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Assessment of yield loss in Rosemary (*Rosmarinus officinalis* L.) and Sage (*Salvia officinalis* L.) plants caused by *Fusarium oxysporum*

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***Rosmarinus officinalis* and *Salvia officinalis* wilt caused by *Fusarium oxysporum* could lead to yield losses, under various conditions. This study was therefore focused on determination of yield loss caused by *F. oxysporum* on *R. officinalis* and *S. officinalis* plants. A greenhouse experiment was conducted at Wondo Genet College of Forestry and Natural Resources (WGCFNR), Ethiopia, from 2015 to 2016. Mixed soil of animal manure, sand and clay loam (1:1:2 ratio), respectively, was autoclaved at 121°C for 2 h and sterilized soil was filled into plastic pots with three replications. Two plants were transplanted into each pot and regularly watered and maintained in the greenhouse at 26 ± 2°C and 50 to 60% relative humidity. Disease severity was recorded at weekly interval, from the first appearance of symptoms till harvest. Fresh leaf and stem weights, essential oil content and yield were recorded at the time of harvesting. Generally, fresh leaf and stem weights and essential oil content and yield of both plants from treated soil significantly increased compared to the untreated ones. Essential oil yield decreased by 53.86 and 35.36% in *R. officinalis* and *S. officinalis,* respectively compared to the treated ones.**

Key words: Essential oil yield, *Fusarium oxysporum,* Rosemary, Sage, Yield loss

INTRODUCTION

Rosemary (*Rosmarinus officinalis L*.) is an aromatic and medicinal shrubby herb that belongs to the family Labiatae (Türe et al., 2009). It is endogenous to Europe, Asia and Africa, originally grows in southern Europe (Pintore et al., 2002). Its herb and oil are mostly used as spice and flavoring agents in food processing for its desirable flavor, and use folk medicinal value as antimicrobial agent (Lo et al., 2002). Rosemary is used traditionally for relieving visceral muscle spasms in renal colic, menstrual pain, bronchial asthma and gastrointestinal colic, all over the world (Singh et al., 2009; Tironi et al., 2009). In Ethiopia, rosemary, most of the time is used for flavoring food. It has also some therapeutic value in the treatment of disorders like, peptic ulcers, inflammatory diseases, hepatotoxicity, atherosclerosis, ischemic heart disease, cataracts, and cancer (Valenzuela et al., 2004; Katerinopoulos et al., 2005).

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As well, *Salvia officinalis L.* is a perennial low shrub aromatic and medicinal plant belonging to the family Lamiaceae which has about 900 species from genera *Salvia*, native to Mediterranean region (Zervoudakis et al., 2012). Essential oil is added as supplement to meat, sausage, poultry stuffings, fish, soups, canned foods and other food products. Its essential oil is applied in different pharmaceutical, cosmetics, sanitary and food industries owing to its odour and biological effects such as antiseptic, antibacterial, antioxidant activity, etc., (Bakkali et al., 2008).

However, Antracnosis, Ascochitosis, and root rot, caused by *Colletotrichum dematium*, *Ascochyta sclarea* and *Rhizoctonia solani*, respectively are economically important disease of sage in European countries (Subbiah et al., 1996; Voltolina, 2001). Similarly, *Phomopsis sclarea, Phodosphaera inequalis, Erysiphae polygoni* and *Sclerotinia sclerotiorum* are economically important yield loss causing pathogens to the plant in Italy, Spain, California and USA. Massive dying out of sage seedlings infected by *Fusarium oxysporum* was also observed in these countries (Subbiah et al*.,* 1996). *R. solani*, *Phytophthora citrophthora* and *P. cinnamoni* were reported as main root rot causal soil borne pathogens (Alvarez et al*.*, 2007; Verhoeven et al*.,* 2008). Therefore, integrated pest management development requires precise and accurate information on damage caused by pests. Disease damage reduces both quantity and quality of yield. Disease and crop loss assessments are necessary for identification of economic impact of a disease and to develop management and control strategies.

