

Expression of Survivin and Apoptotic Index in Salivary Gland Tumours

Thesis

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I dedicate this thesis to my
family.

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who supported me and provided me
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along the course of this study.

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Rehab.

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

ظهور السيرقيين و مقياس الموت المبرمج للخلايا في أورام الغدد اللعابية

رسالة مقدمة توطئة للحصول على درجة الدكتوراه في علوم طب
الأسنان الأساسية فرع أمراض الفم (باثولوجيا الفم)

مقدمة من

الطبيبة / مرحاب فونري محمود قاسم

مدرس مساعد بقسم باثولوجيا الفم
كلية طب الفم والأسنان
جامعة القاهرة

كلية طب الفم والأسنان
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المشرفون

الدكتورة / أمينة كامل أمين

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بِسْمِ اللّٰهِ الرَّحْمٰنِ الرَّحِیْمِ

نرفع درجات من نشاء و فوق

كل ذي علم عظيم

صدق الله العظيم

آية (٧٦) من سورة يوسف

The present study was conducted to investigate the expression of the novel anti-apoptotic protein survivin and its association with tumour cell apoptosis in 93 archival salivary gland neoplasms (23 benign and 70 malignant neoplasms). Five normal serous and mucous glandular tissues were used as a control group. Specimens were subjected to immunohistochemical staining by anti-survivin monoclonal antibody using Biotin-Strept Avidin detection system. Thirty specimens of malignant tumours were used for histochemical assessment of apoptosis by TUNEL technique.

Quantification of survivin positivity was performed using image analyzer computer system and then evaluated in the form of total score which was graded as follows: scores 1&2 indicated mild expression, scores 3&4 indicated moderate expression and scores 5&6 indicated strong expression. The TUNEL positive apoptotic cells were detected by light microscope and were quantified using the image analyzer for the assessment of the apoptotic index. Significant inverse correlation was found between survivin immunoeexpression and apoptotic index in the control group and malignant salivary gland tumours.

The results showed predominant cytoplasmic survivin positivity in 11/23(47%) benign and in 61/70(87%) malignant salivary neoplasms. However, 13/72(18%) tumours showed both nuclear and cytoplasmic survivin positivity. Normal salivary tissues were negative for survivin immunostain. Survivin immunoeexpression showed significant increase from benign to malignant salivary tumours and from low grade to high grade malignant tumours. Survivin expression correlated with histopathologic malignancy grade in salivary cancers.

Key words: survivin, salivary gland tumours, TUNEL, apoptosis, apoptotic index.

LIST OF TABLES

<i>Table number</i>	<i>Title</i>	<i>Page number</i>
(1)	Members of the IAP family, name, gene location and protein structure.	17
(2)	IAP members: basic function.	18
(3)	Survivin expression in a range of human cancers and its apparent relevance to patient's prognosis	37
(4)	Histopathological types and some clinical data of the salivary gland tumours included in the study.	73
(5)	Difference in apoptotic index between normal salivary tissue (control) and malignant salivary tumours.	75
(6)	Correlation between apoptotic index and survivin expression in low grade and high grade mucoepidermoid carcinoma.	76
(7)	Correlation between apoptotic index and survivin expression in histopathological patterns of adenoid cystic carcinoma.	77
(8)	Correlation between total survivin score and the apoptotic index in normal salivary tissue (control) and malignant salivary tumours.	78
(9)	Immunoreactivity of survivin in benign salivary gland tumours.	83
(10)	Immunoreactivity of survivin in malignant salivary gland tumours.	84
(11)	Difference in the total score of survivin immunoreactivity between benign and malignant salivary gland tumours.	85
(12)	Difference in the total score of survivin immunoreactivity between low grade and high grade malignant salivary gland tumours.	86
(13)	Difference in the total score of survivin immunoreactivity between the histopathological patterns of adenoid cystic carcinoma.	87
(14)	Difference in the total score of survivin immunoreactivity between the histopathological grades of mucoepidermoid carcinoma.	88
(15)	Difference in the total score of survivin immunoreactivity between pleomorphic adenoma and carcinoma ex-pleomorphic adenoma.	89

