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FURTHER STUDY OF HEMATOLOGICAL CHANGES IN HUMANS CHRONICALLY EXPOSED TO LOW LEVEL GAMMA RADIATION

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ABSTRACT

This report summarizes the apparent hematological changes in 10 individuals, exposed to total-body gamma radiation in near tolerance amounts, over a period of four and one-half years. Two control groups, in addition to the exposed group, were used for comparative purposes. Control group I received essentially no gamma or beta radiation. Control group II received exposures at least a factor of 10 below those of the exposed group. The average total leukocyte count, absolute neutrophil value, and absolute lymphocyte value, were determined for each individual and for all groups. Analysis of data indicates a significant depression of the absolute lymphocyte value of the exposed group as compared with those of the control groups.

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FURTHER STUDY OF HEMATOLOGICAL CHANGES IN HUMANS CHRONICALLY EXPOSED TO LOW LEVEL GAMMA RADIATION

1. Introduction

A previous report[1] covered in detail the hematological changes observed in 10 individuals chronically exposed to low level gamma radiation for a period of three years at the Los Alamos Scientific Laboratory. Since the publication of the original report, data have been collected on the same individuals for an additional 18 months. This report covers the additional data and summarizes the observations during the entire four and a half years of the study.

For the purpose of analysis, the total body study period of 54 months has been divided into six consecutive periods of nine months each, designated as Periods I through VI, respectively. Various time intervals during the 54 months will be referred to by their appropriate period designation. The 54 months include the time between December 1946 and June 1951. The initial report[1] gave the observations for Periods I through IV.

The composition of the group of individuals exposed to the ionizing radiations and of the groups selected for control purposes has remained essentially the same as reported previously.[1] Conditions of exposure and methods of data collection and analysis changed somewhat in Periods V and VI These variations in experimental method are covered below.

2. Individuals Studied

2.1 Exposed Group

Ten individuals were selected for study since their work included exposure to total body gamma radiation in relatively large amounts. These individuals handled radioactive substances, both beta and gamma emitters.

The gamma ray spectrum was considered to be approximately that of radium. During Period V, one of the 10 individuals left the employ of the Los Alamos Scientific Laboratory. Another individual received only one blood count during Period VI. Hence, while the data on all 10 individuals from Period I to Period V were available, only eight of the original group were studied for Period VI.

Periodic physical examination of the 10 individuals has indicated that their general health is good. An average weight gain of five pounds has been noted in the group during the four and a half years of study. No unusual incidence of upper respiratory infection or other minor illnesses has been noted.

2.2 Control Groups

Both of the control groups originally selected for this study were used for Periods V and VI. Control group I, consisting of males whose work involved no exposure to beta or gamma radiation, originally included 46 individuals. Certain members of this group left the employ of the Los Alamos Laboratory during Periods V and VI, and by Period VI the size of the group was reduced to 21 individuals. Several members of this group were assigned new work during the later portion of the study, and their exposure under the new working conditions included beta and gamma radiation. The average level of exposure was, however, at least a factor of 10 below that of the personnel of the exposed group; therefore, these men were not dropped from the control group. Periodic physical examinations have indicated that the general health of the control group remained good.

The "random" Control group II, added to the study during Period IV, originally consisted of 46 males whose hematology records were selected at random from general files in the hematology section. No consideration was

given the exposure the individuals received. While numerous members of the group received both beta and gamma exposure, all exposure levels were as le a factor of 10 below those encountered in the exposed group. Because some dividuals left the project, the number of individuals in Control group II had dropped to 30 by Period VI.

No individuals were added to either control group during the time period covered in this report, since sufficient blood counts had been done on the remaining individuals to give a satisfactory sample of normal total white blood counts, absolute neutrophil, and absolute lymphocyte, levels for the Los Alamos area. As mentioned in the previous report, all control individuals had resided at the altitude of Los Alamos for as long as the expos individuals and enjoyed generally comparable working conditions.

3. Collection of Data

All blood counts were done between 8 a.m. and 10 a.m. The same technicians who did the counting in previous periods continued to do so in Periods V and VI. Capillary blood was used, and pipettes calibrated by the National Bureau of Standards were employed. One pipette and one side of the standard counting chamber were employed for each count. Differential counts normally included 100 cells but occasionally 200 were observed. During Periods I through IV, all individuals in both the exposed and control groups received their blood counts at a central laboratory. During Periods V and VI, however, the exposed group and several members of the control groups received blood counts at their places of work.

Members of the exposed group, as well as those members of the control groups receiving incidental radiation exposure, were monitored with body film badges and pocket ionization chambers. Film badge estimates of dose are used

throughout this report. Excellent agreement was observed between the film badge and ionization chamber values.

