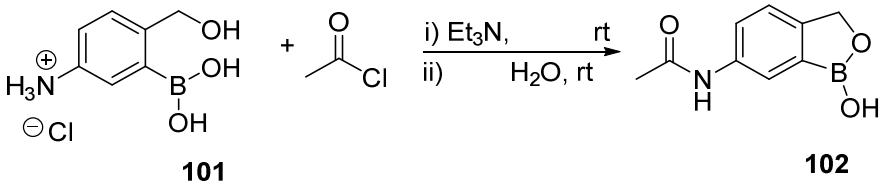
A. fumigatus IC_{50} = 19.4 µM, MIC = 64 µg/mL



97 98 99 100

Scheme 1.19: A Structural Basis for the Rational Design of Antifungal Benzoxaboroles

Plattner *et. al.* recently reported boron containing small molecules as *anti*-protozoal agents. The synthesis involved acetylation of 3-borono-4-(hydroxymethyl)benzenaminium chloride **101**, using acetyl chloride in presence of triethylamine in dichloromethane followed by acidic hydrolysis, which provided N-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)acetamide **102** (**Scheme 1.20**).[17]



Scheme 1.20: Synthesis of *N*-substituted aminobenzoxaboroles

Based on our long-standing interest[18] in the development of boron containing compounds as therapeutic agents, we undertook the project involving the synthesis of functionalized benzoboroxoles as anti-tubercular agents. Our results are reported in the succeeding chapters.

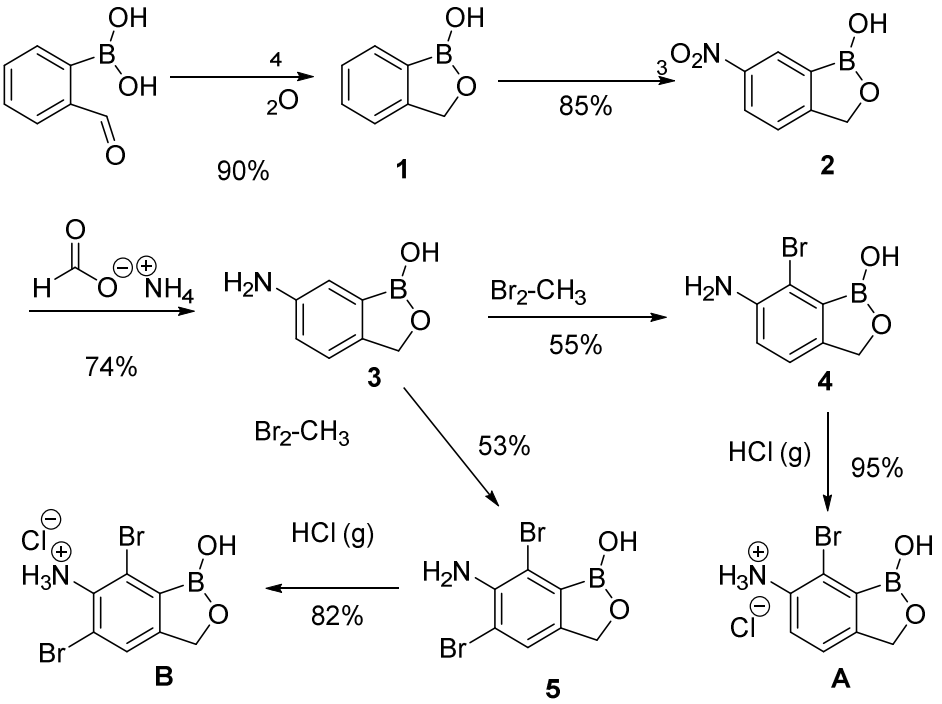
Chapter 2: Synthesis of Functionalized Aminobenzoboroxoles

Introduction

Benzoboroxoles are relatively new class of compounds which have been known for ~60 years. These molecules are highly utilized in materials and medicinal chemistry.[19,20] Owing to our interest in the synthesis of antimicrobial agents, we extended our long-interest in functionalized aminobenzoboroxoles as *anti*-tubercular agents.[21] Based on the recent evaluation of benzoboroxoles in our lab as well as in literature reports, the substitution on methylene group in benzoboroxole resulted in the complete loss of biological activity. Hence, we focused on the substitution on aromatic ring to study structure activity relationship of substituted benzoboroxoles. We utilized aminobenzoboroxole as a starting material for the functionalization of these derivatives. Aminobenzoboroxole was prepared starting from *o*-boronobenzaldehyde.[22]

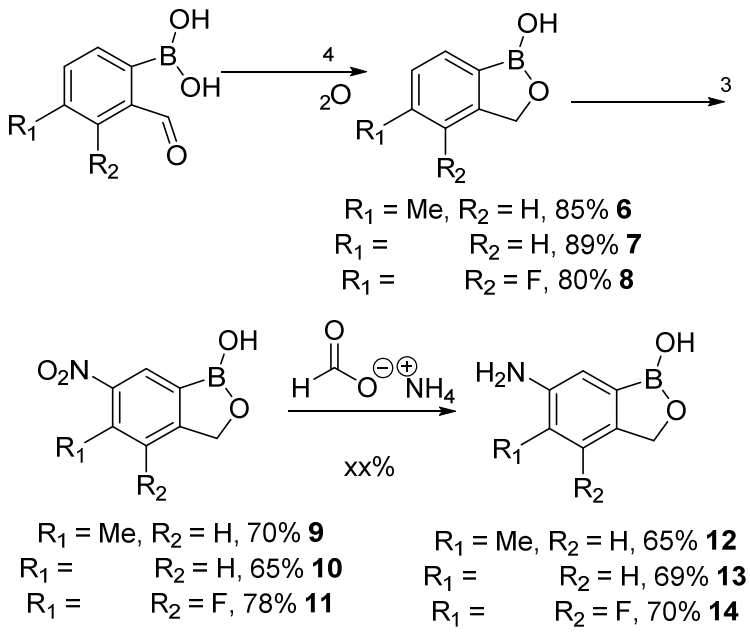
Results and Discussion

Benzoboroxole **1** was synthesized upon reaction of o-boronobenzaldehyde with sodium borohydride. Nitration of **1** with fuming nitric acid followed by reduction with Palladium-carbon in presence of ammonium formate provided aminobenzoboroxole **3**. Electrophilic aromatic bromination of **3** with bromine in acetic acid provided bromobenzoboroxole **4** in 55% yield. Hydrogen chloride gas was passed through **4** to convert it into hydrochloride salt to increase the solubility. **3** was brominated using excess bromine in presence of acetic acid to provide the dibrominated aminobenzoboroxole derivative **5**, which was further treated with HCl gas to obtain corresponding hydrochloride salt (**Scheme 2.1**).



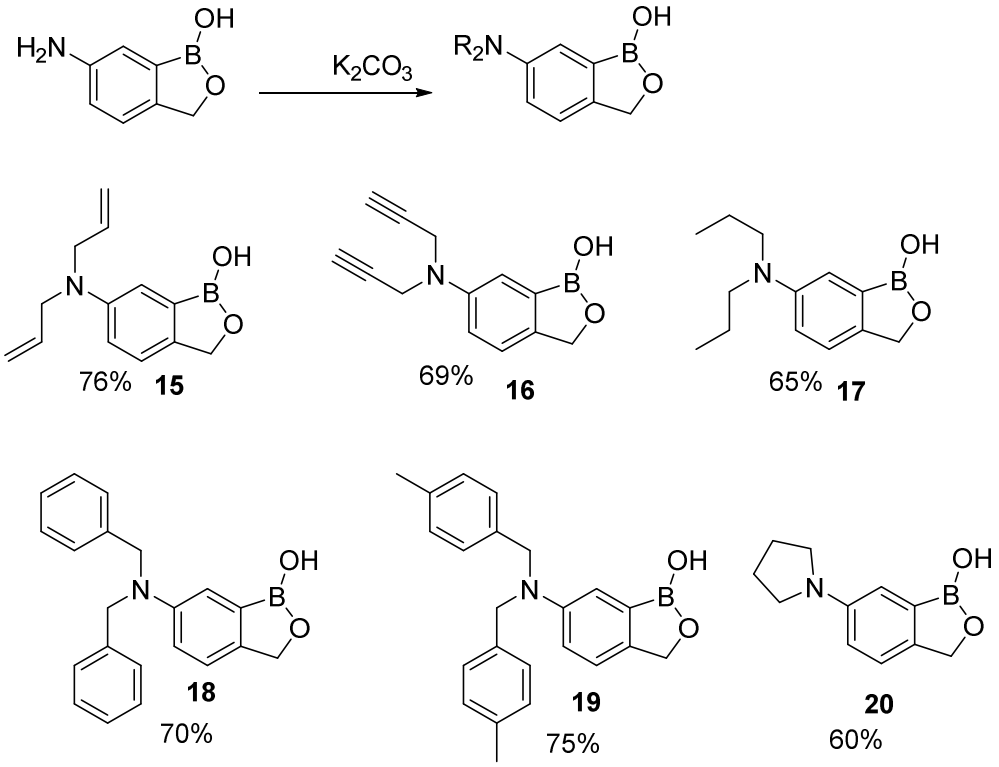
Scheme 2.1: Preparation of Functionalized Benzoboroxoles.

Alkyl and alkoxy substituted aminobenzoboroxoles were prepared upon reaction of corresponding boronobenzaldehydes with sodium borohydride, followed by nitration and reduction (**Scheme 2.2**).



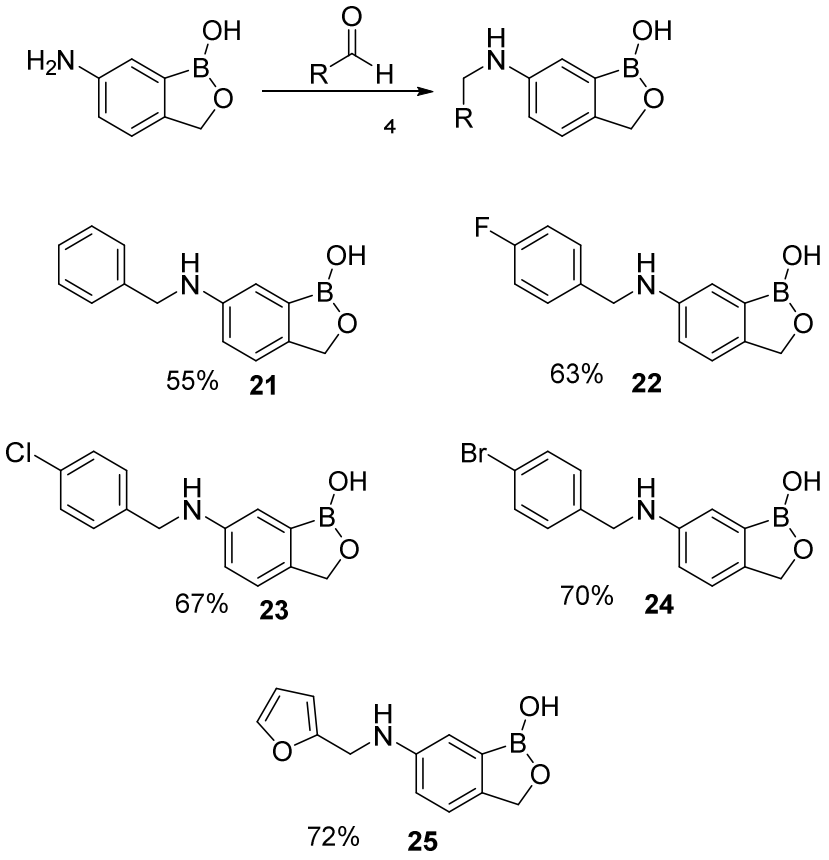
Scheme 2.2: Preparation of Functionalized Benzoboroxoles.

Dialkylaminobenzoboroxoles **15-20** were prepared via S_N2 alkylation of aminobenzoboroxole with alkyl halides in presence of a base (**Scheme 2.3**).



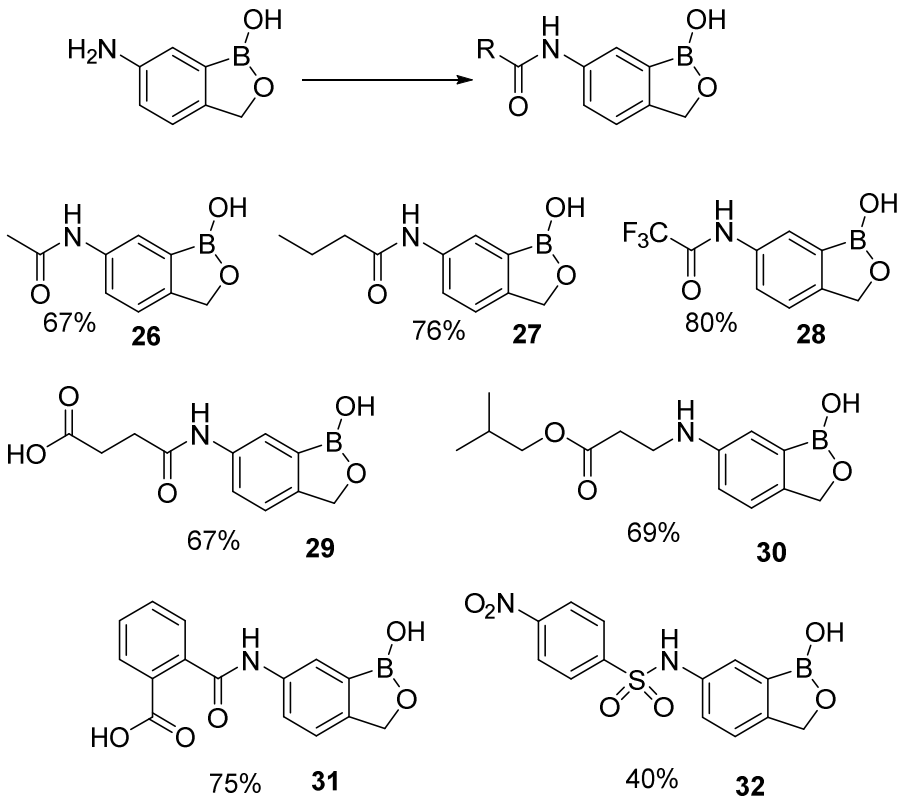
Scheme 2.3: Alkylation of Aminobenzoboroxoles

The monoarylaminobenzoboroxoles were synthesized using reductive amination. Treatment of the aminobenzoboroxole with corresponding aldehydes followed by sodium borohydride reduction gave the desired products **21**-**25** (**Scheme 2.4**).



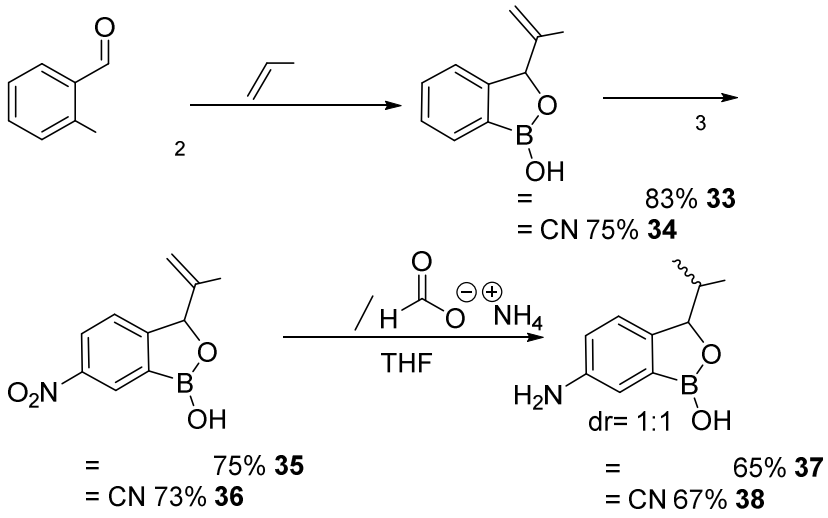
Scheme 2.4: Reductive Amination of Aminobenzoboroxoles

Amidobenzoboroxole derivatives were prepared by the treatment of corresponding anhydrides **26-32** (**Scheme 2.5**).



Scheme 2.5: Synthesis of Amidobenzoboroxoles.

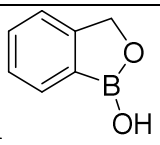
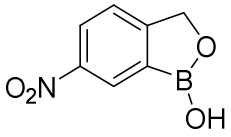
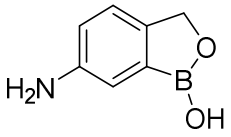
Some of the functionalized aminobenzoboroxoles **37, 38** were synthesized using Baylis-Hilman type reactions. The standard conditions were employed using o-boronobenzaldehyde with methyl acrylate and acrylonitrile in presence of the hindered catalyst DABCO resulted in compound **33** and **34** respectively. Both of these derivatives were nitrated using fuming nitric acid followed by Pd-C/NH_4HCOO reduction to yield corresponding aminobenzoboroxoles **37** and **38** (**Scheme 2.6**).

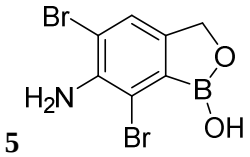
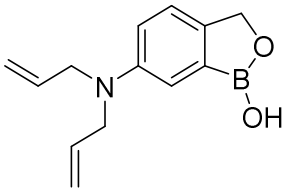
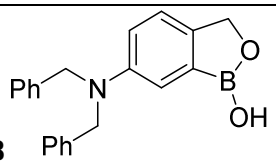
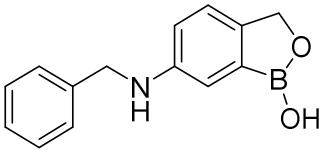
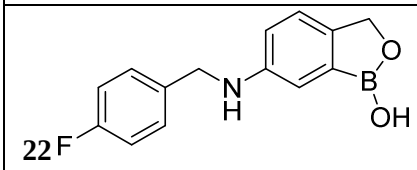
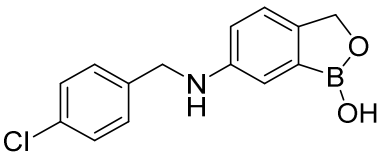
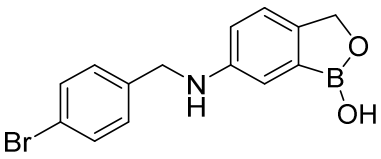


Scheme 2.6: Preparation of substituted Benzoboroxoles

To determine the specificity of the synthesized aminobenzoboroxole derivatives in microbial organisms versus cancerous cell lines, these compounds were tested for their cytotoxicity against MCF-7 breast cancer cell line using sulphorhodamine-B assay. The compounds did not exhibit any cytotoxic effects up to 50 µM concentration. Owing to their non-toxic profile, these compounds were initially tested for their activity in *Mycobacterium Smegmatis* (*M. Smeg*.) using Kirby-Bauer disk method. This strain is chosen for the initial studies due to their fast growth rate and non-pathogenic nature. Most of the compounds exhibited significant zones of inhibition. Among these compounds, two derivatives aminobenzoboroxole **3** and bromoaminobenzoxoborole **4** showed very high activity with zone of inhibition > 5 cm at 100 µM. Their individual minimum inhibitory concentration (MIC_{99}) were determined. MIC_{99} gives information about the ability of the test compounds to inhibit growth of aerobic, logarithmically growing cells.

Table 1: Zone of inhibition on *Mycobacterium Smegmatis*

Compound	Zone of inhibition at 100µM (in cm)
 **1**	3
 **2**	1.1
 **3**	5

 **5**	5.4
 **15**	1.2
 **18**	1
 **21**	3.4
 **22**	2.2
23 	2.1
24 	1.5

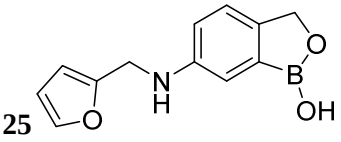
25 	3.4
Ethambutol	5
Isoniazid	4.4
Pyrazinamide	1
Rifampin	2.4

Table 2: Minimum Inhibitory Concentration

#	Compound	MIC (7H9) (µM)		MIC (GAST) (µM)	
		Week 1	Week 2	Week 1	Week 2
1	1	62.5	62.5-125	NG*	62.5
2	3	15.6	31.25	NG	15.6
3	3.HCl	15.6	31.25	NG	15.6-31.25
4	4	3.9	7.8	NG	1.9
5	A	3.9	7.8	NG	1.9
6	INH	0.7	0.7	NG	0.15

*No growth

Minimum bactericidal concentration (MBC) assay was utilized to determine whether the test compound is static or bactericidal. This assay defines the efficacy of the compound to kill aerobically growing log-phase Mtb versus inhibition of bacterial growth. A compound must have at least a 3-log kill of Mtb from the day 0 inoculum to exhibit MBC.

Table 3: Minimum Bactericidal Concentration

#	Compound	Conc.	CFU/mL	Conc.	CFU/mL
1	Untreated	-	2.3×10^5	-	-
2	DMSO	1.25 µL	3.8×10^8	50 µL	1.06×10^8
3	RIF	0.1 µM	290	1.0 µM	165
4	3	4 µM	4.59×10^5	40 µM	750
5	5	4 µM	5.51×10^4	40 µM	1475

Minimum anaerobicidal concentration (MAC) values were determined for the synthesized compounds. MAC is the minimum concentration of the compound required to cause a 1-log kill from the initial inoculum under conditions of non-replicative persistence. This assay was carried out under the conditions of dormancy to determine the potency of test compounds, where, even the most potent compounds like INH lose their activity due to the absence of metabolic activity.

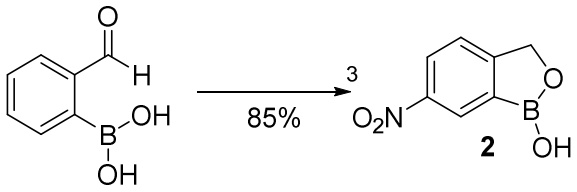
Table 4: Minimum Anaerobicidal Concentration

#	Compound	Conc.	CFU/mL	Conc.	CFU/mL
1	Untreated	-	7.8×10^6	-	-
2	DMSO		1.32×10^7	-	-
3	MTZ	100 µM	1.73×10^5	-	-
4	INH	10 µM	1.36×10^7	-	-
5	**3**	10 µM	7.13×10^6	100 µM	4.81×10^6
6	**5**	10 µM	6.55×10^6	100 µM	4.30×10^6

Conclusions

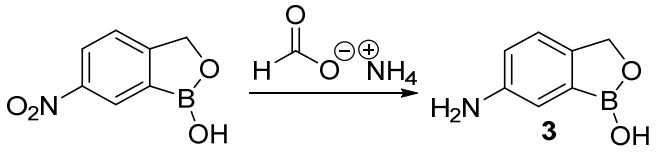
In conclusion, several aminobenzoboroxole derivatives have been prepared starting from *o*-boronobenzaldehyde. Some of the protocols employed in this project include reductive amination, alkylation, and Baylis Hillman reaction. These derivatives have been evaluated for their *anti*-tubercular activity on mycobacterium tuberculosis H37Rv using 7H9 and GAST protocols and these studies are reported in the next chapter. Among all these the synthesized compounds, aminobenzoboroxole **3** exhibited good antifungal properties based on the zone of inhibition and MIC values and have been designated as the lead compound. More structure-activity relationship should be conducted based on the lead compound to obtain more active derivative for the treatment of tuberculosis.

Chapter 3: Experimental Procedures



Representative Procedure for Nitration of Benzoboroxoles:

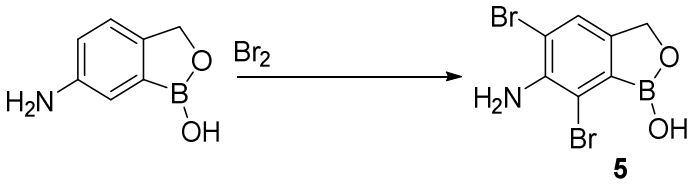
To a stirred solution of fuming nitric acid at -40^{o}C, benzoxaborole (1mmol) was added in small portions and stirred for 0.5 h. Upon completion (TLC), reaction mixture was poured into ice and stirred to obtain solid. The solid was filtered and washed with water and to obtain pure nitrobenzoboroxole **2** (85%). ^{1}H NMR (500 MHz, DMSO-d_6): 10.0 (s, 1H), 8.63 (s, 1H), 8.41 (d, *J* = 8.0 Hz, 1H), 7.65 (d, *J* = 8.0 Hz, 1H), 6.59 (s, 1H), 6.43 (s, 1H), 6.07 (s, 1H); ^{13}C NMR (125 MHz, DMSO-d_6): 159.0, 148.7, 135.8, 127.0, 126.2, 124.2, 122.5, 116.2, 80.0; CHN Analysis: (Found: C, 51.97.00%; H, 3.19%; N, 12.35%; $C_{10}H_7BN_2O_4$ requires: C, 52.22%; H, 3.07%; N, 12.18%).



Representative Procedure for Reduction of Nitrobenzoboroxoles:

To a stirred solution of nitrobenzoboroxole (1mmol) in THF, were added Pd-C and ammonium formate (5mmol) and refluxed at 60^{o}C for 2 h. Upon completion (TLC),

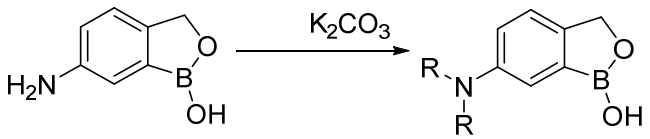
reaction mixture was filtered over celite and washed with THF. The organic layer was evaporated to obtain aminobenzoboroxole **3** (74%).



