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MINI REVIEW ARTICLE

Immunohistochemistry of Epithelioid Soft Tissue Sarcomas, Literature Review Based on Case Studies

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Abstract:

Neoplasms with epithelioid histology may be diagnostically challenging. Immunohisto chemistry (IHC) can aid in confirming the differential diagnosis of mesotheliomas, melanomas, lymphomas, and soft tissue sarcomas, all tumors that can present with an epithelioid histology. Immunohistochemistry can also assist in confirming the type of sarcomas. Using cases diagnosed in a community hospital setting over a ten year period, the use of IHC in sarcomas will be illustrated.

Introduction:

Epithelioid neoplasms tend to have bland/non-specific histological features, which can make diagnosis especially challenging. Cells vary in morphology based on tumor location, but characteristically appear as a monomorphic proliferation of cuboidal to polygonal cells with extensive eosinophilic cytoplasm thus resembles somewhat epithelial cells. Morphology of specific epithelioid tumors will be discussed later for each example.

In addition to mistaking epithelioid neoplasms for more common epithelial neoplastic processes, pathologists may mistake their bland appearance and a heavy epithelioid cell infiltrate for more common granulomatous pathologies or microbial infections [1- 2]. Clinical history and special stains for particular organisms can help in such situations and infectious processes such as *Mycobacterium avium* infections and leprosy can thus be ruled out.

Soft tissue sarcomas have presented with different morphologic features including epithelioid histology. Most patients will present at a community hospital setting with a mass lesion. The mass is often mistaken for a non-sarcomatous lesion, as sarcomas are fortunately extremely rare. In 2011, according to the National Cancer Institute, only 10,980 people in the United States will be diagnosed with a sarcoma. These estimates have been made based on data supporting the fact that the incidence of soft tissue sarcomas has been stable over the last 30 years [3]. Since sarcomas can be overlooked it is imperative that pathologist retain soft tissue sarcomas as a differential diagnosis until they can be ruled out with certainty.

Examination of histological features alone can leave the pathologist with an extensive list of differential diagnoses. Fortunately, epithelioid neoplasms, and soft tissue sarcomas often express characteristic patterns of markers that can be used to identify such tumors. Thus, the use of immunohistochemistry is often essential in the formulation of a definitive diagnosis.

The examples presented are from a surgical pathology practice at a 200 bed community hospital in the US. In many cases, only a hematoxylin and eosin stain was performed in the initial diagnostic work ups with further characterization obtained through consultative immunohistochemistry. These cases are supplemented with information regarding immunohistochemical markers gathered from a thorough literature review.

Case 1: Epithelioid Melanoma :

86 year old female has presented with bleeding and a urethral lesion

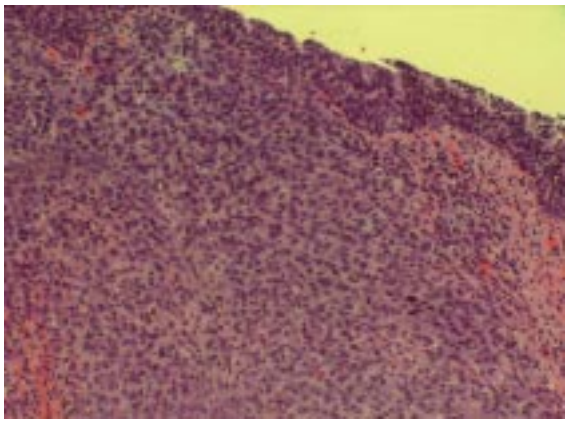

Fig. 1: Urothelium with an underlying tumor infiltrating large sheets. H&E; 10X.

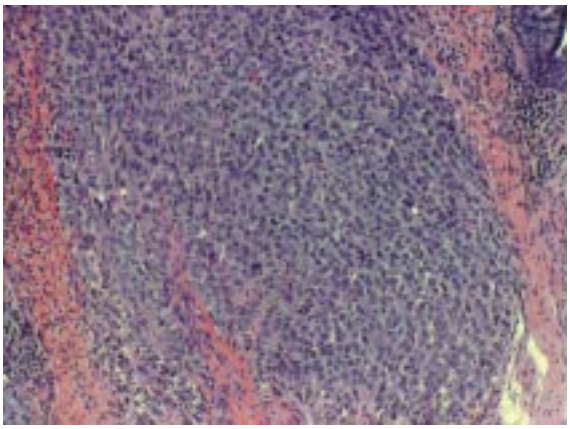

Fig. 2: Higher power showing cells with abundant cytoplasm. H&E; 40X.

This case shows overlying urothelial epithelium, with an invasive tumor infiltrating sub epithelial tissue in broad sheets. The pattern of invasion is unusual for an urothelial carcinoma. On higher magnification, the tumor cells show abundant cytoplasm, central nuclei and prominent nucleoli. Though the tumor mimics an epithelial malignancy, it is indeed a melanoma. The difficulty in diagnosis is due to the unusual location of the melanoma, and the fact that it is a non-pigmented melanoma. A prior history of a melanoma at this site, made the diagnosis easier in this instance.

Although melanomas are not most commonly found in the urethra, melanoma of the genitourinary tract is most likely found in the urethra [4-5]. Melanoma of the genitourinary tract is rare and accounts for less then 1% of all melanomas [6]. Not much is known about the cause or pathogenesis of melanoma of the urethra; however, epidemiological studies have shown that these lesions are more common in black women between the ages of 50 and 70. The clinical presentation is nonspecific and consists of dysuria or hematuria due to their presence in the distal urethra. Other sites of melanoma in the genitourinary tract include the glans penis and labia which may progress to a distal urethra melanoma. Interestingly on examination not all melanomas may produce melanin and the cells may be papillary or spindled making diagnosis difficult [5]. The prognosis for malignant melanomas is poor even in cases where the lesion is localized as in the case of urethral melanoma seen in a community hospital [7].

Histologically, melanomas can be classified as epithelioid and/or spindled in appearance. Cells

tend to be large and round with abundant/ eosinophilic cytoplasm, features that often mimic poorly differentiated malignant neoplasms [8-9] and make diagnosis challenging. To make a diagnosis, most physicians utilize immunohistochemistry [10] in melanomas occurring in unusual sites, such as internal organs and viscera.

A conventional panel of S100 antibody and HMB45 antibody is often used in the clinical setting to identify malignant melanoma [8, 11]. Recently, however, several other markers have been identified that may have equal or greater sensitivity and specificity. One recent study found that an immunohistochemical panel consisting of Melan-A and microphthalmia transcription factor (MiTF) had a sensitivity and specificity of 95% and 100%, respectively, while a panel consisting of S100 protein and HMB45 had a sensitivity of 80% and a specificity of 100% [8]. MiTF is indicative of melanin synthesis [12-13] and can be used to determine the amount of intraepidermal melanocytes [10]. These results are promising; however, a larger scale study is needed. WT1 is expressed in 88% of epithelioid melanomas and 100% of spindled cell and desmoplastic melanomas [11]. However, WT1 is also expressed in several other neoplasms limiting its specificity, and does not have the ability to

Table 1: Immunostaining in Epithelioid Melanomas and Common Differential Diagnoses

IHC Profile	Epithelioid Melanoma [8-11,28,10, 60]	Benign Melanocytic Nevi [26,25,10]	Spindled Cell Melanoma [8,11]	Spitz Nevi (28,10)	GIST [29]
S100	+	+	+		-
S100á		+			
S100â		-			
HMB-45 (epidermal)		+			
HMB-45 (dermal)		-			
HMB-45	+		-		
Diffuse HMB-45				+	
Melan-A	+	+			+/-
A-103	+	+			+/-
Tyrosinase	+	+			
MiTF	+	+			
WT1	+		+		
CyclinD1	- (84%)			+ (74%)	
p21	- (73%)			+ (91%)	
CD117 (c-kit)					+
Other Diagnostic Clues	High Ki-67 proliferative index	Low Ki-67 proliferative index			

distinguish between epithelioid and spindle cell melanomas. CXCR4 is a marker that is believed to be involved in angiogenesis and metastasis of cutaneous melanoma, and was recently found to be predictive of epithelioid type uveal melanoma, indicating a worse prognosis relative to the non-epithelioid type [14]. It is unknown whether these results can be extrapolated to cutaneous melanoma and used as a similar prognostic marker. Distinction of skin lesions based on morphology and clinical findings remains a challenge. Immunohistochemistry may assist this differentiation in some circumstances; however, specific markers for many pathological entities have yet to be discovered. A summary of common immunohistochemical markers expressed in epithelioid melanoma, as well as common differential diagnoses, is displayed below in Table 1.

Current work is being done to develop the optimal set of immunohistochemical markers to diagnoses melanoma. It has been suggested by Preito and Shea that the use of a "pan-melanocytic cocktail" containing HMB-45, anti-MART1, anti-tyrosinase, is widely used; however, a different mixture containing anti-MART1 and anti-Ki-67 may more easily illustrate the amount of melanocytes undergoing mitosis [10]. Other markers of increasing clinical value include the following: NKI-C3 [12], p16 [13], galectin-3 [15], Cox-2 [16], TRP [17], surviving [18], and claudin-1[19].

Melanoma can be differentiated from a nevus using HBM-45 and Ki-67. This is due to the fact that these markers are usually present with in the epidermis and periepithelial dermis so if the stain remains located to these areas and there are more Ki-67 positive cells at the top of the lesion than at the bottom, the lesion is most likely a nevus. After determining that the lesion is a nevus, it must be deduced as to whether it is a Spitz nevus or a blue nevus. Spitz nevi generally stain diffusely for HMB-45 but do not stain Ki-67. Blue nevi mimic the presentation of malignant blue nevi however they can be differentiated by gp100 which is focal in malignant lesions but diffuse in a benign lesion. How invasive the lesion is can also be determined using these markers as the more invasive lesions have higher rates of proliferations and differentiation. Desmoplastic melanoma can be distinguished from a desmoplastic nevus using MART1 which is not usually present in melanomas but S100 protein most likely would be present. S100 can also be used to gauge invasion however it should not be confused with scar tissue [20-21]. The amount of S100 positivity in scars is lower than that seen in desmoplastic melanomas and scars do not express p75 [22]. The presence of Lentigo Maligna or melanoma in situ can stain positively for anti-MART1 and HMB-45 in a converging pattern as opposed to pigmented actinic keratosis which has a less confluent pattern. The usefulness of anti-MART1, however, has been challenged due to its ability to detect melanocyte dendrites and thus mimic a converging pattern of labeling which may be misleading [23]. This confusion can be avoided by staining with HMB-45[24] or anti- MiTF [10].