During Period V, the general level of exposure of the exposed group was essentially the same as it had been during Periods III and IV. During Period VI, however, it was possible to reduce the exposure of nearly all exposed group individuals to levels comparable to those received by members of the control groups mentioned above. In view of the observed decrease in absolute lymphocytes in members of the exposed group during Periods I through IV, this change in exposure level was viewed with great interest by the investigators. With the prospect that the majority of the individuals would no longer receive relatively large radiation doses, the opportunity existed to determine whether the absolute lymphocyte values of these individuals would return to control levels. The demonstration of such a return to normal levels would strengthen the postulate that the observed lymphopenia was radiation-induced. Persistent lymphopenia would make the postulate less tenable.

4. Method of Statistical Analysis of Data

The average total white blood cell count, absolute neutrophil value, and absolute lymphocyte value, were determined for each individual for each exposure period. The mean of the above values for each group was then calculated using the mean figures for each individual in the group. Thus the period-group averages reported consist of the average of the determinations in 8 to 10 individuals in the case of the exposed group and of 21 to 46 individuals in the control groups. The significance of the difference of the mean values for control and exposed groups are expressed in this report in terms of the "t" test.

In addition to comparing the control and exposed groups, an analysis was made of the variations in absolute lymphocyte values for members of the exposed group. A least squares line, each point weighted directly as the number of observations and inversely as the variance of the observations about their mean, was fitted to the mean absolute lymphocyte values for each individual over the six periods. The regression coefficient of this line expressed the absolute number of lymphocytes lost or gained per nine-month period over the entire duration of the study. The significance of each of these regression coefficients was determined and is expressed in terms of its "t" value. The average regression coefficient for all 10 individuals was determined, and the hypothesis that this average regression coefficient differed significantly from zero was tested in the following manner. Where b_i is the regression coefficient for any one individual and b_o is the average regression coefficient for all individuals in the exposed group, the sum of the square of the standard deviate, s^2, is given by

$$s^2 = \frac{(b_i - b_o)}{9}$$

and "Student's" t-value is given by

$$t = \frac{\sum b_o}{s\sqrt{10}}$$

In the case of individuals 1 and 6, who left the project in Period V, the regression coefficient is based on the values for Periods I through V only.

5. Experimental Results

Table 1 gives the total and average exposure for all 10 individuals in the exposed group for Periods I through VI. The mean total white count, the absolute lymphocyte value, and the absolute neutrophil value, for each individual in the exposed group and for the group as a whole are given in Table 2 for all six periods, together with the number of counts each man had in each

period. Average values for the control groups for all six periods are given in Table 3. The significance of the differences between values for the various periods within the control groups and exposed groups themselves are given in Table 4. The significance of the differences between means of the exposed group and the control group for each period is tabulated in Table 5. Data on the regression coefficients of least squares lines of fit to the absolute lymphocyte values for each man in the exposed group are presented in Table 6.

As was the case in the analysis of data on Periods I through IV presented in the original report, it is seen that significant results occur only in the lymphocyte values for the exposed individuals and their depression below the absolute lymphocyte values for the control groups. The significance of the depression of lymphocyte values within the exposed group from Period I to Period VI carries a probability of less than 1 per cent that it is due to chance alone.

6. Discussion

The percentage of lymphocytes found in both control groups over the six study periods (average 29.3 per cent, range 27.9 - 30.7 per cent) is lower than the values previously reported for individuals residing at 5000 - 8000 ft altitudes. Peterson found an average of 36.26 per cent lymphocytes in differential counts on young adults at 5755 ft (Butte, Montana),[2] and Stammers reported an average of 36.8 per cent in 171 young adults at 5750 ft (Johannesburg, South Africa).[3] Ruppanner found 38.25 per cent in 8 men residing at 5741 ft and 36.6 per cent in 12 men residing at 7382 ft in the Swiss alps.[4] Peterson suggested that the higher ultraviolet content of sunlight at these altitudes might be responsible for the lymphocytosis, and

Stammers pointed out that the Johannesburg area received an average of 73 per cent of the total annual available sunlight. The Los Alamos area has an equally favorable climate, yet at the Los Alamos elevation, 7200 ft, no lymphocytosis was seen.

Nor was the percentage of lymphocytes found in the present study as high that noted by Osgood in studies of normal young adults near sea level,[5,6] although our findings agree well with the values reported by Wintrobe.[7] It is possible that technical differences between laboratories may account for some of the differences noted.

The values for the total white blood cell count in this study are slightly lower than those reported by Osgood, Ruppanner, and Peterson, but the differences seen are not statistically significant.