Fusarium species are the main soil borne plant pathogens that is economical important disease in agricultural productions all over the world (Saremi, 2000). The primary symptom of Fusarium wilt is characterized by discoloration of the vascular tissue which mostly starts from the ground and expands to the above parts, wilting the stem and leaves, sometimes followed by leaf abscission and plant death (Schwartz et al., 2005). Fusarium wilt caused by *F. oxporum* is economically an important soil borne disease which affects plant yield severely in susceptible varieties. Depending on crop age losses due to Fusarium wilt disease vary from 0 to 100% (Kannaiyan and Nene, 1981). Similarly, Fusarium wilt is an economically important prioritized disease of Rosemary and Sage plants that significantly affects the cultivation of these plants under various conditions at Wondo Genet. This study was therefore focused on determination of yield loss caused by *F. oxysporum* on *R. officinalis* and *S. officinalis* using sterilized and unsterilized soils under greenhouse conditions*.*

MATERIALS AND METHODS

Experiment site and yield loss estimation

The greenhouse experiment was conducted at Wondo Genet College of Forestry and Natural Resources (WGCFNR), Ethiopia, from 2015 to 2016.

Mixed soil of animal manure, sand and clay loam (1:1:2 ratio), respectively was autoclaved at 121°C for 2 h and 3 kg of sterilized soil, with pH 6.8, was filled into plastic pots (20 × 15 cm) with ten replications. Unsterilized soil of the same amount was used as control for both experiments. Two seedlings of each plant species were transplanted into the individual pot and regularly watered and maintained in greenhouse at 26 ± 2°C and 50 to 60% relative humidity. The plants were regularly observed for Fusarium wilt symptom development, characterized by discoloration of the vascular tissue. The symptom mostly starts from the ground, wilting the stem and leaves, sometimes followed by leaf abscission and plant death. Data were recorded on disease severity at weekly interval from the first appearance of symptom till harvest, 3 months after planting date. The disease severity was assessed based on visual assessment key (0 - 4 scale), where 0, 1, 2, 3 and 4 represent 0 to 24%, 25 to 49%, 50 to 74%, 75 to 99% and 100% (dead plant), respectively of wilted leaves. Plant height, branch number, fresh leaf and stem weights and essential oil content and yield were recorded at the time of harvesting. Percent EO content was determined on fresh weight (w/w) basis from 250 g of fresh composite leaves and EOY was calculated by multiplying the fresh weight biomass and the essential oil content. Essential oil was extracted by hydro distillation as illustrated by Guenther (1972).

The percent essential oil yield losses were calculated by the formula:

$$EOY\ loss\ (\%) = \frac{Yield\ of\ healthy\ plant - Yield\ of\ diseased\ palnt}{Yield\ of\ healthy\ plant}$$

Statistical analysis

The data were statistically analyzed using analysis of variance (ANOVA) and differences between treatments means was assessed using Duncan's Multiple Range Test, using SAS statistical software (SAS, 2002).

RESULTS AND DISCUSSION

The soil treatment had significant effects on essential oil content and yields and growth parameters of both Sage and Rosemary plants. Analysis of variance and mean comparison revealed significant differences ($P < 0.05$) in plant heights, branches numbers and essential oil content and yield between plants from treated and untreated soils in both experiments (Tables 1 and 2). Average plants heights from unsterilized soil were reduced by 19.7 and 14.9 cm in *R. officinalis* and *S. officinalis,* respectively. Similarly, branch numbers from untreated soil were reduced by 84.33 and 36.73% in *R. officinalis* and *S. officinalis,* respectively on unsterilized soil. Likewise, fresh leaf weights reduced by 17.3 kg/p (60.28%) and 7.21 kg/p (27.99%) in *R. officinalis* and *S. officinalis,* respectively compared with plants from treated soil (Tables 1 and 2).

