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Importance of hemoglobin concentration to exercise: Acute manipulations☆

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Abstract

An acute reduction of blood hemoglobin concentration ([Hb]), even when the circulating blood volume is maintained, results in lower $\dot{V}_{O_2max}$ and endurance performance, due to the reduction of the oxygen carrying capacity of blood. Conversely, an increase of [Hb] is associated with enhanced $\dot{V}_{O_2max}$ and endurance capacity, that is also proportional to the increase in the oxygen carrying capacity of blood. The effects on endurance capacity appear more pronounced and prolonged than on $\dot{V}_{O_2max}$. During submaximal exercise, there is a tight coupling between O_2 demand and O_2 delivery, such that if [Hb] is acutely decreased muscle blood flow is increased proportionally and vice versa. During maximal exercise with either a small or a large muscle mass, neither peak cardiac output nor peak leg blood flow are affected by reduced [Hb]. An acute increase of [Hb] has no effect on maximal exercise capacity or $\dot{V}_{O_2peak}$ during exercise in acute hypoxia. Likewise, reducing [Hb] in altitude-acclimatized humans to pre-acclimatization values has no effect on $\dot{V}_{O_2peak}$ during exercise in hypoxia.

Keywords: Exercise; O_2 transport; Hemoglobin; Performance; Endurance; Hypoxia

1. Introduction

This review focuses on the effects that acute changes in blood hemoglobin may have on maximal aerobic power and endurance capacity. Despite the enormous literature on blood doping, few studies have provided the key data necessary to explain how the acute alteration of blood hemoglobin (Hb) concentration influences maximal aerobic power. Most of the studies have been performed using whole body

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exercise models, particularly exercise on the cycle ergometer and running. Less is known about the influence that changes in blood hemoglobin may have on peak aerobic power and endurance capacity during exercise with a small muscle mass. The latter, would allow a more clear separation between central and peripheral effects of blood Hb concentration on oxygen transport and utilization. The acute alteration in blood Hb concentration is also associated with compensatory changes in the cardiovascular and respiratory response to exercise that, in this review, are treated only briefly.

2. Acute reduction of hemoglobin concentration and exercise performance

The effect of a direct manipulation of the total amount of hemoglobin in healthy volunteers was first examined by Karpovich and Millman (1942), who showed that a reduction of total hemoglobin caused by bleeding decreased performance of athletes to a greater extent in long than in brief sport events. This finding was later confirmed by Balke et al. (1954), who observed a 9% decrease in maximal oxygen uptake ($\dot{V}_{O_2max}$), 1 h after a blood donation of 500 ml. However, pre-donation $\dot{V}_{O_2max}$ values were re-established 2–3 days later despite the fact that blood Hb concentration was still reduced by 8% (Balke et al., 1954). Studies carried out half a century ago by Astrand (1952) showed a close relationship between total Hb mass and $\dot{V}_{O_2max}$ such that the differences in maximal oxygen intake between adults and children and between men and women corresponded to differences in total hemoglobin. This relationship gave support to the idea that $\dot{V}_{O_2max}$ and endurance capacity depend on the oxygen carrying capacity of the blood. Nonetheless, the influence of blood Hb concentration on endurance remained unclear. Some authors reported that anemia had no influence on physical performance of women (Robbe, 1958; Cotes et al., 1969) or children (Hermansen, 1973). In contrast, the classic work of Sproule et al. (1960) found $\dot{V}_{O_2max}$ to be about 40% lower in 11 anemic patients (mean Hb concentration of 70 g l^{-1}) than it was in normal subjects, while cardiac output at exhaustion was similar in both groups. In theory, in chronic anemia, particularly in iron-deficiency anemia, endurance capacity may be independently reduced by other factors in addition to the low blood Hb concentration, due to the important role played by iron in several iron-containing proteins involved in the transport and utilization of O_2 (Dallman, 1986). However, Celsing et al. (1986) showed that in healthy subjects, inducing iron deficiency by repeated venesections during a period of 8–11 weeks did not affect endurance when transfusion of red blood cells was performed in order to exclude the influence of a low Hb concentration. In agreement, oral iron supplementation did not enhance blood Hb concentration, $\dot{V}_{O_2max}$, Wingate test performance or the exercise intensity corresponding to the ventilatory threshold in non-anemic women which serum ferritin value rose from a mean of 12.4 ± 4.5 to 37.7 ± 19.7 ng ml^{-1} (Newhouse et al., 1989). Although in patients with severe iron-deficiency anemia, the reduction in maximal exercise capacity may be explained in a large part by the reduction of blood Hb (Edgerton et al., 1981), a contribution by other non-Hb related biochemical changes is also likely.