Chamberlain has pointed out the tendency of physicians to repeat blood counts on individuals when abnormal values are found, thus biasing the data in favor of high or low values with individuals showing transitory or persistent abnormal white cell levels.[8] He has pointed out that this may be a legitimate criticism of Knowlton's study of leukopenia and lymphopenia in individuals exposed to gamma radiation over many months.[9] We feel confident that no such bias has appeared in the data collected on the members of the exposed group in this study. At no time was the counting schedule of the exposed group altered during the course of the study because of abnormal values as the counting frequency was high enough to make special repeat determinations unnecessary in the reviewing physicians' opinion.

The absence of repeat counts was not true in the case of the control groups, however, in which there was a low counting frequency. When an abnormal white cell count was found, a repeat determination was usually made in a matter of days after the original count. Since in the majority of

instances, the repeat determination was made because the original value found was below 4000 cells, values of the total white cell count for control groups may be slightly lower than they truly should be. This may account for the differences between our control white cell count values and the values given by other authors noted above. No attempt has been made at this writing to remove repeat counts from the control group data, for we feel that the effect of such adjustment of the data would be slight, and would tend to increase rather than decrease the significance of the differences between control and exposed group means.

The six time periods used in this study should have minimized any effect which seasonal blood count variations would have on the changes seen. Each time period of nine months included three of the four annual seasons, the three included varying within any consecutive group of four periods. In our data, no significant differences between the determinations made on control groups appeared between any periods. Nor did any change appear in the total white cell count of the exposed group which might be explained on the basis of possible seasonal variations. This is contrary to the changes in the total white cell count reported by Chamberlain, who found lower total white cell values in the summer months.[8]

The large number of reports dealing with hematological observations in humans chronically exposed to low intensity ionizing radiations attests the general interest in this subject.[8-17] In general, the most consistent finding reported has been a granulocytopenia. Opinion is divided on whether or not lymphocytes are decreased or increased with doses near present tolerance levels. With relatively large continued exposure, lymphopenia is evident,[11] and in the most complete data given on chronic low level exposures to date, lymphopenia was also the most striking finding in certain cases.[10] In his analysis of the

data collected in AEC laboratories in this country, Moshman[16] stated that granulocytopenia appeared to be the most consistent finding in personnel exposed to ionizing radiation, but he also noted a general downward trend in the total white cell count and the lymphocyte values in certain exposure groups. Chamberlain has concluded that decrease in the circulating lymphocytes is the only significant finding in personnel exposed to slightly more gamma radiation than was received by the individuals in this study.[8] His group included individuals with possible internal as well as external radiati

Since the original report of the present study[1] the mean lymphocyte value for the group has continued to decrease. In the analysis of the first four periods, the average absolute lymphocyte loss per individual per period was 94 cells. In the present analysis including the data from the entire six periods now available, the average loss is 66 cells per period. Thus the decrease in the last two periods was not as large as would have been anticipated on the basis of the changes seen in the first report. This tendency of the absolute lymphocyte value to level off, rather than to continue its rate of decrease, is encouraging, especially in view of the marked reduction in the exposure of the individuals during Period VI.

No significant variations in the hematological observations made, other than the lymphocyte change, has been noted to date. No change in working conditions, other than the marked reduction in exposure, has occurred. In the authors' opinion, the possibility of internal hazard has remained slight. No other known toxic agents were encountered in the working environment of the individuals studied.

7. Summary

Hematological changes observed in 10 individuals chronically exposed

to low-level gamma radiation over a period of 4-1/2 years have been limited to an apparent persistent lymphopenia. The average exposure of each individual over this period of time has been 146 mr/wk. No factor in the working conditions of these individuals other than the ionizing radiation exposure has been discovered which could account for the apparent hematological changes seen.

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TABLE 1

GAMMA EXPOSURE OF EXPOSED GROUP IN ROENTGENS

Individual Period	1	2	3	4	5	6	7	8	9	10	Total	Weekly Avg. Per Man
I	11.62	13.84	9.72	10.11	13.67	12.77	9.19	8.73	8.43	9.56	107.64	0.276
II	2.33	8.31	3.98	3.51	5.98	5.73	6.13	3.87	4.33	7.99	52.16	0.134
III	2.97	4.24	3.92	3.21	8.65	4.66	3.55	5.74	5.78	7.19	49.91	0.128
IV	4.25	2.69	6.90	3.35	7.25	6.28	5.42	7.25	8.11	8.15	59.65	0.153
V	3.21	6.66	7.25	2.46	7.71	7.97	7.83	8.31	7.65	8.94	67.99	0.174
VI	0.00	0.25	0.17	0.00	0.21	0.00	0.18	0.41	0.25	1.27	2.74	0.009
Total	24.38	35.99	31.94	22.64	43.47	37.41	32.30	34.31	34.55	43.10	340.09	0.874
Weekly Avg. per Man	0.125	0.154	0.136	0.097	0.186	0.192	0.138	0.147	0.148	0.184	1.507	0.146

