

## NATURE-INSPIRED HYBRIDS TARGETING NRF2-PATHWAY EXERT ANTI-INFLAMMATORY EFFECT ON HUMAN MONOCYTIC CELL LINE

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**Introduction:** Natural products offer a wide chemical diversity and provide a great source of therapeutics. By combining molecular fragments deriving from garlic and curcumin into new chemical entities, we synthesized new nature-inspired hybrids targeting Nuclear factor (erythroid-derived 2)-like 2(Nrf2)-pathway, with a significant antioxidant activity (Simoni et al., 2017). Since data from literature suggest that several natural compounds, including curcumin, exhibit anti-inflammatory activity, we tested whether, beside their well-established antioxidant activity, our nature-inspired molecules exert anti-inflammatory activity in a view of multi-target pharmacological agents, by possibly triggering Nrf2-pathway.

**Materials and methods:** Using an undifferentiated human monocytic cell line (THP1), we first evaluated the ability of our nature-inspired hybrids to induce the nuclear translocation of Nrf2 by performing Western Blotting. To investigate the anti-inflammatory potential of our molecules, THP-1 cells were pretreated with the hybrids for 24h and then exposed to lipopolysaccharide (LPS) for 3h in order to promote the inflammatory response. The anti-inflammatory capability of the hybrids were determined using RT-PCR to assess the modulation in the expression of pro-inflammatory cytokines such as tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and interleukin-8(IL-8). By performing ELISA test, we then tested the effect of the molecules on the release of the same pro-inflammatory cytokines (TNF $\alpha$ , IL-8). To establish whether the anti-inflammatory action of the molecules relies on the activation of Nrf2, THP1 cells were subjected to Nrf2silencing and the expression and release of TNF $\alpha$  and IL-8 were determined by RT-PCR and ELISA test, respectively.

**Results and discussion:** Our results suggest that the nature-inspired hybrids significantly suppress the LPS-induced increases in TNF $\alpha$ , and IL-8mRNA expression, as well as the release of these pro-inflammatory cytokines in undifferentiated THP-1 cells. Such a reduction in the mRNA expression and release of pro-inflammatory cytokines was also observed after Nrf2silencing, thus suggesting that our nature-inspired hybrids exert an anti-inflammatory activity not mediated by the activation of Nrf2. Further experiments are needed to investigate the molecular pathway through which the nature-inspired hybrids exert the anti-inflammatory activity.

**Conclusions:** Overall, our data demonstrate the anti-inflammatory activity of our nature-inspired molecules and provide evidence of their therapeutic potential as anti-inflammatory agents.