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MEDIA STEAMING AND COCO-COIR ENHANCE GROWTH OF ROUGH LEMON (*Citrus jambhiri* L.) STOCK

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Essential nutrient profiling of natural and steam sterilized soil and soilless media components was performed to obtain better rough lemon seedling growth in containers. Steam sterilization of potting media components did not alter physico-chemical properties of the media. Three fold higher seed germination (95%), two fold better seedling growth 9.17 cm plant height and more leaf area 14.4 cm^{-2} was observed in sterilized sand media compared with conventional nursery media containing sand, silt and farm yard manure (w/w, 1:1:1). Media amendment with coco coir enhanced plant growth; up to three fold including seedling survival (90%), shoot length 5.25 cm and stem girth 0.63cm. Two fold increase in leaf size 21.17 cm length and 14 cm width, three fold higher numbers of leaves 6.33, two fold more number of roots 30.17 and root length 12.65 cm were obtained in coir media compared with control and other organic media amendments. These findings suggest a wider application of cost-effective environment friendly steam sterilization and coco-coir as organic media amendment for enhanced nursery growth in healthy environment in containers.
Keywords: Potting media, rootstock, citrus, plant growth, nursery

INTRODUCTION

Citrus industry in Pakistan is dependent on conventional commercial nurseries which are being managed unscientifically without considering soil hygiene. Owing to this, citrus orchards are highly vulnerable to soil pests like *Phytophthora*, *Fusarium* and *Pythium* sp. (Rehman *et al.*, 2012; Youpensuk *et al.*, 2012). Availability of sanitized citrus rootstock is pre-requisite for the maintenance of citrus industry and establishing new orchards. Rough Lemon (*Citrus jambhiri* L.) rootstock is commonly used for producing good quality Kinnow mandarin in major citrus growing areas of Pakistan (Khan *et al.*, 2006). It provides deep root system with significant drought resistance and high vulnerability to *Phytophthora* and *Fusarium* (Cao *et al.*, 2013). These pathogens are known to cause citrus decline and ~38% of these pathogens were found in orchard soil (Rehman *et al.*, 2012). Conventionally, the nurseries are being raised in the same infested soil suggesting a dire need to shift to containerized nursery system. In this kind of nurseries, sterilized seeds of rootstocks are raised in different types of containers including plastic pots and polyethylene bags filled with sterilized potting media to block dissemination of soil pests from nursery to the orchards. The rootstocks are grafted with bud wood obtained from sanitized mother plants maintained under screen houses (Usman and Fatima, 2013).

Container grown nursery offers salient advantages like a) manipulation of different potting media ingredients for better plant growth b) better media hygiene to suppress soil pests using different sterilization methods c) easy plant portability without disturbing its root system d) plant production and propagation under controlled environmental conditions throughout the year (Khan, 2005; Ouma, 2007). In California and Florida, citrus nursery is produced in Citra-pots containing sterilized potting media and citrus propagation is in greenhouses to exclude disease and pest infestation.

Conventionally, the toxic pesticides like methyl bromide were used to control soil born plant diseases which is responsible for Ozone depletion (Ajwa *et al.*, 2003). Banning of toxic fumigants like Methyl bromide under Montreal protocol, an international treaty, promoted use of steam sterilization as alternative environment friendly method (UNEP, 2001) to control soil pests in fruits and vegetable nurseries (Yakabe *et al.*, 2010; Sharma *et al.*, 2011). Steam penetrates more efficiently into the media and is more effective and safe in controlling soil pests compared with dry heating and using soil fumigants (Hansen *et al.*, 2011). Steaming is recommended particularly for warm and dry climates (van Leonen *et al.*, 2003). It ensures supply of clean and healthy nursery plants to the growers leading to orchards with long productive life, higher yields and quality fruits. In Natal Sweet orange, fruit yield was 21% higher from healthy nursery plants compared with infected plants (Fabricio-Packer *et al.*, 2011).

Plant growth in containers depends upon nutrient filled porous soil or soilless media for better root development in a limited space. Different natural growth media have been analyzed for nutrition and their effects on plant growth in

Sour orange and rough lemon rootstocks (Ma *et al.*, 2000; Bhagat *et al.*, 2013). The physico-chemical properties greatly vary among soil and soilless media components (Ahmad *et al.*, 2012). Both types of media components are usually infested with pathogens and weed seeds thus need proper sterilization. However, information about the effect of sterilization on physico-chemical properties of the potting media in container grown nurseries is lacking. Therefore, the study was aimed to investigate effect of steam sterilization on chemical properties of different potting substrates and standardization of a cost effective potting mix for efficient plant growth.

MATERIALS AND METHODS

***Potting media sterilization and chemical analysis*:** Standard soil mix comprising of fine river sand, silt and farm yard manure (1:1:1; control media) being used by commercial fruit nurseries, modified soil mixes and organic media components (Table 1) were analyzed for chemical changes before and after steam sterilization.

