

Liquid-chromatographic product purification of highly blue-emitting carbon dots (CDs)

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Carbon dots (CDs) are fascinating fluorescent particles < 10 nm, which combine high fluorescence quantum yields (QY) and good biocompatibility and can be synthesized by cheap, green and simple techniques. In literature it was already proposed that CDs consist of a carbon core and highly fluorescent fluorophores which are supposed to be attached to the surface of the CDs.

As there are still a lot of debates about the exact structure of the CDs and the composition of the synthesized CD solution, an efficient process for separation of CDs from by-products is required for deeper investigations of the CD's structure. We used isocratic glass column chromatography in previous work to separate the diverse product fractions. By measuring the optical and structural properties of the product fractions by several techniques, we were yet able to show that the CD solutions contain a series of subsets of fluorescent molecules and particles. [1]

For more precise separation of the different product species we applied step gradient elution and transferred the separation process to normal-phase High Performance Liquid Chromatography (HPLC). Thus, we were able to improve the separation quality significantly and could associate the product fractions of a step-gradient HPLC elution with hydrophobic carbon cores, free molecular fluorophores and fluorophore polymers or carbon dots with linked fluorophores.

As some product fractions still consist of numerous different species we started investigations on temperature and pH dependency of the chromatographic process and it became clear that also temperature gradients and lower pH values can contribute to successful purification.

[1]: V. Hinterberger, C. Damm, P. Haines, D.M. Guldi, W. Peukert, Purification and structural elucidation of carbon dots by column chromatography, *Nanoscale* 11 (17) (2019), 8464-8474.