SYNTHESIS AND <u>IN VITRO</u> BIOLOGICAL ACTIVITY OF NEW NON-STEROIDAL PLATINUM (II) COMPLEXES DESIGNED FOR THE TREATMENT OF BREAST CANCER

YUEHUA HE

SYNTHESIS AND IN VITRO BIOLOGICAL ACTIVITY OF NEW NON-STEROIDAL PLATINUM (II) COMPLEXES DESIGNED FOR THE TREATMENT OF BREAST CANCER

by

YUEHUA HE

A thesis submitted to the

School of Graduate Studies

in partial fulfilment of the

requirements for the degree of

Master of Science

School of Pharmacy
Memorial University of Newfoundland
St. Jonh's, Newfoundland

January, 1995

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ABSTRACT

Breast cancer is the leading form of cancer among women in North America. The development of resistance to endocrine therapy as well as chemotherapy is presently the major obstacle to successful treatment of advanced breast cancer. Therefore, more potent and selective chemotherapeutic agents should be designed. An attractive solution to this problem is to combine both endocrine therapy and chemotherapy in a single agent. It may result in a more powerful approach to advanced breast cancer treatment.

In order to achieve this goal, a series of new triphenylethylene platinum (II) complexes **39a-d**, **40a-c** and **41** have been designed and synthesized. The commercially available benzyl, 4-hydroxyphenyl ketone was efficiently transformed in eight steps into the platinum (II) complexes **39a-d** with an overall yield of around 30%. In a similar sequence of reactions, the complexes **40a-c** and **41** were also synthesized, the overall yield exceeded 40%. All new compounds were fully characterized by their infrared and ^{1}H, ^{13}C nuclear magnetic resonance and mass spectra. The final compounds **39a-d**, **40a-c** and **41** also passed element analysis.

The biological activity of the complexes **39a-d**, **40a-c** and **41** were evaluated in vitro on both ER^{+} and ER^{-} breast cancer cell lines: MCF-7 and MDA-MD-231. The complexes **40b-c** showed promising antitumor activity. Their IC_{50} is up to 28 fold lower than tamoxifen on MDA-MD-231, and 3 fold lower on MCF-7. However, there was no evidence of selective antitumor activity on ER^{+} breast cancer cell in vitro.

ACKNOWLEDGEMENTS

I would like to extend my sincere appreciation to my supervisor, Dr. Gervais Bérubé, for his many hours of helpful instruction and discussion during the course of my research project.

Special thanks are given to Ms. Roberts, Dr. Kantha Arunachalam, Achilles Lau, Stephen Sherman, Cida Zhao, Dongyuan Yao, Sue Ellen Maher and John Bautista for their support, encouragement, and friendship.

I am grateful to Ms. Natalie Brunet for 300 NMR spectra, Ms. M. Baggs, Dr. Liang Tang and Dr. Feimeng Zhou for mass spectra. Special acknowledgement is given to Dr. René C.-Gaudreault and Mr. Jacques Lacroix for biological testing. I thank my supervisory committee, Dr. Ford and Dr. Loomis for their support during my study. I would like to thank Département de Chimie-Biologie, Université du Québec à Trois-Rivières for providing me laboratory space to accomplish my research work.

Sincere thanks are due to Mrs. Daphne Efford for her help and friendship. I am deeply grateful to my husband, my parents for their love, support, and encouragement.

The financial support from School of Pharmacy, Memorial University of Newfoundland in the form of a Memorial Graduate Fellowship and Medical Research Council of Canada is gratefully acknowledged.

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GLOSSARY OF ABBREVIATIONS

d	doublet
dd	double doublet
DHP	dihydropyran
DMF	N, N-dimethylformamide
DMSO	dimethyl sulfoxide
E_2	estradiol
EGFR	epidermal growth factor receptors
ER	estrogen receptor
ER^+	estrogen receptor positive
ER^-	estrogen receptor negative
ERBA	estrogen receptor binding affinity
ERE	estrogen response elements
Et	ethyl
hr	hour
IGF	insulin like growth factors
IR	infrared spectroscopy
m	multiplet
MAb	monoclonal antibody
Me	methyl
mp	melting point
MS	mass spectroscopy
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide

NMR	nuclear magnetic resonance spectroscopy
PBS	phosphate-buffered saline
Ph	phenyl
PPTS	pyridinium, para-toluenesulphonate
***p*TSA**	para-toluenesulphonic acid
q	quartet
RPMI	Roswell Park Memorial Institute
s	singlet
TGF	tumor growth factors
THF	tetrahydropyranyl
THP	tetrahydrofuran
TLC	thin layer chromatography
TMS	tetramethylsilane
TPA	triphenylacrylonitrile

TO MY HUSBAND AND MY PARENTS

YOU MAKE IT ALL WORTH IT

Chapter 1

INTRODUCTION

Breast cancer is the most common form of cancer among women in North America. In 1994, it is estimated that 17,000 women will be diagnosed with breast cancer in Canada, representing approximately 30% of new cancer cases for all sites. Of the 27,600 cancer deaths, approximately 5,600 (20%) will be caused by breast cancer.[1] Therefore, finding an effective method to treat breast cancer is an important and urgent matter.

Currently, surgery with adjuvant radiotherapy is quite effective to treat breast cancer when the tumour has not metastasized by the time of treatment. However, even in the best circumstance, ten year survival rates of 50% have been unusual and some "clinical cures" may recur with fatal outcome as late as twenty years with such local treatment.[2] Therefore, a systemic approach such as chemotherapy plays an important role for a more effective cancer management.

At present, different types of drugs are used in chemotherapy, such as alkylating agents, DNA-intercalating agents, antibiotics, antimitotic agents, antimetabolites and so on. An ideal anticancer drug would eradicate cancer cells without harming normal tissues. Unfortunately, no currently available agents meet this criterion and clinical use of these drugs involves a weighing of benefits against toxicity in a search for a favorable therapeutic index.[3]

Another major problem in cancer chemotherapy is drug resistance, which means that tumors no longer respond to presently available chemotherapeutic agents. It is estimated that over 90% of all cancer death are, in some measure, influenced by the problem of drug resistance.[4] Some mechanisms of drug resistance have already been identified in human tumor cell lines. They include: decreased transport,[5] altered drug activation,[6] altered DNA repair,[7] gene amplification,[8] defective drug metabolism,[9] altered target proteins and altered intracellular nucleotide pools.[10] Faced with the complexity of the drug resistance mechanisms already identified, one might conclude that circumventing drug resistance is not a likely possibility.

An attractive solution which is being considered to solve these two problems is the use of drug targeting. The aim of drug targeting is to deliver drugs only to those sites needing treatment. When this objective is met, not only toxic side effects will be minimized, but the efficacy of the treatment will be improved. Therefore, the tumor might be eradicated rapidly before any sign of resistance occurs.

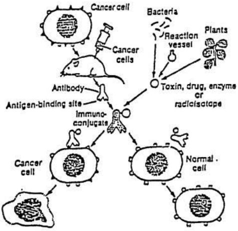
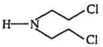


Fig.1 Immunotargeting with MAb for the treatment of cancer.

The concept of drug targeting was first suggested by Paul Ehrich in the early 1900's.[11] He proposed that chemotherapeutic agents might be covalently joined to ligand substrates which had affinity for and selectivity to a target tissue such as malignant tumors. Since then, some biological and chemical molecules have been tried as ligand substrates for anticancer drug targeting, such as monoclonal antibodies (MAb). By covalently linking antitumor agents to MAb reactive with tumor-associated antigens, these drugs can be targeted to the tumors (Fig.1).[12] Numerous scientists are still working on this area.

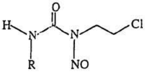
The presence of substantial amounts of estrogen receptors (ER) in many human breast tumors is well known and is being used to select the most appropriate therapy such as endocrine therapy and/or chemotherapy for breast cancer patients.[13] These receptors provide a potential targeting mechanism by which agents with estrogen receptor binding affinity (ERBA) could be concentrated selectively in the tumor tissue. For example, several groups have developed γ-emitting estrogens as diagnostic imaging agents for human breast cancer.[14] It might also prove possible to prepare conjugates of molecules with ERBA and cytotoxic agents, which would bind to ER and would thus concentrate their cytotoxic activity within ER-containing cells, sparing cells in nontarget tissues. It is this prospect of achieving a selective ER mediated killing of ER positive (ER^+) breast cancer cells that has concerned scientists in the area of breast cancer. This thesis is also devoted to preparing this type of antitumor agent.

The development of antitumor agents with ERBA for breast cancer has some encouraging experimental precedent. As early as the 1950s, several scientists have been trying to link nitrogen mustard (**1**), an alkylating agent, to a variety of steroid nuclei such as cholesterol (**4**), estrone (**5**), testosterone (**6**),



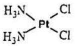
Nitrogen Mustard

1



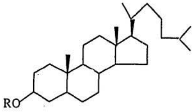
Nitrosoureas

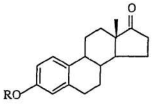
2



Cisplatin

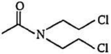
3

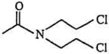


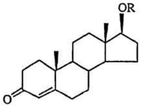


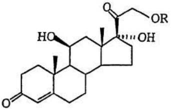
4 R=H Cholesterol

5 R=H Estrone

8 R= 

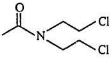
9 R= 

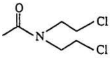




6 R=H Testosterone

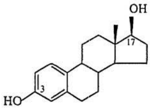
7 R=H Hydrocortisone

10 R= 

11 R= 

and hydrocortisone (**7**) to treat hormone dependent breast cancer.

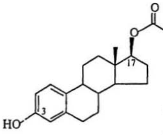
Unfortunately, the compounds obtained, i.e., **8**,[15] **9**,[16] **10**[17] and **11**[18] displayed only moderate antitumor activities. It is believed that these steroid nuclei are not the most appropriate ligand for the ER.



12 Estradiol (E_2)

Since the discovery of estradiol (**12**), the endogenous ligand for the ER, numerous attempts were made to conjugate it to alkylating agents. In the early attempts, alkylating agents such as nitrogen mustard (**1**), nitrosoureas (**2**), and cisplatin (**3**) were linked to the 3- and 17-position of the estradiol skeleton. However, the compounds obtained **13**,[19] **14**,[19] **15**,[20] **16**,[21] **17**[22] and **18**[23] showed only low biological activities against ER+ breast cancer.

Structure activity relationship studies of various estrogenic compounds have shown that in order to obtain the highest ERBA, the estrogenic steroid hormone with an estra-1,3,5(10)-triene-skeleton must have the 3- and 17β-hydroxy groups available, presumably to form hydrogen bonds with the receptor protein at the binding sites.[24] Therefore, it has been suggested that the relatively weak antitumor activities of those agents against breast cancer were partly due to their inability to bind to the ER and thereby to accumulate in ER+ tissues.[25] Simply, the cytotoxic moiety was inappropriately linked to the binding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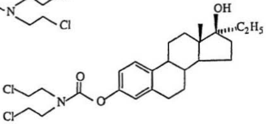
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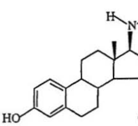
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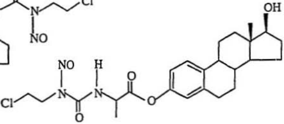
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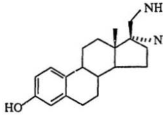
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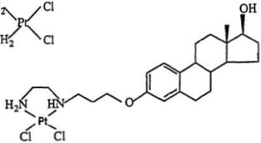
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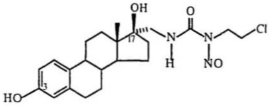

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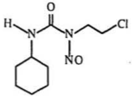


20 Lomustine

sites of estradiol, namely carbon 3 and/or 17.

In order to retain the 3- and 17β-hydroxy groups on the estradiol (E_2) nucleus, a nitrosourea (**2**) moiety was introduced at the 17α-position.[20] As expected, the resulting compound **19** had higher ERBA value than the 17β-derivative **15** (**19**=1.8%, **15**=0.41%, E_2=100%).[26] The order of ERBA also correlates with the order of cytotoxicity against ER^+ breast cancer. Moreover, compound **19** was more active than the mixture of estradiol and lomustine (**20**), the latter being a clinically useful antitumor nitrosourea.[26] Unfortunately, the estrogenic activity induced by this product made it less attractive since it could cause cardiovascular and other toxic side effects.[27]

With the various studies of hormonal influence on breast cancer cell growth, one question has been put forward: "Is an estrogenic molecule such as estradiol a good carrier of antitumor agents ?"

There is considerable evidence to suggest that estrogen has direct and indirect effects on proliferation of ER^+ breast cancer cells (Fig.2).[28,29] Estrogen can bind to the estrogen receptor to unmask the DNA binding domain. This domain can bind to the estrogen response elements (EREs) on the DNA to initiate the transcription of estrogen-sensitive genes and protein synthesis. Estrogen can also increase the production of tumor growth factors alpha (TGFα) that possibly interacts with epidermal growth factor receptors (EGFR) in an autocrine loop. Similarly, estrogen causes a decrease in some members of the TGFβ family which is a tumor suppressor.

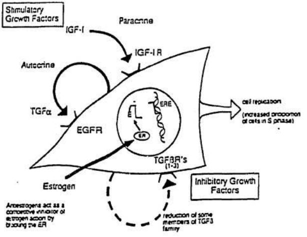


Fig.2 Estrogen effects on breast cancer cell.

Alternatively, antiestrogen has ERBA, but no estrogentic activity. Therefore, antiestrogen can act as a competitive inhibitor of estrogen binding to the ER to prevent estrogen-stimulated changes in cellular biochemistry.[29] Until now, three mechanisms of antiestrogen action were proposed: antiestrogen, A. reduces DNA binding by interfering with receptor dimerization (Fig.3, **A**);[30] B. induces conformational changes of the receptor that allow binding to DNA but do not promote events needed for gene transcription (Fig.3, **B**);[31] C. causes a rapid disappearance of the ER from the target tissue, resulting in an insufficient amount of ER to bind the native ligand and elicit agonistic responses (Fig.3, **C**).[32] Consequently, the blockage of estrogen action with antiestrogen remains a generally accepted method of treatment of ER+ breast cancer.

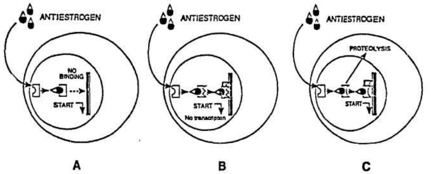
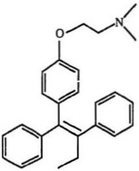


Fig.3 The mechanism of antiestrogen action for the treatment of ER+ breast cancer cell.

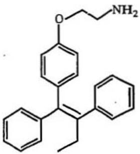
From these points of view, Katzenellenbogen thought that[33] the syntheses of new conjugates containing both antiestrogenic and cytotoxic moiety could be of prime interest. Simply, an antiestrogenic moiety would not only be used as the carrier of cytotoxic agents, but also could produce potential antitumor activity by itself. It may result in a powerful drug for the treatment of ER+ breast cancer.

Therefore, Katzenellenbogen and his co-workers selected tamoxifen (**21**), an antiestrogenic agent which is widely used for the treatment of ER+ breast cancer, as a carrier of toxic moieties to produce new anticancer drugs.[33,36] Alkylating agents such as nitrogen mustard, nitrosourea, nitrosocarbamate were linked on the alkoxy side chain of tamoxifen because the ethyl side chain was found necessary for antiestrogenic activity.[34] Two analogues, the nitrogen mustard-tamoxifen **22** and the nitrosocarbamate-tamoxifen **24** showed dose related cytotoxicity in both ER+ and ER- breast cancer cell lines: MCF-7 and

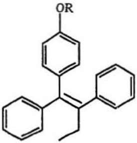
MDA-MB-231, which was not suppressed by estradiol. However, tamoxifen-nitrosourea **23** demonstrated marked cytotoxicity in MCF-7 cells that was blocked by estradiol, whereas its activity in MDA-MB-231 cells was unaffected by estradiol. It seemed that the selective toxic activity of **23** against ER^+ cell line was mediated via the ER. However, recent evidence suggested that the cytotoxicity of **23** resulted from the antiestrogenic effect of its hydrolysed product, bisdesmethyl tamoxifen **25**.[35]



21 Tamoxifen



25 Bisdesmethyl Tamoxifen



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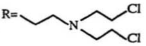

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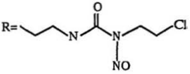

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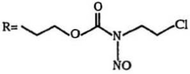
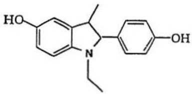


The reason for these analogues of tamoxifen to be devoid of selectivity for ER^+ breast cancer cells was rationalized by their low ERBA. As mentioned by Katzenellenbogen,[36] for a cell to be killed using a receptor uptake process, an adequate dose of drug must be delivered by the receptor system. However, the capacity of the ER uptake system is quite limited. From clinical assays on human breast tumors, it is known that the range of receptor content is 1000-10000 per cell. The calculation based on the number of ER and the possible drug concentration per cell demonstrates that the ERBA value of drugs should be at least 1% of estradiol (E_2=100%).[37] But the ERBA assay of the compounds **22**, **23** and **24** on MCF-7 cell lines showed the ERBA values of only 1.2%, 0.35% and 0.19% respectively.[33,36] From this point of view, tamoxifen might not be a good carrier of antitumor agents for ER^+ breast cancer since derivatives based on a relatively low affinity ligand would also have relatively low affinity (The ERBA value of tamoxifen is only 1.8% of E_2).[36]

More recently, German scientists, Knebel and co-workers, thought that a non-steroidal structure, 5-hydroxy-2-(4'-hydroxyphenyl)-3-methylindole (**26**) might be a suitable derivative to link a cytotoxic agent to because of its structural similarities with zindoxifene (**27**, ERBA=9.5% of E_2),[38] a drug which has been developed as an antiestrogen.[37] Moreover, it also showed high ERBA, i.e., 33% of E_2.[39] They found that the nitrogen atom was the best position for the introduction of bulky substituents into the indole skeleton without much interference with the important binding sites of the molecule (C-5-OH and C-4'-OH).[41] The cytotoxic moiety was linked to the indole skeleton by a spacer group in order to avoid strong steric interaction of the ligand with the ER binding sites. In their model, a cis-(diaminoalkane) dichloroplatinum (II) complex was used as the cytotoxic moiety. The parent compound cisplatin (**3**) is a potent antineoplastic agent against solid tumors, especially testicular cancer,[42] but has low activity against breast cancer.[43] Moreover, cisplatin induces very serious side effects such as nephrotoxicity[44] and ototoxicity[45]. They believed that the affinity of the new platinum complexes for the ER might increase the activity of such an agent on mammary tumors and reduce its toxic side effects. The compounds synthesized (**28a-c**) showed some specific binding affinity for the ER.[46] The relative ERBA values of the compounds **28a**, **28b**, **28c** were 1.0%, 1.3%, 6.5% respectively. The order of their ERBA values correlated with the order of their cytotoxicity.[47] In vitro, only the growth of ER^+ MCF-7 mammary tumor cells was inhibited, whereas ER^- MDA-MB-231 cells did not respond. In vivo, a strong inhibitory effect was observed in ER^+ MXT mammary tumors of the mouse. The complex **28c**, with six carbon atom side chain, reduced the tumor weight by 89% after six weeks of treatment (The dosage was 3x20 mg/Kg body weight/ week.). The effect on ER^- tumors was weaker than on ER^+ tumors.[47] The complexes **29** and **30** also showed inhibitory effect similar to the complex **28c** (the ERBA value of **29** and **30** = 5.2% and 4.4%, E_2=100%).[48,49] Moreover, there was no sign of nephrotoxicity observed in these experiments (usually cisplatin induces a serious nephrotoxic side effect).

