



Effect of biosorption of Fe^{2+} by *Rhodotorula mucilaginosa* YMM19 on the physiology of *Lupinus albus*, *Triticum aestivum*, *Vicia faba*, and *Zea mays* seedlings

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Abstract

The goal of this study was to look into growth changes and a number of metabolic events in Fe^{2+} treated *Lupinus albus*, *Triticum aestivum*, *Vicia faba*, and *Zea mays* plants and assess the role of biosorption of Fe^{2+} by *Rhodotorula mucilaginosa* YMM19 to alleviate these changes. The effect of Fe^{2+} concentrations (untreated and treated with biosorbent) on seed germination was studied. *T. aestivum* plumule was stimulated with a low dose of Fe^{2+} . However, the application of Fe^{2+} reduced the elongation of plumule, and radicle of all seeds during germination. High doses of Fe^{2+} treated with biosorbent significantly increased seedling weights (fresh and dry) of all plants. Also, after 20 days, the height, and weight of seedlings of *L. albus*, *T. aestivum*, and *Z. mays* were increased with biosorbed Fe^{2+} solution. In addition, biosorption of Fe^{2+} enhanced total carbohydrate and protein accumulation in both *T. aestivum* and *Z. mays* radicles. Moreover, Fe^{2+} caused slight suppression of α - and β -amylase in *L. albus* and *Z. mays* seeds after 2 days, but the opposite effect was observed in *T. aestivum*. Thus, the biosorption by *R. mucilaginosa* YMM19 is an efficient system for removing the negative effect of excess Fe^{2+} from water.

Keywords Crop seedlings · Biosorption of Fe^{2+} by *Rhodotorula mucilaginosa* YMM19 · Growth and metabolic activities

Introduction

Because of their long-term persistence in the environment and poisonous nature, heavy metals constitute a severe environmental issue (Ali et al. 2019). They include iron, silver, lead, cadmium, nickel, and cobalt, among others (Nagajyoti et al. 2010). Some of the heavy metals (e.g., Zn, Fe, Cu, Ni and Co) are micronutrients for plants (Reeves and Baker 2000; Wintz et al. 2002). Iron is a necessary element for all plants and has many vital roles. It is also a significant component of cellular oxidation systems (Marschner 1995). Also, Fe is a micronutrient for all plants. It performs vital task in respiration, photosynthesis, and as enzymes cofactor (Rout and Sahoo 2015). However, excessive absorption of these plant micronutrients leads to toxic effects (Blaylock 2000; Monni et al. 2000). Although Fe^{2+} is abundant in soil

minerals, signs of iron poisoning appear in flood conditions due to high plant absorption of Fe^{2+} (Becker and Asch 2005). Fe^{2+} toxicity includes the formation of toxic radicals that destroy cellular structures such as proteins, DNA, and membranes and reduce photosynthesis (Arora et al. 2002; De Dorlodot et al. 2005; Sinha et al. 1997). Therefore, removing excess Fe^{2+} from irrigation water is an important issue (Shamshuddin et al. 2013; Shokoohi et al. 2009). Several systems have been aimed to eliminate Fe^{2+} from the aqueous solution, including physical, chemical, and biological processes (Shamshuddin et al. 2013; Shokoohi et al. 2009). However, new eco-friendly methods have emerged with cost-effective and straightforward implementation (Quintelas and Tavares 2002; Shamshuddin et al. 2013; Shokoohi et al. 2009). Several microbial isolates are used as bio-sorbents (Aksu et al. 1999; Kanamarlapudi and Muddada 2019; Quintelas and Tavares 2002; Verma et al. 2017; Wang and Chen 2006). Moreover, several studies have used *R. mucilaginosa* in the bioremediation of heavy metals (Jiang et al. 2013; Jin et al. 2020; Lopez-Fernandez et al. 2019; San and Dönmez 2012).

The present study aimed to study the biosorption of Fe^{2+} from solutions by *R. mucilaginosa* YMM19. In addition,

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factors governing the biosorption, such as biomass dosage, pH, adsorption period, and initial Fe^{2+} concentration were analyzed. Moreover, to evaluate the influence of bio-treated water on the germination, growth, and development of *Lupinus albus*, *Triticum aestivum*, *Zea mays*, and *Vicia faba* compared to untreated water.

Materials and methods

Yeast biosorbent

The fungus (strain YMM19) was isolated and purified from the fruits of *Morus nigra* L. at Beheira governorate, Egypt. According to their morphological and molecular characteristics (GenBank accession numbers of MN960158) the fungus (YMM19) was recognized as *R. mucilaginosa* via a significant identity of 99.28%.

Preparation of yeast biosorbent

The PDB medium was used for yeast biosorbent formation and was made according to the manufacturer's instructions (Oxoid, Basingstoke, Hampshire, RG24 8PW, UK). An aliquot of 1% (v/v) of yeast strain (10^5 cell/ml), grown in PDB at 200 rpm for 3 days, 28 °C and pH 6.5, were used as an inoculum for biosorbent production. To prepare the biosorbent, *R. mucilaginosa* strain YMM19 was cultured in a flask (Erlenmeyer) of 250 mL size (contains 50 mL of PDB) at pH 6.5, 200 rpm, and 28 °C for three days. First, yeast cell masses were obtained by centrifugation of the PDB (at 6000 rpm for 15 min) and washed well twice with distilled water. Then, the harvested live yeast cells were dried for one day in 60 °C in an electric oven and powdered with a mortar and pestle.

Biosorption of Fe^{2+} using different doses of yeast biosorbent

The dried yeast biomass (1–7 g/L) was subjected to direct contact with aqueous solutions (20 ml) containing 125 mg/L of Fe^{2+} at 28 °C, pH 7 and 200 rpm. Metal solution without biomass addition was used as a control. After 3 h of contact time, the biomass was aseptically filtered using Whatman (No. 1) paper. Then, the residual Fe^{2+} ions in the supernatants were analyzed after dilution with a Perkin-Elmer 2380 atomic absorption spectrophotometer. Finally, the biosorption (%) of the Fe^{2+} ions was calculated by the equation:

$$B(\%) = 100 \times (C_i - C_f) / C_i$$

where $B = \text{Fe}^{2+}$ ion biosorption (percentage), $C_i = \text{Fe}^{2+}$ in solution initial concentration (mg/L), and $C_f = \text{Fe}^{2+}$ in solution final concentration (mg/L).

Effect of time on Fe^{2+} biosorption

The dried biomass (1 g/L) of the YMM19 strain was examined for its biosorption of Fe^{2+} from aqueous solutions (20 ml) containing 125 mg/L of Fe^{2+} ions at 28 °C, pH 7 and 200 rpm for 3 and 6 and 8 h.

Effect of pH on Fe^{2+} biosorption

The YMM19 strain was tested for its Fe^{2+} (125 mg/L) biosorption using a 1 g/L biosorbent under a pH of 3–8 at 200 rpm, 28 °C for 3 h. The tested pHs were regulated by freshly prepared 0.1 N HCl or 0.1 N NaOH solutions.

Effect of Fe^{2+} on its biosorption

The YMM19 strain was tested for its biosorption of 125 and 500 mg/L Fe^{2+} using 1 g/L biosorbent at pH 7, 28 °C and 200 rpm for 3 h.

