

Antioxidants Reverse Reduction of the Human Hypoxic Ventilatory Response by Subanesthetic Isoflurane

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Background: In subanesthetic concentrations, volatile anesthetics reduce the acute hypoxic response (AHR), presumably by a direct action on the carotid bodies but by an unknown molecular mechanism. To examine a possible involvement of reactive oxygen species or changes in redox state in this inhibiting effect, the authors studied the effect of antioxidants on the isoflurane-induced reduction of the AHR in humans.

Methods: In 10 volunteers, the authors studied the effect of antioxidants (intravenous ascorbic acid and oral α -tocopherol) on the reduction by isoflurane (0.12% end-tidal concentration) of the AHR on a 3-min isocapnic hypoxic stimulus (hemoglobin oxygen saturation $86 \pm 4\%$). All subjects participated in three separate sessions in which the effects of the antioxidants (session 1), placebo (session 2), and sham isoflurane plus antioxidants (session 3) were tested on the (sham) isoflurane-induced effect on the AHR.

Results: Isoflurane reduced the acute hypoxic response from $0.82 \pm 0.41 \text{ l} \cdot \text{min}^{-1} \cdot \text{\%}^{-1}$ to $0.49 \pm 0.23 \text{ l} \cdot \text{min}^{-1} \cdot \text{\%}^{-1}$ and from $0.89 \pm 0.43 \text{ l} \cdot \text{min}^{-1} \cdot \text{\%}^{-1}$ to $0.48 \pm 0.28 \text{ l} \cdot \text{min}^{-1} \cdot \text{\%}^{-1}$ in sessions 1 and 2, respectively (mean \pm SD; $P < 0.05$ vs. control). This reduction of the AHR was completely reversed by antioxidants (AHR = $0.76 \pm 0.39 \text{ l} \cdot \text{min}^{-1} \cdot \text{\%}^{-1}$; not significantly different from control, session 1) but not by placebo in session 2 (AHR = $0.50 \pm 0.30 \text{ l} \cdot \text{min}^{-1} \cdot \text{\%}^{-1}$; $P < 0.05$ vs. control). Sham isoflurane or antioxidants *per se* had no effect on the hypoxic response.

Conclusions: The data indicate that isoflurane may depress the AHR by influencing the redox state of oxygen-sensing elements in the carotid bodies. This finding may have clinical implications for patients who are prone to recurrent hypoxic episodes, e.g., due to upper airway obstruction, in the postoperative period when low-dose isoflurane may persist in the body for some time.

A RAPID increase in pulmonary ventilation in response to acute hypoxemia is an important physiologic adaptation of the human body that is initiated by oxygen-sensitive (type D) cells in the carotid bodies. A decrease in oxygen tension in the blood leads to a complex cascade of events in type I cells, in which closure of one or more classes of potassium channels results in membrane depolarization.¹⁻⁴ The mechanistic link between hypoxia and potassium channel closure is unknown; however, potassium channels are sensitive to oxidizing and reducing agents and reactive oxygen species (ROS).^{5,6} Whether ROS plays a mediating role in oxygen

sensing is controversial.⁷ At low subanesthetic doses, volatile anesthetics reduce the acute hypoxic response by 30-50%, depending on the anesthetic involved.⁸⁻¹³

The molecular mechanism by which subanesthetic volatile anesthetics reduce the hypoxic response is unresolved. Most probably, the effect is caused by a direct and preferential action on the carotid bodies.^{14,15} Volatile anesthetics increase the open probability of TASK-1 background potassium channels, thus counteracting the effect of hypoxia.¹⁶⁻¹⁹ However, halothane not only opens TASK-1 channels, but, particularly in hypoxia, also produces ROS^{20,21} that may influence the hypoxic response in a more indirect way. To investigate a potential role of the cellular redox state or ROS in the halothane-induced reduction of the acute hypoxic ventilatory response (AHR), we recently administered an antioxidant cocktail (AOX, consisting of α -tocopherol and ascorbic acid) to human volunteers and found that this treatment completely reversed the effect of halothane on the AHR.²² The more generally used volatile anesthetic isoflurane has a much lower metabolism than halothane, produces much less radical species, and, in subanesthetic concentrations, causes less depression of the hypoxic response.^{12,13,23} This raises the question as to whether the ability of AOX to reverse the effect of halothane may not apply to the isoflurane-induced depression of the AHR, indicating different mechanisms of action of both anesthetics. However, a reversal by AOX of an isoflurane-induced reduction of the AHR could indicate a general property of antioxidants to increase carotid body output when they are in a state of (pharmacologic) inhibition, and this could have potential clinical implications. Therefore, the aim of the current study in healthy volunteers was to examine the effects of AOX on reduction of the acute hypoxic response by subanesthetic isoflurane.

