Hsp90 inhibition protects the hCMEC/D3 brain microvascular endothelial cells against oxidative stress

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Introduction

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- **Blood-brain barrier (BBB) dysfunction is associated with major neurological diseases, including cerebral ischemia/reperfusion (I/R) (stroke) and traumatic brain injury (TBI).**
- **The development of new therapeutical approaches against these devastating disorders is urgent.**
- **BBB endothelial cells interact with the surrounding cells of the CNS, including neurons and astrocytes. Those cells are important for maintaining brain homeostasis, and for preventing the entry of harmful substances.**
- **Recent observations reveal that P53 regulates endothelial barrier function.**
- **P53 supports endothelial barrier function by disrupting the inflammatory RhoA/MLC2 pathway and suppressing the actin-severing activity of cofilin.**

Uddin MA, Barabutis N., *J Cell Biochem* (2019)
Sweeney et al., *Nat Rev Neurol* (2018)

Introduction

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- **Hsp90 is a chaperone protein, involved in fundamental cellular processes including DNA repair, development, and immune responses.**
- **Activation of Hsp90 has been linked to the development of multiple pathological conditions including cancer, inflammation, and viral infection.**
- **Inhibition of Hsp90 is associated with anti-inflammatory responses.**
- **P53 mediates the barrier enhancing effects of Hsp90 inhibitors, al least in part.**
- **This transcription factor reduces APE1/Ref1 (ROS generator) in human tissues.**
- **In our study we employed commercially available hCMEC/D3 cells from human temporal lobe microvessels to investigate the protective role of Hsp90 inhibition towards the H_2O_2-induced brain endothelium breakdown.**

Uddin MA, Barabutis N., *DNA Repair (Amst),*95:102952 (2020)

Methods

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- Cell Culture
  - hCMEC/D3 cells
- In vitro treatments: H_2O_2, Hsp90 inhibitors (17-DMAG, AUY-922)
- Western Blot Analysis
- ROS measurement
  - H_2DCFDA
- Measurement of endothelial barrier function
  - ECIS-TEER measurements

