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Pine litter as substrate for propagation of vegetable transplants in trays

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The study evaluates the potential of pine litter for use as a substrate in the propagation of vegetable transplants in trays. Sub-optimum pH and electrical conductivity of pine litter were addressed by incorporating 10 % feedlot manure by volume. The water holding capacities of pine litter and the pine litter-feedlot manure mixture were higher than the minimum required, but their air filled porosities were below optimum. The pine litter-feedlot manure mixture reduced the germination percentages of cabbage seedlings by 17 % and lettuce seedlings by 13 % relative to composted pine bark, but the germination percentage of tomato seedlings was similar in both substrates. There were no significant differences in the growth of vegetable transplants between the two substrates. In both substrates increasing nutrient availability by adding controlled-release fertilizer had similar positive effects on the growth transplants. Substrate-nutrient availability interactions were only observed in dry shoot mass of lettuce seedlings and dry root mass of tomato seedlings. Composting the pine-litter animal manure mixture could possibly improve the observed low germination percentage of vegetable transplants in pine litter.

Keywords: Feedlot manure, pine bark, pine litter, substrates, vegetable transplants

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Introduction

Soilless potting substrates are commonly used in the production of vegetable transplants. These substrates usually consist of a mixture of organic materials, such as peat moss or composted pine bark, and inorganic substances, such as perlite, vermiculite, sand, or rock wool (Bunt, 1988; Nelson, 1991). Among the organic materials, sphagnum peat moss is favoured, because of its desirable physical and chemical properties (Raviv, Chen & Inbar, 1986). However, the distribution of peat bogs is limited to temperate climates. As a result, many countries including South Africa have to import peat. Moreover, peat is not really a renewable source, because it takes several decades before peat bogs recover fully from being mined (Raviv, Reuven & Zaidman, 1998). This has caused growing environmental opposition to peat harvesting (Atiyeh, Edwards, Subler, & Metzger, 2000).

Worldwide, economic and ecological concerns have encouraged a search for alternative organic materials that can be used as substrates in plant propagation. According to Evans & Stamps (1996), most research into the development of substitutes for peat moss has focused on municipal or agricultural waste. Examples of materials that have been considered include composted kraal manure and grape marc (Inbar, Chen & Hadar, 1986), coir dust (Evans & Stamps, 1996), and worm-worked duck waste (Wilson & Carlile, 1989). In South Africa, milled composted pine bark has become the most important organic substitute for peat (H. Coetzee, 29-01-2001, CAL, Phelindaba). Its use in the propagation of vegetable transplants spread rapidly during the 1980s (Van Schoor, Smith & Davis, 1990), because it was found to perform as well as peat moss (Smith, 1982).

Pine litter, also referred to as pine straw, pine litterfall or pine needles consists of dead leaves and twigs which fall from pine trees throughout the year and accumulate and decompose on the forest floor (Zwolinski & Quicke, 1998). Dames (1996) defines pine litter as the dead plant material

found on top of the mineral soil surface in pine forests, which is composed of needles, branches, twigs, cones and roots. Three layers may be identified in pine litter. The top layer also called the litter layer is composed of intact dead plant tissues. The fermentation layer occurs below the litter layer and is composed of broken up but recognizable organic tissues. The humus layer separates the fermentation layer from the mineral topsoil. It is composed of dark, powdery, organic matter, which no longer resembles the plant tissues from which it originated. Dames (1996) reported that in South African pine forests the humus layer was largely absent.

In pine forest litter fall plays an important role in nutrient cycling (Versfeld & Donald, 1991). Pine litter is also a liability, because it represents a fire hazard, prevents rain from entering the soil and increases soil acidity (Zwolinski & Quicke, 1998). Under circumstances where removal of pine litter from forest floors is desirable, utilization of the litter as a potting substrate could change this product into an asset.

