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RESEARCH ARTICLE

Safety, hemodynamic effects, and detection of acute xenon inhalation: rationale for banning xenon from sport

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Lawley JS, Gatterer H, Dias KA, Howden EJ, Sarma S, Cornwell WK 3rd, Hearon CM Jr, Samels M, Everding B, Bruick RK, Hendrix M, Piper T, Thevis M, Levine BD. Safety, hemodynamic effects, and detection of acute xenon inhalation: rationale for banning xenon from sport. *J Appl Physiol* 127: 1511–1518, 2019. First published August 15, 2019; doi:10.1152/japplphysiol.00290.2019.—This study aimed to quantify the sedative effects, detection rates, and cardiovascular responses to xenon. On 3 occasions, participants breathed xenon (F_iXe 30% for 20 min; F_iXe 50% for 5 min; F_iXe 70% for 2 min) in a nonblinded design. Sedation was monitored by a board-certified anesthesiologist. During 70% xenon, participants were also verbally instructed to operate a manual value with time-to-task failure being recorded. Beat-by-beat hemodynamics were measured continuously by ECG, photoplethysmography, and transcranial Doppler. Over 48 h postadministration, xenon was measured in blood and urine by gas chromatography-mass spectrometry. Xenon caused variable levels of sedation and restlessness. Task failure of the self-operating value occurred at 60–90 s in most individuals. Over the first minute, 50% and 70% xenon caused a substantial reduction in total peripheral resistance ($P < 0.05$). All dosages caused an increase in cardiac output ($P < 0.05$). By the end of xenon inhalation, slight hypertension was observed after all three doses ($P < 0.05$), with an increase in middle cerebral artery velocity ($P < 0.05$). Xenon was consistently detected, albeit in trace amounts, up to 3 h after all three doses of xenon inhalation in blood and urine with variable results thereafter. Xenon inhalation caused sedation incompatible with self-operation of a breathing apparatus, thus causing a potential life-threatening condition in the absence of an anesthesiologist. Yet, xenon can only be reliably detected in blood and urine up to 3 h postacute dosing.

NEW & NOTEWORTHY Breathing xenon in dosages conceivable for doping purposes (F_iXe 30% for 20 min; F_iXe 50% for 5 min; F_iXe 70% for 2 min) causes an initial rapid fall in total peripheral resistance with tachycardia and thereafter a mild hypertension with elevated middle cerebral artery velocity. These dose duration intervals cause sedation that is incompatible with operating a breathing apparatus and can only be detected in blood and urine samples with a high probability for up to ~3 h.

anesthesia; blood pressure; brain blood flow; doping

INTRODUCTION

Recently, the sporting press has speculated on the potential benefits and use of xenon inhalation by Russian athletes to improve endurance performance (5). Xenon is an inert, odorless gas and is, theoretically, not metabolized to toxic metabolites (10, 13), yet xenon preconditioning has convincingly been shown to stabilize hypoxia-inducible factor 1α and hypoxia-inducible factor 2α and elevated kidney erythropoietin in animal models (7, 14, 15, 21, 32). In a recent human clinical trial, breathing F_iXe 30% xenon for 45 min caused small yet consistent increases in plasma erythropoietin for several days after xenon administration (26).

Based on the World Anti-Doping Code, a substance enters the prohibited list if at least two out of the following three criteria are met: *1*) it has the potential to enhance or enhances sport performance; *2*) represents an actual or potential health risk to the athlete; and *3*) violates the spirit of sport. At high concentrations [~63%−71%, 1 minimum alveolar concentration (MAC)], xenon is a known and potent anesthetic agent (4, 8, 13); hence, in the absence of a trained anesthesiologist, the risk of anesthesia is clear. However, scientific data on the level of sedation at different doses and acute inhalation times from a nonpatient-centered viewpoint are lacking, especially as it pertains to dubious doping practices.

From a cardiovascular point of view, xenon inhalation is mostly associated with stable hemodynamics (1, 11), albeit slight hypertension (22) and a reduction in cerebral blood flow (12) are reported during anesthesia. Yet, scientific data on the integrative time course of these changes to different acute doses of xenon from a doping point of view are not clear.

Finally, the ability to detect xenon doping is paramount to effective doping controls. Because of its low solubility, xenon is eliminated quickly, yet traceable levels have been observed consistently up to 24 to 48 h after xenon-based anesthesia (23, 27) and after prolonged inhalation times (26) with the use of a closed-circuit anesthesia machine. Whether these detection rates remain after xenon doping with very short open-circuit

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breathing systems more likely to be used by athletes is unknown.

Thus, this study aimed to quantify the sedative effects, detection rates, and cardiovascular responses to increasing doses of xenon delivered by an open-circuit breathing system, which feasibly would be used by athletes for doping [F_iXe 30% for 20 min (~0.4 MAC); F_iXe 50% for 5 min (~0.7 MAC); F_iXe 70% for 2 min (1 MAC)].

METHODS

Study 1

Participants. Healthy male ($n = 6$, age: 31 ± 3 yr, height: 180 ± 5 cm, weight: 85 ± 13 kg) and female ($n = 2$, age: 33 ± 1 yr, height: 176 ± 2 cm, weight: 68 ± 0 kg) volunteers took part in the study. Informed written consent was obtained after each participant was given a verbal and written explanation of the experimental protocol and fully understood the possible risks involved in taking part in the study. Participants were screened for evidence of chronic disease with a history and physical examination along with a 12-lead ECG and echocardiography. Urine tests in women confirmed the absence of pregnancy. The study protocol was approved by the Federal Food and Drug Administration (FDA investigation new drug application 125194) and the institutional review committee at the University of Texas Southwestern Medical Center and followed the principles from the Declaration of Helsinki. A Data Safety and Monitoring Board was established and consisted of three anesthesiologists (Dr. Charles Whitten, M.D.; Dr. Michael J. Joyner, M.D.; Dr. Thomas Hornbein, M.D.) who were familiar with xenon administration and were experienced in clinical research. To ensure participant safety, the Data Safety and Monitoring Board implemented a sequential procedure for monitoring each dose of xenon, studying four participants at a time before moving on to the next higher dose. Thus, these trials were not randomized.

