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TERT Expression in Pituitary Adenomas

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ABSTRACT

Objective: Although pituitary adenomas have benign histomorphological features, some of them may present in an aggressive manner. To predict the behaviour of these tumours, telomerase reverse transcriptase (TERT) activity in pituitary adenomas has been the subject of a few studies with contradictory results. This study aims to investigate whether immunohistochemical expression of TERT differs in neoplastic and nonneoplastic pituitary tissues and aims to investigate whether TERT expression is related to clinicopathological features of pituitary adenomas.

Material and Method: The study included 48 patients who had been diagnosed with pituitary adenomas and had clinical follow-ups. Nonneoplastic pituitary tissues were obtained from autopsy specimens (n=20). Immunohistochemistry for TERT antibody was performed. Both the nuclear and cytoplasmic expression of TERT antibody was noted, and total combined TERT staining was evaluated according to nuclear and cytoplasmic stainings.

Results: TERT expression did not differ between neoplastic and nonneoplastic pituitary tissues. Neither total (combined nuclear and cytoplasmic) TERT nor nuclear TERT expression revealed any statistically significant relationship with any of the clinicopathological features. Higher cytoplasmic TERT expression was observed in adenomas with recurrence than adenomas without recurrence (p=0.035).

Conclusion: This study introduces the notion that immunohistochemical expression of TERT does not differ in neoplastic and nonneoplastic pituitary tissues. Pituitary adenomas with cytoplasmic immunohistochemical expression of TERT have significantly higher rates of recurrence. Further studies, including combined methods of immunohistochemistry and molecular analyses in larger groups, may reveal applicable results for the clinical significance of TERT in pituitary adenomas.

Key Words: Pituitary adenomas, Telomerase reverse transcriptase, Clinicopathological features

INTRODUCTION

Pituitary adenomas are adenohypophyseal tumours with increasing incidence due to the improving radiological methods and hormone assays for detection (1). The prevalence of these tumours is 14.4% in autopsy series and 22.5% in radiological studies, with an overall estimated prevalence of 16.7% (2). Several clinical outcomes according to tumour size and hormonal activity may occur in these tumours that evolve from a small endocrine gland (1,3). These clinical scenarios may include either local mass effects or systemic effects resulting from endocrine disorders (3).

Pituitary adenomas can be classified according to size as microadenomas (≤10 mm) or macroadenomas (>10 mm) and according to radiological appearance as invasive, noninvasive, or aggressive-invasive (4). The World Health Organization (5) currently classifies pituitary adenomas based on the immunohistochemical demonstration of produced and expressed hormones with clinical reflections. However, the most recent classification of pituitary adenomas is based on an immunohistochemical panel consisting of immune profiling of adenohypophyseal hormones by monoclonal antibodies, cell-specific transcription factors, and low-molecular-weight keratin (CAM5.2), Ki-67, and p53 (6,7).

Although pituitary adenomas have benign histomorphological features, some of these tumours may present in an aggressive manner by invasion of surrounding tissues, recurrences, and resistance to medical therapies (4,8). Thus, the WHO classification defined these tumours as invasive pituitary adenomas with increased mitotic activity, a Ki-67 proliferation index of >3%, and extensive p53 immune staining, namely, 'atypical adenomas'. To predict the behaviour of these tumours, many studies have been performed by investigating various markers related to chromosomal alterations, microRNAs (miRNAs), proliferation markers, oncogenes, tumour suppressor genes, angiogenesis, cell adhesion, growth factors, and their receptors (1,4,9-12). Such studies show that none of these markers may predict the behaviour of these tumours alone,

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but combinations of fibroblast growth factor receptor 4 (FGFR4), matrix metalloproteinases (MMPs), particularly MMP2 and MMP9, Ki-67, p53, pituitary tumour transforming gene (PTTG), and deletions in chromosome 11p seem to have benefits for predicting the aggressiveness of pituitary adenomas (1).

Moreover, investigations continue into several biomarkers other than the suggested panel mentioned above (13, 14). One of these markers is telomerase reverse transcriptase (TERT). Telomerase is a ribonucleic protein complex that includes a catalytic subunit TERT (telomerase associated protein 2) and an RNA component (TERC), and it maintains telomere homeostasis and chromosomal integrity (15). Telomeres are located at the end and inner sides of chromosomes, and shortening of these nucleotide sequences in cell divisions induces apoptosis or cell senescence. On the other hand, lengthening of the telomeres results in prevention of cell replication and thus is assumed to be a part of tumorigenesis, particularly by expression or activation of telomerase (16). TERT expression is suppressed in normal adult somatic tissues, but it can be expressed in embryogenic tissues. Reactivation of TERT has been detected in approximately 90% of human cancers (3,17-20). Thus, TERT activity in pituitary adenomas has been the subject of a few studies that showed contradictory results (3,16,21).

The present study aims to investigate mainly two issues. Initially, the authors aim to investigate whether immunohistochemical expression of TERT differs in neoplastic and nonneoplastic pituitary tissues. Then, the authors will investigate whether TERT expression is related to clinicopathological features of pituitary adenomas such as gender, age at the presentation, tumour size, hormonal activity of the tumour, and recurrence.