On the other hand, 14.65 kg/p (46.14%) and 14.99 kg/p (64.64%) less essential oil yields were recorded in *R. officinalis* and *S. officinalis,* respectively from unsterilized soil. In the same way, less essential oil content, 40% in *R. officinalis* and 44.44% in *S. officinalis,* were recorded

Table 1. Effect of Fusarium wilt on growth parameters, essential oil content and yield on *R. officinalis*.

Treatments	Growth parameters				Oil yield	
	PH (cm)	BN	FLW (kg/p)	FSW (kg/p)	EOC (%)	EOY (kg/p)
Sterilized soil	52.5	45	28.7[a]	24.11[a]	2.5[a]	31.75[a]
Unsterilized soil	32.5	7	11.4[b]	14.09[b]	1.5[b]	17.1[b]
SD	42.5	26	20.05	19.1	2	24.43

Means with the same letter within the same column are not statistically different ($p < 0.05$). PH: Plant height, BN: branch number, FLW: fresh leaf weight, EOC: essential oil content, EOY: essential oil yield.

Table 2. Effect of Fusarium wilt on growth parameters, essential oil content and yield on *S. officinalis*.

Treatments	Growth parameters				Oil yield	
	PH (cm)	BN	FLW (kg/p)	FSW (kg/p)	EOC (%)	EOY (kg/p)
Sterilized soil	70.71	27.66	25.77[a]	27.76[a]	0.9[a]	23.19[a]
Unsterilized soil	56.25	17.5	18.56[b]	16.41[b]	0.5[b]	8.2[b]
SD	63.48	22.58	22.17	22.09	0.7	15.69

Means with the same letter within the same column are not statistically different ($p < 0.05$). PH: Plant height, BN: branch number, FLW: fresh leaf weight, EOC: essential oil content, EOY: essential oil yield.

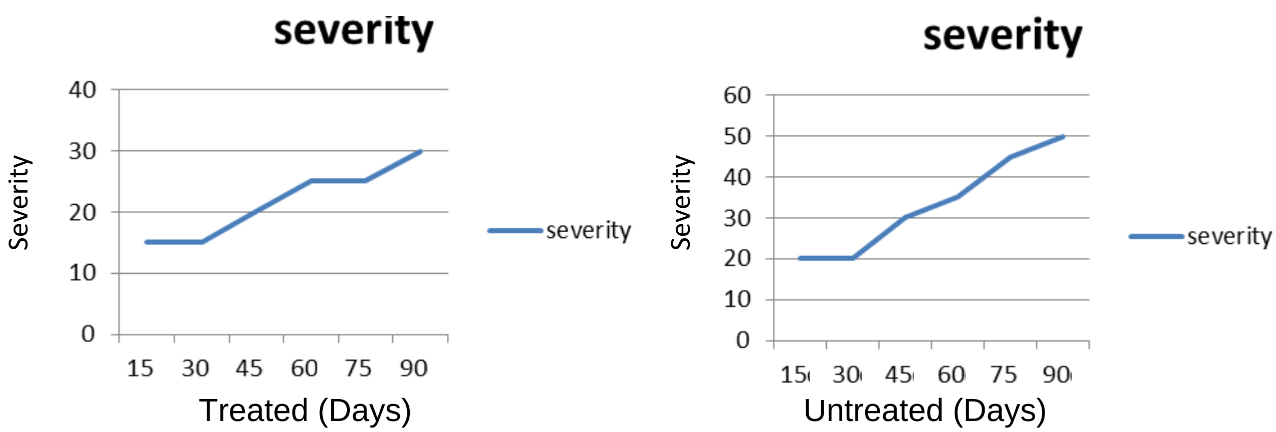

Figure 1. Disease severity of *R. officinalis* on sterilized and unsterilized soils.

in this treatment (Tables 1 and 2). Siddiqui and Akhtar (2007) reported 15 to 20% yield losses of chili caused by *F. oxysporum* in Pakistan. Similarly, significant losses in yield of Watermelon had been reported in Maryland due to the prevalence of Fusarium wilt (Zhou et al., 2003b). Losses can approach 100% if susceptible cultivars are planted in heavily infested fields (Egel et al., 2007).

