

APOE, APOE Promoter, and Tau Genotypes and Risk for Concussion in College Athletes

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Objective: To investigate associations of APOE, APOE promoter (G-219T), and tau protein exon 6 polymorphisms (^{His}47^{Tyr} and ^{Ser}53^{Pro}) and a history of self-reported concussion in college athletes.

Design: Multi-center cross-sectional study.

Setting: Male football and male and female soccer programs at the University of South Carolina, Jacksonville University, Benedict College, and the College of Charleston.

Participants: Active 18- to 30-year-old (n = 195) intercollegiate male football players and male and female soccer players during 2001 and 2002.

Assessment of Risk Factors: Written questionnaires and blood or mouthwash samples for DNA for genotyping by RFLP/PCR.

Main Outcome Measurement: Self-reported history of concussions over the previous 8 years.

Results: A statistically significant, nearly 3-fold increase in risk of a history of concussion for those with the APOE promoter G-219T TT genotype relative to the GG genotype (OR, 2.8; 95% CI, 1.1 to 6.9) adjusted for age, sport, school, and years in their primary sport, a finding that was stronger for Cantu grade 2 and 3 concussions.

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Conclusions: These results suggest that college athletes with an APOE promoter G-219T TT genotype may be at increased risk for having a history of concussions, especially more severe concussions. Although there was some support for the possibility that the tau ^{Ser}53^{Pro} polymorphism may be associated with increased risk of prior concussion (OR, 2.1; 95% CI, 0.3 to 14.5), there was no support for an association with APOE genotypes. The results of this cross-sectional study support the need for a prospective study of genetic factors, such as APOE promoter polymorphisms, and the incidence of and sequelae from concussions in college athletes.

Key Words: concussion, soccer, football, genetic polymorphism APOE, APOE G-219T promoter polymorphism

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INTRODUCTION

An estimated 300,000 mild to moderately severe sports-related traumatic brain injuries occur in the United States annually.¹ Concussion, also termed mild traumatic brain injury (MTBI), can cause significant short-term² and long-term sequelae and may lead to persistent neurocognitive deficits, which some claim as evidence for chronic traumatic brain injury (CTBI).³ Outcomes after TBI have been hypothesized to be genetically influenced.^{4,5}

Several lines of evidence suggest that the APOE and tau genes may play a role in brain injury. The APOE gene, located on chromosome 19 at position q 13.2, has 3 major isoforms coded by 3 alleles (APOE ε2, ε3, and ε4)⁶ and encodes for apolipoprotein (Apo E) production. Apo E and amyloid precursor protein (APP) production is upregulated in experimental studies in response to acute head trauma.^{7,8} Apo E is responsible for lipid transport in the brain, maintaining neural structural integrity,⁹ and recovery after neurological injury.¹⁰

Apolipoprotein (Apo E) is expressed in response to neural injury and repair.^{11,12} Although APOE ε3 promotes neurite outgrowth, APOE ε4 inhibits it, suggesting that APOE ε3 may aid recovery.^{13,14} APOE ε4, a known risk factor for Alzheimer's disease (AD),^{15,16} was also found to be a risk factor for chronic traumatic encephalopathy in boxers.¹⁷ Several studies found APOE ε4 to be associated with poorer outcome after TBI.^{5,18}

APOE ε4 is associated with a poorer outcome after major TBI,⁵ an unfavorable cognitive^{18,19} recovery and functional impairment 6 months after brain trauma.^{5,18,20} APOE may also modify the type or severity of brain damage.⁵

However, a recent large study failed to show a significant association between the APOE $\epsilon 4$ allele and poor outcome in mild to moderate TBI cases.²¹

The G-219T polymorphism in the APOE promoter region 186 bp upstream of the TATA box has been associated with a quantitative impact on APOE levels after TBI,²² increased risk of AD,^{16,23} unfavorable outcome after moderate TBI,²⁴ and increased risk for myocardial infarction.²⁵

The tau gene on chromosome 17, responsible for tau protein production, has 17 polymorphisms^{26,27}; mutations in this gene cause chromosome 17-frontotemporal dementia (FTDP-17)²⁷ and other neurodegenerative diseases. The tau gene has 2 exon 6 polymorphisms, His47Tyr and Ser53Pro, for which no independent associations with AD have been found.²⁸

To our knowledge, there are no previous reports of investigations of associations of APOE or the APOE promoter genotypes with a history of mild TBI in athletes. The association between the tau gene and a history of TBI, to our knowledge, has not been studied in any population. Previous TBI-genetic association studies have been limited by small sample sizes, lack of representation of women and racial/ethnic minorities, few MTBI cases, failure to account for potential confounding factors, and older populations who sustained non-sports-related severe head injuries. To address these gaps in knowledge in the study reported herein, we investigated associations of APOE, APOE promoter, and tau genotypes with a history of concussion in college athletes in a cross-sectional study with a large sample size and adjustment for known potential confounders.

