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Dependency of the O_2 consumption/O_2 transport relationship on amino acid supply in sepsis

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Summary. In sepsis tissue O_2 uptake may be abnormally limited because of a depressed O_2 consumption/O_2 transport relationship. This study has been performed to assess patterns of O_2 consumption, CO_2 production and O_2 transport in septic patients undergoing total parenteral nutrition; more in particular, this study has investigated the interdependence between the patterns of blood O_2 uptake and simultaneous CO_2 release, and the availability of substrates (amino acids, glucose and fat). It has been shown that the O_2 consumption/O_2 transport relationship is significantly influenced by the exogenous amino acid load, which tends to increase O_2 uptake and O_2 consumption at any given O_2 transport, thus suggesting a favourable effect of amino acid administration on energy metabolism. The data on CO_2 production and CO_2 release, in addition to reconfirming the results of previous studies, have shown that the changes in O_2 uptake and in CO_2 production mediated by substrate doses have a quantifiable impact on blood O_2–CO_2 exchange interactions.

Keywords: Amino acids – Sepsis – O_2 transport – O_2 consumption – CO_2 exchange – Metabolism

Introduction

Septic metabolic disregulation is characterized by an impairment in the O_2 consumption/O_2 transport relationship with abnormally low O_2 uptake in spite of adequate or higher than normal O_2 transport. In addition, it is characterized by signs of impaired energy metabolism with progression of abnormalities in substrate utilization, such as intolerance to exogenous glucose, abnormalities in fat metabolism and preferential utilization of amino acids (Siegel et al., 1967, 1979; Cerra et al., 1980; Giovannini et al., 1983). In this study, the relationship

between O_2 consumption, CO_2 production and O_2 transport has been analyzed in a group of septic patients undergoing total parenteral nutrition with glucose, fat and amino acids. The analysis has addressed in particular the effect of substrate infusions on the O_2 consumption/O_2 transport relationship, and on patterns of CO_2 production and exchange, to assess any dependency of the impaired septic O_2 uptake on substrate availability and the simultaneous effect of substrates on blood O_2–CO_2 exchange interactions.

Material and methods

Two hundred-sixtyfive measurements of O_2 consumption and O_2 transport were performed in 82 septic patients (62 males and 20 females) undergoing total parenteral nutrition with glucose, fat and amino acids. Age was 53 ± 17 years (mean ± SD), height 167.5 ± 8.0 cm, weight 64.4 ± 12.9 Kg, ratio of actual to ideal body weight 1.04 ± 0.18 (Metropolitan, 1984), and estimated lean body mass 47.9 ± 6.5 Kg (Hume, 1966). The diagnosis of sepsis was based on the presence of positive blood cultures, or positive cultures from surgical drainage of infected processes (Giovannini et al., 1983). Sixtyfive patients had diffuse peritonitis, the remainder had localized intraabdominal abscesses, or severe biliary or soft tissue infections. All of them were critically ill and were showing signs of metabolic abnormality such as mental confusion, rapid body muscle wasting, a tendency to hyperglycemia, hypoalbuminemia, hypocholesterolemia, high blood urea nitrogen without renal failure and increased urinary urea excretion; however, they were studied at an early and physiologically compensated stage of disease (median distance from the onset of sepsis = 7 days; median sepsis severity score = 15) (Elebute and Stoner, 1983), and were not in multiple organ failure.

O_2 consumption (VO_2, ml/Kg/min) was measured according to a previously developed method (Comroe et al., 1962; Giovannini et al., 1984, 1989a) from the formula:

$$VO_2 = (VE)(F_{ATPS-STPD})\left[\frac{FIO_2(1 - FECO_2) - FEO_2}{1 - FIO_2}\right]$$

in which VE is expired minute volume (ml/Kg/min), FIO_2 is the inspired O_2 concentration fraction, FEO_2 and $FECO_2$ are the mean expired O_2 and CO_2 concentration fractions, respectively, and $F_{ATPS-STPD}$ is the conversion factor from ambient to standard conditions (Comroe et al., 1962). Arterial and central venous O_2 concentrations were determined from the blood O_2 tensions and saturations (measured on a IL 1302 pH-Gas Analyzer and on a IL 282 Co-Oximeter together with hemoglobin concentration) by using a Hufner and Bunsen coefficients for O_2 of 1.36 ml/g and 0.003 ml/100 ml/mmHg, respectively; these were used to obtain arterio-venous O_2 concentration difference (a–vDO_2, ml/100 ml), and to calculate O_2 transport (O_2T, ml/Kg/min) from the product of cardiac output (obtained by the Fick method) times arterial O_2 concentration (Comroe et al., 1962). In patients breathing at increased inspired O_2 concentrations, the respiratory quotient was simultaneously determined from arterial and central venous blood gases and from expired gases in order to test the accuracy of the O_2 consumption measurements (Comroe et al., 1962; Giovannini et al., 1993).