5

Representative Procedure for Dibromination of Aminobenzoboroxoles:

Bromine (2.5 mmol) was added to a solution of aminobenzoboroxole (1 mmol) in acetic acid (5 mL) and the reaction mixture was stirred for 5 h. The reaction was monitored via TLC and the reaction mixture was neutralized by saturated $NaHCO_3$ and extracted with ethyl acetate. The organic layer was dried and vacuumed. The crude product was purified via column chromatography to get pure dibromoaminobenzoboroxole **5** (53%). ^{1}H NMR (500 MHz, DMSO-d_6): 9.11 (s, 1H), 7.51 (s, 1H), 5.24 (s, 2H), 4.85 (s, 2H); ^{13}C NMR (125 MHz, DMSO-d_6): 142.3, 142.0, 125.3, 112.3, 109.9, 68.7; CHN Analysis: (Found: C, 27.12%; H, 2.05%; N, 4.34%; $C_7H_6BBr_2NO_2$ requires: C, 27.41%; H, 1.97%; N, 4.57%)

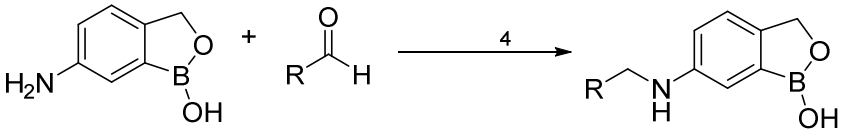


R =

Representative Procedure for Alkylation of Aminobenzoboroxoles:

Benzyl bromide (2.5 mmol) and K_2CO_3 (3.0 mmol) were was added to a solution of aminobenzoboroxole (1 mmol) in DMSO (5 mL) and stirred overnight at room

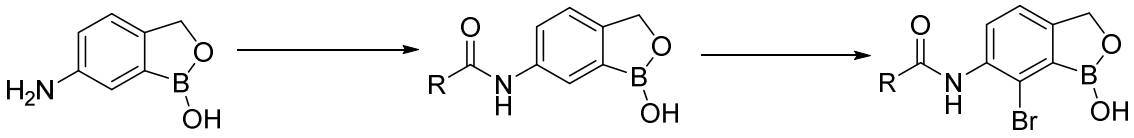
temperature. The reaction was monitored via TLC and the reaction mixture was extracted with water-ethyl acetate. The organic layer was dried and vacuumed. The crude product was purified via column chromatography to get pure dibenzylaminobenzoboroxole **18** (70%). ^{1}H NMR (500 MHz, DMSO-d_6): 8.95 (s, 1H), 7.22-7.35 (m, 10H), 7.12 (d, *J* =8.5 Hz, 1H), 7.07 (s, 1H), 6.84 (d, *J* = 8.0 Hz, 1H), 4.84 (s, 2H), 4.71 (s, 4H); ^{13}C NMR (125 MHz, DMSO-d_6): 148.0, 142.2, 139.4, 128.9, 127.2, 127.1, 122.1, 116.4, 113.6, 69.9, 54.8; CHN Analysis: (Found: C, 76.35%; H, 6.30%; N, 4.16%; $C_{21}H_{20}BNO_2$ requires: C, 76.62%; H, 6.12%; N, 4.25%).



Representative Procedure for Reductive Amination with Aminobenzoboroxoles:

Benzaldehyde (1.1 mmol) and aminobenzoboroxole (1 mmol) were stirred in toluene (5 mL) to facilitate the formation of imine. $NaBH_4$ (1.1 mmol) was added slowly to the above reaction mixture and stirred for 0.5 h. The reaction was monitored via TLC and the reaction mixture was neutralized by 3M HCl and extracted with ethyl acetate. The organic layer was dried and vacuumed. The crude product was purified via column chromatography to get pure N-benzylaminobenzoboroxole (55%). ^{1}H NMR (500 MHz, DMSO-d_6): 8.91 (s, 1H), 7.21-7.37 (m, 5H), 7.07 (d, *J* = 8.0 Hz, 1H), 6.87 (d, *J* = 2.5 Hz, 1H), 6.77 (dd, *J* = 2.5, 8.0 Hz, 1H), 6.23 (bs, 1H), 4.82 (s, 2H), 4.28 (s 2H); ^{13}C NMR (125 MHz, DMSO-d_6): 148.2, 142.0, 140.8, 128.7, 127.6, 127.0, 121.9, 117.0, 112.7,

70.0, 47.1; CHN Analysis: (Found: C, 70.56%; H, 5.78%; N, 5.62%; $C_{14}H_{14}BNO_2$ requires: C, 70.33%; H, 5.90%; N, 5.86%).



Representative Procedure for the Preparation of Amidobenzoboroxoles:

Acetic anhydride (1.2 mmol) was added dropwise to a solution of aminobenzoboroxole (1 mmol) in dioxane (5 mL) and the reaction mixture was stirred overnight at room temperature. The reaction was monitored via TLC and the reaction mixture was neutralized by 3M HCl and extracted with ethyl acetate. The organic layer was dried and vacuumed. The crude product was purified via column chromatography to get pure amidobenzoboroxole **26** (67%). ^{1}H NMR (500 MHz, DMSO-d_6): 9.96 (s, 1H), 9.21 (s, 1H), 8.01 (s, 1H), 7.61 (d, *J* = 8.0 Hz, 1H), 7.33 (d, *J* = 8.5 Hz, 1H), 4.94 (s, 2H), 2.01 (s, 3H); ^{13}C NMR (125 MHz, DMSO-d_6): 168.7, 148.9, 138.6, 122.7, 121.9, 121.3, 70.1, 24.4; CHN Analysis: (Found: C, 56.38%; H, 5.15%; N, 7.18%; $C_9H_{10}BNO_3$ requires: C, 56.60%; H, 5.28%; N, 7.33%)**.**

Cytotoxicity Assay:

The general cytotoxicity of these compounds were determined using sulphorhodamine-B assay. MCF-7 cells were cultured in IMEM supplemented with 10% FBS and 1% penicillin/streptomycin. The cells were seeded at a concentration of 5 x 10^4 cells/mL in 48-well plates and incubated for 18-24 hours at 37^{o}C in 5% CO_2 atmosphere. The growth media was removed and test compounds at desired concentration (100, 50 and 10 µM) were added to the wells and incubated for 72 hours. Taxol was used as positive control, whereas growth media and DMSO were used as negative controls. The growth media was removed and the wells were washed with 1% PBS and dried. 100µL of 0.5% SRB in 1% acetic acid was added to the wells and incubated at 37^{o}C for 45 minutes. The cells were washed five times with 1% acetic acid and dried. To these wells, 400µL of 10mM tris base was added to dissolve the cell protein and absorbance was recorded at 540nm to determine the percentage survival of cells and determine the cytotoxicity of the synthesized compounds. All the compounds were tested in triplicates.

Bacterial strains, media and growth conditions:

M. tuberculosis (Mtb) H37Rv was grown in Middlebrook 7H9 broth medium (Difco) supplemented with 0.2% (v/v) glycerol, 0.05% (v/v) tween 80 and ADC (bovine serum albumin(BSA); 5g/l, dextrose; 2g/l and NaCl; 0.81g/l). When desired Mtb was cultured in glycerol-alanine-salts-tween (GAST) medium consisting of 0.3g/l bacto-casitone, 4g/l dibasic potassium phosphate, 2g/l citric acid, 1g/l L-alanine, 1.2g/l magnesium chloride hexahydrate, 0.6g/l potassium sulfate, 2g/l ammonium chloride, 1% glycerol (v/v) and

0.05% tween 80 (v/v), pH adjusted to 6.6 using NaOH. For generating non-replicating persistent cells Mtb was cultured in Middlebrook Dubos medium (Difco) supplemented with 5g/l BSA, 7.5g/l glucose and 0.81g/l NaCl. For determination of viable cell counts, Mtb cells were plated on Middlebrook 7H11 plates supplemented with 0.5% glycerol (v/v) and OADC (oleic acid; 0.06%, NaOH 4mg/l, BSA; 50g/l, dextrose; 2g/l and NaCl; 0.81g/l). All cultures were incubated at 37°C.

Minimum inhibitory concentration (MIC_{99}):

MIC_{99} was determined in 7H9 as well as GAST medium by micro-dilution broth method as previously described (Wong 2010). Briefly, Mtb was grown to an optical density at 650nm (OD_{650}) of 0.2-0.3 in 7H9 or GAST medium. Cells were diluted to final OD_{650} of 0.0002 (1:1000 of parent culture) in desired medium. Ten-fold serial dilution of the test compounds were made in desired medium in triplicate rows of a 96-well plate. The above dilution of cells was then added to all the wells of the 96-well plate. The assay was performed in duplicates with each compound dilution set-up in triplicate rows per plate. The plates were incubated at 37°C and observations were noted on days 7, and 14. MIC_{99} was noted as the lowest concentration of test compound that inhibited visible growth after 14 days of incubation. Isoniazid (INH), ethambutol (EMB), rifampicin (RIF), moxifloxacin (MOXI), and streptomycin (STM) were used as control for 7H9 MICs while INH was used as a control for GAST MICs.

Minimum bactericidal concentration (MBC):

Mtb was grown to OD_{650}=0.2-0.3 in 7H9 medium. Cells were then diluted to OD_{650}=0.002 (1:100 of parent culture). For enumeration of viable numbers on day 0,

serial dilutions of the diluted cultures were plated in duplicate on 7H11 plates (supplemented with OADC). One-ml aliquots of the diluted culture were then dispensed in duplicate wells of 24-well plates. Treatments with chosen test compounds (#4 and #5) were carried out at 1- (4µM) and 10-(40µM) times the MIC_{99} for 7 days. Rifampicin (0.1 and 10µM) was used at a positive control and DMSO was used as negative control. Serial dilutions were then plated on 7H11 medium (supplemented with OADC). Colonies were counted after 3wk incubation at 37°C and CFU/ml were determined. The data show that compounds #4 and #5 had MBC around 40µM while MBC for RIF is 0.1µM.

Minimum anaerobicidal concentration (MAC):

Non-replicating persistent (NRP) cells of Mtb were obtained by culturing the cells in Wayne model system (Wayne 1996). Briefly, Mtb was cultured in Dubos medium to an OD_{650}=0.2-0.3. Cells were then diluted to OD_{650}=0.005 and dispensed in glass bottles leaving a head-space ratio of 0.5.The bottles were sealed with wax and the bottle was placed on a magnetic stir plate at 37°C. The cells were allowed to stir for 21 days at which point the culture reached NRP2 state. The bottle was removed from the stir plate and opened in an anaerobic chamber. For determination of day 0 cell numbers serial dilutions of the culture were plated on 7H11 plates (supplemented with OADC). One-ml cells were dispensed in duplicate wells of 24-well plates and treated with the desired concentrations (10 and 100µM) of the chosen test compounds (#4 and #5) for 7 days in the anaerobic chamber at 37°C. After 7 days the plates were removed from the anaerobic chamber and serial dilutions were then made and plated on 7H11 plates (supplemented with OADC). Metronidazole (100µM) was used as positive control and DMSO and INH

(10µM) were used as negative controls. Viable numbers were counted after 4wks of incubation at 37°C and CFU/ml was determined.

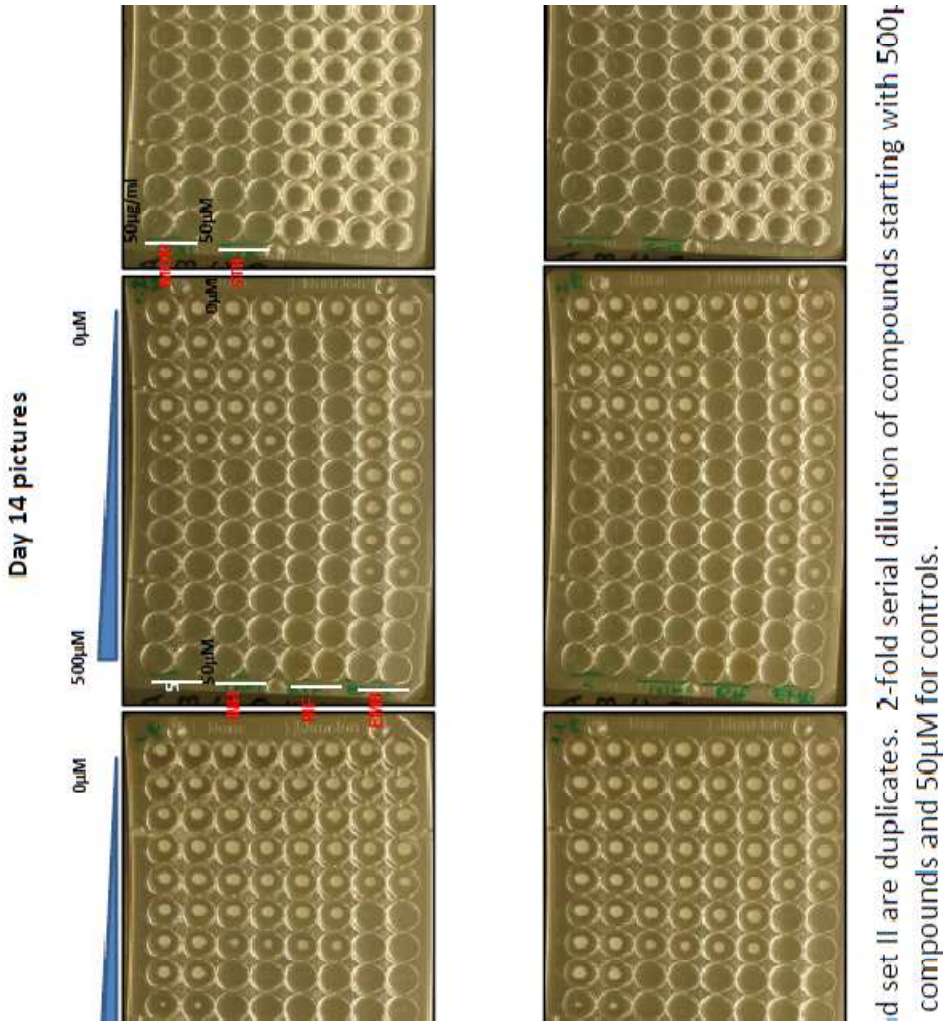


Figure 1: MIC evaluation on Mtb H37Rv

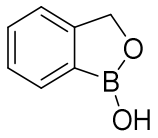
Table 5: MIC values (µM) of the compounds on Mtb H37Rv

Sl. No	Compound	MIC (µM)-Set 1		MIC (µM)-Set 2	
		week 1	week 2	week 1	week 2
1	1	62.5	62.5-12.5	62.5	62.5-125
2	3	15.6	31.25	15.6	31.25
3	HCl salt of 3	15.6	31.25	15.6	31.25
4	5	3.9	7.8	3.9	7.8
5	A	3.9	7.8	3.9	7.8

6	Isoniazid	0.7	0.7	0.7	0.7
7	Rifampin	≤0.045	≤0.046	≤0.047	≤0.048
8	Ethambutol	6.25-12.5	6.25-12.6	6.25-12.7	6.25-12.8
9	Moxifloxacin	0.1 µg/ml	0.1 µg/ml	0.1 µg/ml	0.1 µgml
10	Streptomycin	0.1	0.1	0.1	0.1

Maximum concentration tested: 1 to 5 – 500 µM; 6 to 10 – 50 µM

Spectral Characterization



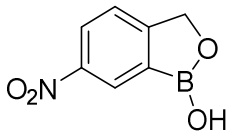
1

Benzo[c][1,2]oxaborol-1(3H)-ol 1:

^{1}H NMR (500MHz, DMSO-

d_6) δ 9.23 (s, 1H), 7.74 (d, *J* = 7.5 Hz, 1H), 7.47 (t, *J* = 7.3 Hz, 1H), 7.41 (d, *J* = 7.5 Hz, 1H), 7.34 (t, *J* = 7.3 Hz, 1H), 4.99 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 154.3, 131.0, 130.9, 127.3, 121.8, 70.4

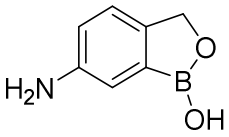


2

6-nitrobenzo[c][1,2]oxaborol-1(3H)-ol 2:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.60 (s, 1H), 8.60 (s,1H), 8.35(d, *J* = 8.5 Hz, 1H), 7.71 (d, *J* = 8.5 Hz, 1H), 5.14 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 161.1, 147.7, 126.1, 126.0, 123.6, 70.6

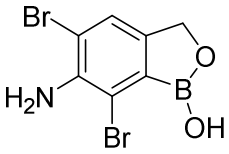


3

6-aminobenzo[c][1,2]oxaborol-1(3H)-ol 2:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.95 (s, 1H), 7.04 (d, *J* = 8 Hz, 1H), 6.90 (s, 1H), 6.72 (dd, *J* = 1.5 Hz, 8 Hz, 1H), 4.97 (br, s, 2H), 4.82 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 148.0, 141.9, 121.9, 118.1, 115.1, 70.1

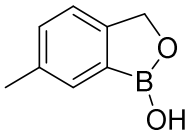


5

6-amino-5,7-dibromobenzo[c][1,2]oxaborol-1(3H)-ol 5:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.11 (s, 1H), 7.51 (s, 1H), 5.24 (s,2H), 4.85(s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 144.3, 142.0, 125.3, 122.0, 112.3, 110.0, 68.8

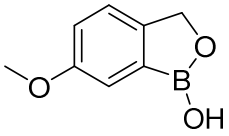


6

6-methylbenzo[c][1,2]oxaborol-1(3H)-ol 6:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.15 (s, 1H), 7.53 (s, 1H), 7.29 (s, 2H), 4.94 (s,2H), 2.35(s, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 151.6, 136.2, 132.0, 131.1, 121.6, 70.2, 21.4

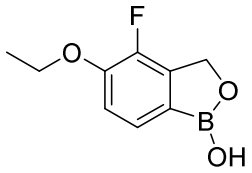


7

6-methoxybenzo[c][1,2]oxaborol-1(3H)-ol 7:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.00 (br, s, 1H), 7.3 (d, *J* = 8 Hz, 1H), 6.97 (s, 1H), 6.91 (dd, *J* = 1.5 Hz, 8 Hz, 1H), 4.93 (s,2H), 3.79 (s, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 162.2, 156.9, 132.1, 114.6, 106.2, 70.1, 55.6

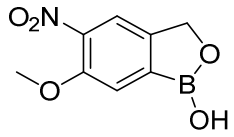


8

5-ethoxy-4-fluorobenzo[c][1,2]oxaborol-1(3H)-ol 8:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.29, (s, 1H), 7.48 (d, *J* = 8 Hz, 1H), 7.18 (t, *J* = 7.25, 1H), 5.03 (s, 2H), 4.16 (q, *J* = *7* Hz, 2H), 1.37 (t, *J* = *6.5* Hz, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 148.6, 147.2, 145.3, 140.5, 127.6, 115.3, 66.8, 65.1, 15.1

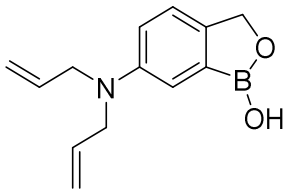


9

6-methoxy-5-nitrobenzo[c][1,2]oxaborol-1(3H)-ol 9:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.80 (s, 1H), 7.70 (s, 1H), 6.14 (s, 2H), 4.23 (s, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 156.2, 136.0, 123.4, 114.9, 70.4, 57.5

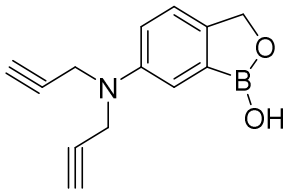


15

6-(diallylamino)benzo[c][1,2]oxaborol-1(3H)-ol 15:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.97, (s, 1H), 7.17 (d, *J* = 8Hz, 1H), 7.03 (s, 1H), 6.84 (d, *J* = 8.5 Hz, 1H), 5.90-5.84 (m, 2H), 5.18-5.12 (m, 4H), 4.87 (s, 2H), 3.94-3.91 (m, 4H).

^{13}C NMR (125 MHz, DMSO-d_6) δ 147.9, 141.9, 134.9, 128.6, 122.0, 116.3, 113.5, 70.0, 53.1

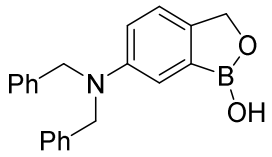


16

6-(di(prop-2-yn-1-yl)amino)benzo[c][1,2]oxaborol-1(3H)-ol 16:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.08 (s, 1H), 7.30 (m, 2H), 7.12 (dd, *J* = 2.5, 8.5 Hz, 2H), 4.12 (s, 2H), 4.14 (s, 4H), 3.16 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 147.1, 145.1, 122.1, 119.7, 116.9, 80.3, 75.6, 70.0, 40.8

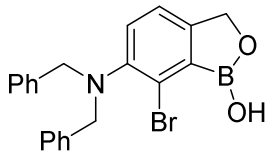


18

6-(dibenzylamino)benzo[c][1,2]oxaborol-1(3H)-ol 18:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.65 (s, 1H), 7.35-7.23 (m, 10H), 7.13 (d, J = 8.5 Hz, 1H), 7.07 (s, 1H) 6.84 (d, *J = 8* Hz, 1H) 4.84, (s, 2H), 4.71 (s, 4H)

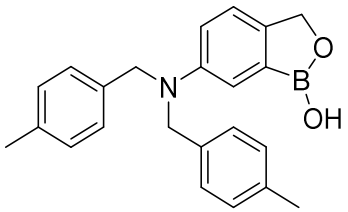
^{13}C NMR (125 MHz, DMSO-d_6) δ 148.1, 142.2, 139.4, 129.0, 127.2, 127.1, 122.1, 116.5, 113.6, 70.0, 54.8



7-bromo-6-(dibenzylamino)benzo[c][1,2]oxaborol-1(3H)-ol

^{1}H NMR (500MHz, DMSO-d_6) δ 7.39 (d, *J = 7* Hz, 4H), 7.30-7.36 (m, 5H), 7.21-7.18 (m, 3H), 4.88 (s, 2H), 4.15 (s, 4H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 151.4, 147.3, 138.6, 128.8, 128.7, 128.5, 128.0, 127.4, 127.3, 124.6, 121.2, 69.1, 57.1, 45.0

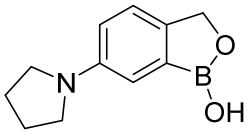


19

6-(bis(4-methylbenzyl)amino)benzo[c][1,2]oxaborol-1(3H)-ol 19:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.97 (s, 1H), 7.21-7.07 (m, 10H), 6.83 (dd, *J* = 2.25 Hz, 8.25, 1H), 4.85 (s, 2H), 4.62 (s, 4H), 2.28 (s, 6H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 148.2, 142.2, 136.3, 136.2, 129.5, 129.1, 127.1, 127.0, 122.1, 116.6, 113.8, 70.0, 63.3, 54.6, 21.2

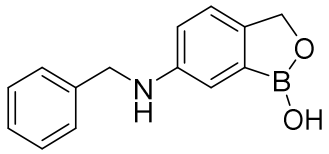


20

6-**(pyrrolidin-1-yl)benzo[c][1,2]oxaborol-1(3H)-ol 20:**

^{1}H NMR (500MHz, DMSO-d_6) δ 8.94(s, 1H), 7.18 (d, J = 8.5 Hz, 1H), 6.86 (s, 1H), 6.69(d, J = 8.5 Hz, 1H) 4.87, (s, 2H), 3.22(t, *J* = 6, 4H), 1.97-1.95 (m, 4H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 147.6, 141.1, 122.0, 115.7, 112.7, 70.0, 48.0, 25.4

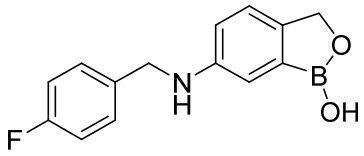


21

6-(benzylamino)benzo[c][1,2]oxaborol-1(3H)-ol 21:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.91 (s, 1H), 7.37-7.30 (m, 4H), 7.22 (t, *J* = **7 Hz,** 1H), 7.08 (d, *J* = 8 Hz, 1H), 6.87 (s, 1H), 6.76 (dd, *J* = 2.5 Hz, 8.5 Hz, 1H), 6.23 (br, s, 1H), 4.82 (s, 2H), 4.28 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 148.3, 142.0, 140.8, 128.7, 127.6, 127.0, 121.9, 117.0, 112.7, 70.0, 47.1

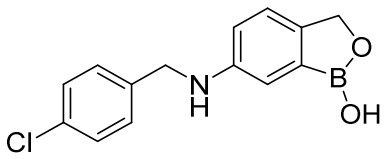


22

6-((4-fluorobenzyl)amino)benzo[c][1,2]oxaborol-1(3H)-ol 22:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.91 (s, 1H), 7.39 (t, *J* = 8 Hz, 2H), 7.14 (t, *J* = 9 Hz, 3H), 7.08 (d, J = 8.5 Hz, 1H), 6.86 (s, 1H), 6.76 (d, J = 8.5 Hz, 1H), 6.24 (t, *J* = 6 Hz, 2H), 4.82 (s, 2H), 4.27 (d, *J* = *6* Hz, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 162.5, 160.6, 148.1, 142.1, 136.9, 129.4, 122.0, 117.1, 115.5, 112.8, 70.0, 46.4

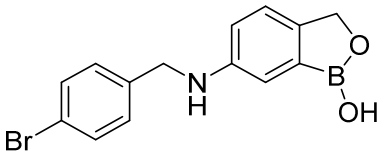


23

6-((4-chlorobenzyl)amino)benzo[c][1,2]oxaborol-1(3H)-ol 23:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.92 (s, 1H), 7.37 (s, 4H), 7.07 (d, *J* = *8* Hz, 1H), 6.84 (s, 1H), 6.74 (dd, *J* = 2.5 Hz, 8.5, 1H), 6.28 (t, *J* = 6.25, 1H), 4.82 (s, 2H), 4.28 (d, *J* = *6* Hz, 4H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 167.8, 148.0, 142.2, 140.0, 131.5, 129.4, 128.7, 122.0, 117.0, 112.8, 70.0, 46.4

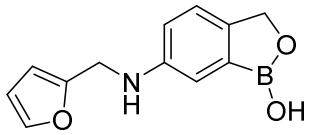


24

6-((4-bromobenzyl)amino)benzo[c][1,2]oxaborol-1(3H)-ol 24:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.92 (s, 1H), 7.51 (d, *J = 8* Hz, 2H), 7.32 (d, *J = 8* Hz, 2H),7.06 (d, J = 8.5 Hz, 1H), 6.83 (s, 1H), 6.74 (dd, *J* = 2 Hz, 8.5, 1H), 6.28 (br, s 1H), 4.82 (s, 2H), 4.26 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 148.0, 142.2, 140.4, 131.6, 129.8, 122.0, 119.9, 117.1, 112.8, 105.0, 70.0, 46.5

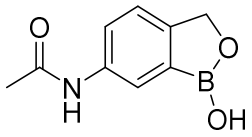


25

6-((furan-2-ylmethyl)amino)benzo[c][1,2]oxaborol-1(3H)-ol 25:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.94 (s, 1H), 7.57 (d, *J* = 1.5 Hz, 1H), 7.10 (d, J = 8.5 Hz, 1H), 6.95 (s, 1H), 6.81 (dd, *J* = 2.5 Hz, 8.5, 1H), 6.38 (dd, *J* = 2 Hz, 3, 1H), 6.28 (dd, *J* = 0.5, 3, 1H), 6.06 (t, *J* = 6.25, 1H),4.84 (s, 2H), 4.25 (d, *J = 6* Hz, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 153.9, 147.9, 142.3, 121.9, 117.1, 112.9, 110.8, 107.3, 70.0, 31.2