METHODS

Participants were recruited from active intercollegiate male football players and male and female soccer players attending the University of South Carolina (Columbia, SC), Jacksonville University (Jacksonville, FL), Benedict College (Columbia, SC), and the College of Charleston (Charleston, SC) during 2001–2002. Exclusion criteria included age less than 18 or over 30, participation in organized boxing, subdural or epidural hematoma, meningitis, brain tumor or abscess, TIA, stroke, temporary paralysis, active seizure disorder, skull fracture or penetrating skull trauma, major neck trauma/fracture, Factor 5 Leiden deficiency, uncontrolled hypertension (average systolic > 170; diastolic > 110), and history of abnormal MRI/CT with intracranial pathology not consistent with sport or non-sport-related TBI. Of 306 potential participants, 301 (98%) agreed to participate and completed concussion/medical history questionnaires. Three participants were excluded, due to boxing, meningitis, and age over 30 years. Concerns about phlebotomy resulted in only 195 (65%) participants providing genetic samples. A chi-square analysis comparing those who did and did not undergo genetic testing showed no significant differences. The study population for analysis included 159 male football and 18 male and 18 female soccer players.

An investigator-assisted medical/concussion history questionnaire was completed by all participants during annual preparticipation exams. The instrument included questions about participants' number of years playing their major sport,

medical/psychiatric history, learning disabilities, attention deficit disorder, alcohol/drug abuse history, and a detailed concussion history. The definition of concussion was explained to each participant, and all occurrences in the last 8 years, whether sport-related or not, were recorded.

The operational definition of concussion for this study was consistent with that provided by the AOSSM Concussion Workgroup²⁹ and those used in the recent literature.³⁰ We used a sensitive case definition: "A concussion occurs immediately after some trauma to the head or due to a force transmitted to the head that may or may not have caused a rapid rotation of the head. The event causes a transient alteration in neurological function that may last 15 seconds or more. You may experience loss of consciousness, memory loss, confusion/disorientation, headache, dizziness, 'being in a fog', nausea, vomiting, increased fatigue, visual disturbances such as flashes of light, or hear 'bells ringing.'" The questionnaire inquired separately about migraine headache history to differentiate this from the typical headache associated with concussion. Alcohol and drug abuse history was also queried.

Completed questionnaires were reviewed 90% of the time during the data collection session by a knowledgeable medical practitioner or research assistant (80% by the PI). Concussions were graded by the primary author. An attempt was made to collect any missing data. Five participants provided no or an incomplete concussion history and were excluded from further analysis.

Clinical scenarios that were consistent with our operational definition of concussion but failed to meet the stricter concussion criteria of the traditional Cantu³¹ and American Academy of Neurology (AAN)³² grading scales were arbitrarily assigned Grade 1 Cantu or AAN concussion grades. The genotype analyses that follow are based on defining concussions by using the AOSSM definition. Concussion severity was classified using the traditional Cantu,³¹ evidence-based Cantu,³² and AAN³³ grading scales.

Biological samples were also collected during the annual preparticipation exams. The 195 samples collected for genotyping included 181 whole blood and 14 buccal cell mouthwash³⁴ specimens. Genotyping could not be done from 3 blood samples and 4 buccal cell specimens; the sample yield was 98% for blood and 78% for buccal cell collections. The author (DX), who completed the genetic analyses, was blinded to the concussion histories.