MATERIAL and METHODS

Patient Selection

The medical reports of patients who were referred to the Department of Pathology were reviewed between August 2007 and August 2014. The study protocol was approved by the local Ethics Committee of the University Hospital. Patients selected for the study had been diagnosed with pituitary adenomas and had clinical follow-ups. In all, 78 patients who fulfilled the criteria were included in the study. Patient data regarding age at the time of diagnosis, sex, and data from clinical follow-ups (recurrence of disease, re-operation) were obtained from the records of the Department of Clinical Endocrinology and Metabolism Diseases. Also, nonneoplastic pituitary tissues were obtained from autopsy specimens among those who died from causes other than endocrine diseases (n=20). Haematoxylin- and eosin-stained slides (Figure 1A), reticulin stained slides (Figure 1B), and immunohistochemical stainings

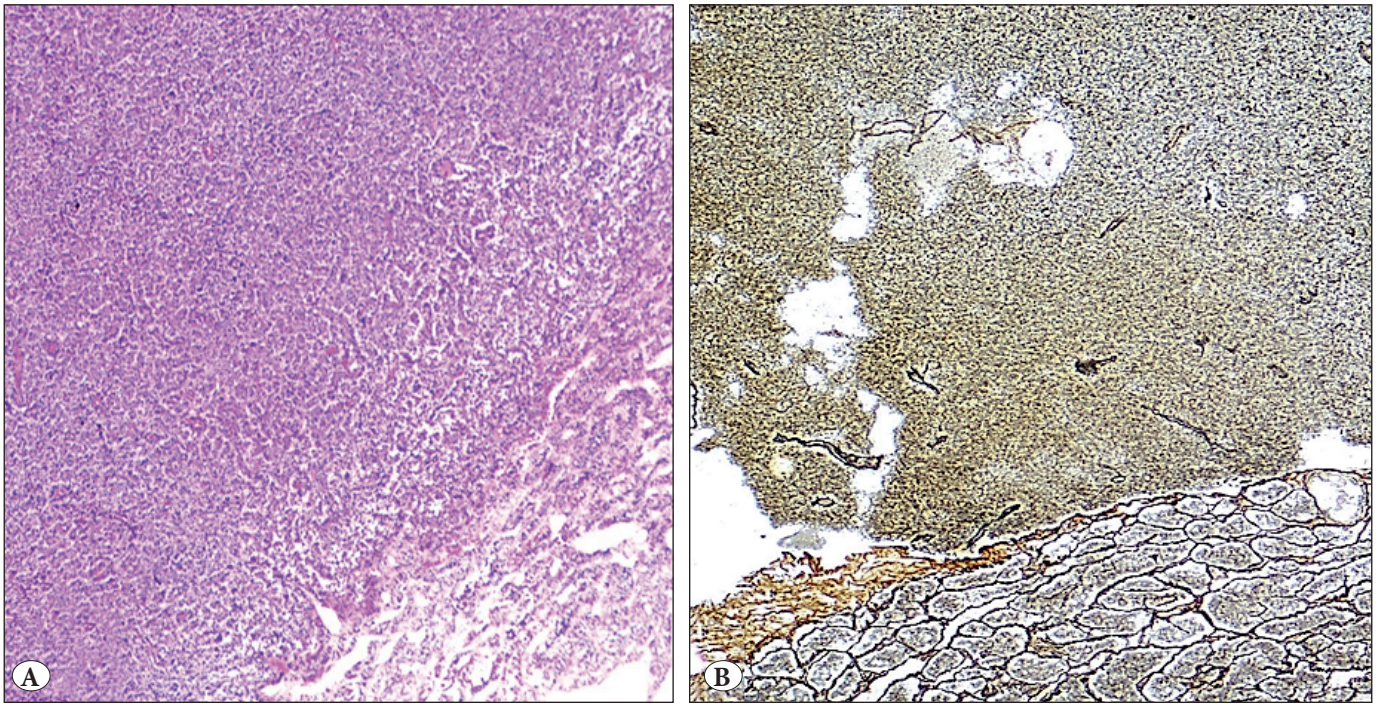

Figure 1: A) Pituitary adenoma surrounded by nonneoplastic pituitary tissue (H&E; x200). **B)** Destruction of reticulin meshwork in pituitary adenoma, note the maintenance of reticulin meshwork in nonneoplastic pituitary tissue (Reticulin; x200).

of adenohypophyseal hormones (growth hormone [GH], prolactin [PRL], adrenocorticotrophic hormone [ACTH], follicle-stimulating hormone [FSH], luteinizing hormone [LH], and thyroid stimulating hormone [TSH]) obtained from paraffin-embedded blocks of specimens were re-evaluated by one pathologist (N.C.) who was blinded to the original pathological diagnosis of the slide and to clinical and prognostic data. The paraffin-embedded blocks containing appropriate tissues representing the adenomas of 48 patients were selected for immunohistochemical studies for TERT. The patients whose specimens did not represent tumour tissue were excluded from the study.

