NEOCYTOLYSIS IN THE ADAPTATION OF RED CELL MASS ON DESCENT FROM ALTITUDE

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RSUMEN: Neocitolisis en la Adaptación de la Masa de Eritrocitos al Descenso de la Altura

Los individuos aclimatizados que descienden de la altura deben adaptarse rápidamente a la policitemia, que es un exceso en la masa de eritrocitos y en el volumen sanguíneo en relación al nuevo ambiente. Creemos que esto se acompana por neocitólisis, que consiste en la destrucción selectiva de los eritrocitos más jóvenes, un proceso fisiológico que descubrimos recientemente a través de estudios en astronautas. Al entrar en microgravedad, la sangre de un astronauta se distribuye centralmente causando policitemia aguda. Luego se produce una disminución reproducible de 10% en la masa de eritrocitos en los primeros días de permanencia en el espacio. La producción de eritrocitos no disminuye en estos primeros días y la supervivencia de los eritrocitos marcados más viejos es normal. La conclusión evidente es que se produce neocitólisis. Estudios realizados hace 45 años en el Perú y en los Himalayas demuestran claramente que hay hemólisis al descender de la altura. Hay un 10% de disminución en la masa de eritrocitos durante los primeros días de permanencia a nivel del mar, además hay un incremento en la bilirrubina sérica y en la urobilina fecal, todo lo que ocurre antes que haya cualquier caída significativa en la producción de eritrocitos. Estudios realizados en sujetos aclimatizados que desciendan de la altura podrán demostrar directamente la neocitólisis,-a través del marcado selectivo de grupos de eritrocitos de diferentes edades. Estos estudios aclararán los mecanismos subyacentes de la neocitólisis, si niveles sub-umbral de eritropoyetina afectan o no la expresión de moléculas de adhesión, así como las interacciones entre fagocitos. La neocitólisis puede tener implicancias amplias en la medicina clínica, incluyendo el aspecto de los regímenes de dosificación de eritropoyetina.

Palabras claves: Eritrocitos, Altura, Adaptación, Neocitólisis, Policitemia

RÉSUMÉ: Néocytolyse dans l'adaptation de la masse d'érythrocytes chez les voyageurs descendant de régions de grande altitude.

Les individus acclimatés qui descendent des hauteurs doivent s'adapter rapidement à la polyglobulie, un excès de la masse d'érythrocytes et du volume sanguin en rapport avec le nouveau milieu. Nous pensons qu'elle est accompagnée de néocytolyse, destruction sélective des érythrocytes les plus jeunes, processus physiologique que nous avons récemment découvert grâce à des études sur les astronautes. En entrant en micro-gravitation, le sang d'un astronaute se distribue centralement, provoquant une polyglobulie aiguë. Survient ensuite une diminution reproduisible de 10 % de la masse d'érythrocytes au début du séjour dans l'espace. Il n'y a pas de diminution de la production d'érythrocytes au cours des premiers jours et la survie des érythrocytes marqués les plus vieux est normale. La conclusion évidente est qu'il se produit une néocytolyse. Des études réalisées il y a 45 ans au Pérou et dans l'Himalaya démontrent clairement qu'il y a hémolyse lorsqu'on descend des hauteurs. La diminution de la masse d'érythrocytes est de 10 % durant les premiers jours de séjour au niveau de la mer, accompagnée d'une élévation de la bilirubine sérique et de l'urobiline fécale, réactions observées avant que ne se produise une chute significative de la production d'érythrocytes. Des études effectuées chez des sujets acclimatés qui descendent des hauteurs pourront démontrer directement la néocytolyse, par le marquage sélectif de groupes d'érythrocytes d'âges différents. Ces études élucideront les mécanismes sous-jacents de la néocytolyse, si les niveaux d'érythropoïétine inférieurs au seuil affectent ou non l'expression de molécules d'adhésion, ainsi que les interactions entre phagocytes. La néocytolyse peut avoir d'amples implications en médecine clinique, y compris l'aspect des régimes de dosification de l'érythropoïétine.

Mots-clés : Erythrocytes, Altitude, Adaptation, Néocytolyse, Polyglobulie.

SUMMARY: .Acclimated individuals descending from high altitude must rapidly adapt to plethora, an excess in red cell mass and blood volume for their new environment. We believe that this is accomplished by neocytolysis, the selective destruction of the youngest red blood cells, a physiologic process we recently discovered through studies on astronauts. On entering microgravity, an astronaut's blood distributes centrally causing acute plethora. There ensues a reproducible 10% decline in red ceil mass in the first several days in space. Red cell production does not decline in these first days, and survival of labeled older red cells is normal. The inescapable conclusion is that neocytolysis ensues.

