Effect of the CB₁ cannabinoid receptor antagonist AM251 on kissorphin protection against amyloid-β neurotoxicity


Introduction: Previous in vitro and in vivo studies demonstrate protective properties of kissorphin (KSO) peptides against amyloid-β (Aβ) neurotoxicity. Overexpression of the KiSS-1 gene, that encodes the KSO peptides, is also neuroprotective. Endocannabinoids and KSO peptides are neuroprotective against Aβ 25-35, but not Aβ 31-35 peptides. The KiSS-1 gene expression is regulated by endocannabinoids. The aim of this study was to determine whether endocannabinoids contribute to KSO protection against Aβ toxicity using a CB₁ cannabinoid receptor antagonist.

Method: This study employed MTT cell viability assays to investigate the effects of the CB₁ antagonist AM281 on KSO 1-6 protection against Aβ 25-35 neurotoxicity in human neuroblastoma SH-SY5Y cells. The effects of AM281 on Aβ 25-35 induced neurotoxicity in KiSS-1 gene overexpressing SH-SY5Y cells (PKiSS) was also investigated. Data was analyzed by one-way analysis of variance (ANOVA).

Results: The CB₁ antagonist AM281 (0.01-10µM) promoted a concentration dependent increase in 10µM Aβ 25-35 induced neurotoxicity in SH-SY5Y cells in the presence of 10µM KSO 1-6 (Figure 1A). The PKiSS protection against 10µM Aβ 25-35 was reversed by the CB₁ antagonist AM281 (10µM) and anti-KSO antibody (1µg/ml). In the presence of anti-KSO antibody 10µM 2-AG was protective against 10µM Aβ 25-35.

Figure 1. (A) Dose-response curves for AM281 in the presence of 10µM Aβ 25-35 with or without 10µM KSO 1-6, on MTT reduction in SH-SY5Y cells. (B) PKiSS SH-SY5Y cells were exposed to 10µM Aβ 25-35 alone; 10µM Aβ 25-35 plus 10µM AM281; 10µM Aβ 25-35 plus 1µg/ml anti-KSO antibody; 10µM Aβ 25-35 plus 1µg/ml control antibody; 10µM Aβ 25-35 plus 1µg/ml anti-KSO antibody with 10µM 2-AG; 10µM Aβ 25-35 plus 1µg/ml control antibody with 10µM 2-AG, and cell viability determined by MTT reduction. Results are mean ± SEM (n=8 for each data point); * = P< 0.05 vs Aβ 25-35 alone (one-way ANOVA).
Conclusion: In conclusion, protection against Aβ 25-35 induced neurotoxicity by KSO and KiSS-1 overexpression in SH-SY5Y cells is reversed by the AM281 CB1 antagonist. Anti-KSO antibodies prevent neuroprotection by KiSS-1 overexpression and 2-AG restores neuroprotection. This suggests KSO neuroprotection against Aβ involves activation of endocannabinoids.

References:


