

# Genetics and Other Risk Factors for Past Concussions in Active-Duty Soldiers

Michael N. Dretsch,<sup>1,2</sup> Noah Silverberg,<sup>3–5</sup> Andrew J. Gardner,<sup>6</sup> William J. Panenka,<sup>7</sup>  
Tanja Emmerich,<sup>5</sup> Gogce Crynen,<sup>5</sup> Ghania Ait-Ghezala,<sup>5</sup> Helena Chaytow,<sup>5</sup> Venkat Mathura,<sup>5</sup>  
Fiona C. Crawford,<sup>5</sup> and Grant L. Iverson<sup>8–10</sup>

## Abstract

Risk factors for concussion in active-duty military service members are poorly understood. The present study examined the association between self-reported concussion history and genetics (apolipoprotein E [APOE], brain-derived neurotrophic factor [BDNF], and D2 dopamine receptor genes [DRD2]), trait personality measures (impulsive-sensation seeking and trait aggression-hostility), and current alcohol use. The sample included 458 soldiers who were preparing to deploy for Operation Iraqi Freedom/Operation Enduring Freedom. For those with the BDNF Met/Met genotype, 57.9% (11/19) had a history of one or more prior concussions, compared with 35.6% (154/432) of those with other BDNF genotypes ( $p=0.049$ , odds ratio [OR]=2.48). APOE and DRD2 genotypes were not associated with risk for past concussions. Those with the BDNF Met/Met genotype also reported greater aggression and hostility personality characteristics. When combined in a predictive model, prior military deployments, being male, and having the BDNF Met/Met genotype were independently associated with increased lifetime history of concussions in active-duty soldiers. Replication in larger independent samples is necessary to have more confidence in both the positive and negative genetic associations reported in this study.

**Keywords:** APOE; BDNF; concussion; DRD2; genetics; mild traumatic brain injury; military; personality

## Introduction

LITTLE IS KNOWN ABOUT GENETIC RISK FACTORS for sustaining a concussion in sports, daily life, or military service. To date, four studies have examined the association between the apolipoprotein E (APOE) genetic polymorphisms and risk for concussion. APOE genotypes containing the  $\epsilon 4$  allele have been linked to worse outcome following traumatic brain injury (TBI); therefore, they have been a logical choice to examine as a possible risk factor for sustaining an injury. All four studies found *no association* between being an APOE  $\epsilon 4$  carrier and concussion risk.<sup>1–4</sup> However, this does not appear to be the case for the APOE *promoter*. The APOE promoter has a common variant, termed APOE G-219T, which

modifies the level of APOE expression. The APOE G-219T promoter has been linked to altered production of APOE following injury,<sup>5</sup> worse outcome after TBI,<sup>6</sup> and central nervous system (CNS) vascular vulnerability to spasm and bleeding.<sup>7,8</sup> Theoretically, this promoter variant could impart a vulnerability such that less mechanical force would be required to reach the minimum threshold for concussion. Two studies have evaluated the effect of the APOE G-219T promoter polymorphism on concussion risk, and both revealed a significant association.<sup>9,10</sup> Other genes that have been studied in relation to concussion risk include various tau genotypes<sup>10,11</sup> and brain-derived neurotrophic factor (BDNF).<sup>12</sup> Of these, the only positive finding was an association between the BDNF Val66Met polymorphism and risk for concussion during a

<sup>1</sup>United States Army Aeromedical Research Laboratory, Fort Rucker, Alabama.

<sup>2</sup>Human Dimension Division (HDD), Headquarters Army Training and Doctrine Command (HQ TRADOC), Fort Eustis, Virginia.

<sup>3</sup>Division of Physical Medicine and Rehabilitation and <sup>7</sup>Department of Psychiatry, University of British Columbia, Vancouver, British Columbia, Canada.

<sup>4</sup>GF Strong Rehabilitation Centre, Vancouver, British Columbia, Canada.

<sup>5</sup>Roskamp Institute, Sarasota, Florida.

<sup>6</sup>Hunter New England Local Health District Sports Concussion Program and Centre for Stroke and Brain Injury, School of Medicine and Public Health, University of Newcastle, Newcastle, NSW, Australia.

<sup>8</sup>Department of Physical Medicine and Rehabilitation, Harvard Medical School, Boston, Massachusetts.

<sup>9</sup>Spaulding Rehabilitation Hospital and Home Base, Red Sox Foundation and Massachusetts General Hospital Home Base Program, Boston, Massachusetts.

<sup>10</sup>Defense and Veterans Brain Injury Center, Bethesda, Maryland.

single deployment to the Middle East in a prospective study of United States soldiers.<sup>12</sup> That particular study, performed by our group, assessed 230 soldiers at both pre-deployment and post-deployment, and revealed that there was a higher frequency of Met/Met carriers in soldiers who sustained a concussion while deployed (16.7%) compared with those who were not injured (4.4%). That study provided preliminary evidence of an association between the BDNF Met/Met genotype and risk for concussion.

