



Influence of organic manure, Minia Azotein, mycorrhizae fungi and active yeast application on growth and chemical constituents of *Taxodium distichum* seedlings

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Abstract

This research was conducted during the two consecutive seasons of 2017/2018 and 2018/2019 to study the effect of Filter mud as organic manure and biofertilizers application, as well as, their interaction on growth and some chemical constituents of baldcypress (*Taxodium distichum*) seedlings. Filter mud was applied at 0, 24, 48, and 72 g/bag. Biofertilizers were control, Active Yeast (AY), Minia Azotein (M.A.), Arbuscular mycorrhizae fungi (AM), AY+ M.A., AY+AM, M.A.+AM and AY+M.A.+AM. The most obtained results were as follows: The addition of filter mud at all levels led to a significant increase in plant growth traits (stem length, stem diameter, leaves fresh and dry weights /plant and stem fresh and dry weights /plant, as well as, the elements of N, P and K % in the leaves), as compared to the check treatment. Utilizing the high level of filter mud (72 g/bag) registered the highest values of these parameters. In relation to biofertilizers treatments, all of them either separately or in combination, except for active yeast, in some cases, led to a significant augment in these previous traits. The application of triple inoculants (AY+ M.A. +AM) proved to be more effective in increasing these above mentioned parameters. In addition, for the interaction, it was significant effect on all examined characters, except for stem diameter. Clearly, most of combined treatments significantly increased all studied traits comparing to untreated ones. The most effective treatments were detected by using filter mud at the high level with the triple inoculants (AY+ M.A. +AM) in comparison with those obtained by other combination treatments.

Keywords: *Taxodium distichum*, filter mud, Minia Azotein, mycorrhizae fungi, active yeast.

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1. Introduction

Baldcypress (*Taxodium distichum*) is a native and deciduous conifer. It is one of the most famous conifer trees because of its economic and ecological importance, as well as, it's highly tolerant to flooding and logging of soil water (Allen *et al.*, 1996; Larzen, 1980; Wilhite and Toliver, 1990). It belongs to family Taxodiaceae which contains 10 genera and 16 species of cone bearing trees native to North America, Asia and Tasmania. The leaves and seeds of *Taxodium distichum* L. Rich. have been used for the treatment of malaria and liver diseases (Sharaf, 1928) and the resins obtained from its cones are used as an analgesic for skin ailments. The essential oil isolated from the female cones is anti-inflammatory and antispasmodic effects, as well as, antibacterial and antifungal activities (El-Tantawy *et al.*, 1999). The wood of *Taxodium* is durable and highly resistant to decay, making it valuable for multiple uses viz. building construction, fine furniture, planking in boats, doors, floorings and cabinetry (Collingwood 1937 and Walker, 1990). Nowadays, organic agriculture is becoming a fermentative alternative to chemical fertilizers because organic manures are very safe for human, animal and environment. Follet *et al.* (1981) suggested the positive roles of organic manure for example: Improve soil properties, water holding capacity and accelerate releasing essential nutrients by microbial decomposition. Filter mud as a local organic fertilizer, it's very beneficial in enhancing the plant growth and productivity. Organic manures contain

growth promoting hormones *i.e.* IAA and GA, macronutrients, essential micronutrients and beneficial microorganisms (Natarajan, 2007; Sreenivasa *et al.*, 2010). The promotion effect of organic manure on improving the growth and some chemical constituents was studied by Ali *et al.* (2001) and El-Sayed and Abdou (2002) on *Khaya senegalensis*, Ahmadloo *et al.* (2012) on cypress seedlings, Abdou *et al.* (2007) on *Ficus elastica* var. Decora Mahmmoud (2014) on *Populus nigra* and Abou El-Makarem (2016), Abdel-Salam (2017) and Hussein (2019) on moringa. Biofertilizers (microbial inoculants) act an important role in improving soil fertility by increasing number of microorganisms and accelerate certain microbial processes in rhizosphere zone. Such microbiological processes can convert the nutrients from the forms of unavailable to available ones (Alaa El-Din, 1982; Rao, 1981). Minia Azotein as biofertilizer containing different strains of N-fixing bacteria. These bacteria capable produce some plant growth hormones (IAA, GA3), thiamin, riboflavin and nicotin (Hartmann *et al.*, 1983; Tien *et al.*, 1979). Phosphate dissolving organisms including mycorrhizae fungi which play an important role on plant growth and chemical constituent traits by production some of growth promotion namely auxins, GA3 and cytokinins, producing phosphate enzymes which are converting organic phosphate to mineral-P which it is possible to absorb by plant, promoting influence on elements absorption (N, S, Zn, Mn, Ca and Cu), protection plant from some diseases and augmenting the immunity against the diseases by

increasing root cortex thickness (Read *et al.*, 1991; Tate, 2001). The positive effect of biofertilizers (N-fixing bacteria and phosphate dissolving organisms) on enhancing the growth and chemical constituent characters was reported by El-Sayed and Abdou (2002), Abdou *et al.* (2006) and Ali *et al.* (2015) on *Khaya senegalensis*, Abdou *et al.* (2004) and Pindi (2011) on *Albizia lebbek*, El-Khateeb *et al.* (2010) on *Chamaedorea elegans*, Ahmed *et al.* (2005) and Mahmmoud (2014) on *Populus nigra*, Abou El-Khair (1993) on *Casuarina glauca* and *Acacia saligna*, Ahmed and Ali (2002) on *Dalbergia sisso* and *Prosopis juliflora* and Abou El-Makarem (2016), Abdel- Salam (2017) and Hussein (2019) on moringa. Active yeast (*Saccharomyces cerevace*) extract is considered as a good source of many natural growth substances (cytokinins), most nutritional elements (P, K, S, N, Ca and Mg), a lot of vitamin B and organic compounds *i.e.* proteins, lipids, carbohydrates and nucleic acids (Nagodawithana, 1991). The enhancement in plant growth and some chemical constituent traits due to applying active yeast was obtained by Ahmed *et al.* (1997) and Akl *et al.* (1997) on grape vine, Ahmed *et al.* (1998) on roselle, Ali (2001) on *Calendula officinalis*, Ahmed *et al.* (2001) on *Ambrosia maritima*, Ahmed (2002) on *Leucaena leucocephala* and Ahmed (2014) on *Ceiba pentandra*. The present work was intended to explore the influence of filter mud as organic manure, Minia Azotein. Mycorrhizae fungi and active yeast, as well as, their interaction on growth and some chemical constituents of baldcypress (*Taxodium*

distichum) seedlings.

