Interrelationship Between Sex, Age, Blood Volume, and $\dot{V}O_{2max}$

Natalie J. Koons; Mithun R. Suresh; Taylor E. Schlotman; Victor A. Convertino

BACKGROUND: Circulating blood volume (BV) and maximal oxygen uptake ($\dot{V}O_{2max}$) are physiological characteristics important for optimal human performance in aerospace and military operational environments. We tested the hypothesis that BV and $\dot{V}O_{2max}$ are lower in older people independent of sex.

METHODS: To accomplish this, a "data mining" effort of an historic database generated from NASA and U.S. Air Force experiments was conducted. BV, red cell volume, plasma volume, hematocrit, and $\dot{V}O_{2max}$ were measured in 84 healthy individuals (24 women, 60 men) across an age range of 23 to 65 yr to assess the interrelationship between sex, age, BV, and $\dot{V}O_{2max}$. Subjects were classified in age groups by $<$ 40 yr and $\geq$ 40 yr; these groups identified women as pre- vs. postmenopausal.

RESULTS: Consistent with our hypothesis, comparisons revealed that men had higher BV, red cell volume, hematocrit, and $\dot{V}O_{2max}$ than women when standardized for body mass. Against expectations, BV was not different in older compared with younger men and women. $\dot{V}O_{2max}$ was not different in older compared with younger women, while $\dot{V}O_{2max}$ was lower in older men.

CONCLUSION: We conclude that physiological mechanisms other than BV associated with aging appear to be responsible for a decline in $\dot{V}O_{2max}$ of our older men. Furthermore, factors other than menopause may also influence the control of BV in the women. Our results provide evidence that aging may not compromise men or women in scenarios where BV can affect performance in aerospace and military environments.

KEYWORDS: aerobic capacity, plasma volume, red cell volume, hematocrit.

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Optimal human performance in aerospace and military operational environments requires tolerance to conditions of acute and chronic central hypovolemia and adequate aerobic capacity. Circulating blood volume (BV) with an adequate oxygen carrying capacity is critical to sustaining adequate tissue oxygenation during hypovolemic challenges and tasks that require high levels of oxygen uptake. Indeed, reduced BV has been associated with orthostatic instability during standing after spaceflight,[5,6,8] while expanded circulating volume is associated with increased maximal oxygen uptake ($\dot{V}O_{2max}$) and $+G_z$ tolerance[9,23] after task specific training.

Sex and age can separately or together contribute to variability of tolerance to pathophysiological conditions of central hypovolemia (e.g., orthostatic stress, hemorrhage) or requiring maximal working capacity. Data from several studies have consistently indicated that women have lower $\dot{V}O_{2max}$ and tolerance to central hypovolemia compared to men.[6,7,20] Additionally, nearly all occupations and positions have become available for participation by women in both military and spaceflight operational environments. As of 2016, the number of women reached 18.3%[27] in the U.S. military and armed services, and 34% in the active U.S. astronaut corps.[26] Consequently, there continues to be a need to understand physiological characteristics of women that may impact performance ranging from the execution of aerospace and military missions to survival from hemorrhage on the battlefield.

From Battlefield Health & Trauma Center for Human Integrative Physiology, U. S. Army Institute of Surgical Research, JBSA Fort Sam Houston, TX.

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Address correspondence to: Victor A. Convertino, Ph.D., U.S. Army Institute of Surgical Research, 3698 Chambers Pass, Fort Sam Houston, TX 78234, Email: victor.a.convertino.civ@mail.mil.

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Previous studies have focused on comparisons of BV or aerobic capacity separately with aging, and in men and women in efforts to identify differences in physiological factors that influence performance in aerospace and military environments. It is clear from the literature that women generally exhibit lower $\dot{V}O_{2max}$ than men,[6,9,12] which may be explained by a lower maximal capacity to deliver oxygen to working muscles as a result of reduced total circulating red cell volume.[1,25,36] Although there are additional studies in which the impact of age on BV and $\dot{V}O_{2max}$ were reported separately in men[14] and women,[21] there is no single study in which the influences of changes in age on BV and $\dot{V}O_{2max}$ have been assessed between sexes. Addressing the interrelationships between BV, $\dot{V}O_{2max}$, sex, and age together is relevant to identifying those individuals with greatest risk of poor outcomes from hemorrhage, as well as assessing the levels of performance competence of individuals deployed in operational environments. In the present investigation, we report and compare for the first time BV and $\dot{V}O_{2max}$ in a cohort of men and women together to test the hypothesis that BV and $\dot{V}O_{2max}$ is lower in older people independent of sex.