In South Africa different species of pine trees are grown, but *Pinus patula* occupies the largest area, covering 48 % of the total area of 620 239 ha planted to pine trees in South Africa in 1999 (R.Godsmark, 09-03-200, FOASA). Mphaphuli (2003) estimated average annual litter production in *P. patula* forests to amount to about 2.4 t ha⁻¹. Consequently, an estimated 833 000 t of *P. patula* litter is produced annually in South Africa. The South African vegetable transplant industry uses approximately 72 000 t (150 000 m³) of pine bark per year (S. Trollip, 12-03-2003, Bark Enterprise, Brits). It follows that annual production of pine litter in *P. patula* plantations is sufficient to be considered adequately available for use as a substrate by the South African vegetable transplant sector.

For commercial exploitation, adequate availability is one of the three requirements of a potential substrate (Evans & Stamps, 1996). Substrates must also be sufficiently uniform (Evans & Stamps, 1996). In pine litter uniformity is achieved

mechanically, during the process of reducing the size of the litter particles with a hammer mill. The final requirement is that the substrate must have favourable physical and chemical properties. Air filled porosity (AFP) and water holding capacity (WHC) are important physical properties of substrates, and pH and electrical conductivity important chemical properties (Bunt, 1988; Ingram, Henley & Yeager, 1990). The objectives of the present study were to determine these physical and chemical properties of pine litter and to assess its suitability for use as a substrate in the propagation of vegetable transplants in trays.

Material and methods

Data collection was subdivided into two phases, viz. laboratory tests and greenhouse experiments. The laboratory tests were used to determine important properties of pine litter in relation to its use as a substrate, and the extent to which these properties needed to be improved. Greenhouse experiments were used to test the suitability of pine litter for use as a substrate in the propagation of vegetable transplants using plant growth as an indicator. Pine litter was collected from the floor of an undisturbed 32-year old *P. patula* forest at the SAFCOL plantation near Belfast in Mpumalanga Province. On average, the pine litter was 89 mm thick and consisted of 29 % litter (L) layer and 71 % fermentation (F) layer on a dry mass basis. The humus (H) layer was absent. The litter was reduced in size and homogenised using an electric hammer mill with a 5 mm screen. The composted pine bark substrate, which had passed through an 8 mm screen, was supplied by Bark Enterprise in Brits. Partially composted cattle feedlot manure, obtained from Chalmar Beef in Bronkhorstspuit, was introduced at a rate of 10 % by volume to modify the low pH and electrical conductivity of the pine litter and to enrich its nutrient content. Feedlot manure was selected, because it is substantially homogeneous in composition, and in Gauteng Province it is available in large quantities. Chemical analysis showed the feedlot manure used in this study to contain 2 % N, 0.6 % P and 1.4 % K on a dry matter basis. The feedlot manure was not sterilized before mixing it with the pine needles.

Electrical conductivity (EC) and pH of the substrates were determined on watery extracts (Lang, 1996), prepared by mixing two volume proportions of the substrate with three volume proportions of distilled water. To standardize their volume, the substrates were compacted in cylinders constructed from transparent perspex tubing with a diameter of 50 mm by applying a pressure equivalent to 500 g cm^{-2} over a period of 60 s. For this purpose two lengths of 60 mm tubing were joined with masking tape. Fly screen with a gauge of 1 mm was glued to the bottom end of the bottom cylinder, whilst the top cylinder was left open. Following compaction the two cylinders were separated with a sharp knife and the standard volume of the substrate contained in the bottom was transferred to a 500 ml reagent bottle. Distilled water amounting to 1.5 times the volume of substrate was added, before sealing the reagent bottle and shaking it at 180 oscillations per min for 30 min, and filtering the mixture through a Whatman V1 filter paper to remove the solid particles. A conductivity meter and a pH meter were used to measure the EC and pH of the watery extracts.

The air filled porosity (AFP) of a substrate, defined as the proportion of the bulk volume of the substrate consisting of pores filled with air after saturation and drainage, and its water holding capacity (WHC), defined as the proportion of the bulk volume of the substrate consisting of pores filled with water after saturation and drainage, are physical properties that are influenced by container height (Ingram, Henley & Yeager, 1993; Fonteno, 1996). Samples for determination of AFP and WHC were prepared in 60 mm perspex cylinders closed with fly screen at one end. The cylinders had the same height as the cavities in the trays that were used in the greenhouse experiments. Compaction of the substrate was achieved by knocking the cylinder three times on a wooden bench. Filling and compaction were repeated until the cylinder was full to the brim, where after the top end of the cylinder was sealed off by means of a plastic lid in which an opening with a diameter of 5 mm had been drilled to enable air to escape. The substrate samples were submersed, left to soak for 16 h, and then placed gauze-end down on a 5 mm mesh-wire sieve to drain for 1 h, before determining their AFP and WHC.

