Genetic, Physiologic, and Behavioral Predictors of Cardiorespiratory Fitness in Specialized Military Men

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ABSTRACT Introduction: Cardiorespiratory fitness (CRF) is a crucial performance requirement of specialized military occupations. Age and physical activity are established predictors of CRF, but it is not clear how these predictors combine with each other and/or with genetic predisposition. The goal of this study was to derive inclusive explanatory models of CRF in US Navy Explosive Ordnance Disposal (EOD) operators, synthesizing conventional (e.g., age, body composition, and physical activity) and novel influences (e.g., genetic variance). Materials and Methods: In this cross-sectional study, 40 male, active duty EOD operators completed a graded exercise test to assess maximal oxygen consumption and ventilatory threshold (VT) using the Bruce protocol. Aerobic performance was further quantified via time of test termination and time at which VT was achieved. Body composition was determined via dual x-ray absorptiometry, and physical activity was assessed by self-report. Genetic variants underlying human stress systems (5HTTLPR, BclI, −2 C/G, and COMT) were assayed. Descriptive analyses were conducted to summarize subject characteristics. Hypotheses were tested with linear regression models. Specifically, separate univariate regression models first determined associations between each of the independent and dependent variables. This protocol was approved by the Naval Health Research Center Institutional Review Board (NHRC.2015.0013). Results: In univariate regression models, age, body composition, physical activity, and 5HTTLPR consistently predicted CRF and/or aerobic performance (R^2 range 0.07–0.55). Multivariate regression models routinely outperformed the univariate models, explaining 36–62% of variance. Conclusion: This study signifies a shift toward inclusive explanatory models of CRF and aerobic performance, accounting for combined roles of genetic, physiologic, and behavioral influences. Although we were able to quantify combined effects, we were unable to evaluate interaction effects (e.g., gene–gene, gene–behavior) due to limited statistical power. Other limitations are that this specialized military population may not readily generalize to broader populations, and the current sample was all male. Considering these limitations, we aim to replicate this study in various populations, both male and female. Despite its limitations, this study reflects a shift toward more comprehensive predictive models of CRF, explaining the unique and shared contributions of genetic predisposition, physiology, and behavior. These findings have implications for assessment, selection, and training of specialized military members, and may also impact mission success and survivability. Future studies are needed to better characterize additive, interactive, and mediated effects.

INTRODUCTION

Cardiorespiratory fitness (CRF) is a crucial performance requirement of military occupations.[1,2] Emerging evidence shows that CRF not only reduces injury risk in military and civilian populations,[3,4] but also mitigates health risks, all-cause mortality, and disease-specific mortality.[5] Recognized correlates of CRF include age, body composition,[6] and physical activity.[7] It is not clear how such predictors combine with each other, and/or with other influences, such as genetic predisposition.

Age is routinely linked to a decline in CRF.[6,8] It is known, however, that many older individuals have aerobic capacities comparable to or greater than those of younger individuals.[9] This implies that age interacts with other contributing factors, such as physical activity, body composition, and genetic predisposition. The nature and extent of such combined effects are not known. Moreover, the influence of the aging process on CRF in relatively young (e.g., 25–45 years), healthy military members is not well understood. Also, physical activity is not only associated with CRF,[9] but is also a dose–response indicator of cardiovascular disease[10] and mortality.[11] For numerous health outcomes, some physical activity is deemed better than none, while

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more is better than some, up to a point of diminished benefit.[9,11] An improved understanding of the link between physical activity and CRF could have important implications for the prevention of cardiovascular and other diseases. Furthermore, in many military populations, CRF is a crucial performance requirement, and physical activity training is routinely administered to improve it. With this in mind, a precise understanding of the association between physical activity and CRF could help to inform and optimize physical training programs for military populations. Moreover, negative associations have been observed between body composition and CRF.[12–14] However, the association between body composition and CRF in relatively young, healthy military members is not well characterized.

