

Macromolecular antineoplastic iron and platinum co-ordination compounds



Hembe Elie Mukaya

A thesis submitted to the Faculty of Science, University of the Witwatersrand, Johannesburg, in fulfillment of the requirements for the degree of Doctor of Philosophy of Science.

Johannesburg, 2013

DECLARATION

I declare that this dissertation is my own unaided work. It is being submitted for the degree of Doctor of Science, in the University of the Witwatersrand, Johannesburg, South Africa. It has not been submitted before for any degree or examination in any other University.

Hembe Elie Mukaya

_____ day of _____, 2013.

ABSTRACT

Chemotherapy, while representing a vital component of cancer treatment modalities, has so far not fulfilled basic expectations with unsatisfactory cure rates and frequent relapse due to limited effectiveness of the therapeutic drugs, severe side effects and resistance problems. The platinum-containing drugs used in present clinical practice are no exception to this generalized finding. While highly effective against a small number of malignancies, they generally share in the deficiencies of other anticancer agents. To address this issue, intense research is being undertaken to develop novel platinum-compounds offering enhanced therapeutic effectiveness. To accomplish this, several new avenues of development are being pursued world-wide, and one of these involving the binding of monomeric anticancer drug systems to water-soluble, biocompatible and biodegradable polymeric carriers, was utilized in the current research. As part of the ongoing research, this dissertation demonstrates the preparation of several water-soluble polymeric carriers bearing pre-synthesized monomers aimed to anchor the platinum drug. The monomers of interest were aspartic acid, *p*-aminobenzoic acid and *p*-aminosalicylic acid derivatives; while the water-soluble carriers were polyaspartamides, prepared by an aminolytic ring-opening process of polysuccinimide. The platination agents were conjugated to the polymer backbone both *via* amine and *via* leaving-group ligands, such as dihydroxylato, dicarboxylato and carboxylatohydroxylato. In order to demonstrate the multidrug-binding capacity of the carriers, platinum complexes were co-conjugated to polymeric conjugates containing ferrocene. The *in vitro* studies against a human breast cancer (MCF-7) cell line showed IC₅₀ values ranging from 48.92 µg.mL⁻¹ to 281.37 µg.mL⁻¹ for the platinum conjugates, 13.18 µg.mL⁻¹ to 149.67 µg.mL⁻¹ for ferrocene conjugates and 6.22 µg.mL⁻¹ to 83.86 µg.mL⁻¹ for platinum/ferrocene co-conjugates; and these values were on average 4 fold more active than the parent drug. The results of these

preliminary tests provide proof of the principle that polymer-drug conjugates can play a role in future cancer therapy.

I will sing to the Lord as long as I will live;
I will sing praise to my God while I have my being.
May my meditation be sweet to Him;
I will be glad in the Lord.
You are He who took me out of the womb;
You made me trust while on my mother's breasts.
I was cast upon You from birth.
From my mother's womb, You have been my God.
Bless the Lord, O my soul;
And all that is within me, bless His holy name!
Bless the Lord, O my soul;
And forget not all His benefits.

Psalm 104:33-34; 22:9-10; 103:1-2 (NKJV)

DEDICATION

To the Lord Almighty God for His unfailing love.

ACKNOWLEDGEMENTS

The author expresses his deepest gratitude to:

- The Lord Almighty God, my creator, for the gift of life.
- Professor Eberhard W. Neuse, for his extreme kindness, guidance, confidence, patience and unending encouragement throughout this project. Professor Neuse's unquenchable passion for science, incredible energy and encyclopedic style is highly communicable. To study under his supervision is a rare fortune I will forever cherish.
- Professor Robyn van Zyl, my co-supervisor, for helpfulness and tireless guidance and for the cell culture testing.
- Dr. M. Tutu, Analytical laboratory, for the atomic absorption analysis.
- Dr. Richard Mampa of the School of Chemistry for the countless NMR spectra scanned for me, and for his kindness and friendship.
- Mr. Chien Teng Chen from the Pharmacology Department for the cell culture testing.
- De Bruyn Spectroscopic Solutions for the Platinum analysis of the free drug and prodrugs.
- Research Office and the School of Chemistry of the University of the Witwatersrand for their financial assistance.
- My colleagues in the Polymer Laboratory, particularly Jacques K. Diainabo, your friendship and sense of humor will always be remembered.
- My parents for being supportive and trusting the Lord God for me to come this far.
- Families Mukaya and Simon, and friends for their love, support and their gift of prayer.
- My spiritual family, your prayers were not worthless.
- Last but not least, my beautiful wife Lydie K. Mukaya, your love, assistance and understanding will always be remembered.