2. Materials and methods

This study was conducted during the two successive seasons of 2017/2018 and 2018/2019 at Monshaat Al-Amary, Luxor Governorate, Egypt to examine the influence of organic manure (filter mud) and biofertilization treatments (Minia Azotein, mycorrhizal fungi and active dry yeast), as well as, their interaction on plant growth and chemical constituent parameters of baldcypress (*Taxodium distichum*) seedlings. Baldcypress (*Taxodium distichum*) seedlings were obtained From Horticulture Research Institute, Giza, Egypt. One-year-old of baldcypress seedlings were planted on February 10th for the two seasons in poly ethane bags of 20 × 30 cm. The seedlings were healthy and uniform, average seedling length was 20–23 cm. and seedling diameter was 0.35 – 0.39 cm. for both seasons, respectively. Each bag was filled with 10 kg of a mixture of sand + clay 2:1 (v:v) and contained one seedling. Chemical properties of the experimental soil were determined according to Jackson (1973) and are show in Table (1). The experimental design of this work was a split plot with four replications and five seedlings / replicate, filter mud treatments at four levels of 0, 24, 48 and 72 g/bag were considered as the main plots (A) and the eight biofertilization treatments occupied

the sub-plot (B), therefore the interaction treatments (A × B) were 32 treatments. Filter mud as organic fertilizer was obtained from Sugar Factory, Armant, Luxor Governorate, Egypt and added with the soil before transplanting the seedlings. Chemical properties of filter mud used were estimated according to Black *et al.* (1965) and are listed in Table (2). Biofertilization treatments were control (Uninoculated seedlings), Active Yeast (AY), Minia Azotein (M.A.), Arbuscular mycorrhizae fungi (AM), AY+M.A., AY+AM, M.A.+AM and AY+M.A.+AM. Minia Azotein (M.A.) contains N-fixing bacteria (1ml M.A. contains 10⁷ cells) was added twice as soil drench around each seedling at 50 ml /bag at one month interval starting March 7th in both seasons, then seedlings were irrigated immediately. Arbuscular mycorrhizae fungi (AM fungi) [*Glomus mosseae* NRC31 and *Glomus fasciculatum* NRC15] 275 spores/g media " peat, vermiculite and perlite was applied once at 3 g / seedling via touch seedling root during transplanting the seedlings. This mycorrhizal inoculum originally isolated from Egyptian soils

and multiply on peat: vermiculite: perlite (Badr El-Din *et al.*, 1999). Both of Minia Azotein and Arbuscular mycorrhizae fungi were obtained from Biofertilization Unit, Agricultural Research Center, Giza. The application of active yeast (5 g/l.) was used as foliar spray at three times at two weeks interval starting March 14th in both seasons. All other agricultural practices were followed as usual. At the end of the experiment (the last week of January) of each season, the following data were recorded: stem length, stem diameter, leaves fresh and dry weight / plant and stem fresh and dry weight / plant. The studied elements of N, P and K % were determined in the dried leaves as follows: Nitrogen (%) was estimated according to the method of modified micro Kjeldahl as described by Wilde *et al.* (1985), Phosphorus (%) was estimated colorimetrically according to Chapman and Pratt (1975) and Potassium (%) was determined by Flam photometer according to Cottenie *et al.* (1982). The data were tabulated and statistically analyzed according to MSTATE-C (1986) using the L.S.D test at 5 % according to Gomez and Gomez (1984).

Table (1): Chemical properties of the used soil (average of the both seasons).

K ppm	P ppm	N ppm	Cations and anions (meq/l)								CaCO ₃ %	TDS %	PH
			K	Na	Mg	Ca	SO ₄	Cl	HCO ₃	CO ₃			
47	15	10	0.6	1.2	0.2	0.2	0.0	1.2	1	-	10.7	0.58	7.8

Table (2): Chemical properties of the used Filter mud (average of the both seasons).

Properties	PH	OM %	Total			Ca %	Total				EC (ds/m)	CN
			N %	P %	K %		Fe ppm	Mn ppm	Zn ppm	Cu ppm		
Values	6.2	72.5	2.65	2.1	0.66	2.8	5000	323	102	180	4.72	18:25

3. Results and Discussion

3.1 Stem length (cm)

The obtained results in Table (3) indicated that stem length of *Taxodium distichum* seedlings was significantly increased due to the application of filter mud as organic manure at all levels, during the two experimental seasons, as compared to the check treatment. Obviously, with increasing the level of filter mud stem length was significantly augmented in both seasons. Therefore, the longest plants were detected by using filter mud at the high level (72 g / bag) as ranged 28.8 and 42.6 % over unfertilized plants, during the two growing seasons, respectively. The role of organic manure in increasing stem length was also discussed by Ali *et al.* (2001), El-Sayed and Abdou (2002) on *Khaya senegalensis*, Ali *et al.* (2002), Ahmed *et al.* (2006) and Mahmmoud (2014) on *Populus nigra* and Siraji *et al.* (2001), Abdou *et al.* (2004), El-Khateeb *et al.* (2006), Abdou *et al.* (2007) and Saleh (2000) on *Ficus spp.* As for biofertilization treatments the data in Table (3) revealed that the use of them either separately or in combinations, except for active dry yeast, during the two