Plant material

Giza Agricultural Research Station in Giza, Egypt, provided seeds of *Lupinus albus* (cv. Giza1), *Triticum aestivum* (cv. Masri 2), *Zea mays* (cv. Cairo 1), and *Vicia faba* (cv. Gizah 3). The healthy seeds were chosen for their size and shape uniformity, then surface sterilised (2.5 percent Clorox for 5 min) and rinsed completely in distilled water.

Germination and seedling experiment

Germination of four seedlings (*Lupinus albus*, *Triticum aestivum*, *Zea mays*, and *Vicia faba*) was carried out in 0.02 m² plastic plates containing 780 g acid-washed sterilised sandy soil at ambient temperature (23 °C). According to the Fe^{2+} concentrations used, seeds were divided into 5 groups. (0.0, 125, 125B, 500 and 500B mg/L); “B” meaning biosorption of Fe^{2+} by *R. mucilaginosa*. Each dish included ten seeds that were irrigated with various iron solutions that were either biosorption treated or not. The light/darkness photoperiod was 10:14 h. The germinating seeds were collected at 10-days-old to determine germination (percentage) and growth characteristics (radicle and plumule lengths, fresh and dry weights), and activity of alpha- and beta-amylases were assayed in seeds at 2-day old (MacGregor 1978) (Fig. 1). The seedlings (*Lupinus albus*, *Triticum aestivum*, and *Zea mays*) were harvested at 20 days old to discover pigment content, total carbohydrate content, and total protein content are all growth metrics

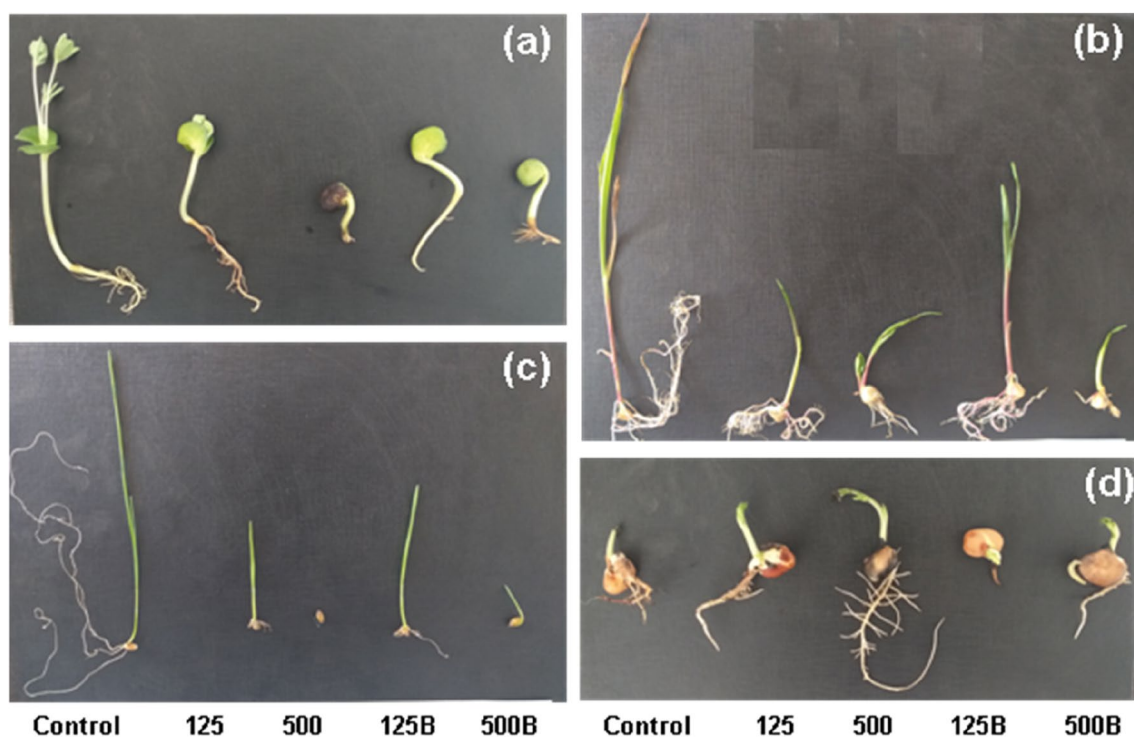


Fig. 1 The seedlings of 10-day old **a** *Lupinus albus* **b** *Zea mays*, **c** *Triticum aestivum* and **d** *Vicia faba* seedlings as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125, 500, 125B and 500B ppm); B: biosorbed by *R. mucilaginosa*

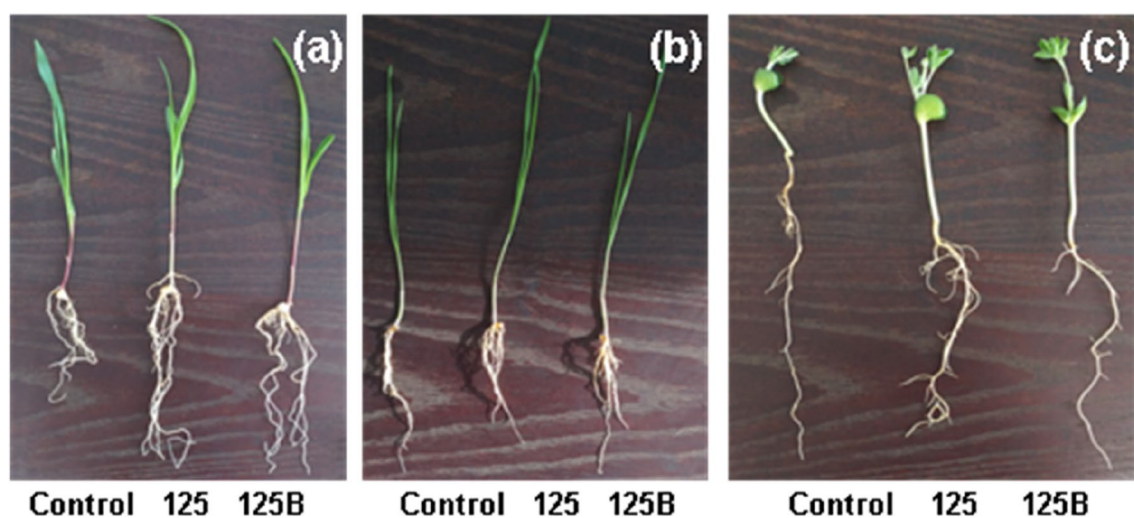


Fig. 2 The seedlings of 20-day old **a** *Zea mays*, **b** *Triticum aestivum* and **c** *Lupinus albus* seedlings as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125, 500, 125B and 500B ppm); B: biosorbed by *R. mucilaginosa*