Experimental design. On three occasions separated by a washout period of at least 2 wk, participants breathed a single dose of increasing concentrations of xenon in a nonblinded design. Medical-grade xenon [F_iXe 30% for 20 min (~0.4 MAC); F_iXe 50% for 5 min (~0.7 MAC); F_iXe 70% for 2 min (~1 MAC)] and oxygen ($F_{I_{O_2}}$ 21%) with the balance nitrogen was used for all breathing procedures. The inspired oxygen concentration was kept at 21% to maintain normoxic oxygen saturation. These concentrations were chosen based on speculative potential dosing strategies, as direct communication with Russian trainers involved in delivering xenon to Russian athletes led to no conclusive insight into dosing regimens but suggested that the concentration may range from 30% to 70% xenon (personal communication). The MAC of an inhaled anesthetic is the point at which 50% of individuals would not respond to a surgical incision. For xenon specifically, 1 MAC is ~63%–71%. Although a number of factors (e.g., age, gender, hypothermia, hyperthermia, circadian rhythm, and use of other stimulants/depressants) can affect the MAC, the depth of anesthesia at or below the MAC is critically dependent on time. As our aim was to test a range of xenon concentrations, the inhalation time was reduced with increasing concentrations to avoid anesthesia. These factors were not taken into account on an individual basis, but given the results (Fig. 2*B*), our strategy seemed effective, as despite sedation, no subjects were anesthetized.

OPEN-CIRCUIT XENON BREATHING. Xenon was administered through an open-circuit breathing system in which a 250-liter Douglas bag (filled with the appropriate dose inspirate) was attached to a 2-way Hans Rudolph valve and leak-free facemask via a 1-m length of Falconia tubing. Participants lay in a semi-recumbent position under the direct supervision of a board-certified anesthesiologist (Fig. 1). Although the xenon doses administered were subanesthetic (≤1 MAC), subjects were required to withhold oral intake of food for 8 h before their dosing time, during which time they were permitted to ingest clear liquids only. For the 2 h preceding their dosing time, they were required to withhold oral intake of all liquids. After xenon inhalation, participants were instructed not to drive home, to be accompanied by a responsible adult when using public transportation (taxi, bus, train), and to not be alone the first night at home.

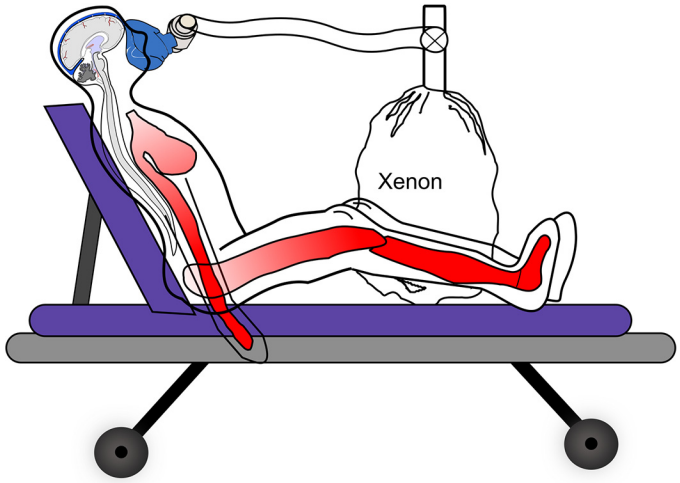

Fig. 1. Schematic overview of open-circuit rebreathing system used to deliver xenon. Participants wore a leak-free facemask connected to a two-way respiratory valve (Hans Rudolph, Shawnee, KS) with the inspiratory port connected to a 150 l Douglas bag containing 21% O_2 with either 30%, 50%, or 70% xenon.

Measurements. SYMPTOMS RATING. The level of sedation was monitored before, during and 1 h after xenon inhalation by clinical interview using standard symptom questionnaires (Richmond Agitation and Sedation Scale and the Continuum of Depth of Sedation Scale) by a trained anesthesiologist.

HEMODYNAMICS. All beat-by-beat hemodynamic parameters were obtained continuously for 10 min before xenon inhalation, throughout xenon inhalation, and after 1 h of recovery. Heart rate was determined from three-lead ECG (Tram-rac 4A, Marquette). Beat-by-beat arterial pressure was assessed noninvasively by using finger photoplethysmography (Nexfin; BMEYE, the Netherlands) corrected to cuff pressure with changes in cardiac stroke volume calculated using the Modelflow method (24, 29). Cardiac output was calculated as the product of heart rate and stroke volume, and total peripheral resistance was calculated as mean arterial blood pressure divided by cardiac output. Cerebral blood flow was also estimated from continuous recordings of middle cerebral artery velocity (ROBOTC2MD, Multigon Industries, New York, NY). To locate the middle cerebral artery, the sample volume was initially placed at a depth of 55 mm while performing a classic transcranial searching pattern. After identification of the middle cerebral artery, simultaneous multidepth Doppler and M-modes were used to scan the length of the middle cerebral artery and identify the optimal depth. The probe was fixed in place at a constant angle with a headset. The cerebral vascular conductance index was calculated as mean arterial pressure divided by middle cerebral artery blood flow velocity to assess changes in cerebral vascular function. Oxygen saturation was measured continuously by pulse finger oximetry (Nellcor N-595 OxiMax, Puritan Bennett). Collection of expired gas was obtained over 1 min using the Douglas bag technique at baseline and during the final minute of each xenon dose and ventilatory volumes measured by a Tissot spirometer. All continuous hemodynamic variables were recorded as analog data at 250 Hz, transferred to a laptop using a standard analog-to-digital converter (MP150, Biopac), and analyzed via commercially available software (Acknowledge 4.0). Data were analyzed over 5 min at baseline and then over the final 30 s of each 5-min period during 30%

xenon inhalation and at the end of each minute period during 50% and 70% xenon inhalation.

