

Immunohistochemical Expression of Human Epidermal Growth factor Receptor- 2 (HER2) In Gastric Cancer

Thesis

Submitted for partial fulfillment of Master Degree
in Pathology

By

Lobna Abd El Fattah Mohammad Hasan Attia

M.B., B.Ch.

Under The Supervision Of

Prof. Dr. Zeinab Abd El Rahman Kamar

Professor of Pathology

Faculty of Medicine – Ain Shams University

Prof. Dr. Hamed Mohammad Gamal El Din

Professor of Pathology

Faculty of Medicine – Ain Shams University

Prof. Dr. Naglaa Samir Ahmad

Professor of Pathology

Faculty of Medicine – Ain Shams University

**Faculty of Medicine
Ain Shams University
2016**

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قالوا

لسبحناك لا علم لنا
إلا ما علمتنا إنك أنت
العليم العظيم

صدق الله العظيم

سورة البقرة الآية: ٣٢



Acknowledgement

*First of all, Thanks are all to **Allah**, by the grace of whom, this work was possible.*

I am greatly indebted to all my supervisors for their great effort, advice and support throughout the preparation of this work,

*I would like to express my unlimited gratitude and appreciation to my **Prof. Dr. Zeinab Abd El Rahman Kamar**, Professor of Pathology, Faculty of Medicine, Ain Shams University, for her valuable comments, continuous guidance and tremendous effort during this work,*

*My deep appreciation and thanks to **Prof.Dr. Hamed Mohammad Gamal El Din**, Professor of Pathology, Faculty of Medicine, Ain Shams University, for his valuable advice, encouragement and support throughout this work,*

*My greatest thanks and gratitude to **Prof.Dr. Naglaa Samir Ahmad**, Professor of Pathology, Faculty of Medicine, Ain Shams University, for her supervision, great care and helpful ideas during this work,*

Finally, I am truly grateful to my family and colleagues who helped and supported me to complete this work,

 **Lobna Abd El Fattah**

Table of Contents

Subject	Page No.
List of Abbreviations.....	i
List of Tables.....	iv
List of Figures	vii
Introduction	1
Aim of the Work.....	4
Review of Literature	
Anatomy and Histology of the Stomach	5
Gastric Cancer	28
Prognosis and Treatment of Gastric Carcinoma	78
Human Epidermal Growth Factor Receptor2 (HER2)....	88
Material and Methods.....	97
Results.....	103
Discussion	142
Summary	161
Conclusion.....	166
Recommendations	168
References	170
Arabic Summary	—

List of Abbreviations

<i>Abb.</i>	<i>Full term</i>
AFP	Alpha Fetoprotein
AG	Atrophic gastritis
AJCC	American Joint Committee on Cancer
ASC	Adenosquamous cell carcinoma
ATM	Ataxia telangiectasia mutated
BE	Barrett's esophagus
CagA	Cytotoxin associated gene A
CD	Cluster of Differentiation
CDH1	E-cadherin gene
CEA	Carcinoembryonic antigen
CHK2	Checkpoint kinase 2
CIMP	CpG island methylator phenotype
CIN	Chromosomal instability
CISH	Chromogenic in-situ hybridization
CK	Cytokeratin
CLDNs	Claudins
DAB	Diaminobenzidine
DNA	Deoxyribonucleic acid
DPD	Dihydropyrimidine dehydrogenase
DSBs	Double-strand breaks
DU	Duodenal Ulcer
DupA	Duodenal ulcer promoting gene A
EAC	Esophageal adenocarcinoma
EBV	Epstein Barr Virus
EC	Endocrine cells
ECL	Enterochromaffin-like
EGF/R	Epidermal growth factor/receptor
EMMPRIN	Extracellular Matrix Metalloproteinase Inducer
ENCI	Egyptian National Cancer Institute
FAP	Familial adenomatous polyposis
FGF/R	Fibroblast growth factor/receptor

