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ORIGINAL ARTICLE

Improving water quality, growth performance, and modulating some stress physiological biomarkers in *Cyprinus carpio* using raw date nuclei as a zinc adsorbent agent

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Abstract

Adsorption of heavy metals by affordable adsorbents has recently become one of the most often applied method for removing these metals from contaminated water. The purpose of this study was to evaluate the efficacy of using raw date nuclei (RDN) as a natural and inexpensive adsorbent to remove Zn ions from contaminated water and reduce its impact on water quality, Zn bioaccumulation levels in fish organs, growth performance, and some physiological aspects of common carp (*Cyprinus carpio*) under sublethal concentrations. Five experimental groups were examined for eight weeks; CNT, control group rearing in clean water; RDN_0, fish group rearing in Zn-contaminated water; RDN_3, fish group rearing in Zn-contaminated water and treated with 3 g RDN/L; $RDN_{4.5}$, fish group rearing in Zn-contaminated water and treated with 4.5 g RDN/L; RDN_6, fish group rearing in Zn-contaminated water and treated with 6 g RDN/L. Water quality measures were monitored in a continuously manner without removing fish excreta or renewing water. The results showed that sublethal concentration of Zn alone significantly elevated the level of antioxidant biomarkers (SOD, superoxide dismutase; GST, glutathione transferases; GPX, glutathione peroxidase; CAT, catalase and MDA, malondialdehyde), significantly declined fish performance, increased Zn-accumulation in fish organs (gills and flesh) and increased the ammonia secretions in fish ponds compared to the control group. Zn concentrations in RDN-containing aquariums were significantly lower than in RDN-free aquariums, and Zn removal from polluted water was dependent on RDN dosage. Consequently, fish performance and serum bio-indicators enhanced in the presence of RDN in a fish group reared under sublethal Zn concentrations. In conclusion, although Zn caused toxicity in *C. carpio* at sublethal levels, treating Zn-contaminated water with RDN at a dosage of 6 g per liter effectively prevented oxidative stress and the harmful impacts of Zn exposure.

Keywords Adsorption · Bio-indicators · Common carp · Raw date nuclei · Zinc bioaccumulation

Introduction

In accordance with the industrial, agricultural, and urban revolutions, abandoned waste materials that have not been refined have grown increasingly hazardous to the aquatic environment (Ismael et al. 2021). Non-treated wastes entering aquatic habitats have an impact on water quality, and although they induce habitat modification or death in sensitive species, they accumulate in aquatic organisms with a high tolerance and cause metabolic, physiologic, and biochemical disorders (Farag et al. 2021). These non-treated wastes include heavy metals such as Pb, Cd, Hg, Zn, and Cu (Mehana et al. 2020).

Zn is abundant in the natural habitat and is known as a vital trace element for aquatic animals and human beings (Lall and Kaushik 2021). The primary sources of Zn

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pollution in aquatic water were found as tire dust, electroplating, motor oil, hydraulic fluid, and galvanized metal surfaces industries (Sörme and Lagerkvist 2002). When the Zn contamination level exceeds a certain limit, the aquatic creatures may suffer negative consequences (Li et al. 2019). It is obvious that aquatic creatures are significantly more sensitive to Zn concentrations in water than other creatures (Abdelghany et al. 2020). Zn might kill fish at acutely toxic amounts via damaging gill tissues (Oğuztürk et al. 2018). However, at chronically toxic levels, it may cause oxidative stress, which can lead to sudden mortality (Giardina et al. 2009). The toxic effects of Zn depend on its level, water physiochemical properties (pH, total alkalinity, and total hardness), fish species, life history, and it is non-specific (Li et al. 2019).

Molecular indicators of oxidative stress have found widespread use in environmental toxicity processes in aquatic creatures exposed to diverse chemical contaminants (Livingstone 2001). The aquatic environment receives large quantities of environmental pollutants including Zn daily, which can cause oxidative stress in aquatic organisms due to the free radical mechanism (Hao et al. 2013). Consequently, physiological disorders induced in fish related with oxidative stress can be used as biomarkers for environmental pollution (Khafaga et al. 2020; Naiel et al. 2020).