Total CO_2 production (VCO_2, ml/Kg/min) was determined from the product (VE) $(F_{ATPS-STPD})(FECO_2)$ (Comroe et al., 1962). Venoarterial CO_2 concentration difference (v–aDCO_2, ml/100 ml) and the fraction of v–aDCO_2 which was involved in the Haldane effect (or O_2-linked CO_2 exchange) were calculated from blood gas data by a recently developed model (Giovannini et al., 1993). This allowed to quantify the amount of totally produced CO_2 which was transported and exchanged on the basis of the Haldane effect (VCO_2–H, ml/Kg/min) separately from the remainder (VCO_2–NH, ml/Kg/min).

Total parenteral nutrition was based on amino acid (Solamin Pierrel, Table 1) and glucose solutions, which were infused at constant rates over any 24hr period, and on a supplemental fat emulsion (Intralipid Kabi), which was infused over 6 to 8 hours periods.

Table 1. Composition of Solamin Pierrel amino acid solution (g/100 g amino acids)

Aspartic acid	1.6	Isoleucine	5.6
Threonine	4.0	Leucine	8.8
Serine	2.0	Tyrosine	0.6
Glutamic acid	1.0	Phenylalanine	8.8
Proline	2.0	Tryptophane	2.0
Glycine	17.0	Lysine	6.4
Alanine	2.0	Histidine	9.0
Valine	6.4	Arginine	14.0
Methionine	8.8		

Amino acid dose was 1.10 ± 0.56 g/Kg/24hr, glucose dose 5.49 ± 2.72 g/Kg/24hr and fat dose 1.06 ± 0.48 g/Kg/24hr (means ± SD). Doses of glucose, amino acids and fat were decided on a clinical basis. Doses of glucose and amino acids could change independently in different days and in different patients; when such changes occurred, the measurements were performed at least 6 hours after the new infusion had been started; with reference to the infusion of fat, the measurements were performed either before starting it or at least an hour after the infusion had been started.

Analysis and validation of the results was performed by using least squares multiple regression analysis with skewness and kurtosis control and best-fit procedures (Scheffé) (Seber, 1977). Regression analysis was used to assess the effects of doses of substrates (considered as independent variables) on metabolic variables and O_2T, and to assess the relationships with VCO_2–H and VCO_2–NH. Since the infusion of fat took place intermittently (and thus its dose could not be considered as a continuously distributed variable), its effect was assessed from changes in regression intercept for measurements done before or during the infusion of fat (Giovannini et al., 1989a). The distribution of variables was assessed by skewness and kurtosis control. A "best-fit" procedure was used for selecting functions of the independent variables which were yielding in each regression the more adequate fit to the data, based on the value of r^2 (coefficient of determination), Mallow's Cp and distribution of residuals (Seber, 1977).

Results

O_2 consumption (VO_2, ml/Kg/min) for all measurements was 4.15 ± 1.25, O_2 transport (O_2T, ml/Kg/min) was 15.18 ± 6.97, and arterio-venous O_2 concentration difference (a–vDO_2, ml/100 ml) was 4.00 ± 1.30. Regression analysis showed that VO_2 was directly related to O_2T, and that this relationship was significantly influenced by the amino acid dose (AAD, g/Kg/24hr):

$$VO_2 = 1.84(\log^* O_2T) + 0.64(AAD) - 1.33 \quad (1)$$

*(natural logarithm, best fit) $r^2 = 0.50$, $p < 0.001$ (Fig. 1).

O_2T accounted for 40% of the variability of VO_2 (partial $r^2 = 0.40$, $p < 0.001$), and AAD for an additional 10% (total $r^2 = 0.50$, $p < 0.001$). Inclusion in the regression of glucose dose (GD, g/Kg/24hr) and of fat infusion as independent variables did not increase the r^2 and did not contribute to increase the explained variability of VO_2.

The separate effects of individual substrates on the absolute values of VO_2 and O_2T were also assessed. It was reconfirmed that AAD was a significant