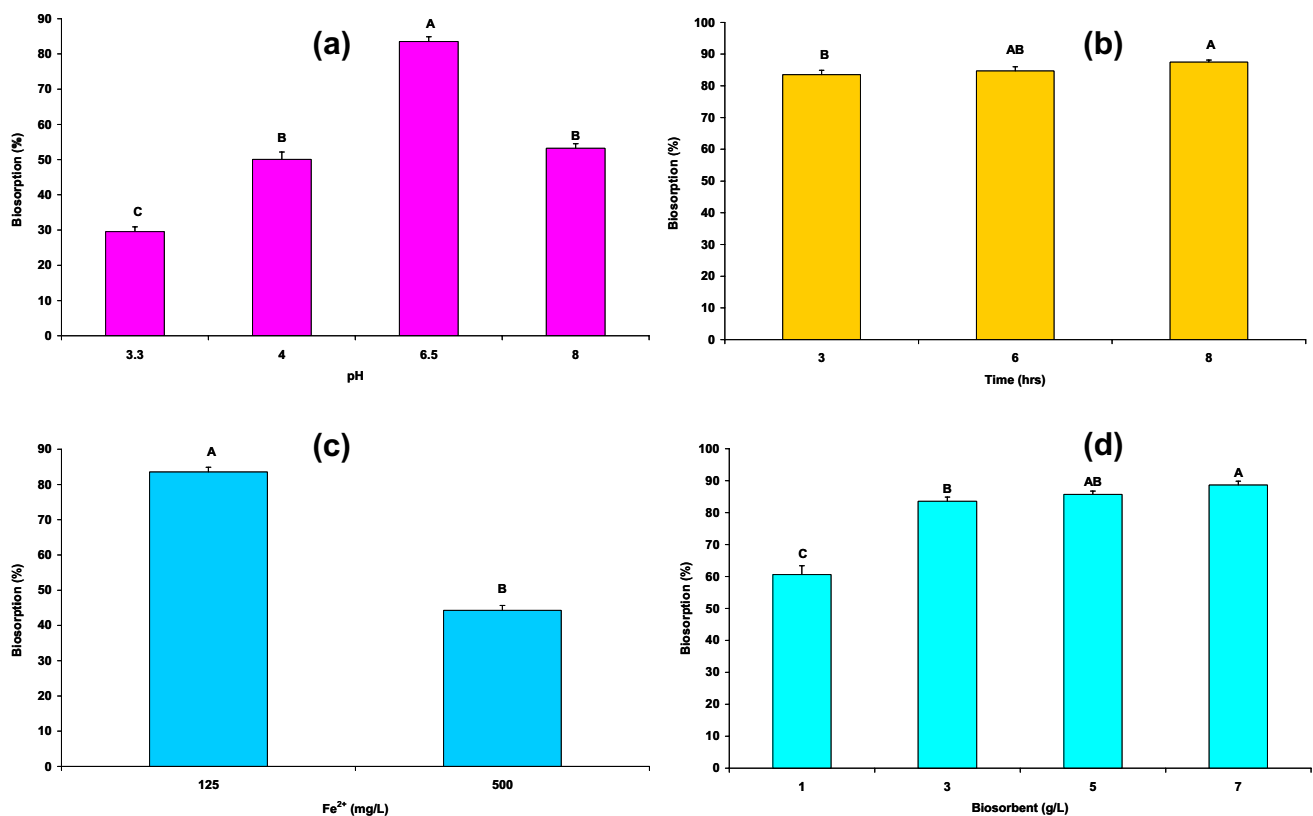


Fig. 3 The biosorption (%) of Fe^{2+} by *Rhodotorula mucilaginosa* YMM19 at different **a** pH values **b** Times (hrs), **c** Fe^{2+} concentrations (mg/L) and **d** biosorbent doses (g/L). Different letters (A-C) on

the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)

(radicle and plumule lengths, fresh and dried weights) (Fig. 2).

Estimation of pigments

Freshly weighed leaves were homogenised in 5 ml 85 percent cold acetone right away, centrifuged for 15 min at 3000×g, and kept in the refrigerator overnight. The colour intensity of the acetone extract was measured at 663, 644, and 452.5 nm once it was completed to the required volume (Metzner et al. 1965). Pigment fractions were expressed as $\mu\text{g/g}$ fresh weight.

Estimation of carbohydrates and total proteins.

Aliquots (100 mg) of fine powdered dry plumules and radicles were ground into a fine powder (100 mg), extracted in borate buffer (28.63 g boric acid + 29.8 g potassium chloride + 3.5 g sodium hydroxide in a liter of distilled water). The mixture pH was regulated to 8.0 and kept standing for 24 h at 4 °C before centrifugation for 15 min at 3000 × g. The residues were cleaned different times and dried at 80 °C for polysaccharide estimation. The supernatant and residue washings were collected and used to estimate soluble sugars and proteins.

Carbohydrates were extracted in a 0.1 dry mass (10 cm³ buffer)⁻¹ borate buffer with a pH of 8. Carbohydrates were estimated quantitatively using Nelson (1944) together with certain amendments was attained by Naguib (1963). Briefly, 10 mg of dry residues of the plant were extracted in a borate solution and mixed with acetate (0.1 ml of mixture containing 6 ml acetic acid (0.2 N) and 4 mL 0.2 N sodium acetate) and 0.2 ml amylase (0.1%; w/v), completed until 3 ml using distilled water, and left for one day at 25 °C, the starch can then be estimated quantitatively after centrifugation for 15 min at 3000 rpm. According to Lowry et al. (1951) total proteins were measured.

Statistical analysis

To evaluate the degree of significance between treatments, the data were statistically examined. The data was analysed using a one-way analysis of variance (ANOVA; factorial). The least-significant difference method (LSD) at 5% was used to compare means. All the experiments were run in triplicates.

Table 1 Percentage of germination, undried and dehydrated weights and radicle and plumule lengths of 10-day old *Lupinus albus*, *Triticum aestivum*, *Zea mays* and *Vicia faba* seedlings as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 (MN960158) and unbiosorbed (Control, 125, 500, 125B and 500B ppm); B: biosorbed by *R. mucilaginosa*

Seedling	Fe^{2+} (mg/L)	Germination (%)	Seedling FW (g)	Seedling DW (g)	Plumule length (cm)	Radicle length (cm)
<i>Lupinus albus</i>	C	98.67 ± 0.94 ^A	2.68 ± 0.33 ^A	0.24 ± 0.03 ^{B C}	13.33 ± 0.94 ^A	5.83 ± 1.31 ^{AB}
	125	80.00 ± 3.27 ^B	2.15 ± 0.31 ^{AB}	0.32 ± 0.04 ^A	6.83 ± 0.24 ^B	7.50 ± 0.41 ^A
	500	60.00 ± 2.45 ^D	1.05 ± 0.29 ^C	0.20 ± 0.03 ^C	2.33 ± 0.85 ^C	0.57 ± 0.17 ^C
	125B	98.67 ± 1.25 ^A	1.71 ± 0.34 ^{BC}	0.31 ± 0.02 ^{AB}	2.50 ± 1.08 ^C	5.17 ± 1.18 ^B
	500B	70.00 ± 4.08 ^C	1.41 ± 0.26 ^C	0.27 ± 0.03 ^{AB C}	2.00 ± 1.08 ^C	2.33 ± 0.24 ^C
<i>Triticum aestivum</i>	C	70.00 ± 2.45 ^C	0.15 ± .002 ^A	0.03 ± .002 ^B	6.57 ± 0.66 ^A	8.87 ± 0.45 ^A
	125	80.00 ± 3.27 ^B	0.11 ± 0.01 ^B	0.03 ± .001 ^B	2.25 ± 0.12 ^B	0.37 ± 0.12 ^C
	500	0	0	0	0	0
	125B	90.00 ± 1.63 ^A	0.10 ± 0.02 ^B	0.02 ± .004 ^C	7.30 ± 1.06 ^A	3.43 ± 0.12 ^B
	500B	80.00 ± 4.90 ^B	0.09 ± 0.01 ^B	0.04 ± .002 ^A	2.47 ± 0.41 ^B	0.23 ± 0.05 ^C
<i>Zea mays</i>	C	99.00 ± 0.82 ^A	0.87 ± 0.02 ^A	0.15 ± 0.01 ^B	20.5 ± 2.27 ^A	14.5 ± 2.45 ^A
	125	80.00 ± 1.63 ^B	0.67 ± 0.02 ^C	0.15 ± .002 ^B	7.33 ± 0.62 ^C	7.17 ± 0.62 ^B
	500	80.00 ± 3.27 ^B	0.73 ± 0.01 ^B	0.23 ± 0.01 ^A	6.00 ± 1.08 ^{CD}	2.50 ± 0.82 ^C
	125B	98.67 ± 0.94 ^A	0.84 ± 0.02 ^A	0.14 ± .004 ^B	12.17 ± 0.85 ^B	7.00 ± 1.08 ^B
	500B	70.00 ± 2.45 ^C	0.65 ± 0.02 ^C	0.23 ± .002 ^A	3.63 ± 0.33 ^D	0.83 ± 0.24 ^C
<i>Vicia faba</i>	C	80.00 ± 1.63 ^B	2.20 ± 0.08 ^D	0.8 ± 0.02 ^C	1.17 ± 0.24 ^B	3.83 ± 2.25 ^{BC}
	125	99.00 ± 0.82 ^A	2.82 ± 0.03 ^B	0.77 ± 0.02 ^C	2.67 ± 0.47 ^{AB}	6.83 ± 0.24 ^{AB}
	500	98.33 ± 1.25 ^A	2.79 ± 0.04 ^B	0.79 ± 0.01 ^C	3.33 ± 1.65 ^A	10.00 ± 4.55 ^A
	125B	20.00 ± 0.82 ^C	2.65 ± 0.04 ^C	0.95 ± 0.01 ^A	0.83 ± 0.62 ^B	1.500 ± 1.08 ^C
	500B	99.33 ± 0.47 ^A	2.97 ± 0.02 ^A	0.91 ± 0.02 ^B	2.33 ± 0.24 ^{AB}	5.5 ± 0.71 ^{ABC}

