

ENHANCEMENT OF SENSITIVITY IN SEPARATION TECHNIQUES WITH FLUORESCENCE DETECTION USING DEUTERATED WATER

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Fluorescent molecules with emission maxima above 500 nm are often dynamically quenched by OH oscillators present in water molecules. We have changed H₂O for D₂O to obtain higher fluorescence intensity signal. Our experiments carried out on spectrofluorometer have proved that anthracyclines can achieve about 300 % higher response in D₂O as a solvent. The affect can be employed in various analytical methods including HPLC and capillary electrophoresis coupled to fluorescence detection. Fluorescence intensity in D₂O based mobile phase of doxorubicin had increased about 2.87 times. An advantage of capillary electrophoresis compared to HPLC is the lower consumption of background electrolyte due to small volume of capillary. We achieved the separation of daunorubicin and epirubicin using capillary electrophoresis coupled with laser induced fluorescence detection (Ar⁺ laser 488 nm). The separation was carried out using borate buffer (105 mM, pH 9.0) prepared in H₂O and D₂O. We obtained 2.3 times larger peak area in buffer prepared in D₂O. Changing normal water for deuterium water is a simple way to increase LODs in separation of anthracyclines, drugs used in cancer chemotherapy, by HPLC or CE coupled with fluorescence detection.