Clinicopathological Criteria

Tumours were grouped according to the preoperative radiological size and the secreted hormone(s) that caused clinical signs. Tumours sized ≤10 mm were classified as microadenomas, whereas tumours sized >10 mm were classified as macroadenomas. Tumours were grouped according to the clinical symptoms and dominant immune reaction of secreted hormones as in the following (since adenohypophyseal transcription factors could not be performed, the classification was performed according to the status of adenohypophyseal hormones):

- GH producing/expressing adenomas: Somatotroph adenomas
- PRL producing/expressing adenomas: Lactotroph adenomas
- FSH-LH producing/expressing adenomas: Gonadotroph adenomas
- ACTH producing/expressing adenomas: Corticotroph adenomas
- TSH producing/expressing adenomas: Thyrotroph adenomas
- GH and PRL producing/expressing adenomas: Mixed somatotroph and lactotroph adenomas
- Nonproducing adenomas: Null cell adenomas

The clinical features considered in the statistical analysis included age at the time of diagnosis, sex (male or female), tumour size (≤10 mm, >10 mm) and recurrence (absent/present).

Immunohistochemistry

Immunostaining for the TERT antibody was performed with a fully automated immunohistochemistry and in situ hybridization (IHC/ISH) staining machine (Ventana BenchMark XT, USA). The following primary antibody at the indicated dilutions was used for TERT immune staining (TERT polyclonal antibody, unconjugated, 1:100; Bioss, USA, Catalogue No. bs-1411R). A single pathologist (N.C.) who was blinded to the clinical assessments of each case evaluated the expression in tissues with a Nikon Eclipse 80i microscope. Both nuclear and cytoplasmic expression of TERT antibody was noted (22) and scored. Then, a total (combined nuclear and cytoplasmic) score was obtained from the sum of cytoplasmic and nuclear scores. The scoring schema is presented below.

The scoring of cytoplasmic TERT expression:

- Cytoplasmic score 0: Cytoplasmic staining in <10% of the tissue (Figure 2A),
- Cytoplasmic score 1: Mild cytoplasmic staining in ≥10% of the tissue (Figure 2B),
- Cytoplasmic score 2: Moderate cytoplasmic staining in ≥10% of the tissue (Figure 2C),
- Cytoplasmic score 3: Significant cytoplasmic staining in ≥10% of the tissue (Figure 2D).

The scoring of nuclear TERT expression:

- Nuclear score 0: Nuclear staining in <10% of the tissue,
- Nuclear score 1: Dot-like nuclear staining in ≥10% of the tissue (Figure 3A),
- Nuclear score 2: Complete nuclear staining in ≥10% of the tissue (Figure 3B).

Total (combined nuclear and cytoplasmic) TERT expression:

- Negative for TERT expression: The sum of nuclear and cytoplasmic scores <2,
- Positive for TERT expression: The sum of nuclear and cytoplasmic scores ≥2 (Figures 3C-3D).

Statistical Analysis

Statistical analysis was carried out using SPSS v20.0 software (IBM SPSS, Inc., Chicago, IL, USA). Appropriate chi-square tests (Pearson, Yates, or Fisher) were used to compare the total TERT expression with clinicopathological features such as tissue type, gender, tumour size, hormonal tumour type, and recurrence. The Mann-Whitney U test and Kruskal-Wallis test were used in the comparisons of numerical data (nuclear/cytoplasmic TERT score with tissue type, gender, tumour size, hormonal tumour type, and recurrence). A p value of <0.05 was considered statistically significant.

RESULTS

Clinicopathological Features of Patients in the Study Group

The clinicopathological features of the patients are summarized in Table I. The median age of the patients was 52.7 years (ranging from 18 to 79 years). Of the 48 patients, 18 (37.5%) were female and 30 (62.5%) were male. Tumour type according to the hormonal status was somatotroph adenoma in 13 (27.1%), lactotroph adenoma in 8 (16.7%), gonadotroph adenoma in 3 (6.3%), corticotroph adenoma in 5 (10.4%), thyrotroph adenoma in 1 (2.1%), mixed somatotroph and lactotroph adenoma in 9 (18.7%), and null cell adenoma in 9 (18.7%) of the patients.

Table I: Clinicopathological features of patients

		n (%)
Gender	Female	18 (37.5)
	Male	30 (62.5)
Hormonal type	Somatotroph adenoma	13 (27.1)
	Lactotroph adenoma	8 (16.7)
	Gonadotroph adenoma	3 (6.3)
	Corticotroph adenoma	5 (10.4)
	Thyrotroph adenoma	1 (2.1)
	Mixed somatotroph and lactotroph adenoma	9 (18.7)
	Null cell adenoma	9 (18.7)
Tumor size	≤ 10 mm	8 (16.7)
	> 10 mm	40 (83.3)
Recurrence	Absent	44 (91.7)
	Present	4 (8.3)

