SPECIAL ARTICLES

THE ABO BLOOD GROUPS*

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The ABO blood group system enjoys the privilege of being as old as genetic research. It was discovered that blood could be classified according to certain antigenic properties at about the same time Mendel's discoveries were brought to light again. The foregoing applies to only three of the four basic phenotypes; the phenotype AB escaped discovery until 1902.

More than 20 years passed before the genetic hypothesis, which is still valid, was put forward by Bernstein. In accordance with his theory, we describe the system as monomeric (although this is now known to be not quite accurate), that is determined by the alleles at one locus only, the ABO locus. We do not know which of the chromosomes carries this locus, except that it is not one of the sex chromosomes. We know that one of the loci in the neighborhood of the ABO locus has some influence on the normal development of the nails and some parts of the osseous system, because a mutant allele at this locus causes the nail-patella syndrome. The distance between the two loci has been estimated to be about 10 centimorgans. Furthermore, we know that the loci determining the other blood group systems and serum group systems studied until now cannot be in the neighborhood of the ABO locus. On the other hand, it has not yet been definitely excluded that one or perhaps even more than one of them may be on the ABO chromosome, even if they are far removed from the ABO locus. We know of quite a number of monomeric, pathological traits whose genes or loci must be at least some distance from the ABO locus.

We usually think of only three or four allelic genes at this locus : A, B and O, or A_1, A_2, B, and O. It is well known the presence of just one A gene or one B gene in an individual may be determined directly through the corresponding antibodies, but we possess no direct means of disclosing the presence of a single O gene in an

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individual. This is without doubt a great obstacle in gaining more extensive knowledge, for instance, about the pleiotropic effects of the ABO genes. And, in countries like Denmark, where paternity cases are to a very large extent based on blood group evidence, our inability to distinguish between the homozygous AA or BB individual and the heterozygous AO or BO individual, has cost quite a number of men quite a lot of money, as they could not be excluded as fathers even if they were not, e.g., if they were in reality of genotype AA and the mother and child in question of genotype OO. So this problem of finding a direct method for the determination of a single O gene is of great scientific as well as practical interest. The prognosis for this kind of work will be touched on later. The problem has been attacked from many directions, by physicists, chemists, biologists, geneticists, and so on, but no absolutely convincing results have appeared so far.

The nomenclature, the symbols of the genes, is by no means fixed. Personally, I see no reason to introduce the symbols I°, I^{A}, and I^{B} for the genes, as is done by many scientists, mainly geneticists. The reason for doing this is obvious : it squares better with the kind of symbols used in plant and animal genetics. However, as long as there is no general agreement among serologists and human geneticists, and as long as there is no real danger of misunderstandings, I find it easier to denote the genes by single letters.

For quite a long time only little was added to the genetic theory put forward by Bernstein. In 1930, Friedenreich, in Copenhagen, extended the three-gene theory to include a fourth allele. His conclusions were based on the observation that red cells of group A could be divided by means of specific antibodies. He showed that red cells of most people of group A reacted with an antibody found in the serum of some AB people, while a minority of people with group A red cells did not react with this serum. The difference was thought to be due to the presence of a specific antibody, later called alpha-1, which is detectable in the serum of some of those individuals who have red cell group A_2B or MA_2.

Apart from this observation, which may obscure the picture a bit, the rule of the reciprocal relationship is quite true : the presence of antigen A in the red cell excludes the appearance of the antibody anti-A in the serum of the same individual. Likewise, the presence of antigen B excludes the appearance of anti-B, whereas the presence of the so-called antigen O does not exclude the appearance of either anti-A or anti-B in the serum of the individual in

question. Thus, with few exceptions, red cells of group A are found together with anti-B in the serum, red cell group B and anti-A appear together in the serum, red cell group AB has no antibody in the serum, and red cell group O is accompanied by anti-A and anti-B in the serum.

It is not clear whether the production of these iso-antibodies is called forth by the action of specific genes or by stimuli from outside the organism. At birth, we find very few if any signs of these antibodies. They usually do not appear until after the third month of life, and this seems to support the exogenous theory. Another important observation in studies of twins has shown that the intra-pair variation in titer is about the same in monozygous and dizygous twins, indicating that hereditary factors influencing the strength of these antibodies are difficult to demonstrate, if they do indeed exist.

One very interesting fact has emerged from studies of the so-called blood group chimeras, mostly dizygous twins who have had vascular anastomoses with each other during fetal life. Such studies show that the presence of, for instance, antigen A on the red cells of an individual, caused not by the genes at the ABO locus of the individual in question, but by implanted red cell precursors at an early stage in the embryonic life and coming from the partner, prevents the production of anti-A throughout the lifetime of the individual. Finally, it may be that the production of the antibodies is initiated by somatic mutation in some red cells, but we have no proofs of this so far. From investigations on animals, it would appear that environmental stimulation is necessary for the production of these antibodies.

In addition to anti-A and anti-B antibodies, there is also an anti-H antibody, which reacts with almost all red cells, the strength of the reaction depending on the nature of the antigen present in the red cells. The strongest reactions occur with red cells of group O and A_2. Thus anti-H seems to react with a substance common to all antigens. No real anti-O antibody exists, if we use the term to mean that such an antibody should react with all red cells from individuals having one or two O genes at the ABO locus. The so-called anti-O sera react with the red cells in the same way as anti-H; the distinction between anti-O (which is often found in animals) and anti-H is based on the fact that the activity of anti-H is inhibited by the saliva of secretors, while anti-O is not.