TABLE OF CONTENTS

Declaration	ii
Abstract	iii
Dedication	vi
Acknowledgement	vii
Table of contents	viii
List of figures	xv
List of schemes	xvii
List of tables	xix
List of abbreviations	xxi
CHAPTER 1: INTRODUCTION	1
1.1 Cancer overview.....	1
1.2 Objectives of the study.....	3
CHAPTER 2: LITERATURE REVIEW	5
2.1 Treatment of cancer: an overview.....	5
2.1.1 Surgery.....	5
2.1.2 Radiation therapy.....	6
2.1.3 Chemotherapy.....	8
2.1.3.1 Chemotherapeutic agents.....	9
2.1.3.1.1 Alkylating agents and platinum antitumor compounds.....	10
2.1.3.1.2 Antibiotics.....	18
2.1.3.1.3 Antimetabolites.....	24
2.1.3.1.4 Ferrocenyl drugs.....	26
2.2 Polymer-Drug Conjugation concept.....	29
2.2.1 Requirements for polymeric drug carrier.....	30
2.2.1.1 Solubility.....	30
2.2.1.2 Degradability.....	31

2.2.1.3 Non-immunogenicity.....	31
2.2.1.4 Drug anchoring	32
2.2.2 Pharmacokinetic benefits of macromolecular conjugates.....	33
CHAPTER 3: RESULTS AND DISCUSSION.....	37
3.1 Synthesis of carrier polymers.....	37
3.1.1 Synthesis of poly-DL-Succinimide.....	40
3.1.2 Synthesis of poly-α,β-DL-aspartamides.....	40
3.1.3 Synthesis of copolyaspartamides.....	41
3.1.4 Copolyaspartamides containing primary amine-terminated side chain as drug anchoring site.....	44
3.1.4.1 Synthesis of <i>p</i> -aminobenzamidopropylamine.....	45
3.1.4.2 Copolyaspartamides containing primary amine-terminated side chain.....	46
3.1.5 Copolyaspartamides containing carboxylatohydroxylato ligand as drug anchoring site.....	52
3.1.5.1 Synthesis of <i>p</i> -N-(4,8-diaza-octanoyl)salicylic acid (PASA-PDA) and <i>p</i> -N-(4,8-diaza-heptanoyl)salicylic acid (PASA-EDA).....	52
3.1.5.2 Copolyaspartamides containing carboxylatohydroxylato ligand....	55
3.1.6 Polyaspartamide carriers bearing dihydroxyl-functionalised side chain as drug anchoring site.....	61
3.2 Polymer platinum conjugation.....	68
3.2.1 Monoamineplatinum(II) conjugates.....	73
3.2.1.1 First route.....	73
3.2.1.2 Second route.....	75
3.2.2 Carboxylatohydroxylato-platinum(II) chelated conjugates.....	78
3.2.3 Dicarboxylato-platinum chelated conjugates.....	81
3.3 Polymer multi-drug anchoring.....	84
3.3.1 Synthesis of 4-ferrocenylbutamidopropylamine (M6).....	84
3.3.2 Co-conjugates coordinated to monoamine platinum and ferrocene.....	86