seasons, led to a significant increase in stem length of baldcypress, comparing to untreated ones. The combined treatments either double or triple inoculants gave better results than alone inoculant ones, in both seasons. In this connection, supplying active yeast + *Minia Azotein* + mycorrhizae fungi gave the longest stems reached 23.3 and 22.4 % over uninoculated plants in the first and second seasons, respectively. The efficiency of applying biofertilizers on enhancing stem length was also reported by El-Sayed and Abdou (2002), Abdou *et al.* (2006) and Ali *et al.* (2015) on *Khaya senegalensis* Ahmed and Ali (2002) and Dash *et al.* (2013) on *Dalbergia spp.* and Ahmed *et al.* (2005) and Mahmmoud (2014) on *Poplar trees* and Sudhakar *et al.* (2000) on *Morus alba* and Moustafa (2008) on *Chorisia speciosa* Abdou and Ashour (2012) on jojoba seedlings for N-fixing bacteria. Munro *et al.* (1999), Badram *et al.* (2003), Sarr *et al.* (2005), Ndiaye *et al.* (2011) and Ndoye *et al.* (2013) on *Acacia spp.* Abdou *et al.* (2007), El-Tayeb and El-Sayed (2010) on *Ficus spp.*, On *Chamaedorea alagena*, El-Khateeb *et al.* (2010) regarding phosphate resolving bacteria. While, Ahmed (2002) on *Leacaena leucocephala*, Ahmed *et al.* (2001) on *Ambrosia maritime* and Ahmed *et al.* (1998) on roselle plant and Akl *et*

al. (1997) on Red Roomy grape vine and Ahmed (2014) on *ceiba pentandra*, Ali (2001) *Calendula officinalis* concerning active yeast. In relation to the interaction effect between the two examined factors, it was significant on stem length of baldcypress, during the two consecutive seasons. Clearly, all combined treatments, except for 0 filter mud with active dry yeast and 0 filter mud plus Minia Azotein,

caused a significant augment in stem length as compared to untreated plants , during the two seasons, However, the application of filter mud at the high level (72 g / pot) with active dry yeast + Minia Azotein + mycorrhizae fungi, in both seasons proved to be more effective in increasing stem length than those obtained by other combination treatments, as clearly shown in Table (3).

Table (3): The effect of filter mud and biofertilizers treatments on stem length (cm.) of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)										
	Control	Stem length (cm)				Mean (B)	Control	Stem length (cm)			Mean (B)
		24	48	72	Mean (B)			24	48	72	
	First season					Second season					
Cont.	38.8	42.4	44.3	46.4	43.0	40.0	43.7	48.1	52.3	46.0	
AY	39.9	43.3	44.7	48.3	44.0	40.3	44.5	47.8	53.7	46.6	
M.A	40.3	44.7	46.4	49.7	45.2	42.3	46.3	50.1	58.7	49.4	
A.M	42.3	44.5	49.1	54.3	47.5	44.6	45.7	52.3	65.1	51.9	
AY+M.A	41.8	44.3	46.9	52	46.2	43.8	47.2	50.9	59.1	50.3	
AY+A.M	42.8	45.7	50.3	56.7	48.9	45.1	47.7	54.3	67.6	53.7	
M.A+A.M	43.8	46.7	53.1	57.9	50.4	46.9	49.3	55.7	70.2	55.5	
AY+M.A+A.M	44.3	48.7	55.1	64.0	53.0	46.4	50.9	56.0	72.0	56.3	
Mean (A)	41.7	45.0	48.7	53.7		43.7	46.9	51.9	62.3		
LSD 0.05%	A: 1.6		B: 1.3		AB:2.5		A: 1.3		B:1.7 AB:3.3		

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbuscular mycorrhizae.

3.2 Stem diameter (cm)

The presented data in Table (4) cleared that fertilizing *Taxodium distichum* seedlings with filter mud at all levels, in both seasons, significantly increased stem diameter as compared to the check treatment. Obviously, there was a gradual, significant augment in such trait with increasing the level of filter mud during the two seasons. Therefore, the use of the high level of filter mud (72 g/ pot) registered thicker stems which

increased it by 25.00 and by 28.81 % over control plants during the two growing seasons, respectively. Many investigators came to similar results that organic manure treatments increased stem diameter in many plants such as Ali et al. (2001), El-Sayed and Abdou (2002) on *Khaya senegalensis*, and Abdou et al. (2008) and Abdou and Ashour (2012) on jojoba seedling and Ali et al. (2002), Ahmed et al. (2006) and Mahmmoud (2014) on *Populus nigra* and Abdou et al. (2004), El-Khateeb et al. (2006),

Abdou et al. (2007) and Saleh (2000) on *Ficus spp.* In regard to biofertilizers application, the listed data in Table (4) showed that all used treatments either alone or mixed, except for active dry yeast, in both seasons, resulted a significant increase in stem diameter comparing to uninoculated ones. However, double or triple inoculant treatments were better than separately ones, in both seasons. In connection, triple inoculants (active yeast + Minia Azotein + mycorrhizae fungi) proved to be more effective in increasing stem diameter as ranged 27.27 and 26.22 % over untreated plants, during the tow experimental seasons, respectively. The beneficial effect of biofertilizers on enhancing stem diameter could be explained through the findings of El-Sayed and Abdou (2002), Abdou et al. (2006) and Ali et al. (2015) on *Khaya senegalensis* Ahmed and Ali (2002) and Dash et al. (2013) on *Dalbergia spp.* and

Ahmed et al. (2005) and Mahmmoud (2014) on *Poplar trees* and Sudhakar et al. (2000) on *Morus alba* and Moustafa (2008) on *Chorisia speciosa* Abdou and Ashour (2012) on jojoba seedlings, regarding N-fixing bacteria, Al-Hadad et al. (2014) on *Eucalyptus comaldulensis* and Abdou et al. (2007), El-Tayeb and El-Sayed (2010) on *Ficus spp.* Also, Munro et al. (1999), Badran et al. (2003), Sarr et al. (2005), Ndiaye et al. (2011) and Ndoye et al. (2013) on *Acacia spp.*, regarding phosphate resolving bacteria. While, Ahmed (2002) on *Leacaena leucocephala*, Ahmed et al. (2001) on *Ambrosia maritime* and Ahmed et al. (1998) on roselle plant and Akl et al. (1997) on Red Roomy grape vine and Ahmed (2014) on *Ceiba pentandra*, Ali (2001) *Calendula officinalis* for active yeast. Accordingly, the combination between the two examined factors on stem diameter, during the two seasons, had no significant effect (Table 4).

