

Strong association of HMB-45 expression with renal angiomyolipoma

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ABSTRACT

Objective: Angiomyolipoma (AML) is a benign neoplasm consisting of varying mixtures of smooth muscle, blood vessels and fat. Although, most of these tumors are easy to recognize, some may pose a diagnostic dilemma due to unusual histologic features. Recently, it was suggested that melanosome-associated protein (HMB-45) immunoreactivity may be used for diagnostic confirmation of several neoplasm. The aim of this study is to analyze the diagnostic efficacy of HMB-45 in patients with AML.

Methods. This study was carried out at the Faculty of Medicine, Department of Pathology, Dicle University, Diyarbakir, Turkey, during the period January 2000 to September 2003. HMB-45 immunoreactivity was analyzed in 6 patients with AML and in 34 patients with other renal and retroperitoneal pathologies, including 10

nephrectomized patients for non-neoplastic reasons by means of immunohistochemistry.

Results: Patients with AML were positive for HMB-45. Whereas, HMB-45 immunoreactivity was negative in all of the histologic specimens from the patients with renal cell carcinoma, retroperitoneal sarcomas, Wilms' tumor, lipoma, leiomyoma, and nephrectomized kidneys of non-neoplastic reason. The association of AML with HMB-45 immunoreactivity was highly significant ($p < 0.001$).

Conclusion: Our findings suggest that HMB-45 may not be a melanocyte-restricted marker, and can be useful in differential diagnosis between AML and other tumors seen in kidney and retroperitoneal region.

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Angiomyolipoma (AML) is an unusual benign tumor that occurs most frequently in kidney, followed by liver, and in other organs.¹ Angiomyolipoma is a hamartomatous disorder composed of varying proportions of adipose tissue, blood vessels and smooth muscle, the relative proportion of which vary widely.² Many cases are associated with tuberous sclerosis; however, the majority of patients do not have this disease complex.^{3,4} Histologically AMLs are heterogeneous, some showing a benign appearance and others exhibiting nuclear hyperchromatism, cell pleomorphism, mitotic activity and presence of bizarre cells with abundant clear or unusual morphology. In the last group, the possibility exists

that these tumors may be misdiagnosed as sarcoma, renal cell carcinoma, leiomyoma or lipoma.^{5,6} Hartwick et al⁷ reviewed the literature and reported that 12% of AMLs had a variety of pseudomalignant features. Immunohistochemically, melanosome-associated protein (HMB-45) (previously reported as melanoma) has been recently shown to stain renal AML in a few study.^{8,9} Some authors drew attention to the fact that the HMB-45 is also present in angiomyolipoma.^{9,10} The staining is mainly in large polygonal cells with eosinophilic or clear cytoplasm, and the staining pattern is typically cytoplasmic and granular.¹¹ Kaiserling et al¹² found HMB-45 reactivity in spindle-shaped smooth muscle cells as well. It has

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also been found in fat cells.¹³ We aimed to evaluate the role of HMB-45 in different diagnosis of AML of kidney and benign and malignant tumors of retroperitoneal region.

Methods. We analyzed HMB-45 immunoreactivity in 6 patients (4 female, 2 male) with AML, 5 patients with renal cell carcinoma (RCC), 5 patients with retroperitoneal sarcoma (RS), 6 patients with Wilm's tumor (WT), 5 patients with lipoma, 3 patients with leiomyoma and 10 patients underwent nephrectomized for non-neoplastic reasons. The patients' age were ranged from 25-45 years for AML, 38-55 years for RCC, 1-5 years for WT, 45-55 years for lipoma, 35-41 years for leiomyoma, and 17-58 years for nephrectomized patients with non-neoplastic pathology. No patients with AML had a tuberous sclerosis. All the specimens of the patients for histopathological examination have been obtained from the archives of Department of Surgical Pathology, Dicle University Hospital, Diyarbakir, Turkey, during the period January 2000 to September 2003. The surgical specimens were fixed in a neutral-buffered 10% formalin. They embedded in paraffin wax using the standard method. Parallel 4 mm sections were cut and placed on silan-coated glass slides. One of the parallel sections was stained with hematoxylin-eosin. Following deparaffinization and hydration, the sections were treated 2 times in a microwave oven for 8 minutes for antigen retrieval. The avidin-biotin peroxidase complex method was used as described previously.¹⁴ For primary antibody, we used monoclonal HMB-45 (1:1000, Dako, CA, USA). The slides were examined under light microscopy by 2 investigators. Tumors in which more than 10% of

