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39

Canine Liver Homotransplants

The Effect of Host and Graft Irradiation

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With homografts of various tissues and organs, systemic histologic changes in the host animal are not usually detectable. Recently, however, in studying whole-organ homografts of the canine liver, widespread changes which were coincident with rejection were found in host organs.[6] The changes consisted of round cell infiltration and mesenchymal tissue proliferation. They were found in bone marrow, lung, kidney, and other tissue and organs which are constituents of the reticuloendothelial system.

From the information available, it was not possible to determine if these changes were part of an exaggerated host-versus-graft rejection process, or if they resulted from an attempt by the graft to repudiate the host (graft-versus-host reaction). In the present report, an attempt was made to study this point by rendering either the graft or host animal incapable of immunologic activity by preoperative ionizing irradiation.

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Methods

Adult mongrel dogs weighing 15 to 20 kg. were used. The animals were dewormed and immunized against distemper and infectious canine hepatitis. Using a previously described technique, liver homografts were transplanted with normal anatomic reconstruction of vena cava and portal veins.[5] The host spleen was removed. Blood chemistry and hematologic data were obtained before and after operation. Autopsy was performed promptly after death.

Ionizing irradiation was given either to the prospective recipient animal or to the dog from which the liver homograft was to be obtained. All animals

were given 1,400 r total body irradiation in a single dose. The dose was measured on the surface of the dog, including back scatter; 700 r to the right lateral side, 700 r to the left lateral side. Factors were 280 kv. hvl., 1 mm. Cu, 20 ma, target dog distance 167 cm. Dose rate at the surface of the dog was 14.2 r per minute. The dose to the center of the dog's abdomen was calculated to be 900 to 1,000 r.

Results

Irradiation of Host.—Six recipient dogs were given 1,400 r. Eighteen to 40 hour later total hepatectomy was performed and a liver transplanted from a normal dog. All animals died within 36 hours from diffuse gastrointestinal hemorrhage or hemorrhagic pneumonitis. The combination of operative trauma and irradiation appeared to preclude even temporary success.

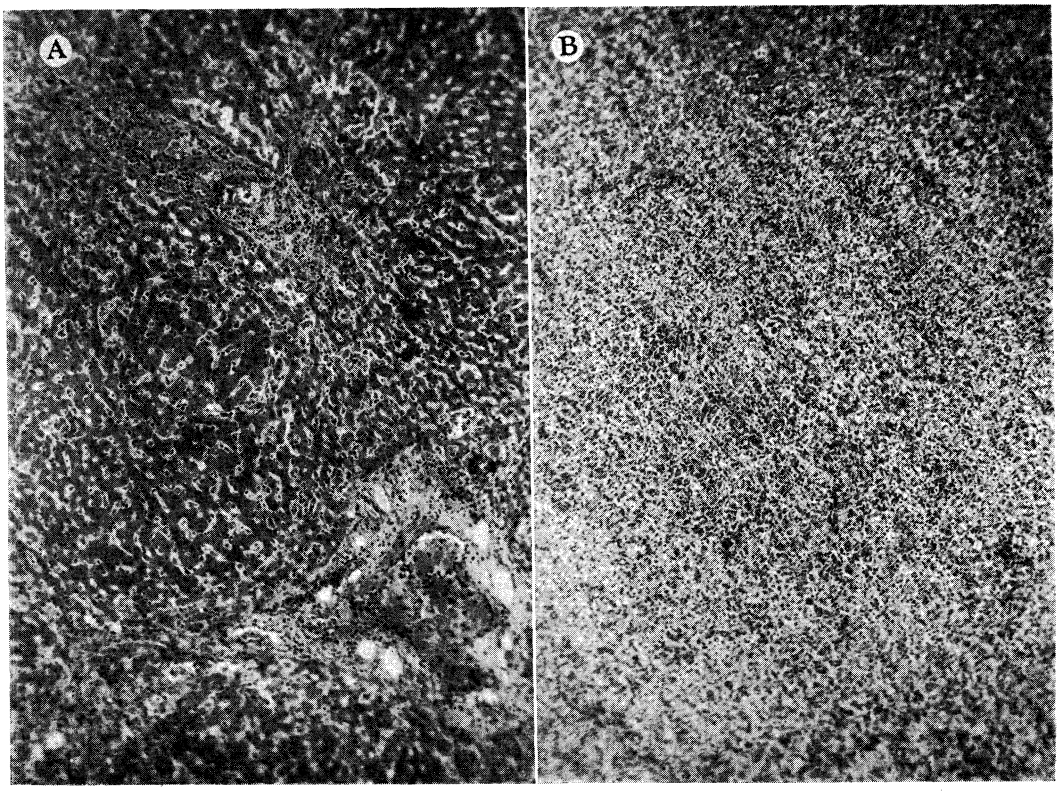
Irradiation of Graft.—Twelve donor dogs were given 1,400 r. Eighteen hours later, their livers were transplanted to normal hosts. Six dogs died within 30 hours of hemorrhagic gastroenteritis. The other 6, which constitute the bulk of the report, recovered and were studied until their later death.

