RESEARCH ARTICLE

Transducer-like Enhancer of Split 1 as a Novel Immunohistochemical Marker for Diagnosis of Synovial Sarcoma

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Abstract

Background: Synovial sarcoma is a mesenchymal neoplasm that accounts for around 10% of all soft tissue sarcomas. The diagnosis of synovial sarcoma can be a challenging task, particularly with small biopsy specimens. Aim: We investigated transducer-like enhancer of split 1 (TLE1), monoclonal antibody, expression by immunohistochemical analysis in a group of 74 synovial sarcoma cases, 20 cases of MPNST, 12 cases of neurofibroma, 15 cases of schwannoma, 5 cases of MFH, 10 cases of lieomyosarcoma and 10 cases of solitary fibrous tumor. Materials and Methods: Whole tissue sections were examined: (39 biphasic and 35 monophasic). Nuclear immunoreactivity was scored as negative (<5% of cells positive), 1+(mild /5-25%), 2+ (moderate/25-50%), and 3+ (strong >50%). Results: Overall, 71 (96%) of 74 synovial sarcomas were positive for TLE1, including 37 biphasic (95%) and 34 monophasic (97%) tumors. Other spindle cell tumors showed very low or absent staining of TLE1. Conclusions: We conclude that TLE1 is a sensitive marker and can be a useful diagnostic marker for synovial sarcoma, particularly the monophasic forms.

Keywords: TLE-1 - synovial sarcomas - spindle cell sarcomas

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Introduction

Sarcomas are rare (less than 1 % of all malignancies) mesenchymal neoplasms that arise in bone and soft tissues. The tumors are usually of mesodermal origin, although a few are derived from neuroectoderm, and they are biologically distinct from the more common epithelial malignancies. Sarcomas affect all age groups, 15% being found in children younger than age 15 and 40% occurring after age 55. Soft tissue include muscle, tendons, fat, fibrous tissue, synovial tissue, vessels and nerves (Sadighi & Raafat, 2003). Synovial sarcoma is a clinically and morphologically well defined entity that, despite its name, is extremely uncommon in joint cavities and is encountered in areas with no apparent relation to synovial structures. It occurs primarily in the particular regions of the extremities, usually in close association with tendon sheathes, bursae and joint capsules It represents 5-10% of of all soft tissue sarcomas (Fisher, 1998).

Histologically, there are two major categories of synovial sarcoma: biphasic and monophasic types. Biphasic synovial sarcoma has distinct epithelial and spindle cell components in varying proportions. The monophasic synovial sarcoma is diagnosed by small spindle cell sarcoma areas (Goldblum, 2014).

Diagnosing biphasic synovial sarcoma is generally straightforward, owing to distinctive histologic features. However, monophasic synovial sarcomas can be difficult to distinguish from other spindle cell sarcomas as malignant peripheral nerve sheath tumor (MPNST), neurofibroma, schwannoma, malignant fibrous histiocytoma (MFH), lieomyosarcoma and solitary fibrous tumor. A carefully selected immunohistochemical panel can aid in differential diagnosis but does not always yield a definitive diagnosis. Ancillary methods such as reverse transcription-polymerase chain reaction, fluorescence in situ hybridization, and traditional karyotyping may be used to demonstrate the specific translocation t(X;18)(SS18-SSX1/2) and thereby confirm the diagnosis. However, such techniques are expensive, are not available in many laboratories, and may require fresh or frozen tissue. Thus, there has been continued interest in the development of novel immunohistochemical markers for diagnosis (Coindre et al., 2003; Kawauchi et al., 2005; Amary et al., 2007).

A number of gene expression profiling studies of synovial sarcoma have identified overexpression of a number of members of the Transduction-Like Enhancer (TLE) gene family in synovial sarcoma (Terry et al., 2007). TLE1 is one of 4 TLE genes that encode human transcriptional repressors homologous to the Drosophila corepressor groucho. TLE proteins are expressed in embryogenesis where they are involved in developmental processes including neurogenesis, body patterning, and hematopoesis particularly TLE1. For that reason, immunostains for TLE1 protein may be helpful in the recognition of synovial sarcoma (Yao et al., 2000).

Here, we investigate the protein expression of TLE in

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synovial sarcoma using immunohistochemistery, to assess the value of TLE as a diagnostic marker for this sarcoma.

