

IgM, Not 7S-IgM, as Cause of Increased Serum IgM Concentrations in Goiter Patients (36142)

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Circulating concentrations of immunoglobulin-M (IgM) are increased in about half of patients with endemic or sporadic nontoxic goiter (1). In the single radial immunodiffusion technique used to test those serums (2), the diameter of the precipitin ring developed against the anti-IgM serum in the agar is proportional to the logarithm of the amount of IgM in the well.

Our attention has been called by Dr. William D. Terry of the National Institutes of Health to the possibility that a 7S-subfraction of IgM first identified in some patients with systemic lupus erythematosus (3), termed 7S-IgM, might produce large rings in the agar plates because of the more rapid spread than that of the intact IgM antibody. As a result, spuriously high estimates of IgM concentrations would be made. To exclude the presence of 7S-IgM, therefore, we have reexamined several serums previously determined to have elevated IgM concentrations.

Nine serums were tested: 4 from patients with endemic nontoxic nodular goiter [Patients 1, 2, 4 and 5 (Table I)]; 2 with sporadic goiter (Patients 6 and 7); and 3 with toxic diffuse goiter (Patients 3, 8 and 9). Serum IgM concentrations have been reported by Mahaux *et al.* to be increased (4) in patients with active Class 2 through 6 eye changes of Graves' disease. (5).

To distinguish 7S-IgM from intact IgM, sucrose density gradient ultracentrifugation (6) and Sephadex G-200 column chromatography were employed. In the former procedure (6) effluent fractions were collect-

ed and protein and IgM concentrations of every second tube through tube 14 determined, using 0.02 cc of the effluent fractions by Folin method (7). The tubes were then combined into three fractions. The first fraction contained only 19S material, the third fraction 7S material, and the second fraction a mixture of the two.

IgM was determined by the technique of single radial immunodiffusion (2) on plates obtained commercially from Kallestad Laboratories.

Sephadex gel filtration using Sephadex G-200 (8) was carried out in columns 75×2.7 cm, with 0.5–1.0 ml serum applied to the column. The effluent was passed through an LKB Uvicord scanner and pooled into four major fractions consisting of the first protein peak, the fraction between the first and second protein peaks, the second peak, and the albumin peak. Fraction 1 contained 19S material, fraction 3, 7S material, and fraction 2, a mixture of the two. Fractions were concentrated to a final volume of 1–2 ml under nitrogen and IgM concentrations determined by single radial immunodiffusion.

IgM concentrations in the fractions tested have been expressed in a 0–4+ basis in view of the obvious qualitative nature of the procedure. In 1 of the 9 samples tested by the sucrose gradient method (Table I), a positive result (++) was found in the 7S fraction but not with the column procedure (Patient 2, Table I); oppositely, a different sample (1 of 6 so tested) was positive (++) by gel filtration but not by sucrose gradient (Patient 4, Table I). Otherwise, all samples were positive for 19S IgM only.

In summary, in the small sampling of serums studied, the increase in IgM concentrations previously found in serums from endemic and sporadic goiter appears to be due to intact 19S IgM and not to the presence of

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TABLE I. IgM Concentrations in Fractions from Sucrose Gradient Ultracentrifugation and Sephadex Column Chromatography.

	Patient no.								
	1	2	3	4	5	6	7	8	9
Tube no.	Sucrose gradient ^a								
2	+	++	++++	++	++	++	+	++	+
4	0	+	++	+	0	0	+	+++	++
6	0	0	+	0	0	0	+	+	++
8	0	0	0	0	0	0	0	0	0
10	0	0	0	0	0	0	—	—	—
12	0	0	0	0	0	0	—	—	—
14	0	0	0	0	0	0	—	—	—
Combined tubes ^b									
1-4	++++	+++	+++	+	+	+			
5-6	+	+	0	0	0	0			
7-19	0	++	0	0	0	0			
Fraction no. ^c	Sephadex column chromatography ^a								
1	++	+++	++++	+	+	+			
2	0	+	+	0	0	0			
3	0	0	0	++	0	0			
4	0	0	0	0	0	0			

^a + = 50-149 mg/100 ml; ++ = 150-299 mg/100 ml; +++ = 300-449 mg/100 ml; ++++ = 450- mg/100 ml.

^b Tubes 1-4 incl. 19S IgM, 5 and 6 mixed 19S and 7S IgM, 7-19 7S IgM.

^c Fraction 1 19S IgM, fraction 2 mixed 19S and 7S IgM, fraction 3 7S IgM, fraction 4 albumin.

a subfraction of IgM, 7S-IgM. Similarly, serums from several patients with toxic diffuse goiter with increased serum IgM concentrations were also shown to contain 19S IgM and not 7S-IgM.

1. Werner, S. C., Bora, S., Koutras, D. A., and Wahlberg, P., *Science* **170**, 1201 (1970).

2. Fahey, J. L., and McKelvey, E. M., *J. Immunol.* **94**, 84 (1965).

3. Rothfield, N. F., Frangione, B., and Franklin,

E. C., *J. Clin. Invest.* **44**, 62 (1965).

4. Mahaux, J. E., Delcourt, R., Nagel, M., Chamla-Soumenkoff, J., Dourov, N., and Levin, S., *Rev. Med. Bruxelles* **25**, 485 (1969).

5. Werner, S. C., *J. Clin. Endocrinol. Metab.* **29**, 982 (1969).

6. Edelman, G. M., Kunkel, H. G., and Franklin, E. C., *J. Exptl. Med.* **108**, 105 (1958).

7. Lowry, O. H., Rosebrough, N. J., Farr, A. L., and Randall, R. J., *J. Biol. Chem.* **193**, 265 (1951).

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