TABLE 16-18. (Continued)

Analysis of NKCA 50/1 Net Response (cpm)

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[a]	p-Value
e) Minimal						0.480[b]
(n=191)	≤18.6	467.7 (21)	376.0 (47)	501.4 (22)	19.3 (21.8)	0.380[c]
(R^2=0.355)	>18.6	367.8 (25)	378.0 (47)	405.5 (29)	-1.9 (21.7)	0.932[c]
f) Maximal						0.277[b]
(n=268)	≤18.6	438.9 (38)	398.6 (67)	441.7 (30)	9.6 (14.9)	0.521[c]
(R^2=0.385)	>18.6	409.7 (24)	394.6 (67)	394.7 (42)	-13.1 (14.5)	0.368[c]

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
g) Minimal						0.253[b]	RACE (p=0.068)
(n=190)	≤18.6	516.1 (21)	463.9 (47)	569.8 (22)	21.8 (20.1)	0.278[c]	PACKYR (p=0.080)
(R^2=0.467)							ALC (p<0.001)
	>18.6	453.5 (25)	481.8 (46)	472.1 (29)	-9.9 (20.4)	0.629[c]	
h) Maximal						0.060[b]	ALC (p<0.001)
(n=267)	≤18.6	426.4 (38)	391.1 (67)	432.5 (30)	12.0 (14.1)	0.394[c]	CSMOK*PACKYR (p=0.036)
(R^2=0.465)	>18.6	437.7 (24)	411.4 (66)	387.5 (42)	-25.6 (13.9)	0.067[c]	

[a]Slope and standard error based on NKCA 50/1 net response versus log_2 dioxin.

[b]Test of significance for current dioxin-by-time interaction (current dioxin and time continuous).

[c]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-18. (Continued)

Analysis of NKCA 50/1 Net Response (cpm)

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean	Contrast	Difference of Means (95% C.I.)	p-Value
Background	291	416.7	All Categories		0.266
Unknown	126	423.3	Unknown vs. Background	6.6 (-35.5,48.7)	0.759
Low	71	373.6	Low vs. Background	-43.1 (-95.2,9.0)	0.106
High	72	387.9	High vs. Background	-28.8 (-82.2,24.6)	0.291
Total	560		(R^2=0.347)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean	Contrast	Difference of Adj. Means (95% C.I.)	p-Value	Covariate Remarks
Background	291	414.2	All Categories		0.299	ALC (p=0.021) CSMOK*PACKYR (p=0.003)
Unknown	126	425.2	Unknown vs. Background	11.0 (-30.5,52.6)	0.604	
Low	70	377.8	Low vs. Background	-36.4 (-87.9,15.1)	0.167	
High	72	386.3	High vs. Background	-27.9 (-80.2,24.5)	0.297	
Total	559		(R^2=0.377)			

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

NKCA 50/1 Percent Release

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

In the unadjusted analysis of NKCA 50/1 percent release and initial dioxin, the association was not significant under both the minimal and maximal assumptions (Table 16-19 [a] and [b]: p=0.813 and p=0.575).

Under the minimal assumption, there was a significant interaction between initial dioxin and current cigarette smoking and a significant interaction between initial dioxin and lifetime alcohol history (Table 16-19 [c]: p=0.036 and p=0.037, respectively). To investigate the interactions, the four categories of current smoking (never, former, 20 cigarettes or less per day, and over 20 cigarettes per day) were examined with two categories of dichotomized lifetime alcohol history (less than or equal to 40 drink-years and greater than 40 drink-years). For Ranch Hands who never smoked, and Ranch Hands who were former smokers but had more than 40 drink-years of lifetime alcohol history, there were nonsignificant negative associations between NKCA 50/1 percent release and initial dioxin (Appendix Table O-1). For the other strata combinations of current cigarette smoking and lifetime alcohol history, there were nonsignificant positive associations. Without the two interactions in the model, the association between NKCA 50/1 percent release and initial dioxin was not significant (Table 16-19 [c]: p=0.748).

Under the maximal assumption, NKCA 50/1 percent release and initial dioxin were not significantly associated (Table 16-19 [d]: p=0.714).

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

For both the minimal and maximal assumptions, the unadjusted analysis of NKCA 50/1 percent release did not contain a significant interaction between current dioxin and time since tour (Table 16-19 [e] and [f]: p=0.735 and p=0.745, respectively); thus, the slopes did not differ significantly between time strata.

For each assumption, the adjusted analysis of NKCA 50/1 percent release also indicated that the current dioxin-by-time interaction was not significant (Table 16-19 [g] and [h]: p=0.465 and p=0.558, respectively); therefore, the adjusted slopes did not differ significantly between time strata.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

In the unadjusted analysis of the NKCA 50/1 percent release, the overall contrast of the four current dioxin categories was not significant (Table 16-19 [i]: p=0.199). The mean NKCA 50/1 percent release for Ranch Hands in the low current dioxin category was marginally lower than that of Comparisons in the background current dioxin category (p=0.072, 32.4 percent versus 35.9 percent).

The adjusted analysis of NKCA 50/1 percent release contained a nonsignificant overall contrast of the four current dioxin categories (Table 16-19 [j]: p=0.202).

TABLE 16-19.

Analysis of NKCA 50/1 Percent Release

Ranch Hands - Log_2 (Initial Dioxin) - Unadjusted

Assumption	Initial Dioxin	n	Mean	Slope (Std. Error)[a]	p-Value
a) Minimal (n=191) (R^2<0.001)	Low	44	34.8	0.228 (0.960)	0.813
	Medium	95	32.7		
	High	52	35.6		
b) Maximal (n=268) (R^2=0.001)	Low	64	36.2	-0.391 (0.695)	0.575
	Medium	133	33.8		
	High	71	34.7		

Ranch Hands - Log_2 (Initial Dioxin) - Adjusted

Assumption	Initial Dioxin	n	Adj. Mean	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
c) Minimal (n=190) (R^2=0.144)	Low	44	38.5**	0.293 (0.912)**	0.748**	INIT*CSMOK (p=0.036) INIT*DRKYR (p=0.037) RACE (p=0.073) ALC (p<0.001)
	Medium	94	37.8**			
	High	52	39.7**			
d) Maximal (n=267) (R^2=0.060)	Low	64	35.8	-0.247 (0.674)	0.714	CSMOK (p=0.013) ALC (p=0.001)
	Medium	132	34.2			
	High	71	34.8			

[a]Slope and standard error based on NKCA 50/1 percent release versus log_2 dioxin.

**Log_2 (initial dioxin)-by-covariate interaction ($0.01<p\leq0.05$); adjusted mean, adjusted slope, standard error, and p-value derived from a model fitted after deletion of this interaction.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.

TABLE 16-19. (Continued)

Analysis of NKCA 50/1 Percent Release

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[a]	p-Value
e) Minimal						0.735[b]
(n=191)	≤18.6	36.9	32.7	36.6	0.718 (1.498)	0.632[c]
(R^2=0.003)		(21)	(47)	(22)		
	>18.6	34.5	31.5	35.4	0.047 (1.292)	0.971[c]
		(25)	(47)	(29)		
f) Maximal						0.745[b]
(n=268)	≤18.6	36.8	34.3	34.2	-0.062 (1.040)	0.953[c]
(R^2=0.002)		(38)	(67)	(30)		
	>18.6	35.5	33.4	34.8	-0.528 (0.987)	0.593[c]
		(24)	(67)	(42)		

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
g) Minimal						0.465[b]	AGE (p=0.104)
(n=190)	≤18.6	38.9	37.6	41.6	1.491 (1.472)	0.313[c]	RACE (p=0.091)
(R^2=0.124)		(21)	(47)	(22)			PACKYR (p=0.064)
	>18.6	38.5	37.3	39.3	0.119 (1.276)	0.926[c]	ALC (p<0.001)
		(25)	(46)	(29)			
h) Maximal						0.558[b]	CSMOK (p=0.012)
(n=267)	≤18.6	35.9	34.4	34.0	0.186 (1.009)	0.854[c]	ALC (p=0.001)
(R^2=0.061)		(38)	(67)	(30)			
	>18.6	36.1	34.0	35.1	-0.631 (0.957)	0.510[c]	
		(24)	(66)	(42)			

[a]Slope and standard error based on NKCA 50/1 percent release versus log_2 dioxin.

[b]Test of significance for current dioxin-by-time interaction (current dioxin and time continuous).

[c]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-19. (Continued)

Analysis of NKCA 50/1 Percent Release

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean	Contrast	Difference of Means (95% C.I.)	p-Value
Background	291	35.9	All Categories		0.199
Unknown	126	36.8	Unknown vs. Background	0.9 (-2.2,4.0)	0.562
Low	71	32.4	Low vs. Background	-3.6 (-7.4,0.3)	0.072
High	72	34.6	High vs. Background	-1.3 (-5.2,2.5)	0.499
Total	560		(R^2=0.008)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean	Contrast	Difference of Adj. Means (95% C.I.)	p-Value	Covariate Remarks
Background	291	35.2	All Categories		0.202	ALC (p=0.037) RACE*CSMOK (p=0.006)
Unknown	126	36.6	Unknown vs. Background	1.4 (-1.7,4.5)	0.364	CSMOK*PACKYR
Low	70	32.2	Low vs. Background	-3.0 (-6.8,0.9)	0.132	(p=0.023)
High	72	33.7	High vs. Background	-1.5 (-5.3,2.3)	0.447	
Total	559		(R^2=0.049)			

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

NKCI 50/1 Net Response

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

In the unadjusted analysis of the NKCI 50/1 net response, the association with initial dioxin was nonsignificant for both the minimal and maximal assumptions (Table 16-20 [a] and [b]: p=0.790 for each).

Under both assumptions, the adjusted analysis of NKCI 50/1 net response were nonsignificant (Table 16-20 [c] and [d]: p=0.551 and p=0.665, respectively).

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

In the unadjusted analysis of NKCI 50/1 net response, the interaction between current dioxin and time since tour was not significant under the minimal assumption (Table 16-20 [e]: p=0.151).

Under the maximal assumption, the unadjusted analysis of NKCI 50/1 net response contained a marginally significant interaction between current dioxin and time (Table 16-20 [f]: p=0.056). For Ranch Hands with time of 18.6 years or less, there was a marginally significant positive association between NKCI 50/1 net response and current dioxin (p=0.080). Within this time stratum, the average NKCI 50/1 net responses were 806.1, 789.3, and 854.8 cpm for low, medium, and high current dioxin. For Ranch Hands with time over 18.6 years, there was a nonsignificant negative association (p=0.312).

The adjusted analysis of NKCI 50/1 net response contained a significant interaction among current dioxin, time, and race (Table 16-20 [g]: p=0.040) under the minimal assumption. To explore the interaction, adjusted analyses were performed for Blacks and non-Blacks separately (Appendix Table O-1). For Blacks, the interaction and time strata associations were reported and are based on sparse numbers within current dioxin and time categories. For non-Blacks, the current dioxin-by-time interaction was significant (p=0.033). For non-Black Ranch Hands with time of 18.6 years or less, there was a significant positive association with current dioxin (p=0.015) and a nonsignificant negative association with current dioxin for the other time stratum (p=0.680). A followup model without the interaction exhibited a marginally significant current dioxin-by-time interaction (Table 16-20 [g]: p=0.073). For Ranch Hands with time of 18.6 years or less, there was a significant positive association between NKCI 50/1 net response and current dioxin (p=0.027). For that time stratum, the average NKCI 50/1 net response for low, medium, and high current dioxin were 808.6, 798.7, and 910.7 cpm. For Ranch Hands with more than 18.6 years since their tour, there was a nonsignificant negative association (p=0.886).

In the adjusted analysis of NKCI 50/1 net response under the maximal assumption, there was a significant current dioxin-by-time-by-race interaction (p=0.008). Similar to the interaction analyses under the minimal assumption, adjusted analyses were again performed for Blacks and non-Blacks separately. For Blacks, the interaction and the time strata associations were reported and are based on sparse numbers (Appendix Table O-1). For non-Black Ranch Hands, there was a significant interaction for current dioxin and time (p=0.017). Non-Black Ranch Hands whose time since tour was 18.6 years or less displayed a significant positive association between NKCI 50/1 net response and current dioxin

TABLE 16-20.

Analysis of NKCI 50/1 Net Response (cpm)

Ranch Hands - Log_2 (Initial Dioxin) - Unadjusted

Assumption	Initial Dioxin	n	Mean	Slope (Std. Error)[a]	p-Value
a) Minimal	Low	44	817.2	2.6 (9.7)	0.790
(n=195)	Medium	97	806.2		
(R^2=0.896)	High	54	830.9		
b) Maximal	Low	63	802.9	1.6 (5.8)	0.790
(n=270)	Medium	134	803.7		
(R^2=0.898)	High	73	801.1		

Ranch Hands - Log_2 (Initial Dioxin) - Adjusted

Assumption	Initial Dioxin	n	Adj. Mean	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
c) Minimal	Low	44	812.2	5.7 (9.6)	0.551	DRKYR (p=0.112)
(n=194)	Medium	96	806.4			
(R^2=0.903)	High	54	838.6			
d) Maximal	Low	63	803.5	2.5 (5.7)	0.665	DRKYR (p=0.068)
(n=269)	Medium	133	803.1			
(R^2=0.903)	High	73	802.2			

[a]Slope and standard error based on NKCI 50/1 net response versus log_2 dioxin.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.