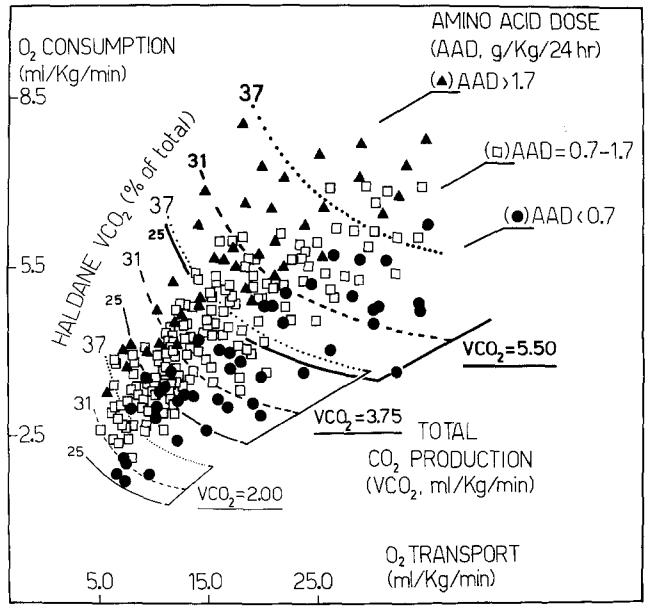

Fig. 1. Increase in O_2 consumption, at any given O_2 transport, with increasing amino acid dose. Simultaneous changes in the percent CO_2 production involved in the Haldane effect, at any given total CO_2 production, are also indicated (curves calculated from regression 5 in the text; curves extend beyond the distribution of measurements only to ease collocation of the scales)

determinant of VO_2:

$$VO_2 = 0.51(AAD) + 3.59 \qquad r^2 = 0.11,\ p < 0.001 \tag{2}$$

with the 95% confidence limits for slope and intercept being ± 0.30 and ± 0.61, respectively. By taking into account the difference in units (VO_2 = ml/Kg/min, AAD = g/Kg/24hr), the slope corresponded to an incremental ratio of 734 ml O_2/g amino acid ($p < 0.001$, 95% confidence limits = ± 431). The effect of GD on VO_2, when GD was included in regression 2 as an independent variable, was only of near borderline significance ($p = 0.058$), and the effect of fat was not significant.

On the contrary, O_2T was significantly related only to GD:

$$O_2T = 0.59(GD) + 11.93 \qquad r^2 = 0.12,\ p < 0.001 \tag{3}$$

with the 95% confidence limits for slope and intercept being ± 0.42 and ± 3.48, respectively. By taking into account the difference in units (O_2T = ml/Kg/min, GD = g/Kg/24hr), the slope corresponded to a mean incremental ratio of 852 ml O_2/g glucose ($p < 0.001$, 95% confidence limits = ± 604). The effects of AAD and of fat infusion, when included as independent variables in regression 3, were not significant.

These patterns, evidenced by using least squares regressions on the whole sample of measurements, were also reconfirmed by analyzing sequential measurements in individual patients. Twelve cases, in which AAD was greater than +2.0 SD from the mean dose (i.e., greater than 2.22 g/Kg/24hr), were also evaluated separately: in these, VO_2 and O_2T were 4.94 ± 1.28 and 14.81 ± 6.95,

respectively, and a–vDO_2 was 4.64 ± 1.08, thus reconfirming the tendency to maintain a higher O_2 uptake and VO_2, at any given O_2T, for the patients who were receiving the higher AAD.

VO_2 was the main determinant of CO_2 production ($VCO_2 = 3.76 \pm 1.11$ ml/Kg/min) and explained 52% of its variability ($r^2 = 0.52$, $p < 0.001$). Their relationship was not significantly dependent on the level of O_2T ($p > 0.05$). At any given VO_2, however, VCO_2 was directly and significantly related to GD, which explained an additional 10% of the variability of VCO_2 (thus raising the total r^2 to 0.62), and not significantly related to AAD or to fat infusion:

$$VCO_2 = 0.540(VO_2) + 0.125(GD) + 0.836 \qquad r^2 = 0.62,\ p < 0.001 \qquad (4)$$

The AAD-mediated increases in O_2 uptake and VO_2 at any given O_2T, and the GD-mediated increase in VCO_2 at any given VO_2, were associated with changes in blood O_2 and CO_2 transport and exchange interactions. The amount of totally produced CO_2 which was transported and exchanged on the basis of the Haldane effect (VCO_2–H = 1.17 ± 0.37 ml/Kg/min) was quantified separately from the remainder (VCO_2–NH = 2.59 ± 0.92 ml/Kg/min) (Giovannini et al., 1993). VCO_2–H, as a percent of total VCO_2 (mean VCO_2–H/VCO_2 = 31%), was found to increase significantly with the amino acid-mediated increase in VO_2 at any given O_2T ($p < 0.001$), and to decrease significantly with the glucose-mediated increase in VCO_2 ($p < 0.001$): such changes were explained by the relationship binding VCO_2–H, O_2T, VO_2 and total VCO_2 (Fig. 1):

$$VCO_2\text{–H}(\%\text{ of total}) = -VCO_2\,[17.85/(VO_2) + 40.46/(O_2T)] + 60.45$$

$$r^2 = 0.80,\ p < 0.001 \qquad (5)$$

Re-analysis of regression 4, by accounting separately for the VCO_2–H and VCO_2–NH components, also showed that VCO_2–H was equal to 0.276 (VO_2), with rather strict 95% confidence limits (= ±0.02), and showed that most of the variability of VCO_2 and the effect of GD in increasing VCO_2 were related to changes in VCO_2–NH.