Means that do not share a letter are significantly different

FW fresh weight, DW dry weight

Results and discussion

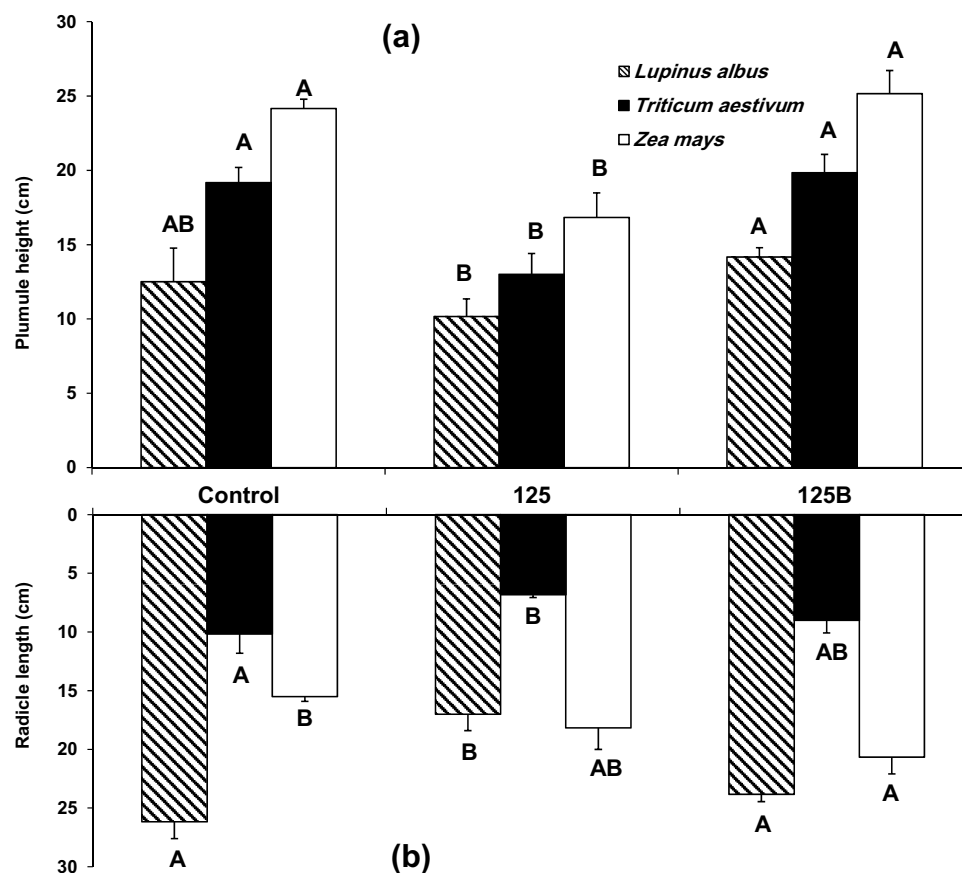
Biosorption of Fe^{2+} from solution

Pollution with heavy metallic elements is a serious problem that threatens natural ecosystems (Tchounwou et al. 2012). Heavy metals, radionuclides, and earth elements are isolated through biosorption, commonly by dead biomass (Kotrba 2011). Several studies have been declared to exclude heavy elements by *Rhodotorula* spp. (Grujić et al. 2017; Jiang et al. 2013; Jin et al. 2020; Lopez-Fernandez et al. 2019). Many factors influence the heavy elements adsorption such as biomass concentration, time, pH, and metal ion concentration (Torres 2020; Veglio and Beolchini 1997; Wang and Chen 2006). In the this study, the optimum condition for Fe^{2+} biosorption (%) was pH 6.5, 7 g/l of biosorbent, 125 mg/L of Fe^{2+} , and for 8 h (Fig. 3). However, no significant biosorption difference was between 3, and 6, and between 6 and 8 h biosorption time. Similarly, no considerable biosorption difference was between 3, and 5, and between 5 and 7 g/l of biosorbent (Fig. 3). Innumerable papers have given similar results (Elsherif 2017; Elsherif et al. 2018; Jin et al. 2020; Rozman et al. 2020; Sundararaju et al. 2020; Verma et al. 2017).

Germinating percentage and growth criteria

The current results shown that biosorption of Fe^{2+} at 125 mg/L induced the same trend applied to the seed germination (%) of *Lupinus albus*, *Triticum aestivum*, *Zea mays*, and *Vicia faba* seedlings of 10-day old (Table 1). The seedlings' fresh weight data demonstrated that biosorption of Fe^{2+} at 500 mg/L induced a significant raise in *Vicia faba* seedlings compared to control. Also, the results of Table 1 denoted that in seedling's dry weight, the biosorption of Fe^{2+} at both concentrations induced a significant raise in *Lupinus albus*, *Vicia faba*, and *Zea mays* seedlings. The data of plumule length more prominently at biosorption of Fe^{2+} at 125 mg/L in *Triticum aestivum* compared to control, but in *Zea mays* the plumule length is more pronounced at biosorption of Fe^{2+} at 125 mg/L compared to the same concentration without biosorption. Under all conditions, the plumule radicle lengths were increased in *Vicia faba* seedlings regardless, at the treatment of biosorption of Fe^{2+} at 125 mg/L compared to control. The Table 1 exposed a considerable decrease in radicle length at all concentrations of *Lupinus albus*, *Triticum aestivum*, and *Zea mays* seedlings compared to control. Figure 4a, b shows the shoot height besides root size of *Lupinus albus*, *Triticum aestivum*, and