TABLE 16-20. (Continued)

Analysis of NKCI 50/1 Net Response (cpm)

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[a]	p-Value
e) Minimal						0.151[b]
(n=195)	≤18.6	830.6	798.5	887.8	19.4 (14.1)	0.172[c]
(R^2=0.899)		(22)	(49)	(23)		
	>18.6	818.8	806.5	791.9	-9.0 (14.0)	0.524[c]
		(24)	(47)	(30)		
f) Maximal						0.056[b]
(n=270)	≤18.6	806.1	789.3	854.8	15.8 (8.9)	0.080[c]
(R^2=0.901)		(38)	(70)	(31)		
	>18.6	826.7	791.1	785.0	-9.1 (8.9)	0.312[c]
		(23)	(65)	(43)		

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
g) Minimal						0.073**[b]	CURR*TIME*RACE
(n=194)	≤18.6	808.6**	798.7**	910.7**	32.6 (14.5)**	0.027**[c]	(p=0.040)
(R^2=0.916)		(22)	(49)	(23)			AGE (p=0.102)
	>18.6	797.2**	798.4**	802.3**	-2.0 (14.0)**	0.886**[c]	DRKYR (p=0.028)
		(24)	(46)	(30)			
h) Maximal						****	CURR*TIME*RACE
(n=269)	≤18.6	****	****	****	****	****	(p=0.008)
(R^2=0.913)		(38)	(70)	(31)			DRKYR (p=0.028)
	>18.6	****	****	****	****	****	
		(23)	(64)	(43)			

[a]Slope and standard error based on NKCI 50/1 net response versus log_2 dioxin.

[b]Test of significance for current dioxin-by-time interaction (current dioxin and time continuous).

[c]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

**Log_2 (current dioxin)-by-time-by-covariate interaction ($0.01<p\leq0.05$); adjusted mean, adjusted slope, standard error, and p-value derived from a model fitted after deletion of this interaction.

****Log_2 (current dioxin)-by-time-by-covariate interaction ($p\leq0.01$); adjusted mean, adjusted slope, standard error, and p-value not presented.

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-20. (Continued)

Analysis of NKCI 50/1 Net Response (cpm)

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean	Contrast	Difference of Means (95% C.I.)	p-Value
Background	298	808.9	All Categories		0.843
Unknown	123	802.3	Unknown vs. Background	-6.6 (-32.6,19.4)	0.620
Low	72	817.5	Low vs. Background	8.6 (-23.1,40.2)	0.596
High	74	802.6	High vs. Background	-6.3 (-38.5,25.9)	0.701
Total	567		(R^2=0.819)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean	Contrast	Difference of Adj. Means (95% C.I.)	p-Value	Covariate Remarks
Background	298	****	All Categories		****	DXCAT*RACE (p=0.016)
Unknown	123	****	Unknown vs. Background	****	****	DXCAT*ALC (p<0.001)
Low	71	****	Low vs. Background	****	****	RACE*PACKYR (p<0.001)
High	74	****	High vs. Background	****	****	RACE*DRKYR (p=0.018)
Total	566		(R^2=0.845)			CSMOK*PACKYR (p=0.020)

**** Categorized current dioxin-by-covariate interaction ($p \leq 0.01$); adjusted mean, confidence interval, and p-value not presented.

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

(p=0.020). For the other time stratum, the association between NKCI 50/1 net response and current dioxin was negative and nonsignificant (p=0.271).

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted analysis of NKCI 50/1 net response indicated that the overall contrast of the current dioxin categories for Ranch Hands and Comparisons was nonsignificant (Table 16-20 [i]: p=0.843).

The adjusted analysis of NKCI 50/1 net response contained a significant interaction between categorized current dioxin and race and an interaction between categorized current dioxin and current alcohol use (Table 16-20 [j]: p=0.016 and p<0.001, respectively). Because of sparse data on Blacks, the interactions were explored only for non-Blacks having one drink or less per day and non-Blacks having more than one drink per day. For non-Blacks who had one drink or less per day, the overall contrast of the current dioxin categories was nonsignificant (Appendix Table O-1: p=0.387). For non-Blacks who had more than one drink per day, the overall contrast of current dioxin categories was significant (p=0.010) and Ranch Hands with low current dioxin had a significantly higher NKCI 50/1 average net response than did the Comparisons with background current dioxin (p=0.002).

NKCI 50/1 Percent Release

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

For the unadjusted analyses of NKCI 50/1 percent release, the association with initial dioxin was not significant under both assumptions (Table 16-21 [a] and [b]: p=0.894 and p=0.758, respectively).

The adjusted analysis of NKCI 50/1 percent release exhibited nonsignificant associations with initial dioxin under both assumptions (Table 16-21 [c] and [d]: p=0.345 and p=0.421, respectively).

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

The unadjusted analysis of NKCI 50/1 percent release exhibited a nonsignificant interaction between current dioxin and time since tour under the minimal assumption (Table 16-21 [e]: p=0.176).

Under the maximal assumption, the unadjusted analysis contained a marginally significant interaction between current dioxin and time (Table 16-21 [f]: p=0.063). For Ranch Hands with time of 18.6 years or less, there was a marginally significant positive association with current dioxin (p=0.077) and a nonsignificant negative association (p=0.355) with current dioxin for Ranch Hands with earlier tours of duty. For time of 18.6 years or less, the average NKCI 50/1 percent releases for low, medium, and high current dioxin were 65.8, 65.0, and 69.7 percent.

In the adjusted analysis of NKCI 50/1 percent release, there was a significant interaction of current dioxin, time, and race (Table 16-21 [g]: p=0.033) under the minimal assumption. To explore this interaction, separate analyses were performed for Blacks and

TABLE 16-21.

Analysis of NKCI 50/1 Percent Release

Ranch Hands - Log_2 (Initial Dioxin) - Unadjusted

Assumption	Initial Dioxin	n	Mean	Slope (Std. Error)[a]	p-Value
a) Minimal (n=195) (R^2=0.693)	Low	44	66.3	0.1 (0.8)	0.894
	Medium	97	64.9		
	High	54	66.9		
b) Maximal (n=270) (R^2=0.705)	Low	63	65.6	0.1 (0.5)	0.758
	Medium	134	66.0		
	High	73	65.6		

Ranch Hands - Log_2 (Initial Dioxin) - Adjusted

Assumption	Initial Dioxin	n	Adj. Mean	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
c) Minimal (n=194) (R^2=0.722)	Low	44	65.2	0.8 (0.8)	0.345	AGE (p=0.109)
	Medium	96	65.1			DRKYR (p=0.065)
	High	54	68.1			
d) Maximal (n=269) (R^2=0.728)	Low	63	65.4	0.4 (0.5)	0.421	AGE (p=0.107)
	Medium	133	66.0			PACKYR (p=0.144)
	High	73	66.0			DRKYR (p=0.030)

[a]Slope and standard error based on NKCI 50/1 percent release versus log_2 dioxin.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.

TABLE 16-21. (Continued)

Analysis of NKCI 50/1 Percent Release

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[a]	p-Value
e) Minimal						0.176[b]
(n=195)	≤18.6	67.7 (22)	64.5 (49)	71.4 (23)	1.4 (1.1)	0.206[c]
(R^2=0.703)	>18.6	65.9 (24)	64.8 (47)	63.7 (30)	-0.7 (1.1)	0.531[c]
f) Maximal						0.063[b]
(n=270)	≤18.6	65.8 (38)	65.0 (70)	69.7 (31)	1.3 (0.7)	0.077[c]
(R^2=0.714)	>18.6	67.9 (23)	64.7 (65)	64.3 (43)	-0.7 (0.7)	0.355[c]

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[a]	p-Value	Covariate Remarks
g) Minimal						0.078**[b]	CURR*TIME*RACE
(n=194)	≤18.6	65.6** (22)	64.3** (49)	73.2** (23)	2.6 (1.1)**	0.025**[c]	(p=0.033)
(R^2=0.758)							AGE (p=0.052)
	>18.6	63.9** (24)	63.9** (46)	64.4** (30)	-0.1 (1.1)**	0.942**[c]	DRKYR (p=0.018)
h) Maximal						****	CURR*TIME*RACE
(n=269)	≤18.6	**** (38)	**** (70)	**** (31)	****	****	(p=0.009)
(R^2=0.756)							AGE (p=0.080)
	>18.6	**** (23)	**** (64)	**** (43)	****	****	PACKYR (p=0.104)
							DRKYR (p=0.009)

[a]Slope and standard error based on NKCI 50/1 percent release versus log_2 dioxin.

[b]Test of significance for current dioxin-by-time interaction (current dioxin and time continuous).

[c]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

**Log_2 (current dioxin)-by-time-by-covariate interaction ($0.01<p\leq0.05$); adjusted mean, adjusted slope, standard error, and p-value derived from a model fitted after deletion of interaction.

****Log_2 (current dioxin)-by-time-by-covariate interaction ($p\leq0.01$); adjusted mean, adjusted slope, standard error, and p-value not presented.

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-21. (Continued)

Analysis of NKCI 50/1 Ratio Percent Release

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean	Contrast	Difference of Means (95% C.I.)	p-Value
Background	298	66.6	All Categories		0.821
Unknown	123	65.9	Unknown vs. Background	-0.7 (-2.8,1.3)	0.488
Low	72	66.8	Low vs. Background	0.2 (-2.3,2.7)	0.867
High	74	65.7	High vs. Background	-0.9 (-3.4,1.7)	0.500
Total	567		(R^2=0.499)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean	Contrast	Difference of Adj. Means (95% C.I.)	p-Value	Covariate Remarks
Background	298	61.9**	All Categories		0.845**	DXCAT*RACE (p=0.013)
Unknown	123	61.5**	Unknown vs. Background	-0.4 (-2.4,1.6)**	0.679**	DXCAT*ALC (p=0.021)
Low	71	62.3**	Low vs. Background	0.4 (-2.1,2.8)**	0.758**	DXCAT*DRKYR (p=0.027)
High	74	61.1**	High vs. Background	-0.9 (-3.3,1.6)**	0.488**	RACE*PACKYR (p<0.001)
Total	566		(R^2=0.575)			CSMOK*PACKYR (p=0.023)

**Categorized current dioxin-by-covariate interaction (0.01<p≤0.05); adjusted mean, confidence interval, and p-value derived from a model fitted after deletion of this interaction.

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

non-Blacks (Appendix Table O-1). For Blacks, the interaction and time strata associations were reported and were based on sparse numbers. For the non-Blacks, the interaction of current dioxin and time was significant (p=0.034). For non-Blacks with time of 18.6 years or less, there was a significant positive association between NKCI 50/1 percent release and current dioxin (p=0.013) and a nonsignificant negative association for time over 18.6 years (p=0.733). A followup adjusted model without the current dioxin-by-time-by-race interaction displayed a marginally significant interaction between current dioxin and time (Table 16-21 [g]: p=0.078). For time of 18.6 years or less, there was a significant positive association (p=0.025) with current dioxin and a nonsignificant negative association (p=0.942) for time over 18.6 years. For the former time stratum, the average NKCI 50/1 percent releases for low, medium, and high current dioxin were 65.6, 64.3, and 73.2 percent.

Under the maximal assumption, the adjusted analysis also contained a significant interaction for current dioxin, time, and race (Table 16-21 [h]: p=0.009). To explore this interaction, separate analyses were again performed for Blacks and non-Blacks (Appendix Table O-1). For Blacks, the interaction and time strata associations were reported but are based on sparse numbers. For the non-Blacks, the interaction of current dioxin and time was significant (p=0.010). For non-Blacks with time 18.6 years or less, there was a significant positive association between NKCI 50/1 percent release and current dioxin (p=0.007) and a nonsignificant negative association for time over 18.6 years (p=0.396).

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted analysis of NKCI 50/1 percent release indicated that current dioxin categories for Ranch Hands and Comparisons were not significantly different (Table 16-21 [i]: p=0.821).

The adjusted analysis of NKCI 50/1 percent release contained three significant interactions between a covariate and the current dioxin categories. The covariates involved in the interactions were race, current alcohol use, and lifetime alcohol history (Table 16-21 [j]: p=0.013, p=0.021, and p=0.027, respectively). To investigate these interactions, current alcohol use was dichotomized into one drink or less per day and over one drink per day and lifetime alcohol history was dichotomized as 40 drink-years or less and over 40 drink-years. Because of sparse numbers for Blacks, the interactions were explored only for non-Blacks.

Regardless of their lifetime alcohol history stratum, non-Blacks who had one drink or less per day displayed nonsignificant overall contrasts for NKCI 50/1 percent release (Appendix Table O-1). For non-Blacks who had more than one drink per day and had lifetime alcohol history of 40 drink-years or less, the overall contrast of current dioxin categories was nonsignificant but the low versus background contrast was marginally significant (p=0.067). For that contrast, Ranch Hands exceeded the Comparisons on the average NKCI 50/1 percent release. For non-Blacks who had more than one drink per day and also had a lifetime alcohol history over 40 drink-years, the overall contrast of current dioxin categories was nonsignificant. The low versus background contrast was significant (p=0.033) with the Ranch Hands having the higher means for NKCI 50/1 percent release.