Few investigators, however, have directly addressed the effect of a reduction in Hb concentration on $\dot{V}_{O_2max}$ and performance (Ekblom et al., 1972, 1976; Woodson et al., 1978). Ekblom et al. (1972) showed that 2 days after the withdrawal of 800 ml of whole blood, Hb concentration was lowered by 13% resulting in a 10% reduction of $\dot{V}_{O_2max}$, while endurance time was reduced by 30%. However, 2 weeks after the venesection, when blood Hb concentration was still reduced by 10%, $\dot{V}_{O_2max}$ was already recovered. Endurance time was normalized in one subject while it remained reduced by 13% in the two other subjects, included in that study (Ekblom et al., 1972). In a subsequent study, they observed that 48 h after the withdrawal of 800 ml of blood hemoglobin was reduced by 12% and $\dot{V}_{O_2max}$ and endurance time by 6 and 20–25%, respectively (Ekblom et al., 1976). In a group of four subjects, similar results were obtained by Woodson et al. (1978) who observed that a 34% acute isovolemic reduction of blood Hb concentration was associated with a 16% decline in $\dot{V}_{O_2max}$. The authors maintained the subjects chronically anemic during 2 weeks by withdrawing 50–100 ml of blood every 1–2 days. Strikingly, after these 2 weeks, the $\dot{V}_{O_2max}$ was further reduced (30% less than before the withdrawal of blood) despite that the Hb concentration remained at

the same level as in the acute manipulation. In contrast, Rowell et al. (1964) reported that repeated phlebotomies (in total 700–1000 ml) over 5 days, resulting in a 14% lower Hb concentration had no significant effect on $\dot{V}_{O_2max}$, measured 3 days after the last phlebotomy, i.e. 8 days after the start of the sequential bleeding.

In general, the studies mentioned above show that an acute reduction of blood Hb concentration has a pronounced effect on $\dot{V}_{O_2max}$ and endurance which is proportional to the reduction of the oxygen carrying capacity of blood (Fig. 1). One to two weeks after the acute reduction of blood, Hb concentration $\dot{V}_{O_2max}$ is almost restored to normal in some instances, while endurance time is only partly restored, despite Hb concentration being still reduced. Ekblom et al. (1972) also studied four additional subjects, who were submitted to three consecutive venesections with an interval of 4 days between each. Blood Hb concentration, 3 days after the venesections, was reduced by 10, 15 and 18%, while corresponding $\dot{V}_{O_2max}$ values were reduced by 6, 10 and 16%. Endurance times were also decreased by 13, 21 and 30%, after the withdrawal of 400, 800 and 1200 ml of whole blood, respectively. In a follow up study, Kanstrup and Ekblom (1984) observed in five subjects that the day after removing 900 ml of blood, blood Hb concentration was lowered by 11%, $\dot{V}_{O_2max}$ by 9% and endurance time at the intensity eliciting $\dot{V}_{O_2max}$ was reduced by 40%. Thus, Ekblom and co-workers studies showed very clearly that the acute reduction of blood Hb mass and blood Hb concentration causes an acute reduction of $\dot{V}_{O_2max}$ and endurance times, the effect being more accentuated on endurance time than on $\dot{V}_{O_2max}$.

