IMMUNITY TO HOMOLOGOUS GRAFTED SKIN. II. THE RELATIONSHIP BETWEEN THE ANTIGENS OF BLOOD AND SKIN.

P. B. MEDAWAR.

From the Department of Zoology and Comparative Anatomy, University of Oxford.

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THE idea that there exists an intimate relationship between the forms of incompatibility revealed by blood transfusion and tissue transplantation has been more widely accepted than any concrete evidence for it allows. (Arguments for and against the theory have been summarized by Medawar, 1943.) Immunity to " transplanted " blood appears to be of two sorts : innate (as with the A-B-O antigen system of the blood of human beings and anthropoid apes), or newly acquired in response to an antigenic stimulus. The latter represents the more usual state of affairs, and it is to this category that skin transplantation immunity belongs (Gibson and Medawar, 1943 ; Medawar, 1944, 1945*a*).

The problem dealt with in this paper can be expressed in the following restricted but nevertheless workable form : do the cellular constituents of blood share antigens in common with the antigens of skin epithelium ? The case of red cells may be considered first. If red cells and skin epithelium share antigens in common, then (*a*) the grafting of foreign homologous skin should in a proportion of cases elicit *pari passu* the formation of red-cell isoagglutinins or isolysins ; and (*b*) the transfusion or grafting of red cells should confer an appreciable degree of immunity towards skin later grafted from their donor. In theory, these corollaries are equally applicable to the case of leucocytes ; but the former, (*a*), is not workable because immunity to leucocytes cannot be demonstrated by an *in vitro* reaction. Immunity to homologous grafted skin may be recognized, however, by the suppression of cell-division in skin transplanted to a specifically immunized animal and by its greatly accelerated breakdown (Gibson and Medawar, 1943 ; Medawar, 1944, 1945*a*, 1946).

Full-grown rabbits from heterogeneous stock have been used in all the experiments described in this paper.

IMMUNITY TO RED CELLS AFTER GRAFTING FOREIGN HOMOLOGOUS SKIN.

In each of 15 trials with 15 independent pairs of rabbits, skin was grafted in high dosage from a donor D to a recipient R in the manner described by Medawar (1944). After these homografts had broken down—but in no case before the 12th day from grafting—blood was withdrawn from the marginal ear vein or median ear artery of animal R by means of a pipette with a stab point, allowed to clot spontaneously, and spun for serum. At the same time about 0·2 ml. whole blood was withdrawn from the marginal ear vein of D, and at once mixed

with not less than 10 ml. 0·4 per cent sodium citrate in 0·85 per cent NaCl solution. The blood cells were spun down, taken up in citrate saline, again spun down, and finally resuspended in about 10 ml. normal saline.

Agglutination test.

One drop of serum from R was mixed in a standard agglutination tube with two drops of the " 1 per cent " saline suspension of blood cells from D. (Slide tests, at first done in parallel with the tube tests, were early abandoned.) The tubes were incubated for 4 hours at 38° C. before their first examination, and left overnight at room temperature before their second. Microscopical examination was made in all trials, though due attention was paid to the nature of the packing and cohesiveness of the red-cell sediment.

No trace of agglutination or haemolysis was recorded in any of these tests. For the sake of completeness it should be added that two of the recipient animals were subjected to "adjuvant" immunization with native chicken plasma. The chicken plasma precipitin titre on the day of the agglutination test was greater than 1000.

A second test made use of a group of 5 rabbits, each one of which received a graft from each one of the others, and from 19 other rabbits as well. (These were animals Nos. 1–5 of the cross-grafting test described in full by Medawar, 1945*a*, pp. 165–168). Fifteen days after this multiple grafting operation the serum of each animal was tested in the manner described above against the red cells of the other four. This experiment, like its predecessor, gave uniformly negative results. It is clear that the formation of red-cell agglutinins is not a normal accompaniment to the reaction provoked by the grafting of foreign homologous skin.

