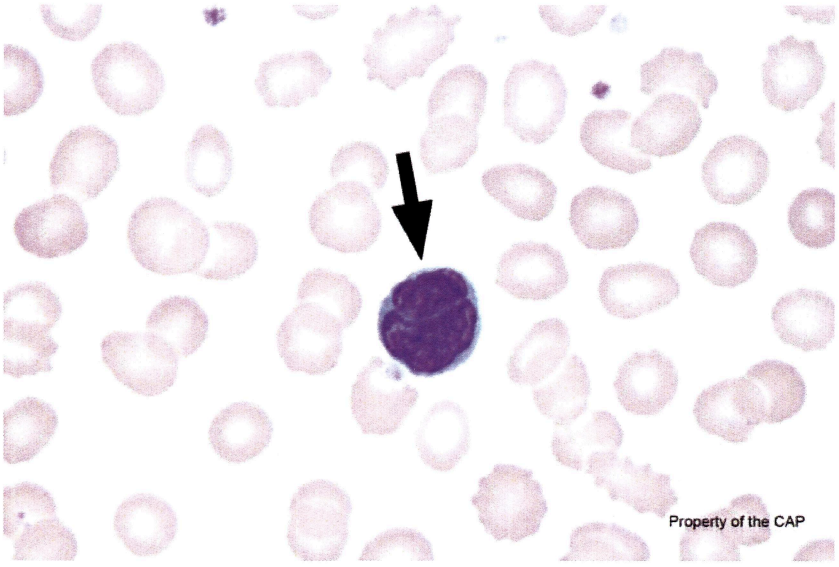
Blood Cell Identification – Ungraded

BCP-30



Identification	Referees No.	Referees %	Participants No.	Participants %	Evaluation
Malignant lymphoid cell (other than blast)	57	62.6	3157	56.2	Educational
Lymphocyte	24	26.4	1639	29.1	Educational
Lymphocyte, reactive (to include plasmacytoid and immunoblastic forms)	7	7.7	443	7.9	Educational
Immature or abnormal cell, would refer for identification	2	2.2	204	3.6	Educational
Monocyte, immature (promonocyte, monoblast)	1	1.1	42	0.8	Educational
Blast cell	-	-	26	0.5	Educational
Lymphocyte, large granular	-	-	23	0.4	Educational
Monocyte	-	-	21	0.4	Educational
Nucleated red blood cell, normal or abnormal morphology	-	-	14	0.3	Educational
Metastatic tumor cell or tumor cell clump	-	-	12	0.2	Educational
Plasma cell (to include morphologically mature, abnormal, containing inclusion, eg, Dutcher body, Russell body, etc)	-	-	10	0.2	Educational
Megakaryocyte (normal, abnormal, or nuclear fragment)	-	-	7	0.1	Educational
Neutrophil with dysplastic nucleus and/or hypogranular cytoplasm	-	-	3	0.1	Educational
Neutrophil, metamyelocyte	-	-	3	0.1	Educational
Platelet satellitism	-	-	3	0.1	Educational

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The arrowed cell is a malignant lymphoid cell (Sézary cell), as correctly identified by 62.6% of the referees and 56.2% of the participants. Sézary cells are classically found in patients with leukemic manifestations of mycosis fungoides, a form of primary cutaneous T-cell lymphoma. These cells are usually round to oval, but they can be irregular. They range in size from 8 - 20 µm, and their N:C ratio varies from 7:1 to 3:1. Smaller Sézary cells, as seen in this peripheral blood smear, are slightly bigger than normal lymphocytes and have folded, grooved, or convoluted nuclear membranes, which may give them a cerebriform appearance. The chromatin is dark and hyperchromatic without visible nucleoli. Larger Sézary cells can be more than twice the size of normal lymphocytes. The nucleus is also convoluted and cerebriform appearing with hyperchromatic chromatin. Often, the nuclear membrane is so folded that the nucleus may appear lobulated or even similar to a cluster of berries. Some cells may exhibit a small nucleolus, although this is not a prominent feature. Both large and small Sézary cells have scant, pale blue to gray agranular cytoplasm, and they may contain one or several small vacuoles that lie adjacent to the nucleus. While the appearance of Sézary cells is distinctive, other T-cell lymphomas and some cases of B-cell lymphoma can mimic Sézary cells. Small populations of Sézary-like cells have been reported in normal, healthy individuals, comprising up to 6% of lymphocytes.

