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# ABSTRACT BOOK

## BUFFALO RICOTTA PEPTIDE REDUCES OXIDATIVE STRESS IN INTESTINAL EPITHELIAL CELLS AND ANGIOTENSIN II-INDUCED VASOCONSTRICTION ON MICE MESENTERIC ARTERIES BY INDUCTION OF NRF2 TRANSLOCATION

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Buffalo dairy products are an important source of bioactive peptides. These are inactive, since encrypted in their parent sequences, but turn active when released by fermentation or ripening during food processing, or by digestive enzymes during gastrointestinal transit.<sup>1</sup> Once released, the bioactive peptides are able to exert either local or systemic pharmacological effects, involving specific biochemical pathways and leading to the identification of undisclosed drug-target interactions. This is why food-derived bioactive peptides, particularly from dairy products, are attractive tools for drug discovery campaigns.<sup>2</sup>

In the present study, to evaluate the biological activities of encrypted peptide sequences from buffalo ricotta cheese, a simulated gastrointestinal (GI) digestion of the raw material was performed.<sup>3</sup> Chemical and pharmacological characterization of the digest leads to the identification of a novel peptide endowed with antioxidant and anti-hypertensive action. The GI digest was fractionated by Semiprep-HPLC and fractions were tested against reactive oxygen species (ROS) release in H<sub>2</sub>O<sub>2</sub>-treated intestinal epithelial cell line. UHPLC-PDA-MS/MS analysis revealed the presence of an abundant  $\beta$ -lactoglobulin peptide (*SFNPTQL*, **BPR2**) in the most active fraction. The peptide was synthesized *via* Fmoc chemistry solid phase peptide synthesis and pharmacologically characterized. Pharmacological assays revealed the antioxidant activity of **BPR2**, involving ROS reduction, Nuclear factor erythroid 2-related factor 2 (Nrf2) activation and cytoprotective enzymes expression. Bioavailability studies through Caco-2 cell monolayer<sup>3</sup> revealed equal bi-directional transport and linear permeability of **BPR2**, consistent with a passive diffusion mechanism. In addition to its local effects, administration of **BPR2** on mice mesenteric arteries counteracts the Angiotensin II-induced vasoconstriction by Nrf2 nuclear translocation, reduction of active form of Ras-related C3 botulinum toxin substrate 1 (Rac1) and NADPH oxidase activity. These data suggest a specific further highlight the role of buffalo ricotta cheese-derived peptides against oxidative stress related diseases and suggest their health promoting potential. Further studies are ongoing to identify the specific **BPR2** structure-activity relationship and to identify its biological targets.

### References

1. Basilicata, M. G.; Pepe, G.; Sommella, E.; Ostacolo, C.; Manfra, M.; Sosto, G.; Pagano, G.; Novellino, E.; Campiglia, P. *Food. Res. Int.* **2018**, *105*, 1003-1010.
2. Basilicata, M. G.; Pepe, G.; Adesso, S.; Ostacolo, C.; Sala, M.; Sommella, E.; Scala, M. C.; Messori, A.; Autore, G.; Marzocco, S.; Campiglia, P. *Int. J. Mol. Sci.* **2018**, *19*, E1955.
3. Pepe, G.; Sommella, E.; Ventre, G.; Scala, M. C.; Adesso, S.; Ostacolo, C.; Marzocco, S.; Novellino, E.; Campiglia, P. *J. Funct. Foods* **2016**, *26*, 494-505.