Aging: Impact Upon Local Cerebral Oxygenation and Blood Flow With Acute Isovolemic Hemodilution

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Abstract: Data from the neurosurgical critical care arena demonstrate a correlation between cerebral oxygenation, survival, and cognitive function. Transfusion may increase and hemodilution decrease cerebral oxygenation. Both acute and chronic anemia have been associated with cognitive dysfunction. Aggressive blood conservation protocols have been instituted across all age groups without conclusive evidence for their impact upon outcome. Aged subjects are at the greatest risk of cognitive sequelae after major surgery associated with significant blood loss. We hypothesize that cerebral physiologic changes associated with "normal" aging may compromise cerebral oxygenation in the presence of severe anemia.

Fischer 344 rats, the NIH National Institute of Aging normal aging rat model, underwent a stepwise isovolemic hemodilution protocol. Age groups (Age Grp) studied were as follows: Age Grp-A (3 months), n = 14; Age Grp-B (9 to 12 months), n = 14; and Age Grp-C (24 months), n = 14. Brain oxygen tension ($P_{Br}O_2$), laser Doppler flow, and mean arterial pressure were measured. Final hemoglobin averaged 6.1 ± 0.9 g/dL. $P_{Br}O_2$ levels decreased from a baseline of 18.1 ± 4.1 to 17.5 ± 6.8 mm Hg ($P = 0.49$), and laser Doppler flow increased by 18 ± 20% ($P < 0.0001$) after hemodilution. Employing repeated measures multiple regression, Age Grp ($P = 0.30$) was not a significant controlling covariate of $P_{Br}O_2$ in response to isovolemic hemodilution. $P_{Br}O_2$ levels were actually higher in Age Grp-C animals at all time points of the hemodilution protocol, although this was not statistically significant. Aged animals were also fully capable of mounting a robust local cerebral hyperemic response to the anemic challenge that was not separable from the response of younger animals.

Key Words: aging, anemia, cerebrovascular, brain, oxygen

(*J Neurosurg Anesthesiol* 2006;18:125–131)

Severe anemia may complicate multiple trauma and major surgery, and also chronic disease and aging.[1] Aggressive blood conservation protocols have been widely implemented in the management of surgical patients with conflicting reports as to their impact upon clinical outcome.[2–8] Chronic[9,10] and acute[11] anemia have been associated with deterioration in cognitive performance in nonsurgical settings. Severe intraoperative anemia has recently been found to negatively impact long-term cognitive function even in the very young.[12] Not surprisingly, therefore, anemia has been identified as a potential cause of cognitive dysfunction in adult and geriatric surgical settings.

Invasive monitoring of cerebral oxygenation is slowly gaining popularity in the neurosurgical critical care arena, secondary to data demonstrating a correlation between cerebral oxygenation and survival.[13–15] Cognitive function[16] after head trauma may also be impacted by cerebral oxygenation during recovery. Limited clinical data in this setting suggest that transfusion may increase[17] cerebral oxygenation. Furthermore, hemodilution has been largely abandoned in the management of acute stroke because of unproven benefit and even acute neurologic deterioration associated with its implementation.[18] Outside neurosurgery, hemodilution during orthopedic[19] and cardiac surgery[20,21] is associated with cerebral deoxygenation, which in turn may be associated with postoperative cognitive dysfunction[22] and stroke.[7]

Given the aging of our population, the importance of blood conservation in the operative and nonoperative settings, and the frequency with which anemia is found in the elderly,[23] we are particularly interested in the impact of senescence upon cerebral oxygenation in the face of anemia. Studies in humans and aged animal models provide evidence of diminished baseline cerebral blood flow (CBF),[24] decreased cortical microvascular density,[25] and impaired vasoreactivity to multiple stimuli including hypercarbia and hypoxia.[26] Regional CBF responses to cognitive tasks are also diminished in the elderly.[27,28] Diminished expression of endothelial nitric oxide synthetase and neuronal nitric oxide synthetase (nNOS) in specific regions of the cerebral cortex with aging[29] may

Received for publication September 2, 2005; accepted November 29, 2005.