Recently, worldwide organizations are doing their high efforts to identified easy, safe, applicable and inexpensive technologies to purifying contaminated water (Naiel et al. 2022). While, the adsorption process is regarded as the most effective water treatment processes due to the convenience, ease of operation and simplicity of design (Abd El-hameed et al. 2021). Additionally, it can be applied for the removal of soluble and insoluble contaminants and biological pollutants with a removal efficiency of around 99% (Bhatnagar and Minocha 2006).

The raw materials of some agricultural and industrial wastes are increasingly used by many researchers as low-cost adsorbents (Gupta et al. 2009). Nowadays, the biomass of date palms becomes of great importance as adsorbents for the elimination of different types of pollutants because of its low cost and its important adsorption potential (Ahmad et al. 2012; Al-Ghamdi et al. 2013; Feng et al. 2015). Also, date seeds have been shown to have excellent nutritional quality due to the high quantity of minerals, vitamins, fats and proteins (Habib and Ibrahim 2009; Ahmed et al. 2014; Aljaloud et al. 2020). Furthermore, ALrajhi et al. (2019) investigated that the extraction of date seeds by ethyl acetate and hexane has a high antibacterial effect. In addition, it is also eco-friendly, economically feasible and could be readily desirable adsorbent for water treatment.

Owing to the abovementioned information, this experiment proposes that raw date nuclei (RDN) might be used to purify fish rearing water from Zn pollution without the need of any additional chemical treatments as a natural and low-cost adsorbent that farmers could easily use. Thus, the water physiochemical features, growth performance, and physiological alters of common carp (*C. carpio*) grown in Zn-contaminated water were assessed under the treatment with graded levels of RDN. Also, the adsorption capacity and removing efficacy of RDN for the removal of Zn contaminants in fish rearing water were evaluated by measuring Zn accumulation levels in both fish tissue and water.

Materials and methods

Experimental fish and rearing conditions

Two hundred and fifty apparently healthy Common carp, *C. carpio*, fingerlings (initial weight, 3.75 ± 0.21) were obtained from the fish hatchery belonging to the Central Laboratory for Aquaculture Research (CLAR), Abbassa, Abo-Hammad, Sharqia, Egypt. The common carp fingerlings were acclimated for 14 days in an indoor fiberglass tank and fed a commercial diet containing 30% crude protein (CP) until apparent satiation (Table 1) twice a day at 09:00 am and 16:00 pm. At the end of the adaptation period, the fish were randomly divided into five equal groups in 25 glass aquaria. Each group has five replicates, and each aquarium has ten fish. The control group was raised in clean water, while the other four treatments were raised in Zn-polluted water (1/10 LC50 = **0.366** mg/L). The all-polluted treatments were subjected to a series of graded doses of RDN (0.0, 3, 4.5, and 6 g/L). The examined level of RDN was chosen based on the findings of the Shafiq et al. (2018) investigation. During the study period, the crushed RDN was kept in permeable bags and remained unaltered in the aquariums. The water was exchanged on a regular basis, and the metal dosage was blended into a small portion of the water before being expanded into the aquarium.

Acute toxicity test

Zinc sulfate ($ZnSO_4 \cdot 7H_2O$) as a source of Zn was purchased from Sigma (Egyptian International Center for Import, Cairo, Egypt). Acute toxicity test was done using graded levels of Zn (0, 1, 2, 4, 6, 8, and 10 mg/L) as ascribed by Hao et al. (2013). Each level included the allocation of nine fish into a 100 L glass aquarium with three replicates. During the acute toxicity test, each aquarium received continuous air supply via air stone connected with electrical air pump. During the assay, neither the supplied fish nor the specimens were fed, and the test medium was not replenished. The dead fish were recorded at different exposure periods (24 h, 48 h, 72 h, and 96 h) for calculating the mortality rate. The value of LC50 was the metal level that induced 50% mortality in

