PER OS TREATMENT OF EXPERIMENTAL DIABETES MELLITUS

S. G. Genes, A. A. Plavskaia and M. Z. Iurchenko

From the Division of Pathological Physiology (Head — Honored Scientist Prof. S. G. Genes) of the Ukrainian Institute of Experimental Endocrinology (Director — Candidate Med. Sci. S. V. Maksimov), Khar'kov

(Received November 4, 1957. Presented by Active Member of the AMN SSSR V. N. Chernigovskii)

At the beginning of 1957, in the Ukrainian Institute of Experimental Endocrinology, was synthesized 4-methylbenzenesulfonylbutyl urea (T. F. Sysoeva and N. I. Makhnenko). In the Soviet Union this preparation is called butamid; abroad it is called D-860, orinase or tolbutamide (in the USA), rastinon or artosine (in Germany) and dolipol (in France). Like the preparation B-55 (nadisan, invenol, glucidoral or carbutamide) it possesses a hypoglycemic action on per os administration, but in contrast to nadisan it is practically without any antibacterial properties. Treatment of patients with diabetes mellitus lasts throughout life, and so the absence of antibacterial properties is an important advantage of butamid. It is, in addition, less toxic than nadisan.

As previous investigations (1957) have shown, butamid in a dose of 0.5 g per 1 kg body weight lowered the blood sugar of normal rabbits for 24 hours by 33-52%, while the blood sugar level in control animals fell by 8-10% and in dogs (in a dose of 100 mg per 1 kg body weight) — by 23% and 36% compared with 15 and 10% in control experiments. Butamid acted differently on the blood sugar of dogs with alloxan diabetes. In some animals the hypoglycemic sulfonamides lowered the blood sugar and in others its level was not altered. The same thing was observed in dogs with alloxan diabetes after injection of these animals with butamid. This effect evidently depends on the amount of residual islet tissue: the more it is damaged, the weaker the hypoglycemic action of the sulfonamides will appear.

We subsequently set out to discover whether butamid reinforces the action of insulin when injected from outside.

EXPERIMENTAL METHOD

Experiments were carried out on dogs with alloxan diabetes and subjected to total pancreatectomy, requiring considerable amounts of insulin. The animals spent the whole time in metabolism cages. Because of this, their daily dietary intake could be calculated and the excreted carbohydrate estimated, from which the utilized carbohydrate could be determined. The effectiveness of butamid was judged by the intensity of fall of hyperglycemia and glycosuria, by the value of utilized carbohydrate and by the amount of insulin which could be dispensed with by means of butamid. The blood sugar was determined by the Hagedorn-Jensen method, and the sugar in the urine polarimetrically. The investigation was carried out on 4 dogs with alloxan diabetes and receiving large amounts of insulin: 16, 24, 32, and 40 units per day. This quantity of insulin did not abolish the hyperglycemia and glycosuria. Butamid was injected into all the dogs in a dose of 100 mg per 1 kg body weight. 84 experiments were performed on each dog. In some experiments (control) the effects were studied on hyperglycemia, glycosuria and assimilation of carbohydrate of insulin alone, and in others — of insulin and butamid. Experiments of a similar type were carried out also on dogs subjected to total pancreatectomy. These received daily 150 g of raw pancreas in addition to their usual diet. We performed 134 experiments on the dog Mal'chik, and 66 on Pushok. Insulin was injected twice daily into the animals — at 9 a.m. and 4 p. m., and butamid — at 9 a.m. (except in a few experiments when it was injected at 2 p.m.).
EXPERIMENTAL RESULTS

The results in Table 1 demonstrate the considerable reinforcement by butamid of the insulin effect. The same results were obtained in comparable experiments on all 4 dogs.

Later on we attempted to discover the intensity of action of butamid — the amount of insulin which could be replaced by butamid. For this purpose the doses of insulin injected into the dogs were gradually reduced to values which, together with butamid, would give the same effect as large doses of insulin alone.

### TABLE 1

<table>
<thead>
<tr>
<th>Dogs</th>
<th>Sivka</th>
<th>Torba</th>
</tr>
</thead>
<tbody>
<tr>
<td>Data of experiments</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of experiments</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Dose of insulin, units</td>
<td>24</td>
<td>24+B</td>
</tr>
<tr>
<td>Hyperglycemia, in mg%</td>
<td>349</td>
<td>211</td>
</tr>
<tr>
<td>Glycosuria, g</td>
<td>45</td>
<td>5</td>
</tr>
<tr>
<td>Assimilation of carbohydrate, g</td>
<td>119</td>
<td>148</td>
</tr>
</tbody>
</table>

*In this and the subsequent tables, B — butamid.

The figures in Table 2 show that butamid reinforces the action of insulin by 25, 33 and 50%. This means that with the aid of butamid it is possible to reduce the insulin requirement of alloxan-diabetic dogs by 25-50% respectively.

What is the mechanism of reinforcement of the insulin effect by butamid?

The sulfinilamides are known not to influence the blood sugar level of animals from which the pancreas has been removed. On this basis Loubatieres put forward the hypothesis that the sulfinilamides stimulated the β-cells of the insulin apparatus. This hypothesis was supported by several authors [1, 4, 5, 6, 9].

Although in the lowering of the blood sugar by sulfanilamides, depression of the glucose-6-phosphatase activity of the liver may possibly have some importance, together with a reduction in the absorption of glucose from the intestine and other factors, Holt et al. [1] nevertheless assert that the main role in this activity belongs to stimulation of the β-cells and not to extrapancreatic reinforcement of the insulin effect. Some authors have tried to prove the importance of suppression of the α-cells in the hypoglycemic effect of the sulfinilamides. However, investigation of the factors quoted as evidence in favor of this action of the sulfinilamides failed to confirm them.

Nevertheless there appeared recently a paper by Schmidt and Blobel [10], which showed that under the influence of D-860, rats always showed a reduction (statistically significant) in the nuclear volume of the α-cells of the insulin apparatus.

If depression of the α-cells is really of importance in the hypoglycemic effect of the sulfinilamides, it is not clear why butamid, like nadisan, does not affect the blood sugar level in animals with severe alloxan diabetes. It is not clear why the effect of sulfinilamides is not shown in patients with severe diabetes mellitus. From the point of view of the importance of the β-cells in the hypoglycemic effect of these preparations, its absence in some of them may be due to the small number of β-cells which are left or to severe weakening of their function.

But how do we explain the reinforcement of the insulin effect in all the alloxan-diabetic animals? It seemed to us that this effect in the dogs with severe alloxan diabetes depends not on the action of butamid on the pancreas but on its action on the insulin effect itself. We verified this hypothesis on totally depancreatized dogs receiving a definite amount of insulin which did not abolish the hyperglycemia and glycosuria (Table 3).