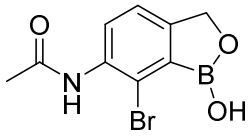


26

N-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)acetamide 26:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.97 (s, 1H), 9.24 (s, 1H), 7.99 (s, 1H), 7.60 (dd, *J* = 2 Hz, 8.5, 1H), 7.33 (d, *J* = 8.0, 1H), 4.94 (s, 2H), 2.06 (s, 3H)

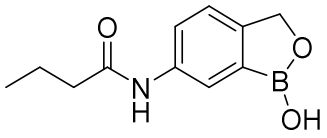
^{13}C NMR (125 MHz, DMSO-d_6) δ 168.8, 149.0, 138.6, 122.7, 122.0, 121.3, 70.1, 24.4



N-(7-bromo-1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)acetamide

^{1}H NMR (500MHz, DMSO-d_6) δ 9.47 (br, s, 1H), 9.12 (s, 1H), 7.63 (d, *J = 8* Hz, 1H), 7.38 (d, *J* = 8.0, 1H), 4.98 (s, 2H), 2.09 (s, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 169.0, 153.0, 135.8, 130.7, 121.3, 69.3, 23.6

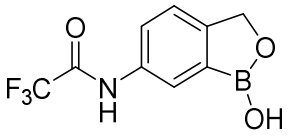


27

N-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)butyramide

^{1}H NMR (500MHz, DMSO-d_6) δ 9.89 (s, 1H), 9.20 (s, 1H), 8.02 (s, 1H), 7.63 (d, *J = 8* Hz, 1H), 7.33 (d, *J = 8* Hz, 1H), 4.94 (s, 2H), 2.31 (t, *J* = 7.25, 2H), (q, *J* = 7.5, 2H), 0.94 (t, *J* = 7.25, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 171.5, 148.9, 138.6, 122.8, 121.9, 121.5, 70.1, 19.1, 14.1

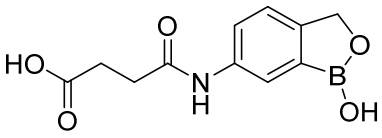


28

2,2,2-trifluoro-N-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)acetamide 28:

^{1}H NMR (500MHz, DMSO-d_6) δ 11.30 (br, s, 1H), 9.32 (s, 1H), 8.07 (s, 1H), 7.68 (d, J = 8.5 Hz, 1H), 7.46 (d, J = 8.5 Hz, 1H), 4.99 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 151.6, 135.5, 124.6, 123.6, 122.4, 105.0, 70.2

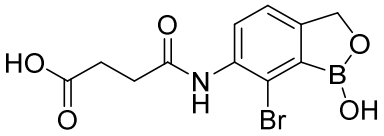


29

4-((1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)amino)-4-oxobutanoic acid 29:

^{1}H NMR (500MHz, DMSO-d_6) δ 12.1 (br, s, 1H), 9.99 (s, 1H), 9.20 (s, 1H), 8.01 (s, 1H), 7.61 (s, 1H), 7.33 (d, *J* = 4.5 Hz, 1H), 4.94 (s, 2H), 2.55 (t, *J* = 16 Hz, 4H)

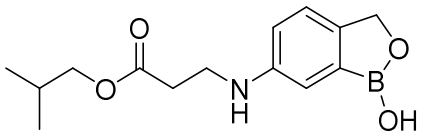
^{13}C NMR (125 MHz, DMSO-d_6) δ 174.35, 170.46, 148.88, 138.59, 122.60, 121.93, 121.32, 70.15, 31.47, 29.34



4-((7-bromo-1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)amino)-4-oxobutanoic acid

^{1}H NMR (500MHz, DMSO-d_6) δ 9.11 (s, 1H), 7.51 (s, 1H), 7.34 (s, 1H), 5.04(s,2H), 2.87,(s, 4H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 176.7, 157.1, 133.3, 131.7, 125.3, 122.2, 69.5, 29.1

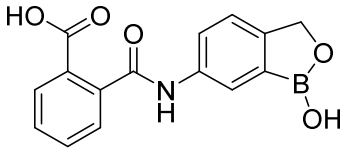


30

isobutyl-3-((1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)amino)propanoate 30:

^{1}H NMR (500MHz, DMSO-d_6) δ 8.95(s, 1H), 7.12 (d, *J = 8* Hz, 1H), 6.91(s, 1H), 6.75(d, *J = 8* Hz, 1H), 5.58(br s, 1H), 4.85(s, 2H), 3.85(d, *J = 6.5* Hz, 2H), 3.34 (t, *J* = 19 Hz, 4H), 2.62(t, *J* = 6.25, 2H), 1.912-1.86(m, 1H), 0.89(d, *J = 6* Hz, 6H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 172.2, 148.1, 142.2, 122.0, 117.1, 112.5, 105.0, 70.2, 70.1, 34.2, 27.7, 19.4

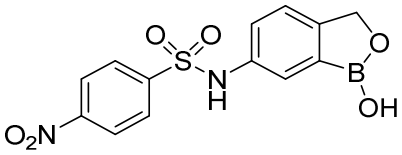


31

2-((1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)carbamoyl)benzoic acid 31:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.36 (s, 1H), 8.00 (dd, *J = 3,* 5.5 Hz, 2H), 7.94 (dd, *J = 3 Hz* 5.5, 2H), 7.78 (s, 1H), 7.59-7.55 (m, 2H), 5.08 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 167.67, 154.09, 135.18, 132.09, 131.16, 130.27, 129.83, 123.91, 122.46, 70.38

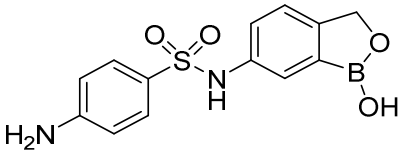


32

N-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)-4-nitrobenzene sulfonamide 32:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.34 (s, 1H), 7.97 (dd, *J* = 3.25 Hz, 5.25, 2H), 7.93-7.90 (m, 2H), 7.76 (s, 1H), 7.57-7.52 (m, 2H), 5.06 (s, 2H)

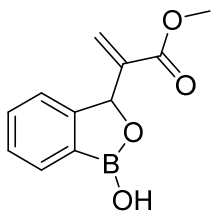
^{13}C NMR (125 MHz, DMSO-d_6) δ 167.7, 154.1, 135.2, 132.1, 131.1, 130.3, 129.8, 123.9, 122.4, 70.4



4-amino-N-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-6-yl)benzene sulfonamide

^{1}H NMR (500MHz, DMSO-d_6) δ 9.34 (s, 1H), 7.98 (dd, *J = 3*, 5.5 *Hz*, 2H), 7.91 (dd, *J* = 3.25 Hz, 6.25, 2H), 7.76 (s, 1H), 7.55 (br, s, 1H), 7.54 (d, *J* = 2 Hz, 1H), 5.06 (s, 2H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 167.7, 154.1, 135.2, 132.1, 131.1, 129.8, 123.9, 122.4, 70.4

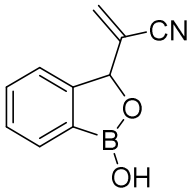


33

Methyl 2-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-3-yl)acrylate 33:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.39 (s, 1H), 7.75 (d, *J* = 7.5 Hz, 1H), 7.46 (t, *J* = 7.5 Hz, 1H), 7.37 (t, *J* = *7* Hz, 1H), 7.30 (d, *J* = 7.5, 1H), 6.20 (s, 1H), 5.98 (s, 1H), 5.84 (s, 1H), 3.72 (s, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 166.2, 155.6, 140.8, 131.5, 131.3, 128.1, 126.6, 122.3, 79.3, 52.6

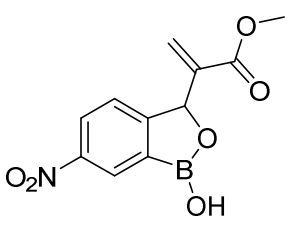


34

2-(1-hydroxy-1,3-dihydrobenzo[c][1,2]oxaborol-3-yl)acrylonitrile 34:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.63 (s, 1H), 7.80 (d, *J* = *7* Hz, 1H), 7.56 (t, *J* = *7* Hz, 1H), 7.46 (t, *J* = *7* Hz, 1H), 7.35 (d, *J* = *8* Hz, 1H), 6.49 (s, 1H), 6.32 (s, 1H), 5.87 (s, 1H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 153.0, 134.3, 131.8, 131.2, 128.8, 124.0, 122.4, 116.5, 80.2



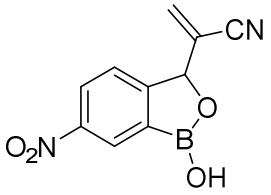
66

35

methyl 2-(1-hydroxy-6-nitro-1,3-dihydrobenzo[c][1,2]oxaborol-3-yl)acrylate 35:

^{1}H NMR (500MHz, DMSO-d_6) δ 9.79 (br, s, 1H), 8.58 (s, 1H), 8.31 (dd, *J* = 2.25 Hz, 8.25 Hz, 1H), 7.55 (d, J = 8.5 Hz, 1H), 6.30 (s, 1H), 6.09 (s, 1H), 6.03 (s, 1H), 3.67 (s, 3H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 165.4, 161.6, 148.0, 138.9, 128.7, 126.5, 125.9, 123.5, 79.7, 52.4



36

2-(1-hydroxy-6-nitro-1,3-dihydrobenzo[c][1,2]oxaborol-3-yl)acrylonitrile 36:

^{1}H NMR (500MHz, DMSO-d_6) δ 10.04 (s, 1H), 8.62 (s, 1H), 8.41 (d, J = 8.5 Hz, 1H), 7.64 (d, J = 8.5 Hz, 1H), 6.59 (s, 1H), 6.43 (s, 1H), 6.07 (s, 1H)

^{13}C NMR (125 MHz, DMSO-d_6) δ 159.0, 148.7, 135.8, 127.0, 126.2, 124.2, 122.5, 116.2, 80.0

References

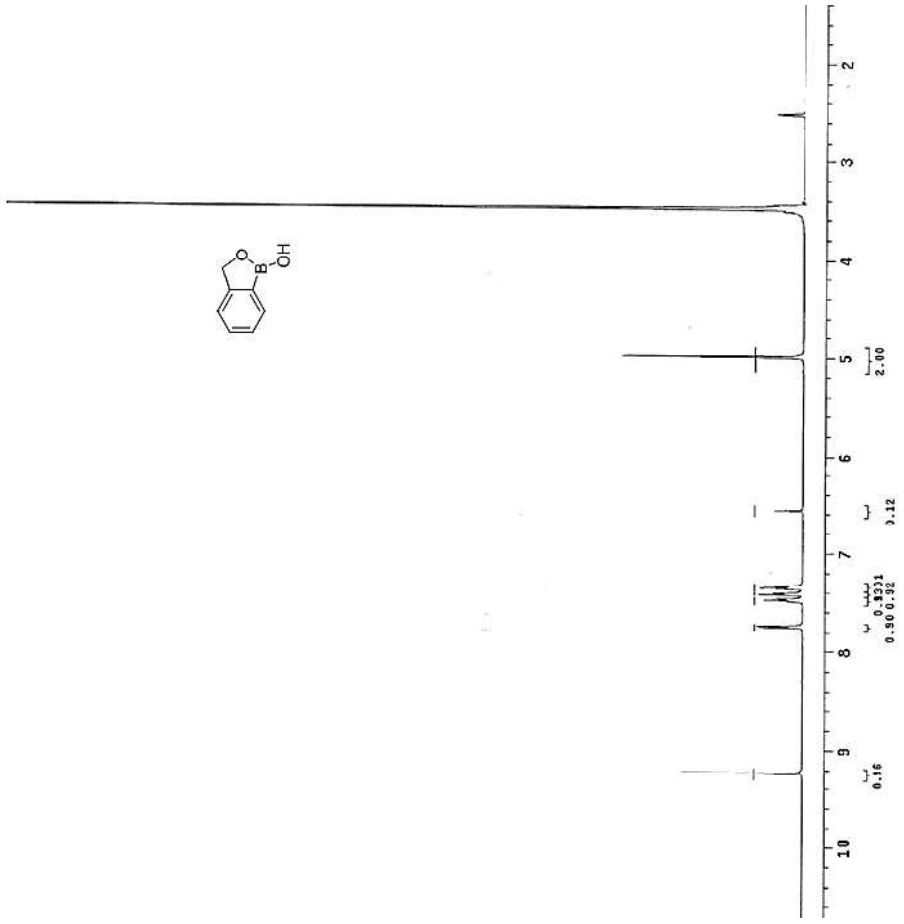
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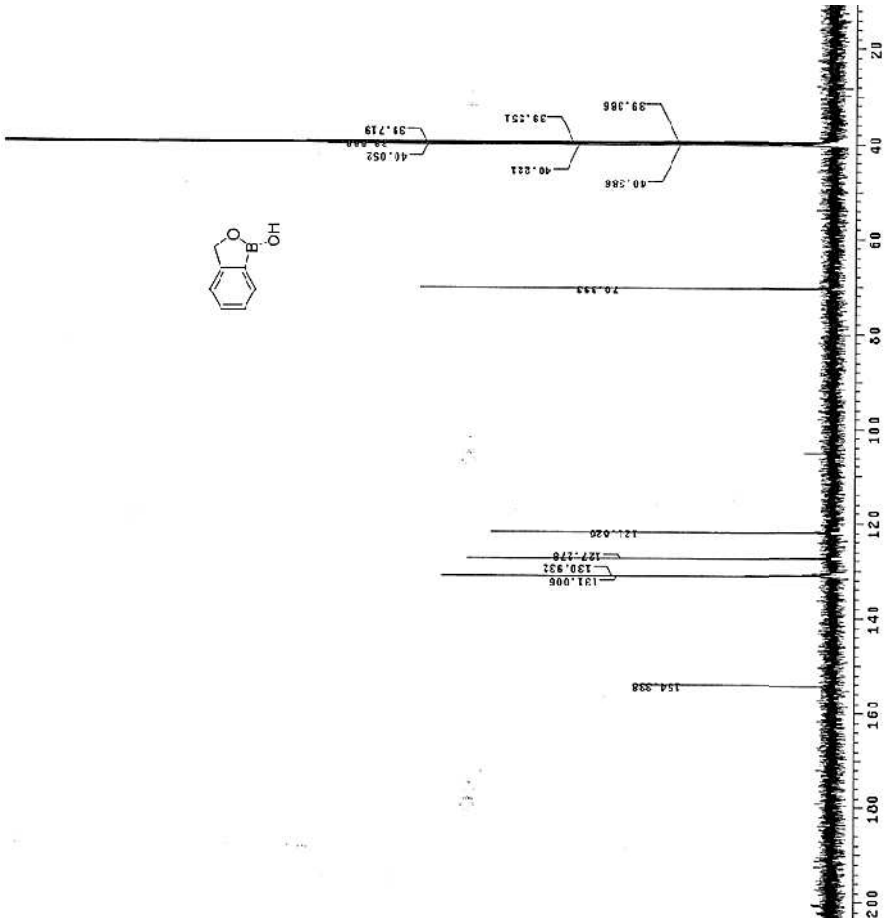
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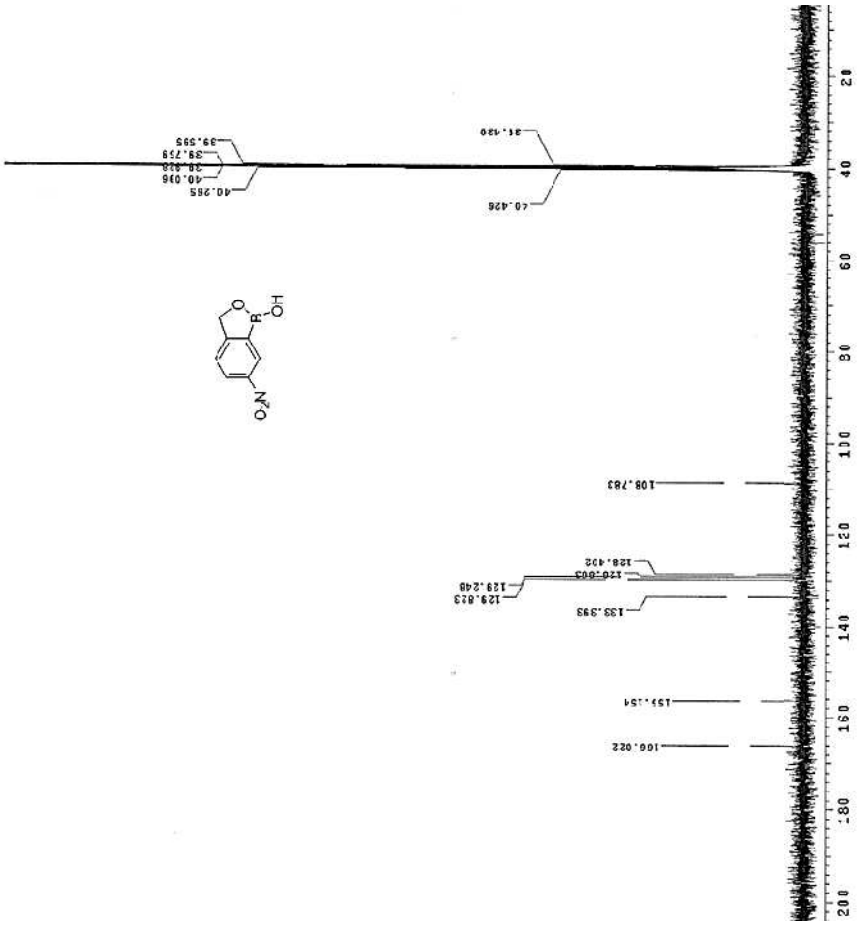
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APPENDIX

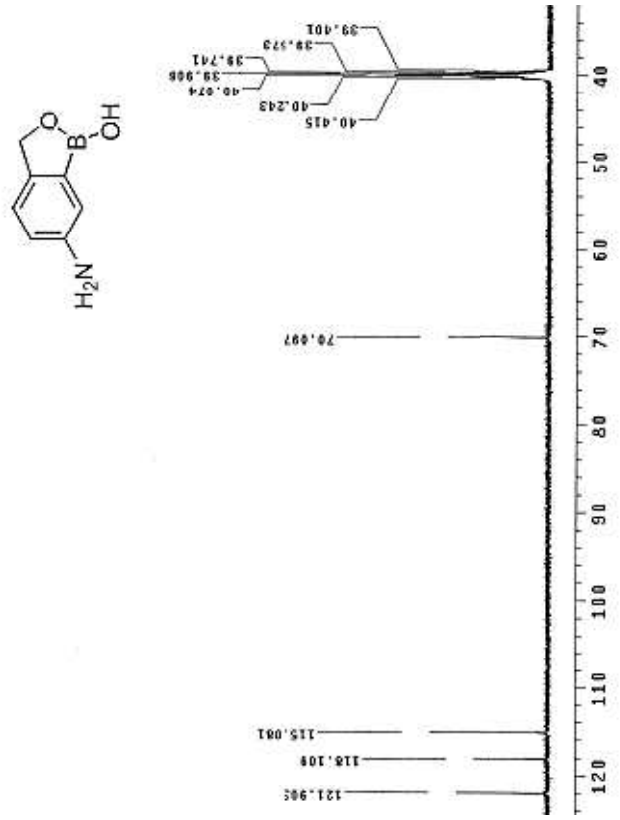

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B
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0.32
0.90
0.16

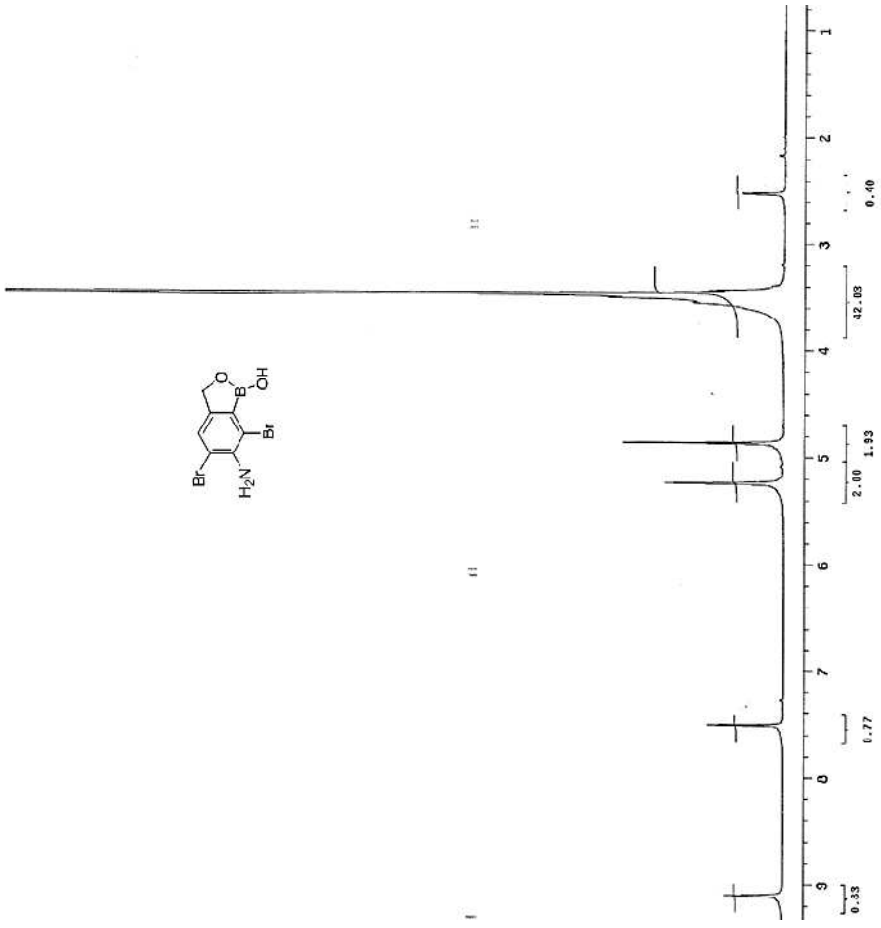

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40.052
40.221
40.386
70.193
121.020
127.278
130.932
131.006
154.338

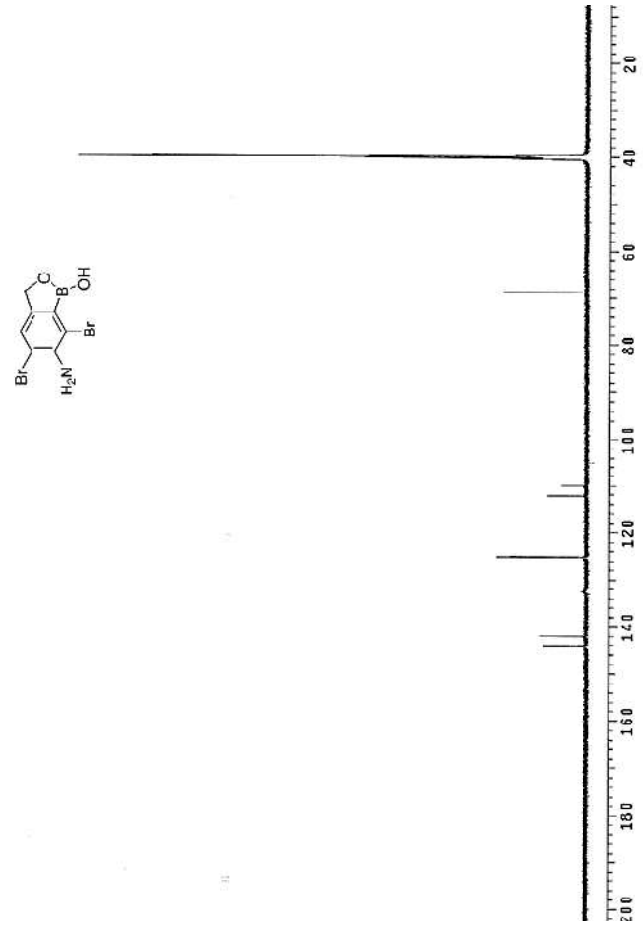
O2N
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1.00
0.56
0.83
1.02

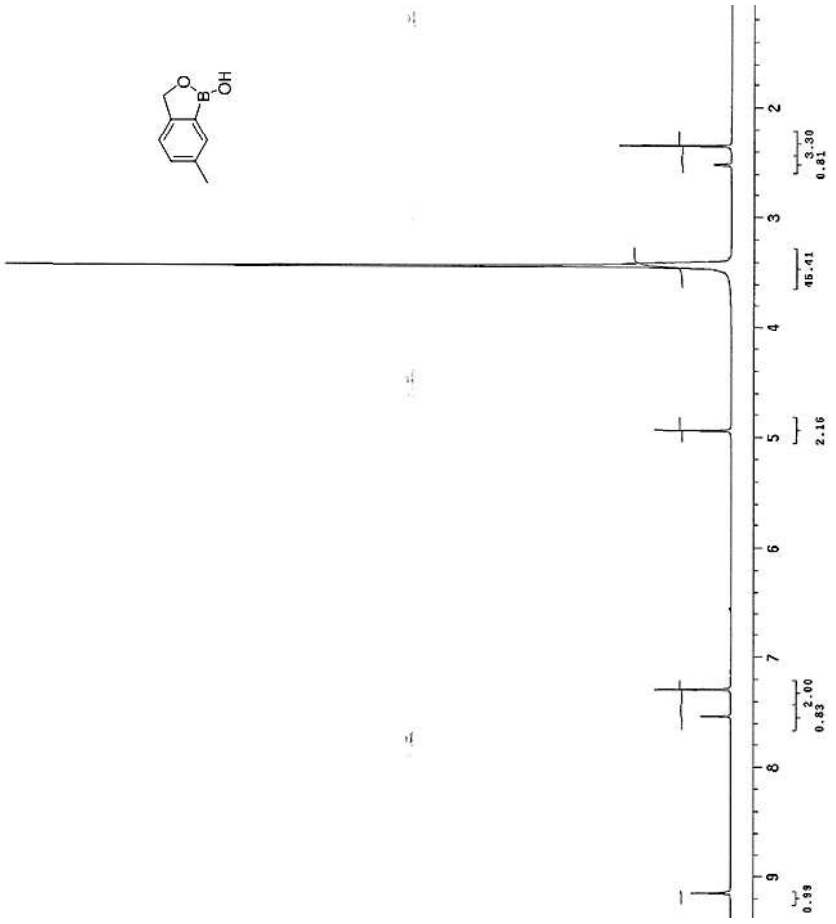

40.285
40.036
39.779
39.523
39.266
31.430
40.426
108.783
128.402
128.003
129.249
129.823
135.393
155.154
166.022

