Histology Studies on the Cerebroprotective Potential of Resveratrol against Ischemia-reperfusion Injury

12.1 Introduction

Resveratrol is reported to have inhibition of delayed neurological damages and decreased glial cell activation (Wang et al. 2002). Astrocytes play a key role in maintainence of neuronal environment and are crucial for the survival of neurons. After ischemic injury, astrocytes were activated and infarcted tissue was observed. Furthermore, activated astrocytes altered anti-oxidative defense systems and inflammatory responses in the ischemic tissue and infarction size of the tissue was increased. In the infarcted tissue histological characters were examined by staining with hematoxylin and eosin (H&E). The present study evaluated the cerebroprotective action of resveratrol by the examinations of histological characters.

12.2 Experimental protocols

Wistar rats of either sex weighing between 250 to 300 g were used in the study. Experimental protocols was as follows

Group-1 Sham control
Group-2 I/R (Rats recived 30 min BCCA occlusion and 4 hr reperfusion)
Group-3 Resveratrol treated (20 mg/kg)

Vehicle and Resveratrol (in 10 % DMSO) were administered i.p. 5 min before reperfusion. Each group consisted of 6 animals. Animals in all groups were anesthetized with thiopental sodium (30 mg/kg i.p) and were subjected to BCCA occlusion for 30 min and 4 hr reperfusion as described in Chapter 8 was followed. At the end of experiment, the brains were removed and subjected to histological examination as described.
12.3 Results

The histopathology of the brain sections of Sham control, I/R and Resveratrol treated groups were shown in Figure. From the histopathological studies, dense eosinophilic cytoplasm and dark stain triangular nucleus were observed in brain regions of I/R rats. These abnormalities were decreased in the resveratrol treated (20 mg/kg) rats.

![Histology of coronal sections of rat brain](image)

**Fig:** Histology of coronal sections of rat brain

A: indicates images show eosinophilic cytoplasm and B: indicates images show neuron nucleus

I/R indicate ischemia-reperfusion. RSV indicates resveratrol treated (20 mg/kg i.p.)

In I/R, appearance of triangular nucleus and dense eosinophilic cytoplasm and significant recovery from formation of dense eosinophilic and triangular nucleus in resveratrol treated group.
12.4 Discussion

Irreversible cerebral ischemia induces two types of morphological injury: selective neurological damage and infarction (Sun et al. 2009). Ischemic neuronal damage has been characterized as karyolysis, pyknosis and karyorrhexis with loss of hematoxylin affinity or cytoplasmic eosinophilia in light-microscopic examination (Nedergaard 1987; Garcia et al. 1995; Fujioka et al. 1999; Guimiot et al. 2008).

The present study evaluated the histological changes in cerebral tissue. Some of the researchers demonstrated that dense eosinophilic cytoplasm and dark stain triangular nucleus were found in I/R injured rats (Della-Morte et al 2009; Liu et al. 2001; Elmore 2007). Dense eosinophilic cytoplasm and dark stain triangular nucleus were observed in I/R injured rats. In contrast, these abnormal changes were diminished in resveratrol treated rats. These results demonstrated that the resveratrol has cerebroprotective action. Therefore it was suggested that resveratrol might be having cerebroprotective action by alteration of histological abnormalities.

12.5 Conclusion

Resveratrol treated group rats showed tremendous recovery from abnormalities in ischemic reperfusion insult. Therefore we suggest that resveratrol has potent cerebroprotective action against cerebral ischemia-reperfusion injury.
References


Summary and Conclusions

Summary

The most successful therapy for cerebral ischemic disease is reperfusion. It produces restoration of blood supply to ischemic tissue. While prompt reperfusion of cerebral ischemic brain tissue is critical for restoring normal function and the ultimate survival of the tissue, it may also exacerbate cerebral injury and causes deleterious biochemical changes, these changes triggered to antagonize the beneficial effects of reperfusion leading to cerebral injury called as cerebral reperfusion injury. Research on cerebral reperfusion injury treatment is a new era for acute stroke therapy. Despite a wealth of experimental knowledge and several promising reports in both experimental and clinical out studies, there is yet no pharmacological therapy which is considered to be a good standard for use in treatment of ischemia-reperfusion injury.

In our study, cerebral infarction was determined using TTC staining technique. Severe infarction was noticed in I/R injured rats and significant reduction of infarction was observed in resveratrol treated I/R injured rats. Resveratrol showed dose dependent cerebroprotection in I/R injured rats. Furthermore, 20 mg/kg dose of resveratrol was used for estimation of biochemical parameters in I/R injured rats.

Previous investigations suggested that in cerebral ischemia-reperfusion, oxidative stress and inflammation has key pathological role to cause tissue apoptosis. Resveratrol has potent anti-oxidative and anti-inflammatory actions. By this virtue we have gone for the evaluation of cerebroprotective actions of resveratrol.
In our study we found that, MDA levels were decreased, SOD and CAT levels were increased in resveratrol treated I/R rats. These results supported that the anti oxidant effect of resveratrol is responsible for its cerebroprotective action. Resveratrol treatment noticeably reduced inflammation characterized by decreased levels of MPO, TNF-α, IL-6 and ICAM as well as increased levels of IL-10 in I/R injured rats. These results supporting the anti-inflammatory role of resveratrol which is responsible for its cerebroprotective action.

Conclusions

1. Bi lateral common carotid artery occlusion (30 min) and reperfusion (4 hr) is the experimental model was used to induce Global cerebral infarction in the present study.

2. Resveratrol showed significant cerebroprotective action against I/R injury in terms of reduction in percent cerebral infarct volume.

3. Resveratrol (5, 10, 20 and 30 mg/kg) showed dose dependant cerebroprotective actions against I/R injury. Resveratrol 20 mg/kg was selected for biochemical estimations (oxidative and inflammatory markers) and histology examination in I/R injured brain tissues.

4. The anti oxidant role of resveratrol in cerebroprotective action was confirmed by estimation of MDA, SOD and Catalase levels in I/R injured brain tissue. MDA levels were significantly decreased; SOD and Catalase levels were significantly increased in resveratrol treated group of rats.

5. The anti-inflammatory role of resveratrol in cerebroprotective action was confirmed by estimation of MPO, TNF-α, IL-6, ICAM-1 and IL-10 levels in I/R brain tissue. MPO, TNF-α, IL-6 and ICAM-1 levels were
significantly decreased, IL-10 levels were significantly increased in resveratrol treated group of rats.

6. Resveratrol exhibited significant cerebroprotective potential against cerebral ischemia and reperfusion induced cerebral infarction in rats.

7. The possible mechanisms involved in the cerebroprotective potential of resveratrol might be anti oxidant and anti-inflammatory.

8. Therefore the present study demonstrated the antioxidant and anti-inflammatory potential of resveratrol in BCCA occlusion and reperfusion induced Global cerebral infarction in rats.

Scope for future aspects

1. However, further studies are needed to investigate other possible mechanisms involved in cerebroprotective potential of resveratrol.

2. The chronic studies are needed to investigate other possible resveratrol mechanisms in cerebral ischemia-reperfusion injury.