medical journal armed forces india xxx (2017) xxx–xxx

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Original Article

# Study of acute hypoxia markers in healthy subjects: Utility in post-crash investigation

S.R. Santhosh[a,*], Wg Cdr S. Sampath[b], Flt Lt P. Sannigrahi[c], Lt Col A. Gupta[d], Lt Col D.K. Khukhar[e]

[a] Associate Professor (Aviation Pathology and Toxicology), Institute of Aerospace Medicine, IAF, Bangalore, India
[b] Assistant Professor, Department of Biochemistry, Armed Force Medical College, Pune 411040, India
[c] Medical Officer, Air Force Station, Kalaikunda, Kharagpur, West Bengal, India
[d] Assistant Professor (Aviation Pathology and Toxicology), Institute of Aerospace Medicine, IAF, Bangalore, India
[e] Assistant Professor (High Altitude Physiology), Institute of Aerospace Medicine, IAF, Bangalore, India

ARTICLE INFO

*Article history:*
Received 7 October 2016
Accepted 24 April 2017
Available online xxx

*Keywords:*
Aircrash investigation
Hypoxia markers
ELISA and Hypobaric chamber

ABSTRACT

*Background:* Lactic acid is being routinely used as a marker of hypoxia in aircrash investigation. Since lactic acid estimation as a marker of hypoxia in postmortem samples for aircrash investigation is prone to many interfering factors, like the postmortem production and hemolysis. A study was carried out to evaluate other hypoxia markers other than lactic acid which could be later added as markers of hypoxia in postmortem investigations of aircraft accidents.

*Methods:* 25 healthy males of age 20–40 yrs volunteered participants were subjected to an simulated altitude of 15,000 ft for 30 min and the mean plasma concentration of Hypoxia Inducing Factor 1α (HIF 1α), Erythropoietin (EPO), Vascular Endothelial Growth Factor (VEGF) and lactic acid (LA) were analyzed from their venous blood sample collected at 4 intervals viz. Ground level pre exposure, 15,000 ft at 15 min, 15,000 ft at 30 min and Ground level 3 h post exposure.

*Results:* Statistical analysis revealed significant increase in mean plasma concentration of lactic acid, HIF-1α and EPO on exposure for duration of 15 min and 30 min at an altitude of 15,000 ft.

*Conclusion:* Our study reveals that HIF-1α and EPO are sensitive to hypoxia exposure as compared to lactic acid and can be used in association with LA as hypoxia markers. However stability of these proteins in postmortem conditions needs to be studied and the potential for estimation of mRNA transcripts of HIF-1α and EPO, which would be stable in postmortem conditions, can be explored.



* *Corresponding author.*
E-mail address: santhuvet4u@gmail.com (S.R. Santhosh).

http://dx.doi.org/10.1016/j.mjafi.2017.04.003

## Introduction

Acute hypobaric hypoxia had been recognized as one of the foremost physiological threats since humans ventured into the sky in balloons.[1] Accidents due to hypoxia are rare, but hypoxia incidents are common. When fatal accidents occur, hypoxia may not be recognized as a primary cause among the multitude of other potential causes. Retrospective studies conducted after the Second World War give an account of a significant number of unexplained military aircraft accidents that had been suspected to be because of hypoxia.[2–4] Detection of possible hypoxia exposures during postmortem investigation of aircraft accidents has implications while determining flight safety. A study conducted by Tripathi et al from 1986 to 1995 in Army Aviation helicopter flying high altitude sorties revealed 29 accidents and hypoxia was a contributing factor in 24% of all accidents.[5] Pilot incapacitation attributable to hypoxia has been confirmed as the cause of crash of IAF MiG 29 at Sirsi, Karnataka dated 11 Apr 2002.[6]

Lack of a suitable histopathological criterion prompted several studies to direct their attention towards the search of a biochemical marker and subsequent establishment of lactic acid as an indicator of antemortem hypoxia. Elevated lactic acid is used as a marker of antemortem hypoxia during analysis of post-mortem samples during Aircraft Accident Investigations at this Institute. However, being an end product of anaerobic glycolytic pathway, there is always the interference by lactic acid produced during postmortem in the specimens and this may confound the interpretation. Moreover, postmortem hemolysis and presence of certain drugs including barbiturates, amphetamines, opioids, cocaine metabolites, etc. in the blood sample also result in false positive increase in lactic acid levels. Thus, the need for a reliable indicator for the postmortem diagnosis of hypoxia has assumed importance. There have been very few reported studies on markers such as Hypoxia-inducible Factor 1-alpha (HIF-1-alpha), Vascular Endothelial Growth Factor (VEGF), Erythropoietin (EPO); furthermore, these studies have been conducted at a maximum exposure of 4000 m (13,120 ft) only. This study is unique as the markers have been studied after exposure to hypoxia at 4572 m (15,000 ft).