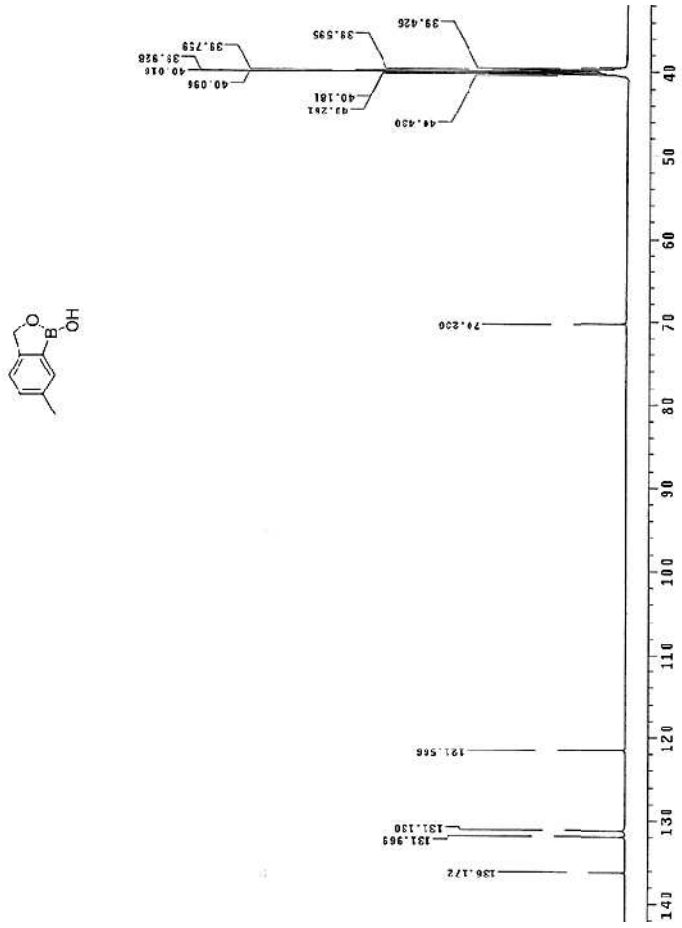
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OH
H2N
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1.00
1.05
1.86
2.31

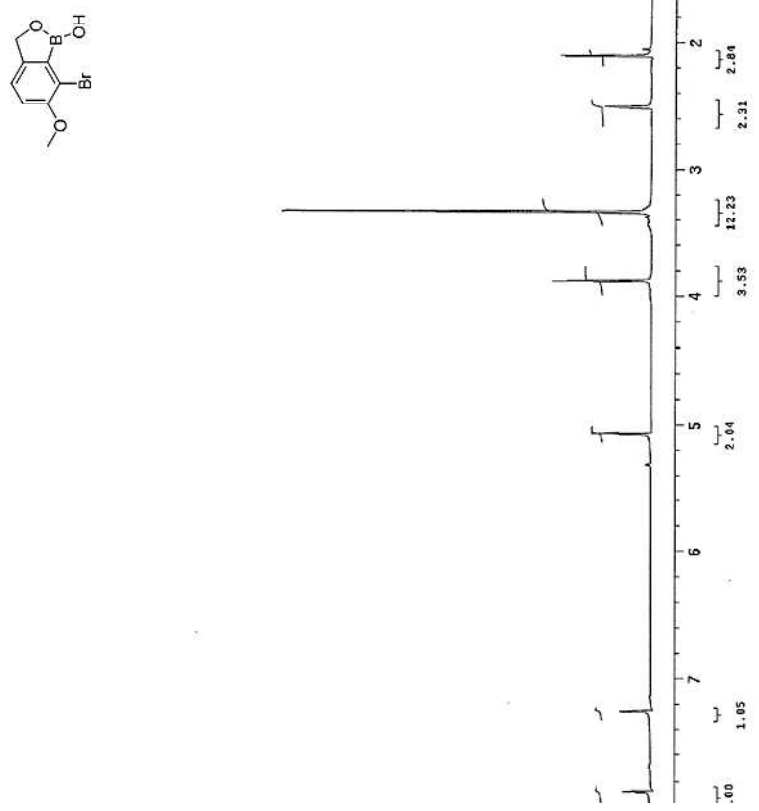

O
B
OH
H_2N
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39.573
39.741
39.908
40.074
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70
80
90
100
110
120

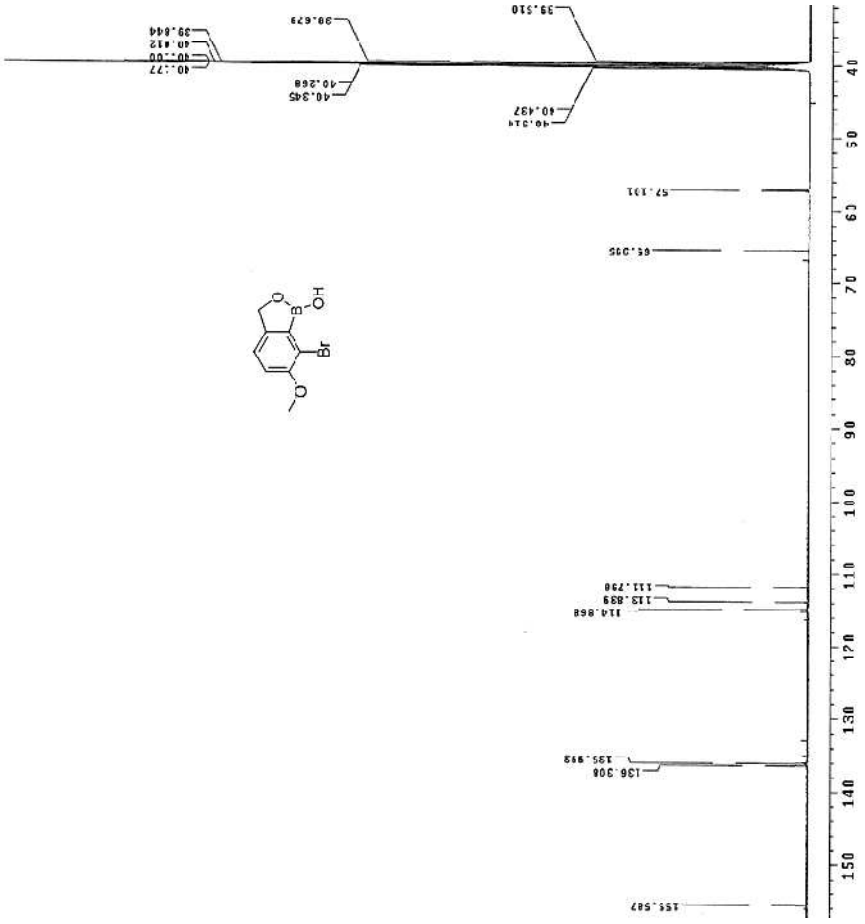

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B
OH
Br
Br
H2N
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0.40
2.00
1.93
0.77
0.33

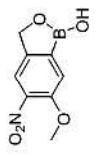

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H2N
Br
B
OH
20
40
60
80
100
120
140
160
180
200

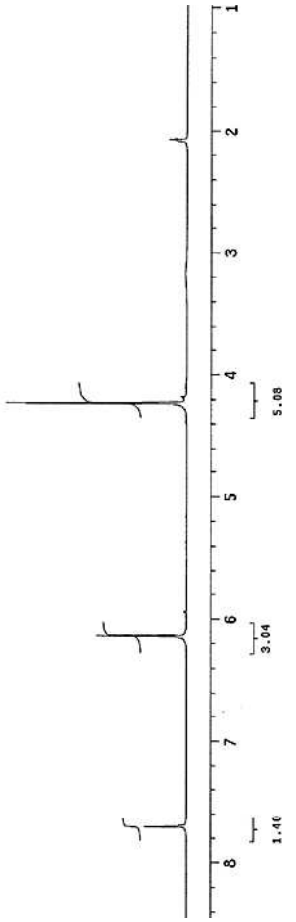

O
B
OH
9
8
7
6
5
4
3
2
0.99
2.00
0.83
2.16
15.41
3.30
0.81

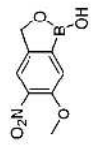

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136.172

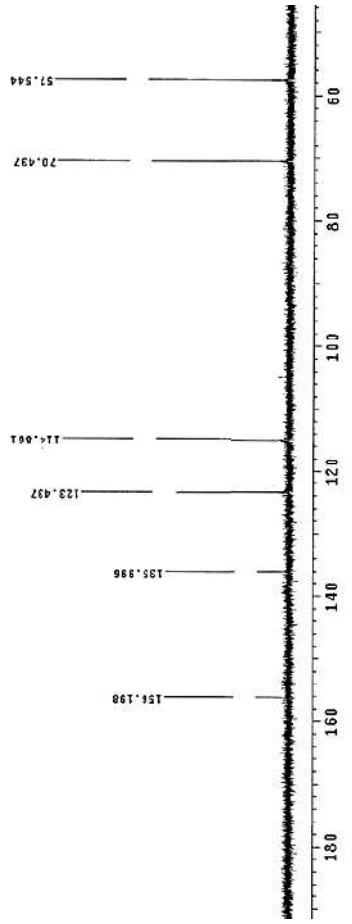

OH
B
O
Br
O
1
2
3
4
5
6
7
2.84
2.31
12.23
3.53
2.04
1.05
.00

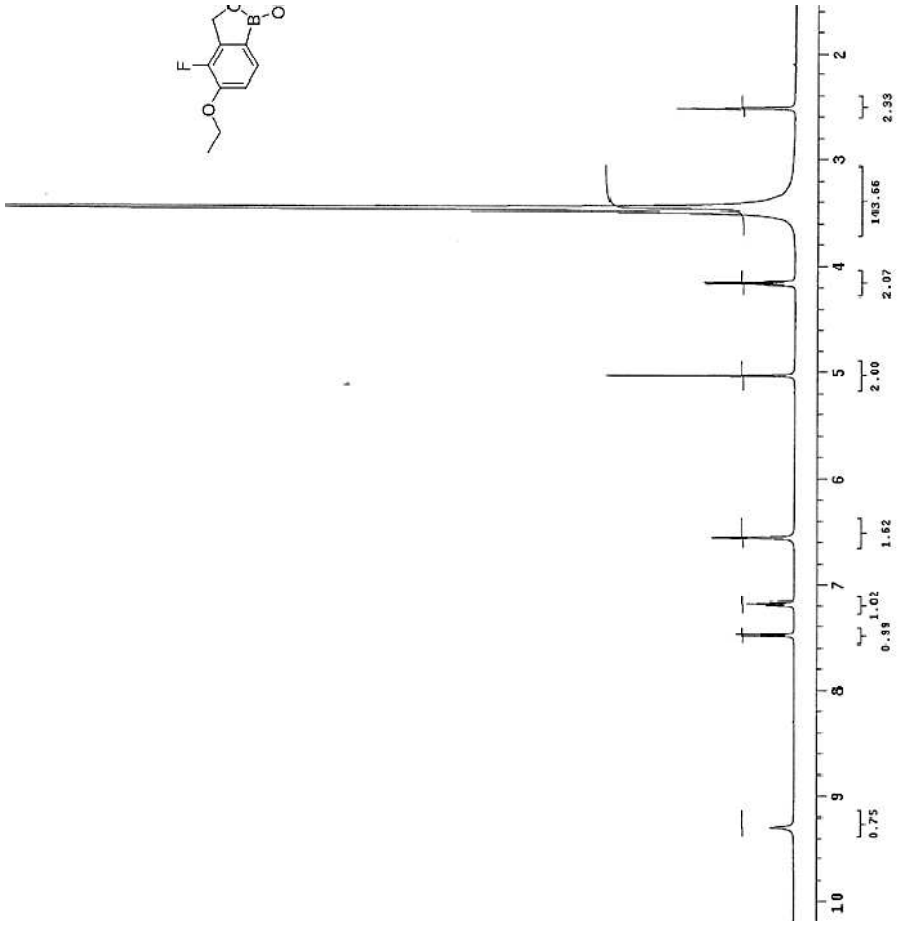

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40.437
40.577
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57.131
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118.868
135.998
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120
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140
150
B
OH
Br
O

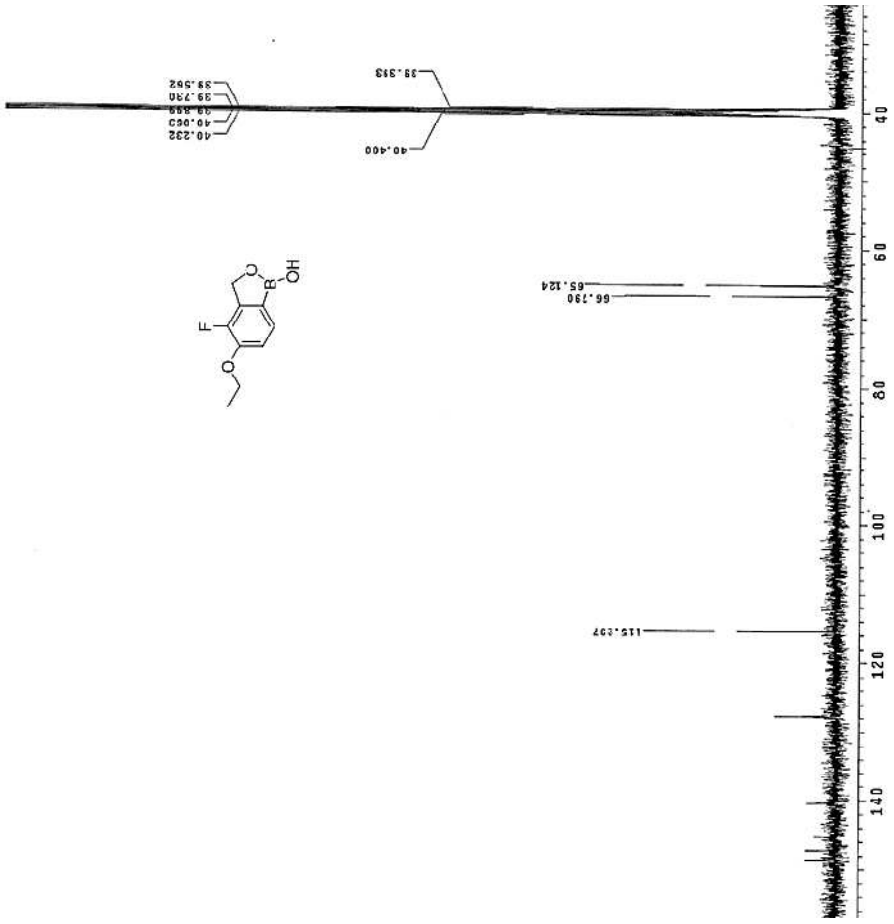

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B
OH
O2N
O

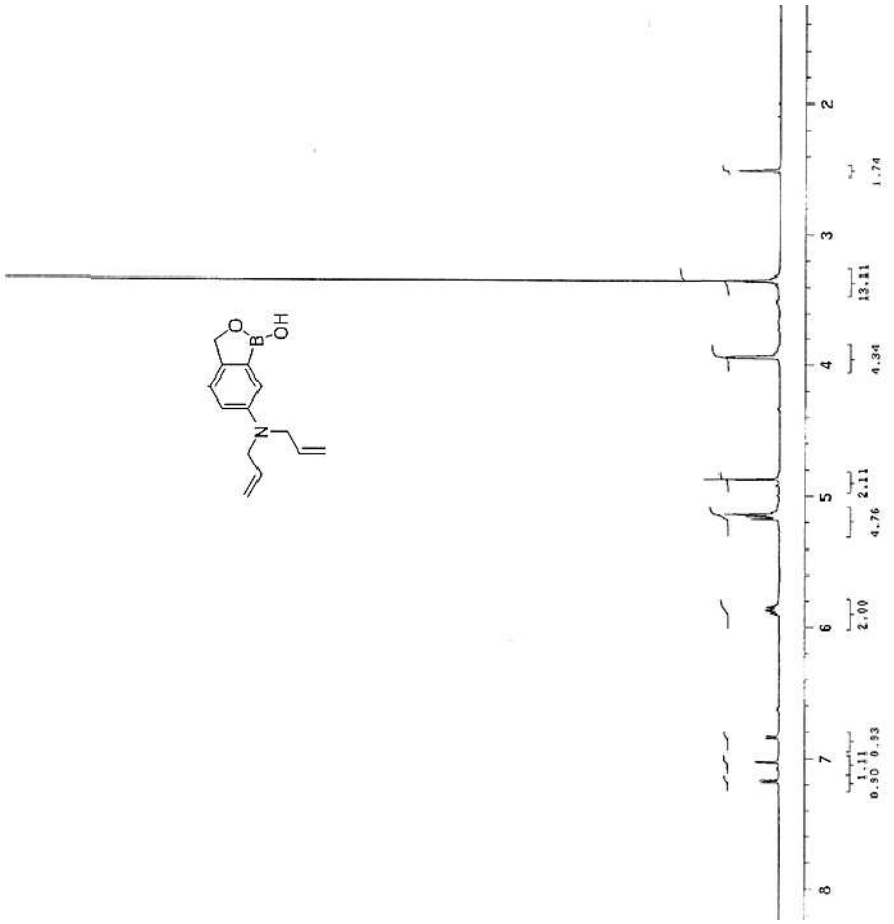

1
2
3
4
5.08
5
6
3.04
7
8

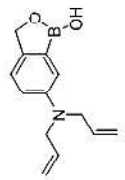

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B
OH
O2N
O

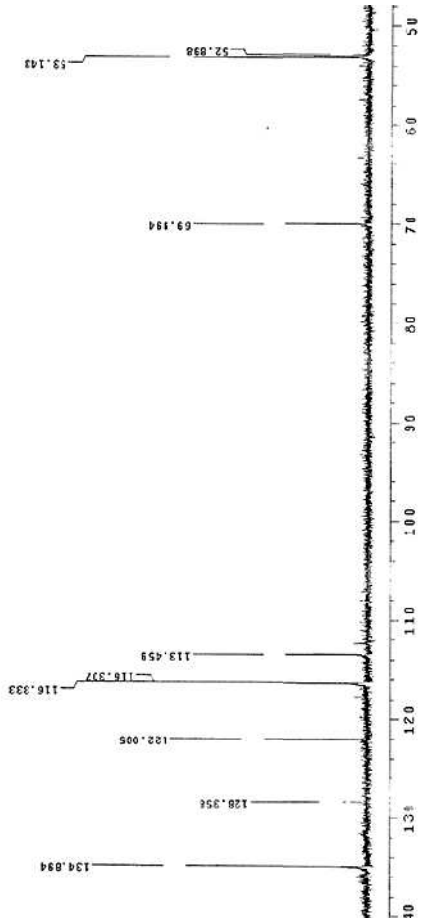

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123.437
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80
100
120
140
160
180

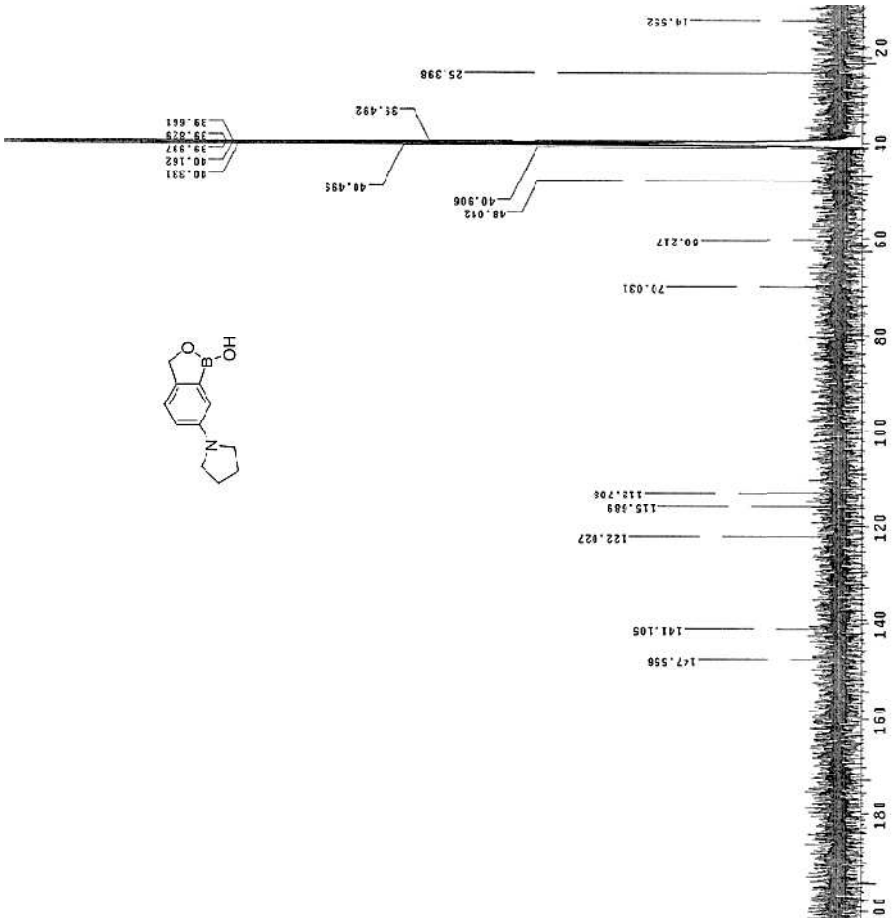

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O
B
O
2.33
143.66
2.07
2.00
1.62
1.02
0.99
0.75

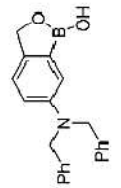

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115.037
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80
100
120
140
O
B
OH
F
O

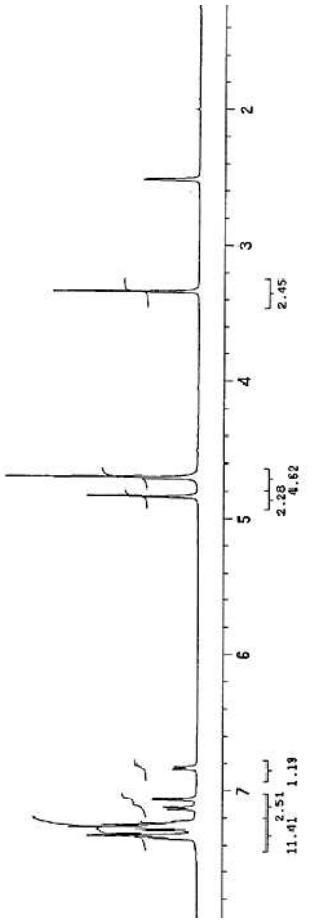

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B
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N
2
3
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8
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0.93
1.74

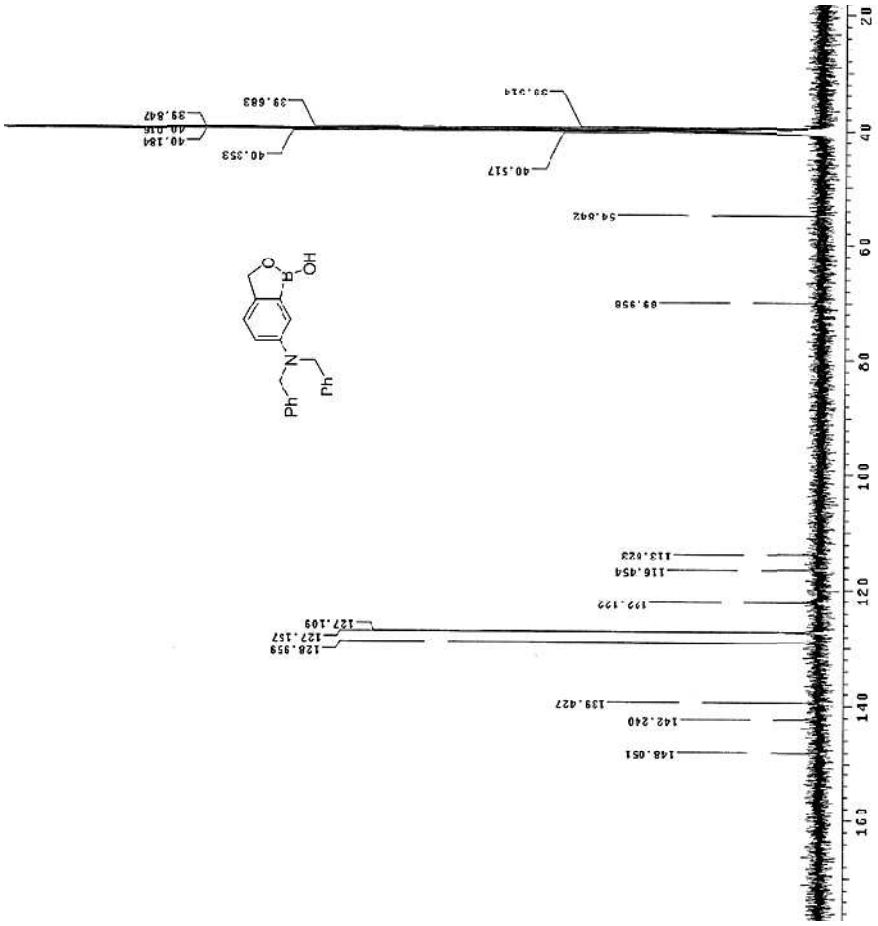


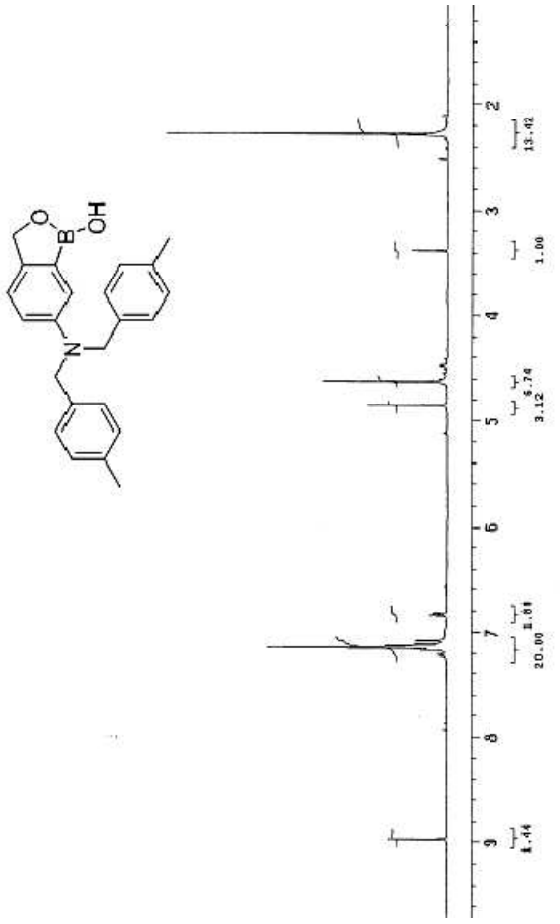

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50
60
70
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90
100
110
120
130
140