Several personality and lifestyle factors have been associated with risk for TBI, such as aggression and hostility,<sup>13–16</sup> impulsivity and sensation seeking,<sup>17–20</sup> and substance use.<sup>21,22</sup> Multiple studies have shown a relationship between alcohol use and TBI (of all severities).<sup>23</sup> Between one third and one half of trauma patients who present to emergency departments with TBI are intoxicated with alcohol, and 18–50% have a history of substance misuse.<sup>24–29</sup> However, the relationship may not be unidirectional, because a history of TBI may also predispose an individual to substance use, especially if the TBI occurred at a younger age.<sup>30</sup> Impulsivity and sensation seeking are personality traits that may directly (by increasing risky behavior) or indirectly (by influencing other TBI risk factors such as substance abuse) lead to TBI.<sup>31</sup>

The primary aim of this study was to examine whether polymorphisms of APOE and BDNF are associated with increased lifetime risk for concussion in active-duty United States soldiers. The D2 dopamine receptor gene (DRD2) was included as part of our larger research program relating to traumatic stress;<sup>12</sup> therefore, it was also examined in this study for exploratory purposes. A secondary aim was to explore associations and interactions among genetic polymorphisms, gender, personality traits (impulsive-sensation seeking and aggression-hostility), alcohol use, and deployment history in relation to lifetime concussions.

## Methods

### Participants and procedures

Data were collected from 469 active-duty United States Army soldiers from two brigade combat teams preparing to deploy to Iraq and Afghanistan. Approval was attained from brigade commanders. Soldiers were given the opportunity to voluntarily participate after receiving a study brief by the principal investigator at a designated facility on the military installation. Informed consent was obtained for all participants with the presence of an ombudsperson. Consented participants were then escorted as a group to a separate classroom to complete questionnaires and computerized cognitive testing (as part of a larger study) and/or to phlebotomy stations where several tubes of blood were drawn for genotyping and stored in a dry ice freezer prior to delivering to a –80°C medical specimen freezer at the end of each day. Participants received a check for \$50 for each blood draw. The study was conducted in accordance with the latest version of the Declaration of Helsinki and the protocol and procedures were approved by the Institutional Review Board at Headquarters United States Army Medical Research and Materiel Command, Fort Detrick, Maryland.

### Measures

#### DNA genotyping

**APOE.** For amplification and digestion of the relevant sequence of the APOE gene (position 44908684) from extracted DNA, we used a direct APOE kit (EzWay Direct APOE Genotyping Kit, Koma Biotechnology), following the manufacturer's instructions. Genotype-specific fragments were separated by electrophoresis in a 3% metaphor agarose gel, stained with ethidium bromide.

**BDNF.** Each sample (0.5  $\mu$ L extracted DNA) was amplified at the BDNF Val66Met region (position 27658369) using 0.125  $\mu$ L iTaq<sup>™</sup> polymerase enzymes and 0.5  $\mu$ L BDNF-specific primers (Eurofins). Primers for amplification of the sequence containing the Val66Met polymorphism were as follows: forward 5' AAA CAT CCG AGG ACA AGG TG 3' and reverse 5' ACG TGT ACA AGT CTG CGT CC 3'. Reaction volume was 25  $\mu$ L with 0.75  $\mu$ L 50 mM MgCl<sub>2</sub>, 0.5  $\mu$ L 10 mM deoxynucleotide (dNTP) mix, and 2.5  $\mu$ L iTaq 10X Buffer. Polymerase chain reaction (PCR) conditions were: 5 min at 94°C, followed by thirty 30 sec cycles of 94°C, 60°C, and 72°C. The PCR was terminated at 72°C for 10 min and held at 4°C. The product of this amplification was digested with 1  $\mu$ L Pml I enzyme (Biolabs) at 37°C for 16 h into allele-specific fragments, which were then separated by electrophoresis in a 3% metaphor agarose gel, stained with ethidium bromide.

**DRD2.** Each sample (0.5  $\mu$ L extracted DNA) was amplified at the DRD2 region using 0.125  $\mu$ L iTaq polymerase enzymes and 0.5  $\mu$ L DRD2 (Taq1A; position 113400106) A1/A2-specific primers (Eurofins). Primers for amplification of the DRD2 A1/A2 polymorphism were as follows: forward 5' - CCG TCG ACG GCT GGC CAA GTT GTC TA - 3' and reverse 5' CCG TCG ACC CTT CCT GAG TGT CAT CA 3'. Reaction volume was 25  $\mu$ L with 0.75  $\mu$ L 50 mM MgCl<sub>2</sub>, 0.5  $\mu$ L 10 mM dNTP mix, and 2.5  $\mu$ L iTaq 10X Buffer. PCR conditions were: 5 min at 94°C, followed by 35 cycles of 1 min-1 min-1.5 min at 94°C, 55°C, and 92°C. The PCR was terminated at 72°C for 10 min and held at 4°C. The amplified sequence was 310 bp long (PCR not shown). The PCR products were digested with the 0.5  $\mu$ L Taq I $\alpha$  enzyme (Biolabs) for 16 h at 65°C, which produced DNA fragments corresponding to the A1 and A2 alleles which, upon separation by electrophoresis in a 3% metaphor agarose gel stained with ethidium bromide enabled genotype evaluation.

**Lifetime concussions.** Participants were asked to report the total number of times that they had sustained a concussion from childhood to the present, based on a diagnosis from a medical professional or meeting the criteria of the Brief Traumatic Brain Injury Screen (BTBIS<sup>32</sup>). The BTBIS requires a brain-injury-related event and, at minimum, an altered state of consciousness (e.g., being dazed, confused, or seeing stars; post-traumatic amnesia; or loss of consciousness <20 min). To reduce the potential for under-reporting, participants were reminded that the data were being used for research purposes only, and that their injury history would not be put in their medical record nor would their command be informed.