Laboratory Methods

Genomic DNA was extracted from frozen white blood cells and buccal cells. APOE genotyping was carried out by PCR amplification (30 s at 94°C, 30 s at 68°C, and 30 s at 72°C each cycle for 35 cycles); 212 bp of the *Apo E* polymorphic region was amplified with an upstream primer, 5'-TCCAAGGAGCTGCAGGCGGCGCA-3', and a downstream primer, 5'-GCCTGGTACTGCCCAC-3'. The PCR products were subsequently digested with *Afl III* and *Hha I*, subjected to electrophoresis on a 4% agarose gel (BMA, Rockland, ME), and then visualized under ultraviolet light after staining with ethidium bromide. The *ApoE2*, *E3*, and *E4* alleles were determined as follows: 2 fragments of 54 bp and 158 bp for *ApoE2*, 3 fragments of 17 bp, 54 bp, and 141 bp for

ApoE3, and 2 fragments of 17 bp and 195 bp for *ApoE4*. The APOE promoter G-219T polymorphism -186 bp of the APOE gene TATA box was detected by PCR-RFLP methods as described by Lambert et al.³⁵ The 2 tau exon 6 polymorphisms, His⁴⁷Tyr and Ser⁵³Pro, were detected by PCR-RFLP methods as described by Poorkaj et al.³⁶

Statistical Analyses

All statistical inquiries were conducted using SAS Software version 8.2e from the SAS Institute (Cary, NC). Descriptive comparisons of cases and controls were conducted using chi-square tests for categorical variables and analysis of covariance (ANACOVA) for continuous variables. Logistic regression was used to calculate odds ratios (ORs) and corresponding 95% confidence intervals (CI). Several risk factors were examined as possible confounders or effect modifiers of the genotype-concussion association. These included age, sex, race, school, sport, total years in sport, sport position, SAT score, and migraine headaches. Criteria for inclusion of any covariate in the final model included: (1) biological plausibility, (2) whether it fit the model at $P \leq 0.1$, and (3) whether it altered the OR for the primary exposure variable, genotype, by 10% or more. The final regression model adjusted only for age, school, sport, and number of years in primary sport to estimate the strengths of associations between APOE, APOE promoter, and tau genotypes and risk for concussion. A test for trend was calculated across genotypes to detect a pattern of association. To examine separate effects of genotype and certain risk factors, stratified analyses were conducted in which continuous variables were dichotomized on the median values among controls.

Ethical Considerations

The study was approved by the Institutional Research Boards of the University of South Carolina and all participating schools. Signed informed consent was obtained from all subjects before participation.

RESULTS

Descriptive Analysis

Selected baseline characteristics of participants with and without a history of one or more concussions are presented in Table 1. From the 196 study questionnaires, a total of 97 concussions were reported; 72 participants reported a history of 1 or more concussions, and 124 reported no concussions. A history of concussion was reported slightly more frequently in men than in women ($P = 0.06$). On average, those with a history of concussion reported participating in their sport for more years ($P = 0.05$). There were no substantial or statistically significant differences in race or distributions of genotypes between the 2 groups. Genotype distributions did not deviate from the Hardy-Weinberg equilibrium (calculations not shown). Genotype frequencies for polymorphic APOE, APOE promoter, and tau for both cases and controls were consistent with other study populations.^{26,37}

The concussion histories of cases are summarized in Tables 2 and 3. A total of 97 concussions were reported, with 7 (7.2%) occurring for non-sport-related reasons. Multiple

TABLE 1. Selected Characteristics of Participants

	Hx of Concussion (N = 72)	No Hx Concussion (N = 123)	P*
Demographic factors			
Age (yr)†	19.8 (1.4)	19.9 (1.7)	0.53
Sex (%)			
Male	95.8	87.8	
Female	4.2	12.2	0.06
Race (%)			
White	54.2	48.8	
Black	40.3	41.5	
Other	5.6	9.8	0.53
Risk factors			
Sport (%)			
Football	87.5	78.1	
Soccer	12.5	21.9	0.10
Sport years	9.6 (3.8)	8.3 (4.5)	0.05
School (%)			
Benedict	2.8	12.2	
College of Charleston	5.6	8.1	
Jacksonville University	45.8	38.0	
USC	45.8	40.6	0.12
Genetic factors			
APOEE genotypes (%)			
22	1.4	1.7	
23	11.6	9.2	
24	2.9	0.8	
33	68.1	70.8	
34	15.9	14.2	
44	0.0	3.3	0.57
APOEE promoter (%)			
GG	25.0	29.2	
GT	39.7	48.3	
TT	35.3	22.5	0.16
Tau-Hist (%)			
CC	4.4	4.2	
TC	41.2	42	
TT	54.4	54.2	0.99
Tau-Ser (%)			
CC	2.9	3.5	
CT	19.1	18.6	
TT	77.9	77.8	0.97

*Based on chi-square test for categorical variables and analysis of covariance (ANACOVA) for continuous variables.