Materials and Methods

Whole tissue sections from cases diagnosed between 2011 and 2014 were retrieved from the archives of the Department of Pathology, Faculty of medicine, Tanta university and the private laboratories. A total of 74 synovial sarcomas were evaluated and divided into the following: 39 monophasic and 35 biphasic. TLE-1 immunostaining was also carried out on 20 cases of MPNST, 12 cases of neurofibroma, 15 cases of schwannoma, 5 cases of MFH, 10 cases of lieomyosarcoma and 10 cases of solitary fibrous tumor. The study samples were available in form of formalin-fixed, paraffin-embedded tissue blocks, with or without stained slides (52), biopsy specimens (15) and tumor resection specimens (7). Hematoxylin and eosin stained (H & E) sections were accessible in all cases. All synovial sarcoma cases had classic clinical presentation, histopathological features and IHC profile, including at least positive expression of the IHC markers, namely EMA and/ or CK, BCL2 and negative expression of CD34.
Immunohistochemical staining for TLE1 was performed following antigen retrieval (PBS buffer; pH 7.2) performed by immunoperxoidase method using a rabbit polyclonal antibody (1:200 dilution; Labvision Biotechnology) and Universal detection kit, labvision, including 3'-3'-diaminobenzidine tetrahydrochloride (DAB) as the chromogen. Nuclear immunoreactivity was graded according to extent as 0 negative (<5% of cells positive), 1+(mild /5-25%), 2+ (moderate/25-50%), and 3+ (strong >50%) (Terry et al, 2007).

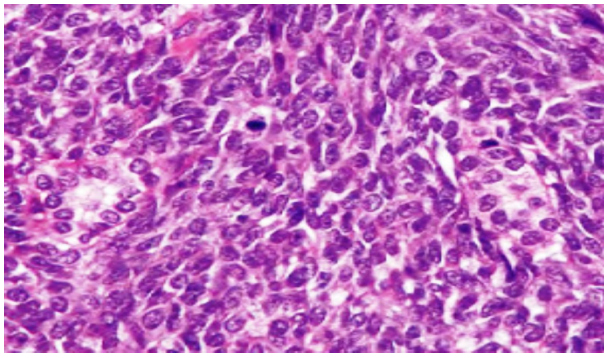

Figure 1. Biphasic Synovial Sarcoma Composed of an Admixture of Monomorphous Spindle Cells and Glandular Structures (H&E, ×400)

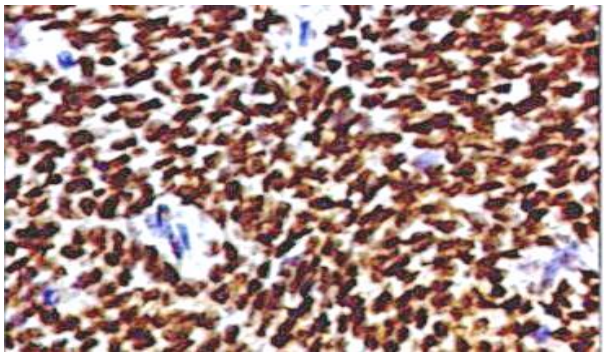

Figure 2. Tumor Cells in Both Components Show Diffuse Nuclear Staining for Transducer-like Enhancer of Split 1, with somewhat Stronger Staining in the Epithelial Component (×400)

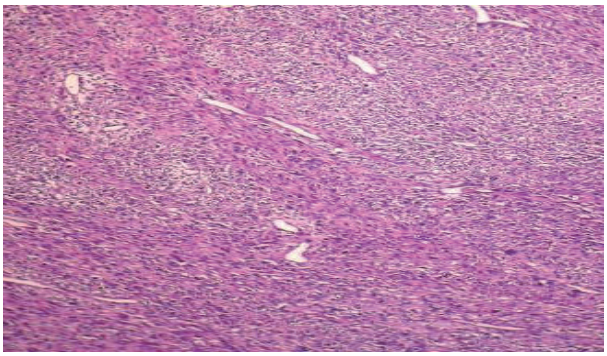

Figure 3. Monophasic Synovial Sarcoma Composed of Highly Cellular Short Fascicles of Uniform Spindle Cells (H&E ×100)

