

## CONTENTS

	Page
ACKNOWLEDGEMENTS.....	i
PREFACE .....	iv
AIMS OF THIS STUDY .....	ix
CONTENT PAGES .....	xii
LIST OF TABLES .....	xxii
LIST OF FIGURES.....	xxvi
CHAPTER ONE: THE BIOCHEMISTRY OF THYROID HORMONES.....	1
1.1 Iodines and Iodoproteins .....	1
1.2 Thyroid Hormone Synthesis .....	4
1.2.1 Iodide Transport .....	5
1.2.2 Mechanisms of Iodination .....	8
1.2.3 Coupling Mechanisms of Iodo- -thyronine Formation.....	10
1.2.4 Proteolysis of Thyroglobulin and Hormone Release .....	12
CHAPTER TWO: EPIDEMIOLOGY OF ENDEMIC GOITRE AND ENDEMIC CRETINISM .....	17
2.1 Historical Background .....	18
2.2 Classification of Goitre .....	26
2.3 Prevalence .....	27
2.3.1 Europe .....	28
2.3.2 Latin America .....	30
2.3.3 Africa .....	33

2.3.4	Asia .....	35
2.4	Clinical Implications .....	45
<b>CHAPTER THREE: THE ROLE OF THYROXINE-BINDING PROTEINS IN HORMONE DELIVERY .....</b>		<b>56</b>
3.1	Thyroxine-Binding Proteins .....	56
3.1.1	Thyroxine-Binding Globulin (TBG) .....	57
3.1.2	Thyroxine-Binding Pre-Albumin (TBPA) ...	59
3.1.3	Human Serum Albumin (HSA) .....	61
3.2	Mechanisms of Hormone Delivery .....	62
3.2.1	Kinetics of Hormone Delivery to Target Tissues .....	64
3.2.1.1	Tait-Burstein Model .....	64
3.2.1.2	Keller-Richardson-Yates Model .....	65
3.2.1.3	Robbins-Rall Model .....	66
3.2.1.4	Pardridge Model .....	67
3.2.1.5	Ekins-Edwards-Newman Model .....	68
3.2.2	Physiological Implications.....	71
<b>CHAPTER FOUR: THYROID HORMONES IN FETAL DEVELOPMENT</b>		<b>76</b>
4.1	Introduction .....	76
4.2	Effects of Thyroid Hormones on Growth .....	79
4.3	Effects of Thyroid Hormones on Differentiation....	81
4.3.1	Skeletal Growth .....	82
4.3.2	Pulmonary Development .....	82
4.3.3	Palatal Development .....	85

<b>CHAPTER FIVE: THYROID HORMONES AND BRAIN DEVELOPMENT..</b>	<b>88</b>
5.1 Introduction .....	88
5.2 Quantitative Growth of Human Brain .....	91
5.3 Thyroid Hormone Receptor in Brain Development.....	93
5.4 Thyroid Hormones in Brain Differentiation .....	97
5.4.1 Cell Proliferation and Cell Migration...	99
5.4.2 Neurite Growth, Synaptogenesis. and Myelination .....	101
<b>CHAPTER SIX: PLACENTAL TRANSPORT OF THYROID HORMONES</b>	<b>104</b>
6.1 Placenta .....	104
6.1.1 Phylogeny .....	105
6.1.2 Descriptive Definition of the Human Placenta .....	107
6.1.3 Morphological Development and Maturation of the Human Placenta .....	107
6.1.4 Immunologicalm Implicationsm of Placentation.....	111
6.1.5 Placental Barrier .....	111
6.1.6 Placenta as a Filter .....	113
6.2 Placental Synthesis .....	115
6.3 Placental Transfer .....	117
6.3.1 Placental Transfer of Iodide .....	118
6.3.2 Placental Transfer of Thyroid Hormones..	121
6.4 Placental Deiodinase Activity .....	127