Of note, Spindled cell melanoma also exhibits WT1 and S100 positivity, but is routinely negative for HMB45.

Benign melanocytic nevi are commonly positive for all markers of melanogenesis (MelanA, Tyrosinase, MiTF, and HMB45); however, the staining pattern for HMB45 differs from malignant melanoma. In benign nevi, HMB45 stains strongly in the epidermis but show a loss of HMB45 expression with progressive descent into the dermis [25]. Some cases have shown aberrant dermal expression of HMB45, however, making distinction difficult. One case series showed that á subunit of S100 is commonly expressed in the junctional nests of dysplastic junctional nevi, however á subunit expression does not occur in benign nevi [26]. The authors theorized that á subunit expression relates to vertical progression of malignant melanomas. Proliferative index (Ki-67) is another useful tool that helps the clinician to differentiate between benign and malignant lesions [27]. Normally, no more than 1% of cells will test positive for Ki-67; however, in melanomas, the mean proliferative fraction is greater than 10% of the lesion [10].

Spitz nevus is another melanocytic lesion that is difficult to distinguish from malignant melanoma. One case series has reported the most significant difference being overexpression of cyclin D1 and p21 in Spitz nevi compared with non-spitzoid melanomas (74 vs. 16% and 91 vs. 27%, respectively) [28]. Some Spitz nevi lesions may also stain completely for HMB-45 [10]. Morphology and clinical findings may be needed in conjunction with immunohistochemical findings to make an accurate diagnosis.

Stage IV melanoma has a 26-58% probability of metastasizing to the gastrointestinal tract, where it can mimic epithelioid gastrointestinal stromal tumor (GIST) [8, 29]. Epithelioid GISTs can be positive for Melan-A, favoring a diagnosis of metastatic melanoma, however, Melan-A reactive cases have been found to be negative for S100 and positive for CD117 [29], confirming the diagnosis of GIST.

Case 2: Malignant Mesothelioma:

Patient has presented with a lumbar mass and an enlarged lymph node.

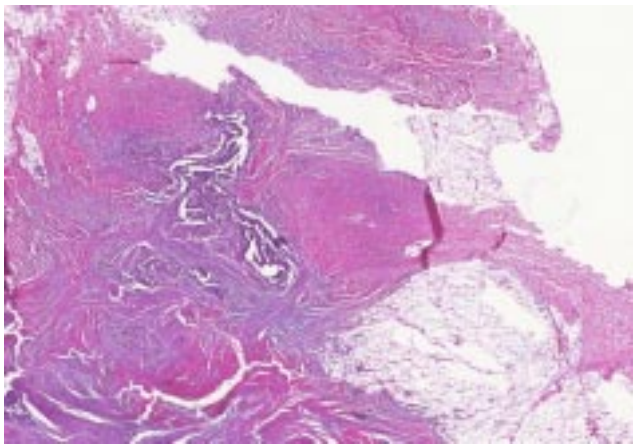

Fig. 3: Malignant mesothelioma.: Desmoplastic reaction surrounding a papillary tumor pattern. H&E; 10X

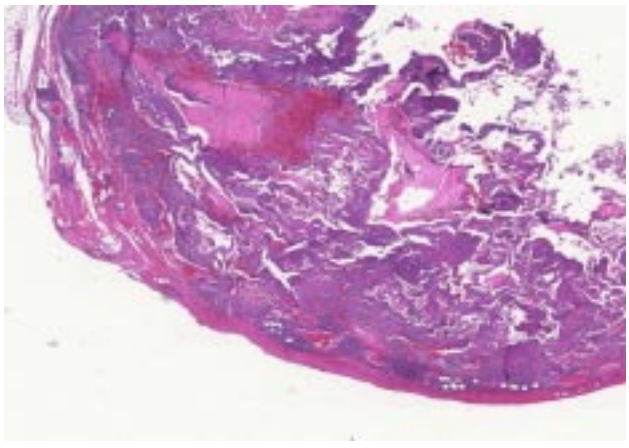

Fig. 4: Malignant mesothelioma involving a lymph node. H&E; 10X

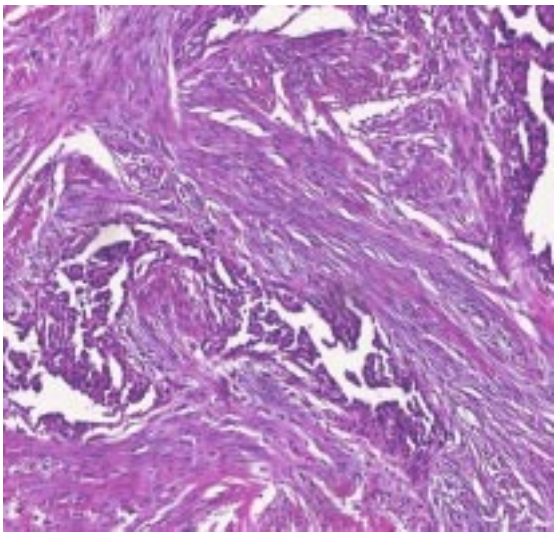

Fig. 5: High Power view showing the invasive tumor with desmoplastic reaction; H&E; 20x

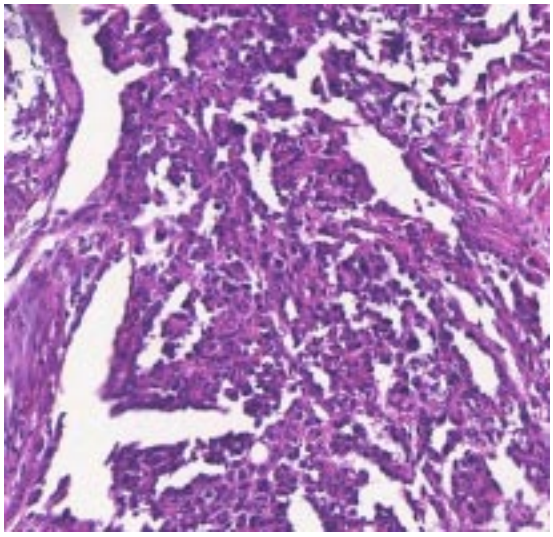

Fig. 6: High Power view showing Papillary tumor. H&E; 40x

The patient from whom the figures above have been obtained, has presented to the hospital with a lumbar mass and lymphadenopathy. Upon resection and histological processing it is evident that the patient has a malignant neoplasm due to the invasive nature of the growth and the desmoplastic reaction that can be seen in the figures above. The cells present in the mass are not sufficient to diagnose the lesion or determine the primary source of the neoplasm. Immunohistochemical stains are ordered to obtain a definitive diagnosis of malignant mesothelioma. Involvement of a lymph node by malignant mesothelioma is rare, and presentation of mesothelioma as an enlarged lymph node is even rarer. On H&E sections, it is difficult to discern the malignant mesothelioma cells within the lymph node; however, on immunohistochemical stains the lymph node involvement is obvious. Without prior knowledge of malignant mesothelioma in the patient, diagnosing the lymph node as involved by mesothelioma would have been difficult.

Malignant mesothelioma (MM) is a rare neoplasm usually of the visceral or parietal pleura that often spreads widely along the pleura and may invade adjacent thoracic structures. Malignant mesothelioma may also arise from other serous surface of the body, such as the peritoneum and pericardium, however the most common location is the pleura [30]. There are three commonly described subtypes: epithelioid, sarcomatoid, and biphasic [31]. Epithelioid mesothelioma often shows dense cellularity with stromal invasion and occasional necrosis, with cells displaying scalloped borders [32].

It is often difficult to distinguish MM from several other pathological entities given its bland morphology, diffuse and invasive nature, and penchant for mimicking other more common neoplasms clinically & radiologically. These include lung adenocarcinoma, reactive mesothelial cell hyperplasia, and metastasis to the pleura or peritoneum from other primary

sites. Table 2 below helps distinguish epithelioid mesothelioma from morphologically similar clinical entities based on immunohistochemical profile.

The most difficult, and frequently encountered, entity to distinguish from malignant mesothelioma is lung adenocarcinoma. It is especially challenging to make a definitive diagnosis, since lung adenocarcinoma may involve the periphery of the lung and spread along the pleura, mimicking mesothelioma radiologically [33]. Immunohistochemistry is an integral tool that must be used to help make this distinction [32]. Several studies have attempted to define the most sensitive and specific immunohistochemical marker panel to make this distinction, since a unique marker for mesothelioma has yet to be found. However, consensus is lacking on which panel is best, although the use of both positive and negative mesothelial markers is widely suggested [32, 34]. One study has recommended that MOC-

Table 2: Immunostaining in Epithelioid Mesothelioma & Common Differential Diagnoses

IHC Profile	Mesothelioma [35, 34, 60]	Pulmonary Adenocarcinoma [34-36]	Reactive Mesothelial Cell Hyperplasia [61]	Metastasis from Other Primary Site
WTI	+	-		Use tissue specific immunomarkers for suspected site
CK5/6	+	-		
D2-40	+	-		
Podoplanin	+	-		
Calretinin	+	-		
Mesothelin	+			
Vimentin	+			
MOC31	-	+		
BG8	-	+		
TTF1	-	+		
Factor VIII	-			
CD31	-			
CD34	-			
Fli-1	-			
SMA	+ (42.3%)		- (90.9%)	
Calponin	+ (38.9%)		- (95.5%)	
Desmin	- (90.0%)		+ (86.4%)	
Other Diagnostic Clues				*Clinical and radiological findings suggest metastasis

31, BG8, CK5/6 and WT1 be used to diagnose epithelioid mesothelioma [35]. However, it is noted that WT1 and CK5/6 do not distinguish between benign and malignant mesothelial cell proliferations. WT1 is also expressed in ovarian serous carcinomas and some renal neoplasms, while CK5 is also expressed on squamous epithelia reducing the utility of these as specific markers for mesothelioma [8]. D2-40, podoplanin, and caveolin-1 have been described in some case series as having utility in differentiating mesothelioma from adenocarcinoma, however, further research is needed [36,37].

