

IMPACT OF BIO-COMPOSITE ON GROWTH AND CHEMICAL COMPOSITION OF THREE PINE SPECIES SEEDLINGS AS COMPARING WITH SOME GROWING MEDIA

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ABSTRACT

This study was conducted at the Nursery of Timber Trees Department, Horticulture Research Institute, Agricultural Research Center, Giza, Egypt, during two successive seasons (2008/ 2009 and 2009/ 2010). The aim of this study was to investigate the effect of different growing media (sand, clay, mixture of clay and sand at the ratio of 1:1(V/V) and Bio-composite) on the growth and chemical composition of *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings in the nursery. The recorded data indicated that the most effective growing medium was Bio-composite, which gave highest values for all recorded growth characteristics, particularly *Pinus halepensis* which reached to (63.7cm, 16.3g and 6.3g in plant height, Fresh weight and Dry weight respectively), whereas the least effective media were sand and clay (when each was used alone). The selection of a growing medium is one of the important decisions in most containers culture of nursery plants. The physical, chemical and biological characteristics of a growing medium affect not only seedling growth but also other aspects of nursery operations.

Keywords: Growing media, Bio-composite, *Pinus halepensis*, *Pinus brutia*, *Pinus pinea*.

INTRODUCTION

Pine is a genus of family pinaceae which is a most important family, embracing about 300 different species, varieties and cultivars. The large order (Pinales) contains many trees which have great economic importance, as the forest pines of the world furnish most of mans requirements in softwood timber and wood pulp besides various oils. The wood that comes from it is also cold pine with many uses. It is often used in furniture in boat building, constructions and for other purposes where its resistance to decay is advantageous, also for carving, pattern- making, hobby work. This is because of its natural durability and it is less expensive than hardwoods Walker (1990).

The effect of soil media on growth and chemical composition of tree seedlings has been studied by many workers including Shafiq *et al.*, (1979) on *Eucalyptus camaldulenssis*, *Pinus brutia* and *Casuarina equisetifolia*, reported that a mixture of sand +clay +farmyard manure (1:1:1 v/v) gave the best growth of the seedlings . Pool and Conover, (1986) reported that increasing levels of woodchip sludge compost changed the physical characteristics of the medium but plant growth of *Brassai actinophylla* was unaffected by these changes. Hammad, (1994) on *Cupressus sempervirens* reported that the best potting medium tested for growth was sand: clay: foam 3:1:1 by volume. AL- Kahal *et al.*, (2003) observed that the level from

olive cake : sand (1:6 v/v) led to increase all the growth parameters of *Swietenia macrophylla*. Hammad *et al.*, (2004) on *Ceratonia siliqua* found that the effective growing medium was clay + compost at ratio of (1:2 v/v). Bio-composite consisted of composed separated manure mixed with vermiculite at the rate of 1:1 v/v and was inoculated with plant growth promoter rhizobacteria as microorganism source. Many a symbiotic diazotrophic bacteria have been described and tested as bio-fertilizers Kennedy *et al.*, (2004). Growers should carefully consider both biological and operational aspects when evaluating different types of growing medium. The mixing process is critical to producing custom growing media, the quality of the best components is compromised if the growing medium is improperly mixed. A variety of materials are routinely added to growing media during the mixing process; these include fertilizers, lime, surfactants, super absorbents, and mycorrhizal inoculums. The uniform incorporation of these materials is important because plant roots have access to only a limited volume of growing media in the relatively small containers used in native plant nurseries, Whitcomb, (2003) emphasized that improper mixing is one of the major causes of variation in container plant quality. A typical growing medium is a composite of two or three component. Mixtures of organic and inorganic components are popular, because these materials have opposite, yet complementary, physical and chemical properties. The world uses huge quantities of synthetic nitrogenous fertilizer for growing plants, this dependence creates certain dangers for the global economy and especially for the environment (Smil, 1997). Anything that can be done to utilize nitrogen fixed naturally from the atmosphere where it occurs as molecular nitrogen N₂ as a substitute for fertilizer N, will benefit all people. Rhizobacteria able to fix atmospheric N₂ and because of this feature they are often introduced to manage agricultural ecosystem to improve their organic fertility, economy or farming system flexibility.

So, the aim of this study was to investigate the effect of different growing media (Sand, Clay and mixture of clay and sand ratio (1:1 v/v) and Bio-composite) on the growth and chemical composition of { *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* } seedlings in the nursery.