Materials and Methods

Subjects, Procedures, and Apparatus

Ten volunteers (five men and five women, aged 18-39 yr) participated in the protocol after approval was obtained from the human ethics committee of the Leiden University Medical Center, Leiden, The Netherlands. All subjects were healthy and did not smoke or use any illicit drugs. The subjects performed a series of test experiments to familiarize them with the apparatus and experimental procedures. The subjects did not eat or drink for at least 6 h before the experiments. They were naive with respect to respiratory physiology and anesthesia

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but were informed regarding the intention of the study and possible risks.

After arrival in the laboratory, an intravenous catheter was inserted in the right antecubital vein for drug infusion. Subsequently, electrodes (BisSensor[®]; Aspect Medical Systems, Newton, MA) for electroencephalographic monitoring using and an A-2000 electroencephalographic monitor (Aspect Medical Systems) were placed on the head (AT1-FP1). After an initial resting period (15–20 min), the antioxidant or placebo cocktail was administered. Next, a facemask was applied over nose and mouth. The mask was connected to a gas mixing system that received oxygen, carbon dioxide, and nitrogen from three mass flow controllers (Bronkhorst; Hi-Tec, Veenendaal, The Netherlands). The total flow was set to 50 l/min. The mass-flow controllers were steered by a personal computer allowing the composition of inspired and expired gas to be adjusted to force end-tidal gas tensions to follow a specific pattern in time. In this study, we applied steps in end-tidal partial pressure of oxygen (P_{O_2}) while end-tidal partial pressure of carbon dioxide (P_{CO_2}) was maintained constant. Part of the nitrogen (5 l/min) passed through a (recently calibrated) isoflurane vaporizer (Dräger 19.2, Lübeck, Germany) *via* a fourth mass flow controller. During control studies, the vaporizer was kept in the off position.

The inhaled and exhaled gas flow was measured using a pneumotachograph (Fleish, Lausanne, Switzerland) that was connected to a pressure transducer and electronically integrated to yield a volume signal. This signal was calibrated with a motor-driven piston pump (stroke volume 1 l at a frequency of 20 strokes/min). Corrections were made for the changes in gas viscosity due to changes in oxygen concentration of the inhaled gas mixture. The oxygen and carbon dioxide concentrations of inspired and expired gas were measured with a gas monitor (Multicap; Datex-Engstrom, Helsinki, Finland) that was calibrated before each experimental session with gas mixtures of known concentration delivered by a gas mixing pump (Wösthoff, Bochum, Germany). The isoflurane concentration was measured at the mouth with an anesthetic gas monitor (Capnomac Ultima; Datex-Engstrom) that was calibrated with a gas mixture of known concentration (Quick Cal; Datex-Engstrom). The arterial hemoglobin-oxygen saturation was measured *via* a finger probe (Sp_{O_2}) by pulse oximetry (Satellite Plus; Datex-Engstrom).

Inspired minute ventilation, end-tidal oxygen, carbon dioxide and isoflurane concentrations, and Sp_{O_2} were collected breath-by-breath using a custom-built data-acquisition system (RESREG) and stored on disc for further analysis.

Study Design

Each subject participated in three sessions. In session 1, the sequence of three hypoxic studies (runs) was

control hypoxia (run 1), hypoxia during isoflurane inhalation (run 2), hypoxia during isoflurane inhalation after antioxidant pretreatment (run 3); in session 2, the sequence was control hypoxia (run 1), hypoxia during isoflurane inhalation (run 2), hypoxia during isoflurane inhalation after pretreatment with placebo (run 3); and in session 3, the sequence was control hypoxia (run 1), hypoxia during sham isoflurane inhalation (run 2), hypoxia during sham isoflurane inhalation after antioxidant pretreatment (run 3). Subjects were blinded to the administration of isoflurane. With respect to the antioxidants and placebo, the study had a double-blind, randomized, crossover design with at least 2 weeks between sessions.