The media were sterilized using locally manufactured unit for steam sterilization (Mistquay Int., Pakistan) at 80-90°C for 25 minutes at 15 psi following Allan *et al.*, (1981) with certain modifications. The temperature and pressure conditions were optimized since most of the fungi, bacteria, insects, nemtaodes and weed seeds are killed up to 70-80°C (Baker, 1957; Usman and Fatima, 2013). The media were immediately cooled down and filled in transparent plastic pots. Total quantity of potting media was taken as 1000 g w/w basis in each combination. Nitrogen in potting media was determined using Kjeldahl method (Jackson, 1962; Bremner, 1996). Phosphorous was extracted from potting media by Olson method (Olsen *et al.*, 1984) and was determined using spectrophotometer. Potassium was extracted from potting media using ammonium acetate method and was determined using flame photometer (Jenway PFP-7). The pH values of media were recorded using Beckman pH meter (USA) in 250 g of media saturated with distilled water. Electric conductivity of the saturation extract (ECe) was measured by electric conductivity meter (CM-1 Mark v, Kent) in $\mu S\ cm^{-1}$ and moisture percentage was noted (Sparks *et al.*, 1996).

***Seed sterilization and germination*:** Rough lemon fruits were collected from the University of Agriculture, Faisalabad (UAF) fruit gardens and Citrus Research Institute (CRI) Sargodha. Fruit were horizontally cut into two halves; seeds were extracted and washed in tap water to remove pulp and other ricinis material. The seeds were put in masculine cloth and dipped in stirring hot water at 52°C for 10-15 minutes for uniform seed treatment to remove seed coat borne fungi like Phytophthora sp. (Bridges and Youtsey, 1966). The seeds were cooled down; dipped in 10% NaOCl solution for treatment against fungal infections for 15-20 min and washed 2-3 times with sterilized distilled water to remove excessive fungicide. The fungicide coating of seeds protects against recontamination during handling and storage of seeds. Seeds were tumble dried for 5 minutes before sowing in sterilized media or storage at 4°C. Sterilized rough lemon seeds were sown in seed germination media containing soil and organic matter OM (T_o-T_4; Table 1) for germination. The pots were kept under 16 hr. photoperiod and 25±1°C temperature for optimal seed germination. Sterilized distilled water was supplied through trays underneath the pots at alternate day intervals to avoid any microbial infestation. Additionally autoclaved Hoagland's solution was applied with sterile distilled water (1:10 v/v) fortnightly. Seed germination (%) and seedling growth pattern was noted in different potting media after four weeks of sowing.

***Seedling growth behaviour in sterilized soil and soilless potting media*:** Different soil and soilless sterilized potting media (Table 2) were tested for their effect on plant growth after transplanting under long summer days (16/8 hours day/night cycle) under greenhouse conditions. The quantity of different potting media components was taken as 3 kg (w/w) in total in each black color polyethylene container (14 x 10 inches size). Seedlings of uniform age (12-14 weeks) were transplanted in different media (Table 2) and placed on elevated trays (30 inches above ground) in the greenhouse. The canal water, stored in a metallic tank, was sterilized by passing pressurized steam from steam generator through water till boiling temperature to avoid microbial infections. After 70 days of transplanting, data were collected for

Table 1. Media formulations for nutrient profiling and chemical analysis before and after steam sterilization

Media types	Treatments	Sand (g)	Silt (g)	FYM (g)	Coconut coir (g)	Compost (g)	Spent mushroom compost (g)	Poultry manure (g)
Soil + Organic matter (OM)	T_o	330	330	330	-	-	-	-
	T_1	250	250	500	-	-	-	-
	T_2	500	250	250	-	-	-	-
	T_3	500	500	-	-	-	-	-
	T_4	1000	-	-	-	-	-	-
OM components	T_5	-	-	-	1000	-	-	-
	T_6	-	-	-	-	1000	-	-
	T_7	-	-	-	-	-	1000	-
	T_8	-	-	-	-	-	-	1000

Table 2. Plant growth media formulations for Rough lemon growth under greenhouse conditions

Treatments		Sand (kg)	Silt (kg)	FYM (kg)	C. Coir (kg)	P.W (kg)	S.C.M (kg)	Compost (kg)
Sand: Silt: FYM (1:1:1)	T_o	1	1	1	-	-	-	-
Sand: Silt: FYM (2:1:1)	T_1	1.5	0.75	0.75	-	-	-	-
Coconut coir (CC)	T_2	-	-	-	3	-	-	-
FYM: CC (1:1)	T_3	-	-	1.5	1.5	-	-	-
Sand: FYM: CC (1:1:1)	T_4	1	-	1	1	-	-	-
Sand: FYM: Poultry waste PW (1:1:1/10)	T_5	1.45	-	1.45	-	0.1	-	-
Sand: FYM: PW (1:1:1/20)	T_6	1.4	-	1.4	-	0.2	-	-
Sand: FYM: SMC (1:1:2)	T_7	0.75	-	0.75	-	-	1.5	-
Sand: FYM: SMC (2:1:1)	T_8	1.5	-	0.75	-	-	0.75	-
Sand: Silt: FYM: CC: PW (1:1:1:1: 1/20)	T_9	0.737	0.737	0.737	0.05	0.737	-	-
Compost	T_{10}	-	-	-	-	-	-	1

survival (%), seedling growth, plant height (cm), stem girth (mm), internodal distance (cm), root length (cm) and number of primary roots. Total number of leaves was counted in seedlings. Leaves were measured along midrib for leaf length and at right angles at the widest point of the leaf blade for leaf width (cm). The curved leaves were measured along the arc and L/W ratio was calculated (Teich and Speigel-Roy, 1972).

