THE EFFECT OF NEMBUTAL ON THE EOSINOPENIC RESPONSE TO SEVERAL COMPOUNDS

By

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It is well known, that many drugs are capable of promoting the secretion of corticotrophin by the pituitary gland. The mechanism by which this release is brought about, however, is still obscure.

Recant et al. (1950) have shown that nembutal-anesthesia prevented the eosinopenic response to certain stimuli such as the handling of animals and tail cutting, but not the eosinopenia induced by adrenaline. This suggests a difference between the mode of action of adrenaline and other kinds of stressors in the activation of the pituitary-adrenal axis. As sodiumsalicylate (van Caermenberge & Betz, 1952; Hetzel & Hine, 1951; Cronheim et al., 1952; Eades & Stanton King, 1953) and pyramidone (Frommel et al., 1945; Blanchard, 1950) have a potent corticotrophin releasing activity, it seemed of interest to investigate whether these substances act in the presence of nembutal. Both compounds also have an antipyretic and analgesic action. As sulfonamides also exert some antipyretic effect but in quite a different way it also seemed of interest to include a representative of this class of compound, namely sulfamerazine, in this investigation.

This report deals with the effect of these compounds as compared with the effect of l-adrenaline on the number of circulating eosinophils in non-anesthetized rats as well as in animals under nembutal anesthesia.

MATERIAL AND METHODS

Adult male rats weighing 150–300 gm. from an inbred strain were used. About half the amount of a lethal dose of sodiumsalicylate (Hetzel & Hine, 1951) and pyramidone (in our own unpublished experiments 30 mg./100 gm. proved to be lethal) dissolved in 1 ml. of saline were injected. The dose of sulfamerazine was 100 mg./100 gm. In mice the lethal dose of this drug was 200 mg./100 gm. (Schmidt et al., 1944). A dose of
1-adrenaline was chosen that caused a marked eosinopenia in this strain of rats (Keuskamp, 1956; Louwerens & Smelik, 1953). All substances were administered intraperitoneally.

In order to determine whether the eosinopenic response to the compounds used, might be caused by the mere administration of a concentrated solution, the effect of a pharmacologically inactive substance l-arabinose (Goldstein et al., 1953) was investigated. Hence, 33 mg. l-arabinose per 100 gm. bodyweight was injected intraperitoneally.

For the anesthesia 3.25-4 mg. nembutal was injected prior to the administration of the compound to be investigated. The variation in the amount of nembutal to be given depended on the weight of the rats and on the number of experiments they had undergone previously.

Blood samples were obtained by venipuncture of one of the tail veins at zero hour and 3 hours later. The eosinophils were counted in Fuchs-Rosenthal counting chambers after diluting 0.1 ml. of blood with 0.9 ml. of the diluent of Randolph as modified by Henneman (1949).

The number of eosinophils in the second sample of blood is expressed in percent of the initial value. A more detailed description of the procedure used in this investigation is given in a paper by Keuskamp (1956).

The statistical analysis was carried out with the aid of Wilcoxon’s two sample test (Wilcoxon, 1945; Mann & Whitney, 1947). A difference was considered as statistically significant, if the double tail probability, k, was < 0.05.

*Table 1.*

Comparison of the effect of various substances on the number of eosinophils in nembutalized and non-anesthetized rats.

