CHAPTER 15

GENETICS AND IMMUNOLOGY

Both genetics and immunology deal with highly specific and very numerous reacting substances, and in both fields, efficient methods are available for analyzing the effects of these substances individually. It was, therefore, of considerable importance when the two approaches began to be applied to the same system. So far as genetics is concerned, this joint attack led to some of the first hopeful advances in the study of the manner of action of genes.

The beginning of this interaction dates from the discovery of the human blood groups by Landsteiner (1900). Early attempts at the clinical use of blood transfusion had frequently led to disastrous effects on the patient, so the practice had been given up, until after the work of Landsteiner and his followers was widely known and understood.

Landsteiner knew of the occurrence of reactions when blood of different species was mixed, and so he was led to look for differences between the bloods of individuals of the same species. He separated the red cells and the serum from the bloods of a number of human subjects and made cross-tests. He found that, in many combinations, such mixtures led to the agglutination of the red cells, and he recognized three different kinds of individuals. In 1901 he showed that there are two kinds of *agglutinins* (α and β) in the sera, and two kinds of *agglutinogens* (A and B) on the cells. When the fourth possible type of individual was found by Decastello and Sturli in 1902, the now familiar "ABO" blood-group system emerged. There were, however, several different systems of naming the groups, which were confusing at the time and still make some of the earlier literature difficult to follow. Table 2 shows the relations:

TABLE 2

Current Designation	Jansky System	Moss System	Agglutinogens on Cells	Agglutinins in Serum
O	I	IV	None	α, β
A	II	II	A	β
B	III	III	B	α
AB	IV	I	A, B	None

It was suggested by Ottenberg and Epstein (1908) that the blood groups are inherited, but the family data of these blood groups were not extensive enough to be convincing. However, von Dungern and Hirszfeld (1910) did succeed in demonstrating the point; they showed that any agglutinogen present in an individual was present in at least one of his parents (A and B are each dependent on a single dominant gene). They supposed that these two genes were independent in inheritance, as their data indicated. On this basis, Group O had the composition *aa bb*; Group A included both *AA bb* and *Aa bb*; Group B included *aa BB* and *aa Bb*; Group AB was of four kinds—*AA BB*, *Aa BB*, *AA Bb*, or *Aa Bb*.

That this view was incorrect finally became evident as an outgrowth of the observations of L. and H. Hirszfeld (1919). The Hirszfelds were army physicians in the Balkans during World War I and determined the blood groups of large numbers of soldiers of diverse races and nationalities in the armies there. They found clearly significant differences in the relative frequencies of the four groups among the sixteen races and nationalities studied, thus beginning the use of blood-group determinations in the study of population problems.

As the data from various populations accumulated, it was natural to compare them with the proportions expected from the algebra of random mating populations (see Chapter 17). I am certain that I was not the only one to realize that these proportions were not in agreement with the von Dungern-Hirszfeld scheme; but this did not seem surprising, since the algebraic analysis assumed random mating; and it is obvious that large human populations do not mate at random, for economic, religious, and geographical reasons.

The discrepancy led Bernstein (1925) to try other genetic interpretations, and he found that a system of triple alleles with only one locus concerned did give equilibrium frequencies in agreement with the ob-

served frequencies, in populations with very different absolute frequencies. According to this scheme, the four groups have the following genotypes: O is *OO,* A is *AA* or *AO,* B is *BB* or *BO,* AB is always *AB.*

There was a serious difficulty here, as Bernstein recognized: On this scheme AB and O can never be related as parent and offspring, yet the published pedigrees included many examples of such relationships.

In 1929 Snyder presented a summary of the published records for the offspring of one of the critical matings—that between O and AB parents. If these are divided into two groups, those published before and those after Bernstein's paper, the totals are as shown in Table 3.

TABLE 3. OFFSPRING RECORDED FROM MATINGS BETWEEN O AND AB

Published Records	O	A	B	AB	Number of Papers
Von Dungern and Hirszfeld, 1910	2	2	2	3	1
All authors up to 1925 (including von Dungern and Hirszfeld)	27	80	59	24	18
All authors, 1927–1929	2	228	234	1	6

On the von Dungern-Hirszfeld interpretation, if nonrandom mating is assumed, no definite proportions can be calculated, although the second row certainly includes too few AB individuals; on the Bernstein interpretation the expected proportions are 0 : 1 : 1 : 0. The three exceptions in the last row of the table are all listed as being under suspicion of illegitimacy. The 51 in the second row were recorded in eleven of the eighteen papers that are summarized.

