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SURVEY OF CONTEMPORARY KNOWLEDGE OF
BLOOD GROUPING AND ITS IMPORTANCE IN
BLOOD TRANSFUSION

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Blood grouping has emerged from an obscure position to one of foremost importance to medical science. At the same time it has become of practical usefulness in ethnological study, in the determination of parentage, and in the science of criminology. Consideration of the subject is confined herein to blood groups in relation to blood transfusion since that is perhaps the most practical application of our present knowledge of the groups, although it is possible that in the future blood grouping may become equally important to pathology and physiology.

As in the case of most contributions to science the subject has had an interesting history of discovery and development. While working in a medical laboratory in Vienna, in 1900, Dr. Karl Landsteiner discovered that when the red blood cells of an individual were mixed with the serum of another's blood, agglutination sometimes occurred. This reaction happened only when the red cells of certain persons were mixed with the serum of certain others. Landsteiner recognized this isohemagglutination as a congenital antigenic difference between individuals of the human species. Upon further study he found that there were two normal antigens in erythrocytes and two corresponding antibodies in human sera. Since a person might have one of these antigens in his cells, or he might have the other, or both, or neither, it was determined that there are four kinds of persons in the world in respect of the two antigens, and thus four blood groups. Obviously whatever antigen a person has in his cells, the corresponding antibody is missing in his serum;
otherwise he would agglutinate his own cells. Also whenever an antigen is absent, the corresponding antibody is present in the serum. Unfortunately Moss in America and Jansky in Europe independently numbered the groups and reversed groups I and IV. The resulting confusion led to adoption of Landsteiner's letter system which is now increasing in use. However since 76% of American hospitals still use the Moss system, the problem is not entirely settled. The average distribution of the groups among Europeans and Americans, using the letter classification, is: O 45%, A 42%, B 10%, and AB 3%. The amount of experimental work which the several blood-group discoverers did in order to arrive at their conclusions is astounding. Jansky's classification is based upon 3160 "cross-agglutinations" in which he used sera from 30 individuals and corpses from 90. Other persons besides Landsteiner, Jansky, and Moss who made early discoveries of the blood groups and contributed to the knowledge of them are Shattock, Decastello, Sturli, Hektoen, Bernstein, and Hirszfeld.

For the practical application of blood groups to transfusion heredity is of importance because with a knowledge of the formulas of heredity we can understand that relatives are suitable as donors for blood transfusions only when they possess compatible blood groups. In the past many fatalities have occurred due to ignorance of this fact. That the two agglutinogens are Mendelian dominant characteristics has been demonstrated by von Dungern and Hirszfeld. The inheritance of blood groups has been studied extensively among all races and found to be explainable on the basis of multiple allelo-morphs.

Often mothers and infants have been found to belong to incompatible blood groups. Regardless of this fact, agglutination between the blood of the mother and that of the fetus in the placenta never occurs. This fact could lead one to the dangerous false conclusion that it is unnecessary to use compatibility tests in transfusing blood into infants. The fact that agglutination does not take place between bloods of mother and infant does not mean that isoagglutination cannot occur between an infant and any other individual. For there may be selective permeability of the placenta which excludes the possibility of mother-fetus isoagglutination, thus making an exception to the rule of incompatibility. The total absence of agglutinins in the serum of the newly born, however, cannot be upheld. A baby's serum, therefore, may agglutinate the transfused cells of a donor of an incompatible group, and a baby's erythrocytes may well be agglutinated by the serum of an incompatible donor of high titer.

Isoagglutinogens are usually detectable in the red cells at birth but occasionally this tentative group may be changed. Once they are permanent, blood groups are believed to be stable throughout life. Contrary to agglutinogens, isoagglutinins are rarely present in the serum at birth, but may become established at any time during the first year or two of life. From this one may see that while in many cases no reaction would occur in the absence of compatibility tests, the fact remains that young children may have the blood group definitely established. Thus the same careful procedure that is used in choosing donors for adults must be observed in preparing to transfuse blood into infants.