Another study has found that using calretinin, BG8, and MOC-31 provides over 96% specificity & sensitivity for distinguishing epithelioid mesotheliomas from adenocarcinoma of the lung [34]. Calretinin is expressed on mesothelial cells, while BG8 and MOC-31 are used as negative mesothelial markers. Mesothelin and WT1 are excluded from this panel, given that they are also expressed in ovarian serous carcinomas.

Distinguishing epithelioid mesothelioma from reactive mesothelial cell hyperplasia also presents a clinical challenge. To date no single marker specific to either condition has been identified, and again a panel of markers is required. A study during 2010 has found that desmin is expressed in 86.4% of non-neoplastic mesothelial cells (NMC), while it is only expressed in 10% of epithelioid mesothelioma (EM) tumor cells [7]. The same study has found that smooth muscle actin (SMA) is expressed in 42.3% of EM cells and 9.1% of NMC, while muscle specific actin (MSA) is only expressed in EM; however the positivity rate is low. A 2009 statement from the International Mesothelioma Interest group confirms the utility of desmin, and also suggests the use of EMA and p53, which are more often positive in benign proliferations of mesothelial cells [32].

Finally, primary malignant mesothelioma must be distinguished from primary malignant neoplasms of other sites that have metastasized to the pleura. Malignant tumors from the breast, ovary, prostate, colon, and kidney commonly metastasize to the pleura, and thus it is important to use tissue specific immunohistochemical markers [5]. For example, TTF-1 can determine the lung origin of carcinoma, while RCC Ma can identify those of renal origin [36].

Case 3: Lennert's lymphoma:

46 years old female has presented with a right groin lymph node.

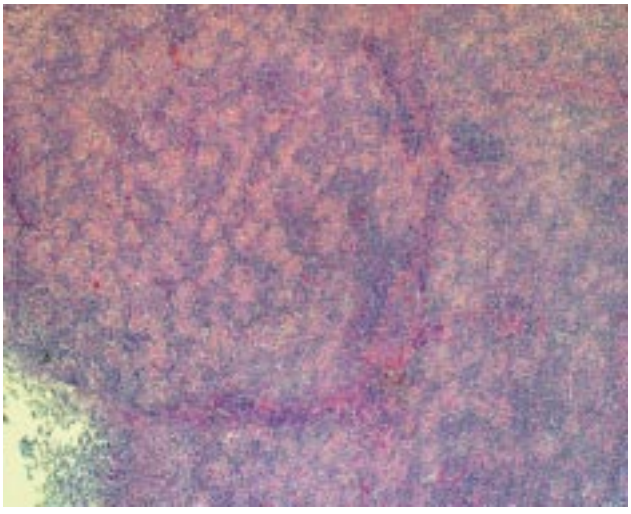

Fig. 7: Lymph node showing numerous clustered epithelioid histiocytes, imparting it a moth-eaten appearance, H&E; 10X

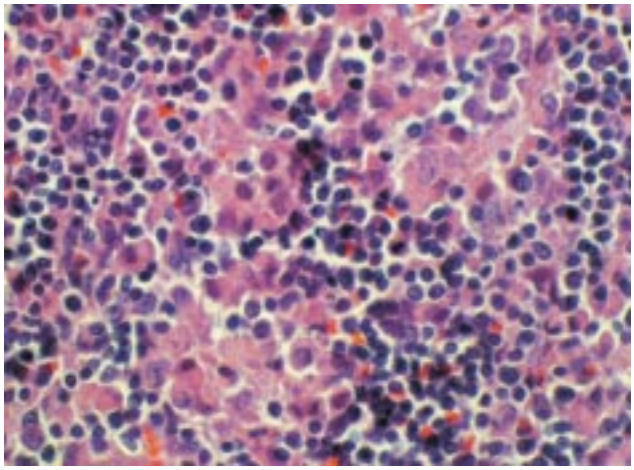

Fig. 8: High power view showing epithelioid granulomas. H&E; 40X

H&E sections show lymph node fragments with complete effacement of normal architecture by an infiltrate of predominantly small to medium-sized lymphocytes with condensed chromatin, irregular contours, and scant cytoplasm. Occasional large lymphoid cells and eosinophils are admixed. Of note, numerous clustered epithelioid histiocytes are present throughout the lesion, imparting it a moth-eaten appearance (Figure-7). The H&E stained sections show clusters of cells with abundant eosinophilic cytoplasm with large nuclei and prominent nucleoli. The initial diagnosis has been a reactive lymph node with sinus histiocytosis. However, immunohistochemical markers to rule out a monoclonal population have revealed a Lennert's lymphoma. The diagnosis would have been missed without the IHC stains. PCR studies have shown clonal rearrangement of the TCR gamma gene. IHC studies have revealed the majority of the lymphoid infiltrate to be comprised of T-lymphocytes with diffuse expression of CD3, CD5, and CD2. Overall, the morphologic, immunophenotypic, & molecular findings have been consistent with involvement by a peripheral T-cell lymphoma, not otherwise specified. The morphology is consistent with the entity historically described as lymphoepithelioid lymphoma by Lennert.

Lennert's lymphoma is classified by the World Health Organization as "lymphoepithelioid cell variant of the peripheral T-cell lymphomas, unspecified" due to the lack of consensus regarding from which cell this lymphoma is derived. Several conflicting studies have suggested that this lymphoma may be CD4, CD8 or even helper-T cell derived [38-40]. Histologically, Lennert's lymphoma appears as a proliferation of atypical small lymphocytes with a large presence of admixed epithelioid histiocytes [38, 41].

It is important to differentiate Lennert's lymphomas from other, more common, lymphoma variants, as well as other pathologies that have a prominent epithelioid histiocyte infiltrate. Table 3 below demonstrates the immunohistochemical profiles of Lennert's lymphoma and clinical entities for which it may be mistaken.

As it is evident from the table, similarly presenting diseases can be ruled out using a specific panel of immunohistochemical markers. Hodgkin lymphoma is positive for CD15 and CD30, unlike Lennert's lymphoma, and shows characteristic Reed Sternberg cells. The neoplastic cells in SHML and LCH are both positive for S100 and CD68, while only the background epithelioid cells in Lennert's lymphoma may be positive for these markers (the neoplastic lymphocytes will be negative).

Table 3: Immunostaining in Lennert's Lymphoma and Common Differential Diagnoses

IHC Profile	Lennert's Lymphoma [38-41,1]	Hodgkin Lymphoma [41-62]	Sinus Histiocytosis with Massive Lymphadenopathy [63,64,41]	Langerhans Cell Histiocytosis [63, 64, 41]
CD1a			-	+
CD2	+			
CD3	+			
CD4	+/-			
CD5	+			
CD8	+/-			
CD15	-	+		
CD20	-			
CD23			-	
CD30	-	+	-	
CD31				+
CD35			-	
CD56	-			
CD68			+	+
S100	-		+	+
Other Diagnostic Clues	Background Epithelioid	Reed Sternberg Cells	Emperipolesis	

Case 4: Leiomyoma:

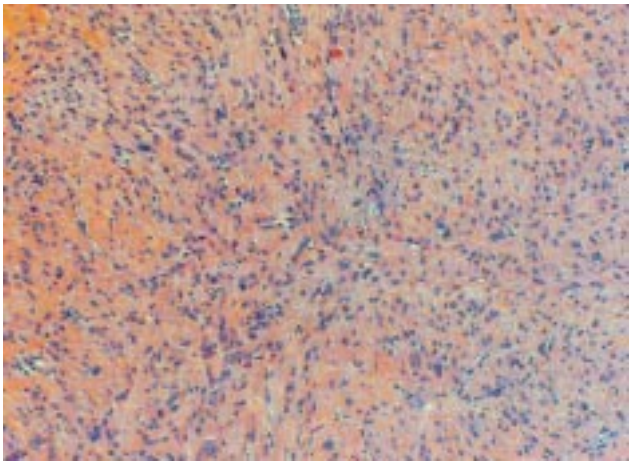

Fig. 9: Epithelioid cells arranged in a spindled pattern. H & E 20X

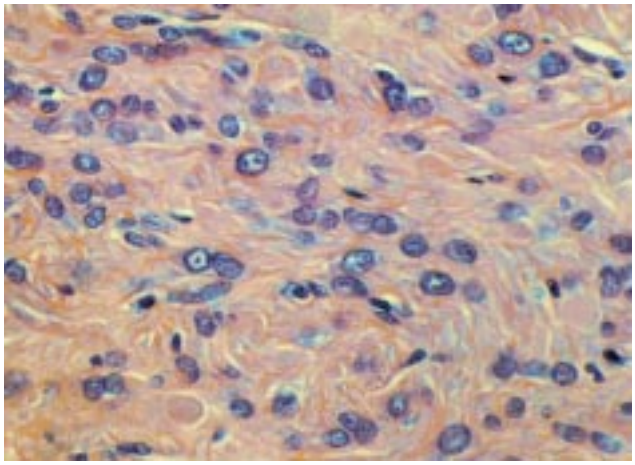

Fig. 10: High Power view showing cells with abundant cytoplasm. H&E; 40x

48 year old premenopausal woman has presented with menorrhagia, with a potential diagnosis of leiomyoma. Figures 9 and 10 illustrate a neoplasm that is not a typical leiomyoma. The abundant cytoplasm and cellularity may lead to a mis-diagnosis of a leiomyosarcoma. Leiomyoma is a benign neoplasm of smooth muscle origin. It most commonly appears as spindle cells arranged in a whorled or interlaced fascicular pattern, though cells can be pleomorphic, epithelioid, or with multinucleated giant cell features [42-43]. The epithelioid cells are often plump; spindle shaped and show deeply eosinophilic cytoplasm, and may dissuade one from the diagnosis of a benign leiomyoma. Table 4: below illustrates typical immunohistochemical profiles of leiomyoma and other neoplasms that may arise as differential diagnoses.

Leiomyosarcoma is a malignant neoplasm that is also of muscular differentiation. It is sometimes positive for smooth muscle antigen (SMA), although has been found to show no staining for both estrogen receptor (ER) and progesterone receptor (PR) in one case series [44]. Spindle cell carcinoma can be excluded via immunostains for cytokeratin and EMA, which are negative in leiomyoma. Perivascular epithelial cell tumor can be excluded via demonstration of negative staining for melanogenesis markers (MiTF, tyrosinase, MelanA).

