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# Cobalt Alters the Seedling Vigour on *Oryza sativa* (L.). Under Soilless Cultivation

Kaliyamoorthy Jayakumar<sup>1,\*</sup> and T. Nedumaran<sup>2</sup>

<sup>1</sup>Department of Botany, A.V.C College (Autonomous), Mannampandal 609 305, Tamil Nadu, India

<sup>2</sup>Department of Plant Biology and Plant Biotechnology, Sir Theagaraya College, Chennai 600 021, Tamil Nadu, India

\* Author to whom correspondence should be addressed; E-Mail: jk81botany@yahoo.com

Article history: Received 13 March 2019, Revised 26 July 2019, Accepted 29 July 2019, Published 5 August 2019.

**Abstract:** Application of cobalt alters the seedling vigour on *Oryza sativa* (L.). under Soilless cultivation. All the germination percentage, vigour index, tolerance index, percentage of phytotoxicity and growth parameters, such as, root length, shoot length and dry weight were increased at 5mg l<sup>-1</sup> cobalt concentration of paddy plants. The above parameters were decreased at higher (10, 25, 50, 75 and 100mg l<sup>-1</sup>) concentrations of cobalt solution.

**Keywords:** Cobalt, Seed germination, Growth parameters, *Oryza sativa*, Vigour index, Tolerance index, Percentage of phytotoxicity

## 1. Introduction

Heavy metals are defined as metallic elements that have a relatively high density compared to water (Fergusson, 1990). With the assumption that heaviness and toxicity are inter-related, heavy metals also include metalloids, such as arsenic, that are able to induce toxicity at low level of exposure (Jayakumar and Viyayarengan, 2006a). Results of these activities end up in outlets and wastes where heavy metals are transported to the environment by air, water or deposits, thereby increasing the metals concentrations in the environment. For example, the metal concentration in river water has been shown to be increased several thousand folds by effluents from mining wastes (Jayakumar *et al.*, 2006 and

Jayakumar and Vijayarengan, 2014). Metals supplied to the environment are transported by water and air ultimately reaching the soil and sediment where they become bound. However, the time taken for them to become bound may be fairly long and it has been shown that the bioavailable fraction of metals in soils is high at the beginning of the binding period, but decreases with time (Martin and Kaplan, 1998). Thus, there is probably a bigger problem with anthropogenically supplied metals, with high levels of bioavailable metals, than with high background levels originating from bedrock with slow weathering (Jayakumar and Vijayarengan, 2009a).

### 1.1. Application of Cobalt

Cobalt and its salts are used in variety of processes to make superalloys which maintain their strength at high temperature, in paint as a dryer, in porcelain enamel finishes as a drying agent, as an ingredient of coloured pigments and in formulating vitamin B<sub>12</sub>. Some radioactive isotopes of cobalt, such as cobalt 60, are used in treating patients in nuclear medicine and in research.

*Oryza sativa*, rice, is originated in India, Thailand, and southern China, was domesticated and diversified in ancient times, and is now cultivated in wet tropical, semi-tropical, and warm temperate areas around the world for the production of its cereal grain. Rice is one of the two most important cereal crops world for human consumption; the other is wheat, *Triticum* species. Rice is cultivated on an estimated 3% of the world's agricultural land, and serves as a primary source of calories for over half the world's population. Rice has also been important as a model system in plant biology, and is the first plant species for which the genome has been fully mapped.

*Oryza sativa* has hundreds of cultivars with different grain color, size, and shape, as well as environmental tolerances and seasonality the types are generally categorized as valley rice, upland rice, spring rice, and summer rice. It is generally grown in fields that are flooded for part of the growing season whether from irrigation, rain fed or floodplain systems which help reduces competition from other plants, among other benefits; some upland varieties can be grown without flooding, but they account for only 4% of rice cultivated worldwide.

Keeping these points in view the present investigation has been made to study the extent of cobalt tolerance and toxicity in paddy (*Oryza sativa* L.) cultivar ADT-38.

### 1.2. Laboratory Studies

Laboratory experiments were conducted to assess the different concentrations (0, 5, 10, 25, 50, 75 and 100 mg l<sup>-1</sup>) of cobalt on seed germination, seedling growth, vigour index, tolerance index and percentage of phytotoxicity of paddy.

## 2. Materials and Methods

The present investigation has been carried out to find out the effect of cobalt on seed germination and growth of paddy (*Oryza sativa* L.) cultivar ADT-38.

### 2.1. Materials

#### 2.1.1. Seed

The experimental plant, the paddy (*Oryza sativa* L.) cultivar ADT-38 belongs to the family Poaceae is one of the important crops of India. Seeds used in the experiments were obtained from the Tamil Nadu Rice Research Institute, Tamil Nadu Agricultural University, Aduthurai. Seeds with uniform size, colour and weight were chosen for experimental purpose.