A followup adjusted model without the three interactions was also used to examine the NKCI 50/1 ratio percent release values among categories of Ranch Hands and Comparisons.

The overall contrast of the four current dioxin categories was nonsignificant (Table 16-21 [j]: p=0.845). Individual contrasts were also nonsignificant.

Laboratory Examination Variables: Quantitative Studies—Quantitative Immunoglobulins

IgA

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

The unadjusted analysis under the minimal assumption exhibited a nonsignificant association between IgA and initial dioxin (Table 16-22 [a]: p=0.109). Under the maximal assumption, there was a significant positive association (Table 16-22 [b]: p=0.009). The unadjusted means for IgA were 195.7, 213.1, and 213.7 mg/dl, for the low, medium, and high initial dioxin categories.

In the adjusted analysis under the minimal assumption, there was a significant positive association between IgA and initial dioxin (Table 16-22 [c]: p=0.019). Age and race were covariates retained in the model. The adjusted IgA means for low, medium, and high initial dioxin were 219.0, 235.8, and 245.7 mg/dl.

Under the maximal assumption, the adjusted analysis also displayed a significant positive association (Table 16-22 [d]: p=0.003) with the adjusted IgA means for low, medium, and high initial dioxin at 213.5, 229.7, and 234.7 mg/dl.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

In the unadjusted analysis of IgA under the minimal assumption, the interaction of current dioxin and time since tour was not significant (Table 16-22 [e]: p=0.613); thus, the slopes describing the relationship between IgA and current dioxin did not differ significantly between time strata.

Under the maximal assumption, the unadjusted analysis also contained a nonsignificant interaction between current dioxin and time (Table 16-22 [f]: p=0.594). However, the slope for time greater than 18.6 years was positive and was of borderline significance (p=0.056).

Within that time strata, the mean levels of IgA were 190.4, 225.1, and 220.5 mg/dl for low, medium, and high current dioxin.

In the adjusted analysis under the minimal assumption, there was a significant interaction among current dioxin, time, and current cigarette smoking (Table 16-22 [g]: p=0.017). The interaction was investigated separately for Ranch Hands who never smoked, formerly smoked, smoked 20 cigarettes or less per day, and smoked over 20 cigarettes per day. For each of these smoking strata, the current dioxin-by-time interactions were not significant. For the first two smoking strata, there were nonsignificant positive associations for both times. For the other two smoking strata, there were nonsignificant positive associations between IgA and current dioxin for time at most 18.6 years and nonsignificant negative associations for time over 18.6 years. Without the above interaction in the model,

TABLE 16-22.

Analysis of IgA (mg/dl)

Ranch Hands - Log_2 (Initial Dioxin) - Unadjusted

Assumption	Initial Dioxin	n	Mean[a]	Slope (Std. Error)[b]	p-Value
a) Minimal (n=504) (R^2=0.005)	Low	127	203.7	0.027 (0.017)	0.109
	Medium	252	215.4		
	High	125	218.9		
b) Maximal (n=720) (R^2=0.009)	Low	180	195.7	0.031 (0.012)	0.009
	Medium	363	213.1		
	High	177	213.7		

Ranch Hands - Log_2 (Initial Dioxin) - Adjusted

Assumption	Initial Dioxin	n	Adj. Mean[a]	Adj. Slope (Std. Error)[b]	p-Value	Covariate Remarks
c) Minimal (n=504) (R^2=0.028)	Low	127	219.0	0.040 (0.017)	0.019	AGE (p=0.010)
	Medium	252	235.8			RACE (p=0.011)
	High	125	245.7			
d) Maximal (n=715) (R^2=0.040)	Low	179	213.5	0.035 (0.012)	0.003	RACE (p=0.007)
	Medium	360	229.7			ALC (p=0.071)
	High	176	234.7			AGE*PACKYR (p=0.015)

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on natural logarithm IgA versus log_2 dioxin.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.

TABLE 16-22. (Continued)

Analysis of IgA (mg/dl)

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean[a]/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[b]	p-Value
e) Minimal						0.613[c]
(n=504)	≤18.6	211.4	197.2	213.2	0.026 (0.027)	0.339[d]
(R^2=0.011)		(71)	(127)	(52)		
	>18.6	219.2	227.0	216.7	0.008 (0.022)	0.725[d]
		(54)	(127)	(73)		
f) Maximal						0.594[c]
(n=720)	≤18.6	202.9	199.1	210.7	0.018 (0.018)	0.317[d]
(R^2=0.012)		(104)	(189)	(80)		
	>18.6	190.4	225.1	220.5	0.031 (0.016)	0.056[d]
		(77)	(171)	(99)		

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean[a]/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[b]	p-Value	Covariate Remarks
g) Minimal						0.480**[c]	CURR*TIME*CSMOK
(n=504)	≤18.6	226.1**	216.1**	240.8**	0.046 (0.027)**	0.096**[d]	(p=0.017)
(R^2=0.045)		(71)	(127)	(52)			AGE (p=0.021)
	>18.6	232.5**	244.3**	237.5**	0.021 (0.023)**	0.352**[d]	RACE (p=0.016)
		(54)	(127)	(73)			
h) Maximal						0.688**[c]	CURR*TIME*CSMOK
(n=715)	≤18.6	219.5**	216.3**	233.5**	0.027 (0.018)**	0.147**[d]	(p=0.043)
(R^2=0.050)		(104)	(188)	(79)			RACE (p=0.010)
	>18.6	205.4**	242.3**	240.2**	0.036 (0.016)**	0.027**[d]	ALC (p=0.071)
		(76)	(169)	(99)			AGE*PACKYR (p=0.030)

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on natural logarithm IgA versus log_2 dioxin.

[c]Test of significance for current dioxin-by-time interaction (current dioxin continuous, time categorized).

[d]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

**Log_2 (initial dioxin)-by-covariate interaction ($0.01<p\leq0.05$); adjusted mean, adjusted slope, standard error, and p-value derived from a model fitted after deletion of this interaction.

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-22. (Continued)

Analysis of IgA (mg/dl)

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean[a]	Contrast	Difference of Means (95% C.I.)[e]	p-Value[f]
Background	759	212.1	All Categories		0.013
Unknown	338	195.1	Unknown vs. Background	-17.0 --	0.003
Low	192	210.0	Low vs. Background	-2.1 --	0.769
High	179	216.1	High vs. Background	4.0 --	0.604
Total	1,468		(R^2=0.007)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean[a]	Contrast	Difference of Adj. Means (95% C.I.)[e]	p-Value[f]	Covariate Remarks
Background	759	219.4	All Categories		0.007	AGE (p=0.001) RACE (p=0.097)
Unknown	336	202.1	Unknown vs. Background	-17.3 --	0.003	CSMOK (p=0.066)
Low	190	219.3	Low vs. Background	-0.1 --	0.982	ALC (p=0.137)
High	178	227.8	High vs. Background	8.4 --	0.292	
Total	1,463		(R^2=0.021)			

[a]Transformed from natural logarithm scale.

[e]Difference of means after transformation to original scale; confidence interval on difference of means not given because analysis was performed on natural logarithm scale.

[f]P-value is based on difference of means on natural logarithm scale.

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

the interaction of current dioxin and time was not significant (Table 16-22 [g]: p=0.480). For time less than or equal to 18.6 years, there was a positive association that was marginally significant (p=0.096).

In the adjusted analysis under the maximal assumption, there was also a significant interaction for current dioxin, time, and current cigarette smoking (Table 16-22 [h]: p=0.043). Investigation of the interaction was again undertaken separately for the current cigarette smoking strata. For former smokers, the current dioxin-by-time interaction was nonsignificant. However, for time greater than 18.6 years, there was a significant positive association between IgA and current dioxin (Appendix Table O-1: p=0.001) and a nonsignificant positive association for time 18.6 years or less (p=0.387). The other smoking strata displayed nonsignificant current dioxin-by-time interactions. An adjusted model without the cited interaction term contained a nonsignificant interaction between current dioxin and time (Table 16-22 [h]: p=0.688). For time over 18.6 years, there was a positive association between IgA and current dioxin that was significant (p=0.027).

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted analysis of IgA indicated that the overall contrast of Ranch Hands and Comparisons was significant (Table 16-22 [i]: p=0.013). The IgA means for the background, unknown, low, and high current dioxin categories were 212.1, 195.1, 210.0, and 216.1 mg/dl. The contrast of Ranch Hands in the unknown current dioxin category versus Comparisons in the background current dioxin category was significant (p=0.003).

The adjusted analysis of IgA also exhibited a significant overall current dioxin category contrast (Table 16-22 [j]: p=0.007) and a significant contrast for Ranch Hands in the unknown current dioxin category versus the Comparisons in the background current dioxin category (p=0.003).

IgG

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

Under the minimal and maximal assumptions, the unadjusted analysis was not significant for an association between IgG and initial dioxin (Table 16-23 [a] and [b]: p=0.720 and p=0.195, respectively).

Under the minimal cohort, the adjusted analysis contained a significant interaction between initial dioxin and lifetime alcohol history (Table 16-23 [c]: p=0.037). Exploring the interaction by stratification (0 drink-years, over 0 drink-years to 40 drink-years, and above 40 drink-years), the association between IgG and initial dioxin was positive but not significant for each stratum (Appendix Table O-1). Without the interaction of initial dioxin and lifetime alcohol history in the model, the association between IgG and initial dioxin was not significant (Table 16-23 [c]: p=0.502).

Under the maximal assumption, the adjusted analysis contained a nonsignificant association between IgG and initial dioxin (Table 16-23 [d]: p=0.156).

TABLE 16-23.

Analysis of IgG (mg/dl)

Ranch Hands - Log_2 (Initial) - Unadjusted

Assumption	Initial Dioxin	n	Mean[a]	Slope (Std. Error)[b]	p-Value
a) Minimal (n=504) (R^2<0.001)	Low	127	1,018.0	0.003 (0.009)	0.720
	Medium	252	1,013.3		
	High	125	1,034.2		
b) Maximal (n=720) (R^2=0.002)	Low	180	990.2	0.008 (0.006)	0.195
	Medium	363	1,020.2		
	High	177	1,013.0		

Ranch Hands - Log_2 (Initial) - Adjusted

Assumption	Initial Dioxin	n	Adj. Mean[a]	Adj. Slope (Std. Error)[b]	p-Value	Covariate Remarks
c) Minimal (n=498) (R^2=0.147)	Low	127	1,096.0**	0.006 (0.009)**	0.502**	INIT*DRKYR (p=0.037)
	Medium	248	1,118.2**			RACE (p<0.001)
	High	123	1,132.2**			AGE*ALC (p=0.029)
						CSMOK*PACKYR (p=0.012)
						PACKYR*DRKYR (p<0.001)
						ALC*DRKYR (p<0.001)
d) Maximal (n=711) (R^2=0.143)	Low	178	1,103.5	0.008 (0.006)	0.156	RACE (p<0.001)
	Medium	360	1,128.0			AGE*ALC (p=0.019)
	High	173	1,117.7			CSMOK*PACKYR (p=0.007)
						CSMOK*DRKYR (p=0.009)
						PACKYR*DRKYR (p<0.001)
						ALC*DRKYR (p<0.001)

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on natural logarithm IgG versus log_2 dioxin.

**Log_2 (initial dioxin)-by-covariate interaction ($0.01<p\leq0.05$); adjusted mean, adjusted slope, standard error, and p-value derived from a model fitted after deletion of this interaction.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.

TABLE 16-23. (Continued)

Analysis of IgG (mg/dl)

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean[a]/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[b]	p-Value
e) Minimal						0.815[c]
(n=504)	≤18.6	1,041.8	1,005.5	1,037.0	0.007 (0.014)	0.612[d]
(R^2<0.001)		(71)	(127)	(52)		
	>18.6	994.4	1,023.4	1,023.0	0.003 (0.012)	0.810[d]
		(54)	(127)	(73)		
f) Maximal						0.938[c]
(n=720)	≤18.6	997.4	1,021.0	1,028.7	0.009 (0.009)	0.311[d]
(R^2=0.004)		(104)	(189)	(80)		
	>18.6	962.6	1,020.3	1,013.7	0.010 (0.008)	0.212[d]
		(77)	(171)	(99)		

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean[a]/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[b]	p-Value	Covariate Remarks
g) Minimal						0.429[c]	RACE (p<0.001)
(n=498)	≤18.6	1,116.9	1,105.7	1,132.6	0.014 (0.013)	0.290[d]	AGE*ALC (p=0.016)
(R^2=0.141)		(71)	(126)	(51)			CSMOK*PACKYR
	>18.6	1,084.1	1,125.9	1,115.9	0.0007 (0.012)	0.952[d]	(p=0.018)
		(54)	(124)	(72)			PACKYR*DRKYR
							(p<0.001)
							ALC*DRKYR
							(p=0.002)
h) Maximal						0.811[c]	RACE (p<0.001)
(n=711)	≤18.6	1,104.2	1,122.6	1,144.4	0.011 (0.009)	0.208[d]	AGE*ALC (p=0.019)
(R^2=0.144)		(103)	(188)	(78)			CSMOK*PACKYR
	>18.6	1,085.9	1,133.4	1,120.0	0.009 (0.008)	0.301[d]	(p=0.007)
		(76)	(169)	(97)			CSMOK*DRKYR
							(p=0.010)
							PACKYR*DRKYR
							(p<0.001)
							ALC*DRKYR
							(p<0.001)

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on natural logarithm IgG versus log_2 dioxin.

