SYNTHESIS OF BRANCHED ALKYL SUBSTITUTED POTENTIAL DRUG MOLECULES ACTIVE AGAINST PROSTATE CANCER

by

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Thanks my MOM and DAD

For their unconditional LOVE

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ABSTRACT

SYNTHESIS OF BRANCHED ALKYL SUBSTITUTED POTENTIAL DRUG MOLECULES ACTIVE AGAINST PROSTATE CANCER

Prostate cancer is the second most common malignancy and age related cause of death among males especially in the western world. It is claimed that there is a relation between the probability of getting prostate cancer and a high androgen level. Testosterone and Dihydrotestosterone are two prostate cancer responsible androgens that are synthesized in both testes and adrenal gland. Hence, it is possible to get rid of the prostate cancer via inhibition of the synthesis of androgens both in testes and adrenal glands where CYP17 is the enzyme responsible for the biosynthesis of testosterone and dihydrotestosterone.

In this study, synthesis of branched alkyl substituted potential drug molecules that might inhibit the CYP17 enzyme was targeted. Syntheses started with 4-amino-3-methoxybenzoic acid where first Fisher esterification method was applied. Then, the protection of the amine was achieved with AC_2O. The double addition of n-BuLi to the ester was the key step in obtaining the branched alkyl substituted phenyl derivatives. After the deprotection of the amide, the resulting amines were coupled with naphthoic acid derivatives. For the coupling reactions two different approaches were undertaken. In the first one, naphthoic acid was converted to naphthoyl chloride with $SOCl_2$ and then coupled with the amine derivative in the presence of triethylamine. The second approach was the *in-situ* conversion of the naphthoic acid derivatives to the corresponding naphthoyl bromides with PBr_3 and then coupling with the amine derivatives in the presence of triethylamine in one pot. Similar transformations were also carried out with 4-amino-2-methoxybenzoic as the starting material. The final products were sent to Koç University for the biological testings.

Docking and binding energy calculations of the final products were done by using AutoDock 4 computational program. It was observed that docking and binding energies of the final compounds obtained were better than the lead compound.

ÖZET

PROSTAT KANSERİNE KARŞI AKTİF, DALLANMIŞ ALKİL TAKILI MUHTEMEL İLAÇ MOLEKÜLLERİNİN SENTEZİ

Prostat kanseri tümörü dünyadaki ikinci en yaygın kötü huylu tümördür. Özellikle batı ülkelerinde daha yaygın olmak üzere, erkekler arasında yaşla ilerleyen ve en çok ölüme sebep olan kanserdir. Prostat kanserine yakalanma ihtimali ile yüksek androjen seviyesi arasında bir ilişki olduğu iddia edilmektedir. Testesteron ve dihidrotestesteron testislerde ve böbreküstü bezlerinde sentezlenen ve prostat kanserine sebep olan iki farklı androjendir. Bu nedenle hem testislerde hem de böbreküstü bezlerinde sentezlenen androjenlerin sentezini engelleyerek prostat kanserinden kurtulmak mümkündür. Ayrıca CYP17 testesteron ve dihidrotestesteron sentezinde görevli enzimdir.

Bu projede CYP17 enzimini inhibe edebilecek, dallanmış alkil takılı muhtemel ilaç moleküllerinin sentezlenmesi amaçlanmıştır. Başlangıç maddesi olarak 4-amin-3-metoksibenzoik asit seçildi ve bu maddeye Fischer esterifikasyonu yöntemi uygulandı. Daha sonra AC_2O kullanılarak amin korunması tepkimesi gerçekleştirildi. N-BuLi tepkimesi ile estere ikili bütil eklemesi dallandırılmıs fenil türevlerinin sentezlenmesinde önemli bir basamaktır. Amidden koruyucu grubun uzaklaştırılmasıyla oluşan aminler naftoik asit türevleriyle birleştirildi. Birleştirme tepkimesi için iki farklı yöntem denendi. Birinci yöntemde naftoik acid $SOCl_2$ kimyasalının varlığında naftoil klorüre çevrildi ve trietilamin eklenmesiyle aminle birleştirildi. İkinci yöntemde is naftoik asit çeşidi PBr_3 ile naftoilbromüre çevrildi. Daha sonra trietilamin ve amin türevinin eklenmesiyle birleştirme tepkimesi tek seferde gerçekleştirilmiş oldu. Benzer tepkimeler 4-amin-2-metoksibenzoik asit başlangıç maddesine uygulandı. Son ürünler biyolojik testler için Koç Universitesine gönderildi.

Son ürünlerin yaklaşma ve bağlanma enerjileri AutoDock 4 programı kullanılarak hesaplandı. Son ürünlerin yaklaşma ve bağlanma enerjilerinin öncül molekülden daha iyi olduğu görüldü.

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LIST OF ABBREVIATIONS

ACE	Angiotensin-Converting Enzyme
ADMET	Adsorption, Distribution, Metabolism, Elimination, Toxicity
ADT	Androgen Deprivation Therapy
BPH	Benign Prostatic Hyperplasia
$CDCl_3$	Deutorated chloroform
CH_2Cl_2	Dichloromethane
CYP	Cytochrome P-450
CYP 17	17 alpha-hydroxylase cytochrome P-450 (P-450(17) alpha)
DHT	Dihydrotestosterone
DMSO	Dimethyl sulfoxide
DRE	Digital Rectal Examination
EtOAc	Ethylacetate
IC50	50% Inhibition of Concentration
LHRH	Luteinizing Hormone-Releasing Hormone
MeOH	Methnaol
NMR	Nuclear Magnetic Resonance
PCF	Prostate Cancer Foundation
PSA	Prostate Specific Antigen
QSAR	Quantitative-Structure Activity Relationship
SAR	Structure-Activity Relationship
SBDD	Structure-Based Drug Design
SEER	Surveillance Epidemiology and End Results
TEA	Triethylamine
THF	Tetrahydrofuran

TLC	Thin Layer Chromatography
TRUS	Transrectal Ultrasound
WHO	World Health Organization

1. INTRODUCTION

1.1 Cancer

Cancer is the general name for diseases in which abnormal cells in a tissue grow and divide without control and invade other tissues. Normal cells grow, divide and die in a normal way however, cancer cells grow but do not die, they form new abnormal cells. According to World Health Organization (WHO), cancer is responsible for around 13 per cent of deaths through worldwide and it accounts for 7.6 million deaths. [1] Moreover, more than 11 million people are diagnosed with cancer in each year. It is predicted that, deaths from cancer will proceed by increasing through worldwide and will be 12 million deaths in 2030. WHO also states that; about 72% deaths from cancer occurred in low and middle income countries in 2007. [1] According to Surveillance Epidemiology and End Results (SEER) statistics from 2003 to 2007, 66 years of age is the median age at diagnosis of cancer of all sites. Approximately 1.1% were diagnosed under age 20; 2.7% between 20 and 34; 5.7% between 35 and 44; 14.0% between 45 and 54; 22.3% between 55 and 64; 24.7% between 65 and 74; 21.8% between 75 and 84; and 7.7% 85+ years of age. Estimation of SEER consistent with statistics is that 1,529,560 men and women (789,620 men and 739,940 women) will be diagnosed with and 569,490 men and women will die of cancer of all sites in 2010 [2].

There are two changes that cause more threatening, or malignant tumors formation:

- Cancerous cells demolish healthy organs by administering to travel through organs and lymph systems.
- When a cell starts to administrate to divide and grow, producing new blood vessels to benefit from, angiogenesis arise [3].

Although there is not any plain explanation why some people develop cancer cells but others do not, there are many scientifically supported factors that increase the chance of developing cancer. Cancer causes may be categorized as physical carcinogens such as ultraviolet and ionizing radiation; chemical carcinogens such as asbestos, components of

tobacco smoke, aflatoxin which is a food contaminant and arsenic which is a drinking water contaminant; biological carcinogens such asinfection from certain viruses and bacteria [4]. Moreover followings are determined as risk factors for cancer: [5].

- Tobacco use
- Acrylamide and other chemicals in food
- Weight and physical activity
- Genetics
- Chemicals and other substances
- Growing old
- Radiation
- Alcohol

Treatment of cancer changes according to type of cancer. Surgery, radiation, chemotherapy, immunotherapy, hormone therapy, or gene therapy are categories that are used for the treatment of cancer.

1.2. Prostate Cancer

Prostate cancer is one of the most common malignancy and age related cause of death among males especially in the western world. [6] Actually it is second leading cause of death from cancer after lung cancer according to the American Cancer Society [7].

According to the US National Cancer Institute in 2007, the U.S. proceeding rate for prostate cancer was around 166 new cases diagnosed per 100,000 men; the death rate was approximately 24 deaths per 100,000 men [8].

Prostate cancer often grows very slowly and may not cause significant harm. However, some prostate cancer types are very combative and they can spread very quickly without treatment. Although at the beginning stages it is not possible to observe symptoms of prostate cancer, at later stages symptoms can be stated as follows:

- Often urination
- Difficulty to start and stop urination
- Weak or fitfully urinary flow
- Hurtful sensation during urination
- Blood in urine or semen [9]

Androgens are steroid based compounds that affect masculine characters of males [10]. Androgens are synthesized in testes with approximately 90% and in adrenal glands with 10% [6]. They are thought to be a factor that affects approximately 80% growth of normal prostate tissue and prostate cell [6]. So, androgen deprivation may be considered as an important therapeutic strategy for inhibition of the tumor growth [11].

Testosterone and 5α-dihydrotestosterone (DHT) are two kinds of androgens that are produced in testicles and provide and preserve male sex characteristics [12]. It is suggested that their levels in blood is proportional to the androgen biosynthesis thus to the risk of getting prostate cancer [13, 14]. It is also indicated that giving high serum concentrations of free testosterone to blood may increase prostate cancer risk [15].

According to Prostate Cancer Foundation (PCF) it is possible to delay and prevent prostate cancer by considering some facts like age, race, family history and the living area [16]. Moreover, PCF gives eating and living suggestions such as; eating more fish and broccoli, taking fewer calories and doing exercises, watching vitamins and minerals intake like calcium and avoiding smoking cigarette and stressful life to prevent getting prostate cancer [16].

1.3. Diagnosis of Prostate Cancer

Patients diagnosed with prostate cancer fall into one of the following four stages;

- Stage A: Incidentally discovered and clinically undetectable
- Stage B: Tangible tumor, captive to prostate gland
- Stage C: Locally invading tumor
- Stage D: Metastatic tumor [17].

Prostate specific antigen (PSA) screening, digital rectal examination (DRE), and transrectal ultrasound (TRUS) and biopsy are three methods for diagnosis of prostate cancer [18].

It has been claimed that PSA production is related with androgen, so having prostate cancer risk probability is related with the PSA level. [19] PSA level in the blood is determined via PSA test and the disease is identified progressively at earlier phase [20].

DRE is used to check the health of the internal organs such as to examine whether the urinary and reproductive organs in the pelvic region are in well condition [21]. It is an important method for early detection of lesions and tumors along with the PSA screening [21].

TRUS is used as a diagnostic and locator tool and it is implemented by inserting an ultrasound probe into the rectum and releasing ultrasound waves and reflecting the image of the rectum on a screen [18].

1.4. Treatment of the Prostate Cancer

There are many methods that are used for the treatment of the prostate cancer. Treatment options can be stated as follows;

- Prostatectomy (Surgery)
- Radiation Therapy
- Hormone Therapy
- Chemotherapy
- Cryosurgery
- Biologic Therapy
- High-Intensity Focused Ultrasound
- Proton Beamed Radiation Therapy [22].

Although there are so much treatment options, it is difficult for doctors to choose most suitable one for the patients since patients age, co-morbidity, life expectancy, personal

preference, clinical stage, tumor grade and potential benefits and risks change from person to person [17].

1.4.1. Basis for the Treatment of Prostate Cancer

As stated before, it is thought that androgens most likely contribute to the risk of getting prostate cancer in some men. Testosterone and DHT are important androgens. In males, androgen is mainly synthesized in testes, and after its synthesis it is converted to DHT, which is an active metabolite, in an external, outside tissue [23]. Since the androgen removal cause regression of prostate cancer, aim of the treatment becomes androgen suppression [24]. Thus androgen suppression will lead prostate cancer cells to shrink or grow slower. On the other hand, testes are not the only location where testosterone is synthesized, testosterone is also synthesized in adrenal glands and so, adrenal glands also give rise to androgen production [23]. Hence, for androgen extermination both testes and adrenal glands should be taken into consideration and it can be stated that the target for the treatment of the prostate cancer may be determined as the testes and adrenal glands.

1.4.2. Androgen Removal

The purpose of androgen removal is to reduce the amount of the androgen in the blood stream which leads to the deprivation of many prostate cancer cells off the fuel they need to survive [24]. Androgen removal used to be done with orchiectomy that is a kind of surgery in which one or both of testes are removed so that important source of male hormone production is decreased [22]. As a result of androgen removal it is detected that level of PSA is decreased in the blood stream of patients and also, malignant tumor growth is slowed down and an increase at survival of patients is observed [25]. As declared before testes are not the only source of androgen, adrenal glands also synthesize androgen hormones, so orchiectomy is not capable of removing the androgen synthesize entirely.

1.4.3. Hormone Therapy

Hormone therapy is used either to stop or reduce the amount of excreted hormones that are responsible for the prostate cancer, so that extension of malignant tumor can be slowed down [22]. Ways to interfere with hormones can be stated as following;

- Using antiandrogens drugs that block androgen action
- Using luteinizing hormone-releasing hormone [LHRH] agonists ketoconazole and aminoglutethimide drugs that disrupt testosterone synthesis
- Taking estrogen

Once the androgen deprivation therapy (ADT) is used, there would be three consequences; cancer tumor will die (called apoptosis), it will not die but also it will not grow, so stay the same, or it will continue to grow bigger [23]. Hence, it can be stated that there are such prostate cancer cells which grow independent from the androgen level in the blood stream and hormone therapy alone is not a solution for the treatment of the prostate cancer.

Moreover, the drugs used during hormone therapy have significant side effects, for example ketoconazole, which is an imidazole antifungal agent, intervenes with steroid genesis [26]. Hence besides the interruption of the androgen synthesis, interruption of the estrogens, progesterone, corticoids, cortisol and aldosterone synthesis are also caused [26]. Since ketoconazole have no selectivity on inhibition of the P-450 enzymes and leads dangerous hepatic dysfunction and gastrointestinal disturbances and thus its usage is limited in many clinical studies [25].

Treatment options above shows that they are no smooth solutions for the treatment of the prostate cancer since they have unwanted side effects. Hence, new therapeutic methods which will not have serious side effects need to be developed.

1.5. New Therapeutic Approaches

Androgen deprivation therapy has been one of the focuses in order to block or slow down the androgen synthesis that is synthesized in both testes and adrenal glands for more than last two decades [27]. Hence, testosterone and DHT synthesis in both adrenal glands and testes are being examined.

It is claimed that androgens play a critical role in the development and maintenance of sexual characteristics in human males. However, they are also growth factors for such severe and widespread diseases as benign prostatic hyperplasia (BPH) and prostate cancer [28]. Moreover, since the androgen enzyme inhibition will lead less testosterone and DHT it is also claimed that inhibitors would also be effective for the treatment of diseases related with excess androgen amount in woman [28]. Hence, main therapeutic option for the treatment of the prostate cancer is the termination of the biosynthesis, release or action of androgenic hormones, which are testosterone and DHT, so that malignant tumor will have possibility to shrink, or stop growing.

1.5.1. Testicular and Adrenal Androgen Biosynthesis

Surgical removal of testes decreases 90-95 percent daily testosterone biosynthesis, but it does not stop the growth of malignant tumor because testes are not the only organs that synthesize testosterone [6]. As mentioned before 5-10 percent of androgens are synthesized in adrenal glands and so, antiandrogens were started to be used for further inhibition of the tumor growth. However, as mentioned before they have severe side effects. Hence, new alternatives target the family of cytochrome P-450 (CYP) enzyme system is suggested [6]. P-450 enzyme system has a single iron protoporphyrin IX prosthetic group and it is large and omnipresent family of medium size proteins [29]. Also it is stated that the cytochrome P-450 monooxygenase enzyme system (P-450) is involved in the synthesis and/or degradation of a large number of endogenous compounds and in the biotransformation of drugs and other xenobiotics [29].

Cholesterol is the starting material for the biosynthesis of androgens in both testes and adrenal glands [11]. After several enzymatic transformations that are catalyzed by P-

450 androgenic steroids are synthesized [11]. Through those mentioned enzymatic transformation CYP 17 catalyze two reactions the 17α-hydroxylation of pregnenolone and progesterone to the corresponding 17α-alcohols and the subsequent 17,20-lyase reaction cleaving the C17-C20 bond [6]. Hence, 17-keto androgens androstendione and dehydroepiandrosterone leads to synthesis of testosterone with all other androgens as shown at Figure 1.1 [30].

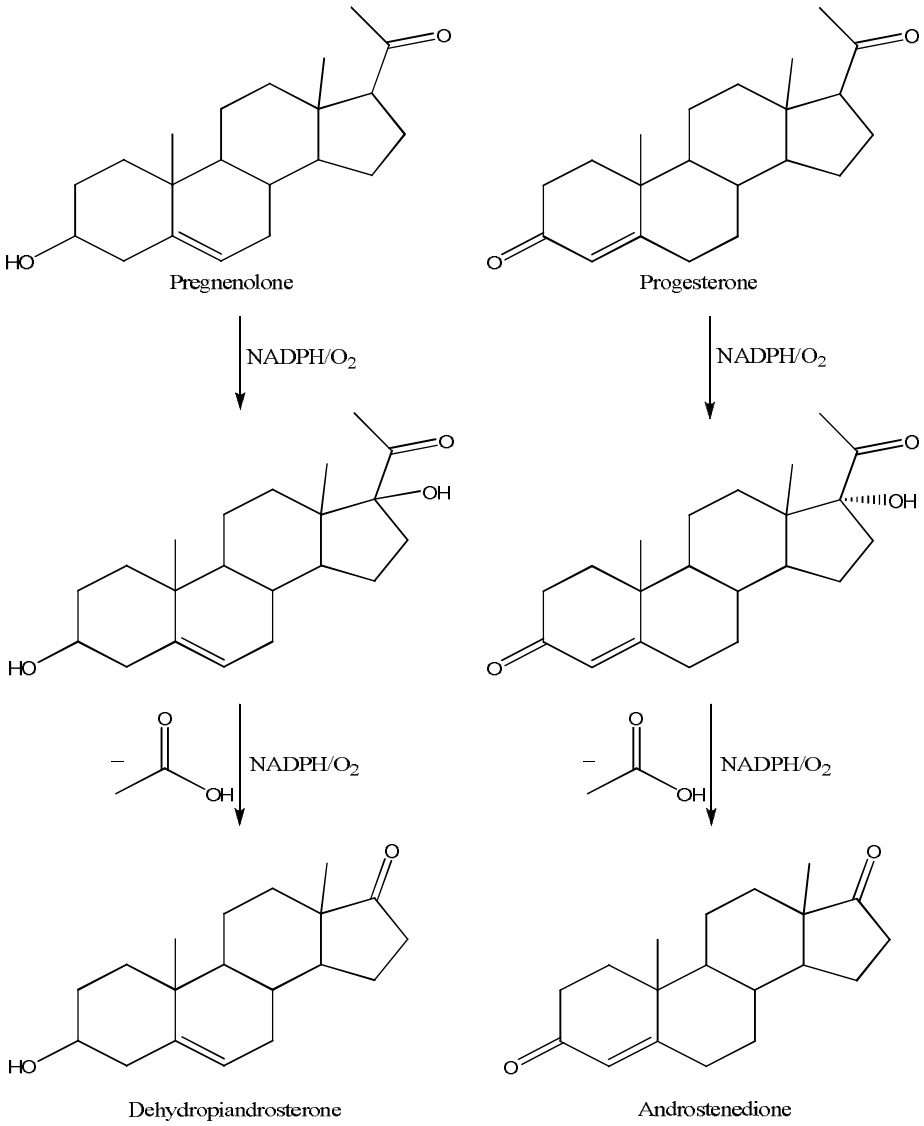


Figure 1.1. CYP 17 catalyzed androgen synthesis.

1.5.2. Identification of the Target Enzyme

Since the androgen receptor antagonists and gonadotropin-releasing hormone analogues have disadvantage like affecting only testes and not diminishing the concentration of androgen, new target is needed such that will both stop the androgen synthesis from testes and adrenal glands [31].

The new target for the androgen suppression was determined as the 17a-hydroxylase-17,20-lyase (CYP 17) [31]. As it is stated before CYP 17 catalyzes the conversion of 17a-hydroxypregnenolone and 17a-hydroxyprogesterone into the weak androgens, dehydroepiandrosterone and androstenedione, respectively, in testes and adrenal glands. Moreover, those weak androgens are turned into the more effective androgens like testosterone and DHT [32]. The relation of CYP 17 enzyme and androgen synthesis in testes and adrenal glands is clearly shown in the following Figure 1.2 [33].

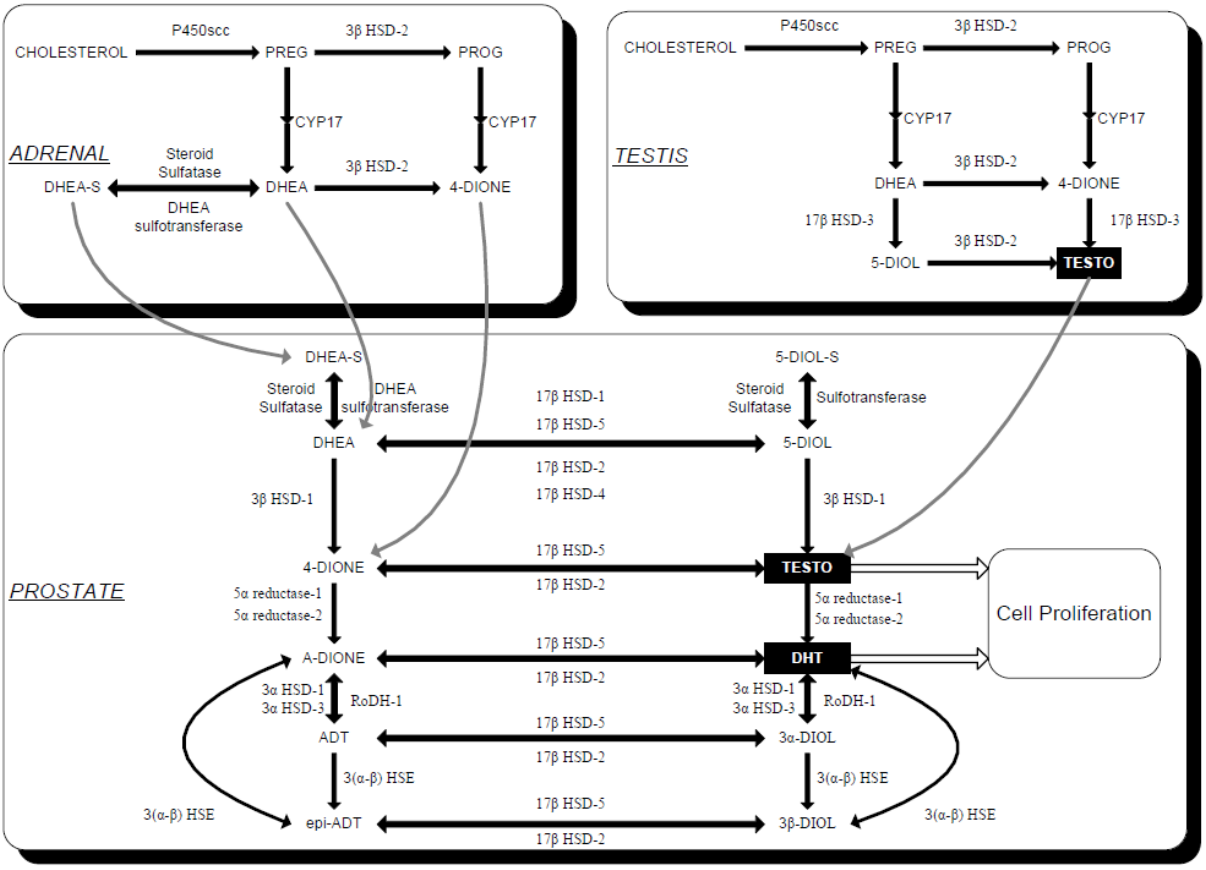


Figure 1.2. Enzymatic pathway of androgen biosynthesis in testes and adrenal gland.

