Serum Dioxin and Diabetes Mellitus in Veterans of Operation Ranch Hand

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Abstract

We studied diabetes in Air Force veterans exposed to Agent Orange and its contaminant 2,3,7,8 tetrachlorodibenzo-p-dioxin (dioxin) during the Vietnam War. The index subjects of the Air Force's ongoing 20-year prospective epidemiological study are veterans of Operation Ranch Hand (N=989), the unit responsible for aerial herbicide spraying in Vietnam from 1962 to 1971. Other Air Force veterans who served in Southeast Asia during the same period but were not involved with spraying herbicides serve as Comparisons (N=1,276). The median dioxin level in the Ranch Hand group was 12.2 parts per trillion (ppt) [range: 0 to 617.8 ppt] and the median dioxin level in the Comparison group was 4.0 ppt (range: 0 to 10 ppt). We found that diabetes prevalence (relative risk=1.5, 95% CI 1.2 to 2.0) and the risk of diabetes requiring oral medication (relative risk=2.3, 95% CI 1.3 to 3.9) increased with dioxin. These results suggest an adverse relation between dioxin exposure and diabetes mellitus.

Introduction

Studies of exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin (dioxin) in animals have found a wide range of species-, strain-, age- and sex-specific effects, including carcinogenicity, immunotoxicity, reproductive and developmental toxicity, hepatoxicity, neurotoxicity, chloracne, and loss of body weight[1], however endocrine toxicity is not widely acknowledged[1,2].

Diabetes is a disorder of the pancreas which results in a number of significant effects on the body, including macroangiopathy, neuropathy and cataracts. Primary risk factors for diabetes are family history, obesity (over 80% of all diabetics are overweight when they are diagnosed) and physical or emotional stress[3]. Little has been reported on diabetes prevalence or glucose and serum insulin levels in Vietnam veterans. We summarize diabetes prevalence and exposure to dioxin in veterans of Operation Ranch Hand, the unit responsible for the aerial spraying of herbicides, including Agent Orange, in Vietnam from 1962 to 1971. These data have been gathered during ten years of follow-up in the ongoing Air Force Health Study (AFHS) from veterans whose exposure in Vietnam occurred from 24 to 35 years ago. This report is derived from a more extensive summary of the relation between diabetes, glucose abnormalities, insulin abnormalities and time to onset[4].

Methods

The study seeks to determine whether veterans of Operation Ranch Hand (the personnel tasked with spraying operations during the Vietnam conflict) have experienced adverse health and whether those health effects, if they exist, can be attributed to exposure to herbicides or their dioxin contaminant. Ranch Hand veterans were exposed to herbicides during flight operations and maintenance of the aircraft and herbicide spray equipment. The study compares the current health and cumulative mortality experience of Ranch Hand veterans with a comparison group of other Air Force veterans who served in Southeast Asia during the same period (1962 to 1971) that the Ranch Hand unit was active and who were not involved with spraying herbicides. Comparisons were matched to Ranch Hands on age, race and military occupation. The study includes periodic analyses of noncombat mortality, in-person interviews and physical examinations. Physical examinations were conducted in 1982, 1985, 1987 and 1992 and additional examinations are planned for 1997 and 2002.

In 1987, blood from willing participants was collected and assayed for dioxin. Participation was voluntary and consent forms were signed at the examination site. Veterans with no quantifiable dioxin result in 1987, those who refused in 1987 and subjects new to the study were also asked to give blood for the assay at the 1992 examination.

Diabetes cases included for analysis were diagnosed during the post-Vietnam period from the end of the veteran's last tour of duty to June, 1995. We report cumulative post-service diabetes and diabetes severity. Each case was verified from medical records and may represent a diagnosis at any of the four physical examinations. Every veteran who attended at least one examination, regardless of his current vital status, was considered for inclusion in the analysis.

We reviewed medical records and laboratory results to determine diabetic status. Veterans who attended at least one examination and had a verified history of diabetes by medical diagnosis or exhibited a 2-hour postprandial glucose laboratory value of 200 mg/dl or greater were classified as diabetic. Veterans not meeting these criteria were defined as nondiabetic.

We defined diabetic severity based on a review of medical records and the latest questionnaire responses. We assigned each veteran with diabetes to one of four categories of control: "Insulin Therapy", "Oral Medications", "Diet Only", or "No Control" and included veterans without diabetes in a category named "No Diabetes". When assessing associations between diabetic severity and dioxin, we considered each severity category separately. When studying insulin therapy, we restricted the analysis to diabetic veterans taking insulin and nondiabetic veterans. We analyzed diabetics on oral medications and diabetics on diet control in a similar fashion. Lastly, we combined "Diet Only", "Oral Medications" and "Insulin Therapy" into a single category named "Any Control" and assessed its association with dioxin category.

