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A comparison of the usefulness of nuclear beta-catenin in the diagnosis of desmoid-type fibromatosis among commonly used anti-beta-catenin antibodies

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Running head

Positivity of B-catenin in desmoid tumor

Abbreviations

None declared.

Abstract (198 words)

Desmoid-type fibromatosis (DF) is a locally aggressive but non-metastatic (myo)fibroblastic neoplasm. A hallmark of the tumor is nuclear positivity for beta-catenin in immunohistochemistry due mostly to *CTNNB1* mutations. However, a recent study has reported that even beta-catenin "nuclear-negative" DFs can harbor *CTNNB1* mutations and that the tissue lesions for which the possibility of DF was considered and compared the sensitivity and specificity of nuclear beta-catenin for the diagnosis of DF among commonly used anti-beta-catenin antibodies, i.e., clone beta-catenin 1, 17C2, and 14. We analyzed 26 cases of DF, 28 cases of benign fibroblastic lesions, and 27 cases of other soft tissue tumors. The sensitivity and specificity of nuclear beta-catenin for the diagnosis of DF were different among antibodies; 54% and 98% in clone beta-catenin 1, 85% and 84% in 17C2, and 96% and 62% in 14. IHC of LEF1 showed comparable results with IHC of beta-catenin, with a sensitivity of 88% and specificity of 76%. Additionally, when beta-catenin 1 was used, DFs showed characteristic dotted cytoplasmic staining, often appearing as rings. Our results might be helpful for making a correct diagnosis of DF.

Keywords: 1: Beta-catenin; 2: CTNNB1; 3: Cytoplasm; 4: Desmoid-type fibromatosis; 5: DNA mutational analysis; 6: Immunohistochemistry; 7: Beta-catenin 1; 8: 17C2; 9: 14; 10: LEF1

Main text (2832 words)

Introduction

Desmoid-type fibromatosis (DF) is a (myo)fibroblastic tumor, which typically arises in deep soft tissues in children and young to middle-aged adults. It exhibits infiltrative growth and local recurrence but lacks metastatic potential. The tumor is composed of long sweeping fascicles of spindle cells without significant nuclear atypia in a collagenous stroma containing prominent blood vessels. Immunohistochemically, it exhibits the nuclear accumulation of beta-catenin, a hallmark of the tumor. This finding is based on the genetic abnormalities involving dysregulation/activation of the Wnt signaling pathway, especially mutations of *CTNNB1* (at codon 41 or 45 in exon 3) or *APC*, which result in the translocation of beta-catenin protein from the cytoplasm to the nucleus (1-3). In addition to lacking metastatic potential, because DF can exhibit spontaneous remission or growth arrest, it is often carefully observed rather than surgically resected (4-7). Thus, it is important for pathologists to differentiate DF from bland-looking but potentially metastatic sarcomas. When the tumor exhibits typical pathological findings, especially nuclear positivity for beta-catenin, the diagnosis is straightforward. However, because not all DFs exhibit this finding (1, 8), making a correct diagnosis of DF is sometimes difficult.

Recently Koike et al. reported that some nuclear beta-catenin-negative DFs harbor characteristic *CTNNB1* mutations, and that positivity of nuclear beta-catenin in DF is different between two anti-beta-catenin antibodies (9). Consistent with this report, we recently encountered two cases of DFs that exhibited typical clinical and histological findings but lacked a nuclear expression of beta-catenin despite harboring *CTNNB1* mutation (Figure 1). We also found that both cases exhibited an apparently unique cytoplasmic beta-catenin staining pattern,

namely, a “dotted” cytoplasmic pattern in immunohistochemistry (IHC) with clone beta-catenin 1, the antibody used in daily practice at our institute; we speculated that this pattern might be a feature of DF.

To further address the staining pattern of beta-catenin in DF, we reviewed our archives of soft tissue tumors or tumor-like lesions for which beta-catenin IHC (clone beta-catenin 1) had been performed. We then performed additional immunostaining with two other commonly used anti-beta-catenin antibodies (clone 17C2 and 14) and an anti-LEF1 antibody, and compared the sensitivity and specificity of nuclear beta-catenin in the diagnosis of DF among these antibodies.

