

Supplementary material

# Antioxidant Potential of Diosmin and Diosmetin against Oxidative Stress in Endothelial Cells

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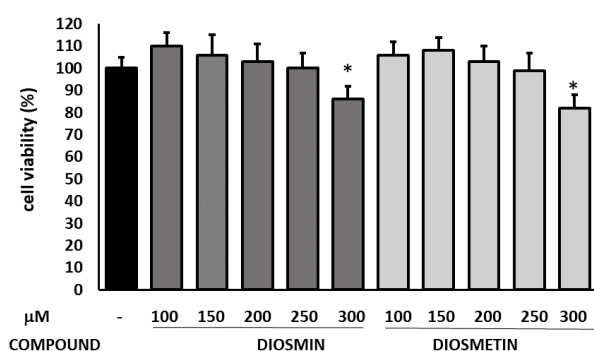
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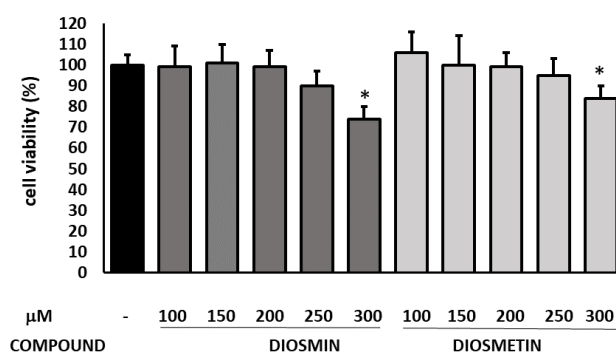
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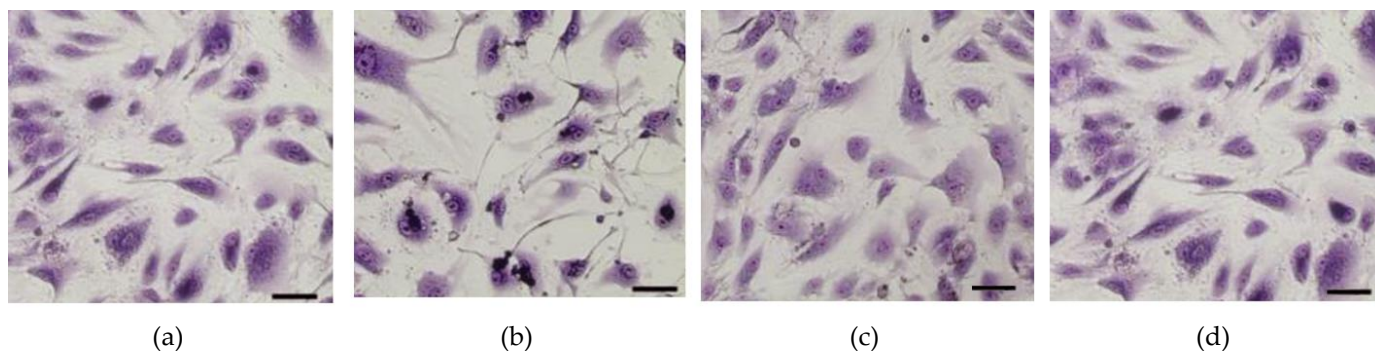


(a)