These findings were rationalized by the specific binding affinity of those complexes. However, the antitumor activities of those complexes were still slightly less active than cisplatin itself which was used in subtoxic dosage (3x1.4 mg/Kg body weight/week). Therefore, it has been suggested that further investigation of steroidal or non-steroidal derivatives to link anticancer agents should be focused on the development of drugs with further improved antineoplastic activity based on enhanced ERBA. This could be achieved by coupling antineoplastic drugs to hormonal derivatives with relatively strong

ERBA.[50]



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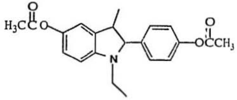
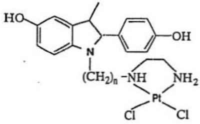
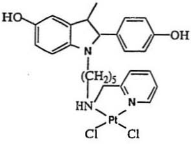


27 Zindoxifene



28

n= 4 (a), 5(b), 6(c)



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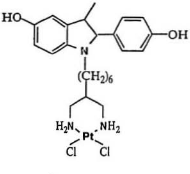
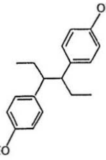


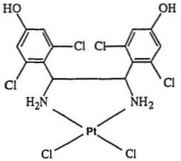
30

In 1987, R. Gust[51] and J. Karl[52] successfully synthesized two new powerful hormonal platinum complexes, namely meso-[1,2-bis(2,6-dichloro-4-hydroxyphenyl) ethylenediamine] dichloroplatinum (II) complex (**32**) and meso-[1,2-bis(2,6-dichloro-4-hydroxyphenyl) ethylenediamine] disulfatoplatinum (II) complex (**33**) for the treatment of ER^+ breast cancer. They selected hexestrol (**31**), a non-steroidal synthetic estrogen as a ligand substrate to link the cytotoxic moiety, platinum (II) complex. In comparative tests on ER^+ and ER^- mammary tumors in cell culture (MCF-7 and MDA-MB-231 cell lines), the complexes **32** and **33** were not obviously selective, inhibiting both ER^+ and ER^- mammary carcinoma. However, In vivo, a strong inhibitory effect of **32** was observed in ER^+ MXT mammary tumors of the mouse. After four weeks of treatment at a dose of 3x6.5 mg/Kg body weight/week, the tumor weight was reduced by 88%, which was significantly more active than cisplatin at the highest tolerable dosage (3x1.5 mg/kg body weight/week). A further increase of efficacy was achieved with the water soluble sulfatoplatinum complex **33**. Moreover, they also displayed inhibitory activity for prostatic cancer.[53]

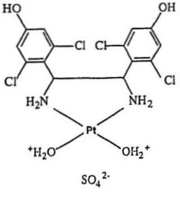
Preliminary biological studies of the complexes **32** and **33** showed that their high antitumor activity for ER^+ breast cancer resulted from their ER mediated enrichment in the nuclei of ER^+ tumor cells. A higher level of Pt in the tumor tissue than in skeletal muscle and blood was found.[52,54] Moreover, both derivatives displayed estrogen-like properties:[55] a) it competed with estradiol for ER binding sites in a competitive manner at 0.1 µM concentration; b) it reduced the number of estradiol binding sites after a 16 hours incubation, and c) it increased the level of progesterone receptor. However, their relatively low ERBA (the ERBA value of **32** and **33** = 0.3%, 0.1% respectively, E_2=100%)[52] is inconsistent with the enrichment theory. It suggests that the 1% ERBA threshold minimum level might not be necessary for selectivity. It is acceptable that the complexes **32** and **33** might bind to an ostrogen-specific nuclear receptor, which also could cause an enrichment in the nucleus giving rise to an increased reaction with DNA.[52] Recent studies suggested that the selective growth inhibitory effects of **32** and **33** also involved immunological factors.[55,56]



Hexestrol **31**



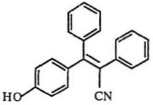
32



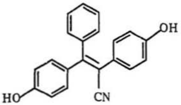
33

Interestingly, the isomers of **32**, namely d,l-[1,2-bis(2,6-dichloro-4-hydroxyphenyl) ethylenediamine] dichloroplatinum (II) complex displayed neither estrogenic activities nor cytotoxic effects for ER+ breast cancer.[52] The exact mechanisms of their actions are still unclear. Numerous scientists are continuing to working in this area.

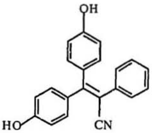
Although the new platinum complexes **32** and **33** display powerful antitumor activity against ER+ breast cancer, their estrogenic properties become a major obstacle in their clinical use.[57] As mentioned before, estrogenic activity can cause cardiovascular side effects. H. Schonenberger and co-workers are trying to modify the structure of the 1,2-diphenyl-ethylenediamine ligand in order to reduce its strong estrogenic potency.[57]



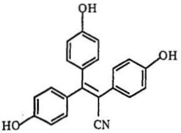
34 **ERBA**= 18



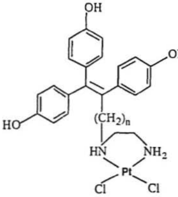
35 **ERBA** = 72



36 **ERBA** = 270

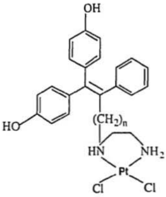


37 **ERBA** = 270



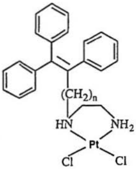
38

n= 6(a), 8(b), 10(c)



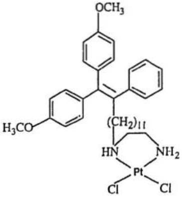
39

n= 6(a), 8(b), 10(c), 11(d)



40

n= 6(a), 8(b), 10(c)



41

In this thesis, we are describing our efforts towards the design of new cisplatinum complexes with good ERBA. According to the enrichment theory, high ERBA is needed to result in a sufficient accumulation of drugs in the ER^+ tumor tissue through the ER mediated transport process.[36,37]

Pons and co-workers investigated the relationships of structure and ERBA of a variety of non-steroidal estrogen derivatives.[58] They found that hydroxylated triphenylacrylonitriles (TPA) had high ERBA, particularly the compounds **36** and **37** (the ERBA value of **36** and **37**=270, E_2=100). These studies suggest that TPA analogues **36** and **37** could be good candidates to link antitumor agents to because according to Von Angerer's findings, derivatives based on relatively high affinity ligand should also possess relatively high affinity.[36]

Therefore, we design a series of new cisplatinum complexes **38a-c** and **39a-d** which are linked to the non-steroidal skeleton, TPA . We hope that these new cisplatinum complexes will have higher ERBA, therefore, higher antitumor activity against ER^+ breast cancer than the platinum complex **28** synthesized by the German scientists.

The cytotoxic moiety, ethylenediamine platinum (II) complex, will be linked on the middle part of TPA skeleton in order to avoid strong steric interference with the important binding sites of the carrier ligand, i.e., the hydroxy groups. Long side chains with six, eight, ten or eleven carbon atoms will be added between the two portions: TPA skeleton and cisplatinum (II) complex. This should allow the two portions to be more flexible to react with the estrogen receptor and DNA respectively (Fig.4). The length and position of the side chain are also based on the structure of the pure steroidal antiestrogens ICI 164,384 and ICI 182,780 (Fig.5) recently described in the literature.[59] Such estradiol with a long alkyl side chain on the 7-α position possesses sufficient ERBA (the ERBA value of ICI 164,384 and ICI 182,780=19%, 89% respectively, E_2=100%)[60] and pure antiestrogenic activity. A pure antiestrogen is devoid of any estrogenic activity, therefore no estrogenic side effects will be induced. This structural analogy should confer upon our new compounds both antiestrogenic and cytotoxic activity.

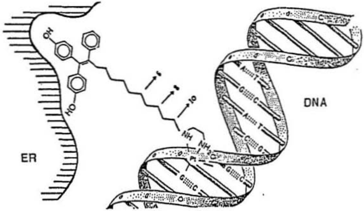
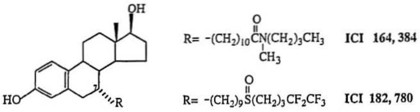


Fig.4 The illustration of the interaction of the non-steroidal Pt (II) complex with the ER and DNA.



R= $-(CH_2)_{10}C(=O)N(CH_3)(CH_2)_3CH_3$ **ICI 164, 384**

R= $-(CH_2)_9S(=O)(CH_2)_3CF_2CF_3$ **ICI 182, 780**

Fig.5 The structures of the new antiestrogens ICI 164,384 and ICI 182,780.

TPA-cisplatinum (II) complexes **40a-c** and **41** without hydroxy groups will also be synthesized. They will be our reference derivatives. Such compounds should have no ERBA and no hormonal activity. Their biological activity will be solely produced by their platinum portion. Therefore, it may help us to understand the possible mechanisms of actions of the compounds **38a-c** and **39a-d** containing hydroxy groups. Moreover, it may also help us to estimate the ERBA values of **38a-c** and **39a-d** more precisely since it is known that platinum complexes might produce non-receptor irreversible binding to proteins in the ER preparation.[61]

In our laboratory, we have already obtained the complexes **38a-c**.[62] In this project, we are going to report the synthesis of the remaining compounds **39a-d**, **40a-c** and **41**, as well as their in vitro biological activities on MCF-7 (ER^+) and MDA-MD-231 (ER^-) human breast cancer cell lines.

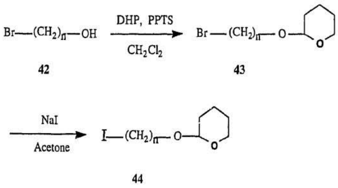
Chapter 2

RESULTS AND DISCUSSION

2.1 Synthesis of Bishydroxy and Bismethoxy Triphenylethylene Platinum (II) Complexes 39a-d and 41.

As shown in Scheme 2, four new platinum (II) complexes **39a-d** were obtained with a 30% overall yield from commercially available benzyl, 4-hydroxyphenyl ketone, after eight steps.

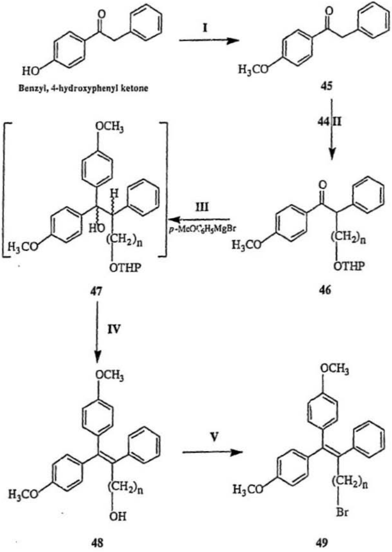
Scheme 1

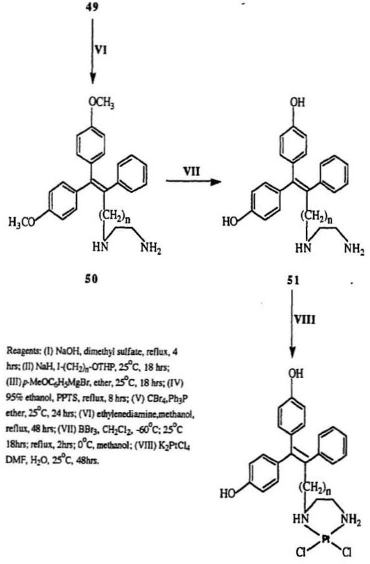


n= 6(a), 8(b), 10(c), 11(d)

Initially the appropriate iodotetrahydropyranyl ethers **44a-d** were prepared. As illustrated in Scheme 1, the alcohols **42a-d** were protected as a tetrahydropyranyl ether to give compounds **43a-d**,[63] which, upon treatment with

Scheme 2


Benzyl, 4-hydroxyphenyl ketone
I
45
44 II
46
III
p-MeOC6H5MgBr
47
IV
48
V
49
HO
H3CO
OCH3
(CH2)n
OTHP
OH
Br



Reagents: (I) NaOH, dimethyl sulfate, reflux, 4 hrs; (II) NaH, I-$(CH_2)_n$-OTHP, 25°C, 18 hrs; (III) p-$MeOC_6H_5MgBr$, ether, 25°C, 18 hrs; (IV) 95% ethanol, PPTS, reflux, 8 hrs; (V) CBr_4,Ph_3P ether, 25°C, 24 hrs; (VI) ethylenediamine,methanol, reflux, 48 hrs; (VII) BBr_3, CH_2Cl_2, -60°C; 25°C 18hrs; reflux, 2hrs; 0°C, methanol; (VIII) K_2PtCl_4 DMF, H_2O, 25°C, 48hrs.

n = 6(a), 8(b), 10(c), 11(d)

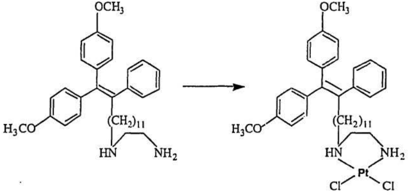
sodium iodide in dry acetone, gave the iodotetrahydropyranyl ethers **44a-d** (95% average yield for the two steps).

Benzyl-4-hydroxyphenyl ketone cannot directly be used as the starting material in alkylating reaction (II, Scheme 2) since the existence of hydroxy group is able to quench the enolation of the ketone.[64] So we protected the hydroxy group as a methyl ether **45** by using dimethylsulfate and sodium hydroxide (I, Scheme 2).[65] The yield for this reaction was around 75% (98% based on the recovered starting material ketone).

Alkylation of **45** with the iodotetrahydropyranyl ethers **44a-d** was achieved using sodium hydride in tetrahydrofuran to give compounds **46a-d** with an average yield of 75% (98% taking consideration **44a-d** recovered (II, Scheme 2). Addition of an excess of *p*-methoxyphenylmagnesium bromide to the ketones **46a-d** (III, Scheme 2)[66] and subsequent treatment of the crude tertiary alcohol intermediates **47a-d** with pyridinium-*p*-toluenesulfonate (PPTS) in ethanol at reflux afforded the triphenylethylene alcohols **48a-d** (IV, Scheme 2) as the result of dehydration of the tertiary alcohols and simultaneous deprotection of the tetrahydropyranyl ethers (85% average yield for the two steps).[63]

With the desired triphenylethylenes **48a-d** in hand, the following sequence of reactions are simple functional group transformations. Initially, alcohols **48a-d** were transformed to the bromides **49a-d** with carbon tetrabromide and triphenylphosphine in dry dimethylether (85% average yield. V, Scheme 2).[67] The amines **50a-d** were obtained with an average yield of 90% by refluxing the bromides **49a-d** in the presence of an excess of ethylenediamine in dry methanol (VI, Scheme 2).[68] Finally, demethylation with boron tribromide gave the intermediate bis-phenols **51a-d** (VII, Scheme 2),[69] which, upon treatment with potassium tetrachloroplatinate (II) in a mixture of dimethylformamide (DMF) and water (VIII, Scheme 2), led to the desired platinum (II) complexes **39a-d** (60% average yield for the two steps).[70]

Scheme 3



50d **41**

Reagents: K_2PtCl_4, DMF, H_2O, 25°C, 48hrs.

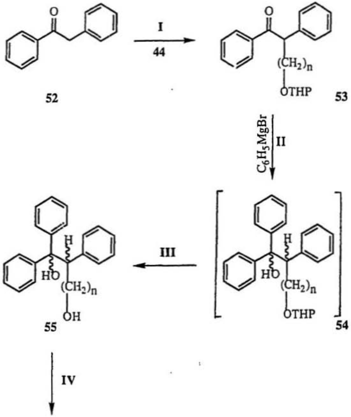
The platinum (II) complex **41** was easily obtained by reacting the amine **50d** with potassium tetrachloroplatinate (II) in a mixture of DMF and water (Scheme 3, 80% yield)).

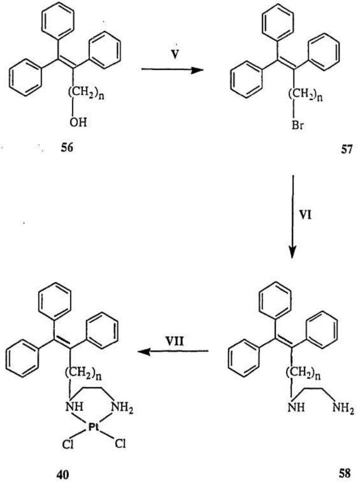
All new compounds obtained were characterized by their IR, ^{1}H NMR, ^{13}C NMR and Mass spectrum. The final products **39a-d**, and **41** passed element analysis (C, H, N).

2.2 Synthesis of Triphenylethylene Platinum (II) Complexes 40a-c.

The triphenylethylene platinum (II) complexes **40a-c** were synthesized from commercially available starting material deoxybenzoin **52** (Scheme 4), in a similar sequence of reactions as used earlier for compounds **39a-d**. The total yield exceeded 40%.

