[2] In the discussion of the valence state of oxygen in reference 1 the energy of the valence state was given incorrectly as $F^0 - \frac{11}{100} F^2$.

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*ACQUISITION OF THE J SUBSTANCE BY THE BOVINE ERYTHROCYTE**

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The antigenic pattern of the bovine erythrocyte is genetically conditioned by numerous genes on at least nine of the thirty pairs of chromosomes.[1, 2] The combinations of these genes permit well over a million blood types. Any two animals taken at random, therefore, will almost invariably possess distinct blood types. A study of bovine twins, however, disclosed a much higher proportion of identical blood types than is expected in view of the relatively low frequency of monozygotous twinning in cattle.[3] Owen pointed out that this high proportion of identical blood types could be accounted for as a consequence of the known frequent union of chorionic blood vessels of twin fetuses, as demonstrated by Lillie.[4] Apparently, embryonal blood cells are interchanged by those twins having a common circulatory system and these primordial cells settle in the hematopoietic tissues where they serve as a source of erythrocytes throughout the life of each twin. The identity of blood types in otherwise genetically dissimilar twins would thus result from a mixture of two kinds of erythrocytes, namely, those produced by (*a*) the twin's own cells and (*b*) cells transplanted from the co-twin. Several lines of evidence, including the *in vitro* separation of the two kinds of erythrocytes comprising the mosaic of blood cells, afford proof of this explanation. It is now found that the cellular character called J constitutes an exception to the general rule in that twins possessing a blood admixture nevertheless may differ in respect to the presence or absence of this blood factor. The present report is concerned with the reasons for this non-conformity in behavior of character J.

Each of the numerous antigenic factors of cattle erythrocytes is recognized by the reaction (hemolysis) of these cells produced by a corresponding reagent (antibodies) and complement (fresh rabbit serum). For example, erythrocytes of an individual that possesses a gene for antigen A are sensitized by the action of A reagent and subsequently destroyed (hemolysed) by the action of the complement, while those of an animal that lacks gene A

are non-reactive in the presence of A reagent and complement. The blood cells of an individual are tested with each of 40-odd reagents for the presence or absence of reactivity (hemolysis). The blood type (antigenic formula) of an animal, as determined by these tests, constitutes the reactions of the cells when tested with each of the reagents. A diagrammatic representation of the techniques involved and references to previous reports are given by Stormont and Cumley.[5]

TABLE 1

BLOOD ADMIXTURE IN A PAIR OF TWINS DIFFERING IN FACTOR J

	BLOOD TYPES													
Twin no. 1:	A	$\overset{\pm}{C_1}$	F	$\overset{\pm}{G}$	H	J	P	$\overset{\pm}{Q}$	$\overset{\pm}{V}$	W	X_2	Z	$\overset{\pm}{D'}$	H′	I′
Twin no. 2:	A	$\overset{\pm}{C_1}$	F	$\overset{\pm}{G}$	H		P	$\overset{\pm}{Q}$	$\overset{\pm}{V}$	W	X_2	Z	$\overset{\pm}{D'}$	H′	I′
	BLOOD TYPES OF THE SEPARATED CELLS FROM TWIN NO. 1													
Type 1:	A	C_1	F		H	J	P	Q	V	W	X_2	Z		H′	I′
Type 2:	A		F	G	H	J	P			W	X_2	Z	D′	H′	I′
	BLOOD TYPES OF THE SEPARATED CELLS FROM TWIN NO. 2													
Type 1*a*:	A	C_1	F		H		P	Q	V	W	X_2	Z		H′	I′
Type 2*a*:	A		F	G	H		P			W	X_2	Z	D′	H′	I′

Generally, erythrocytes that are sensitized with sufficient antibody are completely hemolysed when complement is added. The cells of the twins (table 1), however, exhibited only partial hemolysis when sensitized with antibodies for the factors C, G, Q, V and D′, as indicated by the ± sign above these factors. Cell counts were made following the hemolysis of aliquots of 2.5% suspensions of the erythrocytes of each twin. These counts showed that approximately 65% of the cells of each twin were hemolysed following sensitization with either the G or D′ reagents, whereas about 35% were destroyed following sensitization with the reagents for C, Q, and V, respectively. These results demonstrated that only one kind of erythrocytes in the mixture possessed the antigenic factors G and D′ while the other kind possessed the factors C, Q and V not found in the first. The cells that were not hemolysed following sensitization with the G (or D′) reagent contained the factors C, Q and V as well as the factors common to both types of blood (i.e., A, F, H, P, W, X_2, Z, H′ and I′). Likewise, the other kind of cells, which were not lysed by the C reagent (or by those for Q or V), contained the factors G and D′ in addition to those common to both kinds of cells. The J character obviously did not conform to the behavior of the others, since it appeared in both kinds of the blood cells of twin no. 1 and was not demonstrable in either kind of the cells of the cotwin.

