CD4+/CD56+ Hematodermic Neoplasm ("Blastic Natural Killer Cell Lymphoma")

Neoplastic Cells Express the Immature Dendritic Cell Marker BDCA-2 and Produce Interferon

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Abstract

CD4+/CD56+ hematodermic neoplasm ("blastic natural killer [NK]-cell lymphoma") is a rare highly aggressive neoplasm associated with cutaneous manifestations followed by dissemination to blood, bone marrow, and other tissues. Neoplastic cells exhibit a lineage-negative CD4+/CD56+/CD43+/HLA-DR+ immunophenotype, initially suggesting an NK-cell derivation. The recent discovery of CD123 antigen expression by tumor cells has provided evidence for a relationship to immature dendritic cells (DCs), a group of myeloid and lymphoid early-committed progenitors capable of differentiating into antigen-presenting DCs. Based on flow cytometric analysis, myeloid DCs represent the majority of human peripheral blood DCs and are positive for blood dendritic cell antigen (BDCA)-1 (or CD1c), CD13, CD11c(high), CD33, and CD123(low). Plasmacytoid DCs are BDCA-2+/CD123(high)+/CD13–/CD33– and produce interferon (IFN)-α when triggered by antigens. IFN-α production may be detected in tissue sections using antibodies for myxovirus A (MxA) protein, a surrogate marker. This report describes the clinical, histologic, immunophenotypic, cytogenetic, and molecular genetic findings for 3 cases of CD4+/CD56+ hematodermic neoplasms. In all cases, neoplastic cells were reactive for CD123, BDCA-2, and MxA protein, providing strong evidence for an immature plasmacytoid DC derivation for this rare neoplasm.

"Blastic natural killer (NK)-cell lymphoma" is a rare CD4+/CD56+ neoplasm with aggressive clinical behavior and poor prognosis.[1] Patients typically have skin lesions followed by systemic dissemination, including involvement of peripheral blood, bone marrow, lymph nodes, and other tissues. In view of the primary cutaneous involvement and the absence of any T, B, or myelomonocytic markers, the term lineage-negative CD4+/CD56+ hematodermic neoplasm (HDN) has been used to describe this rare entity.

Although initially thought to be of NK lineage,[2,3] recent recognition of CD123 antigen expression by the tumor cells linked its origin to immature dendritic cells (DCs).[4] Additional studies supporting this derivation have been published during the last several years,[5-8] including the most recent demonstration of immunoreactivity of lesional cells for blood dendritic cell antigen (BDCA)-2 and myxovirus A (MxA), an interferon (IFN)-α–inducible protein.[9] Thus, the tumor cells of HDN are not only immunophenotypically consistent with immature DCs but also maintain some of their functional characteristics.

Immature DCs include plasmacytoid DCs (pDCs), or so-called lymphoid DCs, and myeloid DCs and represent early-committed progenitor cells capable of differentiating into antigen-presenting DCs central to the initiation of primary T cell–based immune responses.[10,11] Antibodies are available that permit detection of these subsets of immature DCs.[12-14] Based on flow cytometric immunophenotypic analysis, myeloid DCs, which represent the majority of human peripheral blood DCs, are positive for BDCA-1 (CD1c), CD11c(high), CD123(low), CD13, and CD33, whereas pDCs are positive for BDCA-2 and CD123(high) and negative for CD13 and CD33. Another characteristic of pDCs is production of IFN-α when triggered by antigens, a feature that can

be assessed in tissue sections by identification of MxA protein, a surrogate marker for IFN-α. The pDCs represent the major source of this cytokine.[15-17]

This report describes the immunohistochemical expression of BDCA-2, MxA protein, and CD123; a broad immunophenotypic profile; cytogenetic and molecular genetic studies; and the morphologic and clinical features in 3 cases of CD4+/CD56+ HDNs and provides further evidence for an immature pDC derivation for these neoplasms. We confirm recently published results and also present expanded immunophenotypic data. Increased awareness of this newly established entity promotes more rapid diagnosis and treatment of this lethal neoplasm.

Materials and Methods

Skin biopsy specimens from 3 patients with cutaneous lesions consistent with CD4+/CD56+ lineage-negative HDN (blastic NK-cell lymphoma) were retrieved from the files of Brigham and Women's Hospital and Children's Hospital (Boston, MA) based on the availability of frozen tumor tissue. Other specimens, including bone marrow biopsies and aspirates and cerebrospinal fluid (CSF), were also reviewed as available.

Immunohistochemical studies were performed manually on cryostat sections and on paraffin-embedded material as previously described.[18] Antibodies used are listed in **Table 1**. A horseradish-peroxidase–labeled polymer detection system (EnVision+, DAKO, Carpinteria, CA, or PowerVision,