CHAPTER SEVEN: MATERIALS AND METHODS.....	133
7.1 Experimental Animals .....	133
7.2 Chemicals .....	133
7.3 Measurement of Radioactivity .....	135
7.3.1 Counting Instrument .....	135
7.3.2 Counting Time .....	135
7.3.3 Background Radioactivity .....	136
7.4 Preservation of Samples .....	136
7.5 Experiments .....	137
7.5.1 Uptake of <sup>125</sup> I-Thyroxine and <sup>131</sup> Iodide...	137
7.5.2 <i>In Vitro</i> Assay of Deiodinase Activity in Fetal and Placental Tissue .....	138
7.5.2.1 Purification of Stock <sup>125</sup> I- Thyroxine.....	138
7.5.2.2 Preparation of Buffer .....	139
7.5.2.3 Preparation of Dithiothreitol (DTT) Solution .....	140
7.5.2.4 Procedure .....	140
7.5.3 Measurement of Protein Content .....	141
7.5.4 Calculations .....	142
7.5.4.1 Uptake of <sup>125</sup> I-Thyroxine and <sup>131</sup> Iodide.....	142
7.5.4.2 <i>In Vitro</i> Assay of Deiodinase Activity.....	142
7.5.4.3 Protein Content .....	143
7.5.5 Statistical Methods .....	144

CHAPTER EIGHT: RESULTS .....	145
8.1 Feto-Placental Growth .....	145
8.1.1 Fetal Weight and Protein Content .....	145
8.1.2 Fetal Brain Weight and Protein Content..	146
8.1.3 Fetal Liver Weight and Protein Content..	151
8.1.4 Placental Weight and Protein Content....	151
8.1.5 Protein Content in Amniotic Fluid .....	155
8.1.6 Relative Growth of Feto-Placental Tissue.....	159
8.1.7 Summary .....	162
8.2 Uptake of <sup>125</sup> I-Thyroxine and <sup>131</sup> Iodide by Entire Tissue .....	162
8.2.1 Uptake of <sup>125</sup> I-Thyroxine .....	163
8.2.1.1 Uptake of <sup>125</sup> I-Thyroxine by Fetuses	163
8.2.1.2 Uptake of <sup>125</sup> I-Thyroxine by Fetal Brains.....	163
8.2.1.3 Uptake of <sup>125</sup> I-Thyroxine by Fetal Livers. ....	166
8.2.1.4 Uptake of <sup>125</sup> I-Thyroxine by Placentae and Amniotic Fluid.....	166
8.2.1.5 Summary .....	170
8.2.2 Uptake of <sup>131</sup> Iodide .....	170
8.2.2.1 Uptake of <sup>131</sup> Iodide by Fetuses ....	170
8.2.2.2 Uptake of <sup>131</sup> Iodide by Fetal Brains.....	173
8.2.2.3 Uptake of <sup>131</sup> Iodide by Fetal Livers.....	173
8.2.2.4 Uptake of <sup>131</sup> Iodide by Placentae and Amniotic Fluid .....	176
8.2.2.5 Summary .....	176

8.3	Uptake of $^{125}\text{I}$ -Thyroxine and $^{125}\text{I}$ Iodide Fractions by Tissues.....	179
8.3.1	Uptake of $^{125}\text{I}$ -Thyroxine in TCA-Soluble and Insoluble Fractions .....	181
8.3.1.1	Uptake of $^{125}\text{I}$ -Thyroxine in TCA- Soluble and Insoluble Fractions by Fetuses.....	181
8.3.1.2	Uptake of $^{125}\text{I}$ -Thyroxine in TCA- Soluble and Insoluble Fractions by Fetal Brains.....	183
8.3.1.3	Uptake of $^{125}\text{I}$ -Thyroxine in TCA- Soluble and Insoluble Fractions by Fetal Livers .....	183
8.3.1.4	Uptake of $^{125}\text{I}$ -Thyroxine in TCA- Soluble and Insoluble Fractions by Placentae .....	186
8.3.1.5	Uptake of $^{125}\text{I}$ -Thyroxine in TCA- Soluble and Insoluble Fractions by Amniotic Fluid.....	186
8.3.1.6	Summary .....	189
8.3.2	Uptake of TCA-Soluble $^{131}\text{I}$ Iodide Fraction by Tissues .....	189
8.3.2.1	Uptake of TCA-Soluble $^{131}\text{I}$ Iodide by Fetuses .....	191
8.3.2.2	Uptake of TCA-Soluble $^{131}\text{I}$ Iodide by Fetal Brains .....	191
8.3.2.3	Uptake of TCA-Soluble $^{131}\text{I}$ Iodide by Fetal Livers .....	194
8.3.2.4	Uptake of TCA-Soluble $^{131}\text{I}$ Iodide by Placentae .....	194
8.3.2.5	Uptake of TCA-Soluble $^{131}\text{I}$ Iodide by Amniotic Fluid .....	197
8.3.2.6	Summary .....	197

