THE ABILITY OF SOME ANABOLIC STEROIDS TO PROTECT RATS AGAINST ETHIONINE-INDUCED ELEVATION OF POSTPRANDIAL BLOOD AMMONIA

By

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ABSTRACT

Compared with control rats, postprandial blood ammonia was significantly higher in ethionine treated rats. This was considered due to abnormal nitrogen metabolism by the injured liver. Pretreatment of rats with anabolic steroids, 17-ethyl-19-nortestosterone and methandrostenolone protected animals against the ethionine effect on blood ammonia. It appears that the primary defect induced by ethionine is one in protein metabolism. The action of steroids, used in this study, on ammonia metabolism is considered due to their anabolic properties.

Ethionine, a metabolic antagonist of methionine induces a periportal fatty liver in animals. It also interferes with synthesis and metabolism of protein, carbohydrate and lipids in the liver (Farber 1959). We found recently that in rats with ethionine induced hepatic injury, postprandial blood ammonia was significantly increased (Kowalewski 1965a, b). This was considered due to abnormal handling of amino nitrogen by the damaged liver (Kowalewski & MacKenzie 1965). Methionine had a protective action against the effect of ethionine on ammonia metabolism (Kowalewski 1965a, b).

It was shown previously that the lipid infiltration caused by ethionine can be effectively blocked by certain testosterone derivatives (Ranney & Drill 1957). Will anabolic steroids also protect the rats against ethionine induced alteration in ammonia metabolism? To answer this question, the following experiment was performed.
METHODOLOGICAL DETAILS

Sprague Dawley female rats (body weight 220-230 g) used for this study, were fed a standard Purina laboratory diet containing 3.4 g of total nitrogen in 100 g of dry weight (Kowalewski 1965a). Steroids were given per os, mixed with the food for three weeks (Kowalewski 1958). Two steroids were used in this experiment: 17-ethyl-19-nortestosterone (ENT), Searle and Co., (Raney & Drill 1957; Drill & Riegel 1958), and Methandrostenolone (MA), Ciba Co., (Kowalewski 1962), both in a daily dose of 2 mg/100 g body weight. Acute liver injury was produced by DL-ethionine intraperitoneally in 3 equal doses, totalling 1.0 mg/g of body weight (Raney & Drill 1957). Rats were divided into seven groups and treated as follows: 1) Controls, no test meal; 2) Controls given test meal; 3) Treated with DL-ethionine; 4) Treated for three weeks with ENT; 5) Treated for three weeks with ENT prior to treatment with DL-ethionine; 6) Treated for three weeks with MA; 7) Treated for three weeks with MA prior to treatment with DL-ethionine. Group I was studied for blood ammonia after a 24 h fasting period. All remaining groups were studied for blood ammonia 4 h after a protein rich meal, containing 4.3 g of total nitrogen in 100 g of wet weight. This test meal consisted of commercial corned beef, given to rats in the amount of 6 g per 100 g of body weight (Kowalewski 1965a). The test meal was given after a 24 h fasting period and in groups 3, 5 and 7, 20 h after the first dose of ethionine. Blood was taken from the vena cava under light ether anaesthesia and the rats were then killed. Total blood ammonia was studied by microdiffusion technique (Conway 1962) and expressed in µg/100 ml of blood.

RESULTS

Table 1 summarizes the results. It is apparent that feeding meat resulted in a marked increase of blood ammonia in normal rats. Ethionine treated rats

Table 1.
The effect of 17-ethyl-19-nortestosterone (ENT) and Methandrostenolone (MA) on postprandial blood ammonia in DL-ethionine treated rats. Sampling after 24 h fasting (F) or 4 h after ingestion of test meal (M). Mean and S.E.

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Sampling Condition</th>
<th>No. of rats</th>
<th>Blood Ammonia µg/100 ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>None</td>
<td>F</td>
<td>18</td>
<td>81.2 ± 0.9</td>
</tr>
<tr>
<td>2</td>
<td>None</td>
<td>M</td>
<td>16</td>
<td>114.0 ± 3.5</td>
</tr>
<tr>
<td>3</td>
<td>DL-ethionine</td>
<td>M</td>
<td>19</td>
<td>*268.0 ± 15.1</td>
</tr>
<tr>
<td>4</td>
<td>ENT</td>
<td>M</td>
<td>16</td>
<td>95.0 ± 4.1</td>
</tr>
<tr>
<td>5</td>
<td>ENT + DL-ethionine</td>
<td>M</td>
<td>20</td>
<td>91.0 ± 5.1</td>
</tr>
<tr>
<td>6</td>
<td>MA</td>
<td>M</td>
<td>20</td>
<td>98.3 ± 4.0</td>
</tr>
<tr>
<td>7</td>
<td>MA + DL-ethionine</td>
<td>M</td>
<td>18</td>
<td>104.0 ± 4.7</td>
</tr>
</tbody>
</table>

* P < 0.001 versus other groups.

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had significantly elevated blood ammonia, as compared with other groups. In animals treated with steroids, postprandial blood ammonia was in the normal range. Ethionine treated rats pretreated with steroids did not show any increase in blood ammonia.

**COMMENT**

In our previous studies (Kowalewski 1965a,b) postprandial blood ammonia was increased in rats treated for 6 weeks with a diet containing ethionine. In the present experiment, acute ethionine intoxication produced the comparable effect. In both cases the animals appeared unable to handle the nitrogen rich test meal and this was considered due to liver injury. Both steroids used in this study were able to protect the rats against the post-ethionine elevation of blood ammonia. The protective action of these steroids is probably associated with their protein anabolic activity.

Ethionine is known to have a general inhibitory effect on protein synthesis in the liver. Since the changes in protein metabolism precede the fatty infiltration of the liver by a few hours, the disturbance of protein metabolism may be the primary effect of ethionine (Farber 1959). Acute inhibition of protein synthesis and fatty livers occur only in female rats and not in males: this difference appears to be sex dependent (Farber 1959; Artom 1959). The inability of ethionine to induce fatty livers in male rats appears to be due to a protection by androgens. In the absence of androgens, ethionine can interfere with the synthesis of certain specific liver proteins which are required for the oxidation of fatty acids in the liver, and perhaps for their mobilization in the plasma (Artom 1959). It was found previously that the protective action of testosterone derivatives against post-ethionine fatty liver does not depend on androgenic or progestational properties of these steroids, but on their anabolic action (Ranney & Drill 1957).

In the present study two observations have been made. Firstly, that female rats with acute liver injury produced by ethionine, have elevated postprandial blood ammonia. Secondly, that anabolic steroids, used in this experiment, were able to protect these rats against postprandial hyperammonaemia. The effect of anabolic steroids on ammonia metabolism is probably due to their ability of affecting nitrogen metabolism.

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