

Excited-state proton transfer in hydroxynaphthaldehydes covalently bound to proteins.

Autorzy

Andrzej Jankowski

Piotr Dobryszcki

J. Lipiński

Piotr Stefanowicz

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The kinetics and equilibrium of excited-state proton transfer (ESPT) in 2-hydroxynaphthaldehyde-1 (HNA-2.1) bound to proteins (bovine serum albumin, cytochrome c, and lysozyme) by an alkylamino bond was studied by means of fluorimetric steady-state and time-resolved methods. The results were compared to analogous data for 1-hydroxy naphthaldehyde-4 (HNA-1.4) bound to proteins and for other 2-naphthol derivatives bound to proteins by a sulfonamide bond. Conclusions concerning the influence on ESPT of the mode of binding and of intramolecular hydrogen bonds occurring in the case of HNA-2. 1 were drawn. An intramolecular hydrogen bond enhances the rate of ESPT but the molecular environment in the protein leads to an opposite effect by increasing reorganization energy during proton transfer. The results obtained prove that the mode of binding and the kind of group linking fluorophores to proteins influence considerably the rate and mechanism of ESPT. In naphthol groups bound to proteins by an alkylamino bond, proton dissociation depends strongly on the molecular environment in the macromolecule. This is due to the short length of the alkylamino bridge and its small interaction with electronic orbitals of the aromatic system. Fluorophores bound to proteins by a sulfonamide bond show a higher rate of ESPT, which is due partly to the electron withdrawing effect of the linking arm. The efficiency of ESPT for naphthol groups bound to proteins by a sulfonamide bond is, in most cases, sufficient for acidification of the medium and influence of the proton gradient in biological membranes.

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