**Zuckerman-Kuhlman Personality Questionnaire (ZKPQ)-Short version.** The 50 item short version of the ZKPQ<sup>33</sup> is a measure of basic dimensions of personality traits used in psychobiology studies of personality.<sup>34</sup> *Impulsive-sensation seeking* measures lack of planning and tendency to act without thinking; preference for thrills and excitement, novelty and variety; and unpredictability. *Aggression-hostility* measures aggression and hostility, anger, lack of inhibitory control, and low social desirability, and is associated with antisocial behavior, vengefulness, and volatility. Cronbach's  $\alpha$  reliability coefficients range from 0.87 to 0.93 for all scales.<sup>33,34</sup>

**Alcohol Use Identification Test (AUDIT).** This 10 item instrument<sup>35</sup> is used to assess alcohol use in the previous 12 months. It has a test-retest reliability coefficient of 0.86, and has excellent sensitivity (0.95) and specificity (0.80). Scores  $\geq 8$  represent "medium" or greater levels of hazardous alcohol use, and scores  $\geq 16$  suggest the need for monitoring and potential counseling.<sup>36</sup>

### Statistical analysis

Eleven of the 469 participants opted to not complete all the questionnaires necessary to be included in the analysis. Therefore, data from 458 of the 469 participants were included in the analysis. The AUDIT was coded as a total score and as low ( $\leq 7$ ), moderate (8–15), and high ( $\geq 16$ ) risk for alcohol-related problems. Basic descriptive statistics were calculated using mean, median, frequency, and percentage. Odds ratios (OR) of prior concussions by genotype were calculated with  $\chi^2$ , and  $\alpha \leq 0.05$  as statistically significant. Mann–Whitney  $U$  tests were used to examine two-group comparisons on the psychosocial variables. Violations in Hardy–Weinberg equilibrium for the genotypes of APOE, BDNF, and DRD2 polymorphisms were assessed based on their respective population frequencies. Preliminary univariate analyses of BDNF, APOE, and DRD2 polymorphisms and odds of a prior concussion were calculated, and only significant univariate genetic predictors of concussion history were included in the multivariate prediction model. A negative binomial regression analysis was used to explore if genotype, gender, age, total deployments, personality (trait impulsive-sensation seeking and aggression-hostility), and/or alcohol use predicted cumulative lifetime history of concussions.

### Results

There were no deviations from Hardy–Weinberg equilibrium for APOE, BDNF, and DRD2 polymorphisms,  $p_s > 0.05$ . Participants' demographics, injury history, genotypes, alcohol use, and personality test scores are reported in Table 1. The sample was composed of mostly men in their 20s and 30s. More than half (57.9%) were preparing for their first military deployment; however, 22.7% had had two or more prior deployments. Medium to high scores on the AUDIT (score  $\geq 8$ ) were reported by 35.8% of the sample. At least one prior concussion was reported by 36.5% of soldiers, and 10.7% reported a history of three or more past injuries. Approximately 38% (162/430) of men reported having one or more prior concussions compared with approximately 18% (5/28) of women.

### Association between genotypes and concussion history

Concussion history stratified by genotypes is presented in Table 2. Of those soldiers who were  $\epsilon 3/\epsilon 4$  or  $\epsilon 4/\epsilon 4$  carriers, 44.1% (41/93) had a history of one or more prior concussions compared with 34.5% (126/365) of those who did not have these polymorphisms, which was not significantly different ( $\chi^2[1] = 2.93$ ,  $p = 0.087$ , OR = 1.50, 95% CI [0.92, 2.44]). Those with  $\epsilon 3/\epsilon 4$  or  $\epsilon 4/\epsilon 4$  were similar (21.5%; 20/93) to those with the other APOE polymorphisms (23.0%, 84/365) with respect to a history of two or more prior concussions ( $\chi^2[1] = 0.10$ ,  $p = 0.757$ , OR = 0.92, 90% CI [0.51, 1.64]). For soldiers who were BDNF Met/Met carriers, 57.9% (11/19) had a history of one or more prior concussions, compared with 35.6% (154/432) of those that were non-Met/Met carriers ( $\chi^2[1] = 3.88$ ,  $p = 0.049$ , OR = 2.48, 95% CI [0.90, 6.92]; or an increased risk of approximately 148%). The likelihood for two or more prior concussions in Met/Met carriers (36.8%; 7/19) was not significantly greater than in those who were non-Met/Met carriers (22.0%; 95/432) ( $\chi^2[1] = 2.29$ ,  $p = 0.130$ , OR = 2.07, 95% CI [0.71, 5.85]). For DRD2, the likelihood of having one or more prior concussions was very similar across the three DRD2 polymorphisms, which ranged from 30.0% to 36.4%,  $p > 0.05$ . Similarly, the rate of having two or more prior concussions across the three DRD2 polymorphisms was very similar and ranged from 22.0% to 24.1%,  $p > 0.05$ . The results of the univariate analyses suggested that only the BDNF Met/Met genotype was associated with risk for prior concussions.

