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Serum titers of IgG antibodies against tetanus and diphtheria toxoids and risk of multiple sclerosis

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Abstract

We conducted a prospective nested case-control study among military service members to investigate whether antibodies against tetanus or diphtheria predict multiple sclerosis (MS) risk. Paired T-tests were used to compare means of anti-tetanus and diphtheria toxoids among 56 MS cases and 112 matched controls. Conditional logistic regression was used to estimate odds ratios (OR). There were no differences between the mean serum IgG antibodies against tetanus (p-value 0.28) or diphtheria (p-value 0.45) in the baseline samples. The OR of MS associated with 1 standard deviation difference in antibody titers was 0.76 (95% CI: 0.48-1.21) for tetanus (SD=4.71) and 1.03 (0.73-1.45) for diphtheria (SD=0.87). Results of this study suggest serum IgG antibodies against tetanus or diphtheria are not predictors of MS risk.

Keywords

Multiple sclerosis; Tetanus; Diphtheria; Etiology; Prospective studies; IgG Antibodies

Introduction

Previous studies have suggested that tetanus vaccination is associated with a decreased risk of multiple sclerosis (MS) (Hernan et al., 2006) and that individuals with MS have lower IgG antibodies against tetanus and diphtheria toxoid (Lamoureux et al., 1976). It has been hypothesized that because the tetanus toxoid contains a universal human T-helper cell epitope, it may shift type 1 (pro-inflammatory cytokines) to type 2 (anti-inflammatory cytokines) responses to environmental stimuli and reduce MS risk (Etlinger et al., 1990; Hernan et al., 2006; Vandenbark et al., 1996). The mechanism by which immunization to diphtheria could alter risk of MS is unclear, however it might be related to intrathecal immunoglobulin

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production (Salmi et al., 1981). To further investigate these hypotheses, we conducted a prospective study to determine whether antibodies against tetanus or diphtheria in healthy individuals predict their future risk of MS. A strength of this study is that antibody titers were measured in blood samples collected several years before the onset of the first neurological symptoms of MS (MS onset).

Methods

We conducted a prospective, nested case-control study among 7 million military service members with serum stored in the Department of Defense Serum Repository (DoDSR). Since 1985, military personnel are routinely tested for HIV at entry into the military and on average, every 2 years thereafter. The residual serum samples from this testing are catalogued and stored by the DoDSR. As previously described (Munger et al., 2006), we have confirmed 315 cases of MS among active duty personnel in the US Army (1993-2004) and Navy (1992-2004) with at least one, but up to three, samples collected before their date of MS onset in the DoDSR. The current study includes 56 of these cases occurring in the US Army between January 1998 and July 2000. The average number of years between baseline sample collection and MS onset was 5.54. Each MS case was matched with two controls on age (± 1 year), sex, race (white, black, Hispanic, other), and date of serum collection (± 30 days). IgG antibodies to tetanus and diphtheria toxoids were measured by indirect ELISA from The Binding Site, San Diego, CA. The assays use the quantitative sandwich enzyme immunoassay technique. The reproducibility of the assays was assessed using blind quality control serum samples, each split into three identical aliquots. These samples were run at the same time as the MS case and control samples. The coefficients of variation for the intra-assay samples for anti-tetanus toxoid was 14.2% and 18.2% for the anti-diphtheria toxoid. Paired T-tests were used to compare sample means between cases and controls and conditional logistic regression to estimate odds ratios (OR) and 95% confidence intervals.

Results

No differences were found in mean levels of serum IgG antibodies against tetanus toxoid or diphtheria between the MS cases and their matched controls (Table).

The OR of MS associated with 1 standard deviation (SD) difference in antibody titers in the baseline sample was 0.76 (95% CI: 0.48-1.21) for tetanus (SD=4.71) and 1.03 (0.73-1.45) for diphtheria (SD=0.87). The OR for tetanus after exclusion of outliers (2 controls with antibody titers > 3 SD of the mean) was 0.88 (95% CI: 0.62-1.25) for 1 SD (SD=3.07).

Discussion

These findings based on prospectively collected serum samples do not support the hypothesis that individuals with higher serum IgG antibodies against tetanus or diphtheria have a lower risk of developing MS than those with lower levels. Because antibody titers increase soon after each booster of tetanus and diphtheria vaccine and then decline gradually over a period of 10 or more years, the lack of association between antibody titers and MS risk also does not corroborate previous results suggesting that MS risk is reduced within 5-years following a tetanus vaccination (Hernan et al., 2006). The present investigation is, however, limited by the small sample size and modest effects cannot be excluded. Further, because recruits are routinely vaccinated, we could not compare the risk of MS between vaccinated and not vaccinated individuals.

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References

- Borrow, R.; Blamer, P.; Roper, MH. The Immunological Basis for Immunization Series Module 3: Tetanus. Geneva, World Health Organization, (WHO/EPI); 2006.
<http://www.who.int/vaccines-documents/DocsPDF07/869.pdf>
- Etlinger HM, Gillissen D, Lahm HW, Matile H, Schonfeld HJ, Trzeciak A. Use of prior vaccinations for the development of new vaccines. *Science* 1990;249:423–425. [PubMed: 1696030]
- Hernan MA, Alonso A, Hernandez-Diaz S. Tetanus vaccination and risk of multiple sclerosis: a systematic review. *Neurology* 2006;67:212–215. [PubMed: 16864810]
- Lamoureux G, Giard N, Jolicoeur R, Toughlian V, Desrosiers M. Immunological features in multiple sclerosis. *Br Med J* 1976;1:183–186. [PubMed: 764920]
- Munger KL, Levin LI, Hollis BW, Howard NS, Ascherio A. Serum 25-hydroxyvitamin D levels and risk of multiple sclerosis. *Jama* 2006;296:2832–2838. [PubMed: 17179460]
- Salmi A, Viljanen M, Reunanen M. Intrathecal synthesis of antibodies to diphtheria and tetanus toxoids in multiple sclerosis patients. *J Neuroimmunol* 1981;1:333–341. [PubMed: 7334086]
- Vandenbark AA, Chou YK, Whitham R, Mass M, Buenafe A, Liefeld D, Kavanagh D, Cooper S, Hashim GA, Offner H. Treatment of multiple sclerosis with T-cell receptor peptides: results of a double-blind pilot trial. *Nat Med* 1996;2:1109–1115. [PubMed: 8837609]

Table
Serum IgG antibodies against tetanus and diphtheria toxoids (IU/ml) in MS cases and controls

		N*	Mean	Median	Standard Deviation	P value**
Tetanus						
Baseline	Cases	55	3.70	2.85	2.75	0.30
	Controls	108	4.46	3.50	4.71	
Average of 2	Cases	24	3.26	3.26	2.05	0.20
	Controls	47	4.56	3.43	4.06	
Average of 3	Cases	24	3.29	2.86	1.69	0.28
	Controls	45	4.11	3.27	2.97	
Diphtheria						
Baseline	Cases	55	0.91	0.73	0.89	0.88
	Controls	108	0.89	0.75	0.87	
Average of 2	Cases	24	0.96	0.71	0.80	0.47
	Controls	47	0.83	0.68	0.70	
Average of 3	Cases	24	0.93	0.73	0.69	0.45
	Controls	45	0.81	0.74	0.64	

* 56 MS cases and 112 controls had serum samples included in these analyses. Smaller number of case and control samples may be present either because an individual did not have all 3 serum samples collected, or because an individual's serum sample was not analyzed in lab in the same batch as his other samples.

** Paired T-test used to compare mean serum samples between cases and controls.