METHODS

Experimental Approach

A "data mining" effort was conducted to identify values for $\dot{V}O_{2max}$ in addition to BV and its component volumes of plasma and red cells. The data were obtained from archived files of experiments conducted from 1978 through 1997. All experimental protocols and measurement procedures were conducted in the same manner by the same investigator and were reviewed and approved by the Institutional Review Boards at NASA-Ames Research Center, Mountain View, CA, and Brooks AFB, San Antonio, TX, where all experiments were conducted.

Subjects

Complete data sets were identified for 84 healthy, nonsmoking, and normotensive volunteer subjects (60 men, 24 women) who participated in these experiments following an evaluation of their medical history and physical examination by a physician to ensure the absence of previous and current medical conditions that would render them exempt from participating. The demographic data of each sex group are presented in **Table I**. Subjects taking prescription medications (except oral contraceptives) were excluded. All women $<$ 40 yr old self-reported that they were premenopausal. Likewise, all women $\geq$ 40 yr old self-reported they were postmenopausal. Additionally, all women completed a pregnancy test to ensure that they were not pregnant prior to the experimental procedures. Subjects received a verbal and written briefing of all protocol procedures and risks associated with the study and were familiarized with the laboratory, protocol, and instrumentation. Finally, subjects were encouraged to ask questions of the investigators before giving their written informed consent to participate in the study.

Maximal Oxygen Uptake Measurement

The exercise tests were performed using a Collins electrically-braked cycle ergometer. The maximal exercise test started with a 1-min warmup at 0 Watts (W) and was followed by a progressively increasing work rate of 30 W every 3 min until volitional fatigue. The end-point was subjective; however, each subject was familiar with the test and encouraged to reach exhaustion.

Oxygen uptake ($\dot{V}O_2$) was measured during the last 30 s of all submaximal and maximal work rates. The subjects used an Otis-McKerrow respiratory valve and the volume of expired gas was measured with a Parkinson-Cowan high-velocity, low-resistance meter. A potentiometer at the gas meter dial transmitted an electrical output to a two-channel recorder (MFE Model M22) to record ventilation volume continuously. Using a semiautomated system, expired gas was collected into a 2-liter anesthesia bag (Ohio Medical, Gurnee, IL) by means of a Dynapump (Scientific Products). The composition of the expired gas was determined with a Beckman E2 oxygen analyzer (Beckman Coulter, Inc., Brea, CA) and a Godart capnograph carbon dioxide analyzer. $\dot{V}O_{2max}$ was calculated using standard equations.

Blood Volume Measurement

Resting plasma volume (PV) was measured with a modified Evans blue dye (T-1824) dilution[17] method. Each subject assumed a supine position for 30 min and a "zero" time (control) blood sample was drawn from the antecubital vein with a 21-gauge needle. Immediately following the zero-time sample, a 0.5% (4.52 mg $\cdot$ ml^{-1}) aqueous stock dye solution (Harvey Laboratories, Inc., Philadelphia, PA) was injected intravenously. The syringe containing Evans blue dye and the needle were accurately weighed before injection. After the injection of dye, the needle was wiped clean and the syringe and needle were weighed immediately. The exact amount of dye injected was determined by the difference in weight of the syringe and needle before and after the injection. Following exactly a 10-min dilution period, a blood sample was taken from the arm opposite that of the injection. After the needle was disconnected from the syringe, the blood sample was delivered slowly into a sodium-heparinized glass tube and was mixed gently for about 1 min. The plasma was separated from the red cells by centrifugation at 3000 g for 12 min.

