21. Granger, G. A., Weiser, R. S., Science, 1964, v145, 1427.
22. Gowans, J. L., Brit. Med. Bull., 1965, v21, 106.
23. Wilson, D. B., J. Cell. Comp. Physiol., 1963, v62, 273.
24. Lieberman, M., Kaplan, H. S., Science, 1959, v130, 387.
25. Gross, L., Acta Haematologica, 1958, v19, 353.
26. Schwartz, R. S., Beldotti, L., Science, 1965, v149, 1511.
27. World Health Organization Tech. Rep. 295, 1965.
28. Sachs, L., J. Nat. Canc. Inst., 1962, v29, 759.

Received April 18, 1966. P.S.E.B.M., 1966, v122.

Immune Globulin Levels in Colostrum and Breast Milk, and Serum From Fomula- and Breast-Fed Newborns. (31335)

ARTHUR J. AMMANN AND E. RICHARD STIEHM* (Introduced by C. F. Piel)
Department of Pediatrics, University of California-San Francisco Medical Center, San Francisco

Breast milk, particularly colostrum, has significant quantities of antibody-containing immune globulins. The antibodies secreted by the breast are chiefly in the γA globulin and parallel the elevated levels of γA globulin in colostrum or breast milk(1).

Studies indicating absorption of intact proteins(2) and antibodies(3) from the gastrointestinal tract led us to reinvestigate whether colostral or breast milk immune globulins, particularly γA, are absorbed intact to any significant degree. We, therefore, compared the serum concentration of γG, γM, γA of breast-fed and formula-fed infants and determined the concentration of immune globulins in colostrum and breast milk.

Methods. Cord serum was obtained from 10 full-term infants who were subsequently breast-fed and from 10 full-term infants given formula feedings. A second serum sample was obtained at 4 days from each infant. During this period the mean weights of the breast-fed infants fell from 3280 to 3160 g and of the formula-fed infants from 3100 to 3060 g. Samples of breast milk from the mothers of these infants, obtained during the first 24 hours after delivery and for 3 days thereafter, were frozen immediately and stored at −10°C.

Immune globulin levels were measured quantitatively by the radial diffusion technic of Mancini and associates(4) with specific antiserum to γG, γM and γA globulin incorporated into agar plates. Purified immune globulins quantified by Kjeldahl nitrogen analysis were used as standards. In this laboratory, the method has an error of 3% when samples are tested in duplicate on the same plate or 10% when duplicate samples are tested on different plates. In adults, the normal values by this method(5) are as follows:

γG	1158 mg/100 ml	± 305
γM	99 mg/100 ml	± 27
γA	200 mg/100 ml	± 61

The student T test was used for statistical comparison.

Results. Fig. 1 illustrates the changes in immune globulin concentration in breast secretions during the first 4 days of transition from colostrum to mature breast milk.

The mean values (ml/100 ml) for initial colostrum (γG 43, γM 159 and γA 1735) compared to fourth-day post-partum breast milk (γG 4, γM 10 and γA 100) indicate a high content of γA in colostrum and a small amount of γG globulin.

The immune globulin levels of formula-fed and breast-fed infants were not significantly different (Table I) nor were there significant differences between cord and 4-day serum. Only 2 infants demonstrated detectable γA in cord and 4-day serum.

Assuming a 25% intravascular distribution (6) and a plasma volume of 50 ml/kg, absorption of 60 mg γA globulin from the gas-

* Present address: Dept. of Pediatrics, Univ. of Wisconsin School of Med., Madison.

TABLE I. Serum Immune Globulin Levels (mg/100 ml).

	Formula-fed				Breast-fed			
	Cord		4-day serum		Cord		4-day serum	
	Mean	Range	Mean	Range	Mean	Range	Mean	Range
γG	1112	720–1296	1000	336–1553	1128	791–1559	1138	902–1331
γM	13	8– 19	15	10– 31	10	6– 14	11	7– 14
γA*	—	0– 3	—	0– 4	—	0– 3	—	0– 3

* Only 1 case had detectable levels.

trointestinal tract in a 3 kg newborn infant would elevate the serum γA concentration to 10 mg/100 ml, a readily detectable level. Even without the high γA level in colostrum, many times this amount of γA would be ingested by a breast-fed newborn in the first days of life. Hence, large quantities of γA globulin do not appear to be absorbed. In contrast, the γG and γM content of breast milk, even if completely absorbed, is too low to detect by the method of immune globulin analysis.

Studies in other species have demonstrated protein absorption: the calf, which has no serum γ-globulin at birth absorbs colostral γ-globulin and other proteins in the first 24 hours of life(2), and the rat absorbs proteins for a longer period of time(7).

The results of previous studies in human neonates suggest that certain antibodies may be absorbed from the gastrointestinal tract. These include anitbodies to *E. coli,* salmonella, tetanus, diphtheria and isoagglutinins. Discordant results have been reported on the absorption of colostral Rh, staphylococcal and vaccinia antibodies(3).

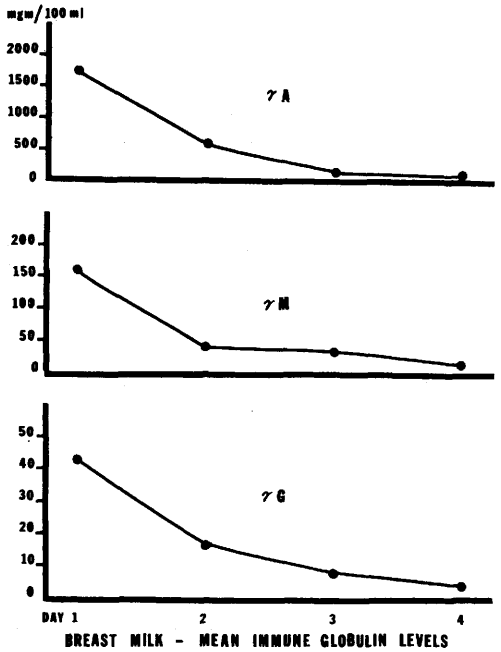

FIG. 1.

