Recurrent Fusions Between YAP1 and KMT2A in Morphologically Distinct Neoplasms Within the Spectrum of Low-grade Fibromyxoid Sarcoma and Sclerosing Epithelioid Fibrosarcoma

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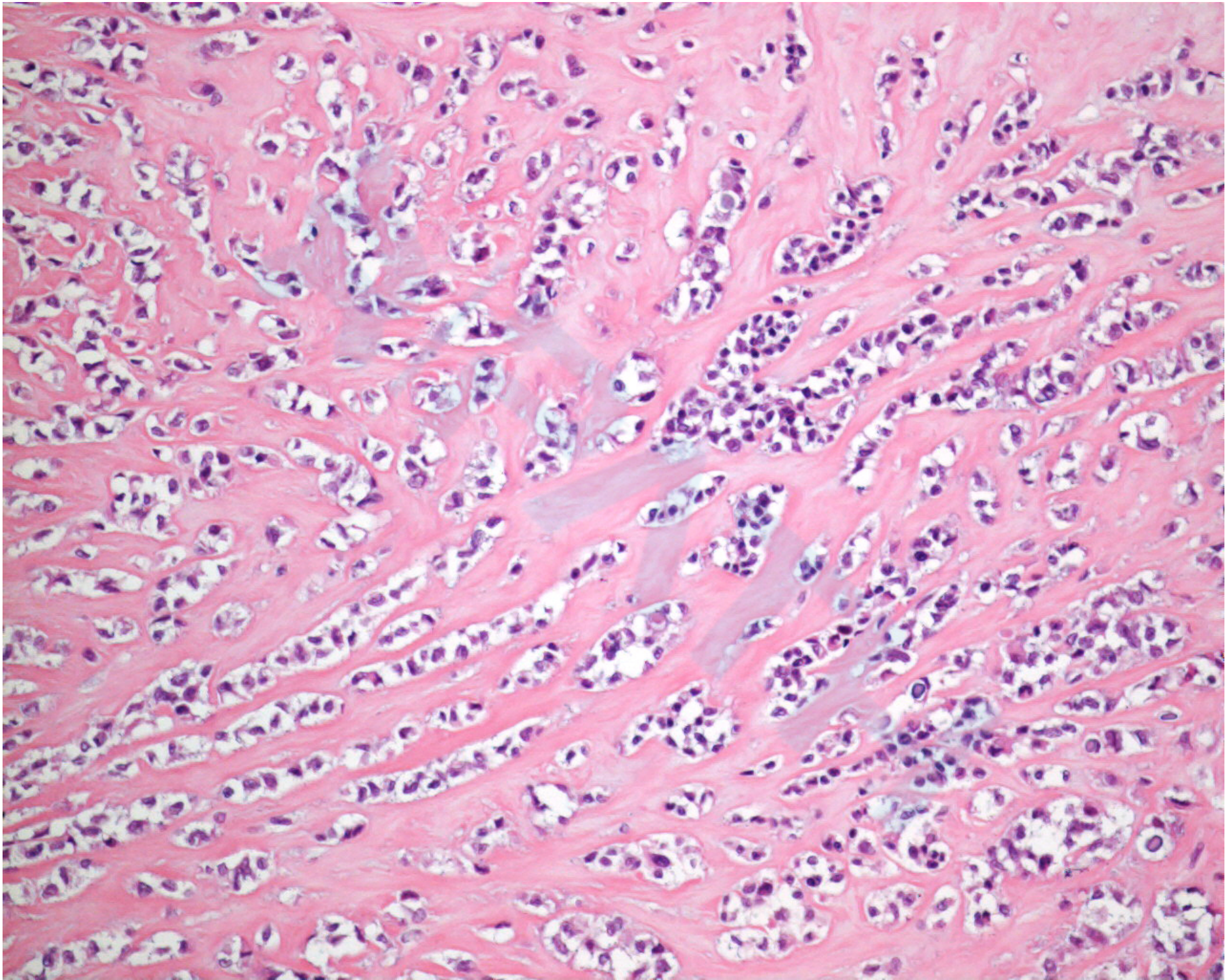
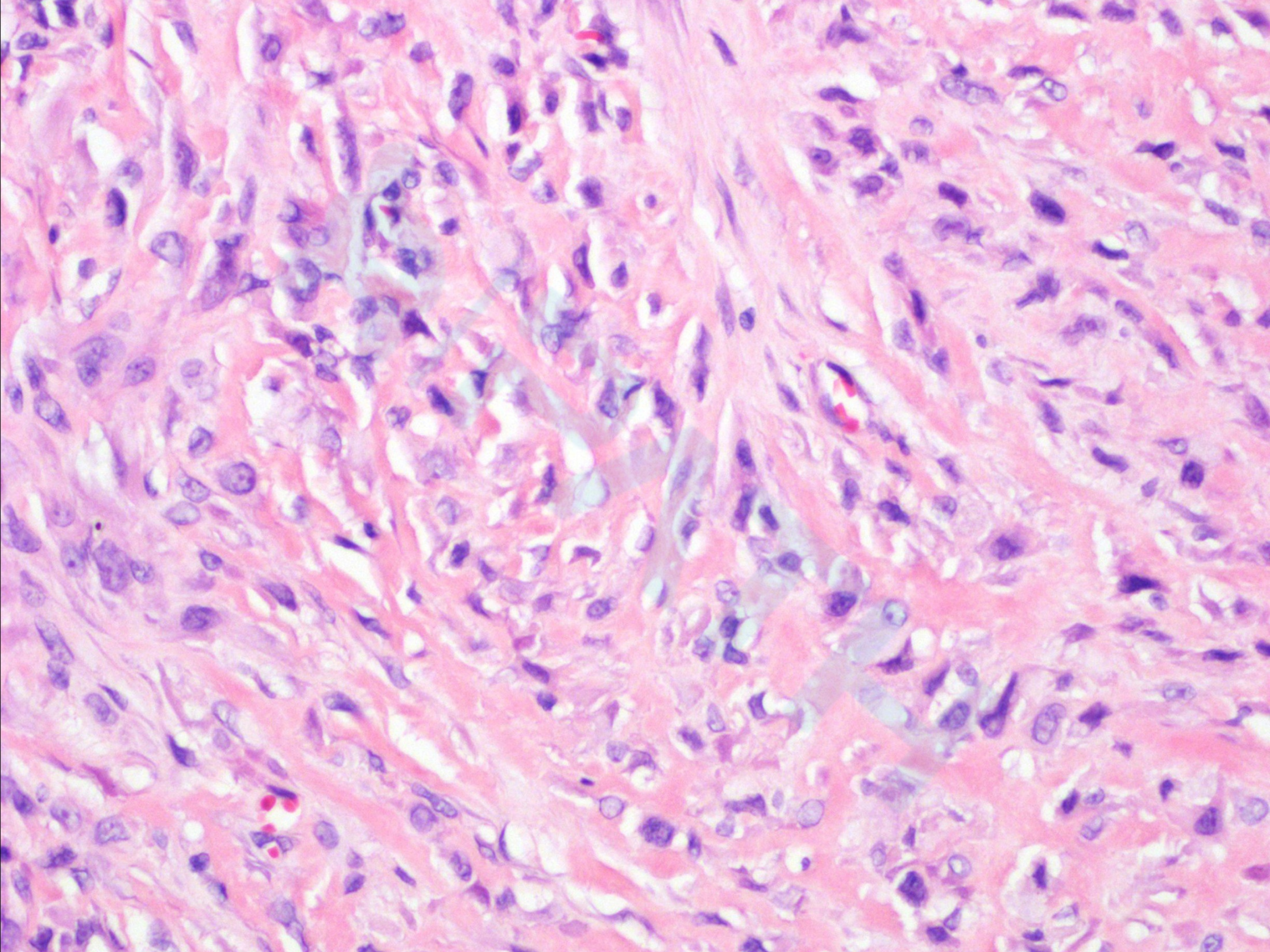
硬化性上皮样纤维肉瘤
（Sclerosing Epithelioid Fibrosarcoma，SEF）

- 临床特征
  - 深部包块
  - 年龄范围宽，中位年龄45岁
  - 下肢、肩带、躯干、头颈部

王主任课件

- 病理特征
  - 境界清楚、分叶状，或多结节，5-10cm
  - 肿瘤细胞上皮样，呈巢状、条索状，偶可见腺泡状排列
  - 肿瘤细胞分布在致密的透明变性间质之中
  - 局部区域总能找到纤维肉瘤的梭形细胞成分
  - 部分肿瘤局部形态与LGFMS形态重叠