8.4	<i>In Vitro</i> Outer-Ring Monodeiodination of $^{125}\text{I}$ -Thyroxine by Fetal and Placental Tissue.....	200
8.4.1	5'- Monodeiodinase Activity in Fetal Tissues .....	200
8.4.2	5'- Monodeiodinase Activity in Placental Tissues .....	202
8.4.3	Summary .....	202
8.5	Uptake Ratio of TCA-Soluble : Insoluble Fractions of $^{125}\text{I}$ -Thyroxine in Tissues.....	204
8.5.1	Uptake Ratio of TCA-Soluble : Insoluble Fractions of $^{125}\text{I}$ -Thyroxine in Fetuses.....	204
8.5.2	Uptake Ratio of TCA-Soluble : Insoluble Fractions of $^{125}\text{I}$ -Thyroxine in Fetal Brains .....	204
8.5.3	Uptake Ratio of TCA-Soluble : Insoluble Fractions of $^{125}\text{I}$ -Thyroxine in Fetal Livers.....	206
8.5.4	Uptake Ratio of TCA-Soluble : Insoluble Fractions of $^{125}\text{I}$ -Thyroxine in Placentae.....	206
8.5.5	Uptake Ratio of TCA-Soluble : Insoluble Fractions of $^{125}\text{I}$ -Thyroxine in Amniotic Fluid .....	210
8.5.6	Summary .....	210
8.6	Uptake Ratios of TCA-Soluble $^{125}\text{I}$ Iodide : $^{131}\text{I}$ Iodide in Tissues .....	213
8.6.1	Uptake Ratios of TCA-Soluble $^{125}\text{I}$ Iodide : $^{131}\text{I}$ Iodide in Fetuses .....	214
8.6.2	Uptake Ratios of TCA-Soluble $^{125}\text{I}$ Iodide : $^{131}\text{I}$ Iodide in Fetal Brains .....	214
8.6.3	Uptake Ratios of TCA-Soluble $^{125}\text{I}$ Iodide : $^{131}\text{I}$ Iodide in Fetal Livers .....	216
8.6.4	Uptake Ratios of TCA-Soluble $^{125}\text{I}$ Iodide : $^{131}\text{I}$ Iodide in Placentae.....	216

8.6.5	Uptake Ratios of TCA-Soluble <sup>125</sup> Iodide : <sup>131</sup> Iodide in Amniotic Fluid .....	220
8.6.6	Summary .....	220
8.7	Uptake Ratios of Fetal Brain : Fetal Liver in the Mother and the Fetus .....	223
8.7.1	Brain : Liver Uptake Ratios of <sup>125</sup> Iodide Released from Injected <sup>125</sup> I-Thyroxine in the Mother and the Fetus .....	223
8.7.2	Brain : Liver Uptake Ratios of <sup>131</sup> Iodide in the Mother and the Fetus .....	224
8.7.3	Summary .....	227
<b>CHAPTER NINE: DISCUSSION .....</b>		<b>228</b>
9.1	The Animal Model in Research in Human Thyroid Physiology .....	229
9.1.1	Introduction .....	229
9.1.2	Relevance of Animal Models to the Study of Thyroid Hormone Deficiency and Human Brain Development.....	230
9.2	Critical Review of Methodology .....	234
9.3	Fetal Growth .....	239
9.4	Placental Uptake of <sup>125</sup> I-Thyroxine .....	240
9.5	Placental Uptake of Iodide .....	245
9.6	The Role of Amniotic Fluid .....	246
9.7	The Fate of Injected <sup>125</sup> I-Thyroxine .....	249
9.8	The Significance of 5'- Monodeiodinase Activity in Placentae and Fetuses .....	253
9.9	Metabolism of <sup>125</sup> I-Thyroxine in the Tissues.....	258
9.10	Conclusions .....	263
9.11	Post Script .....	265



<b>APPENDICES.....</b>	<b>269</b>
Appendix I.    Purification of <sup>125</sup> I-Thyroxine for <i>In Vitro</i> Deiodinase Assay .....	269
Appendix II.   Experiment to Estimate Optimum pH for Deiodinase Assay .....	272
 <b>REFERENCES.....</b>	 <b>276</b>
Chapter One .....	276
Chapter Two .....	282
Chapter Three .....	292
Chapter Four .....	297
Chapter Five .....	362
Chapter Six .....	311
Chapter Nine .....	321