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Supported in part through a grant from the Society of Cardiovascular Anesthesiologists and NIH-RO3 AG023226-01.

Conflicts of interest: None.

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potentially alter the vascular response to anemia. Zarchin et al[30] suggest that structural and functional changes that occur in aging cerebral microvasculature limit their ability to optimally deliver nutrients and oxygen to the brain under conditions of hypoxia. Diffusion of delivered O_2 across the blood-brain barrier[31] could be impaired by age and hypertension-related alterations in the blood-brain barrier, to include basement membrane thickening.[32] Evidence also suggests that cellular oxygen uptake is also diminished with aging.[33,34] These age-related changes are not associated with any particular disease state, such as hypertension, and are part of a process referred to as "normal aging."[35] No studies that we are aware of document the effect of normal aging upon the efficacy of cerebral oxygenation during marked isovolemic hemodilution.

In this study, we test the hypothesis that brain changes accompanying normal aging may be associated with an impairment in cerebral oxygenation under conditions of severe isovolemic anemia.

MATERIALS AND METHODS

Animals

Approval from the Institutional Animal Care and Use Committee at the University of Pennsylvania was obtained before conducting this study. Animals were anesthetized and euthanized in accordance with Institutional Animal Care and Use Committee guidelines.

Experimental Groups

Male Fischer 344 (F344) rats used in this report, the NIH National Institute of Aging aged rat model, were obtained from NIA/HSD (National Institute on Aging). This model was selected as it is characterized by normotension and has been used extensively in research into normal aging in the absence of disease. Survival in the F344 strain is approximately 100% at 12 months and 50% at 24 months of age.[36–38] Aging in the F344 rat is associated with basement membrane thickening of the cerebral vasculature,[39,40] and yet is not associated with chronic hypertension or atherosclerosis.[41,42] Chronic nephropathy[43] and lymphoma[44] can, however, complicate the use of this strain in aging research. Study age groups (Age Grp) were as follows: Age Grp-A (3 months), Age Grp-B (9 to 12 months), and Age Grp-C (24 months).

Experimental Procedures

Rats were anesthetized in a plastic box containing 4% isoflurane and 100% oxygen, tracheotomized with a 14-guage catheter, and ventilated with a tidal volume of 10 mL/kg, at a rate of 40 to 50 breathes/min, and utilizing a volume control rodent ventilator (CWE model MRI-1, CWE, Inc, Ardmore, PA). Anesthesia was maintained with 1.5% to 2% isoflurane in 21% oxygen and α-chloralose (80 mg/kg) injected into the axillary fat pad before craniotomy. Alpha-chloralose was used in this experiment as it has minimal effects on cerebrovascular reactivity as compared with its volatile anesthetic counterparts.[45] Isoflurane was discontinued after craniotomy. Anesthetic management did not include muscle relaxants and was maintained with the previously administered α-chloralose. A period of 45 minutes was allowed to pass to allow for the elimination of isoflurane and for the stabilization of the brain tissue before data collection.

Both femoral arteries were cannulated with PE-50 tubing, the right femoral artery for continuous blood pressure measurement and the left for arterial blood gas measurements and hemodilution. The rat's head was stabilized in a stereotaxic frame and a midline scalp incision performed. The scalp over the frontoparietal cortex was retracted, and the skull was cleaned. A 4×6 mm flap was removed and the dura carefully incised and retracted. After dural retraction, a 450-μm diameter Optronix multiparameter probe (Oxford Optronix, Oxford, UK), was positioned over a point 4 mm posterior to Bregma and 4 mm lateral to the sagittal suture. The multiparameter probe was used to measure laser Doppler flow (LDF), partial pressure of brain oxygen ($P_{Br}O_2$), and brain tissue temperature ($Temp_{Br}$). The probe was stereotactically placed using a micromanipulator, 2 to 4 mm into the parietal association cerebral cortex, under direct vision, and at a magnification of 6X. Care was taken to avoid placing the probe into or next to large pial vessels.