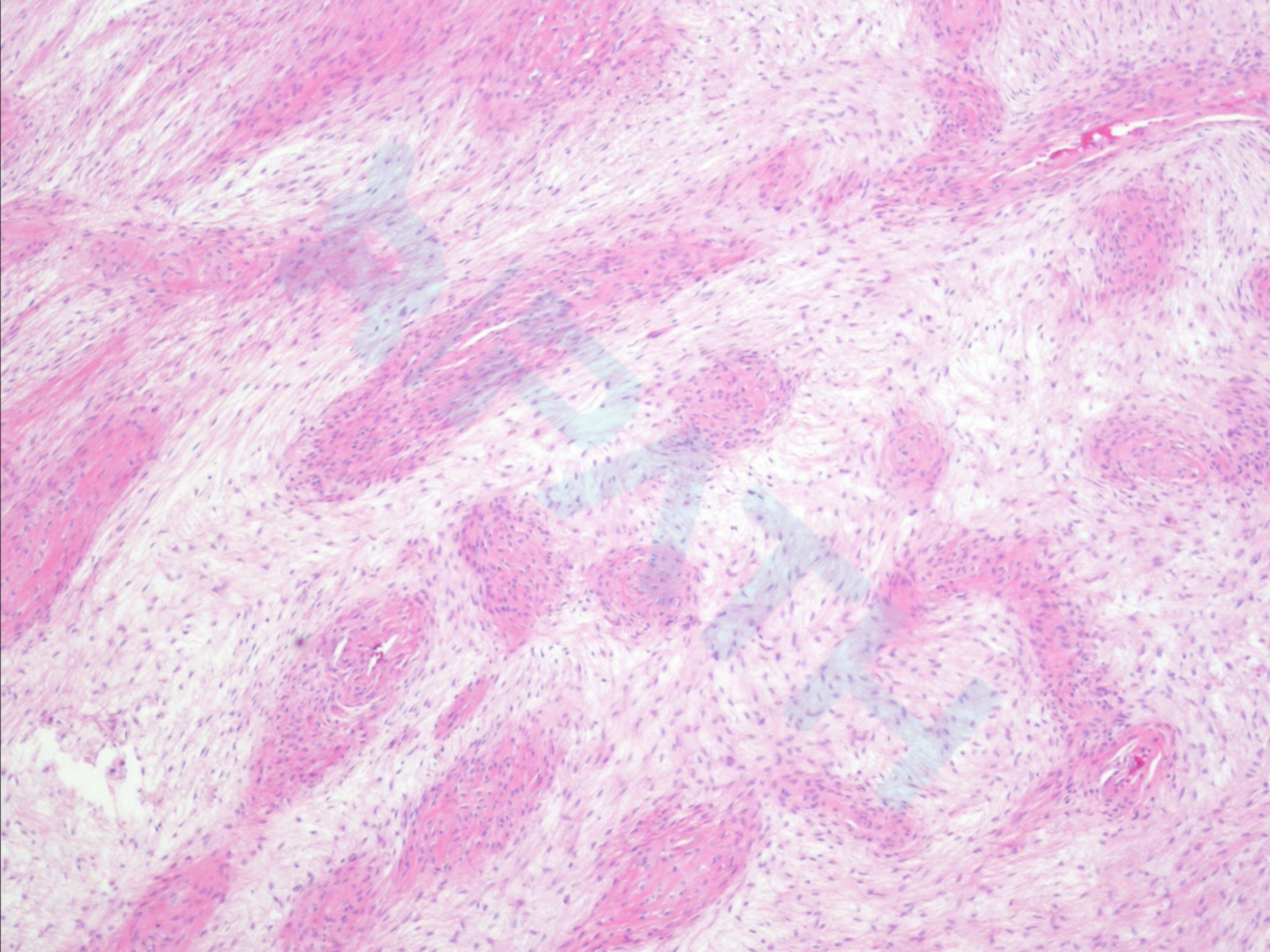
- 免疫组化
  - EMA 50% 病例局灶阳性
  - **MUC4 78%阳性**
    - Am J Surg Pathol 2012; 36(10):1444
- 遗传学
  - *EWSR1-CREB3L1*
  - 少数为*FUS-CREB3L2*（形态与LGFMS重叠）

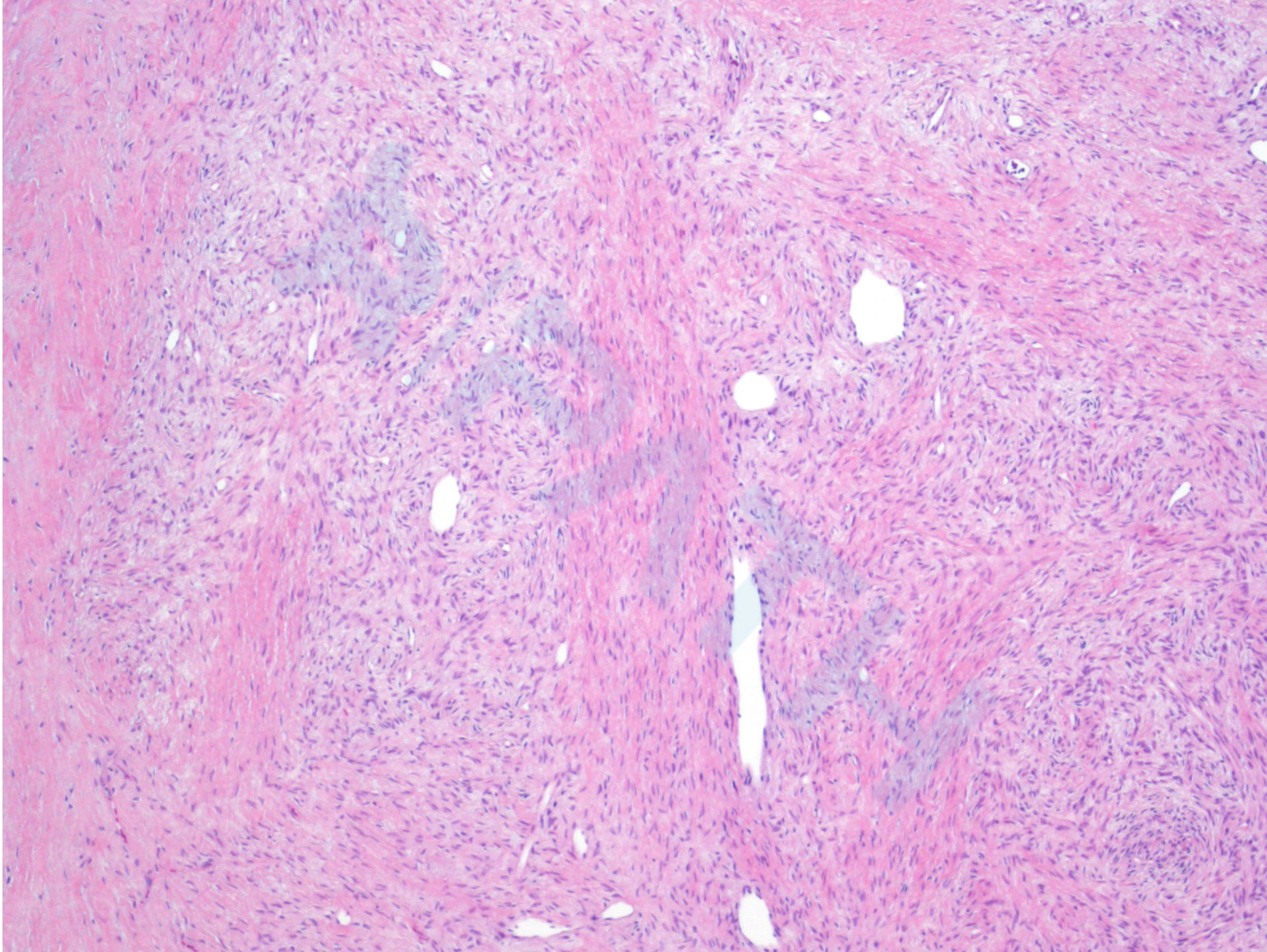
MUC4

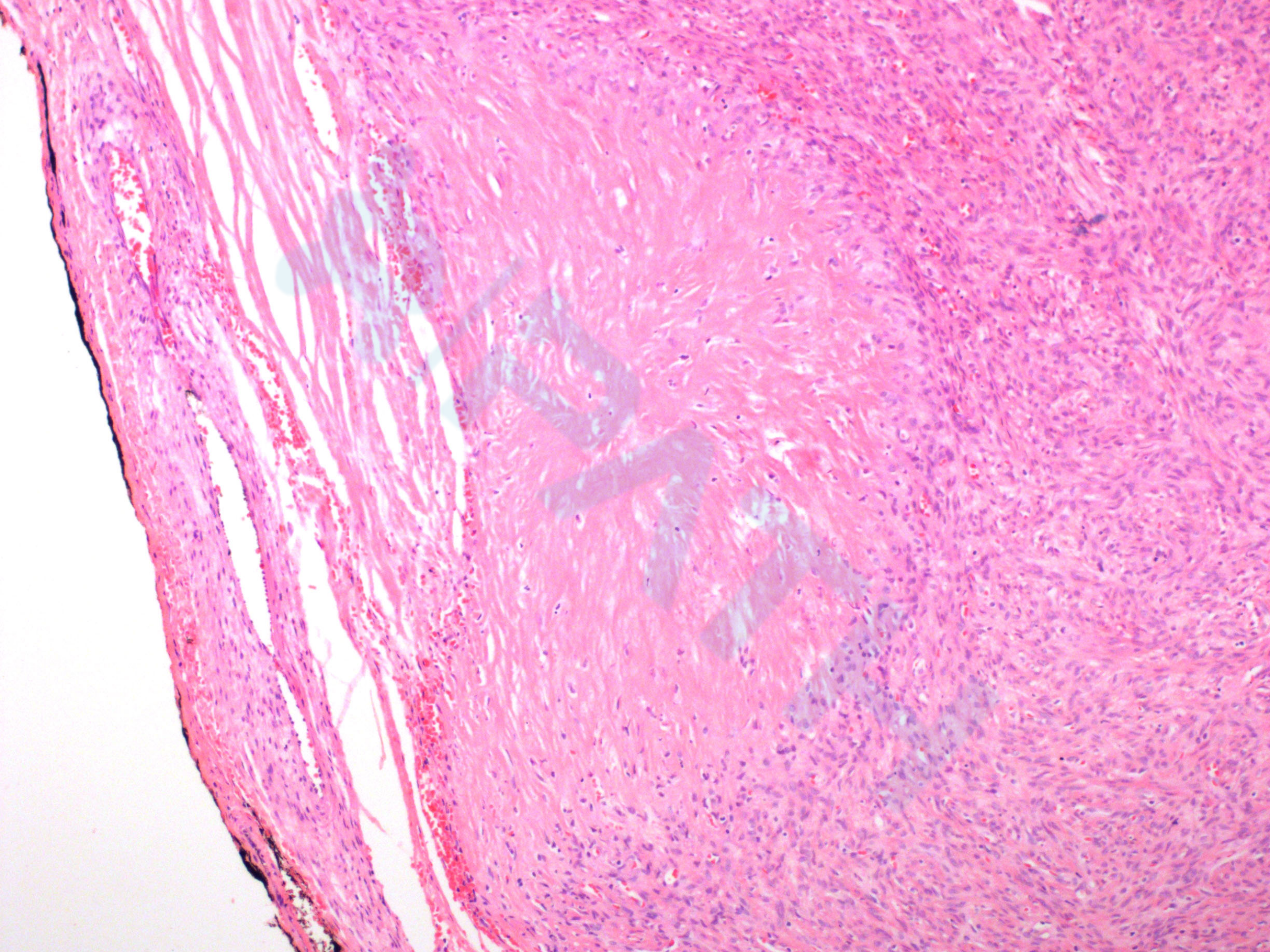
低度恶性纤维黏液样肉瘤
(Low-grade Fibromyxoid Sarcoma,LGFMS)