3.3.3 Co-conjugates coordinated carboxylatohydroxylato-platinum and ferrocene	92
3.3.4 Co-conjugates coordinated to dihydroxylato-platinum and ferrocene	101
3.3.5 Co-conjugates coordinated to dicarboxylato-platinum and ferrocene	106
3.3.5.1 <i>Synthesis of N-(4,8-diaza-octanoyl)aspartic acid</i>	108
3.3.5.2 <i>Co-conjugates coordinated to dicarboxylato- Pt and ferrocene</i> ...	108
3.4 Cell culture testing	114
Chapter 4: EXPERIMENTAL	118
4.1 Chemical synthesis	118
4.1.1 General procedures	118
4.1.2 Solvents and reagents	119
4.1.3 Preparation of monomers	119
4.1.3.1 <i>Synthesis of p-aminobezamidopropylamine M1</i>	119
4.1.3.2 <i>Synthesis of acryloylamidosalicylic acid M2</i>	120
4.1.3.3 <i>Synthesis of M3</i>	120
4.1.3.4 <i>Synthesis of M4</i>	121
4.1.3.5 <i>Synthesis of 4-ferrocenylbutanoic acid M5</i>	121
4.1.3.6 <i>Synthesis of 4-ferrocenylbutamidepropylamine (Fc-PDA or M6)</i> ...	121
4.1.3.7 <i>Synthesis of M7</i>	122
4.1.3.8 <i>Synthesis of M8</i>	122
4.1.4 Poly-DL-succinimide	123
4.1.5 Preparation of polymeric carriers	123
4.1.5.1 <i>Carriers containing M1</i>	123
4.1.5.1.1 <i>Polyaspartamide 1</i>	123
4.1.5.1.2 <i>Polyaspartamide 2</i>	124
4.1.5.1.3 <i>Polyaspartamide 3</i>	124
4.1.5.1.4 <i>Polyaspartamide 4</i>	125
4.1.5.6 <i>Polyaspartamide 5</i>	125

4.1.5.2 Carriers containing PDA.....	126
4.1.5.2.1 Polyaspartamide 6	126
4.1.5.2.2 Polyaspartamide 7	126
4.1.5.3 Carriers containing M3	127
4.1.5.3.1 Polyaspartamide 8	127
4.1.5.3.2 Polyaspartamide 9	127
4.1.5.3.3 Polyaspartamide 10	128
4.1.5.3.4 Polyaspartamide 11	128
4.1.5.3.5 Polyaspartamide 12	129
4.1.5.4 Carriers containing M4	129
4.1.5.4.1 Polyaspartamide 13	129
4.1.5.4.2 Polyaspartamide 14	130
4.1.5.4.3 Polyaspartamide 15	130
4.1.5.4.4 Polyaspartamide 16	131
4.1.5.4.5 Polyaspartamide 17	131
4.1.5.5 Carriers containing dopamine.....	131
4.1.5.5.1 Polyaspartamide 18	131
4.1.5.5.2 Polyaspartamide 19	132
4.1.5.5.3 Polyaspartamide 20	132
4.1.5.5.4 Polyaspartamide 21	133
4.1.5.5.5 Polyaspartamide 22	133
4.1.5.5.6 Polyaspartamide 23	134
4.1.6 Polymer platinum conjugation	134
4.1.6.1 Platination of polyaspartamides 1 to 5	135
4.1.6.1.1 Conjugate 1-Pt	135
4.1.6.1.2 Conjugate 2-Pt	135
4.1.6.1.3 Conjugate 3-Pt	135
4.1.6.1.4 Conjugate 4-Pt	136
4.1.6.1.5 Conjugate 5-Pt	136
4.1.6.2 Platination of polyaspartamides 6-Pt and 7-Pt	136
4.1.6.2.1 Conjugate 6-Pt	136