Hypoxic Studies

Hypoxia was induced with a “dynamic end-tidal forcing” system.¹⁰ End-tidal P_{O_2} was forced as follows: (1) 5–10 min at 109 mmHg, (2) a rapid decrease to 46.5 mmHg within 3–5 breaths, (3) 3 min at 46.5 mmHg, and (4) 5 min at hyperoxia (inspired oxygen fraction > 0.5). End-tidal P_{CO_2} was maintained 2.3 mmHg above control resting values throughout the session.

Isoflurane

During the appropriate studies, the subjects inhaled isoflurane. By manipulating the vaporizer (using an “over-pressure” technique), the end-tidal gas concentration was forced to reach 0.12% within 1 min. Next, the vaporizer was set at 1.1–1.3% (measured at its outflow tract). The flow through the vaporizer (5 l/min) was added to the fresh gas flow (45 l/min), causing a dilution of the inspired gas to 0.11–0.13% (measured at the mouth). After a 7-min equilibration period at which the end-tidal isoflurane concentration was kept at 0.12%, the hypoxic studies started. This procedure ensured that the concentrations applied were in the range as stated in table 1. The alveolar concentration was approximately 0.1 in our studies. During sham isoflurane runs, the subjects inhaled one to two breaths of 0.10–0.15% isoflurane, after which the vaporizer was set at its off position. Eight minutes later, the hypoxic study started.

Antioxidant Cocktail

The antioxidant cocktail consisted of ascorbic acid (Ascorbinezuur CF, Centrafarm, The Netherlands) given in two 1-g intravenous doses and 200 mg α -tocopherol (Organon, Oss, The Netherlands) ingested with a cup of yogurt. The α -tocopherol (or placebo) was given at least 30 min before the appropriate hypoxic study; the ascorbic acid (or placebo) was given 10 and 4 min before the hypoxic run. Placebo consisted of cellulose tablets and 0.9% NaCl manufactured by the local pharmacy.

Data and Statistical Analysis

Analysis was performed on a blinded data set. The breath-to-breath data of the last 10 breaths of normoxia

Table 1. Respiratory Variables of Sessions 1, 2, and 3

	Resting Ventilation, l/min	Resting P_{ETCO_2} , mmHg	Clamped Ventilation, l/min	Clamped P_{ETCO_2} , mmHg	Hypoxic Response, $l \cdot \text{min}^{-1} \cdot \%^{-1}$	Hypoxic Response, % of Control	End-tidal Isoflurane Concentration, %	Hypoxic SpO_2 , %
Session 1								
Control	8.6 ± 1.2	39.0 ± 3.8	10.5 ± 1.5	41.3 ± 3.0	0.82 ± 0.41	—	—	86 ± 4
Isoflurane	8.4 ± 1.1	38.3 ± 3.8	11.5 ± 2.1	41.3 ± 3.0	0.49 ± 0.23*	60 ± 12*	0.12 ± 0.01	86 ± 4
Isoflurane + antioxidants	8.9 ± 1.7	38.3 ± 3.8	10.9 ± 2.2	41.3 ± 3.0	0.76 ± 0.39	92 ± 14	0.12 ± 0.01	86 ± 5
Session 2								
Control	8.9 ± 1.4	39.0 ± 3.8	11.3 ± 2.0	41.3 ± 3.8	0.89 ± 0.43	—	—	86 ± 2
Isoflurane	8.2 ± 1.6	39.0 ± 0.4	11.8 ± 3.3	41.3 ± 3.0	0.48 ± 0.28†	54 ± 22†	0.12 ± 0.00	86 ± 2
Isoflurane + placebo	8.5 ± 1.4	39.0 ± 0.4	10.5 ± 2.5	41.3 ± 3.8	0.50 ± 0.30‡§	56 ± 20‡§	0.12 ± 0.00	85 ± 2
Session 3								
Control	9.2 ± 1.8	39.0 ± 3.0	11.4 ± 2.0	41.3 ± 3.0	0.80 ± 0.44	—	—	85 ± 2
Sham isoflurane	8.9 ± 2.2	38.3 ± 3.8	12.0 ± 2.1	41.3 ± 3.8	0.73 ± 0.36	95 ± 19	—	85 ± 3
Sham isoflurane + antioxidants	9.1 ± 1.8	38.3 ± 3.8	11.4 ± 1.7	41.3 ± 2.3	0.84 ± 0.46	103 ± 11	—	86 ± 2

* $P = 0.0013$ vs. control and isoflurane plus antioxidants (within-session analysis). † $P = 0.0016$ vs. control (within-session analysis). ‡ $P = 0.0002$ vs. control. § $P = 0.0010$ vs. run 3 of session 1 and $P = 0.0009$ vs. run 3 of session 3. All analysis of variance.

