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Vascular tumours of soft tissue and bone

What do we know so far?

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Vascular tumours of soft tissue and bone

What do we know so far?

- Introduction
- Distinct subtypes based on morphology & genetics
  - Epithelioid haemangioma
    - Typical/ classical/ conventional type
    - Cellular/ Atypical epithelioid haemangioma
    - Angiolymphoid hyperplasia with eosinophilia
  - Pseudomyogenic haemangioendothelioma
  - Epithelioid haemangioendothelioma
  - Angiosarcoma
- Multifocallity
- New immunohistochemical panel
- Soft tissue versus bone

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No Conflict of Interest

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Introduction – vascular tumours

- Heterogeneous group of tumours with endothelial differentiation
- Wide histological and clinical spectrum:
  - Haemangioma (benign), different types of haemangioendothelioma (intermediate) and epithelioid haemangioendothelioma + angiosarcoma (malignant)
  - Soft tissue versus bone
- Epithelioid variants remain controversial/ diagnostically challenging
- Differences in clinical behaviour, treatment and prognosis -> important to distinguish them effectively and accurately
- Past decade elucidation of the molecular background of some tumours

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Vascular tumours - distinct subtypes

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1. Epithelioid haemangioma

- Clinically:
  - Erythematous nodules or plaques - ulcerations
  - DD cyst, capillary haemangioma or pyogenic granuloma
- Histology - 3 different subtypes (not included in WHO 2013):
  1. Typical/ conventional/ classical type
  2. Cellular (atypical features) type
  3. *Angiolymphoid hyperplasia with eosinophilia (ALHE)*

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1. Epithelioid haemangioma

Previously: angiolymphoid hyperplasia with eosinophilia or histiocytoid hemangioma

- Benign vascular tumour in soft tissue
- Bone: locally aggressive, rarely metastasizing → intermediate category!
- Most common localization:
  - Soft tissue: head & neck (periauricular), trunk, limbs, penis and deep soft tissue
  - Bone: any place, but up to 40% in the long tubular bones
- Up tot 25% multifocal at presentation
- No (or rarely) metastases, but up to 30% recurrences

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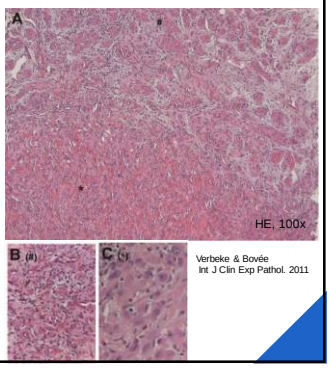
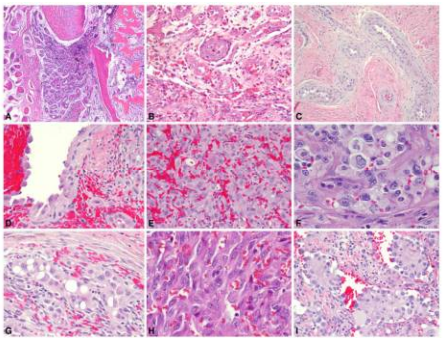
Histology - Typical/ classical/ conventional type

- Circumscribed, lobular proliferation
- Typical zonation:
  - Well-formed capillaries at the periphery (#)
  - Compressed vessels in the centre (*)
- Hobnail, epithelioid endothelial cells
- Nuclear atypia: absent or mild
- In general no necrosis or pleomorphism
- Mitosis: not numerous
- Association mixed inflammatory infiltrate:
  - Lymphocytes
  - Eosinophils
  - Histiocytes
  - Plasma cells

! Might contain areas of spindling or abundant hemorrhage (Hemorrhagic epithelioid and spindle cell hemangioma - Keel et al. Cancer 1999)



Verbeke & Bovée
Int J Clin Exp Pathol. 2011



Errani et al. Clin Orthop Relat Res. 2012; 470:1498-1506

Histology - Cellular/ with atypical features' subtype (2)

- FOSB-rearranged epithelioid haemangiomas (n =9)
  - Lobulated growth with pushing pattern
  - Variable degree of solid growth: diffuse increased cellularity and nuclear crowding
  - At least focal vasoformative features
  - Varying number of blister cells
  - Eosinophils: only focal
  - Some degree of atypical nuclear features (nuclear enlargement, nuclear grooves and indentations)
  - Vesicular nuclei with open chromatin and prominent nucleoli.
  - Mitosis: rare
  - 3/9 showed necrosis + otherwise classical EH features
- FOSB-fusion negative epithelioid haemangiomas (n= 37)
  - Less cellular
  - Less solid components
  - Only occasional atypical cytological features
  - 1/37 (3%) showed focal necrosis.