IMMUNITY TO SKIN HOMOGRAFTS FOLLOWING TRANSPLANTATIONS OF BLOOD CELLS.

The four experiments now to be dealt with represent the converse of those described in the preceding section, their object being to determine whether the transplantation of blood cells from one rabbit to another confers any degree of immunity towards skin later grafted from the same donor. The experiments are reported in the order in which they were done, and they take the following variables into account: the route of the blood cell injection, the type of blood cell, and the cellular dosage.

To test for the existence of a " skin immune state " the blood-cell recipient was grafted, after an appropriate interval of time, with one large pinch graft (modal average weight 0·045–0·055 g., mean diameter of freshly cut grafts after fixation, 8·5 $\pm$ 0·3 mm.) from the blood cell donor. The graft was cut from the outer aspect of the thigh and accurately fitted to the skin overlying the right thoracic wall (Medawar, 1945*a*). Alternative courses were then followed. In Expts. 1 and 2 the graft was allowed to remain in residence for 12 days, and then removed and sectioned to provide a diagnosis of its degree of survival. In Expts. 3 and 4 the graft was removed on the 6th day for mitotic counting, using the technique described in full in the first paper of this series. The second test is the more delicate: it became possible to use it only when the experiments described in the first paper of this series were complete.

First Experiment.

Each of the five similar tests that constitute this experiment made use of three rabbits: blood and skin donor (D), red-cell recipient (R-1), and white-cell recipient (R-2).

Not less than 5 ml. whole blood from the median ear artery of rabbit D was received into a test-tube through a hard glass cannula with a sheared-off stab point (fig. 1*a*). A measured 4 ml. was then at once withdrawn and mixed with 0·4 ml. 4 per cent Na citrate in the fractionation tube illustrated by fig. 1*b*. The fractionation tube was spun, with the central withdrawal tube in position and sealed with wax, until the leucocytes formed a compact layer (the "buffy coat") between the red cells and supernatant plasma. After withdrawing the plasma carefully through the side hole of the outer tube the wax seal on the inner tube was broken, and one-fourth of the cellular deposit, consisting of pure red cells, was withdrawn by means of a long fine pipette and mixed with one-half of the decanted citrate plasma. The pure red-cell suspension, containing the red cells of 1 ml. whole blood, was thereupon injected into the marginal ear vein of animal R-1.

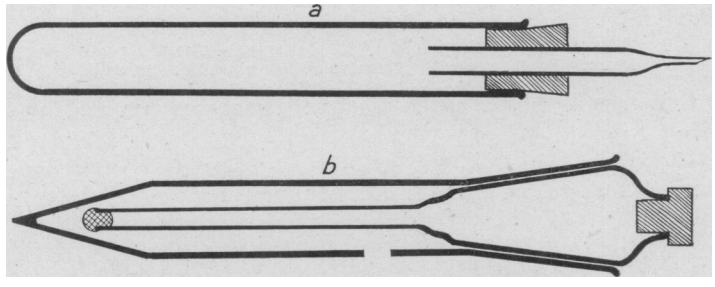


FIG. 1 (*a*).—Test-tube and cannula with sheared-off stab point for collecting blood directly from the median ear artery of the rabbit. A narrow slot is cut in the cork to allow the escape of displaced air. 1 (*b*).—Fractionation tube made from standard ground glass apparatus (B-19 joint). Calibration marks are not shown. See text.

Second and third fractions of the cellular sediment, together comprising one-half of its total volume, were then withdrawn as before and discarded. The final fraction, containing the red cells of 1 ml. whole blood together with the leucocytes of 4 ml. whole blood, was mixed with the remaining half of the decanted citrate plasma and injected into the marginal ear vein of animal R-2.

