

# Accumulation of Cd in the Early Stages of the Development of Rainbow Trout *Oncorhynchus mykiss* Exposed to Cd-Based Quantum Dots and Cd Salt

Živilė Cibulskaitė<sup>1</sup>, Nijolė Kazlauskienė<sup>1</sup>, Kęstutis Jokšas<sup>2,3</sup>, Vytautas Kulvietis<sup>4</sup>, Tomas Makaras<sup>1</sup>, Danguolė Montvydienė<sup>1</sup>, Mantas Stankevičius<sup>4</sup>, Ričardas Rotomskis<sup>4,5</sup>

<sup>1</sup>*Institute of Ecology of Nature Research Centre, Akademijos st. 2, LT-08412 Vilnius, Lithuania*

<sup>2</sup>*Geology and Geography Institute of Nature Research Centre, Akademijos st. 2, LT-08412 Vilnius, Lithuania*

<sup>3</sup>*Vilnius University, Faculty of Chemistry and Geosciences, Naugarduko st. 24, LT-03225 Vilnius, Lithuania*

<sup>4</sup>*Laboratory of Biomedical Physics, National Cancer Institute, Baublio st. 3b, LT-08660 Vilnius, Lithuania*

<sup>5</sup>*Biophotonics Group of Laser Research Center, Vilnius University, Saulėtekio ave. 9, LT-10222 Vilnius, Lithuania*

E-mails: <sup>1</sup>[zivile.cibulskaitė@gmail.com](mailto:zivile.cibulskaitė@gmail.com) (corresponding author)

**Abstract.** The main aims of the present study were: 1) to determine the concentration of Cd in water during the experiment after long-term exposure to rainbow trout embryos and larvae to sublethal concentrations of Cd-based quantum dots (QDs) and Cd chloride salt, and 2) to evaluate accumulation of Cd in the whole body of test-organisms depending on the type of chemical substances (QDs or single Cd) and the duration of exposure. Experimental studies at the early developmental stages of rainbow trout were performed under static conditions. Cd concentration in water during the experiment was significantly higher in QD solution than in CdCl<sub>2</sub> solution, and it declined over time. A comparative analysis indicated that the Cd accumulation in test-organisms from QDs was mostly higher than that from CdCl<sub>2</sub>. The accumulation of Cd from QDs tends to increase with increasing duration of exposure. The bioconcentration factor for Cd increased in embryos and larvae over time until reaching the maximum at the end of the QD exposition.

**Keywords:** Cadmium; nanoparticles; fish; embryos; larvae; accumulation.

**Conference topic:** Environmental protection.

## Introduction

Nanoparticles have unique and specific physicochemical and surface properties, which are related to their size, i.e., mechanical resistance, electronic properties, thermal conductivity, and chemical reactivity (Auffan *et al.* 2009). One type of nanoparticles which have an interest in the last years is quantum dots (QDs). QDs are colloidal nanostructured materials composed of a semiconductor core (e.g., CdSe, CdTe) that can be encapsulated in a shell (e.g., CdS) which enhances optical and electronic properties and reduces core metal leaching (Domingos *et al.* 2011; Zarco-Fernández *et al.* 2016). QDs could have organic coatings that increase their dispersion in water and help direct them to biologic targets (Medintz *et al.* 2005). QDs are now in diverse commercial use such as in optical and electronic devices, biological labeling, photovoltaic, catalysis, light-emitting diodes, and photodynamic cancer therapies (Juzenas *et al.* 2008; Rzigalinski, Strobl 2009).

Like other types of nanoparticles, QDs may eventually find their way into the environment (Domingos *et al.* 2011). Consequently, the increasing release of nanoparticles into the environment has influenced the development of new research about their potential environmental effects (Domingos *et al.* 2011). Current literature reveals that the fate of nanoparticles compared to their dissolved components is an area of importance being rather complicated. QD potential toxicity to aquatic organisms could be caused by the nanoparticle itself or by its free metallic elements generated during the degradation process (Zarco-Fernández *et al.* 2016).

