ISSUE
12
April
2021

QSP Monthly

NEWSLETTER OF THE QUALITY SLIDE PROGRAM

HORIBA Medical

This issue

Last Month's Slides

March 2021 Slide Summaries

Side 1
Follow up of CLL
Expert comment – Evolution of an atypical CLL with 3/5 matures scoring to be compared with the rest of the clinical results

Slide 2
Discreet Eosinophilia

Slide 3
Occasional large platelets

Slide 4
Discreet Basophilia

Slide 5
Multiple alarms on analyser
Expert Comment – Dimorphic RBC, acanthocytes, Macrothrombocytes, Basophilia
Investigate for Myeloproliferative disorder

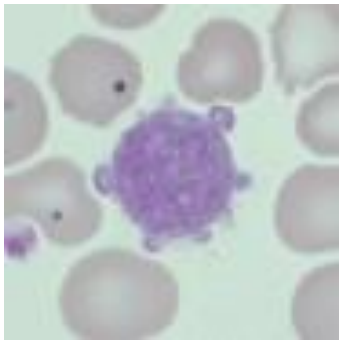
Slide 6
Neutrophilia, presence of myelocytes, metamyelocytes, promyelocytes, nucleated red blood cells

Monthly Digital Case study
Slide 5

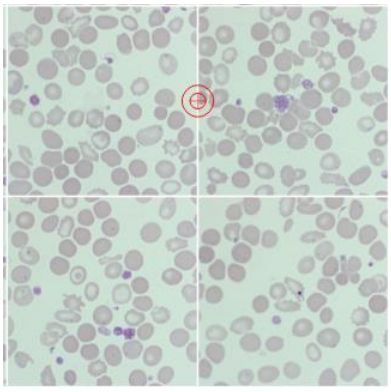
Presentation
Male (65 years old)

FBC Results

WBC	**17.2*(10^3/mm3)**	**Neutrophils**	**76.2 %**
RBC	**4.27* (10^6/mm3)**	**Lymphocytes**	**1.8 %**
HGB	**108* (g/L)**	**Monocytes**	**8.8 %**
HCT	**36.4* (%)**	**Eosinophils**	**4.4 %**
MCV	**85*(fL)**	**Basophils**	**3.5%**
MCH	**25.3* (pg)**		
MCHC	**29.7* (g/dL)**		
PLT	**413*(10^3/mm3)**		
RDW-SD	**87.4 *(fL)**		
RDW-CV	**29.2* (%)**		



Macrothombocyte



RBC picture

Slide review
Multiple alarms on analyser – represented by *
Dimorphic RBC, acanthocytes, Howell Jolly Bodies, target cells Basophilia, Macrothrombocytes (see above).

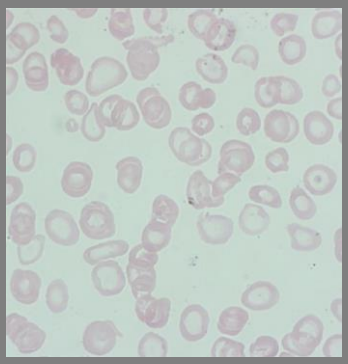
Diagnosis
Myeloproliferative

QSP Quality Slide Program 2.0

Morphology Webinar:

HORIBA recently hosted a webinar on **"An Introduction to Morphology"**, which includes a demonstration on how to use QSP 2.0 in training. You can now watch it on demand **here.**

Last month's Quiz:



Slide B shows a microcytic, hypochromic red cell picture. Red cells are smaller, with a larger than normal central area of pallor. This decrease in redness is due to a disproportionate reduction of haemoglobin in the red cells in proportion to the red cell volume. The colour of the red cells can be evaluated by the MCH (mean corpuscular haemoglobin), and also the MCHC (mean cell haemoglobin concentration).
The most common causes of Microcytic Hypchromic anaemia are iron deficiency and thalassaemia.

Reticulocyte measurement on HORIBA analysers

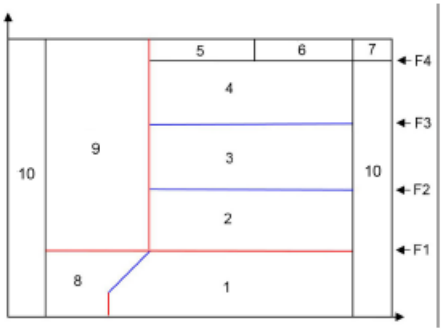
Reticulocytes are immature Red Blood Cells (RBC) that are being constantly released from the bone marrow (approx. 2.5 billion red cells/Kg/day). The stages of red cell development within the bone marrow include pronormoblast, basophilic normoblast, polychromatophilic normoblast, orthochromatic, reticulocyte. Reticulocytes are released from the bone marrow after the nucleus has been extruded at the orthochromatic stage. In normal adults, the red cell has a lifespan of approx. 120 days, therefore new red cells have to be constantly produced. Red cell production is controlled by the hormone Erythropoietin which increases as the blood oxygen levels decrease. Reticulocytes can be identified because they still contain ribosomal RNA required to complete the production of Hb. Reticulocytes mature in the peripheral circulation for between 1 to 2 days finally becoming mature red cells. Reticulocytes tend to be larger than mature Red Cells and often show a blue tinge (Polychromasia). The normal reticulocyte count is 0.5% - 2.5% of Red cells or 20 – 80 $x10^9$/L.