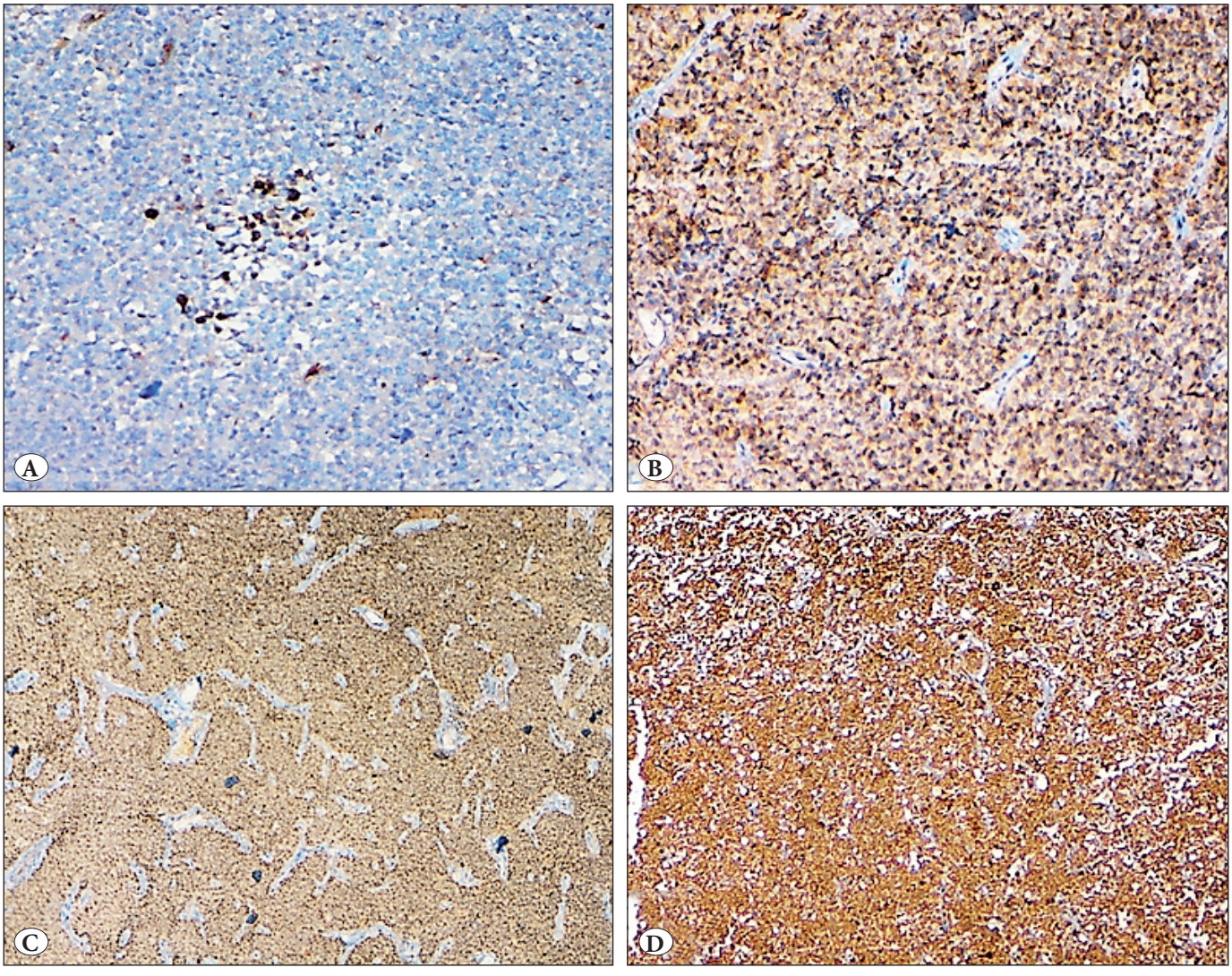

Figure 2: A) Cytoplasmic TERT staining in < 10% of the tissue, score 0 (TERT; x40). **B)** Mild cytoplasmic staining, score I (TERT; x40). **C)** Moderate cytoplasmic staining, score II (TERT; x40), **D)** Significant cytoplasmic staining, score III (TERT; x40).

When the patients were grouped according to tumour size, 8 (16.7%) had microadenomas, while the tumour size was larger than 10 mm in 40 (83.3%) of the cases. Among all of the cases, 4 (8.3%) of the tumours recurred after initial surgery, whereas 44 (91.7%) did not. The mean follow-up period for the patients was 57.5±31.07 months. All of the patients were alive.

Comparisons of TERT Expression in Neoplastic and Nonneoplastic Pituitary Tissues

TERT expression did not significantly differ between neoplastic and nonneoplastic pituitary tissues. According to these results, TERT expression was present in 4 (20%) of 20 nonneoplastic autopsy tissues, whereas it was expressed in 16 (33.3%) of 48 neoplastic tissues. Despite the absence of statistical significance, mean ranks of cytoplasmic and nuclear TERT expression was higher in neoplastic tissues than in nonneoplastic autopsy tissues (p>0.05) (Table II).

Comparisons of Total (Combined Nuclear and Cytoplasmic) TERT Expression with Clinicopathological Features

The results of the comparisons of clinicopathological features with total (combined nuclear and cytoplasmic) TERT expression are presented in Table III. TERT expression did not reveal any statistically significant relationship with any of the clinicopathological features. TERT positivity was present in 30.8% (4/13) of somatotroph adenomas, in 25.0% (2/8) of lactotroph adenomas, in 33.3% (1/3) of gonadotroph adenomas, in 40% (2/5) of corticotroph adenomas, in none (0/1) of the thyrotroph adenomas, in 33.3% (3/9) of mixed somatotroph and lactotroph adenomas, and finally, in 44.4% (4/9) of null cell adenomas. TERT staining was defined in 50.0% (4/8) of microadenomas, while it was defined as 30.0% (12/40)

