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Dynamic ventilatory responses of females and males to acute isocapnic and poikilocapnic hypoxia

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ABSTRACT

The human ventilatory response during acute hypoxia appears to be biphasic (growth and decay), but the effect of sex on the size and timing of this response is not known. We studied the effects of 15 min of poikilocapnic and isocapnic hypoxia ($F_IO_2 \approx 0.10$) on ventilation ($\dot{V}_I$) in 14 healthy female and 13 healthy male subjects. Parameters (amplitudes, time delays, time constants) describing individual $\dot{V}_I$ responses were estimated using a biexponential function. There were no significant effects of sex on any of these parameters. CO_2 regulation significantly altered the amplitudes of growth and decay phases and the onset of the latter phase in females and males. These human data suggest that sex does not affect the biphasic response of ventilation during hypoxia. However, additional evidence in this study suggests that sex influences the breathing frequency response during hypoxia and that this effect depends on the control of CO_2.

1. Introduction

During acute hypoxia, ventilation rises to a peak response within a few minutes before falling to stable levels above those observed during normoxia (Easton and Anthonisen, 1988; Easton et al., 1986; Painter et al., 1993; Steinbeck and Poulin, 2007). This rise and fall in ventilation has been observed during poikilocapnic and isocapnic hypoxia, suggesting that the overall dynamic response consists of a growth and decay phase. The growth phase is linked to the decline in arterial PO_2 and stimulation of the peripheral chemoreflex (Weil et al., 1970), but under poikilocapnic conditions it is also constrained by the fall in arterial PCO_2 (Steinbeck and Poulin, 2007). Mechanisms underlying the decay phase are more obscure, perhaps related to decreased peripheral chemoreflex sensitivity to O_2 (Painter et al., 1993) but not thought to involve CO_2 (Easton and Anthonisen, 1988; Steinbeck and Poulin, 2007). The rise and fall in ventilation is linked predominantly to tidal volume, whereas the contribution of breathing frequency is less clear and might depend on the control of arterial PCO_2 (Easton and Anthonisen, 1988; Easton et al., 1986; Steinbeck and Poulin, 2007).

A large number of human studies have explored the effect of sex on ventilatory responses during hypoxia, but the outcome of this research effort is very unclear. Most of these studies measured ventilation during isocapnic hypoxia and using a 'progressive' protocol (Weil et al., 1970) where the level of hypoxia was continuously increased to a severe level. The sensitivity of changes in ventilation under these conditions has been shown to be similar between females and males (Guenette et al., 2004; MacNutt et al., 2012; Marcus et al., 1994; Regensteiner et al., 1988), greater in females (Aitken et al., 1986) or greater in males (Kunitomo et al., 1988; McCauley et al., 1988; White et al., 1983). By contrast, only one study has assessed the biphasic response of ventilation in females and males during a constant level of hypoxia (isocapnic) sustained for 20 min and reported no effect of sex on ventilation (Sajkov et al., 1997). However, interpretation of this finding is difficult because of the significant, transient fall in end-tidal PCO_2 in females (but not males) that might have blunted their initial ventilation responses. More recent evidence pertaining to the biphasic response from studies of C57BL6 mice exposed to poikilocapnic hypoxia indicated that, although the initial rise in ventilation was similar between sexes, the magnitude of decay in ventilation after the peak response was greater in females than males (Palmer et al., 2013).

Mathematical descriptions of the biphasic response of ventilation provide important information for modelling dynamic processes involved in respiratory control (Ben-Tal and Smith, 2010). Empirical models of ventilation responses during *and* after hypoxia have been developed (Clement and Robbins, 1993; Liang et al., 1997; Painter et al., 1993; Ward et al., 1992) for which physiological constructs such as peripheral and central chemoreflex sensitivities have been attributed to model parameters. Such models have also been designed to account

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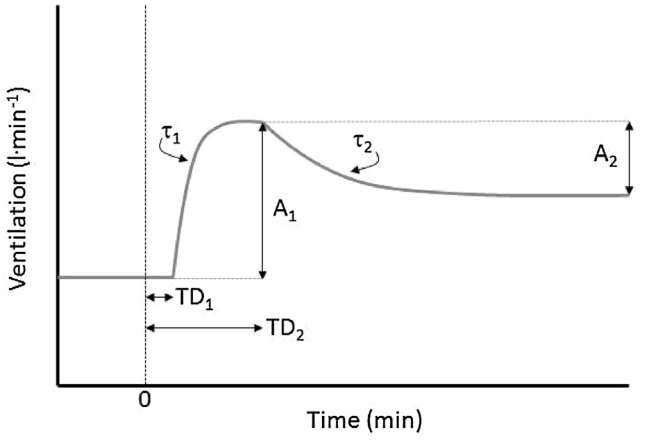

Fig. 1. Conceptual diagram of the biphasic, exponential response of ventilation during acute hypoxia. Hypoxia begins at t = 0 min. The two phases can be described in terms of their amplitudes (A_1, A_2), delays from the beginning of hypoxia (TD_1, TD_2), and rates at which they evolve relative to their amplitudes (τ1, τ2).

for ventilatory suppression after hypoxia. There is, however, a need for a simpler empirical approach which can provide an accurate description of the biphasic response of ventilation during hypoxia and insight into size and timing of phases. This type of approach is used to study dynamic physiological responses during exercise (Lamarra, 1990; Reeder and Green, 2012) and hypoxia (Donnelly and Green, 2013), but to our knowledge has not been used to study ventilation during hypoxia.