TABLE 1. PARTICIPANT DEMOGRAPHICS, CONCUSSION HISTORY, GENOTYPE, AND PERSONALITY TRAITS ( $N = 458$ )

Age (mean, SD)	26.0 (7.0)
Gender (f, %)	
Men	430, 93.9%
Education	
$\leq 12$ years (%)	53.3
$> 12$ years (%)	46.7
Race	
White (%)	76.6
Black (%)	10.0
Hispanic/Latino (%)	7.0
Pacific/Islander (%)	2.8
Asian (%)	0.4
Native American (%)	1.1
Other (%)	2.0
Rank	
Junior enlisted (%)	36.9
Non-commissioned officer (%)	59.6
Senior non-commissioned officer (%)	3.4
Past deployments (f, %)	
None	265, 57.9%
1 prior deployment	106, 23.1%
2 prior deployments	62, 13.1%
3 prior deployments	15, 3.3%
$\geq 4$ prior deployments	9, 1.9%
Number of past concussions (f, %)	
0	291, 63.5%
1	63, 13.8%
2	55, 12.0%
3	19, 4.1%
$\geq 4$	30, 6.6%
Genotypes (f, %)	
APOE	
$\epsilon 2/\epsilon 2$	3, 0.7%
$\epsilon 2/\epsilon 3$	50, 10.9%
$\epsilon 2/\epsilon 4$	9, 2.0%
$\epsilon 3/\epsilon 3$	303, 66.2%
$\epsilon 3/\epsilon 4$	90, 19.7%
$\epsilon 4/\epsilon 4$	3, 0.7%
BDNF	
Met/Met	19, 4.1%
Val/Met	133, 29.0%
Val/Val	299, 65.3%
Missing	7
DRD2	
A1/A1	29, 6.3%
A1/A2	159, 34.7%
A2/A2	268, 58.5%
Missing	2
Alcohol use	
Low	249, 64.2%
Medium	134, 29.3%
High	30, 6.6%
Personality measures (mean, SD)	
Impulsive-sensation seeking	6.1 (2.6)
Aggression-hostility	5.1 (2.6)

APOE, apolipoprotein E; BDNF, brain-derived neurotrophic factor; DRD2, D2 dopamine receptor genes.

TABLE 2. CONCUSSION HISTORY STRATIFIED BY APOE, BDNF, AND DRD2 POLYMORPHISMS IN ACTIVE-DUTY SOLDIERS

		Number of prior concussions							
		0		1		2		≥3	
Genotype	n	f	%	f	%	f	%	f	%
APOE									
ε2/ε2	3	3	100	0	0	0	0	0	0
ε2/ε3	50	32	64.0	6	12.0	9	18.0	3	6.0
ε2/ε4	9	7	77.8	0	0	2	22.2	0	0
ε3/ε3	303	197	65.0	36	11.9	34	11.2	36	12.9
ε3/ε4	90	51	56.7	20	22.2	10	11.1	9	10.0
ε4/ε4	3	1	33.3	1	33.3	0	0	1	33.3
BDNF									
Met/Met	19	8	42.1	4	21.1	5	26.3	2	10.5
Val/Met	133	85	63.9	20	15.0	12	9.0	16	12.0
Val/Val	299	193	64.5	39	13.0	38	12.7	29	9.7
BDNF missing	7	5		0		0		2	
DRD2									
A1/A1	29	20	69.0	2	6.9	3	10.3	4	13.8
A1/A2	159	100	62.9	20	12.6	23	14.5	16	10.1
A2/A2	268	168	62.7	41	15.3	30	11.2	29	10.8
DRD2 missing	2	2		0		0		0	

APOE, apolipoprotein E; BDNF, brain-derived neurotrophic factor; DRD2, D2 dopamine receptor genes.

#### Association among BDNF met/met genotype, alcohol use, personality characteristics, and concussion history

Those who were BDNF Met/Met carriers ( $n=19$ ) were compared with non-Met/Met carriers ( $n=432$ ) on current alcohol use and personality variables. There were no significant differences between groups on AUDIT scores or impulsivity and sensation seeking ( $ps>0.30$ , small effect sizes). Met/Met carriers reported greater aggression-hostility (mean=6.37, SD=3.15) than those without this polymorphism (mean=5.04, SD=2.62;  $p=0.033$ ,  $d=0.50$ ). The difference between Met/Met carriers ( $n=19$ ; mean=6.37, SD=3.15) and Met/Val carriers ( $n=133$ ; mean=5.05, SD=2.69) approached significance ( $p=0.052$ ,  $d=0.49$ ), and the difference between Met/Met carriers and Val/Val carriers was significant ( $n=299$ ; mean=5.04, SD=2.59,  $p=0.033$ ,  $d=0.51$ ), suggesting that greater aggression-hostility was observed in individuals who were homozygous carriers of the Met allele.

There were no significant differences between men and women on AUDIT scores or impulsivity and sensation seeking ( $ps>0.36$ , small effect sizes). Men reported greater aggression-hostility (mean=5.19, SD=2.65) than women (mean=4.00, SD=2.45;  $p=0.021$ ,  $d=0.45$ ).

Those with a history of one or more prior concussions ( $n=167$ ) had greater AUDIT scores (mean=7.12, SD=5.52) than those with no prior concussions ( $n=291$ ; mean=5.99, SD=5.24;  $p=0.021$ ,  $d=0.21$ ); these two groups did not differ on the two personality measures ( $ps>0.24$ , small effect sizes). Those with a history of two or more prior concussions ( $n=104$ ) had greater impulsivity and sensation seeking scores (mean=6.60, SD=2.57) than those with no prior concussions ( $n=291$ ; mean=5.90, SD=2.56;  $p=0.019$ ,  $d=0.27$ ); these two groups did not differ in their current alcohol use or aggression-hostility ( $p>0.05$ , small effect sizes).