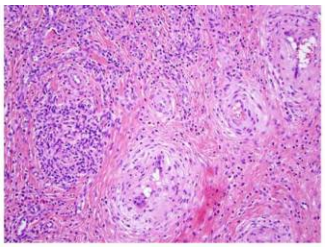
Histology - Cellular/ with atypical features' subtype (1)

- Antonescu et al. Gen Chrom cancer 2014:
- 2 index-cases
  - Worrisome histological (increased cellularity, necrosis, nuclear atypia) and radiological (bone/ cortical destruction) features
  - Less vasoformative, more cellular/ solid or sheet-like growth pattern (>50% of the tumour shows a solid growth)
  - Elevated mitotic activity (2-5/10HPF)
- DD Angiosarcoma
- 44 additional EH
- A subset (~20% = 9/46 cases) show recurrent FOSB gene rearrangement
  - Mostly FOSB gene fused with different partners
  - t(19;19)(q13.2;q13.2) or del19(q13.2-3) resulting in a ZFP36-FOSB gene fusion
  - t(3;19)(q25;q12) resulting in a WWTR1-FOSB gene fusion

HOWEVER: FOSB gene rearrangements are not specific for EH, it can be observed in pseudomyogenic haemangioendothelioma!

Histology – *Angiolymphoid hyperplasia with eosinophilia (ALHE)*

- The epithelioid endothelial cell-lined blood vessels are obscured by a prominent inflammation
  - Lymphoid follicles
  - Eosinophils
- Angiocentric distribution around a lager vessel with evidence of mural damage



Hung et al. Am J Surg pathol. 2016

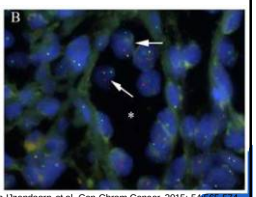
Other molecular changes in epithelioid haemangiomas

1. Huang et al Am J Surg Pathol 2015, 39(10): 1313-1321

- Cohort of 58 EH
  - all lacked FOSB gene abnormalities by FISH
  - Subclassification: typical (25), cellular (21) and ALHE (12)
- Index case: FOS-LMNA gene fusion
- 57 cases -> 17 (=29%) FOS gene rearrangements most in bone (n = 10, 59%) and mostly in cellular variant (12) and also in typical variant (5). None in ALHE.

2. van IJzendoorn et al. Gen Chrom Cancer. 2015; 54:565-574

- 11 EH of bone
  - 1 index case with FOS-MBNL1 translocation
- 2-color FOS break apart FISH
  - 5 FISH break-apart + , only 4 could be scored
- RT-PCR: 1 FOS-VIM fusion and 1 FOS-lincRNA fusion (RT-PCR)



van IJzendoorn et al. Gen Chrom Cancer. 2015; 54:565-574

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Differential diagnosis of epithelioid haemangioma

- Low power examination:
  - Lobular growth pattern?
  - Maturation or vascular channel formation at the periphery?

➡ in favour of EH

- Epithelioid haemangioendothelioma
  - Characteristic strands or cords of epithelioid cells
  - Abundant glassy eosinophilic cytoplasm
  - Prominent cytoplasmic vacuolation
  - Distinctive chondromyxoid or hyalinized stroma
  - Lack of well-formed vascular channels
    - EXCEPTION: TFE3-rearranged EHE (vascular lumen formation, more cytological atypia and strong TFE3 expression)
- (Epithelioid) angiosarcoma
  - Significant cytological atypia: hyperchromasia and nuclear pleomorphism
  - Brisk mitotic activity
  - Infiltrative growth with irregular anastomosing vascular channels

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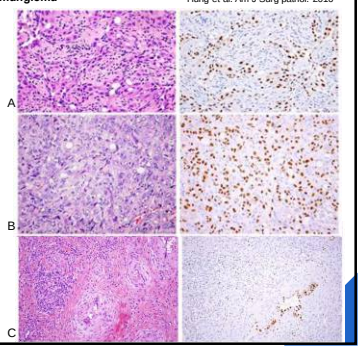
FOSB immunohistochemistry in epithelioid haemangioma

Hung et al. Am J Surg pathol. 2016

- Hung et al. Am J Surg Path. 2016: FOSB IHC in pseudomyogenic haemangioendothelioma and epithelioid haemangioma (control group)
- FOSB expression in 54% (13/24) of the epithelioid haemangioma

A. Conventional EH ~75% (6/8)
B. Cellular EH ~10% (1/10)
C. ALHE ~100% (6/6)



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Treatment and prognosis of epithelioid haemangioma

- Treatment options:
  - Curettage
  - Limited local surgery: marginal en bloc resection
  - No conclusive benefit of chemotherapy or radiation therapy
- Excellent prognosis
  - Errani et al. 2012: 17 patients all NED (<-> WWTR1-CAMTA1 + EHE: 23% died)
  - van IJzendoorn et al. 2015: 11 patients all NED

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2. Pseudomyogenic haemangioendothelioma

- Rare tumour
- Wide age range (14-80y), but mean early to mid-30s (94% between second and fifth decade)
- Marked male predominance (4.6:1)
- Intermediate biological potential: indolent course with low metastatic potential
- Predilection for the extremities (78%), trunk (18%), head and neck (4%)
- Less frequent, but does occur in bone (lower extremity > spine & pelvis >upper extremity)
- Frequent multifocal presentation,
  - Å different tissue planes (up to 2/3)
- Clinically: broad differential diagnosis

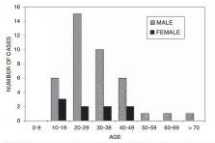

FIGURE 1. Age and sex distribution of 50 pseudomyogenic hemangioendotheliomas.

