Effect of *Ocimum sanctum* on the brain tissue of American cockroach

*Periplaneta americana*

Ranjit Hazari* and Boruah N*

*Animal Physiology Lab, M.C. College, Barbeta-781 301, Assam, India*

*Natural Chemistry Division, Regional Research Lab, RRL Jorhat-785 006, Assam, India*

*Corresponding author: rranjit96@rediffmail.com*

**Abstract**

Methanolic extract of *Ocimum sanctum* with different concentrations ranging from 50 to 200 ppm were exposed to American cockroach *Periplaneta americana* for 96 hrs and observed certain biochemical parameters an increase of brain lipid content were observed ,where as the level of cholesterol content were increased significantly. On the other hand, a significant decrease in the activity of protein and cholesterol was observed in all the test groups. Bio-molecule of plant extract therefore within a short time entering the brain actively become the site of action ,because some major biochemical changes has been observed due to the leaf extract of *O. sanctum*.

**Keywords:** *O. sanctum*, leaf extract, neurotoxic effect, biochemical changes

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**Introduction**

The *O. sanctum* L (labiatae) locally known as, Tulshi is a plant used in the traditional system of medicine. The leaves of the plants are used for the hyperglycemic agent and inflammatory potential and treatment of gastric ulcers (Mandal et al., 1993). In the present instigation, the effect of metabolic leave extract of the *Ocimum sanctum* on the biochemical changes on the brain tissue of cockroach *Periplaneta americana* were studied.

**Materials and Methods**

**Plant material**

*O. sanctum* leaves collected from Nowgong District, Assam on October 2005 and authenticated by the Department of Botany, M.C. College, Assam, India.

**Preparation of Extract**

The sun dried powder leaves of *Ocimum sanctum* percolated in room temperature with 70% methanol. The extract was concentrated 20.3±.8% w/w relative to dried starting material. This residue of *O. sanctum* was dissolved in 2.5% methanol for toxicological study.

**Insects**

Twenty newly emerged adult cockroach average weight male 1.270 gm and female 1.274 gm from inbreed colony were taken for study. Different concentrations ranging from 50 to 200 ppm were prepared by adding desired volume of stock solution in distilled water by vigorous staring.

**Acute toxicity**

Five insects consisting of one group and altogether five groups were kept in insect rearing cage. 6 µl of each concentration ranging from 50 to 250 ppm were injected per insect form ventral side between 3rd and 4th abdominal segment observed for 24 hrs a sets of control was kept side by side in the experiment. After 24 hrs brains were dissected out in physiological saline and biochemical studies were carried out. The Total lipid of brain tissues...
was estimated according to Folch et al. (1951). The cholesterol activity was estimated according to Zlatilis et al. (1953) using ferric chloride as coloring reagent. Cholinesterase activity was determined by enzymatic reaction of acetylcholine buffer salt mixture (Sexena et al., 1978). Total protein in brain was estimated. Folin phenol reagent was used as a coloring agent that results in light green colored complex. The intensity that which gives amount of protein presents BSA was used as a slandered for calculation. The result was express µg/g of the wet tissue.

**Results**

**General and behavioral study**

After exposing the insects indifferent concentrations of leaf extract exhibited abnormal behavioral response. The higher excitability of insects of higher extract concentrations has a great on set threshold as compared with the low dose. In higher concentrations, insects showed rapid jerky movement, convolution, hyper excitability and tendency of escaping from the insect case It was observed that with the increasing concentration the activity was increased expressing the sign of distress. In the later hours insects laid down at the bottom of the insect case.

**Food intake and Body weight**

Food intake was also reduced in the entire test group as compared to control which suggests the effect of leaf extract of tulsi on normal activity including digestive process. Body weight in excremental set so insects at 50, 100, 150 and 200 ppm concentrations were loss in continuation of exposure may be due to the effect of leaf extract and their metabolism the digestive process and in various tissue of the insect.

**Mortality study**

Mortality study on the insect *Periplanta americana* due to the effect of leaf extract of *O. sanctum* showed that average mortality rate was 60% LC50 value according to Read -Munch method , was observed to be 350 ppm at 24 hrs of exposure.

**Biochemical changes in the Brain tissues due to effect of leaf extract of tulsi**

In both sexes, following the fluctuation in content of total protein, cholesterol and cholinesterase enzyme were recorded as compared to control.

a) Concentration with 50 and 100 ppm of leaf extract

1. Lipid: A non-significant increase in lipid at 50 and 100 ppm concentration were recorded in male, where as a significant increase (P<.05) at 50 ppm and at 100 ppm was recorded are recorded in female.

2. Cholesterol: A significant increase (P<.01) in cholesterol content at 50 and 100 ppm is recorded in both the sexes.

b) On the treatment with 150 and 200 ppm of leaf extract

1. Lipid: A significant increase in lipid (P<.001) consent at 150 and 200 ppm in both sexes were recorded.

2. Cholesterol: A significant increase (p<.05) at 150 ppm in both the sexes were observed.

3. Protein: A significant decrease in protein (p<.02) at 150 ppm and significant decrease observed at 200 ppm at both sexes.