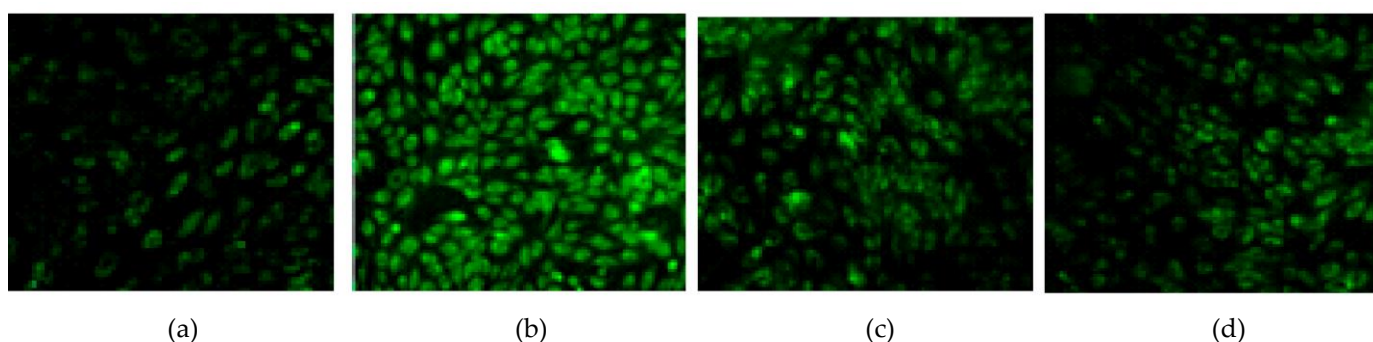


(b)

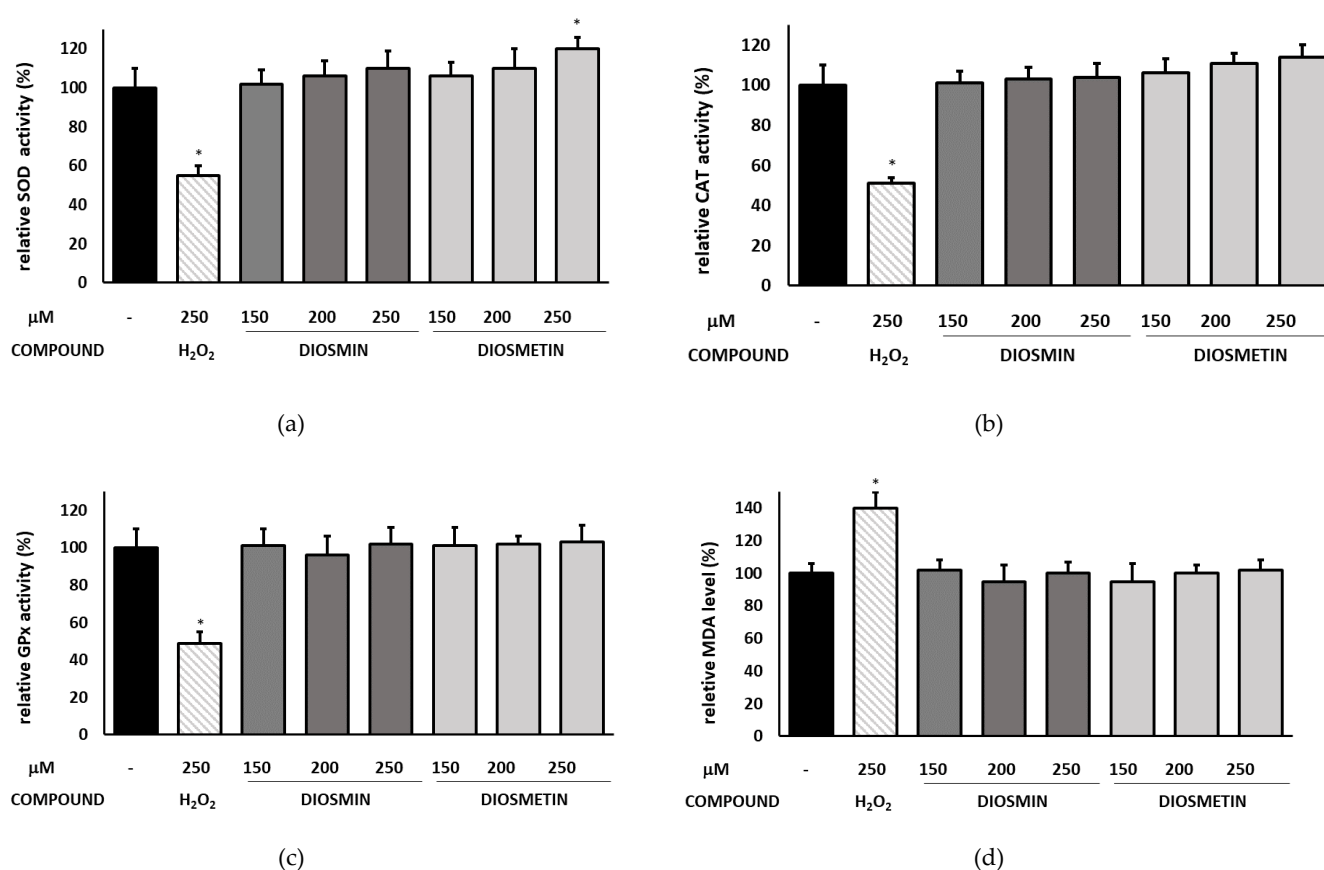
**Figure S1.** Cell viability determined by the MTT (a) and Alamar Blue (b) assay expressed as a percentage of control (0.5% DMSO). Cells were treated with diosmin/diosmetin at different concentrations. The data are means  $\pm$  SD (n = 3). One-way ANOVA followed by Dunnett's multiple comparison post hoc test; \*indicates statistically significant difference ( $p < 0.05$ ) versus untreated controls.