Cadmium (Cd) is a nonessential metallic trace element widely distributed in the aquatic environment (Annabi *et al.* 2013; Pereira *et al.* 2015). In addition, Cd is an extremely toxic metal and its amount has increased over past decades due to its widespread industrial use and also nanotechnology uses (Barjhoux *et al.* 2016). Several field studies demonstrated that Cd contamination could persist for many years in the aquatic environment because of its storage in sediments and its further release into the water column under favorable hydrodynamic conditions (Coynel *et al.* 2007). This fact could be a consequence in a long-term Cd accumulation within the bodies of organisms (Baudrimont *et al.* 2005). The accumulation of a chemical in a sensitive organism is often a good integrative indicator of the environmental contamination, including bioconcentration and biomagnification (Fabrega *et al.* 2011; Lee, An 2015).

Cd exerts a wide range of pathological effects on fish and other aquatic organisms (Al-Asgah *et al.* 2015). Moreover, there are many studies which evaluated the potential toxicity of QDs to fish (Duan *et al.* 2013; Yong *et al.*

2013; King-Heiden *et al.* 2009; Leigh *et al.* 2012; Lewinski *et al.* 2011). QDs could be transferred from protozoa (*A. longa*) to zooplankton (*M. macrocopa*) and to fish (*D. rerio*) (Lee, An 2015).

It has been demonstrated that the feed is the main source of Cd accumulation in tissues (Guinot *et al.* 2012). Furthermore, some studies have shown that QDs degraded in acidic or alkaline environments (Zhang *et al.* 2008), thus accumulation of QDs in the acid environment of the fish intestine (Wiecinski *et al.* 2009) may lead to QD degradation which could result in the release and subsequent absorption of Cd and Se by the fish (Leigh *et al.* 2012). Cd accumulation in the organisms depends on the concentration, route of absorption, environmental conditions and other intrinsic factors (Bowen *et al.* 2006; Guinot *et al.* 2012; Karakoç, Dinçer 2003; Jezierska, Witeska 2006). Meanwhile, accumulation and toxicity of a nanoparticle is based on its composition, size and surface chemistry (Zarco-Fernández *et al.* 2016).

Given the high potential for toxicity of Cd, it is necessary to detect and compare Cd accumulation in organisms from the nanoparticles and salts (Domingos *et al.* 2011). Quantitative information on the bioavailability of Cd to early developmental stages of fish is scarce (Annabi *et al.* 2013). It has been shown that the Cd concentration of fish eggs increased with time and depended of Cd concentration in water (Annabi *et al.* 2013). Hatched larvae are more susceptible to Cd than unhatched embryos are (Hallare *et al.* 2005). A study by Zarco-Fernández *et al.* (2016) demonstrated that Cd from the Cd salts and from the CdSe/ZnS QDs was located in different areas of zebrafish larvae.

Many studies focused on embryonic toxicity induced by nanoparticles rather than assessing the accumulation in embryos and larvae. However, data about the levels of Cd accumulation in early life stages of fish and possible Cd leakage from the QD structure are scarce. King-Heiden *et al.* (2009) demonstrated that metallothionein expression could be useful as a marker of internal Cd exposure, thus providing indirect information on *in vivo* QD degradation: QD did not completely degrade *in vivo*, and some endpoints of toxicity to zebrafish larvae were unrelated to Cd release. Therefore, more studies are needed to better understand the accumulation of Cd from QDs in early life stages of fish.

The main aims of the present study were: 1) to determine the concentration of Cd in water during the experiment after long-term exposure to rainbow trout embryos and larvae to sublethal concentrations of Cd-based quantum dots (QD) (CdSe/ZnS-COOH) and Cd chloride salt (CdCl<sub>2</sub>·H<sub>2</sub>O), and 2) to evaluate accumulation of Cd in the whole body of test-organisms depending on the type of chemical substances (QD or single Cd) and the duration of exposure.

## Materials and methods

Exposure of fish was performed at the Laboratory of Ecology and Physiology of Hydrobionts (Nature Research Centre, Lithuania). Rainbow trout *Oncorhynchus mykiss* (Walbaum, 1792) eggs (eyed-egg stage embryos) were obtained from the Simnas hatchery (Lithuania). Studies were carried out with non-protected life stages in accordance with EU Directive 2010/63/EU. The first test duration was 8 days: starting from eyed-egg stage embryos 4 days before hatching and continued 4 days after hatching and including the hatching period. The duration of the second test performed was 10 days: starting from 1-day-old larvae. The laboratory treatment was carried out in a climatic camera (Bronson PGC-660, Germany) in glass beakers under static conditions according to ISO 7346-1:1996. The dead embryos and larvae were removed daily from the groups.