The still weaker reacting A-antigens are of great interest to the serologist, but these antigens are extremely rare and have so far produced very little new genetic information. The phenotype A_3 was described by Friedenreich who thought it was caused by a new allele at the ABO locus. Although this is the simplest hypothesis, it is not necessarily the true one. The presence of the A_3 phenotype is suspected if only a few red cells are seen to react with a strong and well-defined anti-A, most of the cells being unagglutinated. The diagnosis has, however, to be confirmed in other ways, because more weakly reacting antigens have been observed. The symbols used to denote these phenotypes vary. Some use A_4, A_5, and so on, but the tendency is to accept A_3 (in addition to A_1 and A_2) as a distinct group, and then divide the rest between the two other groups according to their reactions.

Classification of the A-antigens

	Reaction with anti-A	Anti-A +B (from O subject)	Excretion of A substance	(if secretor) H substance
A_1 and A_2	+	+	+	+
A_3	(+)	(+)	+	+
A_x	—or (+)	+	—	+
A_m	—or (+)	—or (+)	+	+

The designation anti-A usually denotes the antibody found in individuals of red cell group B. It is, however, also used to denote one of the two antibodies commonly found in anti-A sera, thus :

			Reaction with A_1 cells	Reaction with A_2 cells
anti-A serum (from a person of red cell type B)	contains	—anti-A antibody	+	—
		—anti-A_1 antibody	+	—

It may be said, then, that A_1 red cells possess two antigens (A_1 and A), whereas A_2 cells possess only one, A.

What is genetically much more interesting is the evidence suggesting that other genes from an unlinked locus affect the eğpression of the ABO genes. This type of effect, which is well-known in plant and animal genetics, had often been suspected in man, but the first exact knowledge of such a phenomenon has come from the study of blood groups. Genes affecting the expres-

sion of other genes are often called modifying genes, and the process is called a modification.

It has been observed a number of times during the last ten years that the red cells of some individuals showed no reaction with anti-A, anti-B, or anti-H, whereas their sera regularly contained anti-A, anti-B, as well as anti-H. The situation was not at all clear in the beginning. A silent allele was of course suspected, but other hypotheses were put forward, involving the idea of a locus possessing modifying genes. Observation of a female of this group, denoted Oh, who had a baby of group AB (the husband being of group A), has given new and valuable information. Additional studies of relatives and more cases similar to the above, support the hypothesis of a suppressor or modifying gene, called x, with the normal allele X, (later changed to h and H, respectively). Other observations made it reasonable to assume that these exceptional individuals were of genotype hh, which prevented the expression of the ABO antigens in their red cells, even if they possessed the A, B, and/or O gene at the ABO locus in their chromosomes and were able to hand them on to their children. This Hh - locus is not closely linked to the ABO locus.

The antigens in the red cells can no longer be described as fully stable and unchangeable by environment. The exceptions to the rule are, however, extremely few. Many of the cases in which the antigens have been observed to change involved patients with leukemia. Whether the changes were due to some chromosomal abnormality is unknown. Such changes are not observed in all patients with conspicuous chromosomal aberrations, but of course, other and minor changes in the chomosomes may be the cause of the antigenic changes.

Another change of a peculiar nature has been observed in ten to 20 individuals, according to the publications, and it is perhaps more common than the changes mentioned above. This is the so-called «acquired B antigen», and expresses the unhappy fact that blood group B can be provoked by environmental factors, and can appear as a phenocopy, i.e., something non-genetical copying a well-known genetically determined event or character. People with the acquired B antigen are generally not normal, healthy people. They have nearly all been patients, suffering from chronic diseases (mainly carcinoma of the colon or rectum), they have practically all been 60 years of age or more, and they have all been of group A genetically. It is obvious from many types of information-family studies, repeated tests, presence of anti-B in the serum, lack of B substance in the

secretion-that these individuals have no B gene at their ABO locus. It looks as if some factor may change the A antigen and give it a B-like character. A few families exist in which this peculiarity seems to be transmitted.

Turning to the population genetics aspects of the ABO blood groups, it may be said that for no other locus in man, animals or any other living things, do we have such an extensive knowledge of the distribution of the alleles and the variations within and between population groups.

Analyses and comparisons of population groups ought to be based upon gene frequencies because they allow a more detailed statistical treatment of the data and a check of the reliability of the techniques used in the determination of the blood groups in the sample. The gene frequencies are estimated from the sample investigated, and of course the size of the sample (in addition to the reliability of the technique) influences the validity of the estimate. Very simple formulae exist, giving rough but highly useful approximations. The following symbols are always used :

p denotes the frequency of the A gene
q denotes the frequency of the B gene
r denotes the frequency of the O gene

The simplest method of computing frequencies is as follows :

$$r = \sqrt{\text{proportion of O people in the sample}}$$

$$q = 1 - \sqrt{\text{proportion of non-B, that is proportion A + proportion O}}$$

$$p = 1 - \sqrt{\text{proportion of non-A, that is proportion O + proportion B}}$$

The frequencies vary not only from country to country and population group to population group, but, even within a country like Denmark, with a rather stable, homogenous population and very few remnants of isolates, large samples show significant differences as we move from one part of the country to another.

The compilation made by Mourant, et al, in 1958 shows that the rather few published reports on the ABO blood groups in Turkey are almost exclusively based on soldiers (a common sourse of samples for such purposes). The gene frequencies in the Asian part of Turkey vary between the following limits : p, 0.242 - 0.366; q, 0.088 - 0.151; r, 0.517 - 0.660. Very little work has been done on the subgroups of A. Although derived from small samples, the figures given above show that there are great variations within the country. Much more work is needed before an accurate description of ABO blood group distributions can be made.

MODEL OF THE ABO BLOOD GROUP SYSTEM (after Ceppellini)