Ki-67 proliferative index also holds some weight in differentiating benign and malignant neoplasms. Leiomyomas typically stain negatively or focally for Ki-67, while more malignant lesions such as leiomyosarcoma and spindle cell carcinoma show diffuse staining for Ki-67 [45-46].

Table 4: Immunostaining in Leiomyoma and Common Differential Diagnoses

IHC Profile	Leiomyoma [42- 45]	Leiomyosarcoma [44,46]	Spindle Cell Carcinoma [45]	Perivascular Epithelial Cell Tumor (PEComa) [65, 10]
SMA	+	+/-		
Vimentin	+			
PR	+	-		
ER	+/-	-		+
Desmin	+/-			
HMB-45	+/-			
Cytokeratins	-		+	+
EMA	-		+	+
S100	-			+
MiTF	-			+
Tyrosinase	-			
A-103				
Melan-A	-			

Case 5: Myxoid Liposarcoma:

87 years old male has presented with a right groin mass

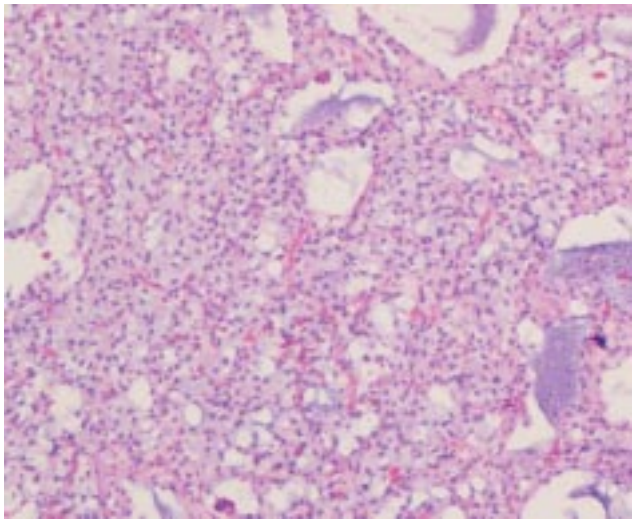

Fig. 11: Cellular neoplasm with microcystic areas. H&E; 20X

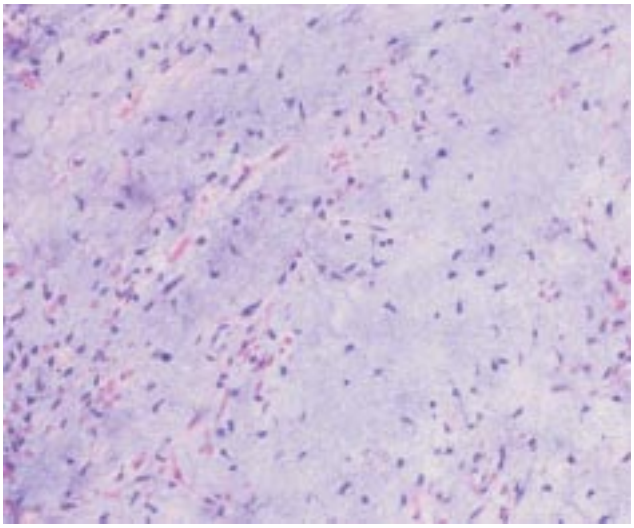

Fig. 12:Tumor heterogeneity with focal areas showing a pure myxoid neoplasm. H&E;40X

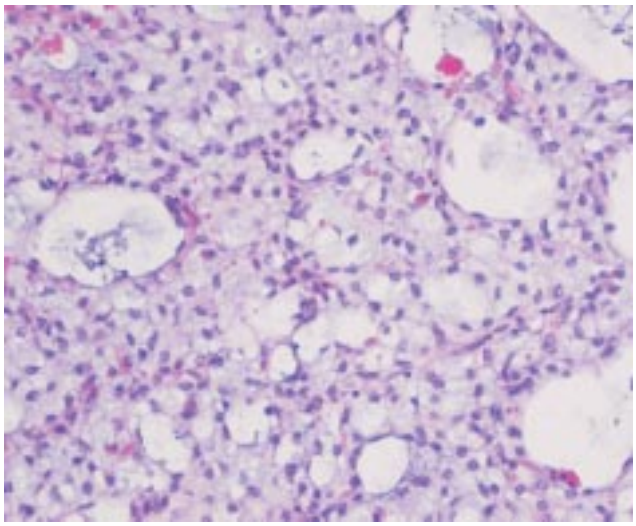

Fig. 13:High power view showing microcystic areas. H&E; 20X

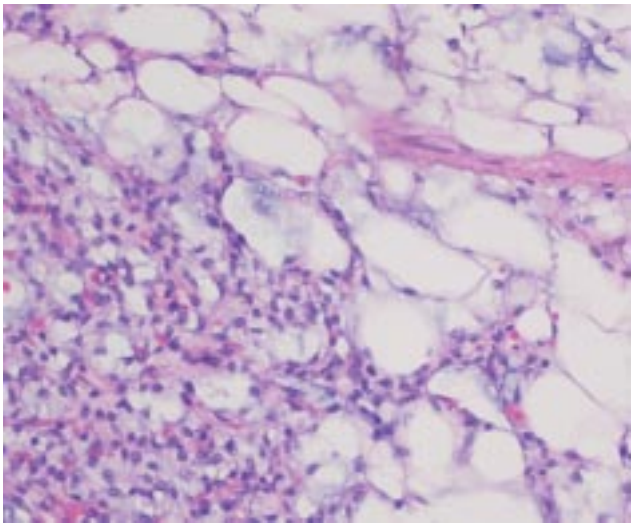

Fig. 14:Lipocytes are seen adjacent to cellular myxoid areas. H&E; 40X

The patient from whom this biopsy has been excised has been an 87 year old man with the mass in his groin. Initially, the mass has been thought to be a hernia. However, at surgery, the surgeon has found it difficult to enucleate completely, and it has been apparent that this has not been a hernia, but has been indeed a neoplasm. The advanced age of the patient has raised the suspicion for malignancy. Soft tissue neoplasms can present at unusual locations, as seen in this instance. Figures 11 through 14 are representative of this myxoid

Table 5: Immunostaining in Myxoid Liposarcoma and Common Differential Diagnoses

IHC Profile	Myxoid Liposarcoma [66,49]	Lymphangioma [67]	Hemangioma [66,68]		Pleomorphic sarcoma NOS or MFH [66,69]
Focal SMA	+				+
Muscle Specific	+				+
Actin	-				
Desmin	-				
S100	-				-
Keratins	-				
CD68	-				+
Factor XIIIa	-				
Podoplanin		+			
VEGFR-3		+			
Flt-4		+			
CD34			+/-		
CD31			+	+/- (involuting)	
Fli-1			+		
PCNA			+ (infantile)		
VEGF			+ (infantile)		
Type IV			+ (infantile)		
collagenase					
vWF				+/- (involuting)	
GLUT1				+ (involuting)	
Factor VIII					
Osteocalcinin					-
ALP					-
Other Diagnostic Clues	No specific IHC Markers are known				

liposarcoma. The microscopic findings of myxoid liposarcoma resemble the morphology seen in developing fetal fat. The adipose cells may be present in different stages of development because the cells attempt, albeit unsuccessfully, to differentiate. The bland fusiform cells form nodules that are surrounded by myxoid matrix containing mostly hyaluronic acid. The abundance of myxoid stroma may create a cribiform pattern. If the cells become flattened and do not stain well, they may be mistaken for a lymphangioma or if there is a hemorrhage within the interstitial space it could be misdiagnosed as a hemangioma. The cells within the neoplasm can also become cartilaginous [47-48], leiomyomatous or osseous (myxoid liposarcoma subtypes). Myxoid liposarcomas may be distinguished from myxomas due to the presence of plexiform capillary vasculature [49]. Myxoid liposarcomas generally arise in the lower extremity during the 6th decade of life. Myxoid liposarcoma expresses a variety of proteins which have been listed above in Table 5. The presence of focal SMA & muscle-specific actin indicate that there is a smooth muscle component to the neoplasm. These, however, are not a differentiating factors due to their presence in pleomorphic sarcoma NOS or MFH. Staining for CD68, a glycoprotein found on monocytes, macrophages, and neutrophils, would indicate that the lesion contains histiocytes & is therefore pleomorphic sarcoma NOS or MFH. Hemangioma can be easily excluded if stains reveal the presence of endothelial cell markers such as factor VIII, CD34, CD31 and VEGF. The positive staining of podoplanin in lymphangioma illustrates that there are lymphatic vessels within the neoplasm which are not present in myxoid liposarcoma. Although there are no specific markers known for myxoid liposarcoma, a combination of stains negative for lymphatic, endothelial and histiocyte markers can be used to exclude the differential diagnosis. Furthermore, the presence of smooth muscle proteins can be used to confirm a myxoid liposarcoma [50].

Case 6: Desmoplastic Small Round Cell Tumor:

21 years old with retroperitoneal adenopathy and abdominal mass.