Table 1 The ingredient and proximate chemical analyses of formulated basal diet (%, DM bases)

Ingredient (%)	
Fish meal	9.10
Soybean meal	50.1
Maize flour	26
Plant oil	1.8
Fish oil	2
Vitamin premix[a]	1.5
Mineral premix[b]	1.5
Total	100
Proximate chemical analysis (%, DM bases)	
DM[c]	91.68
Crude protein	30.58
Ether extract	9.13
Crude fiber	5.49
Ash	8.13
NFE[d]	46.67
GE[e] (kcal/g)	4.729

[a]Vitamin premix (per kg of premix): thiamine, 2.5 g;riboflavin, 2.5 g; pyridoxine, 2.0 g; inositol, 100.0 g; biotin,0.3 g; pantothenic acid, 100.0 g; folic acid, 0.75 g; para-aminobenzoic acid, 2.5 g; choline, 200.0 g; nicotinic acid,10.0 g; cyanocobalamine, 0.005 g;α-tocopherol acetate,20.1 g; menadione, 2.0 g; retinol palmitate, 100,000 IU; cholecalciferol, 500,000 IU

[b]Mineral premix (per kg of premix): $CaHPO_4 \cdot 2H_2O$, 727.2 g; $MgCO_3 \cdot 7H_2O$, 127.5 g; KCl, 50.0 g; NaCl,60.0 g; $FeC_6H_5O_7 \cdot 3H_2O$, 25.0 g; $ZnCO_3$, 5.5 g;$MnCl_2 \cdot 4H_2O$, 2.5 g; $CuCl_2$, 0.785 g; $CoCl_3 \cdot 6H_2O$, 0.477 g;$CaIO_3 \cdot 6H_2O$, 0.295 g; $CrCl_3 \cdot 6H_2O$, 0.128 g; $AlCl_3 \cdot 6H_2O$, 0.54 g; Na_2SeO_3, 0.3 g

[c] DM—dry matter

[d]NFE—Nitrogen-free extract = 100 − (protein% + moisture % + lipid% + ash% + crude fiber%)

[e]GE—Gross energy was calculated according to National Research Council (2012) as 5.65, 9.45, and 4.11 kcal/g for protein, lipids, and carbohydrates, respectively

fish at 96 h, calculated by Finney (1971) Probit Analysis (SPSS Inc., Chicago, IL, USA). The LC50 concentration level was **3.66** mg/L (Table 2).

Adsorbent agent

Raw date nuclei (RDN) (date seeds or pits or stones) were collected from the fruits found in the Aquifer in El-Salhyia Area, Sharkia Governorate, Egypt (latitudes 30 35′ and 30 43′ N and longitudes 31°49′ and 32° 05′ E). The nuclei were separated, washed multiple times with distilled water, and dried for two hours in an 80 °C oven. The dried nuclei were retrieved from the furnace and cooled in a silica gel drier. After that, it was crushed into tiny particles using an electric grinder and packed in an airtight dark plastic bag, which was then kept in a clean desiccator until use.

Water physicochemical parameters assessments

The water samples were examined and recorded daily for estimated temperature and dissolved oxygen using an oxygen meter (YSI model 58, Yellow Spring Instrument Co., Yellow Springs, Ohio, USA). The Multi-parameter Ion Analyzer (HANNA Instruments, Rhodes Island, USA) was used to detect un-ionized ammonia. While, the pH value was measured using a pH-meter (Digital Mini-pH-Meter, model 55, Fisher Scientific, Denver, USA). The water samples used to estimate total alkalinity and total hardness were collected biweekly and quantified using the Diana et al. (2017) technique.

Growth performance

During the 60-days trial, the fish were given a commercial diet until they were satiated. Each aquarium was weighed every two weeks to determine the amount of fish biomass that had survived. The following formulae were used to determine growth performance:

Weight gain,

$$\mathrm{WG}\ (g) = W_2 - W_1$$

Daily weight gain,

$$\mathrm{DWG}\ (g/\mathrm{day}) = \mathrm{WG}/T$$

Specific growth rate,

$$\mathrm{SGR}\ (\%/d) = 100\ (\mathrm{Ln}\ W_2 - \mathrm{Ln}\ W_1)/T$$

where W_1 is initial weight, W_2 is final weight, and T is the experimental period (60 days).