TABLE 2

SUMMARY OF HEMATOLOGICAL DATA OF EXPOSED GROUP

Period	Individual	1	2	3	4	5
I	No. of Counts	35	37	39	24	38
	W.B.C.	8509 ± 223*	7918 ± 206	6491 ± 190	6461 ± 250	6322 ± 230
	Neutrophils	5365 ± 188	5175 ± 154	4014 ± 138	4212 ± 216	3776 ± 176
	Lymphocytes	1743 ± 71	1869 ± 69	1665 ± 68	1387 ± 87	1727 ± 59
II	No. of Counts	27	36	25	13	35
	W.B.C.	7688 ± 247	6635 ± 179	5775 ± 135	5621 ± 418	5433 ± 167
	Neutrophils	4644 ± 235	4500 ± 162	3784 ± 86	3662 ± 275	3270 ± 130
	Lymphocytes	1727 ± 102	1401 ± 67	1290 ± 66	1121 ± 134	1454 ± 59
III	No. of Counts	25	32	20	13	25
	W.B.C.	7655 ± 363	7919 ± 310	6227 ± 398	5500 ± 290	5289 ± 137
	Neutrophils	4677 ± 257	5391 ± 258	3887 ± 233	3445 ± 220	3217 ± 119
	Lymphocytes	1612 ± 101	1568 ± 88	1487 ± 63	1189 ± 101	1426 ± 79
IV	No. of Counts	20	28	34	13	26
	W.B.C.	7590 ± 391	7075 ± 205	5700 ± 169	6269 ± 420	5430 ± 241
	Neutrophils	5036 ± 308	4966 ± 188	3536 ± 110	3991 ± 352	3438 ± 243
	Lymphocytes	1418 ± 95	1388 ± 75	1446 ± 86	1223 ± 87	1287 ± 72
V	No. of Counts	13	30	23	5	27
	W.B.C.	7454 ± 313	8153 ± 355	5700 ± 174	5900 ± 727	4992 ± 148
	Neutrophils	4962 ± 328	5903 ± 337	3754 ± 146	3888 ± 432	3003 ± 148
	Lymphocytes	1384 ± 75	1411 ± 74	1189 ± 66	1143 ± 239	1359 ± 66
VI	No. of Counts	1	31	21	10	26
	W.B.C.	7100	7861 ± 321	5090 ± 168	5650 ± 417	5830 ± 280
	Neutrophils	4757	5604 ± 291	3367 ± 136	4193 ± 427	3895 ± 224
	Lymphocytes	2130	1550 ± 95	1064 ± 63	975 ± 81	1332 ± 100

* Denotes estimated standard error of the mean.

TABLE 2 (Continued)

Period	Individual	6	7	8	9	10	Average (1 - 10)
I	No. of Counts	34	30	39	39	34	
	W.B.C.	7985 ± 249*	7133 ± 202	9452 ± 268	10474 ± 268	7243 ± 180	7797 ± 440
	Neutrophils	4312 ± 205	3918 ± 172	4812 ± 205	7859 ± 228	4688 ± 120	4814 ± 477
	Lymphocytes	2325 ± 142	2021 ± 66	2764 ± 135	1719 ± 76	1601 ± 75	1882 ± 128
II	No. of Counts	28	36	31	34	30	
	W.B.C.	6914 ± 172	6563 ± 150	7798 ± 237	9355 ± 356	6742 ± 144	6853 ± 381
	Neutrophils	3785 ± 168	3455 ± 132	4290 ± 214	7020 ± 313	4765 ± 121	4318 ± 336
	Lymphocytes	1865 ± 110	1955 ± 85	2087 ± 97	1589 ± 79	1165 ± 68	1566 ± 109
III	No. of Counts	24	33	27	27	22	
	W.B.C.	7258 ± 208	6639 ± 150	7613 ± 214	9948 ± 447	6948 ± 209	7101 ± 423
	Neutrophils	4083 ± 166	3460 ± 109	4240 ± 164	7651 ± 428	4804 ± 126	4485 ± 412
	Lymphocytes	1790 ± 94	2012 ± 91	2004 ± 99	1527 ± 102	1253 ± 72	1587 ± 86
IV	No. of Counts	28	32	32	30	28	
	W.B.C.	6886 ± 216	7347 ± 180	8159 ± 204	9263 ± 353	7461 ± 399	7118 ± 366
	Neutrophils	4016 ± 177	4007 ± 150	4892 ± 204	7300 ± 282	5302 ± 301	4648 ± 361
	Lymphocytes	1642 ± 71	2124 ± 89	1918 ± 112	1811 ± 97	1348 ± 98	1560 ± 96
V	No. of Counts	20	32	29	31	33	
	W.B.C.	6860 ± 241	7046 ± 212	8886 ± 280	10350 ± 403	7184 ± 204	7253 ± 507
	Neutrophils	3900 ± 228	4178 ± 192	5592 ± 279	7383 ± 386	5229 ± 162	4452 ± 472
	Lymphocytes	1602 ± 81	1801 ± 89	1844 ± 93	2108 ± 95	1129 ± 63	1507 ± 96
VI	No. of Counts	1	33	31	24	22	
	W.B.C.	5100	6267 ± 159	8458 ± 295	9441 ± 355	7381 ± 431	7022 ± 530**
	Neutrophils	3162	3396 ± 132	5178 ± 229	6931 ± 330	5559 ± 366	4766 ± 443
	Lymphocytes	1581	1852 ± 81	1780 ± 89	1896 ± 121	1161 ± 91	1451 ± 131