As it is simply followed from figure 1.2 CYP17 is the second enzyme that results in the synthesis of testosterone and DHT in both adrenal glands and testes. Hence it can be concluded that CYP17 is new target for therapeutic treatment of prostate cancer since inhibition of CYP17 enzyme inhibits androgen synthesis.

1.6. Drug Discovery, Design and Optimization

1.6.1. Drug Discovery

First step of the drug discovery and design starts with choosing a disease that is important. Choosing and discovering the drug targets is the second step and in that step identification of which biomacromolecules are involved in that disease is crucial [34]. According to that biomacromolecules lead compound choice will be done. Lead compound may be defined as a structure that shows a pharmacological activity and lead compounds can be thought as the starting points for the drug design [34].

1.6.2. Drug Design

The following table shows the possible steps of drug discovery and the average time length corresponding to each step [35].

Table 1.1. Drug Discovery Steps.

STEPS	YEARS
Discovery and lead generation	1-2
Lead optimization	1-2
In vivo and in vitro assays	1-2
Toxicology trails	1-3
Human safety trails	1
Human efficacy trails	1-2
Total development time	6-12

As it is claimed before once the lead compound is identified, drug design takes the next place. So far, random screens or exploiting information about macromolecular receptors were two methods for drug discovery and design [35]. However, nowadays after lead compound structure determination, structure-activity relationship (SAR), which correlates important parts of the molecule to the biological activity, become the new tools of the drug design [34]. Examination of important parts of molecule means changing some functional groups in the lead compounds so that greater selectivity and less chance of side effects will be achieved.

1.6.3. Drug Optimization

Finding a drug molecule that fits into the enzyme is the part of optimizing target interactions. With the purpose of finding best fitting enzyme extension tactics are used. For instance, angiotensin-converting enzyme (ACE) inhibition is achieved with extension of antihypertensive agents. Addition of a phenylalkyl group to a lead compound (I) in Figure.1.3 [34]. increases its inhibition 1000 more. Its indication is that extra aromatic ring binds the hydrophobic pocket and fills inside of that hydrophobic pocket [34].

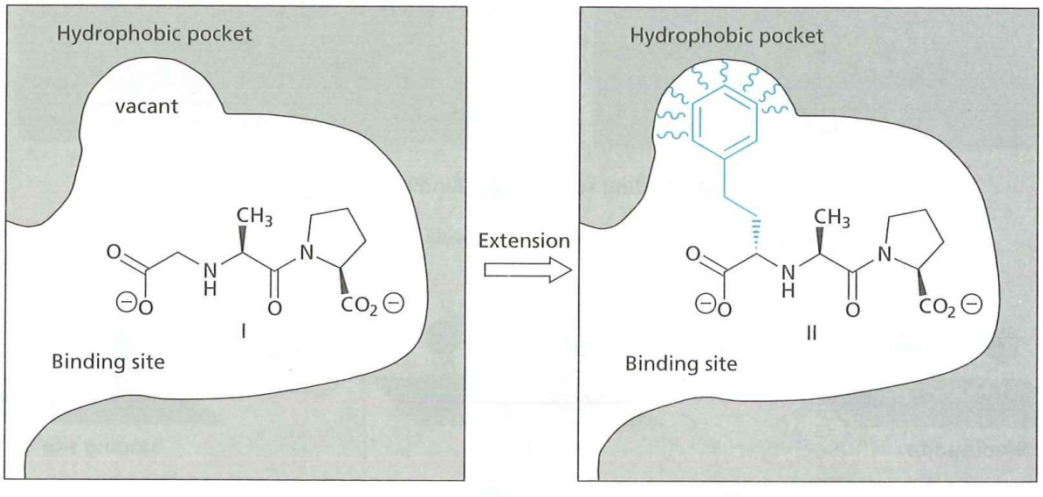


Figure 1.3. Addition of phenylalkyl group to lead compound (I).

Besides addition of new groups to a lead compound, chain extension and ring expansion tactics are also applied to have better inhibition and best fit inside of the target enzyme.

Moreover, changes done on a lead compound of a molecule gives chance of following changes on IC_{50} values. For example changes done on two lead compounds against kinase enzyme (developed by Astex and Vernalis/Novartis respectively) shows that it is possible to increase the activities of the drugs by 1600-2500 times as can be seen in Figue 1.4.

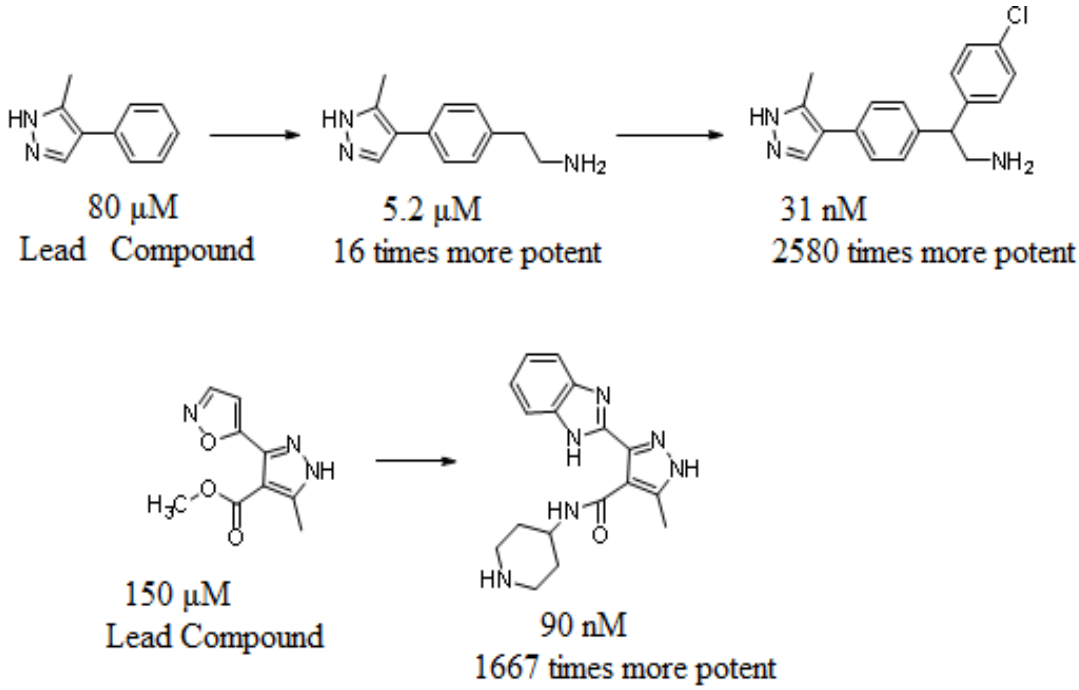


Figure 1.4. IC_{50} value changes according to changes on lead compound.

1.6.4. Structure-Based Drug Discovery and Design

Together with random screens or exploiting information about macromolecular receptors with specialized computer programs introduce structure based drug design (SBDD) [35]. Moreover, the process of structure based drug design requires the followings;

- Reveal a proper protein target
- Constitution of the 3-D structure of the target protein
- Implementation of an easy and reliable, high-throughput screening assay
- Identification of a lead compound
- Determination of computer assisted methods for estimating the relationships of new compounds
- Access to a synthetic route to synthesize the designed compound [36].

3-D structure of the target protein is constituted by X-ray crystallography, nuclear magnetic resonance (NMR), or homology modeling [37]. After identification of target protein, virtual screenings of drug candidates on regulatory or active side of the target protein is done via using docking analysis [37]. Then, steric and electrostatic interactions of candidate molecules are calculated via program AutoDock4 [34]. At next stage selected drug candidates that show relatively high binding and docking energies are tested *in vitro* and *in vivo* assays [37].

The following figure (Figure 1.5) shows the general approach to the structure-based design of inhibitors [35].

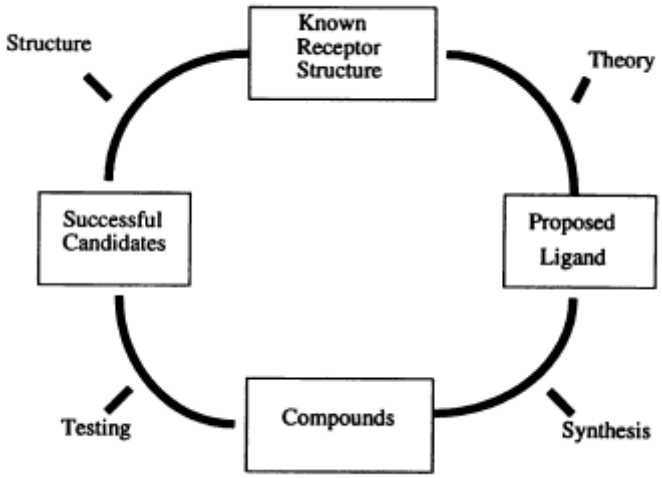


Figure 1.5. General approach to the structure-based design of inhibitors.

1.7. Modeling and Inhibitors of Cytochrome P450

As stated previously, the cytochrome P450 monooxygenase 17αhydroxylase/17,20-lyase (CYP17) is an enzyme that catalyzes the hydroxylation of progesterone and pregnenolone into the corresponding 17 α-products, as well as the cleavage of the C17-C20 bond to yield androstenedione and dehydro-epiandrosterone [38]. These steroids are weak androgens which subsequently are converted by other enzymes to the most potent androgens testosterone and DHT [32].

1.7.1. Computational Modeling of the Enzyme CYP17

Computational modeling of the CYP17 enzyme is done at Koc University by computational chemists. Since the crystal structure of the CYP17 enzyme has not been reported in the literature computer generation of CYP17 enzyme is done according to II P450 crystal structure, P450BMP [38]. The researchers (Prof.Metin Türkay, Assoc. Prof. Halil Kavaklı) also worked on CYP17 enzyme model and identified the interaction types between the enzyme and potential drug candidates. They have also examined other models for cytochrome P450 and concluded that previous models have smaller substrate binding pocket and those models just allow to planar substrates like steroids to fit [38].

According to all computational chemists who studied on CYP17 enzyme structure, CYP17 enzyme consists of a hydrophobic region and a heme group which is common in all P450 enzymes [30]. The following structure, Figure 1.6 [37], is computational generated model of the CYP17 enzyme and named as 2c17 in protein data base and its substrate binding pocket is placed in the cavity above the heme group [38].

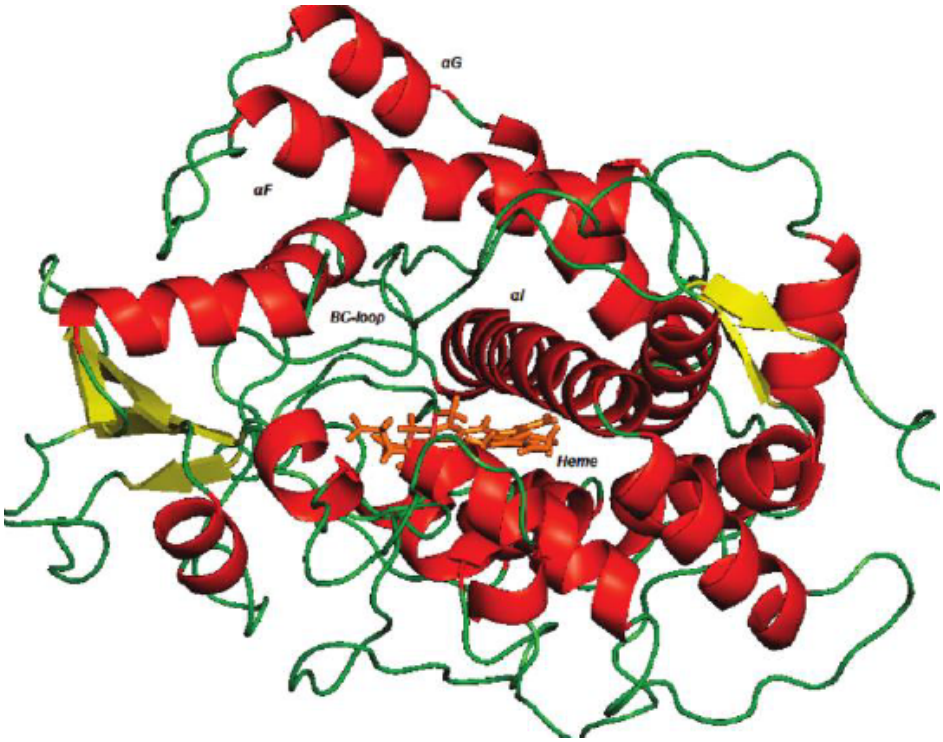


Figure 1.6. Computer generated model for cytochrome P450 with PDB access code, 2c17.

Computer generated model for cytochrome P450 with PDB access code, 2c17.

Enzyme structure is very important in terms of understanding the catalytic activities, substrate and reaction selectivity [38]. It is not possible to optimize substrate without the model of the enzyme to be inhibited. Hence, according to 2c17 structure lead compound optimization will be done so that improvement of the activity and selectivity and minimization of side effects is achieved [34].

1.7.2. Selection of Inhibitors for CYP17 Enzyme

Inhibition of the enzyme CYP17 means finding drug candidates that will bind to the active side of the CYP17 and stop biosynthesis of testosterone and DHT. Drug candidates have different interactions with the active side of the enzyme such as blocking, accelerating, decelerating, reversing or initiating reactions according to treatment type of the diseases [38]. After calculations of the drug candidates' docking and binding energies, ADMET (adsorption, distribution, metabolism, elimination, and toxicity) and IC_{50} calculations are also done. Lastly, lead optimization is completed with quantitative-

structure activity relationship (QSAR) that is used in structurally similar drug candidates which then draws attention to the specific functional groups of the lead compound [37]. According to literature, it can be claimed that steroidal drugs have ability to interact with some other proteins because of strong affinity to the steroidal compounds [11]. It means steroidal drug candidates are not selective and should be replaced by steroidomimetics that will lead to fewer side effects.

After all above calculations and screenings of drug candidates, lead compound in Figure 1.7. was found with docking energy and binding energy values -10.82 kcal/mol and -8.90 kcal/mol respectively and its IC_{50} value is 35μM *in vivo* testing.

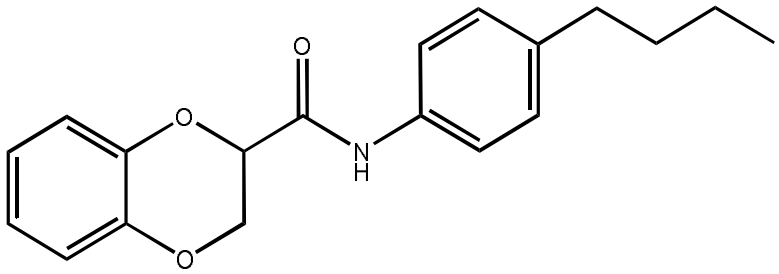


Figure 1.7. Structure of the Lead Compound.

1.7.3. Substituents on the Lead Compound and Their Possible Functions

In order to find best directions for further synthesis, lead optimization have to be done with the help of repeated cycles of design, experimental selection, change in the functional groups of the structure and remodeling and calculations of the affinities [36].

Optimization process was also implemented to our lead compound and some changes on the structure were done. Following Figure 1.8 shows the main functional groups of the lead compound and their possible functions.

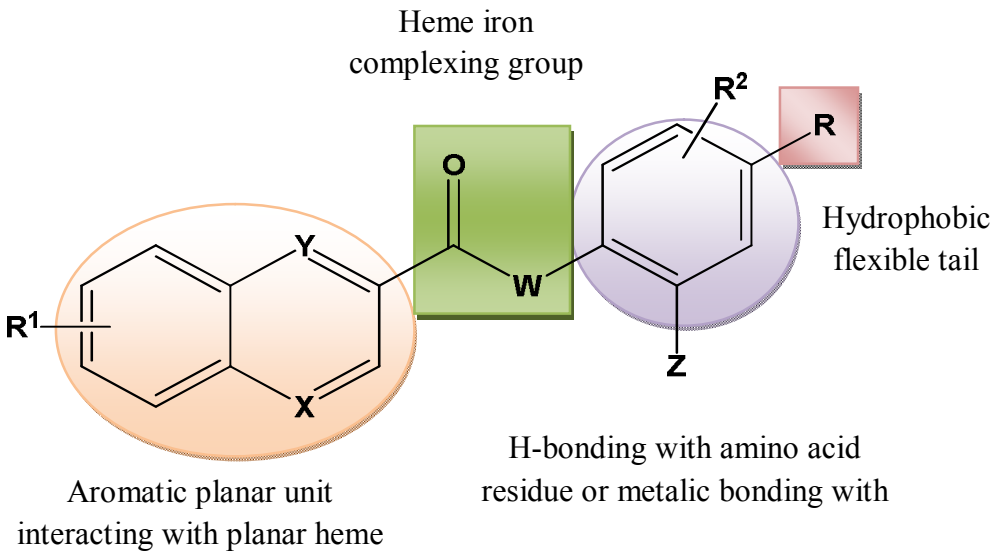


Figure 1.8. Substituent atoms on the lead compound and their possible functions.

Following Table 1.2. shows the possible change that will be done on substituent parts of the lead compound. This stage also may be considered as a part of the lead optimization step.

Table 1.2. Possible changes on substituent parts of lead compound.

X and Y atoms	C, O
W atom	N, O, C
Z group	-OR, -NR_2(R= -H, Alkyl)
R	C2-C6 alkyl or alkoxy, Branching alkyls
R_1	-MeOH, F (at different positions)

1.7.4. Docking and Binding Energy Calculations

Binding energy is the strength of interaction between the enzyme and the candidate drug molecule. Docking energy is related to the approach of the candidate drug molecule to the enzyme. Following table shows docking and binding energies of the lead compound and its' possible derivatives. Calculations were done by using AutoDock 3.05. computational program at Koc University.

Table 1.3. Docking and binding energies of lead compound.

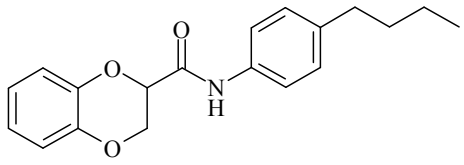
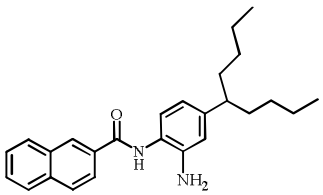
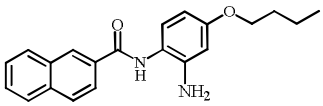
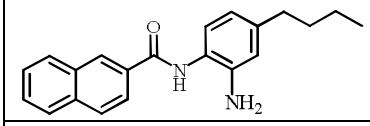
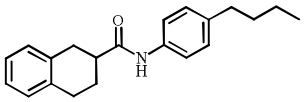
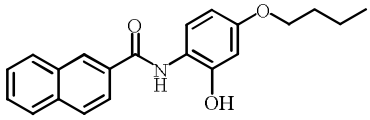
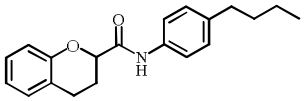
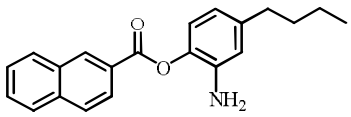
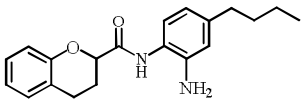
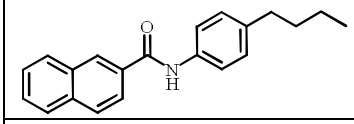
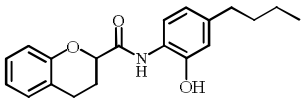
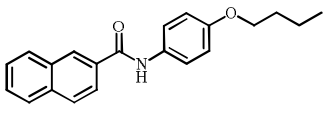
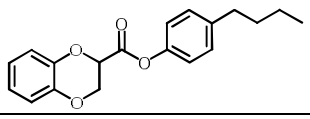
Lead Compound	**Docking** (kcal/mol)	**Binding** (kcal/mol)
	-10.82	-8.90

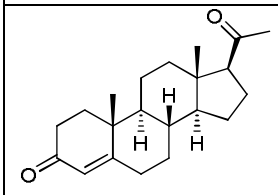
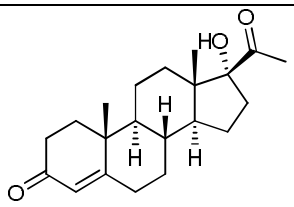
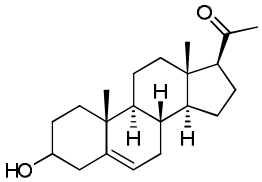
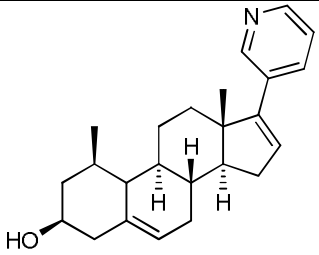
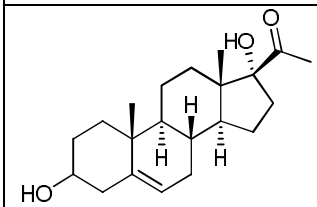
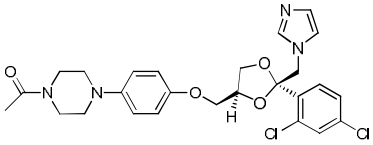
Table1.4. Docking and binding energies of some compounds.

Lead Compound Derivatives	**Docking** kcal/mol	**Binding** kcal/mol	**Lead Compound Derivatives**	**Docking** kcal/mol	**Binding** kcal/mol
	-11.85	-9.44		-10.66	-8.39
	-10.56	-8.47		-9.50	-7.85
	-10.38	-8.10		-9.43	-7.61
	-10.14	-8.26		-9.31	-7.33
	-9.89	-7.77		-9.23	-7.30
	-9.85	-7.63		-8.54	-6.671

1.7.5. Docking and Binding Energy Calculations of Natural Substrates and Current Drug Molecules

In order to compare results of lead compound derivatives, calculations of natural substances and current drug molecules were done. Following Table 1.4. shows docking and binding studies of the natural substrates and the current drug molecules.

Table 1.5 Docking and binding studies of natural substrates and current drug molecules.

Compounds	**Docking** (kcal/mol)	**Binding** (kcal/mol)	**Compounds**	**Docking** (kcal/mol)	**Binding** (kcal/mol)
 Progesterone	-10.04	-9.73	 17hydroxyprogesterone	-9.61	-9.68
 Pregnenolone	-9.71	-9.44	 Abiraterone	-10.15	-10.10
 17hydroxypregnenolone	-9.30	-9.33	 Ketoconazole	-11.13	-9.33

1.8. Selective Inhibition of the Target Enzyme CYP17

Selective inhibition is one of the biggest targets in all type of drug enzyme interactions. In opposite case, not having selective inhibition, may lead to blow up of new diseases.

Selective inhibition of CYP17 is also important, since there are many P450 enzymes and they all have similar parts like a heme part as prosthetic group. P450 enzymes are used for the synthesis of cholesterol, steroids and other important endogenous substrates. In addition to this, they undertake many catalytic reactions such as hydroxylation [28]. Selective inhibition will result in minimization of the side effects. Hence, selective inhibition of the target may be achieved by increasing CYP17 enzyme drug candidate interaction and decreasing interaction of the drug candidate with other CYP enzymes. Interaction may be controlled by testing the drug candidates against CYP11B1, CYP11B2, CYP3A4, and CYP19 enzymes [28].

2. AIM OF THE STUDY

2.1 Derivatization of lead Compound

Docking and binding energy calculations of following compounds were done by using computational program AutoDock 4.00. Following table shows the results of the docking binding energies of final products.

