THE ROLE OF SEROTONIN IN CARBOHYDRATE
METABOLISM III.
THE EFFECT OF SEROTONIN ON BLOOD LACTATE LEVEL
OF RATS

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In the preceding papers (Kobayashi et al., 1960 a, b) serotonin (5-hydroxy-
tryptamine) intravenously injected into rats was found to cause a transient hypo-
glycemia as well as a rapid and marked decrease in liver and muscle glycogen.
If such a fall of glycogen would be a reflect of enhanced glycogenolysis, a eleva-
tion in blood lactate, final metabolite of glycogen in muscle, should be resulted
in the animal treated with serotonin. Phenethyldiguanide (DBI), a synthetic
hypoglycemic agent, which brings forth tissue anoxia and glycogenolysis in muscle
and liver causes an increase in blood lactate (Tyberghein and Williams, 1957).
The observed similarity of serotonin to DBI in hypoglycemic and glycogenolytic
activity also urged the authors to follow the variation of blood lactate level after
serotonin administration.

This paper deals with the effect of serotonin administration on blood lactate
level under various conditions.

METHODS

Male and female rats of Wistar strain weighing 150~250 g were used as in the previous re-
port (Kobayashi et al., 1960a). Estimation of blood lactate was carried out on 0.04 ml blood
withdrawn from the tip of rats' tail according to Barker and Summerson (1941) after depro-
teinization with ZnSO₄ and Ba(OH)₂. When rate of blood glycolysis was followed, decrease of
blood glucose and increase in blood lactate was measured 30, 60, 90 and 120 mins. after
incubation of heparinized blood, glucose was determined by Somogyi-Nelson method
(Nelson, 1944; Somogyi, 1945). Serotonin creatinine sulfate (Nutritional Biochemical Co-op.)
was used throughout the experiments, the dosage being expressed as creatinine sulfate. The
rats employed in this study had been bilaterally adrenomedullated, unless otherwise stated, to
exclude the effect of endogenous epinephrine.

RESULTS

Intravenous injection of serotonin 500 µg per rat caused a rapid rise of blood
lactate in adrenomedullated rats (Fig. 1). A transient but consistent rise, the
maximum value being twice as high as the initial one, was observed at the dosage
as low as 50 µg per rat. At a dose of 10 µg intravenous, serotonin failed to produce

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Fig. 1. Blood lactate level of adrenodemedullated rat following intravenous injection of serotonin

The vertical lines indicate standard error of the mean, figures in parentheses are number of animals. Throughout the present paper this expression holds.

Fig. 2. Blood lactate level of adrenodemedullated rat following subcutaneous injection of serotonin
any significant change in lactate level. Subcutaneous injection of 1 mg serotonin into adrenomedullated animal also brought about a striking increase in blood lactate (Fig. 2), whereas intraperitoneal injection elicited a far less rise in lactate than subcutaneous route (Fig. 3). The comparison made on the difference of

Fig. 3. Blood lactate level of adrenomedullated rat following intraperitoneal injection of serotonin

Fig. 4. Blood lactate level of normal and adrenomedullated rat following intravenous injection of serotonin

--- normal rat

----- adrenomedullated rat
Fig. 5. Blood lactate level of normal and adrenomedullated rat following subcutaneous injection of serotonin

- normal rat, anesthetized
- normal rat, unanesthetized
- adrenomedullated rat, unanesthetized

Fig. 6. Blood lactate level of adrenomedullated rat during infusion of serotonin

- treated with serotonin
- saline-treated control
broken line indicates the period of infusion
blood lactate increase between intact and adrenomedullated rats are presented in Figures 4 and 5. Administration of serotonin into the rats with intact adrenal medulla, both intravenously and subcutaneously, brought about an increase of blood lactate far in excess of that observed on adrenomedullated ones. It may reasonably be deduced that administration of serotonin enhances the release of epinephrine from adrenal medulla or stimulates the action of endogenous epinephrine. Figure 5 also shows that the appearance of the rise in blood lactate under pentobarbital anesthesia was delayed compared with that without anesthesia. Variation of blood lactate level following gradual infusion of serotonin into femoral vein was plotted in Figure 6. Also in this case serotonin was effective on blood lactate.

For comparison of activity between serotonin and epinephrine, the effect of epinephrine alone on blood lactate level of adrenomedullated rats was examined (Fig. 7). Two µg of epinephrine produced approximately comparable effect on blood lactate with 50 µg of serotonin when intravenously injected, and as to the subcutaneous administration, the extent of hyperlactacidemia obtained by 20 µg epinephrine was nearly half as large as that elicited with 1 mg of serotonin.

To exclude the possibility that blood lactate increase would be due to the enhanced glycolysis in blood, several experiments were carried out, the results being presented in Figures 8, 9 and 10. The rate of glycolysis in blood withdrawn from serotonin-treated rats was not significantly different from that of saline-treated control. Moreover, serotonin added in vitro to blood specimen also had no effect on blood glycolysis (Fig. 10).

DISCUSSION

Serotonin creatinine sulfate, as low as 50 µg per rat, caused a striking increase
Fig. 8. *In vivo* effect of serotonin on blood glycolysis; rate of glucose degradation. Serotonin, 100 µg per rat, was intravenously injected into adrenomedullated rat 10 mins. prior to sacrifice. Heparinized blood obtained by decapitation was incubated at 37°C.