***Experimental layout*:** The experiment was laid out according to Completely Randomized Design (CRD). Twenty biological replicates (seedlings) were used per treatment and each experiment was repeated twice. Significance among treatment means was compared using Duncan's Multiple Range (DMR) test (Steel *et al.,* 1997). The delta values were calculated by subtracting the pre-transplanting plant growth values from post-transplanting values to observe net gain in plant growth in response to media. The calculated delta values were plotted in Fig. 6-8.

RESULTS

***Effect of sterilization on nutrient availability and chemical properties of the potting media*:** Media sterilization did not significantly affect the availability of essential elements (NPK), and chemical properties of the media including pH, EC and moisture (%) (Fig. 1a-c). Organic media components poultry manure (PM) and spent mushroom compost (SMC) were found highly rich in N (0.47 mg kg^{-1} - 0.53 mg kg^{-1}) and P (30 mg kg^{-1} to 57 mg kg^{-1}) compared with control media containing sand, silt and composted FYM (1:1:1) and all other media types (Fig. 1a, b). Poultry manure (PM) was also highly rich in available K (517 mg kg^{-1}) followed by media rich in organic components including SMC, compost and sand + silt + FYM (1:1:2). Sand media consistently remained the lowest resource in available N, P and K compared with other media types (Fig. 1a-c). Analysis of chemical properties of the media revealed PM having the highest pH values 8.6-8.7 followed by other organic media components including SMC, compost and sand + silt + FYM (1:1:2) (Fig. 2a). Coco coir (CC) and sand + FYM (1:1) media showed the lowest pH values 6.7-7.0 respectively compared with control and other media types. Electric conductivity was significantly higher in PM 7.6-7.5 µS cm^{-1} compared with control and other media (Fig. 2b). Sand and CC media showed the lowest EC values 3.21-3.82 µS cm^{-1}. Moisture (%) was significantly higher in SMC media 8.6%-10% followed by compost compared with control and other media types (Fig. 2c). The lowest moisture (%) was found in PM and sand media. No significant impact of sterilization was found on nutrient availability and chemical properties of the media, hence, only sterilized media types were compared for seed germination and plant growth studies.

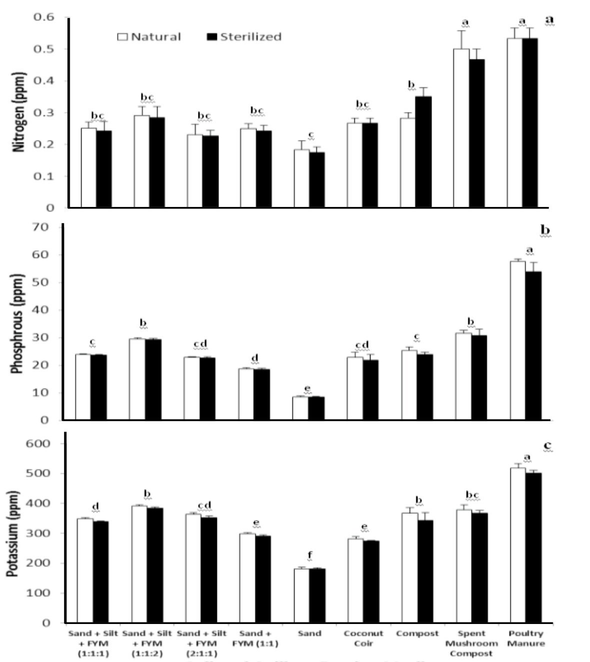

Figure 1. Availability of essential nutrients NPK in different soil media and organic potting media components. The values are means of three technical replicates and bars represent standard errors of the mean. The bars show differences at P <0.01.

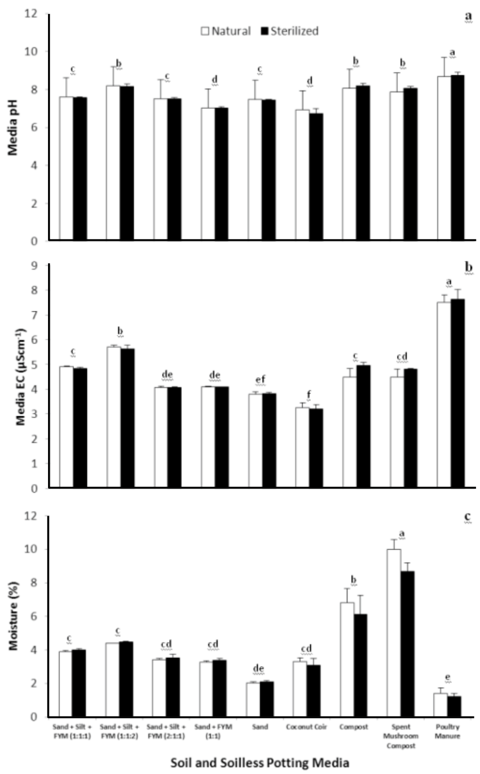

Figure 2. Response of different soil and soilless potting media for pH, EC (µS cm^{-1}) and Moisture (%) content. The values are means of three technical replicates and bars represent standard errors of the mean. The bars show differences at P <0.01.