In the greenhouse, a factorial experiment involving two substrates (main plots) and five nutrient supply levels (split plots) was conducted. The greenhouse had a wet wall and two fans to limit maximum temperature to 25 °C. The experiment was conducted three times using different vegetables as test crops. The experiment employed a split plot design with four replicates. The first main plot treatment consisted of a mixture of 90 % pine litter and 10 % feedlot manure by volume. The second main plot treatment was the control, which consisted of pure composted pine bark. Application of the 70 days controlled-release Hortico 7:1:2 (22) fertilizer, supplied by Ocean Agriculture in Muldersdrift (Gauteng) at five different rates was used to create the five split plot treatments (Table 1). The experiment was conducted in seedling trays with 200 cavities. Each tray of 200 cavities contained a single main treatment, and the five splits were randomly assigned to blocks of 40 cavities. The vegetables planted in the experiments were cabbages (*Brassica oleracea*), lettuce (*Lactuca sativa*) and tomatoes (*Lycopersicon esculentum*). The cultivars used were Gloria (cabbages), Great Lakes (lettuce) and Rodade (tomatoes). The seedling trays were planted using the method described by Atiyeh *et al.* (2000). One seed was planted per cavity at a depth of 10 mm in holes made by a dibbler. Trays were placed on the tables in the greenhouse and left for six weeks under ambient conditions. The plants were watered daily using a hosepipe with a spray attachment.

The germination percentages of the three vegetables were determined 15 days after planting. After six weeks the seedlings were harvested. Ten plants were selected randomly from each treatment to measure growth using oven-dry mass of shoots and roots as indicators. Shoots and roots of sampled plants were separated by cutting the stem immediately above the first root using a surgical knife. Dry shoot mass was determined for all 10 sampled plants and dry root mass for two of the 10. Analysis of variance (ANOVA) of the data obtained in the three experiments was conducted by means of SAS (SAS Institute Inc, 2000).

Table 1 Rates of application of controlled-release Hortico fertilizer used in the substrate evaluation experiments

Treatment	Application rate (number of fertilizer granules applied per cavity)	Mass of fertilizer applied per cavity (mg)	Amount of pure nutrients applied per cavity (mg)		
			N	P	K
1	0	0	0.00	0.00	0.00
2	8	23	3.54	0.51	1.01
3	16	46	7.08	1.01	2.02
4	32	92	14.17	2.02	4.05
5	64	184	28.34	4.05	8.10

Results and discussion

Laboratory tests

Table 2 Important physical and chemical properties of pine litter, pure composted pine bark, feedlot manure and of a substrate composed of 90 % pine litter and 10 % feedlot manure (90 % PN – 10 % FM)

Materials	pH (H ₂ O)	Electrical conductivity (mSm ⁻¹)	Air filled porosity (vol %)	Water holding capacity (vol %)
Pine litter	5.3	30	5.4	72.5
Composted pine bark	5.7	470	6.7	64.3
Feedlot manure (cattle)	7.3	2 250	-	-
90 % PN – 10 % FM	5.7	300	5.4	55.6