In this study, we decided to expose a group of healthy individuals to simulated acute hypobaric hypoxia and study the time course of changes in the plasma concentration of these acute hypoxia markers to check their significance for utilizing them as markers of hypoxia in postmortem samples.

## Materials and methods

Subjects were volunteers from medical officers attending courses at our institute. Informed consent of the participants was obtained before conducting the study. The participants were selected as per the inclusion and exclusion criteria mentioned below.

(a) Inclusion criteria
- i. Healthy individuals
- ii. Gender: Male
- iii. Age 20–40 years

(b) Exclusion criteria:
- i. Cases of anemia
- ii. Any history of acute or chronic respiratory illness
- iii. Smokers
- iv. Intake of alcohol within the last 72 h
- v. Strenuous exercise within the last 72 h
- vi. Intake of tea/coffee within the last 72 h

The Explosive Decompression Chamber (EDC) or Hypobaric chamber in the Dept of High Altitude Physiology & Hyperbaric Medicine was utilized to induce hypoxia. This chamber is custom-made for the institute by KASCO Industries, Pune. The Hypobaric chamber has the following two parts – Main and Air Lock Chamber (MC and ALC, respectively). The MC houses 10 seats and the ALC four seats. The MC can go up to 15,240 m (50,000 ft) and the ALC up to 30,480 m (100,000 ft). The maximum ascent and descent rate is 80 m/s. The chamber has provision for both manual and automated control via a central control unit (work space).

Ear clearance run as per the existing policy was done on subjects before the study. Ear clearance run is given from ground level at Bangalore 914 m (3000 ft) to 3048 m (10,000 ft) at the ascent and descent rate of 914 m per minute. This profile creates a differential pressure of 157 mm of Hg across the middle ear that is well within the TM rupture pressure. This aids the trainees in equalizing the middle ear pressure voluntarily as taught during the briefing sessions. This check also ensures that individuals in the chamber are able to return to ground level in further runs without any difficulty in pressure equalization.

In our study, the participants were taken in the Hypobaric or EDC without supplemental oxygen to 4572 m (15,000 ft) altitude at an ascent rate of 914 m (3000 ft)/min and were observed in the chamber for 30 min. The profile for ear clearance run and hypoxia exposure is as shown in picture (Fig. 1). The subjects were continuously monitored at the control station through camera and by one to one communication system as well as general PA system. The medical attendants inside the chamber were on 100% oxygen throughout the study.

An altitude of 4572 m was chosen because of the comfort of the subjects, as exposure beyond 4572 m and up to 6096 m (20,000 ft) is known to produce a Stage of Disturbance,[7] where

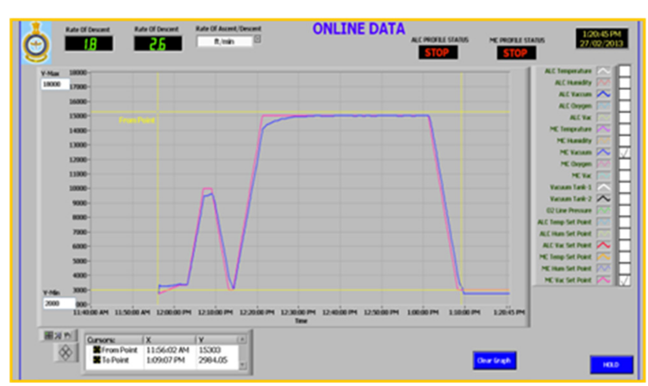


**Fig. 1 – Profile for ear clearance run and hypoxia exposure in this study.**

Please cite this article in press as: Santhosh SR, et al. Study of acute hypoxia markers in healthy subjects: Utility in post-crash investigation, *Med J Armed Forces India*. (2017), http://dx.doi.org/10.1016/j.mjafi.2017.04.003