the neoplastic cells showed intense, granular and cytoplasmic staining were accepted as positive for HMB-45 immunoreactivity.

Results. Of the 6 AML included in this study, 4 had a varying proportions of mature adipose tissue, blood vessels, and smooth muscle components. One of these tumors was composed predominantly of mature adipose tissue with only a few interspersed hyalinized tortuous vessels around which a few spindly or epithelioid cells were arranged. These tumors could easily be misdiagnosed as lipoma. At the other extreme of the morphological spectrum, another tumor could be confused with leiomyoma as it was composed predominantly of spindle smooth muscle cells with a very little fat and vascular component. All the tumors diagnosed as AML were positive for HMB-45 immunoreactivity. **Figure 1** shows typical microscopic features of AML as an admixture of dysplastic vessels spindle or clear smooth muscle cells with copious lipid contents, the relative proportions of which vary widely. **Figure 2** shows cytoplasmic HMB-45 immunoreactivity haphazardly scattered within the tumor mass in the epithelioid, spindle and fat cells except blood vessels. The relationship between HMB-45 immunoreactivity and the presence of AML were highly significant ($p < 0.001$) (**Table 1**). Whereas, the entire specimen from the patients with RS, WT, lipoma, leiomyoma, and normal kidney were negative for HMB-45 immunoreactivity. The intensity of HMB-45 staining varied from moderate to strong. Only one patient with RCC and one patient with liposarcoma showed light cytoplasmic HMB-45 expression in approximately 10% of the cells.

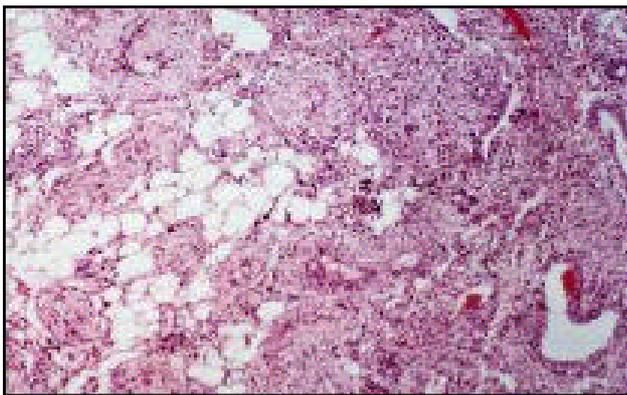


Figure 1 - Typical appearance of renal angiomyolipoma, showing an admixture of adipose tissue, blood vessels and smooth muscle (hematoxylin-eosin, original magnification x 100).

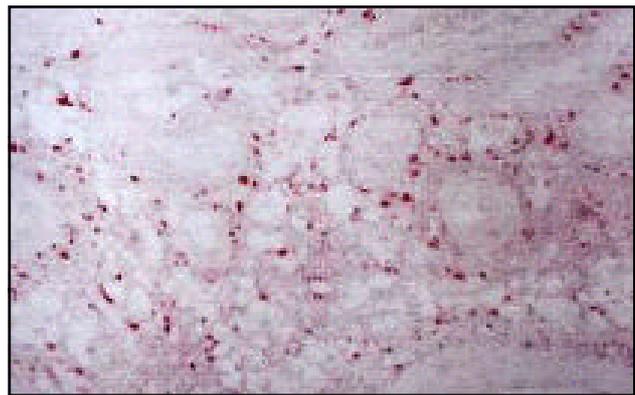


Figure 2 - HMB-45 positivity cells are seen scattered within the tumor mass (immunoperoxidase stain, original magnification x 100).