Finally, recent studies have estimated the heritability of CRF. Schutte et al.'s[15] meta-analysis of VO_{2max} in children and young adults yielded heritability estimates of 59–72%. Other work links the angiotensin-converting enzyme (ACE) insertion/deletion polymorphism to CRF and/or aerobic performance.[16,17] Bueno et al.[16] observed higher VO_{2max} in physically active young men who were ACE II genotype carriers compared with DD and ID genotype carriers. On a closely related topic, some literature explores genetic predictors of aerobic training response[18] and physical activity participation.[19] Williams et al.[18] reviewed 35 candidate gene and genome-wide association studies and identified 97 genes as potential contributors to VO_{2max} trainability. In that study, 13 predictive variants were replicated across at least two studies; these variants primarily underlie lipid and skeletal muscle metabolism, calcium signaling, transcriptional and apoptotic regulation, angiogenesis, and mitochrondrial biogenesis.

Despite this more recent work, there is a knowledge gap regarding the role of genetic regulators of human stress systems (e.g., −2 C/G, BclI) in relation to CRF. This is surprising, given that both sympathoadrenergic[20] and adrenocortical systems[21] respond to acute exercise stress and may adapt to aerobic exercise training.[22] High CRF is linked to more optimal stress hormone profiles,[23] and some evidence supports the "cross-stressor adaptation hypothesis," which asserts that adaptations to aerobic exercise training correspond to adaptations in reactivity to non-physical (e.g., psychological) stressors.[24] In light of this, there is a need to understand the role of genetic variants underlying human stress systems in relation to CRF. Plausible candidate genetic variants underlying human stress processes include BclI, −2 C/G, COMT, and 5HTTLPR. Whereas BclI affects cortisol feedback regulation,[25] hypertension,[26] and abdominal obesity,[27] −2 C/G influences cortisol levels, renin–angiotensin system activation, and systolic blood pressure.[28] Likewise, 5HTTLPR influences both activation and feedback control of the hypothalamic–pituitary–adrenal axis.[29] Finally, COMT regulates the metabolism of catecholamine neurotransmitters (i.e., dopamine, norepinephrine, and epinephrine).[30] To date, we have shown that BclI, −2 C/G, and 5HTTLPR regulate adrenocortical and/or cardiovascular responses in humans exposed to intense military stress.[31] However, such genetic variants have not been examined in relation to CRF.

Altogether, it is important to understand the combined influences of demographic, genetic, physiologic, and behavioral factors associated with CRF. Although there is some preliminary evidence characterizing the combined effects of body composition and various genetic traits on responsiveness to exercise,[19] to our knowledge, this has not been explored in military populations. This line of research may lead to comprehensive predictive models that explain the unique (independent) and shared contributions of genetic predisposition, behavior, training adaptations, and environmental exposure in this unique population. Ultimately, this could have implications not only for the assessment, selection, and training of specialized military members, but may also impact mission success and survivability. The goal of this study was to derive inclusive explanatory models of CRF and aerobic performance in US Navy Explosive Ordnance Disposal (EOD) operators, synthesizing conventional (e.g., age, body composition, and physical activity habits) and novel influences (e.g., genetic variance). US Navy (EOD) operators are the premier combat force for countering explosive hazards and must operate in austere environments. Therefore, EOD operators must regularly demonstrate and maintain superior physical fitness. We expected that both conventional and novel factors would associate with CRF in these men. We further hypothesized that these factors would combine to explain substantial variance (e.g., >30%) in CRF and/or aerobic performance, decisively exceeding any individual predictor alone.

METHODS

Participants

Forty male, active duty EOD operators participated in this study. Participant demographic characteristics are presented in Table I, while independent and dependent variables are displayed in Table II. This protocol was approved by the Naval Health Research Center Institutional Review Board (NHRC.2015.0013).