†Mean (standard deviation) presented unless otherwise indicated.

concussions were reported by 22 participants (30.6% of cases). Loss of consciousness occurred in 23.2% of the total concussions. Traditional Cantu grading Grade 1 (71.1%) and Grade 2 (25.8%) concussions comprised the majority of cases, while AAN Grade 1 concussions (39.2%) and Grade 2 concussions (41.2%) were most common using the AAN scale. Evidence-based Cantu Grade 1 (66.0%) and Grade 2 (29.9%) concussion grades were similar to traditional Cantu grades.

Associations of APOE, APOE promoter, and tau protein genotypes with *history of concussion* are presented in Table 4. The APOE promoter TT genotype, relative to the GG genotype,

TABLE 2. Numbers of Athletes With Concussions

Total No. of Concussions	Number of Athletes, n (%)
1	50 (69.4)
2	20 (27.8)
≥3	2 (2.8)

was statistically significantly associated with a nearly 3-fold increased risk for a history of 1 or more concussions, whereas there was no evidence of risk associated with the heterozygous genotype, GT. The TT genotype-concussion association was stronger for those with 5 or more sports-years of exposure (OR, 2.5; 95% CI, 1.0 to 6.5) and for those who were African-American (OR, 9.8; 95% CI, 1.6 to 59.0) (data not shown). There was no substantial evidence of an association between a history of concussion and APOE genotypes or haplotypes; however, the cell sizes for some of the APOE genotypes were so small that meaningful analysis was not possible. Also, compared to those with the APOE E3/E3 genotype, those with the E2/E3 genotype were at a 60% higher risk for concussion, but the results were not statistically significant (OR, 1.6; 95% CI, 0.5 to 4.8). Although there were no statistically significant associations in relation to the tau protein genotypes, there was a suggestion that the ^{Ser53Pro} polymorphism may be associated with increased risk for a history of one or more concussions (OR, 2.1; 95% CI, 0.3 to 14.5).

Associations of APOE G-219T promoter genotypes with a history of concussion according to the number and grade of concussions are shown in Table 5. There appeared to be substantially stronger associations with a single concussion and with high-grade concussions (Grade 2 or 3 traditional Cantu or AAN).

Some of the clinical scenarios that met our operational definition of concussion did not meet the strict criteria for

TABLE 3. Characteristics of Recorded Concussions

Activity	Concussions, n (%)
Sport-related concussions	
Football	72 (74.2)
Soccer	9 (9.3)
Other	5 (5.2)
Unknown	4 (4.1)
Non-sport related	
AAN concussion grade	
1	38 (39.2)
2	40 (41.2)
3	19 (19.6)
Cantu concussion grade	
1	68 (70.1)
2	25 (25.8)
3	4 (4.1)
Evidence-based Cantu	
1	64 (66.0)
2	29 (29.9)
3	4 (4.1)

TABLE 4. Multivariate-adjusted* Associations of Genotypes and Risk for Concussion in Participants

	Hx of Concuss (n = 72)† n, %	No Hx of Concuss (n = 123) n, %	Multivariate-Adjusted OR‡ (95% CI)§	P
<i>APOE</i> genotypes				
E3/E3	47, 68.1	85, 70.8	1.0	
E2/E2	1, 1.5	2, 1.7	0.6 (0.1 to 7.0)	
E2/E3	8, 11.6	11, 9.2	1.6 (0.5 to 4.8)	
E2/E4	2, 2.9	1, 0.8	1.3 (0.1 to 23.8)	
E3/E4	11, 15.9	17, 14.2	1.1 (0.4 to 2.7)	
E4/E4	0, 0.0	4, 3.3	None	0.88
<i>APOE</i> promoter genotypes				
E3 (33)	47, 68.1	85, 70.8	1.0	
E2 (22, 23)	9, 13.1	13, 10.8	0.9 (0.5 to 1.7)	
E4 (24, 34, 44)	13, 18.8	22, 18.3	1.3 (0.6 to 2.5)	0.82
<i>Tau Ser</i> genotypes				
GG	17, 25.0	35, 29.2	1.0	
GT	27, 39.7	58, 48.3	1.1 (0.5 to 2.5)	
TT	24, 35.3	27, 22.5	2.7 (1.1 to 6.8)	0.03
<i>Tau His</i> genotypes				
TT	53, 77.9	88, 77.9	1.0	
CT	13, 19.1	21, 18.6	1.4 (0.6 to 3.4)	
CC	2, 2.9	4, 3.5	2.1 (0.3 to 14.5)	0.28
CC + CT	15, 22.1	26, 22.8	1.5 (0.7 to 3.5)	0.31
<i>Tau His</i> genotypes				
TT	37, 54.4	64, 54.2	1.0	
TC	28, 41.2	49, 41.5	1.2 (0.6 to 2.5)	
CC	3, 4.4	5, 4.2	1.1 (0.2 to 5.8)	0.61
CC + TC	31, 45.6	54, 45.8	1.2 (0.6 to 2.5)	0.56