The two injections were repeated on the 3rd, 6th, and 9th days, so that each animal received a course of four injections in all. On the 16th day the sera of R-1 and R-2 were tested for donor red-cell agglutinins. None were expected and none were found. Two "large" pinch grafts were then cut from D, one being transplanted to R-1 and the other to R-2. Twelve days after grafting—28 days after the first injection—the grafts were removed, together with the tissue surrounding and underlying them, for histological examination. (It was clear to outward inspection that many had so far survived the homologous transplantation.) The histological reports are based upon the study of adjacent 8μ sections cut from not less than 5 equally-spaced vertical levels in the graft and stained with Ehrlich's haematoxylin, orange-G and eosin, and with Heidenhain's haematoxylin and picro-fuchsin respectively.

The results set out under the column-heading "Survival" in Tables I and II make use of the following notation (Medawar, 1945*a*):

1 = Complete survival of epidermal epithelium.
$\frac{3}{4}$ = The greater part of the epithelium surviving.
$\frac{1}{2}$ = About one-half of the epithelium surviving.
$\frac{1}{4}$ = The greater part of the epithelium destroyed.
0 = Total breakdown.

The propriety of basing a diagnosis of cellular survival upon histological evidence has been discussed in full by Medawar (1944). The "mark" awarded in the last column of the table is based upon a comparison between the pair of specimens afforded by each test: the mark "+" is awarded if the degree of survival in the R-1 graft is higher than in the R-2, "=" if it is the same, and "—" if it is less.

TABLE I.—*The Results of Strictly Paired Experiments Illustrating the Degree of Survival after* 12 *Days of Skin Homografts Transplanted to Rabbits Previously Injected through the Intravenous Route with Red Cells* (*R*-1) *and Red Cells + Leucocytes* (*R*-2).

Test No.	Donor.	Red-cell recipient (R-1).	Survival.	Leucocyte recipient (R-2).	Survival.	Mark.
1	383	384*	1	385*	1	=
2	388	389	$\frac{1}{2}$	390	1	—
3	393	394	1	395	0	+
4	403	404	0	405	$\frac{1}{2}$	—
5	408	409	1	410	0	+

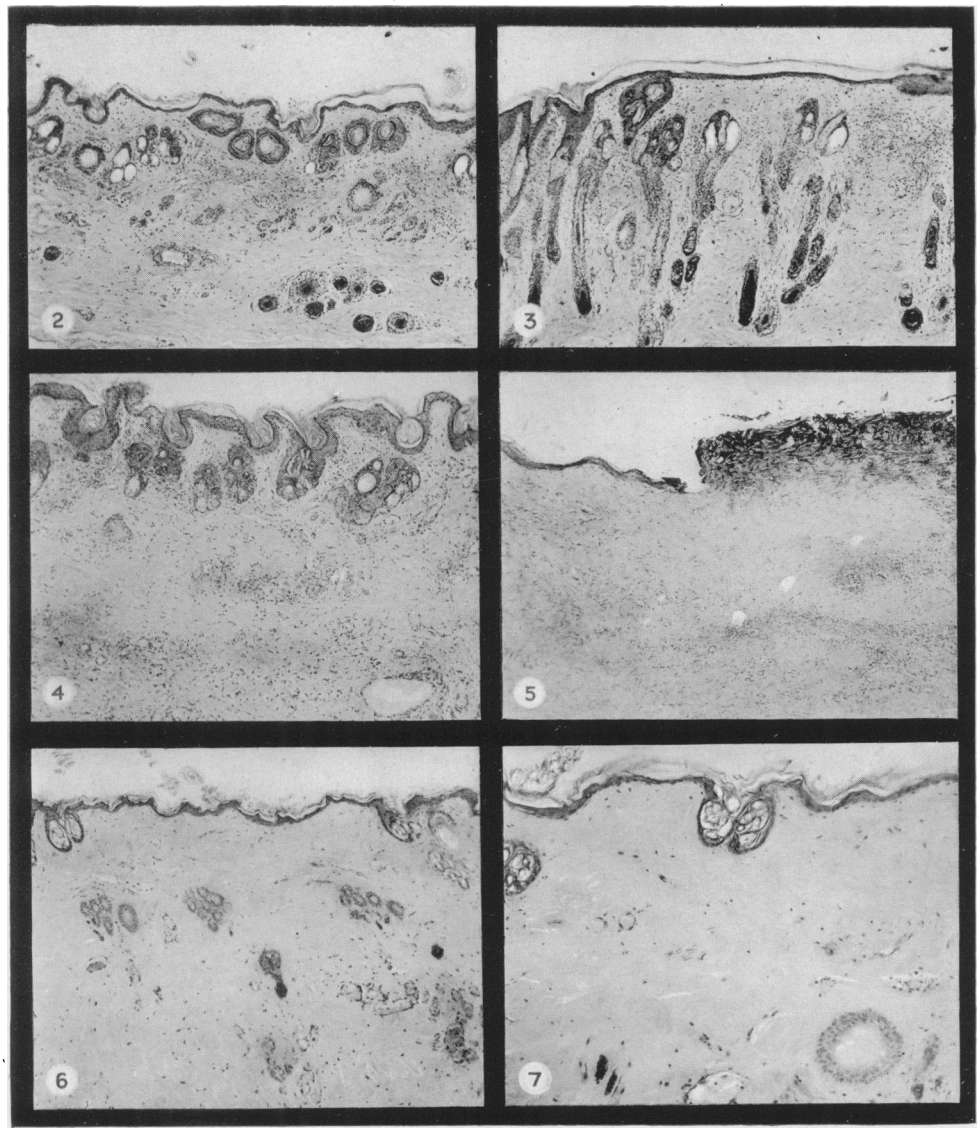
* Fig. 2 and 3.

