1/4 inch in diameter were placed as the core of a pair of coils wound one upon the other. Electric pulses of 5  $\mu$ sec duration were fed into the primary coil. The secondary coil was connected to an oscilloscope. The procedure is similar to the method described by C. P. Bean et al. (2) for measuring the resistivity ratio in metals. With no core or an insulating one inside the coils, the trailing edge of the pulse in the secondary is sharp and steep, like the trailing edge of the primary pulse. In a metallic core, the primary pulse induces eddy currents whose decay rate determines the decay of the secondary pulse. If the metal of the core becomes superconducting, the curve resembles again that of an insulator, since practically no eddy currents are induced.

Figure 1 shows the decay curve for a sample of metallic InSb in the normal conducting state at 4.2°K, indicating a very good conductivity comparable to that of tin or indium at the same temperature. Figure 2 shows the decay curve for the same sample after it has become completely superconducting.

The superconducting transition started at about 2.1°K and was complete



Fig. 1. Decay curve at secondary coil with sample of metallic InSb as a core at a temperature of 4.2°K. Decay is indicative of a sample of very high conductivity, comparable to that of pure tin or indium above their transition temperatures. The vertical scale shows voltage, the horizontal shows time.



Fig. 2. Decay curve at secondary coil with same sample at 1.3°K after complete transition to the superconducting state.

at about 1.6°K. The broadness of the transition is not surprising, considering that the samples are presumably heavily strained. Darnell and Libby report that the structure of metallic InSb is practically identical with that of tin (1). The fact that the observed superconducting transition temperature is so close to that of tin seems to be another confirmation of this fact (3).

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## Synthesis of Chicken Antibodies of High and Low Molecular Weight

Abstract. Two major populations of chicken macroglobulin antibodies in primary- and secondary-response serums are associated with electrophoretically slow- and fast-moving y-globulins. If the fractions separable by electrophoresis are treated with 2-mercaptoethanol, two populations of 7S antibodies are revealed. Primary-response precipitins have the characteristics of macroglobulins.

The synthesis of high concentrations of macroglobulin antibodies (19S)class) in the early phases of antibody production is often followed by the synthesis of increasing amounts of antibodies of low (7S class) molecular weight (1). Generally, 19S antibodies are characterized as  $\gamma_1$  or  $\beta$ -globulins of high anionic binding, which are easily dissociated by sulfhydryl reagents (2). The 7S antibodies are usually associated with the slower-moving globulins of low anionic binding and are more resistant to sulfhydryl reagents. Recently we reported that in the chicken at least three chromatographically distinct macroglobulin antibodies are produced after a single injection of bovine serum albumin (BSA) and that these antibodies remain at relatively high concentrations even after a second exposure to antigen (3, 4). In contrast, the ratio of 19S to 7S antibodies in rabbits drops rapidly in a short period of time after a single injection of BSA, and the conversion from 19S to 7S responses is accompanied by changes in electrophoretic patterns (1). In the present study, the electrophoretic patterns of anti-BSA chicken antibody obtained after a single injection, or after repeated injections, of BSA were similar. This similarity resulted from the initial synthesis of two populations of 19S antibodies which were replaced by 7S antibodies of similar mobilities.

Primary antibody responses were induced in adult Austra hens by a single intravenous injection of 40 mg of BSA (Pentex), and secondary responses were induced by a second similar injection 3 weeks later. Booster serums were obtained by giving four intramuscular injections daily (10 mg of BSA each), 3 to 4 weeks after the secondary injection. Individual and pooled serums, inactivated by heat and adsorbed with sheep red blood cells, were fractionated by starch-block electrophoresis (5). The time of bleeding the hens for serum for electrophoretic analysis was selected on the basis of previous observations (4) that high concentrations of macroglobulin hemagglutinins (HA) were produced 6 days after immunization. Fractions eluted after electrophoresis were assayed for antibody by the tanned-cell method (6).

Hemagglutinins in all serums were found throughout the  $\gamma$ - and  $\beta$ -globulin regions, with some activity associated with the  $\alpha$ -globulins; however, two major antibody peaks were evident (Fig. 1). The  $\gamma_2$  peaks were sharp, and the faster-moving peaks were broad. Most primary serums had activity in the fast-moving fractions (Nos. 25 to 29), and secondary and hyperimmune serums had little or no activity in these fractions. Nevertheless, delineation of primary and hyperimmune serums from the gross patterns was difficult

The residual activities of the fractions after treatment with 0.1M 2-mercaptoethanol (ME) for 24 hours (2) are shown in Fig. 1. Most of the primary hemagglutinins were inactivated. One serum had a small amount of resistant activity in the slowest-moving fraction, and the other primary serum shown had two ME-resistant peaks. Secondary serums had less MEsensitive activity and two reproducible ME-resistant peaks were resolved; the

slowest peak was predominant. Most of the dissociated hyperimmune antibodies were located in rapidly migrating fractions (Nos. 19 to 24).