Table (4): The effect of filter mud and biofertilizers treatments on stem diameter (cm.) of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)										
	Control	Stem diameter (cm)				Control	Stem diameter (cm)				Mean (B)
		24	48	72	Mean (B)		24	48	72	Mean (B)	
	First season					Second season					
Cont.	0.50	0.53	0.55	0.63	0.55	0.53	0.61	0.62	0.69	0.61	
AY	0.51	0.53	0.57	0.61	0.55	0.53	0.6	0.65	0.69	0.62	
M.A	0.55	0.55	0.59	0.68	0.59	0.57	0.66	0.68	0.73	0.66	
A.M	0.57	0.59	0.68	0.73	0.64	0.60	0.66	0.72	0.76	0.68	
AY+M.A	0.55	0.58	0.61	0.68	0.6	0.60	0.65	0.67	0.74	0.66	
AY+A.M	0.59	0.61	0.71	0.72	0.66	0.62	0.69	0.73	0.78	0.70	
M.A+A.M	0.62	0.63	0.75	0.78	0.69	0.64	0.74	0.77	0.82	0.74	
AY+M.A+A.M	0.63	0.65	0.76	0.76	0.70	0.64	0.77	0.80	0.86	0.77	
Mean (A)	0.56	0.58	0.65	0.70		0.59	0.67	0.70	0.76		
LSD 0.05%	A:0.02 B:0.03 AB:n.s					A:0.04 B:0.02 AB: n.s					

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbescular mycorrhizae.

Obviously most of combined treatments led to an increase in such parameter in comparison with those obtained by other combination treatments, in both seasons. The highest values were detected by applying filter mud at the high level (72 g /bag) in combination with active yeast + Minia Azotein + mycorrhizal fungi or filter mud at the high level with Minia Azotein plus mycorrhizal fungi, as compared to other combination treatments, during the two consecutive seasons, as clearly declared in Table (4).

3.3 Leaves fresh weight (g) /plant

As shown in Table (5) that leaves fresh weight /plant of *Taxodium distichum* was significantly increased as a result of supplying organic manure as filter mud at all levels, during the two growing seasons, comparing to unfertilized plants. Such parameter was gradual significantly augmented with increasing filter mud

level, in both seasons. Therefore, the heaviest fresh weight of leaves was obtained by the application of filter mud at the high level (72 g / plant) which increased such trait by 77.6 and by 64.8 % over the check treatment in the first and second seasons, respectively. It worthy mention that inoculating *Taxodium distichum* seedlings with all biofertilizers either individually or together, in both seasons, resulted a significant augment in leaves fresh weight / plant, as compared to uninoculated treatments, except for active dry yeast, in the first season. Apparently, double or triple inoculant treatments gave better results than separately ones, during the two consecutive seasons. Clearly, the heaviest fresh weight of leaves / plant was observed when utilizing the triple inoculants (active yeast + Minia Azotein + mycorrhizal fungi) reached 64.8 and 51.4 % over control plants, during the two experimental seasons, respectively, as clearly postulated in Table (5).

Table (5): The effect of filter mud and biofertilizers treatments on leaves fresh weight (g/plant) of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)									
	Control	Leaves fresh weight (g/plant)								Mean (B)
		24	48	72	Mean (B)	Control	24	48	72	
	First season					Second season				
Cont.	13.2	17.2	19.1	20.9	17.6	12.4	18.6	21.1	21.9	18.5
AY	13.2	18.7	19.9	22.0	18.4	13.2	21.3	22.4	23.2	20.0
M.A	14.3	20.2	23.8	24.3	20.6	15.5	22.0	24.3	24.7	21.6
A.M	13.5	23.7	26.7	28.0	23.0	16.4	24.3	25.6	26.4	23.2
AY+M.A	14.8	22.9	24.2	24.8	21.7	17.0	23.4	25.5	26.0	23.0
AY+A.M	15.7	24.6	27.2	27.8	23.8	17.0	24.3	26.4	27.0	23.6
M.A+A.M	18.1	27.0	30.9	33.0	27.2	18.3	28.7	30.2	32.0	27.3
AY+M.A+A.M	18.7	28.6	33.5	35.0	29.0	19.4	29.3	30.8	32.4	28.0
Mean (A)	15.2	22.9	25.7	27.0		16.2	24.0	25.8	26.7	
LSD 0.05%	A: 0.6 B: 1.0 AB:2.0				A:0.7 B:0.8 AB: 1.6					

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbuscular mycorrhizae.

With respect to the interaction, given data in Table (5) pointed out that it was significant effect on leaves fresh weight / plant, during two growing seasons. The most of combined treatments led to a significant increase in leaves fresh weight / plant, during the two seasons, as compared to untreated ones. In connection, the application of filter mud at the high level (72 g / bag) with triple inoculants (active yeast + *Minia Azotein* + mycorrhizae fungi) proved to be more effective in increasing such trait than those obtained by other combination treatments, during the two growing seasons.