(*All photographs are of sections stained with Ehrlich's hæmatoxylin, orange-G, and eosin.*)

FIG. 2 and 3.—Expt. 1, test 1: 12-day homografts from rabbits injected through the intravenous route with pure red cells (Fig. 2) and red cells + leucocytes (Fig. 3). Neither graft gives any evidence of an earlier immunization of its recipient. New hairs and glandular acini have matured in both, and the epithelium shows the retrograde thinning-out that follows the burst of hyperplasia in autografts and in all homografts that survive so long (Medawar, 1944). The specific vascular and mesenchyme cell reaction that is clearly developing is likewise in every respect characteristic of homografts that have been transplanted to *non*-immunized rabbits. (× 40.)

FIG. 4 and 5.—Expt. 2, test 3. Fig. 4 illustrates a 12-day homograft from a rabbit injected intradermally with a saline suspension of pure red cells. The condition is similar to that illustrated by Figs. 2 and 3, save that the hyperplastic condition of the superficial epithelium is more strongly marked and new hairs have not yet matured. By contrast, Fig. 5 illustrates a 12-day homograft from a rabbit which had received intradermal injections of pure leucocytes. Breakdown is of such long standing that native epithelium (seen on the left of the photograph) has begun to invade and overgrow the deeper levels of the graft dermis. (Fig. 4 × 40; Fig. 5 × 30.)

FIG. 6 and 7.—Expt. 4, test 4, illustrating in low and higher power views respectively the condition of a 6-day homograft from a rabbit immunized by a single intradermal injection of the leucocytes of 4 ml. whole blood. The epithelium of the graft is in a surviving condition and gives some evidence of general amoeboid activity (e.g. the upward migration of follicle epithelium towards the surface). Nevertheless cell division has been almost wholly suppressed: the epidermis is thin and unstratified and the cuticle remains delicate. (Fig. 6 × 40; Fig. 7 × 84.)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Reference to the table shows that the number of grafts showing *any* degree of survival was 4 out of 5 in the case of red-cell recipients and 3 out of 5 in the case of leucocyte recipients. On this score there is nothing to choose between them. The question of whether recipients of either class were to any degree immunized by the blood cell injections may be answered with reference to two independent groups of controls. Of 20 normal untreated rabbits which carried skin homografts in exactly similar dosage to that used in the experiments just described, 13 still bore surviving foreign epithelium on the 12th day (Medawar, 1945*a*). On the other hand (Medawar, 1945*a*), there is only one instance in 30 independent trials of a skin homograft surviving 12 days after transplantation to an animal immunized by an earlier grafting of skin from the same donor. The experimental results recorded here obviously belong to the former of these two categories ; there is no evidence of any immunization whatsoever.