P_{ETCO_2} = end-tidal pressure of carbon dioxide; SpO_2 = hemoglobin oxygen saturation.

(before hypoxia) and of hypoxia (before hyperoxia) were averaged. Hypoxic ventilatory sensitivity (or acute hypoxic ventilatory response; $l \cdot \text{min}^{-1} \cdot \%^{-1}$) was calculated as¹⁰

$$\frac{[V_i(\text{hypoxia}) - V_i(\text{normoxia})]/[SpO_2(\text{normoxia}) - SpO_2(\text{hypoxia})]}{SpO_2(\text{hypoxia})}.$$

The statistical analysis was performed using SPSS version 11.0 for Windows (SPSS Inc., Chicago IL; 2003). To detect a significance of difference among the three treatment levels of each session, a two-way repeated-measures analysis of variance with *post hoc* Bonferroni correction was performed. To assess the effect of the antioxidant cocktail (with and without isoflurane) against placebo pretreatment, a two-way analysis of variance with *post hoc* Bonferroni correction was performed on run 3 of sessions 1, 2, and 3. Values reported are mean ± SD. P values of less than 0.05 were considered significant.

Results

All subjects completed the protocols without adverse effects. In accord with previous studies,^{12,13} we found no effect of low-dose isoflurane on resting normoxic ventilation (table 1, sessions 1 and 2) or end-tidal P_{CO_2} . As the data in table 1 show, neither sham isoflurane nor placebo or AOX (in combination with sham isoflurane) had any influence on baseline ventilation, resting end-tidal P_{CO_2} , or hypoxic ventilatory response. Prehypoxic and hypoxic end-tidal P_{CO_2} were kept at the same level (41.3 mmHg). The effects of (sham) isoflurane, antioxidants, and placebo on the hypoxic response in one individual are shown in figure 1. Isoflurane (0.12%) reduced the acute hypoxic response by approximately 40% and 46% in sessions 1 and 3, respectively, which is somewhat less than the reduction by low-dose halothane.²² Note that this inhibiting effect of isoflurane was consistent across subjects (see fig. 2, in which individual responses are shown). Hypoxic SpO_2 values in this study

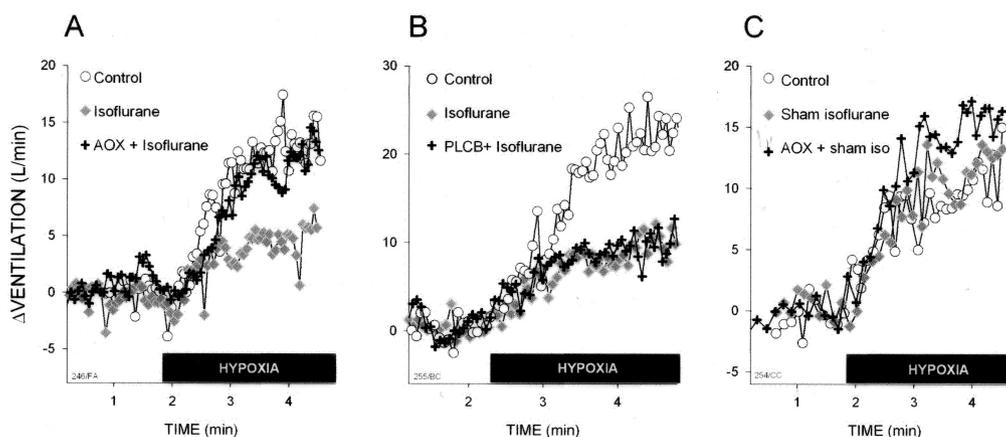


Fig. 1. Effect of (sham) isoflurane, antioxidants (AOXs), and placebo on the acute hypoxic response in one individual. (A) Depression of the hypoxic response by isoflurane and its reversal by antioxidants. (B) Isoflurane reduces the hypoxic response, but placebo (PLCB) does not reverse this. (C) Sham isoflurane or antioxidants do not significantly alter the hypoxic response.