MATERIALS AND METHODS

This study was conducted at the nursery of the Timber Trees Department, Horticulture Research Institute, Agricultural Research Center, Giza, Egypt, during the two successive seasons 2008/ 2009 and 2009/ 2010. On first of October 2008, as much as possible, unified seeds were stored at room temperature for three months. After that seeds were soaked in solution contained gibberellic acid (GA₃) at concentration of 150 ppm then they were sown directly in plastic pots (5. cm in diameter) filled with the used media (sand, clay, clay + sand at ratio 1:1 v/v and bio-composite). On first of February 2009 seedlings were transplanted in plastic pots (15-cm in diameter) filled with the same media. On first of July 2009 seedlings were retransplanted into plastic pots (25-cm in diameter) filled with the same media. Each pot received 1/2 gm N.P.K 19:19:19 (Kristalon) every month from

March to December. Each of the 12 treatments was replicated 3 times, and each replicate was represented by 5 seedlings. The physical and chemical characteristics of the sand and clay used in the study are shown in Table (1).

Bio-composite is produced by processing of different plant residues (peanut shell and saw dust) and natural minerals and rock (bentonite, manganese ore, dust and phosphate) were done via partial acidifying, steam pressure, then composting, afterwards, the product was bore with some beneficial microorganisms to prepare non-traditional bio-organic fertilizer. The physical and chemical characteristics of the bio-composite are presented in Table (2).

The seedlings were placed in shaded area, and after one month from transplanting the seedlings were moved to a sunny area and the common cultural practices (including regular irrigation) were applied.

The layout of this experiment was a randomized complete blocks design. The experiment was started on the first of October 2008 and ended on the end of December 2009, the same work was repeated as second growing season in 2009/2010. The vegetative growth parameters were recorded, including plant height (cm) and fresh and dry weights of the whole shoot [stem, leaves (needles)]. Nutrients were extracted from samples of dried shoots using the method described by Piper (1947). The Nitrogen content was determined using modified micro-Kjeldahl method as described by Pregl (1945), while the phosphorus content was estimated using the method recommended by King (1951).

The data were subjected to statistical analysis of Least Significant Difference (L.S.D)" test at 5% levels, as describe by Steel and Torrie (1980).

Table (1): Physical and chemical characteristics of sand and clay used for growing *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings.

Soil characteristics	Sand	Clay
Physical characteristics		
Course sand (%)	65.60	3.45
Fine sand (%)	31.50	37.03
Silt (%)	0.77	27.75
Clay (%)	2.50	27.25
Organic matter (%)	0.30	0.33
Texture	Sand	Clay
Chemical characteristics		
pH	6.70	8.10
EC (de/m)	0.65	2.13
Na (ppm)	3.45	13.40
K (ppm)	0.58	0.85
Bicarbonate (ppm)	1.95	4.00
Chloride (ppm)	2.80	16.50
Sulphur (ppm)	2.28	0.83
Calcium carbonate (%)	28.00	3.77
Ca ++ (ppm)	2.12	5.25
Mg ++(ppm)	0.88	1.79
C.E.C. (meq / 100g)	5.50	32.60

Table (2): Characteristics of bio-composite used for growing *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings.

Parameter	Value
Moisture content (%)	32
Bulk density (kg/ m ³)	590
Water holding capacity (%)	170
pH in 1:5 extract	6.47
EC in 1:5 extract (dS/ m)	7.19
E4 /E6	3.46
Organic-C (%)	26.77
Total-N (%)	1.31
C/N ratio	20.44
Total-P (%)	1.16
Total-K (%)	0.64
N-NH ₄ (ppm)	697.0
N-NO ₃ (ppm)	165.1
Available-P (ppm)	296.0
Available -K (ppm)	913.0
Total- Fe (%)	0.78
Total- Mn (ppm)	284.3
Total- Zn (ppm)	167.0
Total- Cu (ppm)	38.5
DTPA-extractable-Fe (ppm)	624.8
DTPA-extractable-Mn (ppm)	63.9
DTPA-extractable-Zn (ppm)	39.5
DTPA-extractable-Cu (ppm)	8.2
Dehydrogenase activity (gtpf/g)	142.5
Nitrogenase activity (nmol C ₂ H ₄ /hr)	98.8
Total count of bacteria	14x 10 ⁷
Total count of fungi	13x 10 ⁶
Total count of actinomycetes	2.7x 10 ⁶
Seed germination test %	91.6

RESULTS AND DISCUSSION

1-Effect of growing media on vegetative growth and chemical composition of *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings.