4.1.6.2.2 Conjugate 7-Pt	137
4.1.6.3 Platination of polyaspartamides 8 to 12	137
4.1.6.3.1 Conjugate 8-Pt	137
4.1.6.3.2 Conjugate 9-Pt	138
4.1.6.3.3 Conjugate 10-Pt	138
4.1.6.3.4 Conjugate 11-Pt	138
4.1.6.3.5 Conjugate 12-Pt	139
4.1.6.4 Platination of polyaspartamides 13 to 17	139
4.1.6.4.1 Conjugate 13-Pt	139
4.1.6.4.2 Conjugate 14-Pt	140
4.1.6.4.3 Conjugate 15-Pt	140
4.1.6.4.4 Conjugate 16-Pt	140
4.1.6.4.5 Conjugate 17-Pt	140
4.1.6.5 Platination of polyaspartamides 18 to 23	141
4.1.6.5.1 Conjugate 18-Pt	141
4.1.6.5.2 Conjugate 19-Pt	141
4.1.6.5.3 Conjugate 20-Pt	141
4.1.6.5.4 Conjugate 21-Pt	142
4.1.6.5.5 Conjugate 22-Pt	142
4.1.6.5.6 Conjugate 23-Pt	142
4.1.7 Synthesis of co-conjugates	142
4.1.7.1 Conjugates 24-Fc to 28-Fc	143
4.1.7.1.1 Conjugate 24-Fc	143
4.1.7.1.2 Conjugate 25-Fc	143
4.1.7.1.3 Conjugate 26-Fc	144
4.1.7.1.4 Conjugate 27-Fc	144
4.1.7.1.5 Conjugate 28-Fc	144
4.1.7.2 Conjugates 29-Fc to 33-Fc	145
4.1.7.2.1 Conjugate 29-Fc	145
4.1.7.2.2 Conjugate 30-Fc	145
4.1.7.2.3 Conjugate 31-Fc	146

4.1.7.2.4 Conjugate 32-Fc	146
4.1.7.2.5 Conjugate 33-Fc	146
4.1.7.3 Conjugates 34-Fc to 38-Fc	147
4.1.7.3.1 Conjugate 34-Fc	147
4.1.7.3.2 Conjugate 35-Fc	147
4.1.7.3.3 Conjugate 36-Fc	147
4.1.7.3.4 Conjugate 37-Fc	148
4.1.7.3.5 Conjugate 38-Fc	148
4.1.7.4 Conjugates 39-Fc to 43-Fc	148
4.1.7.4.1 Conjugate 39-Fc	148
4.1.7.4.2 Conjugate 40-Fc	149
4.1.7.4.3 Conjugate 41-Fc	149
4.1.7.4.4 Conjugate 42-Fc	150
4.1.7.4.5 Conjugate 43-Fc	150
4.1.7.5 Conjugates 44-Fc to 48-Fc	150
4.1.7.5.1 Conjugate 44-Fc	150
4.1.7.5.2 Conjugate 45-Fc	151
4.1.7.5.3 Conjugate 46-Fc	151
4.1.7.5.4 Conjugate 47-Fc	152
4.1.7.5.5 Conjugate 48-Fc	152
4.1.7.6 Co-conjugates 24-Fc/Pt to 28-Fc/Pt	152
4.1.7.6.1 Co-conjugate 24-Fc/Pt	152
4.1.7.6.2 Co-conjugate 25-Fc/Pt	153
4.1.7.6.3 Co-conjugate 26-Fc/Pt	153
4.1.7.6.4 Co-conjugate 27-Fc/Pt	153
4.1.7.6.5 Co-conjugate 28-Fc/Pt	154
4.1.7.7 Co-conjugates 29-Fc/Pt to 48-Fc/Pt	154
4.1.7.7.1 Co-conjugate 29-Fc/Pt	154
4.1.7.7.2 Co-conjugate 30-Fc/Pt	154
4.1.7.7.3 Co-conjugate 31-Fc/Pt	155
4.1.7.7.4 Co-conjugate 32-Fc/Pt	155