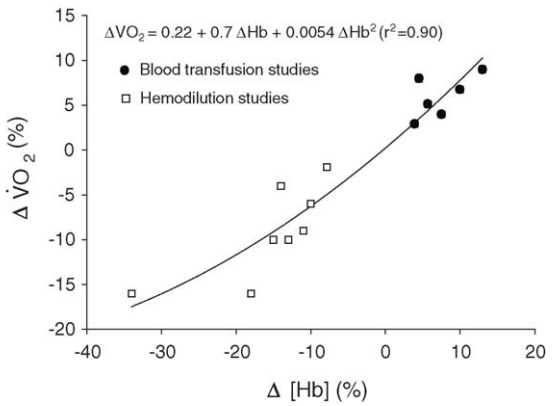

Fig. 1. Relationship between the change (%) in hemoglobin concentration ([Hb]) and the corresponding change (%) in $\dot{V}_{O_2max}$. Each point represents the mean value of one study. Only data obtained during the first 48 h after the manipulation of [Hb] have been included. Drawn using data from Refs.: Balke et al. (1954), Rowell et al. (1964), Ekblom et al. (1972, 1976), Woodson et al. (1978), Buick et al. (1980), Kanstrup and Ekblom (1984), Spriet et al. (1986) and Turner et al. (1993).

Bloodletting causes initially a reduction of blood volume that is progressively compensated for in the following hours by a progressive expansion of the plasma volume. Twenty-four to forty-eight hours after the withdrawal of blood, the blood volume is almost normal (around 4–6% less than before the intervention), while the concentration of hemoglobin is reduced by hemodilution (Ekblom et al., 1976; Kanstrup and Ekblom, 1984). This means that the effect of blood withdrawal on exercise capacity may be mediated by two mechanisms. The main mechanism is the reduction of blood Hb concentration which is associated with a reduction of Ca_{O_2}. The second mechanism is the reduction of circulating blood volume, which could affect venous return, lowering heart filling pressures, and hence, maximal cardiac output. However, the impact that such a small reduction in circulating volume could have on maximal cardiac output and $\dot{V}_{O_2max}$ is likely negligible (Saltin, 1964; Fogelholm, 1994; Moquin and Mazzeo, 2000).

In the studies by Ekblom et al., the diminution of $\dot{V}_{O_2max}$ by blood withdrawal could be explained by a decreased maximal O_2 delivery, since maximal cardiac output could not be increased to offset the fall in Ca_{O_2} (Ekblom et al., 1976). The inability to increase cardiac output above maximal levels during whole body exercise to compensate for decreases in Ca_{O_2} has also been reported when Ca_{O_2} has been lowered by carbon monoxide breathing (Stenberg et al., 1966; Ekblom and Huot, 1972; Ekblom et al., 1975), as well as during exercise in acute hypoxia (Stenberg et al., 1966; Hartley et al., 1973; Calbet et al., 2003a).

2.1. Influence of active muscle mass

Saltin et al. (1986) were the first to study the regional blood flow responses at different exercise intensities in subjects with normal and low Hb concentrations. Their study suggested that low Ca_{O_2} may be offset at submaximal exercise by an increase in muscle blood

flow, so that O_2 delivery is maintained. Their findings at peak exercise, however, were inconclusive. To specifically address the effect of isovolemic anemia during submaximal and maximal exercise, we measured cardiac output and leg blood flow in seven subjects having a normal Hb concentration and a second time after withdrawing about 1 l of whole blood and substituting it with an equal amount of a 5% human albumin solution to reduce Hb concentration by ~20% (from 144 ± 4 to $115 \pm 2\,g\,l^{-1}$). The two-legged knee-extension model was used to avoid limiting the capacity of the heart and allow an increase in cardiac output, if needed, to elevate leg blood flow even at the highest workloads. Isovolemic anemia resulted in a 17% reduction of whole body $\dot{V}_{O_2peak}$ and power output and a 20% lower leg $\dot{V}_{O_2peak}$. This means that $\dot{V}_{O_2peak}$ was reduced almost in the same proportion as blood Hb concentration was reduced. During submaximal exercise, leg blood flow was increased to compensate for the reduction of blood Hb concentration. However, despite that during the control condition leg extension exercise only elicited a peak cardiac output of ~20 l/min, i.e. ~80% of the value reached during an incremental exercise to exhaustion, neither the peak cardiac output nor the peak leg blood flow were increased to compensate for the reduction in oxygen carrying capacity of blood during the maximal exercise in the anemic condition. Consequently, a similar O_2 delivery was achieved during submaximal exercise, but not at maximal exercise, where neither leg blood flow, cardiac output, nor O_2 extraction compensated for a low Hb concentration, resulting in reduced maximal power output and $\dot{V}_{O_2}$. In addition, the blood lactate accumulation curve was steepest after hemodilution, meaning that for a given exercise intensity blood lactate accumulation in blood was greater after hemodilution (Fig. 2). Although in these studies (Koskolou et al., 1997b; Roach et al., 1999), endurance capacity was not directly measured, the facts peak leg $\dot{V}_{O_2}$, the intensity achieved at peak exercise and the higher accumulation of blood lactate during the condition with reduced blood Hb concentration strongly suggest that endurance capacity is also reduced during exercise with a small muscle mass.