Scheme 4



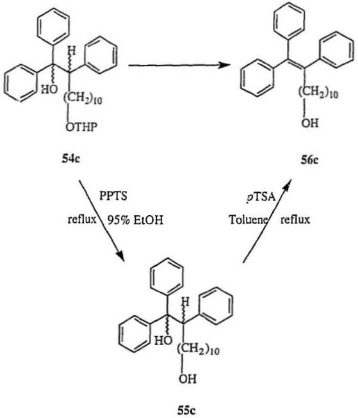


n = 6(a), 8(b), 10(c)

Reagents: (I) NaH, I-$(CH_2)_n$-OTHP, THF, 25°C, 18 hrs; (II) C_6H_5MgBr, diethyl ether 25°C, 6 hrs; (III) 95% ethanol, PPTS, reflux, 3 hrs; (IV) toluene, *p*TSA, reflux, 2 hrs; (V) CBr_4, Ph_3P, diethyl ether, 25 °C, 24 hrs; (VI) ethylenediamine, methanol, reflux, 48 hrs; (VII) K_2PtCl_4, DMF, H_2O, 25°C, 48 hrs.

One reaction which we would like to emphasize here is the dehydration and deprotection of the tertiary alcohols **54a-c** (III, IV, Scheme 4) because it is quite interesting.

Scheme 5



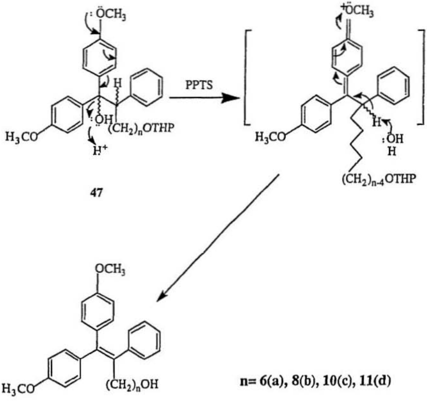
Initially, we followed the same procedure as for the dehydration and deprotection of **47a-d** (IV, Scheme 2). The tertiary alcohol **54c** was allowed to react in the presence of PPTS in 95% ethanol at reflux for 8 hrs (Scheme 5). We expected simultaneous dehydration of tertiary alcohol and deprotection of THP ether to form the alkylalcohol **56c**. Unfortunately it was not the case. The IR spectrum of the product obtained showed a broad absorption at 3600-3300 cm^{-1} suggesting the presence of hydroxy group. If the product was **56c**, a allylic methylene should appear around δ 2.43 in its ^{1}H NMR. Five quaternary carbons should also be observed between δ 150.00 and 135.00 in its ^{13}C NMR. However, there was no signal at δ 2.43 in the ^{1}H NMR of the product obtained. An unexpected double doublet appeared at δ 3.69 (Fig.6, **A**). The ^{13}C NMR showed only three quaternary carbon at δ 146.31, 145.97, and 140.00. Two unexpected signals were observed at δ 80.90 and 54.13 (Fig.7, **A**). Clearly, the product obtained was not **56c**. It was the alcohol **55c**. The signal at δ 80.90 was due to the carbon bearing the hydroxyl and the two phenyl groups, and the one at δ 54,13 due to the carbon to which was attached the ten carbon side chain. Further treatment of **55c** with a stronger acidic catalyst, i.e., *p*-toluenesulphonic acid (*p*TSA) in toluene at reflux (Scheme 5) produced the desired compound **56c**. As expected, its ^{1}H NMR spectrum showed a multiplet at δ 2.43 accounting for the allylic methylene (Fig.6, **B**). Five signals at δ 143.43, 142.93, 142.40, 141.00 and 138.92 were also observed accounting for the five quaternary carbons in its ^{13}C NMR spectrum (Fig.7, **B**).

This interesting result can be explained if we compare the structure of the substances **47a-d** and **54a-c**. An electron donating group is present on compounds **47a-d**, i.e., a methoxy group which can assist the dehydration reaction (Scheme 6). Therefore, the compounds **54a-c** without electron donating group on their aromatic ring need a stronger acidic catalyst *p*TSA and higher reaction temperature to achieve the same reaction.

This result emphasizes that a very subtle change in reaction condition and the chemical structure of the substrate may drastically change the outcome of a chemical reaction.

All new compounds were characterized by IR, ^1H NMR, ^{13}C NMR, MS spectrum. All spectra were consistent with the assigned structures. The final platinum (II) complexes **40a-c** also passed element analysis (C, H, N).

Scheme 6



47

48

n= 6(a), 8(b), 10(c), 11(d)

Fig.6 ^{1}H NMR Spectra of Compounds 55c (A) and 56c (B).

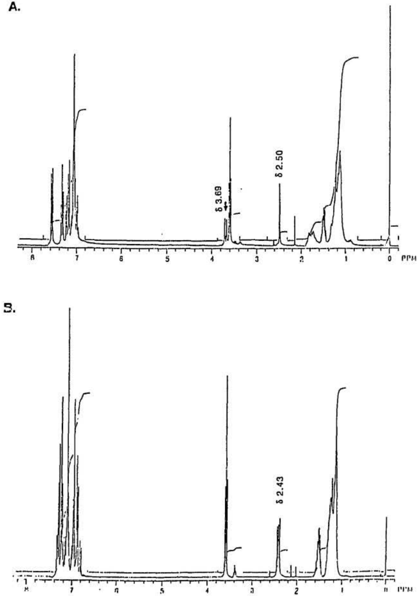
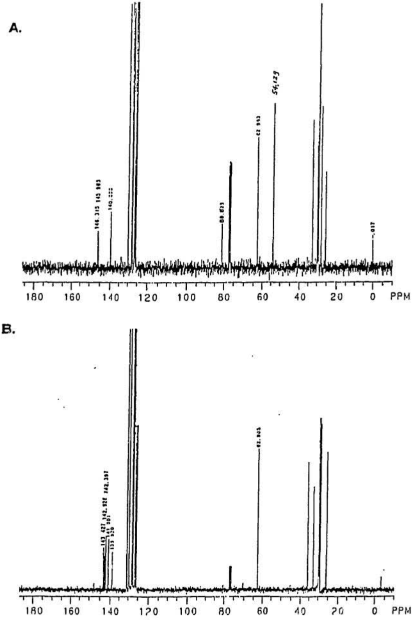
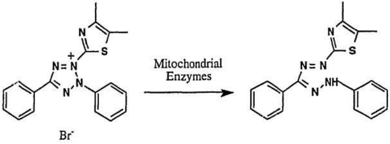


Fig.7 ^{13}C NMR Spectra of Compounds 55c (A) and 56c (B).



2.3 In Vitro Antitumor Activity

Two human breast tumor cell lines were chosen based on their estrogen receptor content, to evaluate the antitumor activities of our new platinum (II) complexes.[71] The cytotoxicity of our compounds was tested along with controls (cisplatin and tamoxifen) on both ER$^+$ (MCF-7) and ER$^-$ (MDA-MD-231) human mammary carcinomas in order to assess the potential selective anti-neoplastic effect on hormone-dependent breast cancer. The antitumor activity was evaluated with a colorimetric assay that uses the ability of viable cells to reduce a colorless tetrazolium salt 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT), into a thiazolyl blue MTT formazan (Fig.8).[72] A recent report indicates that the MTT assay can be used to replace the [^{3}H] -uridine assay for chemosensitivity screening. The colorimetric assay has the advantages of being safer, less costly and simpler than the radiometric assay.[73] The MTT assay is widely used now.[74]



MTT
(colorless)

MTT-FORMAZAN
(thiazolyl blue)

Fig.8 The Reaction Equation of MTT Reduction.

As shown by the MTT assays on two human breast cancer cell lines, our new compounds demonstrated cytotoxicity on both ER$^+$ (MCF-7) and ER$^-$ (MDA-MD-231) cells (Table 1). Clearly, the more lipophilic the compound, the better the cytotoxicity. The compounds **40b-c** without hydroxy groups showed similar cytotoxicity to cisplatin and higher cytotoxicity than tamoxifen, particularly on the MDA-MD-231 (ER$^-$) cell line. This result was and can be explained by the following fashion:[62] a more lipophilic compound could theoretically penetrate the lipophilic cell membranes more easily, therefore concentrate sufficiently in the cell to produce its biological activity.

Table 1. Inhibitory concentration of drug on both ER$^+$ and ER$^-$ breast cancer cell lines.

Drug \ Cell Line	MCF-7 (ER$^+$) IC$_{50}$ (μM)^a	MDA-MD-231 (ER$^-$) IC$_{50}$ (μM)^a
Cisplatin	3.1±0.3	2.4±0.2
Tamoxifen	11±1	28±3
39a	44±4	30±3
39b	34±3	24±2
39c	40±4	26±3
39d	16±2	7.0±0.6
40a	7.0±0.8	4.0±0.4
40b	3.4±0.3	1.5±0.2
40c	4.0±0.4	1.0±0.1
41	14±1	2.4±0.2

a. Concentration inhibiting 50% of cell growth was determined graphically from the cell survival curves (Fig.9, 10). Data represent mean values ± SD for eight wells.

As we expected, platinum (II) complexes with a longer side chain has the tendency to increase the cytotoxic activity. The compound **39d** with eleven carbon atoms side chain was significantly more cytotoxic as compared with compounds **39a-c** containing less carbon atoms. The reason might be as described before: (1) a compound with a longer side chain might allow the platinum (II) complex portion to alkylate DNA more efficiently due to the fewer steric interactions between the triphenylethylene moiety and DNA; (2) the increase of carbon atoms in the side chain could increase the lipophilicity of the compound, therefore might improve its cytotoxic activity.

The complexes **39a-d** with two hydroxy groups showed cytotoxic activities by inhibiting proliferation of the MCF-7 (ER^+) cells, which appears not to be mediated by the ER. This seems to be the case since the proliferation of the MDA-MD-231 (ER^-) cells was inhibited at a lower concentration as it was for the inhibition of MCF-7 (ER^+) cells. However, it is important to indicate that the desired selectivity of the compounds **39a-d** might be expressed more clearly (and possibly only) in vivo as demonstrated previously for the compounds **32** and **33**. The hypothesis of ER mediate selectivity of compounds **39a-d** should be and will be further evaluated in vivo in the future.

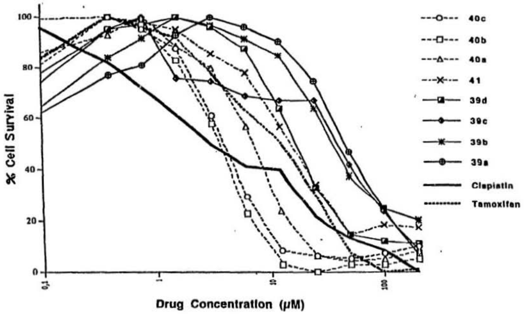


Fig.9 MCF-7 Cell Survival Curves

* Each point represents the mean of eight wells. Error bars have been omitted for clarity.

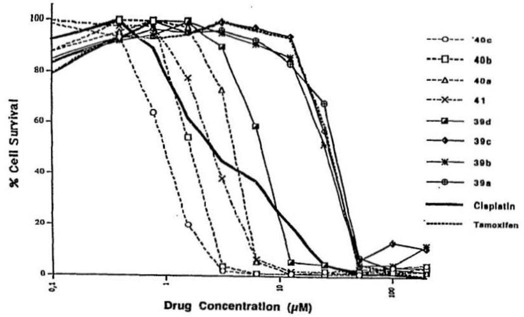


Fig.10 MDA-MB-231 Cell Survival Curves

* Each point represents the mean of eight wells. Error bars have been omitted for clarity.

2.4 Conclusion.

In a conclusion, eight new platinum (II) complexes have been synthesized and tested for their biological activities in vitro. The synthesis for these types of compounds is straightforward and efficient. The lipophilic compounds **40b-c** showed promising antitumor activity for both ER^+ and ER^- human breast cancer cells in vitro.

Chapter 3

EXPERIMENTAL

3.1 Synthesis

3.1.1 General Procedures.

Anhydrous reactions were performed under an inert atmosphere, the set-up assembled and cooled under dry nitrogen. Unless otherwise noted, starting material, reactant and solvents were obtained commercially and were used as such or purified and dried by standard means.[75] Organic solutions were dried over magnesium sulphate ($MgSO_4$), evaporated on a rotatory evaporator and under reduced pressure. All reactions were monitored by UV fluorescence, or staining with iodine or spraying with an aqueous solution of phosphomolybdic acid followed by heating the plate around 135 °C. Commercial TLC plates were Sigma T 6145 (polyester silica gel 60 Å, 0.25mm). Preparative TLC was performed on 1mm silica gel 60 Å, 20x20 plates (Whatman, 4861 840). Flash column chromatography was performed according to the method of Still and co-workers on Merck grade 60 silica gel, 230-400 mesh.[76] All solvents used in chromatography had been distilled. Melting points were recorded on an Electrothermal 9100 apparatus and are uncorrected. The infrared spectra were taken on a Nicolet model 205 FT-IR, or Perkin Elmer model 2000 FT-IR spectrophotometer. Mass spectral assays were obtained using a VG Micromass 7070 HS instrument using an ionisation energy of 70 eV. Nuclear magnetic resonance spectra were obtained in $CDCl_3$ solution, unless otherwise noted, on a General Electric GE 300-NB (300 MHz) instrument: chemical shifts were measured relative to internal standards: tetramethylsilane (TMS, δ 0.0 ppm) for ^1H and $CDCl_3$ (δ 77.0 ppm) for ^{13}C NMR. Multiplicities are described by the following abbreviations: s (singlet), d (doublet), q (quartet), p (pentet), m (multiplet), dd (double doublet), tq (triple quartet), and so on. The NMR assignments were assisted by ^{13}C-^1H correlation (HET-CORR) 2-D spectra.

3.1.2 Conversion of Bromoalcohols to Iodotetrahydropyranyl Ethers.

A. Synthesis of 1-tetrahydropyranyloxy-n-bromoalkane (43).

A solution of bromoalcohol **42** (27.6 mmol), dihydropyran (2.57 g, 30.6 mmol), and pyridinium *p*-toluenesulfonate (PPTS) (10mg, 0.04mmol) in dichloromethane (CH_2Cl_2, 50 mL) was stirred for 5 hrs under nitrogen. Afterwards, sodium bicarbonate ($NaHCO_3$, 500mg) and $MgSO_4$ (5.0 g) were added to the reaction mixture and stirred 15 minutes before being filtered on a short pad of celite / silica gel (1 cm / 4 cm) using CH_2Cl_2 as eluent. The filtrate was evaporated to a viscous oil **43** (98% yield) which was used without further purification in the next step.

1-Tetrahydropyranyloxy-6-bromohexane (43a)
IR, υ_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^1H NMR (δ ppm): 4.57 (1H, t, J=3.2 Hz, -OC<u>H</u>O-), 3.87, 3.74, 3.50, and 3.38 (4H, 4xm, $-C\underline{H_2}OCHOC\underline{H_2}-$), 3.41 (2H, t, J=6.7 Hz, $-C\underline{H_2}Br$), 1.3-2.0 (14H, m, $-C\underline{H_2}-$); MS, m/e: 265 ($M^+ + 1$), 163 (M^+ - OTHP).

1-Tetrahydropyranyloxy-8-bromooctane (43b)

IR, ν_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^{1}H NMR (δ ppm): 4.58 (1H, t, J=3.5 Hz, -OC$\underline{H}$O-), 3.87, 3.73, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.41 (2H, t, J=6.8 Hz, -C$\underline{H}_2$Br), 1.2-2.0 (18H, m, -C$\underline{H}_2$-); MS (m/e): 293(M$^+$ + 1), 191(M$^+$ - OTHP).

1-Tetrahydropyranyloxy-10-bromodecane (43c)

IR, ν_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^{1}H NMR (δ ppm): 4.58 (1H, t, J=3.5 Hz, -OC$\underline{H}$O-), 3.87, 3.73, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.41 (2H, t, J=6.8 Hz, -C$\underline{H}_2$Br), 1.2-2.0 (22H, m, -C$\underline{H}_2$-); MS (m/e): 321 (M$^+$ + 1), 219(M$^+$ - OTHP).

1-Tetrahydropyranyloxy-11-bromoundecane (43d)

IR, ν_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^{1}H NMR (δ ppm): 4.58 (1H, t, J=3.5 Hz, -OC$\underline{H}$O-), 3.87, 3.73, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.41 (2H, t, J=6.8 Hz, -C$\underline{H}_2$Br), 1.2-2.0 (24H, m, -C$\underline{H}_2$-); MS (m/e): 335 (M$^+$ + 1), 233(M$^+$ - OTHP).

B. Synthesis of 1-tetrahydropyranyloxy-n-iodoalkane (44).

Sodium iodide (6.07 g, 40.5 mmol) was added to a solution of the crude bromide **43** (27mmol) in dried acetone. The reaction mixture was stirred at 23°C for 5 hrs. Then, most of the solvent was evaporated and the residue was transferred to an extraction flask with ether (150 mL) and water (100 mL). The organic phase was washed with water (6 X 50 mL), dried, filtrated and concentrated to a viscous liquid. The crude iodide **44** (98% yield) was used as such at the alkylation step.

1-Tetrahydropyranyloxy-6-iodohexane (44a)

IR, υ_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^1H NMR (δ ppm): 4.57 (1H, t, J=3.2 Hz, -OC$\underline{H}$O-), 3.87, 3.74, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.19 (2H, t, J=7.0 Hz, -C$\underline{H}_2$I), 1.3-2.0 (14H, m, -C$\underline{H}_2$-); MS (m/e): 311 (M^+ - H), 211 (M^+ - OTHP).

1-Tetrahydropyranyloxy-8-iodooctane (44b)

IR, υ_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^1H NMR (δ ppm): 4.58 (1H, t, J=3.5 Hz, -OC$\underline{H}$O-), 3.87, 3.73, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.19 (2H, t, J=7.0 Hz, -C$\underline{H}_2$I), 1.2-2.0 (18H, m, -C$\underline{H}_2$-); MS (m/e): 339 (M^+ -H), 239 (M^+ - OTHP).

1-Tetrahydropyranyloxy-10-iododecane (44c)

IR, υ_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^1H NMR (δ ppm): 4.58 (1H, t, J=3.5 Hz, -OC$\underline{H}$O-), 3.87, 3.73, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.19 (2H, t, J=7.0 Hz, -C$\underline{H}_2$I), 1.2-2.0 (22H, m, -C$\underline{H}_2$-); MS (m/e): 367 (M^+ - H), 267 (M^+ - OTHP).