- 临床特征
  - 主要见于青年人，中位年龄34岁
  - 缓慢生长的无痛性深部包块
  - 大部分病例发生于肌肉，少部分见于皮下组织及真皮
  - 下肢最常见，大腿>胸壁>腋窝>臀部>头颈部>大网膜>盆腔>纵膈

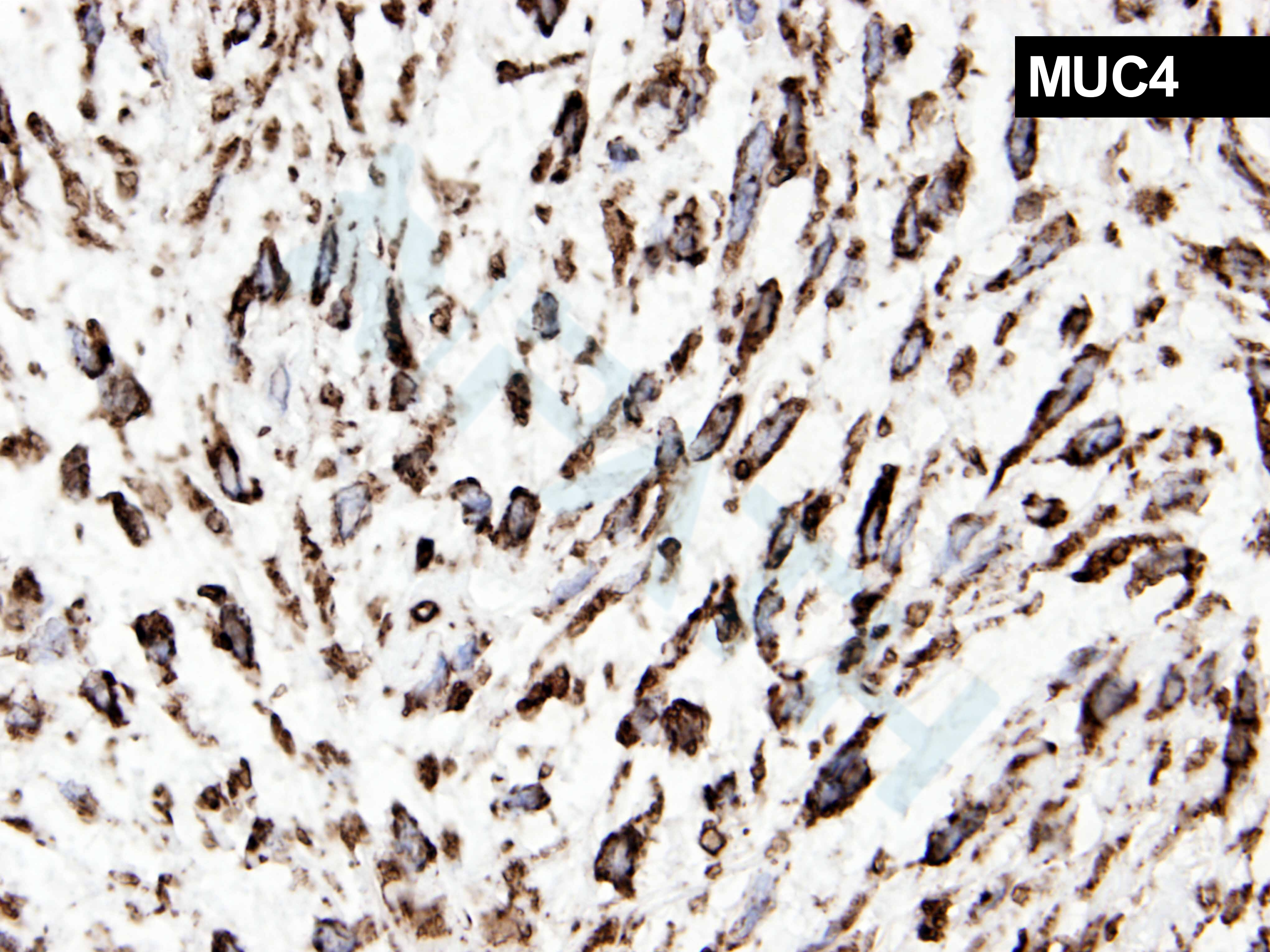
- 病理特征（LGFMS）
  - 大体边界清楚，切面灰白
  - 细胞稀少或中等丰富，梭形细胞，核小深染，染色质团块状，小核仁
  - 胞浆淡嗜酸性，边界不清，轻度多形，核分裂少见
  - 背景为纤维性或粘液样，纤维性区和粘液性区相互交替
  - 粘液样区域常见明显的分支弯曲的毛细血管
  - 局部可能出现细胞丰富的结节和原始细胞







- 免疫组化
  - Vimentin阳性
  - 局灶性SMA、MSA阳性
  - β-catenin阴性
  - **MUC4 阳性**，非常特异（在其他类似组织学形态的肿瘤中阴性） Am J Surg Pathol 2011 35(5):733
- 遗传学
  - t(7;16)(q33;p11) *FUS-CREB3L2*


MUC4

MATERIALS AND METHODS

- Case Selection

  8 cases of MUC4-negative tumors that resemble hybrid LGFMS/SEF

- Histology and Immunohistochemistry

  MUC4、S100、CD34、cytokeratin、EMA、desmin、SMA

- Massively Parallel Sequencing of RNA
- Reverse Transcription-PCR
- Single-Nucleotide Polymorphism Array and Fluorescence in situ Hybridization Analyses

RESULTS

TABLE 1. Clinical Features of *KMT2A/YAP1* and *KMT2D/PRRX1*-rearranged Tumors

Case No.	Age*/Sex	Site, Depth	Size (mm)	Treatment	Follow-up (mo)
1†	91/female	Finger, S	25	MR	DOUC (80)
2	11/male	Supraclavicular fossa, S	50 PL 140 LR	MR of PL, MR of LR	NED (321), LR (111)
3	16/female	Heel/ankle D	30 PL 20-45 LR	MR, RTx, 4 LRs, AMP after fourth LR	NED (184) LR (7, 13, 77, 149)
4	53/male	Foot, D	82 PL	AMP, CTx	DOD (23), Met (4)
5	69/male	Chest wall, D	65 PL	MR	NED (11)
6	51/female	Thigh, S	40 PL	MR	Recent case
7	34/female	Thigh, S	30 PL	MR	Recent case
8	40/male	Knee, D	130 PL	MR	Recent case

*Age at diagnosis
†Previously published in Arbajian et al.[7]
AMP indicates amputation; CTx, chemotherapy; D, deep; DOD, died of disease; DOUC, died of unrelated causes; LR, local recurrence; Met, metastases; MR, marginal resection; NED, alive, no evidence of disease; PL, primary lesion; RTx, radiotherapy; S, subcutaneous/suprafascial.

- There were 4 female and 4 male patients. The age range was wide (11 to 91 y, median: 45.5 y)；
- The sizes of the primary lesions were between 25 and 130mm；
- Of the 4 patients with> 12 months of follow-up (range: 23 to 321, average: 152, median: 132)