The reason why during exercise with a small muscle mass peak blood flow is not increased to compensate for the reduction of blood Hb concentration is not known. Since peak leg blood flow during knee-extension exercise was not increased even when acute hypoxia was added to the anemic condition (Roach et al., 1999), one possible explanation is that during exercise with small muscle mass, peak levels of perfusion are already reached during exercise in normal conditions, leaving little room for further enhancement of perfusion (Calbet, 2000).

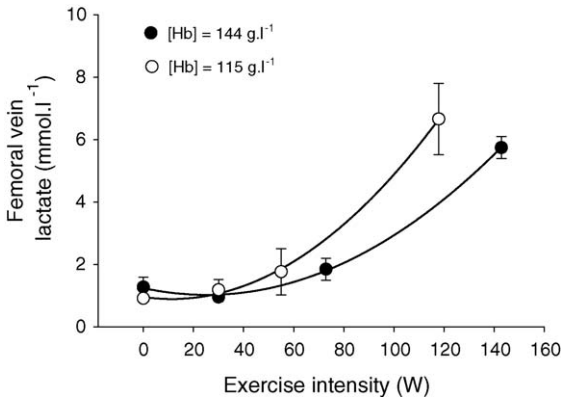

Fig. 2. Blood lactate concentration in the femoral vein during incremental exercise to exhaustion with both legs in the leg extension ergometer. The black circles represent the control condition and the white circles represent the response after isovolemic hemodilution which resulted in 20% lower blood Hb concentration. A detailed description of the procedures may be found in Koskolou et al. (1997b).

2.2. Acute reduction of hemoglobin concentration and exercise at altitude

One of the main adaptations to chronic hypoxia is the increase of blood Hb concentration. During the first 24–48 h of exposure to altitude, Hb concentration is increased due to a reduction of plasma volume. The latter is caused by an enhancement of diuresis and displacement of water from the vascular to the intracellular and extravascular spaces. In addition, at altitude water is lost via sweating and increased ventilation due to the lower environmental humidity. In fact, altitude natives living at 4330 m may have chronically reduced plasma volumes compared to the values observed in natives living at sea level (Sanchez et al., 1970). This relatively rapid development of hemoconcentration allows for an elevation of the arterial Ca_{O_2} to values close to those observed at sea level in 48 h, but more than 3–4 weeks of residence at an altitude above 2500–3000 m are necessary to detect a significant increase in blood cell mass

(Reynafarje et al., 1959; Weil et al., 1968; Hannon et al., 1969).

Accordingly, 9 weeks of residence at 5260 m resulted in an enhancement of resting Ca_{O_2} to values that were 21% higher and blood Hb concentration 36% higher than before the exposure to altitude (Calbet, 2003). This high Hb concentration resulting from altitude acclimatization has the benefit of allowing resting O_2 delivery to reach values similar to those observed at sea level without necessitating a higher cardiac output (Calbet, 2003). However during maximal exercise on the cycle ergometer, systemic O_2 delivery was 10% lower than during maximal exercise at sea level, due to a reduction of peak cardiac output during exercise in hypoxia (Calbet et al., 2002). Although the increase of blood hemoglobin almost restored maximal sea level O_2 transport values, $\dot{V}_{O_2peak}$ was only partly improved and remained 30% below the values observed in normoxic conditions after 9 weeks of acclimatization to 5260 m (Calbet et al., 2003b). This implies that part of the extra systemic O_2 delivery gained with acclimatization is not made available to the exercising muscle during a maximal effort (Calbet et al., 2003b). In addition, it has been questioned whether the polycythemia of altitude is of adaptive value or not. In Tibetan high altitude natives (Beall et al., 1998) and in some high altitude mammals such as the llama (Weiser et al., 1992), all of which have resided at altitude for many generations, Hb concentration is only slightly higher than sea level values. In fact, it has been shown that hemodilution at altitude, in contrast to what is observed at sea level, does not reduce $\dot{V}_{O_2peak}$ during upright exercise on the cycle ergometer in hypoxia (Sarnquist et al., 1986; Schaffartzik et al., 1993; Calbet et al., 2002) or during one leg knee-extension exercise (Saltin, Radegran, Boushel, Calbet, unpublished).

