Natural Deep Eutectic Solvents (NADES) as a strategy to increase plasma levels of blueberry phenolic-derived postbiotics

Silva D.T*[1], Vizzotto M[2], Rodrigues E[3], Barcia T.B[1], Emanuelli T[1].
[1]Universidade Federal de Santa Maria, Santa Maria/RS; [2]Empresa Brasileira de Pesquisa Agropecuária/Embrapa; [3]Universidade Federal do Rio Grande do Sul.
*Doctoral student – daritrivisiolsilva@gmail.com

Natural deep eutectic solvents (NADES) are eco-sustainable solvents that have been recently proposed to be a safe alternative for obtaining ready-to-use phytochemical extracts. Despite their putative biocompatibility, scarce information is available on how NADES would affect the bioavailability and bioactivity of phytochemicals. Among phytochemicals, phenolic compounds (PC) attract great interest because of their health promoting properties. Nevertheless, many health benefits of PC are actually triggered by PC-derived metabolites, which are formed by gut microbiota and are considered postbiotics. The aim of the present study was to evaluate *in vivo* whether PC-derived metabolites of blueberry extract obtained using NADES solvent have a different pharmacokinetic profile than that obtained using organic solvent. Blueberry fruits were extracted using a choline chloride:glycerol:citric acid (0.5:2:0.5 molar ratio, plus 25% water) NADES or an organic solvent (methanol:water:formic acid; 50:48.5:1.5, v/v/v). Wistar rats (200-250 g) were treated (intragastric) with a single dose (10.0 mg of PC kg^{-1} of body weight) of crude blueberry extract obtained using NADES (CE-NADES) or organic solvent (CE-SORG), the latter one being previously dried and reconstituted in water. Animals were euthanized at 15, 30, 60, 120, 240, 360, and 720 minutes after extract administration (n=4/group/per time) to collect cecal feces and intracardiac blood samples. PC-metabolites were analyzed by HPLC-ESI-MS/MS and pharmacokinetic parameters were calculated using a non-compartmental model. Protocatechuic acid (PA) and 3- (3-hydroxyphenyl) propanoic acid (HPPA) were produced from the microbial metabolism of anthocyanin and non-anthocyanin phenolic compounds. PA and HPPA reached peak plasma levels at 56.3 and 60.0 min for CE-SORG, while for CE-NADES group the peak levels were reached at 165.0 and 240.0 min, respectively. The maximal concentrations (C_{max}) of PA and HPPA increased by 37% and 50% in the CE-NADES group when compared to the CE-SORG. Additionally, the $AUC_{0\text{-}720\ min}$ value of PA and HPPA was 1.8 and 2.0-fold higher in the CE-NADES group than in the CE-SORG group. In general, PA and HPPA showed higher fecal concentration in the CE-NADES group, specially at the last time-points (240 to 720 min). This increase was greater than 90% for both analyzed metabolites. Our results demonstrate that NADES, in addition to producing biocompatible extracts, increases plasma levels of biologically active microbial-derived PC metabolites, namely PA and HPPA.

Keywords: green solvent, pharmacokinetic, metabolites, stability intestinal, phenolic acids

Acknowledgments: National Council for Scientific and Technological Development (CNPq) [grant number 303654/2017-1] and by Coordination for the Improvement of Higher Education Personnel (CAPES) [finance code 001].