Materials and Methods

Case selections

We searched the pathological archives of the Department of Diagnostic Pathology, Kyoto University Hospital, between 2011 and 2020. We first enrolled a total of 83 cases of soft tissue tumors or tumor-like lesions for which DF was suspected or considered as a differential diagnosis, and thus IHC for beta-catenin was assessed. This case series contained the two aforementioned DFs, which harbored *CTNNB1* mutations but were negative for nuclear beta-catenin in IHC with clone beta-catenin 1 (Figure 1). The genetic status of *CTNNB1* in other cases was not examined. We reviewed the representative H&E specimen and the IHC of beta-catenin (clone beta-catenin 1) for all cases. We then performed additional IHC of beta-catenin with clone 17C2 and 14, and of LEF1 (lymphoid enhancer-binding factor 1), except for two cases in which tissue size was too small for further staining (i.e., a total of 81 cases were enrolled in this study), and investigated the relationship among the histological subtype, the positive ratio, and the staining pattern.

Considering the clinical and histological relevance and subsequent statistical analysis, we categorized our study series into three groups: DF, benign fibroblastic lesions, and other soft tissue tumors. In the end, the following tumor or tumor-like lesions were enrolled in the study: DF, 26; benign fibroblastic lesion, 28 (palmar/plantar fibromatosis, 5; fibroma of the tendon sheath, 4; desmoplastic fibroblastoma, 2; inflammatory fibrid polyp, 2; calcifying fibrous tumor, 1; dermatofibroma, 1; nodular fasciitis, 1; plexiform fibromyxoma, 1; non-neoplastic, 11 [fibrosis, 2; granulation tissue, 2; bursitis, 1; fibrosing dermatitis, 1; fibrous lesion, 1; fibrous nodule, 1; fibrous tissue with calcification, 1; hypertrophic scar, 1; and reactive fibroblastic proliferation, 1]); and other soft tissue tumors, 27 (schwannoma, 2; angioleiomyoma, 1 [these 3 cases are benign]; leiomyosarcoma, 5; myxofibrosacoma, 3; synovial sarcoma, 3; dedifferentiated liposarcoma, 2; low-grade fibromyxoid sarcoma, 2; solitary fibrous tumor, 2; angiomatoid fibrous histiocytoma, 1; BCOR-CCNB3 sarcoma, 1; dermatofibrosarcoma protuberans, 1; PEComa, 1, undifferentiated pleomorphic sarcoma, 1; spindle cell sarcoma, and NOS, 2 [these 24 cases are malignant, i.e., sarcomas]). All experiments and procedures were approved by the Medical Ethics Committees of Kyoto University Graduate School of Medicine and Kyoto University Hospital.

Immunohistochemistry

IHC was performed on formalin-fixed, paraffin-embedded specimens using an automated immunostainer (Benchmark Ultra; Ventana Medical Systems, Tucson, AZ, USA). For beta-catenin, three anti-beta-catenin antibodies were used; 1) clone, beta-catenin-1; dilution, 1:200; Dako, Santa Clara, CA, USA; 2) clone, 17C2; dilution, 1:50; Leica Biosystems, Wetzlar, Germany; 3) clone, 14; dilution, 1:100; Becton, Dickinson and Company, Franklin Lakes, NJ,

USA. IHC of LEF1 was performed with one anti-LEF1 antibody (clone, EPR2029Y; Abcam, Cambridge, United Kingdom). For three representative cases of DF (Case 1-3), we also performed IHC with clone beta-catenin 1, using a different lot, substrate (alkaline phosphatase [ALP]), and/or autostainer (BOND RX; Leica Biosystems). For IHC with anti-beta-catenin antibodies, we evaluated the presence or absence of nuclear expression. We used the membrane of the surrounding vascular endothelial cells (ECs) as a control, and categorized the intensity from 0 to 3 (0, no staining; 1, weaker than that of ECs; 2, comparable to that of ECs; and 3, stronger than that of ECs). When the tumor cells exhibited an intensity of 2-3, we interpreted it as positive regardless of the proportion; The number and ratio of the positive nuclei were not included in the present evaluation criteria.

For IHC with clone beta-catenin 1, we also evaluated the presence or absence of cytoplasmic dotted staining, irrespective of the proportion and intensity of the positive cells, because it might be characteristic of DF with this clone. When DAB (3,3'-diaminobenzidine)-positive granules aggregated in the cytoplasm and the size of the aggregate was larger than that of half of the nucleus (i.e., not a small aggregate), we defined it as dotted staining. When it looked like a ring (i.e., when the center of the aggregate appeared blank), it was interpreted as a dotted ring. When it looked like a sphere (i.e., when there was no blank in the aggregate), it was interpreted as a dotted sphere. (Figure S1). No other dotted patterns were recognized, and when the other two clones were used, such dotted staining was not observed in any DFs (data not shown).

