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THE EFFECTS OF SODIUM SALICYLATE ON BLOOD GLUCOSE IN THE RAT

BY

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The injection of salicylate into normal male rats has been observed to cause a marked depletion of the liver glycogen level at 4 hr., but the blood glucose concentration remained constant (Lutwak-Mann, 1942; Smith, Meade, and Bornstein, 1952). A similar reduction in the liver glycogen content of mice 1 and 2 hr. after the injection of salicylate was reported by Sproull (1954). No significant alteration in the blood glucose level accompanied this change in the liver glycogen in male mice, but female mice showed a hyperglycaemia.

The effect of salicylate on the blood glucose of male and female rats has therefore been studied to determine if this sex difference also applied to rats. The effect of fasting for 24 hr. before the experiment was also investigated, and, in order to assess the influence of the adrenal, similar experiments were performed on adrenal-demedullated and on adrenalectomized rats.

METHODS

Animals.—Rats of the Wistar strain, weighing 200–300 g. and maintained on a diet of cubes (M.R.C. Diet 41), were used. Adrenal demedullation was performed according to the directions in Farris and Griffith (1949). The rats were given 0.45% (w/v) NaCl soln. to drink instead of water for the first post-operative week only, and were used 40 days after operation. The bilaterally adrenalectomized rats were used eight days after operation, having been maintained on 0.45% (w/v) NaCl soln. instead of drinking water. The animals were used in groups of 20 which were divided into two sub-groups. One sub-group received an intraperitoneal injection of 0.5 mg./g. body weight of a 10% (w/v) solution of sodium salicylate and the other was given a similar injection of an equal volume of 0.9% (w/v) NaCl soln. Blood glucose estimations by the method of Nelson (1944) were made on tail-vein samples obtained immediately before and at intervals of 1, 2, 3, and 4 hr. after the injections. The rats were either deprived of food only from the beginning of the experiment (non-fasted animals) or for 24 hr. previously (fasted animals).

The following groups of rats were used: (1) non-fasted and fasted females; (2) non-fasted and fasted males; (3) non-fasted and fasted adrenal-demedullated males; and (4) non-fasted adrenalectomized males.

RESULTS

The blood glucose results on female rats are given in Table I. In all the following tables the significances of the differences between the means have been analysed by Student's *t*-test, and values of P are included. The minimum acceptable level of significance has been taken as $P=0.02$.

TABLE I

THE EFFECT OF SALICYLATE ON THE BLOOD GLUCOSE OF NON-FASTED AND FASTED FEMALE RATS

Mean values±S.E. (no. of rats in parentheses) for blood glucose levels expressed as mg./100 ml.

Time after Injection (hr.)	Non-fasted			Fasted		
	Control (8)	Salicylate-treated (12)	P	Control (8)	Salicylate-treated (11)	P
0	88·6±4·5	83·6± 9·0	0·1	70·6± 7·0	64·8± 5·4	0·1
1	93·6±6·0	129·1±10·6	0·001	69·3± 7·5	81·5±10·0	0·02
2	91·7±6·0	118·5±15·6	0·001	78·5± 7·5	98·6±11·5	0·001
3	90·6±7·5	105·0± 8·4	0·01	75·0±11·1	90·5±13·2	0·02
4	90·1±6·2	91·6±11·7	0·8	73·3± 5·9	87·0±17·9	0·05

The results show that the injection of sodium salicylate caused a significant but moderate hyperglycaemia in both non-fasted and fasted female rats 1, 2, and 3 hr. after the injection. The effect appeared to be more prolonged in the fasted animals, although the results of the salicylate-treated and control animals at 4 hr. were not significantly different.

The blood glucose results on male rats are given in Table II.

The results are essentially similar to those obtained with the female rats: a hyperglycaemia occurred 1 and 2 hr. after the injection in the non-fasted males and persisted for 4 hr. in the fasted male animals.

