Granular cell tumors overexpress TFE3 without gene rearrangement: Evaluation of immunohistochemistry and break-apart FISH in 45 cases

YANG LIU[1], QIN ZHENG[1], CHEN WANG[1], JINPING WANG[1], JIAN MING[2], YONG ZHANG[3], XIAOMAN LI[4], WILLIAM CHI-SHING CHO[5], LIANG WANG[1*], QING-CHANG LI[1], XUE-SHAN QIU[1] and EN-HUA WANG[1]

[1]Department of Pathology, The First Affiliated Hospital and College of Basic Medical Sciences, China Medical University, Shenyang, Liaoning 110001; [2]Department of Pathology, The 202nd Hospital of Chinese PLA, Shenyang, Liaoning 110003; [3]Department of Pathology, Cancer Hospital of China Medical University, Shenyang, Liaoning 110042; [4]Key Laboratory of Medical Cell Biology, Ministry of Education, China Medical University, Shenyang, Liaoning 110001; [5]Department of Clinical Oncology, Queen Elizabeth Hospital, Kowloon, Hong Kong SAR, P.R. China

Received December 7, 2018; Accepted August 1, 2019

DOI: 10.3892/ol.2019.10995

Abstract. Transcription factor E3 (TFE3) is a useful marker for tumors with Xp11.2 translocation, including alveolar soft part sarcoma and renal cell carcinoma. Recently, TFE3 overexpression was also found in granular cell tumors (GrCTs). However, the case cohorts of these two studies were limited to only 11 and 6 cases. Whether aberrant TFE3 expression is a common feature of Asian patients with GrCT requires further investigation. In the present study, immunohistochemical staining and TFE3 break-apart fluorescence *in situ* hybridization (FISH) assay were performed in 45 samples of GrCTs obtained from Chinese patients recruited from three medical centers in northeast China. Diffusive and marked nuclear staining for TFE3 was identified in 11/45 (24%) cases, which was lower than previously reported. Focal or weak TFE3 staining was identified in 13/45 (29%) cases. The remaining 21 cases were negative stained. In addition, GrCTs in subcutaneous tissue exhibited a relatively higher ratio (8/45, 18%) for TFE3 expression, compared with those in other sites. Furthermore, according to FISH data, no rearrangement or amplification of TFE3 was identified in these cases, whether they were positively or negatively stained for TFE3. The results from the present study demonstrated that part of patients GrCTs exhibited TFE3 overexpression, which suggested that this may not be derived from gene rearrangement.

Correspondence to: Professor Liang Wang, Department of Pathology, The First Affiliated Hospital and College of Basic Medical Sciences, China Medical University, Shenyang 110001, P.R. China
E-mail: cmuwangliang@qq.com

Key words: granular cell tumor, immunohistochemistry, FISH, TFE3, gene rearrangement

Introduction

Granular cell tumor (GrCT) is a type of benign and rare neoplasm, which accounts for approximately 0.5% of soft tissue tumors (1). GrCTs are also known as granular cell schwannoma, granular cell nerve sheath tumor, granular cell myoblastoma and Abrikossoff tumor (2). Previous studies are in favor of neural/Schwann cell origin of GrTC (1-4), however, its exact histogenesis remains unclear (2). GrCTs usually occur in adults between 40 and 60 years of age, are mainly observed in women (ratio women/men, 2:1) and are more prevalent in African-American populations (3,4). Most GrCTs are presented as painless, solitary and circumscribed nodules, of <3 cm in diameter, which essentially occur in the tongue, esophagus, skin, muscle or subcutaneous tissues (3). GrCT can however appear in internal organs of the respiratory, urinary tract or central nervous systems (5). Malignant GrCT is extremely rare and accounts for approximately 0.5-2.0% of all GrCTs (6), with a mortality rate less than 40% and a poor prognosis (7). Malignant GrCTs may cause local recurrence and metastasis in regional lymph nodes, lungs and bones (3,8).