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in blood lactate of adrenomedullated rats. As pointed out by Bogdanski et al. (1956), the concentration of serotonin in whole blood of rat is 0.1~0.2 µg per ml. The value of 0.34 µg per ml was also supposed by others (Humphrey and Jacques, 1954). The amount of serotonin required for increase of blood lactate, therefore, is about 10 to 20 times total amounts in body fluid. When epinephrine was employed to achieve the similar result in blood lactate, 2 µg per rat had to be injected into rats. Though the concentration of epinephrine in blood of rats under physiological conditions now remains to be decided, the reliable value seems to be of the order of or below 1 µg/l of blood (von Euler, 1957; Gaddum and Holzbauer, 1957). If such is the case, 2 µg of epinephrine may amount to some hundreds times the total amount of endogenous epinephrine present in...
Fig. 9. In vivo effect of serotonin on blood glycolysis; rate of glucose degradation and lactate production

--- treated with serotonin

...... control

blood. As to the lactate increasing activity, therefore, serotonin is far more active than epinephrine if considered on the basis of concentration in body fluid. The fact suggests that the dose of serotonin employed to affect blood lactate level is not so enormous one. In fact, no vascular abnormalities such as bradypnea, vasoconstriction or blanching of the extremities, were observed in rats injected with 50 μg serotonin. Thus, rise of blood lactate could not be ascribed to anoxemia caused by large doses of serotonin.

As pointed out in the previous report (Kobayashi et al., 1960a), pentobarbital anesthesia enhanced and prolonged the vasoconstriction caused by large dose of serotonin. As to serotonin induced hyperlactacidemia, however, pentobarbital anesthesia resulted in an inverse effect; peak of blood lactate increase was reduced and delayed under anesthesia. It may be suggested that anoxia resulted from vasoconstriction might not be directly responsible for blood lactate increment and
that vasoconstriction is rather effective in retarding the absorption of the drug from the cite of injection. As to the administration through subcutaneous route, 1 mg of serotonin resulted in a marked increase in blood lactate, whereas the effect was strikingly reduced when the dose was diminished to 200–250 μg per rat (Fig. 2). From this result, therefore, sudden increase in blood concentration of serotonin would possibly be required for rise of lactate. The result shown in Figure 6, however, excluded this possibility, because constant infusion at the average rate of 1–2 μg per min. resulted in a clear-cut hyperlactacidemia. As already demonstrated in liver perfusion experiment (Kobayashi et al. 1960b), serotonin added rapidly to perfusate caused a diminution of blood flow rate, while slow infusion exerted little influence. Together with the evidences mentioned above, the effectiveness of slow infusion favors the interpretation that effect of serotonin in blood lactate is not secondary to vascular effect.
The results shown in Figures 8, 9 and 10 that serotonin had no effect on blood glycolysis may be in favor of the possibility that the site of action of serotonin should be primarily the carbohydrate metabolism in peripheral and/or hepatic tissue. Although the authors found that serotonin inhibited the uptake of lactate by perfused hepatic tissue (Ui et al., 1960), the larger part of increased lactate may be ascribed to alteration of metabolism in extrahepatic tissues because subcutaneous injection caused a more marked hyperlactacidemia than intraperitoneal route (Fig. 3). The low sensitivity of rats to intraperitoneal injection of serotonin suggested that the degradation of serotonin occurred rapidly in hepatic tissue. This is in agreement with general findings that the activity of monoamine oxidase is very high in this organ.

Reid (1952) reported that serotonin (1 µg by close arterial injection) released epinephrine in cats. In the previous paper, the authors confirmed this finding by the glycemic study on rats (Kobayashi et al., 1960a). As represented in Figures 4 and 5, administration of serotonin, either intravenous or subcutaneous, resulted in a hyperlactacidemia in the rat with intact adrenal medulla to a larger extent than in adrenomedullated one. Although it may be deduced that serotonin enhances the release of epinephrine from adrenal medulla in good agreement with Reid’s finding as well as the results obtained by the present authors with glycemic study, the responsiveness of adrenomedullated animal to serotonin in this respect suggests that serotonin-induced hyperlactacidemia is not solely due to the action of secreted epinephrine. A possibility still remains to be ruled out that rise of blood lactate following injection of serotonin is dependent on enhanced release of epinephrine from tissues other than adrenal medulla or exaggeration of action of endogenous epinephrine. To obtain any information in this respect, the effect of ergotamine on blood lactate-increasing activity of serotonin and epinephrine was investigated. No decisive results were obtained because the ergotamine tartrate employed itself caused a severe hyperlactacidemia.

It has been suggested that lesions of right side of the heart in malignant carcinoid might be resulted from rich content of serotonin during its passage from liver to lung (Goble et al., 1956). On the other hand, the possibility was postulated more recently that accumulation of lactic acid might be the primary cause of cardiac damage in anoxia (DeHaan and Field, 1959). Furthermore, in the preceding paper significant increment of cardiac glycogen after large intravenous doses of serotonin was demonstrated (Kobayashi et al., 1960b). These observations along with the result of the present experiment strongly suggest that the lesion of right heart in carcinoid may, at least partly, be the result of hyperlactacidemia caused by excess serotonin. This postulation, of course, awaits further experimental confirmation.

SUMMARY

1. Effect of serotonin on blood lactate level of rats were examined.
2. Serotonin, administered intravenously, subcutaneously or intraperitoneally, caused a rise of blood lactate.
3. Effectiveness of serotonin in causing hyperlactacidemia was far more than
that of epinephrine if considered on the basis of physiological concentration in circulation.

4. Serotonin had no effect on blood glycolysis both in vivo and in vitro.

5. The possibility that serotonin may act primarily on carbohydrate metabolism in peripheral and/or hepatic tissues are discussed.

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REFERENCES