The Optronix tissue O_2 probe employs fluorescence quenching from a ruthenium-based fluorophore. Sampling volumes are thought to be approximately 0.25 to 0.35 mm^3. The most sensitive O_2 measurements occur over the range of 0 to 60 mm Hg. LDF sampling volumes are estimated at 0.3 to 0.5 mm^3. LDF, $P_{Br}O_2$, and $Temp_{Br}$ were monitored using the OXYLAB pO_2-temperature, and OXYLAB LDF monitors (Oxford Optronix, Oxford, UK). Mean arterial pressure (MAP) was monitored utilizing an ADI invasive pressure transducer interfaced with an ADI blood pressure amplifier (ADInstruments Pty Ltd, Castle Hill, NSW, Australia). Rectal temperature was monitored via a probe and maintained with a CWE thermo-regulated heating pad (CWE, Inc, Ardmore, PA), set at 37 ± 0.5 C.

Hemodilution Protocol

After the initial 45-minute period of stabilization, and subsequent to baseline arterial blood sampling, hemodilution was begun. Three withdrawals of 2 to 5 mL(7 mL/kg) of arterial blood occurred that were followed with immediate replacement with normal saline (NS) warmed to 37°C, in a 1(blood):3(NS) ratio. NS was chosen as it is commonly used clinically in the resuscitation of patients with multiple trauma, especially when head trauma is also suspected, and the use of crystalloid over colloid better reflects current clinical practice. After each of the 3 hemodilution steps, a period of 10 minutes was allowed for hemodynamic stabilization. The arterial blood samples were analyzed using a Stat Profile pH Ox system (Nova Biomedical, Waltham, MA). Measurements of pH, arterial partial pressure of carbon dioxide (P_{Art}-CO_2, mm Hg), arterial partial pressure of oxygen

(P_{Art}-O_2, mm Hg), hemoglobin (Hgb, g/dL), and oxygen content (CaO_2, mL/dL) were obtained. Ventilation was adjusted to maintain P_{Art}-CO_2 within the range of 38 to 42 mm Hg.

Data Sampling

All physiologic measures were recorded simultaneously and continuously for the duration of the experiment. These data were integrated through an 8-channel ADI PowerLab acquisition system. Continuous digital recording of data was accomplished by interfacing the ADI PowerLab system with Chart 5.0 software interface (ADInstruments Pty Ltd, Castle Hill, NSW, Australia) on a Power Mac computer (Apple Computer, Inc, Cupertino, CA). All analog data, to include LDF and $P_{Br}O_2$, were sampled digitally at a frequency of 10 Hz.

After the initial 45 minute (after probe placement) and subsequent 10-minute stabilization periods (after each of the 3 hemodilution steps), all measured physiologic parameters were averaged over a 5-minute time period. Thus, data from 4 time points were recorded: time 0, baseline before hemodilution; time –1, after hemodilution no. 1; time –2, after hemodilution no. 2; and time –3, after hemodilution no. 3.

Statistical Analysis

Bivariate analyses of group means were conducted via repeated measures analysis of variance (ANOVA). Apart from comparing group means, this method tests for any changes over time and whether these changes vary by age group. Multiple comparisons of group means were adjusted for using the Tukey-Kramer method. Significance of effects was assigned at a $P \leq 0.05$.

The main outcome of interest, $P_{Br}O_2$, was further examined using a random (or "mixed") effects model[46] for the following reasons: (1) Despite attempts to control for important variables during experimentation, subtle but important variances from designed goals in covariates such as Hgb or P_{Art}-CO_2 may make a direct comparison of end points such as $P_{Br}O_2$ between groups inaccurate; (2) any noninformative missingness in the covariates does not exclude the rat from the model, as happens under a balanced repeated measures ANOVA; this approach allowed all available data to be used while handling the nonindependence of measurements from each rat; and (3) sample size may be reduced by the increased power afforded by such a model.[46]

The random effects model was used to test and adjust for any Age Grp, time (Time) or Age Grp-Time interaction, having adjusted for other potentially confounding covariates such as P_{Art}-CO_2, Hgb, CaO_2, MAP, and for the correlated measurements from each rat (via random intercepts and slopes).