***Seed germination and seedling growth in sterilized potting media under controlled conditions (CC) and in open field (OF)*:** Seeds were sown in sterilized potting media for germination under controlled conditions. An increase up to threefold was observed in seed germination in sand media (95%) compared with the control media (30%) having sand, silt and FYM (1:1:1) as shown in Table 3. Potting media sterilization showed no adverse impact on frequency of seed germination and seedling growth suggesting sterilized sand as better media for rough lemon seed germination (Fig. 3a-b). In sand, plant growth was significantly (P<0.01) enhanced including plant height from 5.30 cm to 9.17 cm, internodal distance from 0.81 cm to 1.58 cm and stem diameter from 1.67 mm to 1.8 mm compared with control media. Seedlings in sand media showed more number of leaves from 8.6 to 14.6 and enhanced leaf area from 4.85 cm^{-2} to 14.43 cm^{-2} compared with control media. Root growth was also markedly enhanced from 6.99 cm to 14.06 cm in sand media compared with control media. Shoot length, root length and no. of leaves were found significantly higher in seedlings raised in sterilized sand media under controlled light and temperature conditions compared with seedlings raised in soil media in the open environment (Fig. 4). These findings suggested that healthy rough lemon stock may be successfully raised under controlled environmental conditions in sand media. Seedling growth rate was much better when compared with plant growth in the open environment. After attaining proper size, seedlings were transferred to large size plastic containers for better plant growth.

***Effect of potting media on plant growth under greenhouse conditions*:** Coco coir media T2 significantly (P<0.01) enhanced seedling survival (90%), plant height (5.25 cm), stem girth (0.63 mm) and internodal distance (0.95 cm) compared with control media and other media types (Fig. 3c-g, 5a-c). These findings indicated that traditionally used control media is not potentially suitable for container grown nursery production in citrus. Similar trend was observed in leaf growth parameters. Number of leaves and leaf growth was significantly (P<0.05) higher on CC media (T2; 6.33, 2.17 cm length, 1.4 cm width) followed by media containing Sand: Silt: FYM: CC: PW (T9; 2:1:1:1/20) compared with control media (2.17 leaves, 0.97 cm length, 0.72 cm width) as shown in Figures 6a-b. Leaf number was at the minimum in Sand: FYM: SMC (1:1:2; T7) media.

Table 3. Seed germination and seedling growth response in sterilized potting media

Media composition	Plant growth characteristics						
	Seed germination	Plant height (cm)	Internodal distance (cm)	Stem diameter (mm)	Number of leaves	Leaf area (cm^{-2})	Root length (cm)
Sand + Silt + FYM (1:1:1; control)	30 ± 3.85 b	5.33 ± 0.09 c	0.81 ± 0.06 c	1.67 ± 0.09 b	9.5 ± 0.58 c	4.85 ± 0.35 c	6.99 ± 0.21 c
Sand + Silt + FYM (1:1:2)	34.25 ± 2.22 b	4.13 ± 0.03 e	0.72 ± 0.07 c	1.33 ± 0.03 c	8.67 ± 0.33 c	5.7 ± 0.05 c	6.85 ± 0.57 d
Sand + Silt + FYM (2:1:1)	41.11 ± 2.22 b	6.07 ± 0.09 b	1.29 ± 0.03 b	1.57 ± 0.07 b	12.5 ± 0.58 b	8.54 ± 0.05 b	9.76 ± 0.03 b
Sand + FYM (1:1)	30.45 ± 2.22 b	4.03 ± 0.07 d	0.76 ± 0.33 c	1.4 ± 0.06 c	8.67 ± 0.08 c	6.09 ± 0.19 c	7.57 ± 0.09 c
Sand	94.45 ± 2.22 a	9.17 ± 0.03 a	1.58 ± 0.03 a	1.8 ± 0.01 a	14.67 ± 0.33 a	14.43 ± 0.11 a	14.06 ± 0.22 a

The values are mean of three technical replicates ± standard error of the mean. Means sharing different letters in a column are highly significant (P<0.01).

Figure 3. Seed germination and seedling growth responses in sterilized potting media. Figures show better seed germination in a) sand compared with b) Sand + FYM (1:1) under controlled conditions after 50 days of sowing. Figures c-i shows seedling growth response in Coconut Coir (T3; c,e) and Coconut Coir + FYM (T4; d,f) under greenhouse conditions. Note better fibrous root system developed in T3 media (g). Figures h-i show sprouted T-grafting in rough lemon and grafted Kinnow plants in the green house.

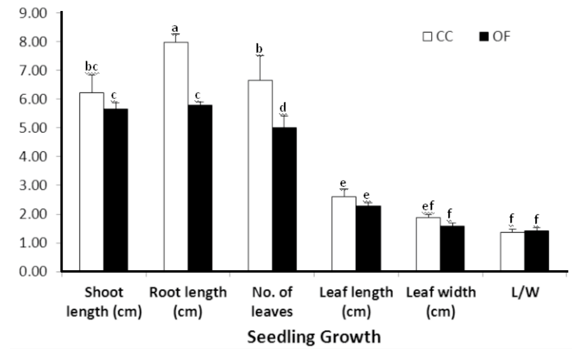

Figure 4. Seedling growth response in sand media under controlled conditions (CC), and seedlings raised in soil in open field (OF). The values are mean of three technical replicates and error bars represent standard errors of the mean. The bars show differences significant at P <0.05.

