**Biochemistry.** — *"On the spreading of the different lipoids from chromocytes of different animals."* By E. GORTER, M. D., and F. GRENDEL. (From the Laboratory of Pediatrics of the University of Leyden, Leyden, Holland). (Communicated by Prof. P. EHRENFEST.)

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In the paper of BEUMER and BÜRGER [1]) we were able to find exact indications on the different lipoids found in the chromocytes of the sheep. They have obtained from 1 Litre of blood

0.5 gm. sphingomyelin
0.5 gm. kephalin etc.
0.5 gm. cholesterol.

Through the kindness of Dr. LEVENE from the Rockefeller Institute we were enabled to determine the spreading value of these substances. He put at our disposal a pure specimen of sphingomyelin and a specimen of kephalin containing 25 % of lecithin.

In order to enable the reader to compare the values, we give not only the spreading per 1 mg. of the material, but also the spreading per molecule.

| | Spreading per molecule | Spreading per 1 mg. |
|---|---|---|
| | sq. cm. | sq. cm. |
| Kephalin (with 25 % lecithin) | $116 \times 10^{-16}$ [2]) | $0.84 \times 10^4$ |
| Sphingomyelin | $46 \times 10^{-16}$ | $0.35 \times 10^4$ |
| Cholesterol | $40 \times 10^{-16}$ | $0.62 \times 10^4$ |
| Cholesterol palmitate | $20 \times 10^{-16}$ | $0.19 \times 10^4$ |

Assuming that the chromocytes do not contain cholesterol in esterform we were able to calculate the total spreading of the lipoids extracted by BEUMER and BURGER from 1 cc. blood of a sheep.

| | |
|---|---|
| Kephalin etc. | $0.42 \times 10^4$ sq. cm. |
| Cholesterol | $0.31 \times 10^4$ ,, |
| Sphingomyelin | $0.75 \times 10^4$ ,, |

[1]) BEUMER and BÜRGER, Biochem. Z., 1913, LVI, 446.

[2]) LEATHES found $114 \times 10^{-16}$ per molecule of lecithin.

We get the impression that the surface is covered by kephalin and a second layer is formed by cholesterol and sphingomyelin combined. Although it is impossible to know the exact numbers of the chromocytes and their size in the blood, that was extracted by BEUMER and BÜRGER, we can solidify this hypothesis by calculating the surface of one cc. blood from the mean values given by KLIENEBERGER [1]) (11.800.000 per sq. mm. and 4.3 $\mu$ as diameter) and one gets :

$$0.44 \times 10^4 \text{ sq. cm.}$$

It seems superfluous to make these calculations from other determinations in different publications, in which the amount of blood used for the extractions is unknown. Therefore we undertook to make ourselves these determinations in some experiments. The most simple appeared to be the exact determination of the cholesterol-content of an extract of blood, that had already served to the purpose of determining the total spreading value.

We made use of the Liebermanntest. The benzene solution of all the lipoids of the red blood cells or a Bloor-extract of the chromocytes of the same blood was evaporated on the waterbath just to dryness, and the warm residue taken up in chloroform. This chloroform-solution served directly to the colorimetric determination of the cholesterol.

All this experiments show conclusively that $^2/_5$ of the surface occupied by all the lipoids of the blood cells, is occupied by cholesterol.

It was impossible to work out a simple method for determining the other lipoid-constituents. Because sphingomyelin as well as kephalin (and lecithin)

| | Animal | Amount of blood | Total surface of the chromocytes (*a*) | Total surface of all the lipoids of the chromocytes (*b*) | Surface occupied by cholesterol of the chromocytes (*c*) | Factor *c* : *b* |
|---|---|---|---|---|---|---|
| | | cc. | sq.m. | sq.m. | sq.m. | |
| 55 | Cavia C | 1 | 0.53 | 1.02 | 0.4 | 0.39 |
| 56 | „ C | 5 | 2.65 | (5.1) | 2 | 0.39 |
| 57 | „ C | 3 | 1.59 | (3.06) | 1.2 | 0.39 |
| 58 | Rabbit D | 5 | 2.55 | 4.60[2]) | 2.1 | 0.45 |
| 59 | Sheep 2 | 10 | 4.19 | 8.07 | 3.4 | 0.42 |
| 60 | Dog B | 10 | 9.8 | 18.4 | 7.4 | 0.40 |
| 61 | Goat 2 | 5 | 1.6 | 3.2 | 1.3 | 0.40 |

[1]) KLIENEBERGER und CARL, Die Blut-Morphologie der Laboratoriums-Tiere, Leipzig, 1912.

[2]) This number probably too small.

both contain phosphorus and the spreadingvalue of these substances differs considerably, the determination of the total lipoid phosphorus would serve no good purpose. Some determinations however gave the result that in the assumption that half of the phosphorus was derived from sphingomyelin and the other half from kephalin, the total spreading was exactly explained but other experiments gave inconstant results.

We therefore only give the numbers of the cholesterol.

We want to add a few words on the different character of kephalin and lecithin on the one side and cholesterol and sphingomyelin on the other side. The first named substances are in the expanded condition [1]), the last mentioned in the condensed form at 37° C. It seems possible, that these properties counterbalance each other in a certain sense on the surface of the chromocytes.

We wish to thank Dr. LEVENE for kindly sending us a sample of kephalin and of sphingomyelin.

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[1]) ADAM, Proc. Roy. Soc. London, Series (A), 1922. CI, 516.