The Cd accumulation in the whole body of embryos and larvae (10 individuals per replicate) and Cd concentration in water during experiments were analyzed in three replications. Sampled organisms were dried on absorbent paper and weighted before being stored at -18 °C until Cd analysis.

Deep-well water of high quality was used for storing control embryos (Svecevičius 2010). The average hardness of dilution water was approximately 284 mg/L as CaCO<sub>3</sub>, alkalinity was 244 mg/L as HCO<sub>3</sub><sup>-</sup>, the mean pH was 8.0, temperature was maintained at 10±0.5 °C, and the oxygen concentration ranged from 8 to 10 mg/L. Dissolved oxygen in the tanks, temperature and pH were measured routinely with a hand-held multi-meter (WTW Multi 340i/SET, Germany).

A water-soluble, red-emitting semiconductor QD [Qdot® ITK™ (Life Technologies, cat. No. A10200)] at a concentration of 4×10<sup>-9</sup> M was used during the accumulation experiments. The QD was made of a core (CdSe) covered with an additional semiconductor layer (ZnS) and with an amphiphilic polymer (methoxypolyethylene glycol, mPEG-COO<sup>-</sup>) coating, and the surface ligands were terminated with carboxyl groups (negatively charged). This QD had a narrow, symmetric emission band with emission maxima at 625 nm. The concentration of the QD was chosen according to the study which showed that CdSe-ZnS (core-shell structure) LC50 values were in the 0.7–4.2×10<sup>-7</sup> M range for zebrafish (Yong *et al.* 2013). A volume of 100 µL of stock solution of 8 µM of the QD was dissolved in the fish media to achieve the final QD concentration of 4×10<sup>-9</sup> M in embryos and larvae incubation solvent before the experiments (Cibulskaitė *et al.* 2016).

Reagent grade cadmium chloride (CdCl<sub>2</sub>·H<sub>2</sub>O) («REACHIM» Company, Russia) was used as a toxicant and stock solutions were prepared by dissolving a necessary amount of salts in distilled water. The concentration of 2 µg Cd/L was chosen according to the 96-hour LC50 for rainbow trout larvae (Kazlauskienė *et al.* 2016).

Cd concentrations were evaluated in the Laboratory of Geoenvironmental Research (Nature Research Centre, Lithuania). For Cd analysis in embryos, the digestion method was used (Thomas, Mohaideen 2015). The content of Cd in the tanks and in the whole body of the fish embryos and larvae was analyzed by an atomic absorption

spectrophotometer SHIMADZU AA-7000 (Japan) with a graphite furnace atomizer GFA-7000 and auto-sampler ASC-7000 (measured wavelength 185 to 900±0.3 nm, high-speed deuterium lamp 185 to 430 nm, heating temperature range 50 to 3000 °C, repeatability 2.5%) according to the analysis method LST EN ISO 15586:2004. Reference materials: Cd standard for AAS 1000 mg/L. Sigma catalog No. 51994. Lot. No. BCBN4966V. Cd detection limit = 0.3 µg/L.

The accumulation of heavy metals in aquatic organisms is an important factor in the estimation of the potential hazards of chemicals to the environment (Tokhun *et al.* 2014). The bioconcentration factor (BCF), defined as the net result of the adsorption, distribution and elimination of chemicals in the aquatic organism after exposure via water, was calculated as a ratio of metal concentration in the organism to the metal concentration in water at equilibrium (Tokhun *et al.* 2014; Zarco-Fernández *et al.* 2016). The results were presented as µg Cd/g wet weight. BCF was determined in the laboratory using the test fish embryos/larvae and was defined as:

$$BCF = \frac{\text{Concentration in organism}(\mu\text{g} / \text{g ww})}{\text{Concentration in water}(\mu\text{g} / \text{L})}$$

Differences between the evaluated characteristics studied were tested by two-way ANOVA at  $p < 0.05$  using Statistica 7.0 software (USA).