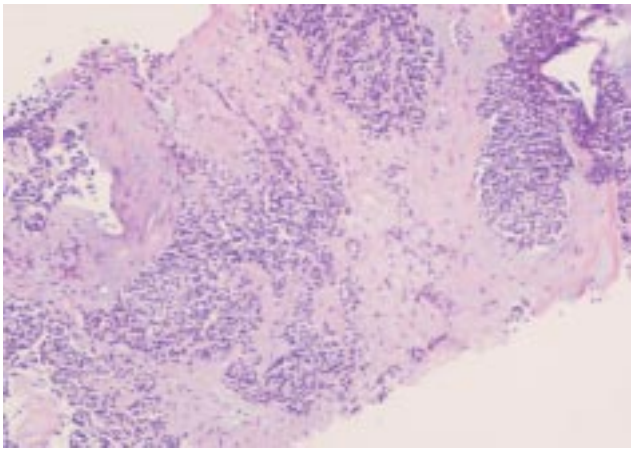

Fig. 15: Core biopsies showing a small round blue cell tumor infiltrating dense stromal tissue; H&E 10X

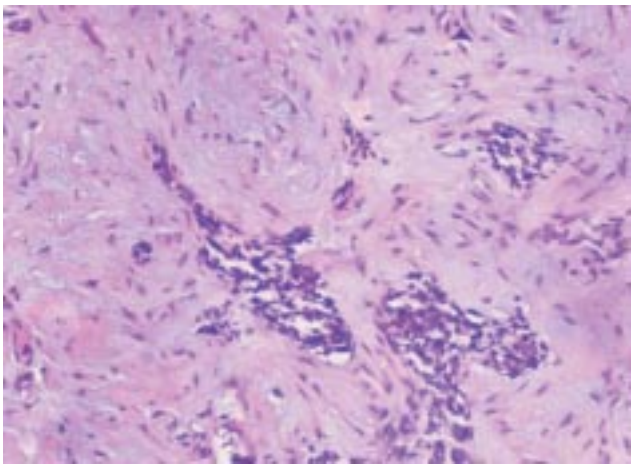

Fig. 16: High power view shows cells with very little cytoplasm. H&E ;40X

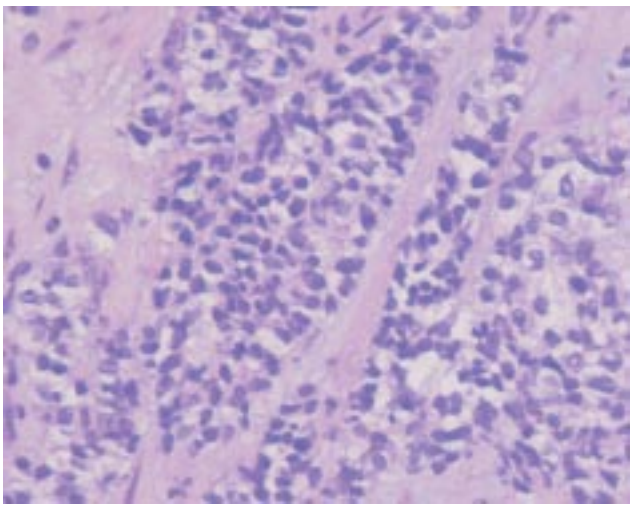

Fig. 17: High power view showing cells with clear cytoplasm in sheets. H&E; 40X

This 21 years old male has presented with a palpable abdominal mass in the emergency room (ER). The patient has been an exchange student from Brazil and with no health insurance. CT scan done in the ER has revealed large retroperitoneal lymphadenopathy and hepatosplenomegaly. The diagnosis after imaging has been an obvious malignancy. Phone calls made to a regional cancer center have asked that referral be made only after a confirmed tissue diagnosis. This has led to an ultrasound guided core biopsy of the mass on an outpatient basis.

At H&E, the small round blue cell tumor has been thought to be extraskeletal primitive neuroectodermal tumors (PNET), rhabdomyosarcoma, neuroblastoma, lymphoma, poorly differentiated carcinoma, small-cell carcinoma, Merkel cell carcinoma. IHC has confirmed the diagnosis as Desmoplastic small round cell tumors. These tumors are usually found in the abdominal and/or pelvic peritoneum of young males as has been seen here. As the name suggests, the tumor is comprised of small round cells surrounded by hypervascular desmoplasia creating rounded nests of cells. The small cells have small dark blue nuclei lacking prominent nucleoli. The cytoplasm of the tumor cells is eosinophilic and not abundant [51]. These characteristics, however, are not diagnostic of DSRCT. Although there is a plethora of differential diagnosis, DSRCT can be distinguished using immunohistochemistry.

Table 6 below lists several different immunohistochemical stains that may be used to diagnose desmoplastic small round cell tumor. EWS-WT1 is a reliable marker when trying to diagnose a desmoplastic small round cell tumor because it is the product of the chromosome 11 and chromosome 22 translocation. More stains need to be conducted if EWS-WT1 results are negative to differentiate between the differential diagnosis of desmoplastic small round cell tumor. PNET neoplasms will contain proteins consistent with neuroendocrine cell origin such as chromogranin and synaptophysin while rhabdomyosarcoma will have muscle cell markers such as myogenin, myoD1, and smooth muscle actin. Lastly Merkel cell carcinoma's epithelial cell origin is apparent due to the positive staining for a variety of cytokeratins which do not stain positively in a case of desmoplastic small round cell tumor.

CK-20 cytokeratin 20, TTF-1 thyroid transcription factor 1, NSE neuron-specific enolase, GrA gromogranin A, NFP neurofilament proteins, CD56 neural adhesion molecule, MAP-2 microtubule associated

Table 6: Immunostaining in Desmoplastic small round cell tumor & Common Differential Diagnoses

Type	DSRCT [70,51]	PNET [66]	Rhabdomyosarcoma [66,71]	Merkel cell carcinoma [72-74,75]
IHC PROFILE	EWS-WT1 + Desmin +/-. If positive will have a dot-like pattern MOC-31 + Ber-EP4 + Leu-M1 + Cytokeratins 5/6 – Thrombomodulin – Cytokeratin 20 – MyoD1 and myogenin – PDGFA+	EWS-WT1 – Fli-1 + Vimentin + S100 + CD56 + Chromogranin + Synaptophysin + Cytokeratin +/- CD99 +	EWS-WT1 – Desmin + Myogenin + MyoD1 + Smooth muscle actin -/+ CK -/+ S100 -/+ Neuroilament -/+ Synaptophysin + (if pure rhabdomyo-sarcoma)	CK8+ CK18+ CK19 + CK20+ (negative in 5-25% of lesions) TTF1 - NSE+ S-100- GrA+/- SYP+/- NFP + CD5+ MAP-2+ LCA-
Other Diagnostic Clues	T(11;22)(q24;q12)			

protein 2, LCA leukocyte common antigen. Other differential diagnoses not included in the table are: neuroblastoma, lymphoma, poorly differentiated carcinoma, small-cell carcinoma.

Case 7: Synovial Sarcoma

65 years old female has presented with large abdominal wall mass measuring 8 x 5.5 x 3.8 cm

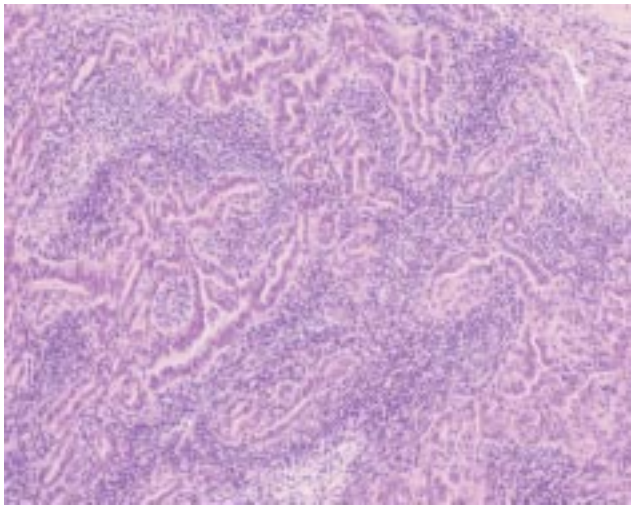

Fig. 18: Biphasic tumor with glandular and stromal elements. H&E; 20X

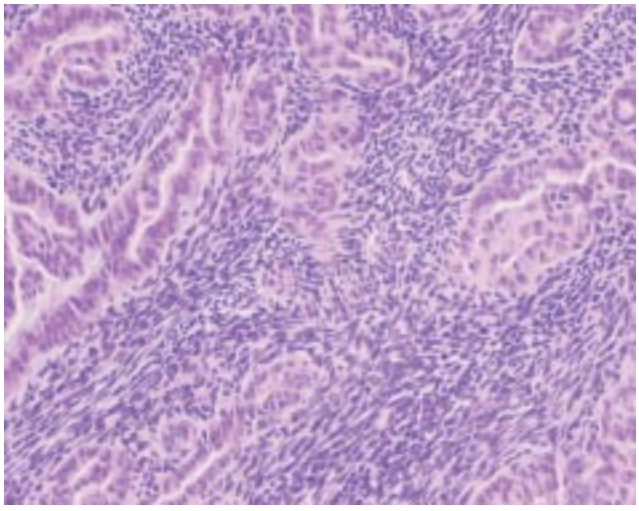

Fig. 19: High power view showing malignant stromal component. H&E; 40X

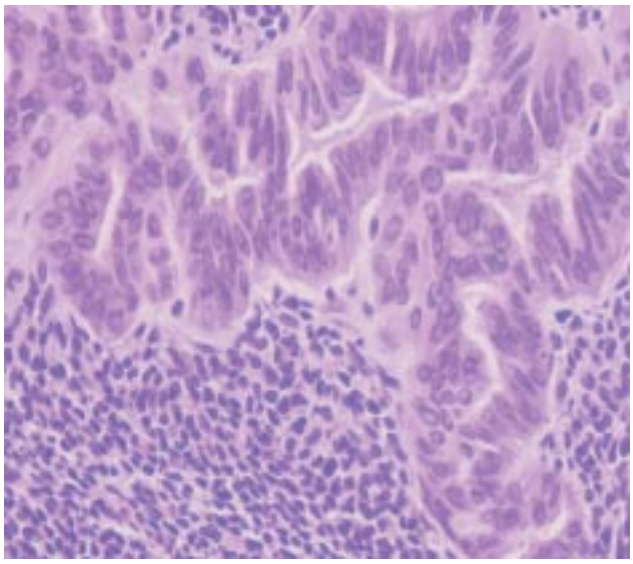

Fig. 20: High power view showing the glandular component. H&E; 40X

This mass has been thought to be metastatic colon carcinoma. At frozen section, a tumor has been diagnosed as a carcinosarcoma. However, IHC stains have revealed it to be a synovial sarcoma.Unlike what their name suggests, synovial sarcomas are not found within the synovial space. Most often these lesions are seen around the joints. Rarely, synovial sarcomas may occur in the parapharyngeal region, retropharyngeal region [52], orofacial region [53], retroperitoneum [54], mediastinum [55], pleura, and heart or, as in the case presented here, in the abdominal wall. In fact this location is very rare for synovial sarcomas. Most abdominal wall sarcomas consist of case reports. Among synovial sarcomas which have been accessioned over a 10 years period at the Armed Forces Institute of Pathology (AFIP), only 2.6% have arisen in the abdominal wall [48].