This conclusion is decisively confirmed by the histological appearance of the experimental grafts. All of them showed either a typical generalized epidermal hyperplasia (Fig. 2 and 3) *or* evidence (e.g. visible acidophilic remains of hyperplastic follicle epithelium, multilayered cuticle, etc.) that hyperplasia had set in before the process of breakdown. Grafts transplanted to specifically immunized animals *never* undergo hyperplastic changes (cf. the first paper of this series). Moreover, three of the R-1 grafts and two of the R-2 grafts had progressed so far as to form new hairs (Fig. 2 and 3)—a state of affairs never once observed in grafts transplanted to immunized animals. Finally, the pattern of vascularization and the nature of the invasion of the grafts by leucocytes of native origin were in all respects characteristic of homografts transplanted to non-immunized animals.

Second Experiment.

In the second experiment the blood cells were introduced through the *intradermal* route. Although the experimental plan was in the main similar to that just described, the red-cell and leucocyte recipients were paired through a common donor in only three of the trials. The following additional modifications were made : (*a*) the cells were suspended in 2·0 ml. normal saline, and not in citrated plasma, before injection ; (*b*) only 0·1 ml. of the sedimented pure red-cell concentrate was administered to the R-1 animals, to reduce the disparity between the numbers of red cells and leucocytes injected ; (*c*) the leucocytes crudely separated by centrifugation in the manner already described were almost wholly freed from red cells by a second and, where necessary, a third centrifugation in tubes of 2 mm. diameter.

The injections were made at 2-day intervals, the R-2 rabbits receiving a course of three injections (and thus the leucocytes of 12 ml. whole blood in all) and the red-cell recipients a course of four. The cell suspension was on each occasion distributed over an average of 10 intradermal " blebs " spaced not less than 2 cm. apart on the shaved skin overlying the right thoracic wall. Care was taken to ensure that the suspension entered the compact collagenous tissue of the dermis, and not the looser " splitting " layer of connective tissue between the true dermis and the panniculus carnosus. The leucocyte injection-points in some animals came to be marked by small hard palpable nodules ; and the injection of red cells was found in some animals, but not in all, to provoke a rapid growth of coarse hair.

Not less than one week after the last injection, one large pinch graft was transplanted from the blood donor to the recipient(s). The grafts were removed for examination on the 12th day thereafter. The results of the experiment as a whole are set out in Table II, which makes use of the same notation as Table I.

TABLE II.—*The Results of Experiments Illustrating the Degree of Survival After 12 Days of Skin Homografts Transplanted to Rabbits Previously Injected through the Intradermal Route with Red Cells (R-1) and Leucocytes (R-2).*

Test No.	Donor.	Red-cell recipient (R-1).	Survival.	Leucocyte recipient (R-2).	Survival.
1	413	414	¼	415	0
2	418	...	...	420	0
3	423	424*	1	425*	0
4	428	429	¾	430	0
5	433	...	...	435	0
6	438	439	¼	...	...
7	438	440	0	...	...

* Fig. 4 and 5.

