# THE FALL IN THE BLOOD LYMPHOCYTES OF THE DOG UNDER CHLORALOSE ANÆSTHESIA.

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THE data presented in this paper form a convenient starting point for a consideration of the factors involved in the maintenance of the "lymphocyte balance". By "lymphocyte balance" is meant the mechanism whereby the blood lymphocytes are maintained at a fairly constant level and prevented from undergoing fluctuations of too extreme a nature.

## MATERIAL AND TECHNIQUE.

The work has been performed entirely on dogs. At various periods after food the animals were anæsthetized with a C.E. mixture, and anæsthesia then maintained by the intravenous injection of chloralose. Immediately after the chloralose injection the first blood count was taken (count A)—in all cases a total and differential white cell count, and in some experiments a red cell count. Blood was obtained in free flow either from a vein on the anterior abdominal wall or from the ear. The animal was then left alone for periods varying from  $2\frac{1}{4}$  to  $4\frac{2}{3}$  hours, after which a second blood count (count B) was made.

### **RESULTS.**

The complete data for a typical experiment are given in Table I.

The fall in the blood lymphocytes is an almost constant phenomenon; the rise in the neutrophiles is usual, but not quite so regular. Table II summarizes the results of thirteen experiments. In each case counts A and B are given both for lymphocytes and neutrophiles. Count A is above the line, count B below. The changes are given both absolutely, in terms of the numbers of cells, and relatively, count B being expressed as a percentage of count A.

| TABLE I. | Animal     | No.4ES    | 70. Dog | g. Female.   | Weight=   | 10 kg. 6  | 3. vi. 35. | No special fe | eding. |
|----------|------------|-----------|---------|--------------|-----------|-----------|------------|---------------|--------|
| Chl      | loralose d | lissolved | in bact | eria-free sa | line. Blo | od from a | abdomin    | al wall vein. |        |

## Anæsthetic begun 11 a.m.

|                   | C                    | hloralose       | Saline           |              |  |  |
|-------------------|----------------------|-----------------|------------------|--------------|--|--|
| Time              |                      | g.              | c.c.             |              |  |  |
| 11.20 a.m.        |                      | 1.0             | 60               |              |  |  |
| 12.00 noon        | 0.2                  |                 | 20               |              |  |  |
| 12.10 p.m.        |                      | 0.2             | 20               | 20           |  |  |
| 1.50 p.m.         | 0.2                  |                 | 20               |              |  |  |
|                   | Cour                 | nt A, 11.30 a.n | a. Count         | B, 2.30 p.m. |  |  |
| Red blood cells   |                      | 5,350,000       | 1                | 5,760,000    |  |  |
| White blood cells |                      | 12,700          |                  | 12,080       |  |  |
| Differential:     |                      |                 |                  | -            |  |  |
| Cells counted     |                      | 480             |                  | 461          |  |  |
| Lymphocytes       | 27.3 р.с.            | 3,470           | 9.33 p.c.        | 1,130        |  |  |
| Neutrophiles      | 62·7 <sup>1</sup> ., | 7,960           | 83·73            | 10,100       |  |  |
| Monocytes         | 1.9 "                |                 | 2.6              | <u> </u>     |  |  |
| Eosinophiles      | 8·1 "                |                 | 4·34 "           |              |  |  |
| Lymphocytes A     |                      | 3,470           | Neutrophiles A   | 7,960        |  |  |
| Lymphocytes B     |                      | 1,130           | Neutrophiles B   | 10,110       |  |  |
| Lymphocyte fall   |                      | 2,340           | Neutrophile rise | 2,150        |  |  |

| TABLE | п.  | Explanation | given | in | text. |
|-------|-----|-------------|-------|----|-------|
| LABLE | 11. | revenuence  | given | щ  | UEX L |