#### Predictors of prior concussions

To examine the association among genetics, alcohol use, personality characteristics and lifetime history of concussion, a negative

binomial regression analysis was conducted with concussions as the dependent variable (observed number of events), BDNF genotype (Met/Met = 1, non-Met/Met = 0), alcohol use (low = 1, medium = 2, high = 3), and sex (women = 0, men = 1) as predictors, and personality (trait impulsive-sensation seeking, aggression-hostility), age, and total deployments as covariates. The model was a good fit (Bayesian information criterion = 1171.83, deviance = 1.11, likelihood ratio  $\chi^2[8] = 33.79$ ,  $p<0.001$ ). BDNF Met/Met (Wald  $\chi^2 = 3.92$ ,  $p=0.048$ ), total deployments (Wald  $\chi^2 = 13.24$ ,  $p<0.001$ ), and gender (Wald  $\chi^2 = 6.21$ ,  $p=0.013$ ) were each significant predictors of prior concussions; aggression-hostility (Wald  $\chi^2 = 0.08$ ,  $p=0.778$ ), impulsive-sensation seeking (Wald  $\chi^2 = 3.34$ ,  $p=0.068$ ), age (Wald  $\chi^2 = 0.92$ ,  $p=0.338$ ), and alcohol use (Wald  $\chi^2 = 2.96$ ,  $p=0.227$ ) were not significant independent predictors of concussion history in this multivariable model. Neither APOE nor DRD2 genotypes were included in the model because the results of the preliminary univariate analysis of concussion frequencies by polymorphism did not indicate significantly increased risk.

The results suggest that deployment was associated with increased risk for a single prior concussion by 1.35 times (B = 0.30, incident rate ratio [IRR] = 1.35, 95% CI [1.15, 1.59]) or ~35%. Being a BDNF Met/Met-carrier was associated with increased risk of a prior concussion by 1.87 times or 87% (B = 0.63, IRR = 1.87, 95% CI [1.01, 3.49]). Being a man was associated with increased risk by 2.81 times (B = 1.03, IRR = 2.81, 95% CI [1.25, 6.34]) or ~181%. None of the interactions were significant predictors of concussion history ( $p>0.05$ ).

#### Discussion

This study examined lifetime history of concussions and risk factors for injury in active duty military service members. A substantial minority of the sample (36.5%) reported a history of one or more prior concussions. The BDNF Met/Met genotype was uncommon in this sample, occurring in only 4.1% of soldiers. Those with this genotype reported more prior concussions than those without this

genotype. They also reported greater trait aggression and hostility. Service members with prior concussion(s) reported greater current alcohol use and greater scores on impulsivity and sensation seeking than those with no prior concussions. There were no significant associations between the APOE or DRD2 genotypes and concussion history. Multivariate modeling revealed that being a BDNF Met/Met-carrier, male, and having been operationally deployed were independently associated with lifetime history of concussions.

One prior published study, by Dretsch and coworkers,<sup>12</sup> used a subgroup of the current sample that was followed prospectively before and after a military deployment to the Middle East, and identified the BDNF Met/Met genotype as a significant risk factor for concussion. To our knowledge, that was the first published study to identify an association between BDNF polymorphisms and risk for concussion. In light of that finding, the pre-deployment data presented in the current study was culled from all participants ( $n=458$ ), which allowed us to retrospectively assess and report here that BDNF genotype was associated with increased risk for *prior* lifetime concussions. As such, both retrospectively and prospectively, the BDNF Met/Met genotype has been shown to be associated with increased risk for concussion.

It is possible that the BDNF Met/Met genotype modulates morphological differences in neuronal and synaptic integrity that may contribute to individual resilience/vulnerability to mechanical strain following injury to the head (for review see, Cohen-Cory and coworkers<sup>37</sup>). The BDNF Met/Met genotype has been linked with abnormal regulation of BDNF in the brain.<sup>38</sup> BDNF is an important neuroprotective and neuroplastic growth factor, which has been shown to modulate neuronal integrity in response to multiple neurological insults including ischemia,<sup>39</sup> excitotoxicity,<sup>39</sup> and trauma.<sup>40</sup> There is evidence in rodents that indirectly increasing BDNF production via tropomyosin-related kinase B signaling improves TBI outcomes.<sup>41</sup> The BDNF polymorphism has been associated with potential TBI-predisposing character traits including hyperactivity-impulsivity,<sup>42,43</sup> proclivity to addictions,<sup>44,45</sup> and high risk sport participation and sensation seeking.<sup>46</sup> Some of these personality traits have been linked to risk for concussion,<sup>13–16,23</sup> and alcohol use also has been associated with risk for TBI.<sup>24–30</sup> In the present study, the BDNF Met/Met genotype was associated with greater aggression and hostility, but not with current alcohol use or impulsivity and sensation seeking. Animal studies provide evidence showing that both BDNF expression and the Met allele, independent of TBI, are linked with increased aggression.<sup>47,48</sup> Overall, one might speculate that the BDNF Met/Met genotype could behaviorally predispose a person to injury, contribute to an aberrant neurochemical cascade leading to a lower threshold for mild TBI, or both.

This study has several limitations. First, individuals homozygous for the BDNF Met allele or the APOE  $\epsilon 4$  allele are difficult to study because these genotypes are uncommon in the general population. Second, although having been deployed was a risk factor for prior concussions, data on whether our participants had sustained their injuries during a prior deployment or at some other point in their lives were not available. Third, there was no way to independently verify the accuracy in reporting lifetime history of concussions, and we had no information relating to the severity of those injuries, when they occurred, or whether they were associated with prolonged recovery times.

## Conclusion

In conclusion, the BDNF Met/Met genotype was associated with greater lifetime history of concussion and with greater aggression

and hostility. In a multivariate model, military deployments, being male, and having the BDNF Met/Met genotype were independently associated with increased lifetime history of concussions in active-duty soldiers. In contrast, the APOE and DRD2 genetic polymorphisms assessed here were not associated with lifetime history of concussions. Replication in larger independent samples is necessary to have more confidence in both the positive and negative genetic associations presented in this study.