TABLE II

THE EFFECT OF SALICYLATE ON THE BLOOD GLUCOSE OF NON-FASTED AND FASTED MALE RATS

Mean values±S.E. (no. of rats in parentheses) for blood glucose levels expressed as mg./100 ml.

Time after Injection (hr.)	Non-fasted			Fasted		
	Control (8)	Salicylate-treated (12)	P	Control (6)	Salicylate-treated (11)	P
0	87·1±7·0	81·4± 6·3	0·1	68·0±3·3	67·4± 5·7	0·9
1	90·4±5·2	127·9±14·2	0·001	69·3±6·2	87·9±13·4	0·01
2	90·5±6·6	118·9±17·7	0·001	68·0±5·6	91·5±13·5	0·01
3	90·4±7·2	95·0±12·1	0·4	75·0±9·3	93·5±13·3	0·02
4	83·8±6·6	83·5± 9·7	0·9	77·3±8·7	94·5± 9·1	0·01

TABLE III

THE EFFECT OF SALICYLATE ON THE BLOOD GLUCOSE OF NON-FASTED AND FASTED MALE ADRENAL-DEMEDULLATED RATS

Mean values ± S.E. (no. of rats in parentheses) for blood glucose levels expressed as mg./100 ml.

Time after Injection (hr.)	Non-fasted			Fasted		
	Control (8)	Salicylate-treated (10)	P	Control (8)	Salicylate-treated (11)	P
0	84·2±8·0	78·1±14·9	0·4	73·1±12·1	66·0±6·2	0·1
1	85·0±6·2	101·3±15·9	0·02	71·1± 5·4	74·1±8·3	0·4
2	88·5±9·1	82·9±17·4	0·5	76·5± 6·3	82·0±7·3	0·2
3	89·9±7·3	78·1±11·4	0·05	74·9± 4·2	78·3±5·3	0·5
4	90·5±6·8	76·9±16·2	0·05	74·4± 7·1	77·2±6·2	0·4

The results on the male adrenal-demedullated rats are given in Table III.

In the non-fasted animals a significant difference was only obtained 1 hr. after the injection; this effect was not observed in the fasted animals.

Table IV gives the results on the non-fasted male adrenalectomized rats.

The injection of salicylate produced an immediate and rapid fall in the blood glucose. This usually proved fatal, 10 of the salicylate-treated animals dying with hypoglycaemic convulsions within 4 hr.

TABLE IV

EFFECT OF SALICYLATE ON THE BLOOD GLUCOSE OF NON-FASTED MALE ADRENALECTOMIZED RATS

Mean values±S.E. (no. of rats in parentheses) for blood glucose levels expressed as mg./100 ml.

Time after Injection (hr.)	Control	Salicylate-treated	P
0	98·0±11·4 (8)	99·1± 6·9 (12)	0·7
1	88·0±13·9 (8)	71·4± 8·2 (11)	0·01
2	82·0±14·1 (8)	51·0±10·7 (8)	0·001
3	72·0±13·3 (8)	43·5±10·9 (6)	0·01
4	70·7±14·0 (7)	43·0± 3·5 (2)	0·05

DISCUSSION

The present work was designed primarily to determine if there is a sex difference in the response of the blood glucose concentration to salicylate in the rat. It was observed that both male and female rats, whether non-fasted or fasted previously to the experiment, showed hyperglycaemia after the injection of salicylate. The rat does not therefore show a sex difference to salicylate similar to that reported for the mouse (Sproull, 1954). In the non-fasted animals the hyperglycaemia did not persist beyond 3 hr.; the present results do not conflict with the observation (Lutwak-Mann, 1942; Smith *et al.*, 1952) that there was no change in the blood glucose concentrations of non-fasted male rats 4 hr. after the injection of salicylate.

