bjh short report

A left shift in the oxyhaemoglobin dissociation curve in patients with severe coronavirus disease 2019 (COVID-19)

Dominik J. Vogel,[1] Federico Formenti,[2,3] Andrew J. Retter,[1] Francesco Vasques[1] and Luigi Camporota[1,2]

[1]Intensive Care Unit, Guy's and St Thomas' NHS Foundation Trust, [2]Centre for Human and Applied Physiological Sciences, Faculty of Life Sciences and Medicine, King's College London, London, and [3]Nuffield Division of Anaesthetics, Nuffield Department of Clinical Neurosciences, University of Oxford, Oxford, UK

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Correspondence: Dr Dominik J Vogel, Intensive Care Unit, St Thomas' Hospital, Westminster Bridge Road, London SE1 7EH, UK.
E-mail: dominik.vogel@gstt.nhs.uk

Abstract

Critically ill patients with coronavirus disease 2019 (COVID-19) present with hypoxaemia and are mechanically ventilated to support gas exchange. We performed a retrospective, observational study of blood gas analyses ($n = 3518$) obtained from patients with COVID-19 to investigate changes in haemoglobin oxygen ($Hb–O_2$) affinity. Calculated oxygen tension at half-saturation (p_{50}) was on average (±SD) 3·3 (3·13) mmHg lower than the normal p_{50} value (23·4 vs. 26·7 mmHg; $P < 0·0001$). Compared to an unmatched historic control of patients with other causes of severe respiratory failure, patients with COVID-19 had a significantly higher $Hb–O_2$ affinity (mean [SD] p_{50} 23·4 [3·13] vs. 24·6 [5.4] mmHg; $P < 0·0001$). We hypothesise that, due to the long disease process, acclimatisation to hypoxaemia could play a role.

Keywords: haemoglobin, oxygen affinity, infection.

Coronavirus disease 2019 (COVID-19) is characterised by hypoxaemia that can precede radiological changes or other clinical symptoms including dyspnoea.[1] Given that the severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) has a vascular tropism,[2] the physiological manifestation of the altered pulmonary perfusion, hypoxaemia is to a degree disproportionate to the severity of parenchymal lung disease. In addition, direct (midbrain[3]) or indirect (via metabolism of angiotensin II on the carotid bodies[4]) viral actions can affect respiratory drive and response to hypoxaemia. The duration of the disease is generally more prolonged compared to acute respiratory distress syndrome (ARDS) from other aetiologies.[1] As reported by Daniel *et al.*[5] in this Journal, haemoglobin (Hb) oxygen (O_2) affinity in 14 patients with COVID-19 was not different from 11 control participants, when affinity was measured *in vitro* with a Hemox analyser, with a standardised pH (7·4) and temperature. These oxygen tensions at half-saturation (p_{50}) values were obtained directly from the blood gas analyser, without adjustments for physiological changes in CO_2 or pH *in vivo*, which could be important in COVID-19. We hypothesised that *in vivo* $Hb–O_2$ affinity could be affected by other factors in COVID-19.

Methods

To assess alterations in *in vivo* $Hb–O_2$ affinity, we performed a retrospective, observational analysis of all arterial and venous blood gases ($n = 3518$) obtained from all intubated and ventilated patients ($n = 43$) with severe COVID-19 in one intensive care unit (ICU), at Guy's and St Thomas' Hospital (London, UK) between 15 April and 15 May 2020. Institutional approval was gained from the local audit committee (project reference: 11013). The need for individual informed consent was waived for this retrospective analysis of data collected prospectively for routine care, with no breach of privacy or anonymity. The study qualified as a service evaluation as defined by the UK NHS Health Research Authority (NHS HRA) and therefore did not require review by a research ethics committee.