For LEF1, it was interpreted as positive when moderately to strongly nuclear positive tumor cells were observed, in accordance with the previous study on DF (10); strong (visible at ×2 objective), moderate (visible at ×4), weak (visible at ×10), and negative (not visible at ×10).

138

139 **DNA mutational analysis**

140 The mutational analysis for codon 41 and 45 at exon 3 of the *CTNNB1* gene was performed for
141 two cases of DF, as previously described (9). Briefly, DNA was extracted from formalin-fixed,
142 paraffin-embedded tissues. The extracted DNA was amplified by polymerase chain reaction
143 (PCR) with two pairs of primers designed to analyze the point mutations in codon 41 or 45 at
144 exon 3 of *CTNNB1*: forward 5'-GATTTGATGGAGTTGGACATGG-3', reverse 5'-
145 TCTTCCTCAGGATTGCCTT-3' and forward 5'-TGGAACCAGACAGAAAAGCG-3', reverse
146 5'-TCAGGATTGCCTTTACCACTC-3'. The amplicon was isolated by gel electrophoresis, and
147 after the purification, the sequence of the product was read by direct sequencing with the above
148 forward primers.

149

150 **Statistics**

151 To compare the ratio of positive cases among three groups in each evaluation, we used the Chi-
152 square test or Fisher's exact test as appropriate. Differences with $P < 0.05$ were considered to be
153 significant. When the difference was significant, we compared the ratio between two groups
154 among the three groups. The corrected *P*-value by the Bonferroni method, $P < 0.0167$ (0.05/3),
155 was considered to be significant.

156

157 **Results**

158 **Clinical findings**

159 Among the 81 cases, the number of male and female patients was 33 and 48, respectively. The
160 ages of the patients ranged from 1 to 87 years, with a median of 48 years. The number of cases

with DF was 26 (8 men and 18 women). The ages of these patients ranged from 9 to 73, with a median of 41 years. All of the cases were regarded as sporadic according to the clinical information. For the other 55 cases, the number of male and female patients was 25 and 30, respectively, and the ages of the patients ranged from 1 to 87, with a median of 51 years.

Pathological findings

Nuclear beta-catenin is almost specific for desmoid-type fibromatosis with clone beta-catenin 1

Consistent with previous reports (8, 11-13), the majority of DF exhibited nuclear positivity for beta-catenin (Figure 1), and the positive ratios between DF and benign fibroblastic lesions and between DF and other soft tissue tumors were significant (DF, 54% [14/26]; benign fibroblastic lesions, 4% [1/28]; and other soft tissue tumors, 0% [0/27]; both $P < 0.001$, Chi-square test) (Table 1-2). Although the sensitivity was not high (54% [14/26]), since only one other lesion (diagnosis, hypertrophic scar) exhibited this staining pattern in our study series, the specificity for DFs reached 98% (54/55). The ratio of nuclear-positive cells in each DF case ranged from 0 to 60%, with a median of 5%.

Characteristic cytoplasmic beta-catenin in desmoid-type fibromatosis with clone beta-catenin 1

In addition to specific nuclear staining, DF exhibited a unique dotted staining pattern (88% [23/26]) with clone beta-catenin 1 (Figure 1). The frequency of dotted staining of DF was significantly higher than that of the other two groups.

For dotted rings (DF, 54% [14/26]; benign fibroblastic lesions, 11% [3/28]; and other soft tissue tumors, 0% [0/27]), the difference was significant between DF and the other two groups

(both $P < 0.001$, Chi-square test). For dotted spheres (DF, 88% [23/26]; benign fibroblastic lesions, 54% [15/28]; and other soft tissue tumors, 26% [7/27]), the difference between DF and the other two groups was significant ($P = 0.005$ between DF and benign fibroblastic lesions; $P < 0.001$ between DF and other soft tissue tumors; Chi-square test) (Table 1). All cases that contain cells showing dotted ring staining also contained cells showing dotted sphere staining (i.e., all dotted ring [+] cases were included in dotted sphere [+] cases). The dotted ring pattern was rather specific for DF (specificity of 95% [52/55]) and was not observed in any tumors of the "other soft tissue tumor" group, which mostly (25/28) consisted of sarcomas (Table 1).