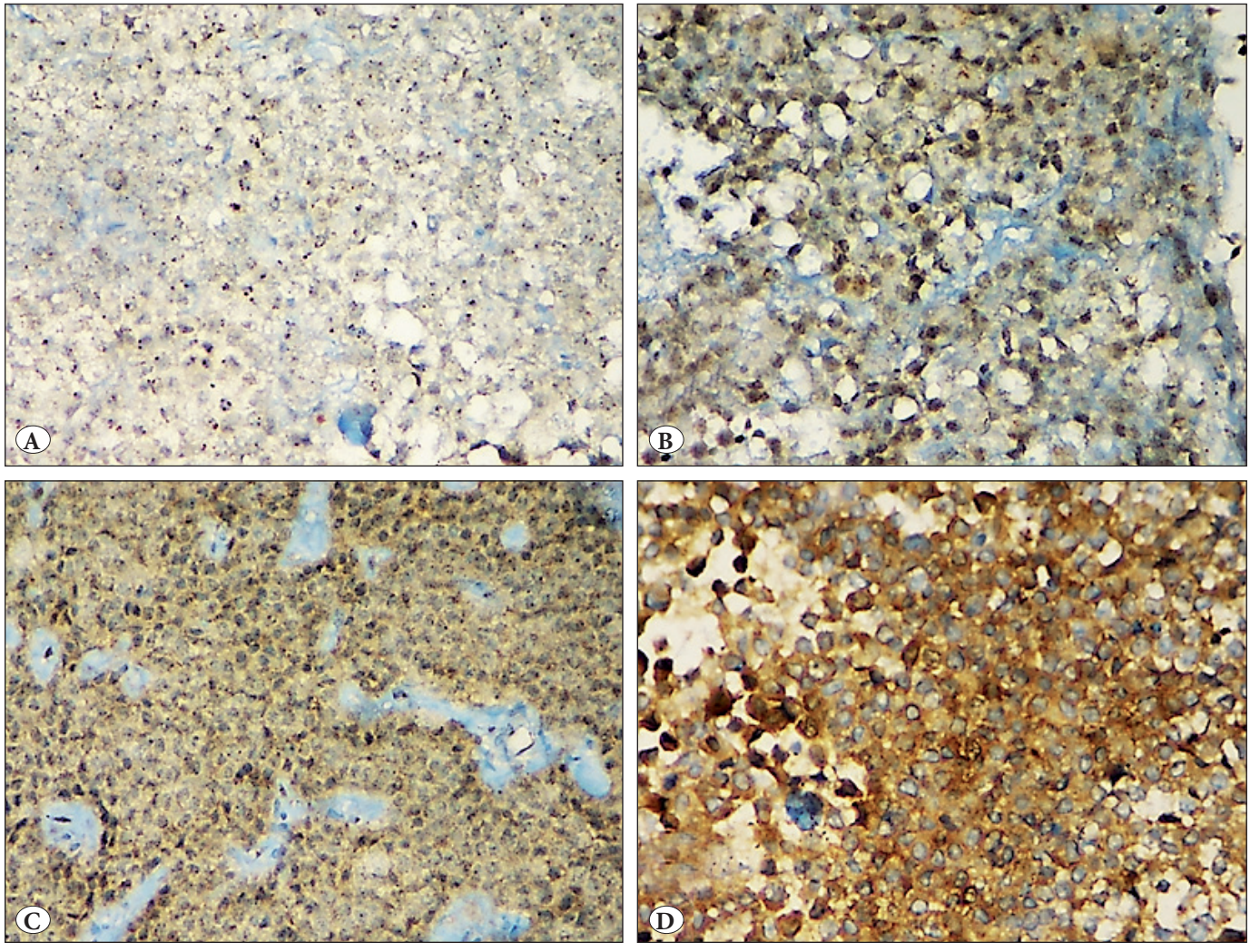

Figure 3: A) Dot-like nuclear staining of TERT; score 1 (TERT; x200). **B)** Complet nuclear staining of TERT; score 2 (TERT; x400). **C)** Dot-like nuclear staining (Nuclear score 1) with mild cytoplasmic expression (cytoplasmic score I) (TERT; x200). **D)** Moderate cytoplasmic expression (cytoplasmic score II) (TERT; x400).

Table II: Comparisons of TERT expression in neoplastic and nonneoplastic pituitary tissues

		TERT expression							
		Combined nuclear and cytoplasmic		p	Nuclear	p	Cytoplasmic		p
		Absent n (%)	Present n (%)		Mean rank		Mean rank		
Tissue type	Nonneoplastic	16/20 (80.0)	4/20 (20.0)	0.272	30.68	0.107	29.25		0.119
	Neoplastic	32/48 (66.7)	16/48 (33.3)		36.09		36.69		

Table III: Comparisons of total (combined nuclear and cytoplasmic) TERT expression with clinicopathological features