4.1.7.7.5 Co-conjugate 33-Fc/Pt	155
4.1.7.7.6 Co-conjugate 34-Fc/Pt	156
4.1.7.7.7 Co-conjugate 35-Fc/Pt	156
4.1.7.7.8 Co-conjugate 36-Fc/Pt	156
4.1.7.7.9 Co-conjugate 37-Fc/Pt	157
4.1.7.7.10 Co-conjugate 38-Fc/Pt	157
4.1.7.7.11 Co-conjugate 39-Fc/Pt	157
4.1.7.7.12 Co-conjugate 40-Fc/Pt	158
4.1.7.7.13 Co-conjugate 41-Fc/Pt	158
4.1.7.7.14 Co-conjugate 42-Fc/Pt	158
4.1.7.7.15 Co-conjugate 43-Fc/Pt	158
4.1.7.7.16 Co-conjugate 44-Fc/Pt	159
4.1.7.7.17 Co-conjugate 45-Fc/Pt	159
4.1.7.7.18 Co-conjugate 46-Fc/Pt	159
4.1.7.7.19 Co-conjugate 47-Fc/Pt	160
4.1.7.7.20 Co-conjugate 48-Fc/Pt	160
CHAPTER 5: CONCLUSION	161
REFERENCES	164
APENDIX	175

LIST OF FIGURES

Figure 2.1: Structures of nitrogen mustards currently used in therapy.....	11
Figure 2.2: Cisplatin, Carboplatin and Oxaliplatin.....	13
Figure 2.3: Structures of platinum antitumor agents that have gained regional approval for use as anticancer drugs.....	14
Figure 2.4: <i>Cis</i> - and <i>trans</i> -platinum(II) isomers.....	15
Figure 2.5: Speciation of <i>cisplatin</i> in aqueous solution.....	16
Figure 2.6: General crosslinked forms of DNA from reaction with <i>cisplatin</i>	17
Figure 2.7: Schematic reaction of <i>cisplatin</i> with DNA.....	18
Figure 2.8: Structure of dactinomycin.....	20
Figure 2.9: Structures of anthracyclines.....	21
Figure 2.10: Structure of mitomycin C.....	22
Figure 2.11: Structures of podophyllotoxin and epipodophyllotoxins.....	23
Figure 2.12: Structure of mitoxantrone.....	24
Figure 2.13: Structures of folic acid, aminopterin and MTX.....	25
Figure 2.14: General structure of a macromolecular carrier as proposed by Ringsdorf.....	33
Figure 2.15: Flow chart describing drug conjugate pharmacokinetics.....	36
Figure 3.1: Structure of polyamide-type carrier.....	38
Figure 3.2: Models of platinum conjugates.....	71
Appendix 1a: ¹ H NMR of M1	176
Appendix 1b: ¹ H NMR of M2	177
Appendix 1c: ¹ H NMR of M4	178
Appendix 1d: ¹ H NMR of M6	179
Appendix 1e: ¹ H NMR of M1	180
Appendix 1f: ¹ H NMR of polyaspartamide 1	181
Appendix 1g: ¹ H NMR of polyaspartamide 2	182
Appendix 1h: ¹ H NMR of polyaspartamide 3	183

Appendix 1i: ^1H NMR of polyaspartamide 4	184
Appendix 1j: ^1H NMR of polyaspartamide 5	185
Appendix 1k: ^1H NMR of polyaspartamide 8	186
Appendix 1l: ^1H NMR of polyaspartamide 9	187
Appendix 1m: ^1H NMR of polyaspartamide 10	188
Appendix 1n: ^1H NMR of polyaspartamide 12	189
Appendix 1o: ^1H NMR of polyaspartamide 13	190
Appendix 1p: ^1H NMR of polyaspartamide 18	191
Appendix 1q: ^1H NMR of polyaspartamide 19	192
Appendix 1r: ^1H NMR of polyaspartamide 20	193
Appendix 1s: ^1H NMR of polyaspartamide 21	194
Appendix 1t: ^1H NMR of polyaspartamide 22	195
Appendix 1u: ^1H NMR of polyaspartamide 23	196