## LIST OF TABLES

	Page
Table 2.1	Clinical features of neurological and myxoedematous cretinism..... 23
Table 2.2	Goitre prevalence in Latin America..... 31
Table 2.3	Prevalence of iodine deficiency disorders (IDD) in developing countries and number of persons at risk..... 36
Table 2.4	Estimate population at risk and prevalence of endemic goitre in eight countries of the WHO South-East Asian region..... 38
Table 2.5	Estimated iodine deficiency disorders (IDD) prevalence in eight IDD affected countries of the WHO South-East Asian region..... 39
Table 2.6	Estimated number and rate of stillbirths and neonatal deaths and their sum (total late reproductive loss) attributable to iodine deficiency in eight countries of the WHO South-East Asian region..... 40
Table 3.1	Some chemical properties of human thyroxine-binding proteins..... 58
Table 8.1	Weights of entire fetuses and their protein content at different periods of gestation..... 147
Table 8.2	Weights of entire fetal brains and their protein content at different periods of gestation..... 149
Table 8.3	Weights of entire fetal livers and their protein content at different periods of gestation..... 152
Table 8.4	Weights of entire placentae and their protein content at different periods of gestation..... 154

Table 8.5	Protein content of amniotic fluid at different periods of gestation.....	157
Table 8.6	Protein content of different maternal tissues taken from all sampling categories.....	160
Table 8.7	Weight ratios between fetus, placenta fetal brain and liver.....	161
Table 8.8	Total uptake of $^{125}\text{I}$ -thyroxine radioactivity by entire fetuses at different periods of gestation.....	164
Table 8.9	Total uptake of $^{125}\text{I}$ -thyroxine radioactivity by entire fetal brains at different periods of gestation.....	165
Table 8.10	Total uptake of $^{125}\text{I}$ -thyroxine radioactivity by entire fetal livers at different periods of gestation.....	167
Table 8.11	Total uptake of $^{125}\text{I}$ -thyroxine radioactivity by entire placentae at different periods of gestation.....	168
Table 8.12	Total uptake of $^{125}\text{I}$ -thyroxine radioactivity by amniotic fluids at different periods of gestation.....	169
Table 8.13	Total uptake of $^{125}\text{I}$ -thyroxine radioactivity by different maternal tissues taken from all sampling categories.....	171
Table 8.14	Total uptake of $^{131}\text{I}$ iodide radioactivity by entire fetuses at different periods of gestation.....	172
Table 8.15	Total uptake of $^{131}\text{I}$ iodide radioactivity by entire fetal brains at different periods of gestation.....	174
Table 8.16	Total uptake of $^{131}\text{I}$ iodide radioactivity by entire fetal livers at different periods of gestation.....	175
Table 8.17	Total uptake of $^{131}\text{I}$ iodide radioactivity by entire placentae at different periods of gestation.....	177

Table 8.18	Total uptake of $^{131}$ Iodide radioactivity by amniotic fluids at different periods of gestation.....	178
Table 8.19	Total uptake of $^{131}$ Iodide radioactivity by different maternal tissues taken from all sampling categories.....	180
Table 8.20	The uptake of $^{125}$ I-thyroxine by fetuses at different periods of gestation measured in TCA-soluble and insoluble fractions.....	182
Table 8.21	The uptake of $^{125}$ I-thyroxine by fetal brains at different periods of gestation measured in TCA-soluble and insoluble fractions.....	184
Table 8.22	The uptake of $^{125}$ I-thyroxine by fetal livers at different periods of gestation measured in TCA-soluble and insoluble fractions.....	185
Table 8.23	The uptake of $^{125}$ I-thyroxine by placentae at different periods of gestation measured in TCA-soluble and insoluble fractions.....	187
Table 8.24	The uptake of $^{125}$ I-thyroxine by amniotic fluid at different periods of gestation measured in TCA-soluble and insoluble fractions.....	188
Table 8.25	The uptake of $^{125}$ I-thyroxine different maternal tissues taken from all sampling categories in TCA-soluble and insoluble fractions.....	190
Table 8.26	The uptake of $^{131}$ Iodide by fetuses at different periods of gestation measured in TCA soluble fraction.....	192
Table 8.27	The uptake of $^{131}$ Iodide by fetal brains at different periods of gestation measured in TCA soluble fraction.....	193
Table 8.28	The uptake of $^{131}$ Iodide by fetal livers at different periods of gestation measured in TCA soluble fraction.....	195