As before, two questions may be asked: Do the R-1 and R-2 animals differ in their responses? And if so, does either group give any evidence of immunization by blood cells? It is evident that the R-1 group has a significantly higher proportion of grafts (4 in 5) bearing some fraction of surviving foreign epithelium than the R-2 group (0 in 5), for mere luck of sampling would not be expected to reveal so severe a bias against the R-2 grafts more often than once in 42 random trials. It is equally clear that the R-1 group, with four surviving grafts in five, is virtually indistinguishable from the control group of 20 animals already referred to, in which 13 grafts were found to have survived. There is no evidence, therefore, that the intradermal injection of red cells in this dosage confers any immunity towards skin later grafted from the same donor. The dosage was small, and in no case was the formation of donor red-cell agglutinins observed; but it was of the same order of magnitude as that which, in the case of skin, is already known to be fully effective.

The leucocyte recipients, by contrast, give the clearest evidence of immunization: the disparity between the proportions of animals bearing surviving grafts in the experimental set (0 in 5) and in the control set (13 in 20) is such as might be expected to occur, by luck of sampling alone, not more often than once in 67 random trials. Histological evidence reinforces this conclusion (fig. 5). Breakdown in the grafts carried by R-2 animals was of such long standing that the surrounding native epidermis had in many cases begun to undermine and nip off the superficial layers of the homograft dermis (cf. Medawar, 1945*a*); the broken-down remains of the homograft epithelium gave no evidence of having at any earlier time undergone proliferation; and the primary blood vessels of the grafts had long since broken down and been replaced by the multiple capillary invasion of the dermis that constitutes a homograft's second cycle of vascularization (Medawar, 1944, 1945*a*). The grafts carried by the red-cell recipients were in all the properties described at the end of the section devoted to Experiment 1 indistinguishable from homografts transplanted to non-immune animals (*cf.* fig. 4).

Third Experiment.

In this experiment an attempt was made to decide whether really massive intravenous injections of whole blood conferred any degree of immunity towards skin later grafted from the blood donor. The diagnosis of the " skin-immune " state was made by the technique of mitotic counting described in the first paper of this series.

The experiment comprises five independent trials, each one making use of an independent pair of animals—donor (D) and recipient (R). An average volume of just over 12 ml. whole blood was withdrawn on each of six occasions at 2-day intervals from the median ear arteries of the donor, and received directly in to a tube containing 1·0 ml. of 4% Na citrate. With the least possible delay, the citrated blood was on each occasion transfused into any one of the ear veins of rabbit R, which received not less than 72 ml. whole blood in all. Nine days after the last injection (19 after the first) the serum of R was tested for red-cell agglutinins, and a single large pinch graft from the donor was fitted to the skin overlying the right thoracic wall. This graft was removed on the *sixth* day for the standardized routine of mitotic counting, and the serum of the recipient was for a second time tested for agglutinins.

TABLE III.—*The Results of Five Similar Tests Illustrating the Degree of Epidermal Proliferation After* 6 *Days in Skin Homografts Transplanted to Rabbits which had Earlier Received Massive Intravenous Injections of Whole Blood.*

Test No.	Donor and recipient.	Mean mitoses per median section ± S.E.	Graft diameter (mm.).	Mean mitoses per mm.	Agglutination test.	
					19 days.	25 days.
1	442 → 444	33·7 ± 2·2	6·9	4·9	0	0
2	445 → 446	35·8 ± 2·0	7·3	4·9	++	+
3	448 → 449	11·8 ± 0·7	7·5	1·6	0	0
4	451 → 452	10·0 ± 1·6	9·0	1·1	0	0
5	454 → 455	56·2 ± 3·4	7·5	7·6	0	0

The results of five such tests are summarized in Table III. It is evident that mitotic activity has not been wholly suppressed, and the grafts did indeed give straightforward histological evidence of some degree of epidermal hyperplasia. The mean of the entries in the column headed " Mean mitoses per mm.," with its standard error, is 4·00 ± 1·19; whereas the mean for homografts transplanted in high dosage to non-immunized animals had earlier been found from 10 independent 6-day estimates to be 7·21 ± 0·96. (The difference of dosage will tell against the control set, if it has any effect at all; for the survival time of homografts varies inversely with the graft dosage.) The recorded difference between the means derived from these two samples is such as might be expected to occur only once in 20 random drafts from the same normal population; so it is not unreasonable to infer that there has been some degree of immunization. Tests on a larger scale would be required to put the matter beyond reasonable doubt. The conclusion that " immunity to skin homografts following massive intravenous transfusions of homologous whole blood is either trivial or absent " does, however, appear to be warranted by the data.