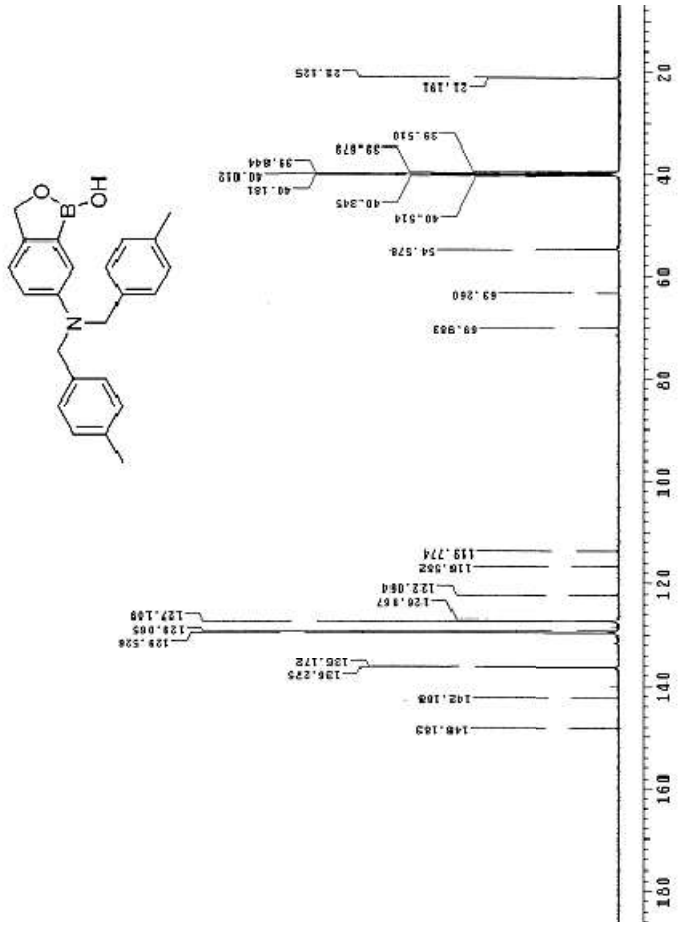

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40.331
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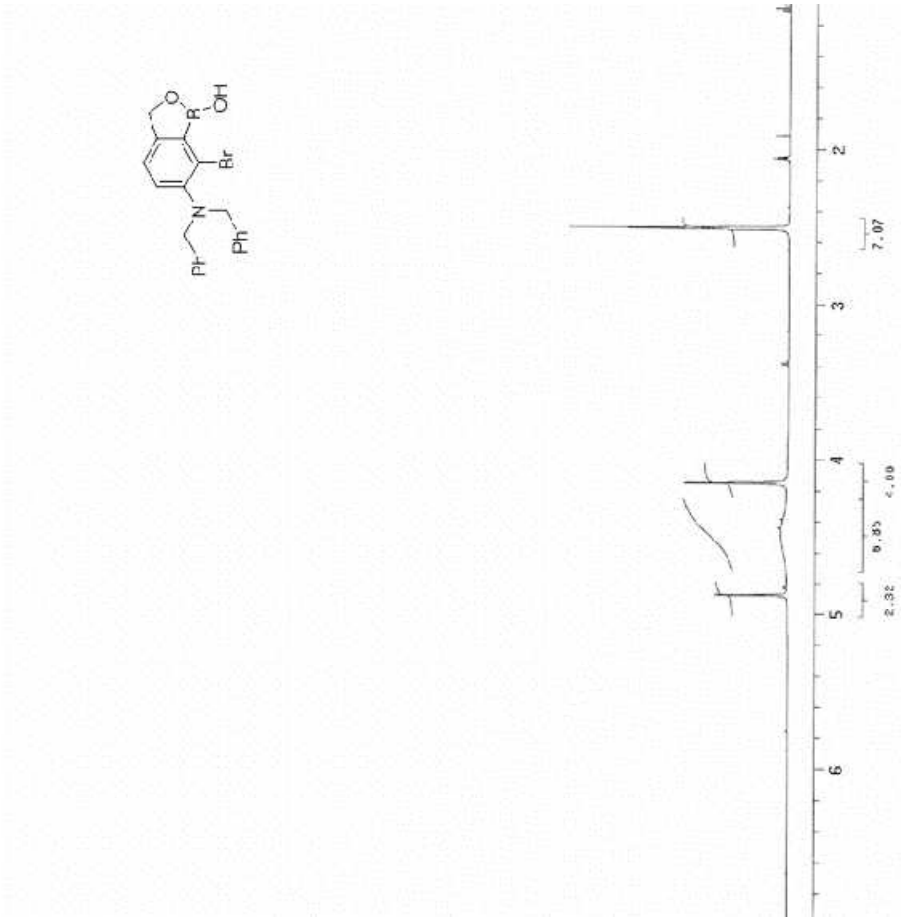

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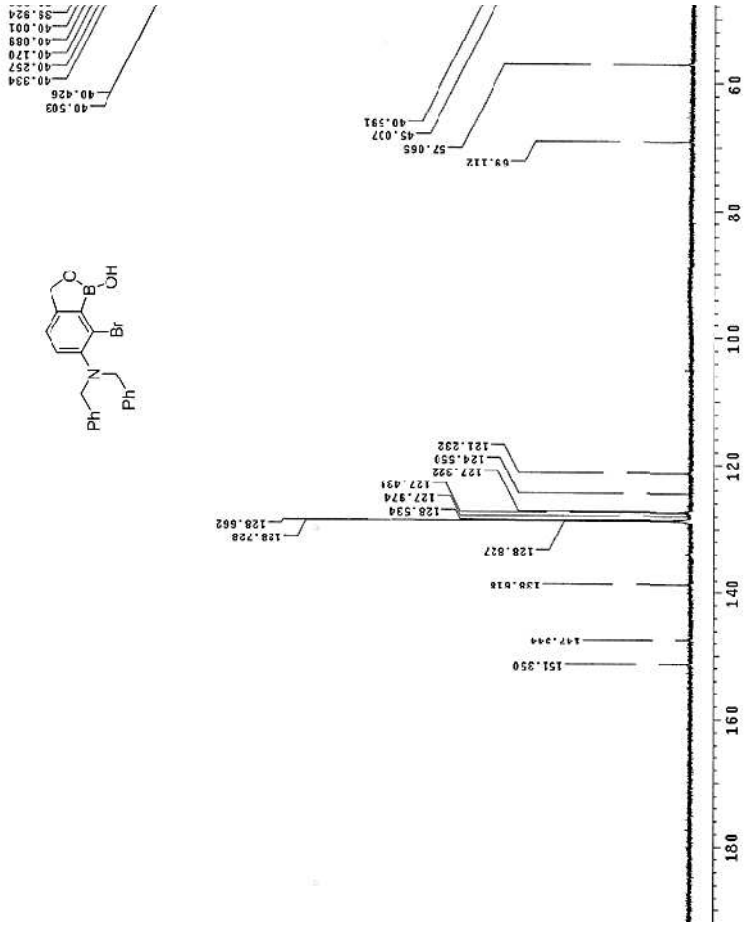

2
3
4
5
6
7
2.45
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4.62
2.51
11.41
1.19

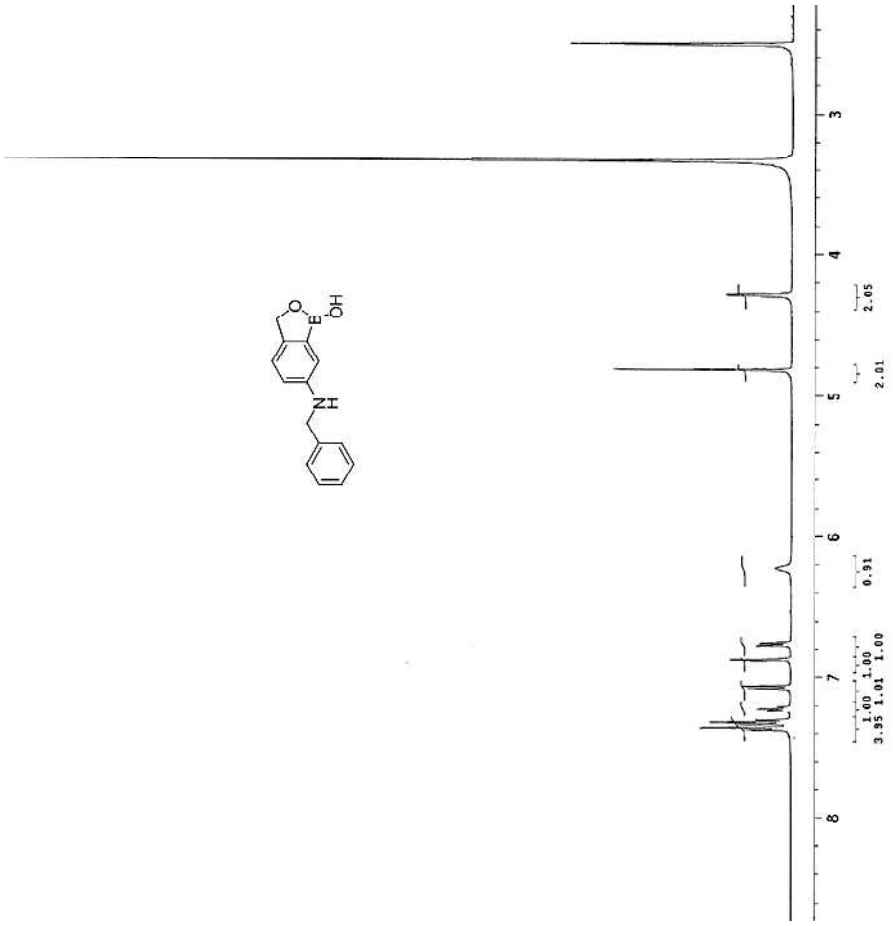

39.214
39.683
39.847
40.016
40.184
40.353
40.517
54.042
89.958
113.023
116.454
122.122
127.109
127.157
128.959
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148.051
20
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60
80
100
120
140
160
HO
B
O
N
Ph
Ph

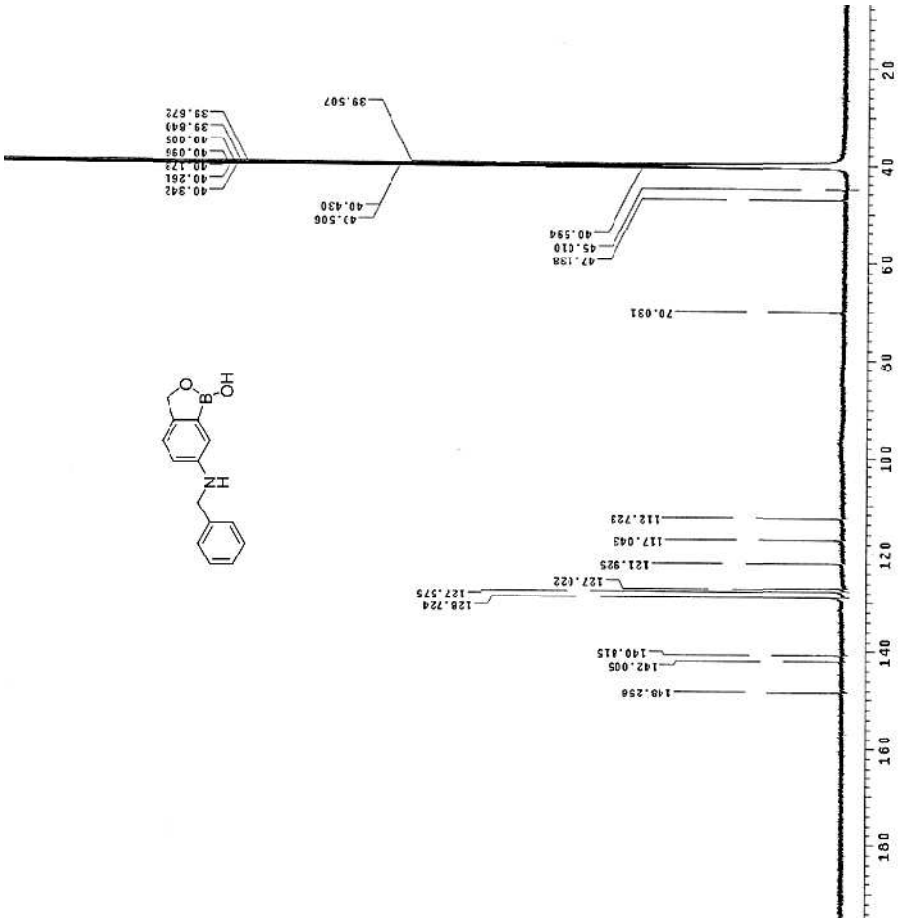

O
B
OH
N
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3
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9
13.42
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6.74
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8.88
20.00
4.44

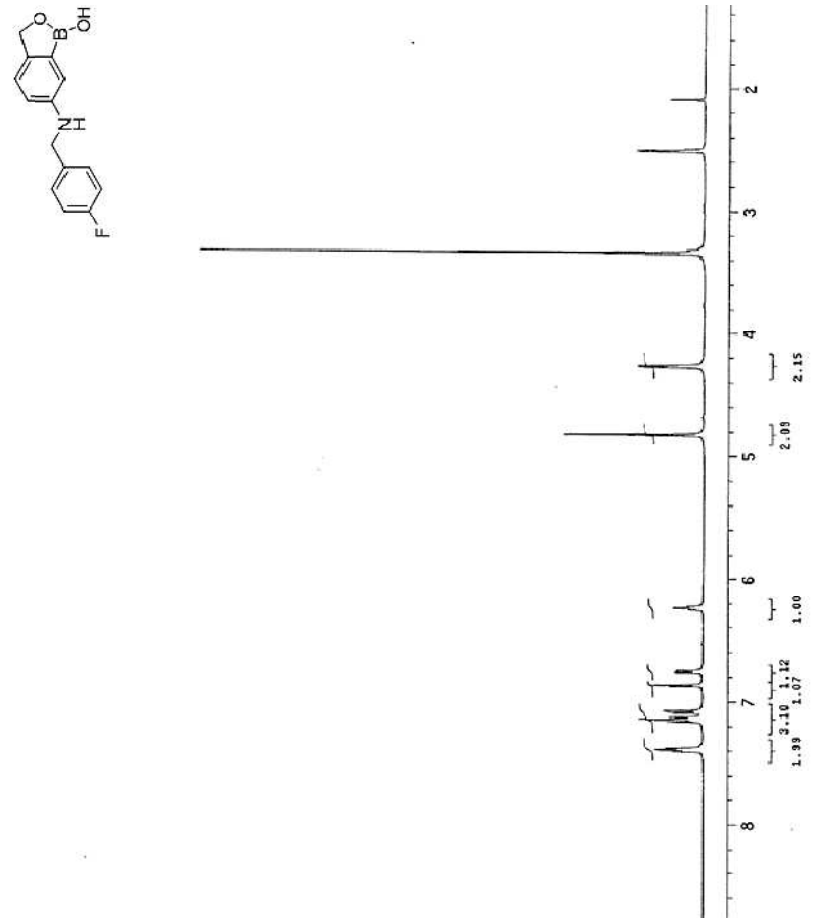

21.191
22.125
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39.679
39.844
40.012
40.181
40.345
40.514
54.578
69.260
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119.774
126.562
122.084
126.967
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142.108
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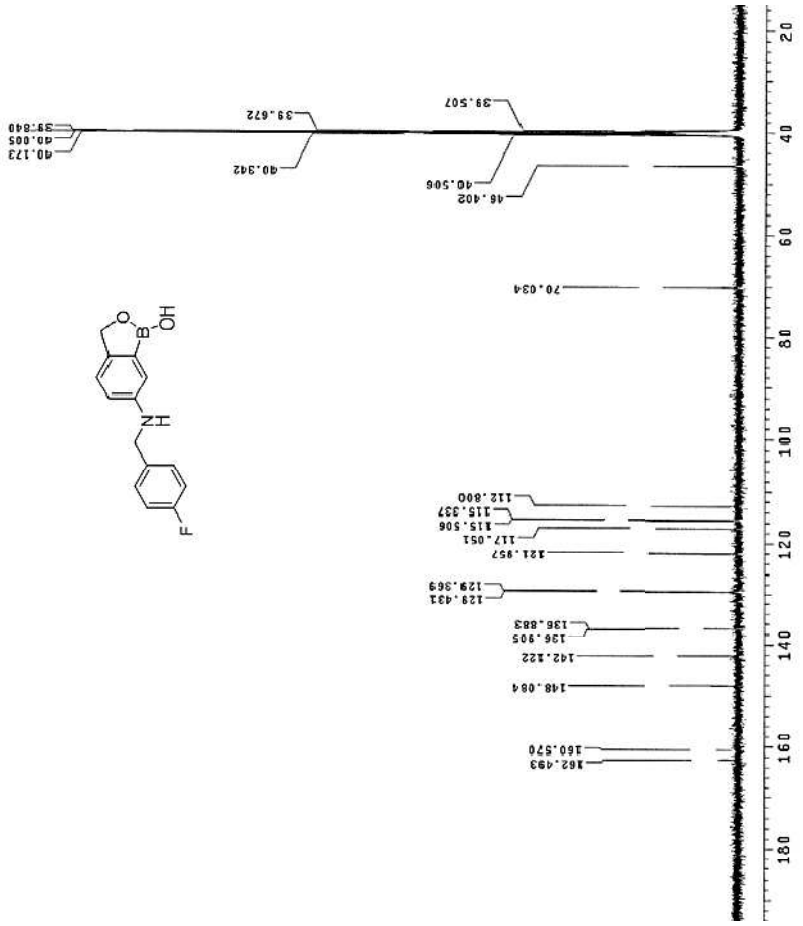

O
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OH
Br
N
Ph
Ph
2
3
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5
6
7.07
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9.83
4.00

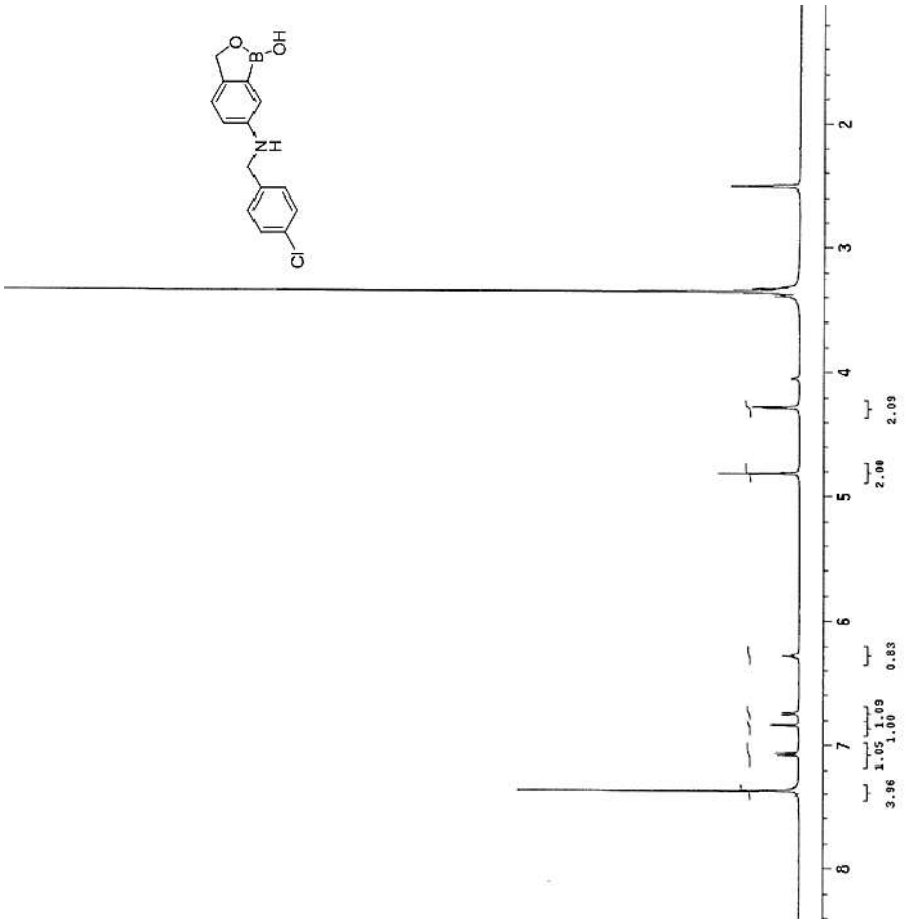

O
B
OH
Br
N
Ph
Ph
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40.334
40.257
40.170
40.001
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40.591
45.037
57.065
59.112
121.232
124.550
127.322
127.431
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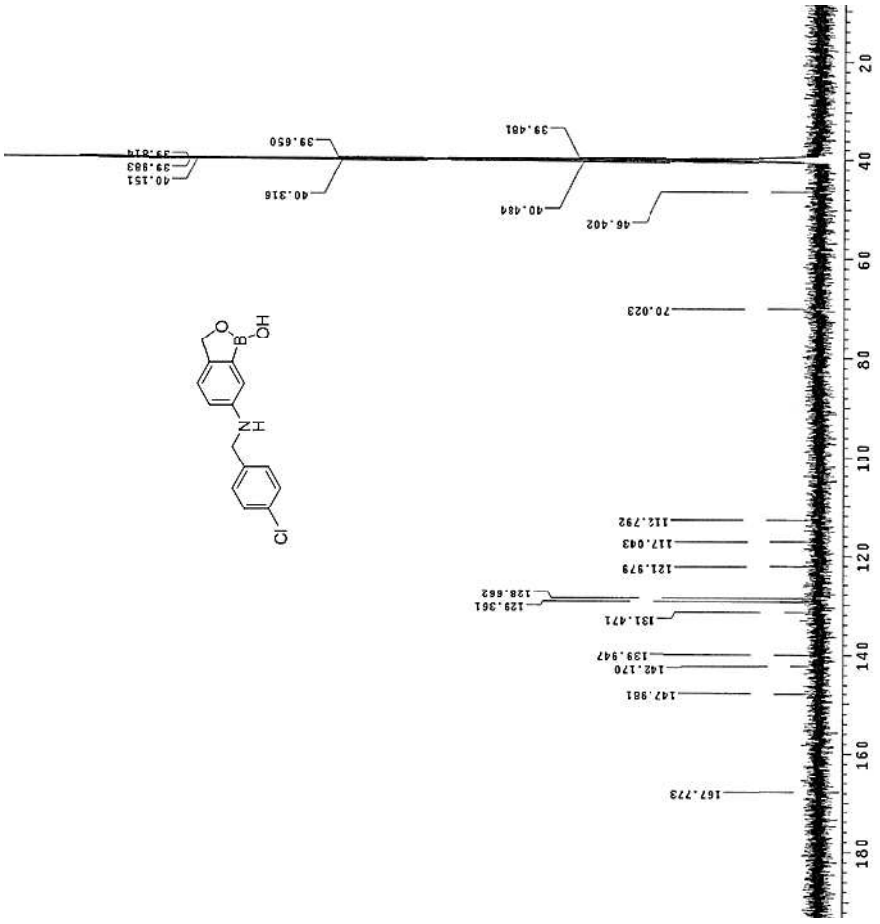

F
OH
NH
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2.01
0.91
1.00 1.00
3.95 1.01 1.00

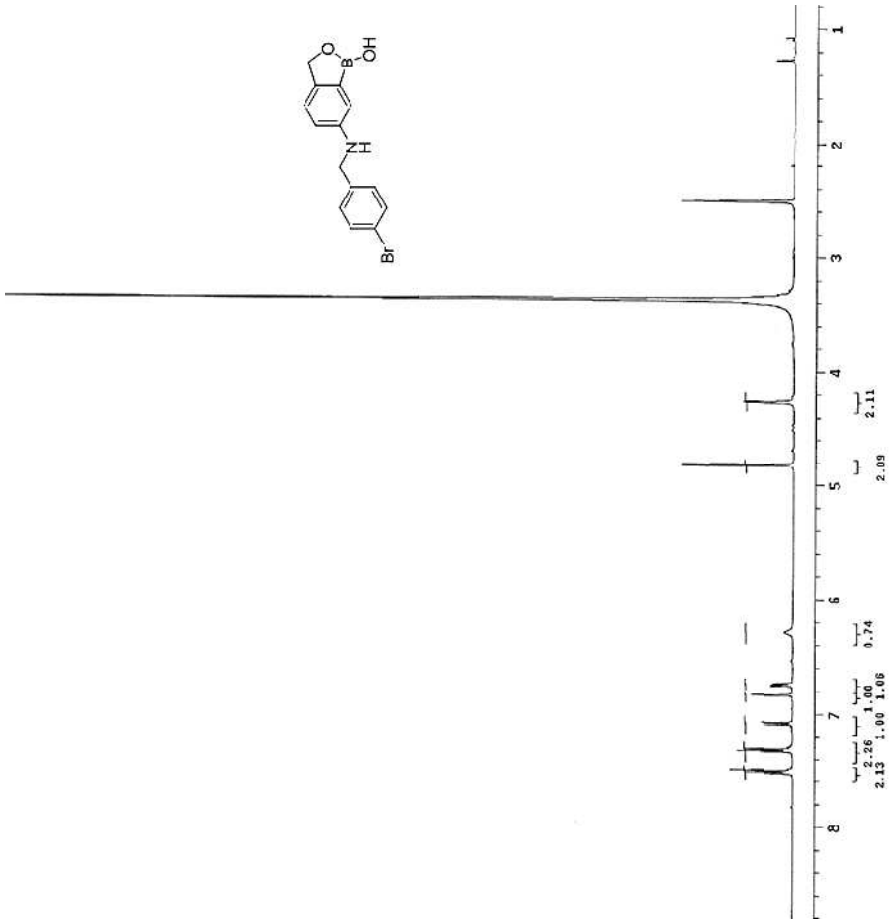

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39.849
40.049
40.096
40.122
40.261
40.342
40.430
43.506
40.594
45.610
47.158
70.031
112.723
117.043
121.925
127.022
127.575
128.724
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140
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O
B
OH
N
H