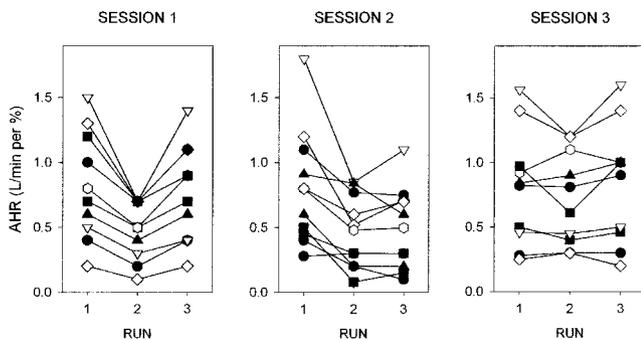


Fig. 2. Hypoxic ventilatory responses (AHRs) in 10 subjects measured during three different sessions. Session 1: run 1 = control AHR; run 2 = AHR during 0.1 minimum alveolar concentration (MAC) isoflurane inhalation; run 3: AHR during 0.1 MAC isoflurane inhalation but after pretreatment with antioxidants. Antioxidant pretreatment reverses the inhibiting effect of isoflurane on the hypoxic response in all subjects. Session 2: run 1 = control AHR; run 2 = AHR during 0.1 MAC isoflurane; run 3: AHR + isoflurane but after pretreatment with placebo. Placebo does not alter the inhibiting effect of isoflurane on the AHR. Session 3: run 1 = control AHR; run 2 = AHR during sham isoflurane; run 3: AHR during sham isoflurane but after pretreatment with antioxidant. There is no influence of sham isoflurane and antioxidant on the AHR.

were kept at 85–86%. In all subjects, administration of the AOX cocktail reversed the depression of the hypoxic response by low-dose isoflurane (session 1), whereas placebo administration (session 2) did not have this effect (table 1 and fig. 2). Similar to our previous study with sham halothane,²² during sham isoflurane conditions (session 3), some subjects showed a smaller AHR compared with control, whereas others showed a similar or somewhat larger response (fig. 3). No significant effect of AOX on the hypoxic response during sham isoflurane conditions could be detected (table 1 and fig. 2). To determine whether the reversal of the isoflurane-induced depression of the hypoxic response by AOX could be explained by a stimulatory effect of the AOX *per se* or by a placebo effect, we show the 95% confidence intervals of the isoflurane-plus-AOX effect relative to isoflurane alone in session 1, the ratio isoflurane plus placebo/isoflurane alone in session 2, and the ratio sham isoflurane plus AOX/sham isoflurane alone in session 3. Figure 3 shows that there is no overlap between these confidence intervals of session 1 on the one hand and those of session 2 and 3 on the other, so that we have no indications that the reversal by AOX of the isoflurane-induced depression of the AHR is explained by a stimulatory effect of the antioxidants *per se* or by a placebo effect.

Discussion

The novel finding of this study is that although AOXs did not have an effect on the acute hypoxic response *per se*, they reversed the depressant effect of approximately 0.1 minimum alveolar concentration (MAC) isoflurane

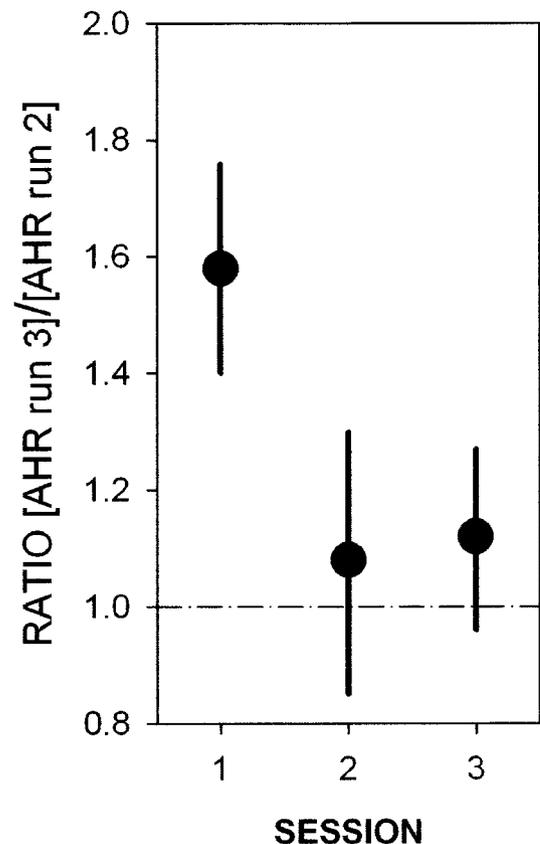


Fig. 3. Effect of antioxidants on isoflurane- or sham isoflurane-induced alterations of the acute hypoxic response (AHR). Values are the ratio of the 95% confidence intervals of the antioxidant effect relative to isoflurane in session 1, the ratio isoflurane + placebo/isoflurane in session 2, and the ratio sham isoflurane + antioxidant/sham isoflurane in session 3. A value of 1 indicates no effect of the antioxidant or placebo on the (sham) isoflurane-induced effect on the hypoxic response. The confidence intervals of session 1 do not overlap with those of session 2 or 3.

