the chromosomes of the developing female mammalian germ cell. The larger bivalents are altered most consistently, but no definite parallel with the work on human leukocytes, in which the pericentric regions and secondary constrictions of group-A chromosomes were broken (*2*), has been demonstrated. Bennett (*6*) has confirmed the presence of secondary constrictions in mouse chromosomes 7, 18, and 20, and it is conceivable on morphologic grounds that one of the frequently altered pairs in the experiments that I have described is No. 7. How Streptonigrin achieves discontinuity and disorganization of female meiotic chromosomes, that have already completed synthesis of DNA (*7*), is not elucidated by these experiments, but, as a tool for studying agents that may produce mutations by acting on intrafollicular ova, the compound and the techniques seem very promising.

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Deficient Complement Fixation by Aggregated Gamma Globulin from Hypogammaglobulinemic Patients

Abstract. *We found that several normal samples of aggregated gamma globulin consistently fixed essentially equal amounts of guinea pig complement, while samples of gamma globulin from three of four hypogammaglobulinemic patients were markedly deficient in this biologic function. Thus, the previously observed limited heterogeneity of gamma globulin in patients with hypogammaglobulinemia is also associated with inefficiency in at least one biological function.*

Hong and Good (*1*) demonstrated that the gamma globulin (IgG) isolated from patients with hypogammaglobulinemic syndromes showed less electrophoretic heterogeneity, variable mobilities, and differences in dye-binding capacity, compared to IgG from normal serum. We find significant deficiency in the complement-fixing capacity of heat-aggregated IgG obtained from the serum of hypogammaglobulinemic patients. Thus, these patients have qualitative as well as quantitative abnormalities of IgG.

By column chromatography (*2*), IgG was isolated from four patients with hypogammaglobulinemia and from 11 normal individuals. Samples of IgG (0.25 and 0.5 mg) in 0.5 ml of 0.15*M* sodium chloride were aggregated by heating at 63°C for 20 minutes (*3*). Complement studies were made with only slight modification of the methods of Mayer (*4*). Guinea pig complement was diluted in barbital-buffered saline containing calcium ($1.5 \times 10^{-4}M$) and magnesium ($5 \times 10^{-4}M$) to a concentration which would provide approximately 30 hemolytic units of complement ($C'H_{50}$) units in 1.0 ml. Samples of 0.5 ml of this complement preparation were added to tubes containing 0.5 ml of the aggregated IgG, thereby making a final volume of 1.0 ml. The mixtures were then incubated at 37°C for 90 minutes, and the remaining complement activity was determined (*4*). The number of units of complement fixed was obtained by subtracting the residual units in the samples reacted with IgG from the number of units in controls incubated at 37°C for the same period of time with 0.5 ml of 0.15*M* sodium chloride lacking IgG. Determinations were done in duplicate.

In repeated examination of normal serums, we found that IgG from individual samples, heat-aggregated in the manner described, showed consistent complement-fixing ability. Results of our study of normals are shown in Table 1. In all instances the unaggregated IgG fixed less than 1.7 units.

In contrast to observations with normal IgG, studies of the complement-fixing ability of aggregated IgG from patients with hypogammaglobulinemia revealed clear differences. The serums of three of the four patients consistently showed inadequate complement-fixing capacity. The aggregated IgG from patient S on two occasions fixed 3.6 and 5.0 $C'H_{50}$ units out of 16, while normal aggregated IgG (studied simultaneously) fixed 11.4 to 13.4 units. Samples of IgG from two other patients fixed 5.5 to 7.5 $C'H_{50}$ units. The IgG from the fourth patient on several occasions fixed amounts of complement equal to or slightly less than normal. In contrast to the others, this individual has never experienced undue susceptibility to infection. His quantitative immunoglobulin abnormality was discovered in a family study in which the propositus was an acquired agammaglobulinemic. The data also show that IgG preparations from these hypogammaglobulinemic patients, unlike similar preparations from normal serum, vary widely from individual to individual in complement-fixing ability (Fig. 1).

Investigations by Ishizaka and Ishizaka (*3*, *5*) have shown conclusively that the complement-fixing (or inactivating) and skin-reactive properties of aggregated human γ-globulin, regardless of the method of aggregation, are essentially identical with those of antigen-antibody complexes. For this reason, we used heat aggregation of our IgG samples to study one aspect of their biologic activity.

Our study revealing that the isolated IgG from some patients with hypogammaglobulinema differs from normal IgG in at least one biologic function and the finding of immunochemical variations from normal in the same IgG samples (*1*) support the thesis that there is a deficient gene product in some of these patients. The data suggest that at least three of the patients we studied are deficient in ability to synthesize a portion or portions of the total IgG population which provides complement-fixing sites when heat-aggregated. The electrophoretic study of the IgG from these same patients shows a limited heterogeneity; this also suggests the presence of only limited segments of the total normal IgG population (*1*). The limited complement-fixing ability of the IgG from our patients might be explained by the absence of a group or groups of IgG molecules which carry complement-fixing sites

Table 1. Complement fixation by heat-aggregated normal human IgG. Eleven samples were studied.