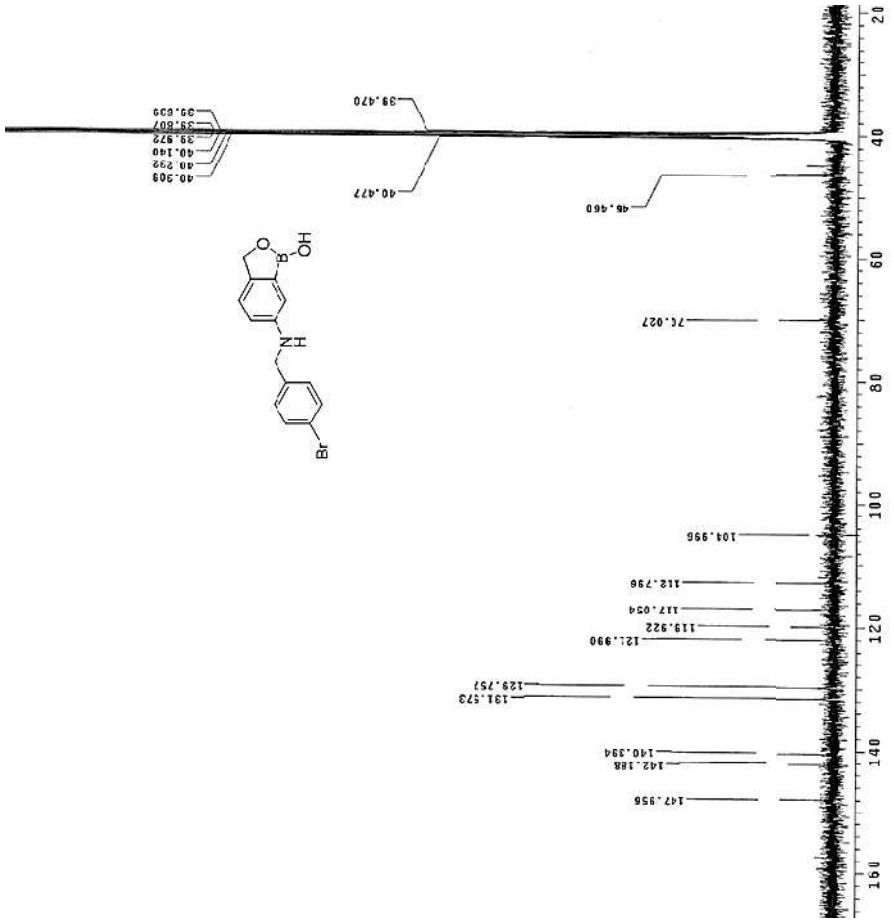

O
B
OH
N
H
F
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3
4
5
6
7
8
2.15
2.05
1.00
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1.07
1.99

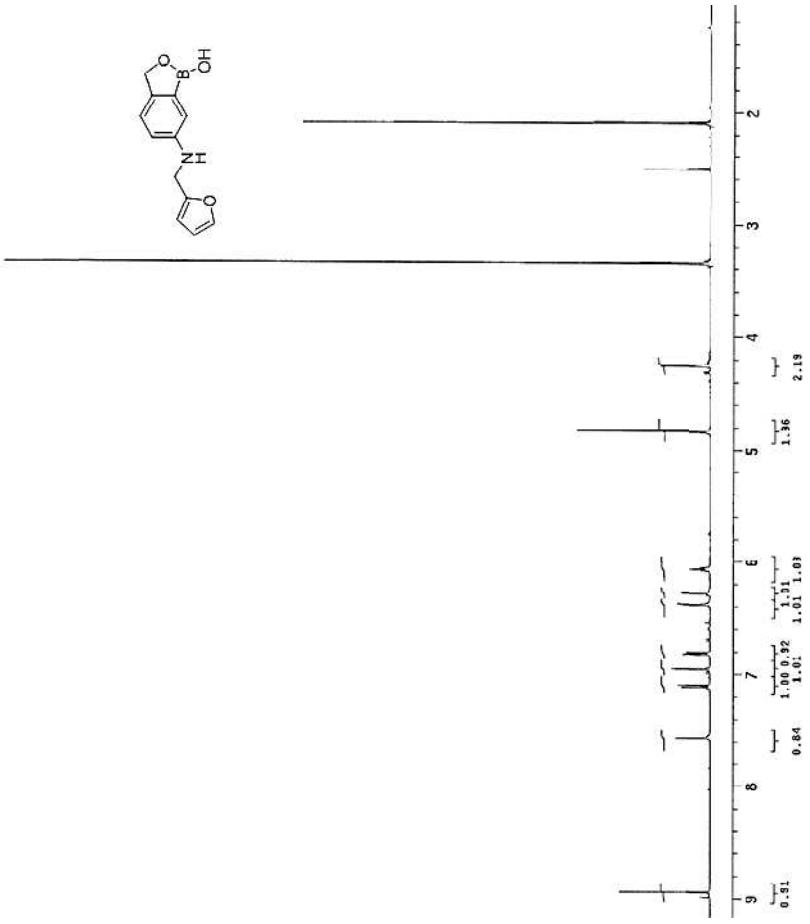

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39.672
40.342
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40.673
40.506
40.340
40.173
40.005
39.840
46.402
70.034
112.800
115.337
115.506
117.051
121.957
129.369
129.431
136.883
136.905
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162.493
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140
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OH
B
O
NH
F

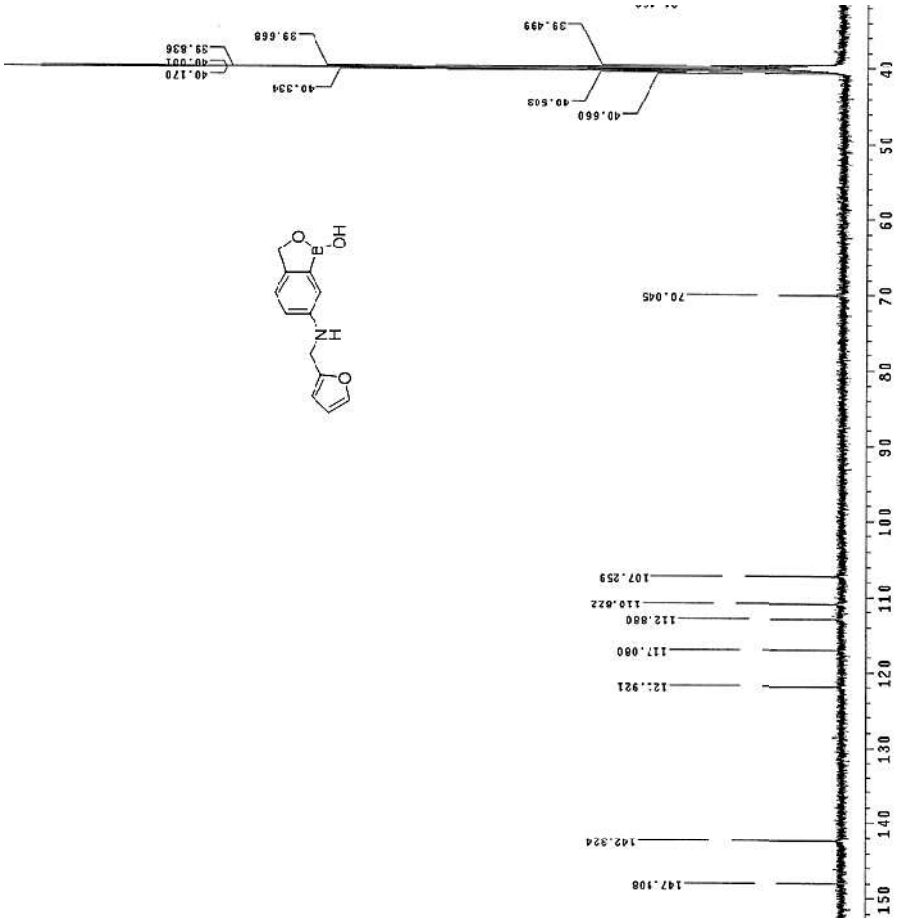

O
B
OH
N
H
Cl
2
3
4
5
6
7
8
2.09
2.00
0.83
1.09
1.00
3.96

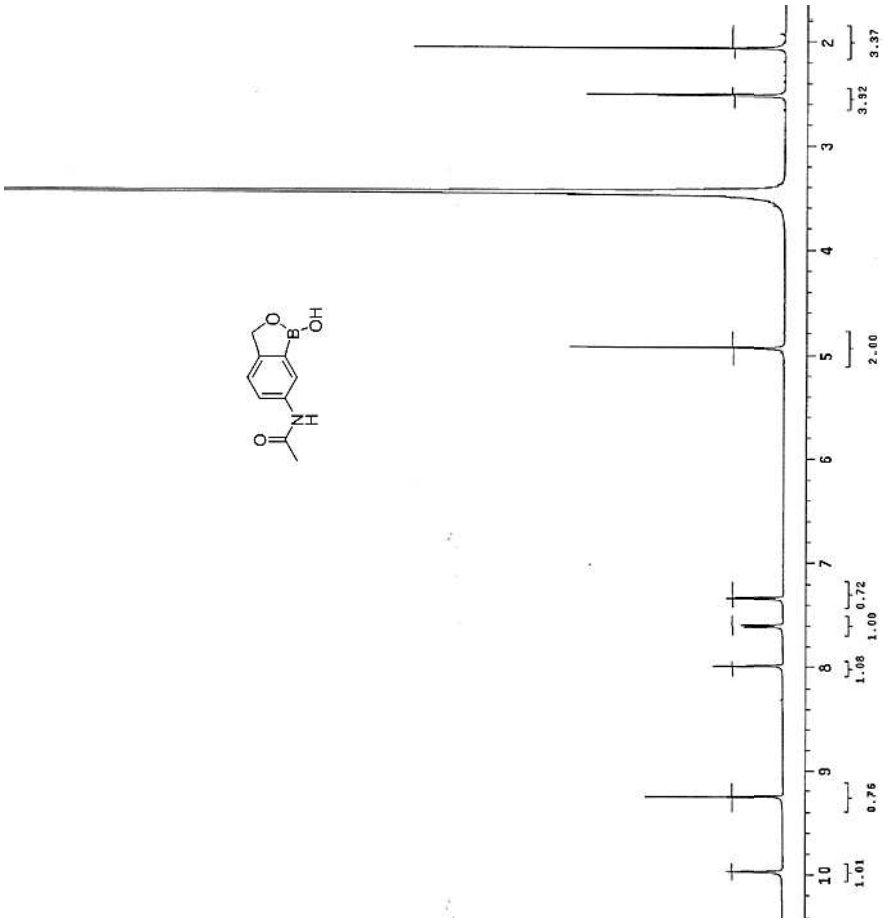

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39.650
39.214
40.151
39.883
40.316
40.484
46.402
70.023
112.792
117.043
121.979
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129.361
131.471
139.947
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167.773
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60
80
100
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140
160
180
O
B
OH
N
H
Cl

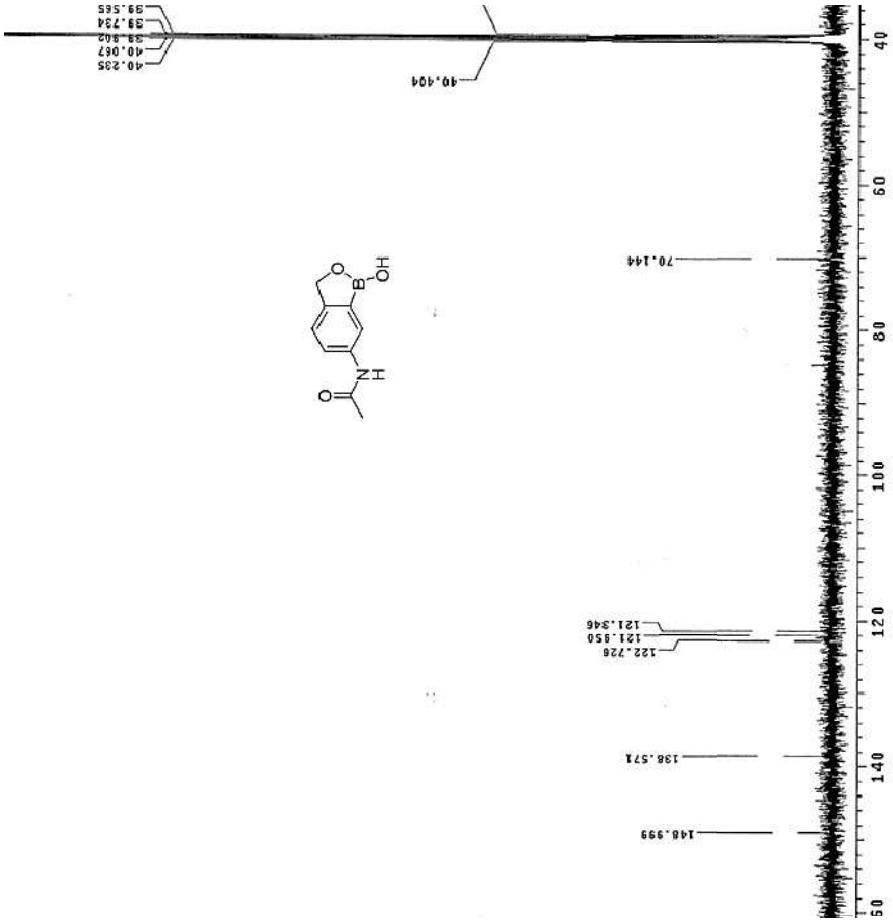

O
B
OH
N
H
Br
1
2
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6
7
8
2.11
2.09
0.74
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2.26
2.13
1.00

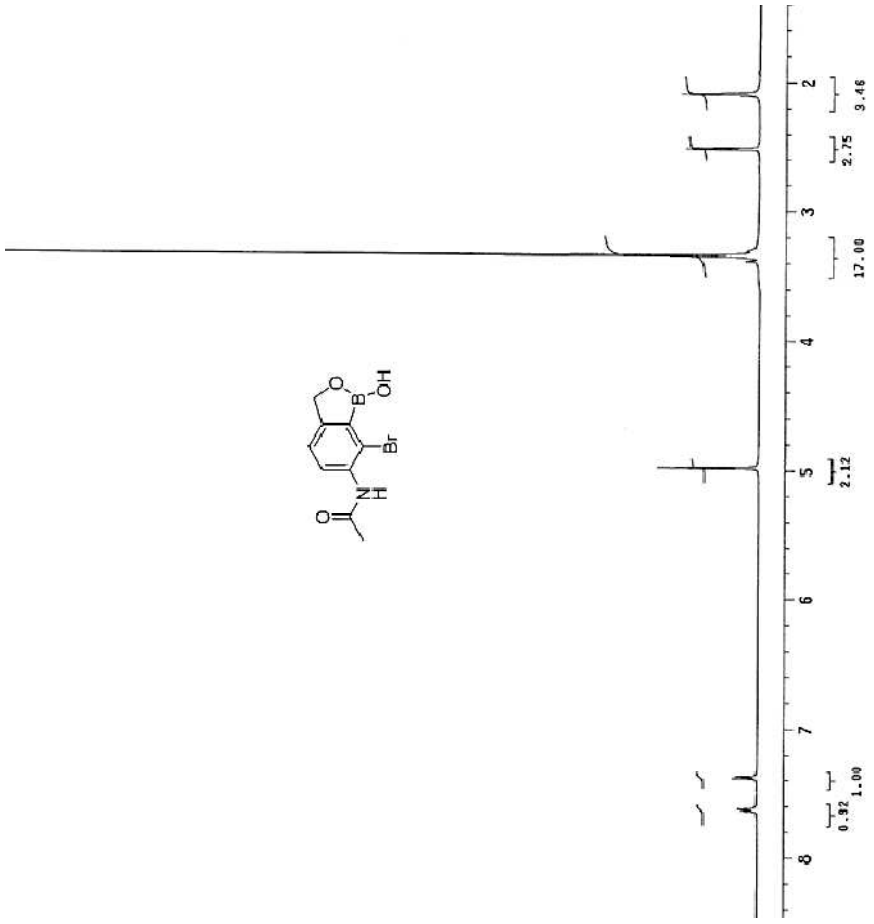

147.956
142.188
140.394
129.757
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121.990
119.922
117.054
112.796
104.995
76.027
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40.477
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40.309
40.292
40.140
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39.807
39.639
O
B
OH
NH
Br

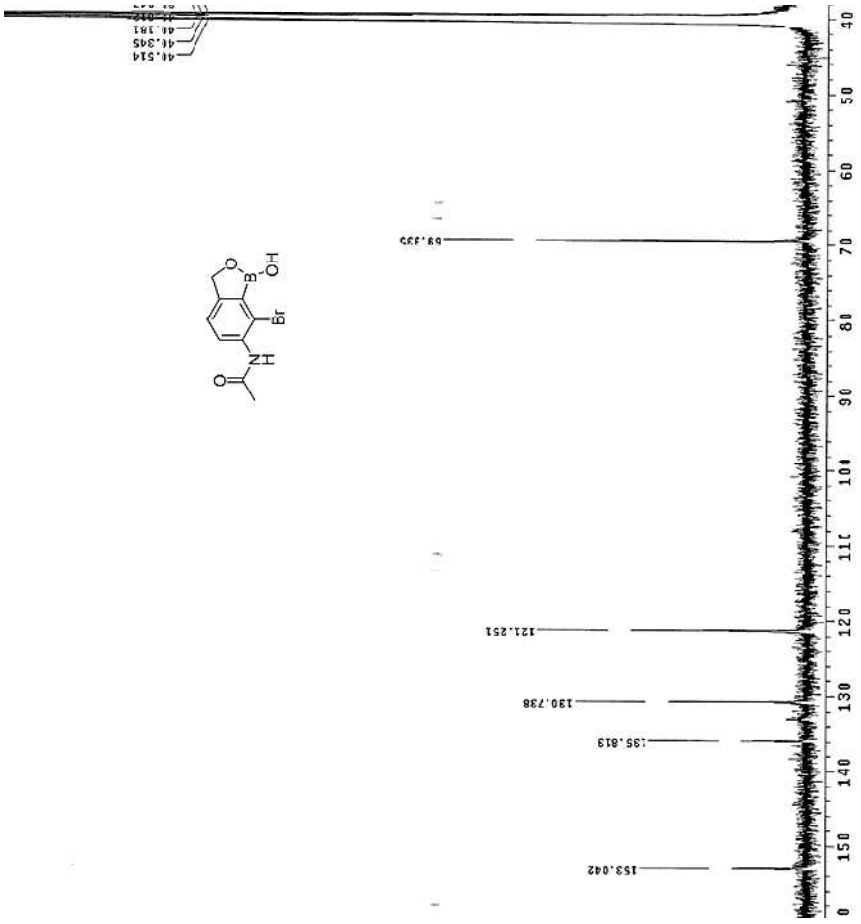


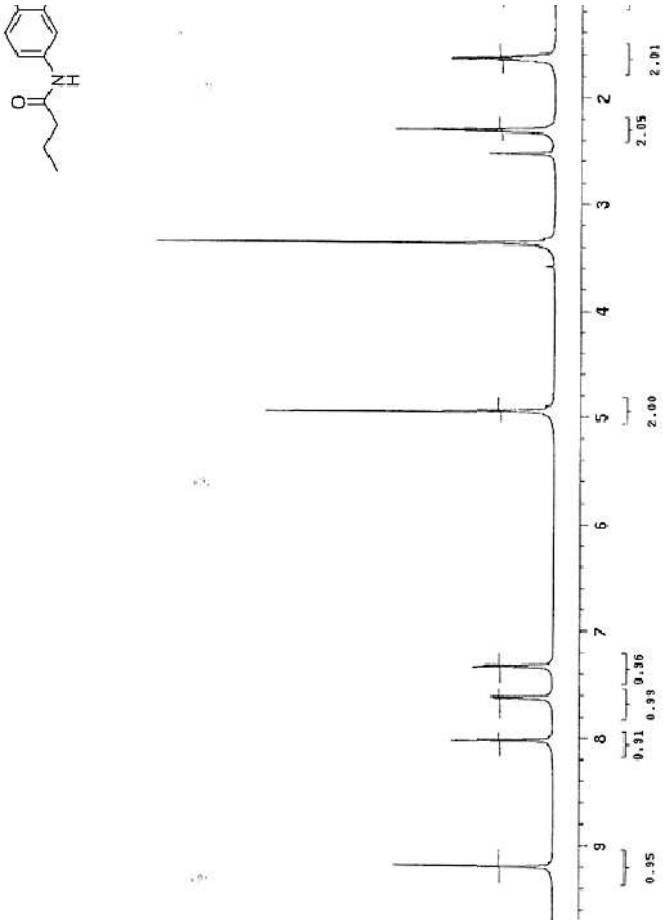

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39.836
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40.001
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40.660
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117.080
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142.324
147.108

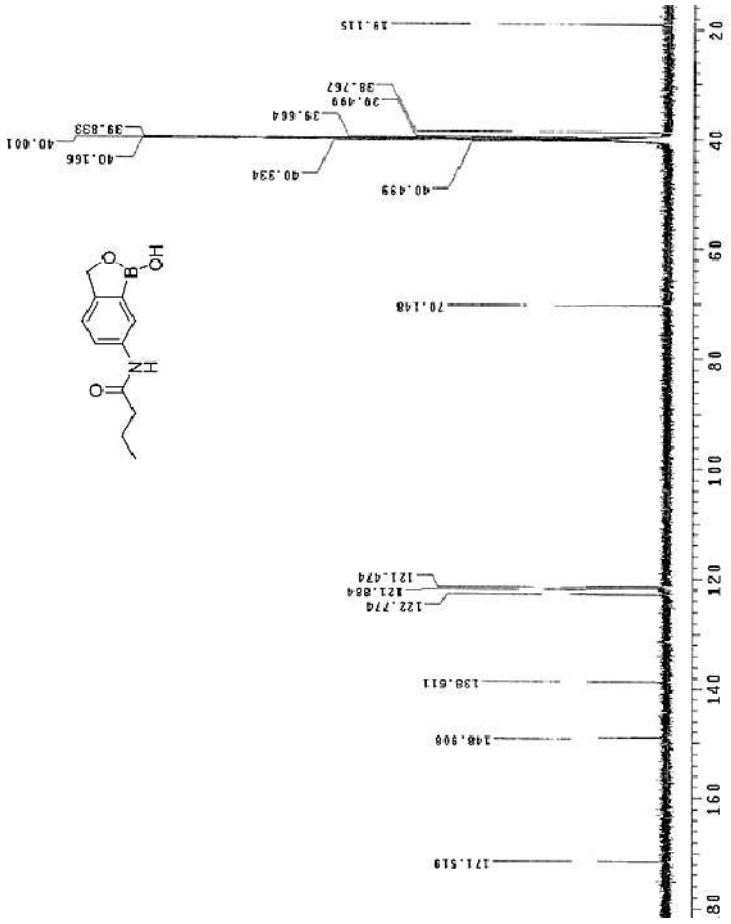

O
B
OH
N
H
O
2
3
4
5
6
7
8
9
10
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3.52
2.00
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1.08
0.76
1.01

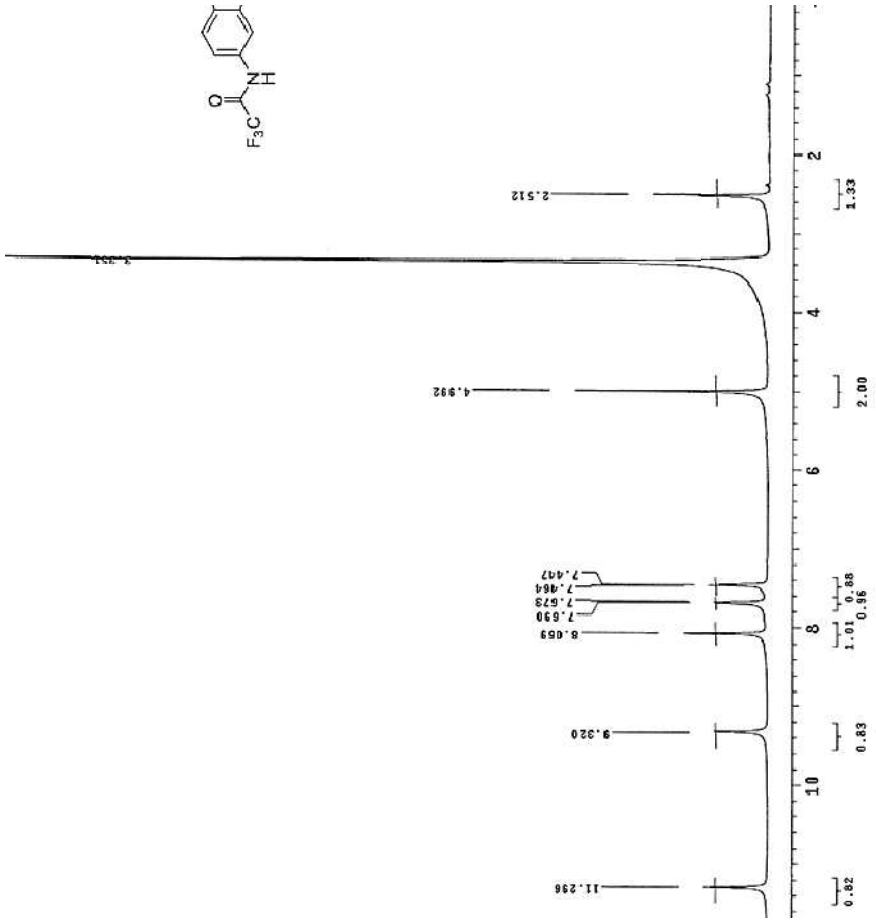

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40.067
39.902
39.734
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19.404
70.144
121.346
121.850
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136.574
148.999
40
60
80
100
120
140
160
OH
B
O
NH
O

