Esculetin ameliorates the lipopolysaccharides-induced inflammation in neuroblastoma cells via the AMPK-NLRP3 pathway

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Aim:
To investigate the effect of esculetin in lipopolysaccharide (LPS) induced neuroinflammation in Neuro-2a cells.

Background:
Since most of the available treatments for neuropathic pain are ineffective or produce unwanted effects, alternative therapeutic approaches are being investigated. This study evaluated the impact of esculetin on LPS-triggered inflammatory cascades using Neuro-2a neuroblastoma cells. We hypothesized that esculetin might produce an anti-inflammatory effect via the AMPK-NLRP3 pathway.

Methodology:
The anti-inflammatory and antioxidant effects of esculetin (10 & 20 µM) were assessed using the Neuro-2a neuroblastoma cells. The Neuro-2a cells were pre-treated with esculetin for 24 h and then exposed to the LPS (1 µg/mL) for one h. Following LPS incubation, 2',7'-dichlorodihydrofluorescein diacetate (DCFDA), immunoblotting and immunocytochemistry assays were performed to examine the protective effect of esculetin. To confirm the therapeutic effect of esculetin in neuropathic pain, a chronic constriction injury (CCI) model was developed in Sprague-Dawley rats.

Results:
As expected, treatment with LPS significantly increased reactive oxygen species (ROS) production compared to control. Esculetin at 20 µM concentration significantly reversed the increased levels of ROS. LPS-exposed Neuro-2a cells also showed an increased expression of nucleotide-binding oligomerization domain leucine-rich repeat and pyrin domain-containing (NLRP3), and decreased expression of nuclear factor erythroid 2-related factor-2 (Nrf-2) and 5' adenosine monophosphate-activated protein kinase-3 (AMPK-3) compared to the control group. Esculetin at both doses (10 & 20 µM) significantly downregulated the expression of NLRP3 and increased the expression of Nrf-2 and AMPK-3 compared to the LPS-treated group. These results indicated the antioxidant and anti-inflammatory effect of esculetin in LPS-induced neuroinflammation in Neuro-2a cells. In a preclinical study, the decreased pain threshold was seen in the paw withdrawal latency (sec), paw withdrawal threshold (g), paw withdrawal pressure (g), allodynia score, sciatic function index and proportion of postural index values in CCI animals compared to the sham group.

Conclusion:
Our results indicate the anti-inflammatory and protective effects of esculetin via the AMPK-NLRP3 pathway. We also suggest that esculetin may reverse the CCI-induced lowered pain threshold.

Biography
Ziaur Rahman is pursuing his PhD in Biological science under the supervision of Dr Manoj Dandaker. He completed his master in Pharmacology and Toxicology from NIPER, Mohali, India. He has published more than five papers in reputed journals and serving as an editorial board member of the Biological Sciences journal. (Up to 100 words)

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