2. Ibid. p. 19.
3. Ibid. p. 19.
Today clinical indications for transfusion are many and varied. This is due to the increased understanding of the blood groups and their application to transfusion, with the resulting advances in the safety of the operation. We can thank the World War for our increased knowledge of blood groups and transfusion. Until then little attention was paid to blood groups in spite of Landsteiner's discovery that nearly all of the severe reactions that had been reported were due to the factors involved in isohemagglutination. During the early part of the war many transfusions were fatal because of the agglutination reaction occurring in the patient's blood vessels due to the use of incompatible donors. The necessity for large numbers of emergency transfusions brought the problem sharply to the fore; this resulted in Landsteiner's long-ignored advice being heeded, and transfusion became the first real practical application of blood grouping. During the war transfusion was only a curative measure to replace blood suddenly lost from the circulation due to wounds. Today, however, transfusion is used often as a palliative measure instead of a cure. Transfusions in cases of placental hemorrhage and other sudden losses of blood in connection with child-birth have been highly successful. It is a wise precaution to have compatible donors available for such obstetrical emergencies. Hemorrhages resulting from "hemorrhagic" diseases are also indications for transfusion. It is well for a hemophiliac to know his blood group and have compatible donors available at all times because a small amount of transfused blood will bring about hemostasis in case he is bleeding. Other indications for blood transfusion are pernicious anemia, malnutrition in infants, septic conditions, and carbon monoxide poisoning.
The methods used by hospitals and doctors for blood grouping are based upon the assumption which I have made so far that there are only four iso-agglutinin groups. These typing tests are devised so that each blood tested falls into one or the other of these groups, thus maintaining the unquestioned belief that every person belongs to one of the blood groupings. The test for the blood groups is made by mixing suspensions of erythrocytes, usually under the microscope, with various serums, each of which is known to have a certain iso-agglutinin. If any serum produces hemagglutination, the cells must contain the agglutinogen which that particular iso-agglutinin agglutinates. In reading the tests, the following rules will enable one to tell the group for certain. If neither test serum agglutinates the cells, the blood is group 0. If only group B serum agglutinates the cells, the blood belongs to group A. If only group A serum agglutinates the cells, the blood belongs to group B. If both serums agglutinate the cells, the blood belongs to group AB.

Recent blood work has shown that the division of human blood into four groups is not as simple as was first believed. The reason for this is that there are three classes of exceptions to the four groups. They are: first, three new agglutinable factors, one of which possesses a newly discovered specific extra-agglutinin; second, a series of phenomena involving atypical reactions such as pseudo-agglutination, auto-agglutination, and "cold"-agglutination; and third, additions to the four original groups in the form of sub-groups, due to the alleged presence in certain bloods of additional iso-agglutinating elements. These exceptions are so complicated that the ordinary hospital technician may probably not understand the theory behind them. These exceptions do not render it necessary to make any definite change in the standard scheme of grouping, however, but only to consider to be possible sub-groups and additional groups. However these exceptions should emphasize this fact:
whatever the ultimate outcome of the question of sub-groups and extra-agglutinins and additional groups, the present uncertain status makes it a practical necessity to always perform direct blood matching tests immediately before transfusion in addition to determining the groups of donor and recipient. For there have been instances in selection of donors for transfusions of irregular agglutination which certainly were due to one of these three exceptions; if these were not discovered before the start of transfusion, such a donor might have been used with perhaps an injurious result. Landsteiner and Levine have found that irregular agglutination reactions are usually weaker than true reactions. With regard to the practice of transfusion, they stated that in several instances in which the serum of the recipient contained irregular agglutinins for the blood of the donor no untoward symptoms at all were observed.

The first exception to the traditional blood groups is the presence of three new agglutinable factors in blood; these are antigens which are distributed among the four major blood groups, and are known as M, W, and F agglutinogens. Their antibodies may be artificially produced by immunizing rabbits. By means of these antibodies it is possible to classify persons as to their M, W, and F antigens. No one has yet been found who lacks both the M and W antigen but the F agglutinogen is found less frequently.