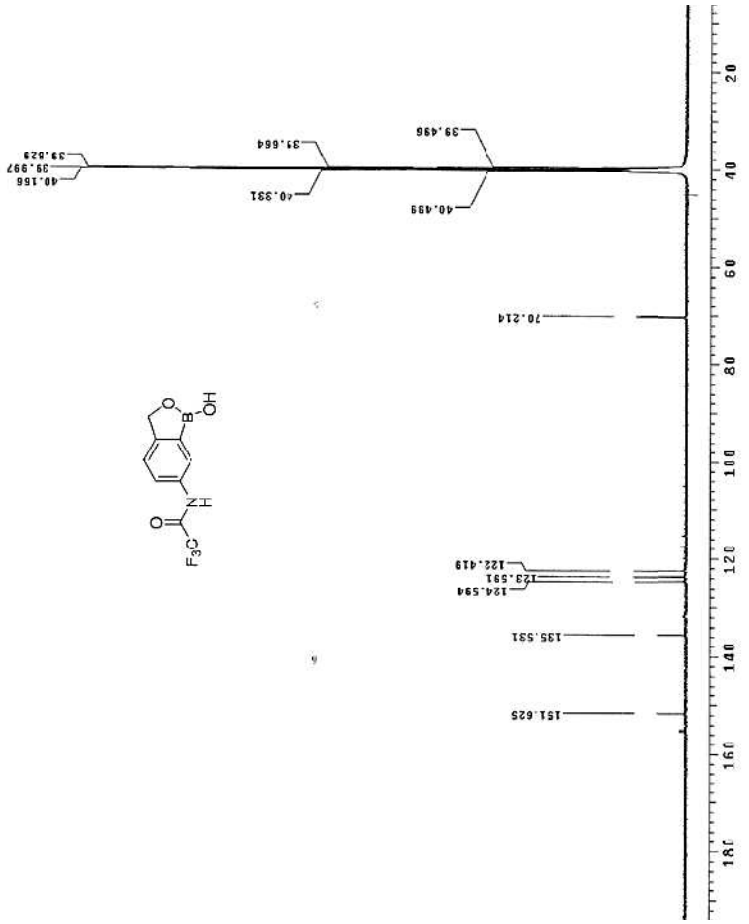

OH
B
O
Br
N
H
O
2.12
17.00
2.75
3.46
0.92
1.00

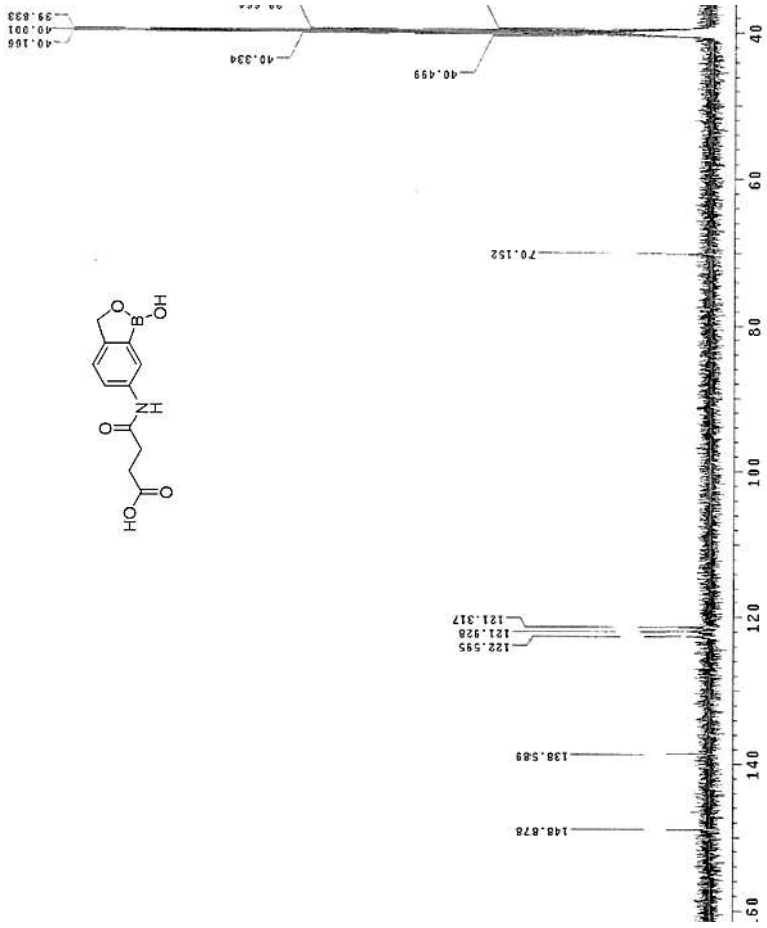

153.042
135.813
130.738
121.251
69.335
41.524
41.345
41.181
Br
B
OH
O
NH
O


NH
O
2.01
2.05
2.00
0.96
0.99
0.91
0.95

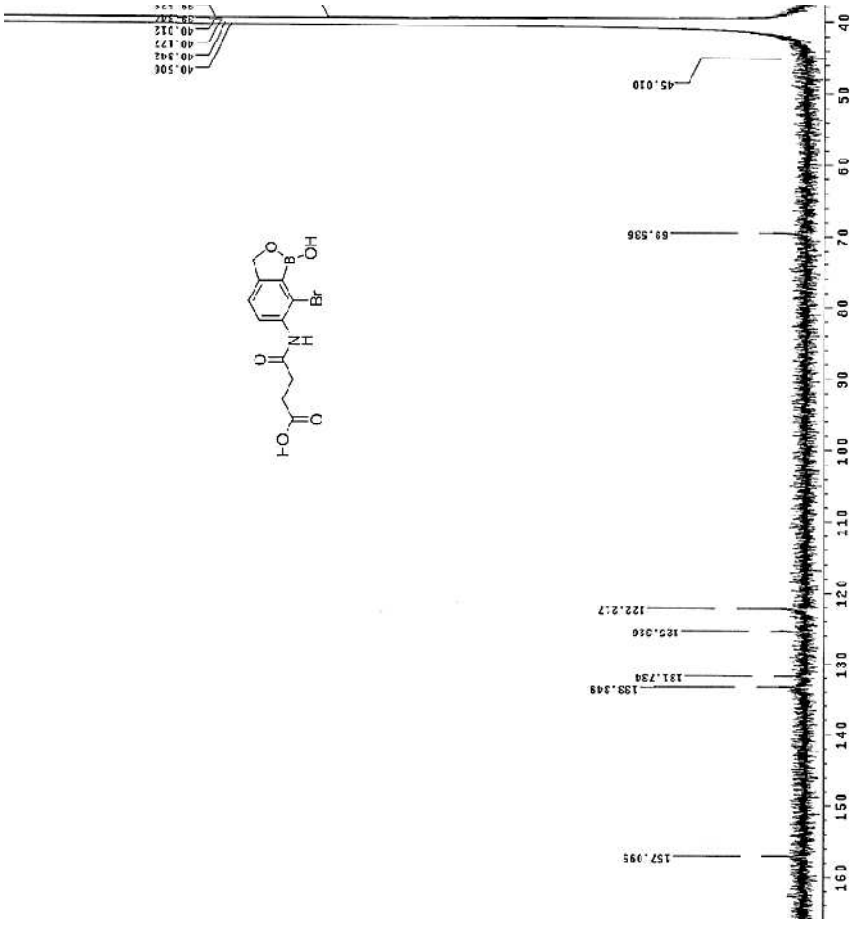

19.115
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40.166
40.334
40.499
70.148
121.474
121.884
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171.519
20
40
60
80
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140
160
180
O
B
OH
NH
O

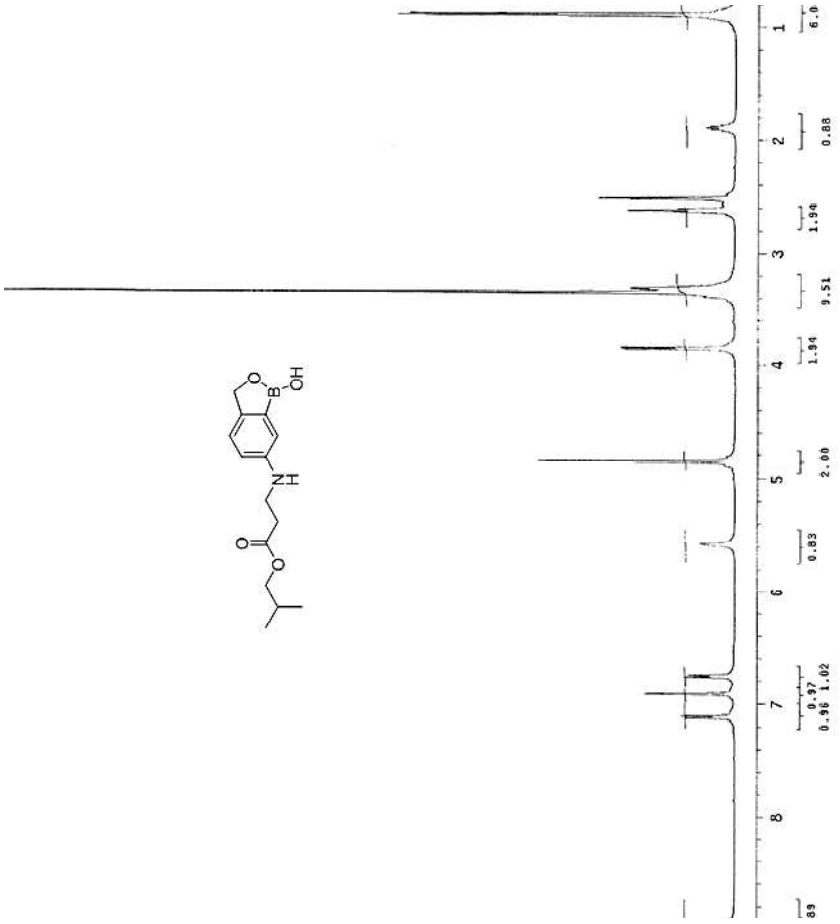

F3C
O
NH
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8.059
7.690
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7.447
4.992
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0.83
1.01
0.88
0.96
2.00
1.33

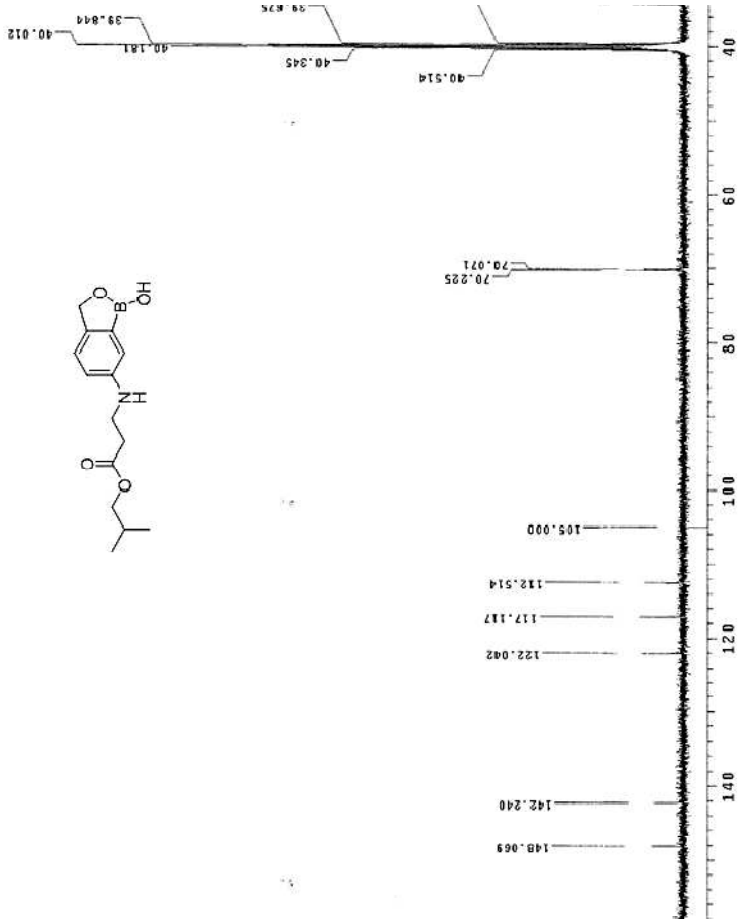

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124.594
135.551
151.625

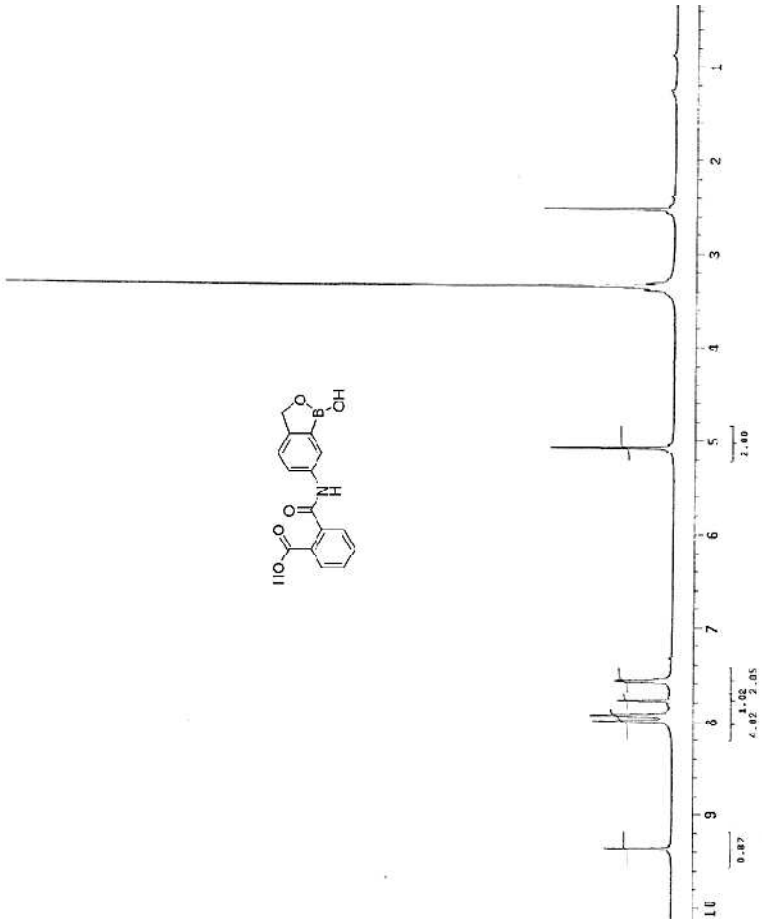

39.833
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40.166
40.334
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121.317
121.928
122.595
138.589
148.878
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140
160

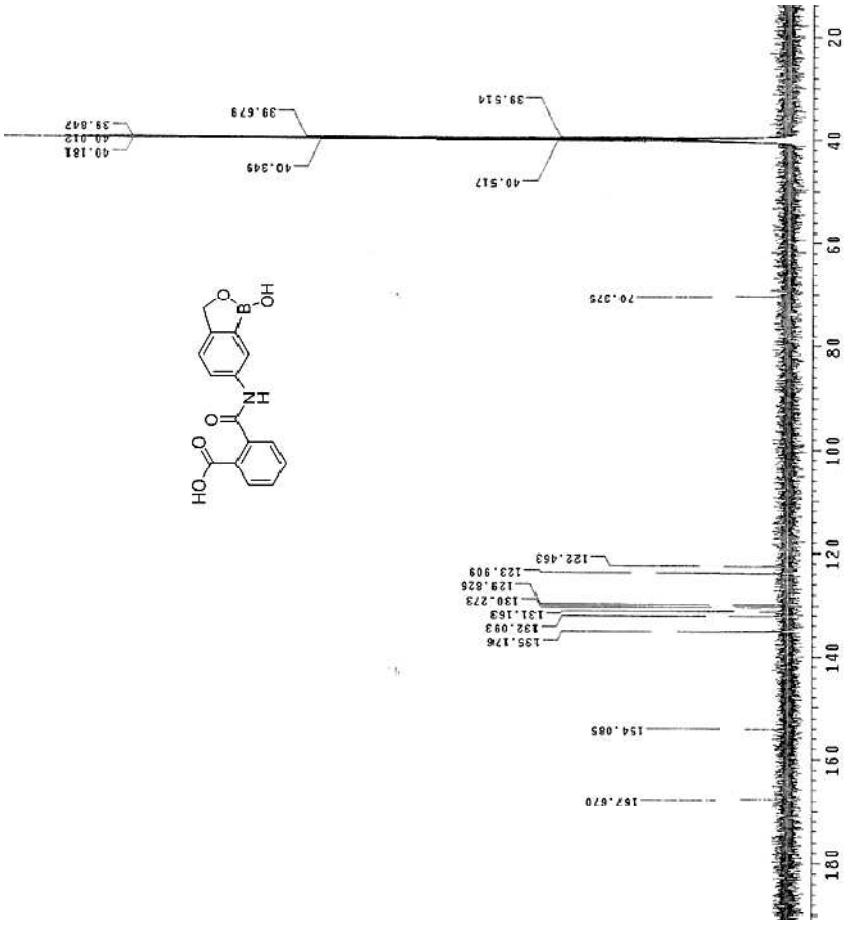
O
B
OH
Br
N
H
O
O
HO
O
1
2
3
4
5
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7
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2.00
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2.72
0.06

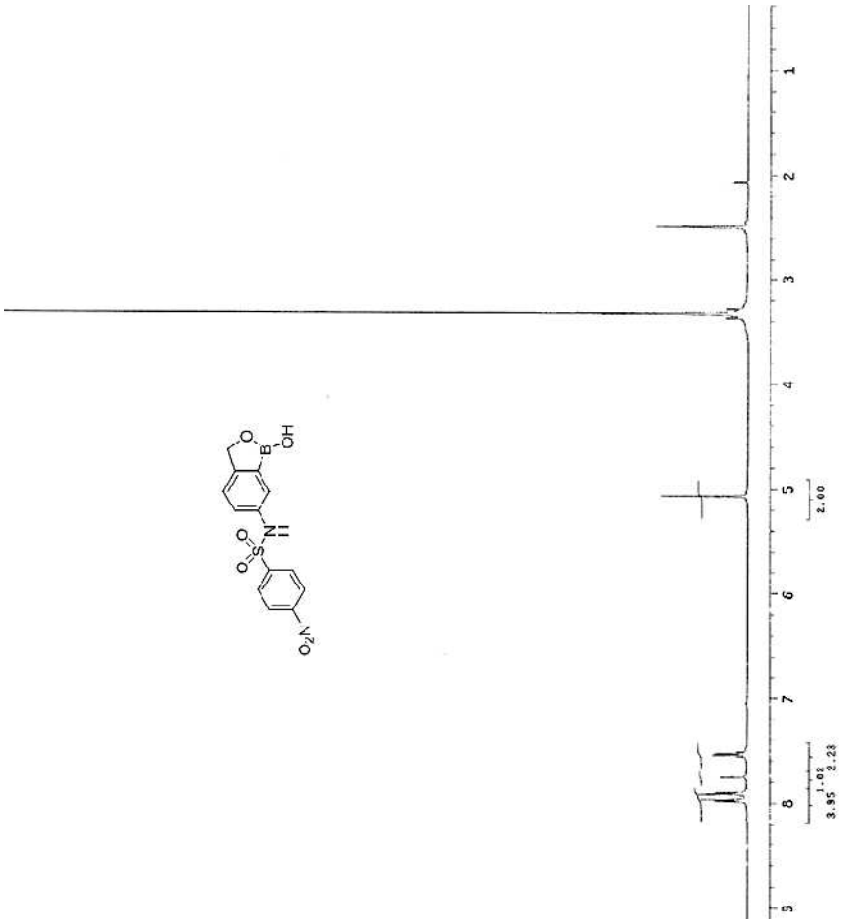

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40.342
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59.586
122.217
125.316
131.736
133.349
157.095
Br
B
OH
O
NH
HO

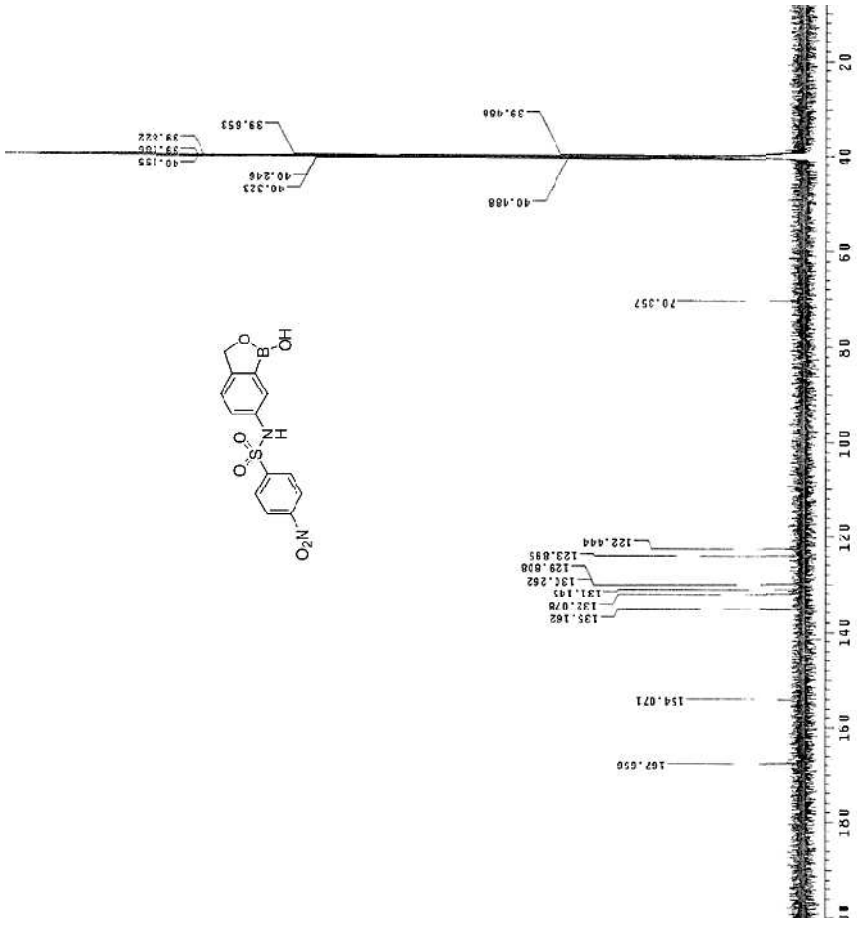


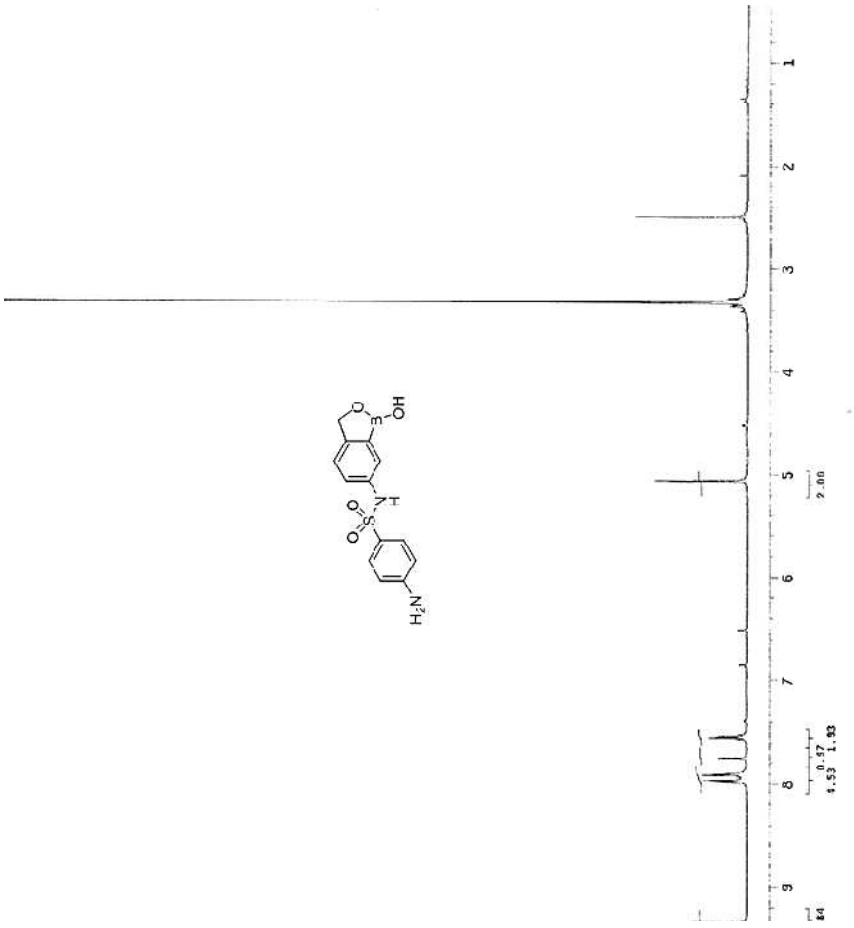

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70.225
70.071
105.000
132.514
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148.069
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140
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H
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O
O