It is curious that red-cell agglutinins developed in only one animal (446, test 2). Their occurence evidently bore no relationship to the mitotic frequency in the corresponding graft.

Fourth Experiment.

The second experiment showed that three intradermal injections of the leucocytes of 4 ml. whole blood were sufficient to establish a complete immunity towards skin later grafted from the same donor. In this section it is shown that a high but not complete degree of immunity may be generated by one such intradermal injection alone. Apart from the fact that only one intradermal injection was administered, the plan of this experiment differed from that governing the leucocyte recipients of Experiment 2, in that (*a*) the technique of mitotic counting was used for the diagnosis of the skin-immune state, as in Expt. 3 ; and (*b*) a different method of leucocyte fractionation was employed. After dilution with an equal volume of 0·8 per cent Na citrate in 0·85 per cent NaCl, the 4 ml. whole blood was spun so gently as to leave the majority of the leucocytes in suspension. The supernatant fluid was decanted and the volume of the blood restored to its original value by adding isotonic citrate saline. After spinning gently for a second and a third time, the leucocyte-containing supernatant fluids were combined and briskly spun down. The pure leucocyte sediment—not always easy to disperse—was finally taken up in 2·0 ml. normal saline, and injected in the form of ten "blebs" into the dermis of the skin overlying the right thoracic wall of the recipient R. Sixteen days after this injection animal R was grafted with the standard dosage of skin from D ; and six days after transplantation the graft was removed for mitotic counting.

This experiment is thus directly comparable with Expt. 3. Its results are summarized in Table IV.

TABLE IV.—*The Results of Five Similar Tests Illustrating the Degree of Epidermal Proliferation After 6 Days in Skin Homografts Transplanted to Rabbits which had Earlier Received Small Intradermal Injections of Leucocytes.*

Test No.	Donor and recipient.	Mean mitoses per median section ± S.E.	Graft diameter (mm.).	Mean mitoses per mm.
1	469 → 477	16·4 ± 2·1	9·0	1·8
2	473 → 480	41·0 ± 1·7	8·9	4·6
3	481 → 483	27·8 ± 1·9	7·5	3·7
4	484 → 486*	4·2 ± 0·5	8·7	0·5
5	478 → 487	0	8·4	0·0

* Fig. 6 and 7.

The mean of the entries in the column headed "Mean mitoses per mm." is 2·21 ± 0·87 ; that derived from the set of ten controls already cited in the discussion of Expt. 3, 7·21 ± 0·96. There can be no question of the significance of this difference : a single intradermal injection of the leucocytes of 4 ml. whole blood produces a high degree of immunity towards skin later grafted from the blood donor. It is not a complete immunity, for if it were so the mitotic frequency would have dropped to zero (see the first paper of this series). But it is certainly not less than that called forth by the *intravenous* transfusion of 72 ml. whole blood (Expt. 3) ; i.e. of a quantity of blood representing 18 times the leucocyte dosage.

Fig. 6 and 7 illustrate in low- and higher-power views respectively the condition of the epidermis in the graft removed from animal No. 486. The cells were clearly "alive" before fixation, but there is little indication of epidermal hyperplasia.

DISCUSSION.

The role of leucocytes.

The fact that leucocytes injected intradermally will immunize their recipient towards skin epithelium later transplanted from their donor shows that these two " tissues " share antigens in common. The " skin-immune " reaction is not, therefore, rigidly tissue-specific. The fact that the operative dose of leucocytes is of the same order of magnitude as that of a sufficient immunizing dose of skin suggests that the two tissues share a good many antigens in common. There is no evidence that they share *all* their antigens in common, and experiments of the type described here could not provide it.