on the hypoxic response in healthy volunteers. The finding that AOXs reverse the inhibiting effects of both isoflurane and halothane²² suggests to us that despite the lack of a significant, ROS-producing metabolism, isoflurane may act by changing the redox environment of the oxygen sensor and that there is no compelling reason to explain the effect of halothane by its potential reductive metabolism.

The design of the current study differed from that of the previous one²² in that the effects of isoflurane, antioxidants, placebo, and sham isoflurane were all measured within the same subjects (divided over three sessions; note the low intraindividual variability in resting parameters between these sessions; table 1). Furthermore, because we did not expect any effect of the combination sham isoflurane and placebo (also, see our previous data²²), we decided not to include a fourth session. To avoid confounding effects of variations in P_{CO_2} , the clamped end-tidal P_{CO_2} values were kept constant at the same levels between runs and sessions (table

1; note also that hypoxic saturation levels were kept at equal levels).

Because variations in arousal level could be a confounding factor in the measurement of a reflex that participates in the metabolic control of ventilation, (*i.e.*, behavioral control superimposed on metabolic control¹²), we measured the raw electroencephalogram in our subjects. Similarly to our previous studies, we observed no electroencephalographic changes indicative for a sleep state or otherwise depressed central nervous system condition (during runs in which [sham] isoflurane was given, all subjects opened their eyes when spoken to in a soft voice). The lack of any signs indicative of central nervous system depression in the presence of a pronounced inhibiting effect on the acute hypoxic response by low-dose isoflurane is in accord with the view that this depressant effect is due to a preferential action on the carotid bodies.^{14,15} Because of the short equilibration period, the brain isoflurane concentration was less than 0.12% (brain equilibration will not be reached before 20–25 min after exposure),¹³ minimizing the occurrence of central effects of the anesthetic.

From figure 2, it is clear that our subjects showed a large intersubject variability of their acute hypoxic responses. A 7- to 10-fold variability of hypoxic sensitivity has been described previously, and it is thought that this wide intersubject variation may be related to genetic factors.^{24,25}

Because we did not measure plasma antioxidant concentrations, we cannot ascribe the effect of our AOX cocktail to one of its components. Our initial purpose was to use a cocktail because of the known synergism between ascorbic acid and α -tocopherol.²² However, given the slow absorption of α -tocopherol by the digestive tract,²⁶ we tend to consider an extracellular effect of the water-soluble ascorbate as the most likely contributor to the effect of our AOX cocktail.

The generally accepted model for oxygen sensing is that low oxygen decreases the open probability of potassium channels in the membrane of carotid body type I cells, which results in depolarization, opening of voltage-gated calcium channels, subsequent influx of Ca^{2+} ions, and a release of neurotransmitters (probably acetylcholine, adenosine triphosphate, or both) that activate nearby afferent fibers of the carotid sinus nerve.^{1–4} In several species, various types of potassium channels are described that may serve as an oxygen-sensing element to initiate the transduction cascade, *e.g.*, K_v channels in rabbit^{27,28} and Maxi-K and TASK channels in rat.^{19,29} It is unknown whether potassium channels have intrinsic oxygen sensitivity, *e.g.*, by possessing a heme moiety. Alternative candidates as actual oxygen sensors—without excluding the role of potassium channels in causing the depolarization—are membrane-bound heme-containing protein complexes such as nicotin-

amide adenine dinucleotide phosphate oxidase (possibly closely associated to potassium channels) or enzymes belonging to the respiratory chain in the mitochondria such as cytochrome *c*-oxidase or enzymes belonging to other complexes.^{1–4} A mitochondrial key role for oxygen sensing and a crucial role for potassium channels are not mutually exclusive: In carotid body cells, a key oxygen sensor in the mitochondria may be functionally linked with K^+ channels.³⁰