Sample size requirements were based on obtaining 80% power, at $\alpha = 0.05$, to detect an overall effect size (ratio of hypothesized difference in change in $P_{Br}O_2$ between groups to the standard deviation of the measurement) of 0.5 between the 3 age groups under an ANOVA model at a given time. This corresponds to an effect size of 2 for the pair-wise multiple comparisons under the Tukey-Kramer adjustment, and resulted in a sample of approximately 14 to 15/group. Note that this is a conservative estimate of the effect size under the random effects model, as more power is obtained under this model.[46]

RESULTS

Bivariate Analyses

A summary of the results of bivariate analyses for all physiologic measurements is shown in Table 1, at each time point and for each age group. Hemoglobin and CaO_2 levels were highly comparable between all groups at each of the 4 time points. Hemoglobin and CaO_2 fell by an average of 4.5 ± 0.8 g/dL and 6.3 ± 1.5 mL/dL, respectively, over the course of the hemodilution steps.

TABLE 1. Comparisons of Physiologic Parameters at Various Stages of Graded Hemodilution

	3 mo	9 to 12 mo	24 mo
Baseline			
Hgb (g/dL)	10.7 ± 0.7	10.3 ± 0.5	11.0 ± 1.3
CaO_2 (mL/dL)	14.6 ± 1.0	13.9 ± 0.9	15.0 ± 1.9
P_{Art}-O_2 (mm Hg)	88.1 ± 14.1	89.9 ± 20.0	84.5 ± 15.1
P_{Art}-CO_2 (mm Hg)	36.0 ± 3.2	37.9 ± 4.3*	33.9 ± 3.4
MAP (mm Hg)	122 ± 15†	129 ± 14†	104 ± 14.8
$Temp_R$ (°C)	37.0 ± 0.5	36.2 ± 1.6	37.0 ± 0.7
$Temp_{Br}$ (°C)	34.2 ± 1.4	34.3 ± 1.9	35.1 ± 1.5
LDF (units)	869 ± 620	1167 ± 498	930 ± 508
$P_{Br}O_2$ (mm Hg)	17.1 ± 4.3	17.6 ± 4.4	19.5 ± 3.4
Hemodilution no. 1			
Hgb (g/dL)	8.2 ± 0.8	7.9 ± 1.3*	8.9 ± 0.9
CaO_2 (mL/dL)	11.2 ± 1.1	11.0 ± 1.8	12.1 ± 1.6
P_{Art}-O_2 (mm Hg)	85.1 ± 11.1	95.6 ± 11.9*	81.9 ± 12.8
P_{Art}-CO_2 (mm Hg)	36.1 ± 3.5	33.5 ± 4.3	36.1 ± 3.8
MAP (mm Hg)	118 ± 11†	130 ± 15†	102 ± 14
$Temp_R$ (°C)	36.9 ± 0.3	36.3 ± 1.2	36.9 ± 0.7
$Temp_{Br}$ (°C)	34.4 ± 1.6	35.1 ± 1.6	35.3 ± 1.4
LDF (units)	924 ± 650	1176 ± 556	1001 ± 483
$P_{Br}O_2$ (mm Hg)	16.9 ± 6.2	18.7 ± 5.1	20.3 ± 5.3
Hemodilution no. 2			
Hgb (g/dL)	7.2 ± 1.0	6.9 ± 0.8	7.8 ± 1.1
CaO_2 (mL/dL)	9.9 ± 1.5	9.5 ± 1.1	10.7 ± 1.6
P_{Art}-O_2 (mm Hg)	87.3 ± 9.6	88.5 ± 12.4	85.5 ± 9.7
P_{Art}-CO_2 (mm Hg)	35.0 ± 2.9	34.1 ± 3.2	34.2 ± 3.4
MAP (mm Hg)	114 ± 15	124 ± 18*	101 ± 14
$Temp_R$ (°C)	37.1 ± 0.2	36.6 ± 0.7	37.0 ± 0.5
$Temp_{Br}$ (°C)	34.6 ± 1.6	35.3 ± 1.3	35.4 ± 1.3
LDF (units)	968 ± 649	1207 ± 578	1018 ± 463
$P_{Br}O_2$ (mm Hg)	16.6 ± 6.9	17.0 ± 5.6	20.4 ± 5.4
Hemodilution no. 3			
Hgb (g/dL)	6.1 ± 0.6	5.7 ± 0.9*	6.6 ± 0.9
CaO_2 (mL/dL)	8.2 ± 0.9	7.4 ± 1.8*	9.1 ± 1.1
P_{Art}-O_2 (mm Hg)	82.6 ± 12.9	84.1 ± 19.4	86.4 ± 7.2
P_{Art}-CO_2 (mm Hg)	36.4 ± 2.8	37.4 ± 6.8	33.3 ± 4.5
MAP (mm Hg)	107 ± 22	108 ± 21	91 ± 16
$Temp_R$ (°C)	37.1 ± 0.3	36.6 ± 0.9	37.1 ± 0.3
$Temp_{Br}$ (°C)	34.9 ± 1.7	35.4 ± 1.6	37.1 ± 1.3
LDF (units)	1025 ± 610	1269 ± 640	1081 ± 427
$P_{Br}O_2$ (mm Hg)	16.8 ± 7.3	16.3 ± 7.3	19.4 ± 5.9