Zn concentration analysis

At the end of the experiment, samples of gills and muscle from rearing fish in each treatment group were collected to determine Zn bioaccumulation. Five fish were dissected each treated group, and pieces of gills and muscle tissue were collected individually. Then, the collected tissues were rinsed in redistilled water, and stored at -20 °C until analysis. From each organ 1 g was placed in crucible and ashed in a muffle furnace (Thermolyne Corporation, Dubuque, Iowa, USA) for 6 h. Ash was digested with 5 ml concentrated H_2SO_4 and gradually kept at 130 °C on a hot plate until complete dryness. Then, the digests were diluted with 2 N HCl to a constant volume. The Zn concentration was determined with an atomic absorption spectrophotometer (Thermo 6600, Thermo Electron Corporation, Cambridge, UK), which was calibrated using Zn standard solutions. The same was done for the raw date nuclei (RDN).

Table 2 The 96 h LC50 of Zinc sulfate ($ZnSO_4 \cdot 7H_2O$) in common carp (*C. carpio*) calculated using Finney's probit analysis

Point	Conc. (mg/L)	95% confidence level		Slope ± S.E	Intercept	Chi-test (χ^2) Sig
		Upper	Lower			
LC 1.0	0.15	−1.402	1.024	1.664 ± 0.101	2.248	0.921
LC 10.0	1.73	0.792	2.286			
LC 25.0	2.64	2.022	3.062			
LC 50.0	**3.66**	3.256	4.059			
LC 75.0	4.67	4.250	5.294			
LC 80.0	4.92	4.470	5.627			
LC 95.0	6.13	5.471	7.282			
LC 99.0	7.16	6.287	8.719			

Control group (theoretical spontaneous response rate) = 0.0000

Bold value indicates the 96 h LC50

For determined the Zn removal level from the rearing water, one liter per aquarium was filtered by 0.8 mm of Millipore acetyl cellulose filter paper (Millipore, Bedford, MA, USA), digested with 10 ml concentrated H_2SO_4 on a hot plate at 70 °C, concentrated to 50 ml and transferred to a volumetric flask. Samples were adjusted up to 100 ml with redistilled water. Owing to Hijab et al. (2021) procedure, the amount of adsorbed Zn was assumed to be equal to the amount of Zn removed from the water (Amount of Zn adsorbed = Amount of Zn removed).

Statistical procedure

After validating the homogeneity and normality using Levene's test, the estimated and collected data were subjected to one-way ANOVA. Tukey's range test was used to identify differences between means at the 5% probability level. All the statistical analysis was done using SPSS version 20 (SPSS, Richmond, USA) as described by Gaur and Gaur (2006).

Results

Water quality measurements

Data in Table 3 reveal that, despite the fact that fish waste was not disposed of and aquaria water was not renewed, water quality measurements (dissolved oxygen, temperature, total alkalinity, and total hardness) did not differ markedly across all experimental groups. Conversely, ammonia levels were lower in aquaria that treated with raw date nuclei compared to the control group (CNT) and the date-free Zn-treated group (RDN_0). However, all water quality endpoints were within the permissible limits for fish production.

Zn accumulation in fish tissues and adsorbent agent

Table 3 shows that the amount of Zn in water reduced as the RDN dosage increased. The water that had not been treated with RDN had a higher level of Zn than the water treated with graded levels of RDN. Furthermore, when compared to fish groups subjected to graded amounts of date nuclei (RDN_3, $RDN_{4.5}$, and RDN_6), fish gills and muscles exposed to a sublethal concentration of Zn alone (RDN0) acquired larger levels of Zn accumulation. Also, Zn accumulation was observed to be lower in muscular tissues of the same fish groups than in gills. Consequently, the presence of RDN in the aquaria decreased Zn accumulation in the gills and muscles of fish (RDN_3, $RDN_{4.5}$, and RDN_6). Moreover, Zn retention was observed to be greater with higher quantities of RDN ($RDN_{4.5}$, and RDN_6) (Table 4).