Determination of Genotypes

We evaluated the following candidate variants in the glucocorticoid receptor (BclI, rs41423247, NR3C1, chromosome 5), mineralocorticoid receptor (−2 C/G, rs2070951, NR3C2, chromosome 4), catecho-O-methyltransferase (COMT, rs737865, chromosome 22), and serotonin transporter genes (5HTTLPR; biallelic; Long [L] vs. Short [S]) and triallelic versions [5HTTLPR/rs25531; Long-Adenine [LA] vs. Long-Guanine [LG]+S, chromosome 17).[31,32] A modified cell extraction method using Gentra® Puregene® Cell Kit (Qiagen, Valencia, CA) was used to isolate the DNA from the saliva samples. For BclI, −2 G/C, and COMT single nucleotide polymorphisms, a TaqMan® genotyping assay (Applied Biosystems™ 7500 Real-Time PCR System, Thermo Fisher Scientific Inc.,

TABLE I. Participant Characteristics

Variable	*n*	%
Age, year		
25–29	8	20.0
30–39	19	47.5
40+	13	32.5
Pay grade		
Enlisted	29	72.5
Officer	11	27.5
E5–E6	6	15
E7–E9	23	57.5
O1–O3	8	20
O4+	3	7.5
Race/ethnicity		
White	38	95.0
Other	1	2.5
Missing	1	2.5
Years in military		
3–5	4	10.0
6–9	7	17.5
10+	29	72.5
Marital status		
Married/cohabitating	28	70.0
Divorced	5	12.5
Never married	7	17.5
Education		
High school	23	57.5
Bachelor's or higher	17	33.5
No. of deployments		
0	3	7.5
1	5	12.5
2	6	15.0
3	7	17.5
4+	19	47.5

Foster City, CA) was employed to amplify and evaluate the two alleles at their respective target locations. To conduct variable number tandem repeat (VNTR) analyses for 5HTTLPR, DNA templates were amplified for the region of interest using polymerase chain reaction (PCR) and VNTR-specific fluorescent-labeled primers. The amplified DNA products were then analyzed by capillary electrophoresis to detect the number of repeats present, which was then used to categorize the VNTR genotype. A 22-base pair (bp) repeat (44-bp insertion/deletion) located in the promoter region was investigated following the method used by Wendland et al.[33]

Body Composition

Body composition assessment was performed by a state certified radiologic technician using dual-energy X-ray absorptiometry (Horizon® A DXA, Hologic, Inc., Marlborough, MA). Data were processed and analyzed with APEX™ software (version 5.0, Hologic, Inc., Marlborough, MA). Participants removed any jewelry and wore clothing free of metal fasteners (e.g., zippers, buttons) that could register as artifact. Quality control and calibration were performed prior to each data collection using a spine phantom made of calcium hydroxyapatite embedded in a lucite block. The percent coefficient of variation for all calibrations was less than 1.5% from the average value.

Physical Activity

Two items assessed participants' physical activity during the past month. Participants were asked, on average, how often they engaged in vigorous physical activity (e.g., jogging or running) for at least 20 min/d, and moderate physical activity (e.g., brisk walking, lawn mowing) for at least 30 min/d. Participants answered on a 6-point Likert-type scale, from 1 (*none/never*) to 6 (*5 or more times per week*). Combined responses were used to determine whether the subject met the physical activity recommendation of the American College of Sports Medicine/American Heart Association (ACSM/AHA) for healthy adults to promote and maintain health (i.e., ≥30 min of moderate intensity physical activity ≥5 d/wk, ≥20 min of vigorous intensity physical activity ≥3 d/wk, or a combination of both).[34] Accordingly, an intermediate variable was computed as [(days per week of moderate physical activity × 1.2) + (days per week of vigorous physical activity × 2)]. Subjects were then classified as meeting/exceeding the ACSM/AHA recommendation with a score of ≥6.