[c]Test of significance for current dioxin-by-time interaction (current dioxin and time continuous).

[d]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-23. (Continued)

Analysis of IgG (mg/dl)

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean[a]	Contrast	Difference of Means (95% C.I.)[e]	p-Value[f]
Background	759	1,024.2	All Categories		0.058
Unknown	338	986.5	Unknown vs. Background	-37.7 --	0.008
Low	192	1,021.3	Low vs. Background	-2.9 --	0.874
High	179	1,020.4	High vs. Background	-3.8 --	0.837
Total	1,468		(R^2=0.005)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean[a]	Contrast	Difference of Adj. Means (95% C.I.)[e]	p-Value[f]	Covariate Remarks
Background	757	1,120.3	All Categories		0.132	RACE (p<0.001) DRKYR (p=0.103)
Unknown	335	1,087.1	Unknown vs. Background	-33.2 --	0.028	CSMOK*PACKYR
Low	190	1,122.4	Low vs. Background	2.1 --	0.915	(p=0.002)
High	175	1,122.1	High vs. Background	1.8 --	0.927	
Total	1,457		(R^2=0.082)			

[a]Transformed from natural logarithm scale.

[e]Difference of means after transformation to original scale; confidence interval on difference of means not given because analysis was performed on natural logarithm scale.

[f]P-value is based on difference of means on natural logarithm scale.

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

In the unadjusted analysis of IgG under the minimal and maximal assumptions, the interaction of current dioxin and time since tour was not significant (Table 16-23 [e] and [f]: p=0.815 and p=0.938, respectively). The nonsignificant interactions indicated that the slopes did not differ significantly between time strata.

The nonsignificant results of the unadjusted analyses remained nonsignificant in the adjusted analyses (Table 16-23 [g] and [h]: p=0.429 and p=0.811, respectively) under the minimal and maximal assumptions.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted analysis of IgG displayed a marginally significant overall contrast of Ranch Hand and Comparison current dioxin categories (Table 16-23 [i]: p=0.058). The IgG means for the background, unknown, low, and high current dioxin categories were 1,024.2 mg/dl, 986.5 mg/dl, 1,021.3 mg/dl, and 1,020.4 mg/dl. The contrast of Ranch Hands in the unknown current dioxin category versus Comparisons in the background current dioxin category was significant (p=0.008).

The adjusted analysis of IgG exhibited a nonsignificant overall category contrast (Table 16-23 [j]: p=0.132). However, a significant contrast for Ranch Hands in the unknown current dioxin category versus the Comparisons in the background current dioxin category (p=0.028) was found with the Comparisons having the higher adjusted IgG mean.

IgM

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

Under both the minimal and maximal assumptions, the unadjusted analysis of IgM displayed a nonsignificant association with initial dioxin (Table 16-24 [a] and [b]: p=0.425 and p=0.471, respectively).

Under the minimal assumption, the adjusted analysis contained a significant interaction between initial dioxin and current alcohol use (Table 16-24 [c]: p=0.029). Exploring the interaction within drinking stratum, there was a nonsignificant positive association (Appendix Table O-1: p=0.183) for Ranch Hands who had zero to one drink per day. For Ranch Hands who had more than one drink per day to four drinks per day, a significant negative association was found between IgM and initial dioxin (p=0.024). For Ranch Hands with more than four drinks per day, a nonsignificant negative association was found (p=0.210). Without the interaction of initial dioxin and current alcohol use in the model, the adjusted model exhibited a nonsignificant association between IgM and initial dioxin (Table 16-24 [c]: p=0.902).

The adjusted analysis under the maximal assumption displayed a nonsignificant association between IgM and initial dioxin (Table 16-24 [d]: p=0.268).

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

In the unadjusted analysis under both minimal and maximal assumptions, the interaction of current dioxin and time since tour was not significant (Table 16-24 [e] and [f]: p=0.725 and p=0.174, respectively). Thus, the association between IgM and current dioxin (i.e., the

TABLE 16-24.

Analysis of IgM (mg/dl)

Ranch Hands - Log_2 (Initial Dioxin) - Unadjusted

Assumption	Initial Dioxin	n	Mean[a]	Slope (Std. Error)[b]	p-Value
a) Minimal (n=504) (R^2=0.001)	Low	127	113.7	0.014 (0.017)	0.425
	Medium	252	106.8		
	High	125	115.2		
b) Maximal (n=720) (R^2<0.001)	Low	180	117.3	-0.009 (0.013)	0.471
	Medium	363	109.6		
	High	177	115.4		

Ranch Hands - Log_2 (Initial Dioxin) - Adjusted

Assumption	Initial Dioxin	n	Adj. Mean[a]	Adj. Slope (Std. Error)[b]	p-Value	Covariate Remarks
c) Minimal (n=501) (R^2=0.024)	Low	127	107.5**	0.002 (0.018)**	0.902**	INIT*ALC (p=0.029)
	Medium	250	99.5**			AGE (p=0.100)
	High	124	104.9**			RACE (p=0.049)
d) Maximal (n=711) (R^2=0.026)	Low	178	108.0	-0.015 (0.013)	0.268	AGE (p=0.048)
	Medium	360	102.2			ALC (p=0.032)
	High	173	105.5			DRKYR*RACE (p=0.040)

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on natural logarithm IgM versus log_2 dioxin.

**Log_2 (initial dioxin)-by-covariate interaction ($0.01<p\leq0.05$); adjusted mean, adjusted slope, standard error, and p-value derived from a model fitted after deletion of this interaction.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.

TABLE 16-24. (Continued)

Analysis of IgM (mg/dl)

Ranch Hands - Log_2 (Current Dioxin) and Time - Unadjusted

Assumption	Time (Yrs.)	Mean[a]/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[b]	p-Value
e) Minimal						0.725[c]
(n=504)	≤18.6	121.0	101.0	109.5	0.0006 (0.028)	0.983[d]
(R^2=0.003)		(71)	(127)	(52)		
	>18.6	112.0	111.9	115.7	0.013 (0.023)	0.567[d]
		(54)	(127)	(73)		
f) Maximal						0.174[c]
(n=720)	≤18.6	125.4	105.8	111.4	-0.033 (0.020)	0.099[d]
(R^2=0.004)		(104)	(189)	(80)		
	>18.6	111.6	112.3	117.9	0.004 (0.018)	0.844[d]
		(77)	(171)	(99)		

Ranch Hands - Log_2 (Current Dioxin) and Time - Adjusted

Assumption	Time (Yrs.)	Adj. Mean[a]/(n) Current Dioxin Low	Medium	High	Adj. Slope (Std. Error)[b]	p-Value	Covariate Remarks
g) Minimal						0.519[c]	AGE (p=0.029)
(n=498)	≤18.6	112.7	90.9	94.3	-0.022 (0.029)	0.456[d]	RACE*DRKYR
(R^2=0.031)		(71)	(126)	(51)			(p=0.021)
	>18.6	104.4	103.5	104.7	0.002 (0.024)	0.934[d]	
		(54)	(124)	(72)			
h) Maximal						0.137[c]	AGE (p=0.021)
(n=711)	≤18.6	114.8	97.3	99.7	-0.045 (0.020)	0.027[d]	ALC (p=0.033)
(R^2=0.032)		(103)	(188)	(78)			RACE*DRKYR
	>18.6	104.4	105.0	108.5	-0.005 (0.018)	0.776[d]	(p=0.034)
		(76)	(169)	(97)			

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on natural logarithm IgM versus log_2 dioxin.

[c]Test of significance for current dioxin-by-time interaction (current dioxin and time continuous).

[d]Test of significance for slope different from 0 (current dioxin continuous, time categorized).

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.

TABLE 16-24. (Continued)

Analysis of IgM (mg/dl)

i) Ranch Hands and Comparisons by Current Dioxin Category - Unadjusted

Current Dioxin Category	n	Mean[a]	Contrast	Difference of Means (95% C.I.)[e]	p-Value[f]
Background	759	110.3	All Categories		0.079
Unknown	338	113.9	Unknown vs. Background	3.6 --	0.301
Low	192	103.1	Low vs. Background	-7.2 --	0.076
High	179	115.0	High vs. Background	4.7 --	0.294
Total	1,468		(R^2=0.005)		

j) Ranch Hands and Comparisons by Current Dioxin Category - Adjusted

Current Dioxin Category	n	Adj. Mean[a]	Contrast	Difference of Adj. Means (95% C.I.)[e]	p-Value[f]	Covariate Remarks
Background	757	103.0	All Categories		0.099	AGE (p=0.005)
						RACE (p=0.004)
Unknown	335	106.6	Unknown vs. Background	3.6 --	0.266	ALC (p=0.005)
Low	190	96.3	Low vs. Background	-6.7 --	0.078	DRKYR (p=0.104)
High	175	105.9	High vs. Background	2.9 --	0.477	
Total	1,457		(R^2=0.021)			

[a]Transformed from natural logarithm scale.

[e]Difference of means after transformation to original scale; confidence interval on difference of means not given because analysis was performed on natural logarithm scale.

[f]P-value is based on difference of means on natural logarithm scale.

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.

slopes) did not differ significantly between time strata. Under the maximal assumption, a negative association between IgM and current dioxin was marginally significant (p=0.099) for time of 18.6 years or less.

In the adjusted analysis under the minimal assumption, the current dioxin-by-time interaction was nonsignificant (Table 16-24 [g]: p=0.519). Thus, the relationships between IgM and current dioxin were not significant between time strata.

In the adjusted analysis under the maximal assumption, the interaction of current dioxin and time was also not significant (Table 16-24 [h]: p=0.137). For time less than or equal to 18.6 years, there was a negative association between IgM and current dioxin that was significant (p=0.027).

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted analysis of IgM indicated that the overall contrast of current dioxin categories for Ranch Hands and Comparisons was marginally significant (Table 16-24 [i]: p=0.079). The IgM means for the background, unknown, low, and high current dioxin categories were 110.3, 113.9, 103.1, and 115.0 mg/dl. The contrast of Ranch Hands in the low current dioxin category versus Comparisons in the background current dioxin category was marginally significant (p=0.076).

Similar to the unadjusted analysis of IgM, the adjusted analysis indicated that the overall contrast of Ranch Hands and Comparisons was marginally significant (Table 16-24 [j]: p=0.099). The contrast of Ranch Hands in the low current dioxin category versus Comparisons in the background current dioxin category was also marginally significant (p=0.078).

Longitudinal Analysis

Laboratory Examination Data

CD4/CD8 Ratio

For the immunologic evaluation, longitudinal analyses were conducted to examine the change in the CD4/CD8 ratio between the 1985 and the 1987 examinations for associations with initial dioxin, current dioxin and time since tour, and categorized current dioxin. Table 16-25 presents the results of these analyses. For a specific longitudinal analysis of the CD4/CD8 ratio (e.g., minimal assumption, initial dioxin analysis), the left side of each subpanel of the table provides the means and sample sizes for participants with laboratory values at each examination. Based on the difference between 1987 and 1985 laboratory values, the right side of each subpanel presents slopes, standard errors, and associated p-values (for models using initial dioxin or models using current dioxin and time since tour), or differences of examination mean changes, 95 percent confidence intervals, and associated p-values (for models using categorized current dioxin). The reported statistics for all three examinations are presented subject to the constraint that participants were at both the 1985 and 1987 examinations.

TABLE 16-25.

Longitudinal Analysis of CD4/CD8 Ratio

Ranch Hands - Log_2 (Initial Dioxin)

Assumption	Initial Dioxin	Mean[a]/(n) Examination 1985	Mean[a]/(n) Examination 1987	Slope (Std. Error)[b]	p-Value
a) Minimal (R^2<0.001)	Low	1.70 (39)	1.98 (39)	0.001 (0.017)	0.968
	Medium	1.66 (88)	2.06 (88)		
	High	1.63 (47)	1.89 (47)		
b) Maximal (R^2=0.006)	Low	1.60 (51)	1.82 (51)	0.014 (0.012)	0.234
	Medium	1.62 (121)	1.97 (121)		
	High	1.67 (64)	1.94 (64)		

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on difference between natural logarithm of 1987 CD4/CD8 ratio and natural logarithm of 1985 CD4/CD8 ratio versus log_2 dioxin.

Note: Minimal--Low: 52-93 ppt; Medium: >93-292 ppt; High: >292 ppt.
Maximal--Low: 25-56.9 ppt; Medium: >56.9-218 ppt; High: >218 ppt.
Summary statistics for 1985 are provided for reference purposes for participants who attended the Baseline, 1985, and 1987 examinations. P-values given are in reference to a contrast of 1985 and 1987 results.

