Meibomian Gland Carcinoma...

Fig 2. Dog – Labrador – Meibomian gland carcinoma – polyhedral cells – Mitotic figure H & E scale bar 25 µm.

Meibomian gland tumours have a characteristic exophytic, pedunculated, cauliflower surface appearance and are based at the Meibomian gland opening or over Meibomian gland itself. (Westermeyer and Hendrix, 2012). These tumours are reported to be more frequent in aged dogs and cats when compared to other domestic animal species and also females have a higher number of adenomas compared to males (Yuksel et al., 2005). In the present case the tumour cells showed the tendency of fat cyst formation. This is in accordance with the observations of Goldschmidt and Hendrick (2002). They reported that Meibomian gland carcinomas are composed of tumour cells that have varying amounts of intracytoplasmic lipid vacuoles.

References


Antioxidant Effect of *Gyrocarpus Asiaticus* on Paracetamol Induced Hepatotoxicity in Zebra Fish

Baisakhi Moharana1, S.P. Preetha, C. Balachandran, M. Parthiban, M.R. Srinivasan, Parag Acharya and Swati Choudhury

Department of Veterinary Pharmacology and Toxicology, Madras Veterinary College, Chennai-600007.

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Abstract

This study was conducted to evaluate the potential of aqueous extract of *Gyrocarpus asiaticus* (100mg/ml) on paracetamol (20mM) induced hepatotoxicity in zebra fish. Result of this study showed that, *Gyrocarpus asiaticus* treated group has got marked reduction in oxidative status.

Key words : Paracetamol, hepatotoxicity, *Gyrocarpus asiaticus*, zebra fish.

The zebrafish is a promising animal for assessing drug induced toxicity in variety of organ systems including liver as zebrafish metabolize drugs using similar pathways as human (Vliegenthart et al., 2014). *Gyrocarpus asiaticus* belongs to family Hernandiaceae and found in various parts of India including Eastern
Ghats of Tamilnadu. According to the perusal of literature there are very scanty reports on hepatoprotective effects of this plant. Vithya et al. (2012) reported on the free radical scavenging activity of this plant which signifies its antioxidant effect. Hence keeping these criteria in mind this research is undertaken to explore the effect of *Gyrocarpus asiaticus* on paracetamol induced hepatotoxicity in zebra fish.

**Materials and Methods**

Stem bark portion of *Gyrocarpus asiaticus* was collected from the Azhagar Kovil hills, Madurai District, Tamilnadu, India. Adult zebra fish of both sexes (*Danio rerio*) were purchased from a local pet shop and acclimated in aerated tanks containing distilled water. Zebra fish were fed with a commercial fish feed twice a day and kept at approximately 28° C with a 14 h: 10h light-dark cycle. Each zebra fish weighed 0.2–0.3 g, and each ten zebra fish were treated in static tanks containing 2.0 liter of water. Fish were divided into four experimental groups of 24 fish each. The hepatotoxicity was induced by 20mM concentration of paracetamol. This was followed by a change of system water for 2-21 hours with or without treatment. Experiments were terminated 5-24 hours after the start of paracetamol exposure (5-24hpe). Group 1 served as control, Group 2 fish were exposed to 20mM concentration of paracetamol for 3 hour. Group 3 and Group 4 were treated with standard silymarin and *Gyrocarpus asiaticus* extract (100mg/ml) for 24 hours following paracetamol induced hepatotoxicity. After treatment for 24hours, zebra fish were anaesthetized using melting ice. Then fish were quickly euthanized in melting ice. The liver tissues were separated and used immediately for analysis. Immediately after sacrificing the fish, the liver was excised and used for the estimation of lipid peroxidation (LPO) (Placer et al., 1976), reduced glutathione (GSH) (Moron et al., 1979), catalase (CAT) (Clai borne, 1985) and glutathione peroxidase (GPX) (Rotruck et al., 1973). The differences between groups were assessed by using the Statistical Package for Social Sciences (SPSS) software package for Windows. The effects of treatments were determined by analyzing the data using one-way ANOVA followed by Duncan’s multiple comparison tests. P values < 0.05 or < 0.01 were considered as statistically significant.

**Results and Discussion**

The antioxidant profile clearly showed a significant (P≤0.01) increase in GSH, CAT, TBARS, GPX in paracetamol induced zebra fish liver while treatment with silymarin and *Gyrocarpus asiaticus* extract restored the altered antioxidant profile to a near normal value (Table-I). Higher GSH in the liver of paracetamol-treated fish indicates severe oxidative damage caused by paracetamol and its metabolite n-acetyl-p-benzoquinone imine (NAPQI). Decrease in GSH in *Gyrocarpus* treated group may be attributed to conjugation of NAPQI under severe oxidative stress. Similarly elevated level of TBARS indicates lipid peroxidation with cell wall damage caused by reactive oxygen species generated during paracetamol metabolism. These findings are supported by another research where oral exposure of paracetamol (500mg/kg) in a fresh water fish for 24 hour duration induces oxidative damage and alter the antioxidant levels (Kavitha et al., 2011). Silymarin and *Gyrocarpus asiaticus* extract showed reduced hepatic lipid peroxidation, suggesting enhanced antioxidant activity that could neutralize the free radical generated under paracetamol exposure. The decrease in catalase activity in treatment groups at par to control inspite of exposure to paracetamol may be attributed to excess of superoxide anion radicals resulted from reduction in SOD activity. GPx detoxifies hydrogen and other organic hydroperoxides by utilizing glutathione (GSH) as cofactor by which it helps the cells from undergoing lipid peroxidation (Shivashri et al., 2013). The increased tissue GPx activity in zebra fish liver under paracetamol exposure reveals severe oxidative stress induction and cells survival instinct from induced toxicity. The increased GPx and GSH concomitantly signify that the cells were in acute stage of toxicity stress and prepared for detoxifying and restoring the cells to normal status. However treatment with silymarin and *Gyrocarpus asiaticus* extract restored the GPx and GSH level up to normal level in liver signifying induction of antioxidant mechanisms which
results in quick restoration of the impaired status.

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References

Table I: Effect of Gyrocarpusasiaticus on liver antioxidant profile in paracetamol induced hepatotoxicity in zebra fishes.

<table>
<thead>
<tr>
<th>Group</th>
<th>GSH (nmoles /mg of protein)</th>
<th>LPO (nmoles of MDA released /mg of protein)</th>
<th>CAT (µmoles of H₂O₂ decomposed/min/ mg of protein)</th>
<th>GPx (nmoles of glutathione (GSH) oxidized/min/ mg of protein)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>48.29 ± 8.88</td>
<td>10.93 ± 2.89</td>
<td>4.19 ± 1.16</td>
<td>497.31 ± 89.59</td>
</tr>
<tr>
<td>Paracetamol</td>
<td>116.14 ± 33.38</td>
<td>50.09 ± 10.27</td>
<td>18.11 ± 3.63</td>
<td>1382.01 ± 298.42</td>
</tr>
<tr>
<td>Paracetamol + Silymarin</td>
<td>41.36 ± 2.39</td>
<td>5.55 ± 0.25</td>
<td>1.27 ± 0.20</td>
<td>405.34 ± 36.14</td>
</tr>
<tr>
<td>Paracetamol+G.A.</td>
<td>29.97 ± 3.30</td>
<td>5.03 ± 0.59</td>
<td>1.56 ± 0.51</td>
<td>279.23 ± 29.34</td>
</tr>
</tbody>
</table>

Means bearing different superscripts in a column differ significantly between groups.