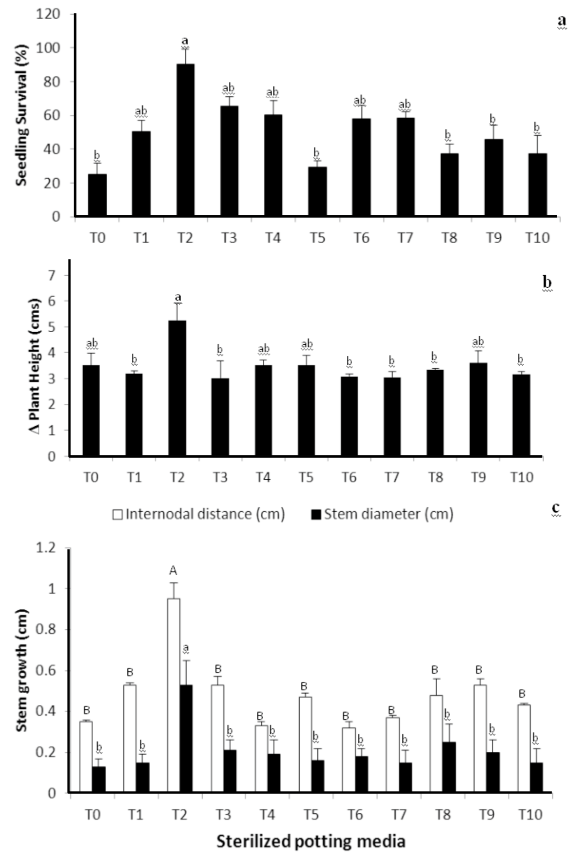

Figure 5. Change in plant growth (cm), stem girth (mm) and internodal distance (cm) in Rough lemon seedlings after 75 days of transplanting in different potting media during summer season. Values are mean of three technical replicates and error bars represent standard error of the mean. The bars show differences significant at P <0.05.

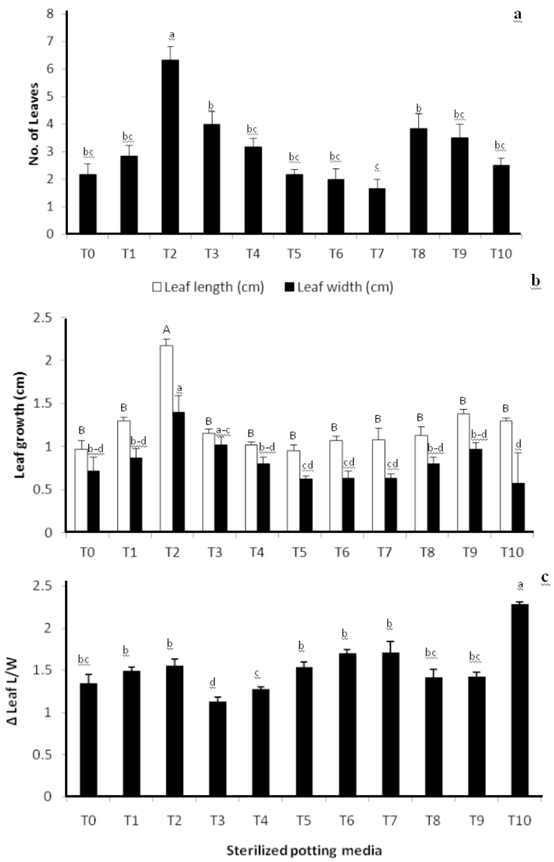

Figure 6. Rise in number of leaves, leaf length (cm) and leaf width (cm) and leaf L/W ratio in Rough lemon seedlings after 75 days of transplanting in different potting media. Values are mean of three technical replicates and error bars represent standard error of the mean. The bars show differences significant at P <0.05.

The leaf L/W ratio was at the highest in PM media 2.28 showing elongated leaves compared with low ratio indicating broader leaf blades in all other media types (Fig. 6c). Addition of CC in FYM (T3) significantly reduced the L/W ratio up to 1.27 indicating an increased leaf width and reduced leaf length compared with other media. These findings indicated that use of CC alone developed maximum number of leaves and enlarged leaf size compared with all other media component combinations. Root length was significantly enhanced in CC in combination with FYM (12.6 cm, T3 media; Fig. 7a). Induction of primary roots was doubled in CC media (T2) only (Fig. 7b) compared with all other media types indicating CC media as a potential media component for higher root biomass development, enhanced secondary root growth and developing a fibrous root system. Use of both CC and sand as a major component of container grown media comprising sand + silt + FYM + CC (40%:10%:10%:40%) markedly enhanced plant growth and development and scion bud sprouting (Fig. 3h, i); and was used for further containerized citrus nursery production.

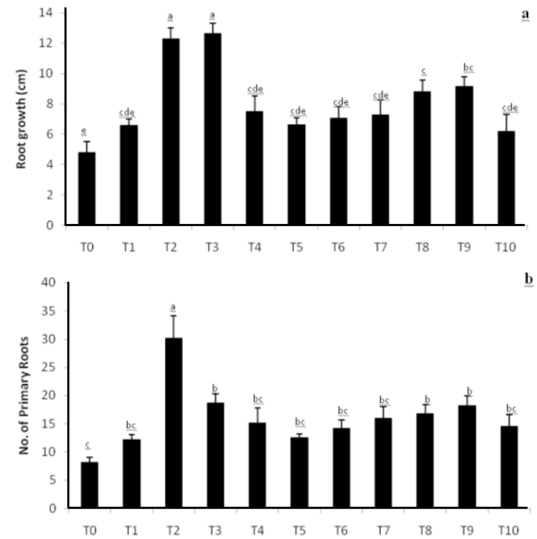

Figure 7. Root growth (cm) and number of primary roots of Rough lemon seedlings after 75 days of transplanting in different potting media during summer season. Values are mean of three technical replicates and error bars represent standard error of the mean. The bars show differences significant at P <0.05.