Studies done 45 years ago in Peru and in the Himalayas clearly demonstrate hemolysis on descent from high altitude. There is a 10% decline in red cell mass in the first several days at sea level, an increase in serum bilirubin and stool urobilin, all occurring before there is any significant fall in red cell production. We preview studies on acclimated subjects descending from high altitude which will directly demonstrate neocytolysis by selectively labeling red cell cohorts of different ages. These studies will elucidate underlying mechanisms of neocytolysis, whether sub-threshold erythropoietin levels affect surface adhesion molecule expression and red cell-phagocyte interactions. Neocytolysis may have broad implications to clinical medicine, including to current dosing regimens of erythropoietin.

Key words: Blood red cels, Hygh-altitude, Adaptation, neocytolisis, Polycythemia.

INTRODUCTION.

Current dogma in hematology holds that red blood cell mass (RBCM) is controlled entirely at the level of red cell production under the influence of the hormone erythropoietin (EPO). EPO works at the level of primitive red cell progenitors, stimulating proliferation of cells committed to an erythroid

maturation pathway, preventing apoptosis of primitive erythroid colony-forming units (1). The actions of EPO are believed to be limited to early progenitor cells, not on more mature normoblasts and erythrocytes. Red cells released into the blood survive 120 days and dogma holds that there are no physiologic mechanisms to shorten red cell survival.

A decrease in RBCM (anemia) is much more commonly observed than plethora. Studies of physiologic responses in normal and anemic individuals gave rise to the principles stated above. There has been relative neglect of physiologic processes that come into play in circumstances of plethora. Through studies of the unusual environment of spaceflight, we became aware of the process of neocytolysis, the selective hemolysis of young red blood cells. In this paper, we concentrate mainly on how this process is manifest when individuals acclimated to high altitude descend to sea level, and on how studies of such descending individuals can shed further light on neocytolysis. Proving and better understanding this process may have broad implications in physiology and clinical medicine.

Spaceflight anemia. It has been known for decades that astronauts returning from space consistently experience a decline in RBCM of about 10% after spaceflights of 8 to 10 days or more. The mechanism responsible for the anemia has finally been clarified by our group's studies on SLS-1 and SLS-2 (2-3). On entering microgravity, the blood that is normally held in the extremities by gravity suddenly shifts centrally. There ensues a very rapid decline in plasma volume (due to a third "spacing" caused by factors not yet defined) and a decrease in EPO secretion. There are two reasons why a hemolytic mechanism must be invoked to explain the fall in RBCM. First, the fall is too rapid to be explained entirely by decreased red cell production. Second, ferrokinetic studies demonstrate that there is little if any decrease in red cell production during the first several days in space. This is just as predicted from our understanding of EPO action; circumstances effecting a decrease in red cell production transpire after a delay of several days (1).

While hemolysis seemed certain, it was disconcerting that ^{51}Cr-labeled red blood cells were repeatedly found to have normal survival in space. This became understandable when we considered that red cells were labeled with ^{51}Cr twelve or more days before launch. The only way to reconcile all observations is to conclude that on entering microgravity, there is selective hemolysis of the youngest red blood cells, a process we call neocytolysis (3,4) (see figure).

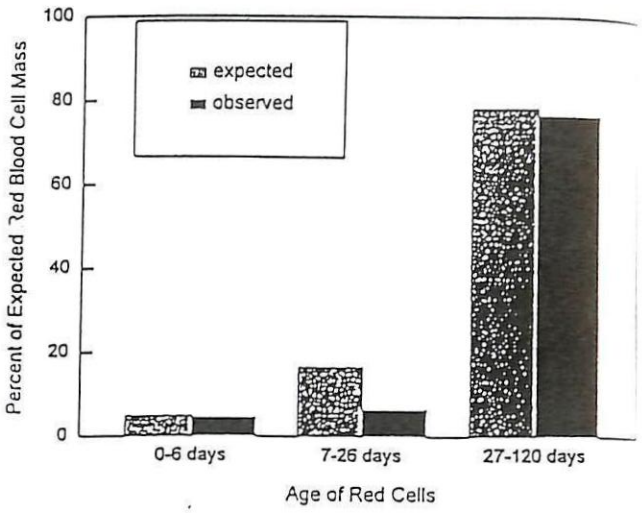

Figure. Red Cell Cohorts Measured Affer Fourteen Days in Space Cells 0-6 days old were labeled with ^{59}Fe on the third day in space. Cells 7-26 days old were unlabeled. These consisted mainly of red cells less than 12 days old at the time of launch. Cells 26-120 days old were labeled with ^{51}Cr. Based on data from SLS-2 astronauts (3).

Prior studies of descent from altitude. Adaptation to the hypoxic environment of high altitude produces polycythemia. Acclimated individuals rapidly descending to sea level find themselves in a situation very similar to astronauts entering microgravity, experiencing plethora maladaptive to their new environment. The changes that occur with descent were studied in Peru in 1950 (5). Merino found that hemoglobin declined by 10 to 18% in the first 10 days after descent. Serum indirect bilirubin and fecal urobilinogen increased markedly with peaks at 6 to 8 days after descent. He concluded that a "very distinct hemolytic process occurred in all cases."