Cardiorespiratory Fitness

Cardiorespiratory fitness was assessed during a graded exercise test (GXT) on a treadmill using the standard Bruce protocol. Target start time for all GXTs was between 1000 and 1400. Participants were given clear, standardized instructions to follow prior to the lab visit. Specifically, they were instructed to drink ample fluids the night before and in the morning prior to the visit, to avoid antihistamines, decongestants, allergy medications, aspirin, ibuprofen, and nonsteroidal anti-inflammatory drugs 24 hours prior; to avoid alcohol 12 hours prior; to refrain from exercise and vigorous physical activity 8 hours prior, and to avoid caffeine, energy drinks, and large meals 2 hours prior to the visit. The maximal oxygen consumption (VO_{2max}) was measured via gas exchange (TrueOne® 2400 metabolic measurement system, Parvo Medics, Sandy, UT) and the time of termination (TT) for the GXT was recorded. Ventilatory threshold (VT) and time of ventilatory threshold (VTT) were automatically calculated during the test and then independently verified by two trained exercise physiologists. Specifically, the minute ventilation (VE) curve [VE/VO_2]; point at which nonlinear increase in ventilation occurs; and V-slope (VO_2 vs. VCO_2); point at which the increase in VCO_2 exceeds the increase in VO_2) were automatically calculated. Both were also evaluated by the trained physiologists, independently and while blinded to the automated results. The physiologists then triangulated across all methods, reconciling differences and reaching consensus regarding the time and VO_2 corresponding to VT.

TABLE II. Independent[a] and Dependent[b] Variables

Variable	*n*	%	Range	Mean	SD
BclI					
CC	4	10.0			
CG	17	52.5			
GG	19	47.5			
−2 C/G					
CC	9	22.5			
CG	23	57.3			
GG	8	20			
COMT					
AA	26	65			
AG	11	27.5			
GG	3	7.5			
5HTTLPR (biallelic)					
LL	14	35.0			
SL	20	50.0			
SS	6	15.0			
5HTTLPR (triallelic)					
LL	8	20.0			
SL	25	62.5			
SS	7	17.5			
Body fat percentage	40		11.4–26.0	17.9	3.7
Vigorous physical activity (d/wk)					
1	4	10.0			
2	10	25.0			
3	6	15.0			
4	14	35.0			
5+	6	15.0			
Moderate physical activity (d/wk)					
1	3	7.5			
2	8	20.0			
3	6	15.0			
4	7	17.5			
5+	15	37.5			
Missing	1	2.5			
ACSM/AHA physical activity guidelines					
Meet/exceed	39	97.5			
Missing	1	2.5			
VO_{2max} (ml/kg/min)	40		37.2–64.2	46.1	6.5
Ventilatory threshold (ml/kg/min)	40		17.4–41.5	26.7	5.0
Time of termination (min)	40		9.6–16.0	12.0	1.5
Time of ventilatory threshold (min)	40		3.8–10.0	5.9	1.3

[a]Independent variables included genetic variance, body composition, and physical activity.
[b]Dependent variables included cardiorespiratory fitness and aerobic performance.
ACSM/AHA, American College of Sports Medicine/American Heart Association; SD, standard deviation; VO_{2max}, maximal oxygen consumption.

Statistical Analysis

Data were analyzed using IBM SPSS software, version 23 (IBM Corporation, Armonk, NY). Descriptive analyses were conducted to summarize subject characteristics. Tests for departure from Hardy–Weinberg equilibrium were performed via chi-square test for goodness of fit. [35] Hypotheses were tested with linear regression models. Specifically, separate univariate regression models first determined associations between each of the independent (age, body composition, physical activity, and genetic variance) and dependent variables (VO_{2max}, VT, TT, and VTT). Dichotomous predictors were coded as 0/1. Significant univariate predictors ($p < 0.05$) of each dependent variable were then entered in multiple regression models. Genotypes were decomposed into alleles reflecting either a dominant (e.g., if hypothesized risk factor = G, then GX + GG vs. XX) or recessive model (if hypothesized risk factor = G, then GG vs. GX + XX).[35,36]

RESULTS

Participant Characteristics

As shown in Table I, most participants (47.5%) were aged 30–39 year, the majority (97.4%) were white, and most (72.4%) were enlisted. The majority (74.1%) had been in the military for 10 or more years, and half (47.5%) had

experienced four or more military deployments. Nearly half (46.6%) had a bachelor's degree or higher education.