FHIT	Fragile histidine triad
FISH	Fluorescence in-situ hybridization
5-FU	5-fluorouracil
GAC	Gastric adenocarcinoma
GAP	GTPase-activating proteins
GC	Gastric Cancer
GHA	Gastric hepatoid adenocarcinoma
GIT	Gastrointestinal tract
GP	Gastric pits
GS	Genomically stable
GSCC	Gastric small cell carcinoma
GSK3β	Glycogen synthase Kinase3 β
GTP	Guanosine triphosphate
GU	Gastric Ulcer
H&E	Haematoxylin and Eosin
H. pylori	Helicobacter pylori
HAC	Hepatoid adenocarcinoma
HCG	Human Chorionic Gonadotrophin
HDGC	Hereditary diffuse gastric cancer
HER-2	Human epidermal growth factor receptor 2
HRP	Horseradish peroxidase
HS	Highly significant
IHC	Immunohistochemistry
IL	Interleukin
IM	intestinal metaplasia
JCGC	Japanese Classification of Gastric Carcinoma
LKB1	Liver kinase B1
LOH	Loss of heterozygosity
LTA	Lymphotoxin-alpha
MAPK	Mitogen-activated protein kinase
miRNAs	MicroRNAs
MLH1	Human MutL homolog
MMR	Mismatch repair
MPC	Micropapillary carcinoma
MSI	Microsatellite instable
MT	Mixed-type

mTOR	Mammalian target of rapamycin
NBI	Narrow band imaging
NECs	Neuroendocrine carcinomas
NETs	Neuroendocrine tumors
NF-κB	Nuclear Factor Kappa Beta
NLR	Neutrophil – lymphocyte ratio
NS	Non significant
OGJ	Oesophago-gastric junction
OLGA	Operative Link for Gastritis Assessment
OS	Overall survival
PA	Pernicious anaemia
PAH	Polycyclic Aromatic Hydrocarbons
PAS	Periodic acid–Schiff
PBS	Phosphate buffer solution
PCR	Polymerize Chain Reaction
PDECGF/R	Platelet-derived endothelial cell growth factor/receptor
PFS	Progression free survival
PG	Pepsinogen
PGC	Primary gastric choriocarcinoma
PI3K	Phosphatidylinositol 3-kinase
PIK3CA	phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha
PJS	Peutz-Jeghers syndrome
PKC	Protein kinase C
PLC	Phospholipase C
PLUNC	Palate, lung and nasal epithelium carcinoma-associated protein
PTAH	Phosphotungstic acid hematoxylin
PTEN	Phosphatase and tensin homolog
PUD	Peptic ulcer disease
RB	Retinoblastoma gene
RHOA	Ras homolog gene family, member A
RTKs	Receptor tyrosine kinases
SC	Squamous cell carcinoma
SCC	Small cell carcinoma

SES	Socioeconomic status
TCGA	The Cancer Genome Atlas
TGF-β/R	Transforming growth factor/receptor- β
TKs	Tyrosine kinases
TNF	Tumor Necrosis Factor
TNM	Tumor- Node-Metastasis
UICC	Union International Cancer Control
VacA	Vacuolating cytotoxin gene
VEGF	Vascular endothelial growth factor
WHO	World Health Organization

List of Tables

<i>Table No.</i>	<i>Title</i>	<i>Page No.</i>
Table (1):	Histologic grading of the gastric adenocarcinoma, intestinal type.....	30
Table (2):	Lymph node status of gastric adenocarcinoma (known in 149 cases).	31
Table (3):	Frequency of the most common genetic alterations found in gastric carcinomas	74
Table (4):	TNM classification of tumours of the stomach Carcinoma of the stomach	77
Table (5):	HER Family of Membrane Receptors	89
Table (6):	Personal data	103
Table (7):	Tumor site	104
Table (8):	Tumor size	105
Table (9):	Tumor type.....	105
Table (10):	Grade of tumor.....	106
Table (11):	pT status	107
Table (12):	pN status	107
Table (13):	LN involvement.....	108
Table (14):	Stage of tumor.....	108
Table (15):	Lymphovascular invasion:.....	109
Table (16):	Perineural invasion:	109
Table (17):	Her2 neu status.....	110
Table (18):	Her2 neu score	111
Table (19):	Relation between Her2 neu status and personal data	111