Hornick et al. Am J Surg Pathol. 2011; 35:190-201

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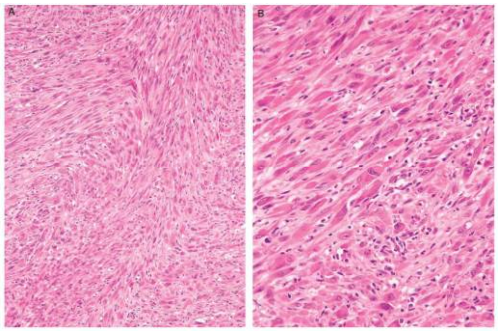

Hornick et al. Am J Surg Pathol. 2011; 35:190-201

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Histology

- Infiltrative margins
- Loose fascicles or sheet of plump spindled and epithelioid cells
  - Spindle cells: vesicular nuclei, variably prominent nucleoli, abundant bright eosinophilic cytoplasm
  - Epithelioid cells (minority)
- Often rhabdomyoblast-like morphology
- Mild to moderate nuclear atypia
- Mitoses ranged from 0 tot 10 / HPF
- Variably present:
  - Å Vascular invasion
  - Å Foci of necrosis
  - Å Focally myxoid stroma
  - Å Neutrophilic infiltrate

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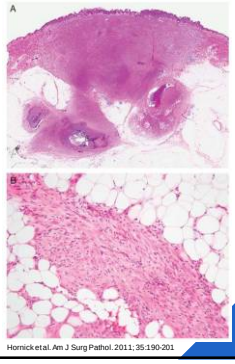
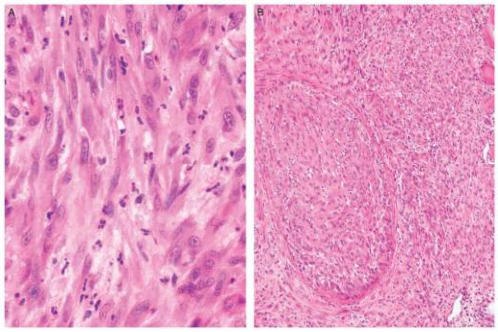


Hornick et al. Am J Surg Pathol. 2011; 35:190-201



Hornick et al. Am J Surg Pathol. 2011; 35:190-201

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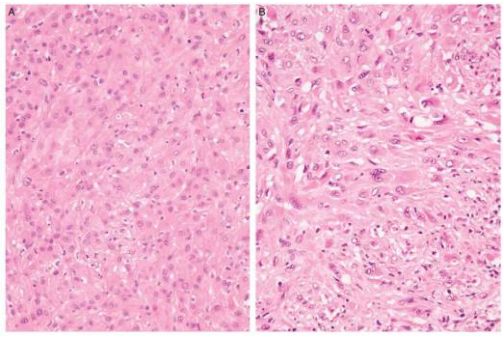


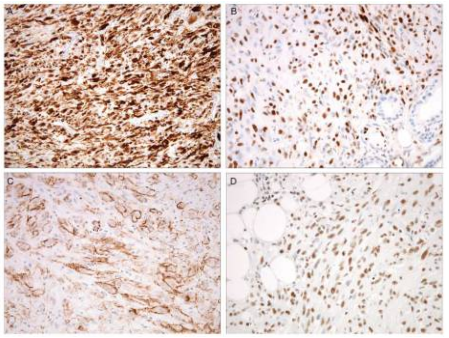
Hornick et al. Am J Surg Pathol. 2011; 35:190-201

CK AE1/AE3

FLI1

CD31

INI1



Hornick et al. Am J Surg Pathol. 2011; 35:190-201

Immunohistochemistry

- Co-expression of endothelial markers (FLI, ERG, CD31) and keratins (AE1/AE3)
- Negative for CD34!
- INI1 (SMARCB1) intact expression

TABLE 3. Results of Immunohistochemical Studies on Pseudomyogenic Hemangioendotheliomas

Antigen	0	1+	2+	3+	4+	Total (%)
PAN-K	46	0	0	1	0	1 (2)
AE1/AE3	0	0	2	3	45	50 (100)
CAM5.2	14	12	7	1	1	21 (60)
EMA	42	2 wk	2 wk	3 wk	0	7 (14)
CD34	50	0	0	0	0	0 (0)
CD31	25	4	10	6	2	22 (47)
FLI-1	0	0	2	0	32	34 (100)
S-100	46	0	0	0	0	0 (0)
SMA	28	2	11	1	0	14 (33)
Desmin	46	0	0	0	0	0 (0)
INI1	0	0	0	0	48	48 (100 intact)

0 indicates no staining; 1+, < 5% tumor cells reactive; 2+, 5 to 25% tumor cells reactive; 3+, 26% to 50% tumor cells reactive; 4+, > 50% tumor cells reactive; EMA, epithelial membrane antigen; SMA, smooth muscle actin.

Hornick et al. Am J Surg Pathol. 2011; 35:190-201

Molecular data in pseudomyogenic haemangioendothelioma

- 2011: first t(7;19)(q22;q13) chromosomal aberration described in PHE
  - Displaying a SERPINE1-FOSB fusion gene
- Analysis of additional cases ï Walther et al. J Pathol. 2014
  - 2 cases with fresh frozen material
  - 10 additional cases (FFPE)
  - FISH analysis: fusion signal in at least 20% of the nuclei (cut-off)
- Also present in bone lesions!