Physiological biomarkers

The findings in Tables 5 showed that the RDN water treatments had a significant ($P < 0.001$) impact on all antioxidative biomarkers evaluated. SOD, GPx, GST, CAT, and MDA levels were significantly higher in fish raised in water that contains sublethal concentrations of Zn compared to the control group (CNT). In contrast, treated fish exposed to Zn-contaminated water with higher levels of RDN (4.5 and 6, respectively) significantly reduced all serum antioxidant activity when compared to fish exposed to Zn-contaminated water alone (RDN_0). The RDN_6 group had reported the normal antioxidant activity that seemed to be similar to the control group (CNT).

Growth indices

Table 6 demonstrates the efficiency of treated Zn-contaminated water with varying amounts of RDN on common carp growth performance indicators (IW, FW, WG, DWG, and SGR). In this study, reared fish in Zn-contaminated water

Table 3 Water quality parameters of Common carp (*C. carpio*) rearing water contaminated with sublethal concentration of zinc (3.38 mg L^{-1}) and treated for 8 weeks with graded amounts of Raw Date Nuclei (RDN)

Measurements	Treatments					*P* value
	CNT	RDN_0	RDN_3	$RDN_{4.5}$	RDN_6	
Temp, °C	25.08 ± 0.18	25.16 ± 0.27	25.49 ± 0.02	25.62 ± 0.19	25.38 ± 0.03	0.215
DO, mg/L	5.01 ± 0.04	4.92 ± 0.02	4.95 ± 0.22	4.98 ± 0.11	4.98 ± 0.01	0.160
pH	7.21 ± 0.22	7.24 ± 0.01	7.22 ± 0.31	7.23 ± 0.21	7.23 ± 0.11	0.596
NH_3, mg/L	0.0096 ± 0.41^{a}	0.0098 ± 0.31^{a}	0.006 ± 0.12^{ab}	0.0054 ± 0.15^{cd}	0.0051 ± 0.11^{d}	< 0.001
T. Alk., mg/L	187.25 ± 0.60	188.17 ± 0.97	187.58 ± 1.21	185.93 ± 0.51	185.93 ± 0.22	0.257
T.Hard., mg/L	225.88 ± 0.33	226.60 ± 0.49	225.26 ± 0.40	225.35 ± 0.87	225.35 ± 0.28	0.375

CNT, control group rearing in clean water

RDN_0, fish group rearing in Zn-contaminated water

RDN_3, fish group rearing in Zn-contaminated water and treated with 3 g RDN/L

$RDN_{4.5}$, fish group rearing in Zn-contaminated water and treated with 4.5 g RDN/L

RDN_6, fish group rearing in Zn-contaminated water and treated with 6 g RDN/L

Temp, temperature; DO, dissolved oxygen; NH_3, un-ionized ammonium; T. ALk, total alkalinity; T. Hard., total hardness

Means with the different letter in the same raw are significantly different at $P < 0.05$

Table 4 The adsorption efficacy of several levels of Raw Date Nuclei (RDN) on zinc concentration in water, gills and muscles of fish, and RDN of common carp subjected to water polluted with sublethal doses of zinc (3.38 mg L^{-1}) for 8 weeks

Measurements	Treatments					*P* value
	CNT	RDN_0	RDN_3	$RDN_{4.5}$	RDN_6	
Water, mg/L	ND	4.71 ± 0.03^{a}	1.79 ± 0.06^{b}	0.81 ± 0.01^{c}	0.82 ± 0.01^{c}	< 0.001
Gills, µg/g	ND	207.03 ± 0.91^{a}	76.36 ± 0.67^{b}	45.77 ± 0.89^{c}	40.77 ± 0.28^{d}	< 0.001
Muscles, µg/g	ND	45.58 ± 2.14^{a}	14.49 ± 0.38^{b}	12.67 ± 0.18^{bc}	11.03 ± 0.25^{c}	< 0.001
RDN seeds, µg/g	ND	NA	210.54 ± 1.06^{b}	215.94 ± 0.80^{a}	215.61 ± 0.47^{a}	< 0.001