1-1- Vegetative growth:

The data presented in Table (3) show that the seedlings of pine species grown in sand or clay media alone gave the lowest values for the different vegetative growth characteristics. Similar results have been reported by Hammad *et al.*, (2004) on *Ceratonia siliqua*, and they found that the use of sand or clay were the poorest media, which gave the lowest characteristics. Sand only gave the poorest results, and this may be attributed to the lowest content of elements, nutrients and organic matter. In contrast, the data presented in table (3) show that the bio-composite medium had generally favorable effect on the vegetative growth characteristics of the seedlings of pine species. In this case, seedlings grown in bio-composite medium have significantly higher values for the different growth parameters in both seasons, compared to seedlings grown in sand or clay media. Bio-composite medium was clearly the most effective one for promoting vegetative growth

of some pine species. seedlings in the first season, giving the highest mean values for plant height, fresh and dry weights/seedling (62.5, 60.3 and 61.7 cm, 15.6, 14.6 and 15.3 g and 6.0, 5.3 and 5.8 g for *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* respectively).

These results were confirmed in the second season, with the same growing medium (bio-composite) giving the highest values for plant height (63.7, 62.2 and 61, 8 cm), as the fresh and dry weights/seedling (16.3, 14.7 and 15.0 g and 6.3, 5.2 and 5.9 g) for (*Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings respectively). In the first season, the percentages of increases in the different growth characteristics as a result of using bio-composite medium, compared to use of clay alone, were 68.8%, 62.0%, and 71. % for plant height and 47.4%, 41.1% and 41.8% for fresh weights / seedling and 46.7%, 50.0% and 34.4% for dry weights/seedling to (*Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings, respectively). Also, the values obtained from seedlings grown in bio-composite were higher by percentages of 87.5%, 84.2%, and 84.6% also, 78.2%, 72.6% and 75.8% added to that 63.3%, 62.3% and 67.2% (for the above-mentioned characteristics, respectively), compared to values obtained from seedlings grown in sand only. In the second season, a similar trend was observed. The superior effect of the bio-composite, compared to any media used, may be attributed to the ability of negatively charged particles of organic matter to attract and hold positively charged cation in the soil solution, and provide the plant roots with these cation (i.e. this medium has a high cation exchange capacity), which allows high availability of water and nutrients to the roots Hartmann *et al.*, (1981), added to that the favorable effect of the bio-composite on vegetative growth, compared to the use of sand may be attributed to an improvement in soil water retention and consequently, to its indirect effect on increasing the uptake of water and nutrients by the plant roots.

Table (2) shows the physical, chemical and biological analysis of the bio-composite, it has appreciable quantities of the total and available macro and micronutrients indicating that the product possess high fertilizer value, particularly in presence of de-composed organic materials which act to prevent nutrients losing or precipitation, also bio-composite has suitable water holding capacity and bulk density for their employing and as growth medium. Free-living micro-organisms exert beneficial effects on plant growth and yields Dobereiner and Pedrosa (1987). Micro-organisms promote root diameter, density and length of root hairs Hadas and Okon (1987), root surface area during early growth 3-4 weeks Fallik *et al.*, (1988), mineral and water uptake and dry-matter accumulation in seedlings Sarig *et al.*, (1988), in ceases indol-3 acetic acid and indole butyric acid content in roots and increases respiration and metabolic root enzyme activities Fallik *et al.*, (1989). Maximal effects depend on competitive rhizosphere population and organic matter content in soil. With respect to the biological properties, it has high biological activity Abdel -Wahab and Ahmed (2003). A number of (Rhizobacteria) strains with apposite effect on plant development Bashan and de Bashan (2002) and Al-kahal *et al.*, (2003) and their ability to induce

plant growth promoting Mekhamar (2001) and Hammad *et al.*, (2011). Also Phytohormones such as indole-3-acetic acid or cytokinins are among the plant growth promoting compounds often produced by bacteria -El-Soud *et al.* (2007).

Table (3): Effect of different growing media on the vegetative growth of *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings.