Since no normal agglutinins are found in human sera against the M and W agglutinogens, their presence need not be considered in blood transfusions. But the presence of the property F must be considered, for Landsteiner and Levine have found an extra agglutinin in some blood sera which is specific

3. Ibid.
for agglutinogen P. These two workers found in 1931 that this new agglutinin, which they named "extra agglutinin 1", appeared occasionally in the serum of types O, A, or B individuals. When a serum of this kind is encountered and compatibility tests are done with it, confusion may result unless the phenomenon is understood. It is especially interesting to know that in rare cases, even group O cells may be agglutinated in cases where this extra agglutinin 1 is present in the serum used.

In a series of experiments with blood containing the factor P, Landsteiner and Levine found that agglutinins specific for factor P are found rather often in normal animal sera and consequently a frequent type of human blood sensitive to these agglutinins can be easily recognized, occurring in each of the four blood groups. One of their experiments made use of twelve horse sera and four human bloods of group O, two of which were not sensitive to the "extra agglutinin 1." The sera were absorbed with one of the two non-sensitive bloods and the fluids were tested with all four bloods. It was found that of the twelve sera, nine showed reaction for the two bloods containing extra agglutinin 1, whereas the other two bloods were not reacted upon.

They also tried rabbit, pig, cat, and cattle sera in this experiment. On the whole, reactions of the various animal and human sera ran parallel. However there were some discrepancies attributable to qualitative variations in the agglutinogens and agglutinins present in the various animal sera. Also reactions of extra agglutinin 1 varied. But the existence of a type P is established by the fact that most sensitive bloods are almost regularly agglutinated by all the reagents, whereas certain other bloods nearly always show negative reactions.

1. Landsteiner, K., and Levine, P.: Journal of Immunology, 1931, 20, 179.
2. Ibid.
Thus the property $P$ is not strictly defined like $A$, $B$ or $M$, but designates a group of related agglutinable factors.

The second class of exceptions to the simple division of blood into four groups is a series of phenomena involving atypical or irregular agglutination reactions. In addition to the scientific interest of these phenomena, they are important because they frequently cause mistakes in typing of blood.

Pseudo-agglutination is one of these phenomena. Rouleaux formation, or the tendency of the red blood cells to adhere together in columns, like piles of coins, is the commonest of pseudo-agglutination phenomena. Doctors and laboratory technicians encounter it often. An untrained worker might mistake rouleaux formation for agglutination, although there are really no agglutinins involved. It is easier to distinguish rouleaux formation under the microscope for there the piling effect of the cells is more easily noticed. Another simple distinction is by means of mechanical agitation. If one taps the suspension of cells in serum, the clumps of cells will be broken up if due to rouleaux formation; if the clumps are due to true iso-agglutination the tapping will only cause them to clump stronger. Another way to identify rouleaux formation is the dilution of the test serum with plain salt solution. Rouleaux formation is stopped by dilution of the serum. In various diseases, such as rheumatic fever, pneumonia, tuberculosis, and heart diseases, and in certain physiologic conditions such as menstruation and pregnancy, the blood shows a tendency to heavy rouleaux formation. In these disturbances it is well to take precautions against errors in reading. One of these precautions is the adding of lecithin or kaolin suspension to the serum to prevent pseudo-agglutination. Formalin and hypotonic salt solution have likewise been suggested for this purpose. In practice the method commonly used for diseased persons as well as normal persons is to dilute the test serum with plain salt solution where rouleaux formation is suspected and see whether the clumping is stopped.
The phenomenon of pseudo-agglutination occurs at low temperatures and may be intensified at blood temperatures of 37°C. Absorption experiments show that you can treat the test serum causing pseudo-agglutination with red cells without causing absorption of the active substance. In true iso-hemagglutination, the active substance of the agglutinin can be absorbed by treating with erythrocytes.

