

Reticulocytes indices in β thalassemia trait individuals

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The thalassemias are a diverse group of microcytic and hemolytic anemias that are characterized by defective synthesis of one of the globin chains of adult hemoglobin. The β -thalassemias are characterized by a reduced (β^+ -thalassemia) or an absence (β^0 -thalassemia) of β globin synthesis which leads to imbalanced globin chain production.^(1,2) This imbalanced synthesis results in a variable degree of anemia which stimulates erythropoietin production, leading to proliferation and expansion of the bone marrow. The β -thalassemias have a considerable phenotypic variation depending on multiple factors, which include the nature of the mutation involved. This leads to a wide range of presentations from profound anemia (requiring lifelong blood transfusions – β -thalassemia major) to extremely mild anemia (β -thalassemia trait).⁽²⁾

Reticulocytes are juvenile red blood cells; they contain remnants of the ribosomal ribonucleic acid which is present in large amounts in the cytoplasm of the nucleated precursors from which they are derived. The number of reticulocytes in the peripheral blood is a fairly accurate reflection of erythropoietic activity assuming that reticulocytes are released from the bone marrow after the 'normal' time, and that they remain in circulation for the 'normal' period of time.⁽³⁾

A total of 152 unrelated adults were included in this study: thirty with the β -thalassemia trait diagnosed by high-performance liquid chromatography (HPLC-Variant, Bio-Rad, Milan, Italy)⁽⁴⁾ with sequencing of the HBB gene using the primers described by Kimura⁽⁵⁾ and Miranda⁽⁶⁾ and 122 individuals recruited during their routine blood counts at the Pharmacy School Laboratory of the Universidade Federal do Rio Grande do Sul. The Ethics Committee of Hospital de Clínicas de Porto Alegre, Rio Grande do Sul approved the study protocol.

Peripheral blood was collected using EDTA as anticoagulant. Hematological and reticulocyte data were obtained in an automated cell counter - Sysmex SE9500 (Sysmex, Kobe, Japan). Table 1 shows the hematological indices for β -thalassemic trait and control individuals.

Individuals with the β -thalassemic trait presented with significantly higher levels (p -value < 0.05) of the following variables compared to controls: reticulocytes (percentage and number), medium fluorescence reticulocytes, high fluorescence reticulocytes and immature reticulocyte fraction. These results are in agreement with those reported by Noronha & Grotto with the exception of the immature reticulocyte fraction, where no

Table 1 - Mean and reference ranges for hematology laboratory values in the Municipal Laboratory of Curitiba, PR

| | Mean (range) | | p-value t-test |
|--|----------------------|----------------------|-------------------|
| | Women | Men | |
| Red blood cells (x 10 ¹² /L) | 4.7 (4.0 - 5.4) | 5.2 (4.3 - 6.1) | <0.05* |
| Hemoglobin (g/L) | 136.2 (118 - 154) | 152.8 (127 - 177) | <0.05* |
| Hematocrit (L/L) | 0.41 (0.35 - 0.46) | 0.45 (0.38 - 0.52) | <0.05* |
| Mean cell volume (fL) | 87.3 (78.0 - 95.1) | 87.9 (78.0 - 97.2) | |
| Mean cell hemoglobin (pg) | 29.3 (25.6 - 32.1) | 29.6 (26.1 - 32.7) | <0.05 |
| Mean cell hemoglobin concentration (g/L) | 335.8 (319 - 354) | 336.9 (322 - 354) | <0.05 |
| Red cell distribution width (%) | 13.7 (11.8 - 16.7) | 13.8 (12.0 - 16.3) | <0.05 |
| White blood cells (x 10 ⁹ /L) | 6.7 (3.84 - 10.4) | 6.7 (3.9 - 10.9) | ** |
| Eosinophils (%) | 3.5 (0 - 11) | 4.3 (1 - 13) | <0.05 |
| Eosinophils (x 10 ⁶ /L) | 228.5 (56 - 682) | 284.6 (65 - 940) | <0.05 |
| Basophils (%) | 0.5 (0 - 1) | 0.6 (0 - 2) | <0.05 |
| Basophils (x 10 ⁶ /L) | 29.6 (0 - 99) | 41.7 (0 - 125) | <0.05 |
| Lymphocytes (%) | 33.2 (21 - 48) | 33.8 (19 - 49) | |
| Lymphocytes (x 10 ⁶ /L) | 2175.3 (1157 - 3500) | 2223.2 (1265 - 3648) | ** |
| Monocytes (%) | 6.9 (4 - 11) | 7.5 (3 - 12) | <0.05 |
| Monocytes (x 10 ⁶ /L) | 455.1 (208 - 807) | 503.2 (192 - 968) | |
| Neutrophils (%) | 56 (40 - 70) | 53.8 (35 - 69) | |
| Neutrophils (x 10 ⁶ /L) | 3777.3 (1804 - 6460) | 3762.7 (1728 - 6820) | |
| Platelets (x 10 ⁹ /L) | 284.1 (175 - 421) | 258.6 (163 - 399) | <0.05** |

* - normal; ** - log-normal distribution evaluated by the Shapiro-Wilks test;

statistical difference was seen between the β -thalassemic trait and control group.⁽⁷⁾ In this study no significant difference was found between the groups for low fluorescence reticulocytes.

The reticulocyte count is used as an indicator of the erythropoietic activity of bone marrow in different anemias.⁽⁷⁾ Manual techniques (such as supravital staining) have great inter- and intra-observer variability and often the results are inaccurate. Automated cell counting has overcome this limitation. The availability of reticulocyte maturation indices, based on the measurement of RNA content extends the clinical utility of reticulocyte determination.

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