		TERT expression		Total	p
		Absent n (%)	Present n (%)	n	
Gender	Female	12 (66.7)	6 (33.3)	18	1.000
	Male	20 (66.7)	10 (33.3)	30	
Hormonal type	Somatotroph adenoma	9 (69.2)	4 (30.8)	13	0.967
	Lactotroph adenoma	6 (75.0)	2 (25.0)	8	
	Gonadotroph adenoma	2 (66.7)	1 (33.3)	3	
	Corticotroph adenoma	3 (60.0)	2 (40.0)	5	
	Thyrotroph adenoma	1 (100.0)	0 (0.0)	1	
	Mixed somatotroph and lactotroph adenoma	6 (66.7)	3 (33.3)	9	
	Null cell adenoma	5 (55.6)	4 (44.4)	9	
Tumor size	≤ 10 mm	4 (50.0)	4 (50.0)	8	0.242
	> 10 mm	28 (70.0)	12 (30.0)	40	
Recurrence	Absent	31 (70.5)	13 (29.5)	44	0.101
	Present	1 (25.0)	3 (75.0)	4	

of macroadenomas. Total TERT expression was observed in 75.0% (3/4) of the patients with recurrence, whereas staining was present in 29.5% (13/44) of the patients without recurrence.

Comparisons of Nuclear TERT Expression with Clinicopathological Features

The results of the comparisons of clinicopathological features with nuclear TERT expression are presented in Table IV. Nuclear TERT expression did not reveal any statistically significant relationship with any of the clinicopathological features. However, mean ranks of nuclear TERT expression were higher in males than in females, in mixed somatotroph and lactotroph adenomas, in lactotroph adenomas, and in adenomas sized >10 mm (p>0.05).

Comparisons of Cytoplasmic TERT Expression with Clinicopathological Features

The results of the comparisons of clinicopathological features with cytoplasmic TERT expression are presented in Table IV. Cytoplasmic TERT expression had a significant relationship with recurrence (p=0.035); namely, higher cytoplasmic TERT expression was observed in some adenomas with recurrence but not in adenomas without recurrence. Cytoplasmic TERT expression was observed in 3 of 4 patients who had recurred. Nuclear TERT expression was not observed in any of the patients with recurrence. Two of these 3 patients were male and 1 was female. Tumours were sized >10 mm in 2 of the patients and sized ≤10 mm in 1 patient. Among these 3 patients, hormonal type was corticotroph adenoma in 1 of the patients, gonadotroph adenoma in 1, and null cell adenoma in 1. The follow-up period ranged between 37 months and 256 months.

There was no significant relationship between cytoplasmic TERT expression and clinicopathological features other than recurrence. Although there was no significant relationship, mean ranks of cytoplasmic TERT expression were higher in males than in females, in corticotroph and lactotroph adenomas than in other types, and in microadenomas than in macroadenomas (p>0.05).

DISCUSSION

Pituitary adenomas have benign histomorphological features, but some of these tumours may present in an

Table IV: Comparisons of nuclear TERT expression and cytoplasmic TERT expression with clinicopathological features

		TERT expression			
		Nuclear Mean rank	p	Cytoplasmic Mean rank	p
Gender	**Female**	23.42	0.556	23.72	0.741
	Male	25.5		24.97	
Hormonal type	**Somatotroph adenoma**	20.81	0.192	18.73	0.692
	Lactotroph adenoma	25.56		24.56	
	Gonadotroph adenoma	Not calculated		Not calculated	
	Corticotroph adenoma	17.50		26.40	
	Thyrotroph adenoma	Not calculated		Not calculated	
	Mixed somatotroph and lactotroph	27.61		23.44	
	Null cell adenoma	19.89		23.00	
Tumor size	**≤ 10 mm**	22.44	0.654	31.50	0.126
	> 10 mm	24.91		23.10	
Recurrence	**Absent**	24.95	0.482	23.34	**0.035**
	Present	19.50		37.25	

aggressive manner by local invasion through surrounding tissues, recurrences, and resistance to medical therapies (4,8). Many studies have aimed to discover a significant predictor of an aggressive clinical course in pituitary adenomas (1,4,9-12). Telomerase reverse transcriptase (TERT), a catalytic subunit of telomerase that includes an RNA component (TERC), therewithal maintains the telomere homeostasis and chromosomal integrity (15). Reactivation of TERT has been detected in approximately 90% of human cancers (3,17-20). This study attempted to investigate whether immunohistochemical expression of TERT differs in neoplastic and nonneoplastic pituitary tissues and aimed to investigate whether TERT expression is related to clinicopathological features of pituitary adenomas. In this context, the results of the present study can be summarised as follows: i) Total immunohistochemical expression of TERT does not differ in neoplastic and nonneoplastic pituitary tissues with significance, ii) Total immunohistochemical expression of TERT does not have any relationship to clinicopathological parameters, and iii) Cytoplasmic immune staining with TERT antibody is significantly more common in pituitary adenomas with recurrence (p=0.035).

Telomerase activity in neoplastic and in nonneoplastic pituitary tissues has been the subject of a few previous studies using various methods of polymerase chain reaction (PCR) (14,21). The mentioned previous studies have reported that there is no difference in telomerase activity and telomere length between neoplastic and nonneoplastic pituitary tissues. It has been suggested that this may be elucidated by the low mitotic activity of pituitary adenomas (14). In agreement with previous studies, the present study showed no significant difference in total TERT expression in neoplastic and nonneoplastic pituitary tissues. Although TERT expression was evaluated by immunohistochemistry targeting only TERT and not TERC in the present study and the comparisons showed a lack of statistical significance (p>0.05), TERT expression rates were higher in neoplastic tissues than in nonneoplastic tissues. The most accurate state of telomerase activity and telomere length in pituitary tumours/nonneoplastic pituitary tissues should be investigated in larger study groups by combined synchronous detection methods.