*Adjusted for age, sport, school, and number of years in sport.

†Number of cases and controls attempted for genetic analysis.

‡Odds ratio.

§95% confidence interval.

concussion advocated by the AAN or Cantu definition and grading system. When these clinical scenarios were reevaluated using these criteria (having the presence of at least one of the following is consistent with a concussion: loss of consciousness, memory loss, or confusion/disorientation), then 24 participants were reclassified into the “no concussion” control group. Repeating the same set of analyses described above for the APOE promoter G-219T polymorphism did not materially alter the magnitude of the associations, but they were no longer statistically significant. For example, using the AOSSM Work Group concussion definition²⁹ and the Cantu concussion grading scale,³¹ the multivariate-adjusted OR for the TT genotype-concussion association was 4.5 (95% CI, 0.7 to 31.4; *P*-trend across TT, CT, and CC genotypes = 0.09). Also, a similar pattern was seen with the tau ^{Ser53Pro} CC (OR, 4.5; 9% CI, 0.7 to 31.4; *P*-trend = 0.09) (data not shown).

DISCUSSION

To our knowledge, this is the first report of an investigation of an association between the APOE promoter gene G-219T polymorphism and a history of MTBI in athletes.

TABLE 5. Multivariate-Adjusted* Associations of APOE Promoter Genotypes and Risk for Concussion in Participants According to Number of Concussions and Concussion Grade

	GG		GT		TT	
	Cases/Controls OR† (95% CI)‡		Cases/Controls OR (95% CI)		Cases/Controls OR (95% CI)	
No. of concussions						
1	11/35	1.0	16/58	1.1 (0.4 to 2.9)	19/27	4.2 (1.4 to 12.1)
>1	6/35	1.0	11/58	1.4 (0.4 to 4.4)	5/27	1.5 (0.4 to 6.1)
AAN grade						
1	6/35	1.0	10/58	0.9 (0.3 to 3.0)	8/27	2.2 (0.6 to 8.0)
2	9/35	1.0	10/58	0.9 (0.3 to 2.8)	9/27	2.6 (0.8 to 8.8)
3	2/35	1.0	7/58	3.2 (0.6 to 17.4)	7/27	7.2 (1.2 to 41.7)
2 + 3	11/35	1.0	17/58	1.3 (0.5 to 1.2)	16/27	3.4 (1.2 to 10.1)
Evidence-based Cantu grade						
1	12/35	1.0	16/58	0.8 (0.3 to 2.1)	16/27	2.4 (0.9 to 6.7)
2 + 3	5/35	1.0	11/58	2.4 (0.6 to 9.5)	8/27	4.5 (1.0 to 19.4)

*Adjusted for age, school, sport, and number of years in sport.
†Odds ratio.
‡95% confidence interval.

The TT genotype was associated with a statistically significant nearly 3-fold increased odds of prior concussion and with a 4-fold increased odds of a prior concussion with loss of consciousness. Although not statistically significant, the results also suggest a trend towards a potential association of a history of concussion with the tau^{Ser53Pro} polymorphism. There was no substantial evidence for an association of a history of MTBI with APOE genotypes.