Results

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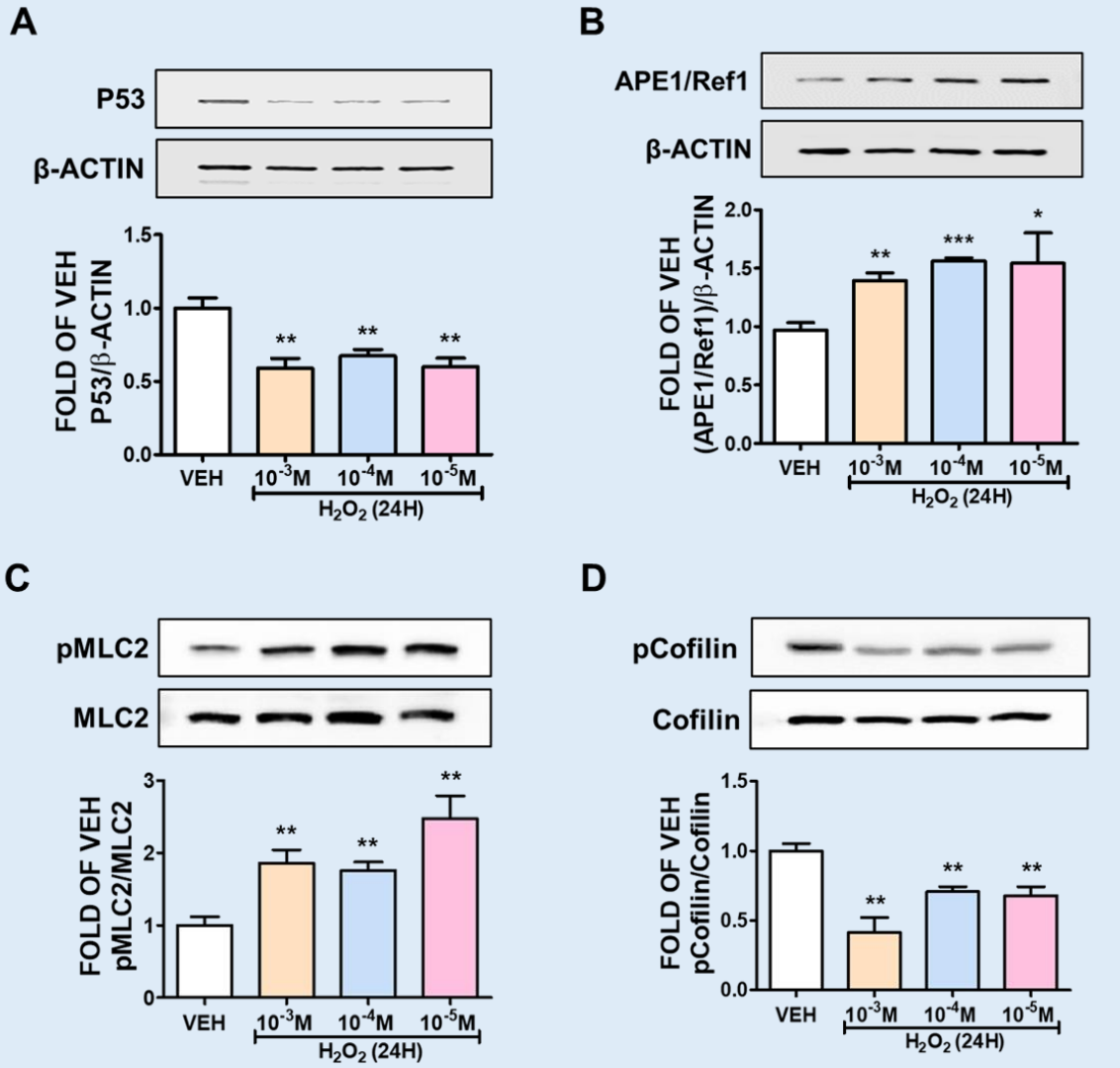
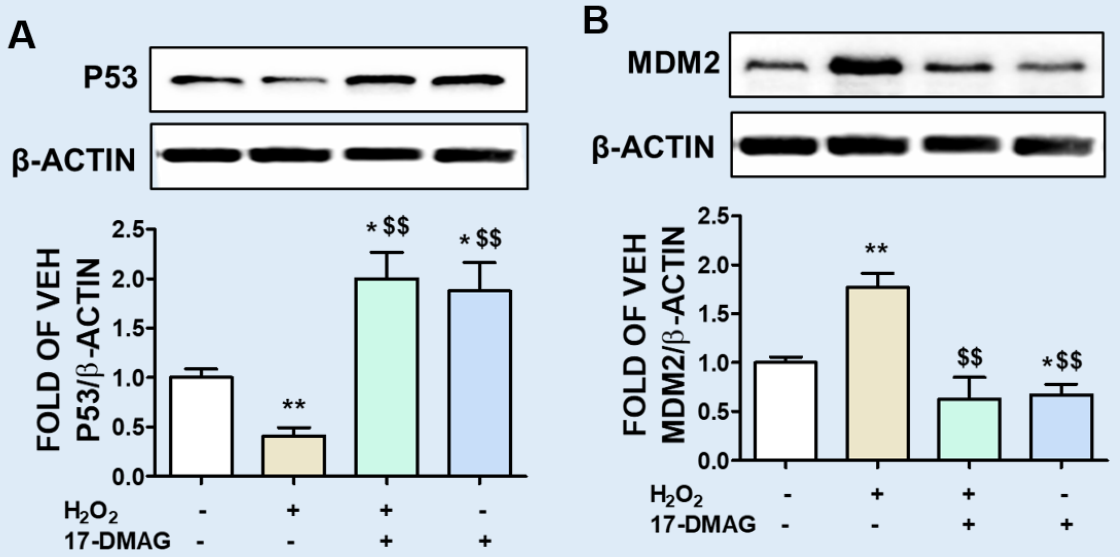
H_2O_2 induces brain endothelial barrier dysfunction