All of the 14 nuclear-positive DFs exhibited cytoplasmic dotted staining (Figure 1). Among the 12 "nuclear beta-catenin-negative" DFs, 9 cases (75%) showed cytoplasmic dotted staining (including two genetically confirmed cases described above), in which 5 cases (42%) showed dotted ring staining. Nine cases (75%) were positive for nuclear beta-catenin with clone 17C2, 11 cases (92%) with clone 14, and 10 cases (83%) for LEF1. In the representative three DFs, these dotted cytoplasmic staining was preserved when a different lot, chromogenic substrate (ALP), and/or autostainer (BOND RX), were used (Figure S2).

Different antibodies produce a different staining pattern in desmoid-type fibromatosis and its mimics

Subsequently, we expanded IHC for beta-catenin with different clones (17C2 and 14) and for LEF1, a cofactor of beta-catenin in the Wnt pathway activation (10). When clone 17C2 was used, 22/26 (85%) of DFs were positive for nuclear beta-catenin (Figure 2), while 5/28 (18%) of benign fibroblastic lesions and 4/27 (15%) of other soft tissue tumors were positive, and the difference of the positive ratios was significant between DF and the other two groups (both $P <$

0.001, Chi-square test) (Table 1-2). When clone 14 was used, 25/26 (96%) of DFs were positive for nuclear beta-catenin (Figure 2), while 8/28 (29%) of benign fibroblastic lesions and 13/27 (48%) of other soft tissue tumors were positive. The difference of the positive ratios was significant between DF and the other two groups (both $P < 0.001$, Chi-square test), although a substantial proportion of "other soft tissue tumor" group (13/27 [48%]) exhibited the nuclear expression (Table 1-2). Accordingly, the sensitivity and specificity of nuclear beta-catenin in the diagnosis of DF were 85% (22/26) and 84% (46/55) in clone 17C2, 96% (25/26) and 62% (34/55) in clone 14. When clone 17C2 was used, the ratio of nuclear-positive cells in each DF ranged from 0 to 70%, with a median of 10%. On the other hand, most DFs showed diffuse nuclear staining with clone 14, and the positive cell ratio in each DF ranged from 0 to 90%, with a median of 75%.

In LEF1 immunostaining, the positive ratio of DFs (23/26 cases [88%]) was higher than those of the other two groups (5/28 cases [18%] in benign fibroblastic lesions, and 8/27 cases [30%] in other soft tissue tumors) (Figure 2), and the difference in the positive ratios was significant between DF and the other two groups (both $P < 0.001$, Chi-square test). The sensitivity and specificity of LEF1 expression in the diagnosis of DF were 88% (23/26) and 76% (42/55) (Table 1-2).

Discussion

Nuclear accumulation of beta-catenin is a useful tool and is practically the gold standard for diagnosing DF. Even though the presence of nuclear beta-catenin-negative DFs has been accepted (1, 8), pathologists may hesitate to make a definitive diagnosis for such cases. However, recent study by Koike et al. demonstrated that nuclear beta-catenin-negative DFs can

harbor *CTNNB1* mutations and that nuclear beta-catenin-positive tumors may lack such mutations (9). They also indicated that the staining pattern of beta-catenin is different for the two antibodies that they used (clone beta-catenin 1 and 17C2). Here, we demonstrated that their observation is reproducible and that DF exhibits a characteristic dotted cytoplasmic staining pattern when clone beta-catenin 1 is used. This information might be helpful for the diagnosis of DF.

In this study, clone 14 showed a lower specificity for nuclear beta-catenin than clones beta-catenin 1 and 17C2, which may contradict a previous study of Ng et al (11). Aside from the different IHC protocols between studies, one reason would be the different cutoff points for nuclear positivity. In this study we interpreted presence of moderately to strongly positive cell(s) as positive regardless of the proportion of percentage of positive tumor cells, because the number of positive cells with clone beta-catenin 1 was often small even in typical cases of DF and we applied the same cutoff point for all the three clones. When using clone 14, Ng et al. reported that high-level staining (>25% of cells having nuclear staining) was seen in only a limited number of non-DF cases, while low-level staining (0-25%) was seen in a variety of tumor types (11). Goto et al. recently reported that with 10% cutoff most scar lesions (95%) expressed nuclear beta-catenin when clone 14 was used, suggesting that clone 14 can actually have lower specificity for the diagnosis of DF (14).