The present study used empirical modelling to test the hypothesis that growth and decay phases are larger in females than males. Each phase was assumed to be exponential and defined by parameters representing its amplitude, time delay and time constant (Fig. 1). The hypothesis predicts that phase amplitudes (A_1, A_2), normalised appropriately, are higher in females. To explore involvement of arterial PCO_2 in sex effects on breathing, we tested the hypothesis during poikilocapnic and isocapnic hypoxia and also examined responses of tidal volume and breathing frequency under these conditions.

2. Material and methods

2.1. Overview and design

To test the hypothesis we used a two-factorial, mixed design with sex and CO_2 control ('CO_2') as main factors. Each subject was screened and familiarised with hypoxia (F_iO_2 = 0.10) on a day prior to the experiment. During the experiment subjects rested in a quiet, darkened room in the lateral decubitus position for ~150 min and were exposed to 15-min periods of poikilocapnic hypoxia (PH) and isocapnic hypoxia (IH). PH and IH were preceded by 15-min periods of normoxia, presented in a single-blinded and counterbalanced manner, and separated by 30–40 min of breathing room air not attached to breathing equipment. Baseline physiological responses were averaged from recordings during the last three minutes of normoxia. Individual responses of ventilation during hypoxia from baseline were smoothed and fitted to a biexponential function to estimate sizes and temporal features of growth and decay phases. A two-way ANOVA (mixed) was used to test for main effects of sex and CO_2 on dynamic response characteristics of ventilation during hypoxia.

2.2. Subjects

Fourteen female and 13 male subjects were recruited from Western Sydney University. Baseline characteristics of these subjects are shown in Table 1. According to self-report, all subjects were apparently healthy, sedentary or recreationally active, non-smokers, and did not have cardiovascular, respiratory, metabolic or neurological disease. Female subjects were premenopausal, had not taken hormonal contraceptives for at least six months prior to testing, and were tested during the first week of the follicular phase of their menstrual cycles. The study was approved by the institutional human research ethics committee and conducted in accordance with the principles of the Declaration of Helsinki (2013).

Table 1
Baseline characteristics (mean ± SD) of females and males, including respiratory variables prior to poikilocapnic hypoxia (PH) and isocapnic hypoxia (IH).

		♀	♂
	Age (y)	26.6 ± 8.6	26.0 ± 8.2
	Height (m)	167.3 ± 6.1*	180.0 ± 7.9
	Weight (kg)	67.0 ± 11.2*	77.2 ± 8.8
	BSA (m^2)	1.75 ± 0.14*	1.96 ± 0.15
PH	$\dot{V}_I$ (l/min)	7.75 ± 0.81*	9.41 ± 1.62
	$\dot{V}_I$ (l/min/m^2)	4.41 ± 0.51	4.79 ± 0.70
	V_t (l)	0.51 ± 0.10*	0.73 ± 0.20
	V_t (l/m^2)	0.30 ± 0.06*	0.37 ± 0.10
	F_b (breaths·min^{-1})	15.7 ± 3.3	13.8 ± 2.8
	$P_{et}CO_2$ (mmHg)	41.4 ± 10.7	41.5 ± 10.6
	S_aO_2 (%)	99.1 ± 1.1	98.9 ± 1.0
IH	$\dot{V}_I$ (l min^{-1})	7.70 ± 1.23*	9.76 ± 2.36
	$\dot{V}_I$ (l/min/m^2)	4.43 ± 0.64	4.97 ± 1.10
	V_t (l)	0.51 ± 0.10*	0.68 ± 0.21
	V_t (l/m^2)	0.29 ± 0.04*	0.34 ± 0.09
	F_b (breaths·min^{-1})	15.8 ± 3.2	15.1 ± 2.5
	$P_{et}CO_2$ (mmHg)	43.8 ± 4.8	44.3 ± 3.2
	S_aO_2 (%)	99.2 ± 1.2	98.9 ± 1.1

* Indicates a main effect of sex at P < 0.05.

2.3. Normoxia and hypoxia

Subjects wore a nose clip, breathed through a one-way non-rebreathing valve (Hans-Rudolph, series 2700) connected to a 2 m respiratory hose, two-way valve and 350 l Tissot spirometer. During normoxia (F_iO_2 = 0.2094) subjects breathed room air and during hypoxia subjects breathed a gas mixture (F_iO_2 = 0.10) from the spirometer. During IH, a 'running' averaged estimate of end-tidal PCO_2 (5-breath average) was monitored and maintained at a normoxic level (mean of 10 min) through manual adjustment of flow of CO_2 mixed well into the inspirate.

2.4. Cardiorespiratory variables

Tidal volume (V_t), breathing frequency (F_b) and minute ventilation ($\dot{V}_I$) were measured using a turbine (VMM-400, Interface Associates, USA) positioned on the inspired side ~30 cm from the mouth. Gas was sampled continuously from a mouth port at 200 ml min^{-1} and analysed for O_2 and CO_2 concentrations (R-2 flow controller, S-3A and CD-3A analysers: AEI Technologies, USA). The maximum values of O_2 and CO_2 (converted to partial pressures) during each breathing cycle represented end-inspired PO_2 (P_iO_2) and end-tidal PCO_2 ($P_{et}CO_2$). Arterial O_2 saturation (S_aO_2%) was measured at the finger using pulse oximetry (Masimo Rad-7, USA). All physiological signals were recorded using three A/D converters (PowerLab 26T, AD Instruments, Sydney) and commercial software (LabChart Pro v8.0, AD Instruments, Sydney) and processed off-line.

2.5. Curve fitting and parameter estimation

Breath-by-breath responses of $\dot{V}_I$ during hypoxia were smoothed (5-breath rolling average) and fitted to the biexponential function,

$$Y(t) = a + A_1(1 - e^{-(t-TD_1)/\tau_1}) - A_2(1 - e^{-(t-TD_2)/\tau_2}) \quad (1)$$