TABLE 16-25. (Continued)

Longitudinal Analysis of CD4/CD8 Ratio

Ranch Hands - Log_2 (Current Dioxin) and Time

Assumption	Time (Yrs.)	Examination	Mean[a]/(n) Current Dioxin Low	Medium	High	Slope (Std. Error)[b]	p-Value
c) Minimal							0.062[c]
(R^2=0.022)	≤18.6	1985	1.75 (20)	1.57 (43)	1.60 (20)	-0.031 (0.026)	0.240[d]
		1987	2.05 (20)	2.09 (43)	1.66 (20)		
	>18.6	1985	1.69 (21)	1.73 (44)	1.63 (26)	0.033 (0.022)	0.131[d]
		1987	1.87 (21)	2.10 (44)	2.02 (26)		
d) Maximal							0.190[c]
(R^2=0.020)	≤18.6	1985	1.70 (30)	1.59 (60)	1.65 (27)	0.003 (0.018)	0.879[d]
		1987	1.93 (30)	2.01 (60)	1.84 (27)		
	>18.6	1985	1.40 (22)	1.69 (59)	1.66 (38)	0.035 (0.016)	0.035[d]
		1987	1.51 (22)	1.97 (59)	2.06 (38)		

[a]Transformed from natural logarithm scale.

[b]Slope and standard error based on difference between natural logarithm of 1987 CD4/CD8 ratio and natural logarithm of 1985 CD4/CD8 ratio versus log_2 dioxin.

[c]Test of significance for homogeneity of slopes (current dioxin continuous, time categorized).

[d]Test of significance for slope equal to 0 (current dioxin continuous, time categorized).

Note: Minimal--Low: >10-14.65 ppt; Medium: >14.65-45.75 ppt; High: >45.75 ppt.
Maximal--Low: >5-9.01 ppt; Medium: >9.01-33.3 ppt; High: >33.3 ppt.
Summary statistics for 1985 are provided for reference purposes for participants who attended the Baseline, 1985, and 1987 examinations. P-values given are in reference to a contrast of 1985 and 1987 results.

TABLE 16-25. (Continued)

Longitudinal Analysis of CD4/CD8 Ratio

e) Ranch Hands and Comparisons by Current Dioxin Category

Current Dioxin Category	Mean[a]/(n) Examination 1985	1987	Contrast	Difference of Examination Mean Change (95% C.I.)[e]	p-Value[f]
Background	1.62 (270)	1.93 (270)	All Categories		0.404
Unknown	1.54 (104)	1.80 (104)	Unknown vs. Background	-0.04 - -	0.637
Low	1.58 (65)	2.00 (65)	Low vs. Background	0.10 - -	0.141
High	1.66 (65)	1.96 (65)	High vs. Background	-0.001 - -	0.918
	(R^2=0.006)				

[a]Transformed from natural logarithm scale.

[e]Difference of 1987 and 1985 examination mean changes after transformation to original scale; confidence interval on difference of 1987 and 1985 examination mean changes not given because analysis was performed on natural logarithm scale.

[f]P-value is based on difference of 1987 and 1985 examination mean changes on natural logarithm scale.

Note: Background (Comparisons): Current Dioxin ≤10 ppt.
Unknown (Ranch Hands): Current Dioxin ≤10 ppt.
Low (Ranch Hands): 15 ppt < Current Dioxin ≤33.3 ppt.
High (Ranch Hands): Current Dioxin >33.3 ppt.
Summary statistics for 1985 are provided for reference purposes for participants who attended the Baseline, 1985, and 1987 examinations. P-values given are in reference to a contrast of 1985 and 1987 results.

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

For both the minimal and the maximal longitudinal analyses, the association between initial dioxin and the change in the CD4/CD8 ratio of the 1987 examination value relative to the 1985 examination value was nonsignificant (Table 16-25 [a] and [b]: p=0.968 and p=0.234, respectively).

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

Under the minimal assumption, the longitudinal analysis detected a marginally significant interaction between current dioxin and time since tour (Table 16-25 [c]: p=0.062). For Ranch Hands with less than or equal to 18.6 years since their tour, there was a nonsignificant decreasing association between current dioxin and the change in the CD4/CD8 ratio between 1985 and 1987 (p=0.240). In contrast, for Ranch Hands with more than 18.6 years since tour, there was a nonsignificant increasing association between current dioxin and the change in the CD4/CD8 ratio between 1985 and 1987 (p=0.131).

Based on the maximal assumption, the longitudinal analysis did not detect a significant interaction between current dioxin and time since tour (Table 16-25 [d]: p=0.190). However, for Ranch Hands with more than 18.6 years since their tour, there was a significant increasing association between current dioxin and the change in the CD4/CD8 ratio between the 1985 and 1987 examinations (p=0.035). The differences of the mean CD4/CD8 ratios from 1985 to 1987 were 0.11, 0.28, and 0.40 for low, medium, and high current dioxin. Thus, for this time stratum, the increases in the mean CD4/CD8 ratio in 1987, relative to 1985, were greater for higher current dioxin levels.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

For the longitudinal analysis of categorized current dioxin, there was no significant difference in the change in the CD4/CD8 ratio (1987 relative to 1985) among the four current dioxin categories (Table 16-25 [e]: p=0.404).

DISCUSSION

Background

Immunologic competence was assessed by analysis of data from cell surface marker studies, immunoglobulin quantitation, functional stimulation assays, and skin tests for delayed hypersensitivity response on a randomized subset of the study population. The absence of a response to a series of skin test antigens is usually indicative of an impaired immune defense mechanism (anergy). Anergy can occur in elderly individuals in the setting of certain viral, bacterial, and fungal infections; or with advanced protein deficiency, underlying malignancy, or treatment with corticosteriods and other immunosuppressive agents. Skin tests for delayed cutaneous hypersensitivity (DCH) are occasionally used to test for anergy as a prognostic indicator in individuals in compromised states such as the acquired immunodeficiency syndrome or those at risk of infection following surgery.

Skin tests for DCH are subject to numerous variables including the dose and method of administration of the antigen and the techniques employed in reading and interpreting the response. Following quality control concerns over the 1985 Air Force Health Study skin test

data, stringent protocols were established to ensure consistent methods and interpretation. In the current study, a premium was placed on uniform and consistent methods of interpretation. There was a 92 percent concordance between readers and duplicate interpretations by the same reader. More than 99.6 percent of the sample population had interpretable skin tests. The 94.9 percent incidence of intact DCH is consistent with clinical experience in the general population (5).

Evaluation of the human immune system is divided into separate segments for humoral and cellular immunity and each of these is further divided into measurements of quality (e.g., cell counts and protein concentrations) and function. Circulating in the plasma phase of blood, the humoral segment consists of the immunoglobulin and complement proteins some of which are also prominent at exposed sites of the body such as mucosal surfaces. The serum immunoglobulins are secreted by plasma cells in the bone marrow and are regulated in a sequence of events modulated by macrophages and memory lymphocytes. The immunoglobulins serve as a defense against bacterial infections and the blood-borne phase of viral infections.

Quantitative analysis of IgG, IgM, and IgA give an overall view of B-cell integrity when related to the expected reference range of values. Selective deficiency of one or more of these antibody classes, whether congenital or acquired, may be associated with increased susceptibility to infections. Elevations of these immunoglobulins in a polyclonal pattern are frequently an indication of chronic infections (perhaps due to impairment of another segment of the immune response) or of a faulty regulation of B-cell responses such as occurs in cirrhosis. Selective elevation of a monoclonal segment of any immunoglobulin (detected by visual examination of serum protein electrophoresis) is a strong indicator of faulty regulation or actual autonomy of plasma cells or lymphocytes and may be an early hallmark of numerous conditions including plasmacytoma, multiple myeloma, chronic lymphocytic leukemia/lymphoma, and smoldering myeloma. Occasionally there may be a cluster of more than one small spike of immunoglobulin in the presence of other normal immunoglobulins. Invariably, this type of oligoclonal banding is associated with some alteration of the immune system (e.g., primary bone marrow involvement, inappropriate regulation, or immunosuppression as in organ transplant recipients). Thus, both quantitative and qualitative parameters of the serum immunoglobulins can give information on the integrity of B-cell responses.

Cellular immunity consists of both granulocytic and lymphocytic processes. Abnormalities of granulocytes can frequently be discerned from examination of the peripheral blood smear as part of the complete blood count. In addition, the infectious history of individuals is usually sufficient to ascertain whether granulocytic deficiency is a consideration. Chapter 13, Hematologic Assessment, discusses the effect of dioxin on the components of these cells.

The lymphocytic segment of the immune response can be broadly evaluated by skin testing against multiple fungal, bacterial, or viral agents. The response to skin tests is dependent in part on the infection exposure history of the patient, and so is probably better used in the diagnosis of specific diseases than in an overall examination of lymphocyte function, although it does have the particular merit of demonstrating the presence or absence of the response in vivo where it must be effective for the patient to remain healthy.

The in vitro functional stimulation tests of PHA and MLC are very valuable for showing explicitly whether there is an impairment of lymphocyte function in response to mitogenic and mixed cellular antigens. Although these assays are typically used in the diagnosis of congenital deficiencies of lymphocyte function (PHA) or for the cross-matching of organ donors and recipients (MLC), they may also demonstrate abnormalities in the cellular immune response associated with a variety of different conditions. Infection with cytomegalovirus, for example, can cause a reduction in the PHA response that reverts to normal in the convalescent phase. In addition, malnutrition, some autoimmune diseases, advanced age, physical stress or trauma, and advanced malignancy can be marked by low PHA response. The assay for natural killer cells with and without IL-2 is useful in evaluating the ability of peripheral blood lymphocytes to destroy dissimilar target cells. This destructive ability has been correlated with the potential of an individual to destroy tumor cells, invasive organisms, and other foreign antigens and presumably is an index of how well an individual's immune system would be in destroying developing tumors.

The total number of circulating lymphocytes provides information relative to the basic cellular quantity of cells that is present and available in the body for mounting an immune response. Examination of the surface marker proteins on the surfaces of these lymphocytes by flow cytometry is an excellent means of evaluating whether the regulatory interactions between T cells, B cells, and monocytes are intact. An alteration in the percentages of any of these categories can be considered presumptive evidence of an inability to recognize and destroy foreign infectious agents or tumor cells. The marker for total T cells was CD2 which is further broken down into the subpopulations of CD4 (helper cells) and CD8 (suppressor cells); CD4 and CD8 should be mutually exclusive. The ratio of CD4/CD8 describes whether the regulation is in balance. Expected values for the CD4/CD8 ratio are roughly 0.9 to 3.5. Ratios substantially below 1.0 are to be expected in patients immunosuppressed with cyclosporine and also those with active human immunodeficiency virus infection that involves primarily the CD4 positive cells. Activation of T cells results in the new synthesis of IL-2 receptor molecules on the surface of lymphocytes. This IL-2 receptor is also designated CD25, and its presence in excess is an indicator of recent stimulus to the immune system by virtually any type of antigen: infectious organism, transplanted organ, etc. The surface marker for B cells, CD20, gives an indication of the balance between cellular immunity and the ability to mount a B-cell response with production of specific antibodies. The CD14 marker is specific for monocytes that are essential for the correct transfer of stimulatory information from the (foreign) antigen processing segment to the antibody turn-on segment of a B-cell response.

Interpretation of alterations in the relative amounts of B cells, T cells and their subsets, and monocytes is based on the expectation that all aspects of the immune system must be intact to prevent infections and to guard against development of tumors with unusual surface antigens. The antibodies specific for tumors can either help to destroy them by binding complement and lysing the cells or stabilize them if those antibodies attach to the tumor surface without binding complement thereby blocking immune recognition and destruction of tumor cells. The T cells also have antigen receptors on their surfaces that similarly call into play the destructive power of the entire lymphocyte cell line in an antitumor attack. T cells stimulated by IL-2 have even greater capacity to attack and destroy foreign cells. Natural killer cells have still greater destructive capacity, but they act on a nonspecific basis and are probably simply recruited into regions of foreign antigens and tumors by the other recognition factors.

Clinical Interpretation of Statistically Significant Results

Immunoglobulins

The concentrations of IgA observed in this study increased with alcohol consumption (5) and are known to occur as an expected consequence of alcoholic cirrhosis, presumably due to diminished regulation of B-cell responses. These changes in cirrhosis lead to polyclonal gamopathy with IgA particularly being synthesized out of proportion to normal. Whether the significant associations between IgA and dioxin seen here are due to a combination of effects from alcohol, tobacco, and dioxin bears examination at other phases of the AFHS. A consistent trend would help to determine the clinical interpretation of these results. The increased IgA levels could represent a chronic inflammatory response to dioxin exposure, as do elevated erythrocyte sedimentation rates (in the general health assessment) and white blood cell and platelet counts (in the hematologic assessment). The analyses of the other immunoglobulins do not indicate the presence of any dioxin-related effects.

PHA Response

PHA (a plant lectin derived from the kidney bean) is a mitogen and as such induces proliferation or blast transformation of normal lymphocytes in cell culture. This response entails induction of new deoxyribonucleic acid (DNA) synthesis that is the basis of quantifying the PHA stimulation according to the incorporation of radioactive thymidine into high molecular weight DNA. The in vitro response to mitogens correlates well with the ability of the body to mount a delayed hypersensitivity response. Various congenital defects of cellular immunity may result in a spectrum of abnormalities in lymphocyte transformation from complete to partial lack of function resulting in increased susceptibility to infections from bacteria, fungi, and viruses. Furthermore, acquired impairment of lymphocyte transformation may be associated with the stress of surgery and anesthesia, aging, malnutrition, major burns, uremia, some malignancies, and other clinical conditions. The in vitro response to mitogens is also reduced in immunosuppression (e.g., cyclosporine) and increased in immunoenhancement (e.g., treatment with interleukins or interferon).