## Results and discussion

### *Cd concentration (µg/L) in water during experiment*

Designed nominal metal concentrations in the tanks were checked during blank tests (without embryos/larvae) with an atomic absorption spectrophotometer. Mean measured Cd concentrations were 2659 µg Cd/L in the QD solution (Cd concentration was  $1 \times 10^{-6}$  M) and 1.71 µg Cd/L in the CdCl<sub>2</sub> solution (Cd concentration was  $1.8 \times 10^{-8}$  M).

Table 1. Cd concentration (µg/L) in water during experiments after rainbow trout embryos (after 1 day of exposure at the first test) and larvae (after 8 days of exposure at the first test and after 7 and 10 days of exposure at the second test) were exposed to QDs and CdCl<sub>2</sub>. The data represents the mean Cd concentration(µg/L) (±SD) for three replicates

First test (Cd µg/L)				
Exposure days	Development stages of rainbow trout	Control	QD	Cd
4	Embryos	0.347±0.027	92.400±49.790 <sup>abc</sup>	1.651±0.566 <sup>ac</sup>
8	Larvae	0.367±0.010	9.270±6.249	0.980±0.104
Second test (Cd µg/L)				
7	Larvae	0.326±0.046	142.133±44.363 <sup>abd</sup>	1.360±0.115 <sup>a</sup>
10	Larvae	0.300±0.000	30.620±16.372	0.949±0.218

<sup>a</sup> Significant difference from the control ( $p < 0.05$ ). <sup>b</sup> Significant difference between the type of chemical substances. Significant difference between the duration of exposure ( $p < 0.05$ ): <sup>c</sup> significant difference from 8 days of exposure; <sup>d</sup> significant difference from 10 days of exposure

Changes of Cd concentrations in water during experiments are shown in Table 1. Cd concentration in the deep-well water from the control was low (did not exceed 0.367±0.010 µg Cd/L) and remained stable throughout the exposure period (Table 1). QDs and CdCl<sub>2</sub> induced a statistically significant ( $p < 0.05$ ) increase in Cd concentrations in water during experiments only at the beginning of both tests (after 4 and 7 days of exposure) in comparison with the control. Cd concentration measurement in water during experiments revealed concentration decrease throughout the exposure period (Table 1). Cd concentration in the QD solution significantly decreased during both tests. Meanwhile, Cd concentration in the CdCl<sub>2</sub> solution significantly decreased only in the first test (after 4 and 8 days of exposure). The mean Cd concentration in water during experiments was significantly higher at QD solution than at CdCl<sub>2</sub> solution after 4 days of exposure at the first test and after 7 days of exposure at the second test, and it mostly declined over time for both treatments (Table 1).

The stated nanoparticle concentration is frequently a dilemma since different studies show nanoparticle concentration by employing inconsistent units or not paying special attention to this parameter (Zarco-Fernández *et al.* 2016). However, to facilitate comparison between study results, it is recommended to estimate QD elemental composition (Tsoi *et al.* 2013; Zarco-Fernández *et al.* 2016). Previous studies on QD toxicity have determined correlation between their chemical stability and toxicity. According to Domingos *et al.* (2011) and King-Heiden *et al.* (2009), the Cd salts and the Cd-based QDs had different biological effects. In zebrafish larvae, Cd salts induced endpoints of sublethal toxicity, including reduced growth, ocular, pericardial, submandibular and yolk sac edema (swelling), and increased axial curvature (bent spine). Meanwhile, zebrafish larvae exposed to CdSe/ZnS QDs

demonstrated endpoints of toxicity related to Cd and endpoints unrelated to Cd, such as apparent necrosis, yolk sac malformation, and malformed tail (King-Heiden *et al.* 2009).

#### Cd accumulation ( $\mu\text{g/g}$ ) in embryos and larvae

The significant ( $p < 0.05$ ) values of Cd concentration in rainbow trout embryos and larvae after 1–10 days of exposure are presented in Table 2. The maximum value of accumulated Cd was found in embryos exposed to QDs after 1 day of exposure ( $3.216 \pm 0.300 \mu\text{g/g}$ ) and in larvae after 4 days of exposure ( $1.189 \pm 0.760 \mu\text{g/g}$ ). Additionally, the maximum value of accumulated Cd was found in embryos exposed to  $\text{CdCl}_2$  after 4 days of exposure ( $0.092 \pm 0.010 \mu\text{g/g}$ ) and in larvae after 4 days of exposure ( $0.025 \pm 0.016 \mu\text{g/g}$ ).