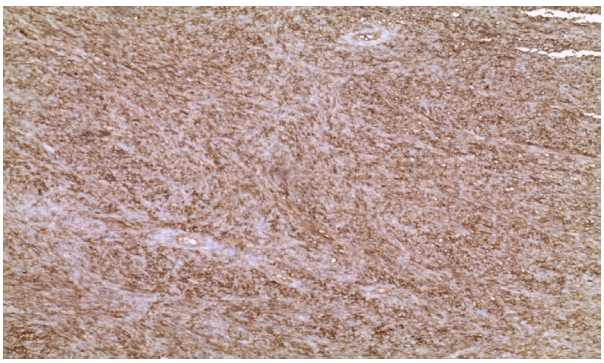

Figure 4. Neoplastic Cells Show Strong, Diffuse Nuclear Reactivity for Transducer-like Enhancer of Split 1 (×100)

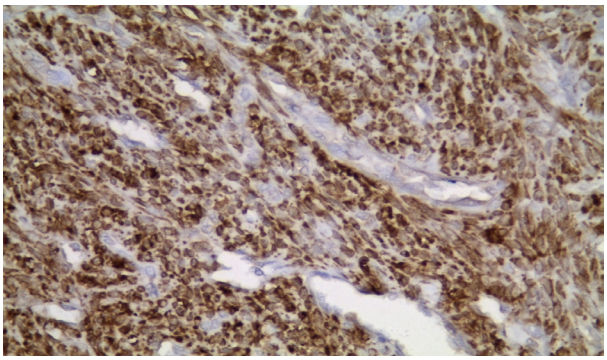

Figure 5. Neoplastic Cells Show Strong, Diffuse Nuclear Reactivity for Transducer-like Enhancer of Split 1 (× 400)

Table 1. Summary for Immunohistochemical Staining Results of TLE1

Tumor type	No. of cases	No. (%) positive cases
Biphasic synovial sarcoma	39	95%
Monophasic synovial sarcoma	35	97%
MPNST	20	10%
Neurofibroma	12	8.30%
Schwannoma	15	6.70%
Solitary fibrous tumor	10	10%
MFH	5	0%
Leiomyosarcoma	10	0%

Table 2. Detailed Immunohistochemical Results of TLE-1 in Synovial Sarcomas

Tumor type	Scoring			
	0 (Negative)	+1 (Mild)	+2 (Moderate)	+3 (Strong)
Biphasic synovial sarcoma	2 (5.1%)	2 (5.1%)	12 (30.8%)	23 (59%)
Monophasic synovial sarcoma	1 (2.9%)	3 (8.6%)	11 (31.4%)	20 (57.1%)

Table 3. Sensitivity and Specificity of GPC3 and GS

	Sensitivity	Specificity	PPV	NPV	Accuracy
TLE1	96%	85%	78%	95%	89%

Results

Nuclear immunoreactivity was observed in 71 synovial sarcoma cases (96%), including 37 (95%) of 39 of the biphasic type (Figure 1, 2), 34 (97%) of 35 of the monophasic type (Figure 3, 4, 5). 23/37 biphasic and 20/34 monophasic synovial sarcoma cases showed strong nuclear positivity (+3) in more than 50% of tumor cells. 12/37 biphasic and 11/34 monophasic synovial sarcoma cases showed moderate nuclear positivity (+2) in less 25-50% of tumor cells. 2/37 biphasic and 3/34 monophasic synovial sarcoma cases showed weak nuclear positivity (+1) in 5-25% of tumor cells. While, 2/39 biphasic and 1/35 monophasic synovial sarcoma cases were negative (0) showing positivity in 0-25% of tumor cells. The immunohistochemical staining results for TLE1 in synovial sarcomas are summarized in (Table 1).

Noteworthy, the cases of biphasic synovial sarcoma showed staining in both the epithelial and spindle cell elements but the glandular elements generally showed stronger staining than did the spindle cell component.

TLE staining was low to absent in other spindle cell tumors in the differential diagnosis of synovial sarcoma. MPNST (2/20), neurofibroma (1/12), schwannomas (1/15), solitary fibrous tumour (1/10) were occasionally positive, whereas MFH and leiomyosarcoma were totally negative for TLE1.

Detailed immunohistochemical results for TLE1 in synovial sarcoma, are summarized in (Table 2).

The overall sensitivity and specificity of TLE1 expression for the diagnosis of synovial sarcoma were 96% and 85% respectively (Table 3).

