DISTRIBUTION OF NEURUTOXICESTERASE IN CERTAIN BRAIN REGIONS OF LABORATOY ANIMALS

M.A. MATIN*, SEEMA SATTAR, S.N. KHAN AND K. HUSAIN

Industrial Toxicology Research Centre, Post Box 80, Lucknow - 226001.

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Summary: Certain organophosphorous compounds caused the inhibition of 'neurotoxicesterase' present in central nervous system. The role of this enzyme is different from that of cholinesterase. The level of neurotoxicesterase in brain, corpus striatum and spinal cord of rats, mice, guineapigs, and hens was measured. Maximum level of the enzyme was found in hens, followed by guineapigs, rats and mice in the order; The concentration of the enzyme was higher in corpus striatum>whole brain>spinal cord. The determination of the normal level of neurotoxicesterase may be useful in monitoring the exposure to organophosphorous compounds.

Key words: Neurotoxicesterase, cholinesterase, brain, corpus striatum, spinal cord

INTRODUCTION

The inhibition of cholinesterase by organophosphorous compounds is accompanied by accumulation of acetylcholine in different brain regions (1, 2) resulting in parasympathomimetic effects. Certain 'late or delayed effects' also appear 3-4 weeks after exposure to certain organophosphorous compounds (3, 4). These 'late' or 'delayed effects' include weakness of hind limbs, ataxia and paralysis which are not related to inhibition of cholinesterase (5). These effects have been attributed to inhibition or phosphorylation of another esterase, "Neurotoxicesterase" which is also present in central nervous system (3). So far less attention has been given to the enzyme neurotoxicesterase by workers; Therefore the determination of the level of neurotoxicesterase activity in certain brain regions and spinal cord of different laboratory animals was undertaken to note the comparative evaluation with cholinesterase activity.

MATERIALS AND METHODS

Rats, mice, hens and guineapigs were used. The animals were maintained on a 12 hr light dark schedule at a temperature of 28±1°C and had food and water at libitum. Food was however, withdrawn 12 hr before experiments. The animals were decapitated and the brain removed quickly; The corpus striatum was dissected according to the method of Glowinski and Iverson (6). The level of neurotoxicesterase was assayed according to the procedure described by Johnson (7) using phenylvalerate as the substrate as reported elsewhere (8). The brain tissue was homogenized (1:10 w/v) in Tris/EDTA buffer, pH 8.0 and preincubated at 37°C for 20 min with paraoxon (40 uM) with either buffer or mipafox (50 uM) in a final volume of 2.0 ml. After preincubation, phenyl valerate (2.0 ml) was added and the incubation continued for 15 min. Reaction was stopped by adding 2 ml of sodium dodecyl sulphate (1% w/v) in buffer containing 4 aminoantipyrine. 1 ml of K₃Fe(CN)₆ (0.4% in water) was added and the stable red colour read at 510 Nm.
The acetylcholinesterase activity was measured according to the method of Ellman et al. (9) using acetylcholine as the substrate (10).

RESULTS

The values of neurotoxicesterase and cholinesterase activities in certain brain regions of animals are given in Table I. The values of both the enzymes were significantly higher in corpus striatum than the rest of the brain. The values were however more in brain than the spinal cord. The concentration of both the enzymes was maximum in hens followed by guineapigs, rats and mice in that order.

DISCUSSION

Results indicated that the acute or parasympathomimetic effects are caused by the inhibition of cholinesterase while the 'late' or 'delayed effects' induced by certain organophosphorous compounds are caused by inhibition of 'neurotoxicesterase' which is widely distributed in the central nervous system of different animals (Table I). It was previously reported that the concentration of cholinesterase was high in corpus striatum, an area of the brain which is important in the regulation of motor function (11). The results presented indicate that the level of neurotoxicesterase was higher in corpus striatum than the whole brain. Further the level of neurotoxicesterase was highest in the brain and spinal cord of hens (Table I) which have been found more susceptible to 'late' or 'delayed' effects of certain organophosphorous compounds (12). Rats

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TABLE I : Distribution of neurotoxicesterase and cholinesterase in the brain, corpus striatum and spinal cords of rats, mice, guineapigs and hens.

<table>
<thead>
<tr>
<th>Animals</th>
<th>Neurotoxicesterase* (mean±SE)</th>
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<tbody>
<tr>
<td></td>
<td>Brain</td>
</tr>
<tr>
<td>1. Mice</td>
<td></td>
</tr>
<tr>
<td>(20±5)</td>
<td>602.46</td>
</tr>
<tr>
<td></td>
<td>±3.95</td>
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<tr>
<td>2. Rats</td>
<td></td>
</tr>
<tr>
<td>(150±10)</td>
<td>840.12</td>
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<tr>
<td></td>
<td>±4.16</td>
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<tr>
<td>3. Guineapigs</td>
<td></td>
</tr>
<tr>
<td>(750±20)</td>
<td>1026.34</td>
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<tr>
<td></td>
<td>±5.95</td>
</tr>
<tr>
<td>4. Hens</td>
<td></td>
</tr>
<tr>
<td>(900±25)</td>
<td>2346.23</td>
</tr>
<tr>
<td></td>
<td>±10.16</td>
</tr>
</tbody>
</table>

Figures in parenthesis indicate the weight of animals in grams. Each figure represents the mean±SE of ten experiments in each group.

*nmol of phenylvalerate hydrolysed/min/g, wet tissue

**mol of acetylthiocholine hydrolysed/min/g x 10^-8, wet tissue.

a, P<.01; b, P<.05; c, <.05, (compared with the corresponding values of other groups).
and mice which are least susceptible to these effects have only a small concentration of neurotoxic esterase in the brain and corpus striatum (13).

Considerable species variation exhibited in respect to 'late' or 'delayed effects' of organophosphorous compounds (14): Since different animal species show varying degree of susceptibility to organophosphate induced "delayed effects", the estimation of the normal level of neurotoxic esterase in animals may help to evaluate the degree of toxicity or exposure to neurotoxic organophosphorous compounds.

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REFERENCES