1-Tetrahydropyranyloxy-11-iodoundecane (44d)

IR, υ_{max} (thin film): 1170-1000 (C-O) cm^{-1}; ^1H NMR (δ ppm): 4.58 (1H, t, J=3.5 Hz, -OC$\underline{H}$O-), 3.87, 3.73, 3.50 and 3.38 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.19 (2H, t, J=7.0 Hz, -C$\underline{H}_2$I), 1.2-2.0 (24H, m, -C$\underline{H}_2$-); MS (m/e): 381 (M^+ - H), 281 (M^+ - OTHP).

3.1.3. Conversion of Benzyl-4-Hydroxyphenyl Ketone to Bishydroxy and Bismethoxy Trisphenylethylene Platinum (II) Complexes 39a-d and 41.

A. Synthesis of benzyl-4-methoxyphenyl ketone (45).

Benzyl-4-hydroxyphenyl ketone (2.12 g, 10 mmol) and sodium hydroxide (0.60 g, 15mmol) was dissolved in 250 mL ethanol by heating. The hot solution was added dropwise with dimethyl sulfate (1.51 g, 12 mmol). The reaction mixture was refluxed for 4 hrs. After evaporation, the residue was diluted with ether (200 mL) and washed with water (5x50 mL). The ethereal phase was dried and evaporated to give a white powder which was purified by flash column chromatography (hexane:acetone, 9:1). The yield was 80% average (98% taking in consideration the starting material ketone recovered). mp: 76.2-77.0 °C; IR, υ_{max} (KBr): 3090-3000 (Ar-H), 1680 (C=O), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.98, 6.90 (4H, 2xd apparent, J=8.87 Hz, $\underline{H}$ in para substituted anisyl group), 7.30-7.21 (5H, m, Ar-$\underline{H}$), 4.20 (2H, s, -$C\underline{H}_2$-), 3.80 (3H, s, -$OC\underline{H}_3$); ^{13}C NMR (δ ppm): 196.06, 163.37, 134.83, 130.79(2), 129.44, 129.25(2), 128.48(2), 126.63, 113.65(2), 55.32, 45.11. MS (m/e): 226 (M^+), 135 (M^+ - $CH_2C_6H_5$).

B. Synthesis of 1-(4'-methoxyphenyl)-2-phenyl-n-tetrahydro-pyranyloxy-alkanone (46).

To a stirred suspension of sodium hydride (448 mg, 11.2 mmol, 60% dispension in mineral oil) in 150 mL dry tetrahydrofuran (THF) the ketone **45** (2.30 g, 10.2 mmol) was rapidly added. The reaction mixture was heated (50 °C) with water bath for 1 hr under a nitrogen atmosphere. After cooling, 1-

tetrahydropyranyloxy-n-iodoalkane **44** (11.2 mmol) was added dropwise and the resulting mixture stirred overnight (18 hrs) at room temperature (23 °C). Most of the solvent was then evaporated and the residue was diluted with ether (200 mL) and treated with water (50 mL). The ethereal phase was washed thoroughly with water (6x50 mL), dried and evaporated to give an oil which was purifed by flash column chromatography (hexane:acetone, 95:5). The yield was 75% average (98% taking into account the alkyl iodide **44** recovered).

1-(4'-Methoxyphenyl)-2-phenyl-8-tetrahydropyranyloxy-octanone (46a)

IR, υ_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1680 (C=O), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.95, 6.85 (4H, 2xd apparent, J=8.93 Hz, H in para substituted anisyl group), 7.32-7.14 (5H, m, Ar-H), 4.55 (1H, t, J=3.51 Hz, -OCHO-), 4.49 (1H, t, J=7.25 Hz, -CH-), 3.84, 3.69, 3.48, 3.34 (4H, 4xm, -CH_2OCHOCH_2-), 3.79 (3H, s, -OCH_3), 2.23-1.18 (16H, m, -CH_2-); ^{13}C NMR (δ ppm): 198.44, 163.12, 140.17, 130.82(2), 129.84, 128.69(2), 128.03(2), 126.73, 113.56(2), 98.73, 67.48, 62.24, 55.30, 53.17, 33.97, 30.70, 29.59, 29.40, 27.62, 25.99, 25.42, 19.60; MS (m/e): no M$^+$, 326 (M$^+$ - DHP), 239 (M$^+$ - C_5H_{10}OTHP).

1-(4'-Methoxyphenyl)-2-phenyl-10-tetrahydropyranyloxy-decanone (46b)

IR, υ_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1680 (C=O), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.96, 6.86 (4H, 2xd apparent, J=8.87 Hz, H in para substituted anisyl group), 7.33-7.16 (5H, m, Ar-H), 4.57 (1H, t, J=3.51 Hz, -OCHO-), 4.49 (1H, t, J=7.24 Hz, -CH-), 3.86, 3.71, 3.50, 3.36 (4H, 4xm, -CH_2OCHOCH_2-), 3.82 (3H, s, -OCH_3), 2.16-1.20 (20H, m, -CH_2-); ^{13}C NMR (δ ppm): 198.60, 163.18, 140.23, 130.91(2), 129.91, 128.74(2), 128.09(2), 126.75, 113.62(2), 98.80, 67.64, 62.33, 55.39, 53.24, 34.06, 30.75, 29.71, 29.56, 29.36(2), 27.75, 26.17, 25.48, 19.66. MS (m/e): no M^+, 354 (M^+ - DHP), 239 (M^+ - $C_7H_{14}OTHP$).

1-(4'-Methoxyphenyl)-2-phenyl-12-tetrahydropyranyloxy-dodecanone (46c)

IR, ν_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1680 (C=O), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.96, 6.84 (4H, 2xd apparent, J=8.89 Hz, $\underline{H}$ in para substituted anisyl group), 7.33-7.16 (5H, m, Ar-$\underline{H}$), 4.57 (1H, t, J=3.51 Hz, -OC$\underline{H}$O-), 4.50 (1H, t, J=7.24 Hz, -C$\underline{H}$-), 3.86, 3.72, 3.48, 3.37 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.76 (3H, s, -OC$\underline{H}_3$), 2.16-1.20 (24H, m, -C$\underline{H}_2$-); ^{13}C NMR (δ ppm): 198.36, 163.03, 140.14, 130.74(2), 129.74, 128.59(2), 127.94(2), 126.62, 113.45(2), 98.62, 67.47, 62.12, 55.15, 53.08, 33.95, 30.62, 29.59, 29.47, 29.35(2), 29.27(2), 27.60, 26.08, 25.34, 19.53; MS (m/e): no M^+, 382 (M^+ - DHP), 239 (M^+ - $C_9H_{18}OTHP$).

1-(4'-Methoxyphenyl)-2-phenyl-13-tetrahydropyranyloxy-tridecanone (46d)

IR, ν_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1680 (C=O), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.96, 6.87 (4H, 2xd apparent, J=8.94 Hz, $\underline{H}$ in para substituted anisyl group), 7.32-7.1 (5H, m, Ar-$\underline{H}$), 4.57 (1H, t, J=3.51 Hz, -OC$\underline{H}$O-), 4.49 (1H, t, J=7.25 Hz, -C$\underline{H}$-), 3.86, 3.72, 3.48, 3.37 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 3.81 (3H, s, -OC$\underline{H}_3$), 2.19-1.17 (26H, m, -C$\underline{H}_2$-); ^{13}C NMR (δ ppm): 198.61, 163.18, 140.26, 130.90(2), 129.98, 128.74(2), 128.12(2), 126.78, 113.65(2), 98.82, 67.67, 62.33, 55.39, 53.26, 34.09, 30.79, 29.74, 29.61, 29.55(4), 29.46(2), 27.77, 26.23, 25.51; MS (m/e): no M^+, 396 (M^+ - DHP), 225 (M^+ - $C_{11}H_{22}OTHP$).

C. Synthesis of x-phenyl-y,y-bis(4'-methoxyphenyl)-x-alken-1-ol (48).

A Grignard reagent, *p*-methoxyphenyl magnesium bromide was prepared from magnesium (432 mg, 18.0 mmol) and 4-methoxyphenylbromide (2.81 g, 15.0 mmol) in the presence of a crystal of iodine in 100 mL of dry ether. The Grignard reagent was usually ready after stirring at room temperature (23 °C) overnight (18 hrs), but sometimes required heating at reflux to initiate the reaction. A solution of the ketone **46** (3.0 mmol) in dry ether was treated with the excess of the Grignard reagent for 6 hrs under nitrogen at room temperature (23 °C) and was then hydrolysed with 50 mL of 10% aqueous ammonium chloride. The ether phase was washed with water (5 x 50 mL), dried and evaporated to give the crude tertiary alcohol intermediate **47**. The oily residue refluxed with 95% ethanol in the presence of PPTS (100 mg, 0.40 mmol) for 8 hrs. After evaporation of the solvent, the residue was taken with ether. The ethereal phase was washed with water (5 x 50 mL), dried and evaporated to an oil. Flash column chromatography (hexane:acetone, 7:1) gave a pure **48** in 85% average yield as a viscous oil.

7-Phenyl-8,8-bis(4'-methoxyphenyl)-7-octen-1-ol (48a)

IR, υ_{max} (thin film): 3340 (br, OH), 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.72 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.82 Hz, $\underline{H}$ in parp substituted anisyl group), 3.82, 3.66 (6H, 2xs, 2x-$OC\underline{H}_3$), 3.55 (2H, t, J=6.60 Hz, -$C\underline{H}_2OH$), 2.43 (2H, m, -C=C-$C\underline{H}_2$-), 1.65 (1H, br s, -O$\underline{H}$), 1.50 (2H, p, J=7.30 Hz, -$C\underline{H}_2CH_2OH$), 1.30-1.10 (6H, m, -$(C\underline{H}_2)_3$-); ^{13}C NMR (δ ppm): 158.16, 157.37, 142.87, 139.73, 138.06, 136.25, 135.73, 131.84(2), 130.56(2), 129.54(2), 127.80(2), 125.85, 113.39(2), 112.66(2), 62.91, 55.18, 54.95, 35.85, 32.60, 29.42, 28.82, 25.32; MS (m/e): 416 (M^+), 329 ($M^+ - C_5H_{10}OH$).

9-Phenyl-10,10-bis(4'-methoxyphenyl)-9-decen-1-ol (48b)

IR, υ_{max} (thin film): 3340 (br, OH), 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.88 (2H, d apparent, J=8.73 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.54 (4H, 2xd apparent, J=8.83 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-$OC\underline{H}_3$), 3.60 (2H, t, J=6.63 Hz, -$C\underline{H}_2OH$), 2.43 (2H, m, -$C{=}C{-}C\underline{H}_2$-), 1.64 (1H, br s, -$O\underline{H}$), 1.51 (2H, p, J=7.30, -$C\underline{H}_2CH_2OH$), 1.30-1.10 (10H, m, -$(C\underline{H}_2)_5$-); ^{13}C NMR (δ ppm): 158.12, 157.31, 142.91, 139.85, 137.94, 136.28, 135.77, 131.85 (2), 130.58(2), 129.55(2), 127.78(2), 125.81, 113.37(2), 112.63(2), 63.00, 55.19, 54.95, 35.91, 32.69, 29.58, 29.22, 29.15, 28.85, 25.61; MS (m/e): 444 (M^+), 329 ($M^+ - C_7H_{14}OH$).

11-Phenyl-12,12-bis(4'-methoxyphenyl)-11-dodecen-1-ol (48c)

IR, υ_{max} (thin film): 3340 (br, OH), 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05, (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.72 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.80 Hz, $\underline{H}$ in para substituted anisyl group), 3.81, 3.66 (6H, 2xs, 2x-$OC\underline{H}_3$), 3.61 (2H, t, J=6.62 Hz, -$C\underline{H}_2OH$), 2.42 (2H, m, -$C{=}C{-}C\underline{H}_2$-), 1.66 (1H, br s, -$O\underline{H}$), 1.53 (2H, p, J=7.20 Hz, -$C\underline{H}_2CH_2OH$), 1.38-1.10 (14H, m, -$(C\underline{H}_2)_7$-); ^{13}C NMR (δ ppm): 158.10, 157.29, 142.89, 139.89, 137.88, 136.27, 135.80, 131.84(2), 130.57(2), 129.53(2), 127.75(2), 125.78, 113.34(2), 112.62(2), 62.95, 55.15, 54.92, 35.91, 32.71, 29.65, 29.48, 29.35(2), 29.20, 28.87, 25.68; MS (m/e): 472 (M^+), 329 ($M^+ - C_9H_{18}OH$).

12-Phenyl-13,13-bis(4'-methoxyphenyl)-12-tridecen-1-ol (48d)

IR, υ_{max} (thin film): 3340 (br, OH), 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.09, (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.87 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.54 (4H, 2xd apparent, J=8.82 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-OC$\underline{H}_3$), 3.63 (2H, t, J=6.62 Hz, -C$\underline{H}_2$OH), 2.42 (2H, m, -C=C-C$\underline{H}_2$-), 1.69 (1H, br s, -O$\underline{H}$), 1.53 (2H, m, -C$\underline{H}_2$CH$_2$OH), 1.39-1.10 (16H, m, -(C$\underline{H}_2$)$_8$-); ^{13}C NMR (δ ppm): 158.10, 157.33, 142.92, 139.92, 137.89, 136.30, 135.84, 131.87(2), 130.60(2), 129.55(2), 127.79(2), 125.79, 113.36(2), 112.66(2), 63.06, 55.20, 54.97, 35.94, 32.75, 29.71, 29.49(3), 29.39, 29.25, 28.90, 25.70; MS (m/e): 486 (M$^+$), 329 (M$^+$ - C$_{10}$H$_{20}$OH).

D. Synthesis of 1-bromo-x-phenyl-y,y-bis(4'-methoxyphenyl)-x-alkene (49).

A solution of the alcohol **48** (2.25 mmol), carbon tetrabromide (2.98 g, 9.00 mmol) and triphenylphosphine (2.36 g, 9.00 mmol) in dry ether (100 mL) was stirred at room temperature (23 °C) for 24 hrs under a nitrogen atmosphere. The triphenylphosphine oxide precipitate was filtrated and the resulting solution was washed thoroughly with water (5x25 mL), dried and evaporated to an oil. The crude material was purified by flash column chromatography (hexane:acetone, 95:5) to give the bromide **49** in 85% yield.

1-Bromo-7-phenyl-8,8-bis(4'-methoxyphenyl)-7-octene (49a)

IR, υ_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.88 (2H, d apparent, J=8.73 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.81 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.66 (6H, 2xs, 2x-OC$\underline{H}_3$), 3.32 (2H, t, J=6.86 Hz, -C$\underline{H}_2$Br), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.74 (2H, p, J=7.30 Hz, -C$\underline{H}_2$CH$_2$Br), 1.40-1.18 (6H, m, -(C$\underline{H}_2$)$_3$-); ^{13}C NMR (δ ppm): 158.23, 157.40, 142.81, 139.59, 138.20, 136.20, 135.69, 131.84(2), 130.54(2), 129.54(2), 127.84(2), 125.89, 113.43(2), 112.67(2), 55.20, 54.96, 35.77, 33.91, 32.62, 28.75, 28.65, 27.80; MS (m/e): 478 (M$^+$), 480 (M$^+$ + 2), 329 (M$^+$ - $C_5H_{10}Br$).

1-Bromo-9-phenyl-10,10-bis(4'-methoxyphenyl)-9-decene (49b)

IR, υ_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.88 (2H, d apparent, J=8.59 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.54 (4H, 2xd apparent, J=8.69 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-OC$\underline{H}_3$), 3.37 (2H, t, J=6.86 Hz, -C$\underline{H}_2$Br), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.80 (2H, p, J=7.33 Hz, -C$\underline{H}_2$CH$_2$Br), 1.42-1.10 (10H, m, -(C$\underline{H}_2$)$_5$-); ^{13}C NMR (δ ppm): 158.14, 157.33, 142.89, 139.78, 137.99, 136.25, 135.77, 131.86(2), 130.57(2), 129.55(2), 127.80(2), 125.83, 113.37(2), 112.65(2), 55.19, 54.94, 35.89, 34.03, 32.74, 29.52, 29.00, 28.80, 28.56, 28.05. MS (m/e): 506 (M$^+$), 508 (M$^+$ + 2), 329 (M$^+$ - $C_7H_{14}Br$).

1-Bromo-11-phenyl-12,12-bis(4'-methoxyphenyl)-11-dodecene (49c)

IR, υ_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.77 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.84 Hz, $\underline{H}$ in para substituted anisyl group), 3.80, 3.64 (6H, 2xs, 2x-OC$\underline{H}_3$), 3.37 (2H, t, J=6.87 Hz, -C$\underline{H}_2$Br), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.82 (2H, P, J=7.32 Hz, -C$\underline{H}_2$CH$_2$Br), 1.40-1.10 (14H, m, -(C$\underline{H}_2$)$_7$-); ^{13}C NMR (δ ppm): 158.10, 157.30, 142.86, 139.80, 137.92, 136.21, 135.73, 131.82(2), 130.54(2), 129.51(2), 127.74(2), 125.77, 113.31(2), 112.60(2), 55.11, 54.88, 35.89, 33.97, 32.74, 29.61, 29.30(2), 29.15, 28.83, 28.66, 28.09; MS (m/e): 534 (M^+), 536 ($M^+ + 2$), 329 ($M^+ - C_9H_{18}Br$).

1-Bromo-12-phenyl-13,13-bis(4'-methoxyphenyl)-12-tridecene (49d)

IR, υ_{max} (thin film): 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.18-7.08 (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.71 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.54 (4H, 2xd apparent, J=8.77 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-$OC\underline{H}_3$), 3.39 (2H, t, J=6.86 Hz, -$C\underline{H}_2Br$), 2.42 (2H, m, -C=C-$C\underline{H}_2$-), 1.84 (2H, P, J=7.34 Hz, -$C\underline{H}_2CH_2Br$), 1.40-1.10 (16H, m, -$(C\underline{H}_2)_8$-); ^{13}C NMR (δ ppm): 158.14, 157.33, 142.94, 139.88, 137.92, 136.26, 135.81, 131.87(2), 130.60(2), 129.56(2), 127.75(2), 125.80, 113.36(2), 112.66(2), 55.17, 54.98, 35.91, 34.02, 32.79, 29.68, 29.44(2), 29.38, 29.21, 28.90, 28.71, 28.13; MS (m/e): 548 (M^+), 550 ($M^+ + 2$), 329 ($M^+ - C_{10}H_{20}Br$).