We excluded from all statistical analyses veterans with a history of diabetes prior to service in Southeast Asia, those with no dioxin measurement, those with a nonquantifiable dioxin result and Comparisons with a dioxin result greater than 10 parts per trillion (ppt), the value we regard as the threshold for background dioxin exposure. Table 1 shows sample size reductions by group (Ranch Hand, Comparison).

Table 1
Sample Size Reduction by Group

	Ranch Hand	Comparison	Total
Fully Compliant at any Exam	1,108	1,494	2,602
Missing Dioxin	(100)	(140)	(240)
Nonquantifiable Dioxin	(17)	(50)	(67)
Diabetes prior to service in Southeast Asia	(2)	(3)	(5)
Current Dioxin > 10 ppt	(0)	(25)	(25)
Net	989	1,276	2,265

We estimated the initial dioxin dose at the end of the tour of duty in Vietnam in Ranch Hands having current dioxin levels above background using a constant half-life of 8.7 years[5] and assigned each veteran to one of four exposure categories, named "Comparison", "Background", "Low" and "High", according to his group, current dioxin level (D) and initial dioxin level (I), defined in Table 2. The cut point separating the Low and High categories (94 ppt) is the median initial dioxin level among all Ranch Hands having current dioxin levels greater than 10 ppt. Table 2 shows sample sizes by dioxin category.

Table 2
Exposure Category Definition and Associated Sample Sizes

Dioxin Category	Definition*	Sample Size
Comparison	$D \leq 10$	1,276
Ranch Hand		
Background	$D \leq 10$	422
Low	$10 < D$ & $I \leq 94$	284
High	$10 < D$ & $I > 94$	283
Total		2,265

* D = current dioxin; I = initial dioxin; in parts per trillion.

We defined percent body fat[6] (PBF) as PBF=1.26×BMI-13.305, where BMI is the body mass index [weight (kg) divided by the square of height (m)] and adjusted all analyses for birth year (born before 1942, born during or after 1942) and PBF at time of dioxin blood draw (25% or less, more than 25%) using stratification[7]. We report relative risk, defined as the ratio of the prevalence of the abnormality or disease in the Ranch Hand cohort to the corresponding prevalence in the Comparison cohort and compute 95% confidence limits using the method of Rothman[7].

Because dioxin half-life increases with PBF measured in 1982[5], we conducted additional analyses by calculating a half-life in each of three strata determined by the tertiles of 1982 PBF and used these to revise our estimate of the initial dioxin level (I) and the Low and High categories. In a separate series of analyses, we matched Ranch Hands in the Background, Low and High categories to Comparisons on age to within one year, race (black, nonblack), percent body fat to within 3%, and military occupation (officer, enlisted flyer, enlisted groundcrew). We computed the point estimates of the relative risk and associated confidence intervals for a varying number of Comparisons matched to each Ranch Hand. We also studied the effect of changing our definition of exposure by using the tertiles of the current dioxin distribution in Ranch Hands to define the dioxin categories.

Results and Discussion

Demographic characteristics of all veterans are presented in Table 3. Ranch Hands in the High dioxin category are younger than Ranch Hands in the Low and Background categories. The median (and range) of current dioxin levels, in ppt, in the Low and High categories were, Low: 15.0 (10.0 to 26.6), High: 46.2 (18.0 to 617.8); the intervals overlap because these categories were defined by initial, not current, dioxin.

Table 3
Distribution of Dioxin and Demographic Characteristics by Dioxin Exposure Category

Characteristic	Comparison	Ranch Hand Background	Ranch Hand Low	Ranch Hand High
Dioxin (ppt)*				
median	4.0	5.7	52.7	197.5
range	0 to10	0 to10	27 to 94	94 to 3,290
Age (years)				
Mean (SD)	53.5 (7.6)	54.6 (7.2)	54.9 (7.6)	50.9 (7.4)
Percent Body Fat				
Mean (SD)	21.8 (5.1)	20.2 (4.5)	22.2 (5.3)	23.4 (5.6)

*Current dioxin levels in the Comparison and Background categories, initial dioxin in the Low and High categories, in parts per trillion.

The percentages of Ranch Hands in the Low (relative risk=1.3) and High (relative risk=1.5) dioxin categories having diabetes are increased relative to the Comparisons (Table 4). The percentage of Ranch Hands in the Background category with diabetes is less than the Comparison percentage (relative risk=0.7).