3.4 Leaves dry weight (g) /plant

The revealed data in Table (6) indicated that the obtained values of leaves dry weight / plant of *Taxodium distichum* seedlings had a trend similar to those of leaves fresh weight / plant in the two experimental seasons. Apparently, receiving the seedlings filter mud as organic manure at all levels, in both seasons, significantly increased in leaves dry weight / plant comparing to the check treatment. Such parameter was gradual significantly augmented with increasing the level of filter mud during the two seasons. Therefore, the high level of filter mud (72 g /plant) resulted the heaviest leaves dry weight / plant as ranged 80.0 and 68.4 % over unfertilized plants, in both seasons, respectively. The capability of organic manure on increasing leaves weight was also demonstrated by Ali et al. (2001), El-Sayed and Abdou (2002) on *Khaya senegalensis*, and Ali et al. (2002), Ahmed et al. (2006) and Mahmoud

(2014) on *Populus nigra* and Siraji et al. (2001), Abdou et al. (2004), El-Khateeb et al. (2006), Abdou et al. (2007) and Saleh (2000) on *Ficus Spp.* And Badran et al. (2003) on *Acacia saligna* and Abdou and Ashour (2012) on jojoba seedling. It is evident from the obtained results that the inoculation with active yeast, *Minia Azotein* and mycorrhizae fungi either alone or in combinations caused a significantly increase in leaves dry weight /plant, as compared to uninoculated ones, during the two consecutive seasons. It is clear that the use of double or triple inoculants application recorded better results than alone applied for the two seasons. Obviously, triple inoculants (active yeast + *Minia Azotein* + mycorrhizae fungi) proved to be more effective in augmenting such parameter than those obtained by other treatments for both seasons. Such superior previous treatment increased leaves dry weight / plant by 65.8 and by 53.5 % over control plants, in the first and second seasons, respectively, as clearly indicated in Table (6). The enhancement in leaves weight due to using biofertilizers was also insured by El-Sayed and Abdou (2002), Abdou et al. (2006) and Ali et al. (2015) on *Khaya senegalensis* and Ahmed et al. (2005) and Mahmoud (2014) on *Poplar trees* and Moustafa (2008) on *Chorisia speciosa* Abdou and Ashour (2012) on jojoba seedlings, Ahmed and Ali (2002) and Dash et al. (2013) on *Dalbergia spp* concerning N-fixing bacteria. Abou El-Khair (1993) and Munro et al. (1999), Badran et al. (2003), Sarr et al. (2005), Ndiaye et al. (2011) and Ndoye et al. (2013) on *Acacia spp.* and Abdou et al. (2007), El-Tayeb and El-Sayed (2010) on

Ficus spp. and Abdou et al. (2003), Meenakshisundaram et al. (2011) and Soliman et al. (2015) on *Delonix regia* for phosphate resolving bacteria. Whereas, Ahmed (2002) on *Leucaena leucocephala*, and Ahmed et al. (1998) on roselle plant and Akl et al. (1997) on Red Roomy grape vine and Ahmed (2014) on *Ceiba pentandra*, Ali (2001) *Calendula officinalis* regarding active yeast. Concerning the interaction, the presented results in Table (6) proved that it was significant effect on leaves dry weight /

plant of *Taxodium distichum*, during the two experimental seasons. Clearly, such trait was significantly augmented, in both seasons, due to applying most of combined treatments, as compared to untreated plants. However, the addition of filter mud at the high level (72 g /plant) with triple inoculants (active yeast + Minia Azotein + mycorrhizae fungi) proved to be more effective in increasing leaves dry weight / plant than those obtained by other combination treatments for the two growing seasons.

Table (6): The effect of filter mud and biofertilizers treatments on leaves dry weight (g/plant) of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)											
	Control	Leaves dry weight (g/plant)								Mean (B)		
		24	48	72	Mean (B)	Control	24	48	72			
	First season					Second season						
Cont.	3.1	4.1	4.5	5.0	4.1	2.9	4.4	5.0	5.2	4.4		
AY	3.1	4.4	4.7	5.0	4.3	3.1	4.8	5.3	6.2	4.8		
M.A	3.4	4.8	5.6	5.7	4.9	3.7	5.1	5.8	5.9	5.1		
A.M	3.2	5.6	6.3	6.5	5.4	3.8	5.7	6.0	6.3	5.4		
AY+M.A	3.4	5.4	5.8	5.9	5.1	4.0	5.5	6.0	6.2	5.4		
AY+A.M	3.6	5.8	6.4	6.6	5.6	3.9	5.7	6.2	6.4	5.6		
M.A+A.M	4.3	6.5	7.3	7.7	6.4	4.3	6.9	7.2	7.6	6.5		
AY+M.A+A.M	4.3	6.7	7.9	8.2	6.8	4.5	6.9	7.3	7.7	6.6		
Mean (A)	3.5	5.4	6.1	6.3		3.8	5.6	6.1	6.4			
LSD 0.05%	A: 0.2			B:0.2		AB: 0.4		A:0.2		B:0.3		AB: 0.5

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbuscular mycorrhizae.

3.5 Stem fresh weight (g) /plant

The illustrated data in Table (7) showed that supplying *Taxodium distichum* seedlings with filter mud at all levels, in both seasons, led to a significant increase in fresh weight of stem / plant comparing to control. By increasing the level of filter mud, such parameter was gradual significantly augmented, during the two growing seasons. Therefore, the heaviest

fresh weight of stem / plant was obtained by using filter mud at the high level which increased it by 72.2 and by 60.0 % over the check treatment, during the two seasons, respectively. In respect to biofertilizes application, the data in Table (7) indicated that there was a significant increase in stem fresh weight / plant, for the two seasons, due to the addition of all biofertilizers either separately or in combination, except for active dry yeast

in the second season, as compared to uninoculated ones. Clearly, double or triple inoculants application achieved better results than individual ones, during the two consecutive seasons. However, applying triple inoculants (active yeast + *Minia Azotein* + mycorrhizae fungi) gave heavier stem fresh weight / plant as ranged 56.1 and 52.4 % over untreated plants in the first and second seasons, respectively. Table (7) appeared that the combined effect between the two studied factors on stem fresh weight /plant was significant for the tow experimental

seasons. Obviously, most of combined treatments, in both seasons, significantly increased stem fresh weight /plant comparing to untreated ones. In this concern, the application of filter mud at the high level in combination with triple inoculants (active yeast + *Minia Azotein* + mycorrhizae fungi) followed by the high level of filter mud plus the double inoculants (*Minia Azotein* + mycorrhizae fungi) proved to be more effective in increasing such trait than those obtained by other combination treatments, during the two experimental seasons.