Table 1. Clinical and genetic data on 12 pseudomyogenic haemangioendotheliomas

Case	Sex*	Age**	Tumour location	Cytogenetics	FISH†	RNA-Seq‡	RT–PCR
1	F	14	Chest wall	t(7;19)(q22;q13)	ND	SERPINE1 nt 100 771 438 FOSB nt 45 973 894	SERPINE1 ex1/FOSB ex2
2	M	14	Foot (bone)	t(7;19)(q22;q13)	29%	SERPINE1 nt 100 770 709 FOSB nt 45 971 519	SERPINE1 ex1/FOSB ex1
3	F	41	Calf	ND	42%	ND	ND
4	M	28	Shoulder	ND	80%	ND	ND
5	M	18	Foot (bone)	ND	20%	ND	ND
6	M	21	Hand	ND	29%	ND	ND
7	M	32	Foot	ND	35%	ND	ND
8	M	43	Penis	ND	32%	ND	ND
9	M	18	Thigh	ND	Failure	ND	ND
10	M	35	Spermatic cord	ND	38%	ND	ND
11	F	30	Finger	ND	51%	ND	ND
12	M	19	Foot	ND	Failure	ND	ND

Walther et al. J Pathol. 2014

FOSB immunohistochemisty in pseudomyogenic haemangioendothelioma

TABLE 1. Summary of Immunohistochemical Staining for FOSB

Tumor Type	Total Cases	FOSB Positive (%)*	0	1+	2+	3+	4+
Pseudomyogenic hemangioendothelioma	50	48 (96)	2	0	0	1	47
Epithelioid hemangioma	24	13 (54)	6	4	1	6	7
Conventional	8	6 (75)	0	1	1	4	2
Cellular	10	1 (10)	6	3	0	0	1
Angiolymphoid hyperplasia with eosinophilia	6	6 (100)	0	0	0	2	4
Other endothelial neoplasms and histologic mimics	200	7 (4)	142	42	9	4	3
Epithelioid angiosarcoma	20	1 (5)	11	7	1	0	1
Spindle-cell angiosarcoma	10	1 (10)	9	0	0	1	0
Epithelioid hemangioendothelioma	20	1 (5)	15	4	0	1	0
Epithelioid angiomatous nodule	10	0	9	1	0	0	0
Epithelioid sarcoma	20	0	10	10	0	0	0
Spindle-cell squamous cell carcinoma	20	0	16	4	0	0	0
Spindle-cell rhabdomyosarcoma	20	0	19	1	0	0	0
Leiomyosarcoma	20	0	18	2	0	0	0
Cellular benign fibrous histiocytoma	20	0	12	4	4	0	0
Nodular fasciitis	20	2 (10)	7	7	4	2	0
Proliferative fasciitis	20	2 (10)	16	2	0	0	2

0, <5%; 1+, 5% to 25%; 2+, 25% to 50%; 3+, 50% to 75%; 4+, 75% to 100%.
*FOSB positivity was defined as moderate-to-strong nuclear staining in at least 50% of cells.

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Hung et al. Am J Surg Pathol. 2017

PITFALL: Wounds and surgical sites!

- a moderate-to-strong nuclear staining in endothelial cells + reactive myofibroblasts in granulation tissue and developing scar tissue
- HOWEVER: mature scare tissue -> only limited nuclear staining in rare cells

IHC for FOSB must be interpreted in the context of histological features and other immunophenotypical findings

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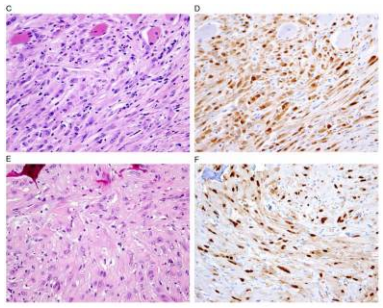
Diffuse nuclear immunoreactivity for FOSB (>50% of cells)



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Hung et al. Am J Surg Pathol. 2017

Differential diagnosis of pseudomyogenic haemangioendothelioma (1)

- Dermis & subcutaneous tissue:
  - Epithelioid sarcoma
    - Lacks plump, myoid-appearing spindle cells
    - No fascicular and sheet-like growth
    - IHC: Keratins+ , but also EMA+ and CD34+ (50%), loss of INI1
  - Spindle cell (sarcomatoid) squamous cell carcinoma
    - Sun-damaged skin of elderly
    - Not Fli1+ and CD31+
  - Cellular benign fibrous histiocytoma
    - Lack of: plump cytomorphology, intense cytoplasmic eosinophilia and neutrophilic infiltrate
    - No keratin expression
  - Other myofibroblastic and smooth muscle neoplasms
    - No strong and diffuse keratin expression, desmin+

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Differential diagnosis of pseudomyogenic haemangioendothelioma (2)

- Deep-seated tumours
  - Epithelioid sarcoma
  - Epithelioid haemangioendothelioma
    - Cords or strand of epithelioid cells
    - Chondromyxoid stroma
    - Intracytoplasmatic vacuoles
    - IHC: some CD34+, keratin+/- (but not strong and diffuse)
  - Epithelioid angiosarcoma
    - Can also grown in sheets
    - Associated with stromal hemorrhage + vasoformative architecture
    - Larger amphophilic epithelioid cells
    - Higher degree of nuclear atypia
  - True myogenic tumours
    - PHE lack expression of desmin and Myf4
  - Nodular and proliferative fasciitis
    - Lack intense cytoplasmic eosinophilia
    - Show less nuclear atypia
    - IHC: no diffuse keratin+, Fli1 and CD31-