DISCUSSION

***Short duration steaming at higher temperature did not affect physico-chemical properties of media*:** Important factors effecting physico-chemical properties of the media in response to steaming include steaming temperature, duration and availability of mineral elements and exchangeable cations in the media. Nutrition profiling of different types of soil mix and soilless media with or without steaming revealed no significant impact on NPK availability, media chemical properties including pH, EC and moisture retention percentage. These findings are corroborative to similar reports in field crops and tree crops showing no marked changes in physico-chemical structure of the soil after steaming (Elsgaarda *et al.*, 2010), oven sterilization of soil at 120°C for 24 hrs (Washa *et al.*, 2012) or soil steaming with or without addition of CaO (Gelsomino *et al.*, 2010). On the contrary, there are reports of increase in NH^{+}_{4}-N level and decrease in NO^{-}_{3}-N level in response to soil sterilization at 60°C for two hours (Yamamoto *et al.*, 2008), increase in K pool in response to shallow steam injection at 40-60°C (Gelsomino *et al.*, 2010) and increase in soil pH, electrical conductivity and Olsen-P (Tesi *et al.*, 2007). It is more likely that prolonged steam application at temperature up to 60°C in previous reports may underlie these changes in chemical properties of the media. In this study, use of steam sterilization at higher temperature up to 90°C for short time 20-25 min did not cause such changes in the media. Differential availability of microbial biomass N in the media in different steam sterilization studies may also give different results. Use of sand in the media enhances its bulk density, reduces the nutrient retention and moisture percentage (Mastalerz, 1977). Enhancing organic supplement of FYM in the sand or soil media reduced pH due to development of carbonic acid in the media and enhanced EC due to rise in the available ionic content in media compared with sand. Media pH was increased by doubling sand proportion in the sand + silt + FYM media (2:1:1; Figure 2a). Moisture retention (%) in sand media was enhanced leading to more soil compaction and reduced porosity when sand was supplemented with silt or FYM. Soil amendments with non-sterilized FYM decreased media pH (Ayesha *et al.*, 2011) while addition of sphagnum peat moss increased the available water holding capacity (Bigelow *et al.*, 2004). We observed similar results in sterilized media. Nutrient profiling of organic media components revealed PM rich in NPK. SMC was rich in N compared with P and K. Coco coir has been used as cheap alternative source to peat moss however coir properties vary from source to source. It is more suitable soilless media and gave better productivity compared with peat media (Islam *et al.*, 2002). Coir was at relatively lowest in pH (6.9) and EC (3.27 µS cm^{-1}) compared with all other media components including control media. Similar qualities of CC are reported with higher water holding capacity, excellent drainage, physical resilience and a renewable resource of potting substrate (Abad *et al*., 2005; Hyder *et al.*, 2008).

Sterilized sand media enhanced seed germination, seedling growth and organic media amendments enhanced plant growth and development**:** It has been established that addition of natural organic manure could change media properties by effects of thermal heating on soil microbiota resulting in steam induced changes in soil chemical properties (Gelsomino *et al.*, 2010). It has been reported that steam sterilized fresh manure reduced accumulation of toxic substances in the media leading to better plant growth and root mass in citrus and avocado (Allan *et al.*, 1981). In contrast, we have observed poor seedling growth in media when amended with decomposed sterilized manure compared to sand media. Sand media enhanced seed germination up to three fold and ruled out use of modified soil mixes for seed germination and initial seedling growth. In live oak, sand beds provided better plant growth compared with control sandy loam soil having higher water holding capacity (Bryan *et al.*, 2011). These findings are corroborative to our results since steam induced increase of available N forms is likely to further increase crop growth. After transplanting in CC media, seedlings showed outstanding growth in all critical plant growth parameters including survival percentage, net plant growth, stem girth and internodal distance, number and size of leaves, root length and number of roots compared with control media and other media amendments. These findings are corroborative to earlier reports in different fruit, vegetable and ornamental crop production on coir media (Ayesha *et al.*, 2011; Ahmad *et al.*, 2012; Tariq *et al.*, 2012). Additionally, steaming exerted no adverse impact on coir nutrient availability evident from better seedling growth in coir compared with other media. On the contrary, Khan *et al.* (2006) reported sand, peat and SMC as better planting natural media for citrus growth. We found CC better than SMC as organic supplement in media for Citrus growth. In addition, coir is reported to suppress 35-90% of soil borne pathogens like Phytophthora sp. causing damping off due to association of this substrate with *Aspergillus terreus* and *Trichoderma viride* (Hyder *et al.*, 2008; Waller *et al.*, 2008). Better plant growth has been reported in sterilized media as soil heat treatment enhanced plant growth, biomass production, quality and yield compared with non-sterilized control plants and reduced sapling wilt in apple nurseries (Raj and Sharma, 2009). Autoclaving of organic sources like peat, perlite and vermiculite soil medium improved plant recovery, ex vitro plant height and diameter and total dry weight in sterile media compared with non-sterile media in Kalopanax (Aggangan and Moon, 2012). Similarly in Troyer citrange, root growth enhanced with media pasteurization treatments (Allan *et al.*, 1981). Soil treatment enhanced number of branches, number of roots and root length in Dalbergia (Washa *et al.*, 2012). Coco coir based organic amendment of soil media in this study enhanced nutrient balance and biological resource restoration. Overtime, enhanced plant growth and root biomass could be attributed to heat induced decomposition of organic biomass in sterilized media compared to non-sterilized media thus enhancing fertility as reported in other organic media as well (Aggangan and Moon, 2012).