The J character is inherited as a dominant in contrast with its absence,[6] as are the other known antigenic components of the erythrocytes of cattle.

It would appear, however, that erythrocytes derived from the primordial blood cells of an individual possessing the gene for J (twin no. 1, table 1) fail to manifest this phenotype in a co-twin-host lacking the J gene. Conversely, those erythrocytes derived from an individual lacking the gene are capable of developing this character in a host possessing the gene.

The unique behavior of J in individuals of multiple births focused attention on other peculiarities of this blood factor that suggested a plausible explanation of this phenomenon.

1. The J character has been detected only by means of normal antibodies present in certain cattle that lack J.[6] Attempts to produce immune antibodies specific for J in cattle and rabbits have not been successful to date. In contrast, all the other blood factors of cattle are recognized by means of immune antibodies.

2. The J character of the erythrocytes seemingly is not present at birth, since the cells of several young calves which originally were J-negative were found in subsequent tests to be J-positive.

3. A soluble J or J-like substance has been found in considerable concentrations in the plasma of J-positive individuals. (This observation was made by Mrs. M. W. Ycas, formerly of this laboratory, who noted that the plasma of J-positive bloods was capable of inhibiting the reaction of J antibodies.) Soluble substances corresponding to the other blood-group-factors of cattle erythrocytes have not been detected.

4. Marked differences in the avidity or reactivity of the cells of different J-positive adults for the same concentrations of J antibodies have been noted. However, the degree of reactivity of the cells of a given individual appears to be relatively constant from month to month. Qualitative differences in the J substance have not been clearly demonstrable by antibody-absorption. On the other hand, the titer of J antibodies varies markedly among individuals and within the same individual from one time to another. As a consequence of such differences, weakly reactive anti-J sera do not contain sufficient antibody to cause visible hemolysis of all J-positive bloods. A similar effect may be obtained by diluting the antibodies of a more strongly reactive normal serum. Two potent normal sera (titer 1:256 and 1:512, respectively) obtained from the same cow (No. 17Z) were used for the tests described in this report. These sera were employed routinely in optimum dilution (1:4 or 1:8) for the detection of the cellular character J.

These observations suggested the explanation that J is primarily a soluble, serologically reactive substance which may be acquired by the erythrocytes of any individual (with or without the gene) on contact with blood plasma containing the soluble substance. To test this assumption, the bloods of donors lacking J, whose cells could be separated and readily

distinguished from those of the recipients, were respectively transfused into selected recipients, as given in table 2.

The cells of donor 41 that were recovered from a blood sample taken from the recipient (cow no. Z21, table 2) just after the transfusion were J-negative. The cells from each of the donors (cows 41 and Z26) recovered at intervals of 2, 4, 7, 9, 14 and 21 days after transfusion of 2.5 liters of whole citrated blood had become J-positive. The cells recovered after the fourth day were indistinguishable from those of the recipients in their degree of reactivity with J antibodies.

The results suggested that the same transformation might be accomplished *in vitro*, if given sufficient time. Washed erythrocytes from J-negative cattle were suspended in freshly filtered (Seitz filter) plasma of J-positive individuals (approximately 0.4 ml. of cells to 10 ml. of plasma) and incubated at 37°C. The cells were changed to fresh plasma at intervals of 2 hours during the day and were kept at refrigerator temperature (±4° C.)

TABLE 2

THE *in vivo* TRANSFORMATION OF J-NEGATIVE ERYTHROCYTES TO J-POSITIVE

EXPERIMENT NO. 1	BLOOD TYPES										
Donor 41		C_1	F	G			X_2	Y_2	Z	E'_1	H′
Recipient Z21	A	C_1	F	G	*J*	W	X_2	Y_2	Z	E'_1	H′
EXPERIMENT NO. 2											
Donor Z26	A	C_1	F			O_3	S	D′		E'_3	H′
Recipient Z18	A	C_1	F	G	*J*	O_3	S	Z D′		E'_3	H′
BLOOD TYPES OF DONORS' CELLS RECOVERED 2 DAYS, AND AT LATER INTERVALS POST-TRANSFUSION											
Donor 41		C_1	F	G	*J*		X_2	Y_2	Z	E'_1	H′
Donor Z26	A	C_1	F		*J*	O_3	S	D′		E'_3	H′

without a further change overnight. The treated cells became J-positive within 24 hours. These experiments are in agreement with the proposal that the J character of cattle is primarily a soluble, serologically reactive substance that is acquired by erythrocytes in contact with this substance in the blood plasma.