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Treatment and prognosis of pseudomyogenic haemangioendothelioma

- Hornick et al. AM J Surg Pathol. 2011
  - Most lesions were treated with local excision
  - ~58% developed local recurrences
  - Only 1 patient had a solitary regional lymph node metastasis
  - 1 patient (1/50) developed distant metastases
- van IJzendoorn et al. Clin Cancer Res. 2018
  - 1 patient with advanced unresectable PHE -> no respons to docetaxel
  - Durable complete remission phase I trial for telatinib
  - Indirectly affects the expression of SERPINE1-FOSB

Conclusion:
- Significant risk for locoregional recurrence
- Overall indolent behaviour
- Low risk of distant metastasis

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3. Epithelioid haemangioendothelioma

- Rare vascular tumour, first described in 1982 (Weiss and Enzinger)
- Occurs both in soft tissue, viscera (liver & lungs) and bone (50% long tubular bones extremities)
- Malignant vascular tumour (low-grade)
- All ages, peak 4-5th decades
- Slight female predominance
- Attempt to stratified into 2 risk groups based on mitotic activity (> 3/50 HPF) and size (> 3 cm)
  - Classic EHE
  - Malignant EHE

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Histology of epithelioid haemangioendothelioma

- Infiltrative growth pattern
- Lack of well-formed vascular channels
- Cords or strands of epithelioid cells
  - Dense, eosinophilic cytoplasm
- Only immature, intra-cytoplasmatic lumina (blister cells)
- Majority has a myxoid/ chondromyxoid/ sclerotic extracellular stroma
- Most tumour low nuclear grade
- Subset has a higher-grade morphology -> ĩmalignant EHEò

- Expression of endothelial markers (CD31, CD34, Fli1, ERG)
- 25-40% epithelial markers (keratins, EMA)

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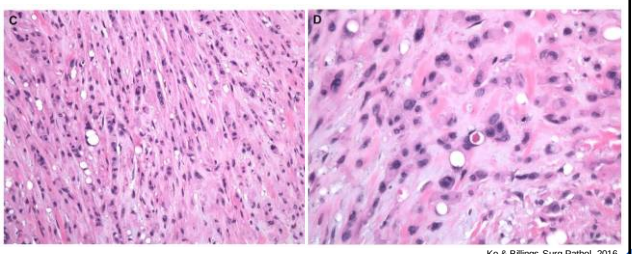

Ko & Billings Surg Pathol. 2016

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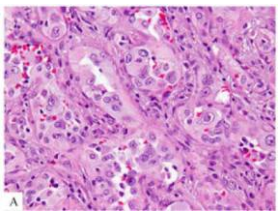
Molecular data in epithelioid haemangioendothelioma (2)

- Small group EHE WWTR1-CAMTA1 fusion ï and have unusual morphological features:
  - Moderate tot voluminous eosinophilic cytoplasm
  - Mature vascular channel formation
  - Some solid growth pattern
  - Minimal intervening stroma
  - At least mild nuclear atypia
  - Mitotic activity < 3/10HPF
  - (necrosis in a minority)
- Other typical EHE features: intra-cytoplasmatic vacuoles
- IHC
  - Strong + for CD31, ERG
  - Nuclear reactivity for TFE3
- 6 patient FU >1 y
  - 5 evidence of metastatic disease
  - 1 died after 17 y



Antonescu et al. Gen Chrom Cancer 2013

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Molecular data in epithelioid haemangioendothelioma (1)

- Mendlick et al. 2001: t(1;3)(p36;q25) in EHE of liver and soft tissue
- Errani et al. 2011
  - identified in all 17 (100%) EHE a rearrangement in CAMTA1 on 1p36.3 and WWTR1 on 3q25.1
  - Not in epithelioid haemangiomas (13), pseudomyogenic haemangioendotheliomas (4) or epithelioid angiosarcomas (5)
- Tanas et al. 2011 analyzed 47 EHE tumours +118 other vascular neoplasms
  - 87% a rearrangement of the CAMTA1 region
  - 89% a rearrangement in WWTR1 region
  - 0% in other 118 vascular neoplasms

➡ Majority of EHE WWTR1-CAMTA1 fusion gene + (prevalence ~90%*)

*Doyle et al. Am J Surg pathol. 2016

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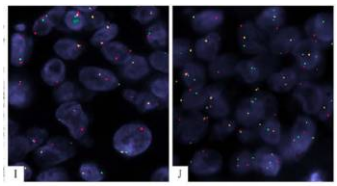
Molecular data in epithelioid haemangioendothelioma (3)

- 10/10 TFE3 break-apart signal
- 8/10 YAP1- break-apart signal



➡ Morphologically and genetically a distinct subgroup: TFE3-rearranged EHE

TFE3 IHC strong and diffuse nuclear staining pattern =>useful marker as screening method of epithelioid vascular tumours

HOWEVER: Flucke et al. 2014 demonstrated TFE3+ in WWTR1-CAMTA1 EHE => antibody dependent!