The procedure for the treatment of plasma samples, the determination of their Evans blue dye concentration, and the subsequent computation of plasma volume were performed in the following manner. The Evans blue stock standard was prepared by diluting 1 ml of the commercial (0.5%) stock dye preparation to 50 ml with distilled water. The internal standard was prepared with 0.2 ml of Evans blue diluted standard solution mixed with 2 ml of control plasma. The quantity of Evans blue recovered from the plasma mixture was analyzed spectrophotometrically and served as the internal standard. The internal standard and plasma samples were mixed thoroughly with 15 ml of Teepol-phosphate, a detergent that displaces the dye from protein. The mixture was transferred gently onto a pulp column

Table I. Subject Demographics and Baseline Physiologic Values.

	MEN	WOMEN	*P*-VALUE	*t* (df)
N	60	24		
Age (years)	45 (1)	48 (2)	0.11	2.65 (1)
Height (cm)	178 (1)	164 (1)	< 0.001	67.4 (1)
Weight (kg)	78.1 (2.1)	63.2 (1.8)	< 0.001	39.9 (1)
Heart Rate (bpm)	68 (1)	71 (2)	0.19	1.74 (1)
Systolic Arterial Pressure (mmHg)	123 (2)	122 (3)	0.67	0.18 (1)
Diastolic Arterial Pressure (mmHg)	74 (1)	71 (2)	0.26	1.27 (1)
Plasma Volume (ml)	3445 (72)	2612 (113)	< 0.001	38.6 (1)
Plasma Volume ($ml \cdot kg^{-1}$)	43.9 (0.9)	41.7 (1.4)	0.20	1.67 (1)
Blood Volume (ml)	5804 (141)	4081 (223)	< 0.001	42.5 (1)
Blood Volume ($ml \cdot kg^{-1}$)	73.9 (1.7)	65.2 (2.7)	0.009	7.18 (1)
Red Cell Volume (ml)	2358 (84)	1466 (132)	< 0.001	32.6 (1)
Red Cell Volume ($ml \cdot kg^{-1} \cdot min$)	29.9 (1.0)	23.5 (0.9)	0.008	12.0 (1)
Hematocrit (%)	44.9 (0.8)	37.8 (1.3)	< 0.001	20.8 (1)
$\dot{V}O_{2max}$ ($ml \cdot kg^{-1}$ BW $\cdot min^{-1}$)	43.9 (1.8)	33.1 (2.2)	0.004	14.3 (1)

Values are means ± SE; N, number of subjects. *t*-value (degrees of freedom).
On average, men were taller, heavier, and had lower diastolic arterial pressure compared to the women. Comparison of average plasma volume, blood volume, red cell volume, hematocrit, and $\dot{V}O_{2max}$ in men versus women. On average, men had higher plasma volume, blood volume, red cell volume, hematocrit, and $\dot{V}O_{2max}$ values compared to women ($P < 0.001$).

prepared with Solka-Floc SW-40A (Brown Co., Boston, MA). The column was rinsed with 5 ml of Teepol-phosphate. Interfering substances such as protein, pigments, and chylomicrons were washed from the column with 2% disodium hydrogen phosphate. With this technique, any additional interference due to hemolysis can be minimized or eliminated completely. The flow rate of the eluate in the chromatographic columns was regulated with a stopcock to about one drop per second. The dye was then eluted from the column with a freshly prepared alkaline acetone-water mixture (1:1) without adjustment of pH. The light absorption of the plasma blanks, standards, and the dye recovered from plasma were measured at 615 mμ with a Beckman DU-2 spectrophotometer. The plasma volume was calculated from the following equation:

$$PV\left(ml\right) = \frac{EBI \times \varepsilon_{std} \times V}{\varepsilon_{pl} \times EB_{std} \times 1.03}$$

where EBI is the weight in grams of the Evans blue injected; ϵ_{std} the absorbance of Evans blue extracted from the plasma standard mixture; V the volume of plasma used for determination (milliliters; ml); ϵ_{pl} the absorbance of Evans blue extracted from plasma; EB_{std} the amount of Evans blue added to the internal standard (ml of Evans blue × 1/50 × 0.2); and 1.03 the correction for the estimated 3% Evans blue absorbed by the tissues during the 10-min dilution period.