Regarding the unique cytoplasmic staining in clone beta-catenin 1, the biggest difference from the other two antibodies is immunogen (i.e., C-terminal beta-catenin-GST [glutathione S-transferase] fusion protein in clone beta-catenin 1, and the C-terminus of the beta-catenin in clone 17C2 and 14). Likely, their epitopes are also different, which may cause different staining patterns. We suspect that the dotted staining pattern seen in IHC with clone beta-catenin 1 is an

antibody-dependent artifact possibly resulting from cross-reaction of the antibody with unknown protein(s) or is a reaction with fragments of cytoplasmic beta-catenin, which may trap the antibody entirely in the cytoplasm in some DF cases. Although the underlying difference between cytoplasmic dotted rings and spheres is also currently unknown, if the above hypothesis is true, it might be because of the quantity of cross-reacted proteins or degradated beta-catenins, or the position of these proteins. Despite this, considering that most DFs with cytoplasmic dotted staining showed nuclear expression with clone 17C2 and 14, and with LEF1, we think dotted cytoplasmic staining, especially dotted ring staining, might be supportive for the diagnosis of DF when clinical and histological settings are consistent.

The typical dotted ring was observed in about half of DFs (15/26), including one with S45F, but that was not the case in one with T41A. DFs with S45F are reported to carry a higher risk of recurrence than those with T41A or the wild-type allele (15), and the DFs with T41A and S45F show different metabolomic profiles, including being related to glutathione (16), although our limited IHC studies did not reveal the association of glutathione S-transferase with the beta-catenin staining pattern (data not shown).

Our study has limitations in that the series is small, with insufficient numbers and types of soft tissue tumors. Further, some of the relevant differential diagnoses, such as low-grade fibroblastic sarcoma and gastrointestinal stromal tumor, were not included. Another comprehensive study is therefore needed to validate our findings.

In summary, we found that the sensitivity and specificity of nuclear beta-catenin in the diagnosis of DF were different among commonly used antibodies when the same cutoff point was applied, and that dotted cytoplasmic staining, especially dotted ring staining, was characteristic of DF when clone beta-catenin 1 was used. Our results might be useful when

considering the diagnosis of DF, although further investigations, such as molecular testing for *CTNNB1,* might be needed to obtain a conclusive diagnosis.

Disclosure statement

None declared.

Author contributions

Conception and design of the study: YY, MH, and AS. Acquisition and analysis of data: YY, KI, and YN. Drafting the manuscript and figures: YY. Correction and approval of the manuscript: All authors.

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Figure legends

Figure 1. Representative H&E sections and immunohistochemistry for beta-catenin with clone beta-catenin 1 in desmoid-type fibromatosis (DF)

(a, d, g, j) DF with *CTNNB1* mutation, S45F (Case 1). The tumor consists of a proliferation of bland spindle cells that form sweeping fascicles (a). The tumor shows bright beta-catenin staining (d, g, j). The tumor cells do not exhibit positivity for nuclear beta-catenin but exhibit dotted cytoplasmic staining, which often creates a ring-like formation (j, N: nucleus) (a: H&E staining; d, g, j: beta-catenin [clone beta-catenin 1] immunohistochemistry). (b, e, h, k) Another DF with *CTNNB1* mutation, T41A (Case 2). The tumor consists of bland spindle cells (b) and shows strong dotted cytoplasmic staining for beta-catenin (e, h, k), which creates a sphere-like formation without nuclear positivity (k, N: nucleus) (b: H&E staining; e, h, k: beta-catenin [clone beta-catenin 1] immunohistochemistry). (c, f, i, l). A nuclear beta-catenin–positive DF (Case 3). Consistent with the histology suggesting DF (c), this tumor exhibits clear nuclear beta-catenin positivity (f, i) but also shows dotted cytoplasmic ring staining in the cytoplasm (f, l) (c: H&E staining; f, i, l: beta-catenin [clone beta-catenin 1] immunohistochemistry)

Figure 2. Immunohistochemistry for beta-catenin with clone 17C2 and 14, and for LEF1 in representative cases of desmoid-type fibromatosis (DF)

(a, d, g, j, m) DF with *CTNNB1* mutation, S45F (Case 1). The tumor cells exhibit positivity for nuclear beta-catenin. No dotted ring pattern is observed (a, d, g, j). LEF1-positive tumor cells are easily observed (m). (b, e, h, k, n) Another DF with *CTNNB1* mutation, T41A (Case 2). The tumor cells also exhibit positivity for nuclear beta-catenin. No dotted cytoplasmic pattern is observed (b, e, h, k). LEF1-positive tumor cells are easily observed (j). (c, f, i, l, o) A nuclear

beta-catenin–positive DF (Case 3). This tumor exhibits nuclear beta-catenin positivity without dotted cytoplasmic staining in IHC with clone 17C2 and 14 (c, f, i, l). The tumor clearly shows LEF1 expression (o).