XENON DETECTION. Venous blood was drawn from an antecubital intravenous catheter at baseline (in duplicate) just before the end of xenon inhalation and 1, 3, 6, 24, and 48 h after xenon inhalation. A urine sample was also obtained at all time points, except at the end of xenon inhalation. Venous blood and urine were stored in sealed vacutainers at 4°C and shipped to the Institute of Biochemistry/Center for Preventive Doping Research (Cologne, Germany) on dry ice and analyzed by gas chromatography as detailed previously (23, 27, 28). The detection limit for this technique is 0.25 μmol/L.

Study 2

Healthy male and female participants (men: $n = 6$, age: 29 ± 3 yr, height: 180 ± 3 cm, weight: 84 ± 10 kg; women: $n = 1$, age: 34 ± 0 yr, height: 177 ± 0 cm, weight: 68.0 ± 0.0 kg) were given a 2-way Hans Rudolph value and instructed via the command "bag off" to turn the valve 1 quarter turn every 30 s during 2 min of the highest dose of xenon inhalation [F_iXe 70% for 2 min (~1 MAC)]. The final time point when each participant was capable of turning the valve was taken as time-to-task failure.

Statistical Analyses

One male participant (the oldest participant) was removed during the 30% xenon trial because of severe sensation of nausea and fatigue (final $n = 7$). The level of agitation-sedation (delta change) was compared by a single-factor one-way repeated measures analysis of variance with follow-up t tests. During the 50% xenon trial, 2 participants were discontinued early (~4.30 min because of observable agitation); agitation-sedation scores were obtained at this point. During the 50% xenon trial, 1 participant's hemodynamic data was lost because of an electrical fault. Each dose was examined by separate one-way linear mixed models with random effects with follow-up planned comparisons to baseline. Ventilation was compared at baseline and at the end of inhalation by paired t test. Xenon concentrations were statistically compared by separate single-factor one-way analyses of variance with follow-up t tests. Single-factor analyses of variance and paired t test were performed on Prism version 7 for Windows. Linear mixed models were carried out on Statistical Analysis System (JMP 12; SAS Institute Inc. Cary, NC) for Windows. All statistical significance was established at $P \leq 0.05$.

RESULTS

Safety of Suspected Xenon Doping Practices

All three doses of xenon caused drowsiness and sedation in all participants. Subjectively, 50% xenon did cause substantial restlessness and agitation in a number of participants, which did not occur in the 30% or 70% condition (Fig. 2*A*). Compared with 30% (20 min) xenon, both 50% and 70% xenon inhalation caused a greater depth of sedation (ANOVA, $P = 0.04$, Fig. 2*B*). In *study 2*, only 1 participant was capable of self-operating a 2-way valve at the end of 70% xenon inhalation (Fig. 2*C*).

Hemodynamics of Suspected Xenon Doping Practices

30% inspired fraction of xenon for 20 min. During 30% xenon, heart rate, stroke volume, and cardiac output remained unchanged through the inhalation procedure. However, heart rate (ANOVA, $P = 0.07$) and cardiac output (ANOVA, $P = 0.01$) were slightly lower after 60 min of recovery. Thirty percent xenon caused an increase in total peripheral resistance (ANOVA, $P = 0.06$) and mean arterial blood pressure (ANOVA, $P = 0.01$). Because of the increase in mean arterial blood pressure, middle cerebral artery velocity (ANOVA, $P = 0.01$) was elevated from 5 to 20 min of xenon inhalation, although an increase in cerebral vascular conductance index, and thus cerebral vasodilation, was also observed (ANOVA, $P = 0.02$). Ventilation ($P = 0.99$) was unchanged at the end of 30% xenon inhalation, and oxygen saturation was maintained throughout (ANOVA, $P = 0.07$, $P = 0.13$, see Table 1 for an overview).

50% inspired fraction of xenon for 5 min. During 50% xenon, heart rate increased (ANOVA, $P = 0.01$), whereas stroke volume was unchanged (ANOVA, $P = 0.58$), causing an increase in cardiac output (ANOVA, $P = 0.01$) during inhalation. Total peripheral resistance (ANOVA, $P = 0.08$) fell in the first minute and thereafter was maintained at or slightly below baseline values throughout the 5 min. Mean (ANOVA, $P = 0.01$), systolic (ANOVA, $P = 0.04$), and diastolic (ANOVA, $P = 0.02$) arterial blood pressures were elevated for

