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Bio-accessible milk casein derived tripeptide (LLY) mediates overlapping anti-inflammatory and anti-oxidative effects under cellular (Caco-2) and *in vivo* milieu

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Abstract

Inflammation and oxidative stress are closely linked patho-physiological processes which occur concurrently in many diseased conditions. Recently, interdependence between these two processes explains the antioxidant paradox associated with failure to select appropriate agents required for prevention of diseases known to be induced by oxidative stress. Present study established the overlapping anti-inflammatory and anti-oxidative potential along with bio-accessibility of milk casein derived tripeptide (LLY). Tripeptide exhibited anti-inflammatory response under *ex vivo* conditions by suppressing ($p < 0.01$) mice splenocytes proliferation and modulating their cytokines (IFN- γ , IL-10 and TGF- β) with improved phagocytosis of peritoneal macrophages. Conversely, tripeptide displayed extraordinary radical scavenging ability and cellular anti-oxidative potential using chemical assays and H₂O₂ induced oxidative stress model on Caco-2 cells. Under cellular assessment, on one hand tripeptide inhibited ($p < 0.01$) intracellular ROS generation and reduced MDA and protein carbonyls but on the other also increased ($p < 0.01$) the activity of anti-oxidative enzyme, catalase without much effect on SOD and GPx. This anti-oxidative potential was further established by studying relative expression of genes (*Nrf-2* and *Keap1*) and Nrf-2 nuclear translocation associated with anti-oxidative signalling in Caco-2 cells. Bio-accessibility of tripeptide and its intact transport across Caco-2 cell monolayer was also found to be $1.72 \pm 0.22\%$ through PepT1 mediated transport mechanism. Besides, tripeptide displayed strong anti-oxidative and anti-inflammatory potential under *in vivo* conditions in mice against

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