Sarnquist et al. (1986) reported similar performance and $\dot{V}_{O_2peak}$ during exercise at altitude after isovolemic hemodilution in four experienced climbers whose hematocrits were lowered from 58 to 51%. Schaffartzik et al. (1993) observed similar whole body $\dot{V}_{O_2peak}$, leg $\dot{V}_{O_2peak}$ and exercise capacity during maximal exercise in hypoxia before and after reducing (isovolemically) blood Hb concentration by 13% in humans acclimatized to 3800 m (Schaffartzik et al., 1993). Leg blood flow and cardiac output responses at peak exercise were similar at high and low Hb levels during both normoxia and hypoxia. However, when Ca_{O_2} was reduced by decreasing the Hb concentration of blood, muscle O_2 extraction rather than blood flow increased to maintain muscle $\dot{V}_{O_2}$ (Schaffartzik et al., 1993). Calbet et al. (2002) reported no effect on $\dot{V}_{O_2peak}$ in nine subjects that resided for 9 weeks at 5260 m when their blood Hb concentration was reduced isovolemically by as much as 20%, i.e. to the values observed before the acclimatization to altitude (Calbet et al., 2002). Although we do not have a definitive explanation for mechanism/s accounting for the lack of effect of moderate hemodilution on $\dot{V}_{O_2peak}$ in chronic hypoxia, we have observed that in chronic hypoxia the maximal blood flow to the active muscle fibers is lower than in normoxia (Calbet et al., 2003b), due to two main factors. First, due to a lower maximal cardiac output and second, because in chronic hypoxia the fraction of the cardiac output directed to the active muscles is reduced. Isovolemic and hypervolemic hemodilution are compensated in chronic hypoxia, in part by increasing skeletal muscle perfusion (Calbet et al., 2003b, 2004), a mechanism that is not available in acute hypoxia (Calbet et al., 2003a), because during maximal exercise in acute hypoxia there is no functional reserve available to deviate a higher fraction of cardiac output to the active muscles.

In contrast to Schaffartzik et al. (1993) and Calbet et al. (2002), Horstman et al. (1980) observed that reducing Ca_{O_2} by removing 450 ml of blood, i.e. by reducing the Ca_{O_2} of maximal exercise by 14%, in four subjects after 3 weeks of acclimatization to 4300 m, resulted in 8% lower $\dot{V}_{O_2peak}$ and 35% lower endurance time during prolonged exercise to exhaustion at an intensity close to 80% of $\dot{V}_{O_2peak}$ observed at altitude before the removal of blood.

The differences between the study of Horstman et al. (1980) and others (Sarnquist et al., 1986; Schaffartzik et al., 1993; Calbet et al., 2002) could be explained by the fact that Horstman et al. performed the withdrawal of blood 1 day prior to the exercise test using a Ringer lactate solution to replace the volume of blood withdrawn. Thus, it cannot be ruled out that the procedures applied by Horstman et al. (1980) also caused some degree of hypovolemia. Another fact that could explain the differences between studies is the exercise protocol used to assess $\dot{V}_{O_2peak}$ in Horstman's et al. (1980) study, which consisted of four runs at constant intensity for 4–6 min duration with 20 min rest periods in between.

2.2.1. Does the altitude acclimatization-induced higher blood hemoglobin concentration confer any physiological advantage for exercise at altitude?

