

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

﴿قَالُوا سُبْحَانَكَ لَا عِلْمَ لَنَا إِلَّا مَا

عَلَّمْتَنَا إِنَّكَ أَنْتَ الْعَلِيمُ الْحَكِيمُ﴾

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Detection of Aberrant Promoter Hypermethylation of Some Tumor Suppressor Genes in Non-Small Cell Lung Cancer in Egyptian Patients

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رسالة مقدمة للحصول على درجة الدكتوراه في فلسفة العلوم في الكيمياء
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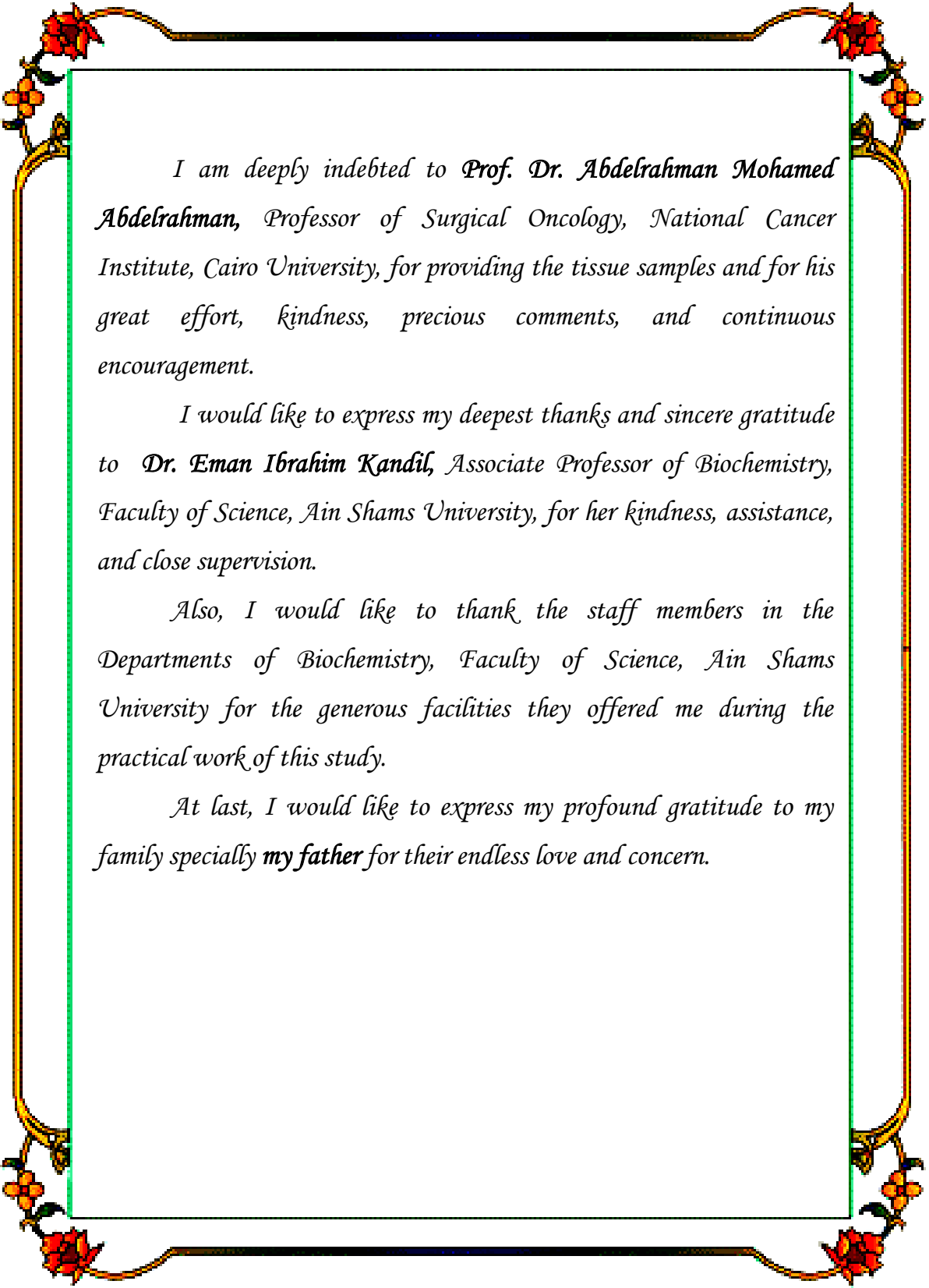
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Abstract

Riham Abdel Hamid Haroun. Detection of Aberrant Promoter Hypermethylation of Some Tumor Suppressor Genes in Non-Small Cell Lung Cancer in Egyptian Patients. Biochemistry Department, Faculty of Science, Ain Shams University.