E. Synthesis of 1-[(2'-aminoethyl)amino]-x-phenyl-y,y-bis(4'-methoxyphenyl) -x-alkene (50).

Under a nitrogen atmosphere, ethylenediamine (900 mg, 15.0 mmol) was added to a solution of the bromide **49** (1.50 mmol) in 80 mL of dry methanol. After boiling for two days under reflux (sometimes longer reaction period was required), the solvent was evaporated. The resulting residue was dissolved in ether (150 mL) and washed with a solution of $NaHCO_3$ (30 mL, 5% aqueous) and with water (5x30 mL). The ethereal phase was dried and evaporated to a viscous oil **50**. The yield was 90%.

1-[(2'-Aminoethyl)amino]-7-phenyl-8,8-bis(4'-methoxyphenyl)-7-octene (50a)

IR, υ_{max} (thin film): 3340 (br, N-H), 3090-3000 (Ar-H), 2930-2860 (C-H), 1660 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.65 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.75 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-$OC\underline{H}_3$), 2.79, 2.64 (4H, 2xt, J=5.92 Hz, -$NHC\underline{H}_2C\underline{H}_2NH_2$), 2.54 (2H, t, J=7.25 Hz, -$C\underline{H}_2NH$-), 2.43 (2H, m, -C=C-$C\underline{H}_2$-) 1.78 (3H, br s, -N$\underline{H}$- and -$N\underline{H}_2$), 1.43-1.18 (8H, m, -$(C\underline{H}_2)_4$-); ^{13}C NMR (δ ppm): 158.14, 157.33, 142.86, 139.76, 137.99, 136.23, 135.73, 131.84(2), 130.55(2), 129.53(2), 127.78(2), 125.81, 113.37(2), 112.63(2), 55.16, 54.93, 52.42, 49.74, 41.58, 35.88, 29.90, 29.60, 28.86, 26.99; MS (m/e): 458 (M^+), 428 (M^+ - CH_2NH_2), 415 (M^+ - CH_2CH_2NH), 329 (M^+ -$C_5H_{10}NHCH_2CH_2NH_2$).

1-[(2'-Aminoethyl)amino]-9-phenyl-10,10-bis(4'-methoxyphenyl)-9-decene (50b)

IR, υ_{max} (thin film): 3340 (br, N-H), 3090-3000 (Ar-H), 2930-2860 (C-H), 1660 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.88 (2H, d apparent, J=8.70 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.54 (4H, 2xd apparent, J=8.83 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-$OC\underline{H}_3$), 2.80, 2.65 (4H, 2xt, J=5.85 Hz, -$NHC\underline{H}_2C\underline{H}_2NH_2$), 2.57 (2H, t, J=7.24 Hz, -$C\underline{H}_2NH$-), 2.42 (2H, m, -C=C-$C\underline{H}_2$-), 1.50 (3H, br s, -N$\underline{H}$- and -$N\underline{H}_2$), 1.46-1.10 (12H, m, -$(C\underline{H}_2)_6$-); ^{13}C NMR (δ ppm): 158.10, 157.33, 142.90, 139.85, 137.95, 136.26, 135.80, 131.87(2), 130.57(2), 129.54(2), 127.77(2), 125.79, 113.34(2), 112.62(2), 55.17, 54.94, 52.61, 49.53, 41.76, 35.92, 30.11, 29.64, 29.41, 29.21, 28.88, 27.27; MS (m/e): 486 (M^+), 456 (M^+ - CH_2NH_2), 443 (M^+ - CH_2CH_2NH), 329 (M^+ - $C_7H_{14}NHCH_2CH_2NH_2$).

1-[(2'-Aminoethyl)amino]-11-phenyl-12,12-bis(4'-methoxyphenyl)-11-dodecene (50c)

IR, υ_{max} (thin film): 3340 (br, N-H), 3090-3000 (Ar-H), 2930-2860 (C-H), 1660 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.18-7.05 (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.67 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.81 Hz, $\underline{H}$ in para substituted anisyl group), 3.81, 3.66 (6H, 2xs, 2x-$OC\underline{H}_3$), 2.83, 2.69 (4H, 2xt, J=5.90, -$NHC\underline{H}_2C\underline{H}_2NH_2$), 2.60 (2H, t, J=7.27 Hz, -$C\underline{H}_2NH$-), 2.53 (3H, br s, -$N\underline{H}$- and -$N\underline{H}_2$), 2.42 (2H, m, -$C=C-C\underline{H}_2$-), 1.48-1.10 (16H, m, -$(C\underline{H}_2)_8$-); ^{13}C NMR (δ ppm): 158.06, 157.26, 142.86, 139.82, 137.84, 136.20, 135.73, 131.80(2), 130.52(2), 129.49(2), 127.71(2), 125.74, 113.28(2), 112.57(2), 55.09, 54.87, 51.98, 49.69, 41.27, 35.88, 29.80, 29.64, 29.44(3), 29.21, 28.85, 27.25. MS (m/e): 514 (M^+), 484 (M^+ - CH_2NH_2), 471 (M^+ -CH_2CH_2NH), 329 (M^+ - $C_9H_{18}NHCH_2CH_2NH_2$).

1-[(2'-Aminoethyl)amino]-12-phenyl-13,13-bis(4'-methoxyphenyl)-12-tridecene (50d)

IR, υ_{max} (thin film): 3340 (br, N-H), 3090-3000 (Ar-H), 2930-2860 (C-H), 1660 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.16-7.08 (7H, m, Ar-$\underline{H}$), 6.87 (2H, d apparent, J=8.62 Hz, $\underline{H}$ in para substituted anisyl group), 6.77, 6.53 (4H, 2xd apparent, J=8.73 Hz, $\underline{H}$ in para substituted anisyl group), 3.82, 3.67 (6H, 2xs, 2x-$OC\underline{H}_3$), 2.81, 2.66 (4H, 2xt, J=5.87, -$NHC\underline{H}_2C\underline{H}_2NH_2$), 2.60 (2H, t, J=7.23 Hz, -$C\underline{H}_2NH$-), 2.42 (2H, m, -$C=C-C\underline{H}_2$-), 1.51 (3H, br s, -$N\underline{H}$- and -$N\underline{H}_2$), 1.48-1.10 (18H, m, -$(C\underline{H}_2)_9$-); ^{13}C NMR (δ ppm): 158.06, 157.29, 142.89, 139.85, 137.84, 136.24, 135.77, 131.84(2), 130.57(2), 129.53(2), 127.72(2), 125.75, 113.31(2), 112.59(2), 55.13, 54.91, 52.51, 49.89, 41.68, 35.90, 30.10, 29.66, 29.52(4), 29.23, 28.87, 27.32. MS (m/e): 528 (M^+), 498 (M^+ - CH_2NH_2), 485 (M^+ -CH_2CH_2NH), 329 (M^+ - $C_{10}H_{20}NHCH_2CH_2NH_2$).

F. Synthesis of 1-[(2'-aminoethyl)amino]-x-phenyl-y,y-bis(4'-hydroxyphenyl) -x-alkene (51).

A solution of **50** (0.665 mmol), in dry CH_2Cl_2 (30 mL) was treated with a solution of boron tribromide (1M in CH_2Cl_2, 1.60 mL, 1.60 mmol) at -60 °C, under nitrogen atmosphere. After the addition, the reaction mixture was allowed to warm at a room temperature (23 °C) and was stirred for 18 hrs. The mixture was refluxed during 2 hrs, then cooled down with an ice bath before adding 10 mL methanol. The resulting solution was adjusted with saturated $NaHCO_3$ solution to pH=7, then evaporated to 2-3 mL, treated with saturated $NaHCO_3$ solution (30 mL), and extracted with ethyl acetate (5x30 mL). The crude yield is around 60-80%. The product **51** was used without further purification in the next step. We obtained the following three compounds:

1-[(2'-Aminoethyl)amino]-7-phenyl-8,8-bis(4'-hydroxyphenyl)-7-octene (51a)

1-[(2'-Aminoethyl)amino]-9-phenyl-10,10-bis(4'-hydroxyphenyl)-9-decene (51b)

1-[(2'-Aminoethyl)amino]-11-phenyl-12,12-bis(4'-hydroxyphenyl)-11-dodecene (51c)

1-[(2'-Aminoethyl)amino]-12-phenyl-13,13-bis(4'-hydroxyphenyl)-12-tridecene (51d)

G. Synthesis of 1-{cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-x-phenyl-y,y-bis(4'-hydroxyphenyl)-x-alkene (39).

A solution of potassium tetrachloroplatinate (II) (219 mg, 0.528 mmol) in

7.5 mL of a mixture of DMF and water (2:1) was added to a warm (35 °C) solution of the diamine **51** (0.528 mmol) in 5 mL of DMF. The resulting mixture (pH=9-10) was stirred in the dark for 2-3 days until the pH value reached 4-5. Then, one drop of N,N-dimethylsulfoxide was added and the stirring was continued for 2 hrs. The solvent was evaporated and the residue was suspended in saturated potassium chloride solution (30 mL). A vigourous stirring was essential in order to pulverized the lumps of platinum (II) complex **39**. The resulting suspension was filtered, washed with water (100-250 mL), and dried in a desiccator. The product can be further purified either by flash column chromatography or by preparative TLC (CH_2Cl_2:CH_3OH, 95:5). The crude yield was around 80%.

1-{Cis-[(2'-aminoethyl) amino] dichloroplatinum (II)}-7-phenyl-8,8-bis(4'-hydroxyphenyl)-7-octene (39a)

mp > 138 °C (dec.); IR, υ_{max} (KBr): 3400-3100 (O-H, N-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (acetone-d_6, δ ppm): 8.32, 8.09 (2H, 2xbr s, 2xAr-O$\underline{H}$), 7.20-7.10 (5H, m, Ar-$\underline{H}$), 7.07, 6.86 (4H, 2xd apparent, J=8.52 Hz, $\underline{H}$ in para substituted phenol), 6.71, 6.48 (4H, 2xd apparent, J=8.62 Hz, $\underline{H}$ in para substituted phenol), 5.68, 5.11, 4.98 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.21, 3.06, 2.77, 2.67 (6H, 4xbr s, -C$\underline{H}_2$NHC$\underline{H}_2$C$\underline{H}_2$NH$_2$), 2.46 (2H, m, -C=C-C$\underline{H}_2$-), 1.78, 1.56 (2H, 2xbr s, -C$\underline{H}_2$CH$_2$NH-), 1.40-1.10 (6H, m, -(C$\underline{H}_2$)$_3$-); ^{13}C NMR (acetone-d_6, δ ppm): 156.88, 156.07, 143.94, 139.72(2), 135.93, 135.62, 132.62(2), 131.30(2), 130.41(2), 128.07(2), 126.55, 115.73(2), 114.93(2), 56.12, 53.42, 47.78, 36.33, 27.64, 26.82 (N.B. 2 carbons are hidden by acetone). Anal. calcd. for $C_{28}H_{34}Cl_2N_2O_2Pt\cdot 11/5H_2O$: C 45.68, H 5.26, N 3.80; found: C 45.72, H 5.28, N 3.70.

1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-9-phenyl-10,10-bis(4'-hydroxyphenyl)-9-decene (39b)

mp > 138 °C (dec.); IR, υ_{max} (KBr): 3400-3100 (O-H, N-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (acetone-d_6, δ ppm): 8.35, 8.13 (2H, 2xbr s, 2xAr-O$\underline{H}$), 7.20-7.10 (5H, m, Ar-$\underline{H}$), 7.07, 6.86 (4H, 2xd apparent, J=8.54 Hz, $\underline{H}$ in para substituted phenol), 6.71, 6.48 (4H, 2xd apperant, J=8.62 Hz, $\underline{H}$ in para substituted phenol), 5.71, 5.10, 4.99 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.24, 3.05, 2.80, 2.72 (6H, 4xbr s, -C$\underline{H}_2$NHC$\underline{H}_2$C$\underline{H}_2$NH$_2$), 2.45 (2H, m, -C=C-C$\underline{H}_2$-), 1.80, 1.58 (2H, 2xbr s, -C$\underline{H}_2$CH$_2$NH-) 1.40-1.10 (10H, m, -(C$\underline{H}_2$)$_5$-); ^{13}C NMR (acetone-d_6, δ ppm): 156.89, 156.08, 144.04, 139.86, 139.66, 135.95, 135.61, 132.59(2), 131.29(2), 130.39(2), 128.56(2), 126.55, 115.68(2), 114.94(2), 56.15, 53.51, 47.83, 36.46, 27.82, 27.21 (N.B. 4 carbons are hidden by acetone). Anal. calcd. for $C_{30}H_{38}Cl_2N_2O_2Pt{\cdot}2H_2O$: C 47.37, H 5.57, N 3.68; found: C 47.40, H 5.62, N 3.73.

1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-11-phenyl-12,12-bis(4'-hydroxyphenyl)-11-dodecene (39c)

mp > 138 °C (dec.); IR, υ_{max} (KBr): 3400-3100 (O-H, N-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (acetone-d_6, δ ppm): 8.31, 8.09 (2H, 2xbr s, 2xAr-O$\underline{H}$), 7.20-7.10 (5H, m, Ar-$\underline{H}$), 7.07, 6.85 (4H, 2xd apparent, J=8.50 Hz, $\underline{H}$ in para substituted phenol), 6.71, 6.48 (4H, 2xd apparent, J=8.62 Hz, $\underline{H}$ in para substituted phenol), 5.70, 5.08, 4.98 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.25, 3.08, 2.78, 2.70 (6H, 4xbr s, -C$\underline{H}_2$NHC$\underline{H}_2$C$\underline{H}_2$NH$_2$), 2.45 (2H, m, -C=C-C$\underline{H}_2$-), 1.82, 1.60 (2H, 2xbr s -C$\underline{H}_2$CH$_2$NH-), 1.40-1.10 (14H, m, -(C$\underline{H}_2$)$_7$-); ^{13}C NMR (acetone-d_6, δ ppm): 156.80, 155.95, 143.94, 139.75, 139.55, 135.85, 135.50, 132.47(2), 131.17(2), 130.27(2), 128.43(2), 126.44, 115.54(2), 114.82(2), 56.04, 53.38, 47.71, 36.38, 27.72, 27.14 (N.B. 6 carbons are hidden by acetone). Anal. calcd. for $C_{32}H_{42}Cl_2N_2O_2Pt\cdot 2H_2O$: C 48.73, H 5.88, N 3.55; found: C 48.65, H 5.78, N 3.61.

1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-12-phenyl-13,13-bis(4'-hydroxyphenyl)-12-tridecene (39d)

mp > 138 °C (dec.); IR, υ_{max} (KBr): 3400-3100 (O-H, N-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^1H NMR (acetone-d_6, δ ppm): 8.31, 8.08 (2H, 2xbr s, 2xAr-O$\underline{H}$), 7.20-7.10 (5H, m, Ar-$\underline{H}$), 7.07, 6.85 (4H, 2xd apparent, J=8.47 Hz, $\underline{H}$ in para substituted phenol), 6.71, 6.49 (4H, 2xd apparent, J=8.54 Hz, $\underline{H}$ in para substituted phenol), 5.70, 5.09, 4.97 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.28, 3.08, 2.75 (6H, 3xbr s, $-C\underline{H}_2NHC\underline{H}_2C\underline{H}_2NH_2$), 2.45 (2H, m, $-C{=}C{-}C\underline{H}_2-$), 1.84, 1.62 (2H, 2xbr s $-C\underline{H}_2CH_2NH-$), 1.40-1.10 (16H, m, $-(C\underline{H}_2)_8-$); ^{13}C NMR (acetone-d_6, δ ppm): 157.46, 156.63, 144.61, 140.43, 140.20, 136.51, 136.17, 133.14(2), 131.84(2), 130.94(2), 129.10(2), 127.13, 116.20(2), 115.49(2), 56.71, 54.04, 48.37, 37.06, 28.39, 27.82 (N.B. 7 carbons are hidden by acetone). Anal. calcd. for $C_{33}H_{44}Cl_2N_2O_2Pt\cdot 2H_2O$: C 49.38, H 6.03, N 3.49; found: C 49.32, H 6.08, N 3.52.

H. Synthesis of 1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-12-phenyl-13,13-bis(4'-methoxyphenyl)-12-tridecene (41).

The Platinum (II) complex **41** was obtained following the procedure of **39** taking **50d** as the starting material. The crude yield was around 80%. The product can be further purified either by flash column chromatography or by preparative TLC (CH_2Cl_2:CH_3OH, 98:2). mp > 173 °C (dec.); IR, υ_{max} (KBr): 3340 (br, N-H), 3090-3000 (Ar-H), 2930-2860 (C-H), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.16-7.08 (7H, m, Ar-H), 6.86 (2H, d apparent, J=8.62 Hz, H in para substituted anisyl group), 6.76, 6.53 (4H, 2xd apparent, J=8.73 Hz, H in para substituted anisyl group), 5.65, 5.05, 4.92 (3H, 3xbr s, -NH- and -NH_2), 3.80, 3.66 (6H, 2xs, 2x-OCH_3), 4.05, 3.18, 2.95, 2.75 (6H, 4xbr s, -$CH_2NHCH_2CH_2NH_2$), 2.43 (2H, m, -C=C-CH_2-), 1.75, 1.52 (2H, 2xbr s, -CH_2CH_2NH-), 1.40-1.06 (16H, m, -$(CH_2)_8$-); ^{13}C NMR (δ ppm): 158.15, 157.34, 142.93, 139.90, 137.93, 136.28, 135.81, 131.88(2), 130.61(2), 129.58(2), 127.80(2), 125.83, 113.40(2), 112.67(2), 55.45, 55.22, 54.98, 53.47, 46.93, 35.98, 29.78, 29.55(2), 29.45, 29.34, 29.24, 28.97, 27.41, 26.56. Anal. calcd. for $C_{35}H_{48}Cl_2N_2O_2Pt$: C 52.89, H 6.09, N 3.52; found: C 52.83, H 6.06, N 3.51.