On histological examination two cell types are apparent: spindle cells and epithelial cells. The ratio of these two cell types had led to subtypes of synovial sarcomas. These subtypes include the biphasic type, monophasic fibrous types, monophasic epithelial type and poorly differentiated (round cell) type. The epithelial cells mimic the epithelial cells within a normal synovium, range from cuboidal to columnar and may arrange themselves in to glands and secrete granular or pink secretions. These epithelial cells may cover papillary structures whose fibrovascular core has been replaced by spindle cells. Due to the epithelial cell content, squamous metaplasia, keratin pearls and keratohyalin granules may be present which leads to the misdiagnosis of squamous cell carcinoma [56]. The spindle cells within synovial sarcoma have scant cytoplasm and dark blue oval nuclei. These cells occur in high densities that can be confused with fibrosarcoma. The characteristics that allow the pathologist to discern that they are in fact looking at a synovial sarcoma are the lack of a herringbone pattern, a more irregular architecture and fewer cells undergoing mitosis [56]. Between the spindle cells and epithelial cells there may be areas of thickened basement membrane, hyaline, myxomatous material or calcifications. The amount of calcification and whether or not this calcification has ossified becomes important when rendering a diagnosis. Other characteristics of biphasic synovial sarcoma include a varying degree of vasculature and the presence of mast cells.

The two ends of the spindle cell and epithelial cell ratio are designated as monophasic fibrous

synovial sarcoma and monophasic epithelial synovial sarcoma. Both contain the characteristics discussed for biphasic synovial sarcomas however the monophasic epithelial subtype of synovial sarcoma is much rarer. Its rarity has led to a discussion about whether this subtype truly exists or whether its low prevalence may be due to the difficulties in excluding other differential diagnoses. These differential diagnoses include metastatic carcinoma, melanoma, adnexal tumors, epithelioid sarcoma, and epithelioid malignant peripheral nerve sheath tumor (MPNST).

The last subtype of synovial sarcoma is the poorly differentiated subtype. All synovial sarcomas independent of the ratio of spindle to epithelial cells may become poorly differentiated. The lack of differentiation poses a problem in diagnosis for the pathologist and worsens the prognosis for the patient. An important differential diagnosis to rule out in a case of poorly differentiated synovial sarcoma is that of malignant hemangiopericytoma due to the abundance of thin walled vasculature.

Table 7: Immunostaining in Synovial Sarcoma and Common Differential Diagnoses

Type	Synovial Sarcoma [66]	Fibrosarcoma [76,77]	MPNST [60]	Hemangiopericytoma [78, 79]	Adnexal carcinoma [80]
IHC PROFILE	Vimentin + EMA + AE1/AE3 + (epithelial cells) CK7 + (epithelial cells) CK8 + (epithelial cells) CK18 (epithelial cells) CK19 (epithelial cells) BerEp4 + E-cadherin + Calretinin +/- S100 +/- CD34 +/- SMA –/+ Desmin -/+ Bcl-2 + (spindle cells) CD99 +	Fibronectin+ collagen type I, III, V + (III>I) collagen type IV- Vimentin + Desmin -	S100+ Factor VIII- CD31- CD34-	CD34+ vimentin+ Actin +/- (focal) SMA +/-(focal) CD 31- cytokeratin- S100 – p75+/-	CK7+ CK20- ER+/- PR+/- GCDFP-15+/- CK5/6 + Podoplanin +
Other Diagnostic Clues	T(x;18) or SYT-SSX fusion transcript				

Poorly differentiated synovial sarcoma may also resemble extraskeletal Ewing's sarcoma/ primitive neuroectodermal tumor (PNET); however, IHC can be utilized in this case.

The abundance of differential diagnoses for synovial sarcoma makes cytogenetic and molecular genetic investigations of suspected cases of synovial sarcoma essential.

Other differential diagnoses not included in the table include carcinosarcoma & mesenchymal chondrosarcoma.

The synovial sarcoma diagnosis is most confidently made using FISH to determine if there is a translocation between the X chromosome and chromosome 18. Immunohistochemical staining can also help determine whether FISH should be ordered.

Depending on the subtype of synovial sarcoma there will be varying degrees of epithelial markers and spindle cell markers may be present. As seen in table 7, neoplasms with a large number of epithelial cells will stain positively for AE1/AE3, CK7, CK8, CK18, and CK19. Therefore monophasic epithelial synovial sarcoma will stain exclusively for these epithelial cell markers. The use of BerEp4 can reliably distinguish epithelial cells from mesothelial cells if the epithelial cells within the synovial sarcoma resemble mesothelial cells [50]. The presence of CD99 within a synovial sarcoma may indicate leukocytic infiltrate or increased vascularization [57].

The figure 22 below shows excised mass from

Case 8: Extraosseous Osteosarcoma of the Chondroblastic Type:

54 years old male has presented with a chin mass.

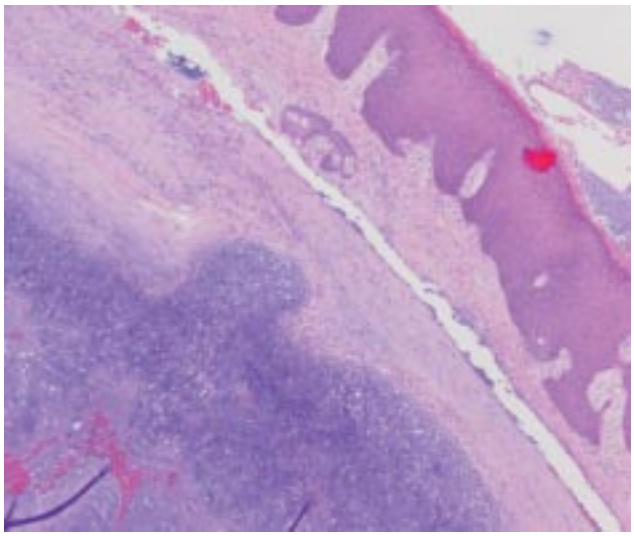

Fig. 21: Sections through the chin mass show overlying skin and a cartilaginous tumor. H&E 10X

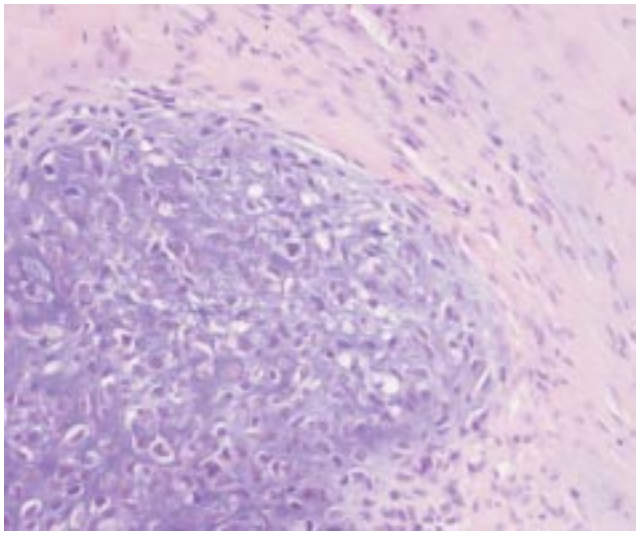

Fig. 22: High power view showing the malignant cartilaginous areas. H&E; 40X

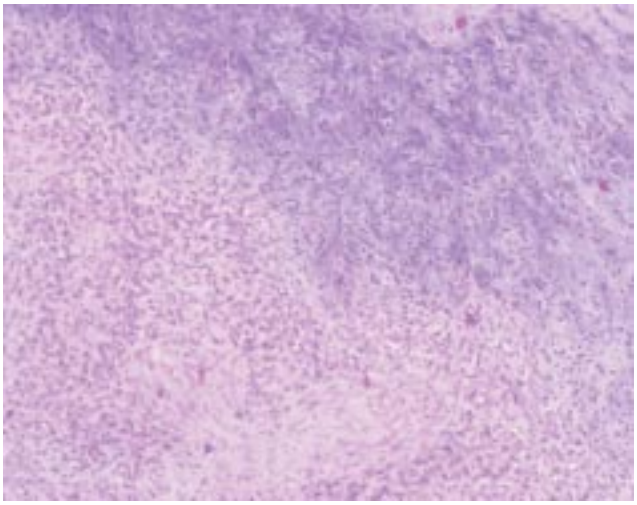

Fig. 23: Cellular areas alternate with myxoid ones. H&E;40X

the chin of a 54 year old male. When examining the biopsy of the tumor stained with H&E, osteoid and bone are visible as well as osteoblasts and fibroblasts. In addition to this typical presentation of an osteosarcoma, the chondroblast type will have atypical chondrocytes with disorganized cellular density, areas of calcification, and myxomatous material. Osteosarcoma of the chondroblastic type is associated with the use of therapeutic radiation. This has become evident due to the presence of chronic radiodermatitis overlying most extraskeletal osteosarcomas. The differential diagnosis for extraskeletal osteosarcoma include myositis ossificans, synovial sarcoma, epithelioid sarcoma, malignant fibrous histiocytoma, liposarcoma, parosteal osteogenic sarcoma, high-grade surface osteosarcoma and malignant melanoma. Luckily, many of these differential diagnosis can be ruled out using H&E because the neoplastic elements of osteoid and bone in these tumors is focally located, the architecture tends to be more organized, and the cells tend to be more differentiated, than that seen in osteosarcoma [58]. One of the more difficult differential diagnoses to exclude is malignant fibrous histiocytoma with metaplastic bone. Studies have suggested; however, that malignant fibrous histiocytoma

Table 8: Immunostaining in Extraosseous Osteosarcoma of the Chondroblastic Type and Common Differential Diagnoses

IHC Profile	Extraosseous Osteosarcoma of the Chondroblastic Type (68,81-84)	Myositis Ossificans (85)	Parosteal Osteogenic Sarcoma (86)
Vimentin	+	+	
Osteocalcin	+		+ (if differentiated)
Osteonectin	+		+
Desmin		-	
SMA		-	
S100		+ (focal)	
A-SMA			+/-
MSA			+/-

with metaplastic bone will have osseous and chondroid elements in the fibrous septa and pseudocapsule [59].

As it can be seen in Table 8, the most reliable marker, when suspecting a diagnosis of extraosseous osteosarcoma of the chondroblastic type is osteocalcin. This stain, however, may be problematic when trying to differentiate extraosseous osteosarcoma of the chondroblastic type from parosteal osteogenic sarcoma because if the parosteal osteogenic sarcoma is well differentiated it will also stain positively for osteocalcin. In this case further stains should be ordered such as A-SMA and MSA to illustrate the presence of muscle cells. A positive result received from a vimentin stain could further negate the likelihood of a parosteal osteogenic sarcoma. Staining for S100 will be focally positive in the case of myositis ossificans so if this pattern of staining is not seen that further indicates the diagnosis of extraosseous osteosarcoma of the chondroblastic type.