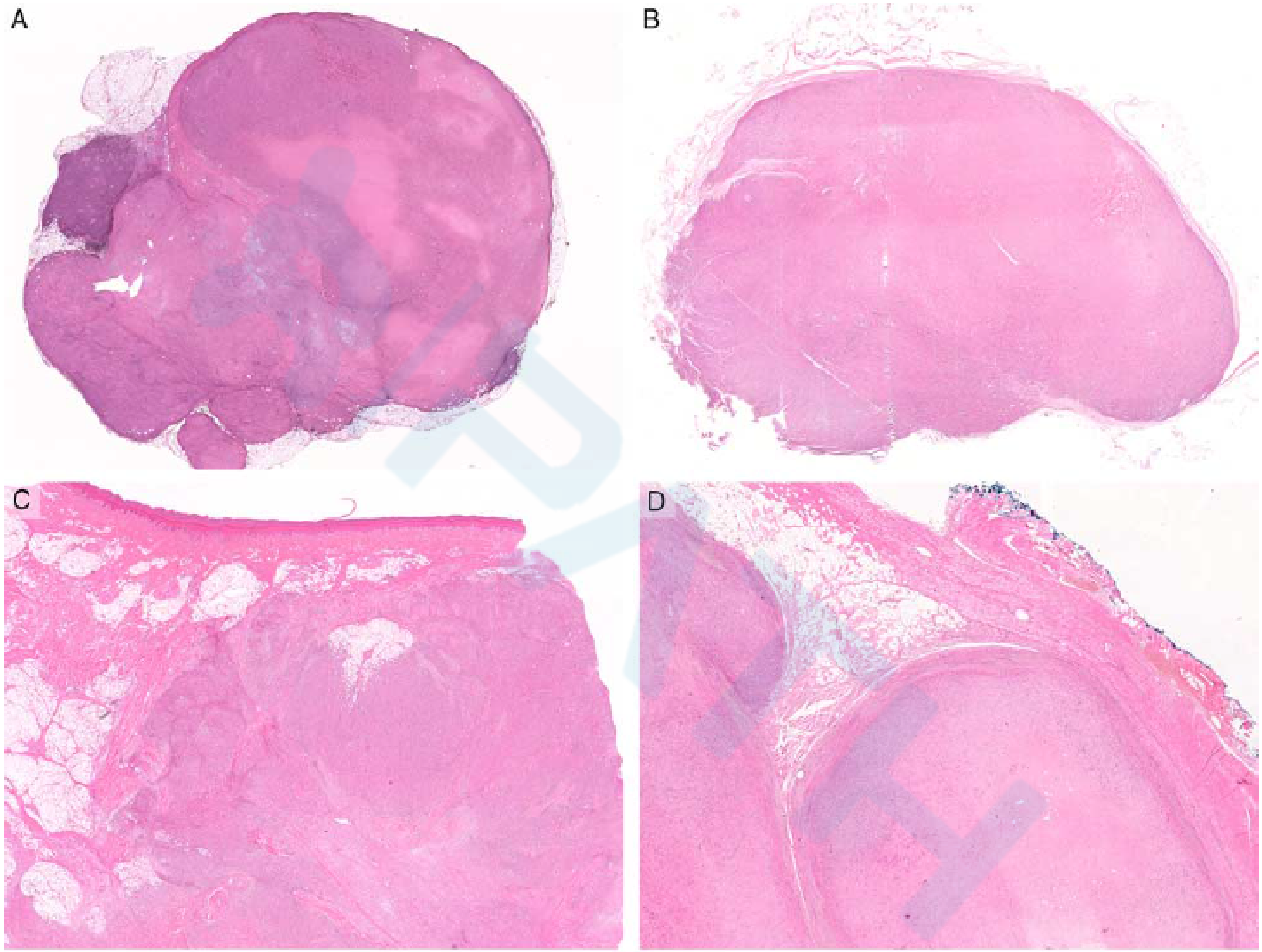
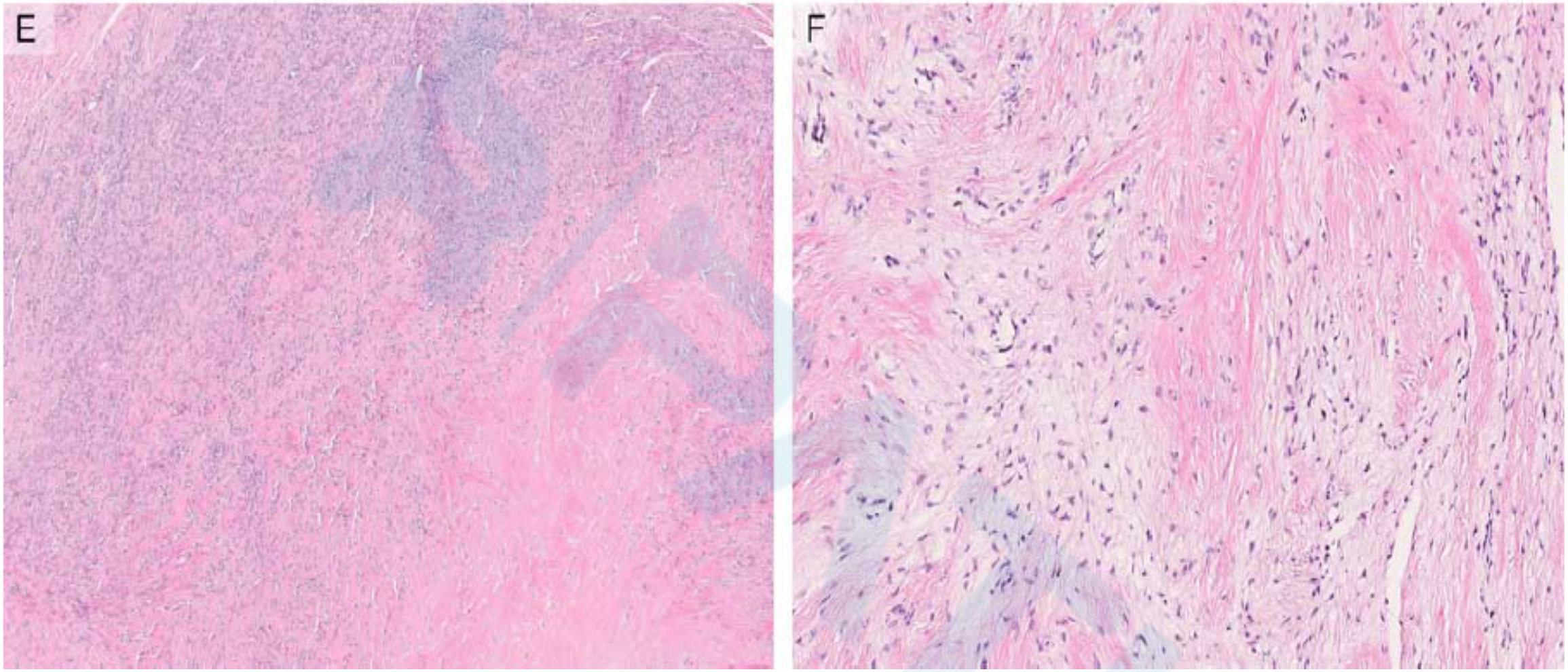


FIGURE 1. **Low-power appearances of tumors with YAP1/KMT2A fusions**. A and B, The subcutaneous, lobulated tumors showed high cellularity at the periphery and more sclerotic areas centrally (cases 6 and 1). C, The extent of highly cellular areas varied between tumors; case 4 was highly cellular throughout and showed infiltrative growth even on low power.



D and E, The deeper tumors also showed variable cellularity (cases 3 and 5). F, Focal LGFMS areas were prominent in several cases (case 7).