Table 2.1. Docking and binding energies of lead compound.

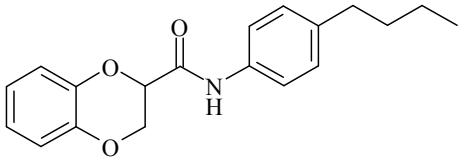
Lead Compound	**Docking** (kcal/mol)	**Binding** (kcal/mol)
	-10.82	-8.90

Table 2.2. Docking and binding energy calculation results of targeted compounds.

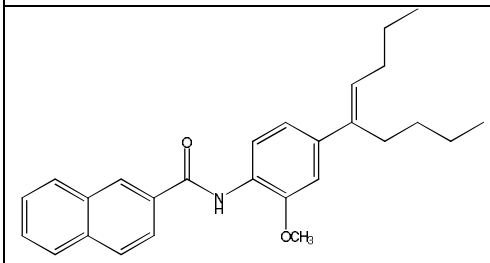
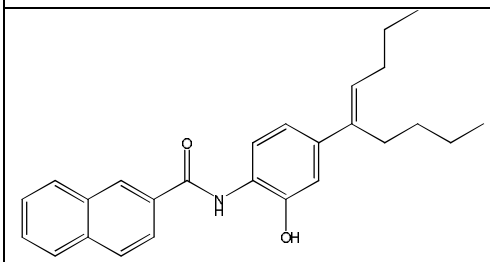
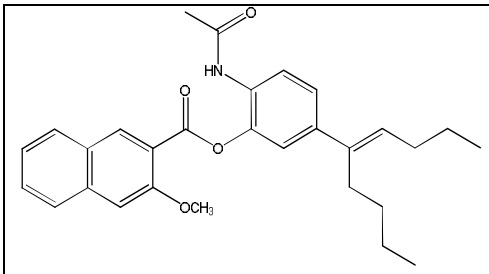
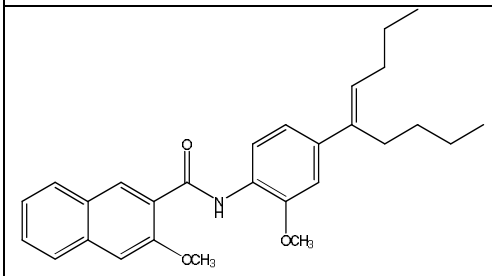
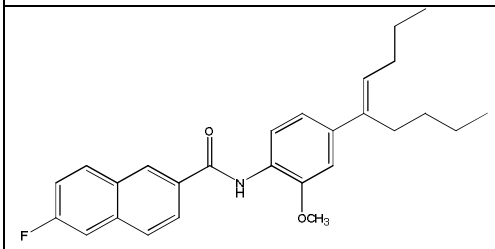
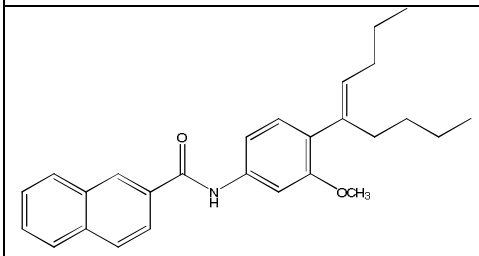
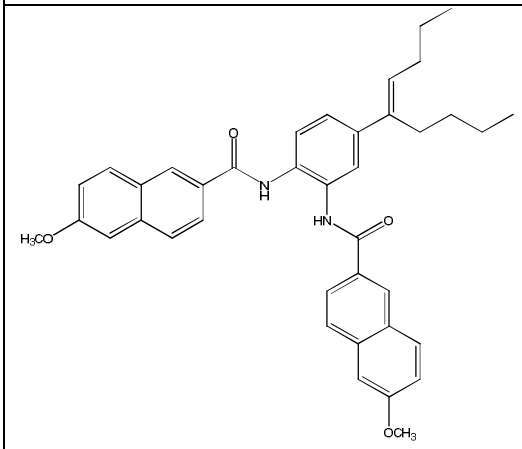
Final product	**Docking energy** (kcal/mol)	**Binding energy** (kcal/mol)
	-12.44	-9.11
	-12.33	-8.67

Table 2.2. Docking and binding energy calculation results of targeted compounds.(contd)

	-15.38	-11.54
	-12.73	-9.28
	-12.68	-9.00
	-11.72	-8.59
	-15.73	-12.17

It is possible to divide the lead compound in two parts where the naphthoic part is at the left hand side and the phenyl at the right hand side of the lead compound. The main target of the present project is to synthesize branched alkyl substituted phenyl groups via derivatization of the lead compound.

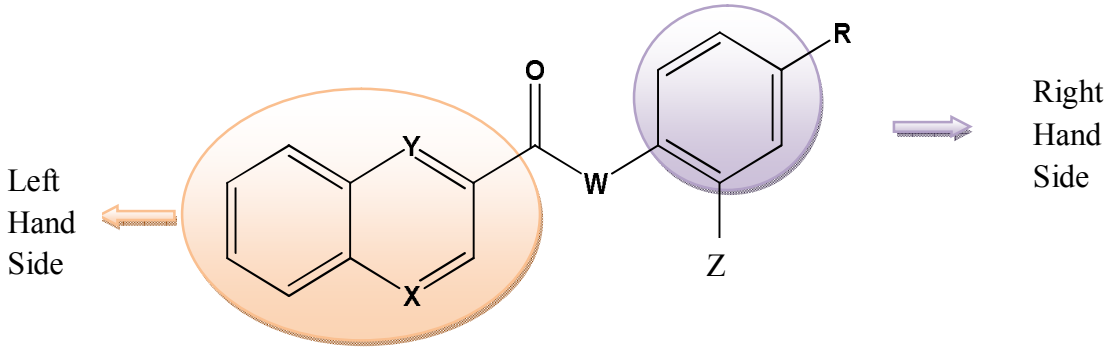


Figure 2.1. Lead Compound Derivatization.

In this part of the project, Y and X were decided to be carbon atoms, thus the right hand side is consisted of naphthoic acid derivatives such as hydrogen, methoxy and floride substituted naphthoic acids.

For right hand side of the lead compound W is determined as N and Z was decided to be OCH_3 or OH. Moreover, in order to derivatize the compound following structure is also thought to be synthesized with coupling from hydroxyl.

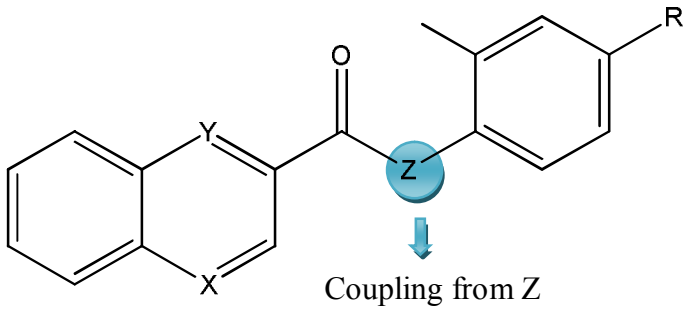


Figure 2.2. Coupling from Z part.

In addition of coupling from Z position of Z was also changed from position three to two so, another derivatization is accomplished as shown Figure 2.3.

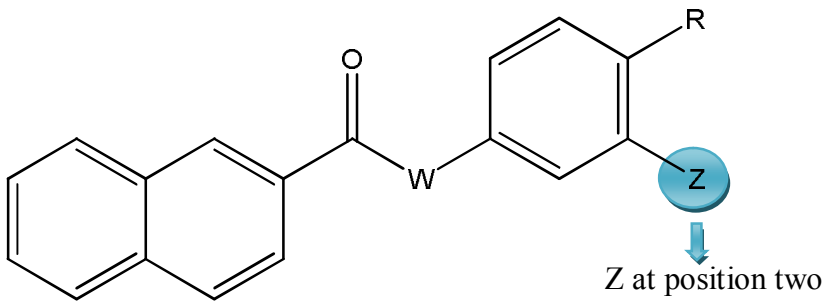


Figure 2.3. Z at a different position.

Lastly R group is targeted as branched butyl group since docking energy and binding energy calculations of branched alkyl substituted drug molecule candidates show better results over linear alkyl substituted drug candidates as shown in the Table 2.1.

Thus, synthesis of branched alkyl substituted phenyl is targeted which will be completed by coupling with different naphthoic acid derivatives.

3. RESULTS AND DISCUSSION

In this project synthesis of branched alkyl substituted phenyl rings and their coupling with various naphthoic acid derivatives were targeted. The branched alkyl substituted derivative shows relatively higher docking and binding energy than linear alkyl substituted ones (Table 1.3, entry 1)

The general strategy followed in the synthesis was as follows; benzoic acid derivatives that had the desired substituents were used as the starting materials and were converted by the fisher esterification method to the corresponding methyl ester (Figure 3.1.). Second implementation became protection of the aromatic amine so that side reactions may be avoided in the next step. Protection of amine was done by using Ac_2O in water. In order to achieve branching on phenyl ring n-Butyllithium (n-BuLi) was used as the nucleophile that will attack the carbonyl of the methyl ester. Since n-BuLi is strong base the reaction was carried out at -78 °C in order to prevent side reactions. Amount of n-BuLi used was threefold with respect to the methyl ester, since two attacks to carbonyl was targeted and hydrogen abstraction from amide part is also possible. Tetrahaydrofuran (THF) was used as the solvent. After the double addition of BuLi an alcohol with two branched alkyl substituent is obtained. With this method it is also possible to synthesize products with longer alkyl chains by employing alkyl lithium reagents such as n-hexyllithium or n-pentyllithium that are commercially available. As next stage deprotection of the amide was targeted so that coupling from amine part would be possible. Deprotection reaction was achieved by the addition of concentrated sulfuric acid in methanol. At the same time dehydration of alcohol also occurred and this change was identified with ^{1}H-NMR Spectroscopy. At the last part coupling of the product with different naphthoic acids were carried out.

For the verification of these transformations ^{1}H-NMR Spectroscopy, ^{13}C-NMR Spectroscopy, Liquid Chromatography-Mass Spectroscopy and Elemental Analysis methods have been used.

3.1. Synthetic Approaches to Synthesize Branched Alkyl 4-Amine-3-Methoxy Substituted Phenyl Derivative

Figure 3.1. shows the synthetic approach used in the synthesis of branched alkyl amine-methoxy phenyl derivative.

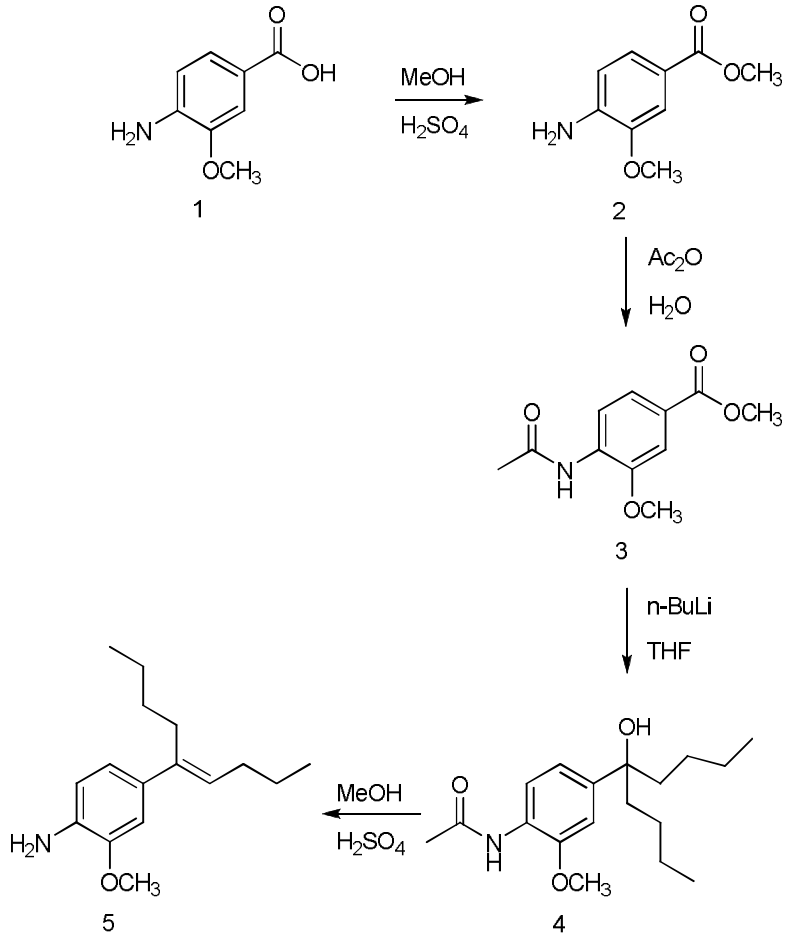


Figure 3.1. Synthetic approach to synthesize branched alkyl, amine-methoxy substituted phenyl.

For the synthesis of branched alkyl and amine-methoxy substituted phenyl derivatives compound 1, 4-amino-3-methoxybenzoic acid, was selected as the starting material. In order to obtain the methyl ester of benzoic acid, Fisher esterification method was used. Reaction was carried out in MeOH in the presence of 20 % H_2SO_4. Light brown product was obtained in 89 % yield.

In the next step amine protection was targeted. Since reducing probability of having side reaction at the organolithium addition step is important, we needed to protect the amine. In order to obtain compound 3 Ac_2O in H_2O was used. Yield of this reaction with compound 2 was 80 %. The reason of having such a high yield is again not having any side products. White colored product was obtained by pouring reactant in icy water and precipitating.

Branched alkyl substitution was done via n-BuLi addition to compound 3 at -78°C in THF. The reason of carrying out reaction at -78°C is not having too much side reactions since n-BuLi is a very strong base. Moreover, this reaction was done under nitrogen since interaction of n-BuLi with air may lead to flame. The reason of selecting THF as solvent is to provide coordination of Li^+ ions with the oxygen of THF. Once the reaction was completed, MeOH was added in small portions in order to get rid of the unreacted n-BuLi. Yield of this reaction was %62 that seems low when compared to previous experiments. The reason of having pretty low yield may be H abstraction of Bu^- from amide part of compound 3.

The last step to obtain branched alkyl and amine-methoxy substituted phenyl is deprotection of the amide. This reaction was carried out at 80°C. While MeOH is used as the solvent, presence of H_2SO_4 supplies hydrogen that was needed for the amine formation. After getting ^{1}H-NMR Spectroscopy of compound 5 it was realized that water elimination occurred together with the amide hydrolysis. Thus we got E and Z isomers of compound 5 with 98% yield.

3.2. Synthetic Approaches for Coupling of Amine with Naphthoic Acid Derivatives

For the synthesis of different final products compound 5 was coupled with different naphthoic acid derivatives.

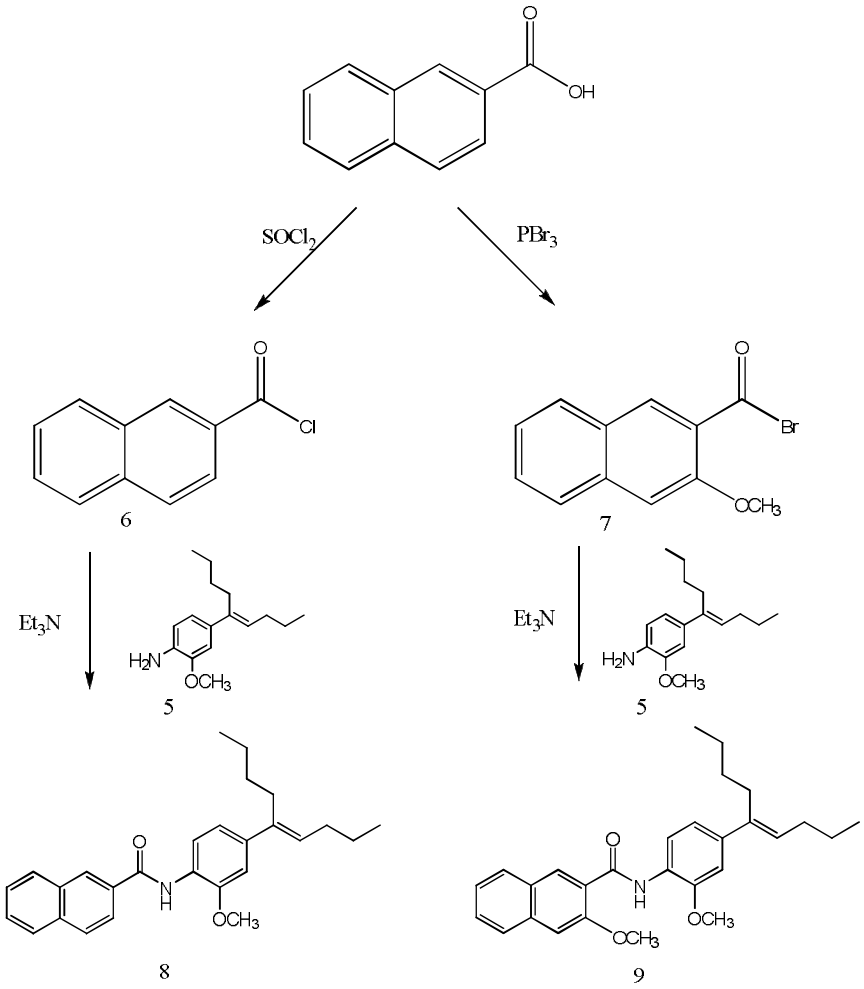


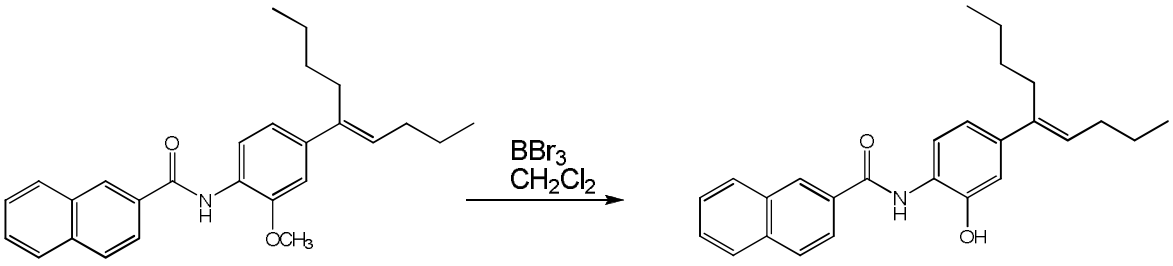
Figure 3.2. Synthetic approach for coupling of amine with naphthoic acid derivatives.

Coupling of compound 5 is the key step for synthesis of drug molecule candidates. Hence two different methods have been tried (Figure 3.2). First reaction was carried out using $SOCl_2$. The reaction was carried out by adding $SOCl_2$ to the naphthoic acid without any solvent. The reaction product was then distilled at 80 °C for 4 hours. After concentrating, compound 6 was obtained in 100 % yield. For the coupling of the amine with naphthyl chloride dry dichloromethane (DCM) was used as the solvent and triethylamine (TEA) was used to neutralize the HCl that will evolve through reaction and activation of the acid chloride through acyl ammonium intermediate formation. After stirring 24 hours at room temperature, reaction was concentrated by evaporation of the solvent and the crude product was purified over silica gel column chromatography. Compound 8 was isolated in 51 % yield. The reason of not getting higher yield may be the lost of some of naphthoyl chloride as the corresponding acid.

The other method used for coupling was done by using PBr_3 under inert conditions by using nitrogen gas and dry solvent. Since this reaction is *in situ* coupling reaction compound 7 was not isolated. Three hours later after addition of PBr_3 to naphthoic acid, TEA and then the amine (compound 5) were added. The purpose of TEA addition is again to neutralize acid that is HBr evolves through reaction. Reaction is carried out at room temperature for five hours. After concentrating and purifying, light brown compound was obtained in 36.8 % yield. Two advantages of this reaction over first one are shorter reaction time and the fact that the latter is done in one pot. The reason of not having higher yield may be not forming enough naphthoyl bromide to react with compound 5.

3.3. Synthetic Approaches Used for Methoxy Deprotection

Methoxy deprotection on the phenyl ring is one of the key steps since deprotection of the methoxy and getting the free phenol gives a new product and it also gives us opportunity of coupling form the phenol as a new derivative. Figure 3.3. shows the synthetic approach to obtain alcohol from methoxy on phenyl ring.



8 10

Figure 3.3. Synthetic approach used for methoxy deprotection.

Methoxy deprotection method was applied on only two products; compound 4 and compound 8. The important part of that reaction is the addition of BBr_3 which was done at -78°C degree. After stirring reaction at room temperature for 20 hours and purification compound 11 was obtained with 63 % yield

3.4. Synthetic Approaches Used for Coupling of Alcohol with Naphthoic acid

Figure 3.4. shows schematic representation of methoxy deprotection of compound 4 and alcohol coupling of compound 11 with a naphthoic acid derivative.

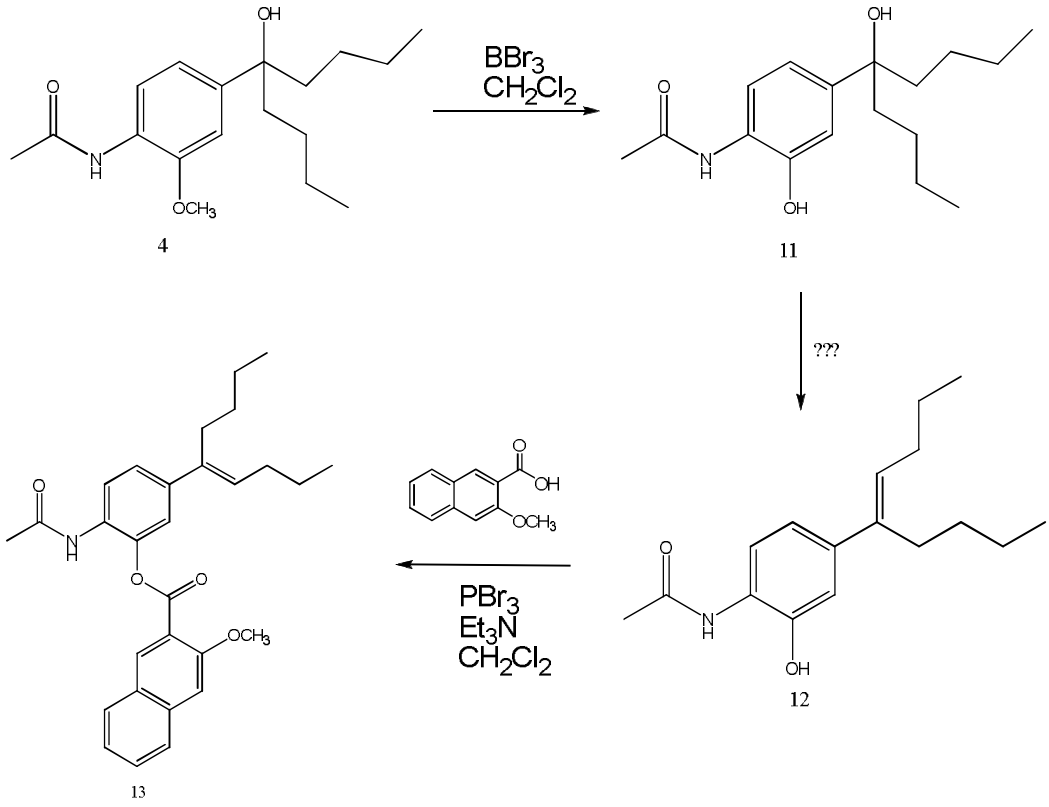


Figure 3.4. Synthetic approach used for alcohol coupling.

Synthetic approach used for methoxy deprotection is the same as the method used in obtaining compound 8. However, yield of this reaction was 65 % better than the previous one.

After obtaining the corresponding phenol, ^{1}H-NMR Spectroscopy of compound showed triplets at 5.63 and 5.23 which indicated the formation of E and Z isomers of 12 instead of compound 11. Then coupling reaction was carried out using PBr_3. Unfortunately yield of this reaction was also low (43 %).