Table 1
Antibodies Used, Specificity, and Sources

Antibody to	Major Immunoreactivity Profile	Clone	Source*
CD2	Pan T cell; NK cells	NA	Lee Nadler, MD
CD3 (surface)	Pan T cell	T3-4B5	DAKO
CD4	Helper/inducer T cells; thymocytes; monocytes	MT310	DAKO
CD5	Pan T cell; B-cell subset	DK23	DAKO
CD7	Pan T cell	DK24	DAKO
CD8	Cytotoxic T cell	DK25	DAKO
TCRα/β	TCR α/β constant region; T-cell subset	BMA031	Endogen
TCRγ/δ	TCRδ constant region; T-cell subset	5A6.E9	Endogen
CD13	Myeloid cells	WM15	BD Biosciences
CD33	Myeloid cells	P67.6	BD Biosciences
CD20	B cell	L26	DAKO
CD19	B cell	4G7	Becton Dickinson
CD56	NK cell; T-cell subset; neural marker	123C3.D5	Cell Marque
CD57	NK cell; T-cell subset; neuroendocrine cells	HNK-1	Becton Dickinson
CD16	NK cell	NA	BD Biosciences
CD123	Interleukin-3 receptor; plasmacytoid dendritic cells	7G3	BD Biosciences
BDCA-1	Myeloid dendritic cells	AD5-8E7	Miltenyi Biotec
BDCA-2	Plasmacytoid dendritic cells	AC144	Miltenyi Biotec
MxA	IFN-α surrogate marker	M143	Otto Haller, MD
TdT	T- and B-cell lymphoblasts; thymocytes	HT-1/3/4; SEN28	DAKO (cryostat); Novocastra (paraffin)
HLA-DR	HLA class II molecules	NA	Lee Nadler, MD
Ki-67	Proliferation antigen	MIB-1	DAKO
bcl-2	Antiapoptotic protein	124	DAKO
Fascin	Mature dendritic cells; Reed-Sternberg cells	55K2	Fumio Matsumura, PhD, and Shigeko Yamashiro, PhD
CD117	Mast cells; progenitor cells; myeloblasts	Rabbit polyclonal	DAKO
CD10	Follicular center cell marker; lymphoblasts	56C6	Novocastra
CD34	Stem cells; blasts; endothelial cells	QBEnd10	Beckman Coulter
CD43	T cells; B-cell subset; NK cells; myeloid cells; monocytes; histiocytes	L60	Becton Dickinson
CD68	Macrophages	KP1	DAKO
CD30	Activation antigen	Ber-H2	DAKO
CD25	Interleukin-2 receptor	ACT-1	DAKO
Myeloperoxidase	Myeloid cells	Rabbit polyclonal	DAKO
Lysozyme	Monocyte/macrophage; myeloid cells	Rabbit polyclonal	DAKO
Granzyme B	NK cells; activated cytotoxic T cells	GrB-7	Biodesign International
TIA-1	NK- and T-cell subset	TIA-1	Coulter
Perforin	NK and cytotoxic T cells	P1-8, Rat monoclonal	DAKO
S-100	Dendritic cells; melanocytes	Rabbit polyclonal	DAKO
CD1a	Dendritic cells; Langerhans cells; cortical thymocytes	O10	Beckman Coulter
Langerin	Langerhans cells	12D6	Novocastra
CD99	Thymocytes; Ewing sarcoma; neuroectodermal tumors	013	Signet Labs

BDCA, blood dendritic cell antigen; MxA, myxovirus A; NA, not available; NK, natural killer; TCR, T-cell receptor; TIA-1, T-cell–restricted intracellular antigen 1; TdT, terminal deoxynucleotidyl transferase.

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ImmunoVision Technologies, Daly City, CA) was used for all studies except fascin, for which an alkaline-phosphatase detection system (ImmunoVision) was used. For immunoperoxidase studies, antibody localization was effected by using a peroxidase reaction with diaminobenzidine (DAB+; DAKO) as the chromogen. Incubation with primary antibody was performed for 1 hour except for BDCA-1, BDCA-2, and MxA protein antibodies, which were incubated overnight. Appropriate positive and negative control experiments were run simultaneously. Chloroacetate esterase studies were performed using standard protocols.

In situ hybridization studies for Epstein-Barr virus (EBV)-encoded small RNAs were performed using commercial nucleic acid probe and negative control probe (Novocastra, Newcastle upon Tyne, England) as previously described.[19] Flow cytometric analysis of bone marrow samples (cases 1 and 2) was performed on a bench-top flow cytometer (FACSCalibur, BD Biosciences, San Jose, CA) and analyzed by using CellQuest software (BD Biosciences). Standard panels of antibodies directed against lymphoid and myeloid antigens obtained from BD Biosciences were used. Conventional cytogenetic studies with G-banding techniques were performed on bone marrow specimens of cases 1 and 2.

Polymerase chain reaction (PCR) was performed on DNA isolated from frozen material in all cases by using fluorescent primers to T-cell receptor (TCR)γ-chain gene segments 1 through 8, 9, 10, and 11 (Vγ1-8, Vγ9, Vγ10, and Vγ11, respectively) and joining regions (Jγ1 and Jγ2) (InVivoScribe Technologies, BIOMED, San Diego, CA). The PCR products were analyzed by agarose (case 1) and capillary (cases 2 and 3) gel electrophoresis.

Results

Clinical Findings

Clinical features for 3 patients with CD4+/CD56+ HDN were as follows:

Case 1

A 48-year-old man had cutaneous lesions in the sacral area with no other apparent sites of involvement. His medical history was not significant and family history included breast and prostate cancer.

Biopsy revealed a CD4+/CD56+ HDN. He was treated with 4 cycles of CHOP (cyclophosphamide, doxorubicin [Adriamycin], vincristine, and prednisone). The lesions cleared but recurred 1 month after completion of treatment. Skin biopsy revealed recurrent disease. The patient was treated with 2 additional cycles of CHOP and total body irradiation with improvement; however, the lesions persisted.

At this point, his peripheral blood and chemistry values were normal, and no evidence of systemic involvement was apparent by physical examination and computed tomography (CT). Six months later, he was admitted for evaluation of chest pain and cervical lymphadenopathy. A bone marrow biopsy showed extensive tumor involvement. Peripheral blood and CSF samples also contained neoplastic cells. The patient received further therapy with regression of disease and 3 months later, underwent matched, related allogeneic bone marrow transplantation.

Approximately 3 months thereafter, he had a cutaneous relapse and was treated by donor lymphocyte infusions and discontinuation of immunosuppression. His laboratory values were normal except for mild elevation of liver function test results attributed to graft-vs-host disease. Restaging CT was negative for visceral disease. He appeared to be in partial clinical remission but rapidly progressive pulmonary symptoms developed, and the patient died of respiratory failure 2.5 years after initial examination. At autopsy, pulmonary fibrosis and diffuse alveolar hemorrhage were present; however, tumor involvement was restricted to the areas of cutaneous relapse.

Case 2

A 16-year-old girl had a solitary, well-circumscribed, red skin papule of 8 months' duration involving the right inner thigh. Her medical history was not contributory; however, family history was significant for a paternal cousin with T-cell acute lymphocytic leukemia at age 5 years.

The cutaneous lesion enlarged progressively to 5 cm in diameter and 2 cm in thickness. Approximately 4 months later, similar lesions appeared on the legs, abdomen, and back. A skin biopsy revealed a CD4+/CD56+ lineage-negative neoplasm. CT staging revealed bilateral axillary lymphadenopathy and multiple soft tissue nodules corresponding to cutaneous and subcutaneous lesions. Bone marrow biopsy demonstrated extensive tumor involvement. The CSF examination results were negative. The results of hematology and chemistry laboratory studies were within normal limits.

The patient was treated with 1 cycle of CHOP and intrathecal chemotherapy resulting in regression of the lesions. Subsequently, however, neutropenia and *Candida* septicemia developed, and the patient died with symptoms of multiorgan failure, about 11 months from the onset of disease and 2 months from the initiation of treatment.

Case 3

A 77-year-old woman had pruritic skin nodules progressively involving multiple sites during a period of about 9 months. Her medical history was significant for diabetes, coronary artery disease, myocardial infarction, and an IgG monoclonal gammopathy of unknown significance. Her family history was noncontributory. Results of the physical examination and laboratory workup were normal. CT scans did not reveal lymphadenopathy, organomegaly, or extracutaneous disease. On admission she had

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dusky, symmetrical nodules and tumors ranging up to 5 cm in diameter with no epidermal changes. Biopsies from left upper arm, left flank, and right upper arm revealed similar changes and were compatible with a CD4+/CD56+ lineage-negative neoplasm. At last follow-up, she was undergoing CHOP chemotherapy and was alive, 11 months from the onset of disease and 2 months from the initiation of therapy.

Histologic and Immunophenotypic Features

Morphologic features in all 3 cases were similar and were characterized by a prominent, diffuse, dermal infiltrate of uniform-appearing mononuclear cells showing epidermal sparing and formation of vague periadnexal and perivascular nodules **Image 1**. The cells were small to intermediate size with round to folded nuclear contours, fine chromatin, distinct nucleoli, and a small to moderate amount of clear to eosinophilic agranular cytoplasm **Image 2**. Mitotic figures were frequent.

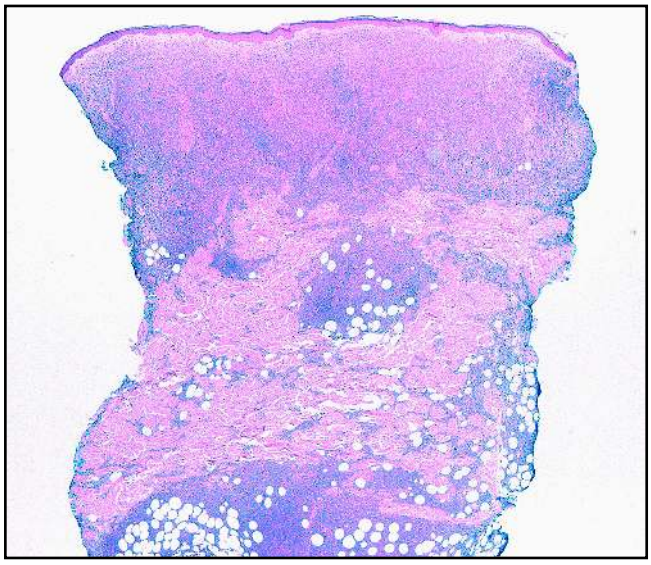

Image 1 (Case 3) Skin biopsy reveals a diffuse, dense, atypical dermal lymphoid infiltrate with epidermal sparing (H&E, ×20).