Measured values of partial pressure of oxygen (pO_2) and oxygen saturation (SO_2) were compared to the standard oxyhaemoglobin dissociation curve (ODC) for normal $Hb–O_2$ affinity.[6] The p_{50} values were calculated using the Hill equation (Eq.)[7,8] (after correcting for pH, temperature and base excess[9]; Hill Eq. 1, see below) and derived from Roche blood

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gas analyser (Cobas system, F. Hoffmann-La Roche Ltd;[10] Roche Eq. 2, see below), and compared to the normal value (for pH 7·4, 37·0°C and pCO_2 40 mmHg) of 26·7 mmHg respectively.[11–13] Results were compared to a historic, unmatched control cohort with an overall total of 15 945 arterial and venous blood gas samples obtained from 828 critically ill patients with acute respiratory failure (pneumonia/pneumonitis, or secondary ARDS, but presumed COVID-19 negative, as these samples were obtained in 2017 and earlier). The one-sample *t*-test was used for comparison between actual means (Eq. 1 and Eq. 2, see below) and normal value. The two-tailed, unpaired *t*-test was used for comparison between means of COVID-19 and control samples. Statistical analysis was performed using Prism (GraphPad Software Inc., La Jolla, CA, USA).

We calculated the p_{50} using two methods:

Hill Eq. 1

$$p_{50} = pO_{2(corr)} \times \left(\frac{1-SO_2}{SO_2}\right)^{\frac{1}{2.711}}$$

$$\text{with } pO_{2(corr)} = pO_2 \times 10^{[0.48(pH-7.4)-0.024(T-37)-0.0013 \times \text{Base Excess}]}$$

Roche Eq. 2

$$p_{50} = 26.7 \times 10^{(\lg pO_2 - \lg pO_2^k)}$$

$$\text{with} \lg pO_2^k = \frac{\lg Q + 4.172}{2.9} \text{ and } Q = \frac{SO_2}{1-SO_2}$$

Results

A total of 3518 blood gas analyses of 43 patients [34 (79%) male; mean (range) age 53 (26–77) years] were obtained (Table I). Figure S1 presents pO_2 and SO_2 values. Figure 1 shows the distribution of p_{50} values derived by the Hill equation (Eq. 1). Compared to the standard p_{50} value of 26·7 mmHg, Eq. 1 presented a difference of 3·3 mmHg [mean p_{50} 23·4 mmHg, 99% confidence interval (CI) 23·23–23·50; $P < 0·0001$] and Eq. 2 a difference of 1·9 mmHg (mean p_{50} 24·8 mmHg, 99% CI 24·68–25·00; $P < 0·0001$). Table I shows the comparison to the control group data. Data on intra- and inter-subject variability, as well as data on temporal trends is shown in the *supplement*.

Discussion

When compared to the control group, patients with COVID-19 had a lower pH and higher pCO_2, but not significantly different temperature. Thus, a lower Hb–O_2 affinity (i.e. a right shift of the ODC) would have been expected. However, we found a lower p_{50} (i.e. a left shift of the ODC) in the COVID-19 group (for both equations). We included two different equations in order to strengthen the methodology, and the results obtained from these equations were in good agreement (Pearson's R^2 0·65). The left shift in the ODC is reflected in the opposing alterations in pO_2 and SO_2. Patients with COVID-19 had a significantly lower pO_2, while showing a higher SO_2. In contrast with typical ARDS, changes in Hb–O_2 affinity could reflect the severity and the duration of hypoxaemia prior to presentation to critical care.

The Hb–O_2 affinity is an important link between alveolar O_2 tension and tissue oxygen supply. The Hb–O_2 affinity is characterised in terms of p_{50} (the O_2 tension where 50% of the Hb is oxygenated), and is the defining factor for binding the O_2 that diffuses from the pulmonary alveoli into the

Table I. Summary of results from patients with COVID-19 and from an unmatched control group of critically ill patients without COVID-19. The ratio of arterial oxygen partial pressure to fractional inspired oxygen (PaO_2/FiO_2) was calculated for arterial samples only. Values are reported as mean, standard deviation (SD) and 99% confidence interval (99% CI).