It is known that a higher blood Hb concentration may enhance endurance capacity at sea level (Ekblom et al., 1972; Buick et al., 1980) and at altitude (Horstman et al., 1980). It has also been shown that isovolemic hemodilution at altitude was associated with a greater blood lactate concentration, lactate release, heart rate and catecholamine concentrations during submaximal exercise strongly indicating that hemodilution decreases endurance capacity at altitude (Calbet et al., 2002). Another possible advantage conferred by the increase in blood Hb concentration with altitude acclimatization is a greater O_2 delivery to tissues other than the contracting skeletal muscles at maximal exercise, particularly the brain and may be the respiratory muscles (see Dempsey et al., in this issue of the journal). However, the fact that $\dot{V}_{O_2peak}$ under hypoxic conditions was not reduced by hemodilution (Calbet et al., 2002) suggests that the increase of blood Hb concentration elicited by chronic hypoxia is not actually mandatory to preserve brain oxygenation in the acclimatized human performing upright whole body exercise at $\dot{V}_{O_2peak}$. A better oxygenation of the respiratory muscles during exercise in chronic hypoxia with a high blood Hb concentration could attenuate fatigue eliciting afferent feedback prolonging endurance time.

Since hemoglobin is an excellent buffer, an increase of the circulating Hb mass contributes to enhance the blood buffering capacity (Cerretelli and Samaja, 2003). A higher blood buffering capacity could facilitate lactate and proton release from the active muscle and enhance the capacity to produce energy via the anaerobic pathways. However, there is no experimental indication that anaerobic energy release is reduced by a reduction of blood Hb concentration up to 20% (Calbet et al., 2002) nor increased with autologous transfusion of blood (Ekblom et al., 1972).

3. Acute increase of hemoglobin concentration and exercise

The influence of an increase in blood Hb concentration on the cardiorespiratory response to exercise was first studied by Pace et al. (1947) in five subjects under conditions of normal hematocrit (46%) and after the transfusion of 1000 ml of red cells resulting in an elevation of hematocrit to 58%. After the blood transfusion, heart rate during submaximal exercise declined in direct proportion to the increase in Ca_{O_2} (Pace et al., 1947), suggesting a lower relative effort to sustain the exercise. In the 1960s and 1970s, new blood transfusion experiments to explore the influence of blood volume and Hb concentration on the hemodynamics during submaximal and maximal exercise were carried out (Robinson et al., 1966; Ekblom et al., 1972, 1975, 1976; Buick et al., 1980; Spriet et al., 1986).

Robinson et al. (1966) observed that the expansion of blood volume by the autologous transfusion of 1000–1200 ml of whole blood decreased maximal heart rate and augmented maximal stroke volume, while maximal cardiac output and $\dot{V}_{O_2max}$ remained unchanged. The lack of effect on $\dot{V}_{O_2max}$ could be due to the fact that the transfusion of blood resulted in only a small increment of the hematocrit (from 42 to 44%).

Ekblom et al. (1972), on the other hand, showed a 9 and 23% increase of $\dot{V}_{O_2max}$ and endurance time, respectively, after increasing hemoglobin by 13% by re-infusion of 800–1200 ml of the subjects' own blood that was withdrawn 4 weeks before the experiment. The maximal values of cardiac output, heart rate and stroke volume were similar regardless of Hb concentration, indicating that the increase in $\dot{V}_{O_2max}$ was possibly due to the elevation of maximal O_2 delivery owing to the increase in Ca_{O_2} produced by the blood re-infusion. Subsequent work by Ekblom et al. (1976) provided further evidence for a tight coupling between $\dot{V}_{O_2max}$ and maximal O_2 delivery, since changes in $\dot{V}_{O_2max}$ correlated closely with changes in Hb concentration ($r = 0.97$). Similar responses were reported when Ca_{O_2} was increased by breathing hyperoxic gases (Asmussen and Nielsen, 1955; Ekblom et al., 1975). Later studies have confirmed since then the enhancing effect of an acute increase of blood Hb concentration on performance, as reflected by either a longer endurance time at a given exercise intensity (Buick et al., 1980) or a greater mean power output during a laboratory time trial (Turner et al., 1993), a higher mean speed to cover a given distance (Williams et al., 1981; Brien and Simon, 1987) or lower blood lactate concentration at a given absolute exercise intensity (Spriet et al., 1986). Buick et al. (1980) studied highly trained athletes (mean $\dot{V}_{O_2max}$ 80 ml kg^{-1} min^{-1}) who responded with marked improvements in endurance time 24 h and

7 days after the transfusion of two units of erythrocytes. However, $\dot{V}_{O_2max}$ remained elevated 16 weeks after the transfusion by the same amount as in the tests performed at 24 h and 7 days, despite the fact that after 16 weeks blood Hb concentration had returned to control levels.