Table 2. Cd accumulation ( $\mu\text{g/g}$ ) in rainbow trout embryos (after 1 and 4 days of exposure at the first test) and larvae (after 8 days of exposure at the first test and after 4, 7 and 10 days of exposure at the second test) exposed to QD and  $\text{CdCl}_2$ . The data represents the mean Cd accumulation ( $\mu\text{g/g}$ ) ( $\pm$ SD) for three replicates

First test (Cd $\mu\text{g/g}$ )				
Exposure days	Development stages of rainbow trout	Control	QD	Cd
1	Embryos	0.002 $\pm$ 0.001	3.216 $\pm$ 0.300 <sup>abcd</sup>	0.068 $\pm$ 0.020 <sup>ad</sup>
4	Embryos	0.004 $\pm$ 0.001	0.026 $\pm$ 0.006	0.092 $\pm$ 0.010 <sup>ad</sup>
8	Larvae	0.008 $\pm$ 0.004	0.134 $\pm$ 0.014	0.017 $\pm$ 0.005
Second test (Cd $\mu\text{g/g}$ )				
4	Larvae	0.007 $\pm$ 0.004	1.189 $\pm$ 0.760 <sup>ab</sup>	0.025 $\pm$ 0.016
7	Larvae	0.009 $\pm$ 0.004	0.961 $\pm$ 0.609 <sup>a</sup>	0.020 $\pm$ 0.009
10	Larvae	0.003 $\pm$ 0.001	1.302 $\pm$ 0.272 <sup>ab</sup>	0.011 $\pm$ 0.006

<sup>a</sup> Significant difference from the control ( $p < 0.05$ ). <sup>b</sup> Significant difference between the type of chemical substances. Significant difference between the duration of exposure ( $p < 0.05$ ): <sup>c</sup> significant difference from 4 days of exposure; <sup>d</sup> significant difference from 8 days of exposure

The QDs induced a significant increase in Cd accumulation in embryos after 1 day of exposure at the first test and in larvae after 4, 7 and 10 days of exposure at the second test in comparison with the control. The same tendency was observed in Cd accumulation in embryos after 1 and 4 days of exposure to  $\text{CdCl}_2$  at the first test (Table 2). However,  $\text{CdCl}_2$  did not induce a significant change ( $p < 0.05$ ) in Cd accumulation in larvae after 4, 7 and 10 days of exposure at the second test in comparison with the control (Table 2).

Obtained results showed that Cd accumulation in embryos and larvae depended on the type of the chemical substance. A significant ( $p < 0.05$ ) difference was found in the Cd accumulation in embryos between chemical substances (QD,  $\text{CdCl}_2$ ) after 1 day of exposure at the first test and in larvae after 4 and 10 days of exposure at the second test (Table 3).

It was found that accumulation of Cd in embryos and larvae exposed to QDs depended on the duration of exposure only at the first test. Cd accumulation in embryos after 1-day exposure to QDs was significantly ( $p < 0.05$ ) higher compared to embryos after 4 days and larvae after 8 days of exposure. Furthermore, the Cd accumulation in embryos and larvae exposed to  $\text{CdCl}_2$  significantly ( $p < 0.05$ ) differed during the first test and depended on the duration of exposure. Cd accumulation in embryos after 1 day of exposure to  $\text{CdCl}_2$  was significantly ( $p < 0.05$ ) higher compared to Cd accumulation in larvae after 8-day exposure. Zarco-Fernández *et al.* (2016) also noticed the differences in Cd accumulation in zebrafish larvae exposed to  $\text{CdSe/ZnS}$  QDs and Cd salt. QDs were not assimilated in the organism, but QDs distributed along the zebrafish larvae surface and the Cd from the Cd salts was located mainly in zebrafish eyes area (Zarco-Fernández *et al.* 2016). The results obtained from Lewinski *et al.* (2011) and Duan *et al.* (2013) suggested that modified QDs could accumulate in the intestines of zebrafish. Duan *et al.* (2013) demonstrated that CdTe QDs could be accumulated in the larvae intestine region rather than in the head or tail region after 96 hours of exposure.