<table>
<thead>
<tr>
<th>No. rats</th>
<th>Nembutal Eosinophils % of initial value</th>
<th>Compounds Dose/100 gm. body-weight</th>
<th>No. rats</th>
<th>No Nembutal Eosinophils % of initial value</th>
<th>k Wilcoxon</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>121 ± 20.0*)</td>
<td>33 mg. l-arabinose</td>
<td>15</td>
<td>89 ± 13.4</td>
<td>0.77</td>
</tr>
<tr>
<td>15</td>
<td>115 ± 17.1</td>
<td></td>
<td>16</td>
<td>102 ± 14.1</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>117 ± 18.9</td>
<td>33 mg. Na-salicylate</td>
<td>20</td>
<td>61 ± 8.5</td>
<td>0.004</td>
</tr>
<tr>
<td>15</td>
<td>115 ± 17.4</td>
<td></td>
<td>21</td>
<td>95 ± 11.3</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>113 ± 11.7</td>
<td>15 mg. pyramidone</td>
<td>12</td>
<td>48 ± 7.1</td>
<td>0.002</td>
</tr>
<tr>
<td>16</td>
<td>159 ± 16.7</td>
<td></td>
<td>11</td>
<td>150 ± 30.9</td>
<td></td>
</tr>
<tr>
<td>16</td>
<td>104 ± 17.8</td>
<td>100 mg. sulfamerazine</td>
<td>17</td>
<td>54 ± 5.4</td>
<td>0.03</td>
</tr>
<tr>
<td>16</td>
<td>128 ± 29.7</td>
<td></td>
<td>16</td>
<td>87 ± 9.7</td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>36 ± 4.7</td>
<td>30 γ l-adrenaline</td>
<td>11</td>
<td>30 ± 5.6</td>
<td>0.0003</td>
</tr>
<tr>
<td>17</td>
<td>124 ± 18.0</td>
<td></td>
<td>12</td>
<td>126 ± 23.9</td>
<td></td>
</tr>
</tbody>
</table>

*) standard error of the mean.

1) double tail probability.
RESULTS

As may be seen from the table, in the non-anesthetized rat sodiumsalicylate, pyramidone, sulfamerazine and 1-adrenaline caused a statistically significant decrease in the number of circulating eosinophils. 1-Arabinose did not have this effect.

In rats under nembutal anesthesia, only 1-adrenaline appeared to cause a statistically significant decrease \( k = 0.00005 \), whereas the eosinopenic response to the other substances was completely inhibited.

DISCUSSION

The experiments show that the eosinopenic response to sodiumsalicylate, pyramidone and sulfamerazine, observed in the non-anesthetized rat, was prevented by nembutal anesthesia whereas 1-adrenaline remained effective in the presence of nembutal. As 1-arabinose failed to elicit an eosinopenia whether nembutal was given or not, the mere administration by the intraperitoneal route of a concentrated solution cannot explain the effect of the compounds used.

Our results are thus in accordance with the experiments of van Cauwenberge & Betz (1952), who observed a decreased response to sodium salicylate in rats anesthetized with dial and are in agreement with the findings of Way & van Peenen (1956), who found that the effect of sodium salicylate and morphine on the adrenal ascorbic acid content was inhibited in rats under nembutal anesthesia, whereas the effect of adrenaline could not be prevented.

Several substances like adrenaline are known to possess a pituitary stimulating activity in the presence of nembutal. Among these are histamine (Briggs & Munson, 1955; Olling & de Wied, 1956; Way & van Peenen, 1956), noradrenaline (Olling & de Wied, 1956), serotonine (Way & van Peenen, 1956), carbachol (de Wied, unpublished data), pitressin (Munson & Briggs, 1955) and a number of adrenaline related phenylalkylamines (Keuskamp, 1956). Two groups of drugs can thus be distinguished, one group consisting of compounds, which are active, and another group, which is inactive in the presence of nembutal. This may mean that the two groups exert their effects at different sites of the central nervous system involved in the release of corticotrophin from the pituitary gland.

SUMMARY

The effect of large doses of sodium salicylate, pyramidone, sulfamerazine, 1-arabinose and 1-adrenaline on the number of circulating eosinophils in normal and nembutalized rats was investigated. All the compounds, except 1-arabinose,
produced an eosinopenia in the non-anesthetized rat. The eosinopenic response to sodium salicylate, pyridoxine and sulfamerazine was blocked in rats anesthetized with nembutal, whereas the effect of 1-adrenaline was not prevented by this anesthesia.

REFERENCES

Wilcoxon, F.: Biometrics 1, 80, 1945.

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