Red blood cell volume (RCV) was calculated by subtracting plasma volume from total blood volume (TBV), where TBV was calculated from the plasma volume value and hematocrit (Hct):

$$TBV\left(ml\right) = PV \times \frac{100}{100 - (0.96 \times 0.91 \times Hct)}$$

Using these procedures in our laboratory, test-retest correlation coefficient for plasma volume was 0.969 ($N = 12$) and the day-to-day variation was 82 ml (1.5%, $N = 17$) over 4 d, 75 ml (1.5%, $N = 19$) over 8 d, and 56 ml (1.1%, $N = 23$) over 15 d.[17]

Data Analysis

Data were analyzed using two-sample, unpaired, two-tailed Student *t*-tests to compare men vs. women baseline demographic data and compare the effect of age in men and women within the < 40 yr and ≥ 40 yr age groups. Two-sample F-tests were conducted to check for equal variance for all variables. The *t*-tests on those variables found to be unequal were adjusted using the Behrens-Fisher problem. These analyses were conducted with JMP 13 (JMP, Version 13. SAS Institute Inc., Cary, NC). The probability that any differences between sex and age groups were not attributable to chance is expressed as exact *P*-values, such that a significant difference occurs at $P < 0.05$. Data are presented as mean ± SE, unless otherwise noted.

RESULTS

Mean (± SE) values for demographic and physiologic data at are presented in Table I. On average, men were taller and heavier compared to the women (all $P < 0.001$).

When comparing physiological values between men and women, men had higher absolute levels of plasma volume (ml), BV (ml), red cell volume (ml), hematocrit, and $\dot{V}O_{2max}$ (all $P < 0.01$). When standardizing the data for body mass, men still displayed higher BV (milliliter per kilogram; $ml \cdot kg^{-1}$) and red cell volume ($ml \cdot kg^{-1}$) compared to the women (both $P < 0.01$).

Across age groups, men < 40 yr old had higher $\dot{V}O_{2max}$ levels compared to men ≥ 40 yr old ($P < 0.01$; **Table II**). However, there were no statistically meaningful differences in plasma volume, BV, red cell volume, or hematocrit values between age groups in the population of men ($P > 0.01$). Likewise, there were no differences in plasma volume, BV, red cell volume, hematocrit, or $\dot{V}O_{2max}$ values between women < 40 yr old and women ≥ 40 yr old ($P > 0.01$).

A linear regression was used to compare $\dot{V}O_{2max}$ and BV between men < 40 yr old, men ≥ 40 yr old, women < 40 yr old, and women ≥ 40 yr old. The linear correlation coefficient between $\dot{V}O_{2max}$ and BV between men and women across age groups is 0.9091 (**Fig. 1**). This linear relationship is illustrated by the equation y = 1.023*X – 34.73.

Table II. Comparison of Plasma Volume, Blood Volume, Red Cell Volume, Hematocrit, $\dot{V}O_{2max}$, as well as Demographic Data in Men and Women < 40 yr Old versus Men and Women ≥ 40 yr Old.