	COVID-19					CONTROL					Group with COVID-19 showed	Unpaired *t*-test (two-tailed *P* value)
	n	Mean	SD	99% CI		*n*	Mean	SD	99% CI			
p_{50} using Eq. 1, mmHg	3518	23·37	3·13	23·23	23·50	15 945	24·59	5·42	24·48	24·70	Lower p_{50}	<0·0001
p_{50} using Eq. 2, mmHg	3518	24·8	3·7	24·7	25·0	15 945	25·7	6·0	25·6	25·8	Lower p_{50}	<0·0001
pH	3518	7·382	0·077	7·379	7·386	15 932	7·397	0·072	7·395	7·398	Lower pH	<0·0001
pO_2, mmHg	3518	77·9	28·0	76·7	79·1	15 945	86·6	62·5	85·3	87·8	Lower pO_2	<0·0001
SO_2, %	3518	94·2	7·9	93·9	94·6	15 945	93·1	10·4	92·9	93·3	Higher SO_2	<0·0001
pCO_2, mmHg	3518	46·1	11·8	45·5	46·6	15 936	43·1	10·1	42·9	43·4	Higher pCO_2	<0·0001
BE, mmol/l	3518	1·2	4·3	1·0	1·3	15 925	0·7	4·2	0·6	0·8	Higher BE	<0·0001
Hct, %	3483	26·5	4·0	26·4	26·7	15 703	29·7	6·7	29·6	29·8	Lower Hct	<0·0001
Hb, g/l	3518	81·2	12·4	80·6	81·7	15 941	93·8	20·2	93·4	94·2	Lower Hb	<0·0001
PaO_2/FiO_2, mmHg	2627	215·7	111·5	210·1	221·3	13,941	299·4	236·5	294·2	304·6	Lower PaO_2/FiO_2	<0·0001
Temperature, °C	3518	36·8	0·8	36·8	36·8	15 945	36·8	0·8	36·8	36·8	NS	0·057

BE, base excess; Hct, haematocrit; NS, no significant difference.

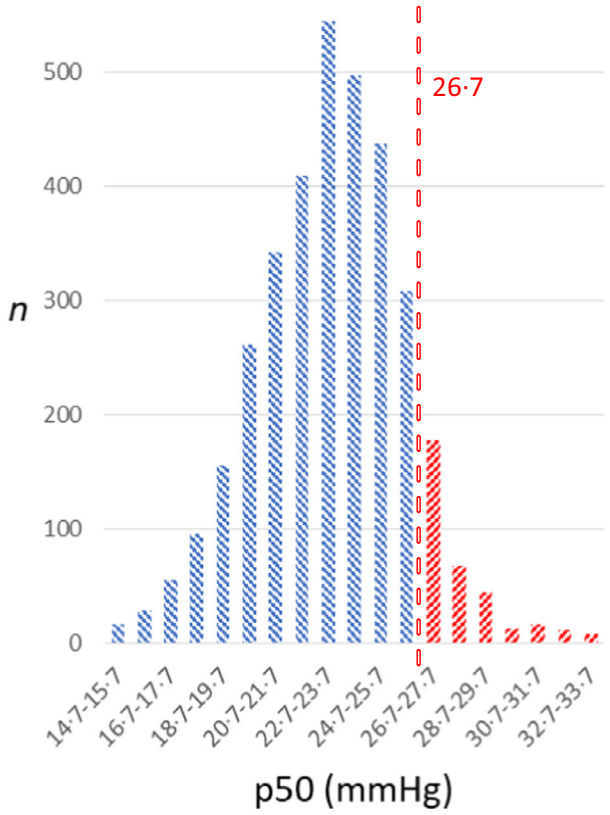

Fig 1. Distribution of p_{50} values calculated using Hill equation (Eq. 1) from measured pO_2 and SO_2 ($n = 3518$). Blue indicates left shift and red indicates right shift of oxyhaemoglobin affinity from the standard p_{50} value. Dashed line at 26·7 mmHg indicates standard value for p_{50}. [Colour figure can be viewed at wileyonlinelibrary.com]

blood and its release in peripheral tissues. Siggaard-Andersen *et al.*[14] have shown agreement between the arterial and the venous values for standard p_{50} based on widely different SO_2 levels. Changes in Hb–O_2 affinity are crucial ways of adjusting both arterial O_2 loading and peripheral O_2 unloading in order to ensure aerobic metabolism when inspired pO_2 decreases and/or O_2 demand increases. The functional properties of Hb may improve tissue O_2 supply with ODC shifts in either direction – increased Hb–O_2 affinity increases O_2-loading under conditions such as severe hypoxia,[15,16] while decreased Hb–O_2 affinity favours the release of bound oxygen from the Hb molecule.