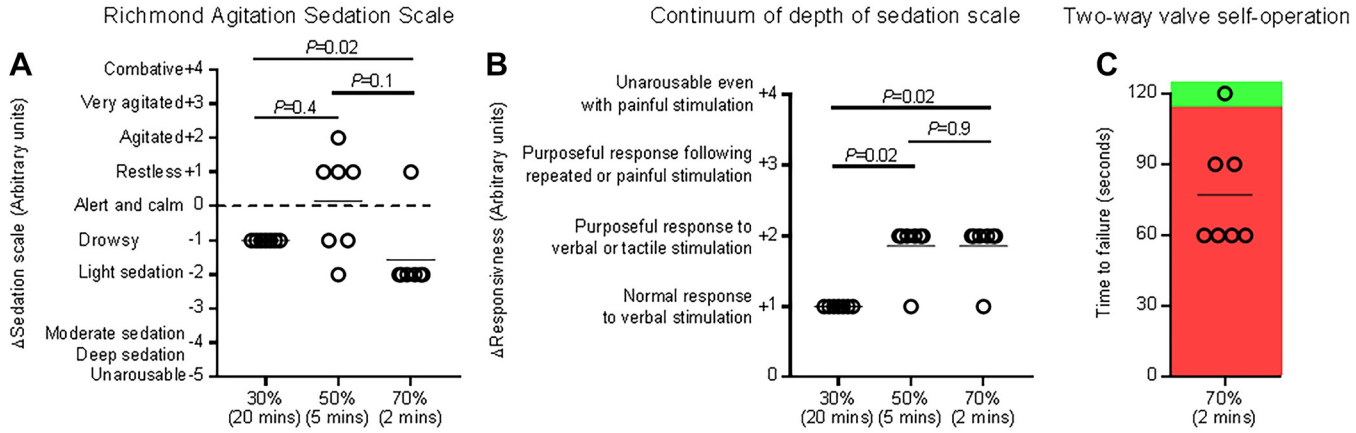

Fig. 2. Sedative properties of acute xenon inhalation. Degree of sedation as measured by the Richmond Agitation-Sedation scale (*A*) and sedation by the continuum of depth of sedation (*B*) at the end of 30%, 50%, and 70% xenon inhalation. Note the time in parentheses on both *x*-axes correspond to the length on xenon inhalation. Time-to-task failure using a two-way Hans Rudolph valve as an objective indication to self-operated breathing apparatus (*C*). Red shaded area indicates failure to self-operate that value, which could result in suffocation. $n = 7$ in *study 1* (*A* and *B*) and *study 2* (*C*), one-way single-factor ANOVA with follow-up t tests.

Table 1. *Hemodynamics of subanesthetic 30% xenon inhalation*

	F_iXe 30% (20 min)					
	Baseline	5 min	10 min	15 min	20 min	Recovery
Heart rate, beats/min	61 ± 8	64 ± 11	60 ± 7	61 ± 8	59 ± 6	55 ± 7
Stroke volume, mL	110 ± 11	114 ± 12	113 ± 13	110 ± 14	109 ± 17	108 ± 15
Cardiac output, liters	6.8 ± 0.8	7.3 ± 1.0	6.8 ± 0.8	6.7 ± 1.0	6.4 ± 0.7	5.9 ± 0.8*
Total peripheral resistance, $mmHg \cdot L^{-1} \cdot min^{-1}$	14.6 ± 2.1	14.7 ± 2.6	15.9 ± 2.7	16.2 ± 1.9#	16.4 ± 2.0*	16.7 ± 2.8*
Mean arterial blood pressure, mmHg	95 ± 11	102 ± 16*	103 ± 17*	103 ± 18*	101 ± 18*	96 ± 14
Systolic blood pressure, mmHg	134 ± 16	143 ± 26	142 ± 24	142 ± 28	139 ± 31	135 ± 19
Diastolic blood pressure, mmHg	76 ± 7	81 ± 12*	83 ± 11*	84 ± 10*	82 ± 12*	77 ± 7
Middle cerebral artery velocity, cm/s	46 ± 10	56 ± 13*	58 ± 9*	55 ± 13*	57 ± 11*	46 ± 10
Cerebral vascular conductance index, $mmHg \cdot cm^{-1} \cdot s^{-1}$	0.49 ± 0.15	0.56 ± 0.19	0.59 ± 0.16	0.56 ± 0.22	0.57 ± 0.17	0.49 ± 0.14
Oxygen saturation, %	99 ± 1	99 ± 1	99 ± 1	98 ± 2	98 ± 1	99 ± 1
Ventilation, L/min^{-1}	9.3 ± 2.7				9.4 ± 3.2	

Data are means ± SD. $n = 7$, one-way linear mixed models with follow-up planned contrasts. *$P \leq 0.05$; #$P \leq 0.1$ (trend toward statistical significance) compared with baseline.

most of the 5 min at 50% xenon. The increase in mean blood pressure caused an elevation in middle cerebral artery velocity (ANOVA, $P = 0.01$) throughout 50% xenon inhalation, although again an increase in cerebral vascular conductance index and thus cerebral vasodilation was observed (ANOVA, $P = 0.03$). Ventilation ($P = 0.01$) was increased at the end of 50% xenon inhalation, and oxygen saturation was maintained throughout xenon inhalation (ANOVA, $P = 0.99$, see Table 2 for an overview).

70% inspired fraction of xenon for 2 min. During 70% xenon, heart rate increased (ANOVA, $P = 0.01$), whereas stroke volume was unchanged (ANOVA, $P = 0.61$), causing an increase in cardiac output (ANOVA, $P = 0.01$) between 1 and 2 min. Total peripheral resistance (ANOVA, $P = 0.01$) fell in the first minute and returned to baseline values after 2 min. Mean (ANOVA, $P = 0.01$), systolic (ANOVA, $P = 0.01$), and diastolic (ANOVA, $P = 0.01$) arterial blood pressure were elevated throughout 70% xenon. Again, the increase in mean blood pressure caused an elevation in middle cerebral artery velocity (ANOVA, $P = 0.01$) throughout 70% xenon inhalation. However, although an increase in cerebral vascular conductance index was also observed in 6 of the 7 participants after 2 min at 70% xenon inhalation, it did not reach statistical significance (ANOVA, $P = 0.17$), likely because of 1 participant having a pronounced fall in conductance index. Ventilation ($P = 0.03$) was increased at the end of 70% xenon inhalation, and oxygen saturation was maintained throughout xenon inhalation (ANOVA, $P = 0.01$, see Table 3 and Fig. 3 for an overview).