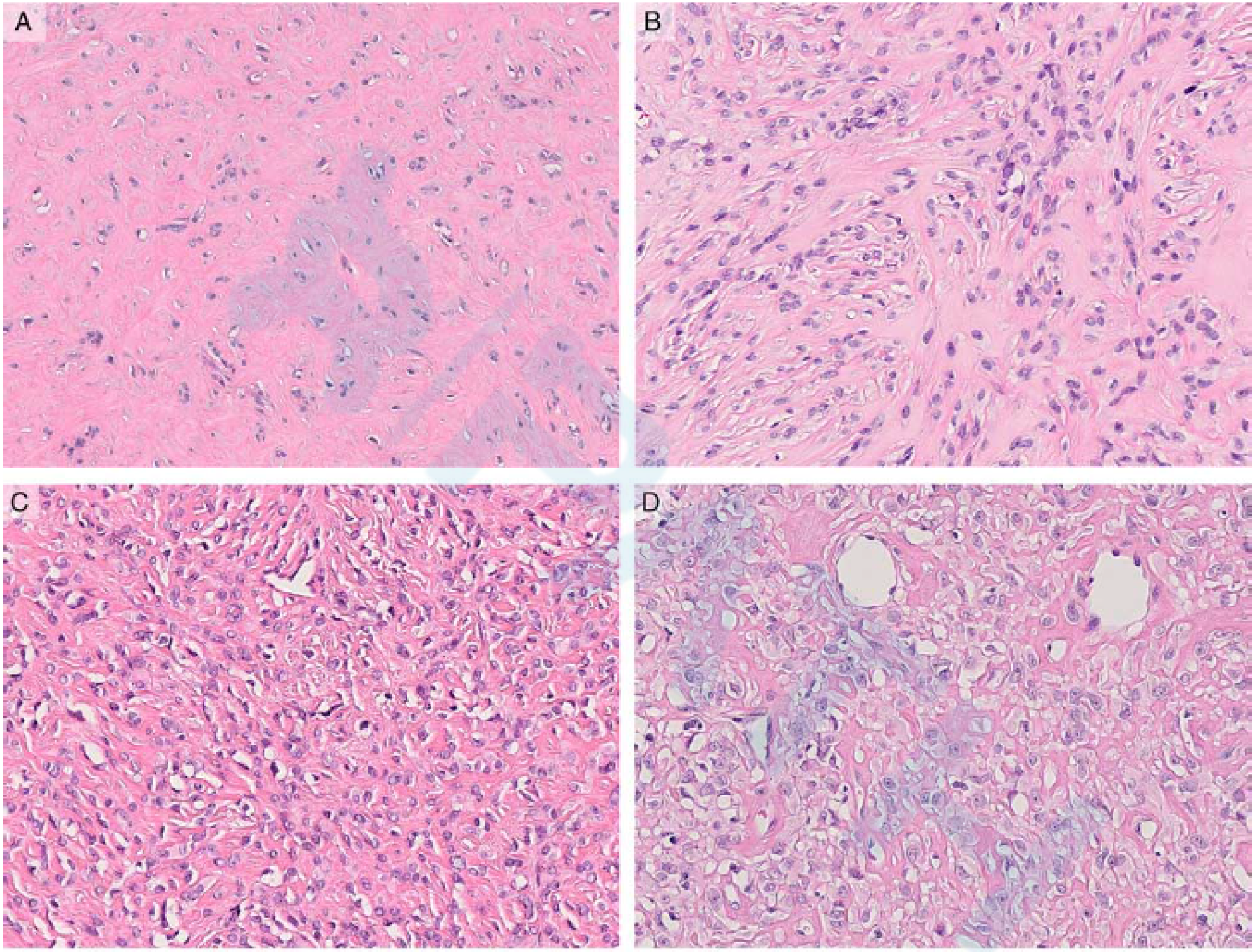
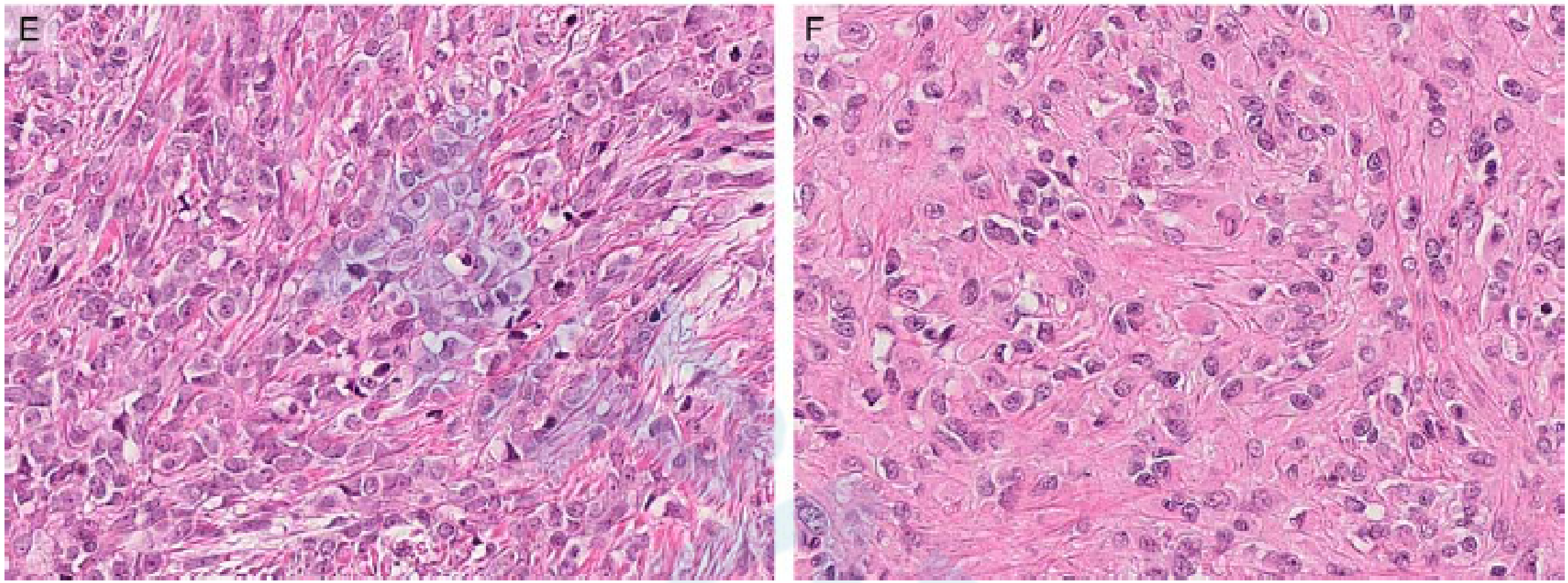


FIGURE 2. **High-power morphology of tumors with YAP1/KMT2A fusions**. A–C, The predominant morphologic picture was small to medium-sized epithelioid cells within the dense collagenous matrix (cases 5, 6, and 1).



D–F, Several tumors showed more prominent cytoplasm and larger cells, simulating epithelioid sarcoma, metastatic carcinoma, or even rhabdoid tumor (cases 2, 3,and 4).

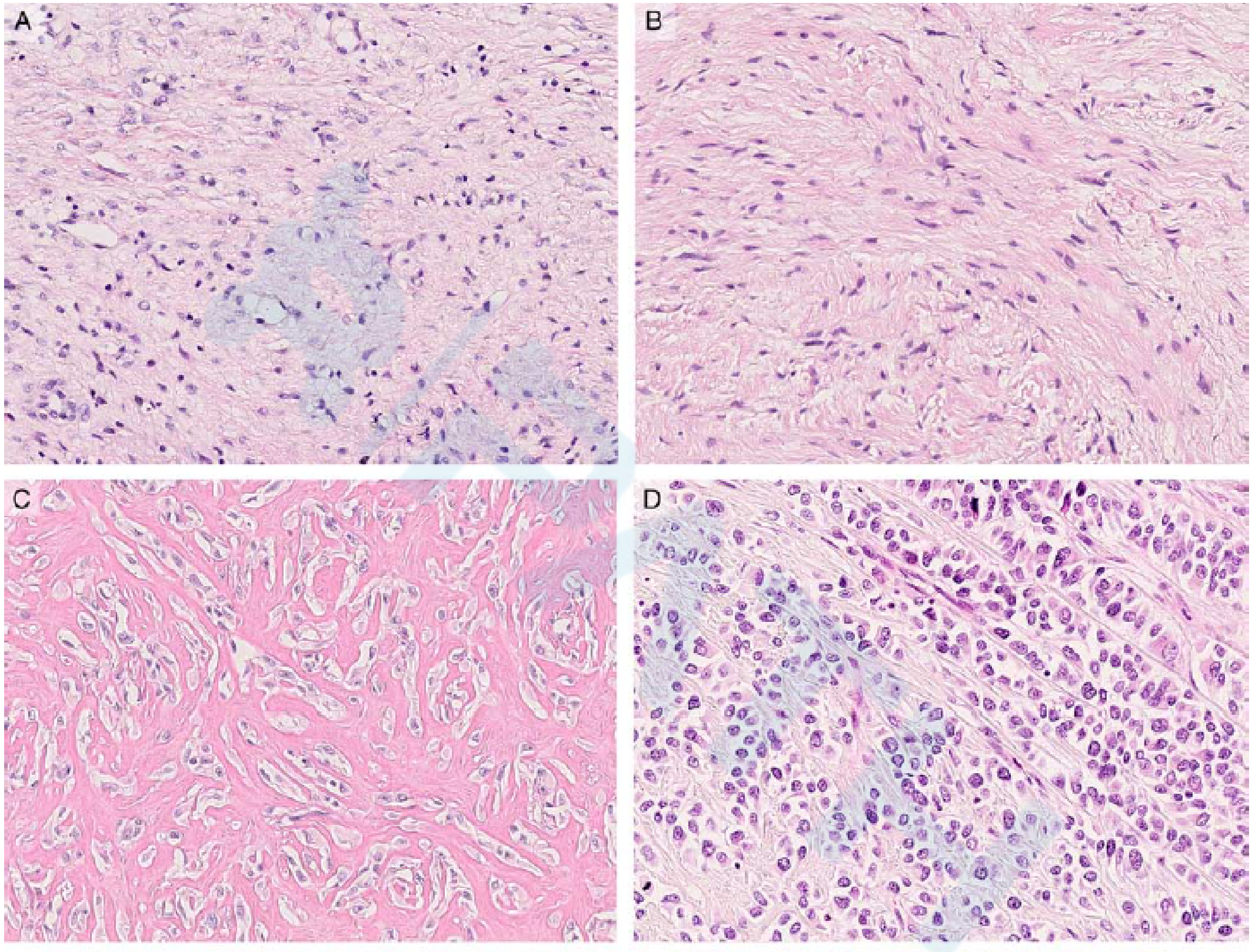
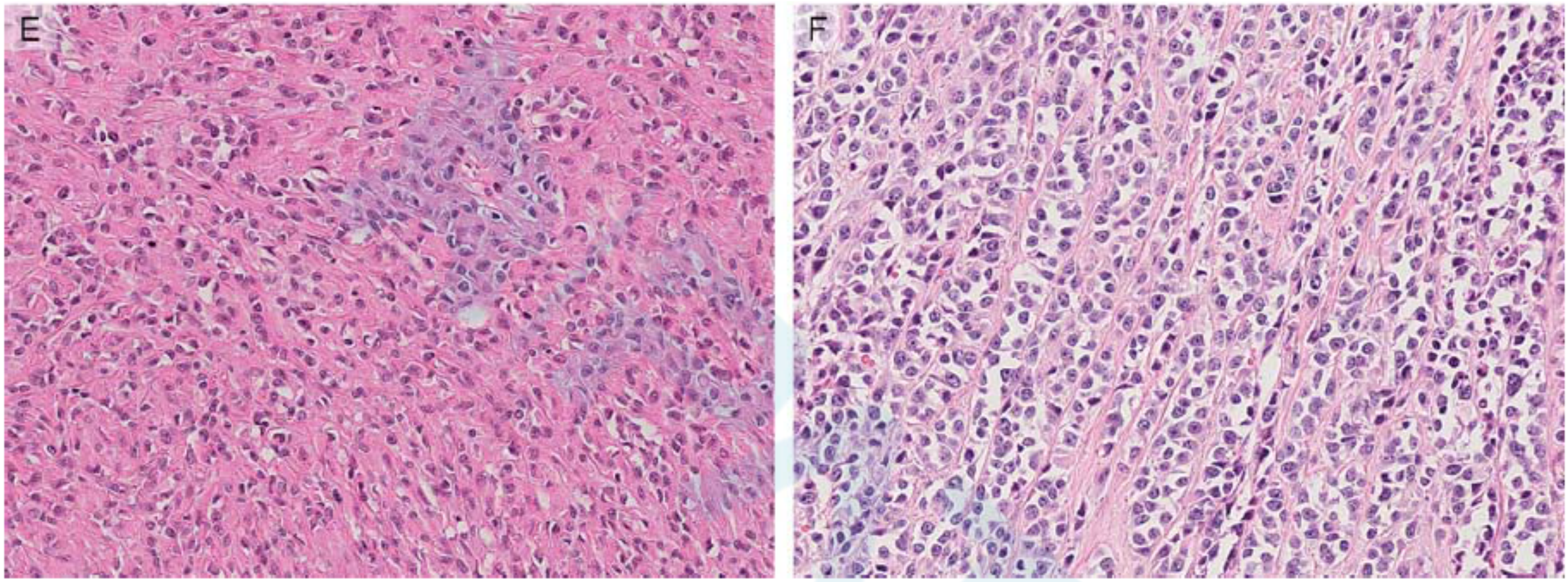


