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Persistent increase in red cell size distribution width after acute diseases: A biomarker of hypoxemia?

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ABSTRACT

Background: A biomarker of hypoxic exposure would be useful in clinical diagnosis and prognosis. Acute hypoxia stimulates large increases in serum erythropoietin (EPO), and EPO induces formation of characteristic enlarged red blood cells (RBCs). The presence of large RBCs perturbs red cell distribution width (RDW).
Methods: Using a >2 M patient medical claims database, the human pathome was scanned for diseases where RDW rose 0–50 days following a new diagnosis. The course of RDW after selected diagnoses was visualized by registering RDW measurements by diagnosis date.
Results: Acute hemorrhage, which provokes EPO-driven erythropoiesis, is followed by increases in RDW but not mean cell volume (MCV). Similar RDW increases follow many acute diseases with risk of hypoxia, including heart failure, pneumonia, atelectasis, pulmonary embolism, pneumothorax, and sepsis. Elevations reach maximum within 1 month after onset and subside to pre-disease levels about 6 months later. Unlike the case with iron-deficiency anemia (IDA), RDW elevations after hypoxia-associated diseases are unaccompanied by discernible clinical macrocytosis (increase in average RBC size).
Conclusions: As predicted by a model risk pathway linking hypoxia to formation of enlarged RBCs via EPO, acute hypoxemia-related disease episodes induce change in RBC size distribution. Further study is needed to explore whether a more sensitive and specific signal can be extracted from the fine structure of the RBC size distribution routinely measured in automated hemocytometers.

1. Introduction

Even brief interruptions of oxygen supply can threaten irreversible tissue injury and may reflect serious underlying pathologies. Acute or chronic hypoxemia presents with shortness of breath, cyanosis, and as decreased peripheral blood oxygen saturation (SaO_2) via pulse oximetry. Hypoxemia that is chronic but intermittent can escape routine clinical examination, yet cause incremental tissue ischemia, provoke pathological compensatory responses, and herald overall poor prognosis. A reliable biomarker for chronically episodic hypoxia would be a useful clinical tool. In this investigation we explore a hypothesis that transient hypoxic episodes can leave a durable residual signal in the form of a subpopulation of unusually large RBCs. Because RDW is sensitive to the presence of even small numbers of large RBCs, this commonly reported measure can be used to probe the linkage of RBC size distribution to hypoxic diseases.

2. Theory

2.1. Model of pathway linking hypoxemia to RDW elevation

Transient oxygen deficiencies stimulate an increase in oxygen carrying capacity through up-regulation of RBC production. In response to decreased oxygen partial pressure (PaO_2), the renal cortex secretes erythropoietin (EPO) into the circulation via hypoxia-inducible transcription factors [1]. EPO increases regardless of the etiology of hypoxemia, e.g., experimentally imposed hypobaria [2], chronic obstructive pulmonary disease (COPD) [3,4], heart failure [5], interstitial lung disease [6], or pulmonary arterial hypertension [7].

EPO amplifies erythropoiesis in bone marrow by suppressing apoptotic wastage of erythroid progenitor cells, resulting in increased RBC production and a higher proportion of reticulocytes. Because erythropoiesis negatively rebounds following EPO withdrawal [8], infrequent EPO pulses can be damped out and have only a modest quantitative effect on measures such as hemoglobin (HGB) and hematocrit. In athletes, recombinant human EPO (rhEPO) increases the fraction of large (>120 fl) RBCs from 0.7% to 1.8% after administration. This signal

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persists for 1 month after treatment offset [9]. Because hypoxemia is accompanied by spikes of endogenous EPO such large cells are a potential biomarker for recent hypoxic experience. Arithmetically, admixture of large RBCs in the 120 fl range can induce mild elevations of RDW (Section 3). The model linking hypoxemia to subsequent increases in EPO and RDW is illustrated in Fig. 1.

We note that the presence of large RBCs in small numbers is distinct from clinical macrocytosis, which is defined as increased average cell volume. To avoid confusion, here we use 'heterocyte' for RBCs falling outside the typical RBC size distribution, and avoid the term 'macrocyte' which is commonly used to specify vitamin-deficiency related megaloblasts.

2.2. A risk-signaling pathway?

Causal pathways transmit signals downstream as observable correlations. Any factor correlated with the source (ancestor) node will also show associations with all the downstream (descendent) nodes. Risk of mortality and poor outcomes are correlated with hypoxemia, especially in patients with pneumonia [10], pulmonary hypertension [11], interstitial lung disease [12], and COPD [13]. The hypothesized causal pathway predicts that this association would also be observed for EPO, heterocytes, and RDW.

2.2.1. Association of mortality and hypoxemia with EPO and heterocyte fraction

Mortality has been associated with serum EPO in patients with sepsis [14,15], heart failure [5], the critically ill [16], and the elderly [17]. The enlarged RBCs associated with EPO treatment fall in a distinct size range that has not been studied as predictor of risk, but another abnormal RBC has been found strongly associated with both hypoxia and mortality. Peripheral nucleated RBCs (nRBCs) occur at low concentrations (0.03–0.1% of RBCs) in normal neonates [18]; more elevated levels are associated with fetal asphyxia [19]. nRBCs are generally absent from the circulation in healthy adults, but small numbers are frequently encountered in critical care patients where they are associated with $SaO_2 < 75\%$ [20]. In a study of intensive care surgical patients, mortality was 4% in nRBC-negative patients, 44% in nRBC-positive patients, and 100% when nRBC exceeded 0.04% of RBCs [21].