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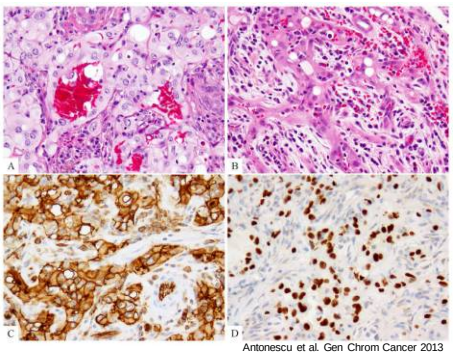
CD31



TFE3

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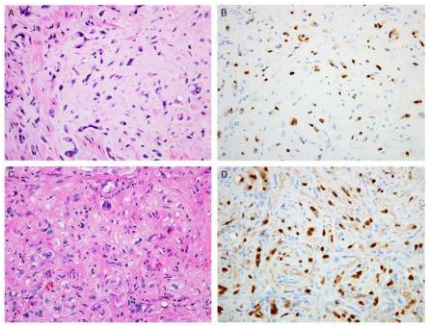
Antonescu et al. Gen Chrom Cancer 2013



CAMTA1

CAMTA1

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Doyle et al. Am J Surg pathol. 2016

Doyle et al. 2016:

- WWTR1-CAMTA1 fusion gene results in overexpression of both genes
  - Â WWTR1: expressed in many different cell types

➡ limited diagnostic potential

- Â CAMTA1 expression: limited to brain

➡ Nuclear expression is a highly sensitive and specific marker for EHE

DD EHE w/ atypical features vs ep. AS

! Importance of antibody selection (previous study all tumours CAMTA1 +)

! 2 cases with conventional EHE morphology, CAMTA1- and TFE3+

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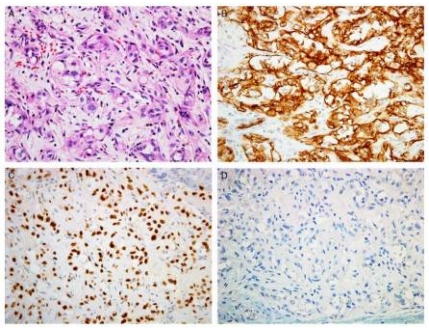
TABLE 1. Summary of IHC Staining for CAMTA1 in EHE and Other Epithelioid Mesenchymal Tumors

Tumor Type	Total Cases	CAMTA1 Positive (%)
EHE	59	51 (86)*
Epithelioid hemangioma	20	0 (0)
Epithelioid angiomatous nodule	10	0 (0)
Epithelioid angiosarcoma	25	1 (4)
Composite hemangioendothelioma	5	0 (0)
Pseudomyogenic hemangioendothelioma	10	0 (0)
Epithelioid sarcoma	25	0 (0)
Sclerosing epithelioid fibrosarcoma	10	0 (0)
Myoepithelial neoplasms of soft tissue	10	0 (0)
PEComa	10	0 (0)
Alveolar soft part sarcoma	10	0 (0)
Ossifying fibromyxoid tumor	10	0 (0)

*Of the 8 CAMTA1-negative tumors, 6 were positive for TFE3. CAMTA1 was positive in 44 of 48 (92%) cases with conventional histology and 7 of 11 (64%) cases with "malignant" histology.

CD31



TFE3

CAMTA1

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Doyle et al. Am J Surg pathol. 2016

Treatment and prognosis of epithelioid haemangioendothelioma

- Treatment: surgery (wide resection)
- Local recurrence rate 12-13%
- Metastatic rate up to 20-30%
- 15% of patients die of their disease
- Based on the risk stratification (mitoses/size):
  - 5y-disease specific survival 59% (> 3/50HPF, > 3cm)
  - 5y-disease specific survival 100%

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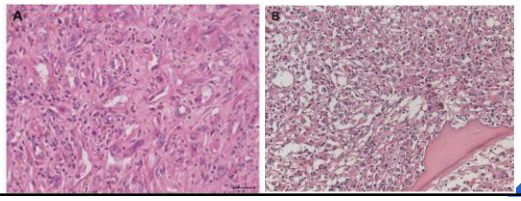
Histology – angiosarcoma (1)

- Multinodular, hemorrhagic mass
- Secondary cystic degeneration and necrosis
- Wide spectrum of morphological appearances + multiple patterns within 1 tumour
  - Areas of well-formed, anastomosing blood vessel
  - Solid sheet of high-grade epithelioid or spindled cells without clear vasoformation



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4. Angiosarcoma

- The most accepted term for high-grade vascular malignancy
- Any body part, but soft tissue (2%) > bone (rare, <1% of all bone tumours)
  - Skin/ Head & Neck
  - Deep muscles of the lower extremities (40%)
  - Retroperitoneum, mediastinum, mesentery
  - Breast: radiation induced angiosarcoma
- Wide age distribution, but peak at 7th decade
- Male > female
- Highly aggressive behaviour
- Dismal overall survival

Panel 1: Risk factors for angiosarcoma

- Radiation
- Chronic lymphoedema (Stewart-Treves syndrome)
  - Postsurgery or radiotherapy
  - Milroy's syndrome
  - Other types of chronic lymphoedema
- Exogenous toxins
  - Vinyl chloride
  - Thorium dioxide
  - Arsenic
  - Anabolic steroids
  - Foreign bodies
- Familial syndromes
  - Neurofibromatosis NF-1
  - Mutated *BRCA1* or *BRCA2*
  - Maffucci syndrome
  - Klippel-Trenaunay syndrome