CNT, control group rearing in clean water

RDN_0, fish group rearing in Zn-contaminated water

RDN_3, fish group rearing in Zn-contaminated water and treated with 3 g RDN/L

$RDN_{4.5}$, fish group rearing in Zn-contaminated water and treated with 4.5 g RDN/L

RDN_6, fish group rearing in Zn-contaminated water and treated with 6 g RDN/L

ND not detected, NA not applicable

Means with the different letter in the same raw are significantly different at $P < 0.05$

Table 5 Changes in some antioxidative biomarkers of *C. carpio* muscle reared in water contaminated with sublethal concentration of zinc (3.38 mg L^{-1}) for 8 weeks in the absence or presence of graded levels of Raw Date Nuclei (RDN)

Measurements	Treatments					*P* value
	CNT	RDN_0	RDN_3	$RDN_{4.5}$	RDN_6	
SOD (IU/mg protein)	32.58 ± 0.85^{c}	106.44 ± 3.52^{a}	52.99 ± 1.94^{b}	33.08 ± 0.83^{c}	32.55 ± 0.06^{c}	< 0.001
GPx (IU/mg protein)	47.68 ± 1.17^{d}	133.87 ± 2.56^{a}	$92.52 \pm .98^{b}$	58.91 ± 1.18^{c}	50.57 ± 0.86^{d}	< 0.001
GST (nmol /min.mg)	154.02 ± 0.77^{d}	190.98 ± 1.33^{a}	168.54 ± 1.02^{b}	163.47 ± 1.28^{c}	155.81 ± 1.54^{d}	< 0.001
CAT						
(IU/mg protein)	11.95 ± 0.23^{c}	24.52 ± 0.54^{a}	16.55 ± 0.35^{b}	12.62 ± 0.26^{c}	11.63 ± 0.26^{c}	< 0.001
MDA (nmol/mg protein)	6.93 ± 0.34^{d}	35.32 ± 0.94^{a}	18.71 ± 0.48^{b}	9.88 ± 0.27^{c}	8.55 ± 0.46^{cd}	< 0.001

CNT, control group rearing in clean water

RDN_0, fish group rearing in Zn-contaminated water

RDN_3, fish group rearing in Zn-contaminated water and treated with 3 g RDN/L

$RDN_{4.5}$, fish group rearing in Zn-contaminated water and treated with 4.5 g RDN/L

RDN_6, fish group rearing in Zn-contaminated water and treated with 6 g RDN/L

SOD, Superoxide dismutase; GPx, Glutathione peroxidase; GST, Glutathione S-transferase; CAT, Catalase; MDA, malonaldehyde

Means with the different letter in the same raw are significantly different at $P < 0.05$

markedly reduced all growth indicators when compared to the control group (CNT). In comparison with the RDN_0 free treated group, treated common carp fish rearing water with higher levels of RDN (6 g followed by 4.5 g RDN, respectively) significantly improved final biomass, weight gain, daily weight gain, and SGR.

Discussion

Fish is rich in a variety of important micro or macro elements and proteins that are easily absorbed and digested by humans (Wang et al. 2021). However, as industrial expands, the public health is challenged with serious Zn water contamination (Liu et al. 2021). Zn was found to accumulate to higher levels in fish flesh, causing serious risks to consumers (El-Moselhy et al. 2014). Also, Zn accumulation causes nutritional disorders as well as induced oxidative stress in fish (Connolly et al. 2016). Therefore, it is critical to develop novel water treatment technologies for eliminating and reducing Zn toxicity (Loro et al. 2012). Date nuclei are gaining more attention as a powerful adsorbent among agricultural wastes for the removal of different contaminants due to their inexpensive cost and higher adsorption capacity (Ahmad et al. 2012; Hegazi 2013). Thus, the primary objective of this study was to assess the efficacy of RDN in improving water quality and removing Zn ions from water and fish flesh, thereby enhancing common carp fish physiological biomarkers and performance.

