Measuring the ventilatory response to hypoxia

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After defining the current approach to measuring the hypoxic ventilatory response this paper explains why this method is not appropriate for comparisons between individuals or conditions, and does not adequately measure the parameters of the peripheral chemoreflex. A measurement regime is therefore proposed that incorporates three procedures. The first procedure measures the peripheral chemoreflex responsiveness to both hypoxia and CO_2 in terms of hypoxia's effects on the sensitivity and ventilatory recruitment threshold of the peripheral chemoreflex response to CO_2. The second and third procedures employ current methods for measuring the isocapnic and poikilocapnic ventilatory responses to hypoxia, respectively, over a period of 20 min. The isocapnic measure is used to determine the time course characteristics of hypoxic ventilatory decline and the poikilocapnic measure shows the ventilatory response to a hypoxic environment. A measurement regime incorporating these three procedures will permit a detailed assessment of the peripheral chemoreflex response to hypoxia that allows comparisons to be made between individuals and different physiological and environmental conditions.

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While several methods have been developed to measure the ventilatory response to hypoxia, a standard method capable of comparing test results between subjects and within the same subject under different conditions remains elusive. Indeed at the recent 15th international hypoxia symposium held at Lake Louise a consensus could not be reached. Nevertheless, such a consensus is highly desirable because it will permit comparisons that will further our understanding of how the ventilatory response to hypoxia changes between and within individuals under different physiological (e.g. changes in cerebral blood flow and central chemoreflex sensitivity) and environmental (e.g. following acclimatization to hypoxia) circumstances, as well as with drug-induced changes. The aim of this article is therefore to describe the current problems associated with measuring the hypoxic ventilatory response (HVR) and propose a method that overcomes them.

The ventilatory response to hypoxia is mediated by the peripheral chemoreflex, a reflex arc from the carotid body sensor to the respiratory muscle effectors (Torrance, 1996). In the range of hypoxia limited by ethical constraints, hypoxia does not affect ventilation through other means such as central depression or excitation (Honda, 1992; Fatemian *et al.* 2003), and is mediated by the peripheral chemoreflex alone (Cunningham, 1987). The HVR can be measured with CO_2 tensions allowed to change (poikilocapnic) or fixed (isocapnic). Figure 1 illustrates the ventilatory response to hypoxia at several isocapnic tensions.

The HVR is time dependent (Easton *et al.* 1986; Powell, 2007) and the isocapnic HVR can be arbitrarily divided into two phases, a first phase (0–5 min) of immediate ventilation increase, followed by a second phase (5–20 min) of slow decline (Steinback & Poulin, 2007). The second phase is referred to as hypoxic ventilatory decline (HVD) (Liang *et al.* 1997). Moreover, HVR may be dependent on the pattern of previous hypoxic exposures and sustained CO_2 tensions (Mateika *et al.* 2004; Harris *et al.* 2006).

The peripheral chemoreflex response to hypoxia consists of an increase in the peripheral chemoreflex sensitivity to CO_2 via changes in $[H^+]$ at the carotid body (Cunningham, 1987; Torrance, 1996; Kumar & Bin-Jaliah, 2007). The ventilatory recruitment threshold of this CO_2 response depends on the level of carotid body activity. In sea level subjects the increase in CO_2 sensitivity dominates the response to hypoxia, with little increase in carotid body activity (Mohan & Duffin, 1997), but in adapted altitude residents the increase in CO_2 sensitivity with hypoxia may be lost (Leon-Velarde *et al.* 2003) (also see Fig. 10). In this case, the response to hypoxia may be a decrease in the ventilatory recruitment threshold due to an increase in carotid body activity.

In most individuals, hyperoxia ($P_{O_2} = 150$ mmHg) effectively silences the peripheral chemoreflex response to CO_2 (Lloyd & Cunningham, 1963; Mohan & Duffin, 1997). It is suggested that hyperoxia be limited to this value during testing to avoid its stimulatory effects (Becker *et al.*

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1996). The time dependency of the ventilatory response to hypoxia is dominated by changes in the ventilatory recruitment threshold of the CO_2 response, reflecting changes in carotid body activity, rather than changes in the sensitivity of the CO_2 response (Duffin & Mahamed, 2003).

Ventilation depends on both the peripheral chemoreflex drive to breathe and the central chemoreflex drive to breathe (Cunningham *et al.* 1986). When hypoxia stimulates the peripheral chemoreflex its effect is assumed to be additive with the central chemoreflex drive already present (Clement *et al.* 1992; Clement *et al.* 1995; St. Croix *et al.* 1996). Determining the peripheral contribution therefore requires subtraction of the central contribution from the measured ventilation. The central chemoreflex stimulus is the medullary $[H^+]$, determined by central P_{CO_2}, with the latter dependent on both arterial P_{CO_2} and cerebral blood flow. Since cerebral blood flow is affected by both CO_2 and hypoxia (Poulin *et al.* 1996; Vovk *et al.* 2002; Meadows *et al.* 2004), the central chemoreflex stimulus is not easily determined from end-tidal P_{CO_2} measurements.

These factors require consideration when devising tests of the ventilatory response to hypoxia. I suggest that current methods of measuring the HVR are inadequate since they do not allow either comparisons between different conditions within the same individual or comparisons between different individuals that are (1) free of possible artefactually induced differences and (2) employ a standardized method of choosing isocapnia. The following sections detail why current methods fail and then suggest a measurement regime that overcomes these difficulties and fully characterizes the ventilatory response to hypoxia.

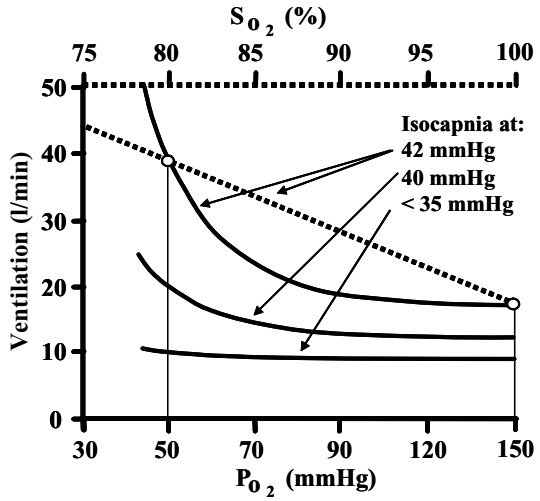

Figure 1. The isocapnic ventilatory response to hypoxia
Ventilation increases rectangular hyperbolically with decreasing P_{O_2} or linearly with decreasing oxygen saturation at any constant P_{CO_2}. At an isocapnic P_{CO_2} below the ventilatory recruitment threshold the response is absent, but as P_{CO_2} exceeds the ventilatory recruitment threshold the response increases.