LIST OF SCHEMES

Scheme 2.1: Oxidation of ferrocene to the ferricenium cation.....	27
Scheme 2.2: Ferrocene in biological environments.....	28
Scheme 3.1: Synthesis of poly-DL-succinimide (PSI).....	40
Scheme 3.2: Synthesis of poly- α,β -DL-aspartamide.....	41
Scheme 3.3: Aminolytic ring opening of polysuccinimide.....	42
Scheme 3.4: Synthesis of <i>p</i> -aminobenzamidopropylamine (PABPA).....	44
Scheme 3.5: Copolyaspartamide containing primary amine-terminated side chains.....	47
Scheme 3.6: Copolyaspartamides containing primary amine (PDA) terminated side chains.....	49
Scheme 3.7: Synthesis of acryloylamidosalicylic acid (M2).....	53
Scheme 3.8: Synthesis of <i>p</i> -N-(4,8-diaza-octanoyl)salicylic acid (PASA-PDA) and <i>p</i> -N-(4,8-diaza-heptanoyl)salicylic acid (PASA-EDA).....	54
Scheme 3.9: Synthesis of copolyaspartamides 8 to 12	57
Scheme 3.10: Synthesis of copolyaspartamides 13 to 17	58
Scheme 3.11: Synthesis of copolyaspartamides 18 to 22	63
Scheme 3.12: Synthesis of copolyaspartamide 23	64
Scheme 3.13: Synthesis of platination agents.....	73
Scheme 3.14: Synthesis of conjugate 6-Pt	74
Scheme 3.15: Synthesis of conjugate 7-Pt	75
Scheme 3.16: Synthesis of conjugates 1-Pt to 5-Pt	76
Scheme 3.17: Synthesis of conjugates 8-Pt to 12-Pt	79
Scheme 3.18: Synthesis of conjugates 13-Pt to 17-Pt	79
Scheme 3.19: Synthesis of conjugates 18-Pt to 22-Pt	81
Scheme 3.20: Synthesis of conjugate 23-Pt	82
Scheme 3.21: Synthesis of 4-Ferrocenylbutamidopropylamine.....	85
Scheme 3.22: Synthesis of conjugates 24-Fc – 28-Fc	88
Scheme 3.23: Synthesis of co-conjugates 24-Fc/Pt to 28-Fc/Pt	92
Scheme 3.24: Synthesis of conjugates 29-Fc to 33-Fc	93

Scheme 3.25: Synthesis of conjugates 34-Fc to 38-Fc	94
Scheme 3.26: Synthesis of co-conjugates 29-Fc/Pt to 33-Fc/Pt	100
Scheme 3.27: Synthesis of co-conjugates 34-Fc/Pt to 38-Fc/Pt	100
Scheme 3.28: Synthesis of conjugates 39-Fc to 43-Fc	102
Scheme 3.29: Synthesis of co-conjugates 39-Fc/Pt to 43-Fc/Pt	106
Scheme 3.30: Synthesis of acryloylaspartic acid (M7).....	107
Scheme 3.31: Synthesis of ACRYASP-PDA (M8).....	107
Scheme 3.32: Synthesis of conjugates 44-Fc to 48-Fc	109
Scheme 3.33: Synthesis of co-conjugates 44-Fc/Pt to 48-Fc/Pt	111