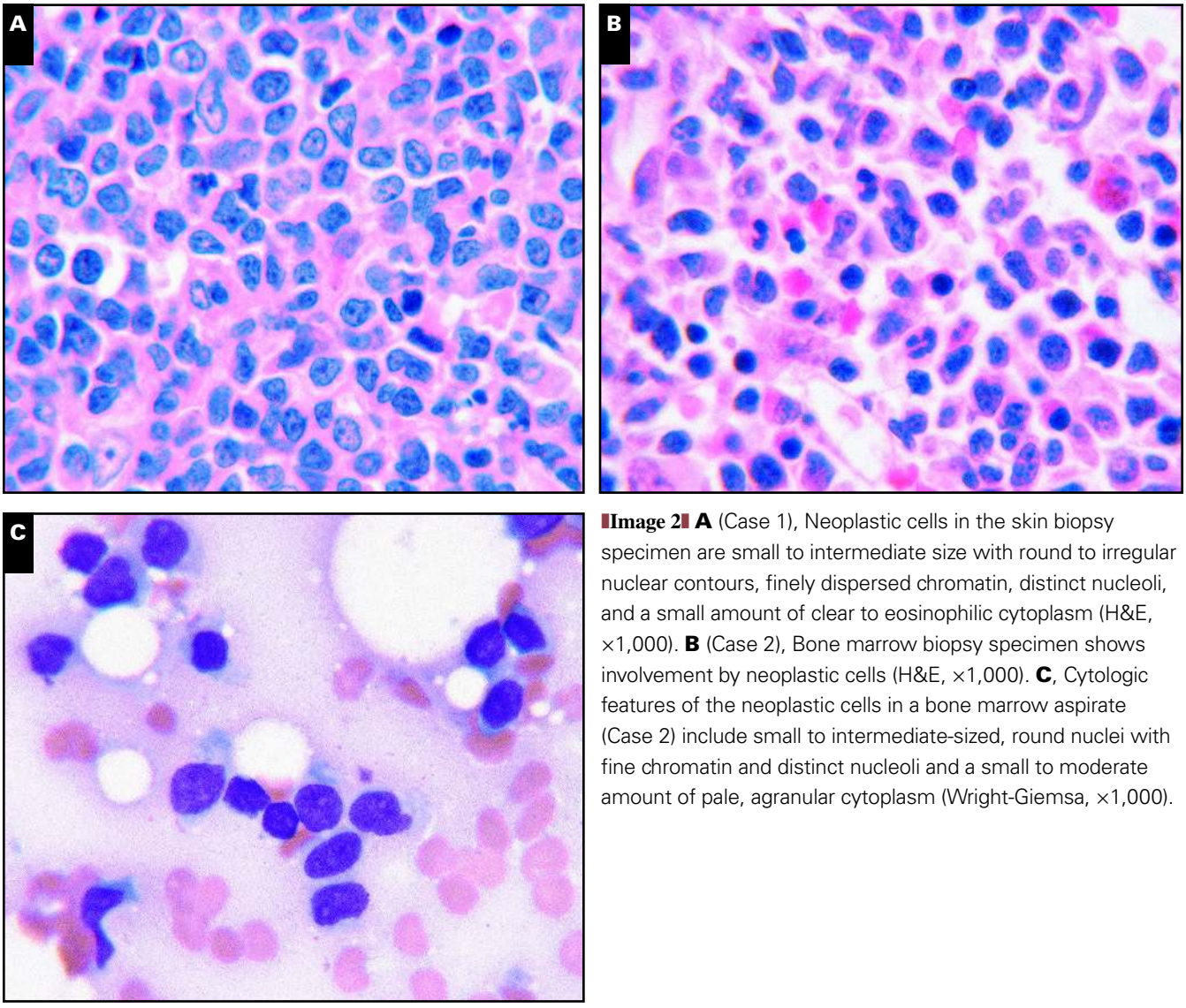

Image 2 **A** (Case 1), Neoplastic cells in the skin biopsy specimen are small to intermediate size with round to irregular nuclear contours, finely dispersed chromatin, distinct nucleoli, and a small amount of clear to eosinophilic cytoplasm (H&E, ×1,000). **B** (Case 2), Bone marrow biopsy specimen shows involvement by neoplastic cells (H&E, ×1,000). **C**, Cytologic features of the neoplastic cells in a bone marrow aspirate (Case 2) include small to intermediate-sized, round nuclei with fine chromatin and distinct nucleoli and a small to moderate amount of pale, agranular cytoplasm (Wright-Giemsa, ×1,000).

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Immunophenotypic characteristics of the lesional cells are summarized in **Table 2** and illustrated in **Image 3**. Immunophenotypically, the neoplastic cells were consistently and uniformly positive for CD4 (Image 3A), CD56 (Image 3B), CD123 (Image 3C), HLA-DR, and BDCA-2 (Image 3D). MxA protein (Image 3E) was expressed strongly in cases 1 and 2 and variably in case 3. Neoplastic cells were negative for fascin (Image 3F), a mature DC marker. Ki-67 (Image 3G) showed nuclear reactivity for about 80% of cells (2 cases) or 30% of cells (1 case). Terminal deoxynucleotidyl transferase (TdT; Image 3H) was expressed for a subset of cells in cases 1 and 2; however, case 3 showed weak and focal nuclear immunoreactivity demonstrable only in paraffin-embedded material (cryostat sections, negative). CD68 (KP1 clone) and CD43 positivity was observed in the majority of the cells. Neoplastic cells were negative for antigens associated with B cells (CD20 and CD19), T cells (CD3, CD2 [1 case positive], CD5, CD7, CD8, TCRα/β, TCRγ/δ), myeloid lineage (CD13, CD33, and CD117), NK cells (CD57, CD16, T-cell–restricted intracellular antigen 1, granzyme B, and perforin), and Langerhans cell phenotype (CD1a, langerin, and S-100). The results of myeloperoxidase (enzymatically and immunophenotypically in cryostat sections) and chloroacetate esterase studies were negative. The results of in situ hybridization studies for EBV-associated mRNA were negative. Results of flow cytometric studies, available in 2 cases, are included in Table 2.