As displayed in Table II, the majority of participants were heterozygous for all of the genetic variants except for COMT. This sample did not depart from Hardy–Weinberg equilibrium for any of the genetic variants. The majority of participants (65%) had engaged in vigorous physical activity 3 or more days per week during the past month, and 37.5% had engaged in moderate physical activity 5 or more times per week. Nearly all participants (97%) met the ACSM/AHA guidelines for physical activity (one participant had missing data). The average body fat percentage was 18% (ACSM normative classification: "good" is 60–65th percentile for 30- to 39-year-old males.[9] Mean VO_{2max} was 46.1 ml/kg/min (ACSM normative classification: "good" is 70–75th percentile for 30- to 39-year-old males).[9]

Genetic, Physiologic, and Behavioral Predictors of Cardiorespiratory Fitness: Univariate Associations

Univariate associations between the independent and dependent variables are summarized in Table III. Age predicted VO_{2max} ($F = 4.3$, $\beta = -0.32$, $p < 0.05$) and VT ($F = 4.7$, $\beta = -0.33$, $p < 0.05$). Accordingly, younger participants (<30 years, $n = 8$) registered higher VO_{2max} (50.2 ± 8.4 ml/kg/min) and VT (29.9 ± 6.9 ml/kg/min) than older participants (≥30 year, $n = 32$; VO_{2max}: 45.1 ± 5.6 ml/kg/min; VT: 25.9 ± 4.1 ml/kg/min). Likewise, 5HTTLPR (triallelic version) predicted both VT ($F = 4.4$, $\beta = -0.32$, $p < 0.05$) and VTT ($F = 8.2$, $\beta = -0.42$, $p < 0.01$). Homozygous L-carriers ($n = 8$) registered higher VT (29.8 ± 6.5 ml/kg/min) and VTT (7.0 ± 1.9 min) than S-carriers ($n = 32$; VT: 25.9 ± 4.2 ml/kg/min; VTT: 5.6 ± 1.0 min). Body composition was a robust predictor of all four endpoints. Namely, lower body fat percentage predicted higher VO_{2max} ($F = 14.0$, $\beta = -0.52$, $p = 0.001$), VT ($F = 16.2$, $\beta = -0.55$, $p < 0.001$), TT ($F = 45.5$, $\beta = -0.74$, $p < 0.001$), and VTT ($F = 14.0$, $\beta = -0.52$, $p < 0.001$). Finally, vigorous physical activity robustly predicted all four endpoints (VO_{2max}: $F = 13.7$, $\beta = 0.51$, $p = 0.001$; VT: $F = 7.1$, $\beta = -0.40$, $p = 0.01$), TT ($F = 14.5$, $\beta = 0.53$, $p < 0.001$), and VTT ($F = 7.2$, $\beta = 0.40$, $p = 0.01$). Moderate physical activity was not evaluated as a predictor of CRF because of the high prevalence of, and substantial overlap with, vigorous physical activity in this sample.

Dose–Response Associations Between Vigorous Physical Activity and Cardiorespiratory Fitness

Dose–response associations between vigorous physical activity and VO_{2max} ($F = 3.9$, $p = 0.01$) are displayed in Figure S1. A threshold effect was implied. Specifically, increases in vigorous physical activity participation from 1 to 3 days did not appreciably alter VO_{2max}. However, vigorous physical activity participation beyond 3 days corresponded to stepwise increases in VO_{2max}. A nearly identical pattern was observed for the other three endpoints: VT ($p = 0.07$), TT ($p < 0.01$), and VTT ($p = 0.08$).

Genetic, Physiologic, and Behavioral Predictors of Cardiorespiratory Fitness: Multivariate Associations

Multivariate regression models are detailed in Tables IV, V, S1, and S2. As shown in Table IV, age, body composition, and physical activity combined to explain 56% of variance in VO_{2max}, while both body composition ($p < 0.001$) and physical activity ($p < 0.01$) contributed uniquely to the model. Age, body composition, physical activity, and 5HTTLPR combined to explain 36% of variance in VT; body composition was the sole independent predictor ($p < 0.001$; Table V). Body composition ($p < 0.001$) and physical activity ($p < 0.01$) combined to explain 62% of variance in TT; both maintained unique predictive value (Table S1). Finally, 5HTTLPR, body composition, and physical activity explained 36% of variance in VTT; 5HTTLPR and body composition each contributed uniquely (Table S2).

DISCUSSION

In this study, inclusive explanatory models of CRF and aerobic performance in specialized military men routinely outperformed univariate models, implying that substantial value is realized by incorporating diverse genetic, behavioral, and physiologic factors in predictive models of CRF.