3.5. Synthetic Approaches to Synthesize Branched Alky 4-Amine-2-Methoxy Substituted Phenyl Derivative

The same synthetic approach with synthesis of 4-amine-3-methoxy substituted phenyl was used in the synthesis of 4-amino-2-methoxy substituted phenyl derivative. Figure 3.5 shows the synthetic approach used in the synthesis of (E)-3-methoxy-4-(non-4-en-5-yl)aniline.

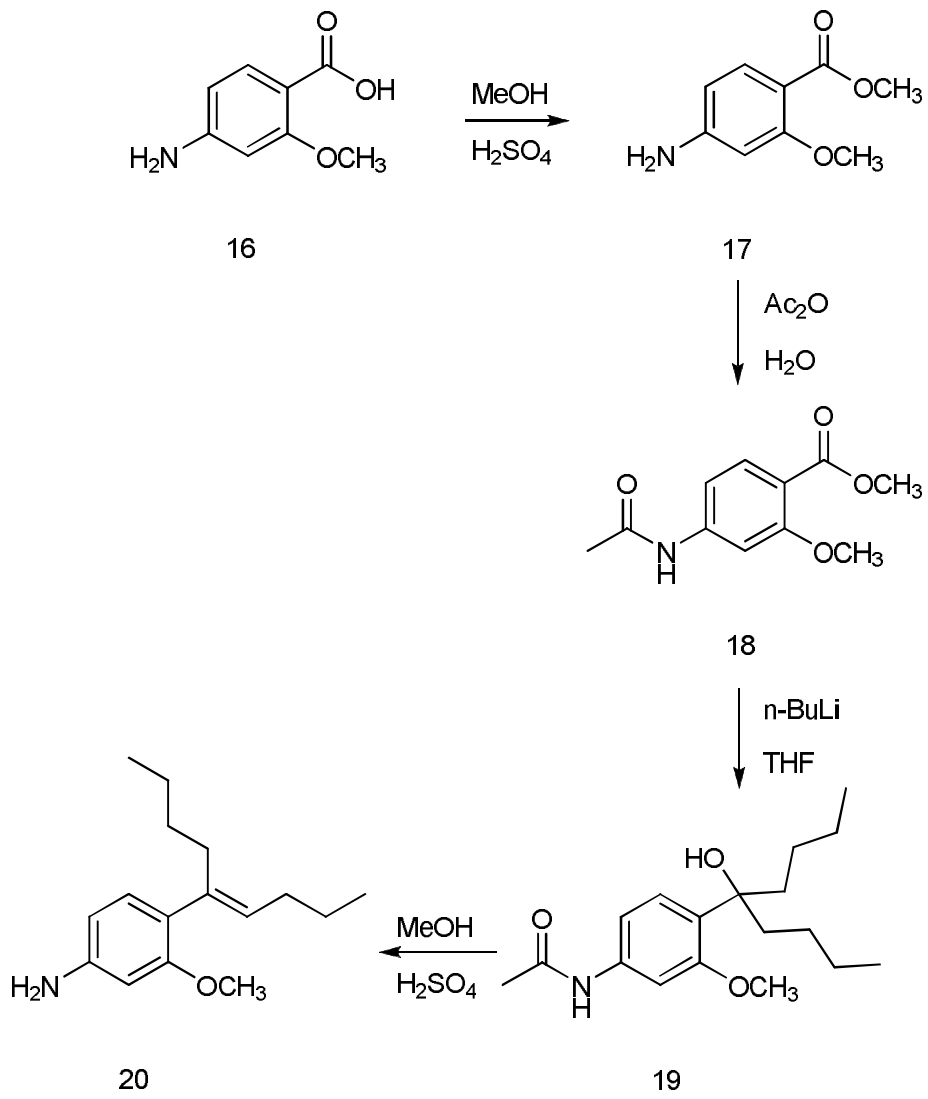


Figure 3.5. Synthetic approach used for the synthesis of 4-amine-3methoxy substituted phenyl.

3.6. Synthetic Approaches to Synthesize (E)-N,N'-(4-(non-4-en-5-yl)-1,2-phenylene)bis(6-methoxy-2-naphthamide)

Compound 22 is one of the compounds that was synthesized by Tuba Şahin one of our group members [39]. In order to synthesize final products compound 22 was deprotected and coupled with naphthoic acid. Through reactions same synthetic approaches with synthesis of compounds 5 and6 are used.

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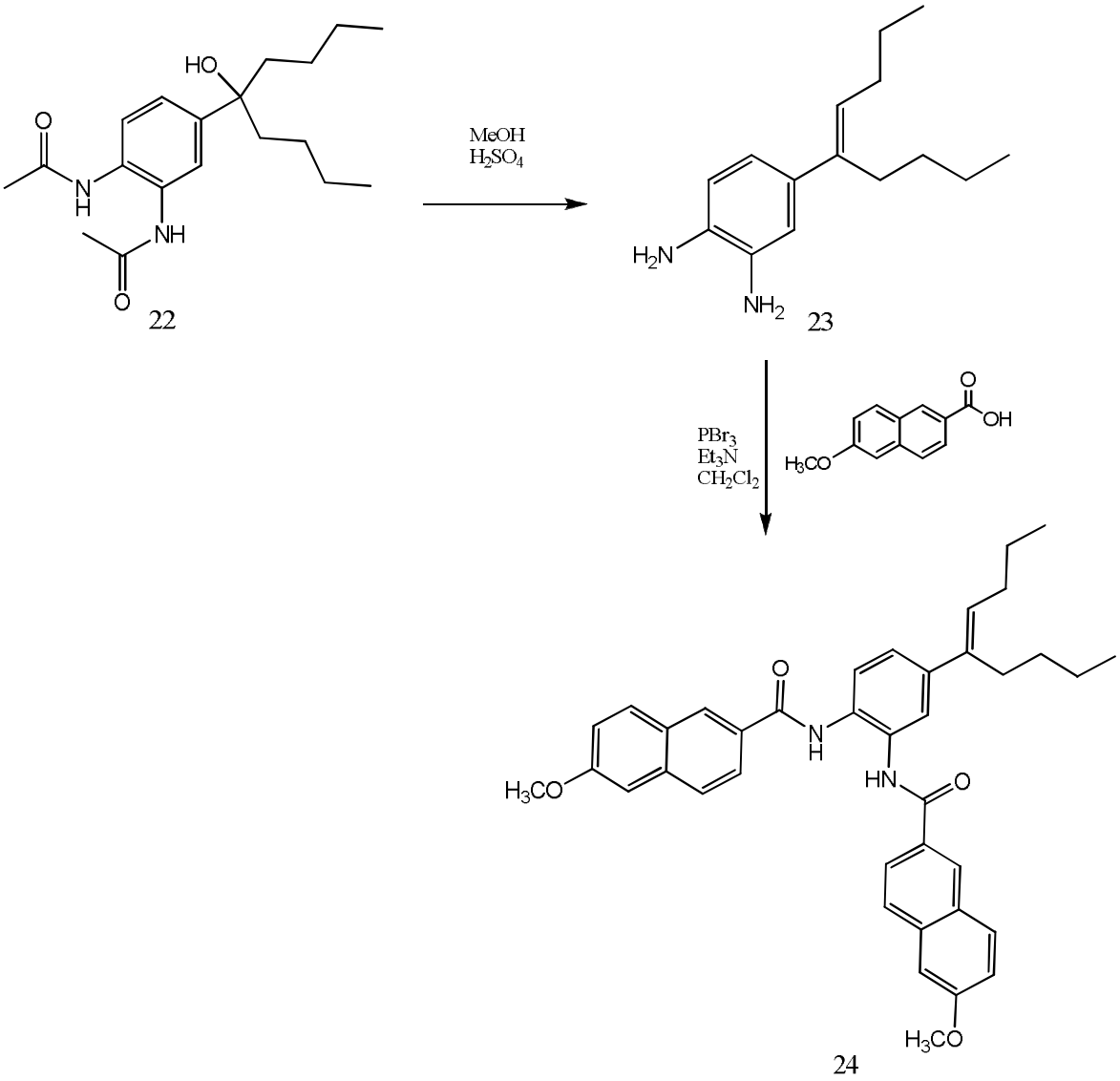


22 23 24

Figure 3.6. Synthetic approach used for synthesis of (E)-N,N'-(4-(non-4-en-5-yl)-1,2-phenylene)bis(6-methoxy-2-naphthamide).

4. EXPERIMENTAL

4.1. Methods and Materials

All chemicals were used as received from the manufacturers (Merck, Aldrich, Alfa Aesar, Riedel de Haen). Dry solvents (CH_2Cl_2, THF, toluene) were obtained from ScimatCo Purification System, other dry solvents were dried of molecular sieves. Column chromatography was performed using silicagel-60 (43-60 nm). Thin layer chromatography was performed using silica gel plates (Kiesel gel 60 F254, 0.2mm, Merck) and aluminum oxide plates.

4.2. Instrumentation

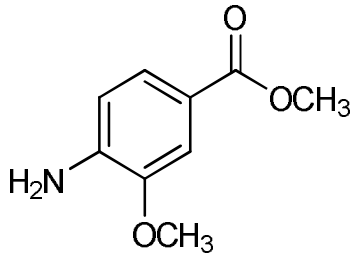
Thin layer chromatograhy plates were viewed under 254 nm UV lamp. Infrared spectroscopy was carried out on Thermo Scientific Nicolet 380 FT-IR spectrophotometer. ^{1}H-NMR and ^{13}C-NMR spectra were recorded using a Varian Gemini 400 MHz spectrometer Varian Associates, Palo Alto, CA) in $CDCl_3$, DMSO and CD_3OD as solvent at the Advanced Technologies Research and Development Center at Boğaziçi University.

4.3. Synthesis of the Derivatives

4.3.1. Synthesis of Methyl 4-amino-3-methoxybenzoate (2)

The reaction was carried using the Fisher Etherification method. The compound 1 (1.00g/ 5.98mmol) and MeOH (10mL) were added into 25mL two necked round bottom flask with a magnetic stirrer. Temperature of the heater was adjusted to 65°C and reflux was started. Concentrated sulphuric acid (1mL) was added drop by drop. The reaction progress was monitored by using TLC silica gel plate and CH_2Cl_2 as the eluent phase. After two hours, starting material was consumed. Solution was added in saturated NaCl solution and extracted with diethyl ether 3 times with 10 mL portions. Then organic layers were collected and washed with 10 mL two portions of water. Organic layer was dried by adding Na_2SO_4 and leaving overnight. The product was pure, no purification process was

done. Product was concentrated and light yellow, 964 mg product was obtained in 89 % yield. ^{1}H NMR (DMSO) δ 7.36 (d of d, J= 8.4, 1H, ArH), 7.26 (d, 1H, J= 1.6, ArH), 6.60 (d, J= 8.4, 1H, ArH), 5.59 (s, 2H, NH_2), 3.77 (s, 3H, CH_3), 3.72 (s, 3H, CH_3) ppm.

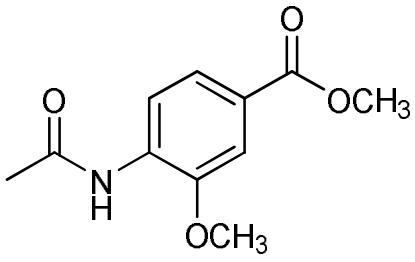


2

Figure 4.1. Methyl 4-amino-3-methoxybenzoate (2).

4.3.2. Synthesis of Methyl 4-acetamido-3-methoxybenzoate (3)

The reaction was done according to literature procedure [40]. The compound 2 (810mg, 4,47mmol) and 5 mL H_2O was added in two necked round bottom flask with a magnetic stirrer and the compound 2 was dissolved. After dissolving of compound, Ac_2O (514,5 mg, 5 mmol) was added to the round bottom flask.. After stirring for 3 hours at room temperature, white precipitation was observed. Then the reaction was monitored by using TLC silica gel plates and CH_2Cl_2 as the eluent phase. One spot on silica gel plate was observed as the only product. The reaction was poured into stirring icy water beaker in order to precipitate all products that weren't precipitated. Suction filtration was next step to remove the water and the remaining Ac_2O. Product was dried in desiccator and 798 mg product obtained in 80 % yield. ^{1}H NMR ($CDCl_3$) δ 2.23 (s, 3H, CH_3), 3.90 (s, 3H, CH_3), 3.94(s, 3H, CH_3), 7.55 (d, 1H, J= 1.6, ArH), 7.68 (d of d, 1H, J= 8.4, ArH), 7.92 (s, 1H, NH), 8.47 (d, 1H, J= 8, CH).ppm.

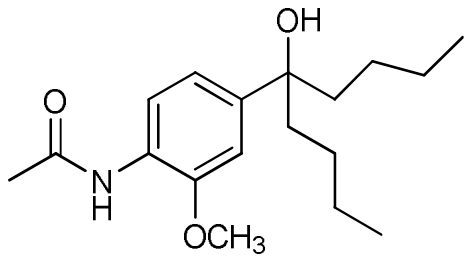


3

Figure 4.2. Methyl 4-acetamido-3-methoxybenzoate (3).

4.3.3. Synthesis of N-(4-(5-hydroxynonan-5-yl)-2-methoxyphenyl)acetamide (4)

The reaction was done according to the literature procedure [41]. The experiment was carried out under inert atmosphere using nitrogen and temperature was adjusted to -78°C in cryostat. The compound 3 (100mg, 0,448mmol) and 5 mL dry THF was added into a two necked round bottom flask with a magnetic stirrer. n-BuLi (0.9mmol, 0.4mL) was transferred to the flask via a deoxygenated syringe in a dropwise manner at four times in 0.1mL portions. After stirring for 4 hours at -78°C, reaction color turned from colorless to yellow. The reaction progress was monitored by TLC using silica oxide gel plates and 1:1 hexane ethyl acetate as the eluent phase. MeOH (15mL) was added in order to get rid of the unreacted n-BuLi. Then, 30 mL H_2O added to the flask and the mixture was extracted 3 times with 20 mL of diethylether portions. The extracts were collected and organic layer was dried over anhydrous Na_2SO_4 by leaving overnight. Then product was concentrated and the crude product was impure. In order to purify product it was dissolved in 5 mL MeOH and poured in 25 mL water and recrystallized. 85 mg light white colored product was obtained in 62% yield. Theoretical elemental analysis values are as follows: C, 70.32; H, 9.51; N, 4.56; O, 15.61 and experimental values are: C, 70.03; H, 9.86; N, 4.60; O, 15.50. ^{1}H NMR (DMSO) δ 8.27 (d, J= 8.4, 1H, ArH), 7.71 (s, 1H, NH), 7.00 (d, J= 2, 1H, ArH) 6.85 (d of d, J= 8.4, 1H, ArH), 3.89 (s, 3H, CH_3), 2.19 (s, 3H, CH_3), 1.77 (m, 4H, CH_2), 1.23 (m, 6H, CH_2), 1.04 (m, 2H, CH_2), 0.83(m, 6H, CH_3) ppm.

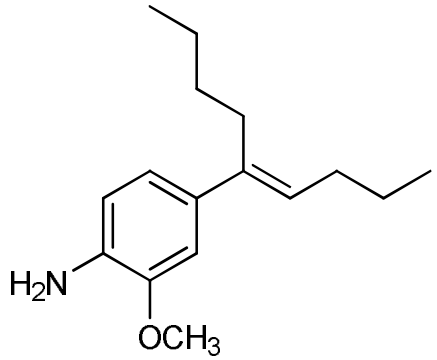


4

Figure 4.3. N-(4-(5-hydroxynonan-5-yl)-2-methoxyphenyl)acetamide (4).

4.3.4. Synthesis of N (E)-2-methoxy-4-(non-4-en-5-yl)aniline (5)

Deprotection and dehydration was done via hydrolysis of the amide group to amine by using 20% H_2SO_4. Compound 4 (80 mg, 0.260 mmol) was dissolved in 5 mL of CH_3OH in a three-necked round bottom flask fitted with a reflux condenser and a magnetic stirrer. 2mL of 20% H_2SO_4 was added drop by drop to the reaction flask and refluxed at 80°C for three hours. The reaction was monitored by using TLC silica gel plates and CH_2Cl_2 as the eluent phase. At the end of three hours the medium was rendered basic with 10mL of 5% Na_2CO_3 then extracted three times with 15 mL of diethylether solutions. The organic layer was extracted once with 20 mL of saturated NaCl solution. Then the organic phase was dried over Na_2SO_4 by leaving overnight. The product was pure so no purification process was done. After concentrating and drying processes 59 mg light brown liquid product is obtained in 92 % yield. ^{1}H NMR ($CDCl_3$) δ 6.80 (d, 1H, ArH), 6.77 (d, 1H, ArH), 6.64 (d, 1H, ArH), 6.58 (s, 2H, NH_2), 5.56 (t, 1H, CH), 3.47 (t, 2H, CH_2), 2.44 (t, 2H, CH_2), 2.14 (m, 4H, CH_2, CH_2), 1.31 (m, 2H, CH_2), 0.95 (t, 3H, CH_3),0.87 (t, 3H, CH_3) ppm.



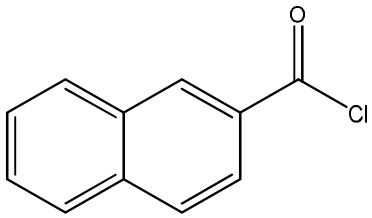
5

Figure 4.4. (E)-2-methoxy-4-(non-4-en-5-yl)aniline (5).

4.3.5. Synthesis of 2-Naphthoyl chloride (6)

The reaction was done according to the literature procedure [42]. 2-Naphtoic acid (1g**,** 5.8 mmol) was added into one necked 50 mL round bottom flask fitted with a magnetic stirrer and condenser. Reaction was carried out under inert atmosphere using nitrogen. Then 8 mL $SOCl_2$ was added under nitrogen and $SOCl_2$ was used both as the

solvent and the reactant. The solution was refluxed at 80 ^{o}C. The reaction progress was monitored by thin layer chromatography using silica gel plates and CH_2Cl_2 as the eluent phase for 4 hours. At the end of four hours one spot product is observed. After evaporation of $SOCl_2$ 1.10 g yellow, solid 2-naphthoyl chloride is synthesized in 100% yield. ^{1}H NMR ($CDCl_3$): δ 8.76 (s, 1H, Ar*H*), 8.04 (2H, Ar*H*), 7.93 (d, 2H, Ar*H*, *J*=8.9 Hz), 7.66 (m, 2H, Ar*H*) ppm.



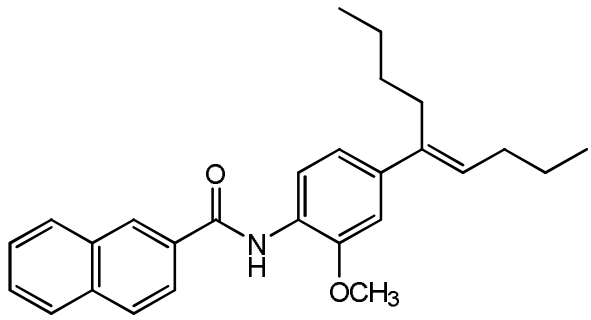
6

Figure 4.5. 2-Naphthoyl chloride (6).

4.3.6. Synthesis of (E)-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (8)

The reaction was done according to the literature procedure [43]. 2-Naphthoyl chloride (51.3mg, 0.27mmol) was added inside of a two necked round bottom flask fitted with a magnetic stirrer under inert atmosphere by using nitrogen gas. 5 mL dry CH_2Cl_2 was added inside the round bottom flask and 2-Naphthoyl chloride was dissolved. (E)-2-metoksi-4-(non-4-en-5-yl)anilin (67 mg, 0.27mmol) and then triethylamine (40.4mg, 0.4 mmol) were added to the round bottom flask under nitrogen gas. Reaction was stirred for 24 hours at room temperature. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 2% MeOH / CH_2Cl_2 as the eluent phase. Solid product was filtered and light brown product was obtained. The product was impure and was purified by using silica gel chromatography. Product was dissolved in minimum amount of CH_2Cl_2 (1 mL), then it was injected through a straight line on silica plate (22×13cm). Silica plate was put inside of a 2% MeOH / CH_2Cl_2 solvent mixture. Desired layer was scratch out, dissolved in 5% MeOH / CH_2Cl_2 solvent and filtered. After concentrating dark brown, solid, pure final product was obtained in 48 % yield. ^{1}H NMR ($CDCl_3$) δ : 0.80 (t, 3H, CH_3), 0.89 (t, 3H, CH_3), 1.12 (m, 2H, CH_2), 1.17(m, 2H, CH_2), 1.40(m, 2H, CH_2), 2.10(q, 2H, CH_2), 2.42(t, 2H, CH_2), 3.87(s, 3H, OCH_3), 6.84(s, 1H,

ArH), 6.94(d, 1H, ArH), 7.48(t, 2H, ArH), 7.80(d, 1H, ArH), 7.85(d, 2H, ArH), 7.89(d, 1H, ArH), 8.32(s, 1H, ArH), 8.41(d, 1H, ArH), 8.58(s, 1H, NH).

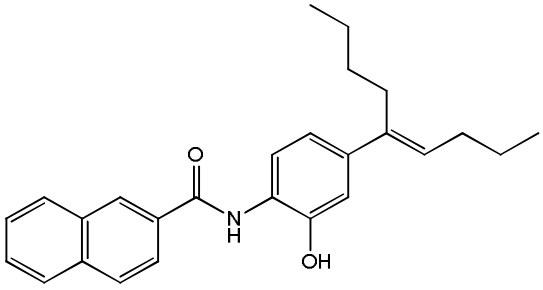


8

Figure 4.6. (E)-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (8).

4.3.7. Synthesis of E)-N-(2-hydroxy-4-(non-4-en-5-yl)phenyl)acetamide (12)

The reaction was done according to the literature procedure [44]. The experiment was carried out under inert atmosphere using nitrogen and temperature was adjusted to -78°C in cryostat. (E)-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (70mg , 0.175 mmol) was added inside of a three necked round bottom flask fitted with a magnetic stirrer and 3.5 mL dry CH_2Cl_2 was added under nitrogen gas. Solution was cooled to -78 ^oC and 0.5 mL (5.2 mmol) BBr_3 was added in 15 minutes drop by drop. Reaction was stirred at room temperature for 20 hours. 5 mL MeOH was added to reaction and stirred for 40 minutes. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 10 % EtOAc / CH_2Cl_2 as the eluent phase. Organic solvent was evaporated under vacuum. The obtained product was dissolved in ethyl acetate and extracted with saturated $NaHCO_3$, saturated NaCl and water respectively. Organic layer was collected inside of an erlenmeyer flask and dried by adding Na_2SO_4 and leaving overnight. After evaporating solvent, crude product was obtained and it was purified by using silica plate chromatography and CH_2Cl_2 as eluent phase. As a result 42 mg pure product was obtained in 63% yield. ^1H NMR ($CDCl_3$) δ :8.78(s, 1H, NH), 8.39(s,1H, ArH), 8.27(d,1H, ArH), 7.88(d, 1H, ArH), 7.81(d, 2H, ArH, ArH), 7.51(t, 2H, ArH, ArH), 7.10(d, 1H, ArH), 7.01(d, 1H, ArH), 6.84(s, 1H, ArH), 5.61(t, 1H, CH), 2.76(m, 2H, CH_2), 2.38(t, 2H, CH_2), 2.09(q, 2H, CH_2), 1.38(m, 2H, CH_2), 1.24(m, 2H, CH_2), 0.78(t, 6H, CH_3, CH_3) ppm.

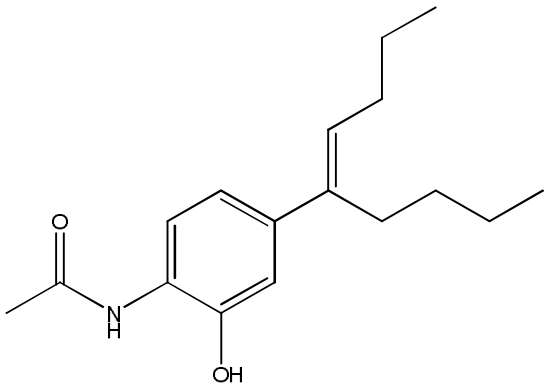


10

Figure 4.7. (E)-N-(2-hydroxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (10).