Huff amplified these observations by studying 11 acclimated natives descending from high altitude in Peru to sea level (6). A fall of 9% in RBCM occurred in 8 to 10 days. Ferrokinetic studies, bone marrow examinations and reticulocyte counts again demonstrated no significant decline in red cell production during the first several days of the RBCM decline, but there was a substantial fall in red cell production later on. Pace's observations on ten men living in the Himalayas and returning to sea level echoed the conclusion that a hemolytic mechanism was necessary to explain the early, rapid fall in RBCM (7).

Thus, observations made on descent from high

altitude complement those made on astronauts entering microgravity and provide further evidence that some type of hemolytic mechanism comes into play in adapting to plethora. Confusing the issue was a study which showed that red cells of rats had normal survival on descent from altitude (8). Just as in the astronauts, the rat red cells were labeled with ^{51}Cr several days before descent. Neocytolysis, hemolysis selectively affecting the youngest red blood cells, was not considered by the investigators of descent from altitude but it would have reconciled all observations.

Proposed mechanism of neocytolysis. Newly-released red blood cells interact intimately with reticuloendothelial phagocytes particularly in the spleen. Inclusions such as Howell-Jolly bodies are pitted and culled, and red cell membrane phospholipid is conditioned. Surface adhesion molecules have been found to be important in red cell maturation (9). Among circulating red cells, adhesion molecules are richest on the youngest cells. It seems likely that neocytolysis is mediated through changes in red cell-reticuloendothelial cell interaction resulting from changes in surface adhesion molecule expression.

EPO is the main regulator of RBCM by up-regulating red cell production in times of need, and there is reason to believe that EPO remains the main regulator of RBCM in adaptation to plethora. A fall in EPO levels below a critical threshold may initiate neocytolysis. Neocytolysis occurs in just the situations where EPO levels are suppressed. How EPO effects changes in adhesion molecule expression remains to be elucidated.

Neocytolysis can be viewed as an example of a general emerging physiologic paradigm, an extension of apoptosis to non-nucleated cells. The body maintains homeostasis and is able to adapt to environmental changes both by regulating celi production and cell death. Cells require lineage-specific growth factors for their birth, their proliferation, and for their survival. Planned studies on descent from altitude. With collaborators at the Institute of Altitude Studies, Universidad Cayetano Heredia in Lima, we are poised to begin studies designed to definitively prove the existence of neocytolysis and to elucidate the underlying mechanism. Individuals acclimated to 14,500 feet will have studies of RBCM, red cell survival and heme turnover in Cerro de Pasco, Peru. Cohorts of red cells of differing ages will be differentially labeled using ^{51}Cr, ^{13}C and ^{15}N, allowing direct determination of which red cells are later hemolyzed. Subjects will be transported to sea level where we will repeat measurements of RBCM, red cell survival and heme turnover. We expect to directly show that only the youngest red cells, less than 7 to 12 days old, will hemolyze. Some subjects will receive daily subcutaneous EPO injections on descent. We expect this to abrogate neocytolysis and prove the role of low EPO levels in initiating the process.

Wider implications. These studies have implications far beyond the unusual situations of descent from high altitude or spaceflight. As one example, neocytolysis should occur in athletes who try to enhance performance by "blood doping" administering supraphysiologic autologous red cell transfusions. Whenever new physiologic processes are defined, perturbations at various steps are soon appreciated which lead to disease. An example might be the congenital hemolytic anemia due to deficiency of pyruvate kinase where it is known that young red cells are selectively destroyed (10). It is likely that understanding and manipulating neocytolysis would allow fresh pathophysiologic thinking and novel therapeutic approaches to a variety of hematologic disorders, such as polycythemias.

The anemia of renal disease is another situation where EPO levels are low, and levels may be low enough in some patients to precipitate neocytolysis. Neocytolysis could contribute to the documented hemolytic component in some patients with the anemia of renal disease. Our theory predicts that currently widely used EPO dosing regimens of three intravenous boluses weekly are highly inefficient because peak levels would stimulate progenitors toward erythroid maturation but neocytolysis may occur at the EPO nadir. This may explain recent empiric observations that much lower doses of EPO are therapeutically effective when given daily subcutaneously (11). Subcutaneous regimens could result in substantial health dollar savings.

In summary our studies on the decline of RBCM with spaceflight led to the inescapable conclusion that adaptation occurred by the selective hemolysis of young red cells, a process we call neocytolysis. We theorized that this should occur as well when individuals acclimated to high altitude descend to sea level. We were surprised to uncover forgotten and ignored data from 45 years ago demonstrating hemolysis on descent. While neocytolysis was not considered by these investigators, it would best explain their data and would be the most efficient way for the body to adapt to acute plethora. Planned studies in Peru will prove and clarify the mechanisms underlying neocytolysis, including the possible role of sub-threshold EPO suppression. Understanding this process will permit a fresh look at hematologic disorders and their therapy.

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