Measurements are mean ± standard deviation.
*Significant difference ($P < 0.05$) between 9 to 12 mo and 24 mo groups.
†Significant difference ($P < 0.05$) between 3 mo and 24 mo groups.

Across all groups, hemodilution resulted in a near instantaneous, but only transient, decline in $P_{Br}O_2$ values. Decrements of 14.5%, 14.7%, and 22.4% after the first, second, and third hemodilution, respectively, were observed. Age group did not significantly affect the magnitude of this transient decline in oxygenation, nor did age group affect the length of time for $P_{Br}O_2$ to return to baseline levels, averaging about 2.2 to 2.5 minutes. Apart from these transient changes in $P_{Br}O_2$ after hemodilution, sustained or steady state $P_{Br}O_2$ values did not change significantly from baseline ($P = 0.49$); nor were there any significant differences in the magnitude of change in $P_{Br}O_2$ measurements across groups ($P = 0.79$).

LDF measurements increased by 24% (Age Grp-A), 9% (Age Grp-B), and 23% (Age Grp-C) from baseline to final hemodilution. These values were not significantly different across groups ($P = 0.09$), however, the rather modest increase in Age Grp-B in comparison with the younger and aged groups is noted.

Tracking the instantaneous but transient changes in $P_{Br}O_2$ measurements with each hemodilution step was the acute, but also transient, decline of MAP values of 22.6%, 30.8%, and 39.6% after the first, second, and third hemodilution, respectively. Age group did not significantly affect this decline or the length of time until return to baseline, averaging approximately 2.2 minutes. Small but significant decreases in MAP occurred between the baseline reading and final hemodilution in all age groups. With respect to age group, the mean fall in MAP from baseline to final hemodilution was 11% (Age Grp-A), 16% (Age Grp-B), and 12% (Age Grp-C), but was not significantly different across groups ($P = 0.43$). Grouping all animals, MAP fell by 1%, 4%, and 13% after the first, second, and third hemodilution, respectively.

$Temp_{Br}$ was significantly lower than rectal temperature at all time points ($P < 0.0001$), with the average difference between the 2 sites of measurement being $1.8 \pm 1.5^{\circ}C$. The construct of the Oxford-Optronix multiprobe is such that the temperature-sensing element resides more than a millimeter above the $P_{Br}O_2$ and LDF elements. The lower brain temperatures recorded might reflect, to some extent, cooling of the brain surface from ambient air temperatures.