LIST OF GRAPHS

<i>Graph number</i>	<i>Title</i>	<i>Page number</i>
(1)	Mean values of the apoptotic index in normal salivary tissue (control) and malignant salivary gland tumours.	75
(2)	Mean values of the apoptotic index and total survivin score in low grade and high grade mucoepidermoid carcinoma.	76
(3)	Mean values of the apoptotic index and total survivin score in histopathological patterns of adenoid cystic carcinoma.	77
(4)	Correlation between the mean values of total score of survivin immunoreactivity and apoptotic index in normal salivary gland tissues (control) and malignant salivary tumours.	78
(5)	Mean values of the total score of survivin immunoreactivity in benign and malignant salivary gland tumours.	85
(6)	Mean values of the total score of survivin immunoreactivity in low grade and high grade malignant salivary gland tumours.	86
(7)	Mean values of the total score of survivin immunoreactivity in the histopathological patterns of adenoid cystic carcinoma.	87
(8)	Mean values of the total score of survivin immunoreactivity in low and high grade mucoepidermoid carcinoma.	88
(9)	Mean values of the total score of survivin immunoreactivity in pleomorphic adenoma and carcinoma ex-pleomorphic adenoma.	89

LIST OF FIGURES

<i>Figure number</i>	<i>Title</i>	<i>Page number</i>
(1)	Convergence of two pathways in the activation of effector caspases.	10
(2)	Basis of TUNEL technique.	23
(3)	Representation of protein survivin structure	26
(4)	(a) Expanded view of one survivin monomer. (b) Survivin dimer.	27
(5)	The role of survivin in apoptosis pathways.	31
(6)	Measurement of area positivity of survivin in high grade MEC.	62
(7)	Measurement of optical density of survivin in PLGA.	63
(8)	Detection of apoptotic cell in high grade MEC.	68
(9&10)	Normal mucous and serous salivary gland tissues.	90
(11&12)	Histopathology of pleomorphic adenoma.	91
(13)	Histopathology of Warthin's tumour.	92
(14)	Histopathology of myoepithelioma.	92
(15,16 &17)	Microcystic and macrocystic cribriform adenoid cystic carcinoma.	93&94
(18&19)	Tubular/trabecular adenoid cystic carcinoma.	94&95
(20)	Solid adenoid cystic carcinoma.	95
(21)	Histopathology of low grade MEC.	96
(22, 23&24)	Histopathology of high grade MEC.	96&97
(25&26)	Histopathology of CEPA.	98
(27)	Histopathology of adenocarcinoma (NOS).	99
(28)	Histopathology of undifferentiated carcinoma.	99
(29)	Glandular pattern of PLGA	100
(30)	Cribriform pattern of PLGA.	100
(31)	Trabecular pattern of PLGA.	101
(32)	Solid sheets of malignant cells in PLGA.	101
(33&34)	Papillary adenocarcinoma.	102
(35)	Histopathology of clear cell carcinoma.	103

<i>Figure number</i>	<i>Title</i>	<i>Page number</i>
(36)	Histopathology of acinic cell carcinoma.	103
(37)	Apoptotic cells and bodies in normal salivary gland.	104
(38)	Apoptotic cell in CEPA.	104
(39)	Apoptotic cells in low grade MEC.	105
(40)	Apoptotic cell in high grade MEC.	105
(41)	Apoptotic cells in cribriform adenoid cystic carcinoma.	106
(42&43)	Apoptotic cells in tubular/trabecular adenoid cystic carcinoma.	106&107
(44)	Apoptotic cell in solid adenoid cystic carcinoma	107
(45&46)	Immunohistochemical staining of survivin in normal serous and mucous salivary gland tissues.	108
(47,48&49)	Immunohistochemical staining of survivin in pleomorphic adenoma.	109&110
(50&51)	Immunohistochemical staining of survivin in Warthin's tumour	110&111
(52)	Immunohistochemical staining of survivin in myoepithelioma	111
(53&54)	Immunohistochemical staining of survivin in cribriform adenoid cystic carcinoma.	112
(55&56)	Immunohistochemical staining of survivin in tubular/trabecular adenoid cystic carcinoma.	113
(57)	Immunohistochemical staining of survivin in solid adenoid cystic carcinoma.	114
(58)	Immunohistochemical staining of survivin in cells invading perineural tissues in adenoid cystic carcinoma.	114
(59)	Immunohistochemical staining of survivin in low grade MEC	115
(60&61)	Immunohistochemical staining of survivin in high grade MEC.	115&116
(62,63&64)	Immunohistochemical staining of survivin in CEPA.	116&117
(65)	Immunohistochemical staining of survivin in adenocarcinoma (NOS)	118
(66)	Immunohistochemical staining of survivin in undifferentiated carcinoma.	118
(67)	Immunohistochemical staining of survivin in glandular pattern of PLGA.	119
(68)	Immunohistochemical staining of Survivin in cribriform pattern of PLGA.	119
(69)	Immunohistochemical staining of survivin in trabecular PLGA.	120
(70)	Immunohistochemical staining of survivin in solid PLGA.	120
(71&72)	Immunohistochemical staining of survivin in papillary adenocarcinoma.	121
(73)	Immunohistochemical staining of survivin in clear cell carcinoma.	122
(74)	Immunohistochemical staining of survivin in acinic cell carcinoma.	122