Molecular and Cytogenetic Studies

TCR gene rearrangement studies were performed by PCR analysis on frozen tissue in all 3 cases and were in a germline configuration.

Cytogenetic studies were available in 2 cases. Case 1 showed a normal chromosome complement except for deletion of chromosome 11, which was interpreted as a random loss. Case 2 showed complex chromosomal abnormalities: 46-47,XX, del(3)(p21), del(11)(q2?3), ?ins(12;?)(q13;?), –15, +21, and +?i(21)(q10)[cp3].

Discussion

In the World Health Organization classification scheme, CD4+/CD56+ HDN corresponds to the lesion designated as blastic NK-cell lymphoma.[1] These neoplasms are associated with typical morphologic and immunophenotypic characteristics and aggressive clinical behavior. All cases reported in the literature and the 3 cases included in our study had cutaneous lesions at initial examination, which disseminated within a few months to involve large areas of the cutis, as well as

Table 2
Immunophenotypic Profile of Neoplastic Cells in Three Cases of CD4+/CD56+ Hematodermic Neoplasms*

	Case No.				Case No.		
Marker	**1**	**2**	**3**	**Marker**	**1**	**2**	**3**
CD2	–†	–†	+	CD20	–#	–#	–¶
CD3	–‡	–‡	–‡	CD10	–¶	–¶	–¶
CD4	+†	+‡	+§	CD16	ND	–	ND
CD5	–‡	–¶	–¶	CD25	–#	ND	–¶
CD7	–‖	–#	–¶	CD30	–¶	–¶	–¶
CD8	–‡	–‡	–§	CD34	–‖	–#	–‖
TCRα/β	–	–	–	CD57	–¶	ND	ND
TCRγ/δ	–	–	–	CD99	ND	–¶	ND
CD123	+	+§	+	CD117	ND	–‖	ND
BDCA-1	–	–	–	Fascin	–	–	–
BDCA-2	+	+	+	CD1a	–¶	–¶	ND
MxA	+	+	+	bcl-2	–¶	ND	–¶
CD56	+†	+‡	+§	Granzyme B	–¶	–¶	–¶
HLA-DR	+†	+†	+	Langerin	ND	–¶	ND
CD43	+§	+§	+¶	Lysozyme	–¶	–¶	–¶
CD68	+	+§	+	Myeloperoxidase	–§	–§	–§
TdT	+	+ (–‖)	– (+¶)	Perforin	ND	–¶	ND
Ki-67 (%)	80	80	30	S-100	–¶	–¶	ND
CD13	–†	–†	–	TIA-1	–¶	–¶	–¶
CD33	–†	–†	–	CAE	ND	–§	ND
CD19	–‖	–‖	–	EBV-encoded small RNAs	–¶	–¶	–¶

BDCA, blood dendritic cell antigen; CAE, chloroacetate esterase; EBV, Epstein-Barr virus; MxA, myxovirus A; ND, not done; TCR, T-cell receptor; TIA-1, T-cell–restricted intracellular antigen 1; TdT, terminal deoxynucleotidyl transferase; +, positive; –, negative.
* Studies were performed on cryostat sections only unless otherwise indicated.
† Cryostat sections and flow cytometry.
‡ Cryostat and paraffin sections and flow cytometry.
§ Cryostat and paraffin sections.
‖ Flow cytometry only.
¶ Paraffin section only.
# Paraffin section and flow cytometry.

