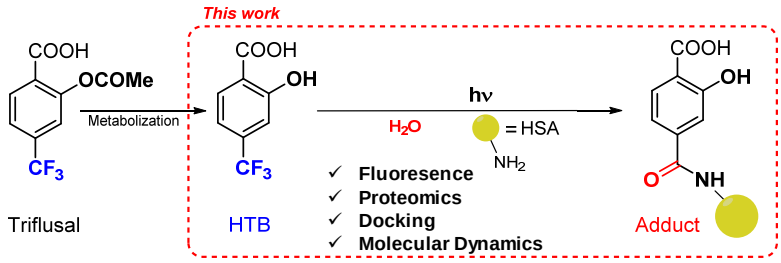
PHOTOBINDING OF TRIFLUSAL TO HUMAN SERUM ALBUMIN INVESTIGATED BY FLUORESCENCE, PROTEOMIC ANALYSIS AND COMPUTATIONAL STUDIES

M. Consuelo Jiménez,[a] Oscar Molins-Molina,[a] Raúl Pérez-Ruiz,[a] Emilio Lence,[b] Concepción González-Bello[b] and Miguel A. Miranda[a]

[a] Dpto. de Química/Instituto de Tecnología Química UPV-CSIC Universitat Politécnica de València, Camino de Vera s/n 46071 Valencia (Spain)
[b] Centro Singular de Investigación en Química Biolóxica e Materiais Moleculares (CIQUS), Departamento de Química Orgánica Universidade de Santiago de Compostela, c/ Jenaro de la Fuente s/n, 15782 Santiago de Compostela (Spain)

Triflusal [1] is a platelet antiaggregant employed for the treatment and prevention of thromboembolic diseases. After administration, it is biotransformed into its active metabolite, the 2-hydroxy-4-trifluoromethylbenzoic acid (HTB). We present here an investigation on HTB photobinding to human serum albumin (HSA), the most abundant protein in plasma, using an approach that combines fluorescence, MS/MS and peptide fingerprint analysis as well as theoretical calculations (docking and molecular dynamics simulation studies).



The proteomic analysis of HTB/HSA photolysates shows that HTB addition takes place at the ε-amino groups of the Lys137, Lys199, Lys205, Lys351, Lys432, Lys541, Lys545, and Lys525 residues and involves replacement of the trifluoromethyl moiety of HTB with a new amide function. Only Lys199 is located in an internal pocket of the protein and the remaining modified residues are placed in the external part. Docking and molecular dynamic simulation studies reveal that HTB supramolecular binding to HSA occurs in the "V-cleft" region and that the process is assisted by the presence of Glu/Asp residues in the neighborhood the external Lys, in agreement with the experimentally observed modifications. In principle, photobinding can occur with other trifluoroaromatic compounds and may be responsible for the appearance of undesired photoallergic side effects.

[1] W. McNeely, W., and Goa, K. L. *Drugs* **1998**, *55*, 823–833.