Fig. 4 Plumule height and radicle length of 20-day old *Lupinus albus*, *Triticum aestivum* and *Zea mays* plants as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125 and 125B ppm); B: biosorbed by *R. mucilaginosa*. Different letters (A–B) on the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)



Zea mays seedlings of 20-day old. It found that biosorption of Fe^{2+} at 125 mg/L induced a considerable growing in plumule height and radicle length compared to the same concentration without biosorption in all three mentioned seedlings above. Concerning Fig. 5a, b the undried and dehydrated weights of the three seedlings, the data revealed that biosorption of Fe^{2+} at 125 mg/L induced a significant increase compared to control. In certain circumstances, high amounts of iron may be a risk. However, It's probably toxic and may stimulate the development of reactive oxygen-based radicals, which can cause lipid peroxidation, which can disrupt essential cellular constituents (e.g., membranes). Acidity, bronzing, and change of color of the plant roots indicate treatment with upper levels of Fe^{2+} (Laan et al. 1991). The bioavailability of iron is greatly influenced by microorganisms. The catalytic chemical reaction of Fe^{2+} by microorganisms takes occur in an acidic environment on their cell walls. The existence of chelating-producing bacteria that create iron chelate complexes through the production of metabolites, in turn, is a crucial component of the Fe^{3+} bioreduction process (Wetzel 2001). Various oxide reduction processes occur as a result of microorganism activity, which, along with fungous organic metabolites, play a vital role in metal circulation biogeochemical cycles, increasing their toxicity while also inactivating them (Urík et al. 2014, 2018).

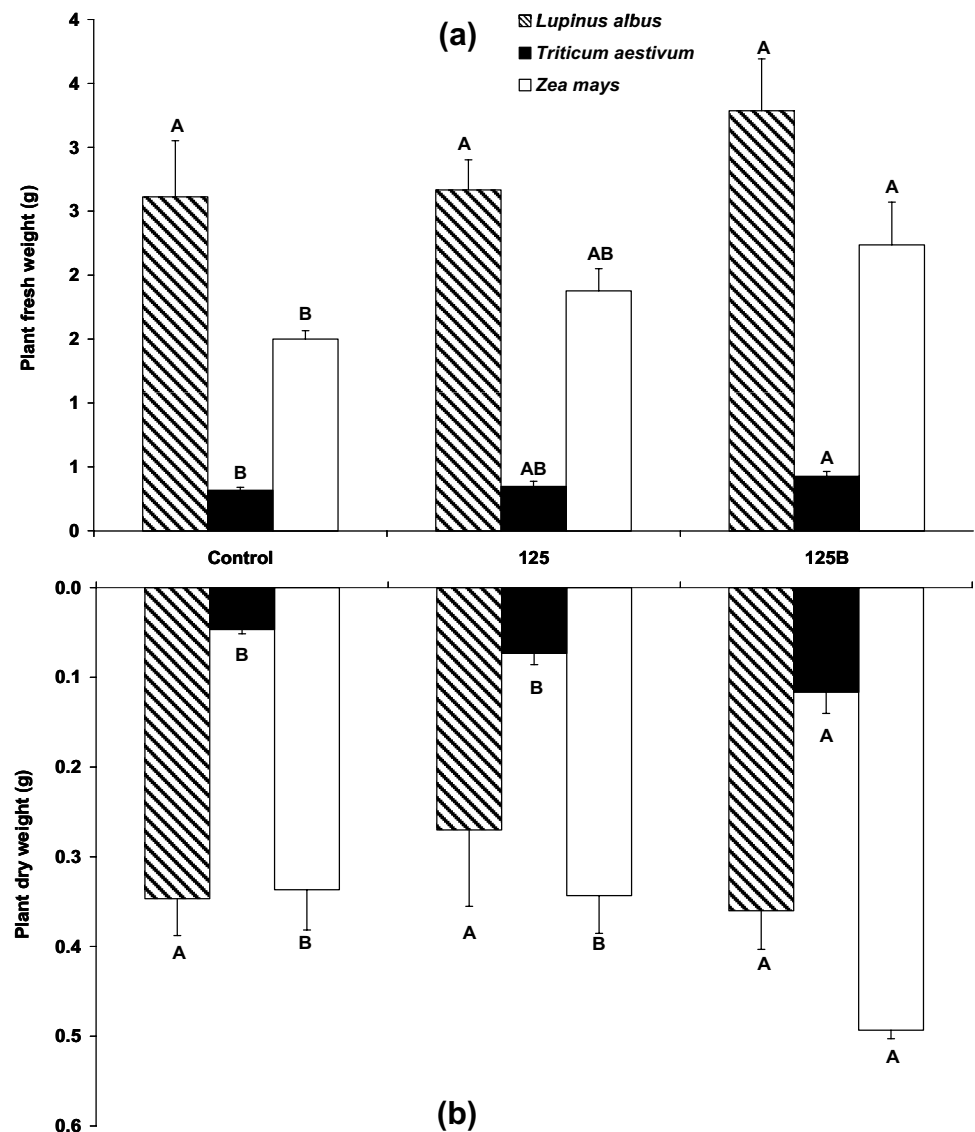
Total carbohydrates content

The data of plumule, and radicle total carbohydrates (Fig. 6a, b), using the control as a reference, demonstrated that at biosorption of Fe^{2+} at 125 mg/L slightly increase in shoot total carbohydrates of *Lupinus albus*, regardless the same treatment at *Triticum aestivum*, and *Zea mays*. However, the root total carbohydrates slightly increased in *Triticum aestivum*, and *Zea mays*. In the cells of those fungi, Fe^{3+} ions resulted in a decrease in monosaccharide content. when compared to control, proving that high levels of Fe^{3+} are toxic to *R. mucilaginosa*. Fe^{3+} ions are required for yeast metabolism and perhaps pathogenicity, indicating that at lower Fe^{3+} concentrations, the quantity of each saccharide and protein within the cell is kept at a manageable level. Glucose and its amine derivatives have been shown to cause virulence in other yeasts, such as *Candida albicans* (Konopka 2012; Rodaki et al. 2009). Therefore, it looks that waters with a high quantity of Fe^{3+} ought to show explicit sanitary threats.

Total proteins content

It is clearly shown from Fig. 7a that, using the control as a reference, the plumule total proteins were decreased in all