Species	Measurements Growing media	First season			Second season		
		Plant height (cm)	Fresh weight (g)	Dry weight (g)	Plant height (cm)	Fresh weight (g)	Dry weight (g)
<i>P. halepensis</i>	Sand	7.8	3.4	2.2	8.3	3.7	2.4
	Clay	19.5	8.2	3.2	19.8	8.4	3.3
	Sand +clay (1:1v:v)	26.0	8.8	3.7	25.8	9.5	4.0
	Bio- Compsite	62.5	15.6	6.0	63.7	16.3	6.3
<i>P. brutii</i>	Sand	9.5	4.0	2.0	9.0	4.0	2.2
	Clay	22.9	8.6	3.4	22.8	8.7	3.5
	Sand+ clay (1:1v:v)	24.7	9.1	4.0	24.5	9.7	4.0
	Bio. compsite	60.3	14.6	5.3	62.2	14.7	5.2
<i>P. pinea</i>	Sand	9.5	3.7	1.9	8.8	4.0	2.0
	Clay	17.5	8.9	3.8	19.5	9.2	3.8
	Sand+clay (1:1v:v)	23.8	9.4	4.1	24.8	9.7	4.0
	Bio. compsite	61.7	15.3	5.8	61.8	15.0	5.9
	L.C.D 5%	3.57	1.09	0.60	0.55	0.38	0.30

1-2- Chemical composition

1-2-1- Total carbohydrates percentage:

Chemical analysis of dry matter samples has revealed that the total carbohydrate content was considerably affected by used bio-composite (Table 4). In both seasons, using sand or clay alone as a medium for growth of *Pine spp* seedlings, resulted in lower mean carbohydrate contents, compared to those obtained in seedlings which were grown in bio-composite medium. Seedlings which were planted in bio-composite medium had the highest content of total carbohydrates (69.4%, 59.3% and 59.6% of *Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings respectively), whereas the lowest value 36.3%, 36.1% and 36.4% for (*Pinus halepensis*, *Pinus brutia* and *Pinus pinea* seedlings respectively) obtained from seedlings which were grown in sand soil.

The seedlings in bio-composite reached the highest content compared to values obtained from seedlings grown in sand or clay, respectively.

The favorable effect of bio-composite as a growing medium on carbohydrate synthesis and accumulation may be attributed to its high cation exchange and water- holding capacities. The high cation exchange capacity of the bio-composite allows the plant roots to take up the potassium needed for activation of the enzymes necessary for photosynthesis, while the high water-holding capacity ensures a sufficient water supply for nutrient absorption and for efficient photosynthesis. The nitrogen absorbed by the seedlings is essential in the structure of chlorophylls and, consequently, the synthesis of carbohydrates. Among the different growing media that were

tested, bio-composite which gave the best results in terms of carbohydrate accumulation, whereas, sand and clay (used separately) were the least effective media in this respect. The results obtained in this work indicate a potential use of microorganisms as inoculant for promoting growth. Yield increases could be due from added to plant growth promoting substances (auxins, cytokinins, or gibberellins) by micro-organisms cells or by respons of plant to bacterial elicitors of hormonal metabolism Okon *et al.*, (1988).

Table(4): Effect of different soil media on chemical composition of *Pinus halepensis* , *Pinus brutia* and *Pinus pinea* seedlings.

Species	Measurements	Total	N	P	K
	Growing media	carbohydrates (%dry matter)	(%dry matter)	(%dry matter)	(%dry matter)
<i>P. halepensis</i>	Sand	36.3	1.00	0.110	0.190
	Clay	38.1	1.01	0.120	0.199
	Sand +clay (1:1v:v)	39.2	1.71	0.160	0.368
	Bio- Composite	69.4	2.90	0.380	0.568
<i>P. brutii</i>	Sand	36.1	1.00	0.110	0.185
	Clay	37.9	1.03	0.120	0.200
	Sand +clay (1:1v:v)	39.5	1.75	0.160	0.370
	Bio- Composite	59.3	2.85	0.380	0.560
<i>P. penea</i>	Sand	36.4	1.00	0.110	0.189
	Clay	38.0	1.03	0.120	0.195
	Sand +clay (1:1v:v)	39.5	1.73	0.160	0.365
	Bio- Composite	59.6	2.77	0.380	0.562

1-2-2- Nitrogen percentage:

The nitrogen content in *Pinus halepensis* , *Pinus brutia* and *Pinus pinea* seedlings was markedly affected by the growing medium Table (4). The data showed that the seedlings which grown in bio-composite medium had a higher N percentage (2.90%, 2.85% and 2.77 % than those grown in other media. Clay or sand media alone had the lowest rates. In contrast, the lowest value (1.00%) was obtained in seedlings grown in sand only for seedlings species.