* Denotes estimated standard error of the mean.

** Mean of 8 individ[illegible]

TABLE 3

SUMMARY OF HEMATOLOGICAL DATA FOR CONTROL GROUPS

PERIODS I-VI

Period	WBC	Neutrophils	Lymphocytes
I	7009 ± 248**	4164 ± 206	1983 ± 66
I*	7654 ± 391	4648 ± 293	2133 ± 108
II	6790 ± 250	4079 ± 204	1927 ± 72
III	6865 ± 263	4001 ± 212	2026 ± 77
IV	6844 ± 327	3994 ± 230	2042 ± 87
IV*	7053 ± 267	4223 ± 245	2049 ± 96
V	6741 ± 300	4045 ± 220	2068 ± 112
V*	7500 ± 332	4427 ± 215	2175 ± 100
VI	6591 ± 362	4079 ± 236	1952 ± 127
VI*	7434 ± 287	4380 ± 198	2264 ± 112

* Denotes random control group.

** Denotes standard error of the mean.

TABLE 5

SIGNIFICANCE OF DIFFERENCE BETWEEN EXPOSED GROUP AND CONTROL GROUP MEANS FOR EACH PERIOD

Periods			W.B.C.		Neutrophils		Lymphocyte	
Exposed	Control	df	t	prob**	t	prob.	t	prob
I	I	54	1.38	20%	1.32	20%	.59	>50
II	II	54	.11	>50%	.51	50%	2.22	4
III	III	54	.39	>50%	.98	35%	2.57	2
IV	IV	44	.42	>50%	1.37	20%	2.78	<1
V	V	37	.87	40%	.88	40%	2.80	<1
VI	VI	27	.64	>50%	1.48	15%	2.26	3%
I	I*	46	.18	>50%	.27	50%	1.13	25%
IV	IV*	54	.11	>50%	.77	45%	2.31	3%
V	V*	47	.35	>50%	.05	50%	3.27	<1%
VI	VI*	36	.66	>50%	.87	40%	3.56	<1%

* Denotes "random" control group used for comparison.

** Probability that difference observed is due to chance alone.

TABLE 6

SIGNIFICANCE OF LYMPHOCYTE DEPRESSION IN EXPOSED GROUP

(ON THE BASIS OF REGRESSION COEFFICIENTS)

Individual	Regression Coefficient* (b_i)	t for test of b
1	- 97.61	10.4
2	- 49.28	5.3
3	- 97.61	10.4
4	- 64.37	6.9
5	- 78.53	8.4
6	-144.46	15.4
7	- 32.44	3.5
8	-134.73	14.4
9	+ 75.28	8.0
10	- 66.97	7.2
Average Regression Coefficient (b_o)	- 69.07	

* In terms of decrease of lymphocytes per nine-month period.

Since the individual regression coefficients differ among themselves somewhat more than would be expected on the variation of the points around each line, the hypothesis that the average slope differs significantly from zero is tested against the variation among the regression coefficients.

The square of the standard deviates, $s^2 = \frac{(b_i - b_o)}{9} = 3812.25$

$$t = \frac{\sum b_o}{\frac{s}{\sqrt{10}}} = - 3.54$$

t with 9 degrees of freedom is significant at between the 1 per cent and .1 per cent levels.