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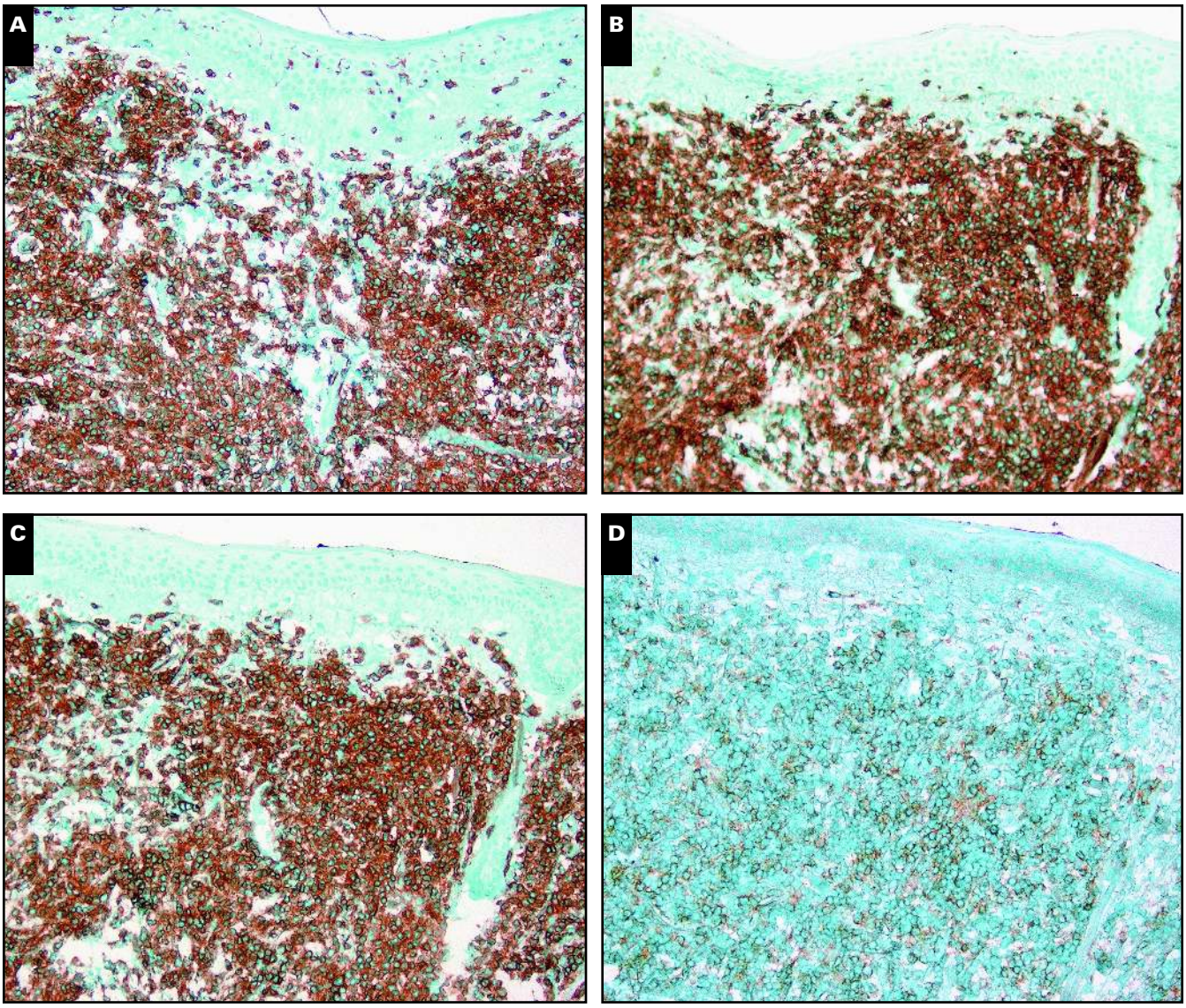

Image 3 (Case 2) Cryostat section studies reveal strong reactivity of the neoplastic cells for CD4 (**A**), CD56 (**B**), CD123 (**C**), and the plasmacytoid dendritic cell marker BDCA-2 (blood dendritic cell antigen-2; **D**). The neoplastic cells also are reactive for myxovirus A protein (**E**), a surrogate marker for interferon-α production, and are negative for fascin (**F**), a mature dendritic cell

blood, bone marrow, and lymph nodes. Progressive disease typically leads to the patient's death within a few years after initial examination.[2,3,5-9,20]

In recent years, accumulating evidence has supported a link between pDCs and the origin of CD4+/CD56+ HDN.[5-9] DCs and precursors are bone marrow stem cell–derived immature antigen-presenting cells constituting approximately 2% of peripheral blood mononuclear cells. They migrate to peripheral sites, including lymph nodes and skin, where they mature on stimulation by cytokines. These immature DCs consist of 2 distinct subsets with separate differentiation pathways: the myeloid DCs resembling myeloid-derived cells and the lymphoid (plasmacytoid) DCs resembling tissue "plasmacytoid monocytes/T cells."[10,21,22]

The pDCs have been recognized in normal tissues, including blood, bone marrow, lymph nodes, tonsils, and thymus, and in various pathologic conditions.[14,23-25] In vitro studies by Grouard et al[26] performed on plasmacytoid T cells isolated from human tonsils demonstrated that these cells correspond to the CD4+/CD11c–/lineage-negative blood DC precursors capable of differentiation into mature DCs on coculture with interleukin (IL)-3 and CD40 ligand. In the same year, Olweus et al[21] demonstrated that antibodies to IL-3 receptor α (CD123) react with DC precursors. Further tissue studies by Cella et al[15] and Facchetti et al[23,27] demonstrated expression of IL-3 receptor α (CD123) and type I IFN by plasmacytoid monocytes. The possible ontogeny of pDCs as DC precursors seems to be established, along with their immunophenotype.[10,22]

