

Essential Oil Components of German Chamomile as Affected by Vermicompost Rates

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ABSTRACT: German chamomile belonging to Compositae family is one of the important medicinal plants. The active principles of chamomile essential oil are α -bisabolol and its oxides and azulenes, including chamazulene. Environmental factors can affect essential oil production. One of the most important environmental factors is growing medium. Vermicomposting is biotechnological process of composting. In this process, earthworms are used to enhance the process of residue conversion. In this study the treatments were different rates of cow manure vermicompost (0%, 2%, 4%, 6% and 8% w/w). The study was conducted using a completely randomized design (CRD) with three replications. Thirty one components were identified in chamomile oil. The major components were α -Bisabolol oxide A, Chamazulene, α -Bisabolone oxide A, En-in-dicycloether and α -Bisabolol oxide B. All treatments altered oil constituents.

Keywords: vermincomposted cow manure, *Matricaria recutita*, chamazulene, bisabolol oxide, organic matter.

INTRODUCTION

Medicinal and aromatic plants use by 80% of global population for their medicinal therapeutic effects as reported by WHO (2008). *Matricaria recutita* L. (syn. *M. chamomilla* L., *Chamomilla recutita* L. Rauschert) is known as true chamomile or German chamomile is from the Compositae family. German chamomile has white ligulate flowers, smells pleasantly of chamomile (typical chamomile smell) and is annual, grows 10 to 80 cm high. (Franke, 2005). Chamomile is widely used throughout the world. Its primary uses are as a sedative, anxiolytic and antispasmodic, and as a treatment for mild skin irritation and inflammation. It has widespread use as a home remedy (Gardiner, 1999). The biological activity of chamomile is mainly due to the phenolic compounds, primarily the flavonoids apigenin, quercetin, patuletin, luteolin and their glucosides, but also to the principal components of the essential oil extracted from the flowers like α -bisabolol and its oxides and azulenes, including chamazulene (Hadaruga et al., 2009). Environmental factors can affect essential oil production. One of the most important environmental factors is growing medium. Conventional farm systems have been characterized by high utilization of chemical fertilizer which decrease quality of soil and products due to reductions in soil organic matter content so, the use of organic matter as nutrient inputs is increasing for crop production with minimal environmental pollution (Singh et al., 2007; Liu et al., 2009; Padel et al., 2009). Vermicomposting is biotechnological process of composting. In this process, earthworms are used to enhance the process of residue conversion. Vermicomposting is faster than composting and the resulting earthworm castings are rich in microbial activity and plant growth regulators, and fortified with pest repellence attributes as well. Vermicomposting converts waste into compost during one months, reduces the C:N ratio and retains more N than the traditional methods of preparing composts so, vermicompost can improve seed germination, growth and yield of crops (Gandhi et al., 1997; Crescent, 2003; Nagavallema et al., 2004). Edwards and Arancon (2004), reported that the vermicompost applications suppressed the incidence of the disease significantly. By increasing levels of compost fertilizer to *Sideritis montana* L., vegetative growth and major components of essential oils increased (El-Sherbeny et al., 2005). The main objective of this experiment was to monitor the effects of vermicompost rates on essential oil components of German chamomile.

MATERIALS AND METHODS

Plant Material and Experimental Conditions

This experiment was carried out on a garden in Shiraz (29°38' N, 52°28' E; 1486 m above sea level), state of Fars, Iran, on September (beginning of autumn), 2011. The pots were filled up by a mixture contained 2/3 soil and 1/3 sand (v/v). The mixture of pots were tested before applying treatments and the texture was sandy clay loam with PH=8.48, organic C=0.29%, total N=0.03%, available P=0.9 mg/kg, available K=274 mg/kg, TNV=53.8% and EC=1.02 dS/m. The mixture of pots was treated with cow manure vermicompost at different rates (0%, 2%, 4%, 6% and 8% w/w). Analysis of vermicompost indicated that the texture was clay loam like with PH=7.54, N=1.57%, P=0.32%, K=0.78%, Cu=40 ppm, Zn=128 ppm, Fe=1850 ppm, Mn=358 ppm and EC=13.18 dS/m. Chamomile seeds were germinated in pots and thinned at 2-4 leaves stage to one plant per each pot. The experiment was conducted using a completely randomized design (CRD) with three replications. Each replicate contained 15 pots. The flower heads were collected each 15 days during one month (three times), and were dried at room temperature.

Essential Oil Extraction

Isolation of essential oils was performed using hydrodistillation of dried sample of flower heads using a Clevenger-type apparatus over 3 hours. The oils were dried over sodium sulphate.