FIGURE 3. Comparison of tumors with YAP1/KMT2A fusions (A, C, and E) with MUC4-positive LGFMS and SEF with FUS/EWSR1-CREB3L1/2 gene fusions (B, D, and F). A, B, Low cellular areas in YAP1/KMT2A fusion tumors resembled conventional LGFMS, although cells were spindled (A, case 7; B, LGFMS). C and D, Sclerotic areas in tumors with YAP1/KMT2A fusions showed strands of cells rather than nests (C, case 2; D,SEF).



E and F, The tendency to form nests was less pronounced when compared with conventional SEF (E, case 4; F, SEF).

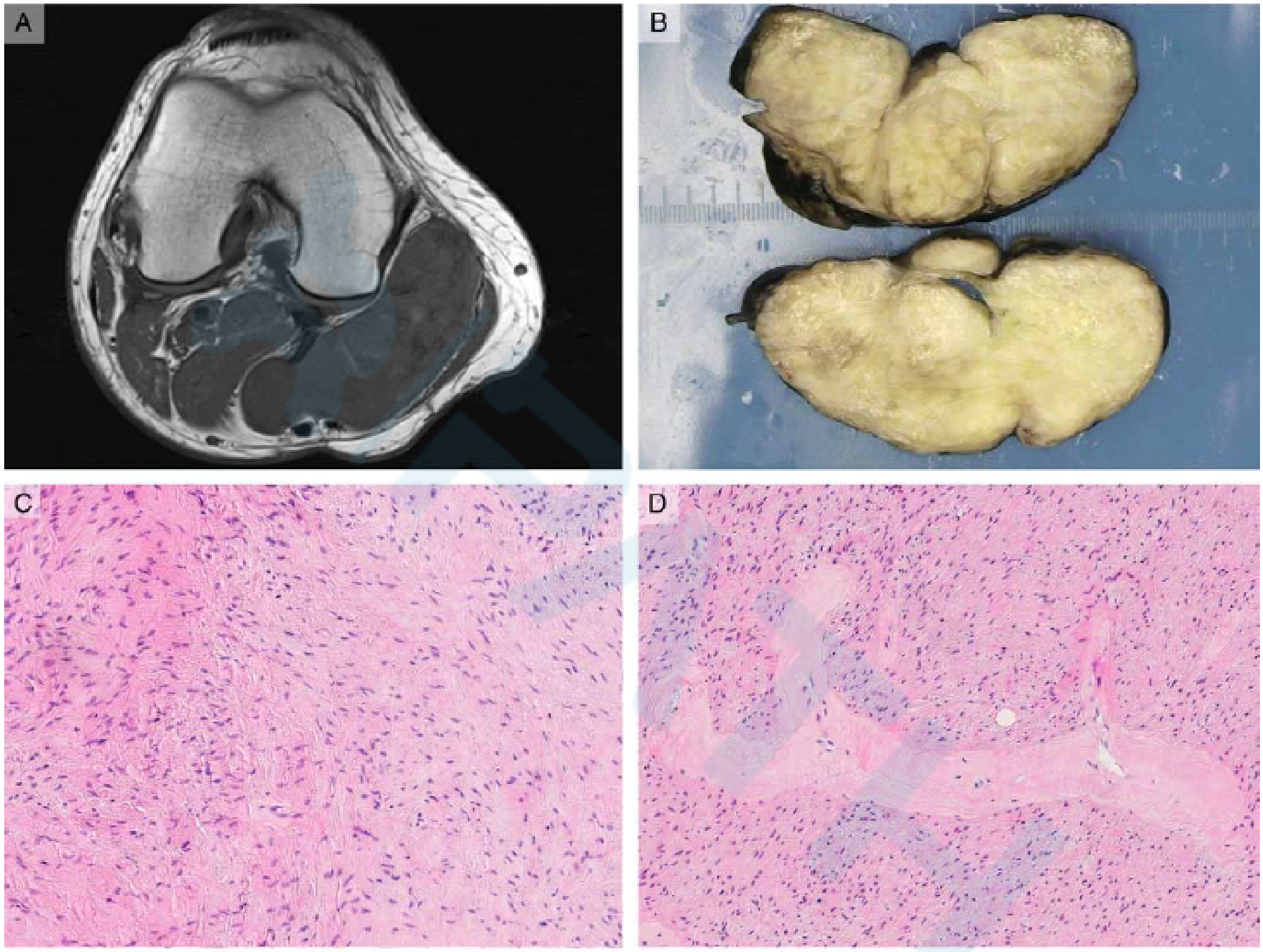
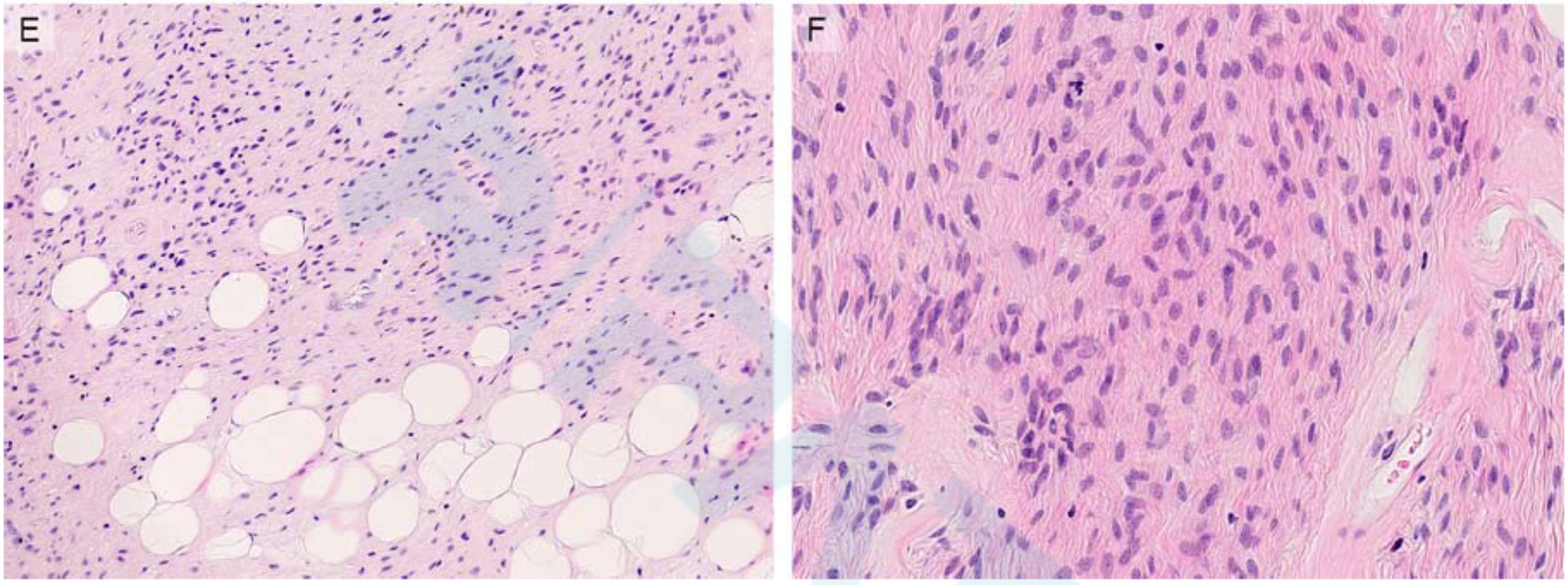


FIGURE 4. **Features of a tumor with PRRX1-KMT2D/KMT2D-PRRX1 fusions**. A, The axial T1-weighted image magnetic resonance showed a sharply demarcated intermuscular soft tissue mass dorsomedial of the knee. B, Macroscopically, the tumor had a firm consistency with a yellowish cut surface and was surrounded by a pseudocapsule. C, The tumor was predominantly hypocellular with bland spindle cells without a distinct growth pattern within a fibrillary collagenous matrix. D, It showed prominent perivascular hyalinization.



E and F, Entrapment of fat was seen multifocally. In a few more cellular areas, the tumor cells retained ovoid nuclei and nondistinct cell borders.