**Discussion**

The finding of the investigation has clearly revealed the nature of toxic effect induced by short term of exposure to sub lethal dose concentration of leaf extract of tulsi on the American cockroach, *Pariplanata amaricana* (L).
Table 1. Lipid content in brain tissue after exposure of methanolic leaf extract of Tulshi (N=20)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Sex</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>2.5±.16</td>
<td>2.4±.14</td>
<td></td>
</tr>
<tr>
<td>50 ml</td>
<td>3±.1.6*</td>
<td>2.9±.34*</td>
<td></td>
</tr>
<tr>
<td>100 ml</td>
<td>4.1±187*</td>
<td>3.5±.216*</td>
<td></td>
</tr>
<tr>
<td>150 ml</td>
<td>5±1.58**</td>
<td>4.9±.387**</td>
<td></td>
</tr>
<tr>
<td>200 ml</td>
<td>6±1.158**</td>
<td>5.9±.387**</td>
<td></td>
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</tbody>
</table>

* p<.001 * p<.001

Table 2. Cholesterol content of brain tissue after exposure of methanolic leaf extract of Tulshi (N=20, M- Male, F- Female)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Sex</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>2.5±0.158</td>
<td>2.3±0.152</td>
<td></td>
</tr>
<tr>
<td>50 ml</td>
<td>5.00±1.58*</td>
<td>4.6±2.166*</td>
<td></td>
</tr>
<tr>
<td>100 ml</td>
<td>8.6±0.21*</td>
<td>5.0±1.55*</td>
<td></td>
</tr>
<tr>
<td>150 ml</td>
<td>9.2±1.92</td>
<td>7.8±0.311</td>
<td></td>
</tr>
<tr>
<td>200 ml</td>
<td>10.00±1.85</td>
<td>7.2±0.192</td>
<td></td>
</tr>
</tbody>
</table>

p<.01 Cholinesterase activity: a significant fall (p< 01) in cholinesterase activity was recorded at both the sexes at 150 and 200 ppm of leaf extract.

Toxicological effect of leaf extract of tulsi has become apparent by change in behavioral pattern of experimental insects treated with variable dose concentrations the sign of restlessness convolution rapid jerky movement were increased. Such type of observation were also reported (Sexena et al., 1978). The behavioral alteration may be due to the effect of leaf extract of tulsi on the central nervous system of the cockroach resulting change of behavior with increasing dose concentration. Food intake capacity was reduced as because the biopesticide directly effect on the digestive system of the insect. On the other hand body weight in the treated group of 50, 100, 150 and 200 ppm concentration of leaf extract were observed to be decreased in comparison to control group. This is due to the leaf extract in digestive system food intake capacity also reduced due to effect of leaf extract.

Fig. 1. Protein content (mg/gm) brain tissue of cockroach after exposure of leaf extract of Tulshi.

Fig. 2. Cholinesterase level in the Brain tissue extract of Pariplanata americana after exposure of leaf extract of Tulsi.

The increase of protein content due to treatment with the leaf extract of tulsi may be either on induction phenomenon or due to blocking of synthesis of messenger for the protein repression which in turn would stimulate protein synthesis by depression which in turn would stimulate protein synthesis by depression. Increase in lipid content may due to liberation of some lipoprotein macromolecules structure of unspecified cell in haemolymph which is responsible for an increased in lipid content in the treated insects. The fat bodies lipid which dissolves and retain the toxin as much as possible. So, like leaf extract of tulsi may be lipophilic in nature and

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accumulated in the fat bodies. Then it passed to the site of action in the brain additional quality of lipids is supposed to the brain probably by fat bodies to resist the toxic action of biopesticide on brain as reported by O’brain (1959). Thus lipid content of the brain of insect increased on the treatment of leaf extract. Similar accumulation of lipid may also be induced by interfering mitochondrial fatty acid oxidation (10). A continuous decrease in enzyme activity in both the sexes for 24 hrs of treatment revealed.

The mechanism involved in toxic action of biopesticide on cockroach depends largely on biochemical processes of the insect. The toxic symptoms along with mild and strong are manifestations of inhibition of cholinesterase activity as result of op pesticide poisoning already been reported by different workers (Matcalf, 1949; Machera, 1992). The mode of action of leaf extract of tulsi is also found in paroxon the similar manner has been reported earlier. The leaf extract may have direct relation to the brain tissue of insect due to effect of biopesticide have been observed present investigation. Besides increase in cholesterol suggest on increased association of phospholipids with the tissue because phospholipids is important in metabolism of cholesterol (Machera et al., 1992) also observed similar finding in the insects. The fact that leaf extract of tulsi in 24 hrs act as biopesticide interfering the normal physiological functioning of insect, Periplanata americana.

References