The effects of telomerase activity and telomere lengths on clinicopathological features in pituitary adenomas have been evaluated in several studies (3,14,16,21,23-25). Some of these papers have concluded that telomerase activity and telomere length do not have any impact on the clinical course of pituitary adenomas based on investigations of telomerase activity and telomere length by various types of polymerase chain reactions (14,16,21,25). On the other hand, some of the previous studies have suggested that TERT expression is associated with an aggressive clinical course, particularly recurrences and invasiveness (3,23,24). Harada et al. (24) have reported telomerase activity by using Southern blotting and reverse transcriptase-chain reaction in a pituitary carcinoma evolving in a background of an initially telomerase-negative PRL-producing benign

adenoma. Yoshino et al. (23) have reported telomerase activity via PCR-based telomeric repeat amplification protocol (TRAP) assay and PCR enzyme-linked immunosorbent assay (ELISA) in 13% of adenomas with invasive features. Ortiz-Plata et al. (3) have examined telomerase activity by TERT immunohistochemistry and have reported that telomerase activity could be a marker for cellular proliferation, angiogenesis and hormonal activity in pituitary adenomas. However, the authors did not inform either cellular localisation or the degree of TERT expression in their study. In the present study, we could not determine any significant relationship between total TERT expression and clinicopathological features, including presentation age, gender, tumour size, recurrence, and hormonal type. But, we could observe that cytoplasmic staining with TERT polyclonal antibody is significantly related to tumour recurrence, as in the case reported by Harada et al. (24). Also, total TERT expression was present in 75% of pituitary adenomas with recurrence without any significance. Although the present study has limitations, including the immunohistochemical evaluation of only the TERT catalytic subunit of telomerase protein complex but not TERC or any detections of mutational status, the authors present the significant increase of recurrence in pituitary adenomas with cytoplasmic TERT expression.

According to the current proposals, a panel of some biomarkers, including PTTG1 and MMPs (MMP1), may predict the aggressive behaviour of pituitary adenomas (1). It is said that higher levels of electron transport system (ETS) transcription factors induce MMP1 expression and cause tumour invasion in pituitary adenomas (1). One of the most recent reports investigating the TERT expression in malignant melanoma has concluded that mutations in TERT promoter create additional binding sites for ETS transcription factors, particularly ETS1, and finally activates the mitogen-activated protein kinase (MAPK) pathway and cell proliferation (19). PTTG1 is a member of the securin family and is highly expressed in hormone-secreting invasive pituitary adenomas (1). Another recent study observing the relationship between PTTG1 and TERT expressions in human mesenchymal stem cells has reported that overexpression of TERT induces PTTG1 expression, and this interaction between TERT and PTTG1 is mediated by Ku70, which is a heterodimeric protein involved in maintenance of telomeres, in an increase in the cell cycle, autophagy, and self-renewal (26).

Consideration of distinct results of the previous studies and the present study about telomerase activity and telomere length in pituitary adenomas and the recent observations revealing interactions of TERT with PTTG1 and ETS1 may require further studies. Such studies should investigate these interactions in pituitary adenomas and may contribute detailed data in the issue of TERT expression as a prognostic predictor in pituitary adenomas.

In conclusion, the present study posits that immunohistochemical expression of TERT does not significantly differ in neoplastic and nonneoplastic pituitary tissues. In addition, pituitary adenomas with cytoplasmic immunohistochemical expression of TERT have significantly higher rates of recurrence. Further studies, including combined methods of immunohistochemistry and molecular analyses in larger groups, may reveal applicable results for the clinical significance of telomerase activity and telomere length in pituitary adenomas.

CONFLICT of INTEREST

The authors declare no conflict of interest.

REFERENCES

1. Mete O, Ezzat S, Asa SL. Biomarkers of aggressive pituitary adenomas. J Mol Endocrinol. 2012;49:R69-78.
2. Ezzat S, Asa SL, Couldwell WT, Barr CE, Dodge WE, Vance ML, McCutcheon IE. The prevalence of pituitary adenomas: A systematic review. Cancer. 2004;101:613-9.
3. Ortiz-Plata A, Tena Suck ML, Lopez-Gomez M, Heras A, Sanchez Garcia A. Study of the telomerase hTERT fraction, PCNA and CD34 expression on pituitary adenomas. Association with clinical and demographic characteristics. J Neurooncol. 2007;84:159-66.
4. Wierinckx A, Auger C, Devauchelle P, Reynaud A, Chevallier P, Jan M, Perrin G, Fevre-Montange M, Rey C, Figarella-Baranger D, Raverot G, Belin MF, Lachuer J, Trouillas J. A diagnostic marker set for invasion, proliferation, and aggressiveness of prolactin pituitary tumors. Endocr Relat Cancer. 2007;14:887-900.
5. DeLellis RA. Pathology and genetics of tumours of endocrine organs. Lyon:IARC Press; 2004.
6. Mete O, Asa SL. Clinicopathological correlations in pituitary adenomas. Brain Pathol. 2012;22:443-53.
7. Gomez-Hernandez K, Ezzat S, Asa SL, Mete O. Clinical implications of accurate subtyping of pituitary adenomas: Perspectives from the treating physician. Turk Patoloji Derg. 2015;31 Suppl 1:4-17.
8. Scheithauer BW, Kovacs KT, Laws ER Jr, Randall RV. Pathology of invasive pituitary tumors with special reference to functional classification. J Neurosurg. 1986;65:733-44.
9. Wierinckx A, Raverot G, Nazaret N, Jouanneau E, Auger C, Lachuer J, Trouillas J. Proliferation markers of human pituitary tumors: Contribution of a genome-wide transcriptome approach. Mol Cell Endocrinol. 2010;326:30-9.