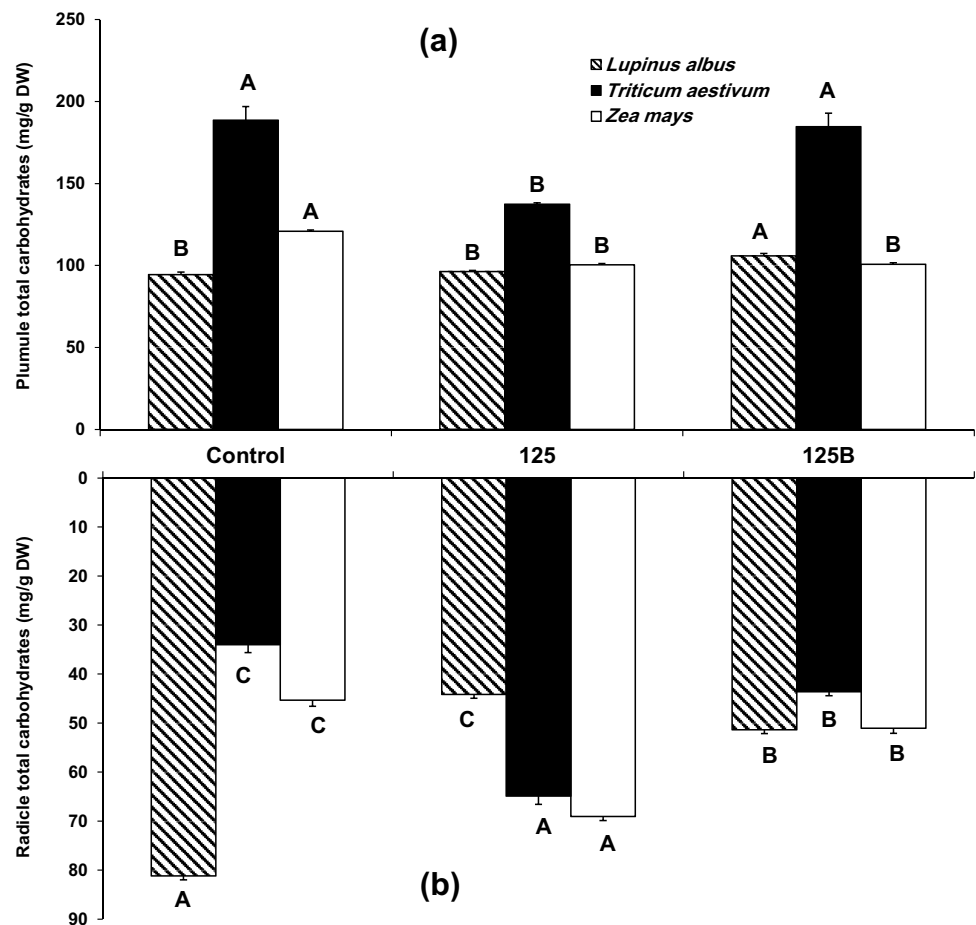
Fig. 5 Seedling undried and dehydrated weights of 20-day old *Lupinus albus*, *Triticum aestivum* and *Zea mays* seedlings as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125 and 125B ppm); B: biosorbed by *R. mucilaginosa*. Different letters (A–B) on the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)



plants at 125 mg/L Fe^{2+} treated with or without biosorption. Still, from Fig. 7b, the total radicle proteins were increased in *Triticum aestivum*, and *Zea mays* seedlings with a slight decrease in the case of *Lupinus albus*. Because of the properties of iron, oxidation–reduction characteristics and its capability to make complexes with numerous ligands, this part may be a component of enzymes and many-electron carriers; thus, It plays a crucial part in the metabolism of plants. On the opposite hand, the low Fe^{2+} solubility in physiological conditions hydrogen ion concentration levels and its high reactivity within the presence of an element that generates hydroxyl harmful radicals denote a serious problem (Hell and Stephan 2003). Fe plays also an indispensable job in protein metabolism. Through iron shortage, the proteins decrease instantaneously and a rise in soluble organic nitrogen compounds. Nitric oxide is a messenger in various cellular physiological routes in plants (Neill et al. 2003).

N_2 -fixation is aided by Fe. and deoxyribonucleic acid repair (Møller et al. 2007). It causes several important functional teams to be replaced at high doses, including lipid peroxidation, cellular damage, and the formation of reactive gas species (Lowry et al.), disturbance within the numerous metabolic reaction by sterilization the catalyst activity (Anjum et al. 2015; de Oliveira Jucoski et al. 2013). Iron is cytotoxic once high levels accumulate. It will be catalyzed by the reaction of Fenton to develop radicals, which might injure lipids, proteins, and DNA. Therefore, plants should reply to iron stress regarding each iron deficiency and iron-storage disease. The incidence of iron toxicity is also influenced by plant species and growth media-related parameters such

Fig. 6 Plumule and radicle total carbohydrates of 20-day old *Lupinus albus*, *Triticum aestivum* and *Zea mays* seedlings as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125 and 125B ppm); B: biosorbed by *R. mucilaginosa*. Different letters (A–C) on the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)



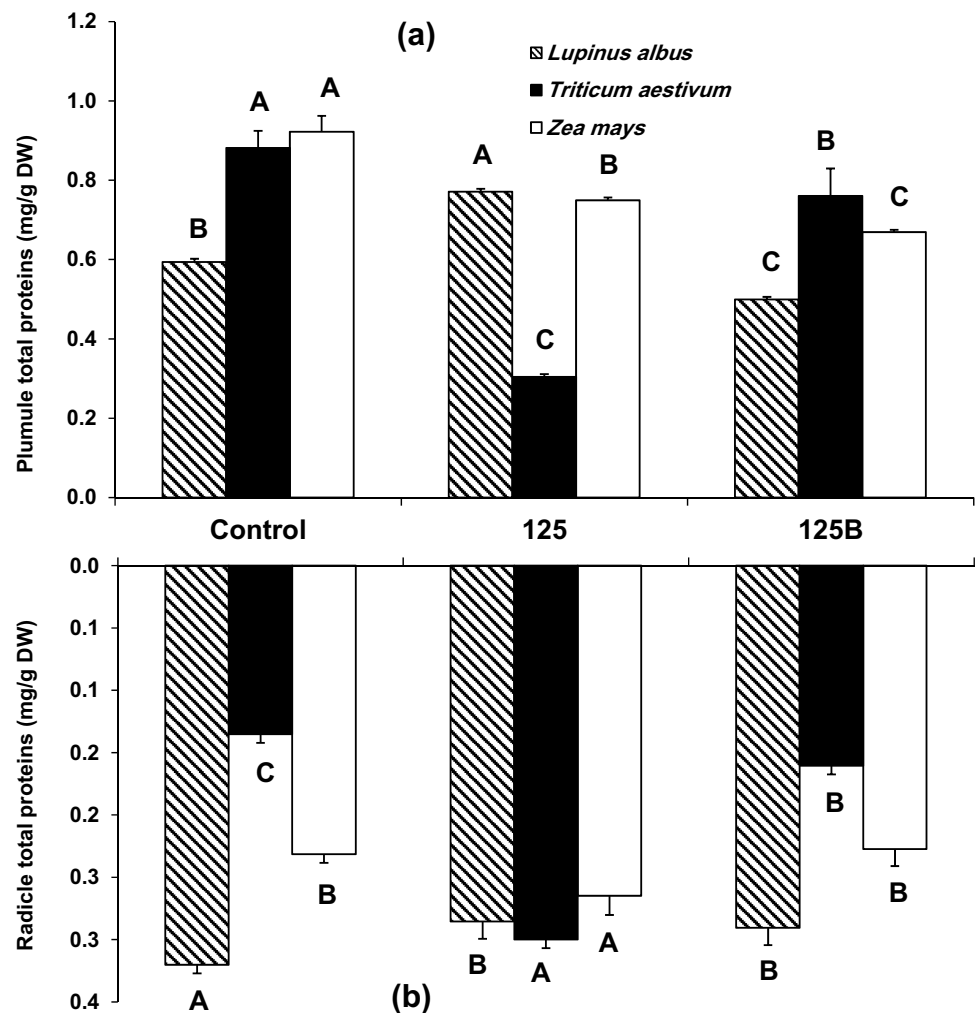
as plant age, sulphide buildup, organic acids, and various reduction products (De Datta et al. 1993; Sahrawat 2005).