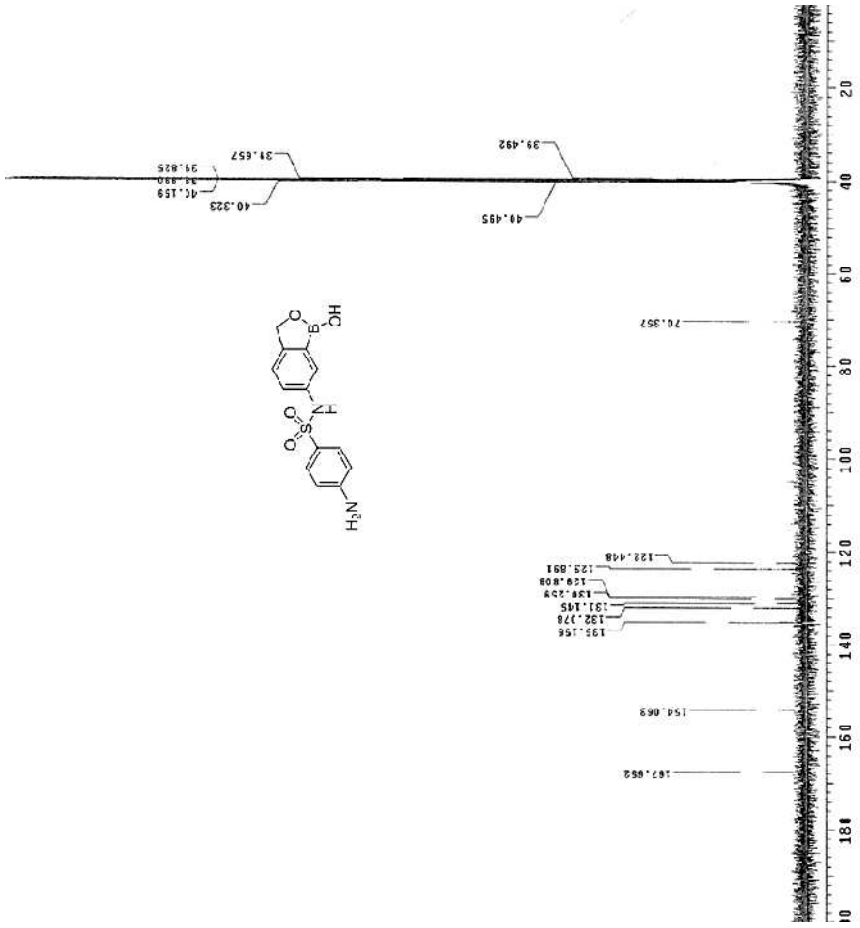


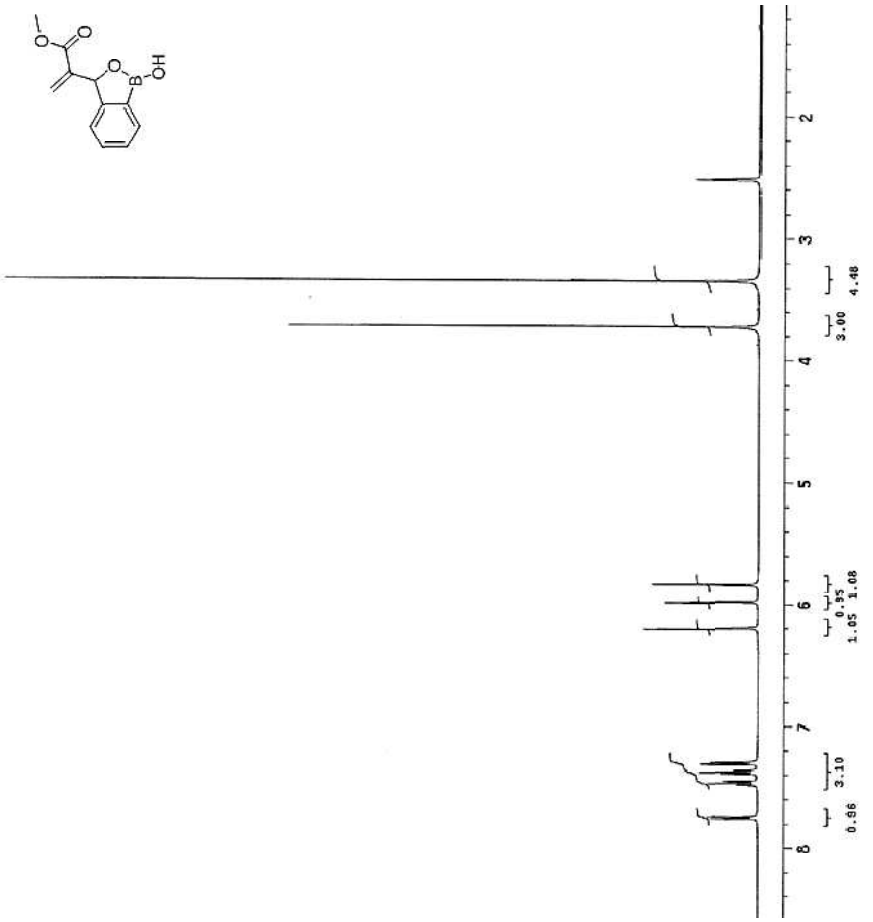

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131.163
132.093
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180
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NH
O
HO
O

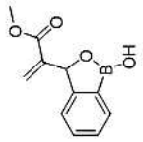

1
2
3
4
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7
8
2.00
1.01
3.95
2.23

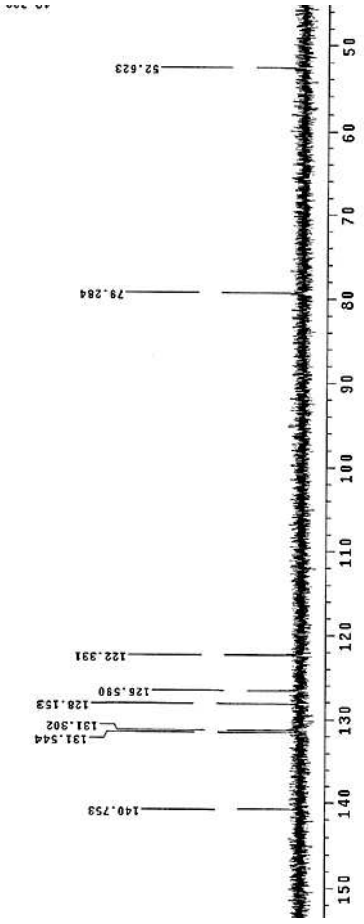

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H
S
O
O
O2N

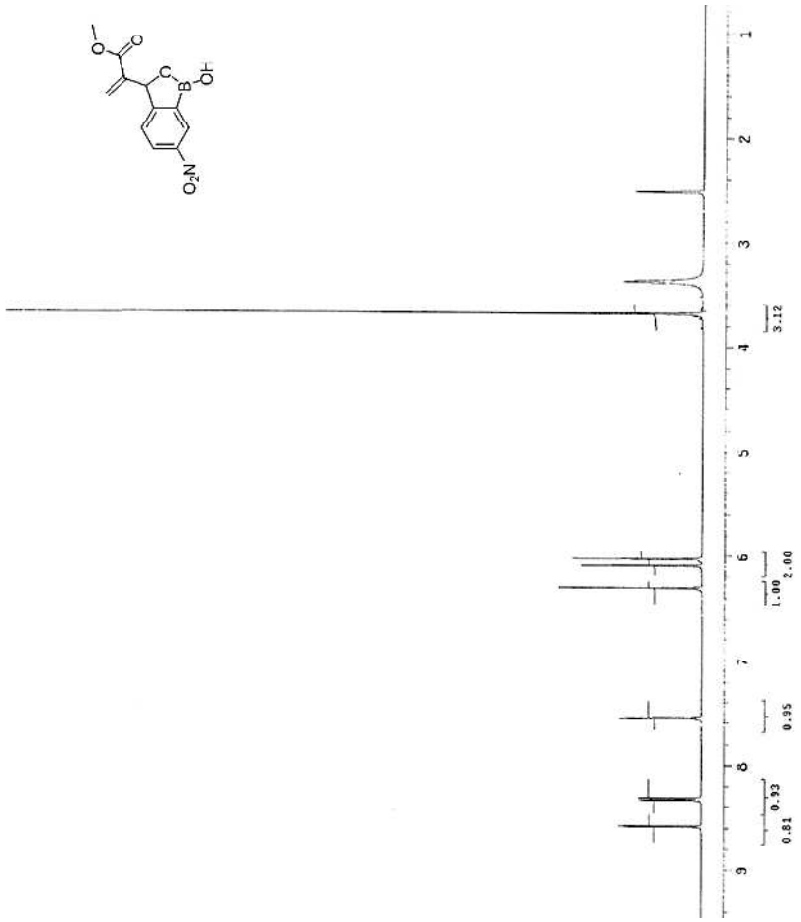


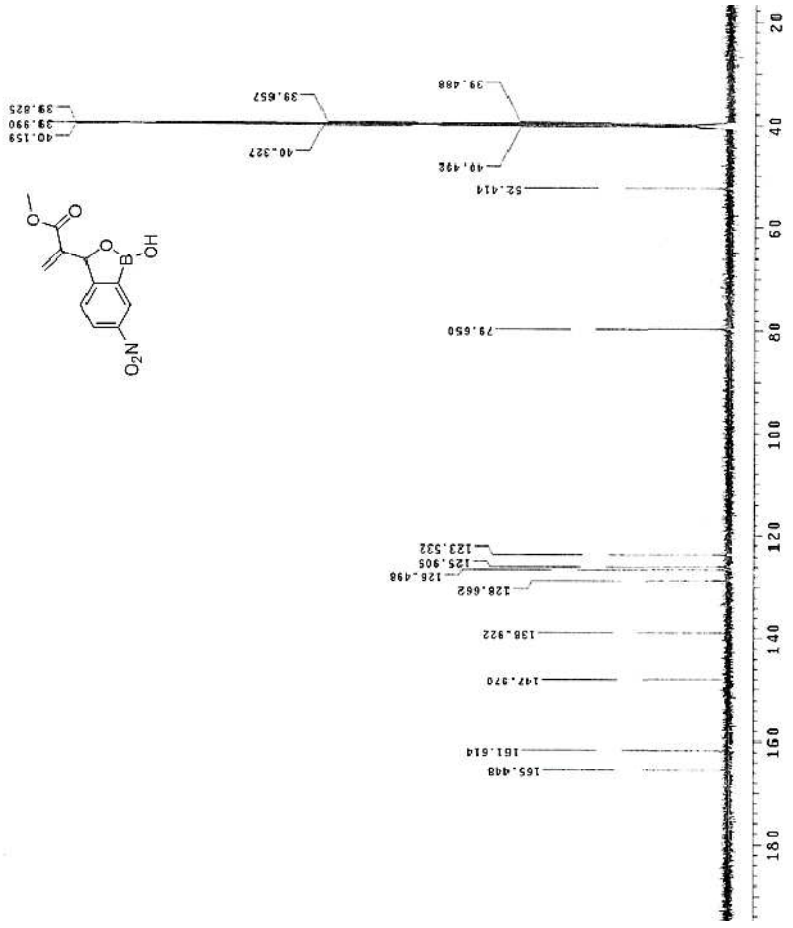

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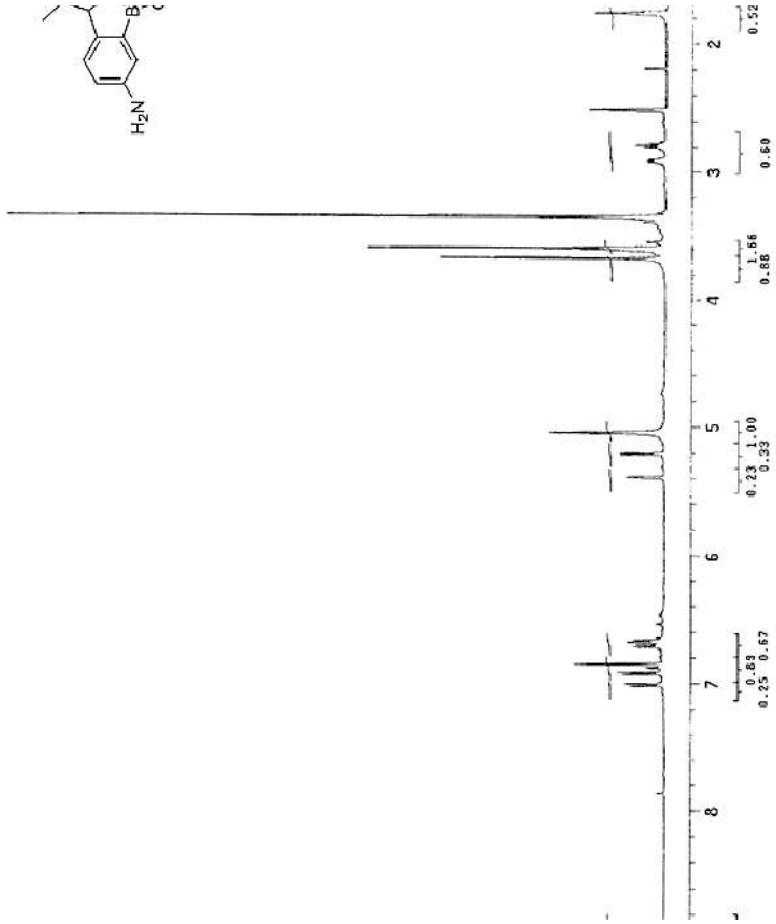

O
O
O
B
OH
2
3
4
5
6
7
8
4.48
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0.95
1.05
1.08
3.10
0.66

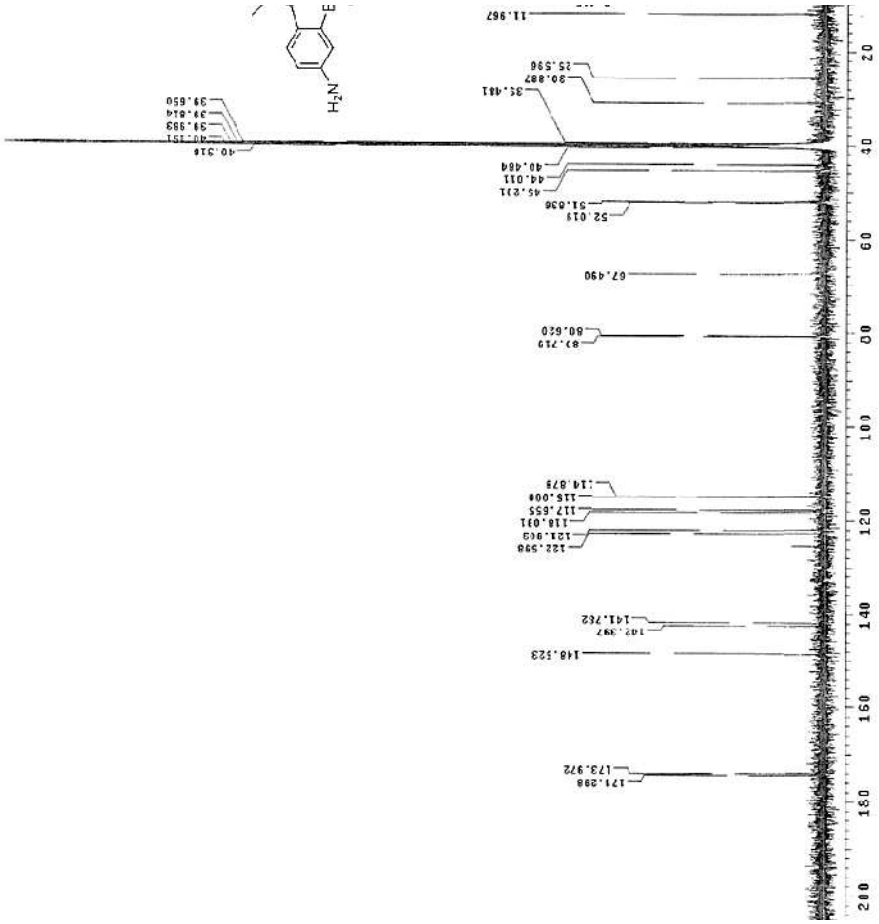


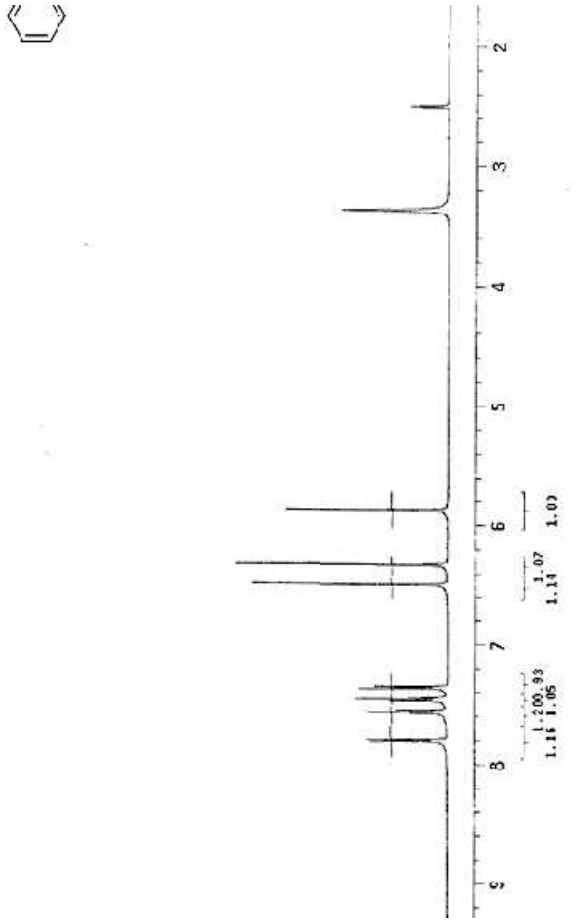

52.623
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150

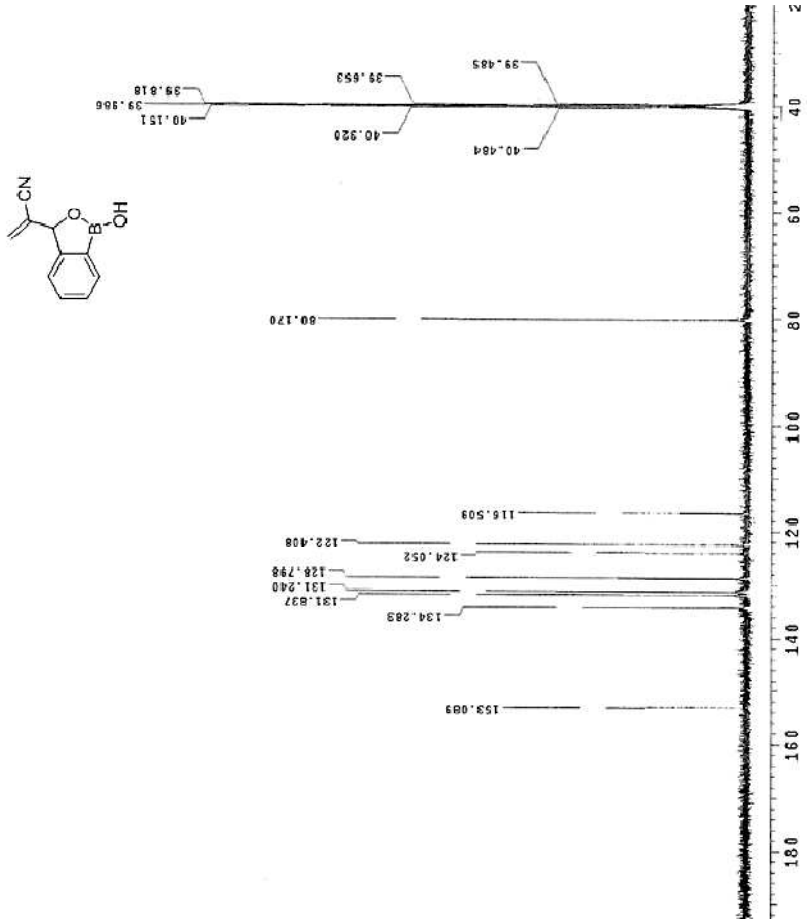

O
O
C
B
OH
O2N
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2
3
4
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6
7
8
9
3.12
1.00
2.00
0.95
0.93
0.81


39.488
39.657
40.327
49.498
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123.532
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126.498
128.662
138.922
147.370
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165.448
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O
O
O
B
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40
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180

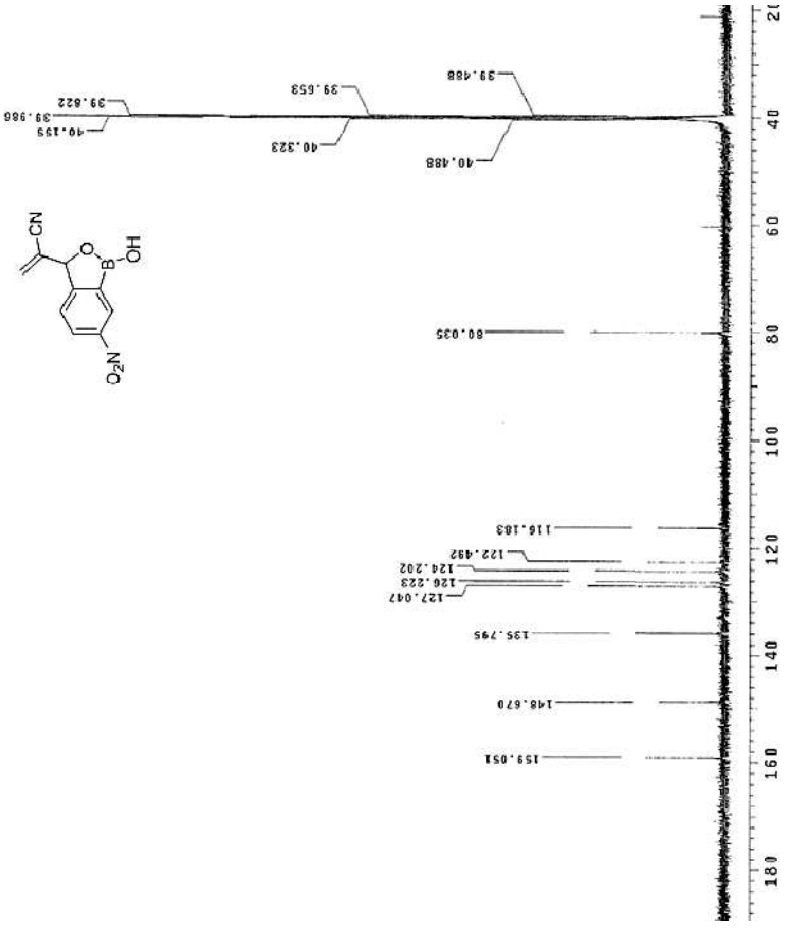

H_2N
2
3
4
5
6
7
8
0.52
0.60
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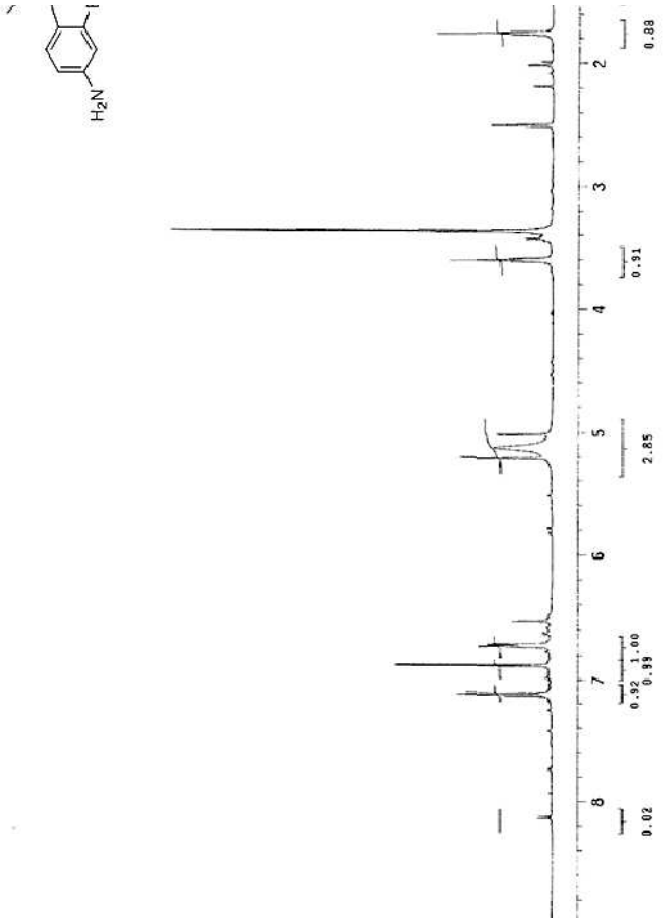

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67.490
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83.719
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118.000
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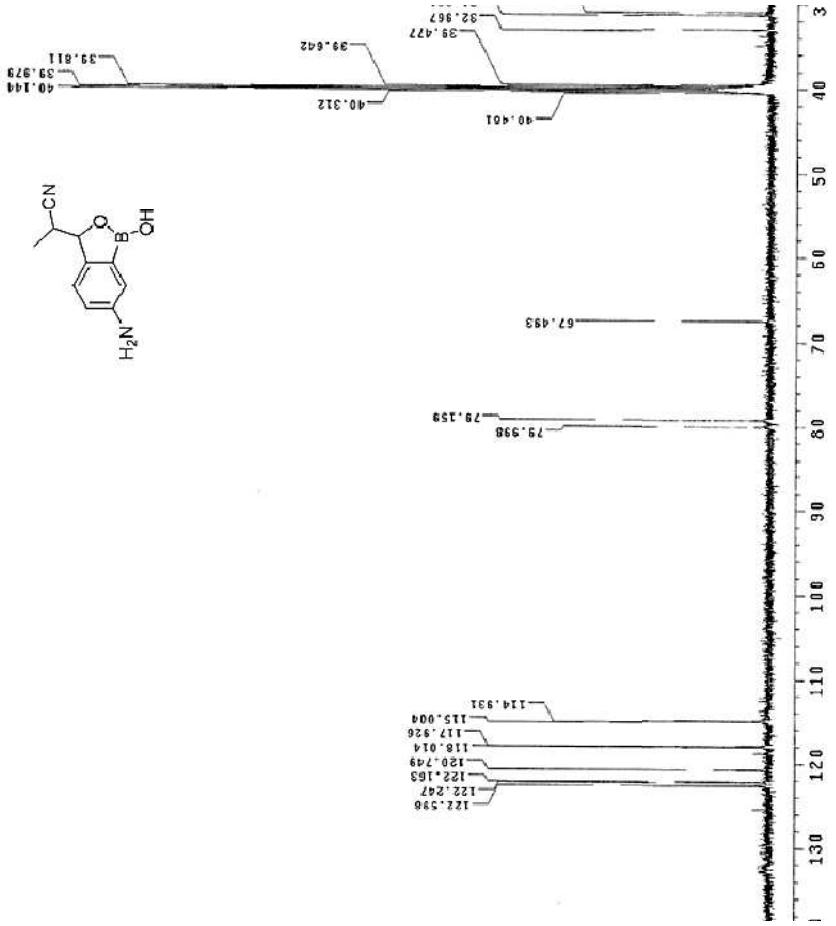

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131.837
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153.089
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80
100
120
140
160
180

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4
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2
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1.20
1.04


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CN
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B
O
O2N


H2N
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0.92 1.00
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CN
OH
B
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H_2N
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