### 2.2. Laboratory Experiments

#### 2.2.1. Preparation of cobalt chloride solution

Cobalt chloride ( $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ ) (M.W = 237.93) is used as a cobalt source. 4.038 g cobalt chloride was dissolved in 1000 ml of distilled water. It is equal to 1000 mg  $\text{l}^{-1}$  Cobalt. From this solution the following concentrations 0, 5, 10, 25, 50, 75 and 100 mg  $\text{l}^{-1}$  were prepared and used for the laboratory experiments.

#### 2.2.2. Seed germination

Seeds were surface sterilized with 0.1 per cent mercuric chloride solution and washed thoroughly with tap water and then with distilled water.

The sterilized seeds were placed equispacially in sterilized petridishes, lined with filter paper. Each petridish was irrigated uniformly by various concentrations of the heavy metal solution in the respective petridishes. In addition to this, petridish containing seeds were irrigated with distilled water and were maintained as control.

All the petridishes were kept under diffused light at room temperature ( $28 \pm 2^\circ\text{C}$ ). The number of seeds germinated in each treatment was counted on the 8<sup>th</sup> day, after which no further germination occurred. The total germination percentage was calculated. The emergence of radicle was taken as a criterion for germination. Six seedlings from each replicate were selected for recording the morphometrical parameters such as length of root and shoot and dry weight of root and shoot.

#### 2.2.3. Dry weight

The plant samples were kept in an oven at  $80^\circ\text{C}$  for 24 hours then the samples were kept in a desiccators and the dry weights were taken using electrical single pan balance.

#### 2.2.4. Vigour index

Vigour index of the seedlings was calculated by using the formula proposed by Abdul Baki and Anderson (1973).

$$\text{Vigour Index} = \text{Germination percentage} \times \text{length of seedlings.}$$

#### 2.2.5. Tolerance index

Tolerance index of the seedling was calculated by using the formula proposed by Turner and Marshal (1972).

$$\text{Tolerance index} = \frac{\text{Mean length of longest root in treatment}}{\text{Mean length of longest root in control}}$$

#### 2.2.6. Percentage phytotoxicity

The percentage phytotoxicity of heavy metal solution was calculated by using the following formula (Chou *et al.*, 1978).

$$\text{Percentage phytotoxicity} = \frac{\text{Radicle length of control} - \text{radicle length of test}}{\text{Radicle length of control}} \times 100$$

(the negative value was taken as stimulation effect)

### 2.3. Statistical Analysis

The statistical analyses of the experimental data were carried out as per the procedure given by Gomez and Gomez (1984).

## 3. Experimental Results

In the present investigation, changes in growth of paddy (*Oryza sativa* L.) cultivar ADT-38 have been studied under cobalt stress.

### 3.1. Laboratory Studies

#### 3.1.1. Germination percentage

Germination percentage values of paddy under cobalt stress are presented in Table 1. The germination percentage of paddy was found to be 100% in 0, 5 and 10 mg l<sup>-1</sup> and it decreased with an increase in cobalt concentrations.

#### 3.1.2. Root length (cm plant<sup>-1</sup>)

Root length of paddy was maximum at 5 mg l<sup>-1</sup> (3.98). When compared with control (3.11). While the minimum root length was found at 100 mg l<sup>-1</sup> (1.18) (Table 1).

### 3.1.3. Shoot length (cm plant<sup>-1</sup>)

The shoot length of paddy was found to be 16.18, 17.89, 15.43, 15.13, 12.69, 10.73 and 8.98 at the various concentrations 0, 5, 10, 25, 50, 75 and 100 mg l<sup>-1</sup> respectively (Table 1).

### 3.1.4. Dry weight of seedlings (g plant<sup>-1</sup>)

#### 3.1.4.1. Root

Dry weight of roots of PADDY seedlings is expressed in Table 1. The result indicated that the maximum root dry weight was observed at 5 mg l<sup>-1</sup> of paddy (0.10). The minimum dry weight of root was recorded at 100 mg l<sup>-1</sup> (0.02).

#### 3.1.4.2. Shoot

Effect of cobalt on shoot dry weight of paddy seedlings is compiled in table 1. Cobalt at 5mg l<sup>-1</sup> (0.22) showed the maximum shoot dry weight of the seedlings when compared to the control (0.19). The shoot dry weight of paddy seedlings decreased further with an increase in the concentration of cobalt.

**Table 1.** Effect of cobalt on seed germination, seedling growth (cm plant<sup>-1</sup>) and dry weight (g plant<sup>-1</sup>) of *Oryza sativa* (L.)