This tabulation raises some disturbing questions. One has the uncomfortable feeling that observers see and report only what they expect to find. The most probable interpretation is that the methods of typing were improved. A study of the history of the clinical use of transfusion might be interesting, since the indicated frequency of misclassification before 1925 should have led to numerous unfavorable transfusion reactions.

The Bernstein interpretation has been consistently confirmed in more recent studies and is now fully established.

In one respect the ABO system is unusual in immunological studies, because the agglutinins are normally present in the blood of all individuals that lack the corresponding agglutinogens. The usual situation is that

the effective serum components—*antibodies*, which may cause agglutination (as in the ABO system), hemolysis, or other reactions—are produced only as a result of the previous introduction of the corresponding *antigen* (agglutinogen in the ABO system) into the blood of an individual that does not itself produce it.

The extensive studies of blood antigens other than A and B in man, and those carried out on other animals, have most often utilized such *induced* antibodies.

If human blood cells are injected into a rabbit, a series of antibodies that will react with any human red cells will be produced in the rabbit's serum. Landsteiner and Levine (1927) carried out this experiment, using a series of rabbits, each injected with cells from one of a series of human donors. They then treated each antiserum with cells from several different donors separately. This procedure resulted in the absorption of all the general antibodies against all human red cells, but in some cases the treatment left antibodies that would still react with the cells from other individuals. They were able to show that there were two such reactive substances in the cells, which they called *M* and *N*. They found three types of individuals—*M*, *N*, and *MN*. These substances have been found to be dependent on a pair of allelic dominant genes: no "inactive" allele, corresponding to the O of the ABO system, is known.* This paper marks the beginning of the study of specific induced antibodies to human blood cells, which has been greatly extended since then.

A modification of this technique was used by Landsteiner and Wiener (1940). They injected red cells from a Rhesus monkey into guinea pigs, and carried out absorption with red cells from different human subjects. The resulting absorbed antisera were, in some cases, reactive with cells from other human subjects. They recognized two types of individuals, Rh positive and Rh negative, the symbol being derived from the name of the original donor species.

The clinical importance of the Rh antigens arises from the fact that human subjects who are Rh negative may develop Rh antibodies which lead to transfusion reactions, if the subjects have previously received transfusions from an Rh-positive donor (Wiener and Peters, 1940)—or, more often, if an Rh-negative mother has had an Rh-positive child (Levine and Stetson, 1939). In the latter case, it is evident that Rh antigens from the fetus enter the maternal circulation, and there induce the formation of anti-Rh in the serum. This antibody may, in turn, enter the

* This series of alleles has since been extended by the discovery of antibodies to "S," and to other properties. Many alleles are now known.

circulation of a later Rh-positive fetus. This results in the condition long known as "erythroblastosis fetalis."

Most of the later work in this field has been based on the study of antisera from such sensitized mothers. The work of Levine and Wiener and their co-workers in this country, of Mourant, Race, Sanger, and their co-workers in England, and of others elsewhere has led to a detailed and complicated analysis of the Rh system—now known to include a whole series of alleles. This work, which is of great clinical, anthropological, and genetic importance, is outside the scope of this book. It may be pointed out, however, that the literature is greatly complicated by the widespread use of two radically different systems of nomenclature, due to Wiener and to Fisher, respectively.

There are a series of other systems of red-cell antigens now known in man; these are also not further discussed in this book.

Absorption methods have been used in the study of the red-cell antigens in several other animals besides man. One of the first genetic studies of this kind was carried out by Todd (1930, 1931) with fowl. He injected the cells of various chickens into other chickens and pooled the resulting antisera. He then absorbed these polyvalent antisera with cells from one or more other birds and tested the resulting absorbed sera against the cells of still other individuals. Among three families of chicks, each from a single pair of a single strain of Plymouth Rocks, the results showed that no bird had antigens that were not present in one or the other of its parents, and that within each family (with 17, 18, and 13 chicks respectively) there were no two chicks of identical antigenic composition.

