Abstract Monotematico SIF Innovazione e Malattie Rare: dalla ricerca pre-clinica al paziente

Title (max 25 parole e incluso il disegno di studio se appropriato):
Potential application of Growth Hormone Secretagogues (GHS) for Amyotrophic Lateral Sclerosis (ALS) treatment: mechanisms of action and neuroprotective effects characterization in human SH-SY5Y SOD1^{G93A} cells

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Aim (state background and specific objective of study): ALS is a rare motor neuron disease for which all interventions are currently only symptomatic and palliative.
The strong need to characterize more effective drugs led us to propose hexarelin and JMV2894, two synthetic compounds belonging to GHS family, as developing molecules for the treatment of ALS.
Hexarelin is a hexapeptide that exerts neuroprotective and cytoprotective effects at the mitochondrial level in cardiac and skeletal muscles, both in vitro and in vivo, while JMV2894 is an agonist of GHS-R1a, which stimulates calcium mobilization in vitro and GH release in vivo, and modulates mitochondria functioning and ROS production.
Methods (briefly describe methods used including 1. design and setting of the study, clear description of interventions and comparisons including drug doses and concentrations; 2. clear description of materials, such as cell lines and conditions; 3. type of statistical analyses used): Human neuroblastoma cells overexpressing SOD1^{G93A} mutated protein (SH-SY5Y SOD1^{G93A} cells) were treated with H_2O_2 (150µM) and GHS (1µM) for 24h. Photomicrographs of stained cells were quantified by skeleton and fractal analysis. The mRNA expression levels of apoptotic and inflammatory markers were quantified by real-time PCR, while protein levels were measured by WB. Data were analysed by one-way ANOVA, followed by Tukey's t-test.
Results (summarize results obtained, report quantitative data, confidence intervals, and p values for comparisons): Morphometric evaluation, mRNA levels and effector proteins quantifications showed that H_2O_2 treatment induced apoptotic activation ($p<0.001$). Both GHS significantly blunted H_2O_2 effects decreasing Bax/Bcl-2 ratio ($p<0.001$), inhibiting the activation of caspases ($p<0.05$) and modulating MAPKs and PI3K/Akt phosphorylation ($p<0.01$). These effects were probably mediated by epigenetic mechanisms as shown by the significantly decreased percentage of γH2AX-positive cells in the GHS-treated group compared to the H_2O_2 one ($p<0.001$).
Conclusions (state conclusions reached): GHS are capable of protecting cells from OS-caused cytotoxicity, suggesting the possibility of developing new neuroprotective drugs with improved therapeutic potential.
Keywords (max 5): ALS, SOD1, GHS, neuroprotection, drug validation