Histologically, GrCTs are characterized by large, oval to round cells with abundant granular eosinophilic cytoplasm (2-4). However, this granular eosinophilic cytoplasm is not unique to GrCTs and similar features are observed in malignant GrCTs and other soft tissue tumors such as schwannoma and oncocytoma (2-4). Distinguishing malignant from benign GrCTs is difficult, and the diagnostic of malignancy must be confirmed by histopathological examination. Although the malignancy criteria remain unclear (9,10), the Fanburg-Smith criteria represent a useful tool that is widely used (11). These criteria include spindling cells, necrosis, large and vesicular nuclei, increased mitotic activity and nuclear to cytoplasmic ratio and pleomorphism. A combination of at least three of these criteria is necessary to determine a GrCT malignancy. The most common misdiagnosis of GrCT is alveolar soft part sarcoma (ASPS) (11). Previous studies have reported that malignant GrCTs were mainly ASPS (2,11).

Although transcription factor E3 (TFE3) is a useful marker for ASPS, the overlap of immunohistochemical staining patterns of TFE3 in other tumors has been addressed along with the increase in case reports (12-16). Previous studies reported that TFE3 is overexpressed in GrCTs (17,18).

In the present study, 42 benign cases of GrCTs and 3 cases of malignant GrCTs were studied. All cases were re-evaluated according to the Fanburg-Smith criteria (11). Immunostaining and fluorescence *in situ* hybridization (FISH) were performed to detect the intensity and expression pattern of TFE3 and to determine whether TFE3 overexpression was caused by TFE3 gene rearrangement.

Materials and methods

Clinical specimens. The present study included 42 benign cases of GrCTs and three cases of malignant GrCTs obtained from patients in three medical centers of Northeast China (The First Affiliated Hospital of China Medical University, the 202nd Hospital of People's Liberation Army of China and the Cancer Hospital of Liaoning Province). All patients were Chinese and were recruited between January 2001 and March 2013 with long-term follow-up data of recurrence and survival (20-214 months). Four cases of ASPS and four cases of Xp11.2 translocation-associated renal cell carcinoma (RCC) were also selected from the database of the First Affiliated Hospital of China Medical University as the positive controls to evaluate TFE3 expression and gene fusion status. Corresponding medical records of all cases were traced. The hematoxylin and eosin (H&E)-stained and immunohistochemical (IHC) slides were analyzed by three independent pathologists. Patient medical records, including basic information, clinical manifestations, therapy and prognosis were reviewed and analyzed in Table SI. Ethical approval for this study was obtained from the institutional ethic review boards of all three medical centers.

H&E and IHC staining. The tumor and the tumor-adjacent tissues were isolated during routine surgeries and fixed in 10% formalin at room temperature for 24 h and embedded in paraffin. Sections (4 μm) were cut from each paraffin block from one patient. One section was stained with H&E, whereas other sections were used for IHC. Briefly, sections were deparaffinized and rehydrated with decreasing ethanol gradient (100, 95, 80 and 70%). Longitudinal sections (5 μm) were stained with hematoxylin for 5 min at room temperature, dipped five times in 1% acid ethanol (1% HCl in 70% ethanol) and washed with distilled water. Sections were then stained with eosin for 3 min, dehydrated with increasing ethanol gradient (70, 80, 95 and 100%) and cleared in xylene. IHC staining was performed using the streptavidin-peroxidase system (Ultrasensitive; MaiXin Inc.) according to the manufacturer's instructions. Briefly, the antigen retrieval was performed by heating sections to 100˚C with citrate buffer (Fuzhou Maixin Biotech Co., Ltd.). The sections were then blocked with 10% goat serum (Fuzhou Maixin Biotech Co., Ltd.) at 37˚C for 1 h. Sections were incubated with commercially available prediluted monoclonal antibodies against TFE3 (cat. no. RMA-0663), vimentin (cat. no. RMA-0547), S100 (cat. no. KIT-0007), serum neuron specific enolase (NSE) (cat. no. MAB-0791), CD68 (cat. no. KIT-0026), phosphohistone H3 (PHH3) (cat. no. RAB-0693), calretinin (cat. no. RMA-0524), inhibin-α (cat. no. MAB-0801) and Ki-67 (cat. no. RMA-0542) (Fuzhou Maixin Biotech Co., Ltd.) at 4˚C overnight, and with the biotinylated goat anti-rabbit IgG secondary antibody at 37˚C for 30 min (1:100; cat. no. KIT-9710; Fuzhou Maixin Biotech Co., Ltd.). Sections were washed three times with PBS, incubated with horseradish peroxidase-conjugated streptavidin-biotin at 37˚C for 30 min (cat. no. KIT-9710; Fuzhou Maixin Biotech Co., Ltd.) and subsequently stained with 3,3-diaminobenzidine tetra-hydrochloride for 1 min at 25˚C (cat. no. KIT-0014; Fuzhou Maixin Biotech Co., Ltd.). Samples were counterstained with hematoxylin at room temperature for 5 min, dehydrated in alcohol (100-70%), and mounted on slides. Appropriate positive (Xp11.2 translocation-associated RCC) and negative (IgG) control slides were included in the IHC assay. To aid the diagnosis of GrCTs, Periodic acid-Schiff-diastase stains were used to stain granules that were recorded as coarsely granular, or demonstrated focal rod-shaped or globular crystalline cytoplasmic inclusions. All slides were evaluated by three independent pathologists (LW, YL and QCL) using Olympus BX51 light microscope (Olympus Corporation; magnification, x200) and scored as either positive or negative based on the presence of specific staining in the appropriate subcellular compartments for each marker. Nuclear staining was evaluated for TFE3, Ki-67 and PHH3, cytoplasmic staining was evaluated for CD68, NSE and inhibin, and nuclear and/or cytoplasmic staining was evaluated for S-100 and calretinin.

Break-apart FISH assay. Paraffin-embedded tissues were cut into 4-μm thick sections. H&E sections were used to confirm that tumor cells were sufficient in number (>60) for FISH analysis and to determine the area to be analyzed. The sections were deparaffinized in xylene twice for 10 min, dehydrated twice with 100% ethanol at room temperature and then pretreated using the ZytoLight FISH-Tissue Implementation kit (cat. no. Z-2028-20; ZytoVision). Slides were digested for 36 min with pepsin (0.5 mg/ml) at 37˚C. TFE3 FISH was performed using ZytoLight® SPEC TFE3 dual color break-apart probe (cat. no. Z-2109-200; ZytoVision). Briefly, slides were incubated for 15 min in pre-warmed citric acid solution (0.1 mM) at 98˚C and dehydrated in 70, 90 and 100% ethanol sequentially, for 1 min each at room temperature. Slides were incubated with the probe (5 μl) overnight at 42˚C in a humidified chamber. Post-hybridization washes were performed in 1X wash buffer A at 37˚C for 10 min (cat. no. Z-2028-20; ZytoVision). Slides were air-dried in the dark and counterstained with 10 μl 4,6-diamidino-2-phenylindole (DAPI)/Antifade-Solution (cat. no. Z-2028-20; ZytoVision) at room temperature for 10 min. All slides were kept at 4˚C in the dark following hybridization. Analysis was performed using a Nikon 80i fluorescence microscope (Nikon Corporation) and IMSTAR Pathfinder Workstation (IMSTAR S.A.) equipped with single and dual band exciters for texas red (561 nm), spectrum green (488 nm) and DAPI (350 nm). Only individual and well-delineated cells were scored. Overlapping cells were excluded from the analysis. Approximately 60 tumor cell nuclei were analyzed in the targeted regions by two independent pathologists. A normal nuclei would exhibit two fusion signals, one which reflects intact TFE3 alleles in a female individual, and one fusion signal that reflects intact TFE3 allele in a male individual, whereas the TFE3 'break-apart' results in

A

tumor location	counts
breast	4
esophagus	3
oropharynx	3
palate	3
subcutaneous tissue	18
thyroid	3
tongue	7
vocal cord	2
vulva	2

B

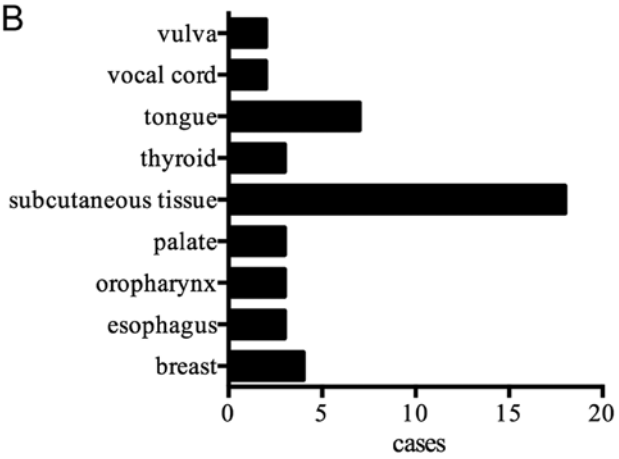

Figure 1. Tumor location distribution for the cases included in the present study. The common sites for granular cell tumors were subcutaneous tissue of the trunk (18 cases) and tongue (7 cases). Other sites included thyroid (3 cases), breast (4 cases), oropharynx (3 cases), palate (3 cases), esophagus (3 cases), vocal cord (2 cases) and vulva (2 cases).

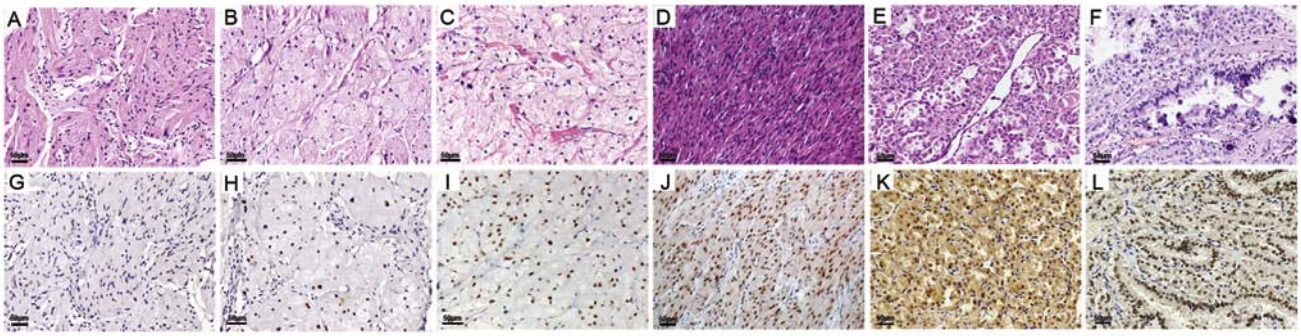

Figure 2. Representative images of GrCTs following TFE3 immunohistochemical staining. (A-C) Representative H&E staining images for the benign GrCTs and (G-I) corresponding TFE3 staining images. (G) Negative, (H) focal positive and (I) diffuse and marked positive TFE3 staining. Representative H&E staining images for (D) the malignant GrCTs with (J) diffuse and marked positive staining for TFE3. Representative H&E staining images for positive controls: ASPS (E) and Xp11.2 translocation-associated RCC (F), with corresponding TFE3 staining images (K and L). Magnification, x200. GrCTs, granular cell tumors; TEF3, transcription factor E3.

two split red and green signal pattern. For TFE3 break-apart signal patterns, 1 red/1 green/1 fusion (yellow) was the most common positive pattern for a balanced TFE3 translocation in a female individual, whereas the signal pattern 1 red/1 green was the most common positive pattern for a balanced TFE3 rearrangement in a male individual. Unbalanced translocations in a female individual yielded a 1 red/2 fusion pattern. To be scored as a break-apart and to avoid false positive, the signals had to be separated by >2 signal diameters. To avoid false negative in a 4-μm section where red or green signal were out of the visible plane of section, a minimum of 60 nuclei were evaluated per case.

Statistical analysis. SPSS version 22.0 for windows (SPSS, Inc.) was used for all analyses. The Pearson's χ^2 test and Likelihood ratio were used to determine the correlations between TFE3 and clinicopathological characteristics of patients with GrCT. Tumors located in tongue, oropharynx and esophagus were grouped in the non-subcutaneous group, whereas tumors located in breast, thyroid and vulva were grouped in the subcutaneous group. $P<0.05$ was considered to indicate statistically significant differences.

Results

Clinicopathological characteristics of patients with GrCT. The 45 patients diagnosed with benign GrCTs (42 patients) or malignant GrCTs (3 patients) between January 2001 and March 2013 included in this study comprised 30 inpatient cased and 15 consultation cases, with a male-to-female ratio of 17:28. The median age of patients was 49 years (range, 9-66 years). Tumors essentially originated from subcutaneous tissue of the trunk (18 cases) and tongue (7 cases), and other sites including thyroid, breast, oropharynx, palate, esophagus, vocal cord and vulva (Fig. 1). Amongst the patients, 34 presented a single swelling mass without symptoms, eight patients felt some pain, three patients were asymptomatic and diagnosed incidentally during endoscopic examination and the remaining three patients were diagnosed with GrCT incidentally during thyroid carcinoma surgery. Tumor size ranged from 0.5 to 6 cm. Amongst the patients, 42 diagnosed with GrCTs accepted the tumor resection. The other three cases were diagnosed with malignant GrCTs via histopathological examination using the Fanburg-Smith criteria (11) and accepted the extended resection. According to long-term follow-up (range, 20-214 months),

43 patients were declared disease-free following tumor resection, and only two patients experienced recurrence (Table SI).

GrCTs in subcutaneous tissue exhibited a higher ratio of TFE3 expression compared with those in other sites. Among the 45 cases of GrCTs, 21 cases (47%) exhibited negative TFE3 staining (Fig. 2A and G) whereas 13 cases (29%) demonstrated focal positive TFE3 staining (Fig. 2B and H). Only 11 cases (24%) of GrCTs in the current study exhibited diffuse and marked positive TFE3 staining (Fig. 2C and I). TFE3 positive staining was observed in all common histological patterns (spindle, solid and acinar-like) for GrCTs. Among the three malignant GrCTs case, one exhibited TFE3 overexpression (Fig. 2D and J). In the control group, the four cases of ASPS (Fig. 2E) and four cases of Xp11.2 translocation-associated RCC (Fig. 2F) exhibited diffusely positive nuclear staining for TFE3 (Fig. 2K and L).

The results demonstrated that, among the 11 TFE3-positive cases, eight cases occurred in the subcutaneous tissue of the trunk, whereas the remaining three positive cases occurred in thyroid, breast and vulva, which were superficial locations that were easily accessible. However, none of the cases from the tongue, oropharynx, palate or esophagus exhibited positive staining for TFE3. It seems that GrCTs had a higher ratio (8/45, 18%) of TFE3 expression in subcutaneous tissues compared with those in other sites. In addition, GrCTs of larger size (≥3 cm) exhibited a higher ratio for TFE3 overexpression compared with the smaller GrCTs. No association was observed between the TFE3 expression and age, sex, histological types or growth patterns in the GrCTs (Table I).

The results of IHC staining performed routinely for rendering GrCTs diagnosis (staining for S100, NSE, CD68, PHH3, calretinin, inhibin-α and Ki-67) were not shown, as they were not related to TFE3 overexpression or gene rearrangement.

TFE3 overexpression in GrCTs was not caused by gene rearrangement or amplification. Considering that some TFE3 rearrangement neoplasm may exhibit false-negative in immunohistochemical staining, a break-apart FISH assay was performed in the 45 cases of GrCTs. The results demonstrated that none of the cases exhibited TFE3 rearrangement or amplification, as presented in Fig. 3A and B. Furthermore, four cases of ASPS and three cases of Xp11.2 translocation-associated RCC exhibited TFE3 gene translocation (Fig. 3C and D).

Table I. Correlation of TFE3 expression with clinicopathological characteristics in GrCTs.

	TFE3		
Characteristics	Negative	Positive	P-value
Age, years			0.793
<49	17	5	
≥49	17	6	
Sex			0.920
Male	13	4	
Female	21	6	
Site			<0.001[a]
Non-subcutaneous tissue	18	0	
Subcutaneous tissue	16	11	
Size, cm			0.003[a]
<3	32	6	
≥3	2	5	
Histological type			0.720
Benign	32	10	
Malignant	2	1	
Growth pattern			0.245
Expansive	32	9	
Invasive	2	2	

[a]P<0.05, indicating statistical significance. GrCTs, granular cell tumors; TFE3, transcription factor E3.

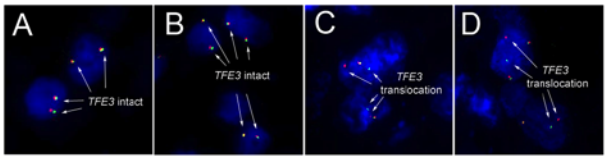

Figure 3. Representative images of GrCTs following TFE3 break-apart FISH assay. GrCTs demonstrated (A and B) negative results with fusional signals. (C) Positive break-apart signals were identified for TFE3 translocation in RCC and (D) ASPS as positive controls. DAPI stained for cell nuclei (blue). Red and green fusional signals indicated intact TFE3, red and green separated signals indicated TFE3 translocation. Magnification, x1,000. GrCTs, granular cell tumors; TEF3, transcription factor E3; RCC, renal cell carcinoma; ASPS, alveolar soft part sarcoma.

Discussion

TFE3 is a member of the helix-loop-helix family of transcription factors and is considered a useful marker in ASPS and Xp11.2 translocation cancers diagnostics, including Xp11.2 translocation-associated RCC, Xp11 translocation perivascular epithelioid cell tumor and melanotic Xp11 translocation RCC (19). Chamberlain *et al* (17) studied the immunophenotypic comparison between ASPS and GrCTs, and revealed that 91% of GrCTs cases have a diffusive and marked positivity for TFE3. Excessive dependence on TFE3 expression, especially in the biopsy with limited tissues, could lead to high ratio of positivity and subsequently to GrCTs misdiagnoses into ASPS or other neoplasms. Schoolmeester *et al* (18) performed FISH on six cases of GrCTs with TFE3 positive immunostaining, and reported that none of the six cases had Xp11.2 rearrangement. This result suggested that genetic alterations, other than TFE3 translocation, may cause TFE3 overexpression in GrCTs. However, these two studies used a limited number of cases, which raised certain points, including whether TFE3 is also overexpressed in Chinese GrCT patients, and whether a small cohort of samples leads to the negative results of FISH.

The present study was designed to answer these questions. The results demonstrated that diffusive and marked nuclear positive TFE3 staining was identified in only 11/45 (24%) cases. Focal or weak positive TFE3 staining was identified in 13/45 (29%) cases. The remaining 21 cases were negatively stained for TFE3. The positive proportion of TFE3 staining in each group was summarized in Table II. The proportions

Table II. Proportions of TFE3 in patients with granular cell tumors.

	The First Affiliated Hospital of China Medical University	The 202nd Hospital of People's Liberation Army of China	Cancer Hospital of Liaoning Province	Chamberlain *et al* (17)	Schoolmeester *et al* (18)
TFE3					
Negative	25	5	4	1	0
Positive	8	2	1	10	6
Positive proportion, %	24	29	20	91	100

TFE3, transcription factor E3.

of positive staining in the cases obtained for the three medical centers in the present study were close, but were much lower than those from the studies of Chamberlain *et al* (17) and Schoolmeester *et al* (18). In addition, the results demonstrated that the cases of TFE3 overexpression mainly occurred in subcutaneous tissues (11/27, 41%), whereas no cases from the tongue, esophagus or oropharynx exhibited TFE3 overexpression. Compared with the immunohistochemical staining results from the aforementioned studies (17,18), TFE3 positive ratio in the present study was much lower. This discrepancy may have been due to ethnic differences or laboratory variation. In the present study, the overnight incubation staining protocol reported by Argani *et al* (12) was used, whereas the aforementioned groups performed TFE3 staining with autostainers (17,18). Notably, Argani *et al* (12) reported a strong nuclear TFE3 labeling in 25% of GrCTs cases (2/8), which was similar to the results from the present study. In addition, this study reported a difference in TFE3 expression between the subcutaneous tissue and non-subcutaneous tissue (tongue, esophagus or oropharynx).

Considering the possibility of false-negative TFE3 staining in some cases, the TFE3 break-apart FISH assay was performed for all samples. No TFE3 rearrangement or amplification was identified, neither in the TEF3 positively nor the negatively stained cases. These results were consistent with those from Schoolmeester *et al* (18) and provided data from a much larger cohort size.

To date, at least nine different types of neoplasms have been reported with TFE3 immunoreactivity (12,14-16,20). The high levels of TFE3 protein expression are commonly due to promoter enhancement, genetic amplification, translocation or dysfunction in protein degradation. Together with those from Chamberlain *et al* (17) and Schoolmeester *et al* (18), the results from the present study indicated that, unlike in ASPS, TFE3 overexpression in GrCTs was not caused by genetic translocation, which suggested that other types of genetic alteration may be involved. As a possible explanation, Chamberlain *et al* (21) proposed that aberrant nuclear TFE3 accumulation could be caused by organelles or intracellular metabolic signaling pathways dysfunctions, which could lead to the typical cytoplasmic accumulation of phagolysosomes in GrCTS. Previous studies reported that TFE3 is involved in lysosome/phagosome synthesis regulation and in the Golgi stress response (22-27). The aberrant TFE3 expression may therefore only represent the degenerative change due to lysosomes cytoplasmic accumulation in these neoplasms and may not be the unique event of GrCTs.

In conclusion, the present study described the clinical and pathological characteristics of 45 GrCTs cases via TFE3 IHC and FISH assay. The results revealed TFE3 overexpression and gene alteration in a large cohort of GrCTs cases. However, TFE3 overexpression in Chinese patients was lower than that in occidental patients according to previous studies, and was not associated with gene rearrangement. Further studies including more cases are required to determine the influence of TFE3 overexpression in GrCTs.

Acknowledgements

Not applicable.

Funding

This study was supported by the National Natural Science Foundation of China (grant no. 81602019 to JM), the Natural Science Foundation of Liaoning Province (grant no. L2013292 to YL) and the Liaoning Technology Research Fund for Social Development and Industrialization (grant no. 2017225010 to LW).

Availability of data and materials

All data generated or analyzed during this study are included in this published article.

Authors' contributions

JM, YZ, WCSC, QCL, XSQ and EHW designed the study. QZ and CW performed the immunochemical staining. JW and XL performed the FISH assay. LW and YL analyzed the data and wrote the manuscript. JM, YZ, WCSC, QCL, XSQ and EHW revised the manuscript. All authors read and approved the final manuscript.

Ethical approval and consent to participate

Ethical approval for this study was obtained from the institutional ethic review boards of the First Affiliated Hospital of China Medical University, the 202nd Hospital of People's Liberation Army of China, and Cancer Hospital of Liaoning province, respectively. Written informed consent was provided by all patients or their guardians.

Patient consent for publication

Informed consents were obtained from all patients for the publication of their cases and any associated images. A copy of the written consent is available for review by the Editor-in-Chief of this journal.

Competing interests

The authors declare that they have no competing interests.

References

1. Rose B, Tamvakopoulos GS, Yeung E, Pollock R, Skinner J, Briggs T and Cannon S: Granular cell tumours: A rare entity in the musculoskeletal system. Sarcoma 2009: 765927, 2009.
2. Rosai J: Tumors of Uncertain Cell Type Granular Cell Tumor. In: Rosai and Ackerman's Surgical Pathology - 2 Volume Set. 10th edition. Expert consult. Mosby/Elsevier, Maryland Heights, MI, USA, pp 2181-2182, 2007.
3. Mindell ER: Benign Tumors of Peripheral Nerves Section Granular Cell Tumor. In: Enzinger and Weiss's Soft Tissue Tumors. Saunders/Elsevier, Philidelphia, PA, USA, pp 838-845, 2014.
4. Fletcher CDM, Bridge JA, Hogendoorn P and Mertens F: WHO Classification of Tumours of Soft Tissue and Bone. 4th edition. IARC Press, 2013.
5. Lack EE, Worsham GF, Callihan MD, Crawford BE, Klappenbach S, Rowden G and Chun B: Granular cell tumor: A clinicopathologic study of 110 patients. J Surg Oncol 13: 301-316, 1980.
6. Elkousy H, Harrelson J, Dodd L, Martinez S and Scully S: Granular cell tumors of the extremities. Clin Orthop Relat Res 191-198, 2000.
7. Torrijos-Aguilar A, Alegre-de Miquel V, Pitarch-Bort G, Mercader-Garcia P and Fortea-Baixauli JM: Cutaneous granular cell tumor: A clinical and pathologic analysis of 34 cases. Actas Dermosifiliogr 100: 126-132, 2009 (In Spanish).
8. Jardines L, Cheung L, LiVolsi V, Hendrickson S and Brooks JJ: Malignant granular cell tumors: Report of a case and review of the literature. Surgery 116: 49-54, 1994.
9. Nasser H, Ahmed Y, Szpunar SM and Kowalski PJ: Malignant granular cell tumor: A look into the diagnostic criteria. Pathol Res Pract 207: 164-168, 2011.
10. Machado I, Cruz J, Lavernia J and Llombart-Bosch A: Solitary, multiple, benign, atypical, or malignant: The 'granular cell tumor' puzzle. Virchows Archiv 468: 527-538, 2016.
11. Fanburg-Smith JC, Meis-Kindblom JM, Fante R and Kindblom LG: Malignant granular cell tumor of soft tissue: Diagnostic criteria and clinicopathologic correlation. Am J Surg Pathol 22: 779-794, 1998.
12. Argani P, Lal P, Hutchinson B, Lui MY, Reuter VE and Ladanyi M: Aberrant nuclear immunoreactivity for TFE3 in neoplasms with TFE3 gene fusions: A sensitive and specific immunohistochemical assay. Am J Surg Pathol 27: 750-761, 2003.