Owing to our results, despite the fact that fish waste was not removed and water aquaria were not refreshed, water quality was not affected by Zn contamination and water treatment with RDN. However, as compared to control and Zn-contaminated water without RDN, treated fish aquarium water with graded amounts of RDN lowered ammonia concentration. The higher levels of ammonia in Zn-contaminated water demonstrated that Zn causes stress in fish, leading them to release more ammonia. These findings were found to be consistent with Wendelaar Bonga (1997) results that increased cortisol levels under stress resulted in increased ammonia production. The same results were obtained when common carp were exposed to Cu toxicity (De Boeck et al. 2007; Kunwar et al. 2009; Yousefi et al. 2020) and Nile tilapia were exposed to Zn toxicity (Mohamed et al. 2019b).

Our data reveal a strong positive relationship between a high RDN dose and lower Zn bioaccumulation in water, gills, and muscles. While, the gills of fish in the Zn-contaminated group were free of RDN had significantly higher Zn accumulation levels than the groups treated with RDN. According to Javed et al. (2008), high concentrations of Zn in gill tissue might be linked to the fact that the gills are the principal sites of Zn absorption in freshwater fish, as well as the large surface area that is in contact with ambient water. Besides, Zn accumulation was lower in muscular flesh of the same fish groups than in gills. This implies that RDN absorbed Zn ions in the water before they accumulated in fish tissue. In addition, it should be demonstrated that the removal of Zn ions and improved water quality were relative to the quantity of RDN used in each aquarium. Actually, the raw date nuclei (RDN) are consisted of three main components, cellulose, hemicellulose and lignin as well as other components such as proteins, oils (El-Juhany 2010). The cellulose and hemicellulose combine to produce lignocellulosic material, which has more oxygen functional groups such as hydroxyl, ether, and carbonyl, while lignin serves as a composite material that keeps cellulose and hemicellulose units together (Pinto et al. 2021). The variety of surface function groups that include oxygen, as well as pore size, are important factors in regulating metal adsorption pathways (Al-Ghouti et al. 2010). These findings were corroborated by Mohamed et al. (2019a) who discovered that RDN powder

Table 6 Growth performance of *C. carpio* reared in water polluted with sublethal concentration of zinc (3.38 mg L^{-1}) for 8 weeks in absence or presence of graded levels of Raw Date Nuclei (RDN)

Measurements	Treatments					*P* value
	CNT	RDN_0	RDN_3	$RDN_{4.5}$	RDN_6	
Initial weight (g)	3.75 ± 0.21	3.75 ± 0.14	3.75 ± 0.54	3.75 ± 0.11	3.74 ± 0.22	0.951
Final weight (g)	20.33 ± 0.14^{c}	13.37 ± 0.27^{d}	22.68 ± 0.94^{b}	26.98 ± 0.13^{a}	27.11 ± 0.29^{a}	0.001
Weight gain (g)	16.58 ± 0.15^{c}	9.62 ± 0.27^{d}	18.93 ± 0.64^{b}	23.23 ± 0.12^{a}	23.36 ± 0.30^{a}	< 0.01
DWG (g)	0.27 ± 0.5^{c}	0.16 ± 0.1^{d}	0.32 ± 0.4^{b}	0.38 ± 0.7^{a}	0.39 ± 0.1^{a}	0.001
SGR (%)	2.81 ± 0.1^{c}	2.12 ± 0.0^{d}	2.99 ± 0.0^{b}	3.28 ± 0.6^{a}	3.29 ± 0.2^{a}	< 0.1

CNT, control group rearing in clean water

RDN_0, fish group rearing in Zn-contaminated water

RDN_3, fish group rearing in Zn-contaminated water and treated with 3 g RDN/L

$RDN_{4.5}$, fish group rearing in Zn-contaminated water and treated with 4.5 g RDN/L

RDN_6, fish group rearing in Zn-contaminated water and treated with 6 g RDN/L

DWG, daily weight gain; SGR, specific growth rate

Means with the different letter in the same raw are significantly different at $P < 0.05$

had a great capacity to decrease heavy metal concentrations in contaminated water. Thus, it is possible to achieve that RDN is an excellent adsorbent for the removal of metal ions from aqueous solutions.