Assuming that tissues which produce soluble J substance, or the cells ancestral to these tissues, are not interchanged by fetuses during embryonic life, the behavior of J in twins having mixed blood types is readily understood. Both kinds of erythrocytes in a twin possessing the gene acquire the soluble substance from the plasma, whereas both kinds of cells of the co-twin that lacks the gene are J-negative.

Holtman[7, 8] demonstrated that bacteria are capable of acquiring serologically reactive substances from culture-media and from the animal host. The acquired substances were from media containing horse serum and from guinea pig hosts. Both these species possess Forssman antigen as well as serologically reactive substances related to A of man. As Holtman pointed

out, certain of the heterophile reactions attributed to bacterial cells may be consequent on the acquisition of these substances from the media or the host.

A definite serological relationship between the J substance of cattle and the A of humans was also established. Normal anti-J sera, that did not contain appreciable agglutinins for the cells of humans of groups O and B, nevertheless agglutinated, in higher titers, the cells of individuals of blood groups A and AB. Moreover, both the saliva and sera of humans of groups A and AB inhibited J antibodies in their reaction with J cells. Presumably, this was due to the presence of a soluble, serologically reactive substance in the saliva and serum corresponding to the A substance of the cells. Likewise, commercial preparations of pepsin and trypsin contained a soluble, heat-stable substance (or substances) which was capable of inhibiting J antibodies. Presumably the J substance is closely allied serologically and thereby chemically with other A or A-like polysaccharides generally distributed throughout the animal kingdom.

With few exceptions, the antigens of blood cells have behaved genetically as simple dominants in contrast to their absence. Moreover, their relative ease of detection by means of antibodies, their apparent constancy under varied environmental conditions and throughout life as well as the fact that these specific properties are contained in the stroma of erythrocytes[9] would suggest that they are produced by the action of genes in maturing erythrocytes or their cellular precursors. Indeed, the idea has been expressed[10, 11] that cellular antigens may be the more or less primary products of their causative genes. While the evidence concerning the J factor of cattle does not constitute an exception, as far as is known, to the above idea, it demonstrates that the synthesis may not always be in the cells in which a particular factor is recognized.

Summary.—Evidence is presented that the J factor of bovine erythrocytes is primarily a soluble, serologically reactive substance elaborated by tissues other than those which produce erythrocytes. Erythrocytes acquire the J substance on contact with soluble J in the plasma.

* Appreciated contributions to various phases of this study have been made by Professor M. R. Irwin. Considerable technical assistance to the project was rendered by Miss M. Claire Busk.

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*FURTHER STUDIES ON THE EFFECT OF INFRA-RED RADIATION ON X-RAY-INDUCED CHROMATID ABERRATIONS IN TRADESCANTIA**

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The time limit within which x-ray-induced breaks in the chromosomes of dividing cells restitute, or recombine non-homologously, has not been established with any degree of exactitude, if, indeed, such a determination is possible. The problem has been variously attacked, but no single generalization adequately encompasses the divergent data obtained from such organisms as Tradescantia, Drosophila and maize. Even from the extensive x-ray studies which have been made on the microspore chromosomes of Tradescantia, the resultant conclusions drawn by the several investigators are only in partial agreement.

Time-intensity and fractional dosage studies on the microspore chromosomes of Tradescantia have led Sax to conclude that the broken ends remain in a condition permitting recombination or restitution for a period no longer than one hour.[1, 2] There is some indication that the time span may be as short as 20 minutes. The fact that temperature changes and centrifugation will modify the x-ray-induced frequency of aberrations in the same material *only* when used within certain time limits supports this conclusion.[3, 4] Data from experiments employing various radiations in combination provide additional supporting evidence. Ultra-violet, used as a pre-treatment, reduces the percentage of x-ray-induced aberrations in both Drosophila and Tradescantia.[5, 6] When used as a post-treatment on the pollen tube chromosomes of Tradescantia, the effectiveness of the ultra-violet in reducing the percentage of detectable aberrations diminishes as