## Acknowledgments

The authors acknowledge the assistance of numerous individuals from the United States Army Aeromedical Research Laboratory and Roskamp Institute for their critical role and diligent effort during the data collection phase. M.N.D. had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. Data analysis was conducted by M.N.D. The authors thank additional individuals from Roskamp Institute including Justin Martin for maintaining the database; Dr. Robert Pelot, Dr. Scott Ferguson, Alex Bishop, and John Phillips for assisting in the sample collection; and Ariel Gonzalez and Dr. Laila Abdullah for assisting with the genotyping. The authors also thank Jeremy Athy, Jinyong Bae, Craig Berlin, Arlene Breau, Jim Chiamonte, Dr. Tim Cho, Elise Corrado, Pedro Cruz, Bradley Erickson, Dr. Arthur Estrada, Allyssa Hathaway, Kathleen Kelley, Melody King, Daniel Lopez, David Lopez, Lana Milam, Jill Parker, Stanslaus Simiyu, Josue Sosa, Elizabeth Stokes, Dr. Kenneth Thiel, Dr. Stephanie Traynham, Melinda Vasbinger, and anyone else from the United States Army Aeromedical Research Laboratory, unintentionally not named, who was instrumental in the logistical process of data collection.

The opinions, interpretations, conclusions, and recommendations are those of the authors and are not necessarily endorsed by the United States Army and/or the United States Department of Defense.

## Author Disclosure Statement

Grant L. Iverson acknowledges support from the INTRuST Posttraumatic Stress Disorder and Traumatic Brain Injury Clinical Consortium funded by the Department of Defense Psychological Health/Traumatic Brain Injury Research Program (X81XWH-07-CC-CSDoD). Andrew J. Gardner acknowledges funding from the New South Wales Sporting Injuries Committee–Sports Research & Injury Prevention Scheme, the Brain Foundation, Australia–Brain Injury Award, Hunter Medical Research Institute (HMRI) early career funding through Anne Greaves, and research fellowship funding from Jennie Thomas, Life Governor of the HMRI. Grant L. Iverson has been reimbursed by the government, professional scientific bodies, and commercial organizations for discussing or presenting research relating to mild TBI and sport-related concussion at meetings, scientific conferences, and symposiums. He has a clinical practice in forensic neuropsychology involving individuals who have sustained mild TBIs (including athletes). He has received honoraria for serving on research panels that provide scientific peer review of programs. He is a co-investigator, collaborator, or consultant on grants relating to mild TBI funded by several organizations. Andrew J. Gardner has a clinical practice in neuropsychology involving the assessment and management of individuals who have sustained sport-related concussion (including current and former athletes). He received travel funding from the Australian Football League (AFL) to present at the Concussion in Football Conference in 2013. He also serves in an unremunerated capacity as a member of the Concussion Advisory Group for the Australian Rugby Union (ARU). The other authors have nothing to disclose.

