Synthesis and photophysical properties of indolenine styrylcyanine dye and its carboxyl-labeled derivative

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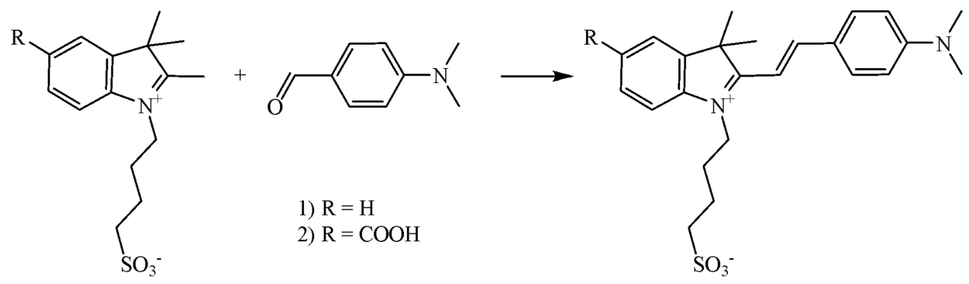
Aim

Organic fluorescent dyes are widely used as a working medium in dye lasers, in analytical chemistry to determine the various trace elements, in photodynamic therapy, tissue optics, and analysis of cells. Here, two indolenine-based styrylcyanine dyes functionalized by the carboxyl group at the indolenine ring were synthesized and characterized as potential probes for biological application.

Chemical synthesis

The indolenine styrylcyanine dyes were synthesized using condensation of the corresponding indolenines with 4-(dimethylamino)benzaldehyde in the presence of an alkaline catalyst.



The functionalization of the dye structure by the carboxyl group leads to a bathochromic shift of fluorescence emission spectra while having no impact on dye specificity. It is established that studied styrylcyanines exhibit no or low preference towards biomacromolecules

Results

1. The spectral-luminescent properties

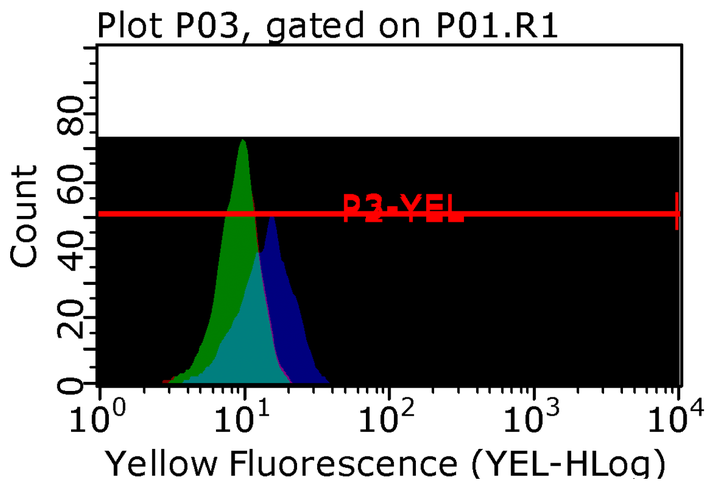
The spectral-luminescent properties of studied dyes in organic solvents, the aqueous solution (buffer pH 7.9), and the presence of different biomacromolecules were investigated.

	buffer				dsDNA				RNA			
	λ_{ex}, nm	λ_{em}, nm	I_0, a.u.	ΔS, nm	λ_{ex}, nm	λ_{em}, nm	I^{DNA}, a.u.	ΔS, nm	λ_{ex}, nm	λ_{em}, nm	I^{RNA}, a.u.	ΔS, nm
Str(1)	550	592	20	42	551	593	24	42	553	593	23	40
Str(2)	561	599	15	38	561	598	15	37	561	597	16	36

