tion of treatment.

1. Herzog, H. L., Nobile, A., Tolksdorf, S., Charney, W., Hershberg, E. B., Perlman, P. L., and Pechet, M. M., *Science,* 1955, v121, 176.

2. Nobile, A., Charney, W., Perlman, P. L., Herzog, H. L., Payne, C. C., Tully, M. E., Jevnik, M. A., and Hershberg, E. B., *J. Am. Chem. Soc.,* 1955, v77, 4184.

3. Herzog, H. L., Payne, C. C., Jevnik, M. A., Gould, D., Shapiro, E. L., Oliveto, E. P., and Hershberg, E. B., *ibid.,* 1955, v77, 4781.

4. Bunim, J. J., and Pechet, M. M., *J. Am. Med. Assn.,* 1955, v157, 311.

5. Perlman, P. L., and Tolksdorf, S., *Fed. Proc.,* 1955, v14, 377.

6. Tolksdorf, S., and Perlman, P. L., *Endocrine Society Meeting,* June 2-4, 1955, 37th Meeting.

7. Speirs, R. S., and Meyer, R. K., *Endocrinology,* 1951, v48, 316.

8. Rosemberg, E., Cornfeld, J., Bates, R. W., and Anderson, E., *ibid.,* 1954, v54, 363.

9. Finney, D. J., *Statistical Method in Biological Assay,* 1952, Charles Griffin and Co., London.

10. Olson, R. E., Jacobs, F. A., Richert, D., Thayer, S. A., Kopp, L. J., and Wade, N. J., *Endocrinology,* 1944, v35, 430.

11. Pabst, M. L., Sheppard, R. S., and Kuizenga, M. H., *ibid.,* 1947, v41, 55.

12. Kahan, J., *Arch. Biochem. and Biophys.,* 1953, v47, 408.

13. Stephenson, N. R., *Canad. J. Biochem. and Physiol.,* 1954, v32, 689.

14. Stafford, R. O., Barnes, L. E., Bowman, B. J., and Meinzinger, M. M., PROC. SOC. EXP. BIOL. AND MED., 1955, v89, 371.

15. Ferrari, V., Ginoulhiac, E., Mosca, M. C., and Tenconi, L. T., *Minerva Medica,* 1955, v46, 51.

16. Huggins, C., Jensen, E. V., and Cleveland, A. S., *J. Exp. Med.,* 1954, v100, 225.

17. Higgins, G. M., Woods, K. A., and Kendall, E. C., *Endocrinology,* 1951, v48, 175.

18. Fried, J., and Sabo, E. F., *J. Am. Chem. Soc.,* 1953, v75, 2273.

19. Byrnes, W. W., Barnes, L. E., Bowman, B. J., Dulin, W. E., Morley, E. H., and Stafford, R. O., PROC. SOC. EXP. BIOL. AND MED., 1956, v91, 67.

20. Borman, A., Singer, F. M., and Numerof, P., *ibid.,* 1954, v86, 570.

21. Chart, J. J., Hetzel, N., and Gaunt, R., *ibid.,* 1956, v91, 73.

22. Nairn, R. C., Masson, G. M. C., and Corcoran, A. C., *Endocrinology,* 1955, v57, 700.

23. Hirschman, R., Miller, R., Beyler, R., Sarett, L., and Tishler, M. T., *J. Am. Chem. Soc.,* 1955, v77, 3166.

24. Swingle, W. W., Baker, C., Eisler, M., Le Brie, S. J., and Brannick, L. J., PROC. SOC. EXP. BIOL. AND MED., 1955, v88, 193.

25. Leatham, J. H., and Wolf, R. C., *ibid.,* 1954, v86, 724.

26. Migeon, C. J., Gardner, L. T., and Crigler, J. F., *Endocrinology,* 1952, v51, 117.

27. Halmi, N. S., and Barker, S. B., *ibid.,* 1952, v51, 127.

28. Bodansky, O., and Money, W. L., *ibid.,* 1954, v55, 173.

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Distribution of Fetal Hemoglobin in Layers by Centrifugation. (22430)

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Hemoglobin of the fetal blood is of 2 varieties, that can be differentiated by the alkali denaturation technic(1). At birth, 50 to 90% of hemoglobin is of the alkali resistant fetal type, while the remainder is of the normal adult type(2). It is not known, however, whether fetal and adult hemoglobins are distributed evenly inside the fetal red blood cells. In order to study this problem washed erythrocytes of the cord blood were centrifuged and fetal hemoglobin concentrations of the upper and lowest layers were compared.

Material and method. Two samples of blood, 20 cc each, were taken from the cord of 29 normal newborns and added to 0.05 cc of sodium heparin (Abbott, in which 10 mg

TABLE I. Fetal Hemoglobin % Concentration in Cord Blood following Centrifugation.

No.	Non-centrifuged sample	Centrifuged Upper layer	Centrifuged Lowest layer	Difference
1	76	68.8	86.1	17.3
2	75	66	82	16
3	55	49	62	13
4	62	50	73	23
5	58	47	72	25
6	68	58.5	79	20.5
7	80	70	91	21
8	75	66	88	22
9	71	63	81	18
10	89	76	97	21
11	78	69	88	19
12	84	74	91	17
13	85	76	92	16
14	90	79	98	19
15	81	70	91	21
16	74	60	83	23
17	80	70	89	19
18	65	59	83	24
19	78	66	82	16
20	91	80	97	17
21	72	60	84	24
22	81	72	87	15
23	89	80	92	12
24	69	58	79	21
25	87	75	92	17
26	82	71	86	15
27	76	69	89	20
28	85	75	89	14
29	89	77	95	18

= 1000 Toronto units per cc). One of the samples was used to determine the initial value of fetal hemoglobin percent concentration. The washed erythrocytes of the other sample were centrifuged in a graduated tube at 2000 rpm for 30 minutes. Fetal hemoglobin percent concentration was then determined in the upper and lowest layers of the washed erythrocytes with the alkali denaturation technic of Singer *et al.*(1).

Results. The results are shown in Table I. Statistically the differences are highly significant and increase inversely with the lowering of the initial values of fetal hemoglobin percent concentration. The Table shows that centrifugation distributes erythrocytes according to their fetal hemoglobin content. Thus, fetal hemoglobin percent concentration at the bottom of the centrifuged blood is distinctly higher than the initial fetal hemoglobin values. The difference is even more significant when fetal hemoglobin values in the upper and lowest layers are compared.

Comment. This finding may point to the possibility that either fetal and adult hemoglobins are contained in different erythrocytes, or that they are found in various proportions inside the fetal red blood cells.

The assumption that centrifugation of fetal erythrocytes distributes them in layers is also supported by our observation that mean corpuscular volume of the red blood cells at the bottom of the tube is distinctly larger than that of the upper layer.*

The higher concentration of fetal hemoglobin at the bottom of centrifuged blood may be of help in diagnosis of pathological conditions in which fetal hemoglobin blood content is only slightly increased.

Summary. Centrifugation distributes fetal erythrocytes in layers according to their fetal hemoglobin content. The fetal hemoglobin percent concentration is distinctly higher in the bottom layer of centrifuged blood.

* This finding will be reported in detail elsewhere.

1. Singer, K., Chernoff, A. I., and Singer, L., *Blood,* 1951, v6, 413.

2. Singer, K., *Yearbook of Pathology and Clinical Pathology,* Chicago, Yearbook Publishers, 1953-1954.

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