One possible mechanism by which volatile anesthetics may reduce the hypoxic response is by a direct effect on potassium channels. This may also offer an explanation of the effect of AOX: (Various) potassium channels not only serve as target for anesthetics but also possess redox sensitivity.^{5,6} In recent years, much interest has been focused on two-pore domain (TASK) potassium channels. By unknown molecular mechanisms, low oxygen reduces the open probability of TASK-1 channels in neonatal rat carotid body cells, whereas halothane directly opens them in these¹⁹ and other cell types.^{16–18} As previously discussed,²² AOX could alter the binding site of halothane/isoflurane to these channels by an unknown mechanism. Alternatively, AOX and isoflurane/halothane could influence potassium channel gating independently: the anesthetics by binding to a specific intracellular binding domain^{16,31} and AOX by changing the redox state of the channel or its environment. Consequently, the inhibiting effects of halothane/isoflurane would not necessarily imply the involvement ROS. Redox sensitivity of the channel limited to a hypoxic condition could explain the absence of an effect of the anesthetics on normoxic ventilation. Other possible targets of halothane/isoflurane could be K_v channels³² or tandem pore domain halothane-inhibited K^+ (THIK)-like channels that have recently been identified in neurons of the glossopharyngeal nerve.³³ By release of the radical species NO (and possibly closing of THIK channels), these cells could play a modulating, negative feedback role on chemoreceptor activity during hypoxia.³³ Inhibition of G protein-coupled inwardly rectifying potassium channels by volatile anesthetics has also been demonstrated,³⁴ but it remains to be studied whether this may play a role in their inhibiting effects on the carotid bodies.

Another possible mechanism by which anesthetics might be able to reduce the hypoxic response is by inhibition of the mitochondrial electron transport chain. Recently, Hanley *et al.*³⁵ reported that halothane and isoflurane inhibit complex I (nicotinamide adenine dinucleotide: ubiquinone oxidoreductase) of the mitochondrial electron transport chain, whereas halothane but not isoflurane also inhibited complex II (succinate dehydrogenase). The effects of halothane and isoflurane on complex I were dose-dependent, resulting in a reduction in activity by 20% at 2 MAC.³⁵ Inhibition of the electron transport chain by isoflurane may result in an altered

production of ROS, which then may influence the oxygen-sensing mechanism.⁴ Upstream (complex I) inhibitors such as rotenone and diphenylene iodonium are known to decrease the concentration of ROS in carotid body cells, but both complex III and IV inhibitors (e.g., antimycin A and cyanide, respectively) increase [ROS].⁷ All uncouplers, however, have a stimulatory action on the carotid bodies^{7,36,37} but, depending on the completeness of electron transport inhibition, may occlude the hypoxic response.³⁸ Because we showed that AOX abrogated the inhibiting effect of isoflurane on the hypoxic response, the question arises as to whether a potential increase in mitochondrial ROS by the anesthetic could be involved in this inhibiting effect. However, because in animal studies a decrease in carotid sinus nerve baseline activity by volatile anesthetics has not been shown^{39,40} and an increase in carotid body mitochondrial ROS by low-dose anesthetics has not been demonstrated, we do not tend to believe that a halothane/isoflurane concentration as low as 0.15 MAC could be responsible for sufficient inhibition of the mitochondrial electron transport chain in the human carotid body to reduce the acute hypoxic response by half.

Our findings may be relevant to patients recovering from anesthesia. After inhalational anesthesia, the concentration of residual anesthetic in the postoperative period may be equal to or larger than used in this study. After exposure to 1 MAC for 1 h, the brain concentration of the volatile anesthetics halothane, isoflurane, and enflurane can be considerably greater than 0.1 MAC for more than 1 h after discontinuation.⁴¹ After anesthesia with 1 MAC halothane, a significant reductive metabolism of this agent has been shown to occur for more than 9 h.⁴² Both in the early and the late (second and third postoperative nights) postoperative periods, hypoxemic episodes are not uncommon.^{43,44} By monitoring the (anti)oxidant status in the clinical setting, it would be interesting to see whether predictions could be made (and precautions taken) as to a patient's ability to respond to hypoxemia in the presence of residual anesthetic. In this context, it is interesting that in women aged 60–80 yr, ascorbic acid supplementation has been found to increase the hypoxic response by 44%.⁴⁵ It remains to be determined whether therapeutic intervention with antioxidants could provide a nonharmful and efficient means to lower the risk of long and recurrent episodes of hypoxia in postoperative patients.

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