Cobalt concentration (mg l <sup>-1</sup> )	7 <sup>th</sup> day				
	Germination percentage	Length (cm)		Dry weight (g/ 10 pts)	
		Root	Shoot	Root	Shoot
0	100	3.11	16.18	0.09	0.19
5	100	3.98 (- 27.97)	17.89 (- 10.56)	0.10 (- 11.11)	0.22 (- 15.78)
10	100	2.94 ( 5.46)	15.43 (4.63)	0.08 (11.11)	0.18 (5.26)
25	98	2.46 (20.90)	15.13 ( 4.635)	0.07 ( 22.22)	0.16 (15.78)
50	95	1.93 ( 37.94)	12.69 (21.56)	0.05 ( 44.44)	0.13 (31.57)
75	93	1.57 (49.51)	10.73 (33.68)	0.03 ( 66.66)	0.11 ( 42.10)
100	91	1.18 ( 62.05)	08.98 (44.49)	0.02 ( 77.77)	0.9 ( 56.39)

(Per cent over control values are given in parentheses)

### 3.1.5. Vigour index

The effect of cobalt on the vigour index of paddy was found to be maximum at 5 mg l<sup>-1</sup> (2187) and minimum in 100 mg l<sup>-1</sup> (1016). When the concentrations of cobalt increases, the vigour index of the seedlings decreased (1837, 1721, 1388, 1143 and 1016) in the order of 0, 5, 10, 25, 50, 75 and 100 mg l<sup>-1</sup> respectively (Table 2).

**Table 2.** Effect of cobalt on vigour index, tolerance index and percentage phytotoxicity of *Oryza sativa* (L.)

Cobalt concentration (mg l <sup>-1</sup> )	Vigour index	Tolerance index	Percentage phytotoxicity
Control	1929	-	-
5	2187 (-13.37)	1.27	1.830
10	1837 (4.769)	0.94	2.164
25	1721 (10.78)	0.79	2.319
50	1388 (28.04)	0.62	2.489
75	1143 (40.76)	0.55	2.605
100	1016 (47.33)	0.37	2.730

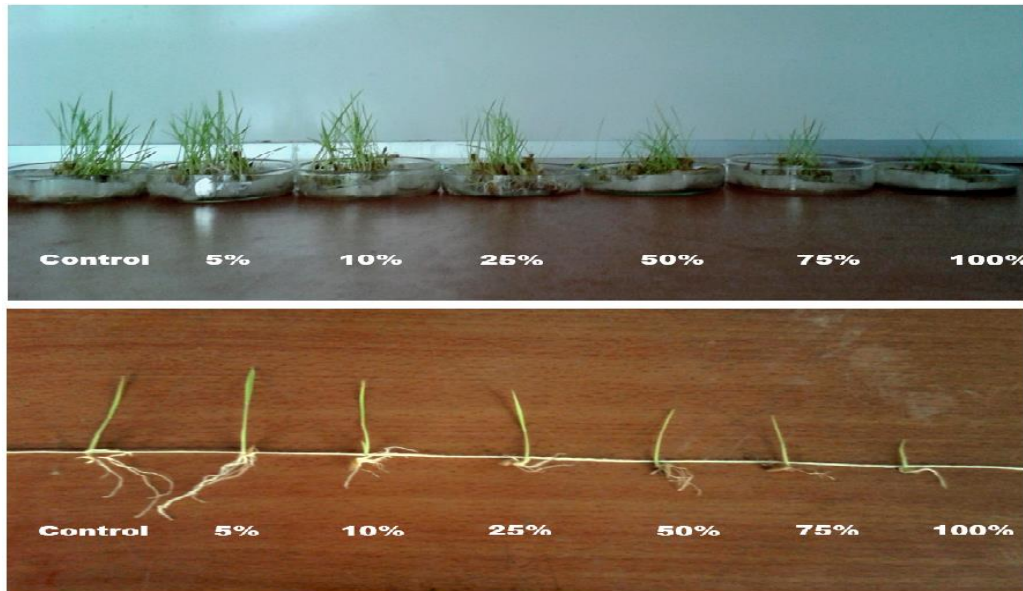
(Per cent over control values are given in parentheses)

### 3.1.6. Tolerance index

Table 2 showed the tolerance index of paddy seedlings under cobalt stress. The maximum tolerance index of paddy was observed in 5 mg l<sup>-1</sup> (1.27) and minimum in 100 mg l<sup>-1</sup> (0.37).

### 3.1.7. Percentage phytotoxicity

The effect of cobalt on the percentage phytotoxicity of paddy seedlings was furnished in Table 2. Percentage phytotoxicity of paddy seedlings was found to be 1.830, 2.164, 2.319, 2.489, 2.605 and 2.730 at 0, 5, 10, 25, 50, 75 and 100 mg l<sup>-1</sup> respectively.