Pigment contents

Figure 8a shows the chloroplast pigments of *Lupinus albus* of 20-day old, at the treatment of biosorption of Fe^{2+} at 125 mg/L neither Chl a nor Chl b exhibit any increasing the slight increase was observed in carotenoids. Figure 8b shows the chloroplast pigments of *Triticum aestivum* of 20-day old, at the treatment of Fe^{2+} at 125 mg/L a noticeable increase in Chl b and total pigments were observed but, the response was reserved with the treatment of biosorption of Fe^{2+} at 125 mg/L regardless, the quantity of carotenoids was increase comparing to control. Figure 8c shows the chloroplast pigments of *Zea mays* of 20-day old, there is a slight decrease in all results at the treatment of Fe^{2+} at 125 mg/L compared to control, but at biosorption of Fe^{2+} at 125 mg/L the same trend was observed. Photosynthesis cells contain approximately eightieth of Fe^{2+} wherever it's essential for the biogenesis of cytochrome molecules, pigment, the electron transport mechanism, and so Fe-S cluster formation (Briat et al. 2007; Hänsch and Mendel 2009). Within the

photosynthetic equipment, in compounds directly related to photosystem II (PS-II), two or three iron atoms are detected, whereas photosystem I has twelve atoms (PS-I), 2 within the ferredoxin and 5 within the hemoprotein complicated (Varotto et al. 2002). Iron allocations are directly concerned with the chemical action of plant's productivity (Briat et al. 2007). Iron may be probably harmful at high concentrations. Because of iron's ability to provide and accept electrons, if it is present in the cell, it will convert peroxide molecules into free harmful radicals. These radicals will harm a healthy form of cellular structure, eventually leading to cell death (Crichton et al. 2002). To prevent this type of harm, life forms have evolved a metabolic protective mechanism that involves protein-iron atom interaction. Plants use antioxidant enzymes and metabolites like ascorbic acid and carotenoids to guard against the harmful effects of reactive radicals on cellular and subcellular structures (Alscher et al. 1997). Excess iron quantities, according to Monteiro and Winterbourn (1988), should have stimulated the creation of active O_2 atom species, which might have changed the chlorophyll and led to a reduction in chlorophyll content. With increased iron concentrations, there is a decrease in chlorophyll and antioxidant content as reported by Arunachalam

Fig. 7 Plumule and radicle total proteins of 20-day old *Lupinus albus*, *Triticum aestivum* and *Zea mays* seedlings as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125 and 125B ppm); B: biosorbed by *R. mucilaginosa*. Different letters (A–C) on the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)



et al. (2009). Because of this pattern, the homogeneity of pigment creation (Wilkinson and Ohki 1988). Carotenoids also serve as antioxidants vital aiding metabolites saving the subcellular and cellular organizations from the toxic properties of those reactive oxygen species (Sarika et al. 2010) by inhibiting the singlet oxygen (Prasad and Bisht 2011). Hemalatha and Venkatesan (2011) found that as iron levels dropped, pigments like chlorophyll and carotenoid levels dropped as well.

Alpha and beta-amylase activities

It is clear from results indicated in Fig. 9a, b that activity of alpha and beta amylase of 2-day old seeds at 125 mg/L Fe^{2+} treated with or without biosorption were higher than control in *Triticum aestivum* seeds but, in *Zea mays*, besides *Lupinus albus* with little adverse effect in both treatments. The existence of a Fenton's catalyst, such as iron or copper ions, which gives birth to highly reactive OH^- radicals in the occurrence of H_2O_2 and O_2^- , determines the toxicity of reactive oxygen species (Chen et al.

1999). DNA, proteins, lipids, chlorophyll, and practically every other organic component of living cells are destroyed by harmful oxygen reactive species (Becana et al. 1998). Amylase is an enzyme involved in the metabolism of carbohydrates. Hofner (1970) discovered that as iron concentrations increased, amylase activity reduced, which he attributed to iron forming complexes with carbohydrate molecules like maltose in plants. Excess iron concentrations also harmed the enzymes amylase, invertase, aspartate aminotransferase, and glutamate synthase in tea plants, according to Hemalatha and Venkatesan (2011). It appears that for yeast, 5 mg/L Fe^{3+} ions are already venomous and cause defensive responses to worry. Extracellularly secreted proteins most likely bind metal ions in the atmosphere or transformed the metals into harmless forms. As a result, they reduce the quality of their products and avoid penetrating cells. The active creation of various exo-metabolites by fungus led to the distribution of metal ions in the aquatic environment varies dramatically as a result of this, as well as their quality, bioavailability, and toxicity (Kolenčik et al. 2014). The creation of reactive

Fig. 8 Pigment contents of the leaves of 20-day old *Lupinus albus*, *Triticum aestivum* and *Zea mays* seedlings as affected by (Fe.²⁺) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125 and 125B ppm); B: biosorbed by *R. mucilaginosa*. Different letters (A–C) on the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)

