Serum microRNA-222 expression for malignancy prediction in euthyroid patients with indeterminate thyroid nodule

Thesis

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Sherihan Abo Elyazed

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18F-FDG-PET	F18-fluorodeoxyglucose-PET
ACE	American College of Endocrinology
ACR	American College of Radiology's
ATA	American Thyroid Association
ATCs	Anaplastic thyroid carcinomas
ATP	Adenosine triphosphate
AUS/FLUS	Atypia of Undetermined Significance /Follicular Lesion of Undetermined Significance
BIRADS	Breast Imaging, Reporting, and Data System
Ca	Calcium
cDNA	Complementary Deoxyribonucleic acid
CE-US	Contrast-enhanced ultrasound
CISH	Chromogenic in situ hybridization
CLL	Chronic lymphocytic leukemia
CT	Computed tomography
D1	Type I iodothyronine deiodinase
D2	Type II iodothyronine deiodinase
D3	Type III iodothyronine deiodinase
DIT	Diiodotyrosine
DNA	Deoxyribonucleic acid
dsRNA	Double-stranded RNA
DTC	Well-differentiated thyroid cancer
DWI	Diffusion-weighted imaging
ELISA	Enzyme-linked immunosorbent assay
FAP	Familial adenomatous polyposis
FCD	Follicular cell derived

List of Abbreviations

FISH	Fluorescence in situ hybridization
FLUS	Follicular lesion of undetermined significance
FN	Follicular Neoplasm
FNA	Fine needle aspiration
FNAB	Fine-needle aspiration biopsy
FNMTC	Familial nonmedullary thyroid carcinoma
FSH	Follicle stimulating hormone
FTAs	Follicular thyroid adenomas
FTC	Follicular thyroid carcinoma
GH	Growth hormone
H&E	Haematoxylin and Eosin
H2O2	Hydrogen peroxide
IFVPC	Invasive follicular variant of papillary carcinoma
K-TIRADS	Korean Society for Thyroid Radiology
MEN	Multiple endocrine neoplasia
MEN2	Multiple endocrine neoplasia type 2
MiRNA	Micro Ribonucleic acid
MIT	Monoiodotyrosine
MRI	Magnetic resonance imaging
MRNAs	Messenger Ribonucleic acid
MTC	Medullary thyroid carcinoma
MTCs	Medullary thyroid carcinomas
NADPH	Nicotinamide adenine dinucleotide phosphate

List of Abbreviations

NFVPTC	Noninvasive follicular variant of papillary thyroid carcinoma
NIFTP	Non-invasive follicular thyroid neoplasm with papillary-like nuclear features
nt	Nucleotides
PCR	Polymerase chain reaction
PET	Positron emission tomography
PGE2	Prostaglandin E2
Pol II	RNA polymerase II
Pol III	RNA polymerase III
pre-miRNA	Precursor miRNA
PRL	Prolactin
PTC	Papillary thyroid carcinoma
PTH	Parathormone
qRT-PCR	Quantitative real-time polymerase chain reaction
RAI	Radioactive iodine
RAIU	Radioactive iodine uptake
RISC	RNA-induced silencing comple
RISC	RNA-induced silencing complex
RNA	Ribonucleic acid
RNAi	RNA interference
RT	Reverse Transcription
rT3	Reverse Triiodothyronine
RXRs	Retinoic acid X receptors
SFN	Suspicious for Follicular Neoplasm
siRNA	Small interfering RNA

List of Abbreviations

SPECT	Single photon emission computed tomography
T2	Diiodothyronine
T3	Triiodothyronine
T4	Thyroxine
TBG	Thyroxin-binding globulin
Tg	Thyroglobulin
TGF-β	Transforming growth factor beta
TIRADS	Thyroid Imaging, Reporting, and Data System
TR	Thyroid receptor
TREs	Thyroid response elements
TRH	Thyrotrophin-releasing hormone
TSH	Thyroid stimulating hormone
US	Ultrasound

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Abstract

Background: The Prevalence of thyroid nodules is rising nowadays, luckily most of them are benign. The risk of malignancy 5-15%, which necessitates the ultimate need to accurately distinguish benign from malignant nodule to avoid unnecessary thyroidectomy with risk of recurrent laryngeal nerve injury, postoperative hypothyroidism and lifetime thyroid replacement therapy, and other complications related to surgery and anaesthesia. Recent evidence suggests that circulating miRNA might have probable advantage as diagnostic or prognostic markers for numerous cancers. Given their reproducible and constant presence in sera, miRNA profiles have emerged as a non-invasive method to categorise a wide variety of human cancers.

Aim of the study: To evaluate a possible relationship between the expression level of circulating miRNA-222 and the histological outcome of euthyroid patients undergoing thyroidectomy for thyroid nodules with indeterminate FNAB cytology.

Subjects and Methods: 45 euthyroid patients with inderteminate thyroid nodules diagnosed with ultrasound and FNAC which planed for thyroidectomy. Quantitative assay of serum micro RNA-222 expression by quantitative Real-Time polymerase chain reaction (qRT-PCR) performed preoperatively, and results compared with postoperative histopathology.

Results: The incidence of thyroid nodules was predominant in female gender in benign group and malignant group. Risk of malignancy increases as TI-RADS and Bethesda scores increases. Also, larger nodule in size has a more risk of malignancy ($p= 0.027$). Expression level of circulating miRNA-222 in serum can't differentiate between benign and malignant patients where there was no significant difference between them statistically ($p=0.905$). Circulating miRNA-222 was a poor predicator for malignant nodules with sensitivity of 50%, specificity of 32.43%, with high negative predictive value (NPV=75%).

Conclusion: Although circulating miRNA-222 has been identified as novel minimally invasive biomarker for preoperative prediction of malignant nodules, but in our study, it did not show a value as a tool for discrimination of malignant nodules. Ultrasound remained important procedure in preoperative prediction and management of thyroid nodules especially when correlating to nodule size which had positive correlation with malignancy in our study.

Key words: Thyroid nodules, microRNA-222, FNAC, Bethesda, TI-RADS

INTRODUCTION

Thyroid cancer is the most common endocrine neoplasms, which accounts for approximately 1.7% of all cancer diagnoses ***(Ferlay et al., 2010).*** However, most thyroid nodules are benign and only 5% harbor malignancy ***(Gharib, 2004).*** Therefore, it is essential to develop safe and accurate test to differentiate between benign nodules and malignant ones ***(Dean and Gharib, 2008).***

Currently, the most important diagnostic method in the detection of thyroid cancer is fine-needle aspiration biopsy (FNAB). Nevertheless, up to 16.6–22.5% of the detections cannot be diagnosed definitely, owing to wrong diagnosis or sampling errors ***(Papini et al., 2002).***

Cibas and Ali published in 2009 the Bethesda Uniform System for Reporting Thyroid Cytopathology. Six different categories were described: nondiagnostic, benign, malignant, atypia/follicular lesion of undetermined significance (FLUS), follicular neoplasm, and suspicious for malignancy. Whereas the first three categories are straight forward to manage, it is the last three categories that pose a management challenge. Biopsy results that include FLUS, follicular neoplasm, and suspicious for malignancy carry a malignancy rate of 5%-15%, 15%-30%, and 60%-75% respectively. The recommended approach for indeterminate FNAB results varies between repeat FNAB,