Background: Methylation of tumor suppressor genes has been investigated in all kinds of cancer. Identification of tumor specific epigenetic alterations can be used as a molecular marker of malignancy, which can lead to better diagnosis, prognosis and therapy. Therefore, the aim of this study was to evaluate the association between gene hypermethylation and expression of fragile histidine triad (FHIT), glutathione S-transferase P1 (GSTP1) and p16 tumor suppressor genes and various clinicopathologic characteristics in primary non-small cell lung carcinomas (NSCLC).

Materials and Methods: The study included 28 primary non-small cell lung carcinomas, where an additional 28 tissue samples taken from apparently normal safety margin surrounding the tumor serving as controls. Methylation-specific polymerase chain reaction (MSP) was performed to analyze the methylation status of FHIT, GSTP1 and p16 while

their mRNA expression levels were measured using a real-time PCR assay with SYBR Green I.

Results: The methylation frequencies of the genes tested in NSCLC specimens were 53.57% for FHIT, 25% for GSTP1, and 0% for p16, and the risk of FHIT hypermethylation increased among patients with NSCLC by 2.88, while the risk of GSTP1 hypermethylation increased among patients with NSCLC by 2.33. Hypermethylation of FHIT gene showed a highly significant correlation with pathologic stage ($p < 0.01$) and a significant correlation with smoking habit and FHIT mRNA expression level ($p < 0.05$). In contrast, no correlation was observed between the methylation of GSTP1 or p16 and smoking habit or any other parameter investigated ($p > 0.05$).

Conclusions: Results of the present study suggest that methylation of FHIT is a useful biomarker of biologically aggressive disease in patients with NSCLC. FHIT methylation may play a role in lung cancer later metastatic stage while GSTP1 methylation may play a role in the early pathogenesis of lung cancer rather than in its later metastatic stage.

Keywords: fragile histidine triad, glutathione S-transferase P1, p16, promoter methylation, non-small cell lung carcinoma.

List Of Abbreviations

5mC	5-methylcytosine
AC	Atypical carcinoid
ADC	Adenocarcinoma
AIS	Adenocarcinoma <i>in situ</i>
ATS	American thoracic society
BAC	Bronchioloalveolar carcinoma
bcl-2	B-cell lymphoma 2
BP	Benzo[a]pyrene
BPDE	Benzo[a]pyrene diol epoxide
BRCA1	Breast cancer 1
CBP	CREB binding protein
CDH1	E-cadherin
CDH13	H-cadherin
CDK	Cyclin-dependent kinase
CDK-4	Cyclin-dependent kinase-4
CDKN2A	Cyclin-dependent kinase inhibitor 2A
cDNA	Complementary DNA
CFSs	Common fragile sites
c-myc	Myelocytomatosis viral oncogene homolog
CpG	cytosine–guanine
CREB	cAMP response element-binding protein

List Of Abbreviations (Cont.)

CYP1A1	Cytochrome P450, family 1, subfamily A, polypeptide 1
DAPK	Death-associated protein kinase
DMN	Dimethylnitrosamine
DMS	Dimethyl sulfate
DNA	Deoxyribonucleic acid
DNMTs	DNA methyltransferases
ED	Extensive disease
EGFR	Epidermal growth factor receptor
ERS	European respiratory society
FADD	Fas-associated death domain
FUS1	Fused in Sarcoma
GAPDH	Glyceraldehyde 3-phosphate dehydrogenase
GSTP1	Glutathione S-transferase P1
GSTs	Glutathione S-transferases
HATs	Histone acetyltransferases
HDACs	Histone deacetylases
HDMs	Histone lysine demethylases
HIT	Histidine triad
HIV	Human immunodeficiency virus
HMTs	Histone methyltransferases

List Of Abbreviations (Cont.)

HPV	Human papillomavirus
hTERT	Human telomerase reverse transcriptase
IASLC	International Association for the Study of Lung Cancer
IGFBP 3	Insulin-like growth factor binding protein 3
JNK	C-Jun-NH2 kinase
K-ras	Kirsten rat sarcoma viral oncogene homolog
LCC	Large cell carcinoma
LD	Limited disease
LOH	Loss of heterozygosity
MAPK	Mitogen-activated protein kinase
MBD	Methyl-cpg-binding domain
MDM2	Mouse double minute 2 homolog
mRNA	Messenger RNA
MSP	Methylation-specific PCR
NSCLC	Non-small cell lung cancer
PAH	Polycyclic aromatic hydrocarbons
RAR-β	Retinoic acid receptor- β
RASSF1	Ras association domain-containing protein 1
Rb	Retinoblastoma gene
RNA	Ribonucleic acid

List Of Abbreviations (Cont.)

RRR	Relative risk ratio
SAM	S-adenosyl methionine
SCC	Squamous cell carcinoma
SCLC	Small cell lung cancer
SEMA3B	Semaphorin-3B
SWI/SNF	Switch/Sucrose nonfermentable
TC	Typical carcinoid
TIMP3	Metalloproteinase inhibitor 3
TSGs	Tumor suppressor genes
WHO	World health organization