Continuing the second class of exceptions, another phenomenon which may be sometimes a possible source of error in blood grouping is "cold"-agglutination, the reaction of the serum of one individual clumping the red cells of another person. Such agglutination occurs mainly from 0 to 5°C, diminishing as the temperature increases, and generally disappearing at body temperature of 37°C. When a person's red cells are agglutinated under such conditions by his own serum, the reaction is called auto-agglutination. Landsteiner showed that in auto-agglutination and other cold-agglutination, the active principle in the serum may be absorbed by the cells, thus showing it to be a true agglutinin. If the cells are warmed the active principle in the serum can be released from absorption. Thus the agglutinin may be obtained in salt solution at room temperature, and its presence demonstrated by adding new red cells to the fluid and lowering the temperature. We have already seen how pathologic disturbances increased pseudo-agglutination. Similarly such diseases as syphilis, hemolytic icterus, pernicious anemia, sickle cell disease, anemia, chronic leukemia, and pneumonia greatly increase auto-agglutination.

The practical significance of cold-agglutination is that one might confuse this "typical agglutination reaction with the typical isohemagglutination if the typing was done in a cold laboratory room. At ordinary room temperature, however, such reactions will not be encountered, and need not interfere with the typical blood grouping scheme.

The third order of exceptions to the four blood groups consists of those forming the sub-groups. When Landsteiner discovered the blood groups he noted exceptions to the general rule. In 1911 von Dungern and Hirsfeld apparently showed by means of agglutinin absorption experiments that persons having agglutinogen A fall into two sub-groups, those that are purely A and those in which the A agglutinogen is linked with a second agglutinogen A'. Likewise they showed apparently that there are two kinds of group B sera, those containing only the agglutinin for the A factor and the other containing the a agglutinin always associated with an a' agglutinin. Landsteiner has demonstrated that many group O sera contain a' as well as a and b agglutinins.

These sub-groups cause anomalous reactions between certain cells and certain sera. For instance, the red cells of various group A persons do not always act alike. Theoretically, when group O serum is absorbed with erythrocytes of group AB, all the iso-agglutinins from the group O serum should be removed, and it should no longer be able to agglutinate the cells of any other group. But sometimes the group O serum is still able to agglutinate the red cells of certain members of group A. This peculiarity is due to sub-groups of the factor A. If they are present in group O serum, absorption with erythrocytes of the first kind of group A leaves the serum still capable of agglutinating cells of the second kind of group A.

Since the a' agglutinin is almost always found associated with the a agglutinin in sera and the A' agglutinogen likewise associated with the A agglutinogen, it is clear that their existence does not make any definite change in Landsteiner's standard scheme of grouping. That is, their presence cannot be detected by routine typing, but only by means of absorption experiments.

2. Landsteiner, K., and Levine, F. The Journal of Immunology, 1929, 17, 1.
Landsteiner and Levine studied the occurrence of atypical isoagglutination reactions of human bloods. They purposely chose a technique which would detect sera of even slight activity. In this way rather numerous irregular reactions were found in a material obtained from patients with mental diseases such as dementia praecox.

Most of the reactions they observed may be termed cold agglutinations since the sera were active only at low temperature. A smaller number of sera (about 3%) contained agglutinins of a type intermediate between cold agglutinins and typical isoagglutinins since their reactions were distinctly noticeable also under the usual conditions of blood grouping.

A survey of their results helps to show the role played in the abnormal reaction by the sera on the one hand and the erythrocytes on the other. They showed that if you have a serum with sufficiently active irregular agglutinins, it will in most cases react not on exceptional but on numerous bloods though with varying intensity. But a converse statement does not hold; that is, if a certain specimen of blood cells displaying irregular agglutination be tested with numerous sera only a few will be found to give distinct abnormal agglutination. Thus one may speak of abnormally reacting sera but hardly of abnormal blood cells; otherwise practically every blood cell ought to be called irregular. Their results also show that there is not a regular relation between the absence of agglutinogens and the presence of corresponding agglutinins.