Because the response of lymphocytes from normal individuals to PHA can vary substantially over the concentration range of the mitogen employed, the PHA assays of this study were done at three different concentrations: PHA concentration 1, 36 µg/ml; PHA concentration 2, 12 µg/ml; PHA concentration 3, 4 µg/ml. One or more of these PHA concentrations will be expected to yield maximal response from normal lymphocytes, and the highest levels of PHA will be associated with some saturation and inhibition from the maximal response. In general, the cpm values were less for PHA concentration 1 (inhibition of response) than with PHA concentration 2 and PHA concentration 3, and values for PHA concentration 2 were generally equal to those for PHA concentration 3 (saturated response). However, none of these differences should be considered clinically different from one another. An acceptable range of variability in the lymphocyte response of a normal individual is from 65 percent to 180 percent of the mean value of a group of normal individuals.

The positive associations that were found to be statistically significant for maximum PHA net response in this study are not supportive of an impaired immune response. Variation in the assay is affected by long-term potency of the mitogen preparation and because of fluctuations in the stability and activity of the lymphocytes, depends on a patient's recent activities and minor medical conditions. Fortunately this degree of normal variability

does not pose a major problem for diagnosing alterations in the immune response because true deficiencies are associated with very low cpm values, perhaps 10,000 cpm or less.

Corroborative information regarding lymphocyte transformation is obtained from the MLC assay in which the foreign mitogen is replaced by antigens from allogeneic mixed lymphocytes inactivated by irradiation. Thus, any true deficiency in lymphocyte response observed in the PHA assay should also be present in the MLC assay. The actual data indicate a statistical difference in PHA data without corroboration from the MLC analysis.

In summary, the indices of immunologic capability analyzed in the current section provide a comprehensive reflection of in vivo and in vitro immune function in the study population. No clinically significant trends were observed relative to the current body burden of dioxin or the extrapolated initial exposure.

SUMMARY

For the immunologic assessment, Tables 16-26, 16-27, and 16-28 summarize the results from analyses based on initial dioxin, current dioxin and time since tour, and categorized current dioxin. All variables, except for the composite skin test diagnosis variable, were continuous in form. Table 16-29 summarizes the covariate interactions from the three models. Each of the interactions listed in Table 16-29 was reviewed along with the corresponding interaction displays in Appendix O for medical plausibility and consistency with the current literature. Based on this review, none of these interactions indicated dioxin-mediated immunosuppression in any subgroup of Ranch Hands.

Physical Examination Variable: Composite Skin Test Diagnosis

For the composite skin test diagnosis, the unadjusted and adjusted analysis of the relative frequency of participants with possibly abnormal tests exhibited nonsignificant associations with initial dioxin. The unadjusted and adjusted analyses of the composite skin test results were also nonsignificant for the models using current dioxin and time since tour. However, the adjusted analysis for the minimal cohort contained a significant current dioxin-by-time-by-age interaction which, when investigated, did not display significant associations with current dioxin for specified age and time since tour strata. For categorized current dioxin, the Ranch Hand and Comparison group contrasts were generally nonsignificant.

Laboratory Examination Variables: Cell Surface Marker (Phenotypic) Studies

The following cell surface marker variables were analyzed using a natural logarithm transformation: CD2 cells, CD4 cells, CD8 cells, CD20 cells, CD14 cells, CD25 cells, HLA-DR cells, and the CD4/CD8 ratio.

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

Except for the CD4/CD8 ratio, which exhibited a marginally significant positive association with initial dioxin under the maximal assumption, the unadjusted analyses of the cell surface marker variables were not significantly associated with initial dioxin (see Table 16-26). A negative association would be expected in the presence of immunodeficiency. For the adjusted analyses, the minimal cohort displayed a significant initial dioxin-by-covariate

TABLE 16-26.

Summary of Initial Dioxin Analyses for Immunology Variables Based on Minimal and Maximal Assumptions (Ranch Hands Only)

	Unadjusted		Adjusted	
Variable	Minimal	Maximal	Minimal	Maximal
Physical Examination				
Composite Skin Test Diagnosis (D)	ns	ns	ns	ns
Laboratory				
CD2 Cells (C)	ns	NS	****	NS
CD4 Cells (C)	NS	NS	NS	NS
CD8 Cells (C)	ns	ns	****	** (ns)
CD20 Cells (C)	NS	NS	** (NS*)	NS
CD14 Cells (C)	NS	ns	****	ns
CD25 Cells (C)	ns	NS	****	****
HLA-DR Cells (C)	ns	NS	****	** (NS)
CD4/CD8 Ratio (C)	NS	NS*	NS	NS*
TLC (C)	ns	NS	** (ns)	NS
Unstimulated PHA Response (C)	NS	NS	NS	NS
PHA Net Response - Conc. 1 (C)	NS	NS	****	** (ns)
PHA Net Response - Conc. 2 (C)	+0.016	+0.008	NS*	NS*
PHA Net Response - Conc. 3 (C)	NS	NS	NS	** (NS)
PHA Net Response - Across Day and Concentration	NS*	NS	** (NS)	--
Maximum PHA Net Response (C)	+0.005	+0.009	NS*	NS*
Unstimulated MLC Response (C)	NS	NS*	NS	NS
MLC Net Response (C)	NS	NS	NS	NS
NKCA 50/1 Net Response (C)	NS	ns	NS	ns
NKCA 50/1 Percent Release (C)	NS	ns	** (NS)	ns
NKCI 50/1 Net Response (C)	NS	NS	NS	NS
NKCI 50/1 Percent Release (C)	NS	NS	NS	NS
IgA (C)	NS	+0.009	+0.019	+0.003
IgG (C)	NS	NS	** (NS)	NS
IgM (C)	NS	ns	** (NS)	ns

C: Continuous analysis.
D: Discrete analysis.
+: Relative risk 1.00 or greater for discrete analysis.
--: Not applicable.
NS/ns: Not significant ($p>0.10$).
NS*: Marginally significant ($0.05<p\leq0.10$).
** (NS)/** ns: Log_2 (initial dioxin)-by-covariate interaction ($0.01<p\leq0.05$); not significant when interaction is deleted; refer to Appendix Table O-1 for a detailed description of this interaction.
** (NS*): Log_2 (initial dioxin)-by-covariate interaction ($0.01\leq0.05$); marginally significant when interaction is deleted; refer to Appendix Table O-1 for a detailed description of this interaction.
****: Log_2 (initial dioxin)-by-covariate interaction ($p\leq0.01$); refer to Appendix Table O-1 for a detailed description of this interaction.
Note: P-value given if $p\leq0.05$.
A capital "NS" denotes slope nonnegative for continuous analysis; a lowercase "ns" denotes relative risk less than 1.00 for discrete analysis or slope negative for continuous analysis.

TABLE 16-27.

Summary of Current Dioxin and Time Analyses for Immunology Variables Based on Minimal and Maximal Assumptions (Ranch Hands Only)

	Unadjusted					
	Minimal			Maximal		
Variable	C*T	≤18.6	>18.6	C*T	≤18.6	>18.6
Physical Examination						
Composite Skin Test Diagnosis (D)	NS	ns	NS	NS	ns	ns
Laboratory						
CD2 Cells (C)	NS	ns	ns	NS	NS	NS
CD4 Cells (C)	NS	ns	NS	NS	NS	NS
CD8 Cells (C)	ns	ns	ns	ns	ns	ns
CD20 Cells (C)	NS	NS	NS	NS	NS	NS*
CD14 Cells (C)	NS	ns	NS	NS	ns	NS
CD25 Cells (C)	NS*	- 0.028	NS	NS	ns	NS
HLA-DR Cells (C)	NS	ns	NS	NS	ns	NS
CD4/CD8 Ratio (C)	ns	NS	NS	ns	NS*	NS
TLC (C)	NS	ns	NS	NS	ns	NS
Unstimulated PHA Response (C)	ns	NS	NS	ns	NS	NS
PHA Net Response (C)	ns*	+ 0.006	NS	ns	NS	NS
Maximum PHA Net Response (C)	ns	+ 0.002	NS	NS	+ 0.049	+ 0.008
Unstimulated MLC Response (C)	ns	NS	NS	NS	NS	NS*
MLC Net Response (C)	ns	NS	ns	NS	ns	NS
NKCA 50/1 Net Response (C)	ns	NS	ns	ns	NS	ns
NKCA 50/1 Percent Release (C)	ns	NS	NS	ns	ns	ns
NKCI 50/1 Net Response (C)	ns	NS	ns	ns*	NS*	ns
NKCI 50/1 Percent Release (C)	ns	NS	ns	ns*	NS*	ns
IgA (C)	ns	NS	NS	NS	NS	NS*
IgG (C)	ns	NS	NS	NS	NS	NS
IgM (C)	NS	NS	NS	NS	ns*	NS

C: Continuous analysis.
D: Discrete analysis.
+: ≤18.6 and >18.6: Slope nonnegative for continuous analysis.
-: ≤18.6 and >18.6: Slope negative for continuous analysis.
NS/ns: Not significant ($p>0.10$).
NS*/ns*: Marginally significant ($0.05<p\leq0.10$).
Notes: P-value given if $p\leq0.05$.
C*T: Log_2 (current dioxin)-by-time interaction hypothesis test.
≤18.6: Log_2 (current dioxin) hypothesis test for Ranch Hands with time since end of tour of 18.6 years or less.
>18.6: Log_2 (current dioxin) hypothesis test for Ranch Hands with time since end of tour greater than 18.6 years.
A capital "NS" denotes relative risk/slope for ≤18.6 category less than relative risk/slope for >18.6 category, relative risk 1.00 or greater for discrete analysis, or slope nonnegative for continuous analysis; a lowercase "ns" denotes slope for ≤18.6 category greater than slope for >18.6 category, relative risk less than 1.00 for discrete analysis, or slope negative for continuous analysis.

TABLE 16-27. (Continued)

Summary of Current Dioxin and Time Analyses for Immunology Variables Based on Minimal and Maximal Assumptions (Ranch Hands Only)

	Adjusted					
	Minimal			Maximal		
Variable	C*T	≤18.6	>18.6	C*T	≤18.6	>18.6
Physical Examination						
Composite Skin Test Diagnosis (D)	**(NS)	**(ns)	**(ns)	NS	ns	ns
Laboratory						
CD2 Cells (C)	NS	ns	ns	NS	ns	NS
CD4 Cells (C)	**(NS)	**(ns)	**(NS)	**(NS)	**(ns)	**(NS)
CD8 Cells (C)	NS	ns	ns	ns	ns	ns
CD20 Cells (C)	NS	NS	+ 0.043	NS	ns	NS*
CD14 Cells (C)	NS	ns	NS	****	****	****
CD25 Cells (C)	NS*	- 0.028	NS	NS	ns	NS
HLA-DR Cells (C)	NS	ns	NS	NS	ns	NS
CD4/CD8 Ratio (C)	ns	NS	ns	ns	NS*	NS
TLC (C)	NS*	ns	NS	NS	ns	NS
Unstimulated PHA Response (C)	NS	ns	NS	ns	NS	NS
PHA Net Response (C)	**(ns*)	**(+0.033)	**(ns)	ns	NS	NS
Maximum PHA Net Response (C)	ns	+0.013	NS	NS	NS	NS*
Unstimulated MLC Response (C)	****	****	****	**(NS)	**(NS)	**(NS)
MLC Net Response (C)	ns	ns	ns	NS	ns	NS
NKCA 50/1 Net Response (C)	ns	NS	ns	ns*	NS	ns*
NKCA 50/1 Percent Release (C)	ns	NS	NS	ns	NS	ns
NKCI 50/1 Net Response (C)	**(ns*)	**(+0.027)	**(ns)	****	****	****
NKCI 50/1 Percent Release (C)	**(ns*)	**(+0.025)	**(ns)	****	****	****
IgA (C)	**(ns)	**(NS*)	**(NS)	**(NS)	**(NS)	**(+0.027)
IgG (C)	ns	NS	NS	ns	NS	NS
IgM (C)	NS	ns	NS	NS	- 0.027	ns

C: Continuous analysis.
D: Discrete analysis.
+: ≤18.6 and >18.6: Slope nonnegative for continuous analysis.
-: ≤18.6 and >18.6: Slope negative for continuous analysis.
NS/ns: Not significant ($p>0.10$).
NS*/ns*: Marginally significant ($0.05<p\leq0.10$).
** (NS)/** (ns): Log_2 (current dioxin)-by-time-by-covariate interaction ($0.01<p\leq0.05$); not significant when interaction is deleted; refer to Appendix Table O-1 for a detailed description of this interaction.
** (NS*)/** (ns*): Log_2 (current dioxin)-by-time-by covariate interaction ($0.01<p\leq0.05$); marginally significant when interaction is deleted; refer to Appendix Table O-1 for a detailed description of this interaction.
** (...): Log_2 (current dioxin)-by-time-by-covariate interaction ($0.01<p\leq0.05$); significant when interaction is deleted; refer to Appendix Table O-1 for a detailed description of this interaction.
****: Log_2 (current dioxin)-by-time-by-covariate interaction ($p\leq0.01$); refer to Appendix Table O-1 for a detailed description of this interaction.

TABLE 16-27. (Continued)

Summary of Current Dioxin and Time Analyses for Immunology Variables Based on Minimal and Maximal Assumptions (Ranch Hands Only)

Notes: P-value given if $p \leq 0.05$.

