Suitability of the Fluorescent Probe Diphenyl-1-Pyrenylphosphine to Measure Lipid and Aqueous Soluble Hydroperoxides in different Matrices

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By means of two alternative batch methods lipid and protein hydroperoxides (HP) were determined by fluorometry using the fluorescent probe diphenyl-1-pyrenylphosphine (DPPP). Fluorescence development was influenced by the type of solvent and HP whereas the presence in the media of antioxidants such as tocopherol and butylated hydroxytoluene had no effect. The addition of chloroform is not advisable although the chloroform:methanol (2:1, v/v) solvent mixture is very efficient for lipid extraction and, in consequence, widely used. As for this reason, this solvent mixture was used to extract and dilute the lipid fraction of samples and, subsequently, this was combined with selected solvents to develop a method with the maximum performance in determining HP in lipid extracts. Using a variety of lipid and lipid extracts, the final method proposed agreed well with the spectrometric method using thiocyanate for HP determination. The method using the DPPP fluorescent probe showed to be very sensitive, precise, accurate, free of interferences and specific for the determination of lipid soluble HP. This probe can be also used to measure HP soluble in hydroalcoholic media. That alternative procedure showed a similar performance to its lipid soluble equivalent and was able to measure hydrogen peroxide, peroxidized bovine serum albumin and water soluble HP in cooked beef protein extract. Moreover, by means of the previous addition of the triphenylphosphine selective reducing reagent to a sample aliquot this method can be specific for the determination of protein HP.