4.3.8. Synthesis of N-(2-hydroxy-4-(5-hydroxynonan-5-yl)phenyl)acetamide (11)

The reaction was done according to the literature procedure [44]. The experiment was carried out under inert atmosphere using nitrogen and temperature was adjusted to -78°C in cryostat. N-(4-(5-hydroxynonan-5-yl)-2-methoxyphenyl)acetamide which is compound 4 (120mg , 0.390 mmol) was added inside of a 25 ml three necked round bottom flask fitted with magnetic stirrer and 4 mL dry CH_2Cl_2 was added under nitrogen gas. Solution was cooled to -78 ^oC and 1.1 mL (11.6 mmol) BBr_3 was added in 15 minutes drop by drop. Reaction was stirred at room temperature for 20 hours. 8 mL MeOH was added to reaction and stirred for 40 minutes. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 10 % EtOAc / CH_2Cl_2 as the eluent phase. Organic layer solvent was evaporated under vacuum. Obtained product was dissolved in ethyl acetate and extracted with saturated $NaHCO_3$, saturated NaCl and water accordingly. Organic layer was collected inside of an erlenmeyer flask and dried by adding Na_2SO_4 and leaving overnight. After evaporating solvent, crude product was obtained and it was purified by using silica plate chromatography and CH_2Cl_2 as eluent phase. As a result 73 mg pure product was obtained in 64% yield. ^1H NMR ($CDCl_3$) δ 7.98 (s, 1H, NH), 6.76 (d, 1H, ArH), 6.72 (d, 1H, ArH), 6.55 (d, 1H, ArH), 5.56 (t, 1H, CH), 2.35 (t, 2H, CH_2), 2.13 (s, 3H, CH_3), 2.06 (q, 2H, CH_2), 1.20 (m, 4H, CH_2, CH_2), 1.18 (m, 2H, CH_2),0.86 (t, 3H, CH_3), 0.76, (t, 3H, CH_3) ppm.



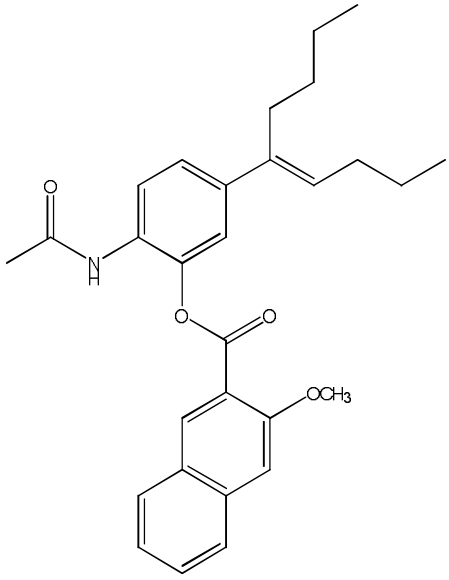
12

Figure 4.8. (E)-N-(2-hydroxy-4-(non-4-en-5-yl)phenyl)acetamide (12).

4.3.9. Synthesis of (E)-2-acetamido-5-(non-4-en-5-yl)phenyl 3-methoxy-2naphthoate (13)

The reaction was done according to the literature procedure [45]. 3-methoxy-2-naphthoic acid (50.5 mg, 0.25mmol) was added inside of a two necked round bottom flask fitted with a magnetic stirrer under inert atmosphere by using nitrogen gas. 3 mL dry CH_2Cl_2 was added into the round bottom flask. PBr_3 (0.036 ml, 0.375 mmol) was added under inert conditions. Reaction was refluxed and stirred at 37 °C for 3 hours. Then TEA (0.75 mmol, 0.12 ml) was added to reaction in 10 minutes drop by drop. When HBr formation stopped N-(2-hydroxy-4-(5-hydroxynonan-5-yl)phenyl)acetamide (73 mg, 0.25mmol) was added to the round bottom flask under nitrogen. Reaction was stirred for 2 hours at room temperature. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 1% MeOH / CH_2Cl_2 as the eluent phase. Solid product was filtered and light brown product was obtained. The product was impure and was purified by using silica gel chromatography. Product was dissolved in minimum amount of CH_2Cl_2 (1 mL), then was injected through a straight line on silica plate (22×13cm). Silica plate was put inside of a 1% MeOH / CH_2Cl_2 solvent mixture. Desired layer was scratch out, dissolved in 5% MeOH / CH_2Cl_2 solvent and filtered. After concentrating the solvent 49 mg dark brown, solid, pure final product was obtained in 43% yield. ^{1}H NMR ($CDCl_3$) δ : 0.82(t, 6H, CH_3, CH_3), 0.88(m, 2H, CH_2), 1.18(m, 2H, CH_2), 1.30(q, 2H, CH_2), 1.47(m, 2H, CH_2), 3.96(t, 2H, CH_2), 4.11(s, 3H, OCH_3), 5.23(s, 3H, CH_3), 5.66(t, 1H, CH), 6.84(d, 1H, ArH), 6.91(s, 1H, ArH), 7.03(d, 1H, ArH), 7.24(s, 1H,

ArH), 7.37(t, 1H, ArH), 7.50(t, 1H, ArH), 7.72(d, 1H, ArH), 7.87(d, 1H, ArH), 8.82(s, 1H, ArH), 10.24(s, 1H, NH)



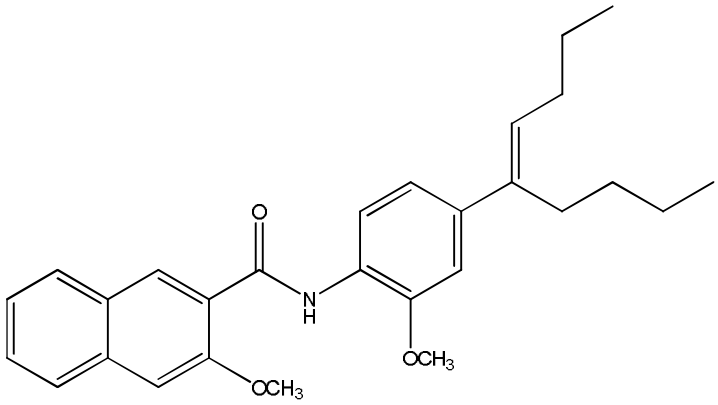
13

Figure 4.9. (E)-2-acetamido-5-(non-4-en-5-yl)phenyl 3-methoxy-2-naphthoate (13).

4.3.10. Synthesis of (E)-3-methoxy-N- (2-methoxy-4- (non-4-en-5-yl) phenyl)-2-naphthamide (14)

The reaction was done according to the literature procedure [45]. 3-methoxy-2-naphthoic acid (30mg, 0.145 mmol) was added inside of a two necked round bottom flask fitted with a magnetic stirrer under inert atmosphere by using nitrogen gas. 2 mL dry CH_2Cl_2 was added inside of round bottom flask. PBr_3 (0.021 ml, 0.22 mmol) was added under inert conditions. Reaction mixture was distilled and stirred at 37 °C for 3 hours. Then TEA (0.435 mmol, 0.070 ml) was added to reaction in 10 minutes drop by drop. When HBr formation stopped (E)-2-methoxy-4-(non-4-en-5-yl)aniline compound 5 (36 mg, 0.145 mmol) was added to the round bottom flask under nitrogen gas. Reaction was stirred for 2 hours at room temperature. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 1% MeOH / CH_2Cl_2 as the eluent phase. Solid product was filtered and light brown product was obtained. The product was impure and was purified by using silica gel chromatography. Product was dissolved in minimum amount of CH_2Cl_2 (1 mL), then it was injected through a straight line on silica plate (22×13cm). Silica plate was put inside of a 20% EtOAc / CH_2Cl_2 solvent mixture. Desired

layer was scratch out, and dissolved in 5% MeOH / CH_2Cl_2 solvent and filtered. After concentrating solvent 23 mg light yellow, solid, pure final product was obtained in 37 % yield. ^{1}H NMR ($CDCl_3$) δ :8.79(s, 1H, NH), 8.53(d,1H, ArH), 7.86(d,1H, ArH), 7.70(d, 1H, ArH), 7.47(t, 1H, ArH), 7.34(t, 1H, ArH), 6.96(d, 1H, ArH), 6.85(s, 1H, ArH), 6.85(s, 1H, ArH), 5.61(t, 1H, CH), 4.10(s, 3H, OCH_3), 3.93(s, 3H, OCH_3), 2.43(t, 2H, CH_2), 2.11(q, 2H, CH_2), 1.41(m, 2H, CH_2), 1.26(m, 4H, CH_2, CH_2), 0.90(t, 3H), 0.81(3H, CH_3, CH_3) ppm.



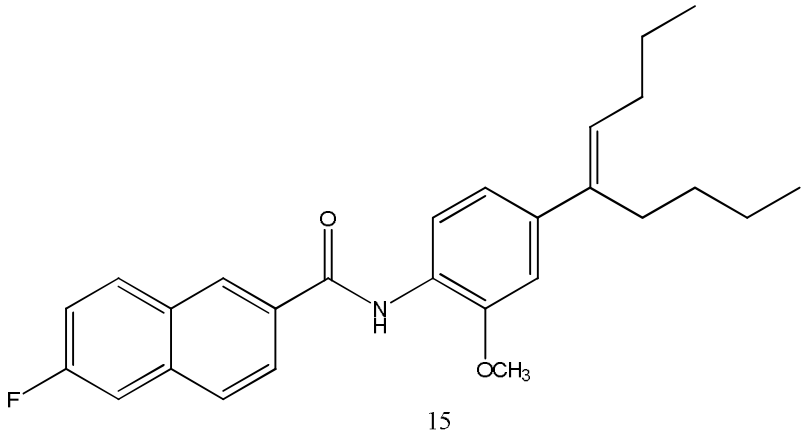
14

Figure 4.10. (E)-3-methoxy-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (14).

4.3.11. Synthesis of (E)-6-fluoro-N- (2-methoxy-4- (non-4-en-5-yl) phenyl)-2-naphthamide (15)

The reaction was done according to the literature procedure [45]. 6-fluoro-2-naphthoic acid (114 mg, 0.6 mmol) was added inside of a two necked round bottom flask fitted with a magnetic stirrer under inert atmosphere by using nitrogen gas. 5 mL dry CH_2Cl_2 was added inside of round bottom flask. PBr_3 (0.14 ml, 1 mmol) was added under inert conditions. Reaction was distilled and stirred at 37 °C for 3 hours. Then TEA (2.00 mmol, 0.28 ml) was added to reaction in 10 minutes drop by drop. When HBr formation stopped (E)-2-methoxy-4-(non-4-en-5-yl)aniline compound 5 (140 mg, 0.57 mmol) was added to the round bottom flask under nitrogen gas. Reaction was stirred for 2 hours at room temperature. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and CH_2Cl_2 as the eluent phase. Solid product was filtered and yellow product was obtained. The product was impure and was purified by using silica gel chromatography. Product was dissolved in minimum amount of CH_2Cl_2 (1 mL), then it

was injected through a straight line on silica plate (22×13cm). Silica plate was put inside of a CH_2Cl_2 solvent. Desired layer was scratch out, and dissolved in 5 % MeOH / CH_2Cl_2 solvent and filtered. After concentrating solvent 97 mg white, solid, pure final product was obtained in 41 % yield. ^{1}H NMR ($CDCl_3$) δ : 0.88 (t, 3H, CH_3), 0.97 (t, 3H, CH_3), 1.31 (m, 2H, CH_2), 1.35(m, 2H, CH_2), 1.48(m, 2H, CH_2), 2.18(q, 2H, CH_2), 2.49(t, 2H, CH_2), 3.96(s, 3H, OCH_3), 5.66 (t, 1H, CH), 6.92(s, 1H, ArH), 7.02(d,1H, ArH), 7.34(t, 1H, ArH), 7.51(d, 1H, ArH), 7.89(d, 1H, ArH), 7.95(d, 1H, ArH), 7.98(d, 1H, ArH), 8.40(s, 1H, ArH), 8.47(d, 1H, ArH), 8.63(1H, NH).



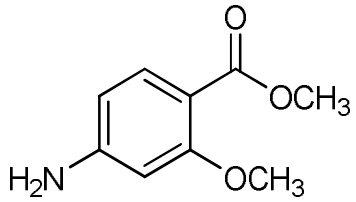
15

Figure 4.11. (E)-6-fluoro-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (15).

4.3.12. Synthesis of Methyl 4-amino-2-methoxybenzoate (17)

The reaction was carried using the Fisher Etherification method. The compound 16 (1.00g/ 5.98mmol) and MeOH (10mL) were added into 25mL two necked round bottom flask with a magnetic stirrer inside of it. Temperature of heater was adjusted to 65°C and reflux is started. Concentrated sulphuric acid (1mL) was added drop by drop. The reaction progress was monitored by using TLC silica gel plate CH_2Cl_2 as eluent phase. After two hours starting material was consumed. Solution was added in saturated NaCl solution and extracted with diethyl ether 3 times in 10 mL portions. Then organic layers were collected and washed with 10 mL two portions water. Organic layer was dried by adding Na_2SO_4 and leaving overnight. The product was pure no purification process was done. Product was concentrated and cream, 932 mg product was obtained in 87 % yield. ^{1}H NMR $CDCl_3$)

δ 7.73 (d, 1H, ArH), 6.24 (d, 1H, ArH), 6.21 (1H, ArH), 6.19 (d, 2H, NH_2), 4.03 (s, 1H, OH) 3.85 (s, 3H, CH_3), 3.82 (s, 3H, OCH_3) ppm.

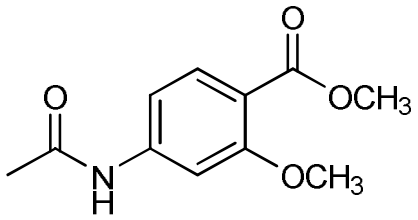


17

Figure 4.12. Methyl 4-amino-2-methoxybenzoate (17).

4.3.13. Synthesis of Methyl 4-acetamido-2-methoxybenzoate (18)

The reaction was done according to the literature procedure [40]. The compound 17 (932 mg, 5.14 mmol) and 2 mL H_2O was added in two necked round bottom flask with a magnetic stirrer and the compound was dissolved. After dissolving of the compound, Ac_2O (561 mg, 5.5 mmol) was added to round bottom flask. . After stirring for 3 hours at room temperature, precipitation could not be observed. However, reaction was monitored by using TLC silica gel plates and CH_2Cl_2 as the eluent phase and one spot was observed as the only product. The reaction was poured into stirring icy water beaker in order to precipitate all products but very small amount of product was precipitated. Aqueous solution with product inside was left to refrigerator for one day and it is observed that all product was precipitated in white color. Suction filtration was done to remove the water and the remaining Ac_2O if exist any. Product was dried in vacuum oven and 723 mg product obtained in 63 % yield. ^{1}H NMR ($CDCl_3$) 7.80 (d of d, 1H, ArH), 7.62 (s, 1H, ArH), 7.36(s, 1H, NH), 6.81 (d of d, 1H, ArH), 3.90 (s, 3H, OCH_3), 3.86 (s, 3H, OCH_3), 2.20 (s, 3H, CH_3).ppm.

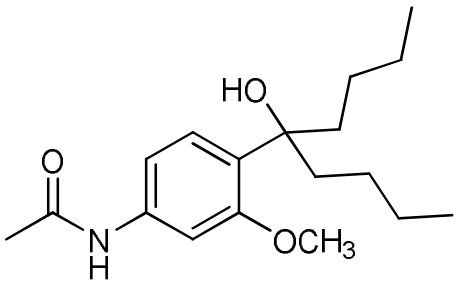


18

Figure 4.13. Methyl 4-acetamido-2-methoxybenzoate (18).

4.3.14. Synthesis of Methyl N-(4-(5-hydroxynonan-5-yl)-3-methoxyphenyl)acetamide (19)

The reaction was done according to the literature procedure [41]. The experiment was carried out under inert atmosphere using nitrogen and temperature was adjusted to -78°C in cryostat. The compound 18 (610mg, 2.74mmol) and 10 mL dry THF were added into a two necked round bottom flask with a magnetic stirrer. n-BuLi (6.25mmol, 2.5mL) was transferred to the flask via a deoxygenated syringe in a dropwise manner at four times in 1mL portions. After stirring for 4 hours at -78°C reaction color turned from colorless to yellow. The reaction progress was monitored by TLC using silica oxide gel plates and 1:1 hexane ethyl acetate as the eluent phase. MeOH (20mL) was added in order to get rid of the unreacted n-BuLi. Then, 30 mL H_2O added to flask and the mixture was extracted 3 times with 20 mL of diethylether. The extracts were collected and organic layer was dried over anhydrous Na_2SO_4 by leaving overnight. Then product was concentrated and the crude product was one spot and pure so, any purification step was not done. 705 mg white colored product was obtained in 89 % yield. ^{1}H NMR ($CDCl_3$) δ 7.94 (d, 1H, ArH), 7.19 (d, 1H, ArH), 7.11 (d, 1H, NH) 6.76 (d of d, 1H, ArH), 3.85 (s, 3H, OCH_3), 3.50 (s, 1, OH), 2.17 (s, 3H, CH_3), 1.96 (m, 2H, CH_2), 1.74 (m, 2H, CH_2), 1.24 (m, 4H, CH_2), 1.09(m, 4H, CH_2), 0.83(t, 6H, CH_3) ppm.

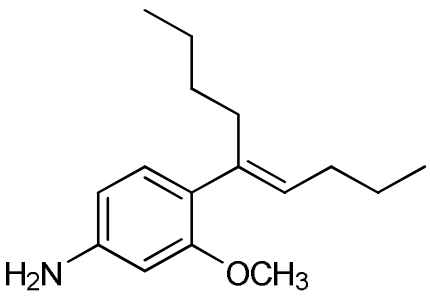


19

Figure 4.14. N-(4-(5-hydroxynonan-5-yl)-3-methoxyphenyl)acetamide (19).

4.3.15. Synthesis of (E)-3-methoxy-4-(non-4-en-5-yl)aniline (20)

Deprotection and dehydration was done via hydrolysis of the amide group to amine by using 20% H_2SO_4. Compound 19 (700 mg, 2.42 mmol) was dissolved in 12 mL of CH_3OH in a three-necked round bottom flask fitted with a reflux condenser and a magnetic stirrer. 3mL of 20% H_2SO_4 was added to the reaction flask and refluxed at 80°C for three hours. The reaction was monitored by using TLC silica gel plates and CH_2Cl_2 as the eluent phase. At the end of three hours the medium was rendered basic with 10mL of 5% Na_2CO_3 then extracted three times with 15 mL of diethylether. The organic layer was extracted once with 20 mL of saturated NaCl solution. Then the organic phase was dried over Na_2SO_4 by leaving overnight. The product was pure no purification process was done. After concentrating and drying processes 488 mg brown solid product is obtained in 82 % yield. ^{1}H NMR ($CDCl_3$) δ 6.80 (d, 1H, ArH), 6.77 (d, 1H, ArH), 6.64 (d, 1H, ArH), 6.58 (s, 2H, NH_2), 5.56 (t, 1H, CH), 3.47 (t, 1H, CH_2), 2.44 (t, 2H, CH_2), 2.14 (m, 4H, CH_2, CH_2), 1.31 (m, 2H, CH_2), 0.95 (t, 3H, CH_3),0.87 (t, 3H, CH_3) ppm.

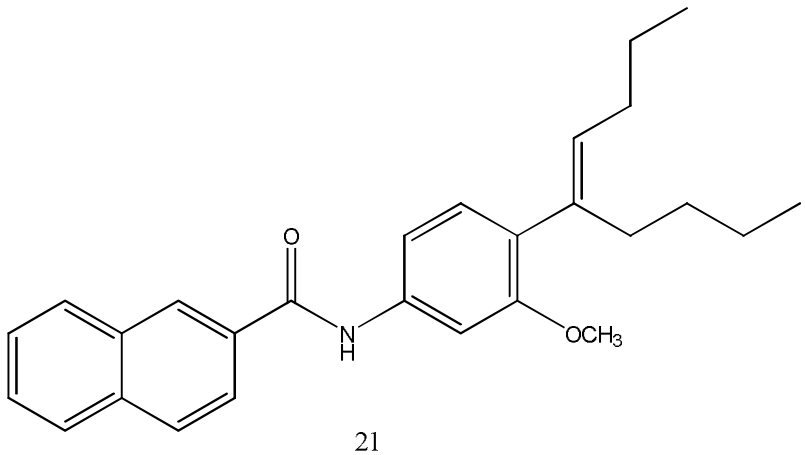


20

Figure 4.15. (E)-3-methoxy-4-(non-4-en-5-yl)aniline (20).

4.3.16. Synthesis of (E)-N-(3-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (21)

The reaction was done according to the literature procedure [45]. 2-naphthoic acid (340mg, 1.97 mmol) was added into a two necked round bottom flask fitted with a magnetic stirrer under inert atmosphere by using nitrogen gas. 2 mL dry CH_2Cl_2 was to the round bottom flask. PBr_3 (0.28 ml, 3 mmol) was added under inert conditions. Reaction mixture was distilled and stirred at 37 °C for 3 hours. Then TEA (5.22 mmol, 0.070 ml) was added to the reaction in 10 minutes drop by drop. When HBr formation is stopped (E)-3-methoxy-4-(non-4-en-5-yl)aniline compound 20 (487 mg, 1.97 mmol) was added to the round bottom flask under nitrogen gas. Reaction was stirred for 3 hours at room temperature. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 1% MeOH / CH_2Cl_2 as the eluent phase. Solid product was filtered and light brown product was obtained. The product was impure and was purified by using silica gel chromatography. Product was dissolved in minimum amount of CH_2Cl_2 (1 mL), then it was injected through a straight line on silica plate (22×13cm). Silica plate was put inside of a 20% EtOAc / CH_2Cl_2 solvent mixture. Desired layer was scratch out, and dissolved in 5% MeOH / CH_2Cl_2 solvent and filtered. After concentrating solvent 276 mg cream, solid, pure final product was obtained in 35 % yield. ^{1}H NMR ($CDCl_3$) δ : 0.84(t, 3H, CH_3), 0.97(t, 3H, CH_3), 1.28(m, 2H, CH_2), 1.32(m, 2H, CH_2), 1.46(m, 2H, CH_2), 2.17(q, 2H, CH_2), 2.45(m, 2H, CH_2), 3.86(s, 3H, OCH_3), 5.39(t, 1H, CH), 6.98(d, 1H, ArH), 7.08(d, 1H, ArH), 7.57(s, 1H, ArH), 7.59(t, 2H, ArH, ArH), 7.90(d, 1H, ArH), 7.92(d, 2H, ArH, ArH), 7.94(d, 1H, ArH), 7.96(s, 1H, ArH), 8.38(s, 1H, NH)

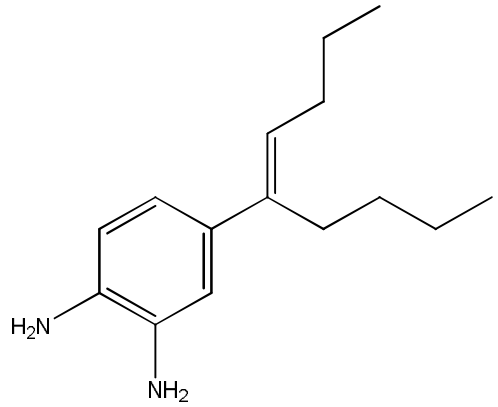


21

Figure 4.16. (E) -N-(3-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide (21).