This variation may be attributed to host factors, such as the presence of disease, individual variations in gastrointestinal maturity, motility or proteolytic enzyme concentration, as well as to the physicochemical characteristics and amount of the protein ingested. If small molecules are preferentially absorbed, the immune globulins γM (m.w. 1,000,000) and colostral or breast milk γA (m.w. 320,000) may not be absorbed as well as serum γA or γG globulin (m.w. 160,000)(8,9).

It is probable that the prime role of the immune globulin in breast milk is to inhibit viral and bacterial multiplication within the gastrointestinal tract. Indeed, breast feeding has prevented successful oral polio immunization(10).

Summary. Immune globulin levels in colostrum and breast milk determined during the first 4 days of nursing show high levels in colostrum, especially of γA globulin, followed by a rapid decline with transition to mature breast milk. After 4 days the immune globulins of breast-fed infants were similar to those of formula-fed infants. This would indicate failure of intact immune globulins to be absorbed by the gastrointestinal tract in sufficient amounts to affect immune globulin levels during this period of life.

1. Chodirker, W. B., Tomasi, T. B., Jr., Science, 1963, v142, 1080.

2. Pierce, A. E., Risdall, P. C., Shaw, B., J. Physiol., 1964, v171, 203.

3. Leissring, J. C., Anderson, J. W., Smith, D. W., Am. J. Dis. Child., 1962, v103, 160.

4. Mancini G., Vaerman, J. P., Carbonara, A. O., Heremans, J. F., in XIV Colloquium, Protides of the Biological Fluids, Peeters, H., ed., Elsevier Publishing Co., Amsterdam, 1964, 370.

5. Stiehm, E. R., Fudenberg, H. H., Pediatrics, 1966, v37, accepted for publication.

6. Stiehm, E. R., Vaerman, P., Fudenberg, H. H., J. Pediatrics, 1966, v68, accepted for publication.

7. Bangham, D. R., Tan, E. M., Solomon, A., v66, 579.

8. Tomasi, T. B., Jr., Tan, E. M., Solomon, A., Prendergast, R. A., J. Exp. Med., 1965, v121, 101.

9. Tomasi, T. B., Jr., Blood, 1965, v25, 382.

10. Warren, R. J., Lepow, M. L. Bartsch, G. E., Robbins, F. C., Pediatrics, 1964, v34, 4.

Received April 18, 1966. P.S.E.B.M., 1966, v122.

Delay of Hereditary Muscular Dystrophy of the Chicken by Oxygen Therapy.*† (31336)

C. ROBERT ASHMORE‡ AND RALPH G. SOMES, JR. (Introduced by Heinz Herrmann)
Department of Poultry Science, University of Connecticut, Storrs

The occurrence of a hereditary muscular dystrophy in the chicken with features similar to that of man has been reported by Asmundson and Julian(1). This muscle abnormality prevents the birds from raising their wings or returning to a standing position when placed on their backs on a flat surface. Chung *et al*(2) concluded that the exhaustion number, which is the number of times a bird can be placed on its back until it can no longer rise, is the best means of early detection of the abnormality.

Many authors have reported biochemical and histochemical alterations of dystrophic tissues. Our laboratory has shown that in 8-week-old, genetically-dystrophic chicks, reduced myoglobin (myoglobin without oxygen) in muscle tissue extracts is approximately 35% higher relative to normal controls. Schapira and Dreyfus(3) have reported a diminution in oxygen consumption in human dystrophic muscle. They further report that many female carriers show an apparent acceleration of the peripheral circulation time. These facts suggested to us the possibility that muscle cell death may be initiated by chronic oxygen starvation.

* This investigation was supported, in part, by a grant from the University of Connecticut Research Foundation.

† Scientific Contribution No. 194, Agri. Exp. Station, Univ. of Connecticut, Storrs.

‡ N.I.H. Predoctoral Fellow, 7-F1-CM-20, 128-O1A1.

Materials and methods. In a preliminary experiment (Experiment I, Table I), 5 chicks with hereditary muscular dystrophy were placed in a high oxygen environment at 1 day of age and maintained there until 28 days of age. Five dystrophic controls from the same hatch and same parents were maintained in an adjacent cage, but without added oxygen. A standard chick starter ration and water were fed to both groups *ad libitum*.

Both cages were about the same size, having approximately 7 square feet of floor space per cage. The cage, which housed the oxygen-maintained birds, was constructed of wood with the top and front covered with polyethylene. In order to allow free circulation of the oxygen, which was admitted *via* a hose from an oxygen tank, and to allow escape of carbon dioxide, the cage was perforated along the bottom. The cage containing the dystrophic controls was a conventional wire battery brooder cage. In both cages, all birds had free access to food and water. There was no noticeable difference in activity between the oxygen-maintained birds and the controls at any time.

Unfortunately, during this preliminary experiment, the oxygen concentration inside of the experimental cage was not monitored. However, oxygen was admitted at a constant rate of 1 liter/cu ft/min throughout the 28-day period of the experiment. Temperatures were generally the same in both cages, although occasional differences were noted.