Cd accumulation values in hatched larvae after 8 days of exposure to QDs at the first test were significantly lower than in embryos after 1 day of exposure ( $0.026 \pm 0.006$  and  $3.216 \pm 0.300 \mu\text{g/g}$ , respectively) (Table 2). The decrease of Cd accumulation in larvae exposed to QDs could be the consequence of the loss of QDs bound to the chorion occurring at hatching. Several studies have previously reported that more than 94% of the total accumulated Cd from salt was located on the chorion of *O. latipes* embryos exposed to 10 mg Cd/L (Michibata 1981) and 93 % more of Cd were in larvae than in embryos exposed to 2 and 20  $\mu\text{g}$  Cd/L (Barjhoux *et al.* 2016). According to Meteyer *et al.* (1988), negatively charged mucopolysaccharides and glutamic acid in the chorion could retain and limit Cd uptake by the embryo. Beattie and Pascoe (1978) noticed that in rainbow trout 98% of total Cd was retained by chorion; however, Cd retained by chorion in zebrafish reached only 61 % (Burnison *et al.* 2006).

### Cd bioconcentration factor (BCF)

Cd bioconcentration factor (BCF) values in the embryos and larvae of rainbow trout under exposure to QDs and CdCl<sub>2</sub> are presented in Figure 1. The estimated BCF value of Cd was significantly higher ( $p < 0.05$ ) in embryos exposed to CdCl<sub>2</sub> (0.059±0.014) compared to BCF value of Cd in embryos exposed to QDs (0.0001±0.00). It was found that the BCF value of Cd in embryos and larvae significantly ( $p < 0.05$ ) depended on the duration of exposure to QDs and CdCl<sub>2</sub>. QD induced a significant increase in the value of BCF in larvae during 10 days at the second test. However, QDs did not cause a significant ( $p < 0.05$ ) increase in BCF in embryos and larvae during 8 days at the first test. Meanwhile, CdCl<sub>2</sub> induced a significant decrease in BCF in embryos and larvae at the first test. The BCF value measured for the rainbow larvae was not statistically different at the second test, indicating that BCF for Cd remained stable over time.

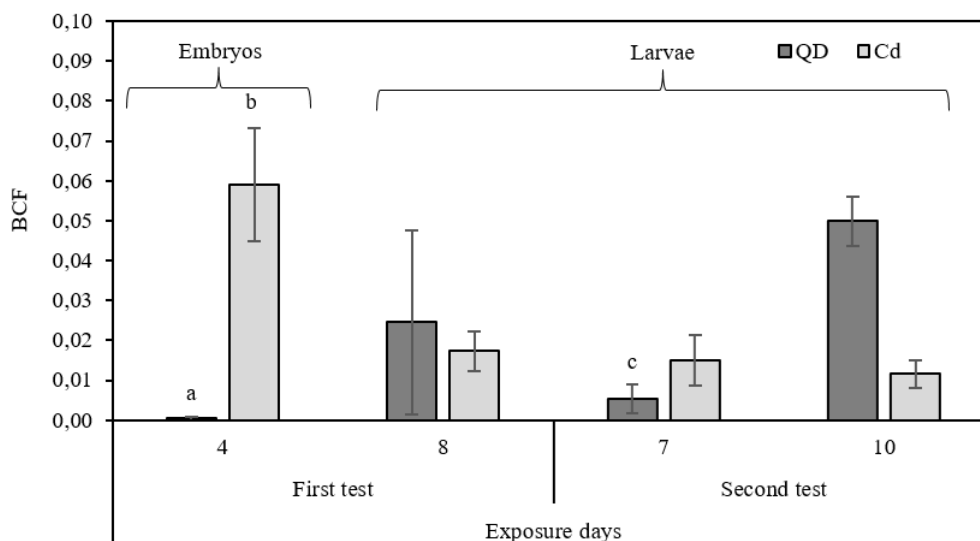


Fig. 1. Cd bioconcentration factor (BCF) values in rainbow trout embryos (after 4 days of exposure at the first test) and larvae (after 8 days of exposure at the first test and after 7 and 10 days at the second test) exposed to QD and CdCl<sub>2</sub>.<sup>a</sup>Significant difference between the type of chemical substances ( $p < 0.05$ ). Significant difference between the duration of exposure ( $p < 0.05$ ): <sup>b</sup>significant difference from 8 days of exposure (the first test); <sup>c</sup> significant difference from 10 days of exposure (the second test)