|                          | Time              | TABLE            | II. Ex           | planation gi | ven in te        | ext.        |                 |
|--------------------------|-------------------|------------------|------------------|--------------|------------------|-------------|-----------------|
| No. of                   | interval          | Lympho-          |                  | Neutro-      |                  | Lymphocyte  | Neutrophile     |
| animal                   | hours             | cytes            | p.c.             | philes       | p.c.             | change      | change          |
| 1 1 2 5 9                | 91                | 2,050            | 100              | 10,670       | 100              | 1 000       | 14.000          |
| 4 120 00                 | 3 <u>7</u>        | 750              | 36               | 25,000       | $\overline{230}$ | 1,300 -     | 14,330 +        |
| 4 10 10                  | 0.1               | 2,640            | 100              | 14,620       | 100              |             |                 |
| 4 ES 50                  | 3 <del>1</del>    | 1,425            | 54               | 13,980       | 96               | 1,215 –     | 640 -           |
|                          |                   | 7.250            | 100              | 16,750       | 100              |             |                 |
| 4 ES 49                  | 3                 | 1.200            | 17               | 29,760       | 178              | 6,050 -     | 13,010 +        |
|                          |                   | 2.980            | 100              | 12,720       | 100              |             |                 |
| 4 ES 48                  | 4 <del>]</del>    | 1.290            | 43               | 4 875        | 38               | 1,690 -     | 7,845 -         |
|                          |                   | 4 620            | 100              | 17 100       | 100              |             |                 |
| $4 \times 847$           | 3 <del>1</del>    | 2 100            | 45               | 16,000       | 100              | 2,520 -     | 200 –           |
|                          |                   | 4 600            | 100              | 10,900       | 99<br>100        |             |                 |
| $4 \operatorname{ES} 46$ | 3 <del>3</del>    | 4,080            | 100              | 8,004        | 100              | 1,620 -     | 3.866 +         |
|                          |                   | 3,000            | 60               | 11,930       | 148              |             |                 |
| $4 \operatorname{ES} 45$ | 3                 | 1,875            | 100              | 9,700        | 100              | 825 -       | $3200 \pm$      |
|                          |                   | 1,050            | 56               | 12,900       | 133              | 020         | 0,200           |
| 4 ES 44                  | 21                | 3,970            | <u>100</u>       | 10,650       | 100              | 9.056       | 4 850           |
|                          | -4                | 1,914            | <b>48</b>        | 6,000        | 56               | 2,000       | 4,030 -         |
| 4 ES 64                  | 2                 | 1,720            | 100              | 8,070        | 100              | <b>50</b> . | 0.010           |
| TROOT                    | 9                 | 1,770            | $\overline{103}$ | 11,980       | 149              | <b>90</b> + | 3,910+          |
| 1 1 2 70                 | 9                 | 3,470            | 100              | 7,960        | 100              |             |                 |
| 4 LO /U                  | 3                 | 1,130            | 32.5             | 10,110       | 127              | 2,340 –     | 2,150 +         |
| 4 12 9 7 1               | 01                | 920              | 100              | 6,830        | 100              |             |                 |
| 46671                    | 2 <del>2</del>    | $\overline{200}$ | 22               | 13,500       | 198              | 720 –       | 6,670 +         |
| 4 13 0 5 0               |                   | 2,750            | 100              | 4.750        | 100              |             |                 |
| 4 Lo72                   | $Z_{\frac{1}{2}}$ | 700              | 26               | 5,890        | 124              | 2,050       | 1,140 +         |
|                          |                   | 2.330            | 100              | 4,680        | 100              |             |                 |
| 4 ES 73                  | $2\frac{1}{2}$    | 1.110            | 50               | 8,910        | 190              | 1,220 –     | <b>4,23</b> 0 + |
|                          |                   | .,               |                  |              |                  |             |                 |

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Thus, to take the summarized data for the first experiment in the table,  $4 \times 58$ : The time interval between counts A and B was  $3\frac{1}{4}$  hours. Lymphocytes A were 2050 per c.mm., and lymphocytes B 750. If lymphocytes A are taken as 100 p.c., then lymphocytes B are 36 p.c. of lymphocytes A. Similarly with the neutrophiles. The last two columns give the actual increase or decrease of lymphocytes and neutrophiles. In this case the lymphocytes fell by 1300 per c.mm., and the neutrophiles rose by 14,330.

## DISCUSSION.

Reference to the column in Table II headed "lymphocyte change" shows that the lymphocytes fell in all the experiments but one, namely  $4 \times 564$ . In this there was a slight apparent rise (+50 per c.mm.). A single change so small in amount possesses no significance seeing that in six consecutive counts made on the same sample of lymph there was an average error of 8.7 p.c. The range of hæmocytometric error therefore is such that one cannot attach importance to changes in cell counts unless they are fairly well marked, and of relatively frequent occurrence.

The biggest lymphocyte fall—as also the highest initial lymphocyte count—occurred in 4ES49. The dog upon which this experiment was performed had a large septic wound upon the face, and this must undoubtedly have been rubbed and massaged during the time the anæsthetic mask was in position. Thus the animal was probably inoculated with a large dose of its own bacteria and toxins, and there is reason to believe from this and other experiments that the abnormal lymphocyte figures are to be correlated with this auto-inoculation.

In 4ES70, 71, and 72, the saline used for dissolving the chloralose was filtered through a Berkefeld filter. The use of this bacteria-free saline does not appear to have influenced either the lymphocyte fall or the neutrophile rise.

The neutrophiles rose in ten out of the thirteen experiments, and fell in three, namely 4ES56, 48, and 47. The most marked fall was in 4ES48. This animal had the astonishing respiration rate of 104 per min., and the abnormal neutrophile reaction may possibly be associated with blood changes consequent upon this rapid breathing.

On reviewing the results of the above experiments, it seems clear that in the dog under the experimental conditions described, a fall in the blood lymphocytes is practically a constant change. From these data alone, however, it is impossible to form any opinion as to the significance of this change, or even to account for the fact that the change takes place. It has been suggested [Yoffey, 1933] that in the normal dog the number of lymphocytes present in the blood is the resultant of a balance between two opposing processes. Newly formed lymphocytes are constantly entering the blood, while others are leaving it. A fall in the blood lymphocytes may mean either an increase in the number of lymphocytes leaving the blood, or a decrease in the number of newly formed lymphocytes entering the blood, or a combination of both these changes. In either case the question can only be solved by the dissociation of these two processes experimentally, and the investigation of each of the processes separately. The results of such an investigation will be presented in a further paper.

### SUMMARY.

Thirteen dogs were anæsthetized by C.E. induction and the intravenous injection of chloralose. The blood lymphocytes were counted immediately after the chloralose injection, and after a period varying from  $2\frac{1}{4}$  to  $4\frac{2}{3}$  hours. In twelve out of the thirteen cases there was a marked fall in the blood lymphocytes, in one there was no significant change.

#### REFERENCE.

Yoffey, J. M. (1933). J. Anat. 67, 250-62.