Measuring HVR

The current acute HVR test consists of establishing an isocapnic P_{CO_2} and then producing a known degree of hypoxia and measuring the resulting change in ventilation before HVD intervenes. The isocapnic P_{CO_2} is usually chosen to be 1–2 mmHg above resting (e.g. Bascom *et al.* 1990). The resulting change in ventilation may be divided by the change in P_{O_2} or S_{O_2} to obtain a measure of hypoxic sensitivity (Fig. 1). The function of this test can be demonstrated using a graphical model of the control of breathing which illustrates the relation between the ventilatory responses to hypoxia and carbon dioxide (Fig. 2). The model features a metabolic hyperbola, describing the dependence of P_{CO_2} on ventilation, and a linear ventilatory response to P_{CO_2}. The latter is the sum of linear ventilatory responses to P_{CO_2} mediated by the central and peripheral chemoreflexes (Fig. 3). The peripheral response slope or CO_2 sensitivity depends on P_{O_2} such that at high P_{O_2} (i.e. at or above 150 mmHg) the sensitivity is neglible, and at low P_{O_2} it becomes significant, varying rectangular hyperbolically with P_{O_2}. The intersection of the metabolic hyperbola and the CO_2 response indicates the equilibrium point defining resting ventilation and P_{CO_2}.

Using this model, Fig. 2 shows the current definition of HVR, and illustrates both its dependence on the choice of isocapnic P_{CO_2} and the central ventilation response to P_{CO_2}. It also illustrates that changes in HVR may be due to alterations in either the slopes or the thresholds of these responses. Therefore, the current test does not show whether a change in HVR is due to an alteration of the interaction of hypoxia with the peripheral chemoreflex CO_2 response, which would affect the slope, or due to an alteration in the carotid body activity, which would affect the ventilatory recruitment threshold.

As stated in the proposal introduced by John W. Severinghaus at the recent 15th international hypoxia conference at Lake Louise (http://www.hypoxia.net/forum/HVR.pdf): 'Peripheral chemosensitivity should be tested with equal central ventilatory drive'. Isocapnic measures of ventilatory responses to changes in P_{O_2} or S_{O_2} rely on the choice of isocapnic P_{CO_2} to maintain equal central ventilatory drives between tests (Harris *et al.* 2006). However, achieving this condition is problematic (Robbins, 2007).

If the isocapnic choice is based relative to resting P_{CO_2}, then changes in the ventilatory recruitment threshold of the P_{CO_2} response due to alterations in acid–base, such as the leftward shift that occurs at altitude (Somogyi *et al.* 2005), produce an artefactual increase in HVR as shown

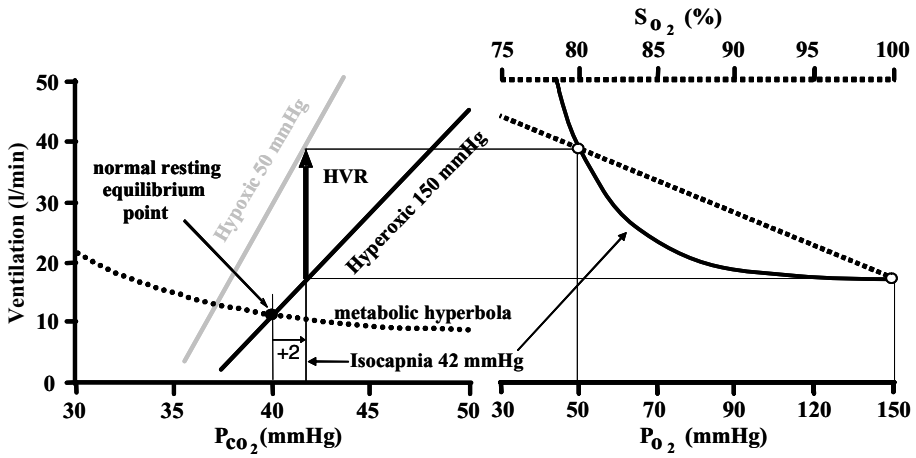

Figure 2. The relationship between hypoxic and hypercapnic ventilatory responses and the current definition of HVR

At a chosen isocapnic P_{CO_2} (here set to 2 mmHg above resting) the ventilation increase (arrow) from the hyperoxic CO_2 response line (assumed to reflect the central chemoreflex response) to any hypoxic CO_2 response line (assumed to reflect the sum of central and peripheral chemoreflex responses) is the HVR. Note that the magnitude of the HVR depends on the choice of isocapnic P_{CO_2}, and that at a given isocapnic tension the mechanisms (change in slope or threshold) responsible for the HVR are indistinguishable from one another.

in Fig. 4. This problem can be overcome by using an isocapnic P_{CO_2} that produces the same hyperoxic ventilation as suggested by (Sato *et al.* 1994), and is illustrated in Fig. 5. However, neither of these methods for choosing the isocapnic P_{CO_2} prevents an artefactual increase in HVR if there is a change in central chemosensitivity between tests, as illustrated in Figs 6 and 7.

A further concern is that no matter what isocapnic P_{CO_2} is used, changes of cerebral blood flow with hypoxia during testing or changes in cerebral blood flow reactivity to CO_2 and hypoxia between and within tests and between individuals will also confound the measurement of HVR by altering the central chemoreflex drive due to CO_2 washout (Ainslie & Poulin, 2004; Xie *et al.* 2006). The effect of a change in cerebral blood flow on HVR measured as the isocapnic change in ventilation with hypoxia is illustrated in Fig. 8. An example of such a change in cerebrovascular reactivity is that which occurs overnight (Meadows *et al.* 2005; Corfield & Meadows, 2006) or upon ascent to high altitude (Ainslie *et al.* 2007).

These scenarios illustrate the difficulty in making a choice of isocapnia that will allow comparisons of HVR between tests. It is therefore necessary to develop measures that will allow such comparisons as well as fully describing the ventilatory response to hypoxia.

Proposed regime for measuring the hypoxic ventilatory response

The following is not intended to mandate the details of a particular experimental setup and the testing conditions, but rather the principles of testing that should be followed so that test results may be compared. A measurement regime consisting of three procedures is proposed. The first determines the immediate (within 5 min) peripheral chemoreflex ventilatory response. The second procedure measures the time course of HVD and the third procedure measures the ventilatory response to environmental hypoxia. Figure 9 outlines the proposed measurement regime, which makes the following assumptions.

(1) HVR is mediated by the peripheral chemoreflex alone (Cunningham, 1987).

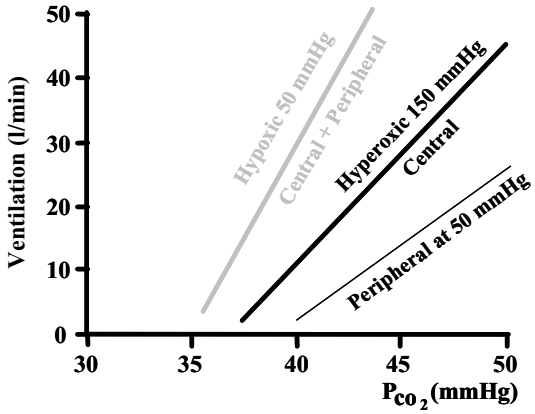

Figure 3. The additive effect of central and peripheral responses to hypercapnia

The hyperoxic line is the addition of a neglible peripheral response and the central response and so reflects the central chemoreflex response alone. The hypoxic line is again the addition of central and peripheral responses, this time with a substantial peripheral contribution.

(2) Hyperoxia (150 mmHg) silences the peripheral chemoreflex response to CO_2 (Lloyd & Cunningham, 1963; Mohan & Duffin, 1997).

(3) In the range of hypoxia limited by ethical considerations, hypoxia does not affect ventilation through other means such as central depression or excitation (Honda, 1992; Fatemian *et al.* 2003).

(4) Ventilation is controlled by both central and peripheral chemoreflexes (Cunningham *et al.* 1986) and their drives are additive (Clement *et al.* 1992; Clement *et al.* 1995; St. Croix *et al.* (1996).

(5) Cerebral blood flow is affected by both CO_2 and hypoxia (Poulin *et al.* 1996; Vovk *et al.* 2002; Meadows *et al.* 2004).

(6) HVR is time dependent (Easton *et al.* 1986; Powell, 2007) and can be arbitrarily divided into two phases; a first phase (0–5 min) of immediate ventilation increase, followed by a second phase (5–20 min) of HVD (Liang *et al.* 1997; Steinback & Poulin, 2007).

Procedure 1. Measuring peripheral chemoreflex responsiveness

For the reasons stated previously, the peripheral chemoreflex response to hypoxia cannot simply be measured as the difference in ventilation produced by changes in isocapnic hypoxic and hyperoxic O_2 tensions or saturations. However, it can be determined by comparing the difference between ventilatory responses to CO_2 at a high constant O_2 tension (isoxic hyperoxic ventilatory response to CO_2) and a low constant O_2 tension (isoxic hypoxic ventilatory response to CO_2). Given the assumptions above, the hyperoxic response measures the

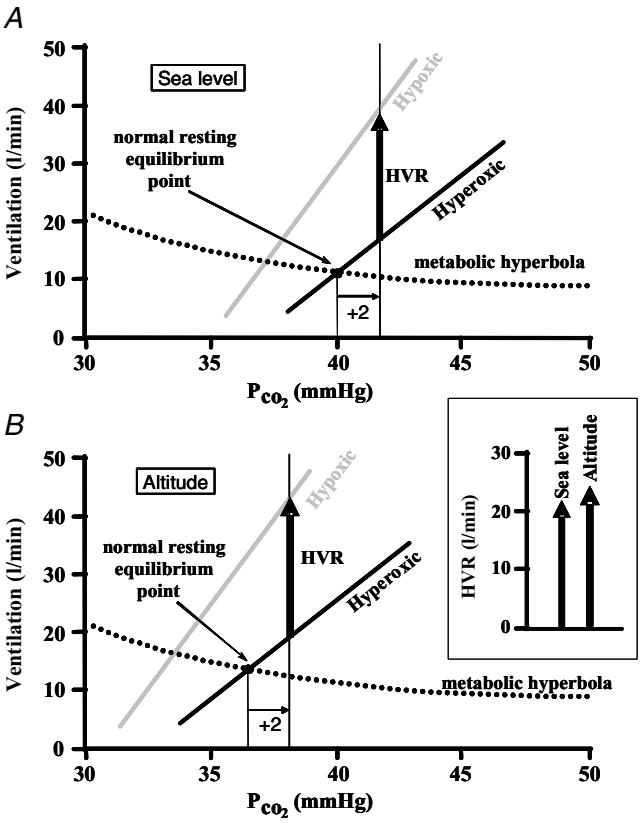

Figure 4. A threshold shift scenario
The HVR (as defined in Fig. 2), is shown at sea level (*A*) and high altitude (*B*). The only difference between the ventilatory responses to CO_2 at sea level and those at altitude at a given isoxic tension (hyperoxic or hypoxic) is a decrease in threshold (leftward shift); the metabolic hyperbola is assumed to be unchanged. If the choice of isocapnia is made relative to the resting P_{CO_2}, then the altitude HVR compared to the HVR at sea level is artefactually increased as the inset box shows in a side-by-side comparison.

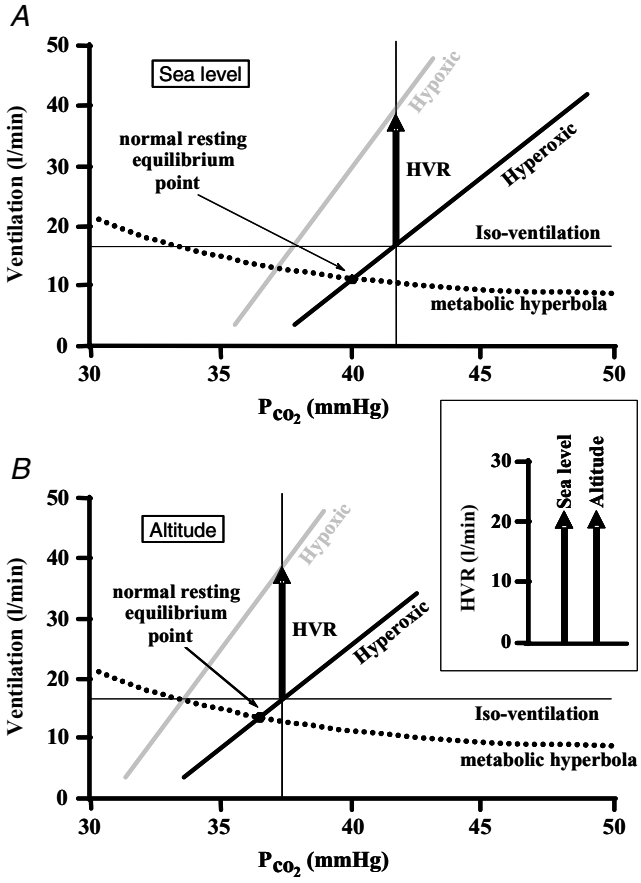

Figure 5. A threshold shift scenario as for Fig. 4
The HVR (as defined in Fig. 2), is shown at sea level (*A*) and high altitude (*B*). The only difference between the ventilatory responses to CO_2 at sea level and those at altitude at a given isoxic tension (hyperoxic or hypoxic) is a decrease in threshold (leftward shift); the metabolic hyperbola is assumed to be unchanged. If a hypercapnic iso-ventilation is used to set the isocapnic P_{CO_2} then the altitude HVR compared to the HVR at sea level does not change and is measured correctly as the inset box shows in a side-by-side comparison.

central chemoreflex and the hypoxic response measures the sum of the central and peripheral responses. Therefore the difference between hyperoxic and hypoxic responses represents the ventilatory contribution of the peripheral chemoreflex.

Such a comparison (Fig. 10) shows that the peripheral chemoreflex response to hypoxia can be characterized by an increase in sensitivity to CO_2, the difference of sensitivities between the isoxic hypoxic and hyperoxic ventilatory responses to CO_2, and a decrease in the ventilatory recruitment threshold from the hyperoxic to the hypoxic isoxic ventilatory response to CO_2, reflecting either an increase in peripheral chemoreflex activity or the increase in CO_2 sensitivity. These two measures thus define the peripheral chemoreflex.

If comparison of peripheral chemoreflex responsiveness is the aim, then the measurements of the isoxic responses to CO_2 can be limited to two defined isoxic tensions: (1) a hyperoxic P_{O_2} of 150 mmHg to determine the central response to CO_2 and (2) a hypoxic P_{O_2} of 50 mmHg, which would add the peripheral response. The differences in sensitivities and ventilatory recruitment thresholds between these tests define a peripheral chemoreflex responsiveness, which can then be compared between subjects and conditions.

However, if the aim is to fully characterize the peripheral chemoreflex, then a series of hypoxic isoxic CO_2 responses at different P_{O_2} tensions can be measured and their differences in sensitivity and ventilatory recruitment threshold from the hyperoxic isoxic CO_2 response defines the complete peripheral chemoreflex response to hypoxia. Such results can be displayed either as a series of isocapnic ventilatory responses to P_{O_2} or as a plot of peripheral chemoreflex CO_2 sensitivity and threshold against P_{O_2} as shown in Fig. 11. The latter relations are useful to modellers (Duffin, 2005; Stuhmiller & Stuhmiller, 2005; Topor *et al.* 2007).

The isoxic hypoxic and hyperoxic ventilatory responses to CO_2 can be measured using either steady state methods,

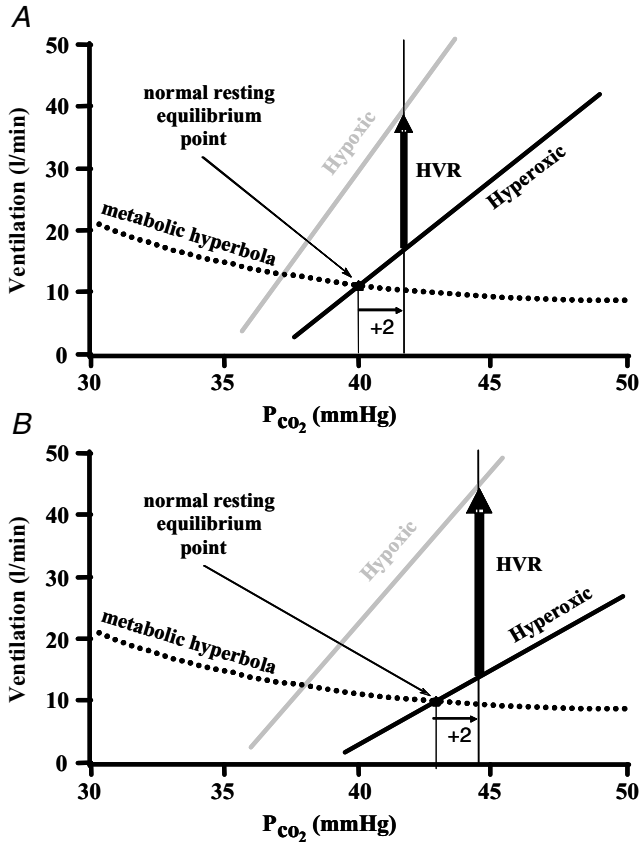

Figure 6. The HVR is as shown in Fig. 2, representing the HVR in the first condition (*A*) and second condition (*B*)
The second condition (*B*) shows a decrease in the slope of the hyperoxic CO_2 response line, but no change in the peripheral chemoreflex contribution to the hypoxic CO_2 response line. If the choice of isocapnia is made relative to the resting P_{CO_2}, then the HVR of condition 2(B) compared to the HVR of condition 1(A) is artefactually increased.

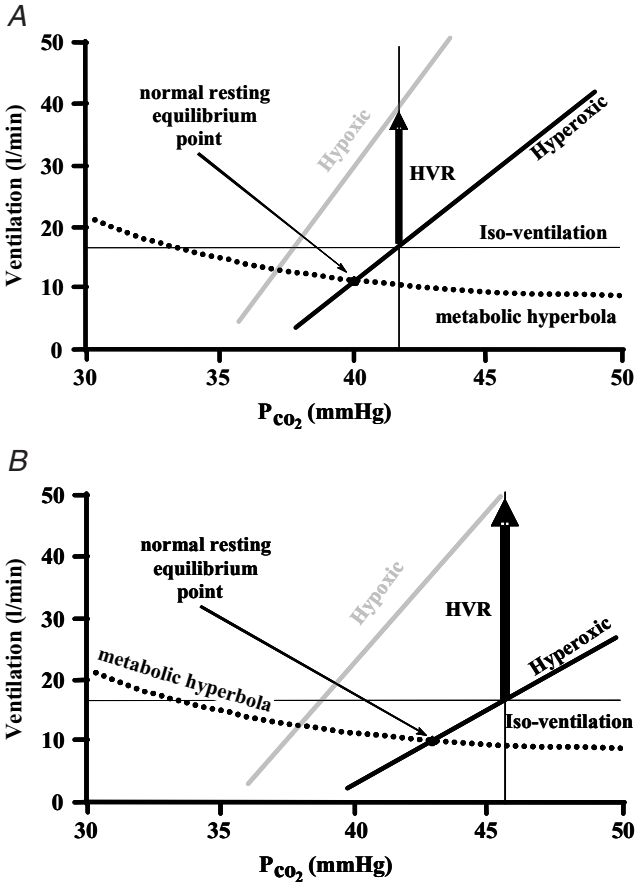

Figure 7
Repeating the scenario pictured in Fig. 6, if a hypercapnic iso-ventilation is used to set the isocapnic P_{CO_2} then the HVR of condition 2 (*B*) compared to the HVR of condition 1 (*A*) is artefactually increased.

including end-tidal forcing, or rebreathing methods, but should be completed within the first 5 min of hypoxic exposure, an arbitrarily set time limit based on the observed time course of the immediate response. Each method has advantages and disadvantages.

The steady state method requires at least three points to determine the sensitivity to CO_2 at each isoxic level, requiring some 5 min per point response time to allow central equilibration of CO_2, and therefore care in limiting the accumulated hypoxic exposure time to avoid HVD. An experimental approach such as the following could be used: in a background of hyperoxia (e.g. 150 mmHg), increase end-tidal P_{CO_2} to three different isocapnic tensions above resting each for 8 min with the last 3 min at specific end-tidal hypoxic tension (e.g. 50 mmHg), with each isocapnic tension separated by a resting period of at least 5 min to return end-tidal tensions to normal. At each isocapnic tension the fifth minute ventilation measures the central response and the eighth minute measures the central plus peripheral response. This approach measures the response relative to arterial CO_2, and may underestimate the sensitivity due to the increase in cerebral blood flow with CO_2 (Berkenbosch *et al.* 1989; Pandit *et al.* 2003). Since the ventilatory recruitment thresholds are not measured, the difference between the hyperoxic and hypoxic responses at resting ventilation can be substituted.

The modified rebreathing method (Mohan *et al.* 1999; Duffin *et al.* 2000) measures both sensitivity and ventilatory recruitment threshold within the initial 5 min of hypoxia. It measures the response relative to venous CO_2, reducing the arteriovenous difference sufficiently to negate any effects of cerebral blood flow changes. It can therefore be used to measure changes in the peripheral chemoreflex response where there are known changes in cerebrovascular activity such as those that occur overnight (Mahamed *et al.* 2005; Meadows *et al.* 2005; Beecroft *et al.* 2006) or at high altitude (Ainslie *et al.* 2007). However, the rebreathing method may overestimate the CO_2 sensitivity if central CO_2 increases faster than mixed venous CO_2 because brain CO_2 production is greater than whole body CO_2 production. Modelling shows this factor to be negligible (Read & Leigh, 1967) but direct experimental evidence for this assumption is lacking.

Procedure 2. Measuring HVD, the isocapnic hypoxic ventilatory response

The peripheral chemoreflex response measurement procedure as described above can be considered a

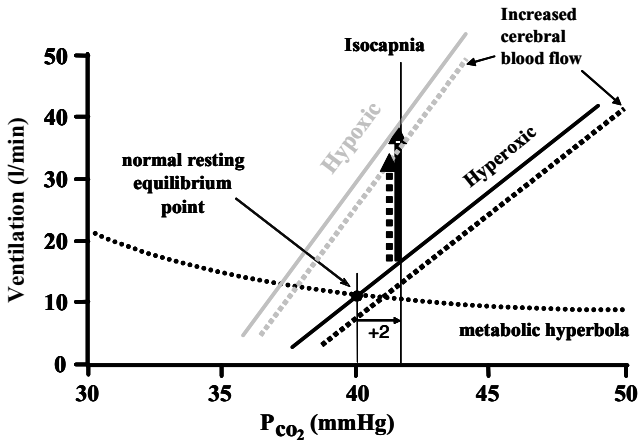

Figure 8. An increase in cerebral blood flow scenario
As in Fig. 2, after determining the ventilation at an isocapnic P_{CO_2} in hyperoxia to measure the central chemoreflex contribution, hypoxia is introduced and the ventilation measured again at the isocapnic P_{CO_2}. Here, however, hypoxia induces an increase in cerebral blood flow, washing out central CO_2 and reducing the central chemoreflex response. The dashed lines show the effect on both hyperoxic and hypoxic CO_2 response lines. As a result the HVR measured (dashed arrow) is artefactually reduced from its true value (continuous arrow).

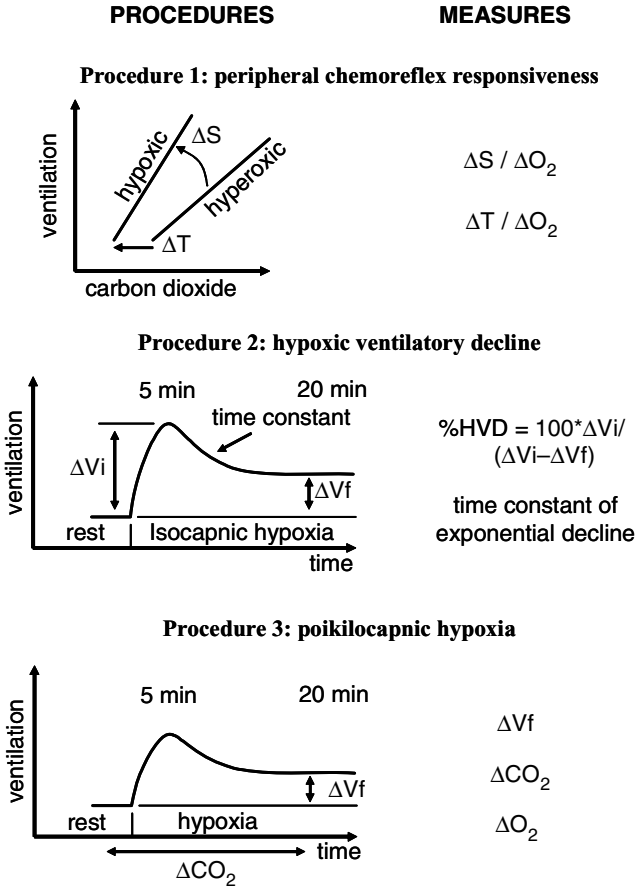

Figure 9. Procedures of the measurement regime
Procedure 1 measures the changes in slopes (ΔS) and thresholds (ΔT) of the ventilatory responses to CO_2 for a given change from hyperoxia to hypoxia (ΔO_2). Procedure 2 measures the time course of the decline in ventilation between 5 and 20 min after the start of isocapnic hypoxia at a specified CO_2 and O_2 in terms of the percentage HVD and the time constant of the fitted exponential decline. Procedure 3 measures the ventilatory response (ΔV_f and ΔCO_2) after 20 min of poikilocapnic hypoxia at a specified O_2.

'snapshot' of the immediate peripheral chemoreflex response to hypoxia. To measure the second phase, HVD, two approaches can be employed. One measures the peripheral chemoreflex response to hypoxia before and after exposure to 20 min of hypoxia (Mahamed & Duffin, 2001) and determines the changes in chemoreflex characteristics according to the first procedure. This approach does not require a choice of isocapnia and if rebreathing is used then cerebral blood flow changes do not affect the measurement; however, the time course of the HVD is not determined.

The other approach measures the complete time course of ventilation during 20 min of isocapnic hypoxia (Steinback & Poulin, 2007) so that the time course of HVD can be modelled (Liang *et al.* 1997). Isocapnic hypoxia may be maintained using a feedback approach to end-tidal forcing (Robbins *et al.* 1982) or a feedforward approach using sequential breathing (Slessarev *et al.* 2007). In this test the choice of isocapnic P_{CO_2} is again problematic, but if comparisons are limited to the time course of the response and the degree of HVD rather than the absolute amplitude, and the effect of any cerebral blood flow changes over the 20 min of measurement can be ignored (Suzuki *et al.* 1989), then HVD can be determined.

Procedure 3. Measuring the poikilocapnic hypoxic ventilatory response

The effect of hypoxia on ventilation, when P_{CO_2} is uncontrolled is not large (Steinback & Poulin, 2007) because any increase in ventilation increases the elimination of CO_2, thereby lowering P_{CO_2} and reducing both the central and peripheral chemoreflex contributions to ventilation. The central chemoreflex contribution is also affected by changes in central P_{CO_2} resulting from changes in cerebral blood flow that occur in response to both hypoxia and the resulting hypocapnia (Poulin & Robbins 1998; Poulin *et al.* 2002). Measuring ventilation during a 20 min poikilocapnic hypoxic exposure thus simulates the response that would be observed on an environmental exposure to hypoxia, for example at altitude. As such it may serve a predictive purpose, relevant to altitude sickness, but this aspect has not been tested.

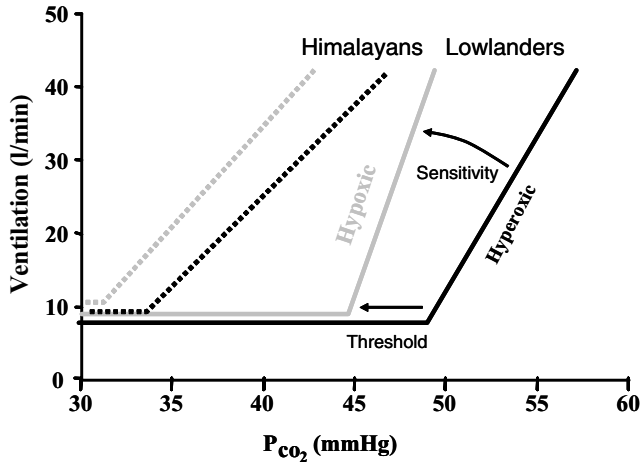

Figure 10. Mean modified rebreathing tests results (Slessarev *et al.* 2007)
A comparison of isoxic hyperoxic (150 mmHg P_{O_2}) ventilatory responses to CO_2 (black lines) with isoxic hypoxic (50 mmHg P_{O_2}) ventilatory responses to CO_2 (grey lines) shows that in lowlanders (continuous lines) an increase in the slope (sensitivity) of the response dominates and most of the change in ventilatory recruitment threshold can be attributed to this change in slope. By contrast, in long-time Himalayan residents (dashed lines) the increase in sensitivity is not significant and a decrease in ventilatory recruitment threshold dominates indicating an increase in the carotid body activity.

Concluding remarks

The measurement procedures described here can be carried out using a variety of equipment and techniques and under a variety of conditions. They are designed to

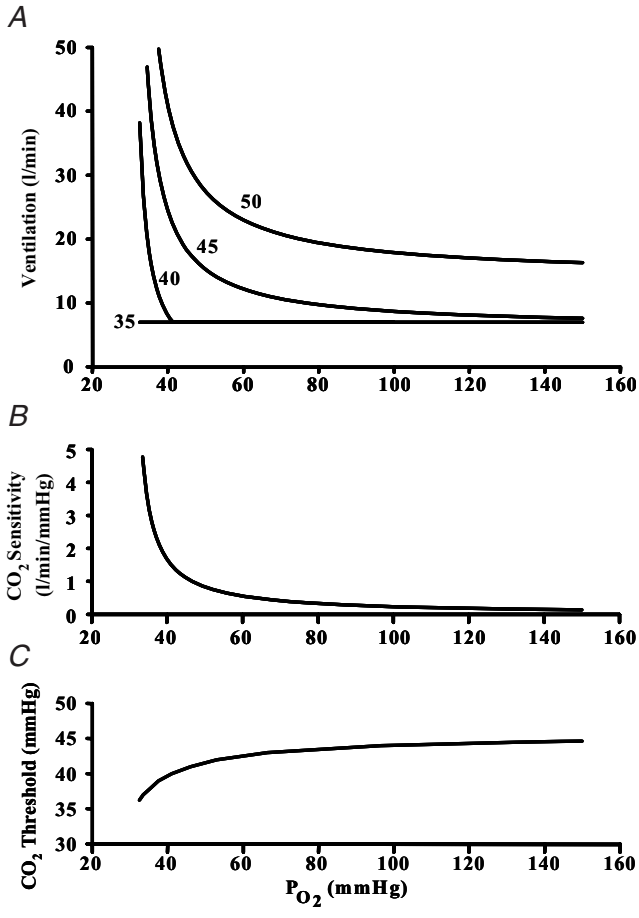

Figure 11
A, isocapnic ventilatory responses to hypoxia at isocapnic tensions of 35, 40, 45 and 50 mmHg. *B*, variation of the peripheral chemoreflex CO_2 sensitivity with P_{O_2}. *C*, variation of the CO_2 response ventilatory recruitment threshold with P_{O_2}.

provide measures of the ventilatory response to hypoxia that can be compared between subjects and within subjects under different conditions. The same principles of methodology can be applied to measuring other physiological responses to hypoxia such as cerebral blood flow (Vovk *et al.* 2002) and cardiovascular changes (Shoemaker *et al.* 2002). It is recognized that the assumptions underlying the tests as stated here may be subject to criticism; they are proposed to stimulate discussion in the hope that a consensus may be reached and standardized methods of measurement developed as a result.

References

Ainslie PN, Burgess K, Subedi P & Burgess KR (2007). Alterations in cerebral dynamics at high altitude following partial acclimatization in humans: wakefulness and sleep. *J Appl Physiol* **102**, 658–664.

Ainslie PN & Poulin MJ (2004). Ventilatory, cerebrovascular, and cardiovascular interactions in acute hypoxia: regulation by carbon dioxide. *J Appl Physiol* **97**, 149–159.

Bascom DA, Clement ID, Cunningham DA, Painter R & Robbins PA (1990). Changes in peripheral chemoreflex sensitivity during sustained, isocapnic hypoxia. *Respir Physiol* **82**, 161–176.

Becker HF, Polo O, Mcnamara SG, Berthon-Jones M & Sullivan CE (1996). Effect of different levels of hyperoxia on breathing in healthy subjects. *J Appl Physiol* **81**, 1683–1690.

Beecroft J, Duffin J, Pierratos A, Chan CT, McFarlane P & Hanly PJ (2006). Enhanced chemo-responsiveness in patients with sleep apnoea and end-stage renal disease. *Eur Respir J* **28**, 151–158.

Berkenbosch A, Bovill JG, Dahan A, DeGoede J & Olievier IC (1989). The ventilatory CO_2 sensitivities from Read's rebreathing method and the steady-state method are not equal in man. *J Physiol* **411**, 367–377.

Clement ID, Bascom DA, Conway J, Dorrington KL, O'Connor DF, Painter R, Paterson DJ & Robbins PA (1992). An assessment of central–peripheral ventilatory chemoreflex interaction in humans. *Respir Physiol* **88**, 87–100.

Clement ID, Pandit JJ, Bascom DA, Dorrington KL, O'Connor DF & Robbins PA (1995). An assessment of central–peripheral ventilatory chemoreflex interaction using acid and bicarbonate infusions in humans. *J Physiol* **485**, 561–570.

Corfield DR & Meadows GE (2006). Control of cerebral blood flow during sleep and the effects of hypoxia. *Adv Exp Med Biol* **588**, 65–73.

Cunningham DJC (1987). Review lecture: studies on arterial chemoreceptors in man. *J Physiol* **384**, 1–26.

Cunningham DJC, Robbins PA & Wolff CB (1986). Integration of respiratory responses to changes in alveolar partial pressures of CO_2 and O_2 and in arterial pH. In *Handbook of Physiology*, section 3, *The Respiratory System*, vol II, ed. Fishman A P, Cherniack N S & Widdicombe JG, pp. 475–528. American Physiological Society, Bethesda, MD, USA.

Duffin J (2005). Role of acid-base balance in the chemoreflex control of breathing. *J Appl Physiol* **99**, 2255–2265.

Duffin J & Mahamed S (2003). Adaptation in the respiratory control system. *Can J Physiol Pharmacol* **81**, 765–773.

Duffin J, Mohan RM, Vasiliou P, Stephenson R & Mahamed S (2000). A model of the chemoreflex control of breathing in humans: Model parameters measurement. *Respir Physiol* **120**, 13–26.

Easton PA, Slykerman LJ & Anthonisen NR (1986). Ventilatory response to sustained hypoxia in normal adults. *J Appl Physiol* **61**, 906–911.

Fatemian M, Nieuwenhuijs DJ, Teppema LJ, Meinesz S, van der Mey AG, Dahan A & Robbins PA (2003). The respiratory response to carbon dioxide in humans with unilateral and bilateral resections of the carotid bodies. *J Physiol* **549**, 965–973.

Harris DP, Balasubramaniam A, Badr MS & Mateika JH (2006). Long-term facilitation of ventilation and genioglossus muscle activity is evident in the presence of elevated levels of carbon dioxide in awake humans. *Am J Physiol Regul Integr Comp Physiol* **291**, R1111–R1119.

Honda Y (1992). Respiratory and circulatory activities in carotid body-resected humans. *J Appl Physiol* **73**, 1–8.

Kumar P & Bin-Jaliah I (2007). Adequate stimuli of the carotid body: More than an oxygen sensor? *Respir Physiol & Neurobiol* **157**, 12–21.

Leon-Velarde F, Gamboa A, Rivera ChM, Palacios JA & Robbins PA (2003). Plasticity in respiratory motor control – Selected contribution: Peripheral chemoreflex function in high-altitude natives and patients with chronic mountain sickness. *J Appl Physiol* **94**, 1269–1278.

Liang PJ, Bascom DA & Robbins PA (1997). Extended models of the ventilatory response to sustained isocapnic hypoxia in humans. *J Appl Physiol* **82**, 667–677.