C*T: Log_2 (current dioxin)-by-time interaction hypothesis test.

≤18.6: Log_2 (current dioxin) hypothesis test for Ranch Hands with time since end of tour of 18.6 years or less.

>18.6: Log_2 (current dioxin) hypothesis test for Ranch Hands with time since end of tour greater than 18.6 years.

A capital "NS" denotes relative risk/slope for ≤18.6 category less than relative risk/slope for >18.6 category, relative risk 1.00 or greater for discrete analysis, or slope nonnegative for continuous analysis; a lowercase "ns" denotes slope for ≤18.6 category greater than slope for >18.6 category, relative risk less than 1.00 for discrete analysis, or slope negative for continuous analysis.

TABLE 16-28.

Summary of Categorized Current Dioxin Analyses for Immunology Variables (Ranch Hands and Comparisons)

Variable	All Categories	Unadjusted: Unknown versus Background	Unadjusted: Low versus Background	Unadjusted: High versus Background
Physical Examination				
Composite Skin Test Diagnosis (D)	NS	NS	NS	NS
Laboratory				
CD2 Cells (C)	NS	ns	NS	NS
CD4 Cells (C)	NS	ns	NS	NS
CD8 Cells (C)	NS	NS	ns	NS
CD20 Cells (C)	NS	NS	NS	NS*
CD14 Cells (C)	NS	ns	ns	ns
CD25 Cells (C)	NS	NS	ns	NS
HLA-DR Cells (C)	NS	NS	NS	NS
CD4/CD8 Ratio (C)	NS	ns*	NS	NS
TLC (C)	NS	ns	NS	NS
Unstimulated PHA Response (C)	NS	ns	NS	NS
PHA Net Response - Conc. 1 (C)	NS	NS	NS	NS
PHA Net Response - Conc. 2 (C)	0.042	ns	NS	+0.025
PHA Net Response - Conc. 3 (C)	NS	ns	NS	NS
PHA Net Response - Across Day and Concentration (C)	NS	ns	NS	NS*
Maximum PHA Net Response (C)	0.037	ns	NS	+0.022
Unstimulated MLC Response (C)	NS*	NS	NS	+0.011
MLC Net Response (C)	NS	NS	NS	ns
NKCA 50/1 Net Response (C)	NS	NS	ns	ns
NKCA 50/1 Percent Release (C)	NS	NS	ns*	ns
NKCI 50/1 Net Response (C)	NS	ns	NS	ns
NKCI 50/1 Percent Release (C)	NS	ns	NS	ns
IgA (C)	0.013	-0.003	ns	NS
IgG (C)	NS*	-0.008	ns	ns
IgM (C)	NS*	NS	ns*	NS

C: Continuous analysis.
D: Discrete analysis.
+: Difference in means nonnegative for continuous analysis.
-: Difference in means negative for continuous analysis.
NS/ns: Not significant ($p>0.10$).
NS*/ns*: Marginally significant ($0.05<p\leq0.10$).
Note: P-value given if $p\leq0.05$.
A capital "NS" denotes relative risk 1.00 or greater for discrete analysis or difference in means nonnegative for continuous analysis; a lowercase "ns" denotes difference in means negative for continuous analysis; a capital "NS" in the first column does not imply directionality.

TABLE 16-28. (Continued)

Summary of Categorized Current Dioxin Analyses for Immunology Variables (Ranch Hands and Comparisons)

Variable	All Categories	Adjusted: Unknown versus Background	Adjusted: Low versus Background	Adjusted: High versus Background
Physical Examination				
Composite Skin Test Diagnosis (D)	NS	NS	NS*	NS
Laboratory				
CD2 Cells (C)	**NS	**(ns)	**(NS)	**(NS)
CD4 Cells (C)	NS	ns	NS	NS
CD8 Cells (C)	NS	NS	ns	NS
CD20 Cells (C)	**(NS)	**(NS)	**(NS)	**(NS)
CD14 Cells (C)	NS	ns	ns	ns
CD25 Cells (C)	NS	NS	ns	NS
HLA-DR Cells (C)	****	****	****	****
CD4/CD8 Ratio (C)	NS	ns*	NS	NS
TLC (C)	****	****	****	****
Unstimulated PHA Response (C)	NS	ns	NS	NS
PHA Net Response - Conc. 1 (C)	NS	NS	NS	NS
PHA Net Response - Conc. 2 (C)	NS	ns	NS	NS*
PHA Net Response - Conc. 3 (C)	****	****	****	****
PHA Net Response - Across Day and Concentration (C)	NS	NS	NS	NS
Maximum PHA Net Response (C)	NS	ns	NS	NS*
Unstimulated MLC Response (C)	NS	NS	NS	+0.038
MLC Net Response (C)	NS	NS	NS	ns
NKCA 50/1 Net Response (C)	NS	NS	ns	ns
NKCA 50/1 Percent Release (C)	NS	NS	ns	ns
NKCI 50/1 Net Response (C)	****	****	****	****
NKCI 50/1 Percent Release (C)	**(NS)	**(ns)	**(ns)	**(ns)
IgA (C)	0.007	-0.003	ns	NS
IgG (C)	NS	-0.028	NS	NS
IgM (C)	NS*	NS	ns*	NS

C: Continuous analysis.
D: Discrete analysis.
+: Difference in means nonnegative for continuous analysis.
-: Difference in means negative for continuous analysis.
NS/ns: Not significant ($p>0.10$).
NS*/ns*: Marginally significant ($0.05<p\leq0.10$).
** (NS)/** (ns): Categorized current dioxin-by-covariate interaction ($0.01<p\leq0.05$); not significant when interaction is deleted; refer to Appendix Table O-1 for a detailed description of this interaction.
****: Log_2 (current dioxin)-by-time-by-covariate interaction ($p\leq0.01$); refer to Appendix Table O-1 for a detailed description of this interaction.
Note: P-value given if $p\leq0.05$.
A capital "NS" denotes relative risk 1.00 or greater for discrete analysis or difference in means nonnegative for continuous analysis; a lowercase "ns" denotes difference in means negative for continuous analysis; a capital "NS" in the first column does not imply directionality.

TABLE 16-29.

Summary of Dioxin-by-Covariate Interactions from Adjusted Analyses of Immunology Variables

Variable	Assumption	Covariate
Model 1: Ranch Hands - Log_2 (Initial Dioxin)		
CD2 Cells	Minimal	ALC
CD8 Cells	Minimal	DRKYR
CD8 Cells	Maximal	ALC
CD20 Cells	Minimal	AGE
CD14 Cells	Minimal	ALC,PACKYR
CD25 Cells	Minimal	CSMOK,PACKYR,DRKYR
CD25 Cells	Maximal	CSMOK,PACKYR,DRKYR
HLA-DR Cells	Minimal	AGE
HLA-DR Cells	Maximal	AGE,ALC
TLC	Minimal	ALC
PHA Net Response - Conc. 1	Minimal	DRKYR
PHA Net Response - Conc. 1	Maximal	PACKYR
PHA Net Response - Conc. 3	Maximal	ALC
PHA Net Response†	Minimal	PACKYR
NKCA 50/1 Percent Release	Minimal	CSMOK,DRKYR
IgG	Minimal	DRKYR
IgM	Minimal	ALC
Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time		
Composite Skin Test Diagnosis	Minimal	AGE
CD4 Cells	Minimal	DRKYR
CD4 Cells	Maximal	AGE
CD14 Cells	Maximal	PACKYR
PHA Net Response†	Minimal	DRKYR
Unstimulated MLC	Minimal	DRKYR
Unstimulated MLC	Maximal	PACKYR
NKCI 50/1 Net Response	Minimal	RACE
NKCI 50/1 Net Response	Maximal	RACE
NKCI 50/1 Percent Release	Minimal	RACE
NKCI 50/1 Percent Release	Maximal	RACE
IgA	Minimal	CSMOK
IgA	Maximal	CSMOK
Model 3: Ranch Hands and Comparisons by Current Dioxin Category		
CD2 Cells	--	AGE,DRKYR
CD20 Cells	--	AGE
HLA-DR Cells	--	AGE
TLC	--	AGE,DRKYR
PHA Net Response - Conc. 3	--	ALC
NKCI 50/1 Net Response	--	RACE,ALC
NKCI 50/1 Percent Release	--	RACE,ALC,DRKYR

†Across mitogen harvest day and mitogen concentration.

interaction for all cell surface marker variables except CD4 and the CD4/CD8 ratio. The adjusted analyses under the maximal assumption generally were nonsignificant. The CD8, CD25, and HLA-DR cells exhibited significant initial dioxin-by-covariate interactions under this assumption. Similar to the unadjusted analysis, the adjusted analysis under the maximal assumption indicated there was a marginally significant positive association between the CD4/CD8 ratio and initial dioxin.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

In general, the unadjusted analysis of the cell surface marker variables under both the minimal and maximal assumptions did not exhibit significant associations with current dioxin and time since tour (see Table 16-27). Under the minimal assumption, the unadjusted analysis of the nonzero CD25 cells displayed a significant negative association with current dioxin ($p=0.028$) for Ranch Hands with later tours (i.e., ≤ 18.6 years) and a nonsignificant positive association with current dioxin for Ranch Hands with early tours (i.e., >18.6 years). The two time strata associations cited above were found to differ marginally from each other. Under the maximal assumption, there were isolated marginally significant positive associations (i.e., for CD20 cells within time over 18.6 years, and CD4/CD8 ratio within time of 18.6 years or less).

For the cell surface marker variables, the adjusted analysis of the nonzero CD25 cells exhibited the same results under the minimal assumption as in the unadjusted analysis (i.e., a marginally significant current dioxin-by-time interaction, a significant negative association for Ranch Hands with late tours, and a nonsignificant positive association for Ranch Hands with early tours) because no covariates were retained in the adjusted model. For CD20 cells, Ranch Hands with an early tour under the minimal assumption exhibited a significant positive association ($p=0.043$). The adjusted analysis of CD4 cells displayed a significant interaction among current dioxin, time, and lifetime alcohol history under the minimal assumption and a significant interaction among current dioxin, time, and age under the maximal assumption. The adjusted analysis of CD14 cells under the maximal assumption exhibited a significant interaction among current dioxin, time, and lifetime cigarette smoking history.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

In the unadjusted analysis of each of the cell surface marker variables, the overall contrast for Ranch Hands in the unknown, low, and high current dioxin categories and Comparisons in the background current dioxin category was nonsignificant (see Table 16-28). In the unadjusted analysis, the CD4/CD8 ratio exhibited a marginally significant difference only for the unknown versus background contrast (the Ranch Hands had the lower CD4/CD8 average) and the unadjusted analysis of CD20 had a marginally significant difference for the high versus background contrast (the Ranch Hands had the higher CD20 average).

In the adjusted analysis of CD2 cells, there were significant interactions between categorized current dioxin and age and between categorized current dioxin and lifetime alcohol history. An additional adjusted model without these interactions displayed a nonsignificant overall contrast for CD2 cells. For CD20 cells, the adjusted analysis contained a significant interaction between categorized current dioxin and age. A followup model without the interaction exhibited a nonsignificant overall contrast as well as nonsignificant Ranch Hand versus Comparison contrasts. The adjusted analysis of HLA-DR cells contained

a significant interaction between categorized current dioxin and age. The adjusted analyses of the other cell surface marker variables were nonsignificant.

Longitudinal Analysis of CD4/CD8 Ratio

For the most part, the longitudinal analyses of the CD4/CD8 ratio showed no significant differences. For the maximal analysis using current dioxin and time, Ranch Hands with more than 18.6 years since their tour displayed a significant increasing association between current dioxin and the change in the CD4/CD8 ratio (1987 ratio relative to the 1985 ratio). This change is opposite to that expected if dioxin caused immunodeficiency.

Laboratory Examination Variables: Quantitative Studies—TLC

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

In the unadjusted analysis of TLC, both the minimal and maximal cohorts displayed a nonsignificant association between TLC and initial dioxin. Under the minimal assumption, the adjusted analysis contained a significant interaction between current alcohol use and initial dioxin. Without that interaction in the adjusted model, the association with initial dioxin was nonsignificant. Under the maximal assumption, the adjusted analysis also indicated that the relationship between TLC and initial dioxin was nonsignificant.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

Under both the minimal and maximal assumptions, the unadjusted analysis indicated that the current dioxin-by-time since tour interaction was nonsignificant for TLC. Also, the adjusted analysis of TLC based on the maximal assumption was nonsignificant for an interaction between current dioxin and time. Under the minimal assumption, the adjusted analysis of TLC exhibited a marginally significant interaction of current dioxin and time for the nonsignificant associations of the two time strata.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted analysis indicated that there were nonsignificant differences for TLC among the Ranch Hands in the unknown, low, and high current dioxin categories and Comparisons having background current dioxin category. The adjusted analysis of TLC for categorized current dioxin contained significant interactions between categorized current dioxin and age and between categorized current dioxin and lifetime alcohol history.

Laboratory Examination Variables: Functional Stimulation Tests

As part of the battery of functional stimulation studies, the following variables were analyzed: unstimulated PHA responses for harvest days 1 and 2 concurrently; an overall simultaneous analysis of six PHA net responses (PHA net response determined for each of 2 mitogen harvest days at 3 mitogen concentrations); the maximum of the six PHA net responses over mitogen harvest day and mitogen concentration; unstimulated MLC response; MLC net response; NKCA 50/1 net response; NKCA 50/1 percent release; NKCI 50/1 net response; and NKCI 50/1 percent release.