Table 4
Diabetes by Dioxin Exposure Category

Condition	Comparison	Ranch Hand Background	Ranch Hand Low	Ranch Hand High
Diabetes				
Number (%)	169 (13.2)	40 (9.5)	49 (17.2)	57 (20.1)
RR	1.0	0.7	1.3	1.5
95% CI		(0.5,1.0)	(1.0,1.7)	(1.2,2.0)

The percentages of diabetic Ranch Hands in the Low (relative risk=1.6) and High (relative risk=1.5) categories on diet control are increased (Table 5), but the percentage of diabetic Ranch Hands in the Background category on diet control is decreased (relative risk=0.5). Diabetic Ranch Hands in the High category are more likely than diabetic Comparisons to be controlling their glucose levels with oral medications (relative risk=2.3), taking insulin (relative risk=2.4) or to be using any control (relative risk=1.8). The risks of insulin therapy (relative risk=2.7) and any control (relative risk=1.4) are increased in the Low category. The increased risks of any control are similar to those in Table 4 because the population using any control comprises veterans with diabetes excluding those using no control, while all diabetics are included in the totals in Table 4.

Table 5
Diabetes Severity and Dioxin Exposure Category

Severity Level	Comparison	Ranch Hand Background	Ranch Hand Low	Ranch Hand High
Diet only				
Number (%)	47 (4.1)	8 (2.0)	16 (6.4)	15 (6.2)
RR	1.0	0.5	1.6	1.5
95% CI		(0.2,1.0)	(0.9,2.7)	(0.9,2.7)

Table 5 continued

		Ranch Hand		
Severity Level	Comparison	Background	Low	High
Oral Medications				
Number (%)	39 (3.4)	2 (0.5)	7 (2.9)	19 (7.8)
RR	1.0		0.9	2.3
95% CI			(0.4,1.9)	(1.3,3.9)
Insulin Therapy				
Number (%)	12 (1.1)	8 (2.0)	7 (2.9)	6 (2.6)
RR	1.0	1.9	2.7	2.4
95% CI		(0.8,4.6)	(1.1,6.8)	(0.9,6.4)
Any Control				
Number (%)	98 (8.1)	18 (4.5)	30 (11.3)	40 (15.0)
RR	1.0	0.5	1.4	1.8
95% CI		(0.3,0.9)	(0.9,2.0)	(1.3,2.6)

Although not shown here, we also found consistent increases in the risk of glucose abnormalities with dioxin and, in nondiabetic Ranch Hands, the risk of abnormally high serum insulin increased with dioxin[4]. Results in other epidemiological studies are mixed. A follow-up study of German industrial workers exposed to dioxin found diabetes less often in the exposed group than among referents[8]. In another study of the same cohort, mean fasting glucose levels in the exposed group appeared to increase with current dioxin, but not back-extrapolated initial dioxin[9]. A study of dioxin-exposed US industrial workers found an increased mean dioxin level in diabetic workers compared with nondiabetic workers and increased mean fasting serum glucose in workers as compared with referents[10]. In the Vietnam Experience Study[11], diabetes prevalence in the Vietnam veteran cohort was similar to that in the non-Vietnam veteran cohort (relative risk=1.1).

When we accounted for a changing half-life by calculating a separate half-life in each of three strata and revising our initial dose the results were similar to those reported here and did not lead us to a different conclusion. We considered the possibility that our method of adjustment may be masking an effect and matched Ranch Hands in the Background, Low, and High categories to Comparisons on a one-to-many basis on age, race, percent body fat, and military occupation. The matched results were similar to those reported here and did not lead us to a different conclusion. Results from analyses that studied the effect of changing our definition of Background, Low, and High dioxin exposure in Ranch Hands by using the tertiles of the current dioxin distribution in Ranch Hands to define the categories produced negligible changes in the results.

Studies of glucose transport in animals dosed with dioxin in the range 0.03 to 1.0 μg/kg have demonstrated reduced glucose transport in adipose, liver and pancreas tissue in Guinea pigs, mice and rats[12]. These doses are biologically relevant to this study because the median dioxin body burden in Ranch Hands is 0.07 μg/kg, the first and third quartiles are 0.03 μg/kg and 0.14 μg/kg, and the 99th percentile is 1.0 μg/kg. That dioxin may be associated with diabetes and glucose and insulin levels in Ranch Hands therefore appears plausible, although a specific mechanism of dioxin alteration of glucose transport has not been established and studies of glucose transport in human adipocytes have not been carried out. Other studies of rats given much higher doses of dioxin (more than 100 μg/kg), found glucose[13] and insulin[14] decreased in exposed animals.

The strengths of this study include high participation and low attrition rates, a Comparison population closely matched to the index population, and 10 years of follow-up. Repetitive examinations and active quality control incorporating double blind entry of data with discordances referred for third-party review and medical review of potential outliers reduced errors that would bias the study toward the null result.

Our ability to detect associations is limited by the fixed size of the Ranch Hand cohort. Since all Ranch Hands have been identified and invited to participate in the study, their number cannot be increased. Thus, the rarity of some abnormalities led to imprecise measures of association, as indicated by wide confidence intervals, and small numbers prevented us from strong inferences on the most heavily exposed Ranch Hands.

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