References:

1. Massone C, Bassot M, Rongiolettit F. Cutaneous presentation of recurrence of lymphoepithelioid T-cell lymphoma (Lennert's lymphoma). Clinical and Experimental Dermatology 2005, 30:155-157.
2. Laskin W, Miettinen M. Epithelioid Sarcoma-New Insights Based on an Extended Immunohistochemical Analysis. Archives of Pathology & Laboratory Medicine 2003, 127: 1161-1168.
3. Weiss SW, Goldblum, JR. Myxoid/Round Cell Liposarcoma. Enzinger and Weiss's Soft Tissue Tumors. 4th edition. Mosby, Inc. 2001: 670-687.
4. Sanders TJ, Venable DD, Sanusi ID. Primary mailignant melanoma of the urethra in a black man: A case report. *J Urol* 1986, 135: 1012-4.
5. Young RH, Srigley JR, Amin MB, Ulbright TM, Cubilla AL. Tumors of the prostate gland, seminal vesicles, male urethra, and penis. Atlas of Tumor Pathology, Fasicile 2, 3rd Series. Washington, DC: Armed Forces Institute of Pathology; 2000: 367-402.
6. Stein BS. Kendall AR. Malignant melanoma of the genitourinary tract. *Ural* 1984, 32: 85.
7. Murphy WM, Grignon DJ, Perlman EJ. Tumors of the Urethra. Atlas of Tumor Pathology, Fasicile 1, 4th Series. Washington, DC: Armed Forces Institute of Pathology; 2004: 367.
8. Sheffield M, Yee H, Dorvault CC, Weilbaecher KN, Eltoum IA, Siegal P, Fisher DE, Chhieng DC. Comparison of Five Antibodies as Markers in the Diagnosis of Melanoma in Cytologic Preparations. American Journal of Clinical Pathology 2002, 118: 930-936.
9. Ascierto P, Mozzillo N, Caracò C, Parasole R, Palmieri G, Melucci MT, Botti G, Castello G. Epithelioid cell-type melanoma as a prognostic factor of poor response to immunological treatment. *Ann Oncol* 2000, 11: 1504.
10. Paul E, Cochran AJ, Wen DR. Immuno-

histochemical demonstration of S-100 protein and melanoma-associated antigens in *Melanocyti nevi*. *J Cutan Pathol* 1988.15(3):161-165.

11. Wilsher M, Cheerala B. WT1 as a complementary marker of malignant melanoma: an immunohistochemical study of whole sections. Histopathology 2007, 51: 605-610.
12. Reed JA, Loganzo F Jr, Shea CR, Walker GJ, Flores JF, Glendening JM, Bogdany JK, Shiel MJ, Haluska FG, Fountain JW. Loss of expression of the p16/ cyclin-dependent kinase inhibitor 2 tumor suppressor gene in melanocytic lesions correlates with invasive stage of tumor progression. *Cancer Res* 1995, 55(13): 2713-2718.
13. Preito VG, Mourad-Zeiden AA, Melnikova V, Johnson MM, Lopez A, Diwan AH, Lazar AJ, Shen SS, Zhang PS, Reed JA, Gershenwald JE, Raz A, Bar-Eli M. Galectin-3 expression is associated with tumor progression and pattern of sun exposure in melanoma. *Clin Cancer Res* 2006; 12(22): 6709-6715.
14. Scala S, Ierano C, Ottaiano A, Franco R, La Mura A, Liguori G, Mascolo M, Staibano S, Ascierto PA, Botti G, De Rosa G, Castello G. CXC chemokine receptor 4 is expressed in uveal malignant melanoma and correlates with the epithelioid-mixed cell type. *Cancer Immunol Immunother* 2007, 56: 1589-1595.
15. Chwirot BW, KuŸbicki £. Cyclooxygenase-2 (COX-2): First immunohistochemical marker distinguishing early cutaneous melanomas from benign melanocytic skin tumours. *Melanoma Res* 2007, 17(3):139-145.
16. Hasimoto Y, Ito Y, Kato T, Motokawa T, Katagiri T, Itoh M. Expression profiles of melanogenesis-related genes and proteins in acquired *Melanocytic nevus*. *J Cutan Pathol* 2006, 33: 207-215.
17. Ding Y, Prieto VG, Zhang PS, Rosenthal S, Smith KJ, Skelton HG, Diwan AH. Nuclear expression of anti apoptotic protein surviving in malignant melanoma. Cancer 2006, 105(5):1123-1129.
18. Cohn ML, Goncharuk VN, Diwan AH, Zhang PS, Shen SS, Prieto VG. Loss of claudin-1 expression in tumor-associated vessels correlates with acquisition of metastatic phenotype in melanocytic neoplasms. *J Cutan Pathol* 2005. 32(8): 533-536.
19. Trejo O, Reed JA, Preito VG. Atypical cells in human cutaneous re-excision scars for melanoma express p75NGFR, C56/N-CAM and GAP-43: Evidence of early Schwann cell differentiation. *J Cutan Pathol* 2002. 29(7): 397-406.
20. Chrony JA, Barr RJ. S100-positive spindle cells in scars: A diagnositic pitfall in the re-excision of desmoplastic melanoma. *Am J Dermatopathol* 2002. 24(4): 309-312.
21. Kanik AB, Yaar M, Bhawan J. p75 nerve growth factor receptor staining helps identify desmoplastic and neurotropic melanoma. *J Cutan Pathol* 1996. 23(3): 205-210.
22. Yan S, Brennick JB. False-positive rate of the immunoperoxidase stains for

MART1/MelanA in lymph nodes. *Am J Surg Pathol* 2004. 28(5): 596-600.

23. Wiltz KL, Qureshi H, Patternson JW, Mayes DC, Wick MR. Immunostaining for MART-1 in the interpretation of problematic intra-epidermal pigmented lesions. *J Cutan Pathol* 2007, 34(8): 601-605.
24. Kim CJ, Pak K, Hamaguchi A, Ishida A, Arai Y, Konishi T, Okada Y, Tomoyoshi T. Primary malignant melanoma of the female urethra. Cancer 1993, 71: 448-51.
25. LeLeux T, Reed J. Pathology of Benign Melanocytic Nevi. Medscape. [Online][Cited:728,2011.] http://emedicine.medscape.com/article/1962932-overview#aw2aab6b9.
26. Cho K, Hashimoto K, Taniguchi Y, Pietruk T, Zarbo RJ, An T. Immunohistochemical study of melanocytic nevus and malignant melanoma with monoclonal antibodies against s-100 subunits. Cancer 1990, 66: 765-771.
27. Ohsie SJ, Sarantopoulos GP, Cochran AJ, Binder SW. Immunohistochemical characteristics of melanoma. Journal of Cutaneous Pathology 2008, 35: 433-444.
28. Garrido-Ruiz M, Requena L, Ortiz P, Pérez-Gómez B, Alonso SR, Peralto JL. The immunohistochemical profile of Spitz nevi and conventional (non-Spitzoid) melanomas: a baseline study. Modern Pathology 2010, 23: 1215-1224.
29. Guler M, Daniels J, Ortiz P, Pérez-Gómez B, Alonso SR, Peralto JL. Expression of Melanoma Antigens in Epithelioid Gastrointestinal Stromal Tumors. Archives of Pathology & Laboratory Medicine 2008, 132:1302-1306.
30. Bridda A, Padoan I, Mencarelli R, Frego M. Peritoneal Mesothelioma: A Review. *Med Gen Med* 2007, 9(2):32. [Online][Cited: 12 14, 2011] http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1994863/
31. Kuman, Abbas, Fausto, Aster. Robbins and Cotran Pathologic Basis of Disease (8th Edition): Saunders Elsevier, 2010. ISBN: 978-1-4377-0792-2.
32. Husain A, Colby T, Ordóñez NG, Krausz T, Borczuk A, Cagle PT, Chirieac LR, Churg A, Galateau-Salle F, Gibbs AR, Gown AM, Hammar SP, Litzky LA, Roggli VL, Travis WD, Wick MR. Guidelines for Pathologic Diagnosis of Malignant Mesothelioma. Archives of Pathology & Laboratory Medicine 2009, 133:1317-1331.
33. Cavazza A, Moroni M, Bacigalupo B, Rossi G, De Marco L, Fedeli F. Malignant epithelioid mesothelioma of the pleura with hyaline globules. Histopathology 2003, 43: 499-505.
34. Yaziji H, Battifora H, Barry TS, Hwang HC, Bacchi CE, McIntosh MW, Kussick SJ, Gown AM. Evaluation of 12 antibodies for distinguishing epithelioid mesothelioma from adenocarcinoma: identification of a three-antibody immunohistochemical panel with maximal sensitivity and specificity. Modern Pathology 2006, 19: 514-523.
35. King JE, Thatcher N, Pickering C, Hasleton P. Sensitivity and specificity of immunohistochemical markers used in the diagnosis of epithelioid mesothelioma: a detailed systematic

analysis using published data. Manchester, UK, Histopathology 2006, 48: 223-232.

