

#### HAEMATOLOGY | September 2023

Leukaemias/malignancies

# Identifying a typical CLL by means of two complementary technologies



## Clinical information and laboratory results

A patient presented to the general practitioner due to feeling unwell. A peripheral blood sample was drawn and sent to the laboratory. Initial analysis with an XN-20 showed a normal PLT count and no anaemia but revealed a markedly increased WBC count of 26.17 ×10<sup>3</sup>/µL, of which 80 % were lymphocytes (LYMPH# 21.15 ×10 $^{3}$ /µL).



Fig. 1 The XN IPU Lab. Only screen shows the differential count and the WDF, WNR and WPC scattergrams.

A blood smear using the DI-60 automated digital imaging analyser revealed small lymphocytes with a high nuclear-to-cytoplasmic ratio and a condensed chromatin nuclear pattern. Smudge cells were also present, which are typically seen in patients with chronic lymphocytic leukaemia [1].

After this suspicion of an abnormal lymphocyte population, the sample was sent to the flow cytometry lab for further clarification and analysis by immunophenotyping. Measurement on an XF-1600 revealed a dim CD19/CD5 dual-positive population. In addition, a monoclonal proliferation of the lambda light chain was observed. The sample also expressed CD20, CD23 and CD79b and was negative for CD10, CD11c and CD38.



Fig. 2 Digital imaging (by a DI-60) showing many smudge cells.



Fig. 3 Scatterplots of lambda, CD19, CD5, CD20 and CD23 from the XF-1600 which shows an abnormal monoclonal proliferation of B cells with lambda immunoglobulins (purple).

## **Result interpretation**

In a typical CLL, abnormal lymphocytes are usually small, mature-appearing cells with low mitotic activity [2]. Neoplastic lymphocytes, which are more mature, have readily permeated membranes, due to the higher lipid content of the cell membranes, and give off a high fluorescence signal after treatment with WPC channel reagents, which can easily enter the cells and bind to the DNA.

The lineage of lymphocytes cannot be reliably determined with a Pappenheim stain, and only some observations can be made morphologically. To determine the lymphocytes reliably, immunophenotyping is necessary. Abnormal lymphocytes in CLL express the surface antigens CD19, CD20, CD5, CD23, CD43 and CD200 with expression of either kappa or lambda light chains [3]. CD10 is found negative. The immunophenotyping results point to a diagnosis of B-CLL.

#### References

- [1] Marionneaux SM et al. (2021): Smudge Cells in Chronic Lymphocytic Leukemia: Pathophysiology, Laboratory Considerations, and Clinical Significance. Lab Med. 52(5): 426–438.
- [2] WHO (2017): Classification of Tumours of Haematopoietic and Lymphoid Tissues, Revised 4th Edition, Volume 2.
- [3] Lanasa MC et al. (2010): Novel insights into the biology of CLL. <u>Hematology Am Soc Hematol Educ Program. 2010: 70–6.</u>