## References

- Zhou, W., Xu, D., Peng, X., Zhang, Q., Jia, J., and Crutcher, K.A. (2008). Meta-analysis of APOE4 allele and outcome after traumatic brain injury. *J. Neurotrauma* 25, 279–290.
- Kassam, I., Gagnon, F., and Cusimano, M.D. (2016). Association of the APOE-epsilon4 allele with outcome of traumatic brain injury in children and youth: a meta-analysis and meta-regression. *J. Neurol. Neurosurg. Psychiatry* 87, 433–440.
- Lawrence, D.W., Comper, P., Hutchison, M.G., and Sharma, B. (2015). The role of apolipoprotein E epsilon (epsilon)-4 allele on outcome following traumatic brain injury: a systematic review. *Brain Inj.* 29, 1018–1031.
- Zeng, S., Jiang, J.X., Xu, M.H., Xu, L.S., Shen, G.J., Zhang, A.Q., and Wang, X.H. (2014). Prognostic value of apolipoprotein E epsilon4 allele in patients with traumatic brain injury: a meta-analysis and meta-regression. *Genet. Test Mol. Biomarkers* 18, 202–210.
- Lambert, J.C., Berr, C., Pasquier, F., Delacourte, A., Frigard, B., Cotel, D., Perez-Tur, J., Mouroux, V., Mohr, M., Cecyre, D., Galasko, D., Lendon, C., Poirier, J., Hardy, J., Mann, D., Amouyel, P., and Chartier-Harlin, M.C. (1998). Pronounced impact of Th1/E47cs mutation compared with -491 AT mutation on neural APOE gene expression and risk of developing Alzheimer's disease. *Hum. Mol. Genet.* 7, 1511–1516.
- Lendon, C.L., Harris, J.M., Pritchard, A.L., Nicoll, J.A., Teasdale, G.M., and Murray, G. (2003). Genetic variation of the APOE promoter and outcome after head injury. *Neurology* 61, 683–685.
- Wu, H.T., Ruan, J., Zhang, X.D., Xia, H.J., Jiang, Y., and Sun, X.C. (2010). Association of promoter polymorphism of apolipoprotein E gene with cerebral vasospasm after spontaneous SAH. *Brain Res.* 1362, 112–116.
- Yin, C., Huang, G.F., Ruan, J., He, Z.Z., and Sun, X.C. (2015). The APOE promoter polymorphism is associated with rebleeding after spontaneous SAH in a Chinese population. *Gene* 563, 52–55.
- Tierney, R.T., Mansell, J.L., Higgins, M., McDevitt, J.K., Toone, N., Gaughan, J.P., Mishra, A., and Krynetskiy, E. (2010). Apolipoprotein E genotype and concussion in college athletes. *Clin. J. Sport Med.* 20, 464–468.
- Terrell, T.R., Bostick, R.M., Abramson, R., Xie, D., Barfield, W., Cantu, R., Stanek, M., and Ewing, T. (2008). APOE, APOE promoter, and Tau genotypes and risk for concussion in college athletes. *Clin. J. Sport Med.* 18, 10–17.
- Terrell, T.R., Bostick, R.M., and Barth, J.T. (2012). Prospective cohort study of the association of genetic polymorphisms and concussion risk and postconcussion neurocognitive deficits in college athletes (abstract). *Clin. J. Sport Med.* 22, 172.
- Dretsch, M.N., Williams, K., Emmerich, T., Crynen, G., Ait-Ghezala, G., Chaytow, H., Mathura, V., Crawford, F.C., and Iverson, G.L. (2016). Brain-derived neurotrophic factor polymorphisms, traumatic stress, mild traumatic brain injury, and combat exposure contribute to postdeployment traumatic stress. *Brain Behav.* 6, e00392.
- Neumann, D., Malec, J.F., and Hammond, F.M. (2015). The association of negative attributions with irritation and anger after brain injury. *Rehabil. Psychol.* 60, 155–161.
- Dyer, K.F., Bell, R., McCann, J., and Rauch, R. (2006). Aggression after traumatic brain injury: analysing socially desirable responses and the nature of aggressive traits. *Brain Inj.* 20, 1163–1173.
- Tateno, A., Jorge, R.E., and Robinson, R.G. (2003). Clinical correlates of aggressive behavior after traumatic brain injury. *J. Neuropsychiatry Clin. Neurosci.* 15, 155–160.
- Baguley, I.J., Cooper, J., and Felmingham, K. (2006). Aggressive behavior following traumatic brain injury: how common is common? *J. Head Trauma Rehabil.* 21, 45–56.
- Goswami, R., Dufort, P., Tartaglia, M.C., Green, R.E., Crawley, A., Tator, C.H., Wennberg, R., Mikulis, D.J., Keightley, M., and Davis, K.D. (2015). Frontotemporal correlates of impulsivity and machine learning in retired professional athletes with a history of multiple concussions. *Brain Struct. Funct.* 221, 1911–1925.
- Cusimano, M.D., Holmes, S.A., Sawicki, C., and Topolovec-Vranic, J. (2014). Assessing aggression following traumatic brain injury: a systematic review of validated aggression scales. *J. Head Trauma Rehabil.* 29, 172–184.
- Kocka, A., and Gagnon, J. (2014). Definition of impulsivity and related terms following traumatic brain injury: a review of the different concepts and measures used to assess impulsivity, disinhibition and other related concepts. *Behav. Sci.* 4, 352–370.
- Hehar, H., Yeates, K., Kolb, B., Esser, M.J., and Mychasiuk, R. (2015). Impulsivity and concussion in juvenile rats: examining molecular and structural aspects of the frontostriatal pathway. *PLoS One* 10, e0139842.
- van Reekum, R., Cohen, T., and Wong, J. (2000). Can traumatic brain injury cause psychiatric disorders? *J. Neuropsychiatry Clin. Neurosci.* 12, 316–327.
- Miller, S.C., Baktash, S.H., Webb, T.S., Whitehead, C.R., Maynard, C., Wells, T.S., Otte, C.N., and Gore, R.K. (2013). Risk for addiction-related disorders following mild traumatic brain injury in a large cohort of active-duty U.S. airmen. *Am. J. Psychiatry* 170, 383–390.
- Riggio, S., and Wong, M. (2009). Neurobehavioral sequelae of traumatic brain injury. *Mt. Sinai J. Med.* 76, 163–172.
- Isokuortti, H., Iverson, G.L., Kataja, A., Brander, A., Ohman, J., and Luoto, T.M. (2016). Who gets head trauma or recruited in mild traumatic brain injury research? *J. Neurotrauma* 33, 232–241.
- Koponen, S., Taiminen, T., Portin, R., Himanen, L., Isoniemi, H., Heinonen, H., Hinkka, S., and Tenovu, O. (2002). Axis I and II psychiatric disorders after traumatic brain injury: a 30-year follow-up study. *Am. J. Psychiatry* 159, 1315–1321.
- Ganti, L., Khalid, H., Patel, P.S., Daneshvar, Y., Bodhit, A.N., and Peters, K.R. (2014). Who gets post-concussion syndrome? An emergency department-based prospective analysis. *Int. J. Emerg. Med.* 7, 31.
- Dikmen, S., Machamer, J.E., Donovan, D.M., Winn, H.R., and Temkin, N.R. (1995). Alcohol use before and after traumatic head injury. *Ann. Emerg. Med.* 26, 167–176.
- Corrigan, J.D. (1995). Substance abuse as a mediating factor in outcome from traumatic brain injury. *Arch. Phys. Med. Rehabil.* 76, 302–309.
- Lange, R.T., Iverson, G.L., and Franzen, M.D. (2007). Short-term neuropsychological outcome following uncomplicated mild TBI: effects of day-of-injury intoxication and pre-injury alcohol abuse. *Neuropsychology* 21, 590–598.
- Bogner, J., French, L.M., Lange, R.T., and Corrigan, J.D. (2015). Pilot study of traumatic brain injury and alcohol misuse among service members. *Brain Inj.* 29, 905–914.
- Kazemi, D.M., Flowers, C., Shou, Q., Levine, M.J., and Van Horn, K.R. (2014). Personality risk for alcohol consequences among college freshmen. *J. Psychosoc. Nurs. Ment. Health Serv.* 52, 38–45.
- Schwab, K.A., Baker, G., Ivins, B., Sluss-Tiller, M., Lux, W., and Warden, D. (2006). The Brief Traumatic Brain Injury Screen (BTBIS): Investigating the validity of a self-report instrument for detecting traumatic brain injury (TBI) in troops returning from deployment in Afghanistan and Iraq. *Neurology* 66, A235.
- Zuckerman, M., Kuhlman, D.M., Joireman, J., Teta, P., and Kraft, M. (1993). A comparison of three structural models for personality: The Big Three, the Big Five, and the Alternative Five. *J. Pers. Soc. Psychol.* 65, 757–768.
- Zuckerman, M. (2002). Zuckerman-Kuhlman Personality Questionnaire (ZKPQ): an alternative five-factor model, in: *Big Five Assessment*. B. De Raad, and M. Perugini (eds.). Hogrefe & Huber Publishers: Boston, pps. 377–396.
- Saunders, J.B., Aasland, O.G., Babor, T.F., de la Fuente, J.R., and Grant, M. (1993). Development of the Alcohol Use Disorders Identification Test (AUDIT): WHO collaborative project on early detection of persons with harmful alcohol consumption—II. *Addiction* 88, 791–804.
- Babor, T., Higgins-Biddle, J., Saunders, J., and Monteiro, M. (2001). *The Alcohol Use Disorders Identification Test, Guidelines For Use in Primary Care*. 2nd ed. World Health Organization: Geneva, Switzerland.
- Cohen-Cory, S., Kidane, A.H., Shirkey, N.J., and Marshak, S. (2010). Brain-derived neurotrophic factor and the development of structural neuronal connectivity. *Dev. Neurobiol.* 70, 271–288.
- Mallei, A., Baj, G., Ieraci, A., Corna, S., Musazzi, L., Lee, F.S., Tongiorgi, E., and Popoli, M. (2015). Expression and dendritic trafficking of BDNF-6 splice variant are impaired in knock-in mice carrying human BDNF Val66Met polymorphism. *Int. J. Neuropsychopharmacol.* 18, pii: pyv069.
- Chen, A., Xiong, L.J., Tong, Y., and Mao, M. (2013). The neuroprotective roles of BDNF in hypoxic ischemic brain injury. *Biomed. Rep.* 1, 167–176.
- Harvey, A.R., Lovett, S.J., Majda, B.T., Yoon, J.H., Wheeler, L.P., and Hodgetts, S.I. (2015). Neurotrophic factors for spinal cord repair: Which, where, how and when to apply, and for what period of time? *Brain Res.* 1619, 36–71.