The arrowed cell is incorrectly identified by 26.4% of the referees and 29.1% of the participants as a lymphocyte and incorrectly identified by 7.7% of the referees and 7.9% of the participants as a reactive lymphocyte. The atypical features (discussed above) exclude a lymphocyte. The key distinguishing feature of reactive lymphocytes is their wide range of cellular sizes and shapes, as well as nuclear sizes, shapes, and chromatin patterns. These cells are reacting to an immune stimulus and are frequently increased in viral illnesses. The classic example is infectious mononucleosis (acute Epstein-Barr virus infection). Reactive or atypical lymphocytes can also be found in a variety of other viral infections (including cytomegalovirus, adenovirus, or acute HIV infection) protozoal infections (such as toxoplasmosis), some drug reactions, connective tissue diseases, and after major stress to the body's immune system. A variety of reactive lymphocyte forms have been described and they are often seen concurrently in the same blood film. These round to ovoid to irregular cells range from 10 - 25 µm in size with an N:C ratio that varies from 3:1 to 1:2. The most common type of reactive lymphocyte resembles a large lymphocyte and corresponds to a Downey type II cell. These cells have round to oval nuclei, moderately condensed chromatin (giving it a smeared appearance), and absent or indistinct nucleoli. They contain abundant pale gray-blue cytoplasm. Granules, if present, are usually small and few in number. Frequently, these reactive lymphocytes have an amoeboid cytoplasm that partially surrounds adjacent red cells and has a darker-staining, furled margin. Basophilia radiating out from the nucleus may also be present. Immunoblasts and immunoblastic-like reactive lymphocytes are large cells (15 - 20 µm) with round to oval nuclei. They have finely to moderately dispersed chromatin with abundant parachromatin and one or more prominent nucleoli. These may resemble lymphoma cells or blasts. Their cytoplasm is moderately abundant and stains deeply basophilic. The N:C ratio is high (3:1 to 2:1). These reactive lymphocytes correspond to Downey type III cells. Another type of reactive lymphocyte is referred to as a Downey I cell. These cells are rare. These cells possess scant to moderate amounts of basophilic cytoplasm. The nuclei often appear indented, folded, or lobulated. The chromatin is condensed. A few small vacuoles may be present. Granules may also be apparent. Plasmacytoid lymphocytes resemble plasma cells and are intermediate in size (10 to 20 µm) and round to oblong in shape. They have round nuclei that are centrally placed or slightly eccentric. The chromatin is slightly to moderately coarse and forms small dense masses or a meshwork of strands resembling that of plasma cells. Nucleoli are generally not visible, but some cells may have one or two small irregular nucleoli. The cytoplasm is moderately abundant, homogeneous, and light blue to deep slate-blue, and it may show a perinuclear clear zone, or hof. Of the common reactive lymphocyte variants, only Downey I cells may resemble Sezary cells, and the distinction between those two may be difficult when examining individual cells, rather than the entire spectrum of lymphocyte morphology from the peripheral blood smear slide. However, since Downey type I lymphocytes are rare, and Sezary cells typically show a distinct population, a comprehensive slide review is helpful in the differential diagnosis.

Case Presentation:

This peripheral blood smear is from a 78-year-old man presenting with leukocytosis and recent progression of skin tumors. Laboratory data include: WBC = 35.6 × 10E9/L; RBC = 4.44 × 10E12/L; HGB = 13.3 g/dL; HCT = 40.0%; MCV = 91 fL; and PLT = 423 × 10E9/L.

(PERIPHERAL BLOOD, WRIGHT-GIEMSA)

Case Discussion: Sézary syndrome

Mycosis fungoides (MF) is a primary cutaneous lymphoma characterized by epidermotropic neoplastic T-cells with characteristic morphologic features (cerebriform nuclei). Sézary syndrome (SS) is defined by the presence of neoplastic T-cells ("Sézary cells") in the peripheral blood of patients with erythroderma and generalized lymphadenopathy. Similar to SS, patients with advanced stage MF demonstrate circulating neoplastic cells (Sézary cells) in the peripheral blood and, in the absence of any clinical history, peripheral blood smears from both conditions show similar morphologic findings.

From an epidemiologic standpoint, MF is the most common type of cutaneous T-cell lymphoma (approximately 50%), and occurs mostly in adult to elderly patients, with a male:female ratio of 2:1. In contrast, SS is a rare disease (5% of cutaneous T-cell lymphomas) and it also has a predilection for affecting older male adults, characteristically over the age of 60.

