the operation state that immediately there is a disappearance of nervous tension. Although memory is intact and conflicts still exist, they cause little concern. They report their patients are able to formulate ideas but lack the accompanying emotional drive that is necessary to put them into effect. As a result their productivity is usually markedly impaired.

Summary. By using small electrodes in electroshock treatment it was possible to lo-

calize the stimulation sufficiently to separate some of the many phenomena that occur with conventional electroshock therapy. The small amount of data thus far accumulated seems to indicate: (1) Memory loss and convulsion are probably not necessary to produce clinical improvement. (2) Stimulation of cortical autonomic centers seems to be the most important factor in producing clinical improvement in the affective disorders with electroshock.

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Diabetes Mellitus in Rabbits Injected with Dialuric Acid.*

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Since the discovery that alloxan produces diabetes when injected into animals¹⁻³ other chemically related substances have been studied for possible diabetogenic action. Until recently no chemical with such action has been found.

Koref, Vargos, Rodriguez and Telchi⁴ reported the production of diabetes with alloxantin and confirmation of this work has already been published from this laboratory.⁵ Recently, Bruckman and Wertheimer⁶ have found that besides alloxantin methylalloxan, dialuric acid, methyl dialuric acid and dimethyl alloxantin produce diabetes when

¹ Bailey, C. C., and Bailey, O. T., J. A. M. A., 1943, **122**, 1165.

² Goldner, M. G., and Gomori, G., *Endocrinology*, 1943, **33**, 297.

³ Dunn, J. S., and McLetchie, N. G. B., Lancet, 1943, 2, 384.

⁴ Koref, O., Vargos, L., Rodriquez, F. H., and Telchi, A., *Endocrinology*, 1944, **35**, 391.

⁵ Bailey, C. C., Bailey, O. T., and Leech, R. S., Bull. New Eng. Med. Center, 1945, 7, 59.

⁶ Bruckmann, G., and Wertheimer, E., Nature, 1945, 155, 267.

injected into rats. The present investigation reports the production of diabetes in the rabbit with dialuric acid and compares the diabetes so produced, clinically and histologically, with the diabetes produced with alloxan.

Experimental. Eight chinchilla male rabbits each weighing 1500-2000 g were injected intravenously with dialuric acid in doses of 150, 200 or 250 mg per kg. The rabbits were not fasted at any time during the experimental period and in all but one instance the injection was made with the animal under nembutal anesthesia. Anesthesia seemed advisable since the dialuric acid was injected as a hot solution, owing to its insolubility at lower temperatures.

The solution of dialuric acid was prepared as a 1.5% solution in nearly boiling distilled water. The solution was cooled to a temperature of $40^{\circ}-50^{\circ}$ C and injected immediately. It was found necessary to keep the solution at this temperature to avoid recrystallization of the dialuric acid which tended to occur at room temperature.

Blood samples were taken from the ear veins before and at frequent intervals following the injection of the dialuric acid.

^{*} This investigation has been aided by a grant from the American Cyanamid Company, who also supplied the dialuric acid.

Determinations of blood sugar were made routinely by the micro procedure of Folin and Malmros⁷ and in one instance (Rabbit No. 1) checked by a micro copper method referred to in an earlier paper.⁸

The chemical relationship between alloxan, dialuric acid and alloxantin is shown in the formulas below:



Dialuric acid is a reduction product of alloxan and may be oxidized to alloxan. That oxidation of dialuric acid to alloxan occurs easily in very dilute solutions in the presence of air has been emphasized by Archibald.⁹ However, by applying Archibald's differentiating phosphotungstic acid test⁹ to a warm 1.5% solution of dialuric acid (the concentration used in the experimental work) we have found less than 10% oxidation to alloxan occurring during the first 15 minutes at room temperature and only about 20% during a half hour at room temperature and reheating.

These results indicate that a solution of dialuric acid prepared as indicated contains during the first 30 minutes after its preparation at least 80% dialuric acid and probably no more than 10-20% alloxan.

Results. Chemical. The blood sugar results of the animals injected with 250 or 200 mg/kg are shown in Fig. 1. Three of these rabbits developed severe diabetes, one (No. 3) developed an initial blood sugar

⁹ Archibald, R. M., J. Biol. Chem., 1945, 158, 347.

curve similar to the alloxan-injected animal but died 12 hours after the injection. Rabbit No. 5, though showing an initial hyperglycemia, did not develop diabetes.

Rabbits No. 6, No. 7 and No. 8, injected with 150 mg/kg, did not develop diabetes. No. 8 showed a tendency toward the transitory type of diabetes with one blood sugar of 278 mg % on the third day following injection, but a glucose tolerance on this rabbit 40 days after the injection was normal.

These results suggest that the rabbit requires a slightly larger dose of dialuric acid to produce permanent diabetes than of alloxan. If dialuric acid is given in 200-250 mg/kg doses, however, the blood sugar changes produced seem to parallel those observed in alloxan-injected animals.