	BSA			OVA			BLG			fBLG		
	λ_{ex}, nm	λ_{em}, nm	I^{BSA}, a.u.	λ_{ex}, nm	λ_{em}, nm	I^{OVA}, a.u.	λ_{ex}, nm	λ_{em}, nm	I^{BLG}, a.u.	λ_{ex}, nm	λ_{em}, nm	I^{fBLG}, a.u.
Str(1)	555	593	25	550	592	20	550	592	18	549	592	15
Str(2)	562	598	15	561	599	15	562	599	15	562	599	14

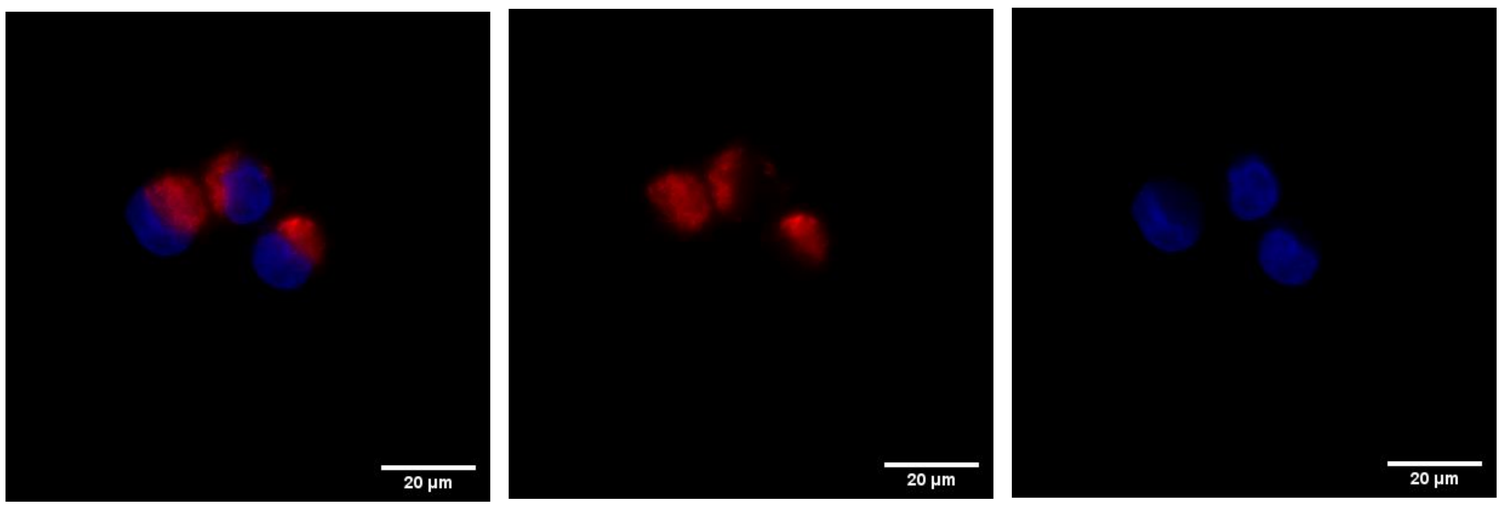
2. Flow cytometry

Cell fluorescence counting by flow cytometry was used to study the ability of dye Str(1) and Str(2) to penetrate the cell membrane. Both dyes were studied at concentrations of 1µM (bottom) and 10µM (top), but higher concentrations decreased the number of cells, which may indicate the toxicity of dyes at high concentrations.



3. Fluorescence microscopy

The human ovarian cancer A2780 cell line was used to study the distribution of the dyes Str(1) and Str(2) in cells with the blue fluorescent standard dye Hoechst binding to nuclear DNA for the co-staining. There is a similar result as in flow cytometry, with the same microscope settings, the signal of the dye Str(2) is almost not observed, while the dye Str(1) is visualized in cells.



Conclusion

- We have shown that studied dyes penetrated the living cells, apparently displaying cytoplasmic staining.
- We can conclude that studied styrylcyanines exhibit no or low preference towards biomacromolecules and no accumulation at any specific site within cells.

Acknowledgements

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