Conclusions**:** No significant change was noticed in chemical properties of soil and soilless potting media in response to short steaming at moderately higher temperatures. Substantial plant growth improvement emphasizes use of hydrothermal, non-pesticidal steam sterilization in commercial citrus nurseries to raise healthy stock with effective pest and weed control. This study provides first report of environment friendly potting media sterilization in Pakistan and development of cost-effective potting mix for commercial container grown fruit plant nurseries. The technology may further be extended to guava and other fruit crops which are highly susceptible to soil pests.

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REFERENCES

Abad, M., F. Fornes, C. Carrión, V. Noguera, P. Noguera, A. Maquieira and R. Puchades. 2005. Physical properties of various coconut coir dusts compared to peat. HortScience 40: 2138-2144.

Aggangan, N.S. and H.K. Moon. 2012. The effects of soil sterilization, mycorrhizal inoculation, and rates of phosphorus on growth and survival of Kalopanax septemlobus microplants during the acclimatization period. Plant Biotechnol. Rep. 7: 71-82.

Ahmad, I., T. Ahmad, A. Gulfam and M. Saleem. 2012. Growth and flowering of Gerbera as influenced by various horticultural substrates. Pak. J. Bot. 44: 291-299.

Ajwa, H.A., S. Klose, S.D. Nelson, A. Minuto, M.L. Gullino, F. Lamberti, J.M. Lopez-Aranda. 2003. Alternatives to methyl bromide in strawberry production in the United States of America and the Mediterranean region. Phytopathologia Mediterranea 42: 220-244.

Allan, P., D. Lamb, D. Chalton and D. Chalton. 1981. Sterilization and pasteurization of soil mixes. S. African Avocado Growers Assoc. Year book 4: 124-127.

Ayesha, R., N. Fatima, M. Ruqayya, K.M. Qureshi, I.A. Hafiz, K.S. Khan, and A. Kamal. 2011. Influence of different growth media on the fruit quality and reproductive growth parameters of strawberry (*Fragaria ananassa*). Med. Plants Res. 5: 6224-6232.

Bhagat, S., A. Thakur and H.S. Dhaliwal. 2013. Organic amendments influence growth, bud ability and budding success in rough lemon (*Citrus jambhiri* Lush.). Biological Agri. & Hort.: An Int. J. Sustainable Prod. Systems 29: 46-57.

Bigelow, C.A., D.C. Bowman and D.K. Cassel. 2004. Physical properties of three sand size classes amended with inorganic materials or sphagnum peat moss for putting green root zones. Crop Sci. 44: 900-907.

Bremner, J.M. 1996. Nitrogen-Total. p. 1085-1121. In: D.L. Sparks (ed.), Methods of Soil Analysis. Part 3. Chemical Methods. No. 5 in the SSSA Book Series SSSA-ASA, Madison, WI.

Bridges, G.D. and C.O. Youtsey. 1966. Improved disease control through hot water treatment of citrus seed. Proc. Florida State Hort. Soc. 79: 114-115.

Bryan, D.L., M.A. Arnoldb, A. Volderb, W.T. Watsonc, L. Lombardinib, J.J. Sloand, A. Alarcóne, L.A. Valdez-Aguilarf and A.D. Cartmill. 2011. Planting depth and soil amendments affect growth of *Quercus virginiana* Mill. Urban For. and Urban Greening, 10: 127-132.

Cao, M., S. Atta, H. Su, X. Wang, Q. Wu, Z. Li, and C. Zhou. 2013. Molecular characterization and phylogenetic analysis of Citrus viroid V isolates from Pakistan. European J. Plant Pathol. 135:11-21.

Elsgaarda, L., M.H. Jorgensenb and S. Elmholt. 2010. Effects of band-steaming on microbial activity and abundance in organic farming. Soil Agri. Ecosystems & Env. 137: 223-230.

Fabrício-Packer, G.A., S. Eduardo-Sanches, D.S. Simone-Rodrigues, R. Eduardo-Toller and A. Lilian. 2011. Role of healthy nursery plants in orange yield during eight years of Citrus Variegated Chlorosis epidemics. Sci. Hortic. 129: 343-345.

Gelsomino, A., B. Petrovi_cová, F. Zaffina and A. Peruzzi. 2010. Chemical and microbial properties in a greenhouse loamy soil after steam disinfestation alone or combined with CaO addition. Soil Biol. & Biochem. 42: 1091-1100.

Hansen, J.D., J.A. Johnson and D.A. Winter. 2011. History and use of heat in pest control: a review. Int. J. Pest Manage. 57: 267-289.

Hyder, N., J.J. Sims and S.N. Wegulo. 2009. *In Vitro* Suppression of Soil borne Plant Pathogens by Coir. HortTech. 19: 96-100.

Islam, M.S., S. Khan, T. Ito, T. Maruo and Y. Shinohara. 2002. Characterization of the physico-chemical properties of environmentally friendly organic substrates in relation to rockwool. J. Hort. Sci. Biotechnol. 77: 143-148.

Jackson, M.L. 1962. Soil Chemical Analysis. Constable and Co. Ltd., London, UK.