13. Folpe AL, Mentzel T, Lehr HA, Fisher C, Balzer BL and Weiss SW: Perivascular epithelioid cell neoplasms of soft tissue and gynecologic origin: A clinicopathologic study of 26 cases and review of the literature. Am J Surg Pathol 29: 1558-1575, 2005.
14. Xia QY, Wang Z, Chen N, Gan HL, Teng XD, Shi SS, Wang X, Wei X, Ye SB, Li R, *et al*: Xp11.2 translocation renal cell carcinoma with NONO-TFE3 gene fusion: Morphology, prognosis, and potential pitfall in detecting TFE3 gene rearrangement. Mod Pathol 30: 416-426, 2017.
15. Hodge JC, Pearce KE, Wang X, Wiktor AE, Oliveira AM and Greipp PT: Molecular cytogenetic analysis for TFE3 rearrangement in Xp11.2 renal cell carcinoma and alveolar soft part sarcoma: Validation and clinical experience with 75 cases. Mod Pathol 27: 113-127, 2014.
16. Calió A, Grignon DJ, Stohr BA, Williamson SR, Eble JN and Cheng L: Renal cell carcinoma with TFE3 translocation and succinate dehydrogenase B mutation. Mod Pathol 30: 407-415, 2017.
17. Chamberlain BK, McClain CM, Gonzalez RS, Coffin CM and Cates JM: Alveolar soft part sarcoma and granular cell tumor: An immunohistochemical comparison study. Hum Pathol 45: 1039-1044, 2014.
18. Schoolmeester JK and Lastra RR: Granular cell tumors overexpress TFE3 without corollary gene rearrangement. Hum Pathol 46: 1242-1243, 2015.
19. Argani P, Zhong M, Reuter VE, Fallon JT, Epstein JI, Netto GJ and Antonescu CR: TFE3-fusion variant analysis defines specific clinicopathologic associations among Xp11 translocation cancers. Am J Surg Pathol 40: 723-737, 2016.
20. Argani P, Aulmann S, Illei PB, Netto GJ, Ro J, Cho HY, Dogan S, Ladanyi M, Martignoni G, Goldblum JR and Weiss SW: A distinctive subset of PEComas harbors TFE3 gene fusions. Am J Surg Pathol 34: 1395-1406, 2010.
21. Chamberlain BK, McClain CM, Gonzalez RS, Coffin CM and Cates JM: Granular cell tumors overexpress TFE3 without corollary gene rearrangement-reply. Hum Pathol 46: 1243, 2015.
22. Martina JA, Diab HI, Li H and Puertollano R: Novel roles for the MiTF/TFE family of transcription factors in organelle biogenesis, nutrient sensing, and energy homeostasis. Cell Mol Life Sci 71: 2483-2497, 2014.
23. Martina JA, Diab HI, Lishu L, Jeong-A L, Patange S, Raben N and Puertollano R: The nutrient-responsive transcription factor TFE3 promotes autophagy, lysosomal biogenesis, and clearance of cellular debris. Sci Signal 7: ra9, 2014.
24. Roczniak-Ferguson A, Petit CS, Froehlich F, Qian S, Ky J, Angarola B, Walther TC and Ferguson SM: The transcription factor TFEB links mTORC1 signaling to transcriptional control of lysosome homeostasis. Sci Signal 5: ra42, 2012.
25. Taniguchi M, Nadanaka S, Tanakura S, Sawaguchi S, Midori S, Kawai Y, Yamaguchi S, Shimada Y, Nakamura Y, Matsumura Y, *et al*: TFE3 is a bHLH-ZIP-type transcription factor that regulates the mammalian golgi stress response. Cell Struct Funct 40: 13-30, 2015.
26. Tsuji K, Ishikawa Y and Imamura T: Technique for differentiating alveolar soft part sarcoma from other tumors in paraffin-embedded tissue: Comparison of immunohistochemistry for TFE3 and CD147 and of reverse transcription polymerase chain reaction for ASPSCR1-TFE3 fusion transcript. Hum Pathol 43: 356-363, 2012.
27. Williams A, Bartle G, Sumathi VP Meis JM, Mangham DC, Grimer RJ and Kindblom LG: Detection of ASPL/TFE3 fusion transcripts and the TFE3 antigen in formalin-fixed, paraffin-embedded tissue in a series of 18 cases of alveolar soft part sarcoma: Useful diagnostic tools in cases with unusual histological features. Virchows Arch 458: 291-300, 2011.