Table legends

Table 1. Pathological diagnosis of our case series and the results of immunohistochemistry for beta-catenin and LEF1

Table 2. Immunohistochemistry of beta-catenin and LEF1 in our case series

Supplementary information

Figure S1. Schemas of dotted cytoplasmic staining

When DAB (3,3'-diaminobenzidine)-positive granules aggregated in the cytoplasm and the size of the aggregate was larger than that of half of the nucleus (i.e., not a small aggregate), it was interpreted as dotted staining. When it looked like a ring (i.e., when the center of the aggregate appeared blank), it was interpreted as a dotted ring (a). When it looked like a sphere (i.e., when there was no blank in the aggregate), it was interpreted as a dotted sphere (b).

Figure S2. Immunohistochemistry of beta-catenin with clone beta-catenin 1 and alkaline phosphatase in representative cases of desmoid-type fibromatosis (DF)

(a, d) DF with *CTNNB1* mutation, S45F (Case 1). The tumor cells exhibit dotted (ring) cytoplasmic staining with alkaline phosphatase as a substrate. (b, e) Another DF with *CTNNB1* mutation, T41A (Case 2). The tumor cells exhibit dotted (sphere) cytoplasmic staining. (c, f) A nuclear beta-catenin–positive DF (Case 3). The cytoplasmic dotted pattern is preserved (c, f).

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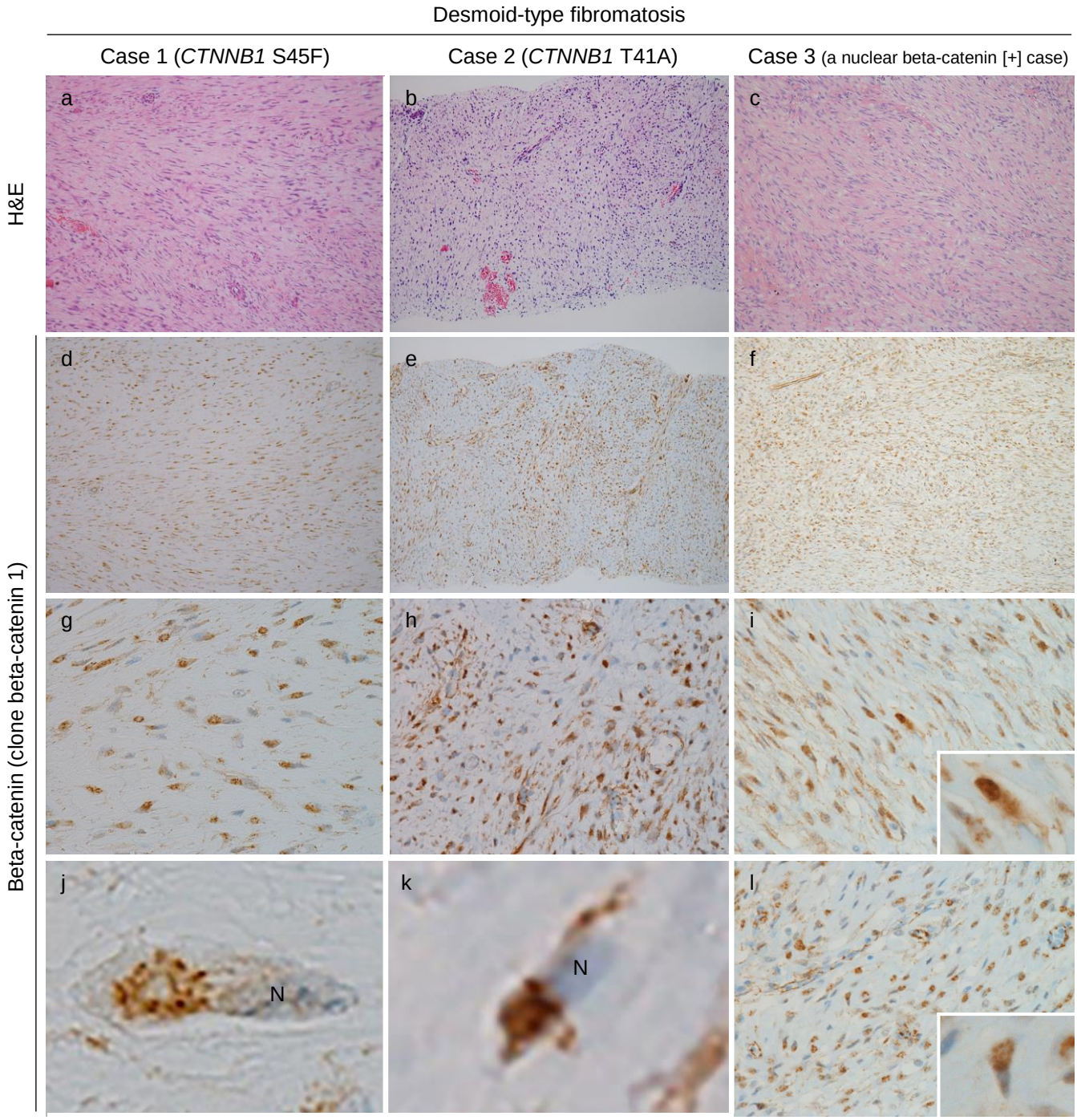