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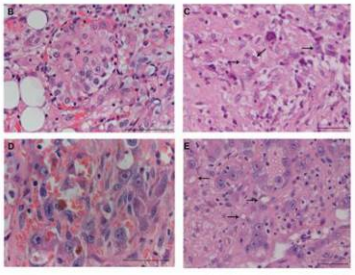
Histology – angiosarcoma (2)

- When predominance of epithelioid cells

-> epithelioid angiosarcoma
  - Abundant amphophilic to lightly eosinophilic cytoplasm
  - Large vesicular nuclei
  - Prominent nucleoli
- DD: metastatic carcinoma

- Other general features:
  - Brisk mitotic activity
  - Necrosis
  - Significant nuclear atypia



Verbeke et al. Histopathology 2011

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Molecular data in angiosarcoma (primary and radiation induced) (1)

- Cytogenetic analysis:
  - Most AS: complex karyotypes without recurrent chromosomal changes
- Gene expression profiling
  - Distinct upregulation of genes related to angiogenesis and endothelial cell receptor
- Recurrent somatic mutations
  - Involving angiogenic signalling pathways ~40% (KDR, PTPRB, PLCG1)
  - Rare mutations: RAS, PTEN, PIK3CA, TP53, FLT4, TIE1
- High-level of MYC amplification
  - Hallmark of most post-radiation and chronic lymphedema-associated AS
    - MYC FISH or IHC dd post-radiation AS versus AVL
  - Small subset of primary AS
- Underlying pathogenesis of most angiosarcomas remains undefined

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Molecular data in angiosarcoma (primary and radiation induced) (2)

Huang et al Am J Surg Pathol. 2016: cohort of 120 AS

- 34 (28%) men and 86 (72%) women
- Mean age 58y (range 13-91y)
- 73 primary AS (breast, H&N, extremity, viscera, trunk, body cavity)
  - Mammary (30%), soft tissue (25%), cutaneous (19%), viscera (15%) and intra-osseous (11%)
- 47 secondary AS
  - Radiation breast cancer (33), radiation other tumours (9) and long-standing lymphedema (5)
  - Interval radiotherapy and AS diagnosis ~11.3 y (range 4-70y)
- Largest molecular investigation:
  - CIC-rearrangement
  - PLCG1/KDR mutations
  - MYC and FLT4 amplifications

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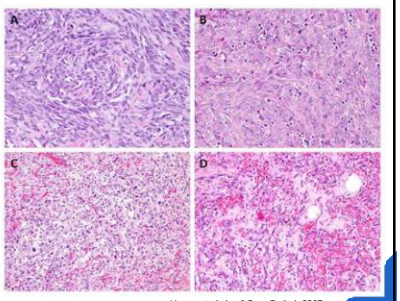
Molecular data in angiosarcoma (primary and radiation induced) (3)

- 9% AS: various **CIC alterations**
  - Gene rearrangements and/or missense mutations
  - Most common in soft tissue tumours
  - Younger patients (mean 41y)
  - 8 primary AS, 1 had radiation therapy 35y ago (no MYC amplification)
  - 1 tumour showed co-existing CIC and PLCG1 mutation
  - Morphology: epithelioid morphology, classic morphology and well-differentiated anastomosing vascular channels



Huang et al. Am J Surg Pathol. 2067

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Molecular data in angiosarcoma (primary and radiation induced) (4)

- 9.5% (11/116) AS harbored **PLCG1 mutation**
  - 10 female, 1 male
  - Mean age 53 y (range 23-82y)
  - Even distribution primary and secondary AS (all MYC amplification +)
  - Mutually exclusive from KDR mutations
  - Wide morphological spectrum (well formed vascular spaces tot solid sheets of spindled to epithelioid endothelial cells
- 7% (8/113) AS harboured a **KDR missense mutation**
  - Exclusively female
  - Mean age 56y (range 23-77y)
  - 5 mutations in primary breast AS, 1 lumbar spine AS, 2 secondary AS (radiotherapy for breast tumour, also MYC amplification+)
  - Wide spectrum of low to high grade phenotype, variable vasoformation and solid growth
- KDR and PLCG1 genes are involved in the VEGFR2 signalling pathway

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Molecular data in angiosarcoma (primary and radiation induced) (5)

- 35% (39/112) AS showed a **MYC gene amplification**
  - Secondary AS
    - sAS-RT for breast tumour: 90% (28/31)
    - sAS-lymphedema ass: 100% (4/4)
    - sAS-RT other tumours: 25% (2/8)
  - Primary AS 7% (5/69)
- 5.5% (6/110) As showed a **FLT4 gene amplification**
  - 5 co-amplified with MYC
  - 5 occurred in secondary AS related tot radiotherapy for breast cancer or lymphedema
  - 1 primary AS (scalp, cutaneous)
  - Mostly solid, high-grade histology with variable epithelioid and spindled cell phenotype

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Treatment and prognosis of angiosarcoma

- Treatment: surgery (wide resection) with or without systemic therapy
- Recurrences are common
- Metastases are common
- 5-year survival ~ 30%