3.1.4 Conversion of Desoxybenzoin to Triphenylethylene Platinum (II) Complexes 40a-c.

A. Synthesis of 1,2-bisphenyl-n-tetrahydropyranyloxy-alkanone (53).

Desoxybenzoin **52** (2.00 g, 10.2 mmol) was rapidly added to a stirred suspension of sodium hydride (448 mg, 11.2 mmol, 60% dispension in mineral oil) in 150 mL dry tetrahydrofuran (THF). The reaction mixture was heated (50 °C) with water bath for 1 hr under a nitrogen atmosphere. After cooling, 1-tetrahydropyranyloxy-n-iodoalkane **44** (11.2 mmol) was added dropwise and the resulting mixture stirred overnight (18 hrs) at room temperature (23 °C). Most of the solvent was then evaporated and the residue was diluted with ether (200 mL) and treated with water (50 mL). The ethereal phase was washed thoroughly with water (6x50 mL), dried and evaporated to give an oil which was purified by flash column chromatography (hexane:acetone, 98:2). The yield was 75% average (98% taking into account the alkyl iodide **44** recovered).

1,2-Bisphenyl-8-tetrahydropyranyloxy-octanone (53a)

IR, υ_{max} (thin film): 3092-3025 (Ar-H), 2933-2858 (C-H), 1683 (C=O), 1585 (C=C) cm^{-1}; ^1H NMR (δ ppm): 8.01-7.15 (10H, m, Ar-$\underline{H}$), 4.55 (1H, t, J=3.55 Hz, -OC$\underline{H}$O-), 4.53 (1H, t, J=7.21 Hz, -C$\underline{H}$-), 3.84, 3.70, 3.47, 3.34 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 2.19-1.22 (16H, m, -C$\underline{H}_2$-); ^{13}C NMR (δ ppm): 199.94, 139.68, 136.83, 132.68, 128.75 (2), 128.53 (2), 128.39 (2), 128.09 (2), 126.83, 98.72, 67.45, 62.25, 53.54, 33.91, 30.68, 29.56, 29.36, 27.55, 25.99, 25.40, 19.62; MS (m/e): 380 (M^+), 296 (M^+ - DHP), 105 (C_6H_5-C=O).

1,2 Bisphenyl-10-tetrahydropyranyloxy-decanone (53b)

IR, υ_{max} (thin film): 3092-3025 (Ar-H), 2933-2858 (C-H), 1683 (C=O), 1585 (C=C) cm^{-1}; ^1H NMR (δ ppm): 8.01-7.15 (10H, m, Ar-$\underline{H}$), 4.56 (1H, t, J=3.53 Hz, -OC$\underline{H}$O-), 4.54 (1H, t, J=7.29 Hz, -C$\underline{H}$-), 3.84, 3.71, 3.47 ,3.35 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 2.16-1.26 (20H, m, -C$\underline{H}_2$-); ^{13}C NMR (δ ppm): 199.93, 139.70, 136.84, 132.65, 128.71 (2), 128.50 (2), 128.35 (2), 128.07 (2), 126.79, 98.67, 67.51, 62.18, 53.53, 33.96, 30.68, 29.61, 29.45, 29.26 (2), 27.60, 26.09, 25.40, 19.60; MS (m/e): 408 (M^+), 324 (M^+ - DHP), 105 (C_6H_5-C=O).

1,2-Bisphenyl-12-tetrahydropyranyloxy-dodecanone (53c)

mp: 59-59.5 °C. IR, υ_{max} (KBr): 3092-3025 (Ar-H), 2933-2858 (C-H), 1683 (C=O), 1585 (C=C) cm^{-1}; ^1H NMR (δ ppm): 8.01-7.15 (10H, m, Ar-$\underline{H}$), 4.57 (1H, t, J=3.50 Hz, -OC$\underline{H}$O-), 4.53 (1H, t, J=7.22 Hz, -C$\underline{H}$-), 3.85, 3.72, 3.49, 3.37 (4H, 4xm, -C$\underline{H}_2$OCHOC$\underline{H}_2$-), 2.15-1.23 (24H, m, -C$\underline{H}_2$-); ^{13}C NMR (δ ppm): 200.12, 139.82, 137.00, 132.74, 128.81(2), 128.61(2), 128.46(2), 128.20(2), 126.88, 98.84, 67.69, 62.34, 53.67, 34.06, 30.80, 29.76, 29.59, 29.51(2), 29.45(2), 27.72, 26.23, 25.51, 19.71; MS (m/e): 436 (M^+), 352 (M^+ - DHP), 105 (C_6H_5-C=O).

B. Synthesis of 1,1,2-trisphenyl-alkan-1,n-diol (55).

A Grignard reagent was prepared from magnesium (432mg, 18.0 mmol) and bromobenzene (2.36 g, 15.0 mmol) in the presence of a crystal of iodine in 100 mL of dry ether. The preparation of Grignard reagent required 4 hrs at room temperature (23 °C). A solution of ketone **53** (3.0 mmol) in dry ether was treated with the excess of the Grignard reagent for 6 hrs under nitrogen at room temperature (23 °C) and was then hydrolysed with 50 mL of 10% aqueous ammonium chloride. The ether phase was washed with water (5 x 50 mL), dried and evaporated to give the crude tertiary alcohol intermediate **54**. The oily residue refluxed with 95% ethanol in the presence of PPTS (100 mg, 0.40 mmol) for 3 hrs. After evaporation of the solvent, the residue was taken with ether. The ethereal phase was washed with water (5 x 50 mL), dried and evaporated to an oil. The crude **55** was used as such for next dehydration step. Flash column chromatography (hexane:acetone, 7:1) could produce a pure **55** (85% yield from **53**).

1,1,2-Trisphenyl-octan-1,8-diol (55a)

IR, υ_{max} (thin film): 3600-3300 (OH), 3090-3015 (Ar-H), 2950-2850 (C-H), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.59-6.96 (15H, m, Ar-$\underline{H}$), 3.69 (1H, dd, J=11.42 Hz, J'=2.86 Hz, -C$\underline{H}$-), 3.50 (2H, t, J=6.62 Hz, -C$\underline{H}_2$OH), 2.57 (1H, br s, CO$\underline{H}$), 1.90-1.06 (11H, m, -(C$\underline{H}_2$)$_5$ and -CH_2O$\underline{H}$); ^{13}C NMR (δ ppm): 146.27, 145.98, 139.97, 129.96(2), 128.10(2) 127.60(2), 127.49(2), 126.60, 126.26, 126.20(2), 126.00, 125.61(2), 80.86, 62.76, 54.13, 32.52, 30.00, 29.22, 27.80, 25.41; MS (m/e): no M^+, 357 (M^+ - OH), 356 (M^+ - H_2O), 183 ($C_6H_5C(OH)C_6H_5$).

1.1.2-Trisphenyl-decan-1,10-diol (55b)

The crude intermediate **55b** was used for next dehydration step without purification.

1.1.2-Trisphenyl-dodecan-1,12-diol (55c)

IR, υ_{max} (thin film): 3600-3300 (OH), 3090-3015 (Ar-H), 2950-2850 (C-H), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.59-6.96 (15H, m, Ar-H), 3.69 (1H, dd, J=11.42 Hz, J'=2.86 Hz, -CH-), 3.60 (2H, t, J=6.62 Hz, $-CH_2OH$), 2.50 (1H, br s, COH), 1.90-1.06 (19H, m, $-(CH_2)_9-$ and $-CH_2OH$); ^{13}C NMR (δ ppm): 146.31, 145.97, 140.00, 129.99(2), 128.14(2), 127.62(2), 127.48(2), 126.60, 126.24(3), 126.02, 125.60(2), 80.90, 62.98, 54.13, 32.72, 30.02, 29.47(2), 29.41, 29.34(2), 27.86, 25.63; MS (m/e): no M^+, 413 (M^+ - OH), 412 (M^+ - H_2O), 183 ($C_6H_5C(OH)C_6H_5$).

C. Synthesis of x,y,y-trisphenyl-x-alken-1-ol (56).

The oily **55** (2.5 mmol) was dehydrated in 100 mL toluene in the presence of *p*TSA (100 mg, 0.56 mmol) at reflux for 2 hrs. After evaporation of the solvent, the residue was extracted with ether and water (5 x 50 mL), dried and evaporated to an colorless product **56**. Flash column chromatograph (hexane:acetone, 9:1) yield a viscous oil (99% yield from **55**).

7,8,8-Trisphenyl-7-octen-1-ol (56a)

mp: 90.5-91.5 °C; IR, υ_{max} (KBr): 3330 (br, OH), 3100-3000 (Ar-H), 2925-2825 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.34-6.86 (15H, m, Ar-$\underline{H}$), 3.55 (2H, t, J=6.60 Hz, -C$\underline{H}_2$OH), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.50-1.19 (9H, m, -(C$\underline{H}_2$)$_4$- and -CH$_2$O$\underline{H}$); ^{13}C NMR (δ ppm): 143.43, 142.92, 142.40, 140.89, 138.92, 130.65(2), 129.52(2), 129.43(2), 128.08(2), 127.76(2), 127.30(2), 126.53, 126.10, 125.67, 62.91, 35.72, 32.58, 29.33, 28.72, 25.30; MS (m/e): 356(M$^+$), 269(M$^+$ - $C_5H_{10}OH$).

9,10,10-Trispheny-9-decen-1-ol (56b)

IR, υ_{max}(thin film): 3330 (br, OH), 3100-3000 (Ar-H), 2925-2825 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.34-6.86 (15H, m, Ar-$\underline{H}$), 3.51 (2H, t, J=6.73 Hz, -C$\underline{H}_2$OH), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.45-1.14 (13H, m, -(C$\underline{H}_2$)$_6$- and -CH$_2$O$\underline{H}$); ^{13}C NMR (δ ppm): 143.22, 142.74, 142.19, 140.77, 138.84, 130.52(2), 129.26(4), 127.89(2), 127.60(2), 127.14(2), 126.36, 125.91, 125.49, 62.41, 35.62, 32.23, 29.35(2), 28.99, 28.58, 25.43; MS (m/e): 384 (M$^+$), 269 (M$^+$ - $C_7H_{14}OH$).

11,12,12-Trisphenyl-11-dodecen-1-ol (56c)

IR, υ_{max}(thin film): 3330 (br, OH), 3100-3000 (Ar-H), 2925-2825 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.34-6.86 (15H, m, Ar-$\underline{H}$), 3.59 (2H, t, J=6.86 Hz, -C$\underline{H}_2$OH), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.52-1.15 (17H, m, -(C$\underline{H}_2$)$_8$- and -CH$_2$O$\underline{H}$); ^{13}C NMR (δ ppm): 143.43, 142.93, 142.40, 141.00, 138.92, 130.65(2), 129.47(2), 129.41(2), 128.01(2), 127.72(2), 127.26(2), 126.48, 126.01, 125.61, 62.91, 35.76, 32.60, 29.58, 29.45, 29.36(2), 29.17, 28.75, 25.63. MS (m/e): 412 (M$^+$), 269 (M$^+$ - $C_9H_{18}OH$).

D. Synthesis of 1-bromo-x,y,y-trisphenyl-x-alkene (57).

A solution of the alcohol **56** (2.25 mmol), carbon tetrabromide (2.98 g, 9.00 mmol) and triphenylphosphine (2.36 g, 9.00 mmol) in dry ether (100 mL) was stirred at room temperature (23 °C) for 24 hrs under a nitrogen atmosphere. The triphenylphosphine oxide precipitate was filtrated and the resulting solution was washed thoroughly with water (5x25 mL), dried and evaporated to an oil. The crude material was purified by flash column chromatography (hexane:acetone, 99:1) to give the bromine **57** in 86% yield.

1-Bromo-7,8,8-trisphenyl-7-octene (57a)

IR, υ_{max} (thin film): 3090-3015 (Ar-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.37-6.86 (15H, m, Ar-$\underline{H}$), 3.31 (2H, t, J=6.84 Hz, -$C\underline{H}_2Br$), 2.43 (2H, m, -C=C-$C\underline{H}_2$-), 1.74 (2H, p, J=7.20 Hz, -$C\underline{H}_2CH_2Br$), 1.34-1.21 (6H, m, -$(C\underline{H}_2)_3$-); ^{13}C NMR (δ ppm): 143.42, 142.88, 142.32, 140.76, 139.23, 130.64(2), 129.53(2), 129.41(2), 128.14(2), 127.79(2), 127.33(2), 126.59, 126.13, 125.71, 35.64, 33.90, 32.60, 28.70, 28.54, 27.79; MS (m/e): 418 (M^+), 420 (M^++ 2), 269 (M^+ -$C_5H_{10}Br$).

1-Bromo-9,10,10-trisphenyl-9-decene (57b)

IR, υ_{max} (thin film): 3090-3015 (Ar-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.37-6.86 (15H, m, Ar-$\underline{H}$), 3.36 (2H, t, J=6.86 Hz, -$C\underline{H}_2Br$), 2.42 (2H, m, -C=C-$C\underline{H}_2$-), 1.79 (2H, p, J=7.36 Hz, -$C\underline{H}_2CH_2Br$), 1.34-1.17 (10H, m, -$(C\underline{H}_2)_5$-); ^{13}C NMR (δ ppm): 143.43, 142.93, 142.40, 140.94, 138.91, 130.67(20, 129.52(2), 129.45(2), 128.07(2), 127.75(2), 127.30(2), 126.53, 126.08, 125.66, 35.76, 34.05, 32.71, 29.45, 28.98, 28.69, 28.52, 28.02. MS (m/e): 446 (M^+), 448 (M^+ + 2), 269 (M^+ - $C_7H_{14}Br$).

1-Bromo-11,12,12-trisphenyl-11-dodecene (57c)

IR, υ_{max} (thin film): 3090-3015 (Ar-H), 2930-2850 (C-H), 1600 (C=O) cm$_{-1}$; ^{1}H NMR (δ ppm): 7.37-6.86 (15H, m, Ar-$\underline{H}$), 3.38 (2H, t, J=6.86 Hz, -C$\underline{H}_2$Br), 2.42 (2H, m, -C=C-C$\underline{H}_2$-), 1.82 (2H, p, J=7.20, -C$\underline{H}_2$CH$_2$Br), 1.34-1.16 (14H, m, -(C$\underline{H}_2$)$_7$-); ^{13}C NMR (δ ppm): 143.48, 142.98, 142.48, 141.05, 139.01, 130.69(2), 129.53(2), 129.46(2), 128.07(2), 127.73(2), 127.30(2), 126.53, 126.07, 125.66, 35.82, 34.04, 32.80, 29.59, 29.33(2), 29.18, 28.77, 28.68, 28.14; MS (m/e): 474 (M$^+$), 476 (M$^+$ + 2), 269 (M$^+$ - $C_9H_{18}Br$).

E. Synthesis of 1-[(2'-aminoethyl)amino]-x,y,y-trisphenyl-x-alkene (58).

Under a nitrogen atmosphere, ethylenediamine (900 mg, 15.00 mmol) was added to a solution of the bromide **57** (1.50 mmol) in 80 mL of dry methanol. After boiling for two days under reflux (sometimes longer reaction period was required), the solvent was evaporated. The resulting residue was dissolved in ether (150 mL) and washed with a solution of $NaHCO_3$ (30 mL, 5% aqueous) and with water (5x30 mL). The ethereal phase was dried and evaporated to a viscous oil **58**. The yield was 90%.

1-[(2'-Aminoethyl)amino]-7,8,8-trisphenyl-7-octene (58a)

IR, υ_{max} (thin film): 3290 (br, N-H), 3090-3015 (Ar-H), 2930-2850 (C-H), 1650 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.35-6.86 (15H, m, Ar-$\underline{H}$), 2.76, 2.61 (4H, 2xt, J=6.06 Hz, -NHC$\underline{H}_2$C$\underline{H}_2$NH$_2$), 2.51 (2H, t, J=7.25 Hz, -C$\underline{H}_2$NH-), 2.43 (2H, m, -C=C-C$\underline{H}_2$-), 1.80 (3H, s, br, -N$\underline{H}$- and -N$\underline{H}_2$), 1.38-1.18 (8H, m, -(C$\underline{H}_2$)$_4$-); ^{13}C NMR (δ ppm): 143.30, 142.80, 142.25, 140.80, 138.91, 130.54(2), 129.38(2), 129.32(2), 127.96(2), 127.64(2), 127.18(2), 126.41, 125.96, 125.54, 52.32, 49.65, 41.54, 35.65, 29.84, 29.41, 28.65, 26.86; MS (m/e): 398 (M^+), 368 (M^+ -CH_2NH_2), 355 (M^+ - CH_2CH_2NH), 269 (M^+ - $C_5H_{10}NHCH_2CH_2NH_2$).

1-[(2'-Aminoethyl)amino]-9,10,10-trisphenyl-9-decene (58b)

IR, υ_{max} (thin film): 3290 (br, N-H), 3090-3015 (Ar-H), 2930-2850 (C-H), 1650 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.34-6.86 (15H, m, Ar-$\underline{H}$), 2.78, 2.65 (4H, 2xt, J=6.03 Hz, $-NHC\underline{H}_2C\underline{H}_2NH_2$), 2.56 (2H, t, J=7.23 Hz, $-C\underline{H}_2NH-$), 2.42 (2H, m, $-C=C-C\underline{H}_2-$), 1.58 (3H, s, br, $-N\underline{H}-$ and $-N\underline{H}_2$), 1.43-1.17 (12H, m, $-(C\underline{H}_2)_6-$). ^{13}C NMR (δ ppm): 143.38, 142.91, 142.36, 140.98, 138.90, 130.62(2), 129.46(2), 129.40(2), 128.00(2), 127.67(2), 127.23(2), 126.44, 126.00, 125.58, 52.49, 49.85, 41.70, 35.74, 30.06, 29.52, 29.34, 29.12, 28.72, 27.22; MS (m/e): 426 (M^+), 396 (M^+ - CH_2NH_2), 383 (M^+ - CH_2CH_2NH), 269 (M^+ - $C_7H_{14}NHCH_2CH_2NH_2$).