Gas Chromatography (GC)

Gas Chromatography analysis was performed on an Agilent technologist model (7890A) equipped with flame ionization detector and capillary column HP-5 (30 m × 0.32 mm, 0.25 µm film thicknesses). The chromatographic conditions were as follows: The oven temperature increased from 60 to 210°C at a rate of 3°C/min then 210 to 240 °C at a rate of 20°C/min. The injector and detector temperatures were 280 and 290°C, respectively. N₂ used as the carrier gas (0.5 ml/min).

Gas Chromatography-Mass spectrometry (GC-MS)

Essential oil was also analysed by GC-MS (Agilent Technologies-5975C-MS, 7890A-GC) equipped with a HP-5 capillary column (phenyl methyl siloxane (30 m × 0.25 mm, 0.25 µm film thickness) with He as the carrier gas and a split ratio of 1:50. The retention indices for all the components were determined according to the Van Den Doll method using n-alkanes as standard. The compounds were identified by comparison of retention indices (RRI- AP-5) with those reported in the literature and by comparison of their mass spectra with the Wiley and mass finder 3 libraries or with the published mass spectra.

RESULTS AND DISCUSSION

The control plants could not grow and produce flowers properly. This can be due to low amount of phosphorous or high value of TNV in the growing medium of control plants. Qualitative and quantitative analyses of essential oils have been shown in Table 1. Thirty one components representing over 99% of chamomile oil composition were identified. The major components were α-Bisabolol oxide A (42.03-46.65%), Chamazulene (14.40-27.01%), α-Bisabolone oxide A (9.14-11.72%), En-in-dicycloether (7.63-8.85%) and α-Bisabolol oxide B (4.97-7.31%). Our data showed that the oil components were affected by vermicompost rates. The highest values of α-Bisabolol oxide B, α-Bisabolone oxide A, Chamazulene and α-Bisabolol oxide A were at 6%, 4%, 2% and 8% rate, respectively. Vermicompost at 2% rate increased chamazulene by 87.57% when compared to 6% rate. Application of 8% rate increased α-Bisabolol oxide A by 10.99% when compared to 2% rate. In our study linalool decreased from 4% to 8% vermicompost rate. According Chand et al. (2011), linalool content decreased with increasing in the organic source of nutrient in Plargonium species.

Table 1. The chemical components of chamomile oil in vermicompost treatments

NO	RI	Compound name	Vermicompost (2%)	Vermicompost (4%)	Vermicompost (6%)	Vermicompost (8%)
1	935	α-Pinene	-	t	0.10 ± 0.02	0.06 ± 0.01
2	947	Camphene	-	0.05 ± 0.00	0.14 ± 0.00	0.12 ± 0.01
3	975	Sabinene	0.12 ± 0.01	0.44 ± 0.01	2.01 ± 0.10	1.35 ± 0.01
4	992	Myrcene	-	-	0.17 ± 0.01	0.14 ± 0.00
5	1003	Yomogi alcohol	-	0.11 ± 0.01	0.20 ± 0.01	0.12 ± 0.00
6	1006	n-Octanal	0.07 ± 0.00	0.40 ± 0.51	0.23 ± 0.00	0.21 ± 0.00
7	1026	p-Cymene	0.08 ± 0.00	0.15 ± 0.00	0.42 ± 0.01	0.42 ± 0.00