36. Ordonez N. Immunohistochemical Diagnosis of Epithelioid Mesothelioma. Archives of Pathology & Laboratory Medicine 2005, 129:1407-1414.
37. Amatya V, Takeshima Y, Kohno H. Caveolin-1 is a novel immunohistochemical marker to differentiate epithelioid mesothelioma from lung adenocarcinoma. Histopathology 2009, 55:10-19.
38. Feller AC, Griesser GH, Mak TW, Lennert K. Lymphoepithelioid lymphoma (Lennert's lymphoma) is a monoclonal proliferation of helper/inducer T cells. Blood 1986, 68: 663-667.
39. Kitamura A, Yamashita Y, Sato Y, Hasegawa Y, Kojima H, Nagasawa T, Mori N. Aggressive Lennert's lymphoma: Report of three cases in comparison to non-aggressive Lennert's lymphoma. Pathology International 2005, 55: 626-631.
40. Yamashita Y, Nakamura S, Kagami Y, Hasegawa Y, Kojima H, Nagasawa T, Mori N. Lennert's lymphoma: a variant of cytotoxic T-cell lymphoma? 2000, American Journal of Surgical Pathology, Vol. 24, pp. 1627-1633.
41. Summers T, Rush W, Aguilera N, Lupton G. Cutaneous involvement in the lymphoepithelioid variant of peripheral T-cell lymphoma, unspecified (Lennert lymphoma). Report of a case and review of the literature. Journal of Cutaneous Pathology 2009, 36: 25-30.
42. Son H, Jang KY, Kim DK, Cho NP, Moon WS. Bizarre Leiomyoma of the soft tissue. Pathology 2005, 37: 318-319.
43. Reddy BVR, Rani B, Anuradha Ch, Chandrasekhar P, Shamala R, Lingamaneni K. Leiomyoma of the mandible in a child 2011, Journal of Oral and Maxillofacial Pathology.15:101-104.
44. Watanabe K, Suzuki T. Uterine leiomyoma versus leiomyosarcoma: a new attempt at differential diagnosis based on their cellular characteristics. Histopathology 2006, 48: 563-568.
45. T, Nishiya M, Ichikawa G, Fujii H Matsumoto. Leiomyoma with atypical cells (atypical leiomyoma) in the. Histopathology 1999, 34:532-536.
46. Amada S, Nakano H, Tsuneyoshi M. Leiomyosarcoma versus bizarre and cellular leiomyomas of the uterus: a comparative study based on the MIB-1 and proliferating cell nuclear antigen indices, p53 expression, DNA flow cytometry, and muscle specific actins. *Int J Gynecol Pathol* 1995, 14:134-142.
47. Siebert JDS, Williams RP, Pulitzer DR. Myxoid liposarcoma with cartilaginous differentiation. *Mod Pathol* 1996. 9:249.
48. Weiss SW, Goldblum, JR. Synovial sarcoma. Enzinger and Weiss's Soft Tissue Tumors. 4th edition. Mosby, Inc. 2001: 1438-1509.
49. Dikhuizen T, Molenaar WM, Hoekstra HJ, Wiersema J, Van den Berg E. Cytogenetic analysis of a case of myxoid liposarcoma with cartilaginous differentiation. *Cancer Genet Cytogenet* 1996, 92:141.
50. Sheibani K, Shin SS, Kexirian J, Weiss

LM. Ber-Ep4 antibody as a discriminant in the differential diagnosis of malignant mesothelioma versus adenocarcinoma. *Am J Surg Pathol* 1991, 15(8):779-84.

51. Weiss SW, Goldblum, JR. Radiation-Induced Extraskeletal Osteosarcoma. Enzinger and Weiss's Soft Tissue Tumors. 4th edition. Mosbe, Inc. 2001:1407-1414.
52. Shmookler BM, Enzinger FM, Brannnon RB. Orofacial synovial sarcoma: A clinicopathologic study of 11 new cases and a review of the literature. Cancer 1982, 50: 269-76.
53. Shmookler BM. Retroperitoneal Synovial Sarcoma. *Am J Clin Pathol* 1982, 77: 686-91.
54. Witkin GB, Miettinen M, Rosai J. A biphasic tumor of the mediastinum with features of synovial sarcoma. *Am J Surg Pathol* 1989. 13:490-9.
55. Prieto VG, Shea CR. Immunohistochemistry of melanocytic proliferations. *Arch Pathol Lab Med* 2011. 135: 853-9.
56. Hein-Hall J, Yohe SL. Application of Immunohistochemistry to Soft Tissue Neoplasms. *Arch Pathol Lab Med* 2008. 138: 476-489.
57. R&DSystems. http://www.rndsystems.com / molecule_detail.aspx?m=3442 [Online][Cited 12 30 2011].
58. Bane BL, Evans HL, Ro JY, Carrasco CH, Grignon DJ, Benjamin RS, Ayala AG. Extraskeletal osteosarcoma: A clinicopathologic review of 26 cases. Cancer 1990. 65:2726.
59. Bosse A, Vollmer E, Bocker W, Roessner A, Wuisman P, Jones D, Fisher LW. The impact of osteonectin for differential diagnosis of bone tumors. *Pathol Res Pract* 1990, 186:651.
60. Hart J, Mandavilli S. Epithelioid Angiosarcoma: A Brief Diagnostic Review and Differential Diagnosis. *Arch Pathol Lab Med* 2011, 135(2): 268-272.
61. Tsukiji H, Takeshima Y, Amatya VJ, Kushitani K, Inai K. Myogenic antigen expression is useful for differentiation between epithelioid mesothelioma and non-neoplastic mesothelial cells. Histopathology 2010, 56: 969-974.
62. Bhushan M, Craven NM, Armstrong GR, Chalmers RJ. Lymphoepithelioid cell lymphoma (Lennert's lymphoma) presenting as atypical granuloma annulare. British Journal of Dermatology 2000, 142: 776-780.
63. Shi Y, Griffin AC, Zhang PJ. Sinus histiocytosis with massive lymphadenopathy (Rosai-Dorfman Disease): A case report and review of 49 cases with fine needle aspiration cytology. Cytojournal 2011, 8: 3
64. Gaitonde. Multifocal, Extranodal Sinus Histiocytosis With Massive Lymphadenopathy. Archives of Pathology and Laboratory Medicine 2007. 131: 1117-1121.
65. Zamecnik M, Michal M. HMB45+ Hyalinized Epithelioid Tumor of the Uterus is Linked to Epithelioid Leiomyoma Rather than to PEC-omas. International Journal of Surgical Pathology 2001, 9:341-343.
66. Roth JA, Enzinger FM, Tannenbaum M.

Synovial sarcoma of the neck: A follow up study of 24 cases. Cancer 1975, 35:1243-53.

67. Cursiefen C, Schlötzer-Schrehardt U, Breiteneder-Geleff S, Holbac LM. Orbital lymphangioma with positive immunohistochemistry of lymphatic endothelial markers (vascular endothelial growth factor receptor 3 and podoplanin). *Graefe's Arch Clin Exp Ophthalmol* 2001, 239: 628–63.
68. Borys D. Pediatric Orthopedic Pathology. [Online] [Cited: 12 22, 2011.] http://www.pedorthpath.com/page04.html
69. Kempson RL, Hendrickson MR. An approach to diagnosis of soft tissue tumors. In: Weiss SW, Brooks JSJ, eds. Monographs in Pathology: Soft Tissue Tumors. Baltimore, Md: Williams & Wilkins; 1996:1-3.
70. Lee SB, Kolquist KA, Nichols K, Englert C, Maheswaran S, Ladanyi M, Gerald WL, Haber DA. The EWS-WT1 translocation product induces PDGFA in desmoplastic small round-cell tumour. *Nat Genet* 1997; 17:309-313.
71. Brooks JSJ. Long course: soft tissue pathology immunohistochemistry of soft tissue tumors. United States and Canadian Academy of Pathology Long Course: Soft Tissue Tumors. 3 15 1995. Toronto, Ontario.
72. Miettinen M, Lehto VP, Virtanen I, Asko-Seljavaara S, Pitkanen J, Dahl D. Neuroendocrine carcinoma of the skin (Merkel cell carcinoma): Ultrastructural and immunohistochemical demonstration of neurofilaments. *Ultrastruct Pathol* 1983, 4:219-225.
73. Moll R, Osborn M, Hartschuh W, Moll I, Mahrle G, Weber K. Variability of expression and arrangement of cytokeratin and neurofilaments in cutaneous neuroendocrine carcinomas (Merkel cell tumors): Immunocytochemical and biochemical analysis of twelve cases. *Ultrastruct Pathol* 1986, 10:473-495.
74. Miettinen M. Keratin 20: Immunohistochemical marker for gastrointestinal, urothelial, and Merkel cell carcinomas. *Mod Pathol* 1995, 8:384-388.
75. Barrett AW, Cort EM, Patel P, Berkovitz BK. An immunohistological study of cytokeratin 20 in human and mammalian oral epithelium. *Arch Oral Biol* 2000, 45:879-887.
76. Smirnov AV. Fibrosarcoma: Immunohistochemical study of the extracellular matrix. Arkhiv Patologii 1988, 50(12):17-24.
77. Ram Kumar Y, Suprakash, Phani Kumar K, Diwakar T. Fibrosarcoma of Mandible-A Rare Case With Cervical Lymphnode Metastasis. *Jr of Orofac Scie* 2009,1(2): 25-27.
78. Middleton LP, Duray PH, Merino MJ. The histological spectrum of hemangiopericytoma: Application of immunohistochemical analysis including proliferative markers to facilitate diagnosis and predict prognosis. *Hum Pathol* 1998, 29(6):636-40.
79. Fanburg-Smith JC, Miettinen M. Low-affinity nerve growth factor receptor (p75) in dermatofibrosarcoma protuberans and other nonneural tumors: a study of 1,150

tumors and fetal and adult normal tissues. *Hum Pathol* 2001; 32(9): 976-83.

80. Preito V. Immunohistochemistry in cutaneous adnexal carcinomas vs metastatic adenocarcinomas. [Online] [Cited 12 23 2011] http://www.uscap.org/site~/98th/pdf/companion01h04.pdf
81. Fanburg-Smith JC, Bratthauer GL, Meittinen M. Osteocalcin and osteonectin immunoreactivity in extraskeletal osteosarcoma: A study of 28 cases. *Hum Pathol* 1999, 30:32.
82. Serra M, Morini MC, Scotlandi K, Fisher LW, Zini N, Colombo MP, Campanacci M, Maraldi NM, Olivari S, Baldini N. Evaluation of osteonetin as a diagnostic marker of osteogenic bone tumors. *Hum Pathol* 1992, 23:1326.
83. Takada J, Ishii S, Ohta T, Koshiba H, Matsuyama T, Usui M, Yamawaki S, Mori M. Usefulness of a novel monoclonal antibody against human osteocalcin in immunohistochemical diagnosis. *Virchows Arch A Pathol Anat Histopathol* 1992, 420:507.
84. United States Department of Health Services. National Institute for Health. A snapshot of Sarcoma. [Online][Cited: 12 19, 2011.] http://nci.nih.gov/aboutnci/servingpeople/snapshots/sarcoma.pdf
85. Smith R, Athanasou NA, Vipon SE. Fibrodysplasia (myositis) ossificans progressiva: clinicopathological features and natural history. *Q J Med* 1996, 89: 445-456.
86. Franchi A, Comin CE, Santucci M. Submicroscopic & Immunohistochemical Profile of Surface Osteosarcomas. Ultra- structural Pathology 1999, 23: 233-240.

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