



# Biosynthesis of cadmium sulfide nanoparticles by cultivating yeast in a fed-batch process



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## Introduction:

Inorganic cadmium sulfide (CdS) nanoparticles show fluorescence and exhibit semiconductor properties, which make them an attractive object for studies and application in microelectronics and nanotechnology (figure 1).



Fig. 1 Size-dependent photoluminescent properties of semiconductor cadmium quantum dots (Q dots) [1]

A promising new dimension in the field of biotechnology is the use of microorganisms for the production of inorganic nanoscale particles. The formation of CdS nanoparticles by the yeast *Schizosaccharomyces pombe* is a result of a specific detoxification mechanism (figure 2), activated by sub-lethal cadmium concentrations in the media [2].

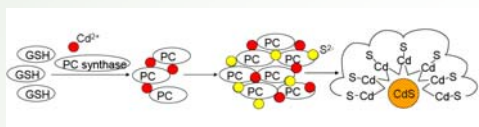


Fig. 2 CdS nanoparticle biosynthetic mechanism involving molecules glutathione (GSH) and phytochelatin (PC)

The final CdS nanoparticle yield of a single cultivation is determined by the final biomass concentration, and the cadmium saturation level, that limits the amount of intracellular CdS nanoparticles in every cell.

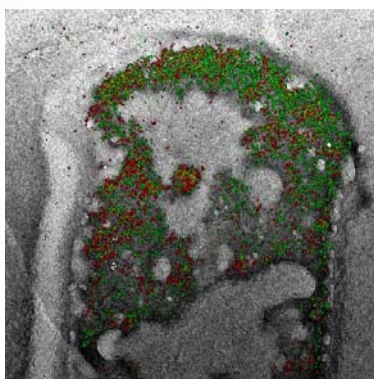


Fig. 3 EFTEM-ESI image of cadmium (red) and sulfur (green) distribution inside *S. pombe* cell

The aim of this scientific work was to develop an appropriate biosynthetic procedure and purification protocol for the production of technically relevant amounts of peptide-stabilized CdS nanoparticles.

## Results and discussion:

*S. pombe* was cultivated in a bioreactor under exponential fed batch conditions (figure 4) to avoid ethanol production during the Crabtree effect and to maximize biomass yield.

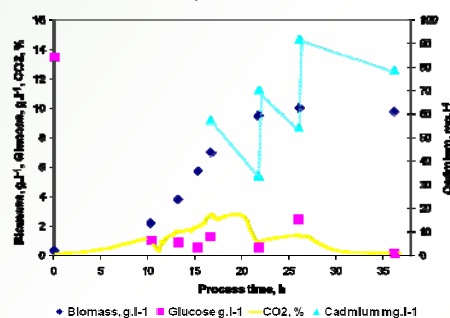


Fig. 4 Fed batch cultivation of *S. pombe* with three-step introduction of cadmium

The cadmium ions as nanoparticle precursors were added according to an optimized strategy for cadmium introduction in three steps at equal six-hour intervals resulting in considerably increased amounts of accumulated heavy metal of up to 19 mg Cd·g<sup>-1</sup> dry biomass [3]. This gave the opportunity for a higher CdS nanoparticle yield.

The intracellular origin of the nanoparticles (figure 3) requires an effective cell disruption technique (FastPrep<sup>®</sup> disintegration kit). The disrupted cell content was subjected to separation and purification by means of Size Exclusion Chromatography (SEC) (figure 5).

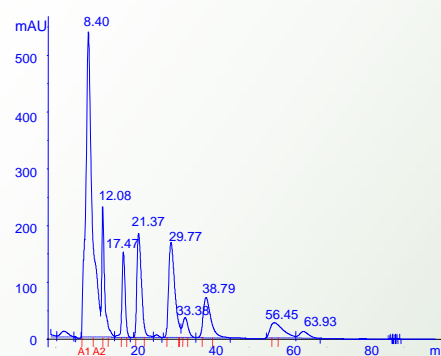


Fig. 5 Fractionation of supernatant after cell disruption by means of SEC

The purification protocol developed resulted in isolation of the whole cadmium amount in the sample within two fractions – A1 and A2 (figure 5).

Mass spectrometric analysis of these fractions revealed presence of peptides PC2 and PC3 (figure 6), intrinsic for the CdS bionanoparticles. These peptides were detected only in the two cadmium containing fractions.

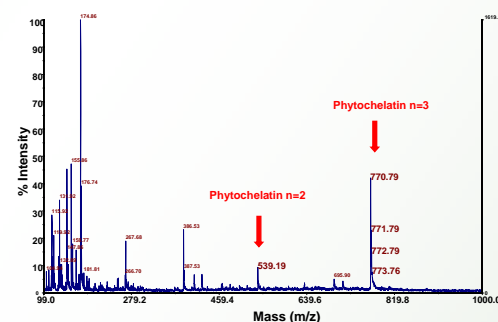


Fig. 6 Electro spray ionisation time of flight (ESI TOF) spectrum of cadmium containing fraction after SEC

The presence of CdS nanoparticles in both fractions A1 and A2 was further demonstrated by photoluminescent analysis (figure 7).

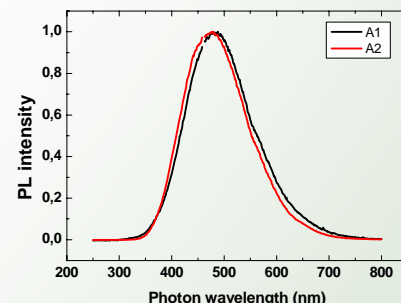


Fig. 7 Fluorescence spectrum of fractions, containing CdS, after SEC excited at 229 nm

The emission maximum at 480 nm together with the absence of emission shift towards higher wavelengths is characteristic for CdS quantum dots and analogous to results reported by other authors [4] or product specification from Lumidot<sup>™</sup> (Sigma-Aldrich) with 2.5 nm core size.

## Conclusions:

The results prove that the cultivation of *S. pombe* under specified conditions and the purification protocol developed is a promising pathway for CdS nanoparticles synthesis. In two isolated fractions all Cd was recovered and the presence of nanoparticles with Q dots properties and PC layer was successfully verified.

## Literature:

- [1] M. Han, X. Gao, J. Z. Su, S. Nie, *Nature Biotech.* 19, 631-635, 2001
- [2] C.T. Dameron, D.R. Winge, *Trends Biotechnol.* 8, 3-6, 1990
- [3] N. Krumov, S. Oder, I. Perner-Nochta, A. Angelov, C. Posten, *J. Biotechnol.*, 132, 481-486, 2007
- [4] Bae, Mehra; *J.Inorg.Biochem.* 69 (1998) 33-43