1-[(2'-Aminoethyl)amino]-11,12,12-trisphenyl-11-dodecene (58c)

IR, υ_{max} (thin film): 3290 (br, N-H), 3090-3015 (Ar-H), 2930-2850 (C-H), 1650 (N-H, bending), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.34-6.86 (15H, m, Ar-$\underline{H}$), 2.80, 2.66 (4H, 2xt, J=6.03 Hz, $-NHC\underline{H}_2C\underline{H}_2NH_2$), 2.59 (2H, t, J=7.23 Hz, $-C\underline{H}_2NH-$), 2.42 (2H, m, $-C=C-C\underline{H}_2-$), 1.64 (3H, s, br, -NH- and $-N\underline{H}_2$), 1.48-1.15 (16H, m, $-(C\underline{H}_2)_8-$); ^{13}C NMR (δ ppm): 143.43, 142.97, 142.41, 141.05, 138.91, 130.66(2), 129.49(2), 129.43(2), 128.03(2), 127.70(2), 127.26(2), 126.48, 126.02, 125.61, 52.49, 49.90, 41.66, 35.79, 30.09, 29.61, 29.49(2), 29.43, 29.21, 28.76, 27.33; MS (m/e): 454 (M^+), 424 (M^+ - CH_2NH_2), 411 (M^+ - CH_2CH_2NH), 269 (M^+ - $C_9H_{18}NHCH_2CH_2NH_2$).

F. Synthesis of 1-{cis-[(2'-aminoethyl)amino]dichloroplatinum(II)}-x,y,y-trisphenyl-x-alkene (40).

A solution of potassium tetrachloroplatinate (II) (219 mg, 0.528 mmol) in 7.5 mL of a mixture of DMF and water (2:1) was added to a warm (35 °C) solution of diamine **58** (0.528 mmol) in 5 mL of DMF. The resulting mixture (pH=9-10) was stirred in the dark for 2-3 days until the pH value reached 4-5. Then, one drop of N,N-dimethylsulfoxide was added and the stirring was continued for 2 hrs. The solvent was evaporated and the residue was suspended in saturated potassium chloride solution (30 mL). A vigourous stirring was essential in order to pulverized the lumps of platinum (II) complex **40** The resulting suspension was filtered, washed with water (100-250 mL), and dried in a desiccator. The product can be further purified either by flash column chromatography or by preparative TLC (CH_2Cl_2:CH_3OH, 98:2). The crude yield was around 80%.

1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-7,8,8-trisphenyl-7-octene (40a)

mp > 210 °C (dec.); IR, υ_{max} (KBr): 3250-3150 (N-H), 3090-3015 (Ar-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^1H NMR (δ ppm): 7.33-6.86 (15H, m, Ar-$\underline{H}$), 5.51, 4.89, 4.72 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.35, 3.09, 2.72 (6H, 3xbr s, $-C\underline{H}_2NHC\underline{H}_2C\underline{H}_2NH_2$), 2.39 (2H, m, $-C{=}C{-}C\underline{H}_2-$), 1.70, 1.54 (2H, 2xbr s, $-C\underline{H}_2CH_2NH-$), 1.40-1.10 (6H, m, $-(C\underline{H}_2)_3-$); ^{13}C NMR (δ ppm): 143.35, 142.86, 142.23, 140.60, 139.21, 130.66(2), 129.52(2), 129.41(2), 128.17(2), 127.82(2), 127.33(2), 126.60, 126.14, 125.71, 55.17, 53.23, 47.03, 35.61, 29.11, 28.44, 27.20, 26.02. Anal. calcd. for $C_{28}H_{34}Cl_2N_2Pt$: C 50.60, H 5.17, N 4.22; found: C 50.63, H 5.20, N 4.19.

1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-9,10,10-trisphenyl-9-decene (40b)

mp > 210 °C (dec.); IR, υ_{max} (KBr): 3250-3150 (N-H), 3090-3015 (Ar-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.33-6.86 (15H, m, Ar-$\underline{H}$), 5.56, 5.30, 5.04 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.38, 3.19, 3.05, 2.72 (6H, 4xbr s, $-C\underline{H}_2NHC\underline{H}_2C\underline{H}_2NH_2$), 2.40 (2H, m, -C=C-C$\underline{H}_2$-), 1.67, 1.48 (2H, 2xbr s, $-C\underline{H}_2C\underline{H}_2NH-$), 1.40-1.10 (10H, m, -(C$\underline{H}_2$)$_5$-); ^{13}C NMR (δ ppm): 143.43, 142.95, 142.40, 140.92, 139.05, 130.69(2), 129.53(2), 129.46(2), 128.10(2), 127.77(2), 127.31(2), 126.56, 126.09, 125.68, 55.20, 53.43, 47.20, 35.79, 29.71, 29.52, 29.02, 28.76, 27.39, 26.37. Anal. calcd. for $C_{30}H_{38}Cl_2N_2Pt$: C 52.01, H 5.54, N 4.05; found: C 52.05, H 5.51, N 4.02.

1-{Cis-[(2'-aminoethyl)amino]dichloroplatinum (II)}-11,12,12-trisphenyl-11-dodecene (40c)

mp > 210 °C (dec.); IR, υ_{max} (KBr): 3250-3150 (N-H), 3090-3015 (Ar-H), 2930-2850 (C-H), 1600 (C=C) cm^{-1}; ^{1}H NMR (δ ppm): 7.33-6.86 (15H, m, Ar-$\underline{H}$), 5.62, 5.01, 4.84 (3H, 3xbr s, -N$\underline{H}$- and -N$\underline{H}_2$), 3.50, 3.15, 2.88, 2.76 (6H, 4xbr s, $-C\underline{H}_2NHC\underline{H}_2C\underline{H}_2NH_2$), 2.40 (2H, m, -C=C-C$\underline{H}_2$-), 1.68, 1.50 (2H, 2xbr s, $-C\underline{H}_2CH_2NH-$), 1.40-1.10 (14H, m, -(C$\underline{H}_2$)$_7$-). ^{13}C NMR (δ ppm): 143.44, 142.97, 142.42, 141.00, 138.91, 130.69(2), 129.51(2), 129.45(2), 128.07(2), 127.75(2), 127.28(2), 126.53, 126.05, 125.65, 55.40, 53.39, 46.86, 35.83, 29.67, 29.43(2), 29.23(2), 28.79, 27.40, 26.54. Anal. calcd. for $C_{32}H_{42}Cl_2N_2Pt$: C 53.32, H 5.89, N 3.89; found: C 53.28, H 5.90, N 3.92.

3.2 In Vitro Antitumor Activity

3.2.1 Materials of Microcytostasis Assay

A. Drugs:

39a-d, 40a-c and 41 synthesized as described previously;

Cisplatin obtained from Sigma Chemical Company, USA;

Tamoxifen obtained from Aldrich Chemical Company, Inc., USA.

B. Cell lines and culture:

Human breast cancer cell lines: MCF-7 and MDA-MB-231 were obtained from the American Type Culture Collection, Maryland, USA. Both MCF-7 and MDA-MB-231 cells were cultured in RPMI-1640 supplemented with 2mM glutamine, 10% fetal bovine serum (Gibcol, Burlington, Ontario, Canada) and 100 U gentamycin/ml (Sigma Chemical Company, USA).

C. Phosphate Buffered Saline (PBS, pH=7.4) prepared from PBS tablets (Oxoid, Unipath Ltd., England), dissolved in water as per manufacturer's instructions.

D. Microtitre Plates, 96 wells obtained from Flow Lab. Inc., McLean, Virginia, USA.

E. MTT and DMSO obtained from Sigma Chemical Company, USA.

F. Plate Reader: Behring Elisa Processor II (Behring, Marburg, Germany).

3.2.2 Method of Microcytostasis Assay

The MTT assay was carried out essentially as described by J. Carmichael and co-workers.[72] Under sterile conditions, the 40 mM solution of drugs in DMSO was diluted with fresh medium (RPMI-1640) to a concentration of 400 μM, then different drug dilutions were prepared in the culture medium (range 0.1- 400 μM). A total of 100 μL cell culture medium RPMI-1640 containing 2000 viable cells was plated per well into 96-well microtitre plate, and preincubated for 24 hours at 37 °C in a 5% CO_2 atmosphere. Then, the medium was removed from the cells, and 100 μL of fresh medium containing various concentrations of a drug was added to the cultures. Tests were performed in 8 wells for each test dilution, with appropriate control wells which received 100 μL of medium only. The cells were incubated with the drug for 72 hours. Next, the medium was removed and the cells were washed with the sterile phosphate-buffered saline (PBS, pH=7.4). Cell survival was evaluated with MTT by the addition of a 50 μL solution containing 2.5 mg/mL in PBS:RPMI-1640 (1:4, v/v). After 4 hours incubation at 37 °C, the solution was aspirated from each well, and 100 μL DMSO was added to dissolve the precipitate of reduced MTT. The plates were shaken on a plate shaker for 15 minutes, then the absorbance was spectrophotometrically determined at 570 and 630 nm with a Behring Elisa Processor II (Behring, Marburg, Germany). Results from the plate reader were expressed as follows:

$$\text{Percentage Cell Survival at Each Dilution}$$

$$= \frac{\text{Mean Absorbance at Each Dilution}}{\text{Mean Control Absorbance}} \text{ X } 100.$$

A dose response curve of percentage cell survival (ordinate) against drug concentration (abscissa) was constructed.

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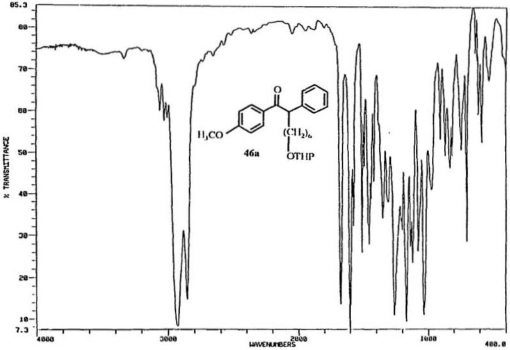
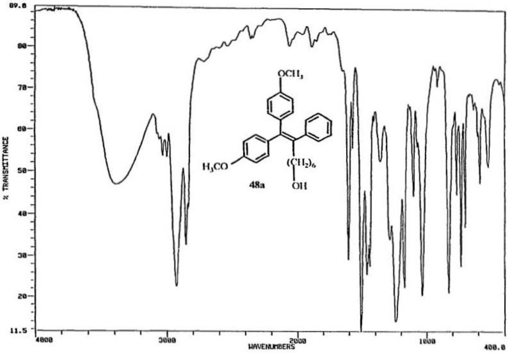
APPENDIX

The selected IR, ^{1}H NMR, ^{13}C NMR of the synthetic samples were arranged according to the order in which they appear in the text. For the instruments employed, see **General Procedures** in ***Charpter 3.***

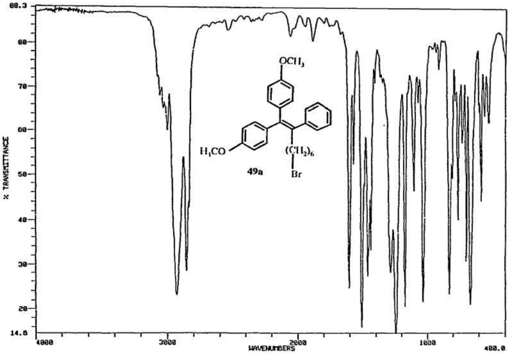
IR Spectrum of Compound **46 a**.



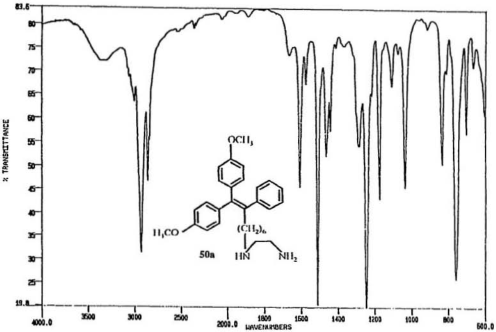
IR Spectrum of Compound **48a**.



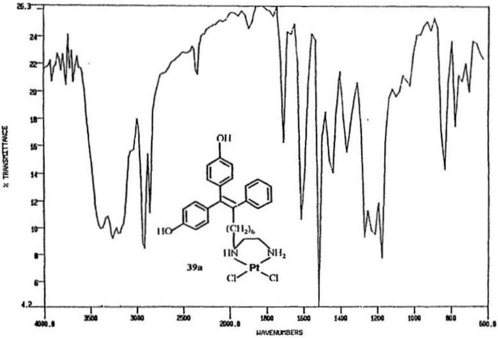
IR Spectrum of Compound **49a**.



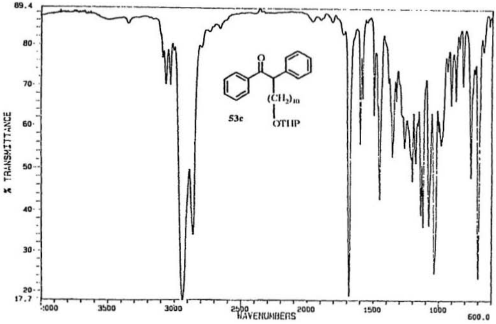
IR Spectrum of Compound **50a**.



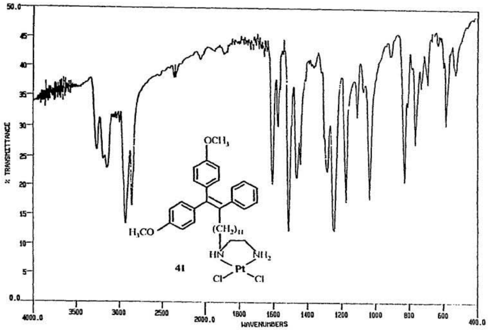
IR Spectrum of Compound **39a** (KBr).



IR Spectrum of Compound **53c** (KBr).

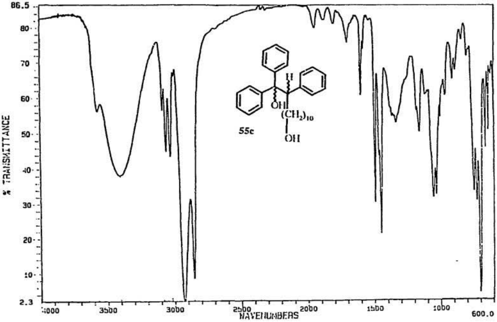


IR Spectrum of Compound **41** (KBr).

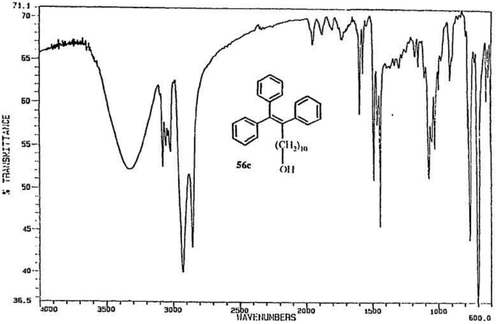


41

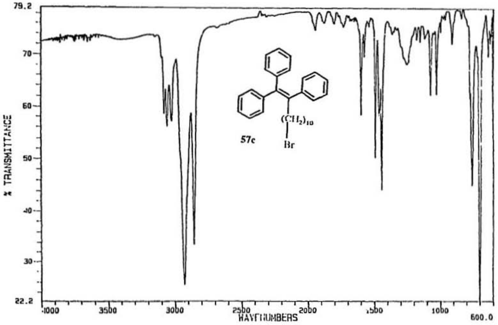
IR Spectrum of Compound **55c**.



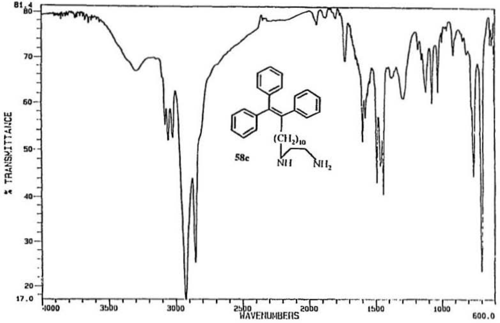
IR Spectrum of Compound **56c**.



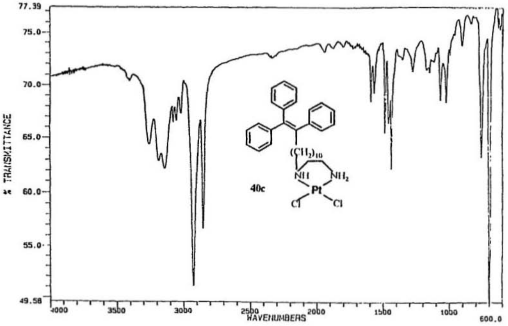
IR Spectrum of Compound **57c**.



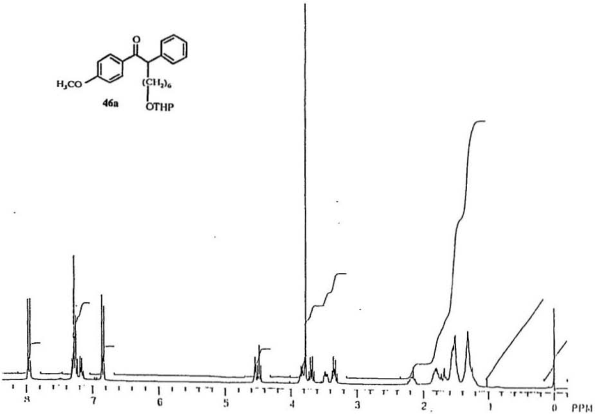
IR Spectrum of Compound **58c**.



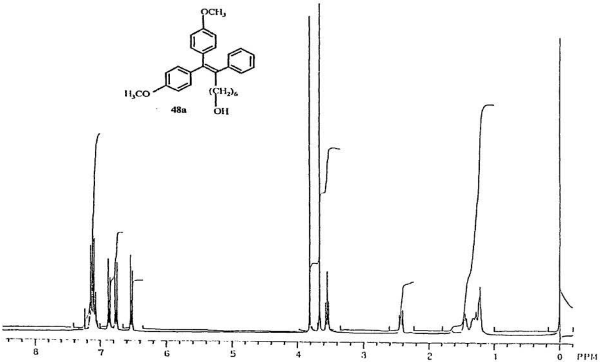
IR Spectrum of Compound **40c** (KBr).