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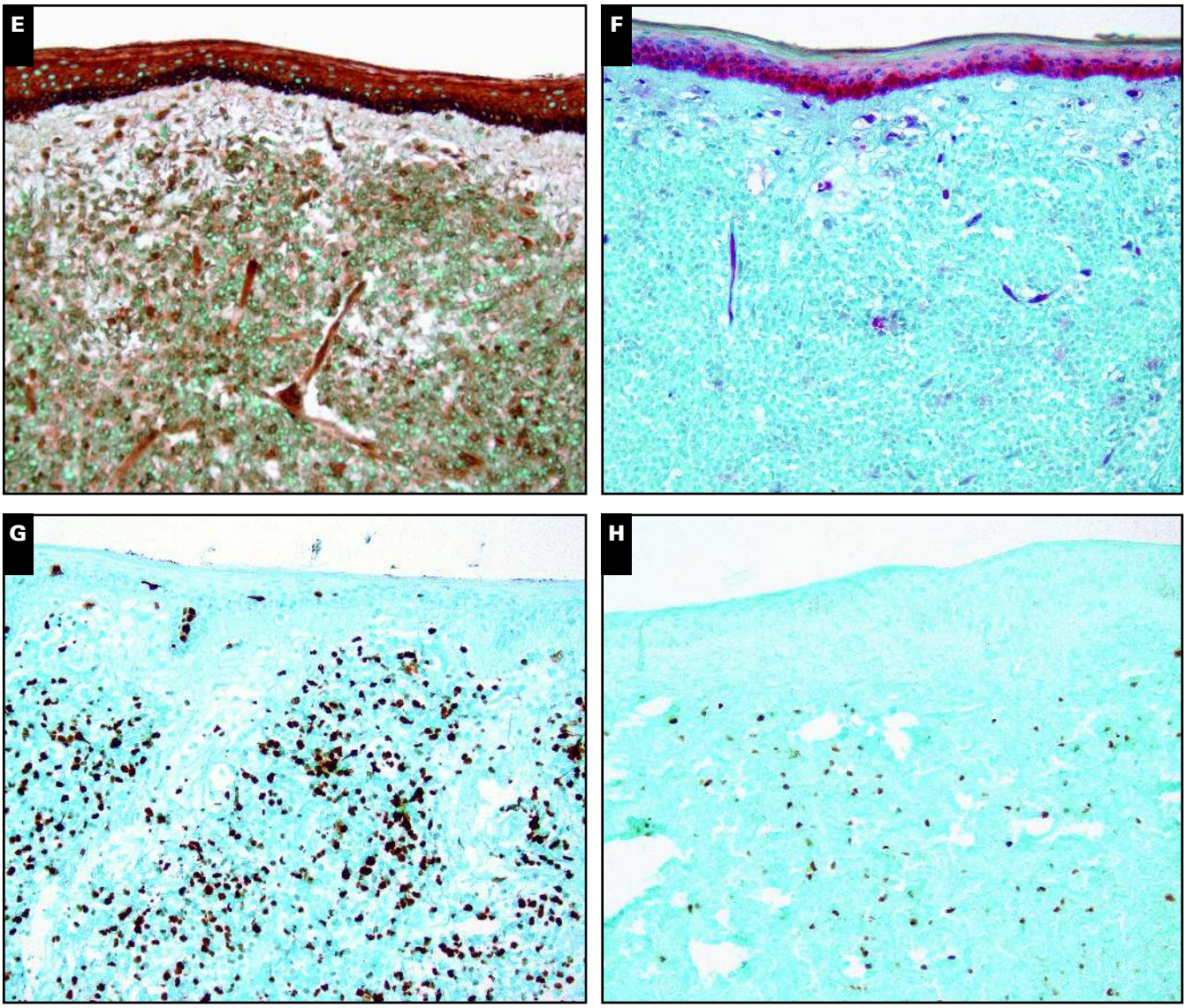

marker. Ki-67 proliferation antigen is seen in about 50% of nuclei (**G**). Nuclear reactivity for terminal deoxynucleotidyl transferase is seen in a subpopulation of the neoplastic cells (**H**) (immunoperoxidase studies, **A**-**E** and **G**-**H**, ×200; **F**, immuno–alkaline phosphatase study, ×200).

In 2001, Chaperot et al[4] made the association between CD4+/CD56+/lineage-negative neoplasms and pDCs based on a study of 7 patients with a CD4+/CD56+/lineage-negative leukemia associated with cutaneous, nodal, and other visceral involvement. In that study, the investigators demonstrated the capacity of the leukemic cells for maturation to DCs on IL-3 and CD40-ligand stimulation along with other functional characteristics, like IFN-α production and stimulation of naive T-cell response, notably T_H2 polarization induction.

Several subsequent studies, as well as our study, provide further evidence for a pDC derivation for HDN based on the immunophenotype.[3,5-9] The pDCs and their neoplastic counterparts are positive for CD45, CD4, CD56, HLA-DR, CD45RA, CD123, CD43, and CD68 and negative for myeloid and monocytic markers (CD13, CD33, CD11c, CD14, myeloperoxidase, chloroacetate esterase, and lysozyme), B and T cell lineage–associated markers (CD19, CD20, CD79a, CD1a, CD3, CD2, CD5, CD7, and CD8), NK-cell–associated antigens (CD16, CD57, T-cell–restricted intracellular antigen 1, and granzyme B), progenitor cell markers (CD34 and c-kit), and lymphoid activation markers (CD25, CD30, and CD71). Other markers not observed for HDN include CD21, CD35, CD36, CD10, S-100, and EBV.[3,5-9] Occasionally, the presence of CD2, CD5, CD7,[3,19] and TdT[8,28] has been reported. Reactivity for CD2 was noted for 1 of our cases (case 3). We also observed TdT positivity for a subset of neoplastic cells. In 1 case, positivity was apparent only in the paraffin section study, and in another

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case, the cryostat section study was positive whereas flow cytometry was negative (Table 2).