TABLE 2. Morphologic Features of *KMT2A/YAP1* and *KMT2D/PRRX1*-rearranged Tumors

Case No.	Margins	Cellularity	MF/10 HPF*	Necrosis	Rhabdoid Morphology	Original Diagnosis
1	Infiltrative	+/++	1	Absent	Absent	SEF
2	Infiltrative	++	2	Absent	Absent	SFT (PL), SEF (LR)
3	Infiltrative	+/++	3	Focal	Absent	SEF
4	Infiltrative	++/+++	8	Present	Present	SEF
5	Infiltrative	+/++	2	Absent	Absent	Unclassified LGFMS/SEF-like
6	Infiltrative	++/+++	18	Absent	Absent	Unclassified LGFMS/SEF-like
7	Infiltrative	+/++	1	Absent	Absent	Unclassified LGFMS/SEF-like
8	Well-demarcated	+	0	Absent	Absent	LGFMS/SEF

*Corresponding to 2.37 mm^2.
+ indicates low; ++, intermediate; +++, high; LR, local recurrence; MF, mitotic figures; PL, primary lesion; SFT, solitary fibrous tumor.

TABLE 3. Immunohistochemical and Molecular Features of *KMT2A/YAP1* and *KMT2D/PRRX1*-rearranged Tumors

Case No.	MUC4	EMA	FISH*	Fusion Transcript	Fusion Junction†
1	Negative	Positive	ND	*YAP1-KMT2A*	ex6-in ex5
				KMT2A-YAP1	ex6-ex9
2	Negative	Positive	Negative	*YAP1-KMT2A*	ex5-ex4
				KMT2A-YAP1	ex6-ex9
3	Negative	Positive	Positive (66%)*	*YAP1-KMT2A*	ex3-ex5
				KMT2A-YAP1	ex6-ex8
4	Negative	Positive	Positive (39%)	*YAP1-KMT2A*	ex5-ex4,
				KMT2A-YAP1	ex6-ex9
5	Negative	Positive	ND	*KMT2A-YAP1*	ex6-ex9
6	Negative	Positive	ND	*YAP1-KMT2A*	ex4-ex5
				KMT2A-YAP1	ex6-ex9
7	Negative	Positive	ND	*YAP1-KMT2A*	ex4-ex5
				KMT2A-YAP1	ex6-ex9
8	Negative	Negative	ND	*PRRX1-KMT2D*	ex1-ex22
				KMT2D-PRRX1	ex34-ex2

*Split signal in 13%, loss of the telomeric signal (a ~256 kbp region that includes the 3′-part of *KMT2A*) in 53%.

†Only the longest fusion transcripts are shown here. All fusion transcripts were compatible with fusion of entire exons, except in case 1, where the *YAP1-KMT2A* fusion had a breakpoint inside exon 5 of *KMT2A*. For details, see Supplementary Table 2 (Supplemental Digital Content 2, http://links.lww.com/PAS/A889).

ex indicates exon; ND, not done.

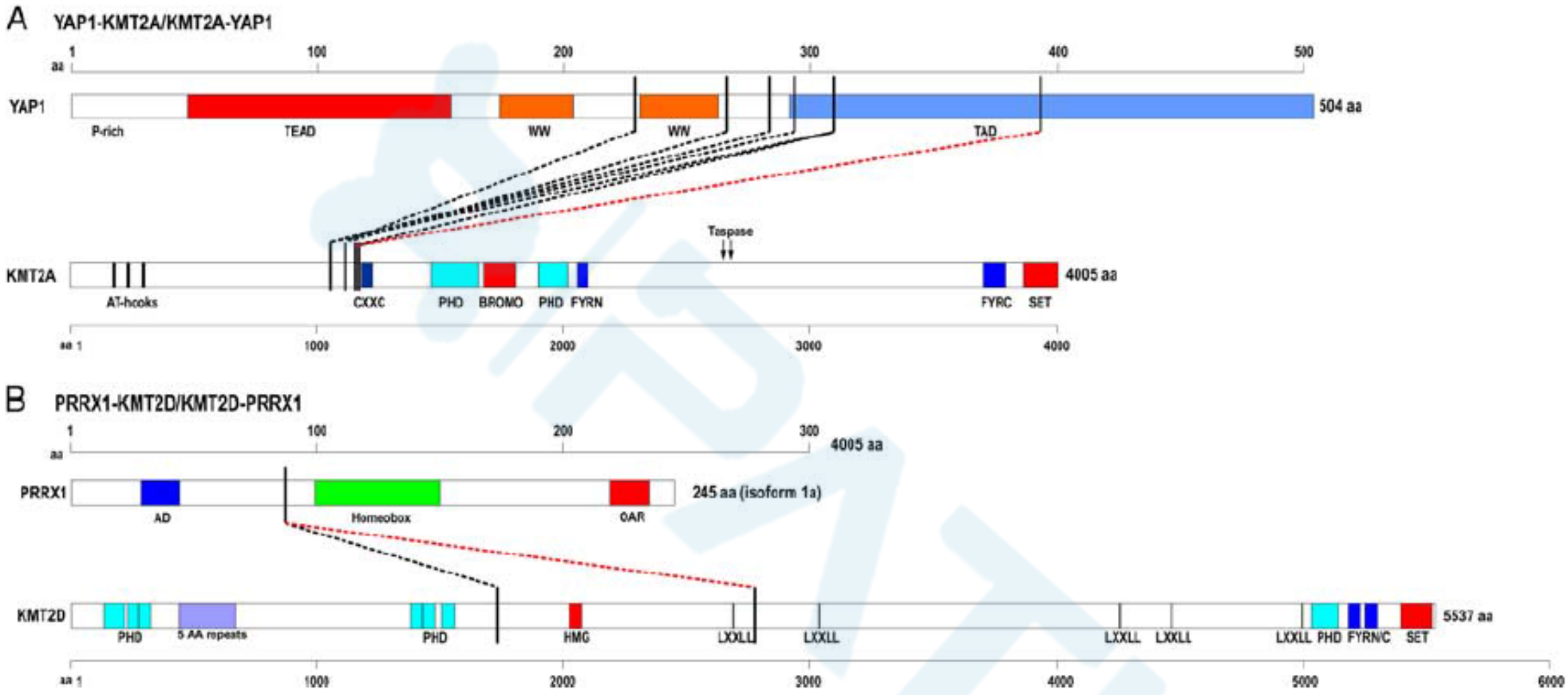


FIGURE 5. Composition of putative YAP1-KMT2A/KMT2A-YAP1 and PRRX1-KMT2D/KMT2D-PRRX1 fusion proteins. A, The different YAP1-KMT2A transcripts (dashed black lines) result in larger fusion proteins incorporating a number of functional domains of both YAP1 (TEAD-binding domain and at least one WW domain) and KMT2A. The putative KMT2A-YAP1 fusion protein (red dotted line) is significantly smaller, incorporating the AT-hooks of KMT2A, and part of the transactivation domain of YAP1. B, Reciprocal transcripts of case 8 could result in both PRRX1-KMT2D (dashed black lines) and KMT2D-PRRX1 (dashed red line) fusion proteins.