As hypothesized, age inversely associated with VO_{2max} and VT, explaining 10% and 11% of variance, respectively. This not only resonates with prior studies showing a link between the aging process and VO_{2max} in diverse populations,[6,8] but it also elucidates two knowledge gaps. Specifically, the established relationship between age and CRF extrapolates to healthy military members within a

TABLE III. Univariate Predictors of Cardiorespiratory Fitness and Aerobic Performance

Variable	VO_{2max}	Ventilatory Threshold	Time of Termination	Time of Ventilatory Threshold
Age[a]	0.10*	0.11*	0.08	0.07
5HTTLPR[b]	0.08	0.10*	0.09	0.18**
Body composition	0.53***	0.30***	0.55***	0.27***
Vigorous physical activity	0.26***	0.16**	0.28***	0.16**

*$p < 0.05$; **$p < 0.01$; ***$p < 0.001$; all values are R^2.
[a]Dichotomized as <30 years versus ≥30 years.
[b]Triallelic version, S-dominant model (dichotomized as 0 = non-carrier, 1 = carrier). Other candidate genetic variants (e.g., BclI) did not associate with cardiorespiratory fitness or aerobic performance.

TABLE IV. Combined Associations of Age, Body Composition, and Physical Activity With Maximal Oxygen Consumption (VO_{2max})

Variable	Standardized β	*t*	*p*
Age	−0.20	−1.8	ns
Body composition	−0.53	−4.6	<0.001
Vigorous physical activity	0.34	3.1	<0.01

Adjusted $R^2 = 0.56$, $F = 17.7$, $p < 0.001$.
ns, not significant.

TABLE V. Combined Associations of Age, Body Composition, Physical Activity, and 5HTTLPR With Ventilatory Threshold

Variable	Standardized β	*t*	*p*
Age	−0.21	−1.6	ns
5HTTLPR	−0.15	−1.1	ns
Body composition	−0.39	−2.8	<0.01
Vigorous physical activity	0.24	1.8	ns

Adjusted $R^2 = 0.36$, $F = 6.5$, $p = 0.001$.
5HTTLPR, a variant of the serotonin transporter gene; ns, not significant.

relatively restricted age range. Second, the results imply that the link between age and CRF extends to *submaximal* fitness indicators (i.e., VT), which may have more relevance to the military context.[2] However, age did not uniquely predict either CRF endpoint in combined models. This is consistent with the fact that many older individuals have aerobic capacities comparable to or greater than those of younger individuals. In turn, this supports the argument that age interacts with other contributing factors, such as physical activity, body composition, and/or genetic predisposition. A logical clinical and public health translation, then, is that maintaining a healthy body composition and an adequate frequency of vigorous activity throughout the aging process has the potential to offset, delay, or mitigate aging effects, even within the relatively restricted age range of the generally healthy men in this study.

Body fat percentage was the most powerful and robust predictor of CRF and aerobic performance. It not only associated distinctly with all four endpoints, but it also contributed substantively and uniquely to every combined model. This is consistent with prior studies in diverse populations involving various methods of body composition measurement.[12,13] Further, the strength and robustness of the current finding summon Schnurr et al.'s[14] evidence that the link between body composition and CRF might be governed by shared genetic etiology. In that study, a genetic risk score comprising alleles of known relevance to body fat percentage and a fat mass/obesity-related allele each associated convincingly with VO_{2max} in a large cohort of men and women. Additional work clarifying the strength and complexity of associations between body fat, CRF, and genetic predisposition are warranted. Synthesized, this work will lead to more inclusive explanatory models of CRF and aerobic performance.

As hypothesized, vigorous physical activity was a consistent predictor of CRF and aerobic performance. It associated with all four endpoints in the univariate models and maintained a unique contribution to VO_{2max} and TT in combined models. While this supports the existing research,[9] we further demonstrated dose–response associations between frequency of vigorous physical activity and CRF. Specifically, a threshold effect was implied; although vigorous physical activity participation for 1, 2, or 3 days did not clearly associate with VO_{2max}, doing vigorous physical activity for more than 3 days associated with stepwise increases in VO_{2max}. This has implications for physical training programming, suggesting that it is vital to exceed 3 days of vigorous physical activity participation to optimize CRF in this unique military population.