Khan, M.M. 2005. Introduction. p. 1-5. In: M.M. Khan, M. Usman and M. Abbas. (ed.) Citrus Nursery Raising: Principles and Practices. MAS Publishers, Faisalabad, Pakistan.

Khan, M.M., M.A. Khan, M. Abbas, M.J. Jaskani, M.A. Ali and H. Abbas. 2006. Evaluation of potting media for the production of rough lemon nursery stock. Pak. J. Bot. 38: 623-629.

Ma, P.Q., X.L. Tang, W. Wen, Y.R. Wei and C.J. Peng. 2000. Effects of the stroma nutritive soil on the growth of the citrus seedlings. South-China-Fruits, 29: 6-7.

Mastalerz, J.W. 1977. The greenhouse environment. John Wiley and Sons, New York.

Nasir, F.R, J.T. Agha and A.R.S. Mohammed. 1990. Effect of mix composition on seed germination and seedling growth of two citrus rootstocks. Meso. J. Agri. 22: 73-80.

Olsen, S.A.R., M.D. Sant, H.R. Gislerod and K. Solbraa. 1984. Nitrogen balance in bark composts used as growing media. Acta Hort. 150: 193-202.

Ouma, G. 2007. Effect of different container size and irrigation frequency on the morphological and physiological characteristics of Mango (*Mangifera indica* L.) rootstock seedling. Int. J. Bot. 3: 260-268.

Raj, H. and S.D. Sharma. 2009. Integration of soil solarization and chemical sterilization with beneficial microorganisms for the control of white root rot and growth of nursery apple. Sci. Hort. 119: 126-131.

Rehman, A., N. Javed, A.U. Malik and S. Mehboob. 2012. Toxin production by *Fusarium solani* from declining citrus plants and its management. Afric. J. Biotech. 11: 2199-2203.

Sharma, S.D., P. Kumar, S.K. Bhardwaj and A. Chandel. 2011. Symbiotic effectiveness of arbuscular mycorrhizal technology and Azotobacterization in citrus nursery production under soil disinfestation and moisture conservation practices. Sci. Hortic. 132: 27-36.

Sparks, D.L. 1996. Methods of Soil Analysis. Part 3-Chemical Methods. Soil Sci. Soc. America Inc. Madison, WI.

Speigel-Roy, P. and E.E. Goldschmidt. 1996. Biology of Citrus. Cambridge Uni. Press, New York.

Steel, R., J.H. Tome and D. Dickey. 1997. Principles and Procedures of Statistics. A Biometrical Approach, 3rd Ed. McGraw-Hill Publications, New York.

Tariq, U., Shoaib-ur-Rehman, M.A. Khan, A. Younis, M. Yaseen and M. Ahsan. 2012. Agricultural and municipal waste as potting media components for the growth and flowering of *Dahlia hortensis* 'Figaro. Turk. J. Bot. 36: 378-385.

Teich, A.H. and P. Speigel-Roy. 1972. Differentiation between nucellar and zygotic citrus seedlings by leaf shape. Theor. Appl. Gen. 42: 314-315.

Tesi, R., A. Gelsomino, A. Baldi, A. Lenzi and A. Peruzzi. 2007. Soil disinfection with steam alone or combined with CaO in a greenhouse radish crop. Adv. Hort. Sci. 21: 75-82.

UNEP. 2001. Report of the Technology and Economic Assessment Panel. United Nations Environmental Program, Nairobi, Kenya.

Usman, M. and B. Fatima. 2013. Nursery sanitation in Citrus and Guava: A way forward. p. 1-22. In: M. Usman and B. Fatima (ed.). Citrus and Guava Nursery Raising: A practical approach. PAK TM publishers Faisalabad, Pakistan.

van Loenen, M.C.A., Y. Turbett, C.E. Mullins, N.E.H. Feilden, M.J. Wilson, C. Leifert and W.E. Seel. 2003. Low temperature-short duration steaming of soil kills soil-borne pathogens, nematode pests and weeds. Eur. J. Plant Pathol. 109: 993-1002.

Waller, P.L., C.R. Thornton, D. Farley and A. Groenhof. 2008. Pathogens and other fungi in growing media constituents. Acta Hort. 779: 361-365.

Washa, W.B.A., A.M.S. Nyomora and H.M.V. Lyaruu. 2012. Improving propagation success of *D. melanoxylon* (African blackwood) in Tanzania (II): Rooting ability of stem and root cuttings of *Dalbergia melanoxylon* (African blackwood) in response to rooting media sterilization in Tanzania. Tanz. J. Sci. 38: 35-42.

Yakabe, L.E. and J.D. MacDonald. 2010. Soil treatments for the potential elimination of *Phytophthora ramorum* in ornamental nursery beds. Plant Dis. 94: 320-324.

Yamamoto, T., U. Venecio, Jr. Ultra, S. Tanaka, K. Sakurai and K. Iwasaki. 2008. Effects of methyl bromide fumigation, chloropicrin fumigation and steam sterilization on soil nitrogen dynamics and microbial properties in a pot culture experiment. Soil Sci. Plant Nut. 54: 886-894.

Youpensuk, S., W. Piwpueak and B. Rerkasem. 2012. Effects of arbuscular mycorrhizal fungi on resistance to *Phytophthora parasitica* of citrus seedlings and on growth of Thai honey tangerine scions on citrus rootstocks. Afric. J. Biotech. 11: 11400-11406.