Detection of Suspected Xenon Doping Practices

During xenon inhalation, plasma xenon concentration increased in all three trials, confirming our experimental design (Fig. 4, *A*, *C*, and *E*).

Wtih an open-circuit non-rebreathing system, 30% xenon inhalation could be detected in plasma (ANOVA, $P = 0.05$) and urine (ANOVA, $P = 0.02$) up to 6 h after xenon administration (Fig. 4, *A* and *B*) in 100% of cases. Moreover, in plasma, xenon could be detected in ~50% of cases 24 h after 30% xenon inhalation (overview in Fig. 4*A*). Xenon inhalation (50%) could be detected in plasma (ANOVA, $P = 0.01$) and urine (ANOVA, $P = 0.01$) up to 6 h post-xenon administration (Fig. 4, *C* and *D*) in ~70%–80% of cases. Xenon could not be detected in plasma or urine at 24 h post-50% xenon inhalation (overview in Fig. 4, *C* and *D*). Xenon inhalation (70%) could be detected in plasma (ANOVA, $P = 0.01$) up to 3 h post-xenon administration (Fig. 4, *E* and *F*) in ~80% of cases. Only one participant had detectable plasma xenon 6 h postinhalation (Fig. 4*E*). Interestingly, xenon could be detected in urine up to 6 h after 70% xenon inhalation in 80% of cases, although the

Table 2. *Hemodynamics of subanesthetic 50% xenon inhalation*

	F_iXe 50% (5 min)						
	Baseline	1 min	2 min	3 min	4 min	5 min	Recovery
Heart rate, beats/min	64 ± 15	89 ± 24*	82 ± 27*	78 ± 22#	83 ± 20*	77 ± 16#	57 ± 9
Stroke volume, mL	113 ± 11	109 ± 23	109 ± 20	107 ± 21	114 ± 17	123 ± 24	108 ± 11
Cardiac output, liters	7.2 ± 1.5	9.7 ± 2.3*	8.7 ± 2.5	10.0 ± 3.2*	9.5 ± 1.5#	9.4 ± 0.8#	6.2 ± 0.9
Total peripheral resistance, $mmHg \cdot L^{-1} \cdot min^{-1}$	13.4 ± 2.0	10.9 ± 3.2#	12.8 ± 3.2	11.7 ± 3.0	11.7 ± 1.4	11.0 ± 1.9	15.1 ± 1.6
Mean arterial blood pressure, mmHg	92 ± 10	99 ± 18	102 ± 12*	104 ± 11*	102 ± 8*	94 ± 11#	88 ± 5
Systolic blood pressure, mmHg	129 ± 11	137 ± 24	141 ± 14#	142 ± 15#	144 ± 13#	135 ± 15	123 ± 8
Diastolic blood pressure, mmHg	73 ± 5	80 ± 13#	83 ± 5*	85 ± 6*	80 ± 9#	74 ± 6	71 ± 5
Middle cerebral artery velocity, cm/s	50 ± 6	59 ± 5*	67 ± 8*	65 ± 10*	66 ± 13*	63 ± 10*	48 ± 9
Cerebral vascular conductance index, $mmHg \cdot cm^{-1} \cdot s^{-1}$	0.54 ± 0.11	0.61 ± 0.17#	0.64 ± 0.13*	0.61 ± 0.13	0.61 ± 0.13#	0.63 ± 0.15*	0.52 ± 0.1
Oxygen saturation, %	99 ± 1	100 ± 0	100 ± 1	100 ± 0	100 ± 0	99 ± 1	98 ± 1
Ventilation, L/min	8.9 ± 2.8					12.3 ± 4.0*	

Data are means ± SD. $n = 6$; one-way linear mixed models with follow-up planned contrasts. *$P \leq 0.05$; #$P \leq 0.10$ (trend toward statistical significance) compared with baseline.

Table 3. *Hemodynamics of subanesthetic 70% xenon inhalation*

	F_iXe 70% (2 min)			
	Baseline	1 min	2 min	Recovery
Heart rate, beats/min	65 ± 11	93 ± 16*	83 ± 18*	57 ± 8
Stroke volume, mL	111 ± 13	112 ± 21	108 ± 19	107 ± 8
Cardiac output, liters	7.2 ± 1.0	10.5 ± 1.6*	8.9 ± 1.5*	6.2 ± 0.8
Total peripheral resistance, $mmHg \cdot L^{-1} \cdot min^{-1}$	14.2 ± 2.6	11.2 ± 3.0*	13.9 ± 2.2	15.3 ± 1.5
Mean arterial blood pressure, mmHg	96 ± 9	107 ± 12*	115 ± 13*	89 ± 10
Systolic blood pressure, mmHg	136 ± 18	152 ± 21*	160 ± 23*	123 ± 14
Diastolic blood pressure, mmHg	76 ± 9	84 ± 15*	92 ± 14*	71 ± 7
Middle cerebral artery velocity, cm/s	46 ± 6	52 ± 9*	62 ± 12*	48 ± 6
Cerebral vascular conductance index, $mmHg \cdot cm^{-1} \cdot s^{-1}$	0.46 ± 0.1	0.47 ± 0.1	0.52 ± 0.1#	0.52 ± 0.1
Oxygen saturation, %	99 ± 1	100 ± 1#	100 ± 0*	98 ± 1
Ventilation, L/min	8.5 ± 1.7		12.3 ± 4.4*	

Data are means ± SD. $n = 7$, one-way linear mixed models with follow-up planned contrasts. *$P \leq 0.05$; #$P \leq 0.1$ (trend toward statistical significance) compared with baseline.

analysis of variance failed to reach classical statistical significance (ANOVA, $P = 0.2$).