Table (20):	Relation between Her2 neu status and tumor site...	112
Table (21):	Relation between Her2 neu status and tumor size	112
Table (22):	Relation between Her2 neu status and tumor type.....	113
Table (23):	Relation between Her2 neu status and grade of tumor	114
Table (24):	Relation between Her2 neu status and PT status	115
Table (25):	Relation between Her2 neu status and PN status	115
Table (26):	Relation between Her2 status and number of LNs involved.....	116
Table (27):	Relation between Her2 neu status and stage of tumor	116
Table (28):	Relation between Her2 neu status and lympho- vascular invasion.....	117
Table (29):	Relation between Her2 neu status and perineural invasion.....	118

List of Figures

<i>Figure No.</i>	<i>Title</i>	<i>Page No.</i>
Figure (1):	Gross appearance of the gastric mucosal surface	7
Figure (2):	Semi-microscopic features of the gastric mucosa visualised by magnifying endoscopy with narrow band imaging	8
Figure (3):	The entire surface of the gastric mucosa is lined by simple columnar mucus secreting cells	11
Figure (4):	Full-thickness view of the body mucosa	12
Figure (5):	Fundic gland mucosa (neck region).....	15
Figure (6):	Fundic gland mucosa (deeper glands).	15
Figure (7):	Full thickness view of antral mucosa.....	17
Figure (8):	Pyloric glands	17
Figure (9):	Cardiac mucosa similar in morphology to the mucosa of the antral and pyloric regions	18
Figure (10):	Endocrine cells in the gastric mucosa.....	20
Figure (11):	Endocrine cells in the body mucosa, as demonstrated by chromogranin	21
Figure (12):	Proliferating cells in the gastric mucosa demonstrated by MIB1immunostaining	22
Figure (13):	Proliferating cells in the gastric mucosa demonstrated by MIB1immunostaining	22
Figure (14):	Illustration of the three layer muscle coat of the stomach.	26

Figure (15):	The interstitial cell of Cajal demonstrated by c-kit (CD117) immunostaining.....	26
Figure (16):	Japanese macroscopic classification of early gastric cancer	43
Figure (17):	Gross appearance of gastric adenocarcinoma: Borrmann type I.....	45
Figure (18):	Gross appearance of gastric adenocarcinoma: Borrmann type III	45
Figure (19):	Gross appearance of diffuse carcinoma of so-called ‘linitis plastica’ type.....	47
Figure (20):	Gastric adenocarcinoma of intestinal type with columnar mucin secreting cells	48
Figure (21):	Gastric adenocarcinoma accompanied by marked histiocytic reaction.....	48
Figure (22):	Diffuse type of gastric adenocarcinoma. An Indian File pattern of infiltration of the muscularis externa	49
Figure (23):	Gastric adenocarcinoma, tubular variant: the neoplasm is composed of irregularly shaped tubules lined by columnar neoplastic cells	51
Figure (24):	Gastric adenocarcinoma, papillary variant	52
Figure (25):	Mucinous adenocarcinoma. Clusters and scattered tumor cells floating in the abundant extracellular mucin pools	52
Figure (26):	Signet ring cell carcinoma: Signet ring carcinoma cells are predominantly at the superficial lamina propria.....	54
Figure (27):	Pseudo-signet ring cells: The cytoplasm of pseudo-signet ring cells are vacuolated and pale	54
Figure (28):	Pseudo-signet ring cells are confined within basement membrane and maintain intact acinar structure with reticulin stain	55