Discussion

The most important physiologic determinant of tissue O_2 uptake is O_2 transport (O_2T). O_2T may become abnormally low (and tissue O_2 uptake may thus be impaired) as a consequence of low blood flow, anemia, hypoxemia, or a combination of these; for comparably low O_2T, anemia and low blood flow also limit O_2 diffusion from blood to tissues, thus causing a greater impairment in O_2 uptake than hypoxemia. In normal or high O_2T conditions, such as in hyperdynamic sepsis, a defect in O_2 uptake may also paradoxically be present; this is thought to be caused by the opening of anatomical shunts in the peripheral circulation, or by the development of a "functional" shunt (that is, by the unability of tissue cells, poisoned by endotoxin or by other mediators, to take up oxygen from the blood) (Siegel et al., 1967, 1979).

The data in this study demonstrate that in sepsis, at any given O_2T, an important determinant of O_2 uptake is also represented by the amino acid dose

(AAD). This finding is consistent with the concept that in sepsis, in the presence of altered glucose and fat metabolism, amino acids may become preferential substrates for oxidation (O'Donnell et al., 1976; Siegel et al., 1979; Cerra et al., 1980). It is also consistent with the concept of "permissible thermogenesis", which addresses the possibility that energy metabolism and thermogenesis may at least in part be conditioned in sepsis by the availability of a readily oxidizable substrate, so that abnormalities in O_2 uptake may result from, and parallel, abnormalities in substrate utilization (Giovannini et al., 1989b). This is also suggested by the large dose-effect relationship, in terms of amino acid-mediated increase in VO_2, observed in our patients: a mean incremental ratio of 734 ml O_2/g amino acid implies a thermogenic effect of amino acids of about 90% (De V. Weir, 1949), a figure which is very close to that found in septic patients in a previously published study (Giovannini et al., 1988). Thus, the upward shift in the O_2 consumption/O_2 transport relationship which has been observed with increasing AAD in our patients (Fig. 1) is likely to result from the increased availability of a preferentially oxidized substrate in a condition of altered energy metabolism.

Increases in total body and lower limb O_2 uptake, with unchanged or slightly increased O_2T, after the administration of high-dose branched chain amino acids in septic patients, have been found to be associated with decreases in respiratory quotient and other metabolic changes, thus indicating improvements in energy metabolism (Chiarla et al., 1988, 1990a, b; Giovannini et al., 1989b). However, while most of the metabolism of branched chain amino acids takes place in the periphery, the splanchnic bed is responsible for most of the use of non-branched chain amino acids (Skeie et al., 1990). Increases in splanchnic O_2 uptake, without changes in O_2T, observed after the infusion of mixed amino acid loads, have been considered indicative of the increased hepatic metabolism of non-branched chain amino acids; this finding was the opposite of the response observed after infusing a pyrogen, in which case there was an increase in splanchnic O_2T without changes in O_2 uptake (Myers, 1954). Although in our patients a fixed ratio of branched to non branched chain amino acids did not allow differentiation of the respective effects, it was in any case remarkable to observe that a commonly ignored factor, such as the AAD, had such an evident impact on the O_2 consumption/O_2 transport relationship. This finding deserves to be better clarified, together with the related, clinically relevant aspects. In recent years, in fact, there has been a growing interest for the implications of the depressed O_2 consumption/O_2 transport relationship of sepsis, for the factors capable to improve it, or for the therapies capable to optimize O_2 uptake in spite of it: a better definition of the role of amino acid supply in this particular issue may provide further relevant information for patient management.

Our data have also shown that the AAD-mediated increases in O_2 uptake and VO_2 at any given O_2T were associated with quantifiable and significant changes in blood O_2-CO_2 transport and exchange interactions. The data on total VCO_2 basically reconfirmed the results of previous studies (which have already shown a GD-related increase in VCO_2, and a VCO_2-sparing effect of fat versus glucose in sepsis) (Askanazi et al., 1979; Giovannini et al., 1989a); in addition, the mean value found by us for the Haldane effect (0.276 ml CO_2/ml O_2) is similar to that

previously found by using different methods (Loeppky et al., 1983). However our study has also evidenced a significant impact of substrate infusions on the dynamics of CO_2 transport and exchange, an aspect which has never been addressed before. Such dynamics have a major role in determining the efficiency of CO_2 transport and distribution within the body, are involved in the regulation of CO_2 tension and pH in venous blood and in the tissues, and thus their changes may have important implications. The investigation on this aspect should be continued in subsequent studies, specifically designed to quantify better all the interactions between substrates and patterns of O_2 and CO_2 exchange, and to improve knowledge of their relevance in different clinical states.

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