DISCUSSION

The main findings of the present study were: *1*) subanesthetic doses of xenon, which could feasibly be used by athletes for doping (xenon 30%, 20 min; 50%, 5 min; 70% 2 min), cause sedation; *2*) 50% and 70% xenon cause a consistent acute fall in total peripheral resistance (~1 min after administration), with an increase in cardiac output and ensuing (mild) hypertension throughout administration (these responses normalized 1 h postadministration); and *3*) with these dosing regimens, xenon can be consistently detected in blood plasma and urine for only up to 3 h postadministration.

Safety of Suspected Xenon Doping Practices

Based on experimental (4, 9, 30, 31) and clinical observations, we chose xenon concentrations and delivery times that were all subanesthetic. Indeed, previous investigations have documented the general tolerability of acute xenon breathing (9, 30, 31), and none of our participants became anesthetized.

This being said, while breathing 30% xenon, participants were very drowsy with a sense of euphoria, which is consistent with previous observations noting the hypnotic effect of low-dose (0.3 MAC) xenon inhalation (30). Moreover, a number of participants became subconsciously restless and agitated during 50% xenon, which was not observed during 70% xenon, likely because of the faster sedative effect and reduced inhalation time. Despite the substantially reduced inhalation times, 50% and 70% xenon caused a greater depth of sedation compared with 30% xenon, which was expected, as 50% and 70% xenon inspirate is anticipated to cause an alveolar concentration close to 1 MAC (3, 16).

Despite these variable levels of sedation and agitation, from subjective reporting, all participants felt a heightened sense of euphoria during xenon inhalation, and it is doubtful any would have been competent to independently operate equipment (valves/stopcocks) necessary to perform open- or closed-circuit breathing systems at any dose. These observations argue in favor of the World Anti-Doping Agency's decision to ban xenon, as in the absence of a certified medical professional, inhaling xenon may cause a severe life-threatening situation. It is also worth mentioning that one participant complained of a severe sense of nausea, and a number of participants subjectively commented that they felt fatigued to a level incompatible with effective exercise training throughout the day post-xenon inhalation.

Peripheral Hemodynamics of Suspected Xenon Doping Practices

From a health perspective, the relatively small hemodynamic changes observed during xenon breathing may be considered unproblematic for athletes under medical supervision, yet consistent transient and sustained changes in cardio- and cerebrovascular hemodynamics are observed.

The 1 MAC of xenon, i.e., the point at which 50% of participants would not respond to surgical incision, is ~63%–71%. However, the depth of anesthesia is dependent on time. With 30% xenon being markedly below the 1 MAC (~0.4 MAC), long inhalation times are possible with subanesthetic affects (26). Over this prolonged inhalation time (20 min), we noted an increase in total peripheral resistance, which was not compensated for by a fall in cardiac output, resulting in elevated blood pressure. In healthy humans, Neukirchen et al. (17) recently noted unchanged muscle sympathetic nerve activity despite elevated concentrations of plasma norepinephrine during prolonged anesthesia with xenon (F_iXe 70%). These data suggest that the mechanism through which xenon increases blood pressure is not heightened sympathetic outflow but an inhibition of norepinephrine reuptake or clearance, which is supported by in vitro inhibition of norepinephrine uptake in xenon-treated cell lines (17). Interestingly, we noted that total peripheral resistance falls rapidly at the onset of xenon inhalation (~1 min), which caused an increase in heart rate and cardiac output, likely via the baroreflex. At present, speculative mechanisms for the rapid fall in total peripheral resistance include central inhibition of sympathetic outflow, modulation of sympathetic transduction pathway, or local peripheral vasodilation. Yet, it is assumed that sympathetic outflow normalizes over time in response to reduced norepinephrine reuptake (17) with maintained baroreflex sensitivity, albeit at a higher set point during prolonged xenon inhalation (17).