Nitrogen exposed to vigorous losing during composting process which caused by volatilization of ammonia and leaching of ammonium and nitrate. The factors which led to nitrogen losses are high temperature, alkaline pH and forced aeration Dewes, (1995). Therefore, composting such organic materials under mild acidic condition may be decreased nitrogen losses. In addition, incorporation of clay minerals such as bentonite with composted materials could increase and maintain the plant nutrients against different losing factors. Bio-composite may behave likewise as bio-fertilizers and plant growth promoting rhizobacteria. The bio-organic compound which represented the major material in production of seedlings as well as production of bio-fertilizer Abdel-Wahab and Ahmed, (2003). The mechanism of the bacterization resulting in yield increase with decrease or no N concentration may be attributed to enhance N₂-fixation or increase N assimilation by plant Mekhamar *et al.*,(2007).

1-2-3- Phosphorus percentage:

Chemical analysis of dried samples taken from seedlings grown in the different growing media Table (4) has shown that the P content was considerably by addition of compost to the clay soil. The P content varied from 0.110 (in seedlings grown in sand only) to 0.380% (in seedlings grown in bio-composite medium, the other clay medium also gave significant decreases in the P content, compared to the use of bio-composite medium, this means that addition of bio-composite to the growing media increase P content in the seedlings. The role of micro-organisms in dissolution of rock phosphate during composting stage may considered with acidic action a comprehensive effect Abdel-Wahab *et al.*, (2003).Table (2) shows the physical, chemical and biological analysis of the bio-composite has suitable C/N ratio for arid and semi-arid conditions which prevailed in Egypt. Such ratio make the decomposition rate of organic materials behave slowly during the growth season without appearance of phytotoxicity effects Abou Bakr, (1994). The bio-composite has appreciable quantities of total and available macro-fertilizer value.

1-2-4- Potassium percentage:

The effect of different soil media on K content in the leaves of pine species seedlings were generally similar to their effect on N and P content Table (4). Plants grown in clay or sand only had the lowest K content, compared to those grown in bio-compost. The most effective media for increasing the K content was bio-composite. The favorable effect of the bio-composite on the accumulation of K in the plants may be attributed to the high K content in the bio-composite, compared to that in clay or sand only.

RECOMMETION

Growers should carefully consider both biological and operational aspects when evaluating different types of growing media. Experiment showed that the selection plant growth medium led to get the best results of growth compared another growth media, sand or mud, which made us to make use of plant wastes economically and commercially in the make bio-composite medium..

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اثر بيئة الكميوست الحيوى على النمو والتركيب الكيماوى لثلاث انواع من شتلات الصنوبر مقارنة ببعض بيئات النمو الاخرى.

حسام حسن حماد

قسم بحوث الأشجار الخشبية بمعهد بحوث البساتين - مركز البحوث الزراعية.

اجري هذا البحث بمشتل قسم بحوث الأشجار الخشبية بمعهد بحوث البساتين بمركز البحوث الزراعية خلال موسمين متتاليين (٢٠٠٨/٢٠٠٩-٢٠٠٩/٢٠١٠) بهدف دراسة تأثير بيئة الزراعة الحيوية المنتجة بمعهد بحوث الاراضي و المياه علي النمو و التركيب الكيماوي لشتلات ثلاث انواع من الصنوبر (صنوبر حلبي و صنوبر بروتي و صنوبر ثمرى) و مقارنة ذلك للتاثير مع بيئات الرمل و الطين و خليط الرمل و الطين بنسبة ١:١ بالحجم اوضحت النتائج المتحصل عليها ان استخدام بيئة الكمبوست الحيوي اعطت اعلي قيم للنمو و كانت الزيادة معنوية في جميع قيم الصفات الخضرية و الكيماوية التي تم قياسها وذلك مقارنة بالشتلات النامية في التربة الرملية او الطينية او بيئة النمو المكونة من خليط الرمل و الطين بنسبة ١:١ بالحجم. وكانت اقل القيم للشتلات النامية في بيئة الرمل يليها بيئة الطين ثم بيئة الخليط من الرمل و الطين . ومن ذلك نري ان يمكن تعظيم الاستفادة من المخلفات النباتية و تحويلها الي بيئة نمو صالحة لانتاج الشتلات علي نطاق تجارى.

قام بتحكيم البحث

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مركز البحوث الزراعية