Discussion

Distinguishing synovial sarcoma from other spindle cell tumors can present a diagnostic challenge, particularly in those cases that do not exhibit biphasic histology. In these situations, immunohistochemical markers can be valuable in confirming the diagnosis of synovial sarcoma. Besides, even though synovial sarcoma is an aggressive sarcoma, it is amenable to treatment modalities, including chemotherapy. Hence, its correct identification is vital. Several IHC markers are employed for its objective diagnosis and in differentiating it from its diagnostic mimics. The diagnostic challenge is further amplified with limited biopsy material, where in focal expression, especially of epithelial markers, might be lacking, thereby creating a challenge in exact recognition, especially of monophasic spindle cell subtype of synovial sarcoma. Although an extensive panel of IHC markers is available for diagnosing a synovial sarcoma, there has been no single, fairly specific and sensitive marker (Pelmus et al, 2002).

We performed this immunohistochemical study on 74 synovial sarcomas (39 biphasic and 35 monophasic) using TLE1 immunomarker. Sixty three (85%) of 74 synovial sarcomas were positive for TLE1, including 34 biphasic (87%) and 29 monophasic (83%) tumors. 19/34 biphasic and 18/29 monophasic synovial sarcoma cases showed strong nuclear positivity (+3) in more than 50% of tumor cells. 9/34 biphasic and 7/29 monophasic synovial sarcoma cases showed moderate nuclear positivity (+2) in less 25-50% of tumor cells. 6/34 biphasic and 4/29 monophasic synovial sarcoma cases showed weak nuclear positivity (+1) in 5-25% of tumor cells. While, 5/34 biphasic and 6/29 monophasic synovial sarcoma cases were negative (0) showing positivity in 0-25% of tumor cells. The immunostaining results for other spindle cell tumors showed very low or absent staining compared t that in synovial sarcoma cases.

In approval with the above mentioned results, Foo et al. (2011) observed nuclear immunoreactivity in 60 synovial sarcomas (82%) out of 73 cases, including 22 (79%) of 28 of the monophasic type , 18 (78%) of 23 of the biphasic type.

Supporting the previous results, in 2012, Rekhi et al. demonstrated that TLE1 immunohistochemical staining performed on 42 cases of synovial sarcoma showed 61.9% positive cases of monophasic spindle cell type and 30.9% of biphasic type. They confirmed that TLE1, in view of its high sensitivity may be a useful marker within the optimal IHC panel.

According to Jagdis et al. (2009), TLE1 gave intense, diffuse nuclear staining in 35 of 35 molecularly confirmed synovial sarcoma cases, and was rare to absent in the 73 other soft tissue tumors examined (positive staining was found only in 1 of 43 malignant peripheral nerve sheath tumors, the 1 tested fibrosarcoma, and 1 pleomorphic sarcoma). TLE1 was more sensitive and specific for synovial sarcoma than other currently available immunohistochemical markers including Bcl2, epithelial membrane antigen and cytokeratins.

Terry et al. (2007), demonstrated that TLE expression was a consistent feature of synovial sarcoma with intense and/or diffuse nuclear staining in 91/94 molecularly confirmed synovial sarcomas. Moderate staining is occasionally seen in schwannoma and solitary fibrous tumor/hemangiopericytoma. In contrast, TLE staining was detected much less frequently and at lower levels, if at all, in 40 other mesenchymal tumors. In addition, Knosel et

al. (2010), analysed the immunohistochemical expression of TLE1 in 259 synovial sarcoma cases with the result of nuclear positive staining in 96% of cases.

On the other hand, in contrast to the results discussed before, Kosemehmetoglu et al. (2009), stated that TLE1 expression is by no means specific for synovial sarcoma, being present in a number of tumors, which enter its differential diagnosis, in particular, tumors of peripheral nerve sheath origin. They also added that TLE1 may be of value in the differential diagnosis of synovial sarcoma, but should be used only in the context of a panel of antibodies.

In conclusion, to sum up, although, molecular testing remains the diagnostic gold standard for a synovial sarcoma, TLE1 could be a useful IHC marker, in small biopsies; in cases with classical histopathological features and for dual confirmation, in conjunction with molecular analysis, whenever necessary. Its inclusion in an optimal IHC panel formed by EMA, BCL2 and CD34 along with CK7, for substantiating a histopathological diagnosis of a synovial sarcoma in cases occurring at unusual sites and with variable histopathological features, could reduce further requests for molecular testing, especially for TLE1 negative tumours.

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