The genetic variant 5HTTLPR explained up to 18% of variance in submaximal CRF, with homozygous L-carriers registering substantially higher VT and VTT than S-carriers. 5HTTLPR also combined with physical activity and body composition to convincingly predict VTT.

To our knowledge, this is the first study to establish an association between genetic variants underlying human stress systems in relation to CRF. We hypothesized that stress-related genetic variants may associate with CRF because sympathoadrenergic and adrenocortical systems associate with CRF, respond to acute exercise stress, and may also adapt to aerobic exercise training. Furthermore, adaptations to aerobic exercise training have been shown to correspond to adaptive changes in reactivity to psychological stressors. We previously identified associations between 5HTTLPR and adrenal and cardiovascular responses to intense military stress, including a period of mock captivity.[31] Namely, 5HTTLPR modulated cortisol (SS > L), heart rate (L > SS), and diastolic blood pressure responses (S > LL). Potential links between 5HTTLPR, cardiovascular/hormonal responses to acute exercise stress, and submaximal/maximal indicators of CRF is a topic of future inquiry in our lab. Also, we aim to evaluate the moderating role of stress-related genetic variants in daily hormone profiles of these men.[36] Synthesized, these studies will help to explain how genetic modulators of human stress systems are tied to CRF.

There are some limitations of this study. Although we were able to quantify combined effects, we were unable to evaluate *interaction* effects (e.g., gene–gene, gene–behavior) due to limited statistical power.[37] Future studies are needed to better characterize additive, interactive, and mediated effects. For example, genetic factors that negatively affect CRF may be mitigated or nullified by engagement in vigorous physical activity beyond 3 days/week (an additive effect), such counterbalancing may be sex-dependent (an interaction effect), or the interrelationships between physical activity and fitness may be explained by telomere length (a mediated effect). Other limitations are that physical activity

was measured by self-report, that this specialized military population may not readily generalize to broader populations, and the current sample was all male. Considering these limitations, we aim to replicate this study in various populations, both male and female. Despite its limitations, this study reflects a shift toward more comprehensive predictive models of CRF, explaining the unique and shared contributions of genetic predisposition, physiology, and behavior. Ultimately, this may have implications not only for the assessment, selection, and training of specialized military members, but may also impact mission success and survivability.

SUPPLEMENTARY DATA

Supplementary data are available at *Military Medicine* online.

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14. ABSTRACT

Cardiorespiratory fitness (CRF) is a crucial performance requirement of specialized military occupations. Age and physical activity (PA) are established predictors of CRF, but it is not clear how these predictors combine with each other, and/or with genetic predisposition. The goal of this study was to derive inclusive explanatory models of CRF in U.S. Navy Explosive Ordnance Disposal (EOD) men, synthesizing conventional (e.g., age, body composition and physical activity) and novel influences (e.g., genetic variance). Forty male, active-duty EOD operators completed a graded exercise test (GXT) to assess maximal oxygen consumption (VO2max) and ventilatory threshold (VT) using the Bruce protocol. Aerobic performance was further quantified via time of test termination and time at which VT was achieved. Body composition was determined via dual x-ray absorptiometry, and physical activity was assessed by self-report. Genetic variants underlying human stress systems (5HTTLPR, BclI, −2C/G, and COMT) were assayed. In univariate regression models, age, body composition, physical activity, and 5HTTLPR consistently predicted CRF and/or aerobic performance (R2 range = .07 to .55). Multivariate regression models routinely outperformed the univariate models, explaining 36-62% of variance. This study signifies a shift toward inclusive explanatory models of CRF and aerobic performance, accounting for combined roles of genetic, physiologic, and behavioral influences. The findings have implications for assessment, selection, and training of specialized military members; and may also impact mission success and survivability.

15. SUBJECT TERMS

graded exercise test, military, explosive ordnance disposal, special operations, serotonin transporter, physical activity, body composition

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