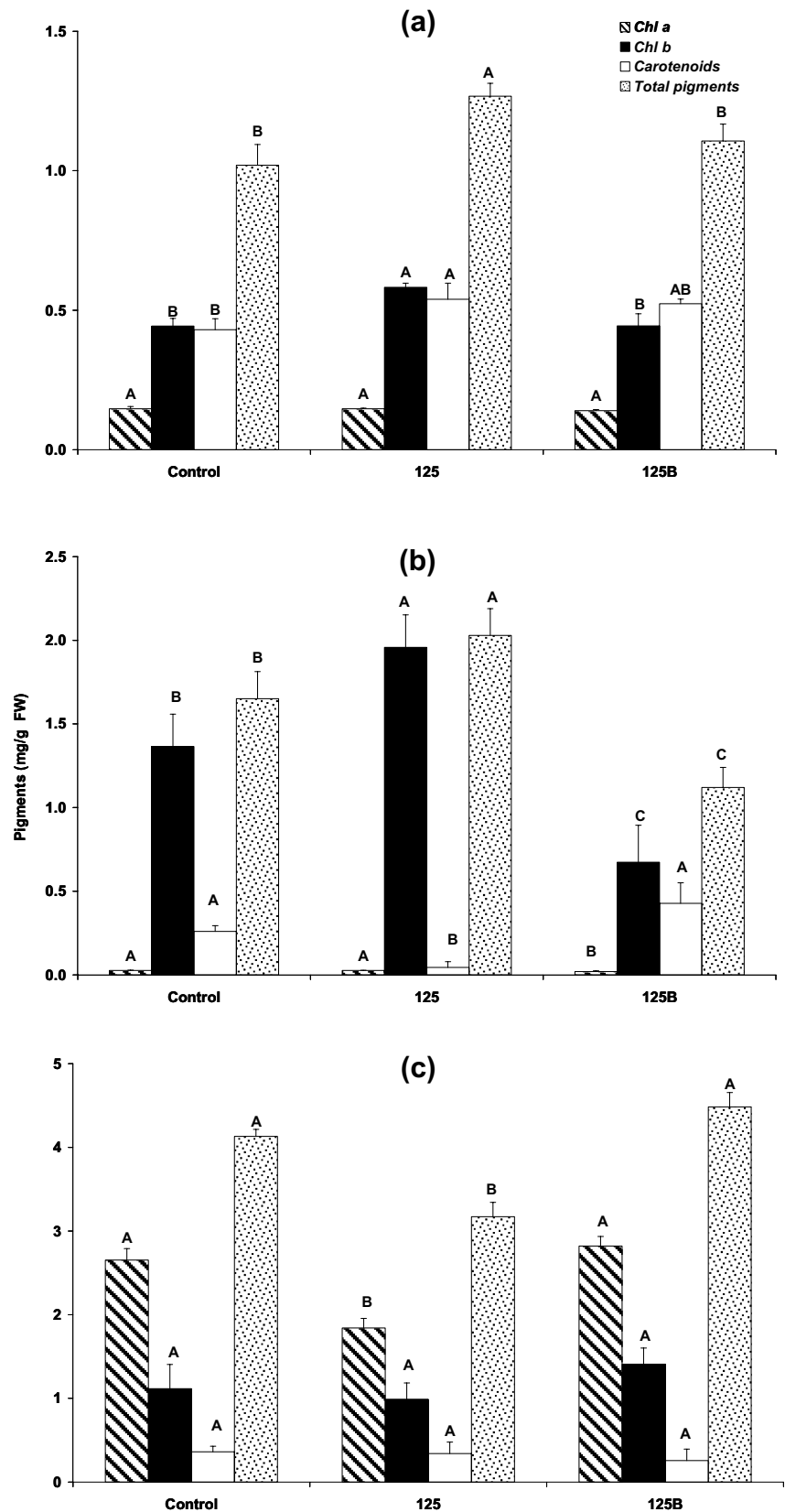
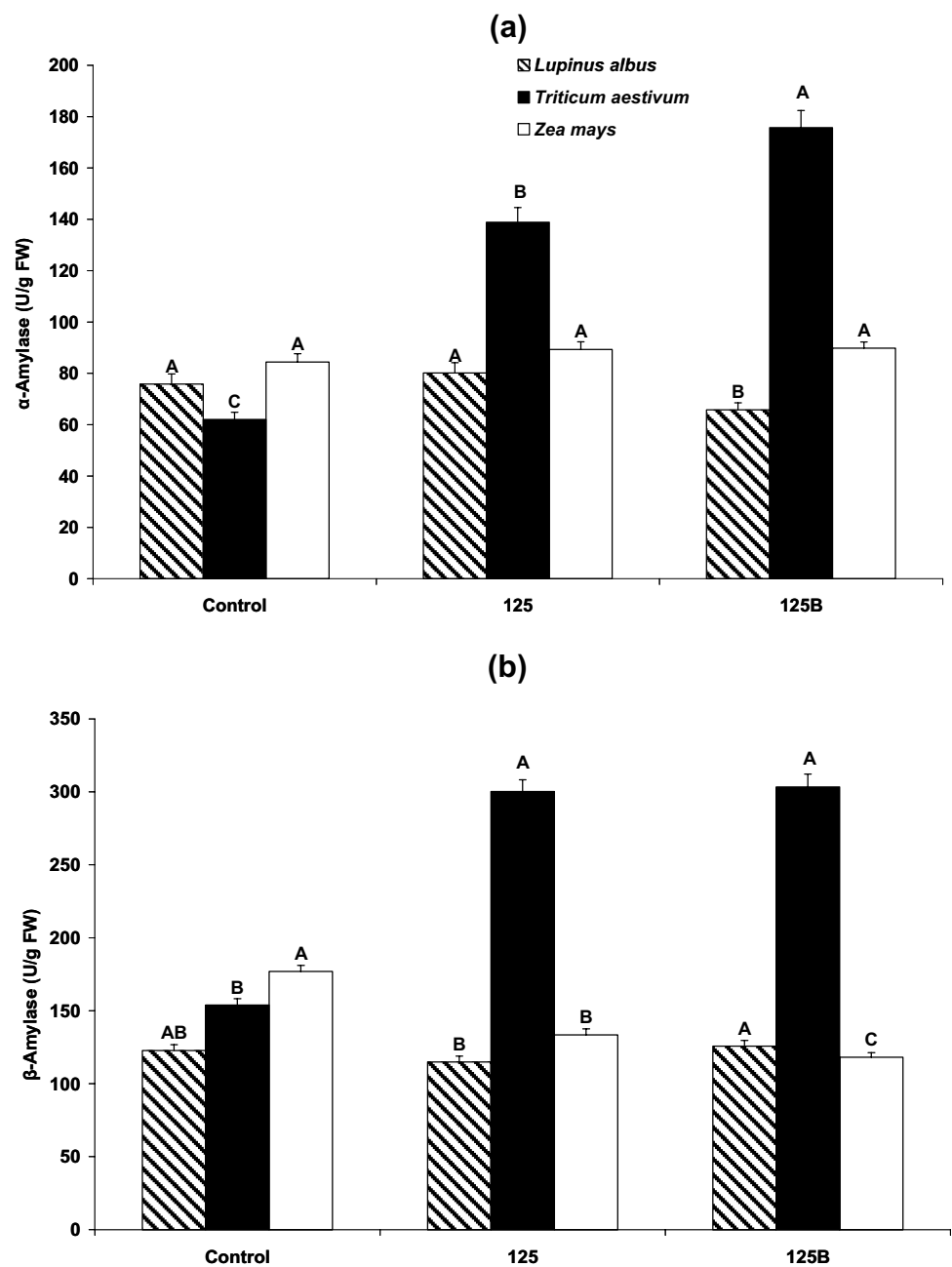


Fig. 9 Alpha (α) and β amylase activities of 2-day old *Lupinus albus*, *Triticum aestivum* and *Zea mays* seeds as affected by (Fe^{2+}) concentrations biosorbed by *R. mucilaginosa* YMM19 and unbiosorbed (Control, 125 and 125B ppm); B: biosorbed by *R. mucilaginosa* Different letters (A–C) on the bars indicate significant differences according to the least significant difference (LSD) test ($P \leq 0.05$)



atomic number 8 species is one of the processes by which cells respond to anxiety produced by heavy metals (Lowry et al. 1951) and therefore the connected aerophilic stress (Azevedo et al. 2007; Pradhan et al. 2015). It was discovered that when higher quantities of Fe^{3+} ions were present in the media, the activity of specified inhibitor enzymes increased in yeast *R. mucilaginosa* cells. This indicates that Fe^{3+} ions have a poisonous effect on plant cells. Moreover, Fe^{3+} ions within the medium within which strains of yeast ended up in stress responses (an increase

in inhibitor enzyme activity and a decrease in the quantity of proteins and simple carbohydrates), and biosorption of Fe^{3+} by cells significantly decreases. In *R. mucilaginosa* cells, intense absorption of Fe^{3+} ions with no stress impact was determined at low concentrations of Fe^{3+} ions in the culture media (up to 1 mg/L). Consequently, Fe^{3+} ions at a level of 1 mg/L enhance *R. mucilaginosa* metabolism. At levels higher than 5 mg/L in the medium, the yeast produced a cellular mechanism that prevented the intake ions of Fe^{3+} (Cudowski and Pietryczuk 2019).

Conclusion

The current study found that biosorption of Fe²⁺ using the *R. mucilaginosa* YMM19 fungus helped iron-stressed seedlings improve their development and metabolic activity.

Author contribution statement Contribution of co-authors: together authors participated equally to this study. YM: and WG: conceived the idea of the study and conducted the experiments. YM: and WG: interpreted the results and wrote the current manuscript. The final form of the manuscript was revised by YM: and WG: who both agreed on it.

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