LIST OF ABBREVIATIONS

<i>Abbreviations</i>	<i>Names</i>
bcl-2	B-cell follicular lymphoma/leukemia-2
TNF	Tumour necrosis factor
IAP	Inhibitor of apoptosis family of proteins
kDa	Kilodalton
Kb	Kilobasepair
TNFR	Tumour necrosis factor receptor
TRADD	Tumour necrosis factor receptor associated with death domain
FADD	Fas associated protein with death domain
Apaf-1	Pro-apoptotic protease activator factor
Smac/DIABLO	Second mitochondrial activator of caspases/Direct IAP-binding protein
IL2	Interleukin 2
VDAC	Voltage dependent anion channel (porin)
ACTH	Adrenocorticotrophic hormone
DISC	Death-inducing signaling complex
TGF- β	Transforming growth factor- β
HSPs	Heat chock proteins
UV	Ultraviolet radiation
TUNEL	Terminal deoxynucleotidyl transferase mediated dUTP nick end labeling.
TdT	Terminal deoxynucleotidyl Transferase
dUTP	Deoxyuridine triphosphate nucleotides
BIRC5	Baculoviral IAP repeat containing protein 5
BIR	Baculoviral IAP repeat
ERP-1	Effector cell protease receptor-1
cDNA	Complementary DNA
siRNA	Small interfering RNA
CPPs	Chromosomal passenger proteins
Thr34	Threonin 34
VEGF	Vascular endothelial growth factor
HER2	CerbB2
INK4	cyclinD/CDK4&cyclinD/CDK6 inhibitors; p15, p16, p18&p19
WAF1/Kip1 family	p21, p27&p57 cyclin dependent kinase inhibitors
Cdk,cdc	Cyclin dependent kinas
mdm2	Murine double minute clone 2
IHC	Immunohistochemistry.
ATP	Adenosine triphosphate.

<i>Abbreviations</i>	<i>Words</i>
pRb	Retinoblastoma protein
HLA	Human leukocyte antigen
RT-PCR	Reverse transcriptase-polymerase chain reaction
ER/PgR	Estrogen/progesteron receptor status
ELIZA	Enzyme-linked immunosorbent assay
dsRNA	Double stranded RNA
dsDNA	Double stranded DNA
ssDNA	Single stranded DNA
CTLs	Cytotoxic T-lymphocytes CD8 ⁺
PBMC	Peripheral blood mononuclear cells
ISEL	In situ 3' -end labeling method
ISET	In situ neck translation
AI	Apoptotic index
MTT assay	methyl thiazolyl tetrazolium assay
PCNA	Proliferating cell nuclear antigen
BTCC	Bladder transitional cell carcinoma
OSCC	Oral squamous cell carcinoma
HCC	Hepatocellular carcinoma
NSCLC	Non small cell lung cancer
MVD	Microvessel density
DCIS	Ductal carcinoma in situ
NB	Neuroblastoma
LI	Labeling index
WHO	World Health Organization
DAB	3,3-diaminobenzidine tetra hydrochloride
PBS	Phosphate buffered saline.
HRP	Horseradish peroxidase
PA	Pleomorphic adenoma
MEC	Mucoepidermoid carcinoma
ACC	Adenoid cystic carcinoma
CEPA	Carcinoma ex-pleomorphic adenoma
NOS	not otherwise specified
PLGA	Polymorphous low grade adenocarcinoma
ANOVA	One way analysis of variance test.
TMA	Tissue microarray
B-SA	Super sensitive improved Biotin Strept Avidin amplified System.
Ab	Antibody