Hsp90 inhibition opposes the H_2O_2 -induced P53 suppression

Results

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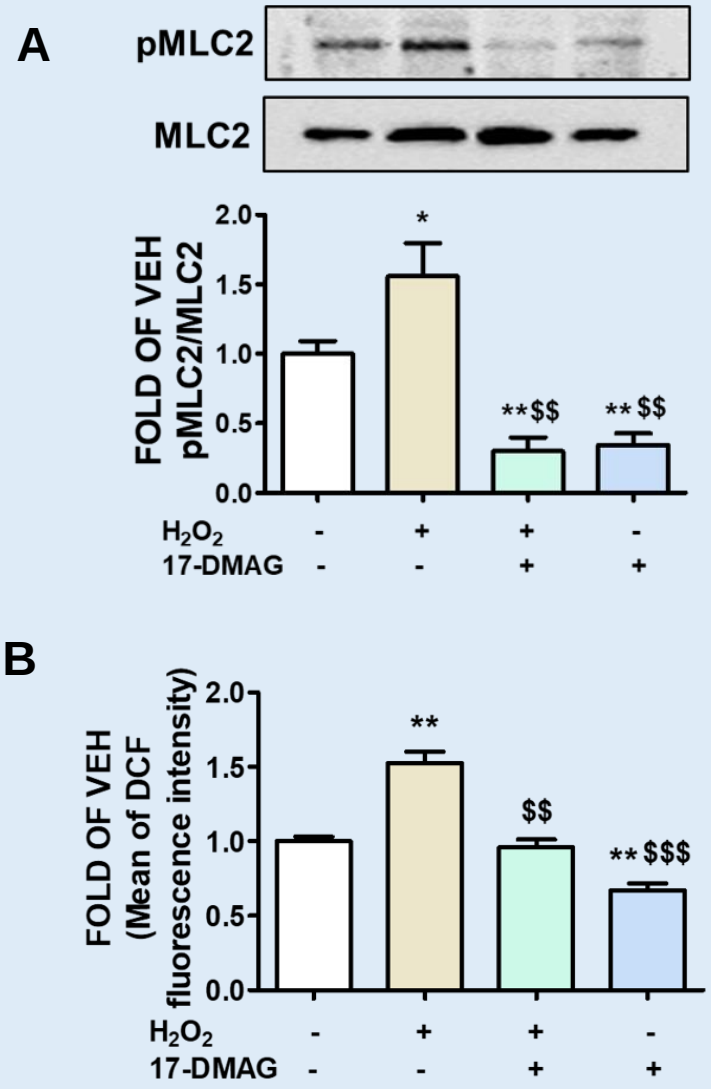

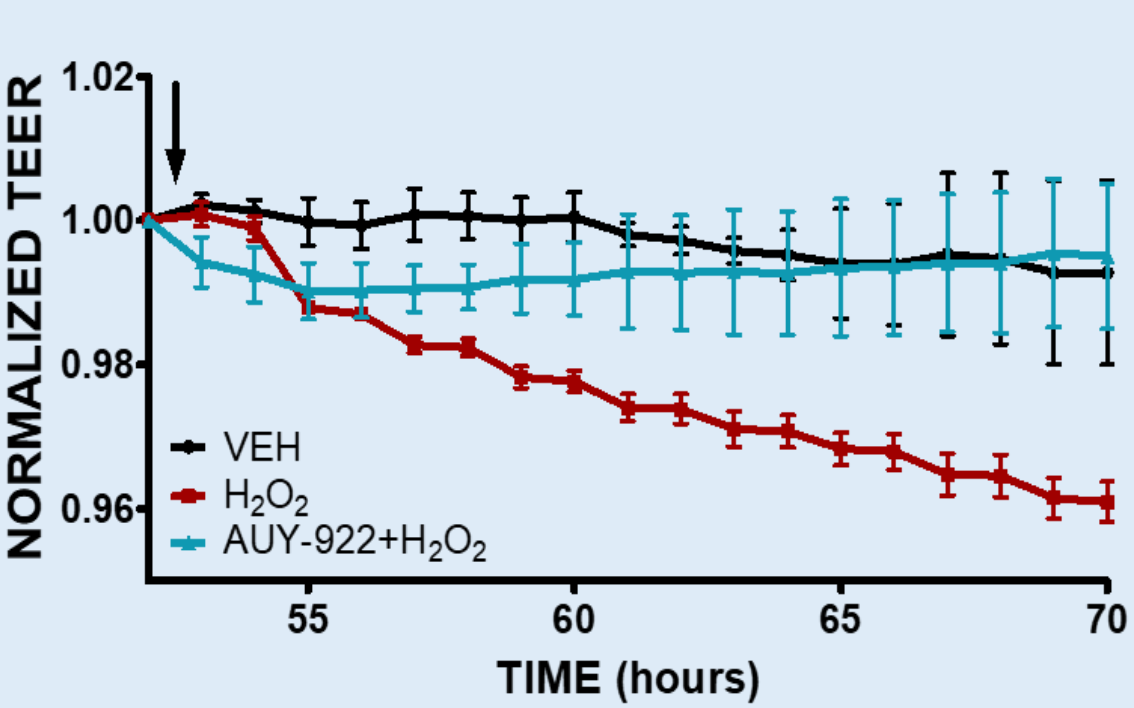

Hsp90 inhibitors protect against H_2O_2 -induced hCMEC/D3 barrier breakdown

Discussion and Conclusions

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- Brain endothelial dysfunction contributes to the development and establishment of serious neurodegenerative diseases and cerebral disorders.
- Herein we report the capacity of Hsp90 inhibitors (P53 inducers) to support the brain endothelial function under severe oxidative conditions.
- Thus, we suggest that these anti-inflammatory agents may deliver new possibilities for the treatment of diseases related to brain endothelial dysfunction, including cerebral ischemia/reperfusion (I/R) (stroke), and traumatic brain injury (TBI).

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