The hyperglycaemia produced by the salicylate may have been due to one or more causes, such as increased liver glycogenolysis, an increased intestinal absorption of glucose, a decreased tissue utilization of glucose and increased gluconeogenesis. Since it was known that salicylate produced a marked depletion of liver glycogen in the rat (Lutwak-Mann, 1942) the possibility was considered that the hyperglycaemia may have been secondary to increased hepatic glycogenolysis. Comparison of the results from the non-fasted and fasted rats (Tables I and II) showed that the hyperglycaemia was more pronounced in the former animals, which may be a reflection of their higher content of liver glycogen. Although the liver glycogen content of fasted rats is relatively low these animals showed a definite hyperglycaemic response to salicylate which appeared to persist for a longer period than in the non-fasted animals. Fasted animals show impaired glucose tolerance curves and are not able to utilize administered carbohydrate as rapidly as the fed animal (Chambers, 1938; Lundbaek, 1948), so that the glucose produced by increased hepatic glycogenolysis in the fasted salicylate-treated rat may not have been removed as rapidly as in the fed rat.

A further difference is the presence of food in the gut of the fed animals; but it has been shown (Smith and Irving, 1955) that salicylate causes a marked delay in the rate of gastric emptying in the fed rat, which is therefore similar to the fasted salicylate-treated animal, in that although food is present in the stomach it is not available for digestion and absorption for at least several hours.

If the hyperglycaemia produced by salicylate was secondary to increased hepatic glycogenolysis it could have been mediated by an increased

release of adrenaline from the adrenal medulla. The fasted adrenal-demedullated rat did not exhibit a hyperglycaemia when treated with salicylate, and the hyperglycaemic response was greatly reduced in the non-fasted salicylate-treated adrenal-demedullated rat. These results suggest that the secretion of adrenaline from the adrenal medulla played an important part in the production of the hyperglycaemic response to salicylate in the rat. However, the presence of a significant degree of hyperglycaemia 1 hr. after the injection of salicylate in the non-fasted adrenal-demedullated animal means that salicylate can also produce an increased blood glucose by some other mechanism. One possible explanation is that salicylate acts directly on the liver, causing increased breakdown of glycogen: it has been found that incubation of liver slices with salicylate *in vitro* causes an increased rate of disappearance of glycogen (Smith, 1955).

Most non-fasted adrenalectomized rats given salicylate died with hypoglycaemic convulsions within 4 hr. of the injection. This hypoglycaemic effect resembles the marked reduction in blood glucose produced by salicylate in the alloxan-diabetic rat (Smith *et al.*, 1952). It has been suggested that salicylate causes an increased rate of tissue glycolysis by interfering with oxidative phosphorylation processes (Smith, 1954).

It is suggested, as a provisional hypothesis, that the injection of salicylate to a normal rat causes an increased secretion of adrenaline from the adrenal medulla. The adrenaline produces increased hepatic glycogenolysis, and the hyperglycaemia is a secondary phenomenon. This is not considered to be a specific effect of salicylate but merely a non-specific stimulation of the adrenal medulla due to a relatively large dose of the drug. There may also be a direct action of salicylate on liver glycogen because of the 1-hr. hyperglycaemia observed in the non-fasted adrenal-demedullated rat. The hypoglycaemic effect of salicylate in the adrenalectomized rat emphasizes the importance of the adrenal cortex in maintaining the blood glucose concentration, and may be due to a more fundamental and specific action of salicylate on carbohydrate metabolism.

Summary

1. The blood glucose concentrations of rats were determined 1, 2, 3, and 4 hr. after the injection of sodium salicylate, 0.5 mg./g. body weight.

2. Moderate hyperglycaemia was observed in normal rats, irrespective of sex, non-fasted and fasted alike, at 1, 2, and 3 hr. following the administration of salicylate; at 4 hr. the blood glucose level returned to normal in females, but remained slightly elevated in fasted males.

3. Hyperglycaemia was less pronounced in adrenal-demedullated rats treated with salicylate; in adrenalectomized rats salicylate caused a marked decrease in blood glucose concentration.

4. Some possible mechanisms are discussed.

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