It is important to measure Reticulocytes as they give an indication as to the state of red cell production. An increase in Reticulocytes (Reticulocytosis) indicates that the bone marrow is responding to the bodies need to produce more red cells. A decrease in blood oxygen levels leads to an increased production of the hormone Erythropoietin, which stimulates the bone marrow to increase red cell production, thereby increasing the number of Reticulocytes (Reticulocytosis) entering the peripheral blood. Reticulocytosis may be present, even if the patient is not clinically anaemic. Causes of reticulocytosis are: haemolysis, blood loss and Iron/B12/Folate deficiency. A low Reticulocyte count is indicative of the failure of the bone marrow to produce RBC and could indicate deficiency of Vitamin B12, Folate or iron, anaemia of chronic disease, Aplastic anaemia etc.

Methodology

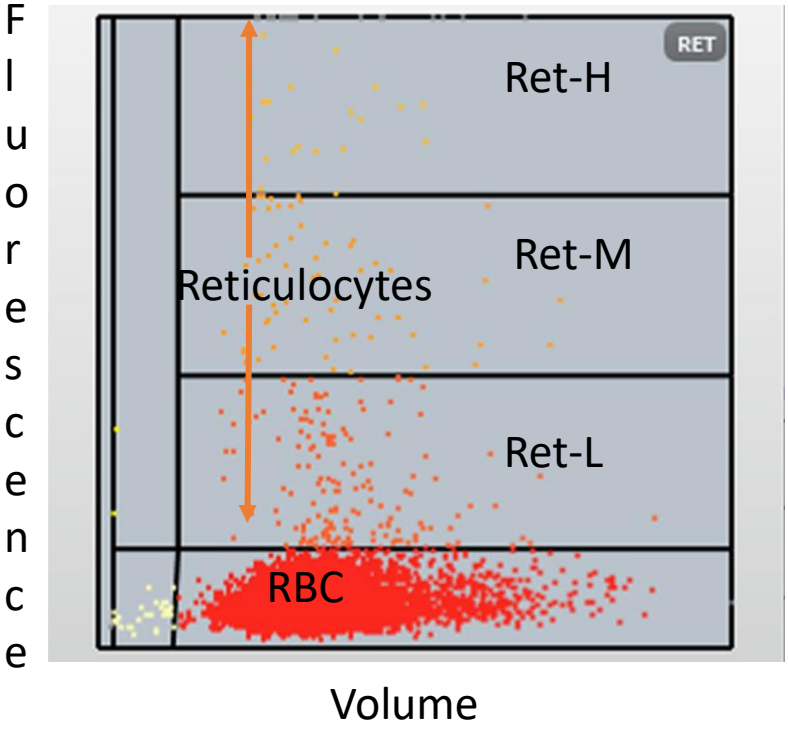
HORIBA Medical uses the reagent Fluocyte which contains the fluorochrome Thiazole Orange to measure reticulocytes. Thiazole Orange which binds to any residual RNA present within the red cell. Therefore, if RNA is present, fluorescence will be induced by a laser (488nm) and be detectable.

0.5 uL blood is incubated with 5mL Fluocyte at 35^0C for 51 sec, then transferred to the flow cell where simultaneous measurement of volume (aperture impedance) and orthogonal fluorescence (laser excitation 488 nm) is performed. Any RNA present on the cell causes an increase in the degree of fluorescence above a motile threshold. A matrix (see below) is produced showing the volume and orthogonal fluorescence of each cell, a threshold separates the Reticulocytes from the mature RBC. The Reticulocyte area is split into 3 areas, indicating the degree of fluorescence and therefore their maturity – the greater the fluorescence the more immature. The Reticulocyte percentage is calculated from the number of cells in the reticulocyte area not including any cells in the NRBC or WBC area.



1 = RBC
2 = RET_L
3 = RET_M
4 = RET_H
5 = IMR
6 = NRBC
7 = WBC
8 = PLT
9 = PLT_RET
10 = Background Noise

Reticulocyte Matrix as displayed on analyser



Reticulocyte Parameters

Percentage Reticulocyte
Absolute Reticulocyte
Reticulocyte Heamoglobin Cellular Content (RHCc)
Mean Reticulocyte Volume (MRV)
Immature Reticulocyte Fraction (IRF)
Mean Fluorescent Index (MFI)
Corrected Reticulocyte count

For more information, watch the video.

BREAKING NEWS

This month our QSP Newsletter, which is part of HORIBA's ongoing commitment to support scientists interested in Blood Film Morphology has reached the milestone of its first year of publication.

On this occasion, we are delighted to announce that **QSP 2.0 multi- site license now works across a Virtual Private Network (VPN),** enabling users to connect multiple QSP PC (10 concurrent) across different sites, even if they are not on the same local area network.

If you would like to find out more about QSP 2.0 and discuss your special requirements, **contact us.**

QSP 2.0

Available as a single use license and a site license which allows up to 10 concurrent users.
To find out more, contact us.

Bibliography

HORIBA Yumizen H2500 user manual

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