Survival and Clinical Behavior: The 6 dogs which survived the immediate effects of surgery lived for 5, 6, 8, 9½, 13, and 13½ days. Clinical behavior was similar to that previously observed with the use of nonirradiated grafts[3,6] and was characterized by fever, eventual lassitude, cessation of oral intake, and jaundice.

Blood and Urine Studies: All dogs had progressive elevation of serum bilirubin and alkaline phosphatase after the fifth and sixth days. Hypoglycemia was frequent late in the course. There were no changes in blood urea nitrogen. After 4 to 6 days, bile was detected in the urine. Before death, all 6 dogs had rises in the white count to as high as 45,000 per cubic millimeter.

Gross Findings: Findings were similar to those obtained with use of nonirradiated grafts.[5] The livers were usually large, firm,

Fig. 1.—Liver in early and late stage of rejection. *A*, dog No. 6: 6 days; *B*, dog No. 9: 9 days. Reduced about 39% from mag. × 100.

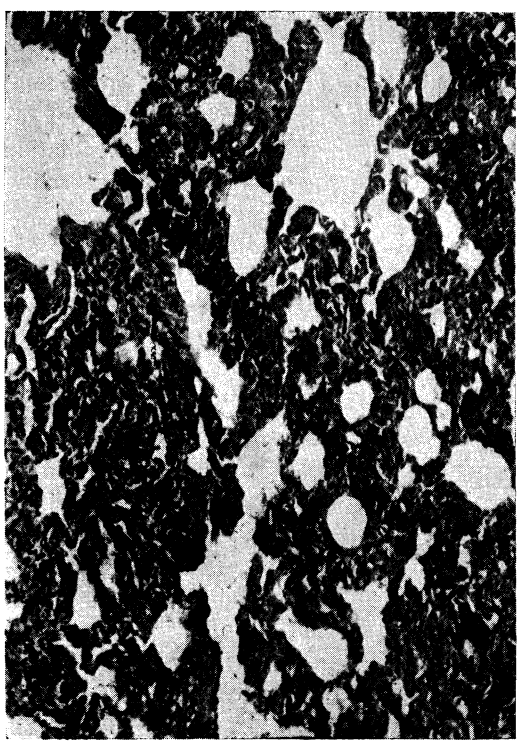


and either pale or mottled. Ascites was variable. In 2 of the animals, there was no obvious cause of death other than graft rejection. In the other 4, massive hemorrhage, ileal intussusception, hemorrhagic ileitis, or duodenal perforation were found and could have been the cause of death. Host lymph nodes were enlarged throughout.

Microscopic Findings: In the animals living 5 days or less, there were no mononuclear infiltrates in the livers. Structure was well preserved. In 6 days, well-defined diffuse and focal infiltrates of plasma cells and lymphocytes were evident (Fig. 1*A*), and in longer surviving animals, these were very prominent. After 6 days, structural preservation varied from an intact architecture (Fig. 1*A*) to almost complete destruction (Fig. 1*B*). The primary area of hepatic parenchymal loss were central lobular and periportal (Fig. 1).

Histologic evidence of host response was found. In 5 of the 6 animals surviving 5 days or more, proliferative changes were found in the alveolar septa (Fig. 2). In the kidneys, there were focal aggregates of mononuclear cells (Fig. 3). Bone marrows were consistently altered. Slight to marked increases in plasma cells, lymphocytes, and reticulum cells were evident in the marrow of each animal. An example of such a change of intermediate severity is shown in Figure 4.

Fig. 2.—Lung from dog No. 13, after 18 days. Note proliferation in alveolar septa. Reduced about 39% from mag. × 250.