Table (7): The effect of filter mud and biofertilizers treatments on stem fresh weight (g/plant) of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)										
	Control	Stem fresh weight (g/plant)				Control	Stem fresh weight (g/plant)				Mean (B)
		24	48	72	Mean (B)		24	48	72		
First season					Second season						
Cont.	7.7	9.2	10.3	12.1	9.8	8.0	9.0	11.8	13.3	10.5	
AY	7.8	9.3	11.1	12.3	10.1	8.0	9.0	12.3	13.7	10.8	
M.A	8.6	10.3	11.9	14.4	11.3	9.8	11.0	13.0	15.0	12.2	
A.M	8.1	10.7	14.1	16.1	12.3	9.8	12.1	14.6	15.8	13.1	
AY+M.A	8.5	10.4	13.5	14.7	11.8	10.0	11.3	14.1	15.5	12.7	
AY+A.M	9.2	11.0	14.8	16.5	12.8	10.2	12.1	14.5	16.3	13.3	
M.A+A.M	10.7	13.6	16.5	18.8	14.9	11.3	13.7	17.2	19.0	15.3	
AY+M.A+A.M	11.2	13.9	17.0	19.2	15.3	12.6	14.3	17.6	19.5	16.0	
Mean (A)	9.0	11.0	13.6	15.5		10.0	11.6	14.4	16.0		
LSD 0.05%	A:0.3 B:0.4 AB: 0.9					A:0.5 B:0.6 AB: 1.2					

Cont.= Untreated plants, AY = Active yeast, M.A.= *Minia Azotein*, A.M = *Arbescular mycorrhizae*.

3.6 Stem dry weight (g) /plant

Data in Table (8) emphasized that the obtained values of stem dry weight / plant had trend similar to those of stem fresh weight /plant for the two seasons. It is appeared that fertilizing *Taxodium distichum* seedlings with filter mud as organic manure at all levels, in both seasons, caused a significantly augment in

stem dry weight /plant as compared to the check treatment. Obviously, there was a gradual, significant augment in such parameter with increasing the level of filter mud, during the two growing seasons. Moreover, the use of filter mud at the high level (72 g /plant) registered the heaviest stem dry weight /plant which reached 77.5 and 65.9 % over the unfertilized ones, for both seasons,

respectively. The promotive influence of organic manures in augmenting stem weight was also detected by Ali *et al.* (2001), El-Sayed and Abdou (2002) on *Khaya senegalensis*, and Ali *et al.* (2002), Ahmed *et al.* (2006) and Mahmmoud (2014) on *Populus nigra* and Siraji *et al.* (2001), Abdou *et al.* (2004), El-Khateeb *et al.* (2006), Abdou *et al.* (2007) and Saleh (2000) on *Ficus Spp.* and Badran *et al.* (2003) on *Acacia saligna*. With respect to the application of biofertilizers, the obtained data in Table (8) indicated that inoculating *Taxodium distichum* seedlings with all examined inoculants, in both seasons, led to a significant increase in stem dry weight, except for active dry yeast, for the two seasons, comparing to uninoculated ones. It could be noticed that utilizing double or triple inoculants gave better results than separately ones, during the two consecutive seasons. Apparently, the addition of triple inoculants (active yeast + Minia Azotein + mycorrhizae fungi) proved to be more effective in

increasing stem dry weight/ plant as ranged 55.5 and 52.0 % over uninoculated plants, during the two growing seasons, respectively. The role of biofertilizers application on enhancing stem weight was also obtained by El-Sayed and Abdou (2002), Abdou *et al.* (2006) and Ali *et al.* (2015) on *Khaya senegalensis*, and Ahmed *et al.* (2005) and Mahmmoud (2014) on *Poplar trees* and Abdou and Ashour (2012) on jojoba seedlings, Ahmed and Ali (2002) and Dash *et al.* (2013) on *Dalbergia spp.*, Moustafa (2008) on *Chorisia speciosa* and Abou El-Makarem (2016) and Abdel-Salam (2017) on *Moringa oleifera* regarding N-fixing bacteria Abo El-Khair (1993) and Munro *et al.* (1999), Badran *et al.* (2003), Sarr *et al.* (2005), on *Acacia spp.* and Abdou *et al.* (2007), El-Tayeb and El-Sayed (2010) on *Ficus spp.* and Abdou *et al.* (2003), Meenakshisundaram *et al.* (2011) and Soliman *et al.* (2015) on *Delonix regia* for phosphate resolving bacteria.

Table (8): The effect of filter mud and biofertilizers treatments on stem dry weight (g/plant) of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)										
	Control	Stem dry weight (g/plant)				Control	Stem dry weight (g/plant)				Mean (B)
		24	48	72	Mean (B)		24	48	72	Mean (B)	
	First season					Second season					
Cont.	3.5	4.1	4.7	5.6	4.5	3.6	4.1	5.4	6.0	4.8	
AY	3.5	4.2	5.0	5.6	4.6	3.6	4.1	5.6	6.2	4.9	
M.A	3.9	4.7	5.5	6.6	5.1	4.4	5.0	5.9	6.8	5.5	
A.M	3.6	4.9	6.4	7.4	5.6	4.4	5.5	6.6	7.2	5.9	
AY+M.A	3.8	4.7	6.1	6.8	5.3	4.5	5.1	6.4	7.1	5.8	
AY+A.M	4.1	5.0	6.8	7.6	5.9	4.6	5.5	6.8	7.4	6.1	
M.A+A.M	4.8	6.2	7.6	8.7	6.8	5.1	6.3	8.0	8.8	7.0	
AY+M.A+A.M	5.0	6.4	7.8	8.9	7.0	5.4	6.6	8.1	9.1	7.3	
Mean (A)	4.0	5.0	6.2	7.1		4.4	5.2	6.6	7.3		
LSD 0.05%	A:0.2		B: 0.2		AB: 0.4		A:0.3		B:0.3		AB: 0.5

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbescular mycorrhizae.