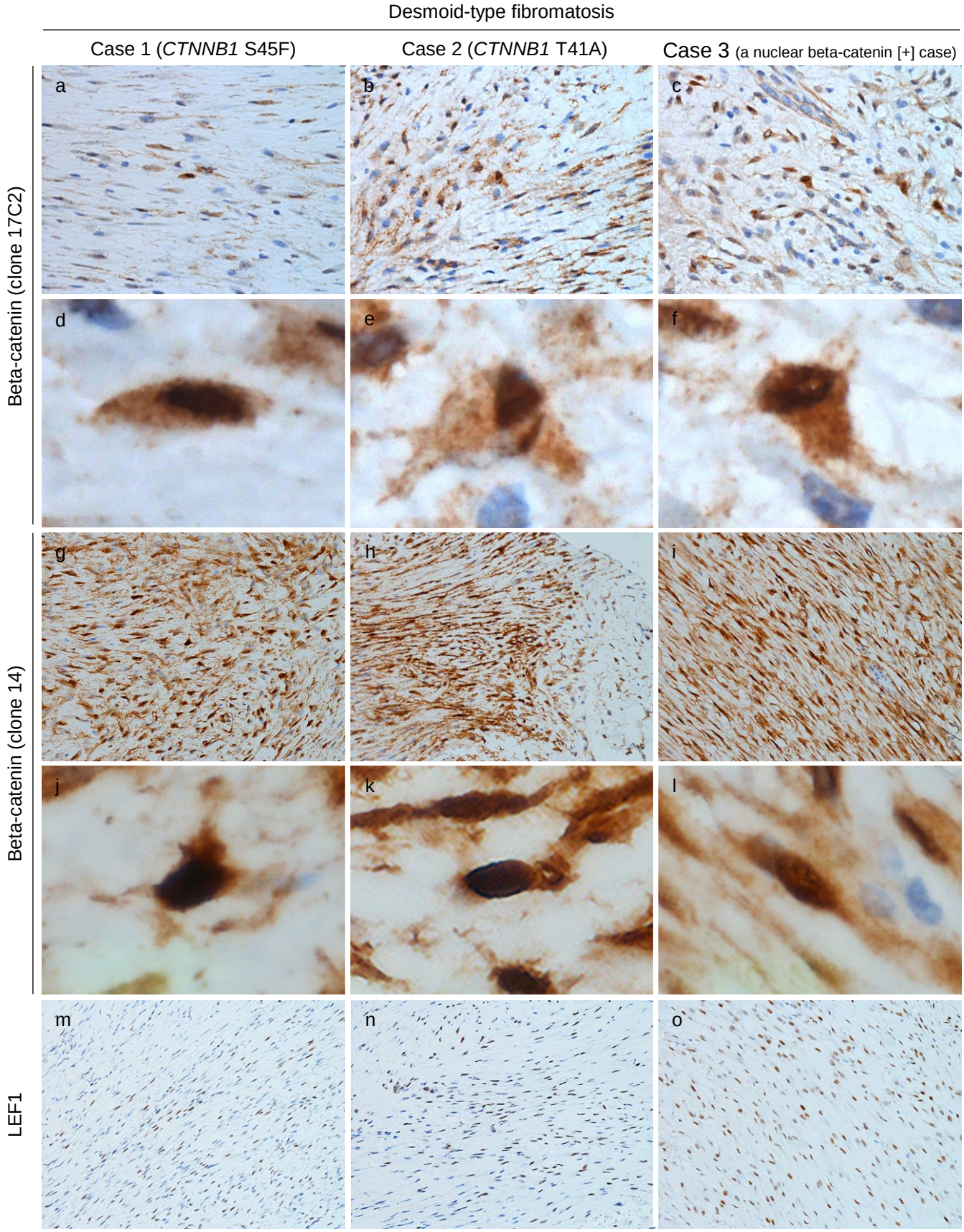

Table 1. Pathological diagnosis of our case series and the results of immunohistochemistry for beta-catenin and LEF1