Results of the long-term tests demonstrated that the QDs and CdCl<sub>2</sub> tested for this study had different BCF values in rainbow trout embryos and larvae. It seems that the BCF values of Cd in test-organisms exposed to QDs increased with exposure time in both tests until reaching the maximum at the end of both tests. According to Zarco-Fernández *et al.* (2016) and Matz *et al.* (2007), zebrafish larvae had a slow elimination rate and did not significantly reduce their Cd body burden, because the absorbed Cd was highly bound to tissues and the accumulation occurred in a dose-dependent manner. Zarco-Fernández *et al.* (2016) found that the BCF in zebrafish larvae was much higher for Cd salt than for QDs after 48 hours of exposure. Other similar studies by King-Heiden *et al.* (2009) showed low accumulation values of Cd in zebrafish larvae exposed to 0.2–20 μM Cd-eq of QDs following 120-hour exposure. In addition, some authors suggested lower accumulation of QDs in fish than Cd accumulation from its salt, indicating a higher bioavailability of QDs over time (Rocha *et al.* 2014; Zarco-Fernández *et al.* 2016). It is in good agreement with our findings that the BCF values of Cd in early stages of development of fish exposed to QDs reached the maximum at the end of both tests.

In order to assess the environmental risk of the newly created nanoparticles, it is necessary to establish their fate in the aquatic environment and to evaluate their quantities and effects of both dissolution products and the intact QDs (Domingos *et al.* 2011). QDs, such as those used in medical imaging, disease treatment and targeted therapy are known to release toxic Cd from the QD core (Leigh *et al.* 2012). Therefore, accumulation and toxicity researches of nanoparticles are fundamental to assess their risk to the environment and human health (López-Serrano *et al.* 2014). In this study, rainbow trout embryos and larvae were exposed to either a soluble Cd salt or QDs at sublethal concentrations. To the best of the authors' knowledge, this is the first study to evaluate the accumulation as well as the Cd concentration in water during the experiment after rainbow trout embryos and larvae were exposed to QDs and Cd singly for a long-term test. Taking Cd measurement as an indicator of evaluating QD accumulation will be more beneficial and comprehensive for the environment safety assessment and biomedical application.

## Conclusions

1. The results of the long-term experiments (8 and 10 days) on rainbow trout at early stages of development showed that the value of Cd concentration in water during the experiment was significantly higher in Cd-based QD solution than in CdCl<sub>2</sub> solution; however, it declined over time in both treatments.
2. The significant ( $p < 0.05$ ) differences between the values of accumulated Cd in embryos and larvae after 1–10 days of exposure were observed, and they depended on the type of chemical substances and duration of exposure. The QDs induced a significant increase in Cd accumulation in embryos and larvae in comparison with the control. However, CdCl<sub>2</sub> did not cause relevant differences ( $p < 0.05$ ) in Cd accumulation in affected larvae in comparison to the control. The Cd accumulation in test-organisms exposed to QDs was mostly higher than in organisms exposed to CdCl<sub>2</sub>.
3. The estimated BCF value of Cd in test-organisms was significantly higher ( $p < 0.05$ ) in embryos exposed to CdCl<sub>2</sub> ( $0.059 \pm 0.014$ ) compared to the BCF value for Cd in embryos exposed to QDs ( $0.0001 \pm 0.00$ ) after 4 days of exposure. QDs induced a significant increase in BCF for Cd in larvae after 10 days of exposure. Meanwhile, BCF for Cd in the rainbow trout larvae did not statistically differ compared with larvae exposed to CdCl<sub>2</sub>.
4. Obtained data suggest that Cd could be a valuable and important indicator for evaluating Cd-based QD accumulation in aquatic organisms and for estimating the toxicity of QDs to organisms, as well as for a safety assessment of a new class of pollutants in freshwater environments.

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## Disclosure statement

The authors declare that they have no conflict of interest.

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