Histology. Pancreas. The early stages of the pancreatic lesion were not studied since our purpose was to compare the fully developed lesions due to dialuric acid and to alloxan. The pancreas of the rabbits rendered diabetic by the injection of dialuric acid was characterized by paucity of islets and the small size of those remaining. А large percentage of the islets had disappeared completely. Their former sites were identified only with difficulty, being marked only by small masses of connective tissue without inflammatory cellular infiltration or residual traces of islet cells. In those islets which remained, the beta cells had entirely disappeared without concomitant infiltration with lymphocytes or polymorphonuclear leuko-Occasional alpha cells of normal cvtes. morphology were seen, but more numerous alpha cells had pyknotic nuclei and agranular cytoplasm at the stage studied. The acinar and ductile tissues, stroma and blood vessels of the pancreas were within normal limits. The lesion was thus extremely similar to that seen in the pancreas in alloxan diabetes,¹⁰ the only distinction being that some degenerating islet cells were present even when the rabbits were well established in diabetes.

Kidneys. There was considerable damage

¹⁰ Bailey, O. T., Bailey, C. C., and Hagen, W. H., Am. J. Med. Sci., 1944, **208**, 450.

⁷ Folin, O., and Malmros, H., J. Biol. Chem., 1929, 83, 115.

⁸ Leech, R. S., and Bailey, C. C., *J. Biol. Chem.*, 1945, **157**, 525.

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to the renal convoluted tubules. The tubular epithelia cells were swollen and the nuclei of many of them were pyknotic. They contained protein precipitate. The glomeruli, convoluted tubules, pelvic epithelium and blood vessels were within normal limits. The character of the renal damage was similar to that seen in alloxan diabetes. Since the extent of the kidney changes varied considerably in our large series of rabbits with alloxan diabetes, it is difficult to compare the degree of renal injury resulting from the injection of the 2 substances. In the material at hand, the renal changes due to dialuric acid seemed to be at least as great and possibly greater than those produced by alloxan.

Other Organs. The liver was somewhat congested, but there was no fatty infiltration. Other organs contained no lesions not found in control animals.

The adrenal showed no infiltration with polymorphonuclear leukocytes or other pathologic changes.

In Rabbit No. 3, which developed severe hypoglycemia and died during the night after injection of dialuric acid, the lesions were at an earlier stage than those of the diabetic animals. In many islets the central beta cells were pyknotic and the cytoplasm agranular, while some alpha cells at the periphery were essentially normal. In other islets all the cells had pyknotic nuclei. Again the degeneration and disintegration of the islet cells were unaccompanied by infiltration of inflammatory cells. The histologic features of other organs were similar to those already described in the diabetic animals.

The rabbit which developed transitory diabetes (No. 8) showed much less marked changes in the pancreatic islets. The alpha cells and many of the beta cells were normal in appearance. Some of the beta cells were pyknotic, while others were judged to have disappeared, since they were less numerous than in control rabbits. There was no inflammatory cellular infiltration. No hvdroptic degeneration or mitoses were seen The epithelial cells of in the islet cells. the convoluted tubules of the kidney were swollen but there was less pyknosis and less protein precipitate in the lumen than in the diabetic animals.

In the rabbits (No. 5, 6, 7) not developing diabetes, either permanent or transitory, there were only a few pyknotic beta cells in some of the pancreatic islets and the other pancreatic changes were absent. The degree of renal damage, however, was as great as in the rabbits developing transitory diabetes.

These studies have been based on tissues fixed in acetic Zenker's solution and stained with phloxine-methylene blue and with Gomori's method for the cells of the pancreatic islets.

Discussion. Clinically the diabetes produced in rabbits by dialuric acid and by alloxan was identical except for the fact that a slightly larger dose is necessary when employing dialuric acid.

The triphasic blood sugar curve consisting of an initial transitory hyperglycemia followed by hypoglycemia and finally a permanent hyperglycemia or diabetes is found after the injection of either dialuric or alloxan. Permanent diabetes may be produced with either drug and with the use of smaller doses transitory diabetes may occur.

The histologic changes described as the result of injection of dialuric acid are closely similar to those due to the injection of alloxan. The few variations mentioned are trifling when considered from the functional point of view. The cardinal features of alloxan diabetes in the rabbit-necrosis of islet cells with survival of a few alpha cells, moderate to mild injury to the renal convoluted tubules, with only minor changes in other organs-are duplicated in dialuric acid diabetes. Such evidence as can be gleaned from these experiments suggests that dialuric acid diabetes, like alloxan diabetes, is due to a lesion of the islets of Langerhans, which is degenerative from its inception.

Since dialuric acid may be obtained as a reduction product of alloxan, it might be questioned whether one of these substances is converted into the other in the body of the experimental animal before the lesions are initiated. The present series of experiments throws no light on this question other than to indicate that the lesions caused by the 2 substances are similar and are consistent with either view.

Dialuric acid is not recommended as a substitute for alloxan as an experimental technic for the production of diabetes, because it must be injected in a warm solution owing to its low solubility. Except for the extra labor involved, however, dialuric acid and alloxan may be regarded as interchangeable as diabetogenic agents.

Summary. Dialuric acid when injected intravenously into rabbits produces diabetes which is indistinguishable clinically or pathologically from alloxan diabetes.