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Multifocality

- Errani et al. 2011: multifocal EHE of the liver -> identical WWTR1-CAMTA1 fusion in different foci
- van IJzendoorn et al. 2015: 2 EH of bone -> multifocal lesions showed identical fusion product
- Monoclonal origin
- Exclusion of the germline-translocation theory
- ñMultifocal regional spreadò instead of ñmetastasisò

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NEW immunohistochemical panel – vascular tumours

- Vascular markers:
  - CD31
  - CD34
  - FLI1
  - ERG
- Pericytes:
  - SMA
- Lymphatic differentiation
  - D2-40 (podoplanin)
  - prox1
- Epithelial markers (especially in epithelioid variants)
  - Keratin
- New markers :
  - FOSB (50% EH, ~ 100% PHE)
  - INI1 (SMARCB1, retained in PHE)
  - CAMTA1 (EHE)
  - TFE3 (CAMTA1- EHE)

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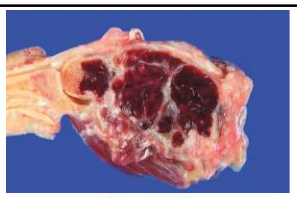
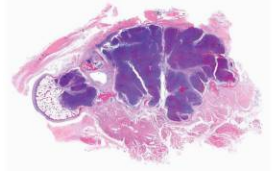
Soft tissue versus Bone

- Most vascular tumours do occur in soft tissue AND bone
- Similar genetic alterations
- Some of these entities have a distinct behaviour in bone, e.g. epithelioid haemangioma
- Vascular tumours of bone are more frequently multifocal





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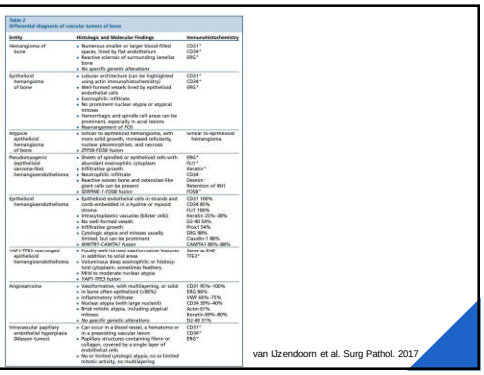

Table 2
Differential diagnosis of vascular tumors of bone

Entity	Histologic and Molecular Findings	Immunohistochemistry
Hemangioma of bone	• Numerous smaller or larger blood-filled spaces, lined by flat endothelium • Reactive sclerosis of surrounding lamellar bone • No specific genetic alterations	CD31+ CD34+ ERG+
Epithelioid hemangioma of bone	• Lobular architecture (can be highlighted by reticulin or immunohistochemistry) • Well-formed vessels lined by epithelioid endothelial cells • Eosinophilic infiltrate • No prominent nuclear atypia or atypical mitoses • Hemorrhagic and sclerotic cell areas can be prominent, especially in central lesions • Rearrangement of *FOS*	CD31+ CD34+ ERG+
Atypical epithelioid hemangioma of bone	• Similar to epithelioid hemangioma, with more solid growth, increased cellularity, nuclear pleomorphism, and necrosis • *FOS* rearrangement	Similar to epithelioid hemangioma
Pseudomyogenic (epithelioid sarcoma-like) hemangioendothelioma	• Sheets of spindled or epithelioid cells with abundant eosinophilic cytoplasm • Infiltrative growth • Neutrophilic infiltrate • Reactive woven bone and osteoidal-like giant cells can be present • *SERPINE1-FOSB* fusion	ERG+ FLI1+ Keratin+ CD10- Desmin- Retention of INI1 SMA-
Epithelioid hemangioendothelioma	• Epithelioid endothelial cells in strands and cords embedded in a hyaline or myxoid stroma • Intracytoplasmic vacuoles (blister cells) • Thrombosed vessels • Infiltrative growth • Cytologic atypia and mitoses usually limited, but can be prominent • *WWTR1-CAMTA1* fusion	CD31 100% CD34 85% FLI1 100% Keratin 25%–30% CAMTA1 100% Prox1 54% ERG 90% Claudin-1 88% CAMTA1 86%–88%
YAP1-TFE3 rearranged epithelioid hemangioendothelioma	• Focally well-formed vascular lumina in addition to solid areas • Vacuolated cytoplasm, eosinophilic or histiocytoid cytoplasm, sometimes feathery • Mild to moderate nuclear atypia • *YAP1-TFE3* fusion	Same as EHE TFE3+
Angiosarcoma	• Vasoformative, with multilayering, or solid growth of epithelioid cells (>90%) • Inflammatory infiltrate • Nuclear atypia (with large nucleoli) • Brisk mitotic atypia, including atypical mitoses • No specific genetic alterations	CD31 90%–100% CD34 70% VWF 60%–75% CD34 39%–40% Actin 67% Keratin 89%–80% ZG-45 31%
Intravascular papillary endothelial hyperplasia (Masson tumor)	• Can occur in a blood vessel, a hematoma or a vascular malformation (>90%) • Papillary structures containing fibrin or collagen, covered by a single layer of endothelial cells • No or limited cytologic atypia, no or limited mitotic activity, no multilayering	CD31+ CD34+ ERG+

van IJzendoorn et al. Surg Pathol. 2017