Leucocytes are of purely mesenchymal origin. Since skin grafts carry with them a complement of dermal mesenchyme cells, it might be argued that the immunizing effect of leucocytes is exerted directly against them, and only indirectly against the homologous skin epithelium. This argument is untenable because (*a*) every test that has been used for the quantitative diagnosis of the " skin immune state " is based upon the reactions of epidermal epithelium alone ; and (*b*) these reactions are not significantly influenced by the presence of dermal mesenchyme. The pure epidermal epithelium which grows outwards from skin homografts transplanted to the middle of a widespread raw area responds in just the same manner as the " resident " epithelium on the surface of the graft itself (Gibson and Medawar, 1943 ; Medawar, 1944).

The route of injection.

The immunizing power of leucocytes towards skin is *at least* 18 times more effective through the intradermal route than through the intravenous. It will be noted that, whereas " every intradermal injection is truly intralymphatic " (McMaster and Kidd, 1937), a properly executed intravenous injection is made at just that point which is physiologically most remote from the lymph nodes (Expt. 3). The experiments reported here do not therefore conflict with the view that lymph nodes play a part in the manufacture of antibodies (McMaster and Hudack, 1935 ; Ehrich and Harris, 1942, 1945), though they can hardly be said to support it strongly.

A wide disparity between the effectiveness of the intradermal and intravenous routes of injection has already been observed in studies on the sensitization of skin to chemical compounds (Sabin and Joyner, 1938 ; Landsteiner and Chase, 1939).

The role of red cells.

In planning these experiments, it was not assumed that the antigens which red cells might share with skin were necessarily those that can be made to express themselves as iso-agglutinogens. It proved, however, that doses of red cells very much greater in bulk and cell number than those which are effective when skin itself is the immunizing agent did not confer skin-immunity of a type appreciable by the methods here described. Nor, conversely, did the grafting of massive doses of skin elicit the formation of homologous red-cell agglutinins. This second group of observations confirms the experience of Kozelka (1933), and Haddow (1934). On the other hand, Gorer (1937, 1938) discovered that a red-cell agglutinogen of Strong A-line mice was identical with one of two or

three antigens which generate immunity towards a sarcoma arising in the same line when it is transplanted to a host that lacks them. (This terminology is not that adopted by cancer research workers, who speak as a rule of genetic "factors for susceptibility.") It follows from Gorer's experiments that tissue cells and red blood cells may sometimes share antigens in common; but the practical consequences of this fact, so far as skin immunity is concerned, are evidently negligible.

Leucocytes and blood transfusion.

The behaviour of leucocytes is commonly neglected in transfusion practice, although fresh whole blood and blood of short storage certainly contains viable white cells. This neglect seems to be empirically well founded. The results of Expt. 3 in the text suggest that no significant leucocyte immunity would develop in response to transfusion through the intravenous route, though it might do so if the sternal route were used. The only clinically relevant effect that leucocyte immunity might be expected to exert is to prejudice the success of skin homografts when used as temporary wound dressings (Medawar, 1945*b*). This effect is of such a special character that it can obviously be neglected in transfusion practice.

SUMMARY

1. The transplantation of massive doses of skin from one rabbit to another does not elicit the formation of red-cell iso-agglutinins.

2. Intradermal and intravenous injections of foreign homologous red cells confer no appreciable immunity towards skin later grafted from the red-cell donor.

3. The intradermal injection of foreign homologous leucocytes confers a typical immunity towards skin later grafted from the leucocyte donor. It is inferred that leucocytes share antigens in common with the antigens of skin, and that tissue transplantation immunity is not rigidly tissue-specific.

4. The immunizing effect of leucocytes is at least 18 times more effective through the intradermal route than through the intravenous.

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