Pathological diagnosis	No.	Beta-catenin 1				17C2		14		LEF1	
		N		C		N		N			
		(+)	(-)	R	S	(+)	(-)	(+)	(-)	(+)	(-)
Desmoid-type fibromatosis	26	14	12	15	23	22	4	25	1	23	3
Benign fibroblastic lesions	28	1	27	3	15	5	23	8	20	5	23
Palmar/Plantar fibromatosis	5	0	5	0	3	1	4	3	2	1	4
Fibroma of tendon sheath	4	0	4	1	2	0	4	0	4	0	4
Desmoplastic fibroblastoma	2	0	2	0	1	0	2	0	2	0	2
Inflammatory fibrid polyp	2	0	2	0	0	0	2	0	2	0	2
Calcifying fibrous tumor	1	0	1	0	1	0	1	0	1	0	1
Dermatofibroma	1	0	1	0	0	0	1	1	0	1	0
Nodular fasciitis	1	0	1	0	1	0	1	0	1	0	1
Plexiform fibromyxoma	1	0	1	0	0	0	1	0	1	0	1
Non-neoplastic	11	1	10	2	7	4	7	4	7	3	8
Other soft tisse tumors	27	0	27	0	7	4	23	13	14	8	19
Schwannoma	2	0	2	0	0	0	2	0	2	1	1
Angioleiomyoma	1	0	1	0	0	0	1	0	1	0	1
Leiomyosarcoma	5	0	5	0	0	1	4	3	2	2	3
Myxofibrosacoma	3	0	3	0	2	0	3	1	2	0	3
Synovial sarcoma	3	0	3	0	0	1	2	2	1	3	0
Dedifferentiated liposarcoma	2	0	2	0	2	0	2	0	2	0	2
Low-grade fibromyxoid sarcoma	2	0	2	0	0	1	1	2	0	0	2
Solitary fibrous tumor	2	0	2	0	1	0	2	0	2	0	2
Angiomatoid fibrous histiocytoma	1	0	1	0	0	0	1	1	0	0	1
BCOR-CCNB3 sarcoma	1	0	1	0	0	0	1	1	0	0	1
Dermatofibrosarcoma protuberans	1	0	1	0	1	0	1	1	0	1	0
PEComa	1	0	1	0	0	0	1	0	1	0	1
Undifferentiated pleomorphic sarcoma	1	0	1	0	0	0	1	1	0	0	1
Spindle cell sarcoma, NOS	2	0	2	0	1	1	1	1	1	1	1

N: nuclear, C: cytoplasmic, R: dotted ring, S: dotted sphere, (+): positive, (-): negative

Table 2. Immunohistochemistry of beta-catenin and LEF1 in our case series

		Beta-catenin											
		Beta-catenin 1			17C2			14			LEF1		
Groups	No.	(+)	(-)	P vs. DF	(+)	(-)	P vs. DF	(+)	(-)	P vs. DF	(+)	(-)	P vs. DF
Desmoid-type fibromatosis	26	14	12		22	4		25	1		23	3	
Benign fibroblastic lesions	28	1	27	<0.001	5	23	<0.001	8	20	<0.001	5	23	<0.001
Other soft tissue tumors	27	0	27	<0.001	4	23	<0.001	13	14	<0.001	8	29	<0.001

DF, desmoid-type fibromatosis; (+), nuclear positive; (-), nuclear negative; P, P-value

Figure S1

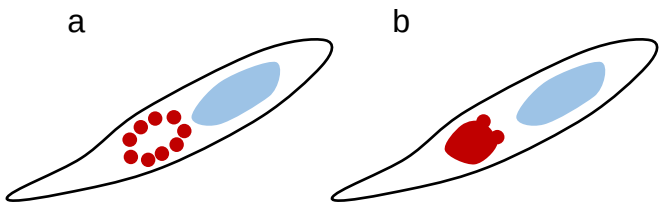

Figure S2