4.3.17. Synthesis of (E)-4-(non-4-en-5-yl)benzene-1,2-diamine (23)

Deprotection and dehydration was done via hydrolysis of the amide group to amine by using 20% H_2SO_4. Compound 22 (36 mg, 0.112 mmol) was dissolved in 3 mL of CH_3OH in a three-necked round bottom flask fitted with a reflux condenser and a magnetic stirrer. 1.5 mL of 20% H_2SO_4 was added to the reaction flask and refluxed at 80°C for four hours. The reaction was monitored by using TLC silica gel plates and CH_2Cl_2 as the eluent phase. At the end of three hours the medium was done basic with 10mL of 5% Na_2CO_3 then extracted three times with 15 mL of diethylether solutions. The organic layer was extracted once with 20 mL of saturated NaCl solution. Then the organic phase was dried over Na_2SO_4 by leaving overnight. The product was pure no purification process was done. After concentrating and drying processes 25 mg light brown liquid product is obtained in 96 % yield. ^{1}H NMR ($CDCl_3$) δ 7.49 (s, 4H, NH_2, NH_2), 7.46 (d, 1H, ArH), 7.26 (s, 1H, ArH), 7.23 (d, 1H, ArH), 5.63 (t, 1H, CH), 2.52 (t, 2H, CH_2), 2.17 (q, 2H, CH_2), 1.46 (m, 2H, CH_2), 1.30 (m, 4H, CH_2, CH_2), 0.96 (t, 3H, CH_3),0.84 (t, 3H, CH_3) ppm.



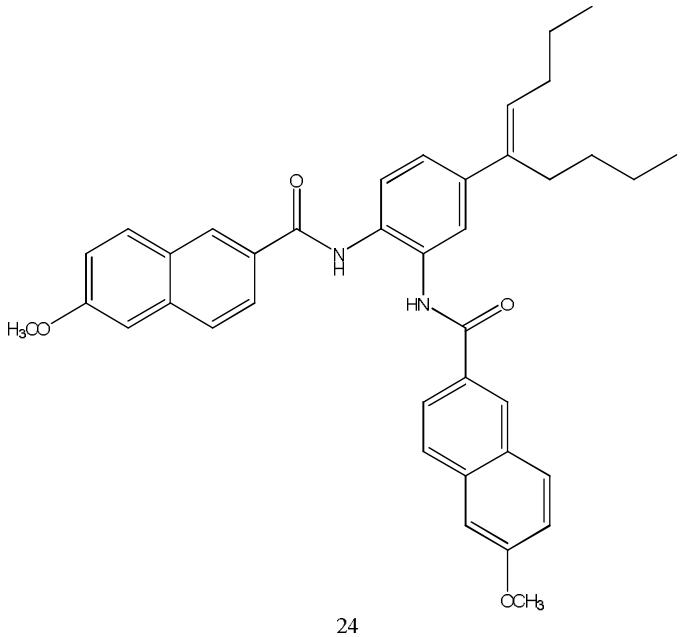
23

Figure 4.17. (E)-4-(non-4-en-5-yl)benzene-1,2-diamine (23).

4.3.18. Synthesis of (E)-N, N'-(4-(non-4-en-5-yl)- 1,2-phenylene) bis(6-methoxy-2-naphthamide) (24)

The reaction was done according to the literature procedure [45]. 6-methoxy-2-naphthoic acid (20.2 mg, 0.1 mmol) was added inside of a two necked round bottom flask fitted with a magnetic stirrer under inert atmosphere by using nitrogen gas. 2 mL dry CH_2Cl_2 was added inside of round bottom flask. PBr_3 (0.0142 ml, 0.15 mmol) was added under inert conditions. Reaction was distilled and stirred at 37 °C for 3 hours. Then TEA

(0.3 mmol, 0.041 ml) was added to the reaction in 10 minutes drop by drop. When HBr formation stopped (E)-4-(non-4-en-5-yl)benzene-1,2-diamine compound 23 (20 mg, 0.086 mmol) was added to the round bottom flask under nitrogen gas. Reaction was stirred for 3 hours at room temperature. The reaction progress was monitored by thin layer chromatography (TLC) using silica gel plates and 2% MeOH / CH_2Cl_2 as the eluent phase. Solid product was filtered and yellowish product was obtained. The product was impure so and purified using silica gel chromatography. Product was dissolved in minimum amount of CH_2Cl_2 (0.5 mL), then it was injected through a straight line on silica plate (22×13cm). Silica plate was put inside of a 2% MeOH / CH_2Cl_2 solvent mixture. Desired layer was scratch out, and dissolved in 5% MeOH / CH_2Cl_2 solvent and filtrated. After concentrating the solvent 34 mg white, solid, pure final product was obtained in 66 % yield. ^{1}H NMR ($CDCl_3$) δ 9.42 (s, 2H, NH_2), 9.37 (s, 2H, NH_2), 8.50(s, 1H, ArH), 8.42 (s, 1H, ArH), 8.02 (d, 1H, ArH), 7.95 (d, 1H, ArH), 7.80(d, 2H, ArH, ArH), 7.75(d, 2H, ArH, ArH), 7.46(d, 1H, ArH), 7.30(s, 1H, ArH), 7.19 (s, 2H, ArH, ArH) 7.15(d, 2H, ArH, ArH), 6.77 (d, 1H, ArH), 5.11 (t, 1H, CH),), 3.88(s, 6H, OCH_3, OCH_3), 1.95(t, 2H, CH_2), 1.63(q, 2H, CH_2), 1.18 (m, 2H, CH_2), 0.91 (m, 4H, CH_2, CH_2), 0.72 (t, 3H, CH_3), 0.58 (t, 3H, CH_3),



24

Figure 4.18. (E)-N,N'-(4-(non-4-en-5-yl)-1,2-phenylene)bis(6-methoxy-2-naphthamide) (24).

5. FUTURE WORK

5.1. Synthetic Approach for Alcohol Reduction

For future work alcohol reduction of compound 4 was targeted in the presence of $HSiEt_3$ and catalytic amount of BF_3 as Lewis Acid. Deprotection will be carried out by adding sulfuric acid in MeOH and coupling reaction will be carried out by using PBr_3. Hence, compound 27 is targeted as a new final product.

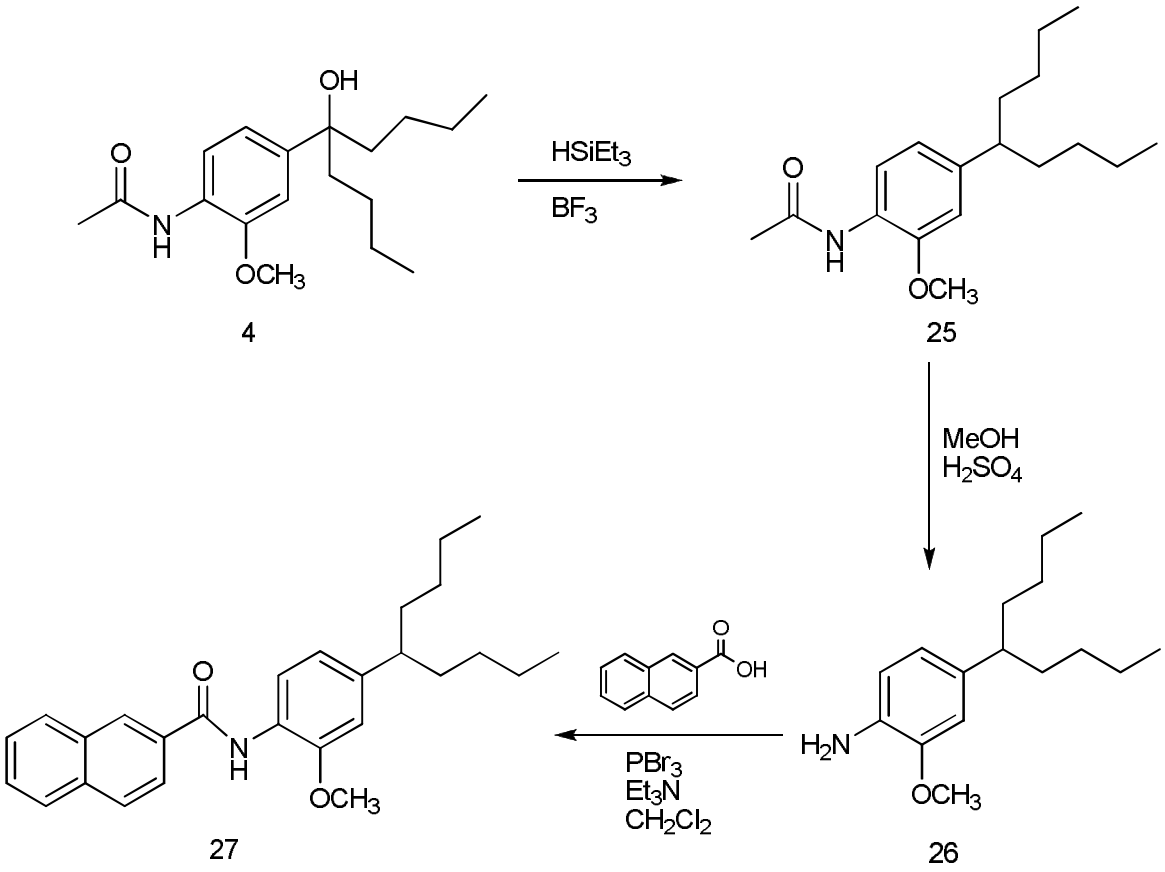


Figure 4.19. Synthetic approach for alcohol hydrogenolysis.

6. CONCLUSION

The aim of this study was to synthesize derivatives of the lead compound 1 and find biologically active drug molecules for the inhibition of CYP17 enzyme. Lead compound have two parts such as left hand side (the naphthyl unit) and the right hand side (the phenyl unit). In this thesis, phenyl rings substituted with a branched alkyl group together with amine and alcohol groups were synthesized successfully. The key step in obtaining the branched alkyl group was the double addition of n-BuLi to a chosen benzoate ester. The final products were purified and sent for biological testings.

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APPENDIX A: SPECTROSCOPY DATA

^{1}H and ^{13}C NMR spectroscopy of the synthesized products are included. Necessary expansions were made on the NMR data.

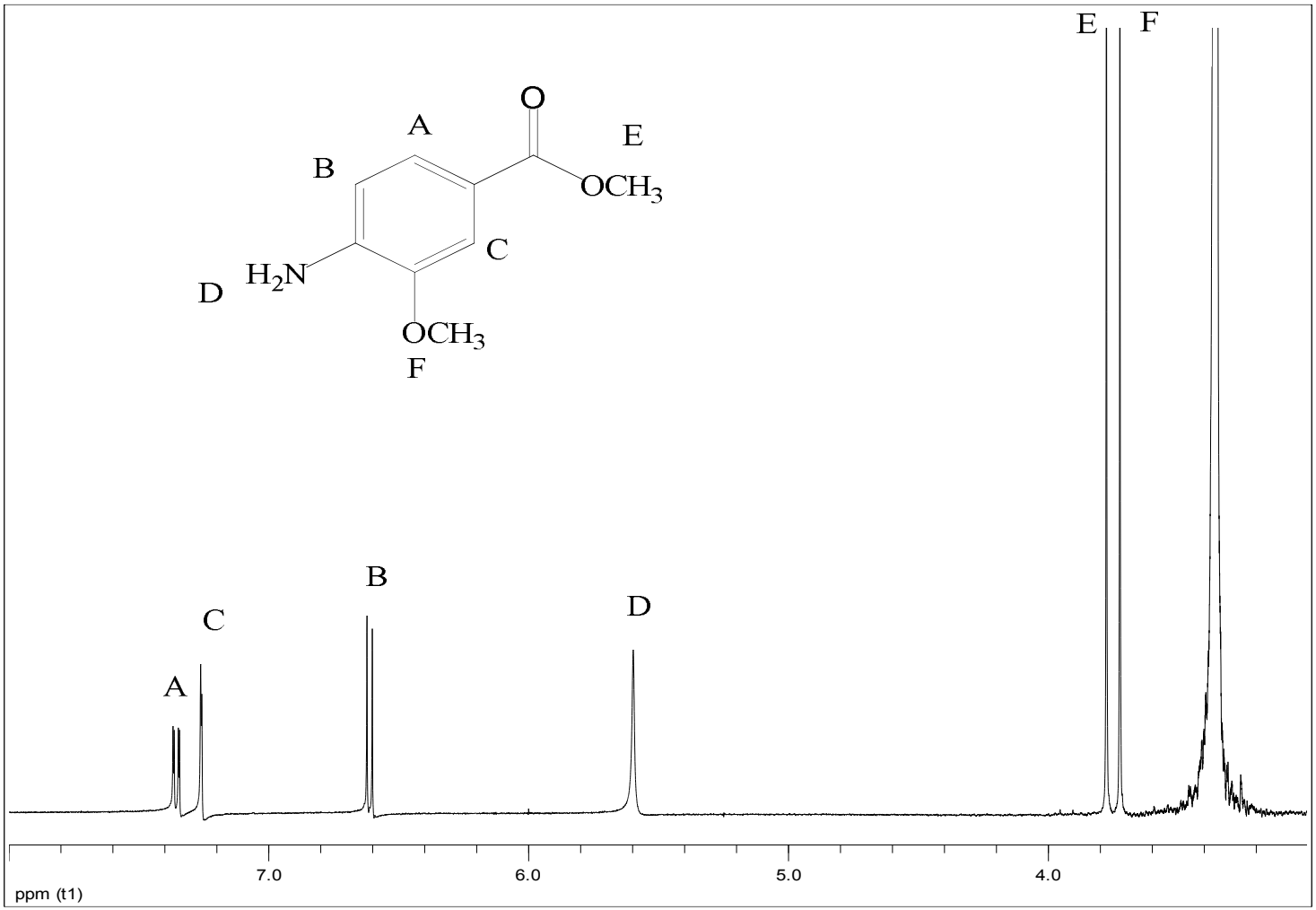

Figure A.1. ^{1}H-NMR of Methyl 4-amino-3-methoxybenzoate.

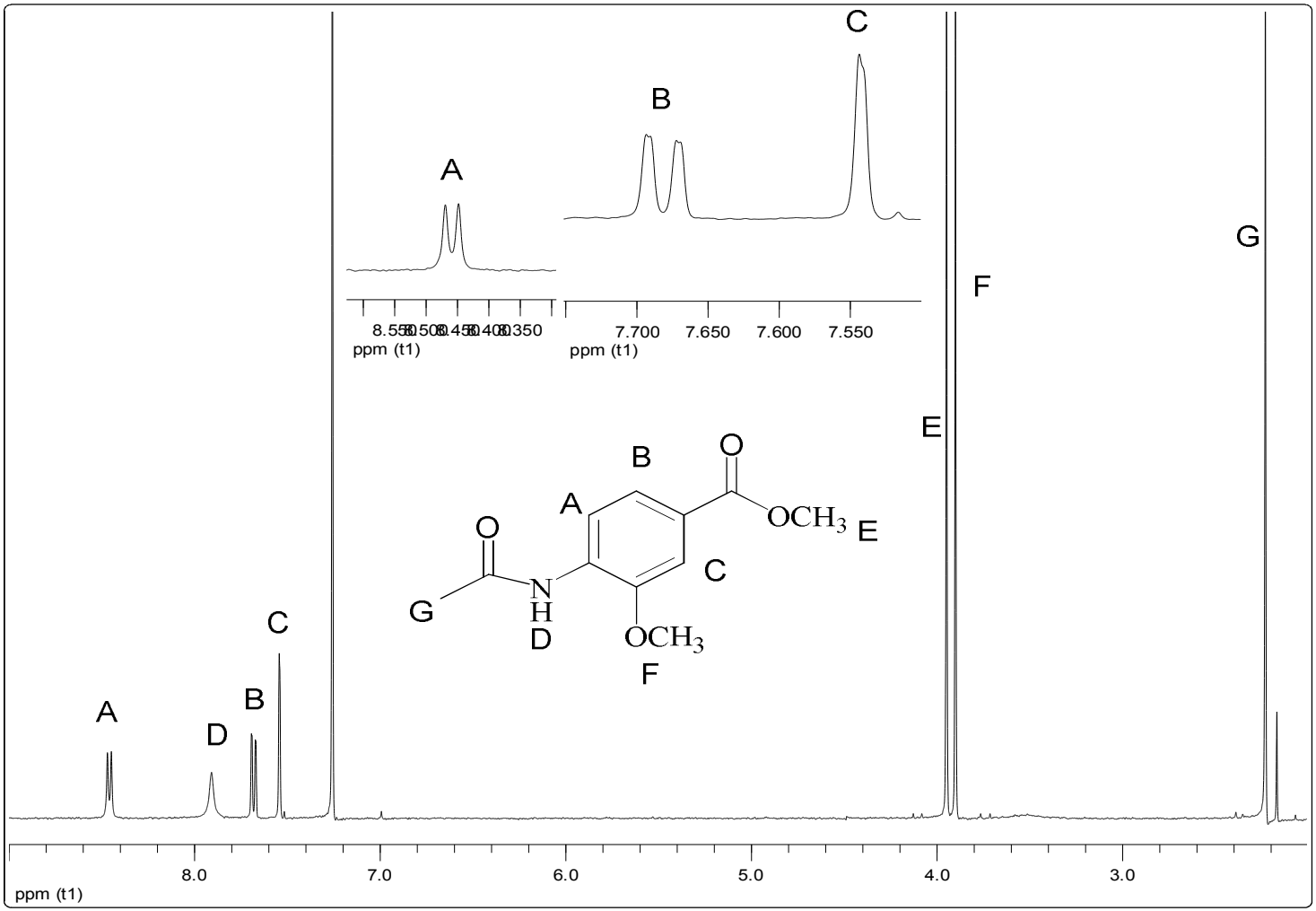

Figure A.2. ^{1}H-NMR of Methyl 4-acetamido-3-methoxybenzoate.

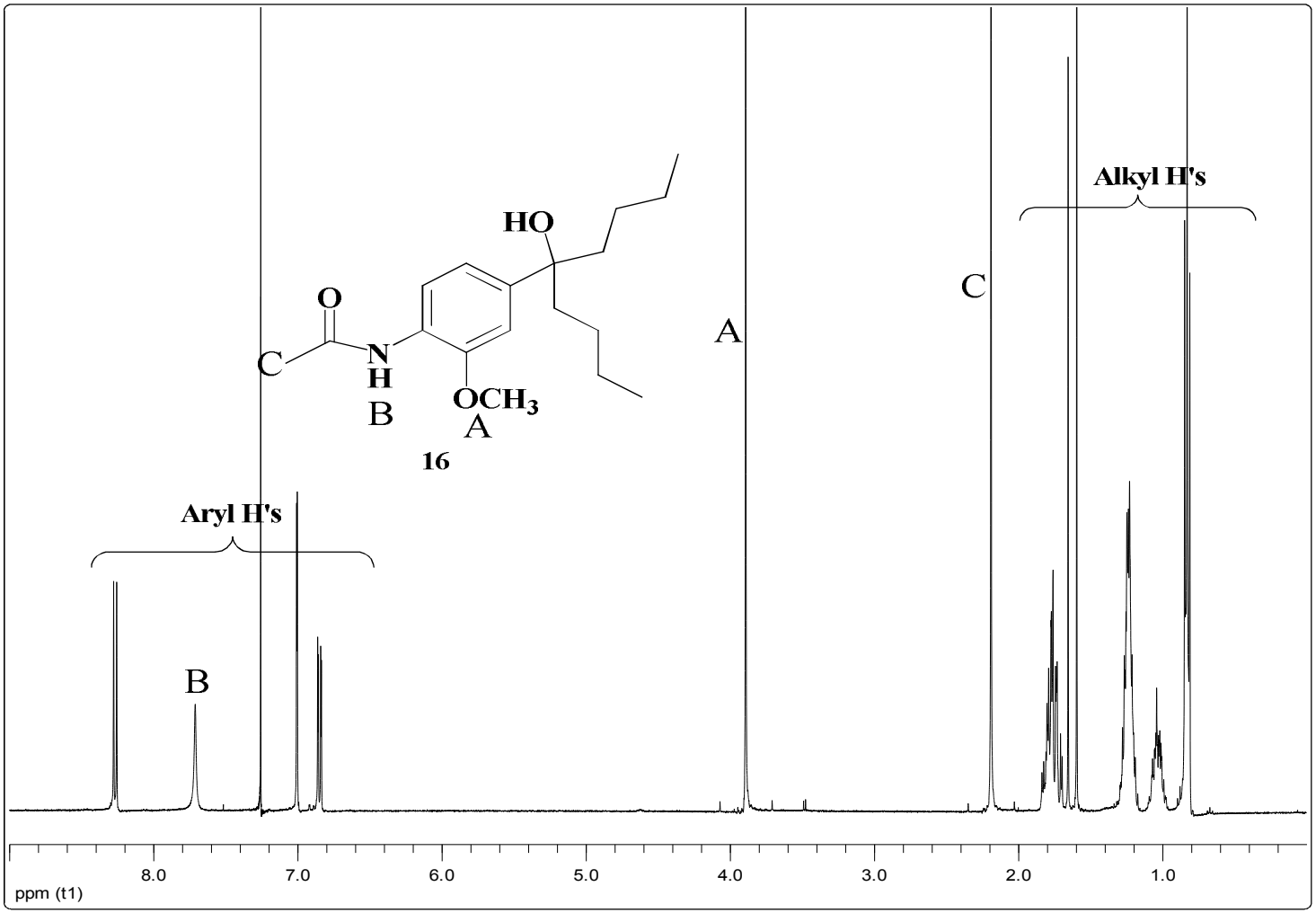

Figure A.3. ^{1}H-NMR of N-(4-(5-hydroxynonan-5-yl)-2-methoxyphenyl)acetamide.

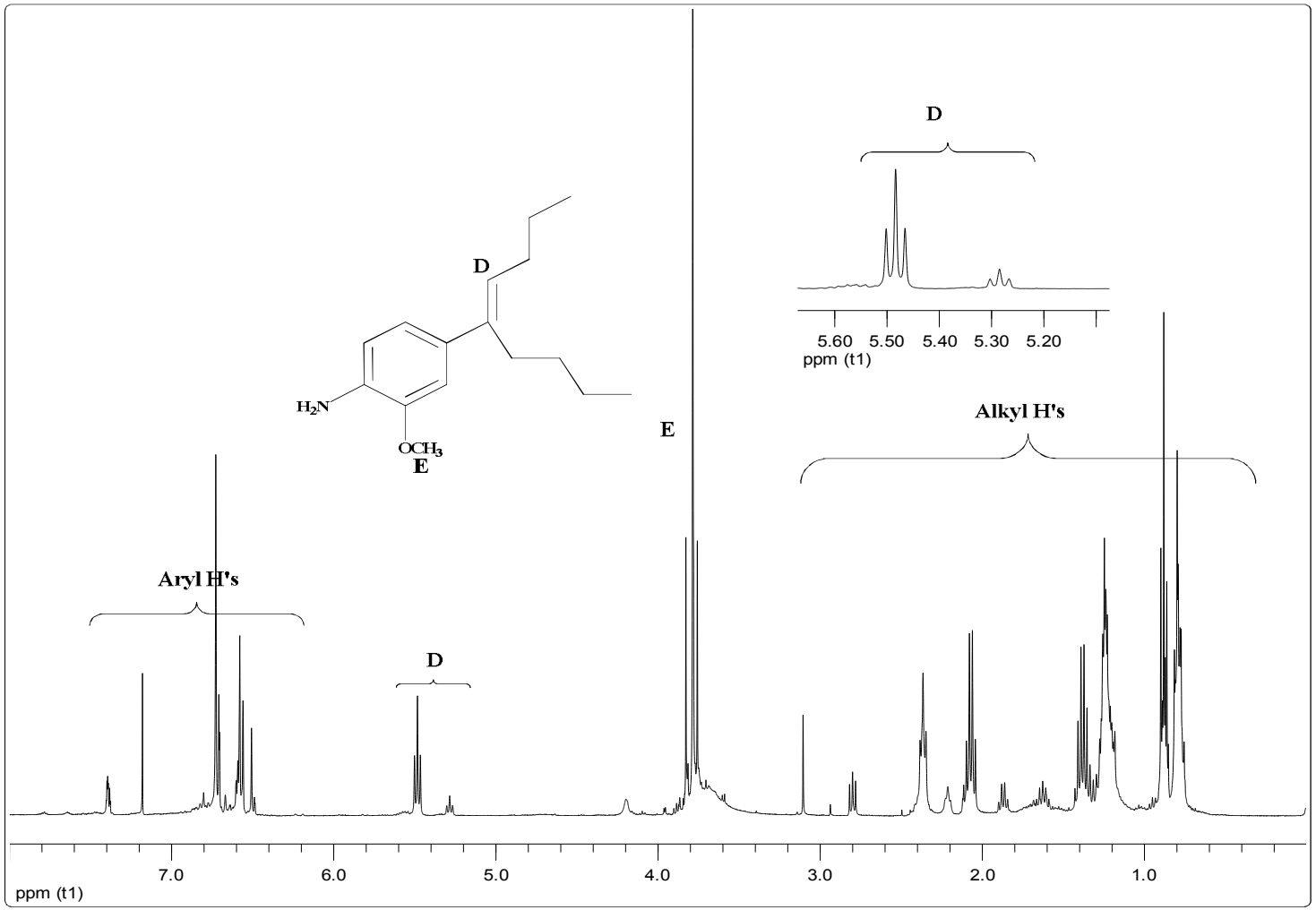

Figure A.4. ^{1}H-NMR of (E)-2-methoxy-4-(non-4-en-5-yl)aniline.

