Diagnosis of Relapsed Burkitt's Lymphoma in a Urine Sample: an Unusual "FISHing" Expedition

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Extranodal involvement of the urogenital tract (UGT) by non-Hodgkin's lymphoma is not infrequent [1]. A tissue sample from the UGT is often required for a de novo diagnosis of lymphoma or confirmation of its relapse. Taking a biopsy from the UGT is an invasive procedure and thus not free of complications. Moreover, UGT biopsy is relatively demanding of hospital resources. Urine analysis, on the other hand, has none of those disadvantages. We describe a case in which an analysis of a patient's urine sample, including cell morphology, flow cytometry and in situ hybridization, yielded the diagnosis of relapsed Burkitt's lymphoma.

PATIENT DESCRIPTION

A 48 year old man was admitted for investigation of pain along the left shoulder and arm accompanied by numbness of the jaw. The physical examination revealed multifocal neurological deficits. The initial laboratory investigation showed mild anemia (hemoglobin 12.6 g/dl), elevated lactate dehydrogenase levels (2972 U/L, normal range 208–378U/L) and normal kidney function. A further workup revealed positive serology for hitherto unknown human immunodeficiency virus (HIV), with a CD4 count of 80 cells/μl. A whole-body computed tomography (CT) scan detected multiple hypodense lesions involving the liver, adrenals and both kidneys. Magnetic resonance imaging demonstrated an extensive paravertebral soft tissue mass penetrating the spinal canal at multiple heights, a significant thickening along the left brachial plexus as well as multiple bone lesions.

A CT-guided biopsy from a lytic lesion in the sacrum detected a heavy infiltration of the marrow by sheets of neoplastic lymphoid cells of intermediate size, with a high mitotic rate and inconspicuous nucleoli. Stains for CD 20, CD 79a and CD 10 were positive in the neoplastic cells and negative for CD3, CD5, CD30, CD34, cyclin D1 and myeloperoxidase. All the cells were positively stained for Ki67. Similar neoplastic lymphoid cells were also present in the fluid of a spinal tap.

The patient was diagnosed as having AIDS-associated Burkitt's lymphoma involving both the central and peripheral nervous systems. He was treated according to the hyper-CVAD-rituximab protocol, together with a highly active antiretroviral regimen, and achieved complete remission based on a marked clinical improvement and a negative PET-FDG scan that showed complete regression of both the soft tissue masses and the lytic lesions. The HIV viral load was undetectable and the CD4 count had improved considerably (650 cells/μl).

Approximately one year later, the patient was admitted to hospital due to fever. The physical examination was unremarkable, but the initial laboratory workup revealed an elevated creatinine level (2 mg/dl). The initial urinalysis detected mild proteinuria with no cellular elements. A CT scan demonstrated bilateral retroperitoneal masses infiltrating the kidneys and obstructing the pelvis of the right kidney, causing hydronephrosis. Since these findings were highly suspicious for relapse of the Burkitt's lymphoma, and in an attempt to confirm the diagnosis histologically, a bone marrow biopsy and a lumbar puncture were performed; both failed to detect any evidence of disease.

At this point, having thus far been unable to document a relapse, we proceeded with an examination of the patient's urine. A morphologic analysis of cells was performed after fresh urine was centrifuged at 700 rpm for 5 minutes, followed by staining of air-dried slides by the May-Gruenwald-Giemsa method. The sediment showed medium-sized lymphoid cells with basophilic cytoplasm, prominent cytoplasmic vacuoles, round nuclei with clumped chromatin, and clearly visible nucleoli [Figure 1A]. To further define these cells, we performed a flow cytometry analysis of the urine, which detected an abnormal B cell population expressing CD 19, 20, 10, 79a, with a kappa light chain restriction.

The patient's urine was also studied by fluorescence in situ hybridization (FISH) with a probe aimed at the 8;14 c-myc translocation [Figure 1B]. Sixty of the 200 analyzed cells (30%) were found to carry the translocation. An ultrasound-guided biopsy taken from the right perirenal mass confirmed the diagnosis of relapsed Burkitt's lymphoma. Bilateral percutaneous nephrostomes were inserted for decompression of the renal collecting

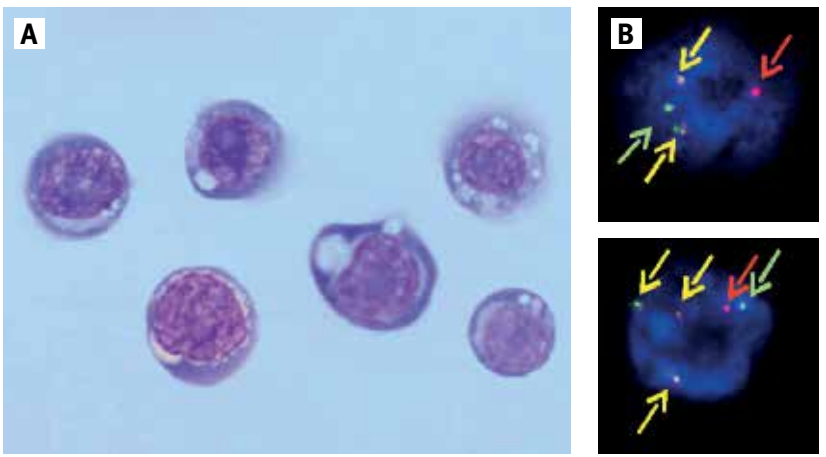

Figure 1. [A] Urine cytospin sediment sample containing medium-sized lymphoid cells with basophilic cytoplasm, cytoplasmic vacuoles, round nuclei, clumped chromatin and visible nucleoli

[B] Urine in situ hybridization. The hybridization pattern of the normal nuclei consists of two red (cMYC) and two green (IgH) signals (2R/2Gconformation). The hybridization pattern of the nuclei with translocation t(8;14) consists of one red (cMYC), one green (IgH) and two fusion yellow [cMYC-IgH Variable on der (8) and IgH Constant – cMYC on der (14)] signals (2Y/1R/1G) (upper panel). More than two fusion signals can be explained by duplication of der(8) or der(14) (lower panel)

system, with subsequent normalization of the creatinine levels. Salvage chemotherapy was initiated using the CODOX-M/IVAC protocol. Unfortunately, the clinical course was complicated by neutropenic fever and the patient died due to severe sepsis.

COMMENT

In an ascending degree of specificity for diagnosing UGT involvement by lymphoma, the initial routine urinalysis can often detect proteinuria, pyuria and hematuria. Urine cytology may identify abnormal lymphocytes and suggest the diagnosis of lymphoma, especially when performed on concentrated urine sediment [2]. Furthermore, such abnormal lymphocytes can be immunophenotyped by using flow cytometry on the urine sample [3,4]. The acidic properties of the urine might potentially blur the morphologic and immunophenotyping properties of lymphoid malignant cells, but this technical obstacle can be overcome if the sample is processed quickly. In certain types of lymphoma, a definite diagnosis is supported by identifying specific chromosomal translocations, such as t8;14 in Burkitt's lymphoma and t11;14 in Mantle cell lymphoma [5].

We believe this to be the first report of a FISH analysis performed on a urine sample for the diagnosis of Burkitt's lymphoma. This case suggests that a meticulous analysis of the urine, which includes morphology, flow cytometry and FISH for specific translocation, may spare a patient with suspected kidney involvement by lymphoma from undergoing an invasive procedure, especially in the setting of suspected relapse.

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