8	1030	Limonene	-	-	0.10 ± 0.00	0.09 ± 0.01
9	1037	1,8-Cineole	0.18 ± 0.01	0.33 ± 0.01	0.78 ± 0.02	0.62 ± 0.00
10	1047	(E)-β-Ocimene	0.38 ± 0.00	0.49 ± 0.03	1.23 ± 0.02	0.94 ± 0.00
11	1059	γ-Terpinene	0.16 ± 0.00	0.29 ± 0.02	0.71 ± 0.00	0.65 ± 0.00
12	1064	Artemisia ketone	0.95 ± 0.03	1.65 ± 0.46	3.02 ± 0.19	2.43 ± 0.00
13	1087	Artemisia alcohol	0.21 ± 0.01	0.43 ± 0.01	0.81 ± 0.02	0.58 ± 0.01
14	1104	Linalool	t	0.18 ± 0.23	0.14 ± 0.02	0.09 ± 0.00
15	1107	n-Nonanal	0.08 ± 0.00	-	0.16 ± 0.00	0.15 ± 0.00
16	1163	(2E)-Nonen-1-al	0.06 ± 0.00	-	-	0.07 ± 0.02
17	1171	Terpinene-4-ol	0.12 ± 0.00	0.26 ± 0.04	0.44 ± 0.01	0.32 ± 0.01
18	1196	α-Terpineol	0.06 ± 0.01	0.13 ± 0.00	0.22 ± 0.01	0.17 ± 0.02
19	1394	β-Elemene	0.13 ± 0.00	0.09 ± 0.01	0.12 ± 0.01	0.13 ± 0.01
20	1422	(E)-Caryophyllene	0.13 ± 0.00	0.11 ± 0.00	0.20 ± 0.00	0.17 ± 0.01
21	1458	(E)-β-Farnesene	2.12 ± 0.04	1.37 ± 0.01	1.99 ± 0.17	2.33 ± 0.12
22	1484	Germacrene D	0.63 ± 0.09	0.43 ± 0.02	0.38 ± 0.01	0.50 ± 0.04
23	1499	Bicyclogermacrene	0.41 ± 0.05	0.31 ± 0.01	0.38 ± 0.01	0.36 ± 0.01
24	1567	(E)-Nerolidol	0.63 ± 0.02	0.64 ± 0.03	0.63 ± 0.03	0.70 ± 0.08
25	1583	Spathulenol	0.62 ± 0.02	0.60 ± 0.01	0.77 ± 0.04	0.86 ± 0.06
26	1589	Caryophyllene oxide	0.12 ± 0.00	-	-	0.12 ± 0.00
27	1663	α-Bisabolol oxide B	4.97 ± 0.06	6.00 ± 0.24	7.31 ± 0.20	6.59 ± 0.24
28	1692	α-Bisabolone oxide A	9.38 ± 0.19	11.72 ± 0.53	11.65 ± 0.59	9.14 ± 0.02
29	1740	Chamazulene	27.01 ± 0.33	22.19 ± 0.14	14.40 ± 0.46	15.61 ± 0.35
30	1763	α-Bisabolol oxide A	42.03 ± 0.18	42.45 ± 0.37	43.12 ± 0.01	46.65 ± 0.45
31	1888	En-in-dicycloether	8.70 ± 0.40	8.85 ± 0.37	7.63 ± 0.33	7.92 ± 0.58
Total (%)			99.39	99.70	99.47	99.59

RI, retention index, All data are means of three replications ± standard deviation, All data show as percent (%)
t, trace (<0.05)

In sweet fennel, the highest anethole content and the lowest contents of fenchone, limonene and estragole of essential oil were obtained in a treatment contained vermicompost (Moradi et al., 2011).

Vermicompost provides all nutrients in readily available form and also enhances uptake of nutrients by plants (Nagavallema et al., 2004). The uptake of nitrogen, phosphorus, potassium and magnesium can improve when fertilizer was applied in combination with vermicompost (Jadhav et al., 1997). Atiyeh et al. (2000), found that compost was higher in ammonium, while vermicompost tended to be higher in nitrates, which is the more plant-available form of nitrogen. It has been reported that applying nutrients can alter oil constituents (Sharafzadeh et al., 2011). N, P and K play important role in essential oil biosynthesis. Besides production of pyruvate, these elements are present in ATP, NADP and CoA structure which terpenoid biosynthesis depend on such coenzymes (Sell, 2003; Kapoor et al., 2004).

The effects of the vermicompost may not only depend on its chemical compounds and physiological properties, but also on its effects on soil physical properties (Wang et al., 2010). It improves soil structure, texture, aeration, and waterholding capacity. The application of vermicompost favorably affects soil pH, microbial population and soil enzyme activities (Maheswarappa et al. 1999) which all of them can affect biosynthesis of compounds. Vermicompost and organic fertilizers increased protein content of peanut and vitamin C in marionberry, strawberry, and corn (Asami et al., 2003; Basu et al., 2008). Phenolic compounds are a large group of plant secondary metabolites. Increases in the levels of phenols have been reported in strawberries and marionberries treated with organic fertilizers (Asami et al., 2003).

vermicompost includes plant-growth regulators which increase growth and yield (Canellas et al, 2002). Excreta of earthworm were rich of Micro-organism especially bacteria and contain large amounts of plant hormones (auxin, gibberellin and cytokinin) which affect plant growth and development (Atiyeh and et al, 2001). One of the factors influencing essential oil production is plant growth regulators or plant hormones. Endogenous levels as well exogenous application could affect essential oil production and chemical composition (Prins et al., 2010). Researchers proposed that the application of growth regulators which affect oil components be due to their effects on enzymatic pathways of terpenoid biosynthesis (Sangwan et al., 2001).

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