It was well known that various metal contaminants had the ability to induce oxidative stress in aquatic creatures including fish (Velkova-Jordanoska et al. 2008; López-López et al. 2011). Oxidative stress disrupts the body's homeostasis by inducing biochemical, physiological, and behavioral alterations (Borković et al. 2005). In the present work, the antioxidant enzymes superoxide dismutase (SOD), glutathione peroxidase (GPx), glutathione transferases (GST), and catalase (CAT) were evaluated as biomarkers for detecting Zn toxicity in Common carp (*C. carpio*). The activity of these enzymes was found to be higher in Zn-contaminated fish groups absent of RDN (RDN_0) compared to the control (CNT). These results are consistent with those of Borković et al. (2005), Fırat et al. (2010), and Karadag et al. (2014), who found that water pollution generated oxidative stress in fish and that oxidative stress-biomarkers increased in response to this stress. As previously stated by Yildirim et al. (2011), the increments in catalase (CAT) and malondialdehyde (MDA) levels was attributable to an increase in lipid peroxidation caused by Zn contaminated water stress. To the best of our knowledge, limited work on date nuclei was done to remove contaminants from polluted solutions (Yazid and Maachi 2008; Nouacer et al. 2015; Bouranene and Sedira 2019). The toxic-Zn-stress was decreased in RDN-containing treatments, leading in a reduction of oxidative stress and, as a result, antioxidants retained to normal levels, and the influence of RDN-adsorption was discovered to be connected to the treated dose. Due to the presence of various functional groups, the RDN becomes acidic in nature and metal ions may form complexes with those surface functional groups of the RDN such as cellulose-OH and phenolic-OH through ion exchange reactions (Soong and Barlow 2004). The most of these functional groups involved in the binding process are located within the cell walls (Demirbas 2008). Therefore, Mall et al. (2006) previously ascribed this increase in adsorption with rising adsorbent dosage to increased adsorbent surface and availability of additional adsorption sites.

Finally, our findings revealed that exposure to a sublethal concentration of Zn (RDN0) had a negative effect on growth performance when compared to the control (CNT). Javed (2012) confirmed the negative impact of Zn on fish growth via reporting that chronic Zn treatment (at one-third of LC50) to three different fish species resulted in significantly reduced final biomass weight and weight gain. In the same manner, Nasri et al. (2020) exhibited that rainbow trout (*Oncorhynchus mykiss*) exposed to higher concentrations of Zn grow slower than those exposed to lower levels of Zn. Furthermore, it was observed that when the Zn levels in the water increased, weight gain and SGR reduced linearly (Nasri et al. 2020). On the other hand, growth performance in fish groups exposed to Zn-contaminated water and treated with graded amounts of RDN exceeded the control group in terms of performance and feed efficiency values. Based on these findings, it is inferred that RDN may adsorb Zn ions from water, hence reducing zinc's undesirable impact on growth performance in rearing carp.

Conclusion

In this study, raw date nuclei (RDN) were effectively used as a low-cost and practical adsorbent for absorbing Zn ions from water and decreasing Zn bioaccumulation in fish organs. In addition, the RDN has enhanced water quality parameters, physiological biomarkers, and final biomass. Therefore, RDN may be suggested for heavy metal ion removal from fish Zn-contaminated water at level 6 g RDN per liter due to their lower cost and higher adsorption capability.

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Data availability All related data were available under reasonable request from the corresponding author.

Declarations

Conflict of interest The authors declare that they have no competing interests.

Consent to participate All the authors equally participated to prepare the manuscript in all stages.

Consent for publication All the authors approved to submit the present manuscript to Fish.

Ethical approval The animal ethics guidelines were followed and approved from the Zagazig university animal ethics committee.

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