**Plate 1:** Effect of cobalt on seed germination and seedling growth ( $\text{cm plant}^{-1}$ ) of *Oryza sativa* (L.)

## 4. Discussion

The present investigation has been carried out to find out the effect of cobalt on seed germination and growth of (*Oryza sativa* L.) cultivar ADT-38. The germination studies were conducted in the Ecology Laboratory, Department of Botany, A .V.C College.

### 4.1. Laboratory Studies

#### 4.1.1. Seed germination

The germination percentage of paddy plants increased at  $5 \text{ mg l}^{-1}$  cobalt level and decreased at high levels of cobalt ( $10\text{-}100 \text{ mg l}^{-1}$ ). Reduction in germination percentage of paddy at higher concentration may be attributed to the interference of cobalt ions. Low concentration of cobalt increase the germination and growth was observed by Jayakumar *et al.* (2008b) Similar inhibition of germination at higher concentration was observed by Pokhrel and Leahak (2001) with lead, Hameed *et al.* (2001) with copper and lead and Mahalakshmi and Vijayarengan (2003) with zinc, Jayakumar and vijayarengan (2006) with cobalt.

#### 4.1.2. Seedling growth

Seedling growth of paddy showed a progressive decline with increase in cobalt concentration. However,  $5 \text{ mg l}^{-1}$  cobalt levels were favorable for the seedling growth. The seedling growth parameter like root and shoot lengths were reduced at higher cobalt levels. Similar results were reported on the effect of aluminium (Shen *et al.*, 1993), cadmium (Kalita *et al.*, 1993; Saravanan *et al.*, 1997), chromium (Corradi *et al.*, 1993), cobalt and zinc (Burhan, 2001), mercury (Ravimycin, 1995) and nickel (Senguttuvel *et al.*, 1998), Jayakumar *et al.*, (2009a&b & 2013a).



#### 4.1.3. Dry weight

The dry weight of paddy seedlings decreased gradually with increase in cobalt concentration. It increased at 5 mg l<sup>-1</sup> cobalt concentration. Minimum dry weight of seedlings was recorded at 200 mg l<sup>-1</sup> cobalt concentration. Dry weight of paddy seedling showed a gradual decline from 10 mg l<sup>-1</sup> onwards. Similar reduction in seedling dry weight under various metal treatments was observed by Dinew and Stancheva (1993) on wheat, rye and triticale and Subramani *et al.* (1997) on blackgram. The dry phytomass yield decrease at higher levels of zinc might be due to poor growth of seedlings and which agrees with the results of Rauno *et al.* (1988) on bushbeans, Aery and Sarkar (1991) on soybean and Jayakumar and Vijayarengan (2006) on blackgram.

Results obtained from the germination studies indicated that the paddy showed higher germination percentage, seedling growth and dry weight at 5 mg l<sup>-1</sup> cobalt level. The values of growth parameters indicated that cobalt had a significant stimulating, beneficiary and nutritional effect at 5 mg l<sup>-1</sup> concentration for paddy. The growth process beyond the concentration indicated that a little excess of cobalt above these levels had an adverse effect. Heavy metals at lower concentrations have been found to stimulate the germination process and seedling growth and to inhibit the growth at higher concentrations was observed by Jayakumar *et al.* (2013b). Similar trend was observed by Balashouri and Prameeladevi (1995) for *Vigna radiata* and *Sorghum bicolor*, Subramani *et al.* (1997) for *Vigna mungo*, Senguttavel *et al.* (1998) for *Arachis hypogaea*, Hemalatha and Francis (1998) for *Oryza sativa* and Jayakumar and Vijayarengan (2006) on blackgram.

## 5. Summary

The present investigation deals with the effect of cobalt on seed germination and seedling growth of paddy plants.

### 5.1. Germination Studies

Paddy (ADT-38) seeds were germinated in different concentrations (5, 10, 25, 50, 75 and 100 mg l<sup>-1</sup>) of cobalt and it was compared with control plants irrigated with distilled water (0 mg l<sup>-1</sup>). The germination parameters like germination percentage root and shoot length, dry weight of seedlings, vigour index and tolerance index showed a decreasing trend with an increase in cobalt concentrations. Whereas the percentage of phytotoxicity value increased with an increase in the concentration of cobalt. However the 5 mg l<sup>-1</sup> concentration of cobalt proved to be beneficial for the various germination parameters of paddy.

## 6. Conclusion



From the present investigation it was concluded that the 5 mg l<sup>-1</sup> level of cobalt solution was beneficial for the growth of paddy crops. The level of cobalt solution above 5 mg l<sup>-1</sup> proved to be toxic. The results indicated that the 5 mg l<sup>-1</sup> cobalt level can be applied for increasing the germination and growth of paddy crops.

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