Hb–O_2 binding is considered co-operative, that is, binding of the first molecule of O_2 to Hb causes an increase in the O_2 affinity of the remaining Hb subunits. According to mathematical modelling,[17] an increase in Hb–O_2 affinity resulting from a p_{50} change of −3 mmHg (as seen in our present data using Eq. 1) only slightly increases SO_2 (by 1%) in arterial blood in normoxia [arterial oxygen partial pressure (PaO_2) 90 mmHg]. However, in hypoxia (PaO_2 45 mmHg), the increased Hb–O_2 affinity increases arterial SO_2 by ~4·5%. While being at a disadvantage under normoxaemia, humans with a high Hb–O_2 affinity (adolescents from a family with Hb Andrew-Minneapolis, a stable β-chain mutant with whole blood p_{50} ~17 mmHg) respond more appropriately to altitude-induced hypoxia.[16] An increased Hb–O_2 affinity results in oxygenation benefits during severe hypoxia and increases survival during acute hypoxia in several animal models.[15,18] Thus, a high Hb–O_2 affinity may be of particular importance for O_2 loading in hypoxic conditions.[19] There are well described existing strategies of shifting the ODC to the left and increasing SO_2 at a given pO_2. A fast increase in Hb–O_2 affinity is mediated by a reduction of CO_2 and increase of pH via hyperventilation under environmental hypoxia. This reversible alteration can occur rapidly within seconds to minutes. A slower mechanism is a decrease in 2,3-diphosphoglycerate (DPG) or other organic phosphates.[17] Reduced 2,3-DPG levels were observed in critically ill normoxaemic patients; however, the effect on p_{50} was diminished potentially due to acidaemia in this cohort.[20]

A hypothetical explanation for our present findings in patients with COVID-19 could be the response to prolonged periods of hypoxia. Patients with COVID-19 often present to hospital after a period lasting on average 15 days, during which patients may suffer from 'happy hypoxia', a term coined for the phenomenon that profoundly low SO_2 levels are found in individuals with relatively little subjective sensation of dyspnoea.[21] It has been hypothesised that SARS-CoV-2 may exert an idiosyncratic effect on the respiratory system via angiotensin-converting-enzyme 2 receptors in the carotid body and the midbrain, and this may lead to attenuation of the perceived dyspnoea.[3,4,22] The patients in our present COVID-19 group might have had unrecognised hypoxia for a significant period prior to their hospital admission. Furthermore, even after hospital admission, many patients with COVID-19 remain relatively stable for a few days before they deteriorate and are admitted to the ICU.[1] Thus, when compared to a general critical care population (e.g. patients with ARDS in whom hypoxia has to develop within 7 days, as per the Berlin definition), patients with respiratory failure secondary to COVID-19 may have a much longer time to 'acclimatise' to hypoxaemia. These changes in p_{50} continued to be present during the length of the stay in ICU, therefore we suspect a sustained response, which could be explained by reduced 2,3-DPG levels.[23] The mechanisms and the importance of this phenomenon require further studies.

Author contributions

Dominik J. Vogel designed the study, collected the data, analysed the data, interpreted the data and drafted the first version of the manuscript. Federico Formenti interpreted the data. Andrew J. Retter interpreted the data. Francesco Vasques interpreted the data. Luigi Camporota designed the study and interpreted the data. All authors read and approved the final manuscript.

Conflict of interests

The authors declare no competing interests.

Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Fig S1. SO_2 with respective pO_2 for 3518 blood gas analyses (circles) and the sigmoid fitting line (green line). For comparison standard oxyhaemoglobin dissociation curve is shown (dashed red line).[1]

Fig S2. Hill plot showing SO_2 with respective pO_2 for 3518 blood gas analyses (circles) and the sigmoid fitting line (green line). For comparison standard oxyhaemoglobin dissociation curve is shown (dashed red line).[1]

Fig S3. Distribution of the mean p_{50} calculated for each subject according to the following equation.

Fig S4. Distribution of absolute difference between each measurement and the respective individual's mean p_{50} calculated with Hill Eq. 1.

Fig S5. Delta between each individual measurement from the mean p_{50} of that subject plotted over time starting from the first measurement.

Fig S6. Distribution of the individual patients' mean p_{50} calculated with Eq. 1 and Eq. 2 plotted according to their length of stay during the observation period.

Data S1. Supplemental results.

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