MEN	< 40 YRS.	≥ 40 YRS.	*P*-VALUE	*t* (df)
N	22	38		
Age (years)	37 (1)	49 (1)	< 0.001	54.2 (1)
Weight (kg)	77.0 (2.2)	78.8 (1.6)	0.54	0.38 (1)
Plasma Volume (ml)	3367 (129)	3489 (98)	0.46	0.57 (1)
Plasma Volume (ml · kg^{-1})	44.0 (1.6)	45.0 (1.2)	0.72	0.13 (1)
Blood Volume (ml)	5834 (266)	5787 (202)	0.89	0.02 (1)
Blood Volume (ml · kg^{-1})	75.6 (3.2)	72.8 (2.4)	0.73	0.12 (1)
Red Cell Volume (ml)	2462 (158)	2298 (122)	0.42	0.67 (1)
Red Cell Volume (ml · kg^{-1})	31.6 (1.9)	29.5 (1.4)	0.39	0.75 (1)
Hematocrit (%)	46.5 (1.6)	44.0 (1.2)	0.22	1.6 (1)
$\dot{V}O_{2max}$ (ml · kg^{-1} BW · min^{-1})	49.7 (2.7)	38.5 (2.6)	< 0.001	8.8 (1)
WOMEN	**< 40 YRS.**	**≥ 40 YRS.**	***P*-VALUE**	***t* (df)**
N	8	16		
Age (years)	33 (2)	56 (1)	< 0.001	123.6 (1)
Weight (kg)	64.5 (3.2)	62.5 (2.3)	0.62	0.26 (1)
Plasma Volume (ml)	2494 (142)	2671 (101)	0.32	1.04 (1)
Plasma Volume (ml · kg^{-1})	39.7 (2.1)	42.8 (1.5)	0.24	1.44 (1)
Blood Volume (ml)	4051 (215)	4096 (152)	0.87	0.03 (1)
Blood Volume (ml · kg^{-1})	64.5 (3.4)	65.6 (2.4)	0.80	0.06 (1)
Red Cell Volume (ml)	1557 (86)	1420 (61)	0.21	1.68 (1)
Red Cell Volume (ml · kg^{-1})	24.9 (1.5)	23.0 (0.7)	0.32	1.03 (1)
Hematocrit (%)	38.3 (1.1)	37.6 (0.8)	0.58	0.32 (1)
$\dot{V}O_{2max}$ (ml · kg^{-1} BW · min^{-1})	29.3 (2.5)	35.0 (1.8)	0.08	3.48 (1)

Values are means ± SE; N, number of subjects. *t*-value (degrees of freedom).
On average, $\dot{V}O_{2max}$ values are larger in men < 40 yr old. There were no significant differences between age groups among women.

DISCUSSION

Previous investigations have consistently identified women as having smaller circulating BV[1,25,36] and lower $\dot{V}O_{2max}$.[6,8,12] While the results from this analysis corroborate previously reported sex differences in BV and $\dot{V}O_{2max}$, this is the first study to provide an assessment of the interrelationship between sex, age, BV, and $\dot{V}O_{2max}$ with direct comparison of men and women. To address this knowledge gap, we strategically utilized an historic database generated from NASA and U.S. Air Force experiments in which BV (including plasma and red cell volume components) and $\dot{V}O_{2max}$ were measured by the same investigator in men and women across an age range of 23 to 65 yr. We anticipated our findings that BV and $\dot{V}O_{2max}$ would be greater in men; however, we also hypothesized BV and $\dot{V}O_{2max}$ would decrease with age in both sexes based on the findings of previous investigations.[14,21] The main finding of this investigation was that the well-established relationship between BV and $\dot{V}O_{2max}$ was unaltered by sex and aging. A novel finding of this study was that $\dot{V}O_{2max}$ decreased in men with increasing age, while BV did not change.

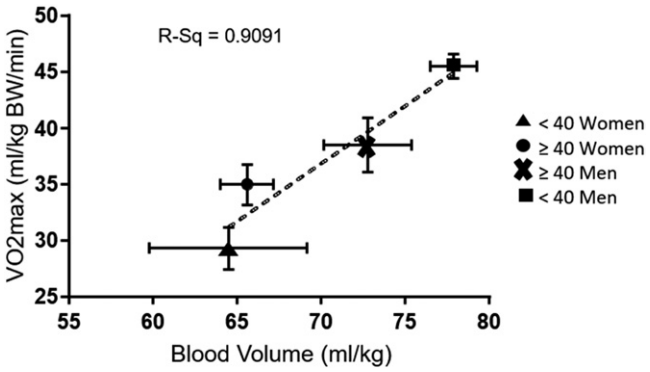

Fig. 1. Linear regression of $\dot{V}O_{2max}$ (ml · kg^{-1} BW · min^{-1}) and BV (ml · kg^{-1}) between men and women in different age groups. The linear regression equation is y = 1.023 * X – 34.73.