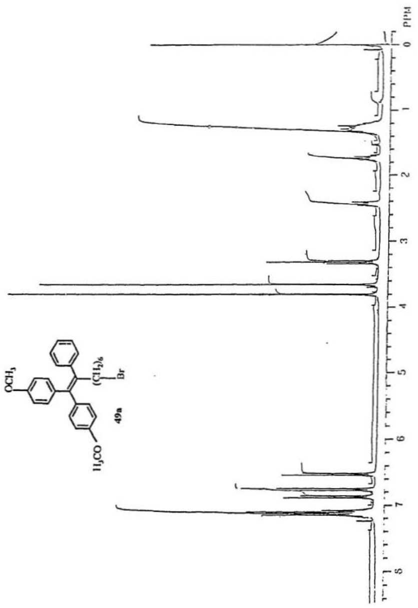
¹H NMR Spectrum of Compound **46 a**.



^{1}H NMR Spectrum of Compound **48 a**.

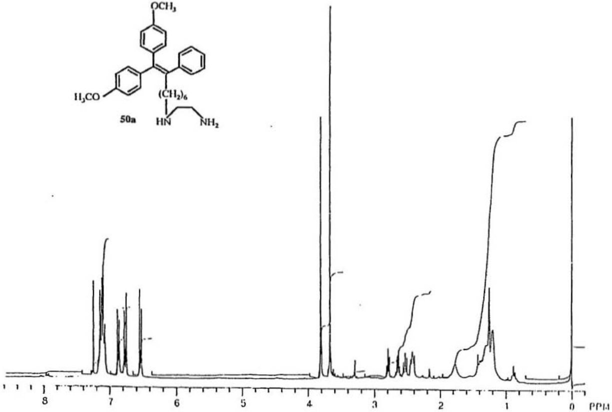


1H NMR Spectrum of Compound 49a.

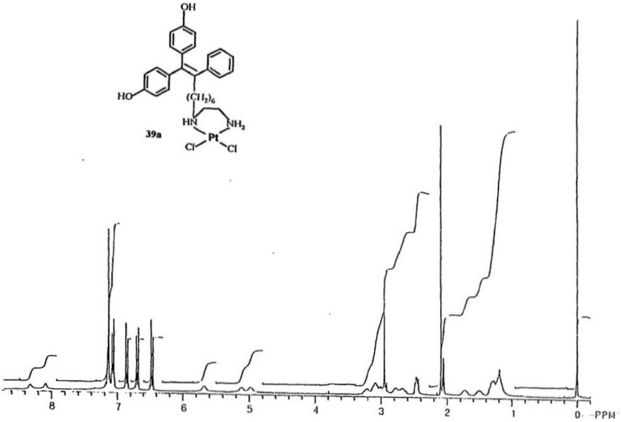


49a

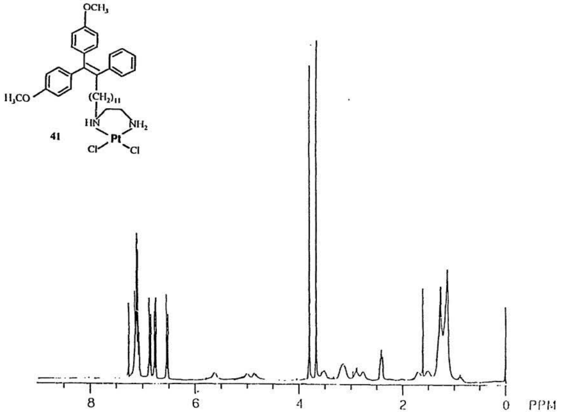
^{1}H NMR Spectrum of Compound **50a**.



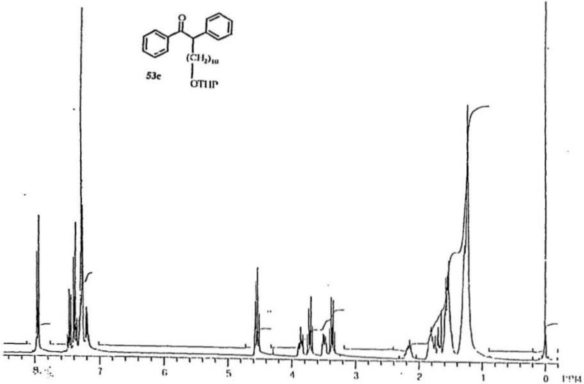
^{1}H NMR Spectrum of Compound **39a** (Acetone-d_6).



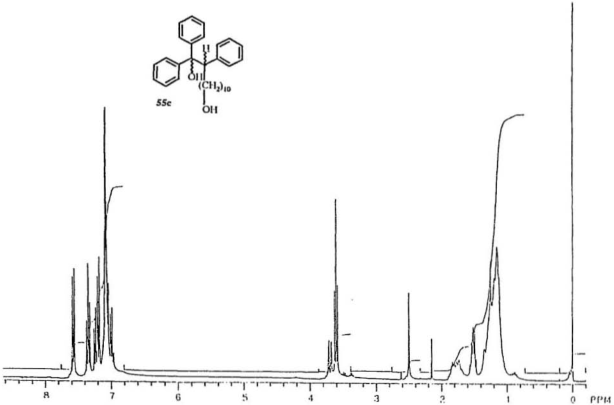
^{1}H NMR Spectrum of Compound **41**.



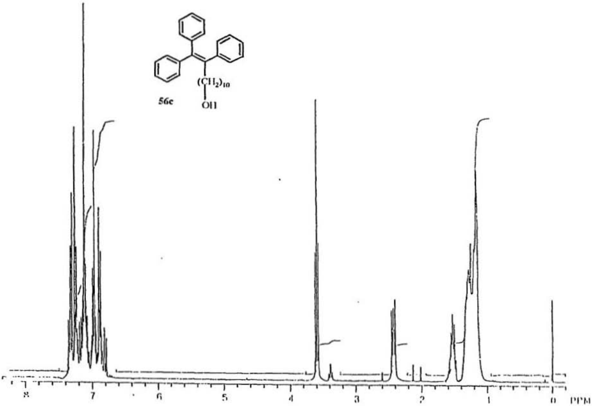
^{1}H NMR Spectrum of Compound **53c**.



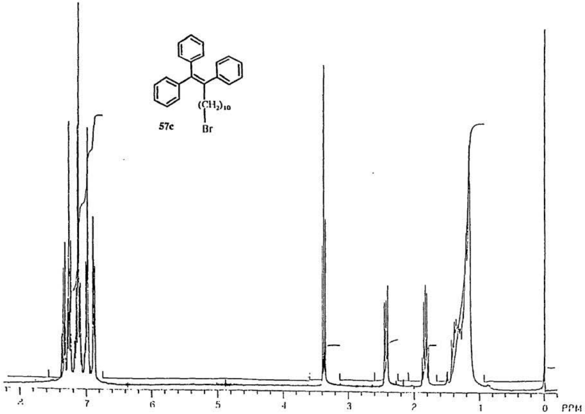
^{1}H NMR Spectrum of Compound **55c**.



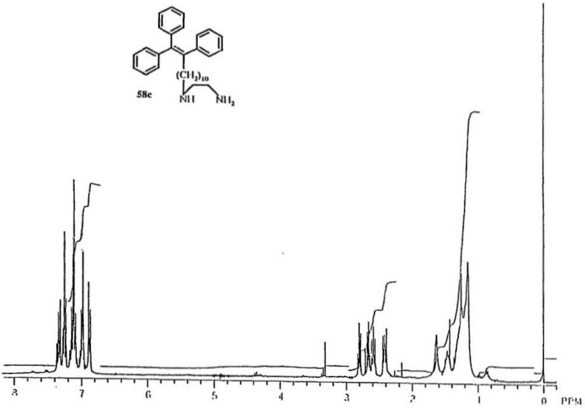
^{1}H NMR Spectrum of Compound **56c**.



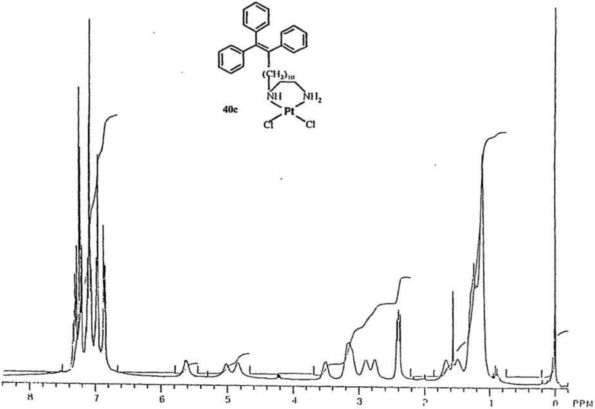
^{1}H NMR Spectrum of Compound **57c**.



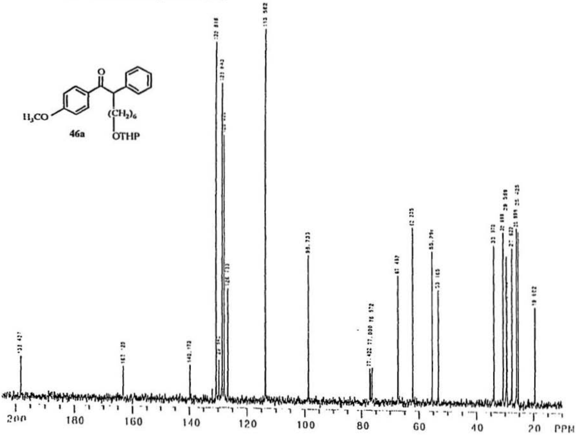
^{1}H NMR Spectrum of Compound **58c**.



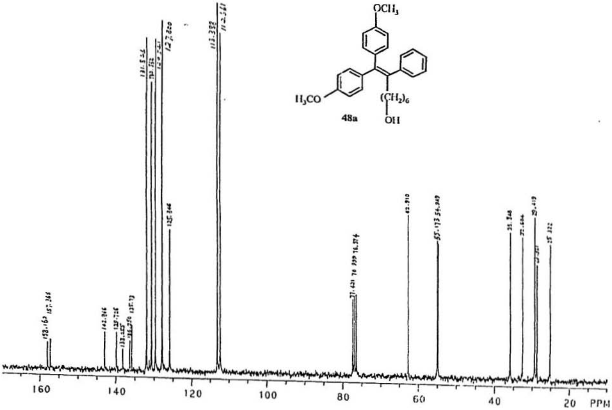
^1H NMR Spectrum of Compound **40c**.


40c

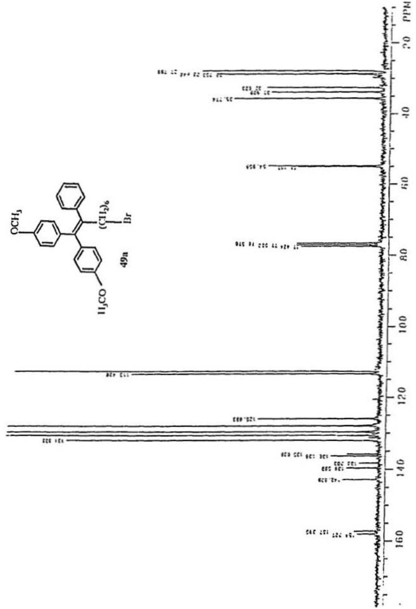
13C NMR Spectrum of Compound **46 a**.



^{13}C NMR Spectrum of Compound **48 a.**

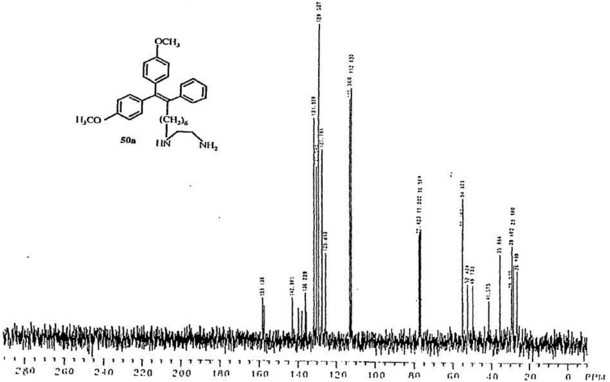

48a

^{13}C NMR Spectrum of Compound 49a.

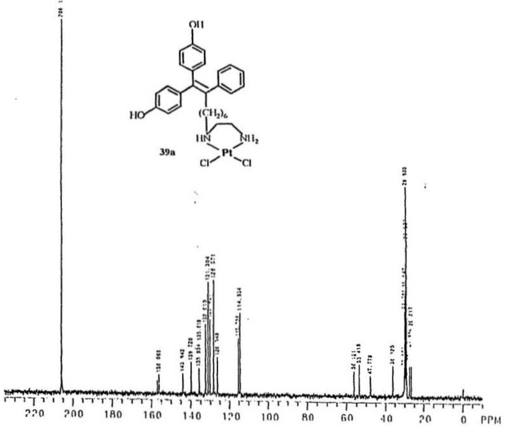


49a

13C NMR Spectrum of Compound **50a**.

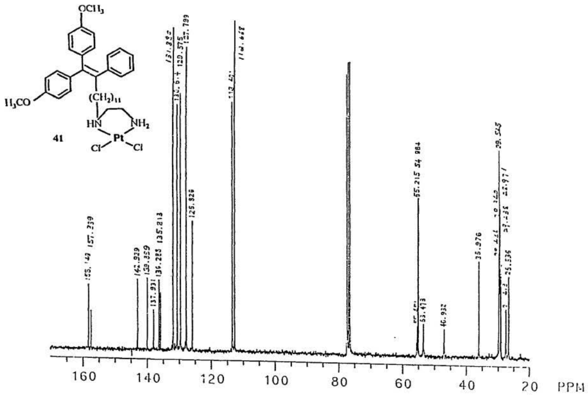


^{13}C NMR Spectrum of Compound **39a** (Acetone-d$_6$).

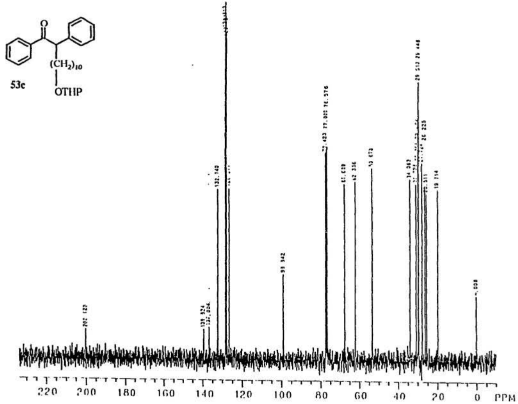


39a

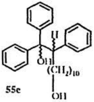
13C NMR Spectrum of Compound 4 1.



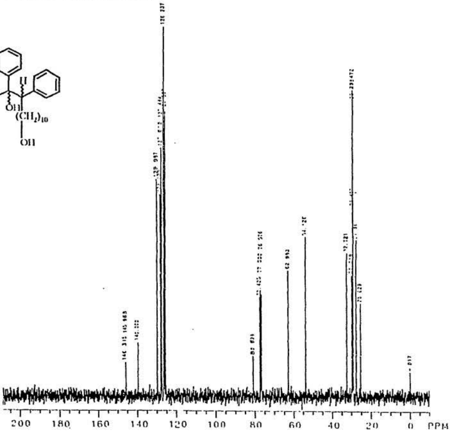
^{13}C NMR Spectrum of Compound **53c**.



^{13}C NMR Spectrum of Compound **55c**.

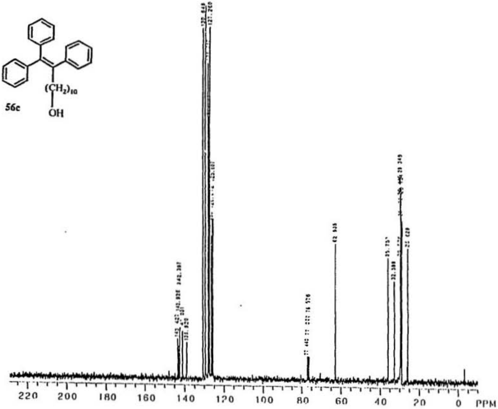


55c

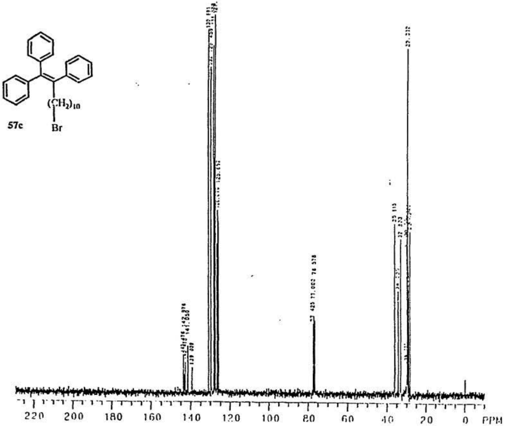


^{13}C NMR Spectrum of Compound **56c**.

56c

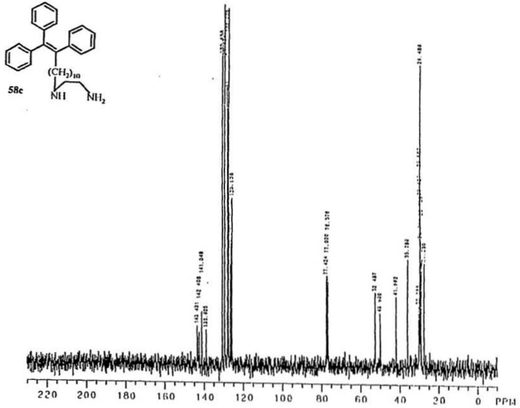


^{13}C NMR Spectrum of Compound **57c**.

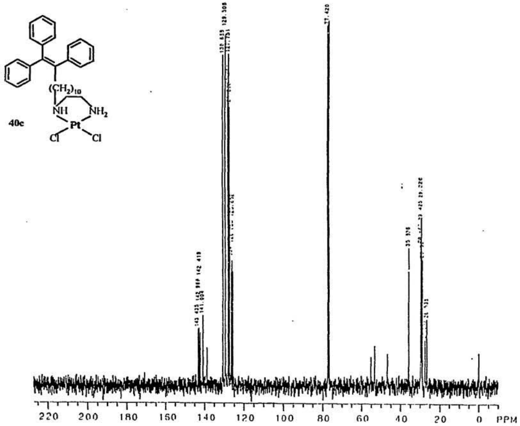


57c

^{13}C NMR Spectrum of Compound **58c**.


58c

^{13}C NMR Spectrum of Compound **40c**.



40c