There are several lines of evidence to support the plausibility of genetic variants of the APOE, APOE promoter, and tau genes in influencing risk for concussion and/or sequelae from concussion. Animal and clinical studies found the APOE promoter and apolipoprotein to be associated with Alzheimer's disease (AD).^{15,16} Tau genotypes were associated with increased risk of AD in some studies²⁶ but no association with AD risk in others.³⁸

The APOE gene and apolipoprotein (Apo E) play important roles in the pathophysiological response to head injury and post-TBI outcome.³⁹ After head injury in animal studies, there is upregulation of Apo E and amyloid precursor peptide (APP) production,^{40,41} and the APOE-mediated lipid transport system is stimulated.³⁹ Amyloid β -protein deposition after TBI is affected by APOE isoform. Greater deposition of amyloid β -protein occurs in APOE ϵ 4 bearers after TBI⁴² and in AD brains.⁴³ TBI victims, boxers with CTBI, and AD patients all develop similar brain neuropathology: amyloid β -protein deposits and neurofibrillary tangles (NFT) comprised of tau protein and amyloid β -protein.²⁴

Relative APOE expression regulates apolipoprotein levels and influences neurological recovery. The genetic variability in APOE expression is due to allelic variation in the APOE promoter region as manifested by G-219T, -491A/T, and other polymorphisms on the basis of *in vitro*²³ and *in vivo*⁴⁴ studies. Specifically, the G-219T TT genotype has been associated with lower APOE transcriptional activity,²³ a weakly less favorable post-TBI outcome (based on the Glasgow Outcome Scale),²⁵ and an increased risk of AD.¹⁶

The number of senile plaques in non-AD patient aging brains in the CA1 and subiculum regions was found to be significantly greater among those with the G-219T TT genotype, although this may not have been independent of the ϵ 4 allele.⁴⁸

The transcriptional activity of the APOE gene also has been correlated with AD risk.^{35,45}

A case-control study showed that plasma Apo E concentration very significantly decrease with the APOE promoter G-219T polymorphism T allele.²⁵ In addition, the APOE promoter G-219T G allele may protect against developing AD and TBI.⁴⁷ We hypothesize that concussion in an APOE promoter TT individual may promote an already genetically predetermined increased rate of neurodegeneration of the brain.

Tau protein deposition is one of the pivotal steps in the pathway that leads to brain neuropathology. The tau gene, encoding tau protein production, may modulate concussion risk. APOE ϵ 4 binds more weakly than APOE ϵ 2 or APOE ϵ 3 to tau protein,⁴⁹ allowing tau to be hyperphosphorylated, preventing normal binding to microtubules. Consequently, tau protein self-assembles into paired helical filaments and NFT.⁵⁰

A number of postulations may be suggested as to why the genes we studied may influence risk of concussion. Teasdale first highlighted the possibility of a "genetically determined variation between individuals in the acute response to brain injury."⁵ Genetic variation may also exist in the acute response to traumatic force to the brain and may dictate whether the head trauma results in a full-fledged case of MTBI.

The previous discussion provides ample evidence of why these genes may influence risk of concussion. Apolipoprotein E has an important role in the response to nervous system injury.³⁹ The same factors that lead to a poorer post-TBI outcome may also contribute to a more vulnerable baseline neurological substrate that is more likely to respond to a given traumatic force with the pathobiological

neurochemical cascade experienced in MTBI. The fact that APOE ϵ 4 allele bearers are more likely to have Amyloid- β deposition after head injury supports a genetic susceptibility to the effects of a head injury that is conferred by the APOE ϵ 4 allele.⁴³ In similar fashion, we postulate that APOE ϵ 4 and the APOE G-219T TT genotype may influence some of the pathobiological changes that occur in the brain after blunt head trauma. The increase in aspartate and glutamate levels, multiple ionic fluxes, and the hypoperfusion/hyperglycolysis mismatch characteristic of the pathobiology of concussion may be regulated by a complex set of gene-gene interactions among the APOE gene, tau gene, APOE promoter, and perhaps the NMDA receptor gene. APOE expression is affected by APOE genotype and G-219T genotype, as well as the inciting head trauma itself. Future research can focus on other genetic and environmental factors that affect APOE expression and the potential genetic regulation of this cascade of events.

Although our study is the first to use a young, mixed sex/race college age athletic population, prior genetic association studies on TBI outcome have focused on moderate/severe TBI in predominantly older male neurotrauma patients. A retrospective study of 87 moderate/severe TBI cases at a neurosurgical center suggested that the APOE promoter polymorphism G-219T TT genotype may lead to a less favorable outcome (death, vegetative state, or severe disability) 6 months after TBI.²⁴ Due to the small sample size, these findings may have been due to chance. Furthermore, the outcome measures are not generalizable to athletic populations.

The association of APOE with TBI outcome was reported from 1 study, a cross-sectional study of professional football players ($n = 53$).⁵¹ Having 1 APOE ϵ 4 allele was directly associated with lower neuropsychological (NP) test scores, a measure of neurocognitive function.⁵¹ The lower scores, however, may have been caused by interindividual differences in native intelligence or in head injury history.

Another study investigated the risk for CTBI in boxers.¹⁷ The cross-sectional study of 30 boxers found that APOE ϵ 4-allele positive, "high-exposure" boxers had significantly higher chronic brain injury scores and may be at higher risk for chronic traumatic brain encephalopathy (chronic TBI).¹⁷ These results may not be generalizable to assessing acute TBI risk or to non-boxing athletic populations.

Two small non-athlete studies on the association of APOE genotype on TBI outcome have been reported. First, a cross-sectional study of 80 mild/moderate TBI patients from a major trauma center found that APOE ϵ 4 may increase risk for an acute head injury with an abnormal computed tomography (CT) scan.⁵² The association was weak and thus possibly due to chance. Moreover, the study population was quite different from an athletic population: 60% had GCS scores of 9 to 14, 59% were MVA victims, there were no recorded sports-related injuries, and 55% had abnormal CT scans. In addition, study population features such as more severe pre-morbid head injury histories may have accounted for the increased number of abnormal CT scans in APOE ϵ 4 patients. In contrast, all of our participants had MTBI with normal CT scans, with 93% of the concussions being sports-related.

This study has several strengths and limitations. This study is, to our knowledge, the first to investigate the association

of the APOE promoter G-219T and the tau gene with concussion. The results are based on genetic testing on the largest population-based sample of mixed sex and race college football and soccer players reported in the literature. In contrast to previous studies, we focused on a homogeneous sample of MTBI cases with no abnormal CT scans or protracted symptoms. The study population is representative of the typical college sports population and represents a cross-section of competitive athletic divisions. The retrospective study design has certain inherent weaknesses, such as recall bias from using historical self-report to obtain concussion histories. However, recent studies substantiate the reliability of self-reported concussion histories.⁵³ The inclusion of the small number of non-sport-related concussions (7%) did not materially impact the study results. Underreporting of concussions by athletes at the collegiate and professional levels is well established.^{54,55} Thus, our study may have underestimated the true number of concussions in our population. However, such misclassification tends to bias associations to the null value; therefore, the association between G-219T and concussion history may have actually been stronger than we estimate. Finally, a history of concussions that occurred more than 8 years from the time of data collection was not collected but is unlikely to have materially affected our results.

We report the first evidence in the literature of an association between the APOE promoter G-219T polymorphism and a self-reported history of concussion in college athletes. The TT genotype was associated with a nearly 3-fold increased risk of a history of all concussions combined, and with a 4-fold increased risk of a history of concussions with loss of consciousness. A large, prospective cohort study is needed to confirm the associations between genotypes of the APOE, APOE promoter, and tau protein genes and a history of concussion and demonstrate that the risk of concussion is increased by the G-219T TT genotype. The authors are currently completing a prospective cohort study.

Given the preliminary nature of these results and the retrospective study design, it would be very premature and inappropriate in our opinion to attempt to apply our reports to clinical practice. The study findings need to be replicated in a large, multi-center, prospective cohort study before any reasonable application to clinical practice should be considered. However, we do agree that this study does raise the prospect that future genetic studies may uncover results that create a legitimate role for genetic analysis in clinical evaluation, management, and treatment of concussion. The American Society of Human Genetics states in its position statement on APOE genotyping that it does not recommend sharing these results with individuals given the fact that APOE ϵ 4 is a risk factor for AD, an incurable disease. This underscores the delicate nature of genetic testing and the profound ethical implications of such an approach. It is conceivable that biomarkers or genetic polymorphisms that predict adverse neurocognitive outcome from concussion, multiple concussions, life-threatening head injury, or post-concussion syndrome may be identified in the future. If that day comes, clinicians and scientists will need to carefully weigh the profound ethical implications of the inappropriate or premature use of unproven genetic testing to unfairly limit or exclude athletes from sports participation.

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