While we found $\dot{V}O_{2max}$ to be unaltered in women over an average period of 23 yr (Table II) compared to > 20% reduction in $\dot{V}O_{2max}$ in men over an average period of only 12 yr (Table II), the explanation for this finding is unclear. Clearly, the lower $\dot{V}O_{2max}$ of the older population in men cannot be accounted for by duration of aging, as the difference in population age was 40% greater in women who showed no alteration in $\dot{V}O_{2max}$. However, the men in the present study had an average $\dot{V}O_{2max}$ > 30% higher than the women, even when standardized for body weight. This is consistent with previous findings[6,8] that men tend to display higher fitness levels than women. Data obtained from previous experiments conducted by NASA investigators on the deconditioning effects of inactivity (bed rest) support the notion that 'fit' individuals with relatively high initial $\dot{V}O_{2max}$ display twice the reduction in $\dot{V}O_{2max}$ compared to less fit cohorts.[4,8] As such, it is likely that one contributing factor to the observation that our older men displayed a significantly lower $\dot{V}O_{2max}$ than the younger men could be a result of a failure to maintain physical activity at a level necessary to avoid a deconditioning effect not observed in the less fit younger and older women populations.

Lower muscle mass has also been associated with lower $\dot{V}O_{2max}$ between sexes[22] and with aging.[3,35] We found no difference in lean mass between our younger and older male subjects, suggesting that the lower $\dot{V}O_{2max}$ in older men could not be explained by lower muscle mass. Contrary to the findings in the men, the older women demonstrated ~20% lower lean mass than the younger women. However, against expectations, we observed no statistical difference in $\dot{V}O_{2max}$ between the younger and older women. As such, the absence of higher $\dot{V}O_{2max}$ in younger women cannot be explained by either differences in blood volume or muscle mass. Although our database did not include any assessment of habitual physical activity, we cannot dismiss the possibility that our older women were inherently more physically active and fit than our younger women.

Data from previous studies have been used to support the notion that changes in BV associated with menopause may be

influenced by varying levels of estrogen and progesterone[15,33]; however, these observations were without reference to changes in fitness with aging. Conversely, we found no difference in BV or $\dot{V}O_{2max}$ of our female subjects when groups between < 40 yr old and ≥ 40 yr old were compared. Given that women in the present investigation were either premenopausal or postmenopausal by self-report, our finding suggests that there may be other factors besides menopause that also have a strong influence in the control of BV.

Additionally, Jones and colleagues reported no difference in BV when the $\dot{V}O_{2max}$ of premenopausal sedentary women was similar to postmenopausal physically active women.[21] The absence of a difference in BV and its components observed in our women was associated with no difference in $\dot{V}O_{2max}$. As such, the evidence from our study taken together with data reported in the literature support the notion that BV may not be influenced by menopause if $\dot{V}O_{2max}$ can be maintained. Thus, the ability of women to maintain aerobic capacity following the onset of menopause appears to be an important influencing factor in avoiding a reduction in BV.

Previous studies have shown total BV (and its plasma volume, red cell volume and hematocrit) decreases with age in healthy men and women.[14,21] Inconsistent with these findings, aging was associated with no change in BV among men and women in the present study. Explanations for the conflicting results could include different age ranges and initial aerobic capacity of the individuals. For the men, the reduced BV in the study of Davey et al.[14] could have been the result of a greater age range (25–66 yr of age) between their younger and older men compared to our study (22–38 yr of age). For the women, the ranges in age and $\dot{V}O_{2max}$ of our women were similar to those of Jones et al.[21] who reported lower $\dot{V}O_{2max}$ with aging when comparing physically-active young and older women to sedentary young and older women. However, the average difference in $\dot{V}O_{2max}$ of our younger and older women ($\Delta = 2.3\ ml \cdot kg^{-1} \cdot min^{-1}$) was statistically similar in magnitude to that of the younger sedentary and older physically active ($\Delta = 4.7\ ml \cdot kg^{-1} \cdot min^{-1}$) women reported by Jones et al.[21] Thus, the ability to sustain a level of activity and $\dot{V}O_{2max}$ with aging in our older women might have accounted for the absence of difference in BV and $\dot{V}O_{2max}$ between our younger and older women.