LIST OF TABLES

Table 3.1: Summary of experimental data for M1	44
Table 3.2: ¹ H NMR characterization of M1	45
Table 3.3: Summary of experimental data polyaspartamide bearing an amine side chain as the drug anchoring site.....	50
Table 3.4: ¹ H NMR of copolyaspartamides 1 to 7	51
Table 3.5: Summary of experimental data for M2 , M3 and M4	54
Table 3.6: ¹ H NMR analysis of M2 , M3 and M4	55
Table 3.7: Summary of experimental data for polyaspartamide bearing a carboxylatohydroxylato-terminated side chain as the drug anchoring site.....	59
Table 3.8: ¹ H NMR analysis of copolyaspartamides 8 to 17	60
Table 3.9: Summary of experimental data Polyaspartamide Bearing a dihydroxylato-ligand Side Chain as Drug Anchoring Site.....	66
Table 3.10 : ¹ H NMR of copolyaspartamides 18 to 23	67
Table 3.11: Preparative and analytical data of conjugates 1-Pt to 7-Pt	77
Table 3.12: Preparative and analytical data of platinum-conjugates 8-Pt to 17-Pt	80
Table 3.13: Preparative and analytical data of platinum-conjugates 18-Pt to 23-Pt	83
Table 3.14: Summary of experimental data for M6	85
Table 3.15: ¹ H NMR of M5 and M6	86
Table 3.16: Preparative data for conjugates 24-Fc – 28-Fc and co-conjugates 24-Fc/Pt – 28-Fc/Pt	89
Table 3.17: Analytical data for conjugates 24-Fc to 28-Fc and co-conjugates 24-Fc/Pt to 28-Fc/Pt	90
Table 3.18: Preparative data for conjugates 29-Fc to 33-Fc and co-conjugates 29-Fc/Pt to 33-Fc/Pt	95
Table 3.19: Preparative data for conjugates 34-Fc to 38-Fc and co-conjugates 34-Fc/Pt to 38-Fc/Pt	96

Table 3.20: Analytical data for conjugates 29-Fc to 33-Fc and co-conjugates 29-Fc/Pt to 33-Fc/Pt	97
Table 3.21: Analytical data for conjugates 34-Fc to 38-Fc and co-conjugates 34-Fc/Pt to 38-Fc/Pt	98
Table 3.22: Preparative data for conjugates 39-Fc to 43-Fc and co-conjugates 39-Fc/Pt to 43-Fc/Pt	103
Table 3.23: Analytical data for conjugates 39-Fc to 43-Fc and co-conjugates 39-Fc/Pt to 43-Fc/Pt	104
Table 3.25: Summary of experimental data for M6 and M7	107
Table 3.26: ¹ H NMR analysis of M7 and M8	108
Table 3.27: Preparative data for conjugates 44-Fc to 48-Fc and co-conjugates 44-Fc/Pt to 48-Fc/Pt	112
Table 3.28: Analytical data for conjugates 44-Fc to 48-Fc and co-conjugates 44-Fc/Pt to 48-Fc/Pt	113
Table 3.29: Antiproliferative activity of conjugates and co-conjugates.....	115

LIST OF ABBREVIATIONS

AEP	2-(2-aminoethyl)pyridine
AF	Activity factor
Asp	Aspartic acid
Calcd	Calculated
d	Day(s)
Dach-Pt	<i>trans</i> -1,2-diaminocyclohexanediaqua platinum(II) dinitrate
DCC	N,N'-dicyclohexylcarbodiimide
DEEA	2-(diethylamino)ethylamine
DHFR	Dihydrofolate reductase
DMEA	2-(dimethylamino)ethylamine
DMF	N,N'-dimethylformamide
DMP	3-(dimethylamino)propylamine
DMSO	Dimethylsulfoxide
DNA	deoxyribonucleic acid
Dopamine	3-Hydroxytyramine (Hydrochloride)
EAB	Ethyl 4-aminobenzoate
EDA	1,2-Diaminoethane
EPR	Enhanced permeability and retention
Et ₃ N	Triethylamine
IC ₅₀	Concentration required to inhibit 50% of the cells
ICP-OES	Inductively coupled plasma-optical emission spectroscopy
IMRT	Intensity modulated radiation therapy
MEA	2-(methoxy)ethylamine
MTD	Maximum tolerated dose
MTT	(3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide)
MTX	Methotrexate
NMR	Nuclear magnetic resonance
PDA	1,3-diaminopropane
1,2-PDA	1,2-diaminopropane

PEG	Poly(ethylene glycol)
ppm	Parts per million
PSI	Polysuccinimide
RNA	Ribonucleic acid
RT	Room temperature
WHO	World health organization