IgG (mg/ml)	Total $C'H_{50}$ units	$C'H_{50}$ units fixed	Mean $C'H_{50}$ units fixed (%)
0.5	16.0 ± 0.5	13.4 ± 1.0	83.6
0.25	16.0 ± 0.5	8.3 ± 1.3	51.8

or the presence of an IgG population which lacks large numbers of these sites, or both. It is also possible that the defect in complement fixation is due to incomplete aggregation of the IgG of the patients. However, this is also consistent with the interpretation that there is a qualitative difference in their IgG which results in the biological variance. Under the conditions used there was no visible evidence of aggregation either in normal samples or in those of the patients. The difficulty of obtaining adequate amounts of isolated IgG from the patients precluded testing for aggregation by other methods.

An interesting and significant parallel for such electrophoretic and functional differences in IgG subgroups is present in the guinea pig. In this species, two distinct types of IgG, one "fast" (γ1) and one "slow" (γ2), were shown by electrophoresis. Each differs in several biologic functions (*6*); for example, the γ1 has been shown not to fix complement, while the γ2 subclass does (*6*). In addition, γ1 antibodies are capable of sensitizing guinea pig lung for antigen-induced histamine release; γ2 antibodies are not capable of this (*6*). Furthermore, populations of IgG molecules differing in heavy-chain antigenic groups vary in biological activity. Terry (*7*) has shown that the γ2a (Ne) subgroup of myeloma proteins do not fix to skin. Müller-Eberhard (*8*) found deficient interaction between myeloma proteins of the γ2d (Ge) and γ2a (Ne) subgroups with C'1q.

Apparently, individual γ-globulin molecules are not biologically equivalent, and the total biological capacity of the humoral antibody system is probably a function of normal population distribution.

Alterations in normal population ratios might result in qualitative defects not explainable on a quantitative basis. While this may be the result of faulty genetic information, the point at which the disturbance in synthesis occurs cannot be defined from these data.

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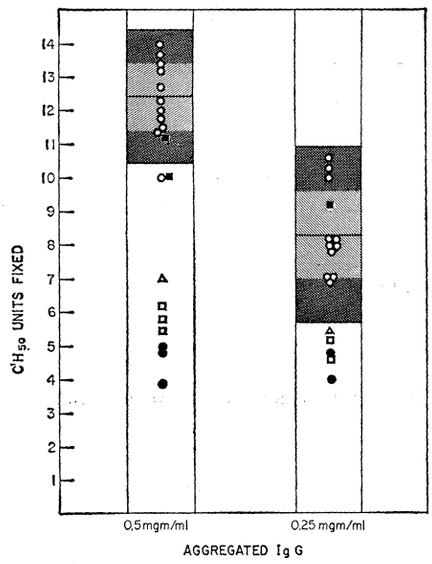

Fig. 1. Number of $C'H_{50}$ units of guinea pig complement fixed from a total of 16 $C'H_{50}$ units by heat-aggregated human IgG. The shaded area indicates a range of two standard deviations from the normal mean based on studies of 11 normals. S.W., S, H.X., and H are four patients with hypogammaglobulinemia. Normal, ○; S, ●; H.X., □; S.W., ■; H, △.

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Reward and Learning in the Goldfish

Abstract. *An experiment with goldfish showed the effects of change in amount of reward that are predicted from reinforcement theory. The performance of animals shifted from small to large reward improved gradually to the level of unshifted large-reward controls, while the performance of animals shifted from large to small reward remained at the large-reward level. The difference between these results and those obtained in analogous experiments with the rat suggests that reward functions differently in the instrumental learning of the two animals.*

The central fact of instrumental learning is that behavior can be controlled by manipulation of its consequences. If a hungry rat is rewarded with a pellet of food for pressing a retractable bar each time the bar is introduced into its cage, it comes to press the bar more and more readily as training proceeds. Large rewards are more effective than small rewards; the animal responds more promptly when, say, ten pellets are given for each press than when only one pellet is given. How does reward work?

One early answer to this question was that reward acts directly to strengthen or "reinforce" connections between contiguously active sensory and motor centers in the brain (as between the centers activated by the introduction of a bar and the motor center for pressing the bar). Performance is better with large rewards than with small rewards because large rewards produce stronger connections. Another early answer was that the animal learns *about* reward—that its increasing readiness to respond as training continues reflects a growing anticipation of reward. Differences in level of performance produced by differences in amount of reward reflect, in this view, differences in the attractiveness of the rewards that are anticipated, the requisite neural connections being assumed to develop as a function of contiguity alone. Although the reinforcement interpretation was widely accepted for a time, contemporary learning theorists are virtually unanimous in preference for the contiguity interpretation (*1*). Of the various experiments whose results dictate this preference, one of the most important is Crespi's (*2*).

Suppose that two groups of rats are