There is compelling and consistent evidence that $\dot{V}O_{2max}$ is associated with BV, with an increase in both BV and $\dot{V}O_{2max}$ induced by chronic exercise training,[4] while a reduction in both BV and $\dot{V}O_{2max}$ accompanies deconditioning with reduced physical activity.[4,11] The finding of the present investigation supports this relationship between BV and $\dot{V}O_{2max}$ (Fig. 1). Despite these established relationships, we observed a significantly ($\sim$23%) lower $\dot{V}O_{2max}$ with $< 4\%$ lower BV in older men compared to their younger cohorts. However, other longitudinal studies have demonstrated that BV expansion is not a prerequisite for an increase in aerobic fitness since they also reported increased $\dot{V}O_{2max}$ with very little change in BV.[30,31,37]

The reduction in $\dot{V}O_{2max}$ with aging in the absence of reduced circulating BV in our men suggests that other mechanisms contributed to maximal delivery and utilization of oxygen in older men. It is well recognized that aerobic performance is reduced with aging.[13,24] Aging is also associated with muscle atrophy,[4,35] reduced systemic oxygen delivery resulting from lowered cardiac function,[28,29] less capillary density,[28] and lowered muscle tissue oxidative capacity reflected by reduced mitochondrial volume density,[3,29] myoglobin,[2] and oxidative enzyme activity.[3,29] In addition, central and peripheral adaptations such as reduced maximum heart rate (HR_{max})[18,19] and lean body mass[19,32] have been proposed to be associated with cardiovascular decline with aging. Any of these factors alone or in combination with reduced physical inactivity (i.e., deconditioning effect) associated with aging could result in reduced $\dot{V}O_{2max}$ without a reduction in BV. The importance of the current study is that the significant reduction in $\dot{V}O_{2max}$ between younger and older men without a difference in BV shows for the first time that total oxygen carrying capacity must be dwarfed in its contribution to the lower aerobic capacity with aging compared to other physiological functions that support total systemic oxygen utilization.

Previous studies with cross-sectional comparisons have failed to show a consistently clear distinction between men and women in total circulating BV.[1,16,34,35] **Table III** presents a summary of studies that address the association of sex with BV and its components. Our values of BV, plasma volume, red cell volume, and hematocrit for women were similar to those reported by Wadsworth.[34] Also consistent with our findings, statistically greater red cell volume and hematocrit in men were reported by Fu et al.[16] and Wennesland et al.[16,34] However, contrary to our findings, both Fu et al.[16] and Wennesland et al.[16,34] reported no sex differences in blood or plasma volumes corrected for body weight. In the case of the Fu study, no sex difference was reported with matched plasma and blood volumes of a relatively small sample size that allowed for experimentally-designed comparison of orthostatic tolerance in men and women. In the case of the investigations conducted by Wennesland et al.[16,34] that included a large cohort of more than 200 men and 100 women, the quantitative values reported for plasma volume were noticeably smaller than those measurements reported in the present study and by other investigators (Table III). This discrepancy could be a result of the more sophisticated purification techniques used in the present study that were not available to Wennesland.[16,34] As such, quantitative classifications, demographics, and interpretations drawn from early cross-sectional comparisons of BV and its components without validation and verified repeatability should be approached with caution.

Like all investigations, our study is not without limitations. Reduced muscle mass is associated with lower $\dot{V}O_{2max}$.[22] Although muscle mass was not directly assessed in all of the subjects studied in the present investigation, a limited data set that included measures of body composition was conducted. With the assumption that differences in muscle mass are reflected by differences in lean mass, our data revealed that the difference in $\dot{V}O_{2max}$ observed between our groups was not altered when expressed per weight of lean body mass. Likewise, our database included information from a physical activity

Table III. Comparison of Standardized values for Plasma Volume, Blood Volume, Red Cell Volume, Hematocrit, and Methodology Among Previous Studies Comparing Blood Volume and Sex versus the Present Study.