Fractionation of electrophoretic fractions by zone ultracentrifugation in sucrose gradients (7) revealed that the major peaks of primary fractions were 19S antibodies, except for a small amount of 7S antibody in the slowestmoving fractions, which corresponded to the ME-resistant hemagglutinin. The relative concentrations of 19S and 7S activities were approximately the same as had been obtained by exposure to 2-mercaptoethanol. As with ME treatment, ultracentrifugal separation of hyperimmune electrophoretic fractions showed that the main peaks were 7Santibody except in the most rapidly migrating fractions.

It is evident that chickens synthesize a heterogeneous population of macroglobulin antibodies which may be separated chromatographically (3, 4), and which occur in both fast- and slowmoving  $\gamma$ -globulin fractions; therefore, the generalization cannot be made in this instance that the macroglobulin antibodies appear only in the faster-



Fig. 1. Primary, secondary, and hyperimmune chicken antibodies to bovine serum albumin fractionated by starch-block electrophoresis. Shaded areas represent loss of activity caused by the addition of 0.1M mercaptoethanol. Primary (top) and hyperimmune serums electrophoresed simultaneously. Circles, untreated; squares, treated with 0.1M mercaptoethanol; triangles, protein.

moving fractions. Furthermore, Dreesman (8) in our laboratory has shown that in the turkey primary anti-BSA serums similarly fractionated had double peaks of antibody corresponding to two 19S activities, and that the slowest peak also contained small amounts of 7S antibody. Apparently the chicken  $\gamma_2$  7S antibody is synthesized earlier than the  $\gamma_1$  7S antibody, but after repeated stimulation by antigen there is synthesized a concentration of the faster-moving antibody equal to or greater than the slower-moving antibody. Ovary and Benacerraf (9) also reported that the slower-migrating 7S guinea pig antibodies appeared earlier during immunization. The fastest-moving macroglobulins which persist in hyperimmune serums account for the small amount of 19S antibody reported in chromatographic and ultracentrifugal fractions of these serums (3, 4).

It is difficult to reconcile these and earlier results (3, 4), with the report that chicken serum macroglobulin is a nonantibody coprecipitating factor (10). In this connecton, immunoelectrophoresis of primary serums with BSA added to the trough give short fuzzy precipitin bands extending out from the origin in the  $\beta$ -globulin region, and resemble the precipitin band given by macroglobulins ( $\beta_{2M}$ ). Hyperimmune serums give distinctly different bands characteristic of the long  $\gamma$ -globulin arc. By indirect methods, similar results have been obtained (11). Analysis of fractions obtained by gradient ultracentrifugation suggests that primary serums contain 7S nonhemaggluti-"univalent" (11) and 19S nating hemagglutinating (precipitating) antibodies, and that hyperimmune serums contain mostly 7S hemagglutinating (precipitating) antibody. The selective precipitation of these antibodies by high and low salt concentrations will be reported elsewhere. In quantitative precipitin experiments, with either 0.15 or 1.5M NaCl concentrations and 0.01M EDTA buffer solution in the reaction mixtures, mercaptans inactivated primary precipitins and only partially inactivated hyperimmune precipitins. Furthermore, dissociation products of ME-treated primary serums inhibited specifically the homologous precipitating system. For example, Table 1 shows that considerable inhibition occurred in the zones of antibody excess and of equivalence if the antigen was incubated for 20 min with 2.0 ml

Table 1. Inhibition of the BSA-chicken primary anti-BSA precipitin reaction by homologous primary serum treated with mercaptoethanol.

]	BSA N added (mg)	Total N precipitated (mg/ml serum)	
		In absence of treated serum	In presence of treated serum
	0.014	0.110*	0.018
	.028	.188†	.040
	.056	.183‡	.088
	.112	.172	.100
	.168	.168	.137
	.280	.164	.136

Antibody excess. † Equivalence. **†** Antigen excess.

of ME-treated primary serum before the addition of the homologous antiserum. The concentrations of NaCl (1.5M) and EDTA (0.01M) were kept constant. The precipitation of and the homologous antigen was not similarly inhibited. Finally, the addition of normal serum, which presumably contains normal coprecipitating macroglobulins, did not restore precipitating ability to dissociated primary antibody. It is clear that in studies with chicken antibody consideration must be given to the duration of immunization. Although there is a change in response from a 19S to 7S antibody, high levels of 7S anti-BSA hemagglutinins are reached slowly and usually require repeated stimulation by antigen (12).

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