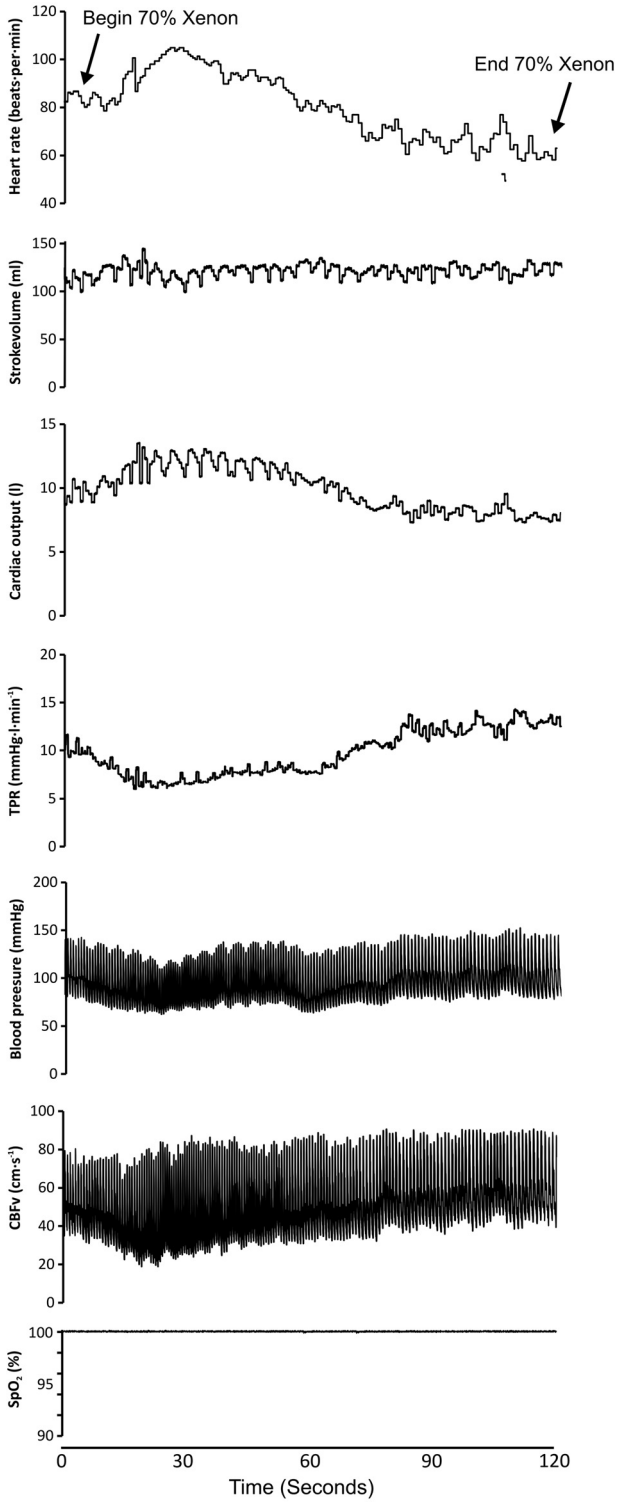

Fig. 3. Hemodynamic profile in 1 individual during 70% xenon inhalation. CBFv, cerebral blood flow velocity; SpO_2, oxygen saturation; TPR, total peripheral resistance.

Cerebral Hemodynamics of Suspected Xenon Doping Practices

Middle cerebral artery velocity increased with each dose of xenon because of an increase in blood pressure and thus incomplete cerebral autoregulation. These data are consistent with most other measurements of acute xenon inhalation (6, 18). However, 50% and 70% xenon caused an acute (1 and 2 min) increase in cerebral vasodilation, as indicated by an increase in cerebral vascular conductance index that was not observed in the 30% xenon condition. It is worth noting that previous reports have noted a reduction in regional cerebral blood flow with xenon inhalation (19), but importantly under conditions of anesthesia, which is associated with a reduction in cerebral metabolism (19, 20). In the current study, participants were in a preanesthetic state with variable levels of sedation, unconscious restlessness, and agitation. Thus, although speculative, under the current conditions it is possible that cerebral metabolism will be elevated, and the cerebral vasodilation is appropriate for effective cerebral metabolic flow coupling. Although we did not incorporate end-tidal carbon dioxide monitoring in this experimental setup, 50% and 70% xenon caused a substantial hyperventilation, and thus it is unlikely that hypercapnia caused cerebral vasodilation.

Detection of suspected xenon doping practices. After all three doses, xenon was eliminated in plasma and urine following a predominantly biphasic profile as previously reported in plasma under conditions of F_iXe 30% [45 min (26)] and F_iXe 60% (23) anesthesia.

In the current study, xenon concentrations fell rapidly over the first few hours in plasma and urine, yet traceable concentrations were detected up to 3 h after xenon inhalation in every participant except one. Thereafter, xenon was traceable in most participants 6 h post-F_iXe 30% and 50% inhalation. However, during F_iXe 70% inhalation, only 1 individual had a traceable plasma xenon concentration, but ~80% of participants had traceable xenon concentrations in urine. These data are at odds with previous investigations (23, 26, 27), whereby xenon could be detected consistently in plasma or urine up to at least 24 h postinhalation.

A conceivable explanation between studies is the different inhalation times and slight differences in apparatus used for xenon delivery. First, although the initial rapid fall in xenon concentration is predominantly due to exhalation from the lungs, the residual detection in blood and urine over time is likely due to release from deep body compartments, including adipose tissue. As with other inhaled anesthetics (25), the slow beta elimination curve from adipose tissue is likely slow for xenon, given its high solubility in oil (2). Thus, the amount of xenon bound to adipose tissue, which is likely dependent on inhalation time, may be a key factor in determining the time window for detectability. Indeed, the data presented herein supports this suggestion with reduced detection rates with decreasing inhalation times. Second, previous studies administered xenon used an anesthesiology closed-circuit rebreathing system, whereas we chose to adopt an open-circuit non-rebreathing technique to simulate suspected doping practices. Indeed, the use of the non-rebreathing technique resulted in substantially lower maximal plasma xenon concentrations compared with those reported with closed-circuit rebreathing [70% xenon: 175 mnol/mL vs. 60% xenon: ~1,000 nmol/mL (23) and 30% xenon: 369 nmol/mL vs. 30% xenon: 4.67 μg/mL (26)]. Taken together, these data suggest that xenon doping can be detected with a high probability up to ~3 h using an open-circuit non-rebreathing system. It should be noted that the samples collected and analyzed within this study did not fulfill all requirements of a regular doping control specimen in