10. Wierinckx A, Roche M, Raverot G, Legras-Lachuer C, Croze S, Nazaret N, Rey C, Auger C, Jouanneau E, Chanson P, Trouillas J, Lachuer J. Integrated genomic profiling identifies loss of chromosome 11p impacting transcriptomic activity in aggressive pituitary PRL tumors. Brain Pathol. 2011;21:533-43.
11. Cornelius A, Cortet-Rudelli C, Assaker R, Kerdraon O, Gevaert MH, Prevot V, Lassalle P, Trouillas J, Delehedde M, Maurage CA. Endothelial expression of endocan is strongly associated with tumor progression in pituitary adenoma. Brain Pathol. 2012;22:757-64.
12. Mete O, Hayhurst C, Alahmadi H, Monsalves E, Gucer H, Gentili F, Ezzat S, Asa SL, Zadeh G. The role of mediators of cell invasiveness, motility, and migration in the pathogenesis of silent corticotroph adenomas. Endocr Pathol. 2013;24:191-8.
13. De Martino I, Visone R, Wierinckx A, Palmieri D, Ferraro A, Cappabianca P, Chiappetta G, Forzati F, Lombardi G, Colao A, Trouillas J, Fedele M, Fusco A. HMGA proteins up-regulate CCNB2 gene in mouse and human pituitary adenomas. Cancer Res. 2009;69:1844-50.
14. Martins CS, Santana-Lemos BA, Saggioro FP, Neder L, Machado HR, Moreira AC, Calado RT, de Castro M . Telomere length and telomerase expression in pituitary tumors. J Endocrinol Invest. 2015;38:1243-6.
15. Martinez P, Blasco MA. Telomeric and extra-telomeric roles for telomerase and the telomere-binding proteins. Nat Rev Cancer. 2011;11:161-76.
16. Kochling M, Ewelt C, Furtjes G, Peetz-Dienhart S, Koos B, Hasselblatt M, Paulus W, Stummer W, Brokinkel B. hTERT promoter methylation in pituitary adenomas. Brain Tumor Pathol. 2016;33:27-34.
17. Xu L, Li S, Stohr BA. The role of telomere biology in cancer. Annu Rev Pathol. 2013;8:49-78.
18. Agostini A, Panagopoulos I, Andersen HK, Johannesen LE, Davidson B, Trope CG, Heim S, Micci F. HMGA2 expression pattern and TERT mutations in tumors of the vulva. Oncol Rep. 2015;33:2675-80.
19. Vallarelli AF, Rachakonda PS, Andre J, Heidenreich B, Riffaud L, Bensussan A, Kumar R, Dumaz N. TERT promoter mutations in melanoma render TERT expression dependent on MAPK pathway activation. Oncotarget. 2016;7:53127-36.
20. Chen C, Han S, Meng L, Li Z, Zhang X, Wu A. TERT promoter mutations lead to high transcriptional activity under hypoxia and temozolomide treatment and predict poor prognosis in gliomas. PLoS One. 2014;9:e100297.
21. Martins CS, de Castro M, Calado RT. Absence of TERT promoter mutations in pituitary adenomas. J Endocrinol Invest. 2016;39:933-4.
22. Kyo S, Masutomi K, Maida Y, Kanaya T, Yatabe N, Nakamura M, Tanaka M, Takarada M, Sugawara I, Murakami S, Taira T, Inoue M. Significance of immunological detection of human telomerase reverse transcriptase: Re-evaluation of expression and localization of human telomerase reverse transcriptase. Am J Pathol. 2003;163:859-67.
23. Yoshino A, Katayama Y, Fukushima T, Watanabe T, Komine C, Yokoyama T, Kusama K, Moro I. Telomerase activity in pituitary adenomas: Significance of telomerase expression in predicting pituitary adenoma recurrence. J Neurooncol. 2003;63:155-62.
24. Harada K, Arita K, Kurisu K, Tahara H. Telomerase activity and the expression of telomerase components in pituitary adenoma with malignant transformation. Surg Neurol. 2000;53:267-74.
25. Hiraga S, Ohnishi T, Izumoto S, Miyahara E, Kanemura Y, Matsumura H, Arita N. Telomerase activity and alterations in telomere length in human brain tumors. Cancer Res. 1998;58:2117-25.
26. Lee HJ, Choi JH, Jung J, Kim JK, Lee SS, Kim GJ. Changes in PTTG1 by human TERT gene expression modulate the self-renewal of placenta-derived mesenchymal stem cells. Cell Tissue Res. 2014;357:145-57.