Here then, within a single strain of a single breed, was a great diversity of antigens; and each was dependent on a dominant gene, with no interaction in the production of the phenotype—either between alleles or between genes at separate loci. This remarkable result was soon interpreted to mean that the antigens were close to immediate gene products, and might furnish useful materials for the study of the action of genes, relatively free of the complications of developmental interactions. It is not clear who first formulated this idea; I first heard it in conversation with Haldane in the winter of 1932–1933. However, the results of this assumption have been of far-reaching importance in the study of the developmental effects of genes (Chapter 16).

The lack of interaction between the products of different genes in the determination of antigen specificities has been found to be a very general relation; there are, however, a few exceptions. Irwin and Cole (1936) reported two cases in species hybrids in doves and pigeons. They crossed

Ring Doves to Pearlnecks and to domestic pigeons, and in both cases found that antisera to F_1 cells were not completely exhausted for antibodies by successive absorption by cells from both parent species. In both cases these F_1 birds were backcrossed through several successive generations to one of the parent species, and several genetically distinct species-specific antigens of the usual noninteracting type were isolated. The "hybrid substance" was, in each series, related to particular ones of these, but as yet the nature of this relation is not entirely clear.

These, and a few more recently discovered examples, do show that interaction of gene products may occur; but the rule is still valid in the great majority of cases, even in the dove and pigeon hybrids. There are also a few clear exceptions in intraspecific crosses. This rule is, as formulated by von Dungern and Hirszfeld in 1910 for the human ABO groups, and by Todd in 1930 for his fowl, that no individual carries a red-cell antigen that was not present in at least one of his parents.

A related series of studies concerns the fate of grafted tissues in vertebrates. The first clear genetic result here was that of Little and Tyzzer (1916). They studied a tumor that could be successfully transplanted into any mouse of a strain of waltzing mice (in which the tumor had arisen spontaneously), but failed to grow in mice of an unrelated strain. When these two strains of mice were crossed it was found that the tumor would grow in the F_1, that is, susceptibility was dominant. But when F_2 mice were tested, only 3 of the 183 tested individuals were susceptible. They concluded that several (about 7?) independently segregating dominant genes must all be present in an individual to make it susceptible. This conclusion, checked in various ways, has since been established for many transplantable tumors—with cases on record for strains differing in one, two, or more genes necessary for the growth of particular tumors.

A similar situation has been studied, using normal tissues for transplantation. It has long been known that most normal mammalian tissues can be successfully transplanted to other parts of the same animal (autotransplants), but that transplants to other individuals are usually unsuccessful. There was evidence that the chance of success was better, though still poor, if the donor and host were closely related. The genetic analysis of this relation dates from the work of Little and Johnson (1921). They used inbred strains of mice, that were largely homozygous, and transplanted spleens. They found that such transplants were usually successful within an inbred line but not between separately inbred ones. When two such inbred lines were crossed, the parental strains would not accept transplants from the F_1, but the F_1 would accept those from either of the parental strains. These results were not very extensive nor wholly con-

vincing, but they were fully confirmed on a large scale by Loeb and Wright (1927), using a series of long-inbred lines of guinea pigs.

This general approach has since been carried much farther in mice, especially by Snell, who has succeeded in genetically isolating a series of genes concerned with graft compatibility, and in locating these on the linkage maps.

One of the most striking results in this field was an outgrowth of the extensive work on the red-cell antigens of cattle. The existence of a great individual diversity in cattle antigens was shown by Todd and R. G. White in 1910, but the genetic analysis was begun by Ferguson (1941) and extended by a group including Irwin, Ferguson, Stormont, and Owen. In 1945, Owen reported on a pair of twins, one of which had a Guernsey sire, the other a Hereford sire. He showed that each of these twins had antigens that could only have come from the sire of the other and, similarly, had two kinds of red cells. That is to say, each had a kind of cell proper to its own genetic composition and another kind proper to that of its twin. This was interpreted as being due to the anastomosis of blood vessels in the placenta, already shown by Lillie (1916) and others to result in the passage of hormones from male embryos to their female co-twins. The new results indicated the reciprocal passage of cells ancestral to red cells; since the cells characteristic of a twin were found to persist into adult life, it followed that these foreign erythropoietic cells persisted and reproduced in the recipient animals.

The consequences of this finding were far reaching, and have led to developments in the study of the immunological basis of individual specificity, acquired tolerance to foreign tissue, and other topics (Owen, Medawar, and their co-workers).