Table 8.29	The uptake of $^{131}\text{I}$ Iodide by placentae at different periods of gestation measured in TCA soluble fraction.....	196
Table 8.30	The uptake of $^{131}\text{I}$ Iodide by amniotic fluids at different periods of gestation measured in TCA soluble fraction.....	198
Table 8.31	The uptake of $^{131}\text{I}$ Iodide maternal tissues taken from all sampling categories in TCA soluble fraction.....	199
Table 8.32	<i>In vitro</i> outer-ring deiodination of $^{125}\text{I}$ -thyroxine in fetal tissue homogenates prepared from samples taken at different periods of gestation.....	201
Table 8.33	<i>In vitro</i> outer-ring deiodination of $^{125}\text{I}$ -thyroxine in placental tissue homogenates prepared from samples taken at different periods of gestation.....	203
Table 8.34	Uptake ratios of soluble and insoluble fractions of $^{125}\text{I}$ Iodide by fetuses in relation to maternal plasma at different periods of gestation.....	205
Table 8.35	Uptake ratios of soluble and insoluble fractions of $^{125}\text{I}$ Iodide by fetal brains in relation to maternal plasma at different periods of gestation.....	207
Table 8.36	Uptake ratios of soluble and insoluble fractions of $^{125}\text{I}$ Iodide by fetal livers in relation to maternal plasma at different periods of gestation.....	208
Table 8.37	Uptake ratios of soluble and insoluble fractions of $^{125}\text{I}$ Iodide by placentae in relation to maternal plasma at different periods of gestation.....	209
Table 8.38	Uptake ratios of soluble and insoluble fractions of $^{125}\text{I}$ Iodide by amniotic fluid samples in relation to maternal plasma at different periods of gestation.....	211
Table 8.39	Uptake ratios of soluble and insoluble fractions of $^{125}\text{I}$ Iodide by maternal tissue in relation to its plasma at different periods of gestation.....	212

Table 8.40	Uptake ratio of TCA-soluble <sup>125</sup> Iodide and <sup>131</sup> Iodide in fetuses in relation to maternal plasma at different period of gestation.....	215
Table 8.41	Uptake ratio of TCA-soluble <sup>125</sup> Iodide and <sup>131</sup> Iodide in fetal brains in relation to maternal plasma at different periods of gestation.....	217
Table 8.42	Uptake ratio of TCA-soluble <sup>125</sup> Iodide and <sup>131</sup> Iodide in fetal livers in relation to maternal plasma at different periods of gestation.....	218
Table 8.43	Uptake ratio of TCA-soluble <sup>125</sup> Iodide and <sup>131</sup> Iodide in placentae in relation to maternal plasma at different periods of gestation.....	219
Table 8.44	Uptake ratio of TCA-soluble <sup>125</sup> Iodide and <sup>131</sup> Iodide in amniotic fluid samples in relation to maternal plasma at different periods of gestation.....	221
Table 8.45	Uptake ratio of TCA-soluble <sup>125</sup> Iodide and <sup>131</sup> Iodide in maternal tissues in relation to its plasma at different periods of gestation.....	222
Table 8.46	Brain/liver <sup>125</sup> I-thyroxine uptake ratios of fetuses of different periods of gestation and of maternal rats of all sampling categories.....	225
Table 8.47	Brain/liver <sup>131</sup> Iodide uptake ratios of fetuses of different periods of gestation and of maternal rats of all sampling categories.....	226

LIST OF FIGURES

	Page
Figure 1 The changes in the rate of weight gain and rate of increase in protein content of fetuses during the period of gestation under study.....	148
Figure 2 The changes in the rate of weight gain and rate of increase in protein content of fetal brains during the period of gestation under study.....	150
Figure 3 The changes in the rate of weight gain and rate of increase in protein content of fetal livers during the period of gestation under study.....	153
Figure 4 The changes in the rate of weight gain and rate of increase in protein content of placentae during the period of gestation under study.....	156
Figure 5 The changes in the rate of increase in protein content of amniotic fluids during the period of gestation under study.....	158