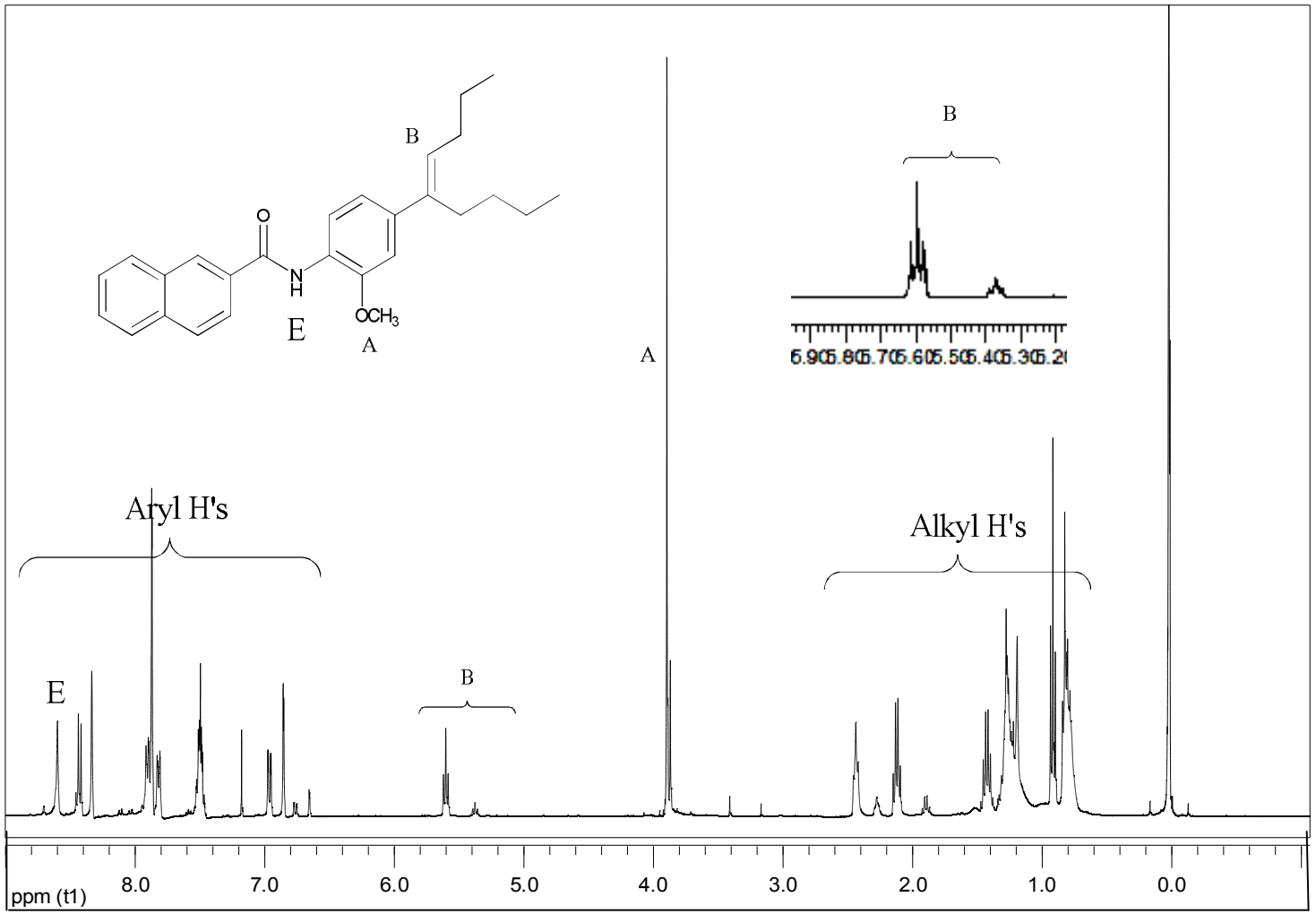

Figure A.5. ^{1}H-NMR of (E)-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide.

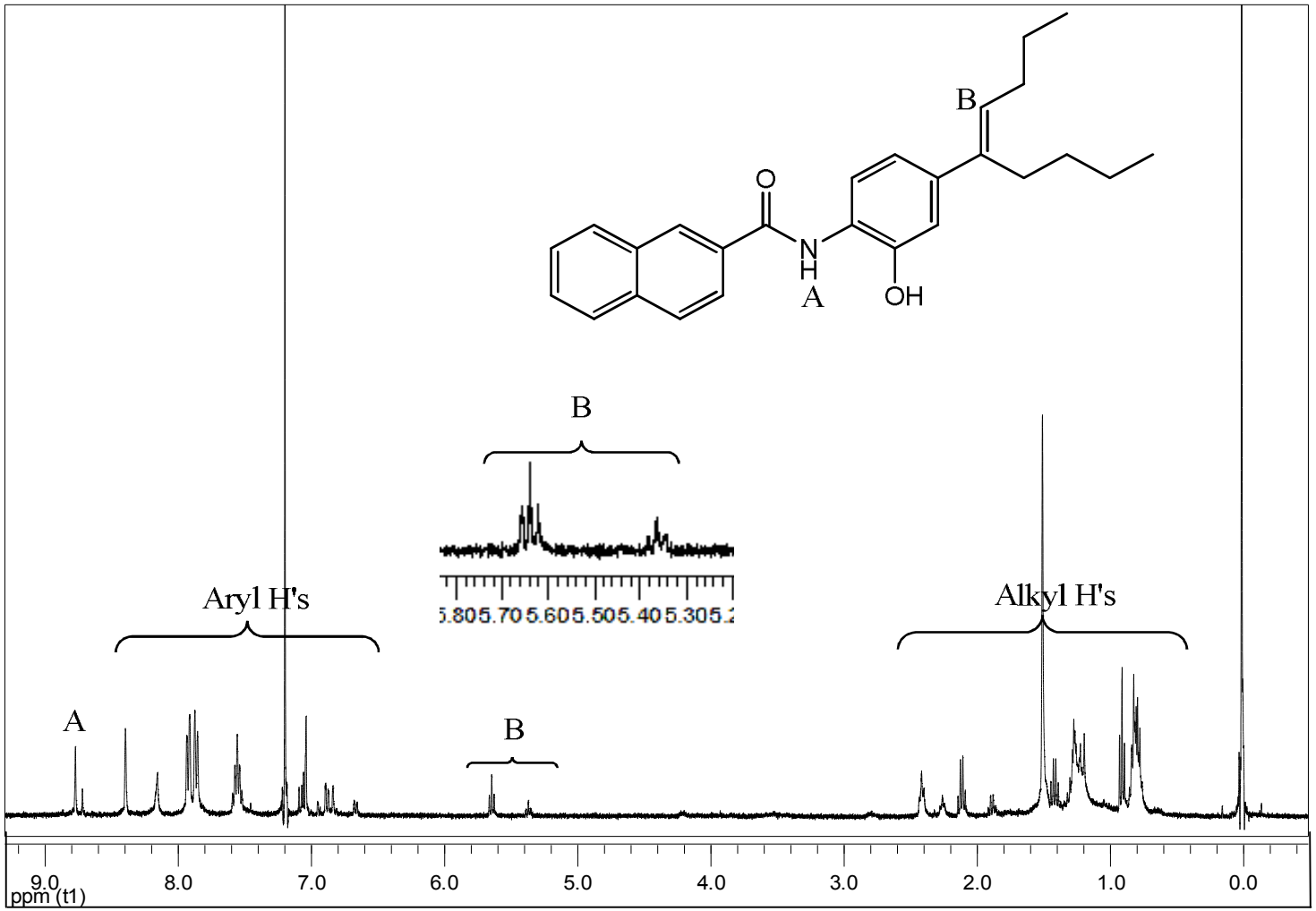

Figure A.6. ^{1}H-NMR of (E)-N-(2-hydroxy-4-(non-4-en-5-yl)phenyl)-2-naphthamid.

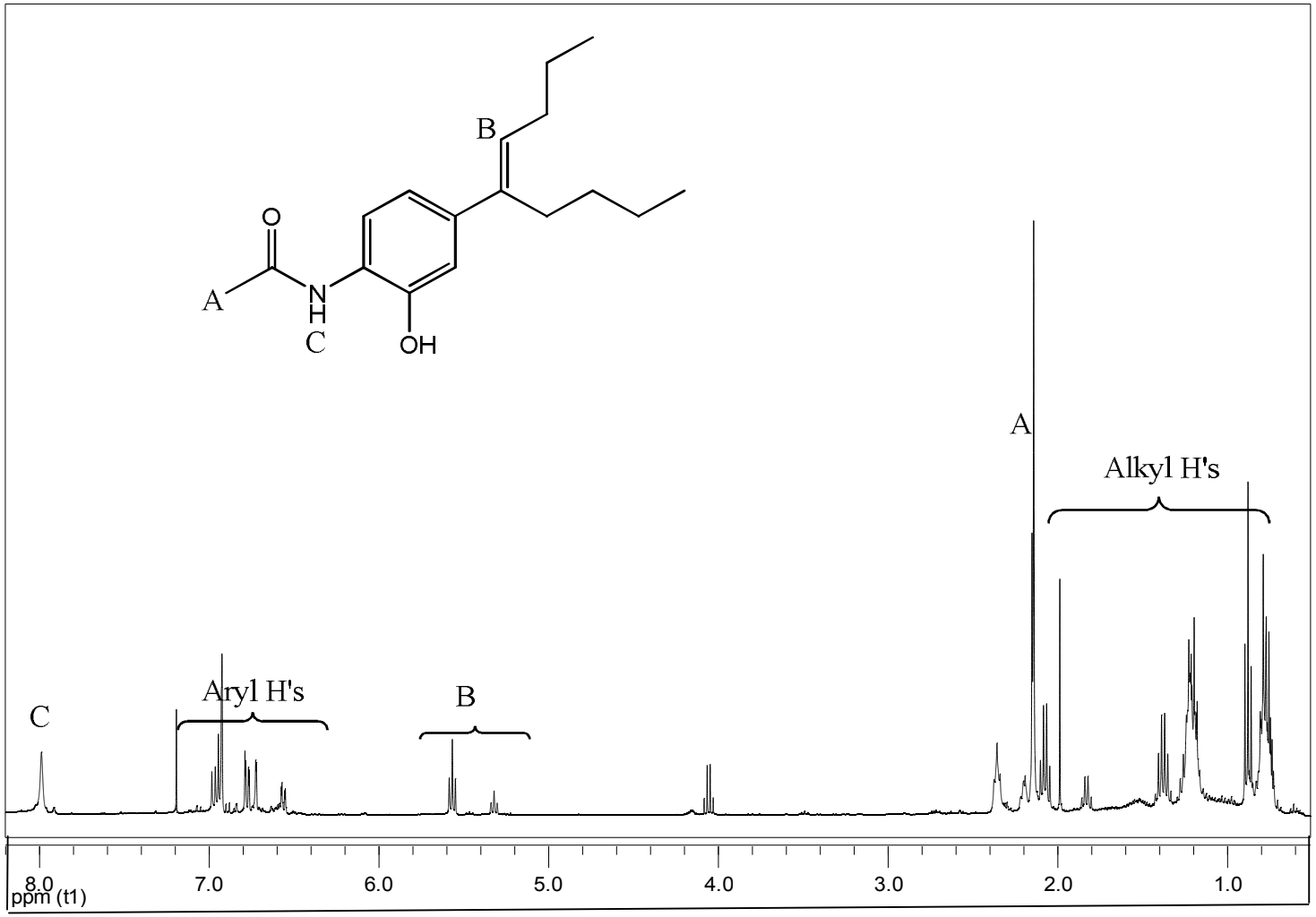

Figure A.7. ^{1}H-NMR of (E)-N-(2-hydroxy-4-(non-4-en-5-yl)phenyl)acetamide.

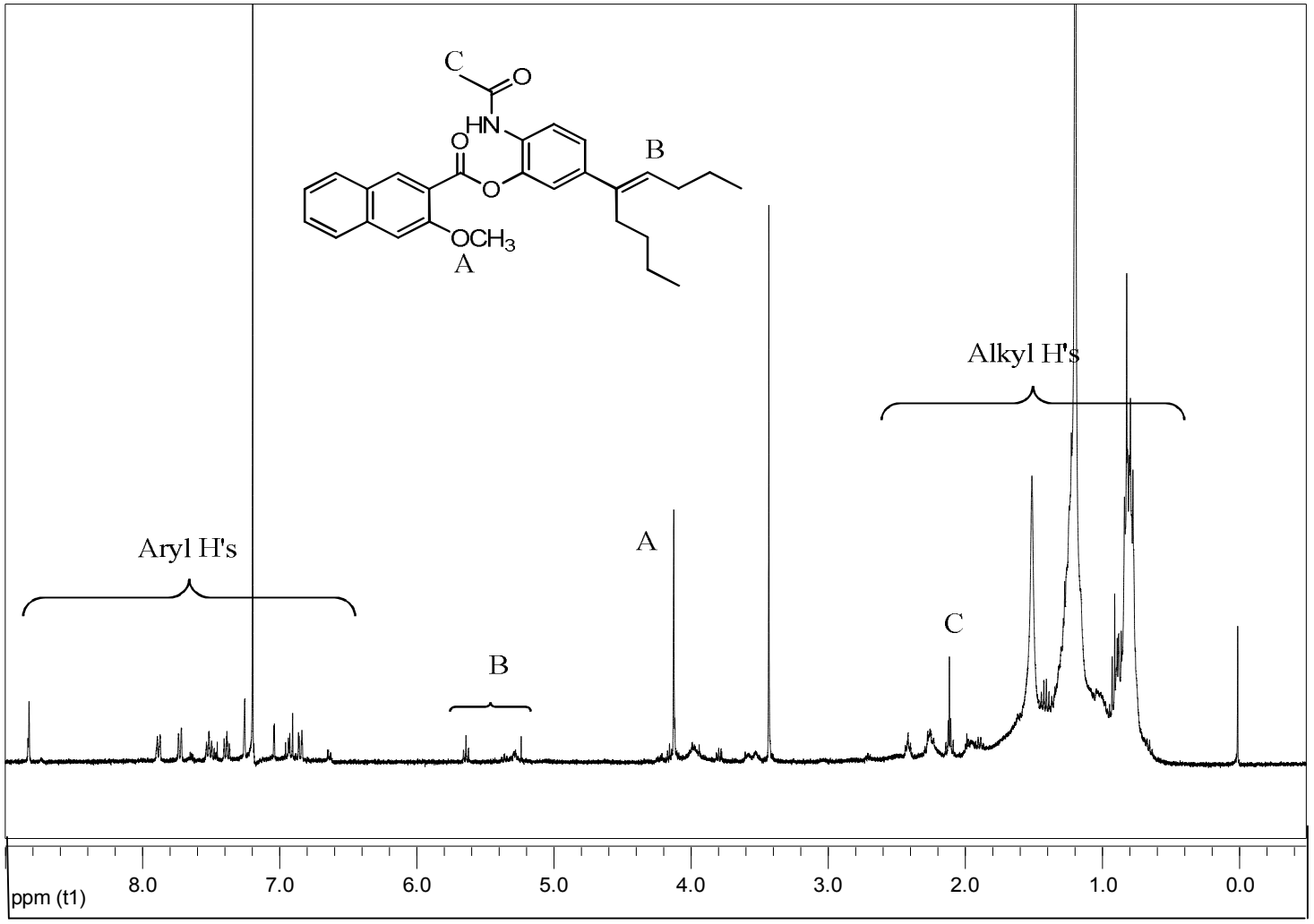

Figure A.8. ^{1}H-NMR of (E)-2-acetamido-5-(non-4-en-5-yl)phenyl 3-methoxy-2-naphthoate.

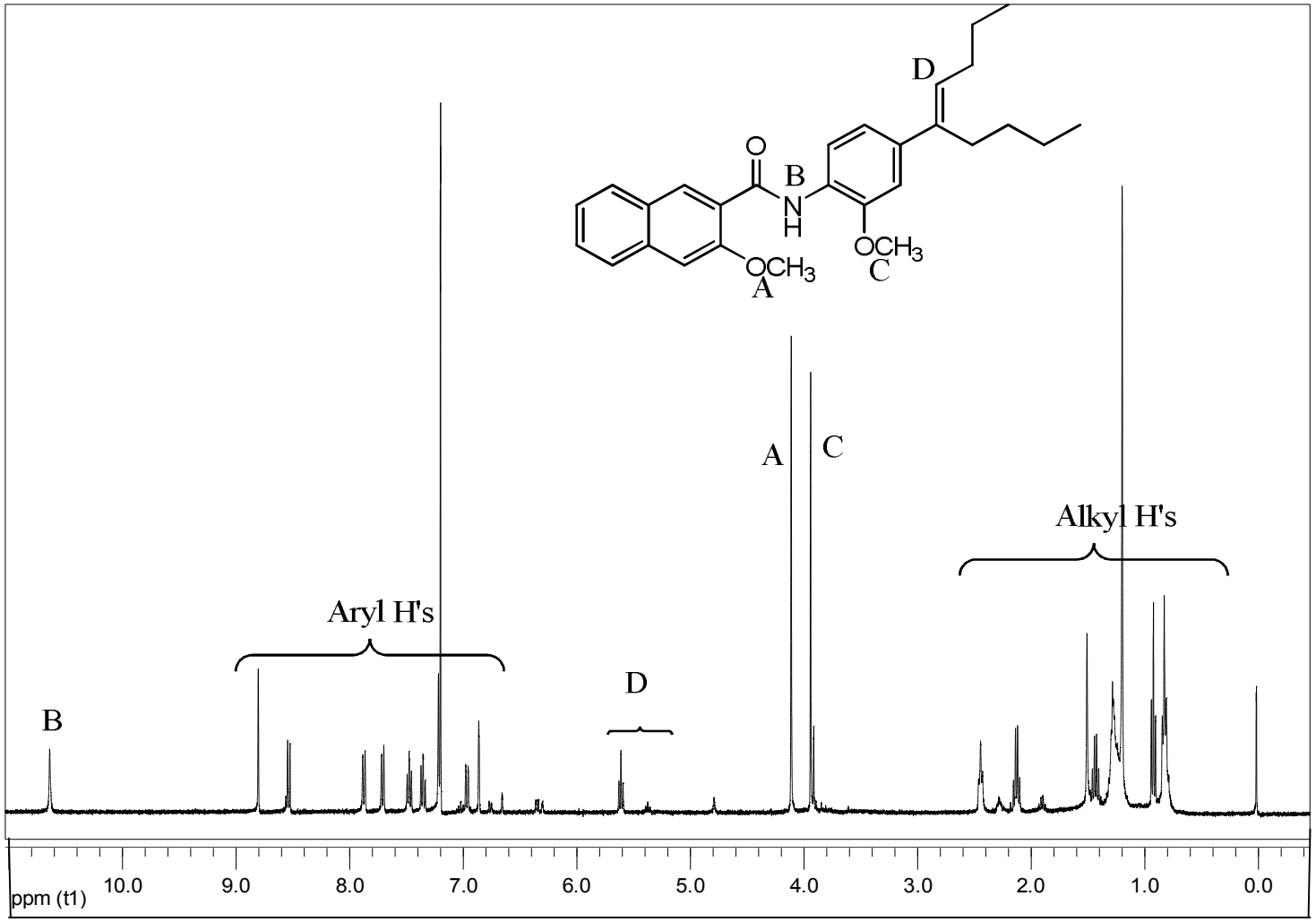

Figure A.9. ^{1}H-NMR of (E)-3-methoxy-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide.

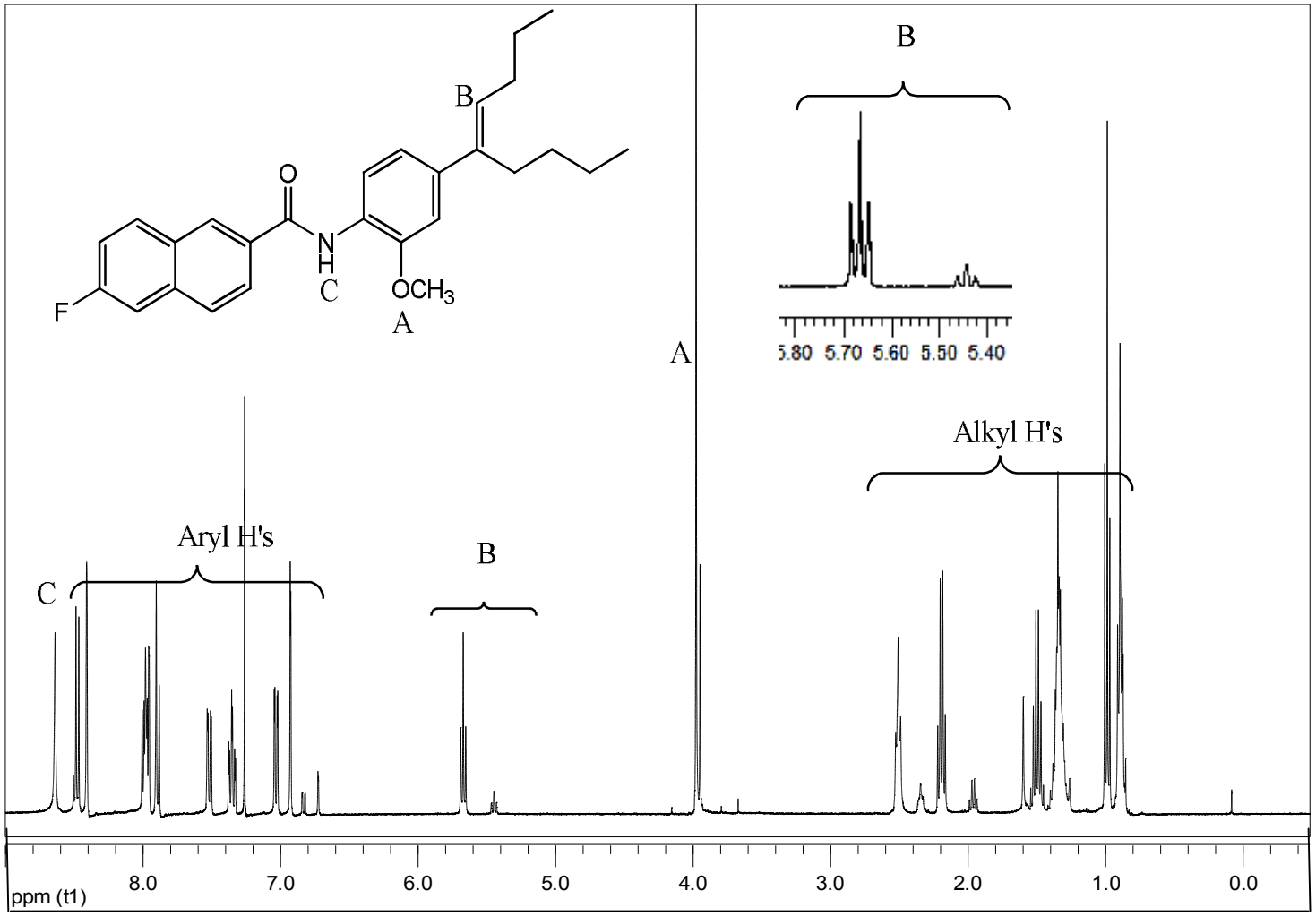

Figure A.10. ^{1}H-NMR of (E)-6-fluoro-N-(2-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide.