Multivariate Analyses

Cerebral oxygenation ($P_{Br}O_2$): Potentially important variables included in the model were Age Grp, CaO_2, MAP, $Temp_{Br}$, and P_{Art}-CO_2. Although we attempted to control $Temp_{Br}$ through warming, and P_{Art}-CO_2 through adjustment of ventilation, these potential confounders can never be perfectly controlled. Particularly concerning to us was the need to control for any effect of change in P_{Art}-CO_2, as a very small change in this variable may have profound effects upon $P_{Br}O_2$. Multivariate analysis allows us to exert further control over these potentially confounding variables.

A summary of the model is seen in Table 2. The $R^2 = 0.88$ suggests that the covariates included in the etiologic model that was tested account for approximately 88% of the variability in $P_{Br}O_2$ during the course of the experiment. The overall contribution of Age Grp to any change in $P_{Br}O_2$ was not significant ($P = 0.30$). Further, no individual age group was found to impact $P_{Br}O_2$ more than another in response to anemia, as shown by the pair-wise comparisons of the relative contribution of Age Grp-2 or Age Grp-3, as compared with the reference Age Grp-1.

TABLE 2. Covariates Summary-multiple Regression Analysis of $PBrO_2$

Covariate	Estimate	Std Error	*P*
Intercept	−1.2	12	0.92
Age Grp (reference-Grp-1)	—	—	0.30
Age Grp-2	−0.77	1.2	0.53
Age Grp-3	0.17	0.36	0.64
CaO_2	0.20	0.19	0.28
P_{Art}-CO_2	−0.062	0.065	0.35
MAP	0.022	0.026	0.40
$Temp_{Br}$	0.47	0.32	0.14
Time	0.21	0.48	0.66
Age Grp × Time (reference-Grp-1)	—	—	0.61
Age Grp-2 × Time	−0.37	0.37	0.32
Age Grp-3 × ime	0.17	0.36	0.64

No effect of time, or duration of the experiment, upon $P_{Br}O_2$ was noted in the model as reflected in Time ($P = 0.66$). We had desired to control for any change in the characteristic of the brain preparation over time with this covariate. The covariate Age Grp x Time was not significant, indicating that there was no age group-specific effect of the duration of the experiment upon the $P_{Br}O_2$ data.

CaO_2, P_{Art}-CO_2, or $Temp_{Br}$ did not impact $P_{Br}O_2$. We attempted to control all of these parameters during the experiment. Arterial pCO_2 can profoundly affect brain pO_2 and was apparently successfully controlled in this experiment. We did not attempt to control MAP during this experiment, yet it remained remarkably steady and did not impact $P_{Br}O_2$ within the range documented.

DISCUSSION

The range of $P_{Br}O_2$ values that we measured is consistent with those reported by others using similar techniques.[47,48] The values for $P_{Br}O_2$ that we reported are those documented after a period of stabilization after the hemodilution phase. Hare et al[47] reported in adult, but not aged, animal that slight (≈ 3 mm Hg) but significant decreases in $P_{Br}O_2$ occurred during the acute phase after hemodilution. Hare also reported that after a brief period of recovery, $P_{Br}O_2$ levels did not differ from baseline. We found similar acute decreases in $P_{Br}O_2$, but these were short lived. We found no evidence for age dependency of cerebral oxygenation during isovolemic anemia.

It is possible that the levels of anemia achieved were not severe enough to unmask an age-related impairment

in cerebral oxygenation; yet our aim in this study was to create conditions that would mirror the clinical experience, and we did achieve levels of anemia consistent with those goals. Van Bommel et al[48] found in pigs that cerebral oxygenation decreased only below a hematocrit of approximately 18%. Even healthy humans have demonstrated cognitive impairment after acute isovolemic hemodilution to Hgb levels of 5 to 6 g/dL,[11] and these extreme levels are not especially clinically relevant.