Much higher disease severity levels (50%) were observed on both *R. officinalis* and *S. officinalis* than the severity levels (30%) of plants from treated soil (Figures 1 and 3). During the first two weeks, no disease symptoms were observed on both *R. officinalis* and *S. officinalis* from sterilized soil. However, after 20 days of planting disease severity gradually increased until the time of harvest. On the other hand, disease severity increased exponentially on *S. officinalis* after the first week of planting on untreated soil (Figures 1 and 2). This finding was in agreement with previous works (Sharma, 1994; Navas et al., 2000; Wharton et al., 2006; Saremi and Amiri, 2010).

For example, the disease severity rapidly progressed from 20 to 50% on both *R. officinalis* and *S. officinalis* on untreated soil. On the other hand, the disease severity increased gradually from 15 to 30% and from 10 to 30% on *R. officinalis* and *S. officinalis,* respectively on sterilized soil (Figures 1 and 3). The occurrence of Fusarium wilt disease on plants cultivated on sterilized soil may most likely be because of the spread of *F. oxysporum*, by irrigation water and contaminated management equipments, from adjacent plants cultivated

Treated Untreated

Figure 2. Sage and Rosemary under greenhouse condition.

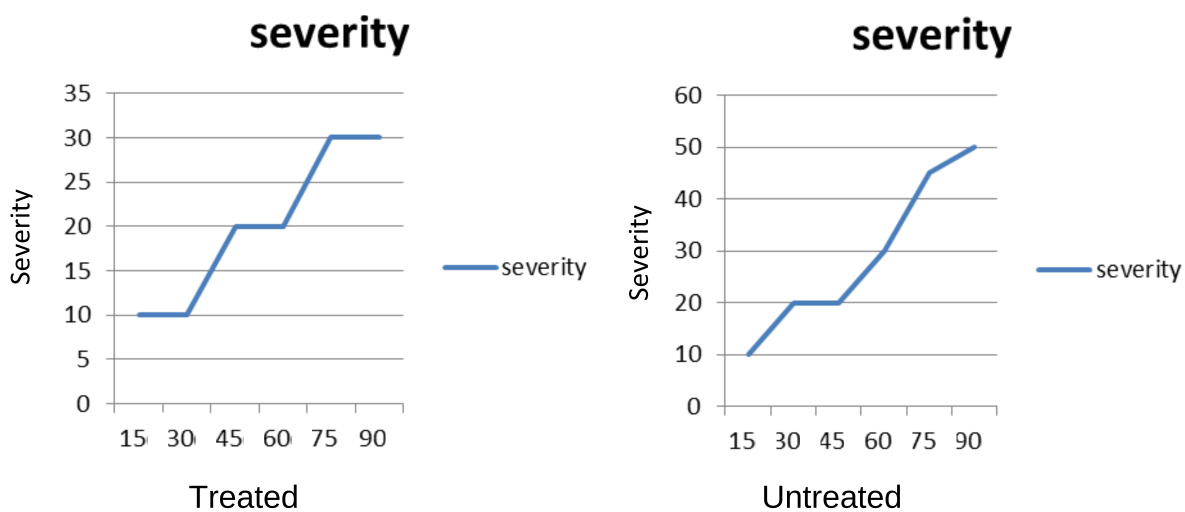

Figure 3. Disease severity of *S. officinalis* on sterilized and unsterilized soils.

on unsterilized soil. Similar result had been reported on *Chrysanthemum* Fusarium wilt, caused by *F. oxysporium.* Fusarium wilt was reported as the most widely spread and destructive disease, causing infection and yield loss from nursery to flowering stage on this plant (Locke et al., 1985).

Conclusion

The result of this study had revealed that soil treatment had a significant effect on growth parameters, essential oil content and yield of *R. officinalis* and *S. officinalis.* Essential oil yield reduced by 53.86 and 35.36% in *R. officinalis* and *S. officinalis,* respectively from untreated soil compared to the treated ones. Therefore, designing an integrated Fusarium wilt management method as alternative control measure for this disease is necessary.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interest

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