The pH, electrical conductivity (EC), air filled porosity (AFP) and field capacity of pure pine litter and pure composted pine bark are presented in Table 2. Ingram *et al.* (1993) regard a pH measured in water ranging between 5.5 and 6.5 as optimal for substrate extracts. The pH of pine litter was below optimum, but that of composted pine bark fell inside the optimum range. To raise the pH of pine litter to within the optimum range partially composted feedlot manure, with a pH of 7.3, was added to the pine litter at a rate of 10 % by volume. The pH of the resulting mixture was 5.7 (Table 2). In substrate extracts an electrical conductivity (EC) ranging between 200 and 350 mSm⁻¹ is regarded as optimum (Hanlon, McNeal, & Kidder, 2002). As with pH, pine litter had an electrical conductivity lower than the optimum, but that of composted pine bark was higher than the optimum (Table 2). Adding 10 % feedlot manure to the pine litter increased the electrical conductivity of the mixture to 300 mSm⁻¹, which was within the optimum range. Bunt (1988) considers an AFP of 10 to 20 % by volume to be desirable for substrates. The AFPs of pine litter and pine bark were both below optimum. Adding 10 % feedlot manure to the pine litter did not increase its AFP. Smith (undated) recommends that substrates used in the propagation of vegetable transplants should have a WHC of at least 20 % by volume. The WHCs of pine litter, composted pine bark, and of the mixture of 90 % pine litter and 10 % feedlot manure were all considerably higher than 20 % (Table 2).

Greenhouse experiments

The germination percentage, dry shoot mass and dry root mass of cabbage, lettuce, and tomato seedlings grown in the

two different substrates at five levels of nutrient supply are presented in Tables 3, 4 and 5, respectively. Mean germination percentages of cabbage seedlings were 92.3 % in pine bark and 75.0 % in the pine litter substrate. Those of lettuce were 82.5 % in pine bark and 69.5 % in pine litter, and of tomatoes 81.9 % in pine bark and 79.1 % in pine litter. According to Jansen (22-07-2002, TerraNova Nursery), commercial producers of vegetable transplants in South Africa consider a germination percentage of 90 % or more as satisfactory and values lower than 90 % as too low. The results obtained indicate that overall conditions affecting germination percentage in this study were sub-optimal, with only the germination percentage of cabbages planted in the pine bark exceeding the industrial minimum requirement. For two of the three vegetables, namely cabbages and lettuce, the germination percentages in composted pine bark were significantly higher than in the mixture of pine litter and feedlot manure. The germination percentages of tomato seed in the two substrates were nearly equal. The low germination percentages recorded in the pine litter-feedlot manure mixture was most likely the result of phytotoxicity caused by secondary metabolites present in the pine needles. Important among these metabolites are oleoresin and phenolics (Kainulainen & Holopainen, 2002). Oleoresin is a complex mixture of volatile terpenes and non-volatile resin acids (terpenoids), which have variable water solubility. The main function of oleoresin is to defend conifers against herbivores and pathogens (Kainulainen & Holopainen, 2002), but monoterpenes in pine litter can also have allelopathic effects on other plants (Wilt, Miller & Everett, 1988). Similarly, phenolics primarily act as toxins and deterrents to pathogens and herbivores (Kainulainen &

Holopainen, 2002), but can also affect seed germination, seedling establishment and nutrition of other plants (Berglund, 2004). High levels of tannins were identified as the cause of poor and inconsistent growth of vegetable seedlings in uncomposted and poorly composted pine bark (Maggs, 1985; Van Schoor *et al.*, 1990). Composting pine bark following the addition of urea or another suitable source of ammonium-N, necessary for active composting and rapid rise of the temperature in the stack to occur, reduced tannin levels to non-toxic levels within a period of 12 weeks (Van Schoor *et*

al., 1990). In pine litter Kainulainen and Holopainen, (2002) report that decomposition in litter bags reduced the concentration of monoterpenes to 6 %, resin acids to 35 % and total phenolics to 17 % of the original concentrations in the litter samples. This suggests that composting the pine litter-feedlot manure mixture before using it as a substrate for the propagation of vegetable transplants may address the problem of low germination percentages. Furthermore, composting is also expected to reduce the risk of introducing any pathogens to the substrate (Handreck & Black, 1994)

Table 3 Germination percentage, dry shoot mass and dry root mass of cabbage seedlings grown in two different substrates at five levels of nutrient supply