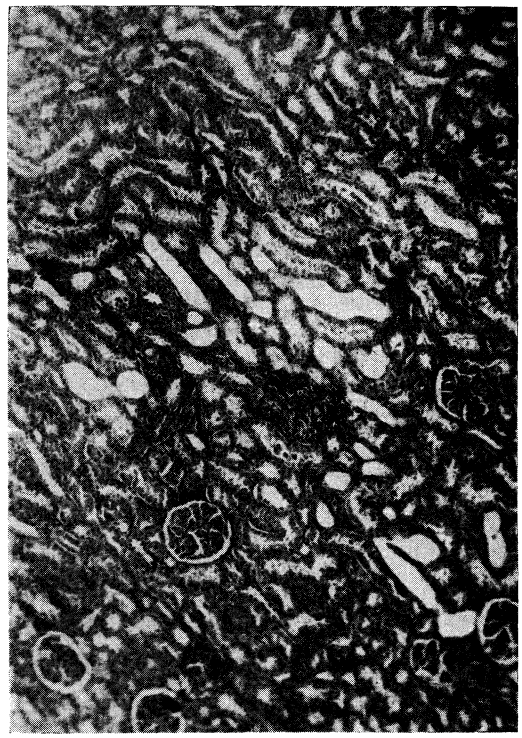


Fig. 3.—Kidney from dog No. 16, after 13½ days. Cellular infiltrate in central part of illustration is focal. Reduced about 39% from mag. × 100.

In all the dogs, there was evidence of increased cellular activity in the mediastinal and mesenteric lymph nodes. The number of plasma cells were increased, especially in the medullary sinuses. In half of the animals, there was distortion or absence of the follicles (Fig. 5). In the other half, the basic lymph node architecture was preserved. The gastrointestinal tract, pancreas, and heart showed variable changes of the type previously related, at least in part, to the trauma of surgery.[5,6] There were no changes in skeletal muscle.

Fig. 4.—Bone marrow after 8 days from dog No. 13 showing increased numbers of plasma cells and lymphocytes. Reduced about 41% from mag. × 430.

Fig. 5.—Lymph node from dog No. 13, an 8 day survivor. Washed out follicular pattern is discernible. Reduced about 39% from mag. × 100.

Controls

Previous control studies demonstrated that the act of transplantation did not by itself result in major histologic changes in host or graft.[6] Controls in the present study were designed to determine the effect of irradiation on the histologic character of the liver. Nine animals were given 1,000-1,400 r, with the same factors of irradiation as described above, and observed until death 4 to 13 days later. In all cases, the platelet and white counts fell precipitately after 3 to 5 days.

The architecture of the liver was well preserved. Infiltrates did not occur. However, small necrotic foci with noninflammatory response were found in 4 animals. Loss of basophilic staining was inconstantly noted.

Comment

Initially, it was hoped to perform experiments in which either the liver homograft or the host received a high dose of irradiation. However, efforts at host irradiation were unsuccessful. Irradiation injury to the bowel, combined with the transient elevation of portal pressure during transplantation, resulted in irreversible intestinal injury and fatal gastrointestinal hemorrhage.

The information gained from irradiation of the hepatic graft is useful in interpreting the interaction between the graft and the host. The graft was irradiated in situ approximately 18 hours before its removal. The calculated dose to the liver was in excess of 1,000 r, enough to effect complete immunologic paralysis of the graft.[2] Having thus eliminated the possibility of a graft-versus-host reaction, the changes in both graft and host can be attributed to the antigenic stimulus of the graft and the resultant response of the host. Survival time, clinical behavior, chemical determination, and pathologic findings were similar to those previously reported in recipients of nonirradiated livers.[3,6]

The origin of the cellular infiltrate in rejected homografts has been the source

of speculation and controversy for several years. It has been suggested by Simonsen [4] and Dempster [1] that the mononuclear infiltrates are of graft origin and represent a graft-versus-host reaction. In the present study, however, the histologic character of the rejected irradiated liver did not differ from that previously observed in nonirradiated homografts.[6] These observations provide support for Hume's contention,[2] derived from studies with renal and hematopoetic homotransplants, that the cellular infiltrate in homografts undergoing rejection is of host origin.

In earlier studies of nonirradiated liver homografts, various host organs displayed alterations which resembled those found in grafts undergoing early rejection. The changes consisted of fixed tissue proliferation and round cell infiltration.[6] It was not possible to be certain if they were part of the host rejection of the graft or if they represented a graft-versus-host reaction. The present study, employing immunologically immobilized hepatic grafts, provides evidence that the abnormalities in the hosts' bone marrow, lungs, kidneys, lymph nodes, and other tissues were due to an exaggerated host response to the massive antigenic stimulus of the foreign liver.

Summary

Whole organ hepatic homografts were performed after massive irradiation of the host animal or of the animal from which the graft was taken. After host irradiation, technical success with grafting was not achieved.

With irradiation and consequent immunologic paralysis of the liver homograft, survival for as long as 13½ days was attained. Length of survival, clinical behavior, chemical determinations, and pathologic findings were essentially the same with the use of irradiated as with nonirradiated livers. These findings indicate that a graft-versus-host reaction is not of immediate significance even with as bulky a graft as the liver. Histologic changes in the liver graft, as well as those in various host organs, appear to be attributable exclusively to host activity in response to the antigenic stimulus of the liver.

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