DISCUSSION. Angiomyolipoma is a rare tumor that is believed to be of hamartomatous nature; however, the precise histogenesis remains controversial.^{4,9} They can be solitary or multifocal, with the kidney being the favored location. Forty percent to 80% of patients with tuberous sclerosis have renal AML that often may be multiple and bilateral, but approximately 80% of renal AML occur in patients without symptoms of the disease.⁴ Histopathologically, the tumor is composed of mature adipose tissue, smooth muscle and blood vessels. The adipose component is formed from uniform fat cells haphazardly arranged in interlacing bundles and sheets. The smooth muscle is consisted of spindle and epithelioid cells. And the vascular component is of long thick walled tortuous blood vessels.² Histogenetically, the tumor is believed to arise from a pluripotent cell that differentiates into adipocytes and smooth muscle. The spindle cell renal AML has been confirmed by ultrastructural studies to be smooth muscle in origin, with some cells showing characteristics of both smooth muscle and adipocytes.¹⁵ Various ultrastructural studies have proposed that the precursor cells of this tumor are derived from a cell residing in the perivascular space.^{16,17} Some angiomyolipomas may demonstrate an unusual morphological appearance. Sometimes it may be quite difficult to differentiate from leiomyomas as in one of our cases. Some others may mimic lipomas. The presence of numerous multinucleated giant cells with foamy cytoplasm may lead to an erroneous diagnosis of liposarcoma or malignant fibrous histiocytoma. In addition, when epithelioid smooth muscle cells with clear cytoplasm arranged in a solid or alveolar pattern are encountered, the possibility of renal cell carcinoma must be excluded. HMB-45 has recently been shown to be a promising marker for renal and hepatic AMLs^{9,18,24} and can be applied to minute samples such as fine-needle aspirates.¹¹ This has led to many case reports and the recognition of some AMLs with unusual morphology which otherwise have been misdiagnosed.^{5,6}

HMB-45 is a monoclonal antibody that was believed to be specific for melanoma and related tumors as melanoma, melanocytic neuroectodermal tumor,^{14,20} clear cell sarcoma²¹ and olfactory neuroblastoma.²² A granular positivity has been reported in normal sweat glands, breast, bronchial epithelium, and plasma cells, as well as in rare cases plasmacytoma and breast carcinoma.²³ Due to its wide range of reactivity, HMB-45 can no longer be considered a melanocyte-specific marker. HMB-45 immunoreactivity were positive in all patients with AML, whereas it was negative in all stained specimens of the patients with RS, WT, lipoma, leiomyoma and underwent nephrectomy for non-neoplastic reasons. The light cytoplasmic expression of HMB-45 in one patient with RCC and in another patient with liposarcoma was quite interesting, and may be due to some technical reasons. Since benign and malignant tumors in kidney and retroperitoneal region were almost always negative for HMB-45, the nature of AML is quite different from those seen in other tumors. The immunoreactivity of AML cells with a melanocyte marker HMB-45 is a consistent finding and thus can facilitate differentiation of this tumor from other lesions. In this study, we provide evidence that immunohistochemical analysis with HMB-45 is useful in differentiating between AML and other neoplasm of the kidney and retroperitoneum. All cases of AMLs tested showed the presence of HMB-45 positive cells while other tumors lacked them.

In conclusion, our study suggests that HMB-45 immunoreactivity is more extensive in AML than we thought it previously, and AML may show melanocytic differentiations or have melanosome such as granules. HMB-45 can be useful in different diagnosis between AML (particularly in those cases of AML exhibiting pleomorphism, cellular atypia or unusual morphology) and other tumors that can be seen in kidney and in retroperitoneal region.

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Table 1 - The association of HMB-45 immunoreactivity with angiomyolipoma (AML).

Patients	HMB-45 positive	HMB-45 negative
AML positive	6	0
AML negative	2	32

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