While, Ahmed (2002) on *Leacaena leucocephala*, Ahmed *et al.* (2001) on

Ambrosia maritime and Ahmed *et al.* (1998) on roselle plant and Ahmed (2014) on *Ceiba pentandra*, Ali (2001) *Calendula officinalis* concerning active yeast. Regarding to the interaction between the two studied factors, it was significant effect on stem dry weight / plant during the two experimental seasons. Clearly, most of combined treatments, in both seasons, resulted a significant increase in stem dry weight / plant, as compared to untreated plants. However, the highest values of such parameter were detected when applying filter mud at the high level (72 g / bag) with triple inoculants (active yeast + Minia Azotein + mycorrhizal fungi) followed by filter mud at the high level with double inoculants (Minia Azotein + mycorrhizal) in comparison with those obtained by other combination treatments, during the two consecutive seasons, as clearly shown in Table (8).

3.7 Nitrogen, phosphorus and potassium percentages

The obtained results in Table (9) cleared that the utilization of filter mud as organic manure at all level, in both seasons, caused a significant augment in the three studied elements N, P and K % in the dried leaves of *Taxodium distichum* seedlings as compared to unfertilized ones. Obviously, significant differences among the levels of filter mud were detected, during the two seasons, except for between the low and moderate levels, regarding P % in the first season and also between the moderate and high levels, in the second season, concerning K %. Apparently, using the high level of filter mud gave the

highest values of N, P and K % as ranged 23.20 and 16.16 % for N %, 25.00 and 17.76 for P % and 21.73 and 31.13 % for K % over control plants, during the two seasons, respectively. The increase in the elements of N, P and K as a result of supplying organic manure was also studied by Ali *et al.* (2001), El-Sayed and Abdou (2002) on *Khaya senegalensis*, and Ali *et al.* (2002), Ahmed *et al.* (2006) and Mahmmoud (2014) on *Populus nigra* and Abdou (2003) on *Washingtonia filifera* seedlings and Abdou *et al.* (2003) on *Dalonix regia*. As for biofertilizers application, the listed data Table (9) declared that inoculating *Taxodium distichum* seedlings with all examined biofertilizers, in both seasons, either separately or in mixture led to a significant increase in N, P and K %, except for active dry yeast, during the two seasons, regarding the three elements (N, P and K %), in addition to the treatment of double inoculants (active yeast + mycorrhizal fungi) concerning N % in the second season, as compared to uninoculated ones. In most cases, applying double or triple inoculants gave better results for N, P and K % than those obtained by separately inoculants, during the two seasons. However, the highest values of N, P and K % were obtained by applying triple inoculants (active yeast + Minia Azotein + mycorrhizae fungi) and double inoculants (Minia Azotein + mycorrhizae fungi) reached 8.85, 6.76, 6.25 and 9.66 % for N %, 37.98, 36.17, 38.75 and 34.75 % for P % and 16.80, 12.71, 15.96 and 13.55 % for K % as compared to uninoculated plants, in the two seasons, respectively. The positive effect of applying biofertilizers on

enhancing the elements of N, P and K was also reported by Abdou *et al.* (2006), El-Sayed and Abdou (2002) and Ali *et al.* (2015) on *Khaya senegalensis*, Abdou *et al.* (2004) on *Albizia lebbek*, Ahmed *et al.* (2005) and Mahmmoud (2014) on poplar trees and Abou El-Makarem (2016) and Abdel-Salam (2017) on *Moringa oleifera*, Moustafa (2008) on

Chorisia speciosa for N-fixing bacteria. Abou El-Kheir (1993), Badran *et al.* (2003) and Sarr *et al.* (2005) on *Acacia spp.*, El-Khateeb *et al.* (2010) on *Chamaedorea elegans* seedlings and Ali *et al.* (2014) *Cassia acutifolia* and Mohan and Radhakrishnan (2012) on *Tectona grandis* concerning phosphate resolving bacteria.

Table (9): The effect of filter mud and biofertilizers treatments on nitrogen percentage of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)											
	Control	Nitrogen (%)				Mean (B)	Control	24	48	72	Mean (B)	
		24	48	72	Mean (B)							
	First season					Second season						
Cont.	1.75	1.86	1.96	2.11	1.92	1.93	1.99	2.18	2.18	2.07		
AY	1.75	1.83	1.98	2.14	1.92	1.90	1.98	2.14	2.21	2.06		
M.A	1.85	1.93	2.05	2.24	2.02	2.06	2.14	2.32	2.38	2.23		
A.M	1.78	1.84	2.05	2.17	1.96	1.93	2.06	2.16	2.32	2.12		
AY+M.A	1.86	1.96	2.15	2.24	2.05	2.02	2.16	2.25	2.40	2.21		
AY+A.M	1.77	1.84	2.10	2.23	1.98	1.94	1.98	2.18	2.09	2.05		
M.A+A.M	1.83	1.88	2.12	2.33	2.04	2.16	2.18	2.32	2.43	2.27		
AY+M.A+A.M	1.88	1.93	2.18	2.37	2.09	1.90	2.20	2.34	2.40	2.21		
Mean (A)	1.81	1.88	2.07	2.23		1.98	2.09	2.24	2.30			
LSD 0.05%	A:0.05		B:0.04		AB: 0.07		A:0.05		B:0.03		AB: 0.06	

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbuscular mycorrhizae.

While, Ahmed (2002) on *Leucaena leucocephala*, Ahmed *et al.* (2001) on *Ambrosia maritime* and Ahmed *et al.* (1998) on roselle plant and Akl *et al.* (1997) on Red Roomy grape vine and Ahmed (2014) on *ceiba pentandra*, Ali (2001) *Calendula officinalis* regarding active yeast. The combined effect between the two examined factors was significant on N, P and K %, during the two growing seasons. Applying most of combined treatments significant increased the three tested elements (N, P and K %) comparing to untreated ones, in both seasons. Clearly, the most effective

treatments were obtained by using filter mud at the high level in combination with triple inoculants (active yeast + Minia Azotein + mycorrhizal fungi) and with double inoculants (Minia Azotein + mycorrhizal fungi) regarding N and P %, both seasons. While, applying filter mud at the moderate level with double inoculants (Minia Azotein) and filter mud at the high level plus triple inoculants (active yeast + Minia Azotein + mycorrhizal fungi) concerning K % were the most effective treatments in both seasons, as clearly shown in Tables (9, 10 and 11).