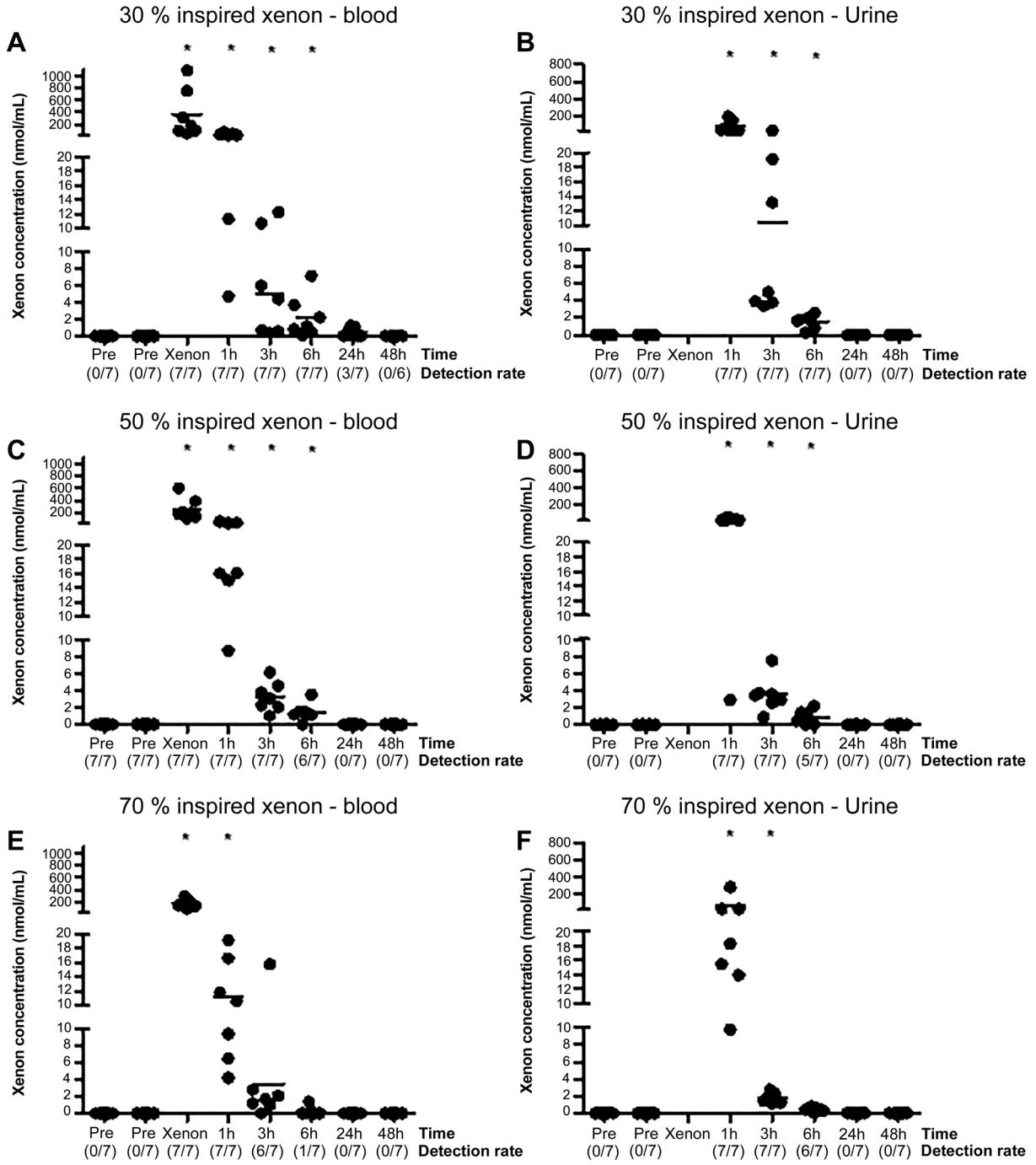

Fig. 4. Detection window of acute xenon inhalation using an open-circuit breathing apparatus. Xenon concentration in blood (*A*, *C*, and *E*) and urine (*B*, *D*, and *F*) over 2 days after 30%, 50%, and 70% xenon inhalation. *$P \leq 0.05$ compared with the average pre-samples. Note that we did not obtain a urine sample on termination of xenon inhalation (*B*, *D*, and *F*). $n = 7$; one-way single-factor ANOVA with follow-up *t* tests.

terms of temperature control and airtight sealing, which can contribute to uncontrolled losses of xenon during transport and storage. Hence, in some individuals, the detection window may be substantially longer.

Conclusions

Xenon has the potential to increase plasma erythropoietin and thus red cell mass. As such, athletes may experiment with xenon inhalation in an attempt to gain an unfair advantage in endurance performance despite possible side effects. We have shown that 3 different conceivable doses of xenon inhalation (F_iXe 30%, 20 min; F_iXe 50%, 5 min; F_iXe 70%, 2 min) caused a level of sedation incompatible with operating a breathing apparatus. All acute doses of xenon caused consistent hypertension and elevated middle cerebral artery velocity. If these dose durations are used for doping, xenon can be detected in blood plasma and urine with a high probability for up to 3 h postinhalation.

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DISCLOSURES

R.K.B. is employed by, and owns equity in, Peloton Therapeutics. None of the other authors have any conflicts of interest, financial or otherwise, to disclose.

AUTHOR CONTRIBUTIONS

J.S.L., E.J.H., S.S., W.K.C., R.K.B., M.H., and B.D.L. conceived and designed research; J.S.L., H.G., K.A.D., E.J.H., S.S., W.K.C., C.M.H., M.S., B.E., M.H., and B.D.L. performed experiments; J.S.L., K.A.D., T.P., M.T., and B.D.L. analyzed data; J.S.L., H.G., K.A.D., E.J.H., S.S., W.K.C., C.M.H., M.S., B.E., M.H., T.P., M.T., and B.D.L. interpreted results of experiments; J.S.L., K.A.D., and B.D.L. prepared figures; J.S.L., H.G., K.A.D., W.K.C., and B.D.L. drafted manuscript; J.S.L., H.G., K.A.D., E.J.H., S.S., W.K.C., C.M.H., M.S., B.E., R.K.B., M.H., T.P., M.T., and B.D.L. edited and revised manuscript; J.S.L., H.G., K.A.D., E.J.H., W.K.C., C.M.H., M.S., B.E., R.K.B., M.H., T.P., M.T., and B.D.L. approved final version of manuscript.

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