Specific ELISAs to follow natural polyphenols in Human biological fluids.

Catherine Bennetau*[1], Valérie Lamothe , and Svitlana Shinkaruk

[1]Acides Nucléiques : Régulations Naturelle et Artificielle – Université de Bordeaux, Institut National de la Santé et de la Recherche Médicale : U1212, Centre National de la Recherche Scientifique : UMR5320 – IECB - 2 rue Robert Escarpit 33607 Pessac., France

Abstract

Introduction. Polyphenols are usually small molecules with potential health effects either through their antioxidant properties or via other cell pathways. However, to exert their effect they should reach somehow their target. The polyphenol concentrations are highly different from plant matter to human biological fluids. Here we show how we developed cheap immunoassays with great specificity and sensitivity to assay several family of active polyphenols in all different matrices.
Material and Methods. ELISAs were developed based on newly synthesized haptens coupled to proteins for immunoglobulin production or to obtain immobilize tracers. The assays follow a competitive process and can be either homologous or heterologous. Specificity is achieved by synthesizing several haptens functionalized on different Carbon atoms and challenged in competition test with close metabolites. Whenever necessary, the specificity is improved by counter selection of IgG harvesting the polyclonal antibody mixture with the undesired cross-reacting compound. Intra and inter-assay variation are assessed. Internal and external standards are used to monitor the extraction procedure.

Results. Several assays were developed against isoflavones including: genistein, daidzein, equol, formononetin, biochanin A and glycitein. Other tools were developed for flavanones including naringenin and hesperetin as well as for the prenyl-flavanol isoxanthohumol. An assay for enterolactone is also proposed. Using the isoflavone assays we were able to monitor plant, urine and plasma concentrations and to give several pharmacokinetic data in human.
Conclusion. As long as the organic chemistry syntheses of the haptens are performed properly, ELISAs against small molecules can be developed for simple use on many different types of samples with a good reproducibility and reliability.

Keywords: polyphenols, active substances, ELISA, bioavailability, food, urine, blood

*Speaker