PHA Response

The expected effect of immunodeficiency on the PHA response is a reduction of cpm values due to impaired lymphocyte proliferation and less incorporation of radioactive precursor nucleotides into newly synthesized DNA. Furthermore, partial immunodeficiency would be reflected by an increase in the PHA concentration at which maximal response is stimulated (i.e., increase in minimal threshold for response).

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

Under both assumptions, the unadjusted and adjusted analysis of unstimulated PHA response exhibited nonsignificant associations with initial dioxin.

For the analysis of PHA considering 2 mitogen harvest days and 3 mitogen concentrations as repeated measure factors, there were significant interactions between initial dioxin and mitogen concentration under both assumptions. Consequently, unadjusted and adjusted analyses were performed at each mitogen concentration.

For mitogen concentration 1, the unadjusted analyses for both cohorts were nonsignificant. For the adjusted analyses of PHA at mitogen concentration 1, the minimal analysis contained a significant interaction between initial dioxin and lifetime alcohol history and the maximal analysis displayed a significant initial dioxin-by-lifetime cigarette smoking history interaction.

For mitogen concentration 2, the unadjusted analyses of PHA exhibited significant positive associations with initial dioxin under both the minimal and maximal assumptions ($p=0.016$ and $p=0.008$, respectively) and marginally significant positive associations with initial dioxin under both assumptions in the adjusted analyses. A negative response would be expected in impaired immunity.

For mitogen concentration 3, the unadjusted analyses of PHA net response contained nonsignificant associations with initial dioxin under both assumptions. The adjusted analysis of PHA net response for the minimal assumption was nonsignificant for an association with initial dioxin. Under the maximal assumption, the adjusted analysis exhibited a significant interaction between initial dioxin and current alcohol use.

Because the minimal cohort had an initial dioxin-by-harvest day interaction with a p-value between 0.01 and 0.05, unadjusted and adjusted analyses were also performed on the six PHA net responses across mitogen harvest day and mitogen concentration. The unadjusted analysis under the minimal assumption had a marginally significant positive association with initial dioxin and the adjusted analysis contained a significant interaction between initial dioxin and lifetime cigarette smoking history. An unadjusted analysis of PHA net response across day and concentration under the maximal assumption was nonsignificant.

For the unadjusted analyses under both assumptions, the maximum PHA net response displayed a significant positive association with initial dioxin ($p=0.005$ and $p=0.009$ for the minimal and maximal assumptions). For the adjusted analyses of the maximum PHA net response, both cohorts exhibited marginally significant positive associations.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

For the unadjusted and adjusted analysis of unstimulated PHA response, the interaction of current dioxin-by-time since tour was nonsignificant under both assumptions.

In the unadjusted analysis of the six PHA net responses under the minimal assumption, there was a marginally significant interaction between current dioxin and time. For time less than or equal to 18.6 years, there was a significant positive association between PHA net response and current dioxin (p=0.006) and a nonsignificant positive association for the other time stratum. Under the maximal assumption, the unadjusted analysis displayed a nonsignificant interaction between current dioxin and time. In the adjusted analysis of PHA net response under the minimal assumption, there was a significant interaction among current dioxin, time since tour, and lifetime alcohol history. A secondary analysis performed without the interaction exhibited a marginally significant current dioxin-by-time interaction. For time less than or equal to 18.6 years, there was a significant positive association (p=0.033) between PHA net response and current dioxin, and for time greater than 18.6 years a nonsignificant negative association. In the adjusted analysis under the maximal assumption, the interaction of current dioxin and time was nonsignificant.

In the unadjusted analysis of the maximum PHA net response under the minimal assumption, the interaction of current dioxin and time was nonsignificant. However, there was a significant positive association with current dioxin (p=0.002) for time less than or equal to 18.6 years. In the unadjusted analysis under the maximal assumption, there were positive associations with current dioxin for both time strata (time≤18.6, p=0.049 and time>18.6, p=0.008) but the interaction of current dioxin and time was nonsignificant. In the adjusted analysis of the maximum PHA net response under the minimal assumption, the interaction of current dioxin and time was nonsignificant but there was a significant positive association (p=0.013) with current dioxin for time less than or equal to 18.6 years. Under the maximal assumption, the adjusted analysis also displayed a nonsignificant interaction between current dioxin and time since tour; however, there was a marginally significant positive association for time over 18.6 years.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

The unadjusted and adjusted analyses indicated that there were nonsignificant differences for unstimulated PHA response among the Ranch Hands in the unknown, low, and high current dioxin categories and Comparisons in the background current dioxin category.

As a result of an interaction between categorized current dioxin and mitogen concentration, the unadjusted and adjusted analyses of PHA net response were performed separately at each mitogen concentration. For mitogen concentration 1, the unadjusted and adjusted analyses contrasting Ranch Hands in the unknown, low, and high current dioxin categories versus the Comparisons were nonsignificant. For mitogen concentration 2, the unadjusted analysis of the PHA net response contained a significant overall contrast (p=0.042) that was accompanied by a significant high versus background contrast (p=0.025). The adjusted analysis of the PHA net response at mitogen concentration 2 exhibited a nonsignificant overall contrast and a marginally significant contrast for the high versus background categories. For the two high versus background contrasts, Ranch Hands exceeded Comparisons on PHA net response. The unadjusted analysis of PHA net response

at mitogen concentration 3 was nonsignificant. The adjusted analysis contained a significant interaction with current alcohol use.

Unadjusted and adjusted analyses were performed without the interaction involving mitogen concentration (i.e., across mitogen harvest day and mitogen concentration). In the unadjusted analysis, the overall contrast was nonsignificant, but the high versus background contrast was marginally significant with Ranch Hands exhibiting a higher response than Comparisons. The adjusted analysis contained a nonsignificant overall contrast.

In the unadjusted analysis of the maximum PHA net response, the overall contrast was significant (p=0.037) and the high versus background contrast was also significant (p=0.022) with Ranch Hands being higher than Comparisons. The adjusted analysis contained a nonsignificant overall contrast and a marginally significant high versus background contrast with the Ranch Hand response again exceeding that of the Comparisons.

MLC

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

For unstimulated MLC response, the unadjusted analysis under the minimal assumption exhibited a nonsignificant association with initial dioxin. Under the maximal assumption there was a marginally significant positive association. The adjusted analysis for both assumptions exhibited a nonsignificant association between unstimulated MLC and initial dioxin.

For MLC net response, the unadjusted and the adjusted analyses under both assumptions were nonsignificant for an association with initial dioxin.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

In the unadjusted analysis of unstimulated MLC under the minimal and maximal assumptions, the interactions of current dioxin and time since tour were nonsignificant. However, there was a marginally significant positive association between unstimulated MLC and current dioxin for time over 18.6 years under the maximal assumption. The adjusted analysis contained a significant interaction among current dioxin, time, and lifetime alcohol history for the minimal cohort. The adjusted analysis of unstimulated MLC for the maximal cohort contained a significant interaction among current dioxin, time, and lifetime cigarette smoking history. Followup analyses without the interaction were nonsignificant for the maximal cohort.

For MLC net response, the unadjusted and the adjusted analyses of both assumptions contained nonsignificant interactions between current dioxin and time since tour.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

In the unadjusted analysis of unstimulated MLC response, the overall contrast of Ranch Hands in the unknown, low, and high current dioxin categories and Comparisons in the background current dioxin category was marginally nonsignificant. The high versus background contrast of unstimulated MLC response was significant (p=0.011), with Ranch

Hands in the high current dioxin category having a higher unstimulated MLC response than Comparisons in the background current dioxin category. In the adjusted analysis of unstimulated MLC response, the overall contrast was nonsignificant but the contrast for high versus background was significant (p=0.038) with the Ranch Hands in the high current dioxin category being greater on unstimulated MLC than the Comparisons.

For both the unadjusted and the adjusted analyses of MLC net response, the overall contrast of Ranch Hands in the unknown, low, and high current dioxin categories and Comparisons in the background current dioxin category were nonsignificant.

Natural Killer Cell

Both stimulated and Interleukin 2 stimulated natural killer cells decline with progression of malignancies and show only a fraction of normal activity with advanced disease. They are probably most important early in carcinogenesis to screen for and destroy tumor cells when their numbers are still small.

Model 1: Ranch Hands - Log_2 (Initial Dioxin)

The unadjusted and adjusted analyses displayed nonsignificant associations between NKCA 50/1 net response and initial dioxin under both assumptions.

Similarly, under both assumptions, the unadjusted analysis of NKCA 50/1 percent release was nonsignificant for an association with initial dioxin. Under the minimal assumption, the adjusted analysis of NKCA 50/1 percent release contained significant interactions between initial dioxin and current cigarette smoking and between initial dioxin and lifetime alcohol history. Under the maximal assumption, the adjusted analysis of NKCA 50/1 percent release was nonsignificant.

Under both the minimal and maximal assumptions, the unadjusted and the adjusted analyses exhibited nonsignificant associations between NKCI 50/1 net response and initial dioxin, as well as NKCI 50/1 percent release and initial dioxin.

Model 2: Ranch Hands - Log_2 (Current Dioxin) and Time

For NKCA 50/1 net response and NKCA 50/1 percent release, the unadjusted analysis under both assumptions exhibited nonsignificant interactions between current dioxin and time since tour. Under the minimal assumption, the adjusted analyses of NKCA 50/1 net response and NKCA 50/1 percent release also contained nonsignificant interactions between current dioxin and time since tour. Under the maximal assumption, the adjusted analysis of NKCA 50/1 net response exhibited a marginally significant current dioxin-by-time interaction. For time less than or equal to 18.6 years, there was a nonsignificant positive association between NKCA 50/1 net response and current dioxin. For time greater than 18.6 years, there was a marginally significant negative association between NKCA 50/1 net response and current dioxin. The adjusted analysis of NKCA 50/1 percent release displayed a nonsignificant current dioxin-by-time interaction under the maximal assumption.

Under the minimal assumption, the unadjusted analysis of NKCI 50/1 net response and NKCI 50/1 percent release displayed nonsignificant interactions between current dioxin and

time since tour. Under the maximal assumption, the unadjusted analysis of NKCI 50/1 net response and percent release displayed marginally significant current dioxin-by-time interactions. For time less than or equal to 18.6 years, there were marginally significant positive associations with current dioxin for both NKCI 50/1 variables. For time greater than 18.6 years, there were nonsignificant negative associations with current dioxin for both NKCI 50/1 net response and percent release.

Under the minimal assumption, the adjusted analyses of NKCI 50/1 net response and NKCI 50/1 percent release both contained a significant interaction among current dioxin, time, and race. Followup analyses without the interactions in the adjusted models displayed marginally significant interactions between current dioxin and time since tour. Under the minimal assumption, both NKCI 50/1 variables displayed a significant positive association with current dioxin for time less than or equal to 18.6 years and a nonsignificant negative association for time over 18.6 years. The adjusted analyses of NKCI 50/1 net response and NKCI 50/1 percent release under the maximal assumption also displayed significant interactions among current dioxin, time since tour, and race.

Model 3: Ranch Hands and Comparisons by Current Dioxin Category

For the unadjusted analyses of NKCA 50/1 net response and percent release and NKCI 50/1 net response and percent release, the overall contrasts of Ranch Hands in the unknown, low, and high current dioxin categories and Comparisons in the background current dioxin category were nonsignificant. Except for a marginally significant low versus background contrast on NKCA 50/1 percent release, the individual Ranch Hand versus Comparison contrasts were nonsignificant for these unadjusted analyses.

In the adjusted analysis of NKCA 50/1 net response and NKCA 50/1 percent release, the overall contrasts of Ranch Hand in the unknown, low, and high current dioxin categories and Comparisons in the background current dioxin category were nonsignificant. The adjusted analysis of NKCI 50/1 net response contained significant interactions between categorized current dioxin and race and between categorized current dioxin and current alcohol use. In the adjusted analysis of NKCI 50/1 percent release, there were significant interactions between categorized current dioxin and each of the following covariates: race, current alcohol use, and lifetime alcohol history. Because the p-values of each of the three interactions exceeded 0.01 but were less than 0.05, a followup model without the three interactions was investigated. The overall contrast for that model was nonsignificant.

Quantitative Immunoglobulins

Serum immunoglobulins may show either increases or decreases related to immunodeficiencies or malignancy. Severe defects of B cells can result in near absence of IgG, IgA, and IgM (hypogammaglobulinemia) or selective deficiency of one or two of these immunoglobulin classes. Hypogammaglobulinemia is frequently (but not always) a consequence of lymphatic cancer due to the replacement of normal immunoglobulin secreting cells with malignant ones. In some B-cell disorders, there is proliferation of a single (malignant) clone of cells that inappropriately synthesize a monoclonal immunoglobulin (IgG, IgA, IgM, IgD, or IgE, with only kappa or lambda light-chain type) (e.g., multiple myeloma, Waldenstrom's macroglobulinemia). In other disorders involving T cells such as AIDS, there is frequently a polyclonal increase of all serum immunoglobulins due to impaired (down) regulation of their synthesis. Liver disease and especially cirrhosis are also well known to