Clinically, MF has an indolent, protracted clinical course (years or decades) characterized by skin lesions evolving through different stages (patches, plaques and, eventually, tumors). Only patients with advanced stages of MF show extracutaneous dissemination, including lymph nodes, liver, spleen and blood. Patients with SS have a more dramatic clinical picture with generalized disease, including general skin rash and leukemic presentation. The characteristic morphology of the neoplastic cells (Sézary cells) is that of small to medium-sized lymphocytes, with irregularly convoluted ("cerebriform") nuclei, powdery chromatin, and small amount of cytoplasm.

The diagnosis of MF/SS is based on clinical findings, morphology, immunophenotyping (by flow cytometry or immunohistochemistry) and genetic/molecular analysis. In recent years, it has been recognized that the degree of peripheral blood involvement by lymphoma cells is an important prognostic indicator in patients with MF/SS. Even though there is consensus on the importance of assessing circulating neoplastic T-cells, there is no universally accepted method to characterize and quantify the number of Sézary cells in peripheral blood. Morphologic identification of abnormal lymphocytes with cerebriform nuclei was previously the standard approach for disease detection and quantification. However, this method is compromised by several drawbacks, including high interobserver variability and difficulty in reliably identifying small Sézary cells. Flow cytometric immunophenotyping has proven to be more reliable than morphology in the detection of circulating Sézary cells, as these cells often have an aberrant immunophenotype, including typically expression of CD2, CD3, CD4, and CD5, and absence of CD8, CD7, and CD26. Figure 1 shows an example of peripheral blood flow cytometry from a patient with SS:

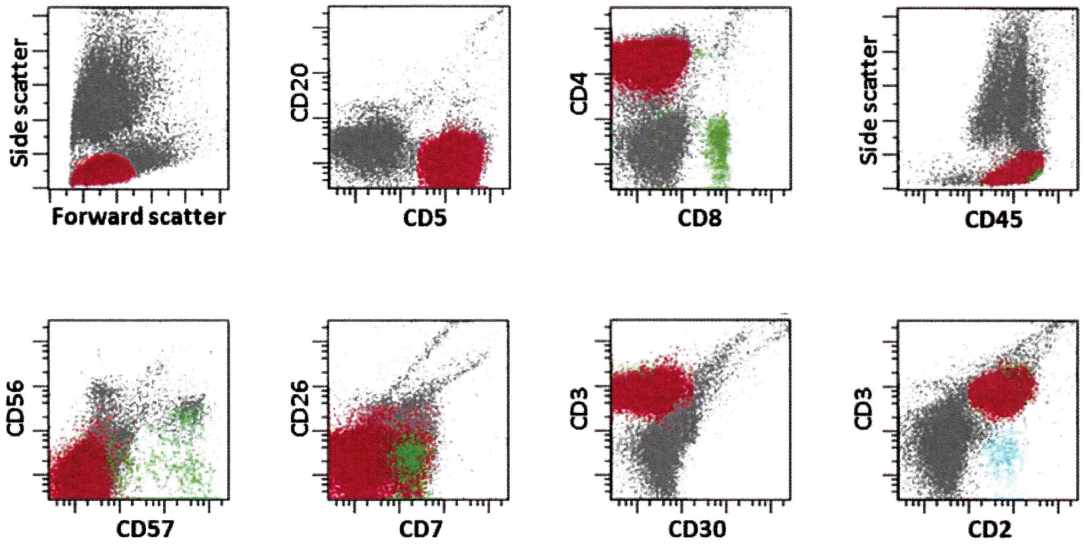

Figure 1: Peripheral blood flow cytometry from a patient with Sézary Syndrome. Immunophenotypic analysis demonstrates a predominant population of aberrant T-cells (in red) that are CD2(+), CD3(+), CD4(+), CD5(+), CD7(partial +), CD8(-), CD26(-), CD30(-), CD56(-), CD57(-). For comparison, normal T-cells (green) and NK cells (cyan) are shown.

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References:

1. Ralfkiaer E, Cerroni L, Sander CA, Smoller BR, Willemze R. Mycosis fungoides. In: Swerdlow SH, Campo E, Harris NL, et al (eds.). *WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues*. 4th ed. Lyon: IARC Press; 2008:190-148.
2. Ralfkiaer E, Willemze R, Whittaker SJ. Sézary syndrome. In: Swerdlow SH, Campo E, Harris NL, et al (eds.). *WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues*. 4th ed. Lyon: IARC Press; 2008:190-148.