Table (10): The effect of filter mud and biofertilizers treatments on phosphorus percentage of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)										
	Control	Phosphorus (%)								Mean (B)	
		24	48	72	Mean (B)	Control	24	48	72		
	First season					Second season					
Cont.	0.115	0.115	0.141	0.145	0.129	0.126	0.140	0.145	0.151	0.141	
AY	0.116	0.127	0.131	0.158	0.133	0.131	0.141	0.149	0.156	0.144	
M.A	0.140	0.141	0.151	0.165	0.149	0.138	0.154	0.160	0.168	0.155	
A.M	0.151	0.167	0.158	0.169	0.161	0.162	0.163	0.179	0.190	0.174	
AY+M.A	0.135	0.145	0.157	0.146	0.146	0.146	0.160	0.168	0.167	0.160	
AY+A.M	0.152	0.164	0.159	0.192	0.167	0.164	0.160	0.174	0.181	0.170	
M.A+A.M	0.157	0.175	0.170	0.213	0.179	0.177	0.180	0.195	0.208	0.190	
AY+M.A+A.M	0.155	0.171	0.174	0.210	0.178	0.174	0.182	0.199	0.212	0.192	
Mean (A)	0.140	0.151	0.155	0.175		0.152	0.160	0.171	0.179		
LSD 0.05%	A:0.005		B:0.005		AB: 0.010		A:0.004		B:0.004		AB: 0.007

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbuscular mycorrhizae.

Table (11): The effect of filter mud and biofertilizers treatments on potassium percentage of *Taxodium distichum* seedlings during 2017/2018 and 2018/2019 seasons.

Bio-fertilizers treatments (B)	Filter mud levels g/plant (A)										
	Control	Potassium (%)								Mean (B)	
		24	48	72	Mean (B)	Control	24	48	72		
	First season					Second season					
Cont.	1.07	1.10	1.28	1.32	1.19	0.98	1.08	1.34	1.34	1.18	
AY	1.05	1.13	1.27	1.31	1.19	1.01	1.10	1.34	1.36	1.20	
M.A	1.08	1.16	1.27	1.41	1.23	1.03	1.21	1.38	1.36	1.25	
A.M	1.16	1.25	1.33	1.43	1.29	1.10	1.27	1.36	1.40	1.28	
AY+M.A	1.18	1.17	1.33	1.31	1.24	1.08	1.24	1.35	1.35	1.25	
AY+A.M	1.20	1.25	1.35	1.41	1.30	1.04	1.28	1.44	1.39	1.29	
M.A+A.M	1.24	1.30	1.51	1.47	1.38	1.10	1.32	1.50	1.45	1.34	
AY+M.A+A.M	1.21	1.33	1.46	1.55	1.39	1.11	1.29	1.45	1.46	1.33	
Mean (A)	1.15	1.21	1.35	1.40		1.06	1.22	1.39	1.39		
LSD 0.05%	A:0.02		B:0.03		AB: 0.06		A: 0.03		B:0.03		AB: 0.06

Cont.= Untreated plants, AY = Active yeast, M.A.= Minia Azotein, A.M = Arbuscular mycorrhizae.

From the obtained results, it could be discussed as follows: The increment in plant growth traits (stem length, stem diameter. Leaves fresh and dry weights and stem fresh and dry weights) and percentages of N, P and K as a result of using filter mud as organic manures might be due to the positive, physiological and biological roles of organic manure on these traits which were explained by

many authors such as, Bohn *et al.* (1985) who indicated that organic matter as a main source of N, P, S and contains high content of B and Mo and also as source of energy for Azotobacter growth. Saber (1997) reported that organic manure minimize the lost of nutrients by leaching. Organic manure resulted an augment in microbial activities in the root zone when applied it to the soil (Taiwo *et al.*, 2002).

The enhancement in the previous parameters due to biofertilizers application could be attributed to the biological and physiological roles of these inoculants. Minia Azotein (contains N-fixing bacteria), Many investigators for example (Dadarwell *et al.*, 1997; Hauwaka, 2000 and Hedge *et al.*, 1999) explored N-fixing bacteria roles as follows: Fixing atmospheric N which causes enhancing available N that increase, consequently the formation of many metabolites, producing adequate photohormones (IAA, GA3 and cytokinins). Increasing different nutrients uptake and enhancing water status which causes an increase in meristematic activities of cell and tissues, consequently augment plant growth protecting their host plants against plant pathogens through production of antibacterial and antifungal substances. Mycorrhizal fungi acts an important roles on plant growth and productivity as follows: Acting as root hairs of plants which led to increasing absorptive area and acts as extension to the root system. Phosphate dissolving organisms including mycorrhizal fungi are capable to produce some growth stimulation substances (auxins, GA3 and cytokinins). Enhancing the roots to secretion of organic acids and CO₂ which lead to increase the solubility of phosphate production of phosphate enzymes that are capable to converting organic phosphate to mineral P which it is possible to absorb by the plants. promotion on elements absorption (N, S, Ca, Zn, Mn and Cu). Increment in the resistance of inoculant plants to the stress due to nutrients deficiency and drought, protecting the plants from some diseases

and increasing the immunity against the diseases via increasing the thickness of root cortex (Read *et al.*, 1991; Tate, 2001). Active yeast plays an important role in enhancing the growth and chemical constituents, Tarrow and Nakase (1975) and Subbo Rao (1984) concluded that active dry yeast contains high amounts of four vitamins, particularly B that acts an important role in enhancing the growth and controlling the incidence of fungi diseases. From the revealed results, it could be recommended to supply the soil of *Taxodium distichum* seedlings with filter mud at 72 g / plant and treating the seedlings with mixture of biofertilizers (Minia Azotein + mycorrhizal fungi + active dry yeast) to improve the growth and some chemical constituents traits under the investigation conditions.

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