Nutrient supply level	Germination percentage			Dry shoot mass (mg)			Dry root mass (mg)		
	M1	M2	Mean	M1	M2	Mean	M1	M2	Mean
1: 0 granules per cavity ¹				2.2	2.1	2.2	0.5	0.3	0.4
2: 8 granules per cavity				5.0	6.3	5.7	1.7	1.0	1.4
3: 16 granules per cavity				11.5	12.5	12.0	2.8	2.0	2.4
4: 32 granules per cavity				20.1	20.6	20.4	4.9	5.1	5.0
5: 64 granules per cavity				31.4	28.8	30.1	19.3	8.6	14.0
Mean	75.1	92.3	83.7	14.0	14.1	14.1	5.8	3.4	4.6
Least significant difference		P=0.05			P=0.05			P=0.05	
Substrate means		6.3			NSD			NSD	
Nutrient level means		-			3.5			1.4	
Nutrient level means in same substrate		-			3.2			NSD	
Nutrient level means in different substrates		-			NSD			NSD	

¹ Refers to the number of granules of controlled-release Hortico fertilizer added to each cavity

² NSD = differences are not statistically significant

M1 = Pine litter substrate consisting of 90 % pine litter and 10 % feedlot manure

M2 = Pine bark substrate consisting of composted pine bark

Table 4 Germination percentage, dry shoot mass and dry root mass of lettuce seedlings grown in two different substrates at five levels of nutrient supply

Nutrient supply level	Germination percentage			Dry shoot mass (mg)			Dry root mass (mg)		
	M1	M2	Mean	M1	M2	Mean	M1	M2	Mean
1: 0 granules per cavity ¹				0.5	2.4	1.5	0.6	1.3	1.0
2: 8 granules per cavity				4.7	7.3	6.0	2.7	2.8	2.8
3: 16 granules per cavity				9.8	12.9	11.4	4.3	3.9	4.1
4: 32 granules per cavity				20.2	17.5	18.9	5.4	16.2	10.8
5: 64 granules per cavity				30.9	21.2	26.7	14.3	20.6	17.5
Mean	69.5	82.5	76.0	13.2	12.3	12.8	5.5	9.0	7.3
Least significant difference		P=0.05			P=0.05			P=0.05	
Substrate means		6.3			NSD			NSD	
Nutrient level means		-			2.5			4.0	
Nutrient level means in same substrate		-			11.6			1.6	
Nutrient level means in different substrates		-			NSD			NSD	

¹ Refers to the number of granules of controlled-release Hortico fertilizer added to each cavity

² NSD = differences are not statistically significant

M1 = Pine litter substrate consisting of 90 % pine litter and 10 % feedlot manure

M2 = Pine bark substrate consisting of composted pine bark

Table 5 Germination percentage, dry shoot mass and dry root mass of tomato seedlings grown in two different substrates at five levels of nutrient supply

Nutrient supply level	Germination percentage			Dry shoot mass (mg)			Dry root mass (mg)		
	M1	M2	Mean	M1	M2	Mean	M1	M2	Mean
1: 0 granules per cavity ¹				0.4	2.0	1.2	0.4	1.2	0.8
2: 8 granules per cavity				4.7	7.3	6.0	2.0	2.9	2.5
3: 16 granules per cavity				8.1	11.9	10.0	4.0	4.9	4.5
4: 32 granules per cavity				17.2	20.9	19.1	5.5	16.6	11.1
5: 64 granules per cavity				31.2	30.9	31.1	10.0	17.2	13.6
Mean	79.1	81.9	80.5	12.3	14.6	13.5	4.4	8.7	6.6
Least significant difference		P=0.05			P=0.05			P=0.05	
Substrate means		6.3			NSD			NSD	
Nutrient level means		-			3.6			6.9	
Nutrient level means in same substrate		-			3.5			5.0	
Nutrient level means in different substrates		-			NSD			NSD	

¹ Refers to the number of granules of controlled-release Hortico fertilizer added to each cavity

² NSD = differences are not statistically significant

M1 = Pine litter substrate consisting of 90 % pine litter and 10 % feedlot manure

M2 = Pine bark substrate consisting of composted pine bark

Once emerged, growth of the vegetable seedlings in the two substrates was very similar. Mean dry shoot mass of cabbage seedlings was 14.1 mg in pine bark and 14.0 mg in pine litter (Table 3). That of lettuce was 12.3 mg in pine bark and 13.2 mg in pine litter (Table 4) and tomatoes 14.6 mg in pine bark and 12.3 mg in pine litter (Table 5). Statistically, mean dry shoot mass of cabbage, lettuce and tomato seedlings did not differ significantly between pine bark and pine litter, suggesting that the concentrations of any phytotoxic compounds that may have been present in the pine litter were reduced as the experiments progressed, possibly as a result of leaching.