In contrast with the performance enhancing effects of an increase of blood Hb concentration at sea level, Young et al. (1996) did not find any significant effect of 700 ml of autologous erythrocyte infusion performed 24 h before the ascent to Pikes peak (4300 m), on the $\dot{V}_{O_2max}$ response 24 h and 9 days after the ascension. In agreement, Pandolf et al. (1998) showed that the administration of 2 units of packed red cells 24 h before the ascent to Pikes peak (4300 m above sea level) produced a small but statistically non-significant improvement in 3.2 km run performance measured after 3 and 14 days of residence at altitude.

It remains to be determined what is the effect of an acute increase of blood Hb concentration on skeletal muscle blood flow. The data available from chronic hypoxia studies suggest that a large increase of blood Hb concentration could be associated with a reduction of peak leg blood flow during exercise (Calbet et al., 2003b). In addition, it is not known whether an acute increase of blood Hb concentration could improve endurance capacity during exercise in acute hypoxia.

4. Hemoglobin concentration and the regulation of blood flow during exercise

In a series of studies carried out by Saltin's group, the effects of Pa_{O_2} and Ca_{O_2} on ventilatory and cardiovascular responses to exercise in humans were determined over wide ranges of Pa_{O_2} and Ca_{O_2} combining several exercise models and intensities (Koskolou et al., 1997a,b; Roach et al., 1999; Gonzalez-Alonso et al., 2001; Calbet et al., 2002, 2003b, 2004). These studies show that Pa_{O_2} alone has a minor role in the regulation of vascular tone in exercising human muscles since similar leg blood flows are observed during submaximal exercise in conditions with nearly identical Ca_{O_2} but widely different Pa_{O_2} ranging from 30 to more than 500 mmHg. However, the mechanism by which changes in Ca_{O_2} elicit the appropriate modification in vascular tone remain unknown. In this context, the Hb molecule has been proposed as an ideal O_2 sensor (Saltin et al., 1986; Calbet, 2000; Singel and Stamler, 2005). The O_2 sensing properties of the Hb molecule appear to involve conformational changes resulting from O_2 desaturation (Buehler and Alayash, 2004). On desaturation, the red blood cells could elicit vasodilation through three independent mechanisms: release of adenosine 5′-triphosphate (ATP) (Ellsworth, 2004), release of *S*-nitrosylated molecules originally bound to β93 cysteine residues of oxyhemoglobin (Jia et al., 1996) and nitrite uptake by the red blood cells and conversion to nitric oxide via the reaction with Hb at the site of ferric (Fe^{3+}) and ferrous (Fe^{2+}) hemoglobin (Cosby et al., 2003; Nagababu et al., 2003). It remains to be established how changes in blood Hb concentration may influence the O_2 sensing capabilities of the Hb molecule in different organs and tissues, particularly in the skeletal muscle, the brain and the lungs during large changes in HbO_2. Understanding these processes may help to unravel, for example, why in chronic hypoxia a lower proportion of the overall O_2 transport capacity is made available to the exercising muscles (Calbet et al., 2003b) and why $\dot{V}_{O_2max}$ appears to be insensitive to variations on blood Hb concentrations of up to 20% during exercise in chronic hypoxia.

In summary, an acute reduction of blood Hb concentration, even when the circulating volume is maintained at the same level, results in a reduction of $\dot{V}_{O_2max}$ and endurance performance, that can be explained by the reduction of the oxygen carrying capacity of blood. Conversely, an increase of blood Hb concentration is associated with an improvement of $\dot{V}_{O_2max}$ and endurance capacity, that is also proportional to the increase in the oxygen carrying capacity of blood. Since these conclusions have been drawn using a rather specific population (in general, young physically active males and in some cases elite athletes), it remains to be determined whether similar effects would be observed in women, children, the elderly and subjects with very low physical fitness. The observation made in some instances regarding the lack of effect of blood removal on $\dot{V}_{O_2max}$ and that normal $\dot{V}_{O_2max}$ values may be achieved before full recovery of the normal Hb concentration also deserve further attention. It is not known if changes in blood Hb concentration may also alter endurance performance during exercise with a small muscle mass. It remains also to be elucidated what is the effect of an acute increase

of blood Hb concentration on endurance capacity at altitude.

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