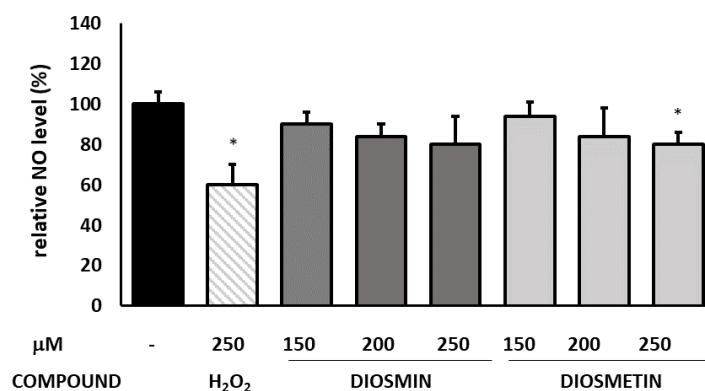
**Figure S2.** May-Grünwald–Giemsa (MGG) staining of endothelial cells: (a) control, (b) cells treated with  $H_2O_2$ , (c) cells preincubated with diosmin prior to  $H_2O_2$  exposure, (d) cells preincubated with diosmetin prior to  $H_2O_2$  exposure. Magnification 200 $\times$ . Bar = 20  $\mu$ m.



**Figure S3.** Intracellular Reactive Oxygen Species ( $\text{H}_2\text{DCFDA}$  assay): (a) control, (b) cells treated with  $\text{H}_2\text{O}_2$ , (c) cells preincubated with diosmin prior to  $\text{H}_2\text{O}_2$  exposure, (d) cells preincubated with diosmetin prior to  $\text{H}_2\text{O}_2$  exposure.



**Figure S4.** Effect of the diosmin/diosmetin treatment and the  $\text{H}_2\text{O}_2$  exposure on the antioxidant enzyme activity calculated as a percentage in comparison with the untreated control. (a)–relative activity of superoxide dismutase (SOD), (b)–relative activity of catalase (CAT) (c)–relative activity of glutathione peroxidase (GPx) (d)–relative malondialdehyde (MDA) concentration. The data are means  $\pm$  SD ( $n = 3$ ). \* indicates a statistically significant difference ( $p < 0.05$ ) versus the control cells. One-way ANOVA followed by Dunnett's multiple comparison post hoc test.



**Figure S5.** Effect of the diosmin/diosmetin treatment and the H<sub>2</sub>O<sub>2</sub> exposure on the NO level calculated as a percentage in comparison with the untreated control. The data are means  $\pm$  SD (n = 3). \*indicates a statistically significant difference ( $p < 0.05$ ) versus the control cells. One-way ANOVA followed by Dunnett's multiple comparison post hoc test.