Lloyd BB & Cunningham DJC (1963). A quantitative approach to the regulation of human respiration. In *The Regulation of Human Respiration*, ed. Cunningham DJC & Lloyd BB, pp. 331–349. Blackwell, Oxford.

Mahamed S & Duffin J (2001). Repeated hypoxic exposures change respiratory chemoreflex control in humans. *J Physiol* **534**, 595–603.

Mahamed S, Hanly PJ, Gabor J, Beecroft J & Duffin J (2005). Overnight changes of chemoreflex control in obstructive sleep apnoea patients. *Respir Physiol Neurobiol* **146**, 279–290.

Mateika JH, Mendello C, Obeid D & Badr MS (2004). Peripheral chemoreflex responsiveness is increased at elevated levels of carbon dioxide following episodic hypoxia in awake humans. *J Appl Physiol* **96**, 1197–1205.

Meadows GE, Kotajima F, Vazir A, Kostikas K, Simonds AK, Morrell MJ & Corfield DR (2005). Overnight changes in the cerebral vascular response to isocapnic hypoxia and hypercapnia in healthy humans: protection against stroke. *Stroke* **36**, 2367–2372.

Meadows GE, O'Driscoll DM, Simonds AK, Morrell MJ & Corfield DR (2004). Cerebral blood flow response to isocapnic hypoxia during slow-wave sleep and wakefulness. *J Appl Physiol* **97**, 1343–1348.

Mohan RM, Amara CE, Cunningham DA & Duffin J (1999). Measuring the central-chemoreflex sensitivity in man: rebreathing and steady-state methods compared. *Respir Physiol* **115**, 23–33.

Mohan R & Duffin J (1997). The effect of hypoxia on the ventilatory response to carbon dioxide in man. *Respir Physiol* **108**, 101–115.

Pandit JJ, Mohan RM, Paterson ND & Poulin MJ (2003). Cerebral blood flow sensitivity to CO_2 measured with steady-state and Read's rebreathing methods. *Respir Physiol Neurobiol* **137**, 1–10.

Poulin MJ, Fatemian M, Tansley JG, O'Connor DF & Robbins PA (2002). Changes in cerebral blood flow during and after 48 h of both isocapnic and poikilocapnic hypoxia in humans. *Exp Physiol* **87**, 633–642.

Poulin MJ, Liang PJ & Robbins PA (1996). Dynamics of the cerebral blood flow response to step changes in end-tidal PCO_2 and PO_2 in humans. *J Appl Physiol* **81**, 1084–1095.

Poulin MJ & Robbins PA (1998). Influence of cerebral blood flow on the ventilatory response to hypoxia in humans. *Exp Physiol* **83**, 95–106.

Powell FL (2007). The influence of chronic hypoxia upon chemoreception. *Respir Physiol Neurobiol* **157**, 154–161.

Read DJC & Leigh J (1967). Blood-brain tissue PCO_2 relationships and ventilation during rebreathing. *J Appl Physiol* **23**, 53–70.

Robbins PA (2007). Role of the peripheral chemoreflex in the early stages of ventilatory acclimatization to altitude. *Respir Physiol Neurobiol* (in press).

Robbins PA, Swanson GD, Micco AJ & Schubert WP (1982). A fast gas-mixing system for breath-to-breath respiratory control studies. *J Appl Physiol* **52**, 1358–1362.

Sato M, Severinghaus JW & Bickler P (1994). Time course of augmentation and depression of hypoxic ventilatory responses at altitude. *J Appl Physiol* **77**, 313–316.

Shoemaker J, Vovk A & Cunningham D (2002). Peripheral chemoreceptor contributions to sympathetic and cardiovascular responses during hypercapnia. *Can J Physiol Pharmacol* **80**, 1136–1144.

Slessarev M, Han J, Mardimae A, Prisman E, Preiss D, Volgyesi G, Ansel C, Duffin J & Fisher JA (2007). Prospective targeting and control of end-tidal CO_2 and O_2 concentrations. *J Physiol* **581**, 1207–1219.

Slessarev M, Prisman E, Ito S, Watson R, Jensen D, Preiss D, Greene R, Norboo T, Stobdan T, Diskit D, Norboo A, Kunzang M, Fisher JA, Duffin J & Appenzeller O (2007). Chemoreflex control of breathing in Himalayan and sea-level residents (Abstracts of the 15th International Hypoxia Symposium). *High Alt Med Biol* **7**, 319–350.

Somogyi RB, Preiss D, Vesely A, Fisher JA & Duffin J (2005). Changes in respiratory control after 5 days at altitude. *Respir Physiol Neurobiol* **145**, 41–52.

St. Croix CM, Cunningham DA & Paterson DH (1996). Nature of the interaction between central and peripheral chemoreceptor drives in human subjects. *Can J Physiol Pharmacol* **74**, 640–646.

Steinback CD & Poulin MJ (2007). Ventilatory responses to isocapnic and poikilocapnic hypoxia in humans. *Respir Physiol Neurobiol* **155**, 104–113.

Stuhmiller JH & Stuhmiller LM (2005). A mathematical model of ventilation response to inhaled carbon monoxide. *J Appl Physiol* **98**, 2033–2044.

Suzuki A, Nishimura M, Yamamoto H, Miyamoto K, Kishi F & Kawakami Y. (1989). No effect of brain blood flow on ventilatory depression during sustained hypoxia. *J Appl Physiol* **66**, 1674–1678.

Topor ZL, Vasilakos K, Younes M & Remmers JE (2007). Model based analysis of sleep disordered breathing in congestive heart failure. *Respir Physiol Neurobiol* **155**, 82–92.

Torrance RW (1996). Prolegomena. Chemoreception upstream of transmitters. *Adv Exper Med Biol* **410**, 13–38.

Vovk A, Cunningham DA, Kowalchuk JM, Paterson DH & Duffin J (2002). Cerebral blood flow responses to changes in oxygen and carbon dioxide in humans. *Can J Physiol Pharmacol* **80**, 819–827.

Xie A, Skatrud JB, Morgan B, Chenuel B, Khayat R, Reichmuth K, Lin J & Dempsey JA (2006). Influence of cerebrovascular function on the hypercapnic ventilatory response in healthy humans. *J Physiol* **577**, 319–329.

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