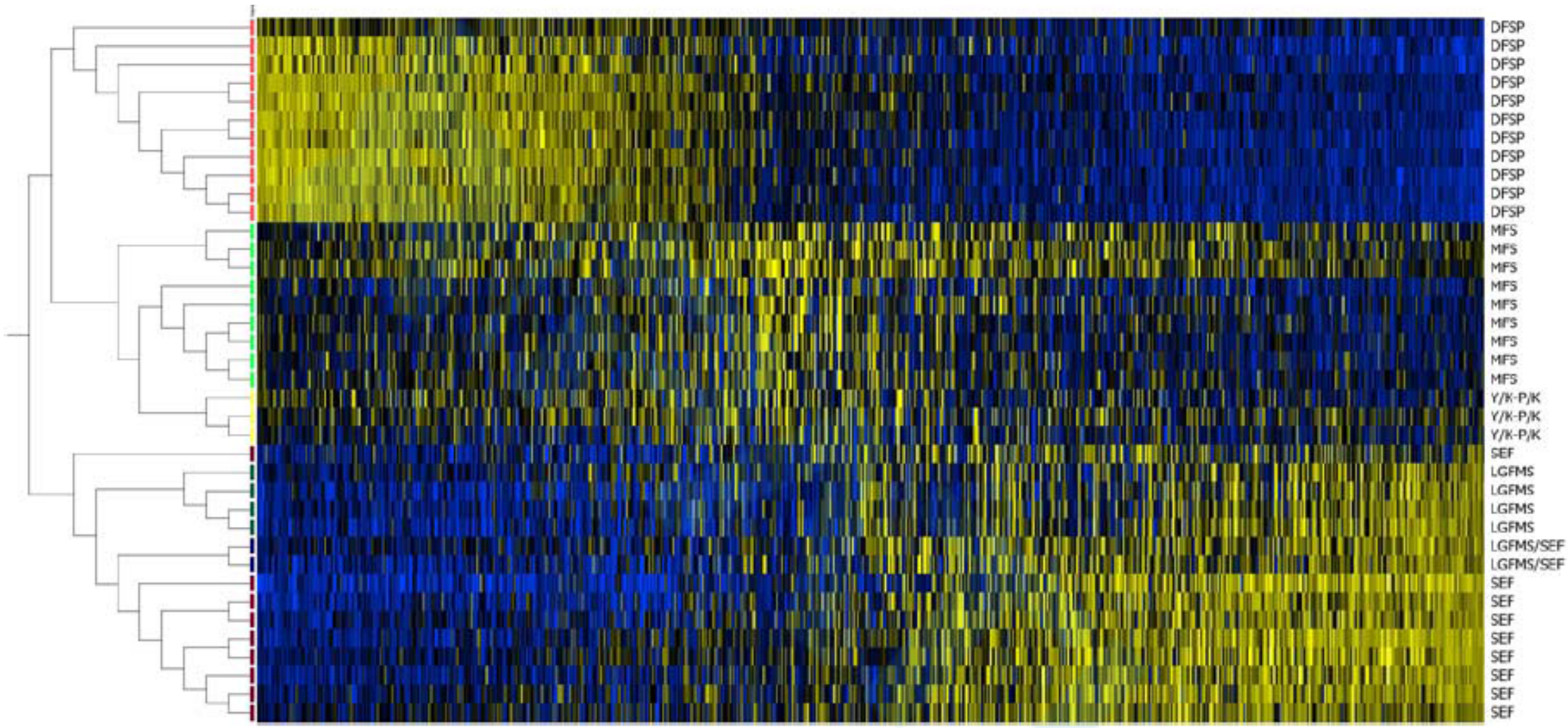


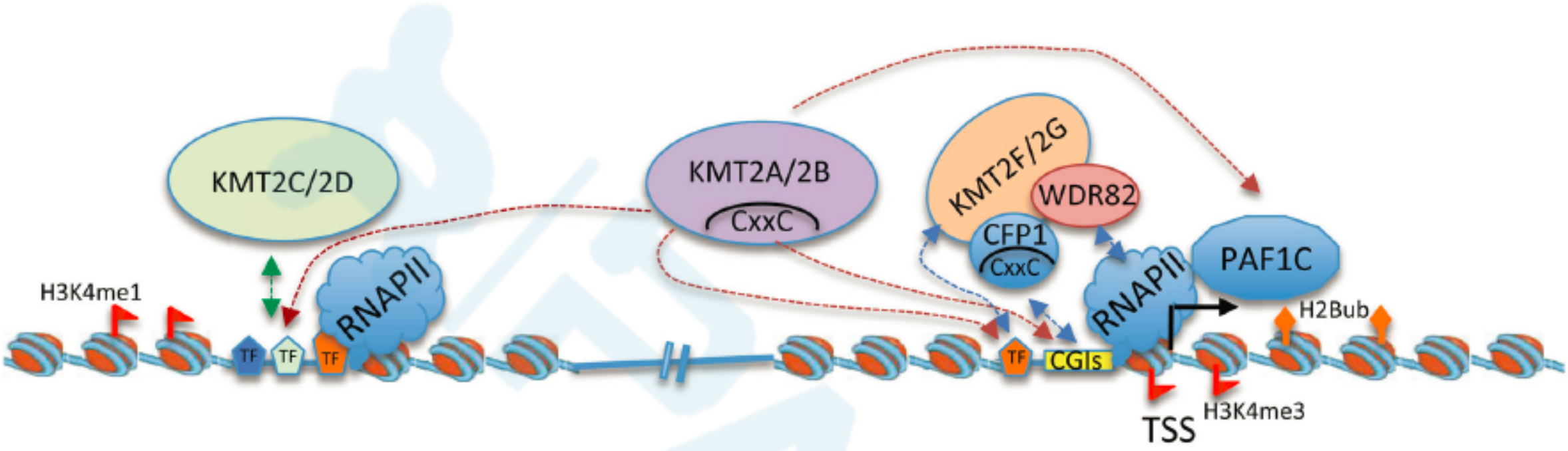
FIGURE 6. Supervised heatmap of the gene expression profiles of 3 tumors with YAP1/KMT2A and PRRX1/KMT2D fusions (Y/K-P/K; yellow) compared with 11 dermatofibrosarcoma protuberans (orange), 9 myxofibrosarcomas (light green), 9 SEFs (dark red), 4 LGFMSs (dark green), and 2 hybrid LGFMS/SEF (dark blue). After filtering of the data (variance ratio, $F = 0.45$, $P < 0.01$; 729 genes), the tumors formed separate clusters.

DISCUSSION

- The 7 tumors with KMT2A and YAP1 fusions had unifying morphologic features. All were variably cellular and composed of small to medium-sized, monomorphic and epithelioid cells situated singly or in short strands within a densely collagenized matrix.
- Examination of larger series with longer follow-up is warranted to further characterize the behavior of LGFMS/SEF-like tumors with KMT2A and YAP1 fusions.
- the tumors clustered separately and closer to myxofibrosarcomas than to LGFMS,SEF, or LGFMS/SEF

KMT2

- KMT2A（MLL）：编码组蛋白赖氨酸甲基转移酶2A，甲基化组蛋白H3赖氨酸4（H3K4），介导与表观遗传转录激活相关的染色质修饰。该基因编码一个转录激活子，在早期发育和造血过程中起到调节基因表达的重要作用。涉及该基因的多个染色体易位是导致某些急性淋巴细胞性白血病和急性髓性白血病的原因。
- KMT2D (MLL2)：胚系突变在大多数歌舞伎综合征（Kabuki syndrome）患者中发现，表现为面部特征明显、心脏和骨骼异常以及智力迟钝。



Nat Rev Cancer. 2015 June ; 15(6): 334–346.

- YAP1：编码yes相关蛋白1，与肿瘤的发生密切相关。它是一个转录增强因子家族的转录共激活因子，该家族包含一个所谓的TEA结构域。Hippo信号的失活导致YAP1去磷酸化在细胞核中聚集，进而上调转录因子的表达，促进细胞的增殖、血管生成和抑制细胞凋亡。对大多数癌症的发生或生长至关重要。
- PRRX1：配对相关同源蛋白1，是一种转录激活因子，在小鼠中，它是颅骨、椎骨、长骨和大动脉生长发育所必需的。在人类，失活突变与耳发育障碍相关。

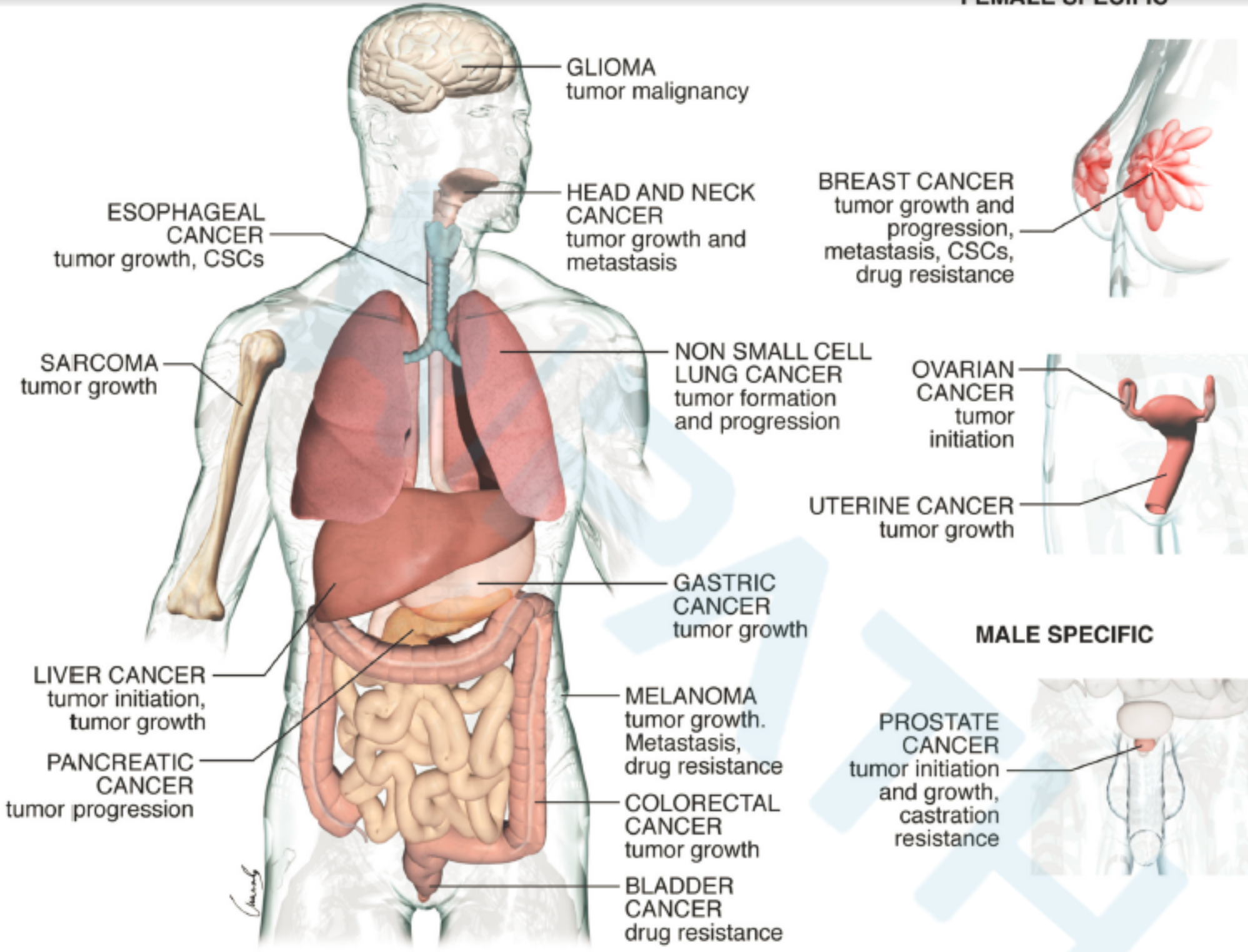


Figure 2. YAP/TAZ in human tumors.

Tumor types for which epidemiological data and functional evidence of YAP/TAZ activation have been reported

Cancer Cell. 2016 June 13; 29(6): 783–803.

SUMMARY

- we describe the clinical, morphologic, and molecular features of tumors within the morphologic spectrum of SEF and LGFMS without MUC4 overexpression.
- Further studies are required to delineate the characteristics of these sarcomas, the relation to conventional LGFMS/SEF, and the functional properties of the fusion genes.

谢谢！