Some workers do not accept these sub-groups or the M, N, and P groups and say that all evidences for them are based on various sources of error, such as pseudo-agglutination, low titer, incomplete absorption, and other quantitative effects. Whatever the ultimate outcome of the question, it bears repeating that the present uncertain status makes it a practical necessity to perform direct blood matching tests immediately before transfusion in addition to determining the groups of donor and recipient.

So far many theoretical considerations of blood groups in their relation to transfusion have been shown. Now the actual choosing of a donor and typing of blood in preparation for a transfusion will be considered.

Blood transfusion serves two main purposes. It restores the bulk of the circulatory fluid, and provides the recipient with red cells for carrying oxygen. Since the usual dosage for adults is 500 cc. and the average adult has approximately 6,800 cc. of blood in his system, the serum of the donor is so diluted that its agglutination power for the recipient's cells is not considered important in most transfusions. The concern is whether the donor's cells are agglutinated by the serum of the recipient. On this basis group O individuals are known as "universal donors" since their erythrocytes are not agglutinated by the serum of any group. Similarly patients belonging to group AB are known as "universal recipients" because they are frequently transfused with blood from donors of any group. This is safe because the serum of type AB contains no agglutinin.

Before transfusion it is very important that a proper donor is chosen. Many factors enter into this choice, such as the physical, physiological, pathological, and even psychological qualities of the various available donors. Besides grouping and direct matching tests, careful routine blood examinations and Wassermann tests should be given each available donor. The Wassermann test is very important because syphilis has been transmitted by transfused blood.
Other diseases which also have been transmitted by transfusion are malaria, measles, smallpox, and allergic conditions.

From the psychological standpoint professional donors are the best. They are listed in large numbers with many hospitals in this country. Although their blood group need only be taken once, they should be given the routine blood examination and Wasserman test each time.

The most important element in the choice of a donor is his blood group. A donor should be chosen from the same group as the recipient if possible. If no donor of the same group is available, a group 0 donor should be used. In cases where universal donors are employed quantitative relationships between cells and agglutinative sera are of special importance in transfusing, since Lettis and Savazzuti have found that quantitative differences may account for many irregular blood differences. The use of universal donors with exceptionally high agglutinin titer is directly contraindicated.

To avoid donors who might cause anomalous reactions, direct compatibility tests with blood of donor and recipient must be made must preceding the transfusion. However, blood matching alone should not be relied on, because the results by themselves are not always sufficient to prove the donor safe.

Normal room temperature of 20°C is accepted as the best temperature at which to perform grouping tests. As the temperature decreases, the chance of "cold"-agglutination increases. After the reading has been made at the end of fifteen minutes, it is sometimes advisable to place the blood being tested in the incubator at 37°C for half an hour, to obviate any chance of missing a weak agglutination due to low titer. Besides the room temperature requirement, the important point to be remembered in grouping bloods is the use of fresh sterile sera of known high titer for testing.

It is important to know the symptoms which a recipient might show if incompatible blood is being transfused into him because when these are noticed transfusion can be stopped before a severe reaction results. Typical reactions are: falling pulse rate, loss of consciousness, hemoglobin-uria, general bodily pain, and heavy and labored breathing. Fullness in the head is often complained of, followed by pains becoming localized in the lumber region. The patient's face is usually flushed and his pulse rate usually low. A severe chill, being followed by high fever with temperature of 103° to 105°F., is characteristic. If more than 100 cc. of incompatible blood is introduced, death usually follows. Since reactions begin to appear early in the transfusion, the operation can usually be stopped before it is too late.

In conclusion of this thesis, let me emphasize two points: first, that at the present time much interest is centering around the applications of blood grouping to clinical medicine other than transfusion, that is, to the genetic relationships of blood groups to various morphologic, pathologic, and physiologic conditions; and second, that my treatment of blood grouping has only been concerned with the importance of the problem to blood transfusion. If I have shown this I feel that my purpose has been accomplished.
BIBLIOGRAPHY


Wigg, Clare: Studies on Agglutinogens of Human Blood. Journal of Immunology, 1930, 19, 1.