FU ET AL. 2003[16]			
	MEN	**WOMEN**	**DIFFERENCE (ml · kg^{-1})**
N	13	10	-
Plasma Volume (ml · kg^{-1})	38.0 (1.0)	42.0 (2.0)	-4.0
Blood Volume (ml · kg^{-1})	63.0 (3.0)	64.0 (3.0)	-1.0
Red Cell Volume (ml · kg^{-1})*	29.0	24.9	4.1
Hematocrit (%)*	44.0 (0.6)	38.9 (0.7)	5.1
Method	Evans Blue Dye		-
WENNESLAND ET AL. 1958[36]/BROWN ET AL. 1962[1]			
	MEN	**WOMEN**	**DIFFERENCE (ml · kg^{-1})**
N	201	101	-
Plasma Volume (ml · kg^{-1})	34.3 (0.3)	35.6 (0.4)	-1.3
Blood Volume (ml · kg^{-1})	62.5 (0.4)	61.7 (0.6)	0.8
Red Cell Volume (ml · kg^{-1})*	28.2 (0.2)	25.1 (0.3)	3.1
Hematocrit (%)*	45.2 (0.2)	40.8 (0.2)	4.4
Method	Cr^{51}/Evans Blue Dye		-
WADSWORTH 1954[35]			
	MEN	**WOMEN**	**DIFFERENCE (ml · kg^{-1})**
N	-	8	-
Plasma Volume (ml/kg)	-	43.1 (1.9)	-
Blood Volume (ml · kg^{-1})	-	66.5 (2.4)	-
Red Cell Volume (ml · kg^{-1})	-	23.4 (0.7)	-
Hematocrit (%)	-	35.1	-
Method	P^{32}/Evans Blue Dye		-
PRESENT STUDY 2018			
	MEN	**WOMEN**	**DIFFERENCE (ml · kg^{-1})**
N	60	24	-
Plasma Volume (ml · kg^{-1})*	43.9 (0.9)	41.7 (1.4)	2.2
Blood Volume (ml · kg^{-1})*	73.9 (1.7)	65.2 (2.7)	8.7
Red Cell Volume (ml · kg^{-1})*	29.9 (1.0)	23.5 (0.9)	6.4
Hematocrit (%)*	44.9 (0.8)	37.8 (1.3)	7.1
Method	Evans Blue Dye		-

Values are means ± SE; N, number of subjects; * indicates $P < 0.01$.

questionnaire on only ~5% of subjects, so we cannot dismiss the potential influence of reduced levels of lifestyle physical activity that can occur with aging.[13] Third, in the absence of estrogen measurements, we were unable to verify pre- and postmenopausal classification by self-reporting; although there is evidence that self-reporting is reliable.[10] Finally, interpretation of results generated from historical data can be influenced by large variability created by differences in data collection technique and various investigators. In contrast, the control of experimental variability was a strength in the present investigation given that all measurements of $\dot{V}O_{2max}$ and blood volume were conducted by the same investigator using the same experimental techniques and protocols.

CONCLUSION

Using an historic database generated from NASA and U.S. Air Force experiments in which BV and $\dot{V}O_{2max}$ were measured in men and women across age, we corroborated previous investigations that women have smaller circulating red cell volumes and hematocrits[1,25,36] and lower $\dot{V}O_{2max}$ [6,8,12] compared to men. However, in contrast to these earlier comparisons,[1,25,36] we observed that plasma and blood volumes were also greater in men compared to women when standardized for body mass. Against our expectations and in contrast to previous findings,[14,21] we report for the first time that increased age was not associated with a reduction in BV in both men and women and appeared to be uncoupled with $\dot{V}O_{2max}$ in men. The absence of difference in $\dot{V}O_{2max}$ between our younger and older women suggests that physical activity with advancing age has a stronger influence in the maintenance of BV than menopause. Our results provide evidence that aging may not compromise men and women in scenarios where BV can affect performance in aerospace and military environments.

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Authors and affiliations: Natalie J. Koons, B.S., Mithun Suresh, Ph.D., M.S., Taylor E. Scholtman, Ph.D., M.S., Victor A. Convertino, Ph.D., M.A., U.S. Army Institute of Surgical Research, San Antonio, TX.

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