The concept of pressure autoregulation of CBF is well known. Much less well-known or investigated, however, is the concept of pressure autoregulation of cerebral oxygenation. In one study, $P_{Br}O_2$ was found to vary by as much as 300% over an MAP range of 50 to 200 mm Hg.[49] The concepts of pressure autoregulation of CBF and of cerebral oxygenation are indeed related, as Hemphill et al[49] determined that $P_{Br}O_2$ is ultimately tightly related to flow. Hemodilution resulted in a decrease of only 13% in MAP over the course of this entire experiment, but more acute and transient changes immediately followed each hemodilution step, and may have been responsible for the coincident, transient, and more marked changes in $P_{Br}O_2$ that also accompanied each hemodilution step. Chronic hypertension may cause greater sensitivity to the effects of anemia and associated hypotension upon cerebral oxygenation.

Brain tissue pO_2 measurements made with probes such as the Optronix and Licox (Integra NeuroSciences, Plainsboro, NJ) devices produce a measurement reflecting contributions of the arteriole, venule, and cellular oxygen levels, and probably represent the results of delivery and also extraction. Documentation of a lack of age dependence of cerebral oxygenation during isovolemic anemia, although important, may not entirely absolve anemia of a role in cognitive dysfunction. Anemia may induce more subtle effects. Neuronally derived nitric oxide is tightly involved in the cellular response to hypoxia.[50] Hare et al[47] has demonstrated increases in nNOS gene expression in response to acute hemodilution associated with only transient and mild decreases in $P_{Br}O_2$ in adult rats. These changes in nNOS expression, in response to anemia, mirrored or exceeded the changes induced by mild hypoxemia. It seems that, when tissue is threatened by even mild hypoxemia or moderate to severe anemia, metabolism, mitochondrial oxygen fixation, and less critical functions such as neurotransmitter synthesis and release may be slowed down to maintain tissue integrity, yet still impacting cognitive function.[51–54] These neuroprotective responses may, however, be age limited.[51,55–57]

LDF changes in our experiments closely tracked the 24% increase measured by Shen et al[58] in adult male Sprague-Dawley rats after a similar hemodilution protocol. We found no evidence on the supply side that the hyperemic regional blood flow response was compromised by healthy aging, as LDF increased as robustly in the aged as it did in the young (3 month) and mature (9 to 12 month) animals. Our human research, in a cardiac surgery population with risk factors for cerebrovascular disease, has also documented a robust CBF response in the aged to a similar anemic challenge,[59] thus suggesting the retained ability of the aging brain to compensate, at least globally via vasodilation, for anemia. Regional effects remain to be determined. Age Grp-B achieved a modest 9% increase in LDF, notably lower, but not significantly lower than that seen in the younger and older cohorts. Further testing is necessary to determine whether this difference may truly be important.

Regarding LDF measurements, the following points should be noted. LDF measurements of changes in blood flow correlate favorably with xenon,[60] microsphere,[61,62] and hydrogen clearance CBF measurements.[63,64] LDF measurements have been reported to overestimate CBF changes during hemorrhage[60] and underestimate[58,65,66] CBF changes during isovolemic or hypervolemic hemodilution. LDF measures the perfusion rate of red blood cells and does not measure whole blood flow rates. Hudetz et al[67] has reported that, with hemodilution, capillary hematocrit does not change, and yet red blood cell velocity and supply rate increase. LDF and whole blood CBF measurements may reflect correlated but distinct and equally important changes in very different quantitative aspects of the cerebral circulation (red blood cell delivery vs. whole blood delivery). LDF has the advantage of allowing continuous measurements[68] versus only intermittent measurements with other techniques.

In conclusion, normal aging, as reflected in the NIH/NIA aged F344 rat model, did not impair cerebral oxygenation or the CBF response in a setting of acute isovolemic anemia where marked hypotension did not occur. The spontaneously hypertensive rat model, because of the occurrence of cerebrovascular disease, may consequently better reflect the patient treated in cardiac and major noncardiac surgery, although not necessarily aging neurosurgery patients. The effect of hemodilution upon cerebral oxygenation, regional blood flow response to anemia, and the pressure dependence of cerebral oxygenation and flow should be further tested in this model. Studies examining the effect of isovolemic anemia upon cognition in these animals are currently underway.

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