Nutrient supply had a significant positive effect on dry shoot mass of all three vegetable seedlings. Dry shoot mass of cabbage seedlings increased from 2.2 mg at the lowest nutrient level to 30.1 mg at the highest. In tomato seedlings it increased from 1.2 mg at the lowest nutrient level to 31.1 mg at the highest, and in lettuce from 1.5 mg to 26.1 mg. In cabbages and tomatoes there was no interaction between substrate and nutrient level. In both cases the growth response of the seedlings to increasing nutrient supply measured in terms of dry shoot mass was very similar in both substrates. In lettuce the interaction between substrate and nutrient supply had a highly significant effect on dry shoot mass of the seedlings. Increases in dry shoot mass to increments in nutrient supply were larger in the pine litter substrate than in the pine bark. However, on the whole, growth of test crop seedlings in the two substrates, measured in terms of dry shoot mass, was very similar.

Mean dry root mass of cabbage seedlings was 3.4 mg in composted pine bark and 5.8 mg in pine litter (Table 3); lettuce 9.0 mg in pine bark and 5.5 mg in pine litter (Table 4) and tomatoes 8.7 mg in pine bark and 4.4 mg in pine litter (Table 5). For all three vegetables differences in mean dry root mass between substrates were not statistically significant. As with dry shoot mass nutrient supply had a significant positive effect on the dry root mass of all three vegetable

seedlings. The dry root mass of cabbage seedlings increased from 0.4 mg at the lowest nutrient level to 14.0 mg at the highest, that of lettuce seedlings from 1.0 mg to 17.5 mg and that of tomatoes from 0.8 mg to 13.6 mg. The interaction between substrate and nutrient supply had a highly significant effect on dry root mass of tomatoes. Yield increases per increment in nutrient supply were steeper in the pine bark than in the pine litter.

The data presented in Tables 3, 4 and 5 also suggested that little if any of the nutrients present in the feedlot manure became available to the vegetable transplants during the 6-week growing period. Evidence to that effect was the absence of statistically significant differences in the mean shoot and root mass between transplants of all three vegetables growing in the composted pine bark and pine litter – feedlot manure substrates that were not enriched with controlled-release fertilizer.

Overall, the results of this study have highlighted the limitations of using pine litter as a substrate for use in vegetable transplant production. The most serious limitation was that seed germination percentages in the pine litter substrate were inadequate. A lesser limitation was that growth of seedlings in pine litter, as indicated by the dry shoot and root mass of the plants six weeks after planting, tended to be slightly less vigorous than in pine bark.

Conclusions

The results obtained in laboratory tests and greenhouse experiments demonstrated that a mixture of 90 % pine litter and 10 % feedlot manure for use in the propagation of vegetable seedlings in trays performed less than composted pine bark, the benchmark substrate of the local vegetable seedling industry, which gave higher germination percentages and tended to provide for better growth of roots, but not of shoots. Incorporation of animal manure in the pine litter improved

the chemical properties of the litter, but adding 10 % feedlot manure to the pine litter did not appear to have a material effect on the nutrient supply to the transplants, necessitating the addition of plant nutrients in the form of a controlled release chemical fertilizer mixture to achieve satisfactory growth of the plants. Additional research is needed to investigate the effects of type and concentration of animal manure on transplants growing in pine litter. Possible adverse effects of phytotoxins in uncomposted pine litter must also be investigated. Furthermore, there is a need to determine the effect of composting and liming pine litter-manure mixtures, before they are used as a substrate in the propagation of vegetable transplants.

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