41. Wu, C.H., Hung, T.H., Chen, C.C., Ke, C.H., Lee, C.Y., Wang, P.Y., and Chen, S.F. (2014). Post-injury treatment with 7,8-dihydroxyflavone, a TrkB receptor agonist, protects against experimental traumatic brain injury via PI3K/Akt signaling. *PLoS One* 9, e113397.
42. Su, H., Tao, J., Zhang, J., Xie, Y., Sun, Y., Li, L., Xu, K., Han, B., Lu, Y., Sun, H., Wei, Y., Wang, Y., Zhang, Y., Zou, S., Wu, W., Zhang, J., Zhang, X., and He, J. (2014). An association between BDNF Val66-Met polymorphism and impulsivity in methamphetamine abusers. *Neurosci. Lett.* 582, 16–20.
43. Bergman, O., Westberg, L., Lichtenstein, P., Eriksson, E., and Larsson, H. (2011). Study on the possible association of brain-derived neurotrophic factor polymorphism with the developmental course of symptoms of attention deficit and hyperactivity. *Int. J. Neuropsychopharmacol.* 14, 1367–1376.
44. Logrip, M.L., Barak, S., Wagnault, V., and Ron, D. (2015). Corticostriatal BDNF and alcohol addiction. *Brain Res.* 1628, 60–67.
45. Greenwald, M.K., Steinmiller, C.L., Sliwerska, E., Lundahl, L., and Burmeister, M. (2013). BDNF Val(66)Met genotype is associated with drug-seeking phenotypes in heroin-dependent individuals: a pilot study. *Addict. Biol.* 18, 836–845.
46. Thomson, C.J., Power, R.J., Carlson, S.R., Rupert, J.L., and Michel, G. (2015). A comparison of genetic variants between proficient low- and high-risk sport participants. *J. Sports Sci.* 33, 1861–1870.
47. Lyons, W.E., Mamounas, L.A., Ricaurte, G.A., Coppola, V., Reid, S.W., Bora, S.H., Wihler, C., Koliatsos, V.E., and Tessarollo, L. (1999). Brain-derived neurotrophic factor-deficient mice develop aggressiveness and hyperphagia in conjunction with brain serotonergic abnormalities. *Proc. Natl. Acad. Sci. U. S. A.* 96, 15,239–15,244.
48. Ito, W., Chehab, M., Thakur, S., Li, J., and Morozov, A. (2011). BDNF-restricted knockout mice as an animal model for aggression. *Genes Brain Behav.* 10, 365–374.

Address correspondence to:

*Michael N. Dretsch, PhD*

*Human Dimension Division*

*Army Capabilities Integration Center*

*Headquarters, Training, and Doctrine Command*

*950 Jefferson Avenue*

*Fort Eustis, VA 23604*

*E-mail: michael.n.dretsch.mil@mail.mil*