Figure (29): Poorly cohesive gastric carcinoma: The cells have a plasmacytoid appearance.....	55
Figure (30): Mixed gastric adenocarcinoma: the neoplasm is composed of tubular and poorly cohesive components	56
Figure (31): Adenosquamous gastric adenocarcinoma.....	58
Figure (32): Hepatoid gastric adenocarcinoma:.....	60
Figure (33): Gastric carcinoma with lymphoid stroma.....	63
Figure (34): Micropapillary variant of gastric adenocarcinoma.....	66
Figure (35): The HER signaling network and HER2 targeted therapy applied in breast cancer.....	89
Figure (36): The main transduction pathways regulated by the four HER family members.....	91
Figure (37): Tumor site (percentage).....	104
Figure (38): Tumor type (percentage)	105
Figure (39): Grade of tumor (percentage)	106
Figure (40): Her2 neu status.....	110
Figure (41): Relation between Her2 neu status and tumor type ...	113
Figure (42): Relation between Her2 neu status and grade of tumor.....	114
Figure (43): Relation between Her2 neu status and perineural invasion.....	118
Figure (44): Well differentiated intestinal type adenocarcinoma, grade I, showing malignant acini invading muscularis propria (H&E X100). Error! Bookmark not defined.	
Figure (45): A high power view showing the malignant acini variable in size and lined by malignant epithelial cells exhibiting hyperchromatic pleomorphic nuclei among the muscle bundles (H&E X200). Error! Bookmark not defined.	

- Figure (46):** Moderately differentiated intestinal type adenocarcinoma, grade II, with part of normal mucosa overlying the malignant neoplastic growth (H&E X40).**Error! Bookmark not defined.**
- Figure (47):** A higher magnification showing irregular intercommunicating malignant acini surrounded by desmoplastic reaction (H&E X100)..... **Error! Bookmark not defined.**
- Figure (48):** Moderately differentiated intestinal type adenocarcinoma, grade II, showing infiltration by malignant glands invading the muscle bundles and surrounded by an inflammatory reaction (H&E X100).**Error! Bookmark not defined.**
- Figure (49):** High power view showing the neoplastic epithelial cells arranged in an irregular acinar pattern and displaying nuclear atypia (H&E X200). **Error! Bookmark not defined.**
- Figure (50):** Higher magnification showing pleomorphic malignant epithelial cells exhibiting pleomorphic vesicular nuclei with prominent nucleoli (H&E X400).**Error! Bookmark not defined.**
- Figure (51):** Moderately differentiated intestinal type adenocarcinoma, grade II, showing clusters and glands of malignant epithelial cells penetrating serosa (H&E X200).**Error! Bookmark not defined.**
- Figure (52):** Poorly differentiated intestinal type adenocarcinoma, grade III, showing infiltration by malignant epithelial cells with occasional glandular differentiation, beneath a part of normal mucosa (H&E X100).**Error! Bookmark not defined.**
- Figure (53):** High power view showing the malignant epithelial cells displaying pleomorphism and nuclear atypia with hyperchromasia and prominent nucleoli (H&E X400).**Error! Bookmark not defined.**

- Figure (54):** Diffuse type adenocarcinoma, grade III, showing infiltration by sheets and cords of malignant epithelial cells with signet ring features, penetrating serosa (H&E X100).**Error! Bookmark not defined.**
- Figure (55):** Diffuse type adenocarcinoma (signet ring carcinoma), grade III, showing sheets of signet ring cells with intracytoplasmic mucin and flattened nuclei (H&E X200).**Error! Bookmark not defined.**
- Figure (56):** Signet ring cell carcinoma showing the signet ring cells among the extracellular pools of mucin (H&E X200).**Error! Bookmark not defined.**
- Figure (57):** Lymph node involvement by moderately differentiated intestinal type adenocarcinoma, grade II (H&E X40).**Error! Bookmark not defined.**
- Figure (58):** Perineural invasion by moderately differentiated intestinal type adenocarcinoma, grade II (H&E X100).**Error! Bookmark not defined.**
- Figure (59):** High power view showing the clusters and acini of malignant epithelial cells surrounding the nerve fibres (H&E X200).**Error! Bookmark not defined.**
- Figure (60):** Perineural invasion by malignant epithelial cells in a moderately differentiated intestinal type adenocarcinoma, grade II, infiltrating muscle bundles (H&E X200). **Error! Bookmark not defined.**
- Figure (61):** Perineural invasion by diffuse type adenocarcinoma..The neoplastic epithelial cells show signet ring features (H&E X200).**Error! Bookmark not defined.**
- Figure (62):** Lymphovascular invasion by diffuse type adenocarcinoma, with clusters of malignant epithelial cells in the vessels, exhibiting pleomorphism and hyperchromatism (H&E X400). **Error! Bookmark not defined.**