Recently, expression of the lymphoid proto-oncogene product T-cell leukemia 1[29,30] and cutaneous lymphocyte-associated antigen[30] has been demonstrated in the majority of cases. T-cell leukemia 1 is seen mainly in T-cell prolymphocytic leukemia as a consequence of 14q32.1 rearrangements.[26] Expression of cutaneous lymphocyte-associated antigen, an E-selectin binding protein, could possibly be responsible for skin homing.[21] CD56 positivity is a characteristic feature of HDN but is typically not observed for pDCs, although a very minor subset of CD56+ pDCs has been identified.[31]

Because HDN is highly aggressive, markers specific for pDCs are of interest for positive identification and patient prognosis. BDCA-2, a novel type II C-type lectin, was proposed as a specific marker of pDCs in human blood.[21,22,27] In the original article, Dzionek et al[12] reported selective expression of BDCA-2 on pDCs using PCR analysis of complementary DNA from multiple tissue arrays and double staining with CD123 and BDCA-2. The pDCs were observed in tonsils, thymus, and T cell–rich extrafollicular areas of reactive lymph nodes. Interestingly, BDCA-2 reactivity was noted for a proportion of cortical thymocytes, which were consistently CD123–.[13] Jacob et al[7] demonstrated positivity for BDCA-2 in HDN along with another pDC marker, BDCA-4, and proposed a new designation of the HDN as early pDC lymphoma/leukemia. Similarly Hallermann et al[6] demonstrated positive staining for BDCA-2 in 1 case of CD4+/CD56+ HDN, and, more recently, Urosevic et al[9] noted a similar finding in a series of 8 cases. Our findings are concordant with their observations and provide further support for an early-immature pDC derivation for these lesions. The neoplastic pDCs apparently retain the functional capacities of their normal counterparts because, in addition to the expression of the IL-3 receptor (CD123), strong immunoreactivity for IFN-α–inducible protein MxA (Image 3E), a surrogate marker of interferon-α production that is a characteristic of pDCs,[15-17] was observed in our cases and the study by Urosevic et al.[9] In addition, tumor cells in our series were negative for fascin, a mature DC marker[32,33] that had not been previously evaluated. In a recent study, Jaye et al[34] investigated 19 archival cases of HDN for expression of BDCA-2, along with other immunophenotypic features. They demonstrated variable expression of BDCA-2 along with an inverse pattern of TdT and postulated a spectrum of maturation among HDNs reflecting functional plasticity of these pDC-derived tumors.[34] This plasticity was observed and reported previously in cases of posttreatment recurrent HDN showing monocytic or myelomonocytic differentiation.[28]

Although the CD4+/CD56+/CD123+ lineage-negative immunophenotype is highly characteristic of HDNs, there are reported cases of cutaneous myelomonocytic or monocytic leukemia coexpressing these markers, making differentiation among these entities difficult.[3,35] An initial study by Bekkenk et al[36] comparing 63 published cases of CD4+/CD56+ HDN and 10 cases of cutaneous myelomonocytic leukemia revealed similarities in clinical manifestations, histologic features, immunophenotype, and prognosis. Differentiation between these entities, based initially on the presence or absence of myeloid/myelomonocytic lineage-associated markers, was improved with the notion that most CD4+/CD56+ HDNs express pDC markers CD123 and BDCA-2 that are usually not seen in cutaneous acute myeloid leukemia.[5,7,9] Also, cells of monocytic derivation are typically positive for lysozyme and nonspecific esterase with variable expression of myeloperoxidase, features not apparent in HDN. A recent study based on gene expression profiling and array-based comparative genomic hybridization analysis demonstrated convincingly that CD4+/CD56+ HDN and cutaneous myelomonocytic leukemia are distinct disease entities with characteristic genetic alterations.[35]

The relationship of pDCs in chronic myelomonocytic leukemia (CMML) remains unclear. The bone marrow in CMML shows expansion of CD123+ DCs in the form of well-defined nodules. These nodules seem to be characteristic of CMML because they are rarely present in other myeloproliferative disorders.[37] Derivation from neoplastic or normal bone marrow progenitor cells is possible; however, their precise origin remains to be elucidated.

Molecular genetic analysis performed on some reported cases[7] and in all of our cases demonstrated *TCR* genes in a germline configuration. Comparative genomic hybridization studies performed on 1 reported case revealed a gain of chromosomes 7q and 22 and loss of chromosomes 3p21.3 and 13q.[6] In most cases, however, complex structural chromosomal abnormalities were seen[2,8] and were also noted in 1 of our cases. In addition, Petrella et al[2] reported frequent deletions and anomalies of chromosome 5q, deletions of 12p, and abnormalities of chromosome 13.

Recent studies and the findings in our 3 cases provide strong evidence for an immature pDC derivation for CD4+/CD56+ HDN. This neoplasm retains the immunophenotypic and functional characteristics of pDCs. Given the accumulating evidence for a pDC origin for this lesion, perhaps consideration should be given to designating this disorder as an early or immature pDC lymphoma/leukemia as initially suggested by Jacob et al.[7] The invariably lethal course mandates an early diagnosis because only bone marrow allograft recipients have shown prolonged survival.[7] Increased awareness of this unique, highly aggressive neoplasm should allow improved and early recognition and aid in defining more effective therapeutic approaches.

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