Pulses of EPO flush immature reticulocytes from the bone marrow. Normally, fluorescent-staining early-stage cells are restricted to the bone marrow and absent from the circulation. Following rhEPO treatment, young and very young reticulocytes reach concentrations of 0.32% and 0.08% of RBCs, respectively, and can comprise about 20% of circulating reticulocytes [22]. Premature expulsion of young reticulocytes from the bone marrow before the normal processes of size reduction and hemoglobin synthesis is coincident with the emergence of long-lived large hypochromic erythrocytes. The co-occurrence of these phenomena suggests that EPO-induced heterocytes may originate as RBCs that are released from the bone marrow prematurely. nRBCs, the immediate precursors of young reticulocytes, that are present in the circulation must also result from premature release.

Evidence linking EPO-induced heterocytes and the premature release of reticulocytes and nRBCs is circumstantial and so the association of large RBCs with mortality risk remains conjectural until more direct measurements are made. However, if heterocytes are associated with mortality risk, this association will also be seen for measures sensitive to RBC size variation, such as RDW.

2.2.2. RDW

Admixture of heterocytes induces elevations in RDW (Section 3). RDW elevation has been shown to be a strong independent predictor of morbidity and mortality in patients with cardiovascular disease [23], heart failure [24,25], pulmonary hypertension [26], venous thromboembolism [27], myocardial infarction [28,29], sepsis [30,31], pneumonia [32–34], pulmonary fibrosis [35], as well as in the critically ill [36,37] and the population at large [38,39]. RDW-associated risk has been found to be independent of anemia [40], serum iron and ferritin concentrations

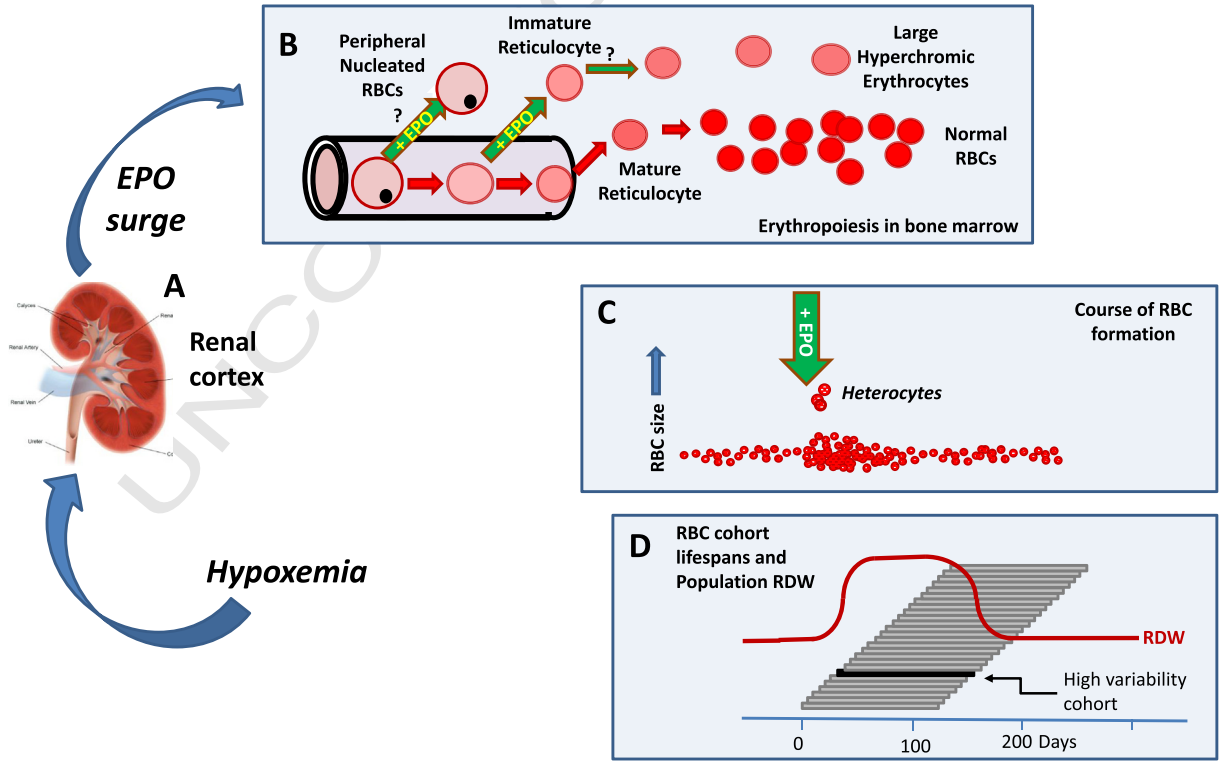

Fig. 1. Schematic of hypoxia–EPO–RDW risk pathway. Hypoxemia triggers EPO secretion (A) in the renal cortex. High serum EPO stimulates the bone marrow endothelium to prematurely jettison nascent RBCs (B) before they have completed normal volume reduction and hemoglobin synthesis, resulting in hypochromic large heterocytes in the circulation (C). An episode of heterocyte release increases RDW for the lifespan of the cohort (D).

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