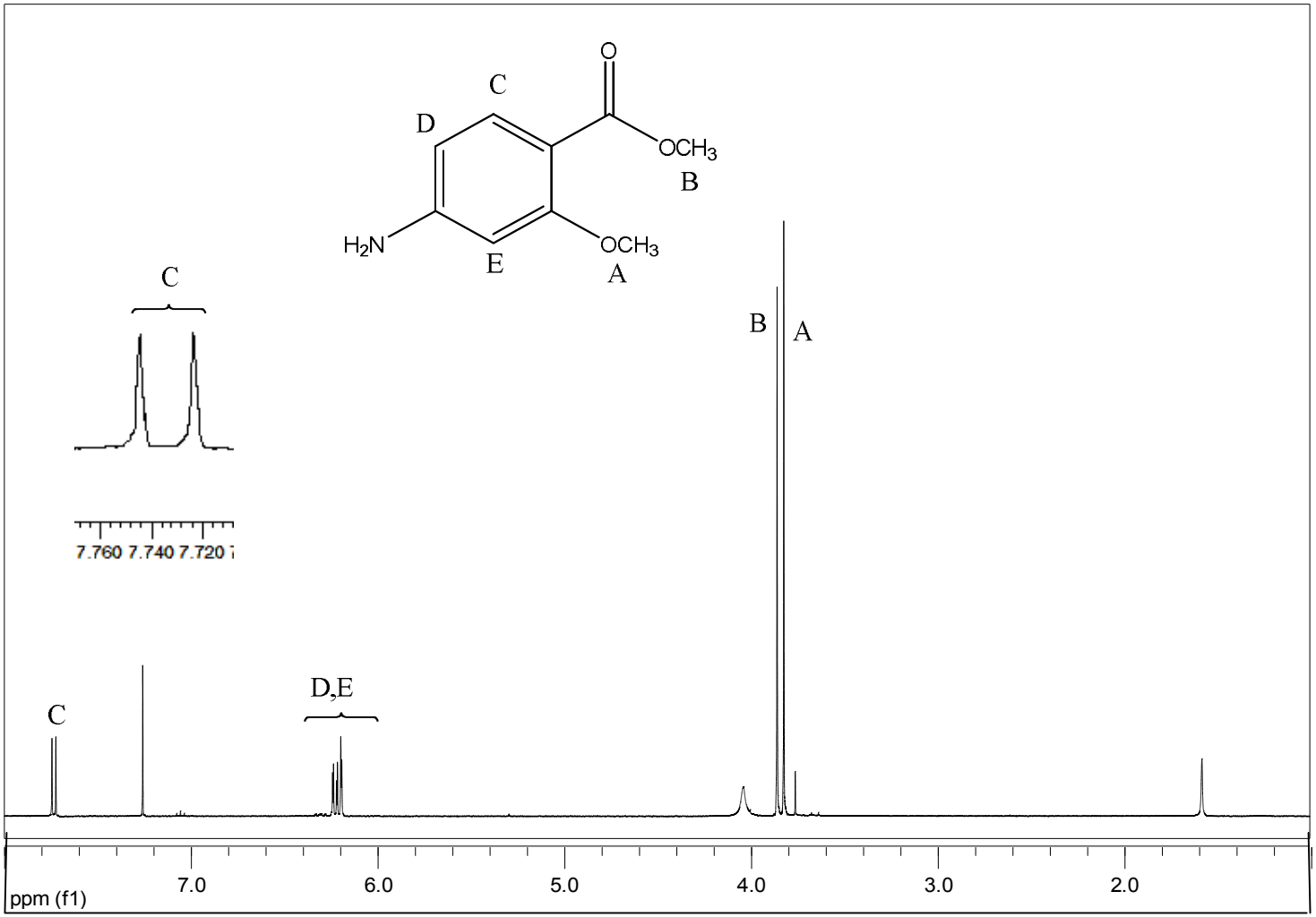

Figure A.11. ^{1}H-NMR of methyl 4-amino-2-methoxybenzoate.

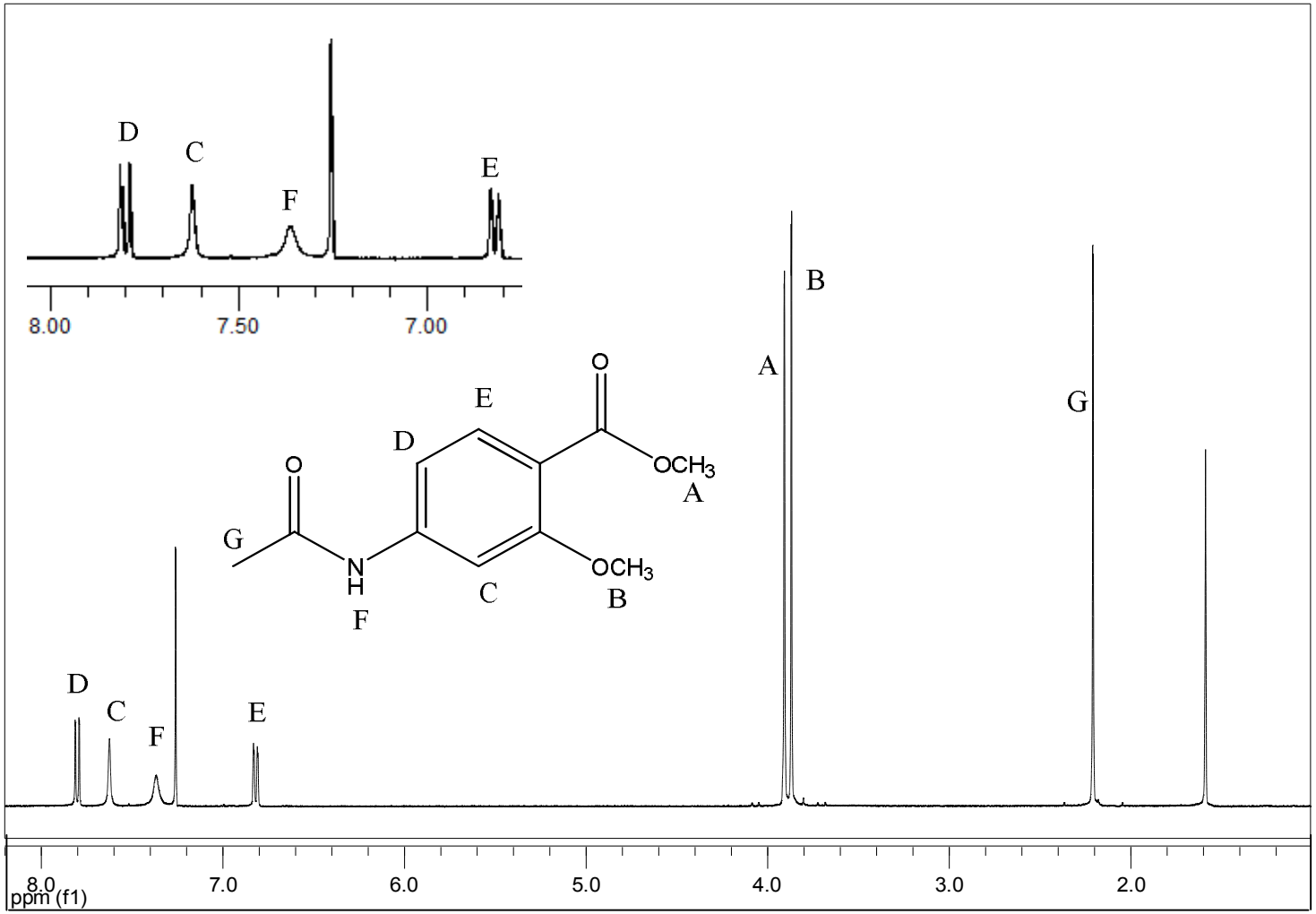

Figure A.12. ^{1}H-NMR of methyl 4-acetamido-2-methoxybenzoate.

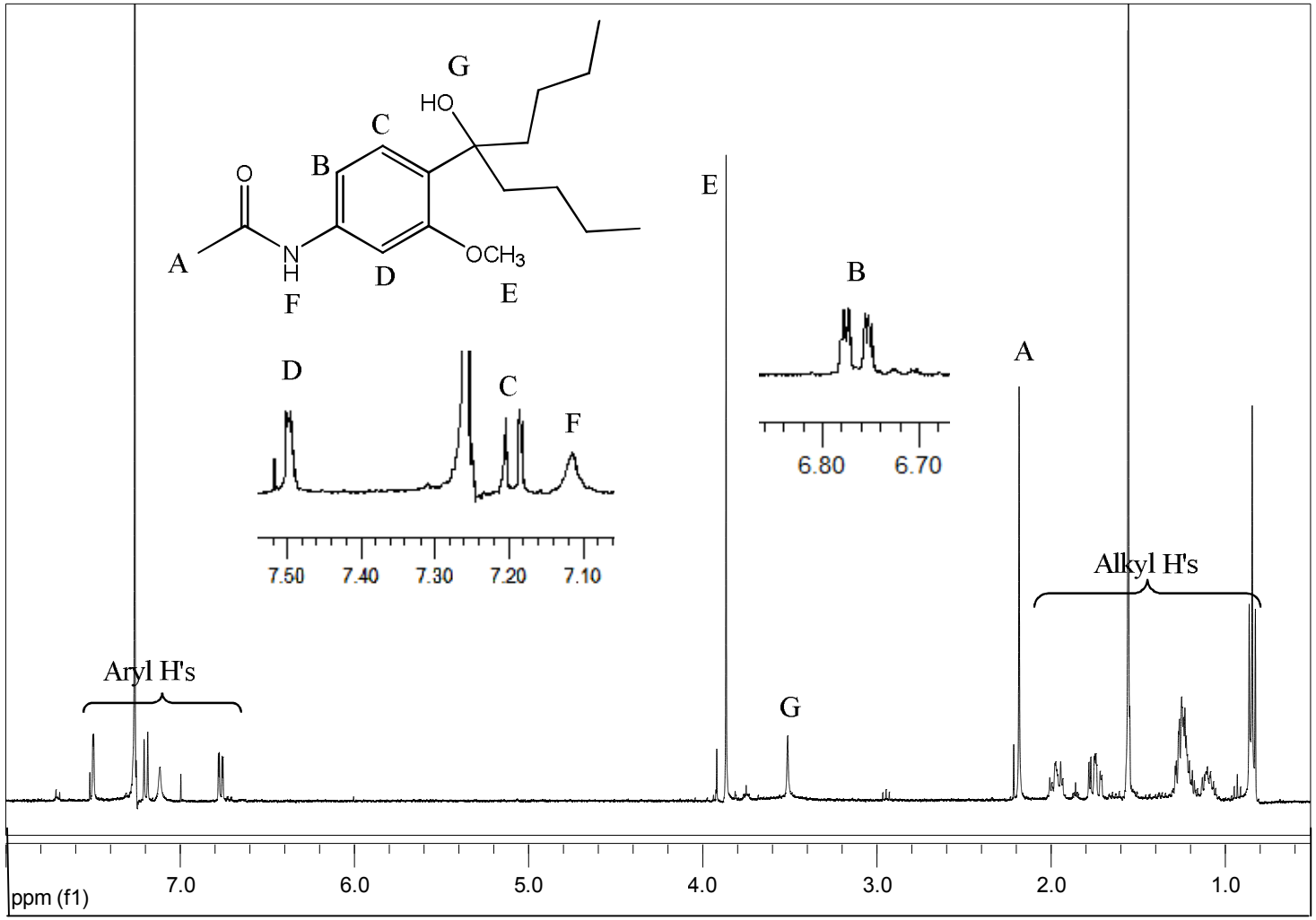

Figure A.13. ^{1}H-NMR of N-(4-(5-hydroxynonan-5-yl)-3-methoxyphenyl)acetamide.

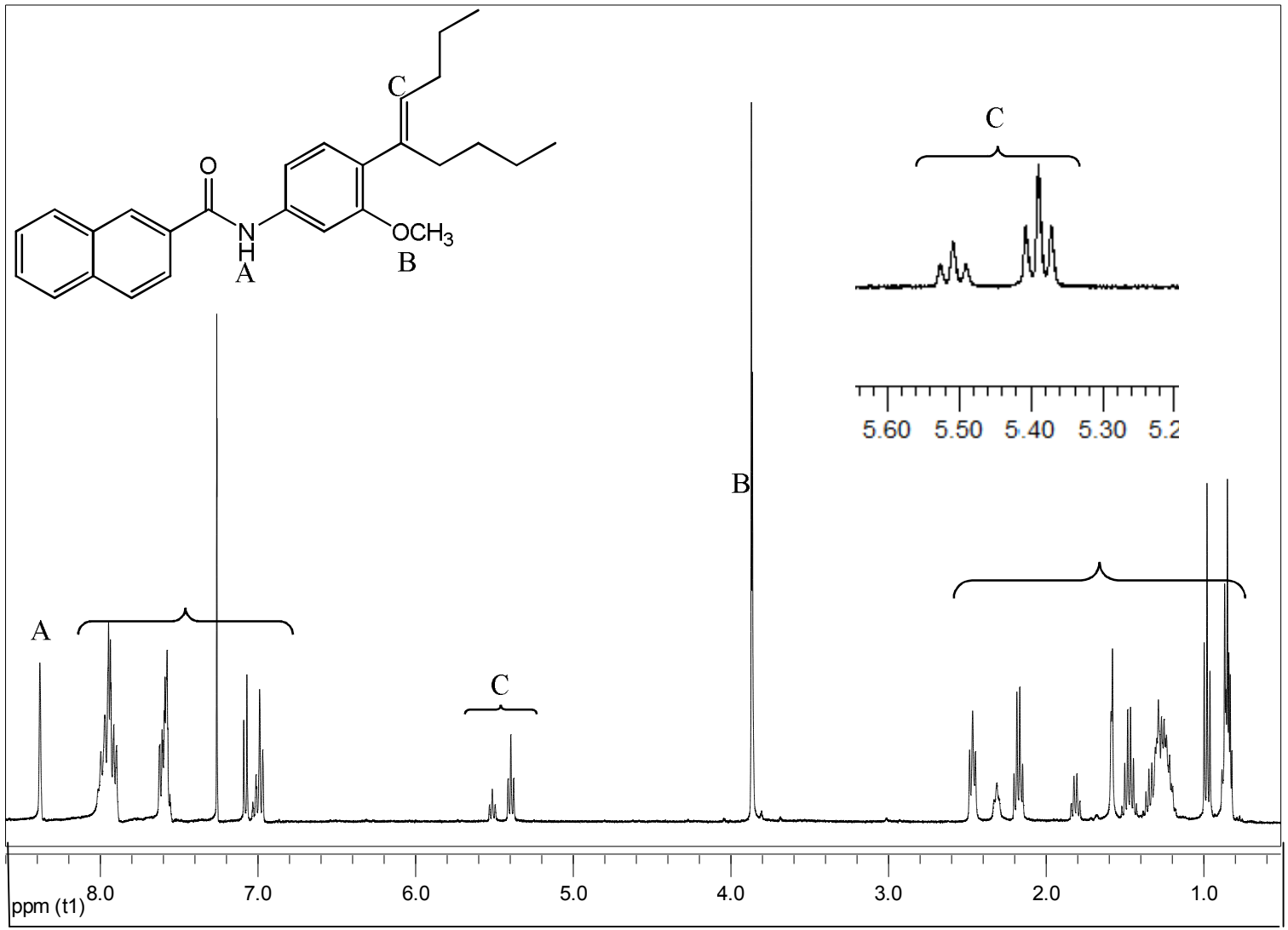

Figure A.14. ^{1}H-NMR of (E)-N-(3-methoxy-4-(non-4-en-5-yl)phenyl)-2-naphthamide.

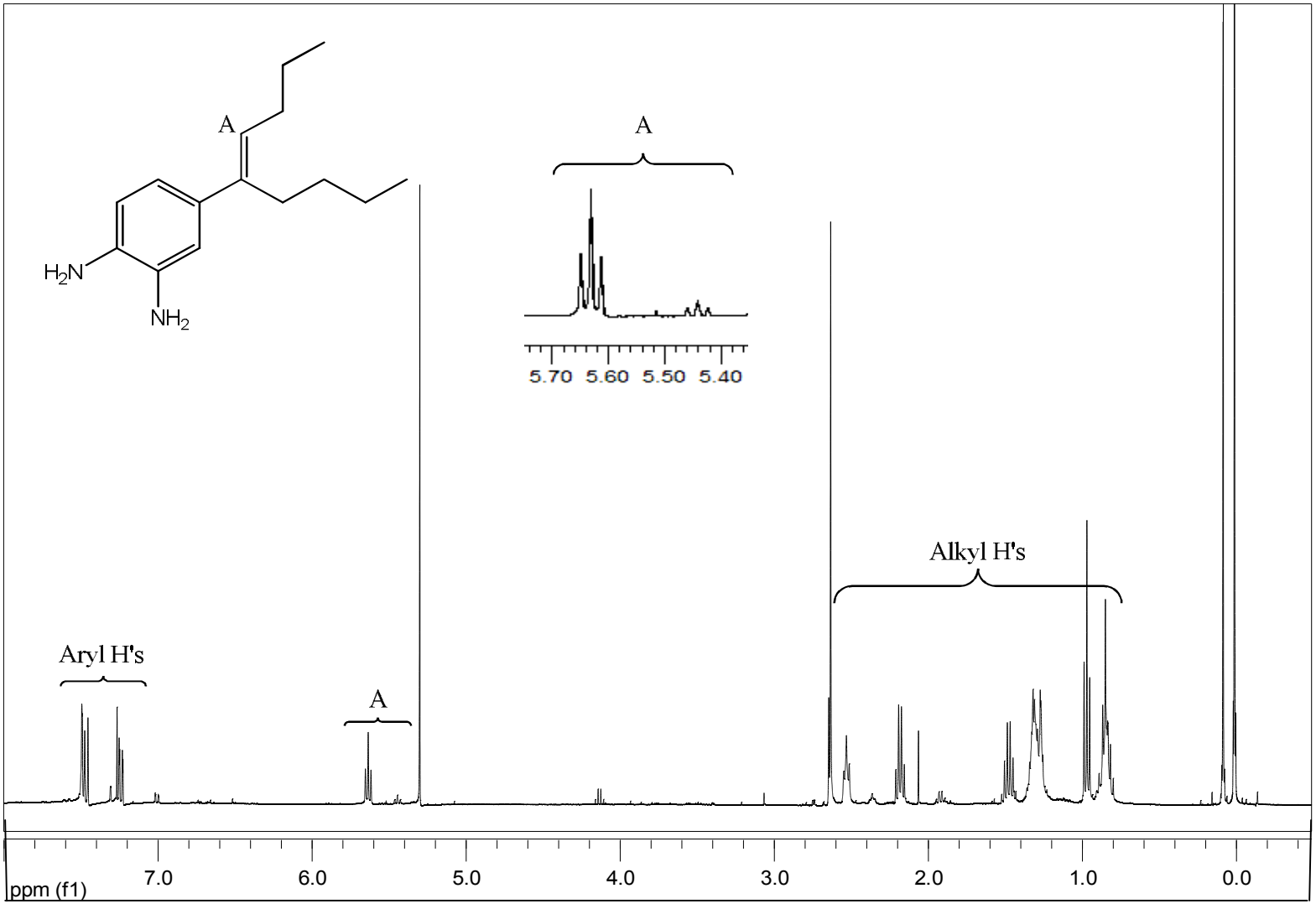

Figure A.15. ^{1}H-NMR of (E)-4-(non-4-en-5-yl)benzene-1,2-diamine.

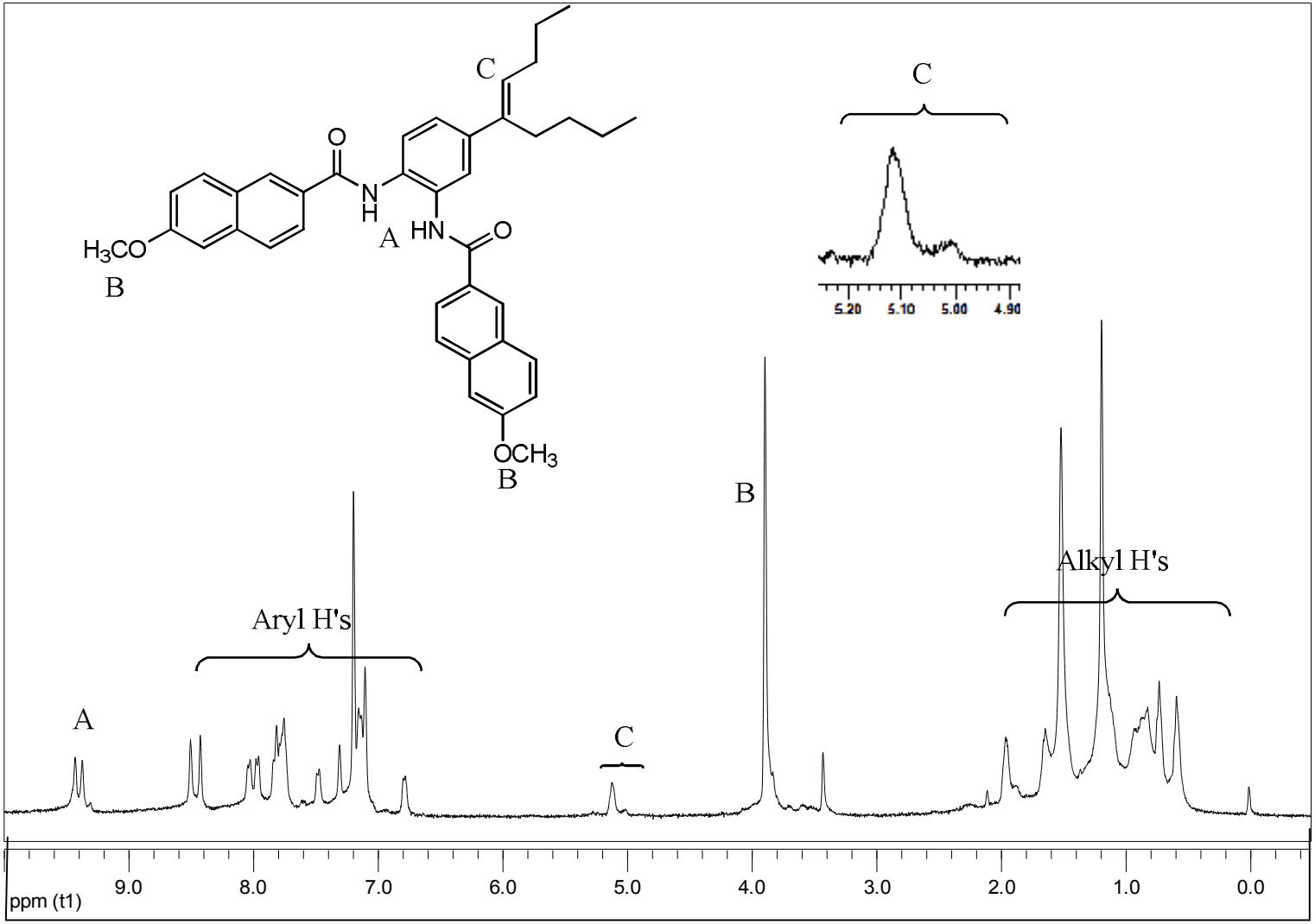

Figure A.16. ^{1}H-NMR of (E)-N,N'-(4-(non-4-en-5-yl)-1,2-phenylene)bis(6-methoxy-2-naphthamide).

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